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Recombination and Point Mutations in Type G Rotavirus Strains: The Challenges of Vaccine Development

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1. Introduction

Active immunity refers to the process of exposing the body to an antigen to generate an adaptive immune response: the response takes days/weeks to develop but may be long lasting—even lifelong. Wild infection with pathogenic agents (eg. Hepatitis A Virus) and subsequent recovery gives rise to a natural active immune response usually leading to lifelong protection. In addition, some infections can be prevented by immunization with vaccines.

The term "vaccine" is derived from the Latin word "vaccinus" which means "pertaining to cows" – a reflection on Jenner's pioneering studies using cowpox vaccinia virus to prevent human smallpox (variola) as discussed previously (Stefan, 2005; Dunn, 1996). Vaccines take advantage of using relatively harmless foreign agents to evoke protective immunity for protection against several important pathogens. Vaccine development has its early roots in the work of Edward Jenner and Louis Pasteur, who discovered how to protect people from smallpox and developed a vaccine to protect from rabies, respectively.

All vaccines contain other substances (termed excipients) that are present because they improve the immune response (an adjuvant), are necessary for ensuring stability of the product (stabilizers and preservatives), are the vehicle for delivering vaccine (carrier) or are a residual of the manufacturing process (for example antibiotics or cell culture components).

Nowadays, many types of vaccines have been proposed and used in vaccine development: Live whole virus vaccines, Killed whole virus vaccines, Subunit vaccines (purified or recombinant viral antigen), Toxoid vaccines, Synthetic vaccines, and DNA vaccines. We will discuss these vaccine types one-by-one.

1.1 Live, attenuated virus vaccines

They are prepared from attenuated strains that are almost or completely devoid of pathogenicity but are capable of inducing a protective immune response. They multiply in the human host and provide continuous antigenic stimulation over a period of time. Primary vaccine failures are uncommon and are usually the result of inadequate storage or administration. Another possibility is interference by related viruses as is suspected in the case of oral polio vaccine in developing countries (Giammanco et al., 1988; Drozdov & Shirman, 1961; Katz & Plotkin, 1968). Several methods have been used to attenuate viruses for vaccine production such as the use of a related virus from another animal (cowpox to prevent smallpox), the administration of pathogenic or partially attenuated virus by an unnatural route, passage of the virus in an "unnatural host" or host cell (e.g. the 17D strain of yellow fever was developed by passage in mice and then in chick embryos (Norrby, 2007) and Polioviruses were passaged in monkey kidney cells (Chezzi et al., 1998) and measles in chick embryo fibroblasts (Katz, 1958), and the development of temperature sensitive mutants (Pringle, 1996).

1.2 Killed/Inactivated vaccines

The term killed generally refers to bacterial vaccines, whereas inactivated relates to viral vaccines (Levine et al., 1997). They were the easiest preparations to use. The preparation was simply inactivated. For viruses, the outer virion coat should be left intact but the replicative function should be destroyed. To be effective, non-replicating virus vaccines must contain much more antigen than live vaccines that are able to replicate in the host. Preparation of killed vaccines may take the route of heat or chemicals (Turner et al., 1970). The chemicals used include formaldehyde or beta-propiolactone (Lo Grippo, 1960; Gard, 1960). The traditional agent for inactivation of the virus is formalin (Weil & Gall, 1940; Kim & Sharp, 1967). Excessive treatment with this detergent can destroy immunogenicity whereas insufficient treatment can leave infectious virus capable of causing disease. Soon after the introduction of inactivated polio vaccine, there was an outbreak of paralytic poliomyelitis in the USA due to the distribution of inadequately inactivated polio vaccine (Prevots et al., 1996). This incident led to a review of the formalin inactivation procedure and other inactivating agents are available, such as beta-propiolactone. Another problem was that SV40 was occasionally found as a contaminant and there were fears of the potential oncogenic nature of the virus (Tam et al., 2004).

1.3 Subunit vaccines

Originally, non-replicating vaccines were derived from crude preparations of virus from animal tissues. As the technology for growing viruses to high titres in cell cultures advanced, it became practical to purify virus and viral antigens. It is now possible to identify the peptide sites encompassing the major antigenic sites of viral antigens, from which highly purified subunit vaccines can be produced. Increasing purification may lead to loss of immunogenicity, and this may necessitate coupling to an immunogenic carrier protein or adjuvant, such as an aluminum salt. Examples of purified subunit vaccines include the HA vaccines for influenza A and B (Bachmayer et al., 1976), and HBsAg derived from the plasma of carriers (Vyas et al., 1984). Subunit vaccines can be further subdivided into those where the antigen is produced using recombinant DNA technology and those based on normal bacteriological growth processes.

Virus proteins have been expressed in bacteria, yeast, mammalian cells, and viruses. *E. Coli* cells were first to be used for this purpose but the expressed proteins were not glycosylated, which was a major drawback since many of the immunogenic proteins of viruses such as the envelope glycoproteins, were glycosylated. Nevertheless, in many instances, it was demonstrated that the non-glycosylated protein backbone was just as immunogenic. Recombinant hepatitis B vaccine is the only recombinant vaccine licensed at present (Yap et al., 1992). An alternative application of recombinant DNA technology is the production of hybrid virus vaccines. The best known example is vaccinia (Smith et al., 1983). The recombinant virus vaccine can then multiply in infected cells and produce the antigens of a wide range of viruses. The genes of several viruses can be inserted, so the potential exists for producing polyvalent live vaccines (Hauser et al., 1988; Hilleman, 1987). HBsAg, rabies, HSV and other viruses have been expressed in vaccinia (Mackett et al., 1985; Panicali et al., 1983; Paoletti et al., 1984; Perkus et al., 1985; Rice et al., 1985; Smith et al., 1983; Kieny et al., 1984; Wiktor et al., 1984).

1.4 Toxoid vaccines

Certain pathogens cause disease by secreting an exotoxin: these include tetanus, diphtheria, botulism and cholera. For these bacteria that secrete toxins, or harmful chemicals, a toxoid vaccine might be the answer. These vaccines are used when a bacterial toxin is the main cause of illness. Scientists have found that they can inactivate toxins by treating them with formalin, a solution of formaldehyde and sterilized water. Such "detoxified" toxins, called toxoids, are safe for use in vaccines. When the immune system receives a vaccine containing a harmless toxoid, it learns how to fight off the natural toxin. The immune system produces antibodies that lock onto and block the toxin. Vaccines against diphtheria and tetanus are examples of toxoid vaccines (Bizzini et al., 1970; Alouf, 1987). These vaccines are: safe because they cannot cause the disease they prevent as there is no possibility of reversion to virulence; the vaccine antigens are not actively multiplying, they cannot spread to unimmunized individuals; they are usually stable and long lasting as they are less susceptible to changes in temperature, humidity and light which can result when vaccines are used out in the community.

1.5 Synthetic peptides

The development of synthetic peptides that might be useful as vaccines depends on the identification of immunogenic sites (Milich, 1990; Hans et al., 2006; Dorothea, 1993; Jonathan, 1987). Synthetic peptide vaccines have been successfully developed for the immunoprophylaxis of infection with foot-and-mouth disease virus (Bittle et al., 1982; Brown, 1990), type A influenza virus (Muller et al., 1982), and poliovirus (Emini et al., 1983). Synthetic peptide vaccines would have many advantages. Their antigens are precisely defined and free from unnecessary components which may be associated with side effects. They are stable and relatively cheap to manufacture. Changes due to natural variation of the virus can be readily accommodated, which would be a great advantage for unstable viruses.

1.6 DNA vaccines

The demonstration by Wolff and colleagues in 1990 (Wolff et al., 1990) that protein could be expressed following direct inoculation of plasmid DNA into muscle tissue unveiled an

exciting, new era in vaccinology and gene therapy. DNA-based vaccination offers a number of advantages over other methods of immunization. It is particularly attractive compared to conventional administration of a preformed protein antigen (Ag) because the immunogen is actively synthesized de novo in cells transfected with DNA. The principle of DNA vaccination has been demonstrated for a variety of bacterial, viral and parasitic diseases (Ulmer et al., 1993). Immune responses have been generated by DNA vaccination against a very wide variety of viral, bacterial and protozoal pathogens and toxins (Donnelly et al., 1994; King et al., 1998). Immune responses against influenza viruses have been demonstrated in chickens, mice, ferrets and non-human primates. For humans, the major concern about DNA vaccines is whether the plasmid DNA integrates into the genome randomly, potentially leading to insertional mutagenesis. In addition, the formal acceptance of this novel technology as a new modality of human vaccines depends on the successful demonstration of its safety and efficacy in advanced clinical trials. Several trials evaluated the efficacy of a DNA vaccine targeting human immunodeficiency virus type 1 (HIV-1) for therapeutic and prophylactic applications (MacGregor et al. 1998). However, the results of these early clinical trials were disappointing. The DNA vaccines were safe and well tolerated, but they proved to be poorly immunogenic.

Vaccines may be monovalent (also called univalent) or multivalent (also called polyvalent). A monovalent vaccine is designed to immunize against a single antigen or single microorganism. A multivalent or polyvalent vaccine is designed to immunize against two or more strains of the same microorganism, or against two or more microorganisms. In certain cases a monovalent vaccine may be preferable for rapidly developing a strong immune response.

2. Rotavirus infection and common associated-genotypes

Infectious acute diarrhea is a significant cause of morbidity and mortality of infants in developing and developed countries and constitutes a major public health problem worldwide. It is estimated that in developing countries (in Africa, Asia and Latin America) 744 million to 1 billion cases of diarrhea and 2.4 to 3.3 million deaths occur annually among children less than 5 years of age, corresponding to 6600 to 9000 deaths per day (Linhares & Bresee, 2000). The viruses are the most common aetiology of these diseases, especially in developed countries, where they cause more than 80% of the cases of acute diarrhea. The most common viral causes of gastroenteritis are rotaviruses and calicivirus (norovirus).

Rotavirus, a member of the *Reoviridae* family, is a highly contagious virus that causes severe and acute dehydrating diarrhea in infants and young children as well as other young animals worldwide (Dhama et al., 2009; Kapikian et al., 2001). Mature rotavirus, non-enveloped virions, contains an 11-segmented, double-stranded RNA (dsRNA) genome enclosed in a triple-layered protein capsid (Hoshino & Kapikian, 2000). The segmented nature of the genome allows rotaviruses to reassort *in vitro* and *in vivo* (Greenberg et al., 1981; Kalica et al., 1981; Gombold & Ramig, 1986).

A dual nomenclature has been used to differentiate rotavirus strains based on their serotype specificities, which are carried by the two outer capsid antigens, VP7 and VP4 (Estes & Kapikian, 2007).

The high disease burden motivated major efforts to develop a suitable rotavirus vaccine. However, the vaccine efficacy is being challenged by the extensive strain diversity of the rotaviruses (Estes, 2001; Green et al., 1987, 1988; Hoshino et al., 1994; Kapikian et al., 2001; Linhares et al., 1999).

Reverse-transcription polymerase chain reaction (RT-PCR) is the most widely used method for rotavirus characterization in surveillance studies. Molecular methods have allowed the detection of many rotavirus G-types (Banyai et al., 2003; Cubitt et al., 2000; Cunliffe et al., 1999; Das et al., 1993a, 2003; Gentsch et al., 1996; Gouvea et al., 1994; Pongsuwanna et al., 2002). Because of the natural variation in the rotaviral gene sequences, G-type-specific-primer based RT-PCR led to the genotyping failure (Adah et al., 1997; Iturriza- Gómara et al., 2000, 2004a; Maunula & von Bonsdorff, 1998; Rahman et al., 2005b). In addition, the accumulation of point mutations through genetic drift at the type-specific primer binding sites has resulted in failures to type strains or in mistyping. For example, the accumulation of point mutations at the G9 type-specific primer binding site was reported as having an impact for the efficient genotyping of rotaviruses (Martella et al., 2004; Santos et al., 2003). Moreover, some nucleotide identity between genotypes lead to primer cross-reactivity between rotavirus strains as the case of rotavirus G3 and G10 strains using primers developed by Gouvea et al. (1990) as discussed previously (Iturriza-Gómara et al., 2004b).

In order to overcome this problem, a modified classification system for VP4, VP7, and NSP4, and a novel classification system for VP1, VP2, VP3, VP6, NSP1, NSP2, NSP3, and NSP5/6 were proposed to be used for international standardization and implementation (Matthijnssens et al., 2008a, 2008b).

Actually, VP4 and VP7 are the main targets for vaccine development strategies. In the present review we will focus on the diversity of VP7 among rotavirus strains and the effect of this variability upon vaccine development.

Similar to most of the group A rotaviruses, the VP7 nucleotide sequence is 1062 nucleotides long. The ORF starts at nucleotide 49 with an AUG (ATG) start codon and ends at nucleotide 1029 with a UAG (TAG) termination codon, comprising 981 nucleotides (Estes & Cohen, 1989; Bellamy & Both, 1990).

The gene segment coding for the VP7 glycoprotein is the basis for genotyping group A rotaviruses into at least fifteen G-genotypes. Studies of intragenotype diversity led to subdivision of the G genotypes into several lineages (two major lineages, designated I and II) and sublineages, distinctly identified by unique genomic as well as epidemiological features. Lineage I was further subdivided into four sublineages, Ia–Id.

Genotypes G1, G2, G3, G4 and G9 are the most common G-types in humans (Gentsch et al., 1996; Liprandi et al., 2003; Martella et al., 2003; Okada et al., 2000; Rao et al., 2000; Sereno & Gorziglia, 1994). Nevertheless, over past decades, type G1 rotaviruses have been the most widespread genotype causing acute gastroenteritis in children from many countries covering all continents of the world (Santos & Hoshino, 2005). Type G2 rotavirus represents a different genogroup which appears to have a cyclic pattern of occurrence and yet little information is available about its genetic variability. Type G3 rotavirus have been found in a broad range of host species, including humans, monkeys, dogs, cats, horses, rabbits, mice, sheep and pigs (Martella et al., 2001; Andrej et al., 2008; Hoshino et al., 1984; Paul et al.,

1988; Fitzgerald et al., 1995). The G9 rotavirus was first reported in the United States in the early 1980s (Clark et al., 1987). It represents the fifth most common G genotype of rotavirus infections throughout the world (Gentsch et al., 2005; Santos & Hoshino, 2005, Khamrin et al., 2006).

3. Rotavirus vaccines

3.1 Monovalent animal and human rotavirus vaccines

Monovalent animal rotavirus vaccines. Research to develop a safe, effective rotavirus vaccine began in the mid-1970s, when investigators demonstrated that previous infection with animal rotavirus strains protected laboratory animals from experimental infection with human rotaviruses (Zissis et al., 1983). Researchers thought that live animal strains that were naturally attenuated for humans, when given orally, might mimic the immune response to natural infection and protect children against disease. Three nonhuman rotavirus vaccines, two bovine rotavirus strains, RIT 4237 (P6[1]G6) and WC3 (P7[5]G6), and a simian (rhesus) rotavirus reassortant vaccine (RRV) strain (P[3]G3), were studied (Christy et al., 1988; Clark et al., 1988; Vesikari et al., 1984). These vaccines demonstrated variable efficacy in field trials and gave particularly disappointing results in developing countries (Hanlon et al., 1987; Lanata et al., 1989; Penelope, 2008). Another monovalent, ovine strain vaccine produced by the Lanzhou Institute and licensed in China in 2000 (Lanzhou lamb rotavirus vaccine (LLR); P[12], G10) was available in some parts of China (World Health Organization [WHO], 2000). Few data are available about the effectiveness of this vaccine and it was not included in national immunization programs in China or elsewhere. Finally, monovalent animal strain vaccines have been mostly abandoned.

Monovalent human rotavirus vaccines. Rotarix®, developed by GlaxoSmithKline Biologicals, Belgium, is a monovalent, P1A[8] G1 rotavirus derived from a human G1 strain (89-12) that yielded high efficacy in early trials in the US and Finland (Bernstein et al., 2002). Its Efficacy has been confirmed in many countries (Ruiz-Palacios et al., 2006; De Vos et al., 2004).

RV3 neonatal strain vaccine, a P2A[6] G3 strain, was first isolated from newborns at the Children's Hospital in Melbourne, Australia (Barnes et al., 1997). Neonates infected with this rotavirus strain in hospital nurseries usually were asymptomatic and were later protected against severe disease in early childhood. However, serum immune responses were poor (Das et al., 1993a). Many attempts were undertaken to increase the titer of this vaccine and return to clinical trials.

Another two Indian neonatal strain vaccines 116E and I321 were proposed as candidate vaccines (Das et al., 1993a, 1993b; Iturriza-Gómara et al., 2004a). Both strains are in preclinical development and human trials are being planned in India, but with the new finding of I321-like strains causing disease in children, both careful epidemiological studies and safety monitoring will be essential prior to licensure.

3.2 Polyvalent rotavirus vaccines

In view of the inconsistency of protection from monovalent animal rotavirus-based vaccines, vaccine development efforts began to use either naturally attenuated human rotavirus strains or reassortant rotavirus strains bearing a human rotavirus gene for the VP7 protein

together with the other 10 genes from an animal rotavirus strain (Midthun & Kapikian, 1996). The next generation of vaccines was formulated to include more than one rotavirus G serotype to provide heterotypic as well as homotypic immunity. The ability of rotaviruses to reassort during mixed infections in vitro allowed the production of reassortant vaccines, termed the "modified Jennerian" approach (Kapikian et al., 1996b). Reassortant viruses contain some genes from the animal rotavirus parent and some genes from the human rotavirus parent. VP7 was thought to be important for protection; therefore, human-animal reassortant rotaviruses for use as vaccines included human VP7 genes to provide protective immune responses.

Quadrivalent RRV-based rhesus-human reassortant vaccine. A RotaShield was the first multivalent live oral reassortant vaccine (tetravalent, reassortant rhesus-human rotavirus vaccine, [RRV-TV]) contained a mixture of four virus strains representing the most commonly seen G types, G1 to G4: three rhesus-human reassortant strains containing the VP7 genes of human serotypes G1, G2, and G4 strains were substituted for the VP7 gene of the parent RRV, and the fourth strain comprised serotype G3 of rhesus RRV (Kapikian et al., 1996a). RRV-TV was extensively evaluated in field trials in the United States, Finland, and Venezuela and proved highly effective (80 to 100%) in preventing severe diarrhea due to rotavirus in each of these settings (Joensuu et al., 1997; Perez-Schael et al., 1997; Rennels et al., 1996; Santosham et al., 1997). Due to the proven efficacy, the RRV-TV vaccine was licensed in August 1998 for routine use in children in the United States at 2, 4, and 6 months of age (Centers for Disease Control and Prevention [CDCP], 1999b). Later, this vaccine was withdrawn from the market in 1999 as a consequence of vaccine-associated intussusception in several cases of vaccinated infants (CDCP, 1999a).

Pentavalent WC3-based bovine-human reassortant vaccine. Rotateq®, manufactured by Merck, Inc, USA, is a pentavalent vaccine containing five reassortants representing the common human VP7 types, G1-4 and the most common VP4 type, P[8] (CDCP, 2006). A large efficacy trial with Rotateq® has been completed, which found 74 and 98% efficacy against all and severe disease, respectively and has efficacy against each of the common circulating serotypes. Compared with the rhesus reassortants, the bovine-human reassortants appear to cause less fever while maintaining immunogenicity (Clark et al., 2004). A large safety trial found no evidence of an increased risk of intussusceptions among vacinees compared with placebo recipients (Vesikari et al., 2006).

Both RotaTeq and RotaRix have been shown to be effective against rotavirus gastroenteritis, however on March 22, 2010 the Food and Drug Administration (FDA) recommended that the use of the Rotarix vaccine be suspended in the United States because of some DNA from a porcine (pig) virus (porcine circovirus type 1) detected in the vaccine. Subsequently, some DNA from this and another porcine virus were also detected in Rotateq. On May 14, 2010 the FDA updated their recommendations for the use of rotavirus vaccines based on a review of the literature and the input from experts. The RotaTeq vaccine was proven to be effective in many countries such as Finland (Vesikari et al., 2010).

Whatever the type of vaccine and strategy of its development, the introduction of a new vaccine faces many hurdles, including cost, production capabilities, safety, and other programmatic issues. For rotavirus vaccines, while there is clearly a need, there are also additional challenges raised by the emergence of new rotavirus genotypes.

4. The prevalence of uncommon G-type rotaviruses and challenges for vaccine development

Genetic variability has been observed for all RNA viruses examined, and their potential for rapid evolution is increasingly recognized as the basis of their ubiquity and adaptability (Holland et al., 1992; Kilbourne, 1991). The molecular mechanisms underlying RNA virus variations are: mutation, homologous and non homologous recombinations, and genome reassortment in viruses with a segmented genome such as reoviruses. The genetic evolution of viruses is an important aspect of the epidemiology of viral diseases and sometimes causes problems in the development of successful vaccines.

The effectiveness of rotavirus vaccines will be dependent upon the immunity conferred against prevalent and emergent variants causing severe diarrhoeal disease. The global effort toward the prevention of rotavirus disease to be successful, special efforts will be required in countries where new genotypes were detected such as G5, G6, G8, G10, G11, and G12 (Figure 1). Nucleotide analysis using CLUSTAL X (version 1.8) of VP7 gene of these uncommon rotaviruses showed high degree of variability with the common G-type viruses (Figure 2). Genomic similarities between rotaviruses from different animal species are regarded as evidence of interspecies transmission of rotaviruses that may occur as a whole virion or genetic reassortment. The high variability of viral sequences due to genetic reassortment and nucleotide substitution are considered the most important mechanisms of evolution for rotaviruses. The antigenic variation within a serotype was known as a mechanism by which variants of rotavirus emerge to escape host immunity. This variability represents considerable potential for impaired vaccine efficacy.

The available information in literature showed that type G5 rotavirus is an important and commonly detected pathogen of swine and has also been identified in equine (Kapikian et al., 2001). However, in 1994, Gouvea and collaborators first demonstrated the occurrence of rotavirus genotype G5 among Brazilian children with diarrhea (Gouvea et al., 1994; Timenetsky et al., 1997). The detection of rotavirus G5 among children with diarrhea has also been reported in Argentina and Paraguay, indicating the spread of this virus across South America (Coluchi et al., 2002; Bok et al., 2001). In addition, the detection of type G5 rotavirus was reported in Cameroon (Esona et al., 2004). Another genotypes, type G6 and G10 strains have been isolated from humans (Dunn et al., 1993; Gerna et al., 1992, 1994; Armah et al., 2010). Although type G6 is the commonest rotavirus G type found in cows and at low frequency in sheep and goats (Kapikian et al., 2001), it was detected from hospitalized children with acute gastroenteritis in Italy during 1987–1988 (Gerna et al., 1992), Australia (Palombo & Bishop, 1995; Cooney et al., 2001), India (Kelkar & Ayachit, 2000), USA (Griffin et al., 2002), Belgium (Rahman et al., 2003), and Hungary (Banyai et al., 2004; Banyai et al., 2003). Type G8 virus, which can be found in cows at relatively high frequency (Kapikian et al., 2001), was first isolated in a study performed between 1979 and 1981, from stool specimens collected from children with diarrhea in Jakarta and Medan (Indonesia) (Hasegawa et al., 1984), Kenya (Nokes et al., 2010), and other countries such as Finland, Italy, Nigeria, Brazil, Malawi, South Africa, Egypt, Australia, the United States, and the United Kingdom (Adah et al., 1997, 2001; Cunliffe et al., 1999; Cunliffe et al., 2000; Gerna et al., 1990; Holmes et al., 1999; Palombo et al., 2000; Parwani et al., 1993; Rao et al., 2000; Santos et al., 1998; Steele et al., 1999). Type G11 rotaviruses are believed to be circulating in pigs, albeit in low numbers mainly in Mexico in 1983 and in Venezuela in 1989 (Ciarlet et al.,

1994; Ruiz et al., 1988). Later, several reports have described the detection of G11 rotavirus strains from humans in India (Banerjee et al., 2007), Bangladesh (Rahman et al., 2005a; Rahman et al., 2007), Nepal (Uchida et al., 2006), Ecuador (Banyai et al., 2009), and South Korea (Hong et al., 2007). Type G12 rotavirus was detected in stool specimens collected from children with diarrhea in the Philippines (Taniguchi et al., 1990), Thailand (Pongsuwanna et al., 2002), USA (Griffin et al., 2002), India (Das et al., 2003), Japan (Shinozaki et al., 2004), Korea (Cheon et al., 2004), Argentina (Castello et al., 2004), Malawi (Cunliffe et al., 2009), and Saudi Arabia (Kheyami et al., 2008). Type G12 has not been detected in animals other than humans.

Furthermore, recombination between human and animal rotavirus constitutes another challenge for vaccine development. Many G-type rotaviruses are considered to be reassortants between human and bovine viruses such as the case of G8 rotaviruses (Browning et al., 1992; Ohshima et al., 1990; Adah et al., 2003). Reassortment among bovine, porcine and human rotavirus strains was also reported (Park et al., 2011).

Other important issues need to be discussed concerning the potential that the vaccine strains themselves may either cause disease or reassort with wild-type rotavirus to produce a virulent strain as reported for RotaTeq reassortant strain in association with acute gastroenteritis (Payne et al., 2010). In addition, the predominance of G2 rotaviruses in Brazil following the introduction of Rotarix (Gurgel et al., 2007; Nakagomi & Nakagomi, 2009) and the predominance of G3 rotaviruses after vaccine introduction in USA after a Surveillance From 2005 to 2008 (Hull et al., 2011) may enhance the study the effectiveness of the available vaccines and the growing need to evaluate their use.

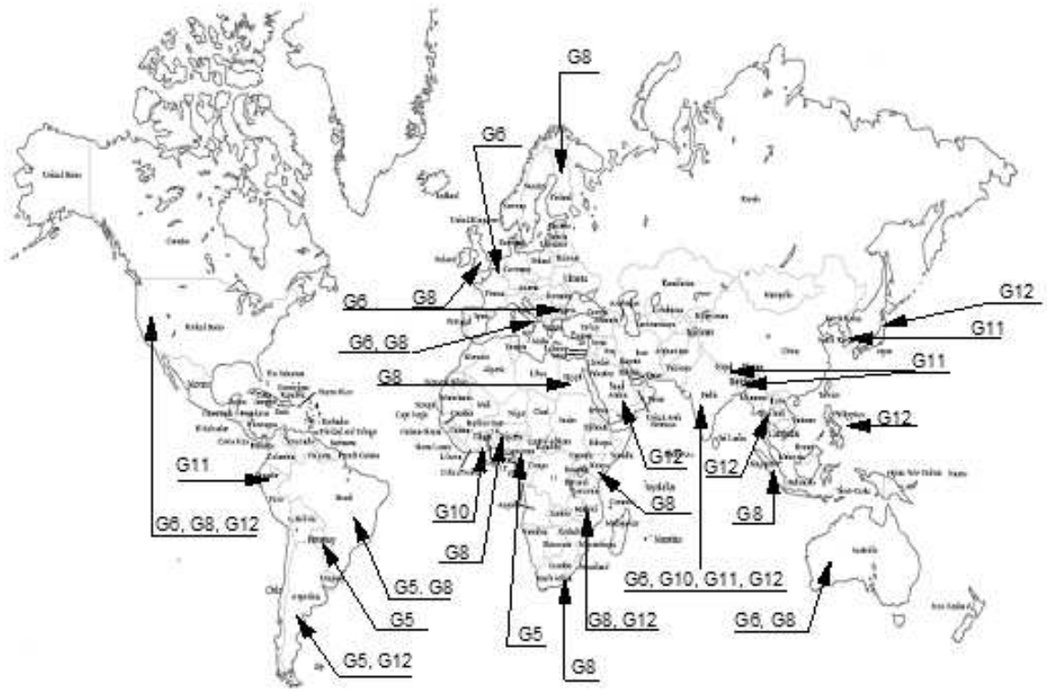
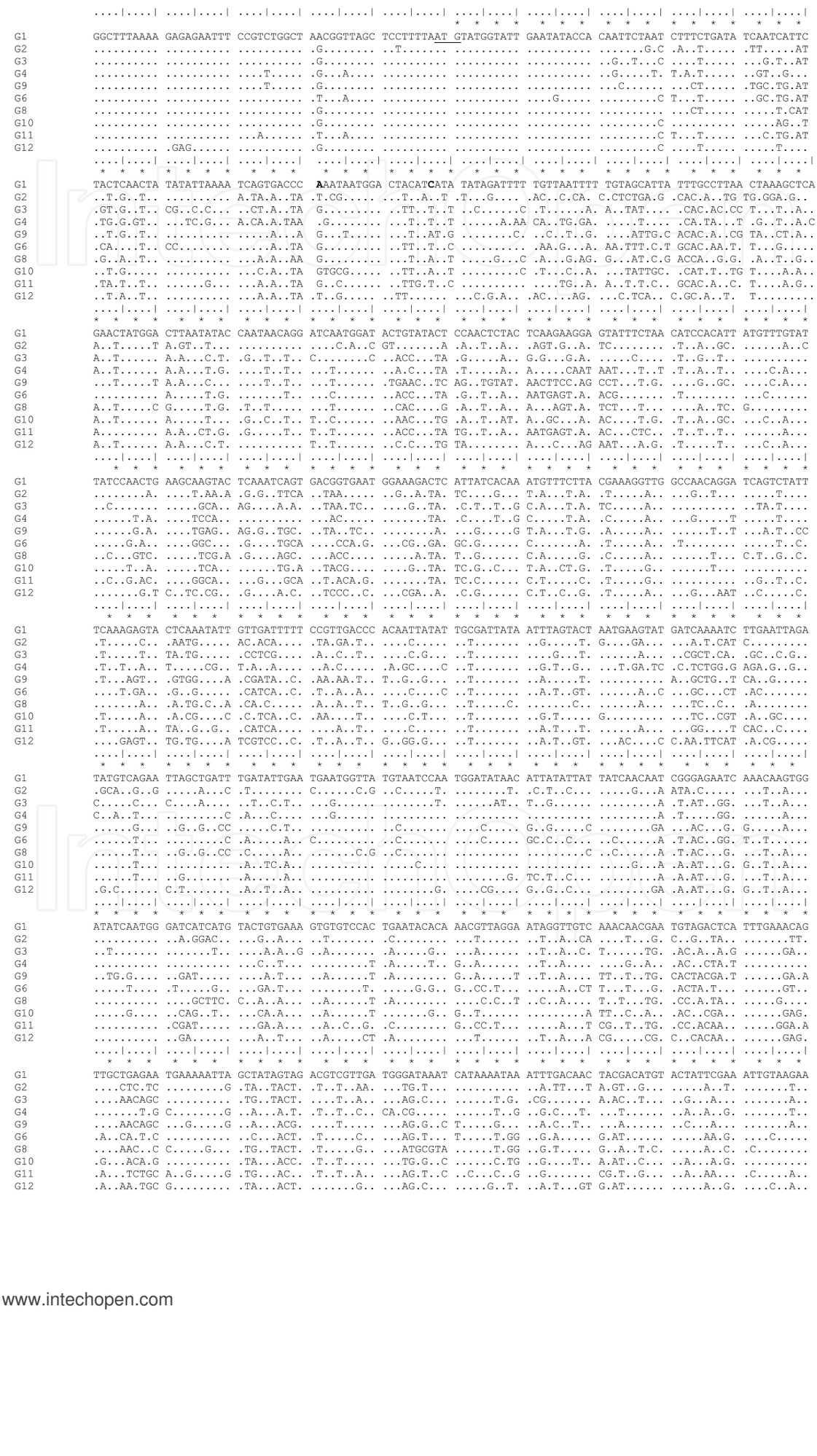


Fig. 1. Distribution of uncommon rotavirus genotypes in the world. The countries where the uncommon rotavirus genotypes were detected are shown by arrows.



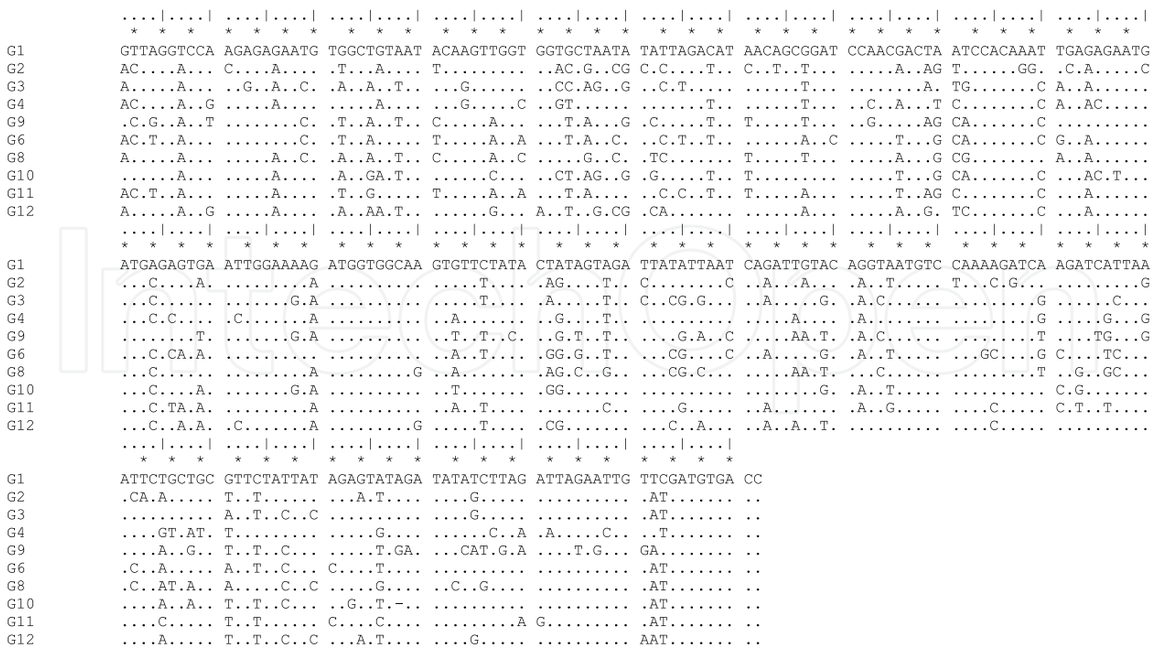


Fig. 2. Nucleotide analysis of rotavirus genotypes. Points indicated no nucleotide change. Nucleotide difference was shown by alphabetical marks. The start codon was underlined at position 49.

In addition, there is evidence of intragenic recombination in rotavirus VP7 genes. The existence of intragenic recombinations between interlineage and intersublineage in G1 rotaviruses was demonstrated (Tung et al., 2007). This variability has led to nucleotide mismatches between the actual VP7 gene and primers and consequently a failure on the detection of the G1 strains (Parra & Espinola, 2006). Other studies have reported the detection of possible new distinct sublineages for G2 genotype (Mascarenhas et al., 2010). Recombination between human rotaviruses and animal rotaviruses were well recognized in G3 rotaviruses (Nishikawa et al., 1989) and the genetic variation in their VP7 gene was reported in China and Japan accompanied with change also on the amino acid level (Wen et al., 1997). In addition, it has been postulated that amino acid substitutions at positions 96 and 213 might be involved in the emergence of G3 rotavirus strains in Japan, China, and Russia from 2001 to 2004 (Trinh et al., 2007). The diversification of rotavirus strains in phylogenetic lineages in G9 strains was reported previously (Santos et al., 2002; Hoshino et al., 2004; Cao et al., 2008; Pattara et al., 2009). Although one amino acid change at position 208 in an antigenic region C of G9 rotaviruses VP7 gene was reported, it is not clear whether this change affected the nature of the viruses in terms of antigenicity, infectivity or pathogenicity.

5. Conclusion

As a conclusion, the use of vaccines with broad and consistent serotype coverage would be important to help decrease the burden of rotavirus in countries with new emergent genotypes. The emergence of new rotavirus strains stresses the importance of a better knowledge of their genotypes and their mechanisms of transmission. Hence the importance

of setting up a vaccine design strategy as well as surveillance programs that should detect the diversity of rotavirus strains before, during and after the introduction of a rotavirus vaccine (Palombo, 1999) in order to detect the possible appearance of a mutants escape strains that probably reflect a selective pressure induced by the vaccine and eventually may pose a challenge to vaccine strategies. Further studies need to be carried out in order to elucidate the mechanism(s) behind the genetic recombination of rotaviruses and the emergence of new genotypes.

6. Reference list

Adah, M.I., Rohwedder, A., Olaleyle, O.D., & Werchau, H. (1997). Nigerian rotavirus serotype G8 could not be typed by PCR due to nucleotide mutation at the 3' end of the primer binding site. *Archives of Virology*, Vol.142, No.9, (September 1997), pp. 1881-1887, ISSN 1432-8798

Adah, M.I., Wade, A., & Taniguchi K. (2001). Molecular epidemiology of rotavirus in Nigeria: detection of unusual strains with G2P[6] and G8P[1] specificities. *Journal of Clinical Microbiology*, Vol.39, No.11, (November 2001), pp. 3969-3975, ISSN 0095-1137

Adah, M.I., Nagashima, S., Wakuda, M., & Taniguchi, K. (2003). Close Relationship between G8-Serotype Bovine and Human Rotaviruses Isolated in Nigeria. *Journal of Clinical Microbioloy*, Vol.41, No.8, (August 2003), pp. 3945-3950, ISSN 0095-1137

Alouf, J.E. (1987). From 'diphtheritic' poison to molecular toxicology. *American Society of Microbiology News*, Vol.53, No.10, (n.d.), pp. 547-550, ISSN 0044-7897

Andrej, S., Mateja P-P., Darja B-M., & Jozica M. (2008). Human, porcine and bovine rotaviruses in Slovenia: evidence of interspecies transmission and genome reassortment. *Journal of General Virology*, Vol.89, No.7, (July 2008), pp. 1690-1698, ISSN 1465-2099

Armah, G.E., Hoshino, Y., Santos, N., Binka, F., Damanka, S., Adjei, R., Honma, S., Tatsumi, M., Manful, T., & Anto, F. (2010). The Global Spread of Rotavirus G10 strains: Detection in Ghanaian Children hospitalized with Diarrhoea. *Journal of Infectious Diseases*, Vol.202, Suppl 1, (September 2010), pp. S231–S238, ISSN 0022-1899

Bachmayer, H., Liehl, E., & Schmidt, G. (1976). Preparation and properties of a novel influenza subunit vaccine. *Postgraduate Medical Journal*, Vol.52, No.608, (June 1976), pp. 360-367, ISSN 0032-5473

Banerjee, I., Iturriza-Gómara, M., Rajendran, P., Primrose, B., Ramani, S., Gray, J.J., Brown, D.W., & Kang, G. (2007). Molecular characterization of G11P[25] and G3P[3] human rotavirus strains associated with asymptomatic infection in South India. *Journal of Medical Virology*, Vol.79, No.11, (November 2007), pp. 1768-1774, ISSN 1096-9071

Banyai, K., Gentsch, J.R., Glass, R.I., & Szucs, G. (2003). Detection of human rotavirus serotype G6 in Hungary. *Epidemiology and Infection*, Vol.130, No.1, (February 2003), pp. 107-112, ISSN 1096-9071

Banyai, K., Gentsch, J.R., Glass, R.I., Uj, M., Mihaly, I., & Szucs, G. (2004). Eight-year survey of human rotavirus strains demonstrates circulation of unusual G and P types in Hungary. *Journal of Clinical Microbiology*, Vol.42, No.1, (January 2004), pp. 393-397, ISSN 0095-1137

Banyai, K., Esona, M.D., Kerin, T.K., Hull, J.J., Mijatovic, S., Vasconez, N., et al. (2009). Molecular characterization of a rare, human-porcine reassortant rotavirus strain,

G11P[6], from Ecuador. *Archives of Virology*, Vol.154, No.11, (September 2009), pp. 1823-1829, ISSN 1432-8798

Barnes, G.L., Lund, J.S., Adams, L., Mora, A., Mitchell, S.V., Caples, A., & Bishop R.F. (1997). Phase 1 trial of a candidate rotavirus vaccine (RV3) derived from a human neonate. *Journal of Paediatrics and Child Health*, Vol.33, No.4, (August 1997), pp. 300-304, ISSN 1440-1754

Bittle, J.L., Houghten, R.A., Alexander, H., Shinnick, T.M., Sutcliffe, J.G., Lerner, R.A., Rowlands, D.J., & Brown, F. (1982). Protection against foot-and-mouth disease by immunization with a chemically synthesized peptide predicted from the viral nucleotide sequence. *Nature*, Vol.298, No.5869, (Jul 1982), pp. 30-33, ISSN 0028-0836

Bellamy, A.R. & Both, G.W. (1990). Molecular biology of rotaviruses. *Advances in Virus Research*, Vol.38, (n.d.), pp. 1-43, ISSN 0065-3527

Bernstein, D.I., Sack, D.A., Reisinger, K., Rothstein, E., & Ward, R.L. (2002). Second-year follow-up evaluation of live, attenuated human rotavirus vaccine 89-12 in healthy infants. *Journal of Infectious Diseases*, Vol.186, No.10, (November 2002), pp. 1487–1489, ISSN 0022-1899

Bizzini, B., Blass, J., Turpin, A., & Raynaud, M. (1970). Chemical characterization of tetanus toxin and toxoid. *European Journal of Biochemistry*, Vol.17, (n.d.), pp. 100-105, ISSN 1432-1033

Bok, K., Palacios, G., Sijvarger, K., Matson, D.O., & Gomez, J.A. (2001). Emergence of G9P[6] human rotavirus in Argentina: phylogenetic relationships among G9 strains. *Journal of Clinical Microbiology*, Vol.39, No.11, (November 2001), pp. 4020-4025, ISSN 0095-1137

Brown, F. (1990). Synthetic peptides as potential vaccines against foot-and-mouth disease. *Endeavour*, Vol.14, No.2, (n.d.), pp. 87-94, ISSN 0160-9327

Browning, G.F., Snodgrass, D.R., Nakagomi, O., Kaga, E., Sarasini, A., & Gerna, G. (1992). Human and bovine serotype G8 strains may be derived by reassortment. *Archives of Virology*, Vol.125, No.1-4 , (n.d.), pp. 121-128, ISSN 1432-8798

Cao, D., Santos, N., Jones, R.W., Tatsumi,M., Gentsch, J.R., & Hoshino, Y. (2008). The VP7 genes of two G9 rotaviruses isolated in 1980 from diarrheal stool samples collected in Washington, DC, are unique molecularly and serotypically. *Journal of Virology*, Vol.82, No.8, (April 2008), pp. 4175–4179, ISSN 0022-538X

Castello, A.A., Jiang, B., Glass, R.I., Glikmann, G., & Genstch, J.R. (2004). Rotavirus G and P genotpe prevalence in Argentina 1999–2003: detection of P[9]G12 strains [abstract P37-4]. *Abstract of 23rd Annual Meeting of the American Society for Virology*, Montreal, Canada, (July 2004)

Centers for Disease Control and Prevention. (1999a). In: *MMWR Morbidity and Mortality Weekly Report*, 1999, Available from: http://www.cdc.gov/mmwr/preview/mmwrhtml/mm4827a1.htm, Intussusception among recipients of rotavirus vaccine—United States, 1998–1999

Centers for Disease Control and Prevention. (1999b). In: *MMWR Morbidity and Mortality Weekly Report*, 1999, Available from:Rotavirus vaccine for the prevention of rotavirus gastroenteritis among children. Recommendations of the Advisory Committee on Immunization Practices (ACIP).MMWRMorb. Mortal. Wkly. Rep. 48:1–20.

Centers of disease Control and Prevention. Prevention of Rotavirus Gastroenteritis Among Infants and Children: Recommendations of the Advisory Committee on Immunization Practices (ACIP). MMWR 2006, 55(RR12); 1-13.

Cheon, D.-S., Lee, K., Kim, W., Lee, S., Choi, W., Ahn, J., Jee, Y., Cho, H. Genetic analysis of the VP7 gene of unusual genotypes of human group A rotavirus strains circulating in Korea [abstract P37-3]. *Abstract of 23rd Annual Meeting of the American Society for Virology*, Montreal, Canada, (July 2004).

Chezzi, C., Dornrnann, C.J., Blackburn, N.K., Maselesele, E., McAnerney, J., & Schoub, B.D. (1998). Genetic stability of oral polio vaccine prepared on primary monkey kidney cells or Vero cells-effects of passage in cell culture and the human gastrointestinal tract. *Vaccine*, Vol.16, No.20, (December 1998), pp. 2031-2038, ISSN 0264-410X

Christy, C., Madore, H.P., Pichichero, M.E., Gala, C., Pincus, P., Vosefski, D., Hoshino, Y., Kapikian, A., & Dolin, R. (1988). Field trial of rhesus rotavirus vaccine in infants. *Pediatric Infectious Disease Journal*, Vol.7, No.9, (September 1988), pp. 645-650, ISSN 1532-0987

Ciarlet, M., Hidalgo, M., Gorziglia, M., & Liprandi, F. (1994). Characterization of neutralization epitopes on the VP7 surface protein of serotype G11 porcine rotaviruses. *Journal of General Virology*, Vol.75, No.8, (August 1994), pp.1867-1873, ISSN 1465-2099

Ciarlet, M.L., Hoshino, Y., & Liprandi, F. (1997). Single point mutation may affect the serotype reactivity of G11 porcine rotavirus strains: a widening spectrum? *Journal of Virology*, Vol.71, No.11, (November 1997), pp. 8213-8220, ISSN 0022-538X

Clark, H.F., Hoshino, Y., Bell, L.M., Groff, J., Hess, G., Bachman, P., & Offit, P.A. (1987). Rotavirus isolate WI61 representing a presumptive new human serotype. *Journal of Clinical Microbiology*, Vol.25, No.9, (September 1987), pp. 1757-1762, ISSN 0095-1137

Clark, H.F., Borian F.E., Bell L.M., Modesto K., Gouvea V., & Plotkin S.A. (1988). Protective effect of WC3 vaccine against rotavirus diarrhea in infants during a predominantly serotype 1 rotavirus season. *Journal of Infectious Diseases*, Vol.158, No.3, (September 1988), pp. 570-587, ISSN 0022-1899

Clark, H.F., Bernstein, D.I., Dennehy, P.H., Offit, P., Pichichero, M., Treanor, J., Ward, R.L., Krah, D.L., Shaw, A., Dallas, M.J., Laura, D., Eiden, J.J., Ivanoff, N., Kaplan, K.M., & Heaton, P. (2004). Safety, efficacy and immunogenicity of a live, quadrivalent human-bovine reassortant rotavirus vaccine in healthy infants. *Journal of Pediatrics*, Vol.144, No.2, (February 2004), pp. 184-190, ISSN 0022-3476

Coluchi, N., Munford, V., Manzur, J., Vazquez, C., Escobar, M., Weber, E., Mármol, P., & Rácz, M.L. (2002). Detection, subgroup specificity, and genotype diversity of rotavirus strains in children with acute diarrhea in Paraguay. *Journal of Clinical Microbiology*, Vol.40, No.5, (May 2002), pp. 1709-1714, ISSN 0095-1137

Cooney, M.A., Gorrell, R.J., & Palombo, E.A. (2001). Characterization and phylogenetic analysis of the VP7 proteins of serotype G6 and G8 human rotaviruses. *Journal of Medical Microbiology*, Vol.50, No.5, (May 2001), pp. 462-467, ISSN 0022-2615

Coulson, B., & Kirkwood, C. (1991). Relation of VP7 amino acid sequence to monoclonal antibody neutralization of rotavirus and rotavirus monotype. *Journal of Virology*, Vol.65, No.11, (November 1991), pp. 5968-5974, ISSN 0022-538X

Cubitt, W.D., Steele, A.D., & Iturriza-Gómara, M. (2000). Characterisation of rotaviruses from children treated at a London hospital during 1996: emergence of strains G9P2A[6] and G3P2A[6]. *Journal of Medical Virology*, Vol.61, No.1, (May 2000), pp. 150-154, ISSN 1096-9071

Cunliffe, N.A., Gondwe, J.S., Broadhead, R.L., Molyneux, M.E., Woods, P.A., Bresee, J.S., Glass, R.I., Gentsch, J.R., & Hart, C.A. (1999). Rotavirus G and P types in children with acute diarrhea in Blantyre, Malawi, from 1997 to 1998: predominance of novel P[6]G8 strains. *Journal of Medical Virology*, Vol.57, No.3, (March 1999), pp. 308-312, ISSN 1096-9071

Cunliffe, N.A., Gentsch J.R., Kirkwood C.D., Gondwe J.S., Dove W., Nakagomi O., Nakagomi T., Hoshino Y., Bresee J.S., Glass R.I., Molyneux M.E., & Hart C.A. (2000). Molecular and serologic characterization of novel serotype G8 human rotavirus strains detected in Blantyre, Malawi. *Virology*, Vol.274, No.2, (September 2000), pp. 309-320, ISSN 0042-6822

Cunliffe, N.A., Ngwira, B.M., Dove, W., Nakagomi, O., Nakagomi, T., Perez, A., Hart, C.A., Kazembe, P.N., & Mwansambo, C.C.V. (2009). Serotype G12 Rotaviruses, Lilongwe, Malawi. *Emerging Infectious Diseases*, Vol.15, No.1, (January 2009), pp. 87-90, ISSN 1080-6059

Das, B.K., Gentsch, J.R., Hoshino, Y., Ishida, S., Nakagomi, O., Bhan, M.K., Kumar, R., & Glass, R.I. (1993a). Characterization of the G serotype and genogroup of New Delhi newborn rotavirus strain 116E. *Virology*, Vol.197, No.1, (November 1993), pp. 99-107, ISSN 0042-6822

Das, M., Dunn, S.J., Woode, G.N., Greenberg, H.B., & Rao, C.D. (1993b). Both surface proteins (VP4 and VP7) of an asymptomatic neonatal rotavirus strain (I321) have high levels of sequence identity with the homologous proteins of a serotype 10 bovine rotavirus. *Virology*, Vol.194, No.1, (May 1993), pp. 374-379, ISSN 0042-6822

Das, S., Varghese, V., Chaudhury, S., Barman, P., Mahapatra, S., Kojima, K., Bhattacharya, S.K., Krishnan, T., Ratho, R.K., Chhotray, G.P., Phukan, A.C., Kobayashi, N., & Naik, N.T. (2003). Emergence of novel human group A rotavirus G12 strain in India. *Journal of Clinical Microbiology*, Vol.41, No.6, (June 2006), pp. 2760-2762, ISSN 0095-1137

De Vos, B., Vesikari, T., Linhares, A.C., Salinas, B., Perez-Schael, I., Ruiz-Palacios, G.M., Guerrero, M.L., Phua, K.B., Delem, A., Hardt, K. (2004). A rotavirus vaccine for prophylaxis of infants against rotavirus gastroenteritis. *Pediatric Infectious Disease Journal*, Vol.23, Suppl 10, (October 2004), pp. S179-S182, ISSN 1532-0987

Dhama, K., Chauhan, R.S., Mahendran, M., & Malik, S.V.S. (2009). Rotavirus diarrhea in bovines and other domestic animals. *Veterinary Research Communications*, Vol.33, No.1, (January 2009), pp. 1-23, ISSN 0165-7380

Donnelly, J.J., Ulmer, J.B., Liu, M.A. (1994). Immunization with DNA. *Journal Immunological Methods*, Vol.176, No.2, (December 1994), pp. 145-152, ISSN 0022-1759

Dorothea, S. (1993). Synthetic peptide vaccines. Journal of Medical Microbiology, Vol.39, No.4, (October 1993), pp. 241-242, ISSN 0022-2615

Drozdov, S.G. & Shirman, G.A. (1961). Interaction of viruses in the intestinal tract of man. I. Interference between wild and vaccine poliovirus strains. *Acta virologica*, Vol.5, No., (Jully 1961), pp. 210-219, ISSN 1336-2305

Dunn, S.J., Greenberg, H.B., Ward, R.L., Nakagomi, O., Burns, J.W., Vo, P.T., Pax, K.A., Das, M., Gowda, K., & Rao, C.D. (1993). Serotypic and genotypic characterization of human serotype 10 rotaviruses from asymptomatic neonates. *Journal of Clinical Microbiology*, Vol.31, No.1, (January 1993), pp. 165-169, ISSN 0095-1137

Dunn, P.M. (1996). "Dr Edward Jenner (1749-1823) of Berkeley, and vaccination against smallpox". Archives of disease in childhood-Fetal and neonatal edition, Vol.74, No.1, (January 1996), pp. F77-F78, ISSN 1468-2044

Dyall-Smith, M.L., Lazdins, I., Tregar, G.W., & Holmes, I.H. (1986). Location of major antigenic sites involved in rotavirus serotype-specific neutralization. *Proceedings of the National Academy of Sciences USA*, Vol.83, No.10, (May 1986), pp. 3465-3468, ISSN 0027-8424

Emini, E.A., Jameson, B.A. & Wimmer, E. (1983). Priming for and induction of anti-poliovirus neutralizing antibodies by synthetic peptides. Nature, Vol.304, (August 1983), pp. 699-703, ISSN 0028-0836

Esona, M., Armah, G.E., Geyer, A., & Steele, A.D. (2004). Detection of an unusual human rotavirus strain with G5P[8] specificity in a Cameroonian child with diarrhea. *Journal of Clinical Microbiology*, Vol.42, No.1, (January 2004), pp.441-444, ISSN 0095-1137

Estes, M.K. & Cohen, J. (1989). Rotavirus gene structure and function. *Microbiological Reviews*, Vol.53, No.4, (December 1989), pp.410-449, ISSN 0146-0749

Estes, M.K. (2001). Rotaviruses and their replication, In: *Fields virology*, Howley P.M., pp. 1747-1786, Lippincott Williams & Wilkins, ISBN 0781718325, Philadelphia

Estes, M.K. & Kapikian, A.Z. (2007). Rotaviruses, In: *Fields virology*, Knipe D.M., Howley P.M., Griffin D.E., Lamb R.A., Martin M.A., Roizman B., & Straus S.E., pp. 1917-1974, Lippincott Williams & Wilkins, ISBN 0781718325, Philadelphia

Gard, S. (1960). Theoretical considerations in the inactivation of viruses by chemical means. *Annals of the New York Academy of Sciences*, Vol.83, (January 1960), pp. 638, ISSN 0077-8923

Giammanco, G., De Grandi, V., Lupo, L., Mistretta, A., Pignato, S., Teuween, D., Bogaerts, H., & Andrè F.E. (1988). Interference of Oral Poliovirus Vaccine on RIT 4237 Oral Rotavirus Vaccine. *European Journal of Epidemiology, Vol.*4, No.1, (March 1988), pp. 121-123, ISSN 0392-2990

Gentsch, J.R., Woods, P.A., Ramachandran, M., Das, B.K., Leite, J.P., Alfieri, A., Kumar, R., Bhan, M.K., & Glass, R.I. (1996). Review of G and P typing results from a global collection of rotavirus strains: implications for vaccine development. *Journal of Infectious Diseases*, Vol.174, Supl. 1, (September 1996), pp. S30-S36, ISSN 0022-1899

Gentsch, J.R., Laird, A.R., Bielfelt, B., Griffin, D.D., Banyai, K., Ramachandran, M., Jain, V., Cunliffe, N.A., Nakagomi, O., Kirkwood, C.D., Fischer, T.K., Parashar, U.D., Bresee, J.S., Jiang, B., & Glass, R.I. (2005). Serotype diversity and reassortment between human and animal rotavirus strains: implications for rotavirus vaccine programs. *Journal of Infectious Diseases*, Vol.192, Supl. 1, (September 2005), pp. 146-159, ISSN 0022-1899

Gerna, G., Sarasini, A., Zentilin, L., Di Matteo, A., Miranda, P., Parea, M., Battaglia, M., & Milanesi, G. (1990). Isolation in Europe of 69M-like (serotype 8) human rotavirus strains with either subgroup I or II specificity and a long RNA electropherotype. *Archives of Virolology*, Vol.112, No.1-2, (n.d.), pp. 27-40, ISSN 1432-8798

Gerna, G., Sarasini, A., Parea, M., Arista, S., Miranda, P., Brüssow, H., Hoshino, Y., & Flores, J. (1992). Isolation and characterization of two distinct human rotavirus strains with G6 specificity. *Journal of Clinical Microbiology*, Vol.30, No.1, (January 1992), pp. 9-16, ISSN 0095-1137

Gerna, G., Steele, A.D., Hoshino, Y., Sereno, M., Garcia, D., Sarasini, A., & Flores, J. (1994). A comparison of the VP7 gene sequences of human and bovine rotaviruses. *Journal of General Virology*, Vol.75, No.7, (July 1994), pp. 1781-1784, ISSN 1465-2099

Gombold, J.L. & Ramig, R.F. (1986). Analysis of reassortment of genome segments in mice mixedly infected with rotaviruses SA 11 and RRV. *Journal of Virology*, Vol.57, No.1, (January 1986), pp. 110-116, ISSN 0022-538X

Gouvea, V., Glass, R.I., Woods, P., Taniguchi, K., Clark, H.F., Forrester, B., & Fang, Z.Y. (1990). Polymerase Chain Reaction Amplification and Typing of Rotavirus Nucleic Acid from Stool Specimens. Journal of Clinical Microbiology, Vol.28, No.2, (February 1990), pp. 276-282, ISSN 0095-1137

Gouvea, V., de Castro, L., Timenetsky, M.C., Greenberg, H., & Santos, N. (1994). Rotavirus serotype G5 associated with diarrhea in Brazilian children. *Journal of Clinical Microbiology*, Vol.32, No.5, (May 1994), pp. 1408-1409, ISSN 0095-1137

Green, K.Y., Midthun, K., Gorziglia, M., Hoshino, Y., Kapikian, A.Z., Chanock, R.M., & Flores, J. (1987). Comparison of the amino acid sequences of the major neutralization protein of four human rotavirus serotypes. *Virology*, Vol.161, No.1, (November 1987), pp.153-159, ISSN 0042-6822

Green, K.Y., Sears, J.F., Taniguchi, K., Midthun, K., Hoshino, Y., Gorziglia, M., Nishikawa, K., Urasawa, S., Kapikian, A.Z., & Chanock, R.M. (1988). Prediction of human rotavirus serotype by nucleotide sequence analysis of the VP7 protein gene. *Journal of Virology*, Vol.62, No.5, (May 1988), pp. 1819-1823, ISSN 0022-538X

Greenberg, H.B., Kalica, A.R., Wyatt, R.G., Jones, R.W., Kapikian, A.Z., & Chanock, R.M. (1981). Rescue of noncultivatable human rotavirus by gene reassortment during mixed infection with ts mutants of a cultivatable bovine rotavirus. *Proceedings of the National Academy of Sciences USA*, Vol.78, No.1, (January 1981), pp. 420-424, ISSN 0027-8424

Griffin, D.D., Nakagomi, T., Hoshino, Y., Nakagomi, O., Kirkwood, C.D., Parashar, U.D., Glass, R. I., & Gentsch, J.R. (2002). Characterization of nontypeable rotavirus strains from the United States: identification of a new rotavirus reassortant (P2A[6],G12) and rare P3[9] strains related to bovine rotaviruses. *Virology*, Vol.294, No.2, (March 2002), pp. 256-269, ISSN 0042-6822

Gurgel, R.Q., Cuevas, L.E., Vieira, S.C., Barros, V.C., Fontes, P.B., Salustino, E.F., Nakagomi, O., Nakagomi, T., Dove, W., Cunliffe, N., & Hart, CA. (2007). Predominance of rotavirus P[4]G2 in a vaccinated population, Brazil. *Emerging Infectious Diseases*, Vol.13, No.10, (October 2007), pp. 1571-1573, ISSN 1080-6059

Hanlon, P., Marsh, V., Shenton, F., Jobe, O., Hayes, R., Whittle, H.C., Hanlon, L., Byass, P., Hassan-King, M., Sillah, H., M'Boge, B.H., & Greenwood, B.M. (1987). Trial of an attenuated bovine rotavirus vaccine (RIT 4237) in Gambian infants. *Lancet*, Vol.329, No.8546, (June 1987), pp. 1342-1345, ISSN 0140-6736

Hans, D., Young, P.R., & Fairlie, D.P. (2006). Current status of short synthetic peptides as vaccines. *Journal of Medical Chemistry*, Vol.2, No.6, (November 2006), pp. 627-646, ISSN 0022-2623

Hasegawa, A., Inouye, S., Matsuno, S., Yamaoka, K., Eko, R., & Suharyono, W. (1984). Isolation of human rotavirus with a distinct RNA electrophoretic pattern from Indonesia. *Microbiology and Immunology*, Vol.28, No.6, (n.d.), pp. 719-722, ISSN 1348-0421

Hauser, P., Thomas, H.C., Waters, J., Simoen, E., Voet, P., De Wilde, M., Stephenne, J., & Petre, J. (1988). Induction of neutralising antibodies in chimpanzees and humans by recombinant yeast-derived hepatitis B surface antigen particle, In: *Viral hepatitis and liver disease*, Zuckerman A.J., pp. 1031-1037, Alan R. Liss., ISBN 978-0471612704, New York

Hilleman, M.R. (1987). Yeast recombinant hepatitis B vaccine. *Infection*, Vol.15, No.1, (January 1987), pp. 3-7, ISSN 0300-8126

Holland, J.J., De La Torre, J.C., & Steinhauer, D.A. (1992). RNA virus populations as quasispecies. *Current Topics in Microbiology and Immunology*, Vol.176, (n.d.), pp. 1-20, ISSN 0070-217X

Holmes, J.L., Kirkwood, C.D., Gerna, G., Clemens, J.D., Rao, M.R., Naficy, A.B., Abu-Elyazeed, R., Savarino, S.J., Glass, R.I., & Gentsch, J.R. (1999). Characterization of unusual G8 rotavirus strains isolated from Egyptian children. *Archives of Virology*, Vol.144, No.7, (n.d.), pp. 1381-1396, ISSN 1432-8798

Hong, S.-K., Lee, S.-G., Lee, S.-A., Kang, J.-H., Lee, J.-H., Kim, J.-H., Kim, D.-S., Kim, H.-M., Jang, Y.-T., Ma, S.-H., Kim, S.-Y., & Paik, S.-Y. (2007). Characterization of a G11, P[4] strain of human rotavirus isolated in South Korea. *Journal of Clinical Microbiology*, Vol.45, No.11, (November 2007), pp. 3759-3761, ISSN 0095-1137

Hoshino, Y., Nishikawa, K., Benfield, D.A., & Gorziglia, M. (1994). Mapping of antigenic sites involved in serotype-cross-reactive neutralization on group A rotavirus outercapsid glycoprotein VP7. *Virology*, Vol.199, No.1, (February 1994), pp. 233-237, ISSN 0042-6822

Hoshino, Y., & Kapikian, A.Z. (2000). Rotavirus serotypes: Classification and importance in epidemiology, immunity, and vaccine development. *Journal of Health Population and Nutrition*, Vol.18, No.1, (June 2000), pp. 5-14, ISSN 1606-0997

Hoshino, Y., Jones, R.W., Ross, J., Honma, S., Santos, N., Gentsch, J.R., & Kapikian, A.Z. (2004). Rotavirus serotype G9 strains belonging to VP7 gene phylogenetic sequence lineage 1 may be more suitable for serotype G9 vaccine candidates than those belonging to lineage 2 or 3. *Journal of Virology*, Vol.78, No.14, (July 2004), pp. 7795-7802, ISSN 0022-538X

Hull, J.J., Teel, E.N., Kerin, T.K., Freeman, M.M., Esona, M.D., Gentsch, J.R., Cortese, M.M., Parashar, U.D., Glass, R.I., & Bowen, M.D. (2011). United States Rotavirus Strain Surveillance From 2005 to 2008: Genotype Prevalence Before and After Vaccine Introduction. *Journal of Pediatric Infectious Disease*, Vol.30, No.1, (January 2011), pp. S42-S47, ISSN 1305-7707

Iturriza-Gómara, M., Green, J., Brown, D.W.G, Desselberger, U., & Gray, J. (2000). Diversity within the VP4 gene of rotavirus P[8] strains: implications for reverse transcription-PCR genotyping. *Journal of Clinical Microbiology*, Vol.38, No.2, (February 2000), pp. 898-901, ISSN 0095-1137

Iturriza-Gómara, M., Kang, G., Mammen, A., Jana, A.K., Abraham, M., Desselberger, U., Brown, D., & Gray, J. (2004a). Characterization of G10P[11] rotaviruses causing

acute gastroenteritis in neonates and infants in Vellore, India. *Journal of Clinical Microbiology*, Vol.42, No.6, (June 2004), pp. 2541-2547, ISSN 0095-1137

Iturriza-Gómara, M., Kang, G., & Gray, J. (2004b). Rotavirus genotyping: keeping up with an evolving population of human rotaviruses. Journal of Clinical Virology, Vol.31, No.4, (December 2004), pp. 259-265, ISSN

Joensuu, J., Koskenniemi, E., Pang, X.-L., & Vesikari, T. (1997). Randomised placebo-controlled trial of rhesus-human reassortant rotavirus vaccine for prevention of severe rotavirus gastroenteritis. *Lancet*, Vol.350, No.9086, (October 1997), pp. 1205-1209, ISSN 0140-6736

Jonathan, R. (1987). Synthetic peptides as vaccines. *Nature*, Vol.330, (November 1987), pp. 106-107, ISSN 0028-0836

Kalica, A.R., Greenberg, H.B., Wyatt, R.G., Flores, J., Sereno, M.M., Kapikian, A.Z. & Chanock, R.M. (1981). Genes of human (strain Wa) and bovine (strain UK) rotaviruses that code for neutralization subgroup antigens. *Virology*, Vol.112, No.2, (July 1981), pp. 385-390, ISSN 0042-6822

Kapikian, A.Z., Hoshimo, Y., Chanock, R.M., & Perez-Schael, I. (1996a). Efficacy of a quadrivalent rhesus rotavirus-based human rotavirus vaccine aimed at preventing severe rotavirus diarrhea in infants and young children. *Journal of Infectious Diseases*, Vol.174, Suppl. 1, (September 1996), pp. S65-S72, ISSN 0022-1899

Kapikian, A.Z., Hoshino, Y., Chanock, R.M., & Perez-Schael, I. (1996b). Jennerian and modified Jennerian approach to vaccination against rotavirus diarrhea using a quadrivalent rhesus rotavirus (RRV) and human-RRV reassortant vaccine. *Archives of Virology. Supplementum*, Vol.12, (n.d.), pp. 163-175, ISSN 1432-8798

Kapikian, A.Z., Hoshino, Y., & Chanock, R.M. (2001). Rotaviruses, In: *Fields virology*, Howley P.M., pp. 1787-1833, Lippincott Williams & Wilkins, ISBN 0781718325, Philadelphia

Katz, S.L., Milovanovic M.V., & Enders J.F. (1958). Propagation of measles virus in cultures of chick embryo cells. *Proceedings of the Society for Experimental Biology and Medicine*, Vol.97, No.1, (n.d.), pp. 23-29, ISSN 1525-1373

Katz, M. & Plotkin, S.A. (1968). Oral polio immunization of the newborn infant; a possible method for overcoming interference by ingested antibodies. *Journal of Pediatrics*, Vol.73, No.2, (August 1968), pp. 267-270, ISSN 0022-3476

Kim, K.S. & Sharp, D.G. (1967). Influence of the physical state of formalinized vaccinia virus particles on surviving plaque titre: Evidence for multiplicity reactivation. *Journal of Immunology*, Vol.99, No.6, (December 1967), pp. 1221, ISSN 0022-1767

King, C.A., Spellerberg, M.B., Zhu, D.L., Rice, J., Sahota, S.S., Thompsett, A.R., Hamblin, T.J., Radl, J., & Stevenson, F.K. (1998). DNA vaccines with single-chain Fv fused to fragment C of tetanus toxin induce protective immunity against lymphoma and myeloma. *Nature Medicine*, Vol.4, (n.d.), pp. 1281-1286, ISSN 1078-8956

Kelkar, S.D. & Ayachit, V.L. (2000). Circulation of group A rotavirus subgroups and serotypes in Pune, India, 1990–1997. *Journal of Health Population and Nutrition*, Vol.18, No.3, (December 2000), pp. 163-170, ISSN 1606-0997

Khamrin, P., Peerakome, S., Wongsawasdi, L., Tonusin, S., Sornchai, P., Maneerat, V., Khamwan, C., Yagyu, F., Okitsu, S., Ushijima, H., & Maneekarn, N. (2006). Emergence of human G9 rotavirus with an exceptionally high frequency in children admitted to hospital with diarrhea in Chiang Mai, Thailand. *Journal of Medical Virology*, Vol.78, No.2, (February 2006), pp. 273-280, ISSN 1096-9071

Kheyami, A.M., Nakagomi, T., Nakagomi, O., Dove, W., Hart, C.A, & Cunliffe, N.A. (2008). Molecular Epidemiology of Rotavirus Diarrhea among Children in Saudi Arabia: First Detection of G9 and G12 Strains. *Journal of Clinical Microbiology*, Vol.46, No.4, (April 2008), pp. 1185-1191, ISSN 0095-1137

Kieny, M.P., Lathe, R., Drillien, R., Spehner, D., Skory, S., Schmitt, D., Wiktor, T., Koprowski, H., & Lecocq, J.P. (1984). Expression of rabies virus glycoprotein from recombinant vaccinia virus. *Nature*, Vol.312, (November 1984), pp. 163-166, ISSN 0028-0836

Kilbourne, E.D. (1991). New viruses and new disease: mutation, evolution and ecology. *Current Opinion in Immunology*, Vol.3, No.4, (August 1991), pp. 518-524, ISSN 0952-7915

Kirkwood, C., Masendycz, P.J., & Coulson, B.S. (1993). Characteristics and location of cross-reactive and serotype-specific neutralization sites on VP7 of human G type 9 rotaviruses. *Virology*, Vol.196, No.1, (September 1993), pp. 79-88, ISSN 0042-6822

Lanata, C.F., Black, R.E., del Aguila, R., Gil, A., Verastegui, H., Gerna, G., Flores, J., Kapikian, A.Z., & Andre, F.E. (1989). Protection of Peruvian children against rotavirus diarrhea of specific serotypes by one, two, or three doses of the RIT 4237 attenuated bovine rotavirus vaccine. *Journal of Infectious Diseases*, Vol.159, No.3, (March 1989), pp. 452-459, ISSN 0022-1899

Levine, M.M., Woodrow, G.C., Kaper, J.B., & Cobon, G.S. (Eds.). (1997). *New Generation Vaccines*, Marcel Dekker, Inc., ISBN 0-8247-0061-9, New York

Linhares, A.C. & Bresee, J.S. (2000). Rotavirus vaccine and vaccination in Latin America. *Revista Panamericana de Salud Pública*, Vol.8, No.5, (November 2000), pp. 305-331, ISSN 1020-4989

Linhares, A.C., Lanata, C.F., Hausdorff, W.P., Gabbay, W.P., & Black, R.E. (1999). Reappraisal of the Peruvian and Brazilian lower titer tetravalent rhesus-human reassortant rotavirus vaccine efficacy trials: analysis by severity of diarrhea. *Pediatric Infectious Disease Journal*, Vol.18, No.11, (November 1999), pp. 1001-1016, ISSN 1532-0987

Liprandi, F., Gerder, M., Bastidas, Z., Lopez, J.A., Pujol, F.H., Ludert, J.E., Joelsson, D.B., & Ciarlet, M. (2003). Novel Type of VP4 Carried by a Porcine Rotavirus Strain. *Virology*, Vol.315, No.2, (October 2003), pp. 373-380, ISSN 0042-6822

Lo Grippo, G.A. (1960). Investigations of the use of β-propiolactone in virus inactivation. *Annals of the New York Academy of Sciences*, Vol.83, (January 1960), pp. 578-594, ISSN 0077-8923

MacGregor, R.R., Boyer, J.D., Ugen, K.E., Lacy, K.E., Gluckman, S.J., Bagarazzi, M.L., Chattergoon, M.A., Baine, Y., Higgins, T.J., Ciccarelli, R.B., Coney, L.B., Ginsberg, R.S., & Weiner D.B. (1998) . First human trial of a DNA-based vaccine for treatment of human immunodeficiency virus type 1 infection: safety and host response. *Journal Infectious Diseases*, Vol.178, (July 1998), pp. 92-100, ISSN 0022-1899

Mackett, M., Yilma, T., Rose, J.K. & Moss, B. (1985). Vaccinia virus recombinants: expression of VSV genes and protective immunization of mice and cattle. Science, Vol.227, No.4685, (January 1985), pp. 433-435, ISSN 0036-8075

Martella V., Pratelli, A., Greco, G., Gentile, M., Fiorente, P., Tempesta, M., & Buonavoglia, C. (2001). Nucleotide sequence variation of the VP7 gene of two G3-type rotaviruses

isolated from dogs. Virus Research, Vol.74, No.1-2, (April 2001), pp. 17-25, ISSN 0168-1702

Martella, V., Ciarlet, M., Camarda, A., Pratelli, A., Tempesta, M., Greco, G., Cavalli, A., Elia, G., Decaro, N., Terio, V., Bozzo, G., Camero, M., & Buonavoglia, C. (2003). Molecular characterization of the VP4, VP6, VP7, and NSP4 genes of lapine rotaviruses identified in Italy: emergence of a novel VP4 genotype. *Virology*, Vol.314, No.1, (September 2003), pp. 358-370, ISSN 0042-6822

Martella, V., Terio, V., Arista, S., Elia, G., Corrente, M., Madio, A., Pratelli, A., Tempesta, M., Cirani, A., & Buonavoglia, C. (2004). Nucleotide variation in the VP7 gene affects PCR genotyping of G9 rotaviruses identified in Italy. Journal of Medical Virology, Vol.72, No.1, (January 2004), pp.143-148, ISSN 1096-9071

Mascarenhas, J.D., Lima, C.S., de Oliveira, D.S., Guerra Sde, F., Maestri, R.P., Gabbay, Y.B., de Lima, I.C., de Menezes, E.M., Linhares Ada, C., & Bensabath, G. (2010). Identification of Two Sublineages of Genotype G2 Rotavirus Among Diarrheic Children in Parauapebas, Southern Para State, Brazil. *Journal of Medical Virology*, Vol.82, No.4, (April 2010), pp. 712–719, ISSN 1096-9071

Matthijnssens, J., Ciarlet, M., Heiman, E., Arijs, I., Delbeke, T., McDonald, S.M., Palombo, A.E., Iturriza-Gómara, M., Maes, P., Patton, J.T., Rahman, M., & Van Ranst, M. (2008a). Full genome-based classification of rotaviruses reveals common origin between human Wa-like and porcine rotavirus strains and human DS-1-like and bovine rotavirus strains. *Journal of Virology, Vol*.82, No.7, (April 2008), pp. 3204-3219, ISSN 0022-538X

Matthijnssens, J., Ciarlet, M., Rahman, M., Attoui, H., Bányai, K., Estes, M.K., Gentsch, J.R., Iturriza-Gómara, M., Kirkwood, C.D., Martella, V., Mertens, P.P., Nakagomi, O., Patton, J.T., Ruggeri, F.M., Saif, L.J., Santos, N., Steyer, A., Taniguchi, K., Desselberger, U., & Van Ranst, M. (2008b). Recommendations for the classification of group A rotaviruses using all 11 genomic RNA segments. *Archives of Virology*, Vol.153, No.8, (July 2008), pp. 1621-1629, ISSN 1432-8798

Maunula, L. & von Bonsdorff, C.H. (1998). Short sequences define genetic lineages: phylogenetic analysis of group A rotaviruses based on partial sequences of genome segments 4 and 9. *Journal of General Virology*, Vol.79, No.2, (February 1998), pp. 321-332, ISSN 1465-2099

Midthun, K. & Kapikian A.Z. (1996). Rotavirus vaccines: an overview. *Clinical Microbiology Reviews*, Vol.9, No.3, (July 1996), pp. 423-434, ISSN 0893-8512

Milich, D.R. (1990). Synthetic peptides: prospects for vaccine development. Seminars in Immunology, Vol.2, No.5, (September 1990), pp. 307-315, ISSN 1044-5323

Müller, G.M., Shapira, M., & Arnon, R. (1982). Anti-influenza response achieved by immunization with a synthetic conjugate. *Proceedings of the National Academy of Sciences USA*, Vol.79, No.2, (January 1982), pp. 569-573, ISSN 0027-8424

Nakagomi, T. & Nakagomi, O. (2009). A critical review on a globally-licensed, live, orally-administrable, monovalent human rotavirus vaccine: Rotarix. *Expert Opinion on Biological Therapy*, Vol.9, No.8, (August 2009), pp. 1073-1086, ISSN 1471-2598

Nishikawa, K., Hoshino, Y., Taniguchi, K., Green, K.Y., Greenberg, H.B., Kapikian, A.Z., Chanock, R.M., & Gorziglia, M. (1989). Rotavirus VP7 neutralization epitopes of serotype 3 strains. *Virology*, Vol.171, No.2, (August 1989), pp. 503-515, ISSN 0042-6822

Nokes, D.J., Peenze, I., Netshifhefhe, L., Abwao, J., De Beer, M.C., Seheri, M., Williams, T.N., Page, N., & Steele, D. (2010). Rotavirus genetic diversity, disease association and temporal change in hospitalized rural Kenyan children. *Journal of Infectious Diseases*, Vol.202, Suppl., (September 2010), pp. S180-S186, ISSN 0022-1899

Norrby, E. (2007). Yellow fever and Max Theiler: the only Nobel Prize for a virus vaccine. *Journal of Experimental Medicine*, Vol.204, No.12, (November 2007), pp. 2779-2784, ISSN 0022-1007

Ohshima, A., Takagi T., Nakagomi T., Matsuno S., & Nakagomi O. (1990). Molecular characterization by RNA-RNA hybridization of a serotype 8 human rotavirus with a super-short RNA electropherotype. *Journal of Medical Virology*, Vol.30, No.2, (February 1990), pp. 107-112, ISSN 1096-9071

Okada, J., Urasawa, T., Kobayashi, N., Taniguchi, K., Hasegawa, A., Mise, K., & Urasawa, S. (2000). New P serotype of group A human rotavirus closely related to that of a porcine rotavirus. *Journal of Medical Virology*, Vol.60, No.1, (January 2000), pp. 63-69, ISSN 1096-9071

Palombo, E.A. & Bishop, R.F. (1995). Genetic and antigenic characterization of a serotype G6 human rotavirus isolated in Melbourne, Australia. *Journal of Medical Virology*, Vol.47, No.4, (December 1995), pp. 348-354, ISSN 1096-9071

Palombo, E.A. (1999). Genetic and antigenic diversity of human rotaviruses: potential impact on the success of candidate vaccines. *FEMS Microbiology Letters*, Vol.181, No.1, (December 1999), pp. 1-8, ISSN 0378-1097

Palombo, E.A., Clark, R., & Bishop, R.F. (2000). Characterization of a "European-like" serotype G8 human rotavirus isolated in Australia. *Journal of Medical Virology*, Vol.60, No.1, (January 2000), pp. 56-62, ISSN 1096-9071

Panicali, D., Davis, S.W., Weinberg, R.L., & Paoletti, E. (1983). Construction of live vaccines by using genetically engineered poxviruses: biological activity of recombinant vaccinia virus expressing influenza virus haemagglutinin. *Proceedings of the National Academy of Sciences USA*, Vol.80, No.17, (September 1983), pp. 5364-5368, ISSN 0027-8424

Paoletti, E., Lipinskas, B.R., Samsanoff, C., Mercer, S., & Panicali, D. (1984). Construction of live vaccines using genetically engineered poxviruses: biological activity of vaccinia virus recombinants expressing the hepatitis B virus surface antigen and herpes simplex virus glycoprotein D. *Proceedings of the National Academy of Sciences USA*, Vol.81, No.1, (January 1984), pp. 193-197, ISSN 0027-8424

Park, S.I., Matthijnssens, J., Saif, L.J., Kim, H.J., Park, J.G., Alfajaro, M.M., Kim, D.S., Son, K.Y., Yang, D.K., Hyun, B.H., Kang, M.I., & Cho, K.O. (2011). Reassortment among bovine, porcine and human rotavirus strains results in G8P[7] and G6P[7] strains isolated from cattle in South Korea. *Veterinary Microbiology*, Vol.152, No.1-2, (August 2011), pp. 55-66, ISSN 0378-1135

Parra, G.I. & Espinola, E.E. (2006). Nucleotide mismatches between the VP7 gene and the primer are associated with genotyping failure of a specific lineage from G1 rotavirus strains. *Virology Journal*, Vol.3, (May 2006), pp. 35, ISSN 1743-422X

Parwani, A.V., Hussein, H.A., Rosen, B.I., Lucchelli, A., Navarro, L., & Saif, L.J. (1993). Characterization of field strains of group A bovine rotaviruses by using polymerase chain reaction-generated G and P type-specific cDNA probes. *Journal of Clinical Microbiology*, Vol.31, No.8, (August 1993), pp. 2010-2015, ISSN 0095-1137

Pattara, K., Aksara, T., Natthawan, C., Pattranuch, C., Shoko, O., Hiroshi, U., & Niwat, M. (2009). Evolutionary consequences of G9 rotaviruses circulating in Thailand. Infection, Genetics and Evolution, Vol.9, No.2, (December 2009), pp. 1394-1399, ISSN 1567-1348

Payne, D.C., Edwards, K.M., Bowen, M.D., Keckley, E., Peters, J., Esona, M.D., Teel, E.N., Kent, D., Parashar, U.D., & Gentsch, J.R. (2010). Sibling Transmission of Vaccine-Derived Rotavirus (RotaTeq) Associated With Rotavirus Gastroenteritis. *Pediatrics*, Vol.125, No.2, (February 1), pp. e438-e441, ISSN 0031-4005

Penelope, H.D. (2008). Rotavirus Vaccines: an Overview. *Clinical Microbiology Reviews*, Vol.21, No.1, (January 2008), pp. 198-208, ISSN 0893-8512

Perez-Schael, I., Guntinas, M.J., Perez, M., Pagone, V., Rojas, A.M., Gonzalez, R., Cunto, W., Hoshino, Y., & Kapikian, A.Z. (1997). Efficacy of the rhesus rotavirus-based quadrivalent vaccine in infants and young children in Venezuela. *New England Journal of Medicine*, Vol.337, (October 1997), pp. 1181-1187, ISSN 1533-4406

Perkus, M.E., Piccini, A., Lipinskas, B.R., & Paoletti, E. (1985). Recombinant vaccinia virus: immunisation against multiple pathogens. *Science*, Vol.229, No.4717, (September 1985), pp. 981-984, ISSN 0036-8075

Pongsuwanna, Y., Guntapong, R., Chiwakul, M., Tacharoenmuang, R., Onvimala, N., Wakuda, M., Kobayashi, N., & Taniguchi, K. (2002). Detection of a human rotavirus with G12 and P[9] specificity in Thailand. *Journal of Clinical Microbiology*, Vol.40, No.4, (April 2002), pp. 1390-1394, ISSN 0095-1137

Prevots, D.R., Sutter, R.W., Quick, L., Izurieta, H., & Strebel, P.M. (1996). Vaccine-associated paralytic poliomyelitis in the United States, l980-1994: current risk and potential impact of a proposed sequential schedule of IPV followed by OPV [Abstract H90]. *Program and Abstracts of the 36th Interscience Conference on Antimicrobial Agents and Chemotherapy*, Washington, (September 1996)

Pringle, C.R. (1996). Temperature-sensitive mutant vaccines. *Methods in Molecular Medicine*, Vol.4, (n.d.), pp. 17-32, ISSN 1543-1894

Rahman, M., De Leener, K., Goegebuer, T., Wollants, E., Van der Donck, I., Van Hoovels, L., & Van Ranst, M. (2003). Genetic characterization of a novel, naturally occurring recombinant human G6P[6] rotavirus. *Journal of Clinical Microbiology*, Vol.41, No.5, (May 2003), pp. 2088-2095, ISSN 0095-1137

Rahman, M., Matthijnssens, J., Nahar, S., Podder, G., Sack, D.A., Azim, T., & Van Ranst, M. (2005a). Characterization of a novel P[25]G11 human group a rotavirus. *Journal of Clinical Microbiology*, Vol.43, No.7, (July 2005), pp. 3208-3212, ISSN 0095-1137

Rahman, M., Sultana, R., Podder, G., Faruque, A.S., Matthijnssens, J., Zaman, K., Breiman, R.F., Sack, D.A., Van Ranst, M., & Azim, T. (2005b). Typing of human rotaviruses: Nucleotide mismatches between the VP7 gene and primer are associated with genotyping failure. *Virology Journal*, Vol.2, (March 2005), pp. 24-28, ISSN 1743-422X

Rahman, M., Sultana, R., Ahmed, G., Nahar, S., Hassan, Z.M., Saiada, F., Podder, G., Faruque, A.S., Siddique, A.K., Sack, D.A., Matthijnssens, J., Van Ranst, M., & Azim, T. (2007). Prevalence of G2P[4] and G12P[6] rotavirus, Bangladesh. *Emerging Infectious Diseases*, Vol.13, No.1, (January 2007), pp. 18-24, ISSN 1080-6059

Rao, C.D., Gowda, K., & Reddy, B.S. (2000). Sequence analysis of VP4 and VP7 genes of nontypeable strains identifies a new pair of outer capsid proteins representing

novel P and G genotypes in bovine rotaviruses. *Virology*, Vol.276, No.1, (October 2000), pp. 104-113, ISSN 0042-6822

Rennels, M.B., Glass, R.I., Dennehy, P.H., Bernstein, D.I., Pichichero, M.E., Zito, E.T., Mack, M.E., Davidson, B.L., & Kapikian, A.Z. (1996). Safety and efficacy of high-dose rhesus-human reassortant rotavirus vaccines—report of the National Multicenter Trial. *Pediatrics*, Vol.97, No.1, (January 1996), pp. 7-13, ISSN 0031-4005

Rice, C.M., Franke, C.A., Strauss, J.H., & Hruby, D.E. (1985). Expression of Sindbisvirus structural proteins via recombinant vaccinia virus: synthesis, proceeding and incorporation into mature Sindbis virions. *Journal of Virology*, Vol.56, No.1, (October 1985), pp. 227-239, ISSN 0022-538X

Ruiz, A.M., Lopez, I.V., Lopez, S., Espejo, R.T., & Arias, C.F. (1988). Molecular and antigenic characterization of porcine rotavirus YM, a possible new rotavirus serotype. *Journal of Virology*, Vol.62, No.11, (November 1988), pp. 4331-4336, ISSN 0022-538X

Ruiz-Palacios, G.M., Pérez-Schael, I., Velázquez, F.R., Abate, H., Breuer, T., Clemens, S.C., Cheuvart, B., Espinoza, F., Gillard, P., Innis, B.L., Cervantes, Y., Linhares, A.C., López, P., Macías-Parra, M., Ortega-Barría, E., Richardson, V., Rivera-Medina, D.M., Rivera, L., Salinas, B., Pavía-Ruz, N., Salmerón, J., Rüttimann, R., Tinoco, J.C., Rubio, P., Nuñez, E., Guerrero, M.L., Yarzábal, J.P., Damaso, S., Tornieporth, N., Sáez-Llorens, X., Vergara, R.F., Vesikari, T., Bouckenooghe, A., Clemens, R., De Vos, B., & O'Ryan, M. (2006). Safety and efficacy of an attenuated vaccine against severe gastroenteritis. *New England Journal of Medicine*, Vol.354, No.1, (January 2006), pp. 11-22, ISSN 1533-4406

Santos, N., Lima, R.C.C., Pereira, C.F.A., & Gouvea, V. (1998). Detection of rotavirus types G8 and G10 among Brazilian children with diarrhea. *Journal of Clinical Microbiology*, Vol.36, No.9, (September 1998), pp. 2727-2729, ISSN 0095-1137

Santos, N., Volotão, E.M., Soares, C.C., Albuquerque, M.C., da Silva, F.M., Chizhikov, V., & Hoshino, Y. (2002). VP7 gene polymorphism of serotype G9 rotavirus strains and its impact on G genotype determination by PCR. *Virus Research*, Vol.90, No.1-2, (December 2002), pp. 1-14, ISSN 0168-1702

Santos, N., Volotão, E.M., Soares, C.C., Albuquerque, M.C., da Silva, F.M., Chizhikov, V., & Hoshino, Y. (2003). VP7 gene polymorphism of serotype G9 rotavirus strains and its impact on G genotype determination by PCR. *Virus Research*, Vol.93, No.1, (May 2003), pp. 127-138, ISSN 0168-1702

Santos, N., & Hoshino, Y. (2005). Global distribution of rotavirus serotypes/genotypes and its implication for the development and implementation of an effective rotavirus vaccine. *Reviews in Medical Virology*, Vol.15, No.1, (January-February 2005), pp. 29-56, ISSN 1099-1654

Santosham, M., Moulton, L.H., Reid, R., Croll, J., Weatherholt, R., Ward, R., Forro, J., Zito, E., Mack, M., Brenneman, G., & Davidson, B.L. (1997). Efficacy and safety of high-dose rhesus-human reassortant rotavirus vaccine in Native American populations. *Journal of Pediatrics*, Vol.131, No.4, (October 1997), pp. 632-638, ISSN 0022-3476

Sereno, M.M., & Gorziglia, M.I. (1994). The outer capsid protein VP4 of murine rotavirus strain Eb represents a tentative new P type. *Virology*, Vol.199, No.2, (March 1994), pp. 500-504, ISSN 0042-6822

Shinozaki, K., Okada, M., Nagashima, S., Kaiho, I., & Taniguchi, K. (2004). Characterization of human rotavirus strains with G12 and P[9] detected in Japan. *Journal of Medical Virology*, Vol.73, No.4, (August 2004), pp. 612-616, ISSN 1096-9071

Smith, G. L., Mackett, M., & Moss, B. (1983). Infectious vaccinia virus recombinants that express hepatitis B surface antigen. *Nature*, Vol.302, (April 1983), pp. 490-495, ISSN 0028-0836

Steele, A.D., Parker, S.P., Peenze, I., Pager, C.T., Taylor, M.B., & Cubitt, W.D. (1999). Comparative studies of human rotavirus serotype G8 strains recovered in South Africa and the United Kingdom. *Journal of General Virology*, Vol.80, No.11, (November 1999), pp. 3029-3034, ISSN 1465-2099

Stefan, R. (2005). Edward Jenner and the history of smallpox and vaccination. *BUMC Proceedings*, Vol.18, No.1, (January 2005), pp. 21-25, ISSN 0899-8280

Tam, D-T., Salaheddin, M.M., Riccardo, P., & Eduardo, L.F. (2004). Polio vaccines, Simian Virus 40, and human cancer: the epidemiologic evidence for a causal association. *Oncogene*, Vol.23, (n.d.), pp. 6535-6540, ISSN 0950-9232

Taniguchi, K., Urasawa, T., Kobayashi, N., Gorziglia, M., & Urasawa, S. (1990). Nucleotide sequence of VP4 and VP7 genes of human rotaviruses with subgroup I specificity and long RNA pattern: implication for new serotype specificity. *Journal of Virology*, Vol.64, No.11, (November 1990), pp. 5640-5644, ISSN 0022-538X

Timenetsky Mdo, C., Gouvea, V., Santos, N., Carmona, R.C., & Hoshino, Y. (1997). A novel human rotavirus serotype with dual G5/G11 specificity. *Journal of General Virology*, Vol.78, No.6, (June 1997), pp. 1373-1378, ISSN 1465-2099

Trinh, Q.D., Pham, N.T., Nguyen, T.A., Phan, T.G., Khamrin, P., Yan, H., Hoang, P.L., Maneekarn, N., Li, Y., Kozlov, V., Kozlov, A., Okitsu, S., & Ushijima, H. (2007). Amino acid substitutions in the VP7 protein of human rotavirus G3 isolated in China, Russia, Thailand, and Vietnam during 2001–2004. *Journal of Medical Virology*, Vol.79, No.10, (October 2007), pp. 1611-1616, ISSN 1096-9071

Turner, G.S., Squires, E.J., & Murray, H.G.S. (1970). Inactivated smallpox vaccine. A comparison of inactivation methods. *Journal of Hygiene*, Vol.68, No.2, (June 1970), pp. 197-210, ISSN 0022-1724

Tung, G.P., Shoko, O., Niwat, M., & Hiroshi, U. (2007). Evidence of Intragenic Recombination in G1 Rotavirus VP7 Genes. *Journal of Virology*, Vol.81, No.18, (September 2007), pp. 10188-10194, ISSN 0022-538X

Uchida, R., Pandey, B.D., Sherchand, J.B., Ahmed, K., Yokoo, M., Nakagomi, T., Cuevas, L.E., Cunliffe, N.A., Hart, C.A., & Nakagomi, O. (2006). Molecular epidemiology of rotavirus diarrhea among children and adults in Nepal: detection of G12 strains with P[6] or P[8] and a G11P[25] strain. *Journal of Clinical Microbiology*, Vol. 44, No.10, (October 2006), pp. 3499-3505, ISSN 0095-1137

Ulmer, J.B., Donnelly, J.J., Parker, S.E., Rhodes, G.H., Felgner, P.L., Dwarki, V.J., Gromkowski, S.H., Deck, R.R., DeWitt, C.M., Friedman, D., Perry, H.C., Shiver, J.W., Montgomery, D.L., & Liu, M.A. (1993). Heterologous protection against influenza by injection of DNA encoding a viral protein. *Science,* Vol.259, No.5102, (March 1993), pp. 1745-1749, ISSN 0036-8075

Vesikari, T., Isolauri, E., D'Hondt, E., Delem, A., André, F.E., & Zissis, G. (1984). Protection of infants against rotavirus diarrhea by RIT 4237 attenuated bovine rotavirus strain vaccine. *Lancet*, Vol.5, No.1, (May 1984), pp. 977-981, ISSN 0140-6736

Vesikari, T., Matson, D.O., Dennehy, P., Van Damme, P., Santosham, M., Rodriguez, Z., Dallas, M.J., Heyse, J.F., Goveia, M.G., Black, S.B., Shinefield, H.R., Christie, C.D., Ylitalo, S., Itzler, R.F., Coia, M.L., Onorato, M.T., Adeyi, B.A., Marshall, G.S., Gothefors, L., Campens, D., Karvonen, A., Watt, J.P., O'Brien, K.L., DiNubile, M.J., Clark, H.F., Boslego, J.W., Offit, P.A., & Heaton, P.M. (2006). Safety and Efficacy of a pentavalent human-bovine (WC3) reassortant rotavirus vaccine. *New England Journal of Medicine*, Vol.354, No.1, (January 2006), pp. 23-33, ISSN 1533-4406

Vesikari, T., Karvonen, A., Allen, F.S., & Ciarlet, M. (2010). Efficacy of the pentavalent rotavirus vaccine, RotaTeq®, in Finnish infants up to 3 years of age: the Finnish Extension Study. *European Journal of Pediatrics*, Vol. 169, No.11, (November 2010), pp. 1379-1386, ISSN 1432-1076

Vyas, G.N. & Blum, H.E. (1984). Hepatitis B virus infection-Current concepts of chronicity and immunity. *West Journal of Medicine*, Vol.140, No.5, (May 1984), pp. 754-762, ISSN 0093-0415

Weil, A.J. & Gall, L.S. (1940). Studies on the immunization of rabbits with formalinized vaccine virus. *Journal of Immunology*, Vol.38, No.1, (January 1940), pp. 1-8, ISSN 0022-1767

Wen, L., Nakayama, M., Yamanishi, Y., Nishio, O., Fang, Z.-Y., Nakagomi, O., Araki, K., Nishimura, S., Hasegawa, A., Müller, W.E.G., & Ushijima, H. (1997). Genetic variation in the VP7 gene of human rotavirus serotype 3 (G3 type) isolated in China and Japan. *Archives of Virology*, Vol.142, No.7, (July 1997), pp. 1481-1489, ISSN 1432-8798

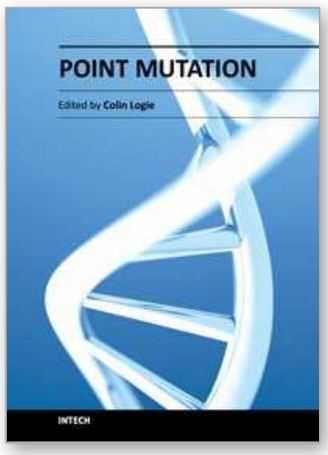
Wiktor, T.J., Macfarlan, R.I., Reagan, K.J., Dietzschold, B., Curtis, P.J., Wunner, W.H., Kieny, M.P., Lathe, R., Lecocq, J.P., Mackett, M., Moss, B., & Koprowski, H. (1984). Protection from rabies by a vaccinia virus recombinant containing the rabies virus glycoprotein gene. *Proceedings of the National Academy of Sciences USA*, Vol.81, No.22, (November 1984), pp. 7194-7198, ISSN 0027-8424

Wolff, J.A., Malone, R.W., Williams, P., Chong, W., Acsadi, G., Jani, A., & Felgner, P.L. (1990). Direct gene transfer into mouse muscle in vivo. *Science*, Vol.247, No.4949, (March 1990), pp. 1465-1468, ISSN 0036-8075

World Health Organization, 2000, Geneva, (February 2000), Available from: www.who.int/vaccines-documents/, Report of the meeting on the Future Directions for Rotavirus Vaccine Research in Developing Countries

Yap, I., Guan, R., & Chan, S.H. (1992). Recombinant DNA hepatitis B vaccine containing Pre-S components of the HBV coat protein-preliminary study on immunogenicity. *Vaccine*, Vol.10, No.7, (n.d.), pp. 439-442, ISSN 0264-410X

Zissis, G., Lambert, J.P., Marbehant, P., Marissens, D., Lobmann, M., Charlier, P., Delem, A., & Zygraich, N. (1983). Protection studies in colostrum-deprived piglets of a bovine rotavirus vaccine candidate using human rotavirus strains for challenge. *Journal of Infectious Diseases*, Vol.148, No.6, (December 1983), pp. 1061-1068, ISSN 0022-1899



Point Mutation

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This book concerns the signatures left behind in chromosomes by the forces that drive DNA code evolution in the form of DNA nucleotide substitutions. Since the genetic code predetermines the molecular basis of life, it could have been about any aspect of biology. As it happens, it is largely about recent adaptation of pathogens and their human host. Nine chapters are medically oriented, two are bioinformatics-oriented and one is technological, describing the state of the art in synthetic point mutagenesis. What stands out in this book is the increasing rate at which DNA data has been amassed in the course of the past decade and how knowledge in this vibrant research field is currently being translated in the medical world.

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