A waterborne outbreak of *Salmonella* Saintpaul

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Abstract

Contamination of a tank water supply system led to an outbreak of *Salmonella* Saintpaul with 28 cases of gastroenteritis amongst over 200 workers at a large construction site. The outbreak was identified following notification of two salmonellosis cases by general practitioners from different towns during March 1999. The source of infection, contaminated drinking water, was identified through environmental sampling and confirmed by epidemiological investigations. Frogs and/or mice may have been the original source of the contamination. This report details control measures, the results of investigations and recommendations for future research. *Commun Dis Intell* 2000;24:336-340.

Keywords: salmonellosis, waterborne, surveillance, frogs, mice

Introduction

Outbreaks due to *Salmonellae* are usually foodborne, though waterborne outbreaks have been reported. In the USA between 1993 and 1997,[1] 357 (54.5%) of the 655 foodborne outbreaks known to be due to bacteria were due to *Salmonellae*. Conversely, in the United States from 1993 to 1998, only one of fifteen (6.6%) of the reported gastroenteritis outbreaks involving drinking water due to bacteria was caused by *Salmonella*;[2,3,4] the likely source of that outbreak, due to *S*. Typhimurium, was one of a pair of storage towers inadequately protected against bird droppings.[2]

S. Saintpaul has been associated with foodborne outbreaks including one due to contaminated paprika.[5] *S.* Saintpaul usually accounts for about 12 per cent of all typed human salmonella isolates from the central Queensland area. According to National Salmonella Surveillance Scheme (NSSS) reports, it is consistently amongst the top 10 serovars in Australia, with Queensland accounting for around two thirds of the nation's total reported cases.

Lizards (including geckos) and other reptiles have been well documented as sources of salmonella,[6-8] and there are reports that amphibians such as frogs and toads are potential salmonella sources.[9-13] NSSS data on *S*. Saintpaul isolates from non-human sources in Queensland January 1990 to April 1999 show a wide range of animal sources, including reptilian, bovine, ovine, porcine, equine, canine, avian and marsupial species. (Personal communication, D Lightfoot, National Salmonella Surveillance Scheme, Microbiological Diagnostic Unit, University of Melbourne).

In this paper we report two laboratory-proven cases of salmonellosis due to *S*. Saintpaul in workers from an isolated construction site in Central Queensland. It was concluded that the source of the outbreak, which involved 28 cases in all, was probably a contaminated water supply in which live green tree frogs were found.

Methods and Results

Clinical investigation

Two cases of salmonellosis were notified by General Practitioners from towns 250 km apart, on 15 March and 24 March 1999. Both worked at the same site and both were faeces culture-positive for salmonella, later typed as *S*. Saintpaul. Telephone follow-up of the first suggested no associated cases, but follow-up of the second did, prompting an immediate site visit and further investigation.

Enhanced surveillance was carried out by asking general practitioners and hospital staff in the nearby town to sample and notify any further cases of gastroenteritis amongst construction site workers. However no further cases were notified by this means.

Environmental investigation

Although numbers fluctuated with the various stages of construction, at the time of this outbreak 212 workers were employed at a construction site in central Queensland. Many were accommodated in temporary living quarters about 2 kilometres away, where they were catered for by a camp kitchen.

The initial site visit (conducted on 25 March 1999) included environmental health assessments, interviews with identified cases and examination of absenteeism records supplied by three main subcontractors (A, B and C). These records highlighted an unusual number of employees off work during March with gastrointestinal illness. Preliminary assessment identified several potential sources of exposure. These included septic tanks that had overflowed in recent weeks, sharing of drinks (particularly 5 litre water bottles, from which workers drank directly), manual handling of ice for water bottles, tank-sourced drinking water, a mouse infestation, and sub-optimal lunch box storage facilities.

On 29 March, samples for bacteriological analysis were collected from two ice machines, three water fountains

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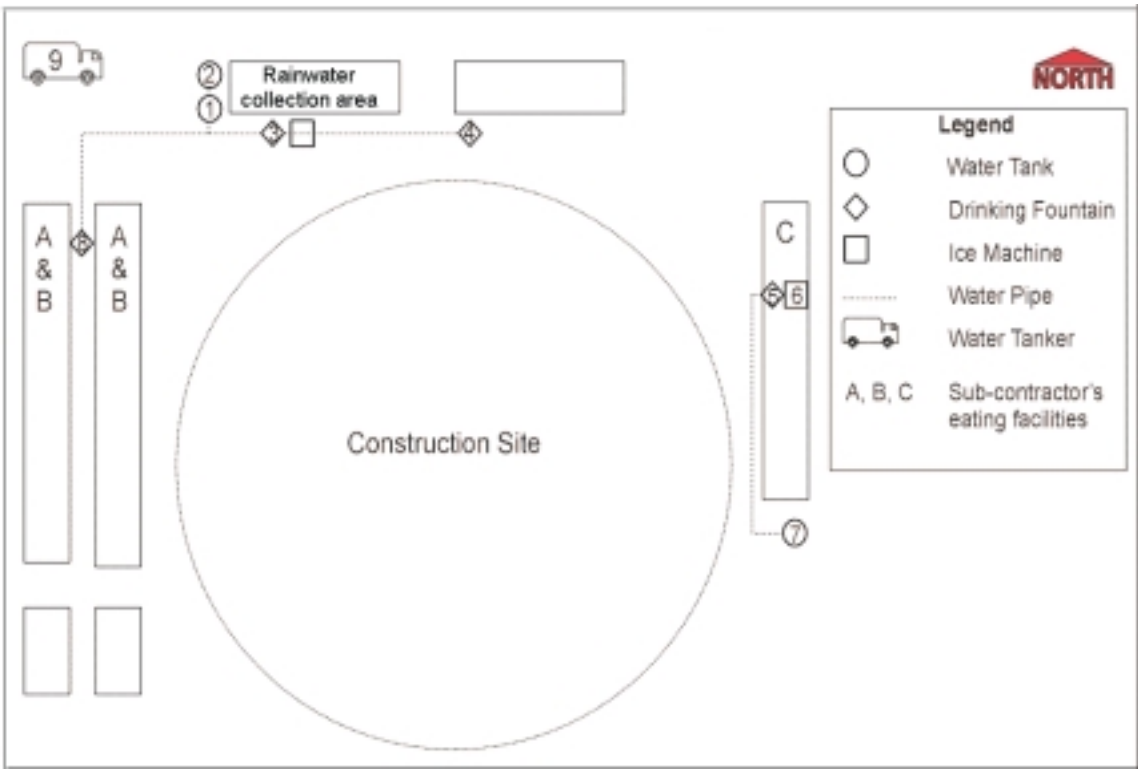
Figure 1. Site diagram showing water sampling locations, 8 April 1999



serving most on-site drinking water needs and food sold from the mobile food van servicing the site daily. More targeted water sampling was conducted, including specifically for *Salmonella* culture, on 8 April, 27 April and 26 May.

An assessment of water carted to the site was also conducted. The carter servicing the site obtained water from a reticulated water supply for which there were no indications of recent problems. Supplementary chlorine was added to it prior to delivery. Samples taken from the carter's tankers complied with NHMRC *Australian Drinking Water Guidelines*[14]. On delivery water was stored without further chlorination in six new 5,000-litre tanks located at three points around the construction site (Figure 1). Two of the six tanks (1 and 2) were interconnected and supplemented with rainwater collected from a workshop roof. One of these tanks had an uncovered inlet.

All ice and water samples taken from the site on 29 March (results available 8 April) failed to comply with NHMRC guidelines in terms of coliform counts. Some samples contained *E. coli* counts of up to 47 organisms/100 mL.

Repeat water samples taken on 8 April (results available 19 April) also showed contamination (Table 1). Other than site 9 (plate count <25 cfu) all sites had Standard Plate Counts greater than 500 cfu. *Salmonella* was cultured from samples taken from tank 2 and from two water fountains sourced from this tank. Subsequent typing identified the serovar as *S*. Saintpaul, the same as the human isolates. Water samples taken on 27 April demonstrated improved water quality whilst those taken 26 May showed no coliforms.

Food safety assessments were conducted on site and at the accommodation camp. These found the accommodation camp kitchen and mobile food van practices to be satisfactory. All food samples were of satisfactory bacterial quality.

Epidemiological investigations

A case-control study was initiated, with most controls being interviewed on 29 March using a modified Queensland Health Foodborne Illness questionnaire. Information was also sought about consumption and source of ice in water bottles, and usual sources of food.

A case was defined as any construction site employee who had suffered diarrhoea, or vomiting and abdominal cramps, or vomiting and fever, during the month of March. Controls were taken from unaffected construction site employees who worked at the site during this period. Data were analysed using Epi Info version 6.04c.

Table 1. Results of water sampling, 8 April 1999

Sample location (refer Figure 1)	Coliform count[1]	*E. coli* count[1]	*Salmonella* culture
1	> 80	23	- ve
2	+ ve[2]	> 48	+ ve
3	> 80	7	Not tested
4	> 80	16	+ ve
5	7	- ve	Not tested
6	5	1	Not tested
7	5	- ve	- ve
8	> 80	19	+ ve
9	- ve	- ve	- ve

1. Colony forming units (cfu) per plate.
2. Detected, but no count due to confluent growth.

Twenty-eight cases and 88 controls were interviewed. Most (if not all) cases were ascertained, so the attack rate was approximately 13 per cent. Illness occurred more frequently amongst employees of two of the three main subcontractor groups (firms 'A' and 'B' had a combined attack rate of 21 per cent, compared with an attack rate of 9 per cent in firm 'C'). Although some employees of all firms stayed at the accommodation camp, there was little on-site contact between employees of firms A and B with those of firm C, either during work or rest breaks. Firms A and B shared lunch areas, ablution facilities and sources of ice that were separate from those of firm 'C' (Figure 1). Gastroenteritis had occurred in both accommodation camp residents and workers living off-site in the nearby town. There were no reported cases in the families of employees and no evidence of secondary spread.

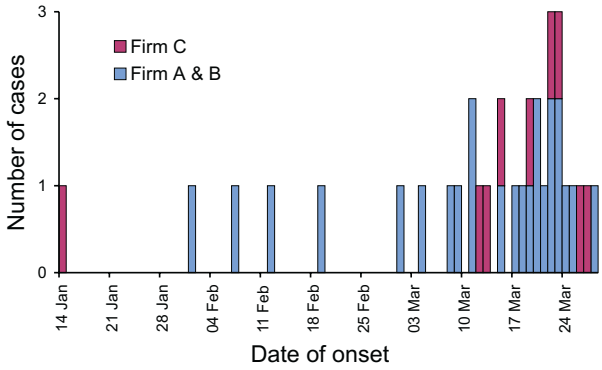
The association between illness and the use of an ice machine was striking, pointing to the ice (or water used to make it) as a source of infection. The association with employer firm was biologically plausible as *Salmonella* contaminated both the ice machine and water tanks used by firms A and B (Figure 1). For each firm separately it appeared the distribution of cases to controls was unlikely to be random, with $p = 0.03$. (Table 2). The same analysis, this time allocating the firms according to which tanks they used, shows that the risk of illness to employees in firms A and B (sharing the same water tanks) was threefold that of employees of firm C, who used the uncontaminated tanks ($p = 0.02$).

Whilst eating at the pievan initially appeared to be a significant risk factor (Table 2), the questions asked of where the employees 'usually' ate were not rigorously defined, nor mutually exclusive, and the interpretation of results based on their responses is uncertain. Stratified analysis of the variables (illness, eating at the pievan, employee firm, sharing of water bottles and using an ice machine) was performed. A much higher proportion of employees in firms A and B (75%) usually consumed food from the pievan than did firm C employees (30%).

Stratified analysis of firm, illness and use of pievan showed crude and summary odd ratios to be different (4.02 versus 2.15) indicating confounding. There was a reduced risk of illness amongst firm C employees whether they ate at the pievan (odds ratio 0.15) or not (odds ratio 0.26).

The epidemic curve (Figure 2) shows a series of sporadic cases in the 6 weeks from the start of February and then a clustering of cases peaking around 24 March.

Figure 2. Gastrointestinal illness amongst construction site workers, 1 February to 31 March 1999, by date of onset and construction firm



Control measures

Following initial environmental health assessments on 25 March, site-managers were advised to ensure all staff used their own water bottles and that ice machine scoops be appropriately provided, stored and cleansed. It was requested that hand washbasins, soap and disposable paper towelling be placed close to meal facilities.

On 31 March 1999, after preliminary analysis of the epidemiological data, the construction site safety coordinator was requested to empty and sanitise the ice machines. On the same day a scheduled shutdown of the entire plant for 8 days over Easter enabled a convenient break in further exposure for workers whilst further results were awaited. On receipt of unsatisfactory water sample results during this period, the tanks and distribution system were re-sampled before being emptied, disinfected, flushed and refilled, measures not initially recommended because

Table 2. Exposures and characteristics of cases and controls, and results of statistical analysis

Exposure category	Cases (n=28)	Controls (n=88)	Odds ratio (OR)	Cornfield's 95% confidence intervals	Probability (P)
Sex	3F:25M*	4F:84M	2.52	0.41< OR <14.83	0.22
Firm (A, B or C)	12A:8B:8C	20A:19B:49C			0.03
Firms sharing water tanks (A&B, C)	20A&B:8C	39A&B:49C	3.10	1.2 < OR < 8.8	0.02
Used an ice machine	25Y:1N*	61Y:20N	8.20	1.05 < OR <175.5	0.01
Shared water bottle	13Y:10N	23Y:58N	3.28	1.13 < OR <9.60	0.01
Usually ate at pievan†	13Y:5N	22Y:34N	4.02	1.10 < OR <15.38	0.01
Usually ate at canteen†	13Y:2N	37Y:18N	3.12	0.42 < OR <4.93	0.35
Usually ate at home†	13Y:2N	37Y:18N	3.16	0.57 < OR <23.06	0.12

* F = female, M = male, Y = yes, N = no

† These categories are not mutually exclusive

obtaining drinking water for the site was difficult. However, during this remediation and whilst awaiting further water sampling results to assess its efficacy, a temporary alternative supply of bottled water was arranged.

Further recommendations included discontinuation of rainwater collection, regular chlorination of stored water, and disinfection of water bottles when not in use. The upsurge in cases ceased abruptly following institution of control measures. Follow-up water samples complied with NHMRC guidelines, and no further cases of gastroenteritis were reported after the end of March.

Exposures in relation to illness were analysed by contingency tables (Table 2).

Discussion

The epidemic curve for this outbreak (Figure 2) is neither consistent with person-to-person spread of a gastrointestinal infection with a relatively short incubation period, nor the 'classic' picture of a point-source outbreak, such as a meal at a function. It could be consistent with a point source of infection with an initial relatively low level of risk, which then increased due to either significantly increased levels of contamination or exposure.

Only two cases had faeces sampled owing to a time lag between onset and recognition of the outbreak, combined with the fact that few cases presented to a medical practitioner and faeces sampling was either not offered or was declined. These cases had no connection with each other apart from their workplace, yet both cultured *S*. Saintpaul. This fact, taken in context of the water sample results, makes it highly likely that the other cases were caused by the same organism. Their onset times also span the period during which the outbreak was at its peak in March, consistent with the other cases being caused by an ongoing (waterborne) source rather than from a single exposure at one point in time.

Since it is unclear exactly when this outbreak began, it is possible our case definition either excluded some earlier cases in February or included unrelated cases early in March (Figure 2). It is also possible that case ascertainment was incomplete to a minor degree if some staff developed their illness after leaving the area upon completion of contracts.

The observed association between illness and sharing water bottles is likely to be explained by confounding rather than person-to-person spread. Firms A and B did not supply individual water bottles. Therefore these employees were more likely to share water bottles; 58 per cent of employees from A and B did this, compared with 16 per cent from firm C. In the case of firms A and B, the shared water was sourced from contaminated tanks.

Owing to constraints in access to on-site staff, we were unable to carry out a cohort study or ask more detailed questions about exposures in a lengthy questionnaire. However, results of microbiological sampling supported by epidemiological findings provide strong evidence that this outbreak was caused by contamination of parts of the worksite reticulation system with *Salmonella* Saintpaul. The tanks with proven salmonella contamination (which collected rainwater as well as being topped up by tanker deliveries) supplied water to the taps, drinking water fountains, ice machine and kitchen/eating areas used by the most-affected work groups.

The most significant rain during February/March was a fall of 125 mm on 6 March, and it is possible this marked the actual beginning of the outbreak. Although the original source of contamination of the water tanks is uncertain, there are at least two plausible explanations.

Salmonellae may have been introduced via mice and/or their excreta washed into the tanks from the roof collection area. Mice were evident during site inspections despite rodent control measures. On cleaning, no dead mice were found inside the salmonella-contaminated interconnected tanks, but a number of live green tree frogs were and these may have introduced contamination directly. Alternatively, salmonella from animal excreta flushed into the tank by the heavy rainfall event multiplied in the frogs and were effectively 'amplified'.

O'Shea et al reported various salmonellae in 19 of 150 cane toads (*Bufo marinus*), but not *S*. Saintpaul.[15] NSSS data for 1985 - 1999 support this finding. The NSSS database records only one isolation of salmonella from a green tree frog (a *Salmonella* Onderstepoort). However, based upon overseas experience, a greater number of isolates and range of salmonellae are likely to be found if frogs were sampled more extensively.

Local Parks and Wildlife staff confirm that inquiries from the public about removal of frogs from water tanks are common. Maintenance of barriers across water tank inspection ports is the obvious (but frequently neglected) intervention to prevent ingress of frogs. Further research may be warranted to investigate the potential role of frogs as vectors in human salmonella infection.

Acknowledgments

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