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Epidemiology of rotavirus in Portugal: G9 as a major cause of diarrhoea in non-hospitalised children

Fernanda Rodrigues [a,*], Miren Iturriza [b], Jim Gray [b], Luis Januário [a], Luis Lemos [a]

[a] *Hospital Pediátrico de Coimbra, Av. Bissaya Barreto, 3000 075 Coimbra, Portugal*
[b] *Enteric Virus Unit, Virus Reference Department, Centre for Infections, Health Protection Agency, 61 Colindale Avenue, London NW9 5HT, UK*

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Abstract

Background: Rotavirus is a major cause of gastroenteritis in children worldwide, but there is no data available on the incidence of rotavirus gastroenteritis or on the strains circulating in Portugal.
Methods: We determined prospectively the incidence of rotavirus infection in non-hospitalised children and the genotypes circulating during one winter season in the central region of Portugal.
Results: Rotavirus was found in 45% of the samples tested. The peak incidence was in February (54% positive) and March (60% positive). Genotyping was performed in 195 samples; unexpectedly, G9P[8] was present in 90% of the cases, a much higher percentage than previously reported in other countries.
Conclusions: These results contribute to the assessment of the burden of disease attributable to rotavirus in Portugal and facilitate preparation for intervention by vaccination. The predominance of G9 in Portugal is unlikely to be a local phenomenon, and may be observed elsewhere in Portugal and Europe. The epidemiology of rotaviruses in Portugal should be monitored in subsequent years.

Keywords: Rotavirus; Gastroenteritis; Genotypes

1. Introduction

Rotavirus (RV) is a major cause of acute gastroenteritis (AG) in children, being responsible for more than 500,000 deaths each year, the majority in developing countries (Lepage, 2006; Parashar et al., 2003; Rheingans et al., 2006). In addition to causing morbidity and mortality in children, RV gastroenteritis imposes a major economic burden on health care systems and families (Rheingans et al., 2006; Soriano-Gabarró et al., 2006).

The European Medicine Evaluation Agency (EMEA) has approved two vaccines for use in Europe. The formulation of a country-specific vaccination policy will require country-specific studies to more accurately understand the burden of disease caused by RV.

There is no data available on the incidence of RV gastroenteritis or on the strains circulating in Portugal.

The purpose of this study was to describe the impact of RV gastroenteritis on paediatric outpatients in the central region of Portugal, and to analyse the most common genotypes of RVs circulating during the annual epidemic season, before the introduction of RV vaccines.

2. Materials and methods

Hospital Pediátrico de Coimbra is a tertiary hospital with an emergency service (ES) that admits children between 0 and 13 years of age from all the central region of Portugal, covering a population of 400,000 children. Approximately, 50,000 children are seen annually. These may be referred

Abbreviations: RV, rotavirus; AG, acute gastroenteritis; ES, emergency service

* Corresponding author. Tel.: +351 239 480 651; fax: +351 239 717 216.
E-mail address: frodrigues@hpc.chc.min-saude.pt (F. Rodrigues).

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from other hospitals or general practitioners, but the majority are brought by their parents. There is no other paediatric ES in the city or in the surrounding area.

2.1. Study period

The study was designed to cover the RV epidemic season, which in this region is typically between January and March. Samples were collected from February to June 2006 (for technical reasons the study did not start in January).

2.2. Case definition

Children under 3 years of age, attending our ES, with symptoms of AG defined as ≥2 watery or looser than normal stools within a 24 h period, lasting <7 days and preceded by a symptom-free period of 14 days. Children were excluded if they had chronic gastrointestinal tract disease or an immunodeficiency. The study included only non-hospitalised children. Children admitted to the Short Stay Unit or to the wards were excluded from the analysis. All infections were considered community acquired.

The study was approved by the Ethical Committee. Informed consent was obtained from a parent/guardian of each child.

2.3. Stool collection and testing

A stool sample was obtained at the visit to the hospital. If a sample was not available, the parent/guardian was asked to bring a specimen to the ES within 24 h. Rectal swabs were not used. Specimens were tested for RV using a rapid test, based on the immunochromatography technique (VIKIA®, Biomerieux, France). Faecal samples positive for RV antigen were sent for genotyping (Virus Reference Department, Health Protection Agency, London). Genotyping was performed using methods published elsewhere (Iturriza-Gómara et al., 2004). Briefly, stool samples were prepared as 10% suspensions in balanced salt solution, and nucleic acid was extracted using guanidinium isothiocyanate and silica. Reverse transcription was performed in the presence of random hexamers and the cDNA used in G and P genotyping hemi-nested PCRs as previously described (Iturriza-Gómara et al., 1999, 2004).

3. Results

During the 5 months studied, 642 children under 3 years of age with AG were seen at our ES. A stool sample was collected for RV antigen detection from 475 (74%) cases. A total of 216 (45%) cases of AG were positive for RV. The proportion of RV-positive cases was as follows: February 73/134 (54%), March 74/124 (60%), April 29/85 (34%), May 18/67 (27%) and June 22/65 (34%).

A total of 195 samples were available for genotyping, of which 9 (4.6%) were negative in the PCR assay, indicating a false-positive in the antigen detection assays. This is within the expected rate of false positivity rate seen with commercially available antigen detection kits (Gray and Iturriza-Gómara, unpublished data). A total of 183 were fully characterised in the genoptyped and three were only partially genotyped. Rotavirus genotyping showed G9P[8] in 165 (90%) cases, G1P[8] in 8 (4.3%), G3P[8] in 7 (3.8%), and 1 each of genotypes G12P[8], G3+9P[8] and G1+9P[8] . G12P[8] was identified in a 3-month-old boy with a 3-day history of diarrhoea and vomiting.

Of the 183 samples, 145 were from our district and 38 (20.4%) from others districts in the Central Region of the country

4. Discussion

RV is a major cause of gastroenteritis in paediatric outpatients practice in the central region of Portugal. AG in children impacts widely on their families and society in general, leading to increased medical expenditure, loss of productivity, extra childcare costs and/or loss of already contracted childcare, and pain and suffering caused to children and their families (Rheingans et al., 2006).

The incidence of individual serotypes in a particular region can fluctuate annually, and within the same country the RV genotype distribution can differ during the same year in different regions (Santos and Hoshino, 2005).

Until recently, four RV strains (G1P[8], G3P[8], G4P[8] and G2P[4]) made up 96% of the globally identified strains. However, several recent surveys highlight the emergence of previously rare types, such as serotypes G5, G6, G8, G10, and in particular G9 (Dennehy, 2005).

The relative frequency of genotype G9 strains, usually detected in association with P[8] or P[6] appears to be increasing, and these strains already represent 4.1% of global RV infection (Santos and Hoshino, 2005).

G9 strains were initially detected in Philadelphia, during 1983–1984 in 9.2% of infants with RV disease (Clark et al., 1987), became undetectable for about one decade, and then re-emerged with an incidence of 50% of rotavirus diarrhoea in 1995–1996 (Clark et al., 2004). In Australia serotype G9 was identified for the first time in the season of 1999–2000. In the seasons of 1999–2000 and 2000–2001, it was the second most common G type, causing 10% and 18.1%, respectively, of the infections in Australia (Masendyez et al., 2000, 2001). In Europe the emergence of G9 strains was first reported in the mid-1990s (Cubitt et al., 2000). The prevalence of G9 RV varies considerably between European countries, from <0.1% to 52% (Desselberger et al., 2006). The occurrence of G9 strains in a Belgium hospital increased greatly between 1999–2000 (4.8%) and 2002–2003 (51.5%) (Rahman et al., 2005). A comparably high G9 prevalence of 53.4% was found in Bari, Italy (Martella et al., 2003). Data from Spain shows