CARACTERÍSTICAS MICROBIOLÓGICAS DE SUSHIS ADQUIRIDOS EM ESTABELECIMENTOS QUE COMERCIALIZAM COMIDA JAPONESA.

MICROBIOLOGICAL CHARACTERISTICS OF SUSHI ACQUIRED IN ESTABLISHMENTS THAT SELL JAPANESE FOOD.

R. A. SATO[1]; O. D. ROSSI JUNIOR[1]; K. P. BÜRGER[1]; P. M. ÉVORA[1]
[1] Universidade Estadual Paulista – UNESP - Câmpus de Jaboticabal.
rafaelsato.vet@gmail.com

Resumo – Com crescimento do consumo da comida japonesa no Brasil surge a preocupação com doenças de origem alimentar, relacionadas à ingestão do pescado cru e erros de manipulação desse tipo de alimentos. Diante dessa realidade, esse trabalho avaliou a qualidade microbiológica de sushis adquiridos em 15 restaurantes especializados em comida japonesa e 15 restaurantes não especializados da região de Ribeirão Preto – SP, quantificando microrganismos mesófilos, psicrotróficos, coliformes termotolerantes, *Staphylococcus* spp. e *S. aureus*, *Escherichia coli;* e presença da *Salmonella* sp. e do *Vibrio parahaemolyticus* . As populações de microrganismos heterotróficos mesófilos e psicrotróficos variaram de 10^2 a 10^7 UFC.g^{-1} e 10^2 a 10^9 UFC.g^{-1}, respectivamente. Todas as amostras analisadas apresentaram *Staphylococcus* sp., com populações variando de 10^2 a 10^5 UFC.g^{-1}, sendo que 23,3% foram caracterizados como *Staphylococcus* coagulase positivo e 13,3% apresentaram valores de populações acima do limite estabelecido pela legislação vigente para pescado cru. Foram encontrados coliformes termotolerantes 60,0% das amostras e 33,3% das estavam acima do limite estabelecido. As presenças de *Staphylococcus aureus* e *Escherichia coli* foram identificadas em 16,7% e 30,0% das amostras, respectivamente. *Salmonella* spp. e *Vibrio parahaemolyticus* não foram isoladas nesse trabalho. Não houve diferença na qualidade microbiológica de sushis oferecidos pelos dois tipos de estabelecimentos. O sushi é um alimento com potencial de veiculação de doenças de origem alimentar. As amostras fora do padrão refletem a necessidade de treinar adequadamente os manipuladores de alimento.

Palavras-chaves: *Escherichia coli.* Microbiologia. Salmão. *Staphylococcus aureus. S*ushi.

Abstract - Food quality control refers to all and any action aimed at improving good practice in hygiene procedures and food handling. Therefore, there is a continuous need to obtain data, study and monitor the production and marketing in the food industry, thus producing a "safe food", free from any contamination that may cause damage to the consumer´s health. Given this need, this study evaluated the microbiological quality of sushi bought in 15 restaurants specialized in Japanese food and 15 non-specialized restaurants from the region of Ribeirão Preto - SP. The populations of heterotrophic mesophiles and psychrotrophic microorganisms ranged from 3,9 x 10^2 to 1,7 x 10^7CFU.g^{-1} and 7,5 x 10^2 to 1,4 x 10^9CFU.g^{-1}, respectively. All samples presented *Staphylococcus* sp., with populations ranging from 2,0 x 10^2 to 3,8 x 10^5CFU.g^{-1}, where 23,3% were classified as positive coagulase *Staphylococcus* and 13.3% had population values above the established limit. Total and thermotolerant coliforms were found in 83,3% and 60,0% of the samples, respectively, and 33,3% of them were above the established thermotolerant coliform limit. The presence of *Staphylococcus aureus* and *Escherichia coli* were identified in 16,7% and 30,0% of the samples, respectively. *Salmonella* sp. and *Vibrio parahaemolyticus* were not isolated in this research. There was no difference in the microbiological quality of sushi offered by the two types of establishments.

Keywords: *Escherichia coli*. Microbiology. Salmon; *Staphylococcus aureus*. Sushi.

INTRODUCTION

The gastronomic sector in Brazil offers a wide variety of cuisine and is responsible for generating many direct jobs in the country. Within this sector, "sushi bars", "fast-foods" and restaurants specialized in Japanese food have been highlighted by the growth in the number of establishments that offer this type of service. This growth is due to the popularization of Japanese food, which currently has the "status" of being healthy and nutritious, therefore conquering new adepts.

The traditional Japanese cuisine is worldwide known for the habit of consuming raw or *in natura* fish. However, from the Public Health´s point of view, the *in natura* ingredient generates a concern related to foodborne diseases.

Foodborne diseases occur when an individual ingests food contaminated with infectious or toxic agents that enter the body. Among the potentially pathogenic microorganisms that can be transmitted through raw fish, we can mention *Aeromonas sp*., *Escherichia coli*, *Salmonella* sp., *Pseudomonas* sp., *Staphylococcus aureus*, *Vibrio cholerae* and *Vibrio parahaemolyticus*.

Given the growth in Japanese food consumption, combined with food contamination risks during production, storage and sale, there is a preoccupation with the quality of products sold in specialized and non-specialized stores.

Thus, the present study aimed to evaluate the microbiological characteristics of sushi sold in restaurants specialized in Japanese food and in non-specialized establishments, by quantifying the presence of mesophilic and psychrotrophic microorganisms; total and thermotolerant coliforms; *Staphylococcus* sp. and *S. aureus; Escherichia coli*; *Vibrio parahaemolyticus* and *Salmonella* spp., and to compare the results with pre-established quality parameters for raw fish from the current legislation.

MATERIAL AND METHODS

The study was performed on 30 samples of *in natura* sushi consisting of seasoned rice and salmon (*Salmo salar*), which may present other ingredients such as algae, sauces and vegetables, preserved refrigerated. Samples were collected from fifteen restaurants specialized in Japanese food and fifteen non-specialized commercial establishments, and a sample was acquired from each location.

The specialized establishments were characterized as restaurants that only work with Japanese cuisine, usually offering the customer the service of “all you can eat”, "a la carte" or "self-service" per kilo. The non-specialized establishments category included ordinary restaurants (fast foods, self-services, steakhouses, etc.) that daily offer customers traditional Brazilian food, but also incorporate other types of cuisine, including Japanese, into the menu.

The samples were collected from the cities Jaboticabal - SP, Ribeirão Preto - SP and Monte Alto - SP. The quantities of sushi acquired varied according to the "menu" offered by the chosen establishment. When the establishment offered the option to sell by weight, approximately 400g of sushi were weighed for purchase. All samples were packed in the ordinary form of sale to the consumer.

Immediately after purchase the samples were conditioned in isothermal boxes, containing blocks of ice, and taken to the Laboratory of Microbiological Analysis of Animal-derived Food and Water of the Department of Preventive Veterinary Medicine and Animal Reproduction of the FCAV / Unesp, where they were submitted to quantification of mesophilic and psychrotrophic microorganisms, thermotolerant coliforms, *Staphylococcus* sp. and *aureus*, *Escherichia coli* and presence of *Vibrio parahaemolyticus* and *Salmonella* sp.. The methodologies described in the American Public Health Association (APHA, 2001) were used for the analyzes.

RESULTS AND DISCUSSION

The results obtained, especially of the microbial groups predicted by the legislation, were compared with parameters pre-established by Resolution - RDC Nº. 12, of January 2, 2001, for raw and similar fish (BRASIL, 2001).

Table 1 shows the distribution of the *in natura* sushi samples according to the exponential of the populations of heterotrophic mesophilic microorganisms, classified according to the type of establishment where the sushi was acquired.

In the Brazilian legislation there is no defined limit on the acceptable number of mesophilic microorganisms, so a comparison was made with values cited in the literature.

According to ICMSF (2009), the population of mesophilic microorganisms found in a food is one of the indicators of its quality, and should not exceed 10^6CFU.g^{-1}. A high population of these microorganisms may indicate excessive contamination of the raw material or during preparation, as well as inadequate cleaning and sanitation conditions. Thus, 20% of the analyzed samples presented values above this limit, indicating failures or lack of good manipulation practices in these products. Errors in the conservation process and transport also contribute to increase the populations of these microorganisms.

Considering mesophilic microorganisms, Gilbert et al. (2000) presented a grading scale for the quality of raw marine fish food where products with populations under 10^3CFU.g^{-1} are considered suitable for consumption, between 10^3 and 10^4CFU.g^{-1} are satisfactory and above 10^4CFU.g^{-1} are considered unsatisfactory. Taking into account this classification, it can be stated that 73,3% of the samples collected in this research correspond to products considered unsatisfactory for commercialization.

Table 2 shows the distribution of the *in natura* sushi samples according to the exponential of the populations of heterotrophic psychrotrophic microorganisms, classified according to the type of establishment where the sushi was acquired.

In Brazilian legislation, there is also no defined limit regarding the acceptable number of psychrotrophic microorganisms. According to the ICMSF (2009) the limit for populations of psychrotrophic microorganisms in food is 10^7CFU.g^{-1}. Taking into account this parameter, it can be stated that 13.3% of the samples collected correspond to unsatisfactory products for commercialization.

According to Reinbold (1983), aerobic psychrotrophic microorganisms population evaluates the deterioration degree of refrigerated food. Considering that for psychrotrophic microorganisms values above 10^4CFU.g^{-1} are relatively high, and that sushi is a food with high deterioration potential (being prepared manually and presenting raw fish as an ingredient), 83,3% of the samples would not have a long shelf-life under refrigeration.

Table 3 refers to the distribution of *in natura* sushi samples according to the exponential population of *Staphylococcus* sp.

Among the 30 analyzed samples all presented *Staphylococcus* sp., as observed in Table 3. Bacteria of this genus are part of the normal human skin microbiota, and food contamination may occur during handling, especially when protective equipment and appropriate forms of asepsis are not used. Sushi is a food that by tradition is handmade, making it a product with high potential of contamination with this microorganism.

Table 4 presents the results of coagulase positive staphylococci populations in the *in natura* sushi samples, distributed according to the type of establishments in which they were acquired, and the samples in which the presence of *Staphylococcus aureus* was confirmed.

Coagulase positive *Staphylococcus* was present in 7 of the 30 samples analyzed, totaling 23,3% of the samples (Table 4). The production of coagulase is an important characteristic used in the identification of *S. aureus*, therefore, the Brazilian legislation indicates the research of this enzyme for the characterization of this microorganism in food. Of these 7 samples, only 4 had population values above $5,0 \times 10^3$CFU.g^{-1}, the maximum limit established by the legislation. Consequently, 13,3% of the total samples analyzed would be unsatisfactory for consumption.

S. aureus has worldwide distribution, and it is estimated that is found on the skin and in the nose of about 25% of healthy people and animals (CDC, 2017). According to Franco and Landgraf (2003), this microorganism is frequently associated with outbreaks of foodborne diseases. The presence of *S. aureus* (Table 4) was confirmed in 5 samples: 1, 4 and 10 from the specialized establishments group, and only in samples 1 and 12 from the non-specialized establishments group. Thus, the presence of *Staphylococcus aureus* was confirmed in 16,7%

of the total samples analyzed. According to Silva Junior (2001), the research of this bacterium in food allows to evaluate the hygienic sanitary quality during its manipulation and preparation. Therefore, the positive samples found represent a risk to Public Health, reflecting the need to improve the production process of sushi, as well as to train the manipulators of this food to perform their tasks correctly.

Table 5 refers to the distribution of *in natura* sushi samples according to the exponential of thermotolerant coliforms (MPN.g^{-1}).

Thermotolerant coliforms are bacteria that indicate fecal contamination and possible pathogenic enterobacteria presence, and it is also used in food´s sanitary evaluation due to the low cost of research of these microorganisms (JAY, 2005). For thermotolerant coliforms, in Table 5, it was verified that 33,3% of the samples presented values above 10^2MPN. g^{-1}, the maximum limit established by the legislation for similar foods. Therefore, these samples would be considered unacceptable for consumption, showing that in some stage of the production chain process of these sushi there were failures related to good hygiene practices, which might have been during handling or obtaining of the raw material.

Table 6 shows the samples in which *Escherichia coli* populations and thermotolerant coliform populations were identified, according to the type of establishment in which they were collected.

In Table 6, it is possible to observe the presence of *E. coli* in samples 1, 8 and 14 of the specialized establishments group and in samples 1, 2, 5, 10, 11 and 14 of the non-specialized establishments group, with populations ranging from 0,15 x 10 to 7,5 x 10MPN.g^{-1}totaling 9 samples from the 30 analyzed (30,0%). Several strains of this microorganism are pathogenic to humans and the presence of *E. coli* in a food confirms fecal contamination (FRANCO and LANDGRAF, 2003). Thus, 30,0% of the sushi analyzed presented fecal residues in some phase of the manipulation, storage or obtaining of the raw material. Franco and Landgraf (2003) recommend that food should not present *E. coli.*

Table 7 presents, in summary, the arithmetic means and standard deviations of the populations of heterotrophic mesophilic and heterotrophic psychrotrophic microorganisms, *Staphylococcus* sp., total coliforms and thermotolerant coliforms, distributed according to the type of establishment where the samples were collected.

The Student t test was applied to the values of the microorganism populations specified in Table 7, and from the statistical point of view there were no significant differences between the means of these populations ($p > 0,05$).

At the beginning of this study, it was expected that there were significant differences between the results obtained in the two groups of restaurants studied. Specialized restaurants work only with Japanese cuisine, and many advertise strict hygiene measures during the preparation of the sushi, as well as the efficiency, quality, tradition and expertise of their "sushimen". These employees are mandatorily trained in workshops to learn how to prepare good quality sushi. Non-specialized restaurants, however, work with various types of cuisine and although they require good cooks, they don´t train the staff for good sanitary practices during their service.

However, the results obtained at the end of this study showed that there were no differences in the quality of the sushi offered to the consumer by the two types of restaurants. Although all the samples analyzed presented excellent visual appearance when collected, the high amounts of heterotrophic mesophilic and psychrotrophic microorganisms, and relatively high populations of total and thermotolerant coliforms reflect inadequate hygienic and sanitary conditions during preparation, handling, or even low-quality raw material used by the commercial establishments. This diagnosis demonstrates the deficiency of the governmental inspection agencies that should be controlling the quality of these products, as well as the lack of training and qualification of food professionals.

Another important factor observed in this study was the presence of *Staphylococcus aureus* and *Escherichia coli*. These microorganisms are important for Public Health because, under ideal conditions for their multiplication, they can cause severe illnesses, being able to affect great part of consumers.

Vibrio parahaemolyticus and microorganisms of the *Salmonella* genus were not found in any of the analyzed samples. Therefore, all samples were kept within the established standard, which is defined by absence of *Salmonella* spp. in 25.0 g of the food and a maximum limit of 10^3MPN.g^{-1} for *V. parahaemolyticus* in dishes that contain raw fish (BRASIL, 2001).

Vallandro (2010), at the end of his work, reported that the most probable source of contamination by *Salmonella* spp. and *Vibrio parahaemolyticus* in salmon sashimi (*Salmo salar*) is the place of fish breeding and/or capture. The fact that the raw material used by the restaurants have well-known origin and are imported from inspected establishments probably contributed to the absence of these microorganisms. As all samples analyzed in this study were necessarily consisted of salmon, the same conclusion can be adopted to explain the absence of these pathogens. It is important to note that sushi, unlike sashimi, is a food composed of several ingredients. Thus, contamination by *Salmonella* spp. could also occur through eggs, sauces, cheeses, vegetables and other varieties of components used for their preparation.

Regarding *Vibrio parahaemolyticus*, Vallandro (2010), presents a hypothesis that the absence of this microorganism in sashimi can be related to the fact that the salmon consumed in Brazil is bred in cold waters, mainly in Chile. According to Forsythe (2002) and quoted by Vallandro (2010), it is probable that this characteristic contributes to the low occurrence of the bacterium or its low presence in the fish, since *V. parahaemolyticus* is normally present in quantities greater than 10^3CFU.g^{-1} in fish and seafood proceeding from warm waters.In addition, the salmon consumed is transported frozen to Brazil, which is also adverse to the bacteria survival, according to Cook and Ruple (1992) and quoted by Vallandro (2010).

Table 8 presents a summary of the non-standard samples for coagulase positive *Staphylococcus*and thermotolerant coliforms, distributed according to the type of establishment where the samples were acquired.

Fisher's exact test was applied between the groups of specialized and non-specialized establishments, comparing the standard and non-standard samples for coagulase positive *Staphylococcus* and thermotolerant coliforms. As results, there were no statistically significant differences between the two groups of establishments (p> 0,05), therefore, there were no differences in the frequencies found between the standard and non-standard samples for the two types of establishments, concerning the microorganisms.

At the end of this work, it was verified that the total of non-standard samples were 11 (36,6%). The chi-square test was applied between the groups of specialized and non-specialized establishments, comparing the standard and non-standard samples. As a result, there was no statistically significant difference between the groups of establishments (x2 <x2c = 0,05), therefore, the frequencies found between the standard and non-standard samples for the 2 types of restaurants were the same.

CONCLUSIONS

The results of this work suggest that sushi can potentially cause foodborne diseases since microbiological isolation and biochemical confirmations of the agents *Staphylococcus aureus* and *Escherichia coli* were possible. These data, added to the amount of non-standard samples found, reflect the need to invest in the training of food handlers in order to raise awareness of how and why to produce food with good hygienic sanitary quality.

The present study also demonstrates the importance of continuing, and deepening the studies on the subject. Japanese food has gained prominence in Brazil, and sushi is a much manipulated food that has several ingredients, one of them being raw material. Therefore, there is a need to study the entire sushi production chain to identify the critical points and seek solutions aiming at the production of a healthy and safe food for the society.

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Table 1. Exponential distribution of mesophilic heterotrophic microorganisms populations. (Jaboticabal, Ribeirão Preto and Monte Alto - SP, 2011 and 2012).

Exponential of mesophilic microorganisms (CFU.g^{-1})	**Numberofsamples (%)**		**Total (%)**
	specialized	**Nonspecialized**	
10^2	2 (13.3)	0 (0.0)	2 (6.7)
10^3	2 (13.3)	4 (26.7)	6 (20.0)
10^4	5 (33.3)	3 (20.0)	8 (26.7)
10^5	2 (13.3)	6 (40.0)	8 (26.7)
10^6	2 (13.3)	1 (6.7)	3 (10.0)
10^7	2 (13.3)	1 (6.7)	3 (10.0)
Total ofsamples	15 (100.0)	15 (100.0)	30 (100.0)

Table 2. Exponential distribution of psychrotrophic heterotrophic microorganisms populations. (Jaboticabal, Ribeirão Preto and Monte Alto - SP, 2011 and 2012).

Exponential of psychrotrophic microorganisms (CFU.g^{-1})	**Numberofsamples (%)**		**Total (%)**
	specialized	**Non specialized**	
10^2	0 (0.0)	1 (6.7)	1 (3.3)
10^3	2 (13.3)	2 (13.3)	4 (13.3)
10^4	6 (40.0)	2 (13.3)	8 (26.7)
10^5	4 (26.7)	4 (26.7)	8 (26.7)
10^6	1 (6.7)	4 (26.7)	5 (16.7)
10^7	1 (6.7)	1 (6.7)	2 (6.7)
10^8	0 (0.0)	1 (6.7)	1 (3.3)
10^9	1 (6.7)	0 (0.0)	1 (3.3)
Total of samples	15 (100.0)	15 (100.0)	30 (100.0)

Table 3. Exponential distribution of *Staphylococcus sp* populations. (Jaboticabal, Ribeirão Preto and Monte Alto - SP, 2011 and 2012).

Exponential of *Staphylococcus sp* populations. ($CFU.g^{-1}$)	**Numberofsamples (%)**		**Total (%)**
	Specialized	**Non specialized**	
10^2	2 (13.3)	1 (6.7)	3 (10.0)
10^3	10 (66.7)	8 (53.3)	18 (60.0)
10^4	2 (13.3)	5 (33.3)	7 (23.3)
10^5	1 (6.7)	1 (6.7)	2 (6.7)
Total ofsamples	15 (100.0)	15 (100.0)	30 (100.0)

Table 4. Populations of coagulase positive staphylococci and the samples in which the presence of *Staphylococcus aureus* was confirmed. (Jaboticabal, Ribeirão Preto and Monte Alto - SP, 2011 and 2012).

Sample number	**Specialized establishments (Pop. $CFU.g^{-1}$)**	**Confirmed *Staphylococcus aureus***
1	7.5×10^4 *	X
4	4.5×10^2	X
10	4.9×10^2	X
	non specialized establishments (Pop. $CFU.g^{-1}$)	
1	5.1×10^4 *	X
5	7.4×10^2	
6	5.7×10^3 *	
12	1.2×10^4 *	X

* Non-standard samples for coagulase positive *Staphylococcus*. Value above $5.0 \times 10^3 CFU.g^{-1}$ according to RDC Nº 12, dated January 2, 2001 (BRASIL, 2001).

Table 5. Exponential distribution of thermotolerant coliform populations (NMP.g^{-1}). (Jaboticabal, Ribeirão Preto and Monte Alto - SP, 2011 and 2012).

Exponential of thermotolerant coliforms (MPN.g^{-1})	**Number of samples (%)**		**Total (%)**
	Specialized	**Non specialized**	
absence (< 3.0)	8 (53,3)	4 (26,7)	12 (40,0)
< 10	1 (6,7)	1 (6,7)	2 (6,7)
10	2 (13,3)	4 (26,7)	6 (20,0)
10^2	1 (6,7)	5 (33,3)	6 (20,0) *
10^3	1 (6,7)	1 (6,7)	2 (6,7) *
10^4	2 (13,3)	0 (0,0)	2 (6,7) *
Total ofsamples	15 (100,0)	15 (100,0)	30 (100,0)

* Non-standard samples for thermotolerant coliforms. Value above 10^2 (MPN. g^{-1}) according to RDC – Nº12, of January 2, 2001 (BRASIL, 2001).

Table 6. Distribution of the samples in which *Escherichia coli* and thermotolerant coliforms were quantified according to the type of commercial establishment.(Jaboticabal, Ribeirão Preto and Monte Alto - SP, 2011 and 2012).

Sample number	**thermotolerant coliforms population Specialized establishments (Pop. MPN.g^{-1})**	***Escherichia coli* population (Pop. MPN.g^{-1})**
1	4,6 x 10^4 *	0,75 x 10
8	3,6 x 10	0,15 x 10
14	1,2 x 10^4 *	7,5 x 10
	nonspecialized establishments (Pop. MPN.g^{-1})	
1	2,1 x 10^3 *	0,39 x 10
2	3,6 x 10^2 *	0,15 x 10
5	3,9 x 10^2 *	0,15 x 10
10	9,1 x 10^2 *	0,2 x 10
11	9,3 x 10	0,15 x 10
14	4,3 x 10	0,15 x 10

* Non-standard samples for thermotolerant coliforms. Value above 10^2 NMP.g^{-1}according to RDC - Nº12, of January 2, 2001 (BRASIL, 2001).

Table 7. Arithmetic means and standard deviations of the populations of mesophilic and psychrotrophic microorganisms, *Staphylococcus sp*., total coliforms and thermotolerant coliforms. (Jaboticabal, Ribeirão Preto and Monte Alto - SP, 2011 and 2012).

Microorganisms	**Arithmetic means of the populations**	
	Specialized	**Non specialized**
Heterotrophic Mesophilic	$2{,}0 \times 10^6 \pm 4{,}6 \times 10^6$	$1{,}8 \times 10^6 \pm 4{,}6 \times 10^6$
Psychrotrophic	$9{,}4 \times 10^7 \pm 3{,}6 \times 10^8$	$1{,}1 \text{ X } 10^7 \pm 3{,}3 \times 10^7$
*Staphylococcus*sp.	$3{,}5 \times 10^4 \pm 9{,}8 \times 10^4$	$3{,}3 \times 10^4 \pm 6{,}7 \times 10^4$
Total coliforms	$1{,}6 \times 10^4 \pm 3{,}8 \times 10^4$	$4{,}9 \times 10^3 \pm 1{,}2 \times 10^4$
Thermotolerant coliforms	$4{,}0 \times 10^3 \pm 1{,}2 \times 10^4$	$2{,}9 \times 10^2 \pm 5{,}6 \times 10^2$

Table 8. Non-standard samples for coagulase positive *Staphylococcus* and thermotolerant coliforms, according to the parameters established by RDC - N°12, of January 2, 2001 for similar foods, distributed according to the type of establishment where they were collected.

Microorganisms	**Non-standard samples**		**Total (%)**
	Specialized	**Non-specialized**	
coagulase positive *Staphylococcus**	1 (A1)	3 (A1, A6 e A12)	4 (13.3)
thermotolerant coliforms**	4 (A1, A12, A14 e A15)	6 (A1, A2, A4, A5, A6 e A10)	10 (33.3)
Non-standard samples for both microorganisms	1 (A1)	2 (A1 e A12)	3 (10.0)
Total of non-standard samples	4 (A1, A12, A14 e A15)	7 (A1, A2, A4, A5, A6, A10 e A12)	11 (36.6)

* Value above 5.0×10^3CFU.g^{-1}. ** Value above 10^2MPN.g^{-1}