Outbreak of Salmonella Partypi B Var Java Due to Contaminated Alfalfa Sprouts in Canada

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During August and September 1999, laboratory-confirmed infections of *Salmonellaparatyphi* B var *java* were reported from several provinces. The investigations identified 51 persons with *S.paratyphi* B var *java* infection across Canada; 43 in Alberta, six in British Columbia and two in Saskatchewan. This report describes the investigation conducted in Alberta.

Background

On 16 September, 1999, the Provincial Laboratory for Public Health (Microbiology), PLPH(M), Edmonton Site (ES) notified Alberta Health and Wellness (AHW) regarding one confirmed *S. paratyphi* B var *java* infection

and nine *Salmonella* species infections, identified in stool between 22 August, 1999 and 11 September, 1999. The PLPH(M)-ES had reported one infection earlier in the year and no infections in 1997 or 1998.

By 17 September, 1999, the PLPH(M)-ES and the PLPH(M), Calgary Site (CS) reported nine confirmed infections of *S. paratyphi* B var *java* infection and 14 *Salmonella* species infections to AHW. Follow-up of these infections by the Calgary Regional Health Authority identified alfalfa sprouts as a potential source. AHW reported the outbreak to Health Canada.

Methods

Epidemiologic-Descriptive: Between mid-August and late September 1999, public health officials with the regional health authorities in Alberta attempted to contact all persons infected with *S.paratyphi* B var *java* by telephone to collect demographic and illness information, animal exposure and food history for the week prior to illness.

A confirmed case was defined as a person with laboratory confirmation of *S. paratyphi* B var *java* from stool with the same pulsed-field gel electrophoresis (PFGE) pattern. Confirmed cases were sub-categorized into primary and secondary. A primary case had an illness onset date in August or September, 1999. A secondary case had contact with an ill household member. Additionally, a suspect case was defined as a person with laboratory confirmation of *S. paratyphi* B var *java* from stool (PFGE results pending or unknown as of 30 September, 1999).

Epidemiologic-Analytic: A case-control study was conducted to test the hypothesis that alfalfa sprouts were associated with infection. Health Canada staff administered a standard questionnaire to controls by telephone on 25 and 26 September, 1999 after the health officials had completed follow-up interviews with 27 primary cases. The questionnaire was designed to collect information about alfalfa sprout consumption within the past week, either in the home or from a food establishment. Controls were matched to cases by age, sex and telephone exchange. Potential controls were identified by dialling the matched telephone exchange followed by a random four digit combination. Age groups for matching were defined as <= 3 years of age, 4 to 17 years of age and >= 18 years of age. Potential controls were excluded if they had experienced an episode of diarrhea in the past 14 days, travelled outside of Canada in the past 14 days or used any antibiotics in the past 14 days. An attempt was made to find two controls per case. Data were entered into Epi Info 6.04b (Centers for Disease Control and Prevention, Atlanta, Georgia, U.S.A., 1995) and analysed using the matched analysis function.

Environmental Investigation: At the time of the interviews, health officials with the regional health authorities retrieved alfalfa sprout packages from cases' homes, if available, for submission for bacterial testing. Health officials also collected various retail samples of alfalfa sprouts and submitted these to the PLPH(M). In addition, sprout samples were taken from one restaurant where a case had identified eating sprouts.

On 21 September, 1999, the Canadian Food Inspection Agency (CFIA) and the local regional health authority conducted an investigation of the suspect alfalfa sprout processing plant to determine the source and lot identification of the alfalfa seeds. Seed samples, in-line samples (product in the process of being grown), environmental swabs of food and non-food contact surfaces and final product samples were collected during the investigation.

Laboratory: The PLPH(M) and the National Laboratory for Enteric Pathogens (NLEP) conducted serotyping, phage typing (PT) and PFGE analysis for human *Salmonella*isolates as well as for isolates from samples of alfalfa sprouts from cases' homes. Samples from the processing plant were sent to the Alberta Agricultural Laboratory for initial testing. The PLPH(M) and the NLEP also sub-typed isolates from retail and processing plant food samples.

Results

Epidemiologic-Descriptive: Between 22 August and 29 September, 1999, 43 laboratory-confirmed infections of *S. paratyphi* B var *java* with PT "Worksop" and one of two very similar PFGE patterns (A1 and A2), were reported to AHW. Alberta health officials successfully contacted and interviewed 42 infected persons. Forty persons were primary and two were secondary. Thirty-eight of the primary cases had PT "Worksop" and the PFGE pattern A1; two had PT "Worksop" and the PFGE pattern A2.

The mean age of primary cases was 29 years of age (median 24.5 years, range 2 to 83 years), 78% being >= 20 years of age. Sixty-six percent were female. Although primary cases were distributed across all areas of Alberta, 46% were residents of Calgary. Onset of illness was between 24 July, 1999 and 19 September. Case counts peaked during the week of 6 September (Figure 1). Of the 40 primary cases who were contacted, 24 (60%) reported consuming alfalfa sprouts in the week prior to their illness, eight (20%) reported not consuming and eight (20%) reported their exposure status as "unsure" or "maybe".

Epidemiologic-Analytic: A total of 27 primary cases and 53 matched controls were enrolled in the case-control study. Three cases and their six matched controls were excluded as cases were unsure of their exposure status. Cases were more likely than their matched controls to have eaten sprouts within the past week (Mantel-Haenszel matched odds ratio 10.83, 95% confidence interval 3.03 – 56.42).

Environmental Investigation: Six opened packages of alfalfa sprouts were taken from cases' homes; however, these packages may not have been the alfalfa sprouts to which the case was exposed prior to illness.Two packages were positive for *S.paratyphi* B var *java*, PT "Worksop", PFGE pattern A1. One package was positive for *S. litchfield*, one for *S. thompson* and one for *S. newport*; one package was negative.

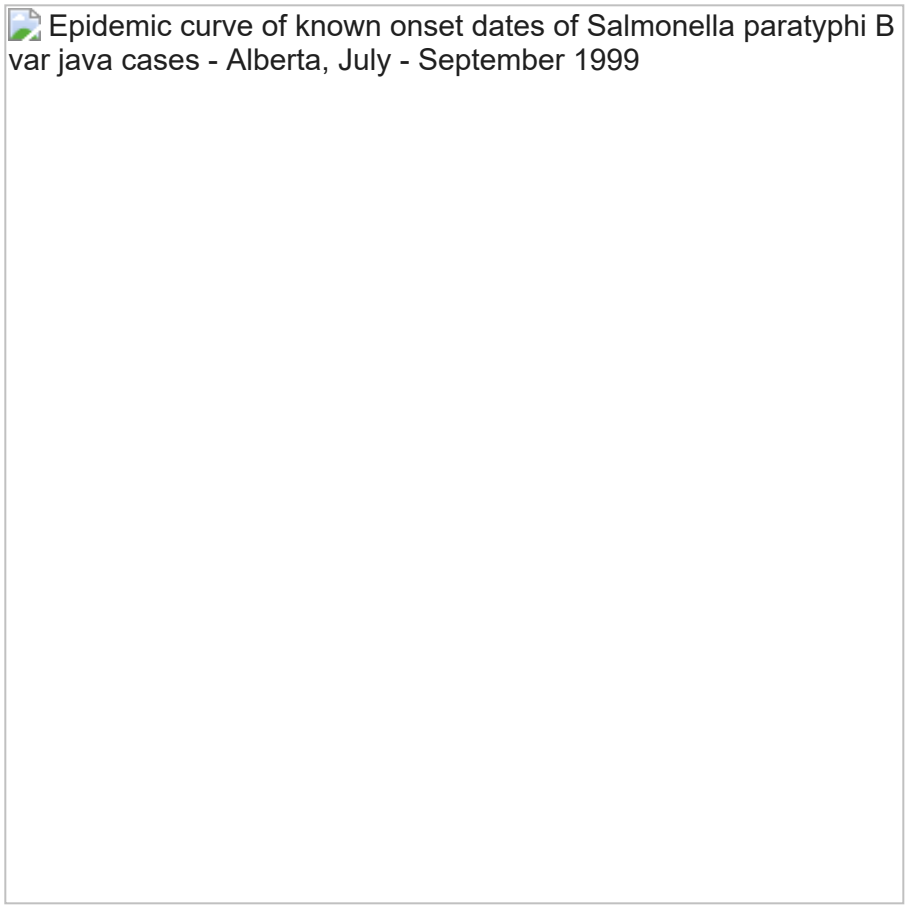
Of 21 retail samples submitted, five were positive for *Salmonella*, including *S. newport*, *S. litchfield* and *S. give*.

An investigation of the alfalfa sprout plant by the CFIA and the regional health authorities revealed that alfalfa sprout seeds were grown on several farms and sent to a seed cleaning plant in Kentucky, USA. The seeds were then mixed into one seed lot and approximately 300 50-pound bags were distributed to a manufacturer located in Ontario, Canada. The Ontario manufacturer forwarded all seeds to the Alberta manufacturer. The Alberta manufacturer then sent seeds to other processing plants in Saskatchewan and British Columbia.None of this seed lot was used at the Ontario plant, nor was it distributed anywhere else within Canada or the United States. The Alberta plant distributed the final product across all regions of Alberta with main points of distribution in Calgary and Edmonton. The final product was also distributed from the Alberta plant to Northwestern Ontario.

On 29 September, 1999, the CFIA issued a Class II recall with a public warning. (A Class II recall is applied to a violative product that may cause temporary adverse health consequences or where the probability of serious adverse health consequences is remote). CFIA detained 86 bags of the remaining seeds from the implicated seed lot. Although CFIA had recommended that the Alberta manufacturer chemically treat the seeds prior to sprouting, the Alberta plant did not have any operational treatment process in place prior to the outbreak. CFIA ordered the manufacturer to discontinue use of the suspected seed lot, discontinue sprout production, implement cleaning and pre-treat their seeds. The manufacturer complied.

Laboratory-processing of samples from the manufacturer; which included seed samples, in-line samples, environmental swabs of food and non-food contact surfaces, and final product samples were all negative for *Salmonella* species, with the exception of one final product sample which was positive for *Salmonella meunchen.*

Figure 1
Epidemic curve of known onset dates of *Salmonella paratyphi* B var *java* cases – Alberta, July – September 1999


Epidemic curve of known onset dates of Salmonella paratyphi B var java cases - Alberta, July - September 1999

Discussion

During the course of this outbreak, 51 persons with *S. paratyphi* B var *java* infection associated with alfalfa sprouts were identified across Canada. Alfalfa sprouts were identified as the suspect cause during investigations and this was further supported by the case-control study. Although not included in the study results, follow up by the Alberta regional health authorities identified nine additional persons during the outbreak period with either ***S. newport*****,** *S. thompson* or *S. litchfield*, some of who also identified exposure to alfalfa sprouts.

Five of six sprout package samples taken from cases' homes were positive for *Salmonella* and of these, two were positive for *S. paratyphi* B var *java*, PT "Worksop" and PFGE pattern A1. The other *Salmonella* serotypes found may reflect that these were not the alfalfa sprouts that were directly related to the cases' exposure; however, these results do provide further evidence that final product from this plant had potential to cause salmonellosis.

The findings from this investigation reinforce the fact that alfalfa sprouts remain a potentially hazardous product for consumption. This outbreak of salmonellosis associated with consumption of alfalfa sprouts was the second to occur in Alberta over a period of 2 years and the third to occur in Canada in the past 5 years. In 1997 an outbreak of infections with *S. meleagridis*[(1)] in Alberta, was responsible for 43 confirmed cases associated with consumption of contaminated alfalfa sprouts from the same alfalfa sprout manufacturer as in this current outbreak. In 1995-1996, an outbreak of infections with ***S. newport*** in British Columbia[(2)] and Quebec, as well as Oregon[(2)], USA was also linked to the consumption of alfalfa sprouts and traced back to one contaminated seed lot.

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Editorial Comment

Although it is unclear how and where the alfalfa seeds became contaminated, the opportunities for contamination from the field, through shipping and storage, to sprouting are numerous and not easily preventable. Furthermore, the sprouting process provides an effective growth medium so that an insignificant number of organisms may increase to become a potential health hazard[1,2]. As alfalfa seed is traded globally as a commodity and may be contaminated regardless of the purported country of origin, all seed should be treated prior to sprouting.

In 1996, Health Canada's Bureau of Microbial Hazards (BMH), Health Products and Food Branch commented on the use of chlorine for treating seeds prior to germination and growth, stating that soaking seeds in a level of 200-300 ppm chlorine for a shorter time of 10 minutes may be as effective as 100-200 ppm chlorine for 30 minutes. Unfortunately disinfection procedures may not reliably eliminate *Salmonella* from alfalfa seeds[1-3]. The Canadian Food Inspection Agency (CFIA), in consultation with Health Canada developed a draft "Code of Practice for the Hygienic Production of Sprouted Seeds and Beans"[4] that will aid sprout manufacturers in minimizing the risk associated with the consumption of sprouted seeds and beans. This code deals with best practices for the agricultural production of seeds and the hygienic production of sprouts. Among several recommendations, the code suggests rinsing and disinfecting seeds in specific ways to maximize efficacy of the treatment and minimize the risk of contamination, as well as ensuring that the seeds are thoroughly rinsed with potable water after the disinfection treatment.

Health Canada and CFIA are also developing a policy that will further aid sprout manufacturers in minimizing the risk associated with the consumption of sprouted seeds. Further research is still required to provide evidence on how best to reduce the risk of illness from the consumption of raw sprouts.

Outbreaks caused by pathogens such as *Salmonella, Escherichia coli* O157, Hepatitis A, *Cyclospora cayetanensis*, *Bacillus cereus* and *Shigella flexneri*, and associated with produce (such as cantaloupes,

raspberries, frozen strawberries, tomatoes, lettuce, bean sprouts and alfalfa sprouts), are a relatively recent phenomena[5]. Three epidemiologic features common to these outbreaks are a widespread geographic distribution of cases and an absence of clustering by household or food-service establishment[6]. These features are barriers to promptly identifying and controlling of a foodborne disease outbreak. It is most commonly, the laboratory-identification of a rare serotype which triggers an investigation. Foodborne outbreaks of a more common *Salmonella* serotype could easily be missed amid the flurry of supposedly sporadic cases[7].

Salmonellosis has been recognized as a “re-emerging” infection[8]. These alfalfa sprout-associated outbreaks provide an example of how global trading of food supplies and consumer demand for perceived “health foods” can be contributing factors in the re-emergence of salmonellosis.

Effective provincial, national and international surveillance networks, both formal and informal, with epidemiologic and laboratory expertise, as well as collaboration from the food industry and regulatory agencies are required for timely public health responses to global foodborne outbreaks. Currently, systematic serotyping of *Salmonella* at the provincial level, reporting of these serotypes at the national level and a national protocol for outbreak investigations ensure efficient communication occurs between epidemiologists, microbiologists, food regulators and food inspectors to rapidly identify foodborne outbreaks.

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