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An outbreak of monophasic and biphasic *Salmonella* Typhimurium, and *Salmonella* Derby associated with the consumption of dried pork sausage in Castellon (Spain)

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ABSTRACT

An outbreak of *S.*Typhimurium occurred in several towns and cities in the province of Castellon (Spain) between 23 February and 27 May 2011. On April 5, the microbiology laboratory of a hospital in Castellon alerted the health authorities to the increase in *S.*Typhimurium isolated in fecal culture of children with gastroenteritis. The serotype and phage-type of 83 positive cases of *S.*Typhimurium isolated in these period included 49 monophasic/biphasic *S.*Typhimurium phage type 138, phage type 193, *S.*Derby, and 34 other *S.*Typhimurium phage-types. The median of age of patients was 4 years with a range of 0.6–80 years, and the 18% of patients were hospitalised. Two incident matched case–control studies were carried out; the first with *S.*Typhimurium phage type 138, 193, and *S.*Derby cases and the second with the other cases. The two studies found that the consumption of brand X dried pork sausage, purchased in a supermarket chain A, was associated with the disease (matched Odds Ratio [mOR] = 13.74 95% Confidence Interval [CI] 4.84–39.06 and mOR = 8.20 95% CI 2.32–28.89), respectively). *S.*Typhimurium phage type 193 and *S.*Derby were isolated in the food taken from the household of two patients and from the supermarket chain's A central warehouse. The pulsed-field gel electrophoresis study confirmed the similarity of the strains from the patients and the food. On May 25 2011, a national food alert led to the withdrawal of the food from the chain A and the outbreak ended.

Un brote de *Salmonella* Typhimurium monofásica y bifásica y *Salmonella* Derby asociado al consumo de longaniza seca de cerdo en Castellón (España)

Palabras clave:
Salmonella typhimurium
Monofásica
Brote
Niños
Longaniza seca cerdo

RESUMEN

Entre el 23 de febrero y el 27 de mayo del 2011, un brote de *Salmonella* Typhimurium ocurrió en varios pueblos y ciudades en la provincia de Castellón (España). El día 5 de abril del 2011 el laboratorio de Microbiología de un hospital de Castellón alertó a las autoridades sanitarias del incremento de aislamientos de *S.*Typhimurium en coprocultivos de niños con gastroenteritis. El serotipo y fagotipo de 83 casos positivos a *S.*Typhimurium aislados en este periodo incluyó 49 casos con monofásica/bifásica *S.*Typhimurium

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fagotipo 138, fagotipo 193, y *Salmonella* Derby y otros 34 casos con distintos *S.*Typhimurium fagotipos. La mediana de los pacientes era de 4 años, con un rango de 0,6 a 80años. Dos incidentes casos-control apareados fueron llevados a cabo, el primero con los casos *S.*Typhimurium fagotipos 138,193 y *S.*Derby, y el segundo con los demás casos. Los 2 estudios encontraron que el consumo de la marca X de longaniza seca de cerdo comprada en una cadena de supermercados A estaba asociado con la enfermedad (odds ratio apareada [ORa]=13,74; intervalo de confianza [IC] del 95%: 4,84-39,06, y ORa=8,20; IC95%: 2,32-28,89, respectivamente). *S.*Typhimurium fagotipo 193 y *S.*Derby fueron aislados en dicho alimento recogido en la casa de 2 pacientes y en el almacén central de la cadena A de supermercados. La electroforesis en gel de campo pulsado confirmó la similitud de las cepas de los pacientes y del alimento. El día de 25 de mayo de 2011 una alerta alimentaria nacional obligó a la retirada del alimento de la cadena A y el brote terminó.

Introduction

In Spain during August 1997, a monophasic *S.*Typhimurium (4,5,12:i:-) was isolated from human and food samples with affecting of 14 of the 17 Spanish Autonomous Communities. All positive food isolates corresponded to pork and pork products and it was indicated that this strain "has emerged and spread to human in Spain, probably with contaminated pork as the source".[1,2] From 1998 to 2000 in New York, a monophasic variant 4,5,12:i:- caused a food poisoning outbreak.[3] In the Europe Union an increase of this monophasic variant of serovar Typhimurium has been observed over the last ten years, and the likely reservoir of infection, as in the Spanish case, is pigs.[4,5]

On April 5 2011, the Microbiology Laboratory of the General Hospital in Castellón (Spain) alerted the Epidemiology Division of the Public Health Center to a major increase of positive fecal culture *S.*Typhimurium in children with gastrointestinal symptoms. In this context, the aim of this study was to identify the cause of the infection and to take measures in order to control and prevention further spread of the disease.

Methods

The outbreak occurred during the local holydays, from March 26 to April 3 2011, in the city of Castellon, the capital of the province of Castellon with 170,000 inhabitants. A descriptive epidemiologic study was begun to considering the case definition and characteristics of patients: clinical symptoms, evolution and time and place of each case and potential risk factors of salmonellosis. All *S.*Typhimurium positive cases from the General Hospital of Castellon were investigated. Two incident matching case–control studies were then begun when the descriptive study generated a hypothesis relating to the sources of infection and before the serotyping and phage typing of the *S.*Typhimuriun were carried out. When the serotyping and phage typing were performed, positive cases of *S.*Typhimurium from the Microbiology Laboratory of the Hospital La Plana in Vila-real, a city of 50,000 inhabitants situated 6 km from Castellon, were included in the study. Once these results had been obtained, two matched case–control studies were performed.

In the first incident matched case–control study, the definition of case was a patient during the period February to May 2011 with positive monophasic/biphasic *S.*Typhimurium phage type 138, *S.*Typhimurium phage type 193 or *S.*Derby isolated from fecal urine or blood cultures. In the second incident matched case–control study, the definition of case was a patient during the period February to May 2011 with positive *S. Typhimurium* phage type 104b, 21, U311, U302, 195, and un-typable phage types isolated from fecal cultures.

The matched case–control studies (1 case:2 controls), were matched by age (less than 1 year) and gender, and each control was chosen at random from the list of patients of the pediatrician or general practitioner who attended the case. Before contact was made with controls, their pediatrician or general practitioners were consulted and verbal consent was obtained. Verbal consent to participate was also obtained from the controls or their parents. An ad hoc questionnaire was used to obtain information on demographic characteristics and potential risk factors; the same questionnaire was used for cases and controls. Information on food exposure was obtained from questions about the consumption of the presumed source foods and other risk factors in a period of up to 15 days before the start of symptoms in the cases; for controls, enrolment was considered equivalent to the day symptoms started in the cases. If the control had suffered gastroenteritis in the 15 days before the interview, he or she was excluded. Information about cases was obtained through face to face or telephone interviews by the epidemiology division staff. Telephone interviews were used with the controls, and if contact had not been established after five telephone calls, another control was randomly chosen from the list.

Microbiological study

- Isolation of *S.*Typhimurium and *Salmonella* spp from human clinical specimens was performed by the Microbiology Laboratories at the Hospital General of Castellon and Hospital La Plana of Vila-real.
- Isolations of *S.*Typhimurium and *Salmonella* spp from food samples were carried out by the Microbiology Laboratory at the Public Health Center and the Microbiology Laboratory at the Valencia Public Health Center.
- Serotyping and phage-typing of *Salmonella* strains from human and food samples. *Salmonella* isolates were serotyped at the Spanish National Reference Laboratory Majadahonda (Madrid) for *Salmonella* by the slide agglutination method using commercial antisera (Bio-Rad; Statents Serum Institut; Izasa).
- Pulsed-field gel electrophoresis. *Salmonella* isolates were characterized by PFGE using XbaI enzyme (Fermentas Life Sciences) for total DNA digestion, following the PulseNet-Europe protocol (http://www.cdc.gov/pulsenet/protocols.htm) at the National Reference Laboratory.

Statistical methods

Descriptive statistics were calculated. Crude and adjusted matched odds ratio (mOR) were estimated as measures of associations between the disease and risk factors with 95% confidence intervals (CI) calculated by conditional logistic regression following the matched case–control study using the Stata® 9.0 program.[6] Epi Info® version 6[7] was used to calculate the dose-response test and attributable risk fraction.

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