

Research Paper

Salmonellosis in the State of Rio Grande do Sul, southern Brazil, 2002 to 2004

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Submitted: December 1, 2011; Approved: September 10, 2012.

Abstract

Salmonella has been identified as the main aetiological agent responsible for foodborne diseases in several countries worldwide, including Brazil. In the State of Rio Grande do Sul (RS), southern Brazil, previous studies analysed official foodborne illnesses data, identifying *Salmonella* as the main bacterial agent of foodborne diseases during the period of 1997 to 2001. The present study aimed to analyse the official epidemiological data on salmonellosis occurred in the State of RS, during the period of 2002 to 2004. Even though data on recent salmonellosis were available, only data concerning the period comprising in 2002 to 2004 were analysed because the official worksheet records presented more consistent information about the salmonellosis outbreaks. Results indicated that, among the 624 foodborne outbreaks officially investigated, 202 (32.37%) were confirmed as salmonellosis. Among them 23,725 people were involved, 4,148 became sick, 1,878 were hospitalized and one person died. The season with the highest incidence of salmonellosis was spring, and the most affected age group was composed of people aged between 20 to 49 years old (56.66%). Animal origin foods - especially eggs and meat products - were very often involved with the outbreaks, however homemade mayonnaise was identified as the main food vehicle for salmonellosis (53.51%). The majority of the cases occurred inside private homes (55.81%) and food services (12.1%), and the main factors contributing to the occurrence of the outbreaks were the consumption of products without sanitary inspection (26.7%) and exposure of food at room temperature for more than two hours (18.58%). Similarly to what was previously reported for the period of 1997 to 2001, *Salmonella* spp. was the most prevalent foodborne disease agent in the State of RS during the years of 2002 to 2004.

Key words: Salmonellosis, Brazil, The State of Rio Grande do Sul.

Introduction

Salmonella has been reported as an important cause of concern in the contemporary world. Global surveillance data has indicated that the number of salmonellosis has increased expressively during the last years, mainly associated with the consumption of raw or undercooked eggs, poultry, meat or dairy products (Brasil, 2010; Caffer and Eigner, 2004; Laconha *et al.*, 2000; Mead *et al.*, 1999; Notermans and Hoogenboom-Verdegaal, 1992; Peresi *et al.*, 1998; Roadford and Board, 1993; Scallan *et al.*, 2011; Lindqvist *et al.*, 2000; Tondo and Ritter, 2012).

During the last decade, salmonellosis has been reported as the most frequent foodborne disease in Brazil, and animal products have been identified as the most frequent food vehicles responsible for such syndromes (Brasil, 2010). Since Brazil is currently the biggest exporter of poultry and red meat in the world, the control of *Salmonella* is of great interest in this country.

In RS, the southernmost State of Brazil, there are many well developed industries for producing poultry, pork and red meat, in which great efforts are made to control *Salmonella*. However, *Salmonella* has been identified as the main cause of foodborne diseases investigated by regula-

tory bodies since 1997 (Brasil, 2010; Mürmann *et al.*, 2008; Oliveira *et al.*, 2007; Rio Grande do Sul, 2001), occurring mainly in private homes and food services.

Aiming to understand the characteristics and causes of salmonellosis outbreaks in the State of RS, improved surveillance services and reporting of outbreaks are constantly necessary, thus making it possible to set priorities and implement preventive measures in order to reduce the occurrence of these illnesses (Costalunga and Tondo, 2002).

The foodborne outbreak investigations began in the State of RS in 1980, making it possible to identify locals where illnesses occurred, food vehicles and other factors related with foodborne diseases (Rio Grande do Sul, 2001). The foodborne epidemiological investigation in RS is a result of the collaborative work of the Division of Epidemiological Surveillance, Division of Sanitary Surveillance (DVS/RS) and Central Public Health Laboratory of the State of RS (FEPPS/LACEN/RS), all bodies coordinated by the Secretary of Health of the State of RS. The structure of this regulatory body in RS is composed of nineteen Regional Health Coordination sectors which work in cooperation with the municipalities, investigating foodborne diseases occurred in a population of more than 10.7 million people (Carmo *et al.*, 2005, Rio Grande do Sul, 2001). Foodborne outbreak investigations start after the regulation bodies are informed, and this generally occurs after larger outbreaks. Whenever possible, sanitary officers collect the suspected foods and send it to be analysed in the Central Public Health Laboratory (FEPPS/LACEN/RS) or in other accredited laboratory close to the site of the outbreak occurrence (Brasil, 2001). However, in many salmonellosis outbreaks it is not possible to collect food samples because the suspected food was either totally consumed or purposely removed (Costalunga and Tondo, 2002, Rio Grande do Sul, 2001). For this reason, many salmonellosis outbreaks are investigated and concluded based on the epidemiological and clinical data registered on the official worksheets (Brasil 2001, Carmo *et al.*, 2005), thus demonstrating the importance of outbreak records. However, this information is not always accessible and it can be better publicized if official data are analysed and published on scientific manuscripts.

The present scientific report is a result of the collaborative work of the Food Science and Technology Institute of Federal University of Rio Grande do Sul (ICTA/UFRGS) and the Secretary of Health of RS, whose aim was to disseminate relevant information about the investigation of salmonellosis carried out in the State of RS. The objective of this work is analysing the official registered data of salmonellosis outbreaks occurred in the State of RS, during the period of 2002 to 2004.

Materials and Methods

Epidemiological data on salmonellosis outbreaks occurred in the State of RS, Brazil, during the period of 2002

to 2004, were analysed by using the database registered on worksheets of epidemiological investigation conducted by Secretary of Health of RS. The final reports were done by the Division of Epidemiological Surveillance, while the investigations of the outbreaks were carried out by the Sanitary Surveillance services of the State of RS and municipalities. Only epidemiological data on salmonellosis occurred between 2002 to 2004 were chosen and analysed for two reasons: 1) to give sequence of previous epidemiological studies about salmonellosis of RS concerning the period of 1997 to 1999 (Costalunga and Tondo, 2002) and 2000 to 2001 (Silveira and Tondo, 2006); 2) data available on the official worksheet records of 2002 to 2004 presented consistent and detailed information about the salmonellosis outbreaks, make it possible this publication.

The data analysis focused on evaluating the following information: number of involved people, age and sex of real patients, number of deaths, most probable causes for the outbreaks, place of consumption (intake), food vehicles, and period of the year when the salmonellosis occurred.

When possible, inspectors of the Sanitary Surveillance Services of the municipalities collected samples of suspect food, feces of sick people and swab samples of food handlers. These samples were microbiologically analysed using methods described by the Food and Drug Administration in The Central Laboratory of Rio Grande do Sul (FEPPS/LACEN/RS), or other accredited laboratories in different regions of RS. Available microbiological results were also investigated in the present study.

Results

In the period between 2002 and 2004, 624 investigated outbreaks were recorded in Secretary of Health of the State of RS. Among them, 202 outbreaks (32.37%) were caused by *Salmonella* spp. In these outbreaks, 23,725 people were involved, 4,148 became ill, 1,878 were hospitalized and one person died (Table 1).

Results showed in Figure 1 demonstrates that springtime (October, November and December) had the highest incidence of salmonellosis in the State of RS (n = 14), while summertime (January, February and March) proved to have the second greatest rate of salmonellosis occurrence (n = 9).

The aging group most affected was adults ageing 20 to 49 years old (56.66%), followed by children and teenag-

Table 1 - General data about outbreaks of *Salmonella* spp. in the State of Rio Grande do Sul, Brazil, 2002 to 2004.

	2002	2003	2004	Total
Confirmed outbreaks	89	57	56	202
Involved people	6504	13810	3438	23752
Ill people	1649	1510	989	4148
Hospitalized people	812	713	313	1838
Deaths	1	0	0	1

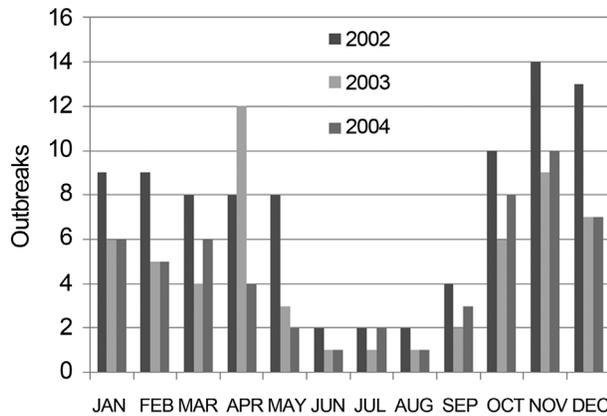


Figure 1 - Monthly occurrence of outbreaks of *Salmonella* spp. in the State of Rio Grande do Sul, Brazil, during 2002 to 2004.

ers ageing 10 to 19 years old (18.15%), as indicated in Table 2. There was no significant difference between the occurrence of salmonellosis between males and females, even though the female group showed a slightly higher incidence (51.93%).

Analysing the results of Table 3, it is possible to verify that 256 food vehicles were identified in the 202 salmonellosis outbreaks. These numbers indicate that more than one food vehicle were involved in several outbreaks.

Potato salad with homemade mayonnaise was the most involved preparation in the outbreaks (53.51%), followed by meat and meat products (roasted beef, roasted chicken, ground beef, raw and cooked beef and minced chicken - 21.48%), and pastry products (pies, cakes, lasagna, capeletti and pastels - 16.41%, Table 3).

Table 4 indicates that the majority of the outbreaks occurred in private homes (55.81%) followed by food services (12.1%). If the results of Table 1 and Table 4 were compared, it is possible to note that there is a small differ-

Table 2 - Age and sex of people involved in outbreaks of *Salmonella* spp. in the State of Rio Grande do Sul, Brazil, during the period of 2002 to 2004.

Age (years)	2002	2003	2004	Total	%
0-1	4	5	1	10	0.24
1-4	56	54	24	134	3.23
5-9	95	109	56	260	6.27
10-19	314	267	172	753	18.15
20-49	951	825	574	2350	56.66
> 50	222	229	148	599	14.44
Unknown	7	22	13	42	1.01
Sex					
Male	762	764	468	1994	48.07
Female	887	736	531	2154	51.93

ence between the total numbers of the outbreaks reported (Table 1: 202 and Table 4: 206). This difference can be explained by the occurrence of the same outbreak involving different places after the intake of a common food preparation.

The major factor contributing to the salmonellosis outbreaks was the use of raw materials without sanitary inspection (*i.e.* eggs, 26.07%) (Table 5). Other factors responsible for the outbreaks were storage of food at ambient (room) temperature for more than 2 hours (18.58%), improper food manipulation (12.5%) and improper refrigeration (9.65%).

Discussion

In Brazil, there were 6,349 notifications of foodborne outbreaks during 1999 to 2009, causing 123,917 ill people and 70 deaths. Among those outbreaks, 42.5% were caused by *Salmonella* spp. (Brasil, 2010). Contrasting these num-

Table 3 - Food vehicles of outbreaks of *Salmonella* spp. in the State of Rio Grande do Sul, Brazil, during the period of 2002 to 2004.

Food vehicle	Number of outbreaks			Total	
	2002	2003	2004	n	%
Salad ¹ with homemade mayonnaise	56	41	40	137	53.51
Pastry products ²	21	12	9	42	16.41
Meat and meat products ³	26	14	15	55	21.48
Milk and dairy products ⁴	1	0	3	4	1.57
River water	1	2	1	4	1.57
Eggs	2	0	1	3	1.17
Corn	0	0	2	2	0.78
Not identified	6	0	3	9	3.51
Total	113	69	74	256	100

¹salad of potato, paste, vegetables and others.

²pies, cakes, lasagna, capeletti and pastels.

³roasted beef, roasted chicken, ground beef, raw and cooked beef and minced chicken.

⁴raw milk, cheese and pudding.

Table 4 - Place eaten of *Salmonella* spp. in the State of Rio Grande do Sul, Brazil, during the period of 2002 to 2004.

Place	Number of outbreaks			Total	
	2002	2003	2004	n	%
Private homes	58	29	33	120	55.81
Commercial food establishments	10	7	8	25	12.1
Clubs and associations	7	5	4	16	8.84
Community rooms	4	8	6	18	10.7
Schools	2	3	1	6	2.79
Refectories	5	4	3	12	5.58
Industrial kitchens	3	3	2	8	3.72
Others	1	0	0	1	0.46
Total	90	59	57	206	100

bers, in the United States, 47.8 million illnesses and three thousand deaths are estimated to occur each year due to foodborne illnesses (Scallan *et al.*, 2011). Of this statistic, 1.4 million cases are estimated to be caused by the primary etiologic agent *Salmonella* (Voetsch *et al.*, 2004). Even though it is very difficult and may be impossible to compare realities found in countries like Brazil and the United States, it is possible to note a strong difference between the numbers of registered outbreaks in both countries. This difference can be attributed to many factors like differences in the production systems, differences in field operations, consumption patterns, among others, however probable one important factor is the precariousness of surveillance system in many regions of Brazil. Brazil is composed by approximately 195 million people distributed in 27 States. The great majority (72%) of the Brazilian foodborne notifi-

cations have been done by only three States of the Southern Brazil, *i. e.* RS (30%), São Paulo (22%) and Paraná (12%) (Brasil, 2010). In several States of Brazil, unfortunately, there is a lack of human and financial resources and there is lack of training for the personnel involved in foodborne investigations.

Between 2002 and 2004 in the State of RS, *Salmonella* was responsible for more than 32% of the outbreaks notified and investigated by health officers, causing 4,148 ill people, 1,878 hospitalizations and one death. Similar results were showed by Costalunga and Tondo (2002) who had demonstrated that salmonellosis accounted for 36% of foodborne disease outbreaks investigated in the State of RS between 1997 and 1999 (Costalunga and Tondo, 2002) and by Silveira and Tondo (2006) who had shown that salmonellosis was the first foodborne disease in the State of RS in the period of 2000 to 2001 (Silveira and Tondo, 2006). These results are in agreement with the Brazilian Ministry of Health that has published approximately 35% of the foodborne outbreaks occurred in Brazil, during 1999 to 2004, ending with laboratory testing, were caused by *Salmonella* spp. (Carmo *et al.*, 2005).

Other countries also have reported data on salmonellosis. For example, in the period of 1995 to 2000, 35% of confirmed outbreaks in Australia were caused by *Salmonella*, of which eight deaths were recorded (Todd, 1997). According to Hughes *et al.* (2007), in England and Wales in the period of 1992 to 2003, it was found that *Salmonella* spp. was responsible for 52% the investigated cases of foodborne outbreaks. The data from Austria, in 2005, showed that *Salmonella* was the cause of 76% of the 606 outbreaks (Much *et al.*, 2007) and, in the same year, in France, *Salmonella* was the leading cause of hospitalization

Table 5 - Factors that contributed to the *Salmonella* spp. in the State of Rio Grande do Sul, Brazil, during 2002 to 2004.

Factors contributing to the outbreak	Number of outbreaks			Total	
	2002	2003	2004	n	%
Use of raw material without sanitary inspection	25	24	24	73	26.07
Holding food at ambient (room) temperature for more than 2 hours	22	18	12	52	18.58
Improper hygiene of food handlers	9	14	12	35	12.5
Inadequate refrigeration	7	11	9	27	9.65
Poor hygiene of equipment and utensils	5	8	6	19	6.78
Cross-contamination	3	2	4	9	3.21
Contamination by infected food handler	3	2	2	7	2.5
Improper warm holding	2	1		3	1.07
Improper cooking	1	1	1	3	1.07
Ingestion of contaminated water	1	2	1	4	1.44
Improper place of food processing	1			1	0.35
Use of wood utensils			1	1	0.35
Unknown	23	4	19	46	16.43
Total	102	87	91	280	100

and death related to foodborne diseases (Vaillant *et al.*, 2005). In China, *Salmonella* was responsible for 19% of the outbreaks caused by bacterial contamination at schools during the period of 1994 to 2005 (Wang *et al.*, 2007). These data confirm the importance of this microorganism as aetiological agent in relation to public health issues worldwide.

During the present research, the period with highest salmonellosis occurrence in the State of RS was comprised in springtime, *i. e.* during the months of October, November and December, tending to decrease in subsequent months of summertime. This higher incidence in springtime could be explained by the wide temperature variation during the day-long period in the State of RS, where very often the morning periods present cold temperatures, whilst afternoons are frequently warm or hot. According to Surveillance Service officers of RS, *Salmonella* outbreaks occur due to the negligence with food temperature control during springtime, when weather temperatures do not seem to be as high, especially in mornings, and people are not concerned about salmonellosis and do not store food in refrigeration. In summertime, the number of salmonellosis outbreaks generally decreases because people store food refrigerated. The subsequent declining number of salmonellosis cases during autumn and winter was a probable consequence of the temperature decline in southern Brazil, where the average temperature is usually around 10° Celsius. In opposite to the results reported for the State of RS, summer was pointed as the season when the highest amount of salmonellosis outbreaks occurred in Argentina (Caffer and Eiguer, 2004) and in the United States (CDC, 2006).

According to Magnus *et al.* (1999) *Salmonella* has no age preference, but generally demonstrates a linear increase in the incidence with increasing age. In the present study the age group most involved in salmonellosis were people above 19 years old (71.1%), followed by children and adolescents between 10 and 19 years old (27.65%). These results can be explained by the fact that people above 19 years old are those representing the economical active population of the State of RS, which mostly made its food consumptions out of home. However, it is necessary to point out that, according to our results, most of the salmonellosis of the State of RS occurred inside private homes, followed by food services. Similar findings were reported by the Brazilian Ministry of Health during the analysis of outbreaks occurred in 1999 and 2004, demonstrating that the private homes (49%) followed by commercial food establishments (19%) were also the sites of the highest rate of *Salmonella* outbreaks (Carmo *et al.*, 2005).

According to the results of the present study and those reported in previous ones (Costalunga and Tondo, 2002; Silveira and Tondo, 2006), the high incidence of salmonellosis in the State of RS seems to be related mainly with the consumption of egg and egg products without proper heating. Other studies have reported similar results (FDA,

2010; Humphrey, 1994). Our work demonstrated that the main food vehicle responsible for salmonellosis was the potato salad prepared with homemade mayonnaise, and similar results were reported previously (Costalunga and Tondo, 2002; Silveira and Tondo, 2006). One probable explanation is the fact that, as a cultural habit of RS population, homemade mayonnaise is generally prepared with raw and cooked eggs well mixed with soya oil and warm cooked potato. Very often the eggs used are those produced in small farms or at home without sanitary inspection, increasing the probability of *Salmonella* contamination. During homemade mayonnaise preparation, *Salmonella* can easily multiply in eggs due to warm temperature provided by cooked potato and subsequent maintenance in inadequate temperatures (De Paula *et al.*, 2005). However, if eggs or egg products are stored in proper refrigeration temperature (Brasil, 2009; Jay, 2005; Rio Grande do Sul, 2006) and thermal processed-egg yolk are used to prepare homemade mayonnaise the risk of salmonellosis decreases significantly.

In the State of RS, several salmonellosis outbreaks were also caused by pastry products, as pies, cakes, lasagna, capeletti and pastels, and by meat and meat products, as roasted beef, roasted chicken, ground beef, raw and cooked beef and minced chicken. Outbreaks caused by pies and cakes also could be attributed to contaminated eggs used in its preparations, whilst salmonellosis caused by lasagna, capeletti and pastels had *Salmonella* in their meat fillings. Salmonellosis caused by meat and meat products have been reported in RS by previous studies (Costalunga and Tondo, 2002; Silveira and Tondo, 2006), and most of them can be attributed to meat contamination and cross-contamination during meat products preparation. The fourth food vehicle group most involved with salmonellosis in RS was milk and dairy products, *i.e.* raw milk and cheeses. The involvement of these kinds of foods with salmonellosis can be explained because some people in RS have the habit of drinking raw milk and others use raw milk to produce a typical cheese called "Colonial cheese". The most common form of production of Colonial cheese involve a maturation period of less than one week and this procedure is not sufficient for the complete inactivation of *Salmonella* by lactic acid bacteria.

In this study, the two mainly factors that contributed to the outbreaks were the usage of raw material without sanitary inspection (for example, eggs) and holding foods in ambient (room) temperature for more than 2 hours. In fact, probably the most important factor to be controlled is the process temperatures because even contaminated eggs will not cause salmonellosis if they were properly cooked and conserved under refrigerated temperatures. Previous studies have indicated homemade mayonnaise as an important cause of salmonellosis outbreaks in the State of RS. For example, during the period of 1997 to 1999, homemade mayonnaise was responsible for 42.45% (Costalunga and

Tondo, 2002) of the salmonellosis cases registered in RS, while in 2000 - 2001 this preparation was responsible for 48.1% (Silveira and Tondo, 2006) of the cases. Results of the present study demonstrated that homemade mayonnaise was responsible for 53.51% of the salmonellosis cases, during 2002 to 2004, indicating that the consumption of homemade mayonnaise was an important cause of salmonellosis in the State of RS.

During the period of 1999 to 2009, the consumption of eggs was identified as the main cause for most of the salmonellosis outbreaks (22.8%) investigated in Brazil (Brasil, 2010). In United States, egg was the food most frequently involved in outbreaks of *Salmonella* spp. reported by the Centers for Disease Control and Prevention (CDC) (2009). Similar results have been reported by several other researchers, as Greig and Ravel (Greig and Ravel, 2009), Hughes *et al.* (2007) and Much *et al.* (2007).

Microorganisms such as *Salmonella*, mainly *S. Enteritidis*, can infect the ovaries and oviducts of chickens, thus contaminating eggs, especially during shell formation. In addition, egg shells can be contaminated with intestinal bacteria during the passage through the chicken cloaca (Forsythe, 2010). Foods prepared with eggs were involved in 72% of *Salmonella* outbreaks investigated in the State of RS in 2000 (Nadvorny *et al.*, 2004). It is important to note that the multiplication of *Salmonella* can be prevented by adequate cooling of foods. According to Malheiros *et al.* (2007), in a study carried out in order to investigate the growth kinetics of different serovars of *Salmonella* in homemade mayonnaise there was not detected growth of any *Salmonella* serovars in homemade mayonnaise stored at 9.5 °C in the first 24 hours. However, *S. Enteritidis* SE86 showed higher counts in the first 6 hours when cultivated at 30 °C.

Other studies investigating the strains of *Salmonella* involved with foodborne outbreaks in RS have reported interesting information about these food pathogens. For example, Geimba *et al.* (2004) have shown that more than 97% of the salmonellosis occurred in the State of RS, during 1999 to 2002, were caused by *S. Enteritidis*. Studies with those *S. Enteritidis* isolates have shown that the majority of the outbreaks were caused by one specific strain of *S. Enteritidis* called SE86. (Oliveira *et al.*, 2006). Giving sequence to these studies, Oliveira *et al.* (2007) used molecular methods in order to compare *S. Enteritidis* strains isolated from stool cultures of salmonellosis victims with *S. Enteritidis* strains isolated from food involved with the outbreaks occurred in RS from 1999 to 2006, and showed that SE86 was involved in most of them. Even though, different *Salmonella* serovars been isolated from diverse types of foods in RS, the majority of the foodborne outbreaks from 1999 to 2006 has been caused by *S. Enteritidis* SE86 (Oliveira *et al.*, 2007).

The results of the present study demonstrated the need for greater attention in food safety issues related to

salmonellosis. The fact that most of the *Salmonella* outbreaks have occurred inside private homes followed by food services indicates the need for public campaigns aiming awareness and more rigid regulatory control, mainly concerning temperature control, usage of foods after sanitary inspection and appropriate food handling.

Acknowledgments

We would like to thank to the staff of Water and Food Microbiology Laboratory of FEPPS/LACEN/RS and to all the Staff of Secretary of Health of Rio Grande do Sul, especially to the M. Sc. Josete Balaiardi Silveira and the Veterinarians Susana Costalunga Lima and Denise Maria da Silva Figueiredo, who greatly works to improve regional Surveillance and Epidemiological Services of the State of RS.

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