and meat should be prepared on a separate counter or cutting board from other food items; 2) all utensils, cutting boards, and counters should be cleaned with hot water and soap after preparing other foods; 3) hands should be washed thoroughly with soap and running water after handling raw poultry or meat; and 4) poultry should be cooked thoroughly to an internal temperature of 82ºC or until the meat is no longer pink and juices run clear [8].

In summary, custard cross-contaminated by chicken served in a school appears to have been the source of infection in this outbreak of *Campylobacter* enteritis. Sanitary education of the food handlers continues to be the main control measure in foodborne outbreaks.


## Acknowledgements

We thank all the clinicians, microbiologists and public health officials involved in the study and control of the outbreak in the Autonomous Community of Madrid.


## References


1. Allerberger F, Al-Jazrawi N, Kreidl P et al. Barbecued chicken causing a multi-state outbreak of *Campylobacter jejuni* enteritis. Infection. 2003 Jan; 31(1):19-23.
2. Peterson MC. *Campylobacter jejuni* enteritis associated with consumption of raw milk. J Environ Health. 2003 May; 65(9):20-1, 24, 26.
3. Harrington P. Outbreak of *Campylobacter jejuni* Infections Associated with Drinking Unpasteurized Milk Procured through a Cow-Leasing Program. Wisconsin, 2001. MMWR Morb Mortal Wkly Rep. 2002 Jun 28; 51(25):548-9.
4. Jones IG, Roworth M. An outbreak of *Escherichia* coli O157 and *campylobacteriosis* associated with contamination of a drinking water supply. Public Health. 1996 Sep; 110(5):277-82.
5. Melby KK, Svendby JG, EggebØ T et al. Outbreak of *Campylobacter* Infection in a Subartic Community. Eur J Clin Microbiol Infect Dis. 2000 Jul; 19(7):542-4.
6. Takkinen J, Ammon A, Robstad O, Breuer T, and the *Campylobacter* Working Group. European Survey on *Campylobacter* surveillance and diagnosis 2001. Euro Surveill. 2003 Nov; 8(11):207-13.
7. Mandell/Douglas/Bennet. Principles and Practice of Infectious Diseases, 3rd Edition.
8. Graves TK , Bradley KK, Crutcher JM. Outbreak of *Campylobacter* Enteritis Associated with Cross-Contamination of Food-Oklahoma, 1996. JAMA. 1998 May 6; 279(17):1341.
9. Kirk M, Waddell R, Dalton C, Creaser A, Rose N. A prolonged outbreak of *Campylobacter* infection at a training facility. Comm Dis Intell 1997; 21:57-61.
10. Tauxe RV. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrial nations. In: Nachamkin I, Blaser MJ, Tompkins L, eds. *Campylobacter jejuni*: current status and future trends. Washington, DC: American Society of Microbiology, 1992:9-19.


ORIGINAL ARTICLES

Outbreak report

# OUTBREAKS CAUSED BY PARVOVIRUS B19 IN THREE PORTUGUESE SCHOOLS


G Gonçalves [1], AM Correia [2], P Palminha [3], H Rebelo-Andrade [4], A Alves [4]



This paper reports the study of outbreaks of an acute exanthematous disease among children of three schools in the municipality of Braga (Portugal). Laboratory tests were performed for five cases, showing that the disease was not due to infection by measles or rubella virus, and infection with parvovirus B19 was confirmed. There were 41 cases in children: 12 in the kindergarten, 17 in the secondary school and 12 in the primary school. There was only one case in a staff member, who worked in the kindergarten. Eight cases were identified among household contacts; two of them were brothers, one from the kindergarten and another from the secondary school, where the outbreak occurred after the kindergarten outbreak. The estimated values of the basic reproduction number $R_0$ were very low and it is very likely that asymptomatic infectious cases have occurred. The local health authority produced written documents and met with staff members and parents. Primary healthcare facilities and the obstetric department of the local hospital were also informed. As we are approaching the elimination of measles in Portugal and the rest of Europe, with very high vaccine coverage, it is very likely that a high proportion of infectious non-vesicular exanthemas will be due to B19 infections. This is to be taken into account in the design and conduct of surveillance activities, in the context of measles and rubella elimination programmes.



Euro Surveill 2005; 10(6): 121-4 Published online June 2005

**Key words:** B19, erythema infectiosum, exanthemas, outbreak, parvovirus


## Introduction

Erythema infectiosum (EI), or 'slapped cheek' disease, is the most common clinical manifestation of infection with parvovirus B19 (B19) in children [1]. Several diseases, like measles, rubella, scarlet fever, erythema multiform and this 'fifth disease' can cause similar acute non-vesicular exanthematous rashes and differential diagnosis is often necessary, especially in the context of measles elimination programmes in Europe [2] and worldwide [3]. Parvovirus B19 infections are usually benign and self-limiting [4] and commonly asymptomatic [1]; nevertheless, they can have important adverse effects among specific risk groups [1], namely pregnant women, immunocompromised individuals and patients suffering from chronic haemolytic anaemia. Thus, some authors have recommended the use of ELISA and PCR tests to confirm the aetiology of outbreaks [4].

In Portugal, EI and other B19 infections are not reportable to the local health authority (LHA), unlike measles and rubella. There are written recommendations on how to investigate suspected outbreaks (and isolated suspected cases) of measles [5]. We do not know of written reports of B19 outbreaks previously studied in Portugal but some serological data were published: in a blood transfusion department of a Portuguese hospital, 66.2% of health adults and 83.3% of haemophiliacs were found to be positive for B19 IgG antibodies [6].


1. Instituto Gulbenkian de Ciência, Oeiras, Portugal.
2. Unidade Operativa de Saúde Pública de Braga, Portugal.
3. Instituto Nacional de Saúde, Unidade de Herpesvirus e Rubéola, Lisboa, Portugal.
4. Instituto Nacional de Saúde, Laboratório Nacional de Referência para o Sarampo, Lisboa, Portugal.
5. Hospital de S. Marcos, Braga, Portugal.

The main purpose of investigating the reported cases of an acute exanthematous illness in three schools was to understand whether it was a dangerous public health situation and if appropriate control measures could be implemented. We were particularly concerned about the possibility of measles or rubella outbreaks. We report here the findings of the study of three outbreaks due to B19, in Braga, Portugal.

### *Alert*

On 14 January 2004, the LHA of Braga (a municipality in the North of Portugal) was informed by the warden of a kindergarten that several cases of an acute exanthematous illness among children were occurring. Similar alert phone calls were made on 17 February (secondary school) and on 26 of April (primary school).

### *Preliminary assessment*

Telephone interviews were conducted with the directors of the three schools and some general practitioners (GP) who had observed the children. Signs and symptoms were so suggestive that some physicians explicitly mentioned erythema infectiosum (EI), as the most likely diagnosis. No laboratory confirmation had been done. The first known case of erythema occurred on 31 December 2003.

## Methods

### *Laboratory study*

Blood samples were collected from five children with clinical manifestations suggestive of EI: two from the kindergarten and three from the secondary school. In the sera from those five children specific IgM and IgG antibodies against measles, and B19 were measured using ELISA techniques; a PCR technique was used to detect B19 DNA. Specific antibodies against rubella were only studied in sera from the three cases in the secondary school. Tests were performed in the laboratory of the hospital of Braga (2 cases from the kindergarten) and in the national institute of health (three cases from the secondary school).

### *Active case finding*

A written questionnaire was given to all staff members and parents from the three schools inquiring about signs, symptoms, medical care and the existence of other cases in the household. Written vaccination records were checked among all children in the kindergarten and all cases in the other two schools.

### *Case definition*

Probable case of EI: erythema on the face, extremities or trunk, between 30 December 2003 and 8 of May 2004, in members of the three schools community (children and staff) and their household contacts.

Confirmed case of EI: probable case with laboratory confirmation (either by ELISA or PCR) of acute infection with B19 or epidemiological link with a confirmed case.

### *Statistical methods*

To compare attack rates [7] we used $\chi^2$ tests. The basic reproduction number $R_0$ has been defined as '*the average number of persons directly infected by an infectious case during the entire infectious period, when he enters a totally susceptible population*' [8]. In order to estimate the basic reproduction number $R_0$ of infection by B19, we used a formula which had already been used during measles outbreaks [9]. This was performed for the kindergarten, the Class 5A of the secondary school and in Class 1 in the primary school. For the purpose of this estimate in these groups we assumed that there were only susceptible children before the outbreak and that transmission (after the primary cases) occurred inside these groups, with homogenous mixing. For the two class groups with 10 symptomatic cases, we made the estimates for the whole range of possible numbers of asymptomatic infections, from zero to sixteen.

## Results

### *Laboratory study*

The five cases with clinical manifestations suggestive of EI were confirmed as being cases of infection with B19. Most cases were positive for both specific IgM and specific DNA [TABLE 1]; in most cases, the results were compatible with what was expected for the time when blood collection took place [1]. The three cases from the secondary school were negative for both measles and rubella IgM search. Four of them had serological markers (positive IgG) for immunity against measles. The one exception was a girl, born in 1993, who had received MMR at 9 months of age, during the last 1993/1994 measles epidemic observed in Portugal; following the standard procedure, she had been revaccinated at 15 months of age and failed to show protective measles antibodies at 11 years of age (last case in Table 1). We do not know the cause of this finding but it could be related to previously expressed concerns about the consequences of vaccinating too early against measles [10].

TABLE 1

**Laboratory results of the study of five children with clinical manifestations of erythema infectiosum. Braga, Portugal**

| Case | Measles | Rubella | Parvovirus B19 | Parvovirus B19 |
|---|---|---|---|---|
| Age / Sex / School | Serology | Serology | Serology | PCR (DNA) |
| 4 / M / kindergarten | IgM negative<br>IgG positive | Not tested | IgM negative<br>IgG positive | Positive |
| 5 / M / kindergarten | IgM negative<br>IgG positive | Not tested | IgM negative<br>IgG positive | Positive |
| 11 / F / secondary | IgM negative<br>IgG positive | IgM negative<br>IgG positive | IgM positive<br>IgG positive | Positive |
| 10 / M / secondary | IgM negative<br>IgG positive | IgM negative<br>IgG positive | IgM positive<br>IgG positive | Positive |
| 11 / F / secondary | IgM negative<br>IgG negative[1] | IgM negative<br>IgG borderline | IgM borderline<br>IgG positive | Positive |

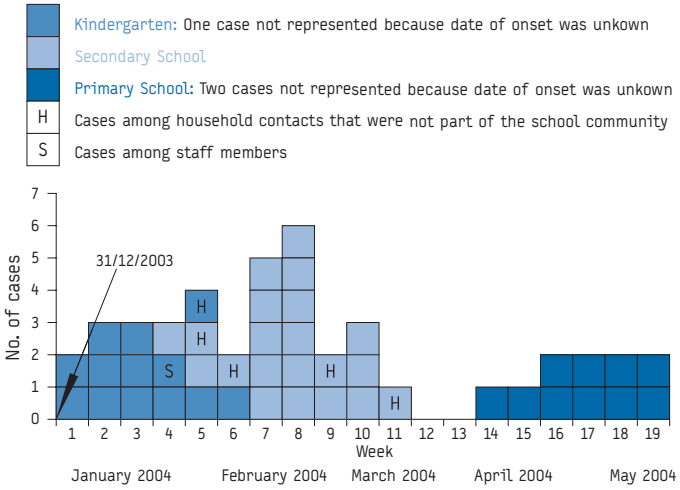
1. This child, born in 1993, had received MMR at 9 months of age during the 1993/1994 measles epidemic observed in Portugal. Following the standard procedure, he was revaccinated at age 15 months.

## Description of the outbreaks

From the laboratory results and questionnaire data, we identified 35 confirmed cases (in the kindergarten and secondary school) and 12 probable cases (in the primary school). We had no questionnaire data from 2 of the 12 cases in the primary school. The most frequent reported clinical manifestations were facial erythema (93.3%), rash on the arms (79.1%), rash on the legs (77.4%), itching (12.2%) and fever (11.1%); only one case reported arthralgia and the facial rash recurred in two cases, 4 and 26 days after the first day of onset. The three epidemic curves, shown in Figure 1, spanned over a period of 19 weeks.

FIGURE 1

**Epidemic curves of three outbreaks of erythema infectiosum in Braga, Portugal, December 2003-May 2004**




The 50 children in the kindergarten were divided in two groups (4 years and 5/6 years), with most activities in separate rooms and meals in a common room. The 12 cases among children occurred between 31 December 2003 and 2 February 2004. There was only one case among the 6 staff members [TABLE 2]: the woman directly in charge of the 4 year old group. The attack rate in the 5/6 year old group (AR=9/25=36%) was significantly higher (P=0.047) than among children in the 4 year old group (AR=1/25=12%). Two cases were reported in household contacts: the mother of the child with onset of disease on 5 January became ill on 27 January; the brother of the child with onset of disease on 17 January became ill on 11 February and was a student of Clasee 5B in the secondary school; these two children had a cousin in Class 5A in the secondary school who was the first case from that school (19 January). All children in the kindergarten had been vaccinated with MMR.

TABLE 2

**Cases of infection with B19 in the three schools in Braga, Portugal**

| School | Children | | Staff | | Household members[1] |
|---|---|---|---|---|---|
| | No. of cases | No. at risk (%) | No. of cases | No. at risk (%) | |
| Kindergarten | 12/50 | 24 | 1/6 | 16.7 | 1[2] |
| Secondary | 17/735 | 2.3 | 0 | 0 | 4[2] |
| Primary | 12/364 | 3.3 | 0 | 0 | 0 |

1. Data on the number of persons by household is not precise or complete enough to estimate attack rates.
2. Household members that were not part of the school communities studied here. In addition to these, a child in the 5-6 years age group at the kindergarten fell ill on 17 January. This child had a cousin in the class 5B who fell ill on 19 January, and a brother in class 5A who fell ill on 11 February.

The 735 children in the secondary school were aged between 10 and 15 years; they were distributed into 28 classes (6, 7, 5, 5, and 5 classes respectively in Classes 5 to 9). The 17 cases in children occurred between 19 January and 5 March 2004; they were aged 10 to 14 years. The attack rate in children at the secondary school was much lower than in the kindergarten [TABLE 2], but big differences were observed between classes within the school. Cases were only observed in four classes, with significantly different (P=0.0015) ARs between them. Observed ARs were 38.5% in Class 5A, 19.2% in Class 5B, and 3.8% in the Class 6F. There was only one case in Class 8C (the brother of one of the cases from Class 5B). No cases were observed in Classes 7 and 9 or in the remaining groups in Class 8. No cases were reported in staff members. Among household contacts, six cases were identified, each from the family of a different case: a father, a mother and four brothers who attended different schools (the kindergarten, another class of the same secondary school, and two other secondary schools not studied in this investigation). All 17 cases had been vaccinated with MMR.

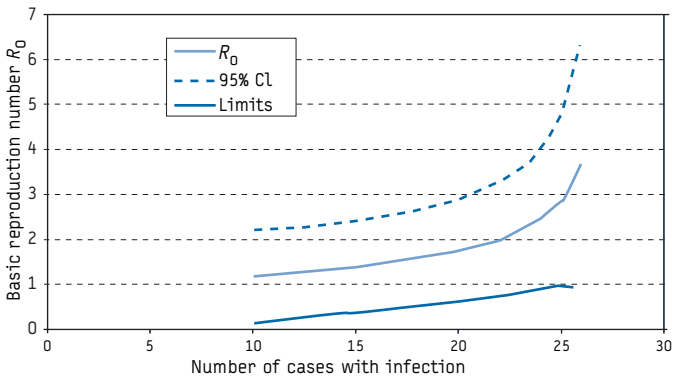
The 364 children in the primary school were aged between 6 and 10 years; they were distributed into 16 classes (4 in each class from Class 1 to Class 4). Ten cases among children from one of the classes in Class 1 (AR of 38.5%), occurred between 2 April and 8 May. Two more cases were observed among children from a single class in Class 3 but we do not have the precise date of onset. No cases were reported among staff members or household contacts. All cases had been vaccinated with MMR.

### *Estimates of the basic reproduction number $R_0$*

The basic reproduction number $R_0$ estimated for the kindergarten was 1.108 (95% CI 0.220-1.996) while for Class 5A of the secondary school and Class 1 of the primary school it was 1.184 (95% CI 0.141-2.227). These estimates assume that no asymptomatic infections had occurred, which is not likely to be true. The real $R_0$ value depends on the number of asymptomatic infections that have occurred [FIGURE 2]; point estimates for $R_0$ varied from 1.184 to 3.706 for asymptomatic infections varying from zero to sixteen.

FIGURE 2

**Estimates of the basic reproduction number $R_0$ of B19 infection in Class 1 of a primary school, and Class 5 of a secondary school, by number of infections[1]**




1. Values were calculated using the formulae published in [9]; the possible number of infections in each group, ranges from 10 (no asymptomatic infections) to 26 children; the actual number of asymptomatic infections is not known.

## Control and prevention measures

Though evidence for effective preventive measures may still be insufficient [7], some actions can be taken [8], to avoid exposure or minimise the consequences of the infection among pregnant women and other high risk groups [1,7]. Procedures were initiated even before laboratory confirmation. Aware of the importance of communication and reassurance [7], the LHA produced and distributed written information to school staff members and parents; written information on the situation and the disease, with a reminder about the risk groups, was also sent to general practitioners, in the municipality, and to the obstetric department of the local (District) Hospital. Except for cases with fever, exclusion from school was not performed because communicability is greatest before the onset of rash and probably not communicable after that [9]. Besides the written information, the LHA met with the staff members of the kindergarten on 15 January, to inform them about the disease, potential consequences and measures to be taken. In the secondary school, with the same purpose, the LHA met staff members and students on 18 February, and parents on 25 February.

## Discussion

After studying these three outbreaks, we had informal knowledge about similar outbreaks occurring in other schools of the same municipality and in a neighbouring one, but we have no information

on whether they were due to B19 infection or not. It was clearly confirmed that the three epidemics of exanthematous disease, studied in Braga, were not due to measles or rubella; they resulted from infection with B19. It is very likely that they were part of a general increase in the incidence of the disease in a wider geographical area in the northern Portugal. It is likely that some children from the kindergarten transmitted the infection to their older siblings who later introduced the infection in the secondary school, but we can not prove that because the alternative explanation that the sibling could just have been infected in his/her own school is possible. No epidemiological link was established between the outbreak in the primary school and the two previous ones, but it is likely that we have missed the detection and study of many isolated cases and institutional outbreaks.

The distribution in time of some cases among household contacts and within the schools suggests that there had been asymptomatic infections, links that were missed in the chain of transmission. This is also suggested by the small values of $R_0$ estimated if no asymptomatic infections are assumed; furthermore, assuming heterogeneity in mixing generally would increase $R_0$ estimates [9]. Other authors have reported the occurrence of asymptomatic infectious infections by B19 [1,11]. Better estimates can be obtained when immune status of people is studied before and after the outbreak [9], which would make it possible to know the precise number of asymptomatic cases.

The age distribution of cases in the primary and secondary schools (in younger age groups) and the lower ARs among staff members is compatible with the existence of immunity due to previous infections. Nevertheless, the higher attack rate in the older group of the kindergarten is not consistent with this explanation. Our data are consistent with seasonal variation observed elsewhere [1], with increased incidence in months of late winter and early spring. But the lack of a surveillance system prevents us to have precise data on the epidemiology of infections due to B19 in Portugal: for example, we do not know about cyclic variation of annual incidence [1,12].

As we are approaching the elimination of measles in Portugal (and Europe), with very high vaccine coverage with MMR and a two dose schedule, it is very likely that a high proportion of infectious non-vesicular exanthems (isolated and in outbreaks) will be due to B19 infections. This is to be taken into account in the design and conduct of surveillance activities [2,5], in the context of measles and rubella elimination programmes: study protocols should include data collection procedures and laboratory tests able to confirm or discard the diagnosis of B19 infections, among other causes of non-vesicular exanthems. High MMR vaccine coverage among children within the age groups in these schools is consistent with routine data from Braga district.

Fortunately, most B19 infections are benign. To minimise the consequences among the small number of people from risk groups [1] guidelines already exist [1,13] that should be consulted and publicised when outbreaks occur, while implementing evaluations of the effectiveness of such recommended procedures.

## Acknowledgements

We acknowledge the parents and staff members from the three schools for their cooperation during the study of the outbreaks. Manuela Bacelar collected the blood samples. Cristina Paulo and Gabriela Gomes made important comments on previous versions of this paper.

## References

1. Heegaard ED, Brown KE. Human Parvovirus B19. Clin Microbiol Rev. 2002; 15(3): 485-505.
2. Ronveaux O, Bosman A, Reintjes R, Conyn-van Spaendonck MAE. Descriptive epidemiology of exanthems in Rotterdam region, January 1997 to June 1998. Euro Surveill. 1998 Dec; 3(12):122-124.
3. Nur YA, Groen J, Yussuf MA, Osterhauss ADME. IgM antibodies in hospitalized children with febrile illness during an inter-epidemic period of measles, in Somalia. J Clin Virol. 1999; 12: 21-25.
4. Hoebe CJPA, Class ECJ, van Steenbergen JE, Kroes ACM. Confirmation of an outbreak of parvovirus B19 in a primary school using IgM ELISA and PCR on thumb prick blood samples. J Clin Virol. 2002; 25: 303-307.
5. Estratégias de luta contra o sarampo (2ª fase). Circular Normativa Nº8/DT de 22/07/1999. Lisbon, Ministry of Health, Direcção-Geral da Saúde, 1999.
6. Araújo FM, Koch MC, Araújo AR. Prevalence of parvovirus B19 infection in Portugal. Blood Coagul Fibrinolysis. 1995 Oct; 6(7):687.
7. Last JM (Editor). A Dictionary of Epidemiology, 2nd edition. Oxford University Press. New York, Oxford, 1988.
8. Giesecke J. Modern Infectious Disease Epidemiology. Edward Arnold. London, Sydney, Auckland, 1994.
9. Mossong J, Muller CP. Estimation of the basic reproduction number of measles during an outbreak in a partially vaccinated population. Epidemiol Infect. 2000; 124: 272-278.
10. Wilkins J, Wehrle PF. Additional evidence against measles vaccine administration to infants less than 12 months of age: altered response following active/passive immunization. J Pediatr. 1979 Jun; 94(6):865-9.
11. Plummer FA, Hammond GW, Forward K, et al. An erythema infeciosum-like illness caused by human parvovirus infection. N Engl J Med. 1985 Jul 11; 313(2):74-9.
12. Van Steenbergen JE. Increase in parvovirus infections in the Netherlands and management of exposed pregnant women. Eurosurveillance Weekly 1998; 2(33): 13/08/1998. http://www.eurosurveillance.org/ew/1998/980813.asp
13. Crowcroft NS, Roth CE, Cohen BJ, Miller E. Guidance for control of parvovirus B19 infection in healthcare settings and the community. Public Health Med. 1999 Dec; 21(4):439-46.