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An outbreak of *Salmonella dublin* infection in England and Wales associated with a soft unpasteurized cows' milk cheese

H. MAGUIRE[1], J. COWDEN[1], M. JACOB[2], B. ROWE[3], D. ROBERTS[4], J. BRUCE[1] AND E. MITCHELL[1]

[1] *Communicable Disease Surveillance Centre,* [3] *Division of Enteric Pathogens,* [4] *Food Hygiene Laboratory, Central Public Health Laboratory, Public Health Laboratory Service, 61 Colindale Avenue, London NW9 5HT*
[2] *Ministry of Health, London*

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SUMMARY

An outbreak of *Salmonella dublin* infection occurred in England and Wales in October to December 1989. Forty-two people were affected, mainly adults, and most lived in south-east England. Microbiological and epidemiological investigations implicated an imported Irish soft unpasteurized cows' milk cheese as the vehicle of infection. A case-control study showed a statistically significant association between infection and consumption of the suspect cheese ($p = 0{\cdot}001$). *Salmonella dublin* was subsequently isolated from cheeses obtained from the manufacturer's premises. Initial control measures included the withdrawal of the cheese from retail sale and a Food Hazard Warning to Environmental Health Departments, as well as a press release, from the Department of Health. Subsequently, a decision was taken by the manufacturer to pasteurize milk used in the production of cheese for the UK market and importation of the cheese resumed in June 1990.

INTRODUCTION

Salmonella dublin is a rare cause of human infection in England and Wales (E & W). Between 1981 and 1990 the PHLS Division of Enteric Pathogens (DEP) made 430 identifications of the organism. In November and the first week of December 1989, 41 identifications of this organism were made, representing a more than fourfold increase above that expected in a 2-month period based on previous experience.

In November 1989, a PHLS laboratory in north-west England isolated *S. dublin* from an imported soft unpasteurized cows' milk cheese, sampled after four people who had eaten the cheese, out of six who attended a dinner party, became ill. No faecal samples were obtained. A second PHLS laboratory referred to the DEP an isolate of *S. dublin* from an unlabelled soft cheese eaten by four ill persons who had *S. dublin* cultured from faecal specimens.

An Incident Committee was set up at the Communicable Disease Surveillance Centre (CDSC) and a PHLS investigation was instituted to identify a possible common source of infection.

METHODS

Primary cases were defined as persons with a gastrointestinal illness who had *S. dublin* cultured from a faecal specimen, the identity of which was confirmed at the DEP Colindale after 1 October 1989. Cases in households where other members had diarrhoea in the 2 weeks before the onset of illness in the case were considered to be secondary.

Case finding was carried out by sending a facsimile transmission letter to all Directors of PHLS Laboratories in E & W, requesting any organisms of antigenic structure 9, 12:G:- to be sent to the DEP for serotyping and a similar request was made on the front page of the Communicable Disease Report (CDR) [1]. *S. dublin* has the antigenic structure 9, 12:g, p:-, and *S. enteritidis* the structure 9, 12:g, m:-.

A detailed semi-structured questionnaire was administered by telephone to affected people, to identify possible common risk factors. Of four primary cases interviewed in this way, two reported that they had eaten the same brand of imported Irish soft cows' milk cheese, a third had eaten soft cheese but was not sure which brand, and the fourth had not eaten any cheese in the week before the onset of symptoms. After these preliminary enquiries CDSC received a further report of diarrhoeal illness in a woman who had apparently eaten an imported soft cows' milk cheese.

A case-control study was undertaken to test the null hypothesis that cases were no more likely than controls to have eaten the particular brand of Irish soft cheese in the 7 days before onset of illness. Three control subjects, matched with cases for neighbourhood, sex, and age, were nominated by persons in the case household. Cases involved in the original clusters and those interviewed in the preliminary enquiries were not eligible for entry to the case-control study. Cases whose date of onset of illness preceded 1 September 1990, whose recall would have been limited, or who had been away from home overnight in the week before the onset of illness were interviewed but were not asked to nominate controls. Cases and controls were interviewed by telephone using a structured questionnaire.

The data were entered onto computer using the Epi-info computer package and the statistical analysis was carried out using an exact one-tailed probability test taking into account variable matched groups [2].

Samples of the suspect cheese and a selection of other soft unpasteurized cheeses on retail sale in London were obtained by CDSC and cultured by the PHLS Food Hygiene Laboratory (FHL). Four PHLS laboratories also obtained samples of the suspect cheese from local retail outlets. The premises of the dairy farm producing the cheese was inspected by Environmental Health Officers (EHOs) within the Republic of Ireland, and also by an EHO from the Department of Health in England, as well as staff from CDSC and the PHLS FHL. Samples of cheese, deep frozen lactic culture, unsalted curd, cheese rennet, brine, and environmental swabs from walls, work surfaces, cheese vats, cheese moulds, and curing shelves were obtained for culture by laboratories including the FHL. Samples were transported in an insulated container by road and air. Subsequently the milking herd were screened for evidence of faecal excretion of salmonella.

At the FHL, 25 gram samples of cheese plus a portion of a 225 ml volume of 1% buffered peptone water plus 0·6% tergitol 7 (sodium heptadecyl sulphate, BDH)

(BPWT) were homogenized and the mixture transferred to the remainder of the BPWT contained in a sterile 2 lb screw-capped glass jar. Tergitol 7 was added to the pre-enrichment medium to improve the dispersion of the fat in the cheese. The jars were incubated at 37 °C for 20 h, when 10 ml volumes of the pre-enrichment culture were transferred to 100 ml volumes each of selenite F, tetrathionate (Rolfe A), and Rappaport-Vassiliadis (RV) broths. After incubation at 43 °C the broths were subcultured after 24 h, and also after 48 h, to Brilliant Green (BG) and deoxycholate citrate with 1% sucrose (DCA) agars respectively. Plates were incubated at 37 °C for 24 h (BG plates) and 48 h (DCA plates). It has been shown that incubation at a higher temperature (43 °C), and the use of a fat emulsifier such as tergitol, increase the likelihood of isolation of salmonella [3–5]. Suspect colonies were confirmed as salmonella by biochemical tests and as *S. dublin* by serotyping at DEP.

RESULTS

Between 1 October and 8 December 1989, the DEP received a total of 44 isolates of *S. dublin* from 42 persons. Thirty-five persons had the organism cultured from a faecal specimen and seven from another type of clinical specimen. Of the 7, 3 had isolates from blood cultures, 1 from joint fluid, 1 from both a joint and blood, 1 from urine, and 1 from a wound (Fig. 1).

Of the 35 persons with faecal isolates 33 were primary cases, the other 2 were known to be symptomless and therefore did not fulfil the case definition. Four persons included in the cluster mentioned in the introduction were not interviewed. It was possible to complete the questionnaire for 25 of the remaining 29 primary cases. Four others could not provide information about exposure prior to onset of illness because of difficulty with the English language in two, and because there was no definite date of onset of illness in the other two (see Fig. 1).

Descriptive epidemiology

Of the 25 primary cases interviewed 11 were male and 14 female. Their ages ranged from 1 year and 5 months to 60 years (median 41 years). The dates of onset of illness were from 20 July to 10 November with a peak in late September to mid-October. Most of the 25 lived in south-east England. Three were admitted to hospital and 18 consulted their General Practitioner. The illness laster 3–21 days (median 5 days). Of these 25, 12 had eaten the suspect brand of soft cheese in the week before onset of illness and 2 had eaten an unspecified soft cheese.

Analytical epidemiology

Of the 25 primary cases, 4 were ineligible for the case-control study because they were involved in the preliminary enquiries which led to hypothesis generation.

Of the remaining 21, a further 13 were excluded for a variety of reasons. One refused to nominate controls, 10 had spent nights away from home in the week before onset of illness, in one the onset date of illness was uncertain; and in another the onset date preceded 1 September. The remaining 8 cases and 18 matched controls were included in the case-control analysis. A significant association was found between illness and the consumption of the suspect brand of Irish soft unpasteurized cheese in the week before the onset of illness, $p = 0{\cdot}001$ (Table 1).

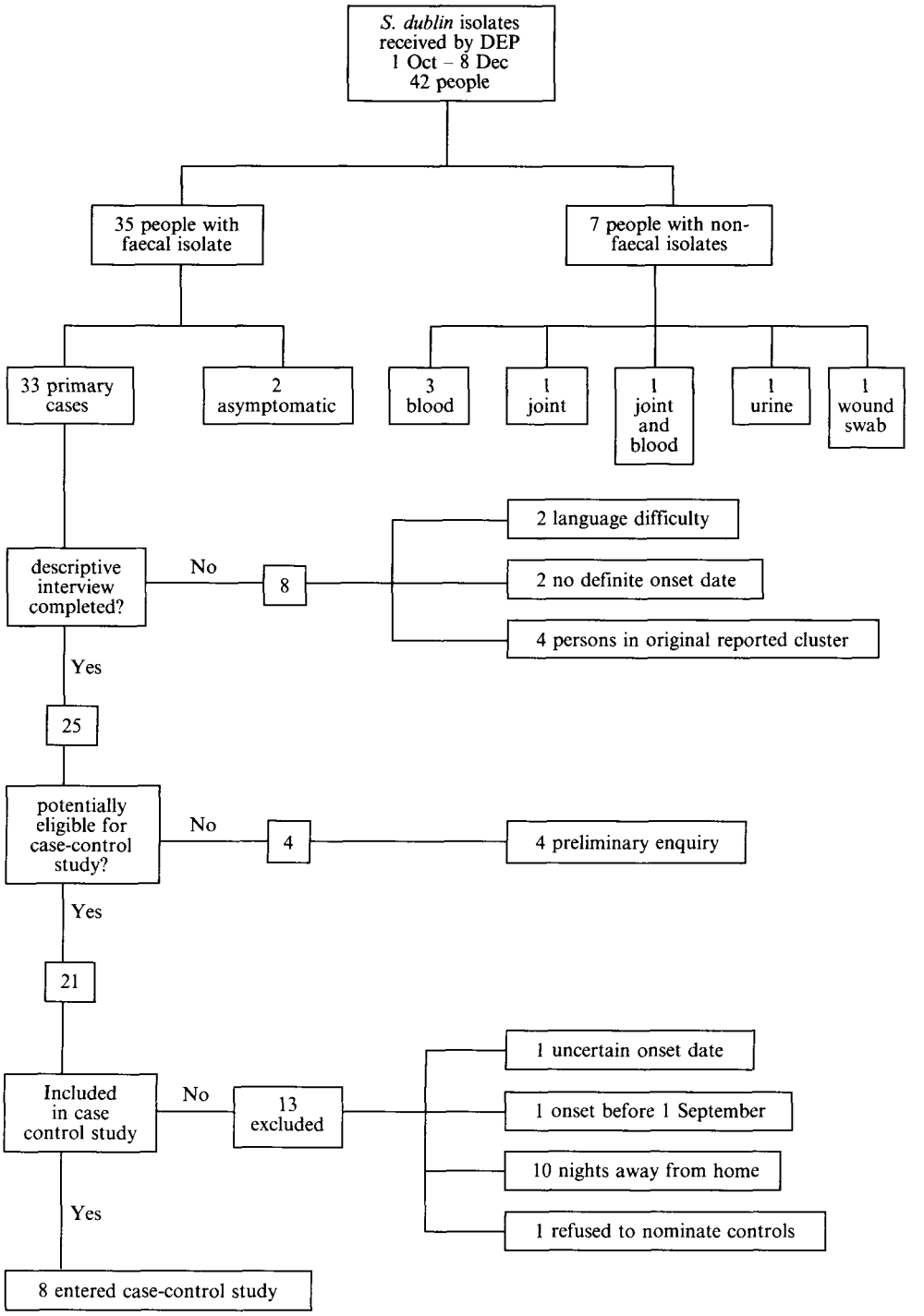

Fig. 1. Schematic diagram of the course of the outbreak.

Table 1. *Consumption of suspect cheese by cases and matched controls*

		Cases	Controls	Totals
Suspect Cheese	Yes	5	0	5
	No	3	18	21
Totals		8	18	26

Matched analysis, p = 0·001.

Microbiology and environment

In addition to the isolation of *S. dublin* from cheeses associated with illness, described in the introductory section, *S. dublin* was cultured, at the FHL, from samples of cheese which were obtained direct from the manufacturer's premises. These included 9 of 15 samples of the cheese, as well as cheese curd, from 4 batches of cheese, manufactured between 3 and 11 December. No antibiotic resistance was found. No salmonella was cultured from suspect or other unpasteurized cheeses purchased in London, or elsewhere in England. No isolations were obtained from environmental samples.

The cheese was produced by a small farm-based family business, and sold through specialist cheese shops and market stalls. About 1200 lb of cheese were produced each week of which approximately 350 lb was imported into the UK.

At the farm, unpasteurized cows' milk was incubated at 30 °C with starter culture for 1 h, rennet was then added, and the mixture held for 30 min for gelling and cutting. The curd was cooked to 34–35 °C to seal it, and then the product was dried and matured in a curing room on open wooden shelving for 2–3 weeks.

The raw milk was tested every 3 weeks at a local laboratory for listeria, salmonella, and coliforms. In addition, a cheese from each batch was tested for salmonella, listeria, and coliforms by tube and plate counts, and for the presence of *Escherichia coli* at a separate 'Quality Assurance' laboratory.

Subsequent screening of the milking herd was reported to have revealed the presence of four cows excreting *S. dublin*. All cows were asymptomatic.

Control measures

The Department of Health issued a Food Hazard Warning to Environmental Health Departments in England and Wales, advising the withdrawal of the cheese from shop shelves, and a press release was made advising members of the public to cease eating the cheese. Importation of the cheese was halted, and after consultation with the Irish Department of Health, manufacture of cheese was stopped by an order of the local health authorities. Following this, the number of identifications of *S. dublin* at the DEP fell to previous levels.

DISCUSSION

This incident was detected because laboratories referred salmonella isolates which they were unable to type to the DEP. *S. dublin* is a rare cause of human infection in the UK, but is a common bovine pathogen constituting between 24 and 43% of all salmonella isolations in the United Kingdom, from cattle, between

1978 and 1989, and 41% in 1989 [6]. *S. dublin* is host adapted to cattle and rarely infects other animals, residing in the gall bladder of the animal to be excreted in faeces [7]. It has a characteristic biotype and can cause severe invasive disease in humans [8].

Of the strains referred to the DEP between 1981 and 1990, 25% were from blood cultures, compared with 1·5% for all serotypes of salmonella [9]. Five of the 42 persons affected in this outbreak had evidence of invasive disease with septicaemia or joint involvement.

In this outbreak the results of a case control study showed a statistically significant association between *S. dublin* infection and eating a particular brand of imported Irish soft unpasteurized cows' milk cheese. Microbiological studies revealed the presence of the organism in a sample of the cheese which was thought to have caused illness, and in a similar but unlabelled cheese which caused gastrointestinal illness in persons from whom *S. dublin* was isolated. Furthermore, samples of cheeses obtained from the dairy, examined at the FHL, yielded *S. dublin*, and *S. dublin* was also reported to have been found in faeces from cows in the milking herd. This suggested that the source of infection was faecal contamination of the milk used in cheese production, which was not pasteurized.

In the USA, from 1971–5 there was a fivefold increase in the annual incidence of reported *S. dublin* infection and nearly 40% investigated cases had drunk raw milk [10]. In California it has been estimated that more than one third of reported *S. dublin* infections during 1980–3 were attributable to raw milk consumption, and that among raw milk consumers, more than 95% infections were acquired from raw milk [11].

In western Europe, particularly in Belgium, an increase in human infections has also been observed, associated with an increase in isolations in cattle [12–14]. In May 1991, an outbreak of *S. dublin* infection associated with drinking unpasteurized milk was reported in England [15]. In the USA and Belgium, the widespread use of antibiotics as prophylactic, therapeutic, and growth-promoting agents in cattle is thought to have contributed to the emergence of tetracycline and chloramphenicol resistance [8]. Although the illegal use of such antibiotics in Ireland was reported to be a source of concern in recent years [16], the strain responsible for this outbreak did not demonstrate antibiotic resistance.

Cheese is not a frequent vehicle of infection in food poisoning or salmonella infections [17]. During 1951–89, in England and Wales, cheese was implicated as the vehicle of infection in 31 outbreaks of communicable disease reported to CDSC [18–22]. Twelve of these involved cheddar cheese and *Staphylococcus aureus* was the organism thought to be responsible in 18 of the 31 outbreaks. Three reports of outbreaks of salmonella infection associated with cheese were received. Cheese associated outbreaks of salmonella food poisoning are also rare in the USA [23–25]. Many of these rare outbreaks were thought to have been related to the fact that the implicated cheese was made from raw milk and that it had been sold and eaten too quickly after it had been made. Several states in the USA have laws requiring that cheese be made only from pasteurized milk and cream, or be ripened or matured for at least 60 days in lieu of pasteurization. This allows time for any organisms present in the cheese to die out.

In order to provide a microbiologically safe product from raw milk, reliance

must be placed on the effects of starter culture competition and of low pH to eliminate pathogens in cheese. Low pH may not be completely effective in destroying pathogens [26, 27]. Indeed, *S. typhimurium* has been found to grow rapidly during manufacture and during the first 2 weeks of maturing in some cheeses. The soft cheese implicated in the outbreak we investigated was matured for between 14 and 21 days.

This outbreak was the second outbreak of food poisoning of national importance in E & W in 1989, investigated by CDSC and found to be associated with eating cheese made from an unpasteurized milk [28]. Both cheeses were produced in small family run dairies. This second outbreak associated with imported Irish unpasteurized cheese highlights a continuing risk to public health. Pasteurization of all milk for human consumption and for the production of cheese has been strongly recommended in England and Wales [29, 30], but there is currently no legislation to ensure this. Thus, pasteurization could not be pre-requisite for resumption of importation of the cheese to the UK. Pasteurization is the most effective method to prevent contamination of cheese made from unpasteurized milk. Ultimately, the dairy owners decided to install a pasteurizer at the farm and samples of pasteurized cheeses tested for the presence of salmonellas at the FHL gave negative results. This pasteurized product has been approved for importation to the UK.

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