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Investigation of food and water microbiological conditions and foodborne disease outbreaks in the Federal District, Brazil

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A B S T R A C T

This is a retrospective study describing data on the microbiological conditions of food and water obtained from analysis reports issued by the Central Laboratory of the Federal District (LACEN-DF), and information on foodborne disease outbreaks investigated by the Office of Water and Food Borne Diseases of the Federal District (NATHA), Brazil, between 2000 and 2010. A total of 4576 analysis reports were evaluated, from which 92.9% of monitoring samples and 7.1% of samples suspected to be involved in outbreaks. Of the total number of samples, 630 did not comply with Brazilian legislation (rejected). Ready-to-eat food, milk/dairy products, water, spices/seasonings, and ice cream/sorbets had the highest rejection rates among the monitoring samples (18.9–11%), with the first two groups having the highest rates among the outbreak samples (23.5 and 21.7%). *Minas* cheese showed to be the food with the highest rejection rate among the samples analyzed by the LACEN-DF. About 9% of the food samples were rejected due to thermotolerant coliforms and/or coagulase-positive staphylococci, and 10.5% of the water samples were rejected due to *Pseudomona aeruginosa*. Ready-to-eat food were the main foods involved in the foodborne disease outbreaks investigated by NATHA (51.3% of the 117 outbreaks with the food identified) and *Bacillus cereus* the most identified etiologic agent (41.2% of the 80 outbreaks with the agent identified). This study indicated that microbiological surveillance programs should focus on ready-to-eat food to prevent the occurrence of foodborne disease outbreaks in the region.

1. Introduction

Food and water represent important vehicles for pathogens of substantial public health concern, including those that cause acute diarrheal illnesses, which account for 1.8 million childhood deaths annually, predominantly in developing countries (WHO, 2008). About 4.3 million cases of acute diarrhea were reported in 2010 in Brazil, with almost 4000 deaths (MS, 2012a).

Over 8500 foodborne disease outbreaks were reported by local/state health authorities to the Brazilian Foodborne Disease Outbreak Surveillance System between 2000 and 2010, involving nearly 180,000 individuals and 88 deaths (MS, 2012b). In 2011, the reporting of foodborne disease outbreaks became compulsory in the country, but only for outbreaks occurring on ships or aircraft (Regulation 104/2011, Brazilian Ministry of Health).

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In addition to laboratory data on sick individuals and disease reports, microbiological analyses of the suspected food items may contribute to the investigation of outbreaks of foodborne diseases (Oliveira, De Paula, & Capalonga, 2010; WHO, 2008). In Brazil, state laboratories are responsible for conducting analyses of food available for consumption in routine sanitary surveillance programs, and of food suspected of involvement in foodborne disease outbreaks. This information may be used by health authorities and food industry professionals to target prevention efforts against pathogens and foods that cause the outbreaks.

The objective of the present study was to identify the critical biological hazards and the associated foods consumed in the Federal District of Brazil, based on food microbiological analyses and epidemiological data of foodborne disease outbreaks that occurred in the region from 2000 to 2010. Covering a total area of 5,801,937 km^2, the Federal District is in the Midwestern region of Brazil, where the nation's capital, Brasilia, is located. In 2009, the estimated population of the Federal District was about 2.6 million inhabitants.

2. Material and methods

2.1. Microbiological analysis reports

This is a retrospective registry-based descriptive study covering data collected from microbiological analysis reports of samples of food ready for consumption (requiring no further thermal processes before consumption) and water analyzed by the Central Laboratory of the Federal District (LACEN-DF) between 2000 and 2010. The samples were classified by the LACEN-DF as monitoring samples (under its Sanitary Surveillance Program) or as outbreak samples (suspected of involvement in a foodborne disease outbreak). The surveillance program samples were randomly collected (non-statistical sampling) from various commercial establishments, including supermarkets, restaurants, catering services and bakeries. The microbiological analyses of all samples were conducted using standard methodologies (APHA, 2001; MAPA, 2003) and included the following cultural analyses: Mesophilic aerobic count (MAC), coliforms, coliforms at 30 °C, thermotolerant coliforms (TTC), coagulase-positive staphylococci (CPS), *Bacillus cereus, Staphylococcus aureus*, *Escherichia coli, Salmonella* spp, *Pseudomona aeruginosa*, and *Enterococcus* spp. In this study, the food products analyzed by the LACEN-DF were classified into food groups, as shown in Table 1.

The conclusions of the analyses were described in the reports as being either compliant or non-compliant with the microbiological parameters established for food samples by Directive 451/1997 (published by the Brazilian Ministry of Health, in effect until December 2000) and Resolution 12/2001 (published by the National Sanitary Surveillance Agency, ANVISA), and by Resolutions 54/2000 and 275/2005 (ANVISA) for bottled water samples. Samples that were non-compliant with legislation were regarded in this work as rejected samples. In addition to the parameters established by legislation, the results of the outbreak samples analyzed by the LACEN-DF were compared with the infectious dose (ID) of detected pathogenic microorganisms (10^5 CFU/g or mL for *S. aureus* and *B. cereus*) (FDA, 2012). Staphylococcal enterotoxin investigation was conducted in four outbreak samples using the RAPD technique (Random amplified polymorphic DNA), followed by Optimum Sensitivity Plate for strain identification (SEA, SED and TSSH-1).

2.2. Foodborne disease outbreak information

Information on foodborne disease outbreaks that took place in the Federal District between 2000 and 2010 were obtained from the Office of Water and Food Borne Diseases (NATHA) of the Epidemiology Department of the Federal District Health Secretariat. Information obtained for each investigated outbreak included the food involved, the etiological agent identified in the food sample and/or the biological sample, number of cases, and age and sex of the individuals affected. Some reports also included the criterion used to conclude the outbreak: laboratory-confirmed, food analysis, laboratory-confirmed/food analysis or clinical-epidemiological, which includes information on symptoms, dietary habits and existence of family members or other consumers with the same symptoms.

2.3. Data analysis

Statistical analysis was performed with STATA® V.12. Exact binomial tests were used to identify significant differences in rejection rates among food groups and food items. Odd ratios (OR) were estimated using Epitools epidemiological calculators (Sergeant, 2009).

3. Results

3.1. Microbiological analysis

The results of the 4576 analysis reports of food samples ready for consumption and water samples analyzed by LACEN-DF between 2000 and 2010 are described and discussed in this study. Over 90% of the samples analyzed were monitoring samples, and 325 were outbreak samples. The number of monitoring samples analyzed decreased along the period under study, with the lowest number in 2004 (Fig.1). The number of outbreak samples analyzed varied, reaching a maximum of 56 samples in 2010.

A total of 630 samples (13.8%) were rejected for not meeting the parameters established by Brazilian legislation, of which 564 (13.3% [CI 95%: 12.2–14.2]) were monitoring samples, and 66 (20.3% [CI 95%: 15.9–24.7]) were outbreak samples. Overall, the chance of an outbreak sample being rejected was significantly higher than that of a monitoring sample (OR = 1.6 [CI 95%: 1.3–2.2]). Fig. 2 shows

Table 1
Food groups and food items analyzed by the LACEN-DF.

Food group	Food
Milk and dairy products	Milk (UHT, pasteurized, fermented and powder); cheese (mozzarella, *Minas*, provolone, Roquefort, parmesan, cottage, brie, *coalho*); milk cream (fresh and canned); *doce de leite*; powder for milky beverages; fruit yogurts; butter.
Ready-to-eat food (from catering establishments, restaurants and bars)	Meals containing rice, beans, vegetables, chicken, cattle, lamb, pork, fish and/or seafood; salmon sashimi; baked and fried *salgados* (with fillings of chicken or cattle meat, ham, cheese and/or shrimp); paste (garlic, tuna, chicken, chickpea, cheddar, ham, salmon, garlic); pasta with various sauces; vegetable salad; potato salad with mayonnaise; potato pure; *pamonha*; hotdogs; sandwiches; pizza; cakes, puddings.
Water	Mineral water, purified water with salts, ice cubes, bottled water
Spices, seasonings, condiments	Salt based condiments; black pepper (powder/ground and whole peppercorns); dried seasonings (curry, oregano, saffron, curcuma, cinnamon, basil), dried garlic; mayonnaise; mustard; ketchup; refined and brown sugar.
Fruit and vegetables, raw or canned	Canned (sweet corn, olive, pea, carrots, eggplant, summer squash, various fruits); fresh fruits and vegetables; minimally processed vegetables; vegetable oil; margarine.
Non-alcoholic beverages	Soft drinks; frozen fruit pulp; concentrated fruit juice; fruit juice ready for consumption; nectars.
Flour, cereals, cakes, industrialized	Corn starch and flour; wheat flour; oat meal and flour; cereal bran; cereal bars; cereal flakes; cookies, crackers; cakes; cake powder.
Ice cream and sorbets	Ice cream and sorbets of various flavors and fruits
Animal products, processed, and eggs	Canned (sardine, tuna, minced meat in tomato sauce, *mocotó*); liver paste; sausages; ham (pork, turkey; chicken); mortadella; eggs (quail and chicken); honey.
Snacks and appetizers	Corn snacks of various flavors; potato chips.
Baby food	Infant formula (up to 1 year old)
Chocolates, candies, sweets	Chocolate, candies, confectionary, candies enrobed in chocolate; vegetable and fruit sweet (such as *bananada* and *goiabada*)

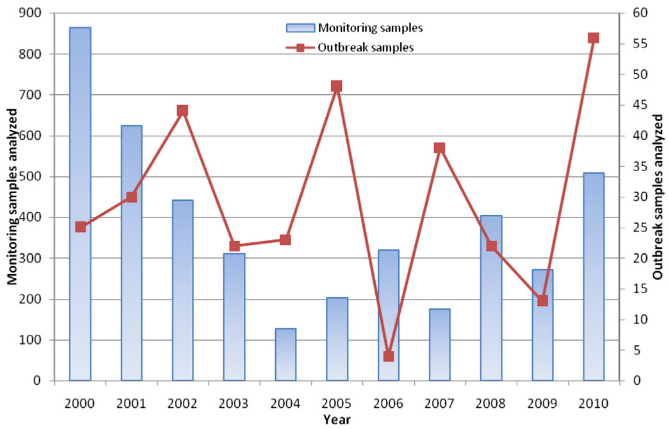

Fig. 1. Number of food ready for consumption and bottled water samples analyzed by the LACEN-DF between 2000 and 2010.

the rejection rates of monitoring and outbreak samples during the period. The highest rejection rate for monitoring samples was registered in 2000 (22.6%), which was also the year with the highest number of samples analyzed (Fig. 1). For the year of 2006, two of the four outbreak samples analyzed (Fig. 1) were rejected.

Monitoring samples from the milk and dairy products group represented 34.7% of all samples analyzed (Table 2). Over 77% of the 325 outbreak samples analyzed were from the ready-to-eat food group. Ready-to-eat food (18.9%), milk/dairy products (16.1%), water (17.4%), spices/seasonings (13.3%) and ice cream/sorbets (11.0%) had the highest percentage of rejection among the monitoring samples, with the first two groups having the highest rejection rate of all outbreak samples (23.5 and 21.7%, respectively) (Table 2).

Within the milk/dairy group, the rejection rate for cheese (439 samples analyzed, including fresh *Minas* cheese, standard *Minas* cheese, mozzarella, cream cheese and cottage cheese) was significantly higher (32.6% [CI 95%: 28.2–37.1]) than for the other foods in the group. Among the milk samples ($n = 990$), pasteurized milk had a significantly higher rejection rate (16.3% [CI 95%: 13.4–19.6]) than UHT (2.2% [CI 95%: 0.8–4.5]) and powdered milk (2.8% [CI 95%: 0.5–7.8]). These frequencies are lower than those found for standard *Minas* cheese (56.9% [CI 95%: 42.2–70.6]) and fresh *Minas* cheese (33.3% [CI 95%: 27–40.1]).

We did not find any significant difference in the rejection rate among the food items from the ready-to-eat food group. However,

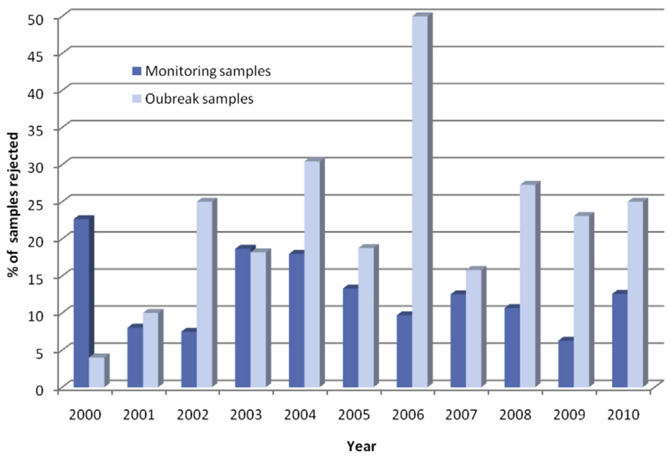

Fig. 2. Percentage of food ready for consumption and bottled water rejected samples according to the Brazilian legislation analyzed by the LACEN-DF from 2000 to 2010.

we did identify that sandwiches ($n = 115$) had the highest rate of rejected samples (40%). All rejected water samples were of mineral water. In the spices/seasonings group, black pepper (ground and whole peppercorns, $n = 157$) had a significantly higher percentage of rejected samples (21.6% [IC 95%: 15.4–28.9]), with a higher chance of being rejected when compared with the other samples of the group (OR = 4.8 [IC 95%: 2.1–10.7]). No samples from the animal products, snacks/appetizers, baby food, and chocolate/candies food groups for the period under study were rejected (Table 2).

Table 3 lists the microorganisms found in the food groups for which at least one sample was rejected for not complying with Brazilian legislation. TTC, *Salmonella* spp and CPS were the parameters most analyzed (74.4, 72 and 38.5% of the total samples collected, respectively). Milk/dairy products, ready-to-eat food, and spices/seasonings groups had the highest % of samples rejected due to TTC (12.0–12.7%). About 9% of the samples analyzed for CPS were rejected (156 samples) for containing counts over the legal limits (500–1000 CFU/g), mainly milk and dairy products (82 samples; 13.8%) (Table 3). About 26–27% of the *Minas* cheese samples were rejected for having TTC and/or CPS counts higher than the legal limits (500 MPN/g and 500 UFC/g, respectively). Almost 60% of the rejected pasteurized milk samples had TTC counts above the legal limits (2 MPN/mL).

Only 0.7% of the samples analyzed for *Salmonella* spp contained this microorganism (all rejected), mainly from the spices/seasonings group (Table 3), mostly black pepper. *B. cereus* was mostly investigated in ready-to-eat food (814 samples), of which 5.8% had microorganism counts above the legal limit (1000 CFU/g) (Table 3), and 40 samples had levels higher than the infectious dose (10^5 CFU/g). Of the 371 samples analyzed for *P. aeruginosa*, 97.6% were bottled water samples, with a 10.5% rejection rate (>2.2 MPN/100 mL; Table 3).

E. coli was present in 64 of the 866 samples analyzed for this parameter (7.4%). The detection rate for spices/seasonings (22.6% [CI 95%: 13.8–33.8]) was higher than for milk/dairy products (7.9% [CI 95%: 4.2–13.5%]) and fruit/vegetables (6.8% [CI 95%: 1.9–6.4]), but similar to ready-to-eat food (14.9% [CI 95%: 8.2–16.2]). Among the 12 samples of milk/milk products containing *E. coli*, 9 were of fresh *Minas* cheese. Brazilian legislation only establishes parameters for *E. coli* in bottled water (where it should not be present), but there was no positive sample for this microorganism among the bottled water samples analyzed by the LACEN-DF.

The presence of *S. aureus* was investigated in 276 samples (6% of all samples collected, 96% until 2005), with 31 positive samples, of which 25 of ready-to-eat food (80%), 5 of cheese samples and one of mineral water. In 13 samples, *S. aureus* was found at levels higher than 10^5 CFU/g and strains isolated from four outbreak samples (all ready-to-eat food from a single outbreak) tested positive for at least one enterotoxin investigated (SEA, SED, and/or TSSH-1).

3.2. Foodborne disease outbreaks reported

A total of 195 foodborne disease outbreaks occurring in the Federal District were notified by NATHA between 2000 and 2010, of which 162 were investigated. Over 40% of the investigated outbreaks occurred in 2009 (33) and 2010 (34). During these two years, 69 samples suspected to be involved in outbreaks were analyzed by the LACEN-DF (outbreak samples, Fig. 1).

The food involved was identified in 117 of investigated outbreaks (72.2%), with 126 samples collected (Table 4). Ready-to-eat food (including sandwiches, mayonnaise and meat food) were the incriminated foods in over half of the outbreaks. The etiological agent was identified in 80 outbreaks, 41.2% of which involved *B. cereus*, 26.2% *S. aureus*, and 22.5% *Salmonella* spp. A total of 104 food samples were collected from these 80 outbreaks (Table 4).

Table 2
Samples analyzed by the LACEN-DF between 2000 and 2010, and percentage of rejection according to Brazilian legislation, *per* food group.

Group	Monitoring samples		Outbreak samples	
	Analyzed, N	Rejected, % (CI 95%)	Analyzed, N	Rejected, % (CI 95%)
Milk and dairy products	1587	16.1 (14.3–18.0)	23	21.7 (7.4–43.7)
Ready-to-eat food	853	18.9 (16.3–21.6)	251	23.5 (18.4–29.2)
Water	385	17.4 (13.7–21.5)	6	0 (0–45.9)
Spices, seasonings, condiments	314	13.3 (9.8–17.6)	0	0
Fruit and vegetables, raw or canned	272	4.0 (1.9–6.9)	4	25.0 (0.6–80.5)
Non-alcoholic beverages	232	2.6 (0.9–5.5)	5	20.0 (0.5–7.1)
Flour, cereals, cakes, industrialized	207	1.0 (0.1–3.4)	9	0 (0–33.6)
Ice cream and sorbets	181	11.0 (6.8–16.5)	0	0
Animal products, processed, and eggs	138	0 (0–2.6)	22	0 (0–15.4)
Snacks and appetizers	30	0 (0–11.5)	0	0
Baby food	26	0 (0–13.2)	2	0
Chocolates, candies, sweets	26	0 (0–16.1)	3	0 (0–7)
Total	**4251**	**13.3 (12.2–14.2)**	**325**	**20.3 (15.9–24.7)**

CI = confidence interval confidence interval at 95%.

The investigated outbreaks involved 3904 cases, with an average of 24.1 sick individuals *per* outbreak; 699 individuals received medical care. The highest number of individuals *per* outbreak occurred in 2003 (305 individuals in 4 outbreaks), but neither the food involved nor the etiological agent was identified in any of these outbreaks. Most of the individuals involved were between 20 and 49 years of age, but in 2007 they were mostly between the ages of 5 and 9 (31 individuals). The only death registered during the period occurred in 2010 due to an outbreak at a foster home that affected 32 children, 11 of whom were under the age of 4. Rotavirus was the agent involved, but the incriminated food was not identified.

The criterion used to conclude the investigation was included in 101 investigated outbreak reports (62.3%). The criterion was clinical-epidemiological in 67.3% of them, laboratory-confirmed in 17.8%, and based on food analysis alone or including laboratory confirmation in 14.8%.

4. Discussion

The results of this investigation have shown that milk and dairy products was the food group most analyzed by the LACEN-DF, and was also the group with the highest rejection rate. Cheese had the highest frequency of rejection within this group, mainly *Minas* cheese. Typically Brazilian, *Minas* cheese is the most consumed cheese in the country (about 30 g/person/day; IBGE, 2011). The fresh type, which can be made with raw milk in artisanal production (Regulation 44864/2008; state of Minas Gerais), has high humidity (maximum of 45.9%), and a maximum shelf life of 9 days under refrigeration. Standard/ripened *Minas* cheese undergoes maturation for 20–30 days before marketing, with a shelf life of about 90 days under refrigeration (Perry et al., 2004).

About 13% of the milk and dairy product samples analyzed by the LACEN-DF were rejected due to high TTC counts, mainly *Minas* cheese samples. Salotti, Carvalho, Amaral, Vidal-Martins, and Cortez (2006) found 75% of the 60 fresh artisanal and industrial *Minas* cheese samples analyzed in São Paulo to be non-compliant with the legal limits for TTC. TTC is an indicator of the possible presence of pathogens such as *E. coli*, which was detected in 7.9% of the milk/dairy samples analyzed by the LACEN-DF for this parameter, mostly fresh *Minas* cheese. The presence of *E. coli* has been widely accepted as an indicator of faecal contamination during food handling (Campos, Kipnis, Porfírio, & Borges, 2006). *E. coli* was present in all 55 samples of *Minas* cheese produced with raw milk in Minas Gerais, and in 70% of the 24 samples of fresh *Minas* cheese in the state of Goiás (Campos et al., 2006). Rosengren, Fabricius, Guss, Sylven, and Lindqvist (2010) found *E. coli* in 34% of the 55 raw milk cheeses analyzed in Sweden; levels in raw milk fresh cheese were significantly lower when starter cultures were used, indicating that contamination occurred later in the cheese production process.

About 26% of the *Minas* cheese samples analyzed by the LACEN-DF were rejected for containing CPS counts higher than the legal limits, a lower rate than that found by Moraes, Vicosa, Yamazi, Ortolani, and Nero (2009; 30.9%) and Brant, Fonseca, and Silva (2007; 92.5%) in samples collected in Minas Gerais. Salotti et al. (2006) found 15% of the 60 samples of fresh *Minas* cheese containing high CPS counts, with higher rejection rates among artisanal cheese. Among the *Staphylococcus* species capable of producing coagulase, *S. aureus* is the most prevalent in

Table 3
Microorganisms found in samples analyzed by the LACEN-DF for which there was at least one rejected sample.

Group	Number of samples analyzed (% rejection due to the microorganism)							
	TTC	*Salmonella*	CPS	*B. cereus*	*P. aeruginosa*	Coliforms	MAC	Coliforms at 30°
All groups[a]	3408 (9.3)	3294 (0.7)	1763 (8.9)	1423 (3.6)	371 (10.5)	885 (10.4)	590 (3.0)	133 (20.3)
Milk and dairy	1137 (12.7)	1011 (0.5)	592 (13.8)	276 (1.4)	2 (50.0)	409 (14.4)	508 (2.9)	108 (25.0)
Ready-to-eat	935 (12.7)	132 (0.7)	772 (8.9)	814 (5.8)	6 (0)	24 (4.2)	8 (0)	19 (0)
Water	360 (1.4)	32 (3.1)	4 (25)	22 (0)	362 (10.5)	363 (5.2)	2 (0)	0 –
Spices/seaso-nings	257 (12.0)	257 (6.6)	4 (0)	47 (0)	1 (100)	1 (0)	2 (0)	0 –
Fruit & veg., raw, canned	186 (6.4)	185 (0)	66 (0)	50 (0)	0 –	2 (0)	9 (0)	1 (0)
Non-alcoholic beverages	145 (0.7)	132 (0)	17 (0)	55 (1.8)	0 –	27 (0)	17 (0)	0 –
Flour, cereals, cakes	197 (0.5)	179 (0)	103 (0.9)	77 (0)	0 –	9 (11.1)	8 (0)	3 (0)
Ice cream/sorbet	70 (4.2)	176 (0)	130 (3.8)	16 (0)	0 –	27 (44.4)	28 (10.7)	0 –

TTC = thermotolerant coliforms; CPS = Coagulase-positive staphylococci; MAC = mesophilic aerobic count.
[a] includes food groups for which none of the samples was rejected.

Table 4
Food and etiologic agents identified in the foodborne disease outbreaks occurring in the Federal District from 2000 to 2010 and investigated by NATHA.

Food	NF_1	%[a]	Agent	NF_2	%[b]
Outbreaks with food identified = 117			Outbreaks with agent identified = 80		
Ready-to-eat food	60	51.3	*B. cereus*	33	41.2
Bakery products	14	12.0	*S. aureus*	21	26.2
Water	11	9.4	*Salmonella* spp	18	22.5
Cheese	7	6.0	*C. perfringens*	15	18.8
Crockets/savory	7	6.0	*E. coli*	5	6.2
Fruits/nuts	7	6.0	*V. parahaemolyticus*	3	3.8
Eggs/egg products	7	6.0	Others	9	11.2
Others	12	11.1	**Total of samples**	**104**	
Total of samples	**126**				

NF_1 = number of food samples collected during the outbreaks with the food identified.
NF_2 = number of food samples collected during the outbreaks with the agent identified.
[a] Relative to 117 outbreaks with the food identified.
[b] Relative to 80 outbreaks with the agent identified.

staphylococcal food intoxication outbreaks worldwide, causing nausea, stomach cramps, vomiting, and diarrhea (FDA, 2012; Hennekinne, de Buyser, & Dragacci, 2012). Cow mastitis, mainly caused by *S. aureus,* is considered one of the main problems in dairy cattle infections, and may affect the sanitary conditions of the produced milk (Zafalon, Arcaro, Filho, Ferreira, & Veschi, 2009). *S. aureus* was only found in cheese samples among the 25 milk/dairy samples analyzed for this parameter by the LACEN-DF.

Salmonella spp and *E. coli* were mostly found in samples from the spices/seasonings group, mainly black pepper. Moreira, Lourenção, Pinto, and Rall (2009) found 5.6% of the 233 spice samples collected in the state of São Paulo containing *Salmonella*, similar to what was found in the Federal District. The authors also found black pepper (and cumin), to have the lowest microbiological quality among the spices. In Brazil, contamination of black pepper with *Salmonella* spp and other enterobacteria occurs mainly due to the lack of good manufacturing practices, mostly during the drying process, which may occur on the floor in an open area to which domestic animals have free access (Duarte & Albuquerque, 2005). A *Salmonella* outbreak associated with salami products made with imported contaminated black and red pepper was recently reported in the USA (Gieraltowski et al., 2012).

About 20% of the ready-to-eat food samples analyzed by the LACEN-DF were rejected, a rate similar to what was found by Cardoso et al. (2010) in ready-to-eat food samples collected at 83 public schools in the state of Bahia (20.4%). The authors also found 2.4% of the samples containing *E. coli,* a much lower frequency than that found in the Federal District (14.9% of the ready-to-eat food samples analyzed for this parameter). High contamination of ready-to-eat food is a worldwide issue and is most likely caused by inadequate cleaning practices or hygienic handling (Christison, Lindsay, & von Holy, 2008; Sospedra, Rubert, Soriano, & Mañes, 2013).

Data obtained from NATHA showed that ready-to-eat food was the food group most involved in the outbreaks investigated in the Federal District, confirming other studies that show that foods that are handled during preparation and are not heated prior to consumption present the greatest risk (Tebbutt, 2007). The etiological agents identified in the outbreaks investigated by the NATHA included *B. cereus, S. aureus*, and *Salmonella* spp., which were also the most prevalent agents found in the outbreak samples analyzed by the LACEN-DF (data not shown).

The clinical-epidemiological criterion was used to conclude almost 70% of the outbreaks investigated by NATHA, a result that was expected as most of the notifications came from the hospitals where the patients were under care. In Brazil, when the food involved in an outbreak is identified by the affected individuals, the food in normally discarded to avoid additional exposure. Hence, very rarely is the food available for analysis. Indeed, food analysis information was used to conclude only 14.8% of the investigations, similar to what was found for the 3737 outbreaks reported nationwide from 1999 to 2004 (15.5%; do Carmo et al., 2005).

In a review to investigate the contribution of microbiological analysis to food safety in England and Wales, Tebbutt (2007) pointed out that, although food microbiological analysis was important during outbreak investigation, routine testing by local authorities is often of limited use and could be improved by more targeted surveillance. In the Federal District, ready-to-eat food, the food group most involved in the outbreaks investigated by NATHA, was the second most analyzed food group under the LACEN-DF surveillance program. However, we found that the results of the outbreak sample analysis conducted by the LACEN-DF were of limited use for the investigation of the outbreaks. Although 126 food items were identified as being involved in the outbreaks investigated by NATHA, only in 15 of the outbreaks did the food analyzed contribute to the conclusion of the investigation. On the other hand, 44 outbreak samples analyzed by the LACEN-DF had either *B. cereus* or *S. aureus* above the infectious dose or tested positive for *E. coli* or *Salmonela* spp. However, these results were not reflected in the outbreak investigations conducted by NATHA.

The increase in the number of foodborne disease outbreaks reported and investigated in the Federal District in 2009 and 2010 indicates a significant improvement in local epidemiological surveillance efforts. However, about one-third of the investigated outbreaks did not have the food involved identified, over half did not have the agent identified, and almost 40% did not have the conclusion criterion reported. Lack of the etiologic agent is mainly due to late notification and sampling, the use of antibiotics by the affected population, and the limited number of routine analyses conducted by the laboratories, including the identification of the enterotoxigenic strains (do Carmo et al., 2005). Only four outbreak samples analyzed by the LACEN-DF during the period under study were tested for staphylococcal enterotoxins.

At the national level, over 8500 outbreaks were reported by the local sanitary authorities to the national epidemiological surveillance system from 2000 to 2011, with 88 deaths (MS, 2012b). The main vehicles involved were mixed foods (~1500 outbreaks) and eggs and egg products (~900 outbreaks); milk and dairy products were involved in over 300 outbreaks. *Salmonella* spp was the main etiologic agent identified (~1700 outbreaks), followed by *S. aureus* (~800 outbreaks; MS, 2012b). This profile is different from what was found in the Federal District, where *B. cereus* was the main agent identified in the investigated outbreaks.

The number of food disease outbreaks reported in Brazil is most likely underestimated. According to do Carmo et al. (2005), over 3 million hospitalizations due to foodborne diseases (ICD 10 A00 to A09) occurred in the country from 1999 to 2004, and 25,281 fatalities from 1999 to 2002. Most of the outbreaks involved in these cases were probably never investigated or reported. Additionally, the reporting of foodborne disease outbreaks only became compulsory in the country in 2011, being restricted to outbreaks occurring on ships or aircraft (Regulation 104/2011, Brazilian Ministry of Health).

The results of this study indicate that the LACEN-DF should prioritize the analysis of ready-to-eat food under its microbiological surveillance programs, due to their frequent involvement in foodborne disease outbreaks. Efforts should be made to improve the outbreak notification and investigation system and the laboratory capabilities in the Federal District so that biological and food samples may be collected in a timely and correct manner so as to identify the etiological agent. Additionally, it is essential that CPS positive samples be tested for *S. aureus* and the toxigenic potential of the isolated strains determined so that the vehicle involved in the outbreaks are identified. This is the first study reporting governmental microbiological data on food and water in Brazil that relates food analyses to outbreak reporting. Comparison with other scenarios in the country is therefore not possible.

This study highlighted the inadequate hygienic-sanitary conditions of the *Minas* cheese available for consumption in the Federal District and other Brazilian regions, and the importance of implementing control measures and good manufacturing practices during the handling and storage of this product.

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