

## Rapid communications

# OUTBREAK OF *SALMONELLA* ENTERICA SEROVAR TYPHIMURIUM IN SWITZERLAND, MAY – JUNE 2008, IMPLICATIONS FOR PRODUCTION AND CONTROL OF MEAT PREPARATIONS

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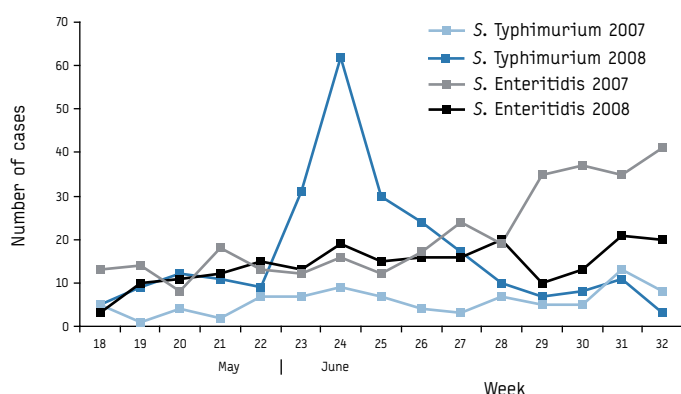
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An increased number of *Salmonella* Typhimurium cases were reported in Switzerland between May and June 2008. Investigations involved 72 cases. Results of PFGE typing identified several outbreak strains, the dominating one present in 43 of the 72 isolates. Strains affecting one third of the cases were also found in animal samples, in particular pork. However, no specific food source could be identified. Outbreaks described in this paper highlight the importance of food safety regulations such as those on minced meat and meat preparations issued by the European Commission and adopted by Switzerland into the national law.

### Introduction

A sharp and countrywide increase of the number of reported *Salmonella* Typhimurium isolates was observed in May 2008 starting in week 19 and peaking in week 24 (Figure 1). Between early May to late June (weeks 19 – 27), 205 cases (2.70 cases / 100,000 inhabitants) were recorded compared to 44 (0.58 / 100,000 inhabitants) in the same period of the preceding year. In week 28, the number of cases returned to the level of 2007.

**FIGURE 1**  
Number of reported *Salmonella* Typhimurium and *Salmonella* Enteritidis cases by week of reception of the stool sample in the laboratory, Switzerland, weeks 18 – 32, 2007 and 2008



### Methods

A total of 72 patient isolates with dates of isolation extending from week 17 to 27 were subjected to molecular analysis using Pulsed Field Gel Electrophoresis (PFGE) [1] by the National Centre of Enteropathogenic Bacteria (NENT) and the Institute for Food Safety, University of Zurich. Minimal inhibitory concentrations for antimicrobial susceptibility testing of representative strains were determined on Mueller-Hinton agar (Becton Dickinson, Sparks, USA) using E test strips (AB Biodisk, Solna, Sweden).

When a private food quality assurance laboratory reported the isolation of *S. Typhimurium* in pork samples, the cantonal authorities of official food control were asked to intensify the sampling and testing activity of meat products and to submit all *Salmonella* isolates from food analyses to the NENT. Subsequently, four official laboratories of food control (Zurich, Vaud, Fribourg, Liechtenstein) analysed 38 samples of raw meat and meat preparations from pork and 15 samples of raw meat and meat preparations from poultry for the presence of *Salmonella*. Furthermore, 55 samples of ready-to-eat raw meat sausages were tested.

Moreover, 24 patients were interviewed by phone between June 25 and July 7, 2008, using a standardised questionnaire. They were asked about food consumed three days before the onset of illness and travel history during the week before the onset of illness.

### Results

#### Epidemiological data

The cases were located in 22 of the 26 Swiss cantons (203 cases) and in the Principality of Liechtenstein (two cases) (Table 1). The distribution of the cases by age (Table 2) in weeks 19 – 27 showed a shift towards the teenage group (23.4% of cases aged 10–19 years) when compared with the period 2000–2007 (13.5%). At the same time, children below the age of five years were much less represented during the outbreak (12.7%) than in the preceding eight-year period (28.0%). The sex ratio male / female seemed to be more even during the outbreak (50.2% / 46.8%) compared to the period 2000–2007 (54.0% [range: 49.1–56.9%] / 42.5% [range: 40.0–44.6%]).

TABLE 1

Number of cases of *Salmonella* Typhimurium and incidences per 100,000 inhabitants in the cantons of residence of the patients, Switzerland, weeks 19 – 27, 2008

Canton	Number of cases	Population	Incidence
Nidwalden	4	40,287	9.9
Grisons	11	188,762	5.8
Uri	2	34,989	5.7
Appenzell Ausser Rhoden	3	52,654	5.7
Lucerne	19	363,475	5.2
Basel-Stadt	8	185,227	4.3
Bern	39	962,982	4.0
Schaffhausen	3	74,527	4.0
Zug	3	109,141	2.7
Basel-Land	7	269,145	2.6
Zurich	31	1,307,567	2.4
Solothurn	6	250,240	2.4
Neuchatel	4	169,782	2.4
Fribourg	6	263,241	2.3
Aargau	13	581,562	2.2
Geneva	9	438,177	2.1
St. Gallen	10	465,937	2.1
Thurgau	4	238,316	1.7
Valais	5	298,580	1.7
Vaud	11	672,039	1.6
Jura	1	69,555	1.4
Ticino	4	328,580	1.2
Total	203	7,593,494	2.7

Note: The Principality of Liechtenstein regularly reports to the Federal Office of Public Health on a voluntary basis. Regarding the outbreak presented here, Liechtenstein reported 2 additional cases, reflecting an incidence of 5.7 cases / 100,000 inhabitants.

TABLE 2

Age distribution of cases of *Salmonella* Typhimurium in the outbreak in weeks 19 – 27 of 2008, and of all cases of *S. Typhimurium* reported in 2000 – 2007

Age group (years)	Percentage of cases in the outbreak weeks 19–27, 2008	Percentage of all cases reported in 2000–2007
0–4	12.7	28.0
5–9	9.8	14.6
10–19	23.4	13.5
20–29	14.6	9.2
30–39	6.3	8.5
40–49	7.8	6.8
50–59	6.3	7.1
60–69	5.4	5.3
70+	13.7	5.9

### Laboratory investigations

The PFGE typing identified several outbreak strains (Figure 2).

The dominating type, designated “strain 2”, was found in 43 of the 72 isolates. It appeared for the first time in week 23 and was obviously responsible for the main phase of the outbreak (Figure 3). However, no matching strains from food isolates have been found. None of the 108 samples of raw meat and meat preparations and ready-to-eat raw meat products analysed by four official laboratories of food control revealed *Salmonella* isolates. Other control laboratories reported no *Salmonella* isolations from foods prior and during the outbreak period within their routine testing programs.

“Strain 1” (11 isolates) was present at the beginning of the outbreak and remained up to week 24. “Strain 3” (six isolates) appeared only in weeks 25 and 26. Both strains matched with isolates from pork samples taken from a meat producer/distributor.

Two further pork-related strains were found in some patients. A strain identified in a spare rib sample from Germany (strain pm - processed meat), was found in three patients with an indistinguishable pattern. A strain identified in a sample taken from a pig at a slaughterhouse (strain sl) was isolated from two patients. Strain sl showed a PFGE profile very similar to that of the outbreak strain 3. In fact, one large band appeared to have been split in two smaller ones by a single genetic difference (Figure 2). Strains 3 and sl might therefore be considered two variants of a single clone.

Finally, seven patient isolates yielded PFGE patterns that were different from each other and from all other strains (although one in week 20 resembled strain 1), and can therefore be regarded as sporadic cases. In total, the pork-related strains 1, 3, sl and pm represented 34% (22/65) of the human cases which were not considered sporadic.

The most prevalent PFGE profiles, yielded by strains 1 to 3, were compared to international databases of Enter-Net, Salm-gene/Pulse-Net [2]. All three types matched profiles in the databases (Table 3). For example, strain 1, indistinguishable from JPXX01.0038, was found in seven patients and three non-human specimens (beef and turtle) in 2008 in the United States [personal communication by P. Gerner-Smidt, Centers for Disease Control and Prevention, US]. In Europe, a very similar profile, but with an extra band at 150 kb, was represented by 34 Pulse-Net entries. Strain 2, the dominant Swiss outbreak clone, was found among European data only once. This single entry in the Salm-gene database was submitted as a human isolate of page type DT 193 by German authorities in 2002. Strain 3 was represented three times in the Pulse-net database [personal communication by J. Threlfall and M. Hampton, Health Protection Agency, United Kingdom]. Interestingly, none of the Swiss outbreak strains corresponded to *S. Typhimurium* U292 which is responsible for a large current outbreak in Denmark.

The outbreak strains 1 to 3, as well as strains sl and pm were fully susceptible to the used panel of antimicrobials (ampicillin, ceftazidime, chloramphenicol, nalidixic acid, streptomycin, tetracycline, and trimethoprim/sulfamethoxazole). In contrast, one randomly chosen isolate from a sporadic case (18/022351) was resistant to ampicillin, chloramphenicol and tetracycline (data not shown).

## Interview results

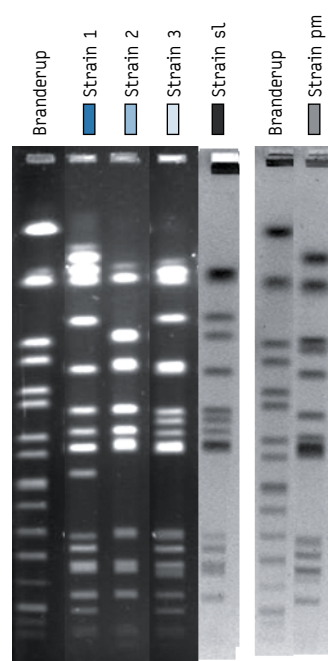
Eight of the 24 interviewed patients were found to be infected with pork-related strains 1, 3 or sl. Six of these patients confirmed having eaten pork, one denied it and one was uncertain. The latter two, however, reported that they had eaten chicken and had taken part in a barbecue event where different sorts of meat were grilled, whereby the possibility of cross contamination should be taken into consideration. In further 15 patients among those interviewed the main outbreak strain 2 was found. Eleven of these reported having eaten pork, nine had consumed beef, six had eaten chicken and seven other kinds of meat (lamb, horse), and four participated in a barbecue. Only one patient reported having travelled (to Germany) in the seven days before onset of illness and having fallen ill while travelling, but this patient was among the sporadic cases.

Interviews were not suggestive of any food item other than those mentioned as a possible common source of infection. The variety of mentioned food items and the variety of identified strains favour the possibility that several outbreaks occurred simultaneously.

## Discussion

The steep rise in cases of *S. Typhimurium* infections in May 2008 was detected by the mandatory reporting system of the Federal Office of Public Health (FOPH) in the context of infectious diseases surveillance in Switzerland. Within a period of nine weeks, the number of registered cases exceeded almost fivefold those of the preceding year. The investigations in collaboration with the National Centre for Enteropathogenic Bacteria (NENT) and the Institute for Food Safety of the University of Zurich confirmed the ongoing of a countrywide outbreak or – more likely – several simultaneous outbreaks caused by different strains of *S. Typhimurium*. On the other hand, microevolution seems to have already gone on, since strains 3 and sl were differentiated by only one or two bands

**FIGURE 2**  
PFGE profiles of the relevant *Salmonella* strains, Switzerland, 2008



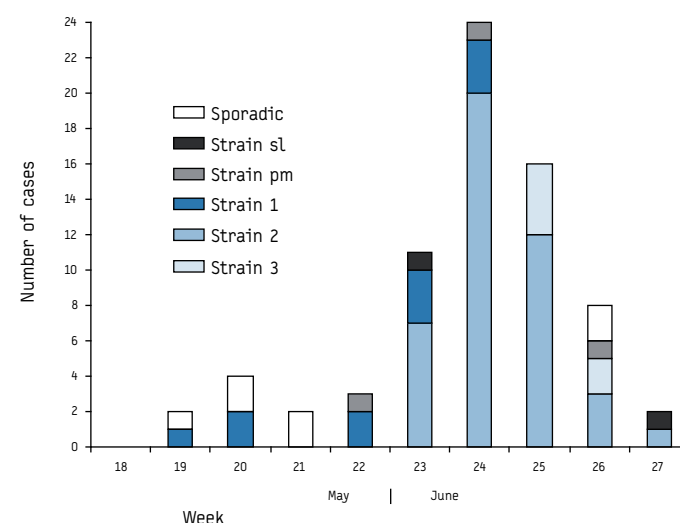
Strain 1 (patient 18/027428); Strain 2 (patient 18/027416); Strain 3 (patient 18/027772); Strain sl (slaughter house); Strain pm (processed meat); Braenderup (*S. Braenderup* (H9812) DNA, restricted with *Xba*I, and used as a size marker [1])

(Figure 3). Therefore, these two strains could be considered two variants of a single clone.

The findings gathered through the patient interviews showed that there was a median delay of six days between onset of disease and date of reception of the stool sample at the laboratory. In addition, a median delay of 10 days was brought about by the elapsed time between reception of the stool sample at the primary diagnostic laboratory and reception of the notification at the FOPH. In total, two to three weeks could have elapsed between the onset of disease and the registration of the infection. This shows that reducing the statutory notification period (currently one week) to 24 hours would improve the timeliness of patient interviews and of potential public health interventions.

About 34% of the human cases were infected with strains which were also demonstrated in quality control samples of pork from a particular company, on a pig carcass from a slaughterhouse and in an imported (from Germany) spare rib sample. Therefore, the evidence by PFGE analysis of human and food isolates, partly

**FIGURE 3**  
Number of *Salmonella* Typhimurium isolates belonging to different PFGE types, Switzerland, weeks 18 – 27, 2008 (n=72)



Strains 1, 2, 3 are different outbreak strains; Strain sl (slaughter house) was identified in a pig; Strain pm (processed meat) was found in a meat sample

**TABLE 3**  
Relatedness of outbreak strains 1 to 3 identified in Switzerland and other *Salmonella* Typhimurium strains deployed in international PFGE databases [2]

Swiss strain	USA <sup>a</sup>	SalMGene / PuLseNet Europe <sup>b</sup>	Denmark <sup>c</sup>
Strain 1	JPXX01.0038	STYMXB.0103	JPXX01.0178.DK
Strain 2	no match	STYMXB.0134	JPXX01.0020.DK
Strain 3	no match	STYMXB.0214	JPXX01.0022.DK

a) Courtesy: P. Genner-Smidt; b) Courtesy: J. Threlfall, M. Hampton; c) Courtesy: S. Ethelberg and R.F. Petersen

supported by patient interviews, allowed the conclusion that about one third of the observed outbreak cases was caused by contaminated pork.

However, in 108 market samples of raw pork and poultry meat, meat preparations and sausages, no *Salmonella* could be isolated. These findings indicated that contamination levels of market products with *Salmonella* must have been low or that the contaminated products were no longer present in the market.

Strain 2 was dominant in the weeks with the majority of cases (43 of 72 cases analysed by PFGE, that may be extrapolated to some 120 of the total 205 cases), but could not be linked to a specific food item. This same profile matched a contemporary cluster of 13 human isolates obtained in Denmark, but was clearly different from strains identified in the large ongoing Danish *S. Typhimurium* U292 outbreak [personal communication by S. Ethelberg and R. F. Petersen, Statens Serum Institute, Denmark]. It also matched at least 18 human isolates in France [personal communication by J. de Valk, Institut de veille sanitaire, France]. In France as well as in Switzerland, this strain was found to be fully susceptible to all tested antimicrobials [3].

The pork-related strains 1 and 3 also found their matches in Denmark where strain 3 represented “a rather common profile”. Infection through contaminated pork products is also the main hypothesis for the U292 and other *S. Typhimurium* outbreaks that occurred this year in Denmark [4].

#### Conclusions in the context of food safety legislation

In outbreaks where a large spectrum of foods, such as meat and meat preparations are potential sources of infection, it is more or less accidental to trace a targeted pathogen successfully with a reasonable number of samples. In the present case, market samples were analysed at the end of the outbreak which possibly was too late. The company which found *S. Typhimurium* in several samples of pork in the context of quality control actions launched a large environmental screening for *Salmonella* in their facilities. These investigations clearly revealed that the strain isolated from pork samples was not persistent in the factory but was introduced by pork imported from other European countries. The contaminated meat was processed into products used for barbecue such as pork sausages. The hypothesis that such products contributed to the outbreak is supported by the fact that younger people were overrepresented among the infected persons. In this age group barbecue parties during the summer months are very popular and frequently practiced. Considering this particular risk, FOPH published a fact sheet on hygienic rules to be applied in barbecue events on its website [5].

To prevent outbreaks such as described in this paper, measures have to be taken at the meat production level as well. The faecal carriage of foodborne pathogens among livestock animals at slaughter is strongly correlated with the hazard of carcass contamination. In order to reduce the risk represented by *Salmonella*, the maintenance of slaughter hygiene is consequently of central importance in meat production. *Salmonella* sampling on carcasses is regulated in view of slaughter hygiene monitoring in the European Commission Regulation (EC) No 2073/2005 [6]. In the same regulation, microbiological criteria are decreed for *Salmonella* in minced meat and meat preparations from poultry meat intended to be eaten cooked and minced meat and meat preparations from other species than poultry intended to be eaten cooked (absence in 10 g; n=5; c=0) [6]. This regulation was adopted by Switzerland into the national law [7]. For companies, there remains in fact only

one option to deal with the new requirements, namely the use of *Salmonella*-free raw materials for certain final products. There are two ways to reach that target. Either only meat that comes from *Salmonella*-free herds is processed or raw meat is analysed with rapid test for the presence of *Salmonella* prior to further processing. If imported meat is used, the producer has to make it clear to the importing company that only *Salmonella*-free meat is accepted. In this way, a certain pressure will build up on farmers and it is there that the problem has to be addressed. For decades, raw meat has been considered unsafe for consumption since it could contain pathogenic bacteria. With the new EU-regulation which demands the absence of *Salmonella* in minced meat or in meat preparations a change of paradigm occurred. There is no doubt that the practical implementation of this regulation will be a costly and long lasting challenge for all involved stakeholders, in particular the livestock keepers who must make efforts to reduce *Salmonella* prevalence.

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