

# Assessing Brazilian food establishments' hygienic handling of leafy vegetables and their microbiological quality

M.R. Ferreira<sup>1</sup>, T.S. Santos<sup>1</sup> and D.F. Maffei<sup>1,2\*</sup> 

<sup>1</sup> Department of Agri-Food Industry, Food and Nutrition, “Luiz de Queiroz” College of Agriculture, University of Sao Paulo (ESALQ/USP), Piracicaba, SP, Brazil

<sup>2</sup> Food Research Center (FoRC-CEPID), Sao Paulo, SP, Brazil

## ORIGINAL RESEARCH PAPER

Received: August 4, 2020 • Accepted: November 19, 2020

Published online: March 22, 2021

© 2020 Akadémiai Kiadó, Budapest



### ABSTRACT

In this study, the hygienic handling of leafy vegetables was evaluated in ten selected food establishments located in south-eastern Brazil. A 14-question survey was applied in these establishments, addressing the origin of the vegetables, washing procedures, and temperature conditions during storage and exposure. In addition, 30 samples of leafy vegetables were collected and submitted to the enumeration of total coliforms, coliforms at 45 °C and tested for *Salmonella*. Based on the responses to the survey, the establishments could be clustered into three groups. Group 1 included four establishments, where the vegetables originated from farms exclusively and were displayed to consumers at room temperature, and where most of the food handlers had participated in courses on good handling practices. Group 2 included two establishments that performed a centrifugation step and where vegetables were not available to consumers in a self-service. Finally, group 3 included four establishments, where vegetables originated from suppliers exclusively, were stored under refrigeration, and displayed to consumers on refrigerated tables. The mean count of total coliforms was  $2.5 \pm 0.7$  log MPN/g, while coliforms at 45 °C were detected in only one sample (0.9 log MPN/g). *Salmonella* was not found in any sample. Microbiological results obtained for leafy vegetables from establishments clustered in the three groups did not differ significantly. These data contribute to a better understanding of the hygienic handling of leafy vegetables in the surveyed Brazilian food establishments.

\* Corresponding author. Tel.: +55 19 3447 8690. E-mail: danielmaffei@usp.br

**KEYWORDS**

food establishments, food safety, handling practices, leafy vegetables, microbiological quality

**1. INTRODUCTION**

Fresh produce is an important component of a healthy diet, since it provides many nutrients, dietary fibres and phytochemicals. Several studies have shown that the regular consumption of fruits and vegetables can help to reduce the risk of chronic diseases and some forms of cancer (Boeing et al., 2012; Dias, 2012; Angelino et al., 2019).

Over the past decades, changes in the lifestyle of the world's population, such as the heavy load of daily activities, short time for food preparation, and increased participation of the women in the labour market, have resulted in an increasing demand for away-from-home food consumption, mainly in developed countries (D'Addezio et al., 2014; Saksena et al., 2018; IBGE, 2019; Yuan et al., 2019).

In Brazil, data from the 2017–2018 Family Budget Survey, carried out by the Brazilian Institute of Geography and Statistics, showed that eating out accounted for 32.8% of all monthly expenses on food consumption during this period (IBGE, 2019). This was higher than what was observed in 2002–2003 and 2008–2009, when monthly expenses on eating out accounted for 24.1% and 31.1%, respectively. In the United States, data from the USDA's Economic Research Service indicate that, between 1987 and 2017, the percentage of income spent on foods purchased at restaurants and other food establishments showed a slight increase from 4.5 to 4.7% (Saksena et al., 2018).

The growing demand of away-from-home food consumption consequently causes an increase in the number of food establishments. These are places where food is handled, prepared, and stored, and where they may be displayed for sale and delivery or consumed on site, including restaurants, bakeries, cafeterias, snack bars, industrial kitchens, and similar places (Brazil, 2004). In Brazil, many of these establishments offer 'à la carte' and buffet meals, in addition to the typical Brazilian food called "pratos feitos" (equivalent to today's specials, although including rice, beans, salad, and some kind of meat).

All food establishments that use vegetables should adopt good practice procedures to ensure proper hygiene of the leaves and, consequently, to offer safer food to consumers. The majority of vegetables (mainly leafy greens) are often consumed raw, increasing the risk of foodborne diseases (FBD) if contaminated with pathogenic microorganisms. In fact, several worldwide FBD outbreaks have been associated with the consumption of fresh produce over the past decade (Callejón et al., 2015; Wadamori et al., 2017; Elias et al., 2018).

According to the Centers for Disease Control and Prevention, between 2014 and 2018, a total of 51 FBD outbreaks linked to leafy greens (mainly lettuce) were reported in the United States, resulting in 1,406 illnesses (CDC, 2020). In Brazil, between 2000 and 2018, nearly 13,163 FBD outbreaks were reported by the Brazilian Ministry of Health, involving 247,570 cases and 195 deaths. For most outbreaks it was not possible to identify the food implicated (45.9%). However, among those identified, 1.1% were associated with the consumption of fresh vegetables. Homes were the main site of FBD occurrence (12.5%), followed by daycare centers/schools (10.6%) and restaurants/bakeries (9.3%) (Brazil, 2019; Finger et al., 2019).

The objectives of this study were to gather information about Brazilian food establishments' hygienic handling of leafy vegetables and to evaluate whether their microbiological quality meets the current legislation.



## 2. MATERIALS AND METHODS

### 2.1. Survey on the handling practices of leafy vegetables

A total of ten food establishments, including restaurants and snack bars (identified as A–J) located in the city of Piracicaba, Sao Paulo, south-eastern Brazil, were selected for this study. A survey including 14-questions about the origin of vegetables, washing steps, temperature conditions during storage, and exposure was applied in these establishments. Additional information about the participation of food handlers in courses on good handling practices, as well as the presence of a standard operating procedure certificate for the sanitation of vegetables, was also recorded. Prior to application, the questionnaire was validated in a pilot study conducted in two establishments.

### 2.2. Sampling

A total of ten water samples (100 mL) were collected in all food establishments (one sample per establishment). These samples were collected from the disinfection tanks, before vegetables were immersed, so that the chlorine test could be performed on site. Free chlorine concentration was measured using a portable photometer (HI96771, Hanna Instruments, USA), following the manufacturer's instructions.

In addition, 30 samples of leafy vegetables (~three per establishment) were collected after washing procedures. These samples included leafy vegetables intended for main meals or snacks, all of which were prepared on the day of the visit. They were placed in plastic bags, transported to the laboratory in plastic boxes (~1 h), and immediately submitted to microbiological tests. Leafy vegetables included arugula ( $n = 4$ ), cabbage ( $n = 1$ ), chard ( $n = 1$ ), chicory ( $n = 1$ ), collard greens ( $n = 3$ ), lettuce ( $n = 16$ ), mustard greens ( $n = 1$ ), mixed leafy vegetables ( $n = 2$ ), and watercress ( $n = 1$ ).

### 2.3. Microbiological analyses

A portion of 25 g of each leafy vegetable sample was transferred to a sterile plastic bag and mixed with 225 mL of 0.1% peptone (Acumedia, Michigan, USA) in a *Stomacher* (ITR Instrumentos para Laboratórios Ltda., Esteio, RS, Brasil) for 1 min. Serial dilutions were prepared in 0.1% peptone water and submitted to enumeration of total coliforms and coliforms at 45 °C using the standard MPN method (Kornacki et al., 2015). An additional portion (25 g) of each sample was mixed with 225 mL of Buffered Peptone Water (BPW) (Kasvi, Italy) for 1 min, followed by incubation at 37 °C for 24 h for detection of *Salmonella* spp. according to the ISO 6579:2002 method (ISO, 2002).

### 2.4. Data analysis

Responses to the questionnaire were compiled to better visualise the procedures adopted in each of the ten food establishments visited. In addition, hierarchical Cluster analysis was performed to identify similarities among the practices adopted in these establishments using the XLSTAT software (version 2019.3.2).

The results of the microbiological analyses were compared to the standards established by the Brazilian Health Regulatory Agency (Anvisa), through Resolution RDC 12/2001, which states a tolerance of 2 logs for coliforms at 45 °C and absence of *Salmonella*/25 g for fresh produce (Brazil, 2001). Moreover, populations of total coliforms in leafy vegetables collected in



these food establishments were treated by One-way ANOVA to determine significant differences ( $P \leq 0.05$ ) based on the clusters obtained. The SigmaStat 4.0 software (Systat Software Inc., USA) was used for statistical analyses.

### 3. RESULTS AND DISCUSSION

The answers to the questionnaire are shown in [Chart 1](#). Of all ten food establishments, five (A, C, G, I, and J) obtained leafy vegetables from farms and the other five (B, D, E, F, and H) from suppliers. The majority reported obtaining vegetables from conventional farming (A, B, C, D, E, H, and J), followed by organic (F and I) and hydroponic farming systems (G). None of the establishments reported obtaining minimally processed fresh-cut-vegetables. After receiving the raw material, five establishments (A, B, C, F, and J) were found to store them in a refrigerator ( $<10\text{ }^{\circ}\text{C}$ ), while the other five (D, E, G, H, and I) kept the vegetables at room temperature ( $\sim 25\text{ }^{\circ}\text{C}$ ) until they were used as main meal and/or snacks.

Pre-washing in running tap water was performed by eight establishments (A, C, D, E, G, H, I, and J). Washing-disinfection was carried out in all ten food establishments through immersion of vegetables in tanks with water containing disinfectant agents under static conditions. Most of the establishments used sodium hypochlorite: bleach (C, E, F, and H, concentration ranging from 34 to 79  $\text{mg L}^{-1}$ ) or chlorine-based commercial products (B, D, and I, concentration ranging from 5 to 42  $\text{mg L}^{-1}$ ). The other three establishments (A, G, and J) used sodium dichloroisocyanurate (concentration ranging from 7 to 99  $\text{mg L}^{-1}$ ). Among these establishments, eight (A, B, C, E, F, H, I, and J) performed rinsing and two of them (C and H) centrifuged vegetables using a salad spinner.

Appropriate washing is one of the most important procedures, aiming to remove dirt and debris and to reduce the microbial load of these products. Furthermore, the addition of sanitisers to the washing water is important to prevent transfer and survival of pathogenic microorganisms during washing (Gil et al., 2015; Jung et al., 2017). In Brazil, the CVC Ordinance no. 05/2013 oversees the technical regulation on good practices for commercial food establishments and food services in the state of Sao Paulo. According to this Ordinance, the washing of fresh produce must be performed using appropriate disinfectants for this purpose and the use of sodium hypochlorite at 200–250  $\text{mg L}^{-1}$  is recommended (Sao Paulo, 2013).

Although washing-disinfection was observed in all ten food establishments visited in the present study, the chlorine concentration was below this recommendation in all of them. The low concentration of sanitiser in the washing water is a concern, since its quality deteriorates rapidly, when organic matter from the soil, leaves, and debris are present in addition to the microbial load associated with produce (Gil et al., 2009). Furthermore, only four establishments (A, C, E, and J) displayed the Standard Operating Procedure (SOP) certificate with instructions for the sanitation of vegetables, as required by the CVC Ordinance no. 05/2013.

Although the use of chlorine is recommended for Brazilian processors to disinfect fresh produce, commercial chlorine-based products are often available for consumers in supermarkets and grocery stores, with information on their labels about how to prepare the sanitising solution at home. In contrast to other countries, the use of sanitisers by consumers is not recommended. The U.S. Food and Drug Administration does not recommend the use of commercial sanitiser products to wash fruit and vegetables (FDA, 2018). Similarly, the use of potable water instead of water with chemical disinfection agents is advocated by some European countries (Gil et al., 2009).



Questions and responses	Food establishment									
	A	B	C	D	E	F	G	H	I	J
1) What is the origin of the vegetables served in this establishment?										
Farms (n=5)										
Suppliers (n=5)										
2) Is there information on whether they are vegetables from conventional, organic, or hydroponic cultivation?										
Conventional (n=7)										
Organic (n=2)										
Hydroponic (n=1)										
3) Does the establishment receive fresh or minimally processed fresh-cut-vegetables?										
Fresh vegetables (n=10)										
Minimally processed fresh-cut-vegetables (n=0)										
4) Under what temperature conditions are vegetables stored?										
Refrigerator (n=5)										
Room temperature (n=5)										
5) In what meals are vegetables served?										
Main meal (n=5)										
Main meal and snacks (n=5)										
6) Is the pre-washing step of vegetables performed in the establishment?										
Yes (n=8)										
No (n=2)										
7) Is the washing-disinfection step of vegetables performed in the establishment?										
Yes (n=10)										
No (n=0)										
8) What is used for disinfection?										
Bleach (n=4)										
Chlorine-based commercial products (n=3)										
Other (n=3)										

Chart 1. Responses to the questionnaire applied in the ten food establishments



9) Is the rinsing step of vegetables performed after disinfection?										
Yes (n=8)	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>
No (n=2)	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
10) Is the centrifugation step performed after rinsing?										
Yes (n=2)	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
No (n=8)	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>
11) Under what temperature conditions are sanitised vegetables stored?										
Refrigerator (n=7)	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>
Room temperature (n=3)	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>
12) Under what temperature conditions are vegetables served to consumers?										
Refrigerated table (n=4)	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
Room temperature (n=4)	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>
n.a. (n=2)	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>
13) Is there a Standard Operating Procedure certificate regarding sanitation of vegetables displayed in this establishment?										
Yes (n=4)	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>
No (n=6)	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>
14) Do food handlers in this establishment have a training course on good handling practices?										
Yes (n=3)	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>
No (n=7)	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>	<input checked="" type="checkbox"/>	<input type="checkbox"/>

Shaded and unshaded boxes mean selected and non-selected replies, respectively.  
 n.a: not applicable (step not performed in the food establishment).

Chart 1. Continued

Concurrently, alternative methods for disinfection of fresh produce have been considered over the past decades, such as the use of chlorine dioxide, ozone, hydrogen peroxide, calcium-based solutions, electrolysed water, organic acids, irradiation, filtration, ultrasounds, ultraviolet light, and cold plasma among others (Meireles et al., 2016; De Corato, 2019). This highlights the demand for further studies on control measures to reduce microbial risks associated with fresh produce and to contribute to the harmonisation of the processing of these products.

In all establishments, washed vegetables were kept in clean plastic containers. The storage was predominantly carried out in domestic refrigerators (<10 °C) by seven establishments (A, B, C, D, F, G, and H), whereas the other three (E, I, and J) kept them at room temperature (~25 °C). Among those establishments, in which vegetables were available as self-service meals (A, B, D, E, F, G, I, and J), only four (D, E, F, and G) included a refrigerated table (<10 °C) to display them.



Only three establishments (G, I, and J) reported the participation of food handlers in courses on good handling practices. Mishandling and disregard of hygienic measures by food handlers may enable pathogenic bacteria to come into contact with food and, in some cases, survive and multiply in sufficient numbers to make consumers sick (Kibret and Abera, 2012). In Brazil, food establishments (e.g. restaurants and bakeries) represented 9.3% of the foodborne outbreak sites reported between 2000 and 2018 (Brazil, 2019; Finger et al., 2019). Thus, food handlers play an essential role in ensuring food safety throughout the entire food chain.

Aiming to identify similarities among the practices observed in the ten food establishments, hierarchical cluster analysis, represented through a dendrogram, resulted in three major groups (Fig. 1): group 1 (establishments A, G, I, and J), group 2 (establishments C and H), and group 3 (establishments B, D, E, and F). The main similarities in the food establishments of group 1 were the origin of vegetables (all of them from farms) and the display of vegetables to consumers at room temperature (A, I, and J). In addition, it was the only group that included food handlers participating in courses on good handling practices (J, G, and I).

The establishments clustered in group 2 were the only ones that performed a centrifugation step after rinsing vegetables. Furthermore, in these establishments, the vegetables were not made available to consumers in a self-service, but rather used as snacks or a set meal. Regarding the establishments clustered in group 3, the main similarities were the origin of vegetables (all of them from suppliers), the storage of vegetables under refrigeration after washing procedures (B, D, and F), and the display of vegetables to consumers on refrigerated tables (D, E, and F).

Microbiological results of leafy vegetables are shown in Table 1. All samples were positive for total coliforms (mean of  $2.5 \pm 0.7$  log MPN/g), while coliforms at 45 °C were detected in only one sample (collard greens) from establishment A (0.9 log MPN/g), but not exceeded the limit of 2 logs established by the Brazilian Health Regulatory Agency, through resolution RDC 12/2001 (Brazil, 2001). Moreover, none of the samples was positive for *Salmonella* spp. in 25 g, which is also in agreement with this resolution.

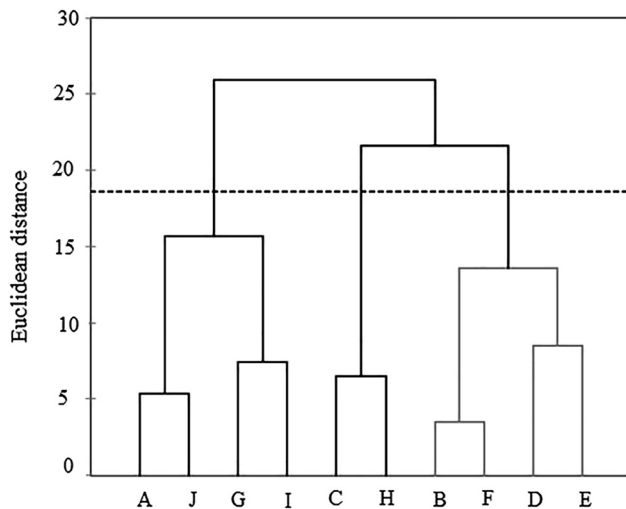


Fig. 1. Cluster dendrogram for the practices recorded in the ten food establishments identified as A–J



Table 1. Numbers of total coliforms and coliforms at 45 °C in the leafy vegetable samples regarding food establishments (A–J)

Food establishment	Number of samples	Total coliforms	Coliforms at 45 °C
A	3	3.0	0.9
B	3	2.7 ± 0.6	<0.5*
C	2	1.8 ± 0.3	<0.5*
D	2	2.9 ± 0.3	<0.5*
E	4	1.4	<0.5*
F	3	2.8 ± 0.4	<0.5*
G	4	2.3 ± 1.2	<0.5*
H	3	2.5 ± 0.3	<0.5*
I	3	2.8 ± 0.4	<0.5*
J	3	2.8 ± 0.4	<0.5*
Mean		2.5 ± 0.7	0.9

Values expressed as mean ± SD (log MPN/g).

\*: Results below the limit of quantification.

The three groups in which the food establishments were clustered were also compared based on the total coliform counts. The results were as follows: group 1 ( $2.7 \pm 0.7$  log MPN/g), group 2 ( $2.2 \pm 0.5$  log MPN/g), and group 3 ( $2.5 \pm 0.7$  log MPN/g). These results did not differ significantly ( $P > 0.05$ ). This shows that the differences in handling practices observed in these groups did not impact the microbial load of leafy vegetables.

## 4. CONCLUSIONS

Certain practices observed in different food establishments go against what is appropriate and recommended by Brazilian authorities, such as: storage of vegetables without refrigeration, low chlorine concentration and no rinsing step, absence of the SOP certificate regarding sanitation of vegetables, and low participation of food handlers in courses on good handling practices. Nonetheless, the leafy vegetable samples collected in these establishments showed satisfactory microbiological quality, in agreement with the microbiological standards established in the current Brazilian legislation. Although the ten food establishments were clustered into three groups, based on the handling practices observed, they had no impact on the microbial load of leafy vegetables. Overall, these data can contribute to a better understanding of the surveyed Brazilian food establishments' hygienic handling of leafy vegetables. This is important to support the government and food services in the implementation of proper control measures to ensure the safety of these foods.

## ACKNOWLEDGEMENT

The authors would like to acknowledge the Sao Paulo Research Foundation (FAPESP, Brazil) through grant #2013/07914-8 and the National Council for Scientific and Technological Development (CNPq, Brazil) through grant #434469/2018-1.



## REFERENCES

- Angelino, D., Godos, J., Ghelfi, F., Tieri, M., Titta, L., Lafranconi, A., Marventano, S., Alonzo, E., Gambera, A., Sciacca, S., Buscemi, S., Ray, S., Galvano, F., Del Rio, D., and Grosso, G. (2019). Fruit and vegetable consumption and health outcomes: an umbrella review of observational studies. *International Journal of Food Sciences and Nutrition*, 70: 652–667.
- Boeing, H., Bechthold, A., Bub, A., Ellinger, S., Haller, D., Kroke, A., Leschik-Bonnet, E., Müller, M.J., Oberritter, H., Schulze, M., Stehle, P., and Watzl, B. (2012). Critical review: vegetables and fruit in the prevention of chronic diseases. *European Journal of Nutrition*, 51: 637–663.
- Brazil (2001). Brazilian Ministry of Health. Resolução RDC n. 12 de 02 de janeiro de 2001. Aprova o regulamento técnico sobre padrões microbiológicos para alimentos. Agência Nacional de Vigilância Sanitária. Available at: [http://bvsmms.saude.gov.br/bvs/saudelegis/anvisa/2001/res0012\\_02\\_01\\_2001.html](http://bvsmms.saude.gov.br/bvs/saudelegis/anvisa/2001/res0012_02_01_2001.html) (accessed on 10 July 2020).
- Brazil (2004). Brazilian Ministry of Health. Resolução RDC n. 216 de 15 de setembro de 2004. Dispõe sobre regulamento técnico de boas práticas para serviços de alimentação. Agência Nacional de Vigilância Sanitária. Available at: [http://bvsmms.saude.gov.br/bvs/saudelegis/anvisa/2004/res0216\\_15\\_09\\_2004.html](http://bvsmms.saude.gov.br/bvs/saudelegis/anvisa/2004/res0216_15_09_2004.html) (accessed on 10 July 2020).
- Brazil (2019). Brazilian Ministry of Health. Doenças Transmitidas por Alimentos: causas, sintomas, tratamento e prevenção. Brasília, DF. Available at: <http://portalms.saude.gov.br/saude-de-a-z/doencas-transmitidas-por-alimentos> (accessed on 27 May 2019).
- Callejón, R.M., Rodríguez-Naranjo, M.I., Ubeda, C., Hornedo-Ortega, R., Garcia-Parrilla, M.C., and Troncoso, A.M. (2015). Reported foodborne outbreaks due to fresh produce in the United States and European Union: trends and causes. *Foodborne Pathogens and Disease*, 12: 32–38.
- CDC. (2020). *Lettuce, other leafy greens, and food safety*. Centers for Disease Control and Prevention. Available at: <https://www.cdc.gov/foodsafety/communication/leafy-greens.html#:~:text=During%202014%20to%202018%2C%2051,were%20linked%20to%20romaine%20lettuce> (accessed on 10 November 2020).
- D'Addezio, L., Turrini, A., Capacci, S., and Saba, A. (2014). Out-of-home eating frequency, causal attribution of obesity and support to healthy eating policies from a cross-European survey. *Epidemiology, Biostatistics, and Public Health*, 11: 1–13.
- De Corato, U. (2019). Improving the shelf-life and quality of fresh and minimally-processed fruits and vegetables for a modern food industry: a comprehensive critical review from the traditional technologies into the most promising advancements. *Critical Reviews in Food Science and Nutrition*, 7: 1–36.
- Dias, J.S. (2012). Nutritional quality and health benefits of vegetables: a review. *Food and Nutrition Sciences*, 3: 1354–1374.
- Elias, S.O., Decol, L.T., and Tondo, E.C. (2018). Foodborne outbreaks in Brazil associated with fruits and vegetables: 2008 through 2014. *Food Quality and Safety*, 2: 173–181.
- Finger, J.A.F.F., Baroni, W.S.G.V., Maffei, D.F., Bastos, D.H.M., and Pinto, U.M. (2019). Overview of foodborne disease outbreaks in Brazil from 2000 to 2018. *Foods*, 8: 434.
- FDA (2018). *Raw produce: Selecting and serving it safely*. Food facts. Available at: <https://www.fda.gov/media/77178/download> (accessed on 10 November 2020).
- Gil, M.I., Selma, M.V., López-Gálvez, F., and Allende, A. (2009). Fresh-cut product sanitation and wash water disinfection: problems and solutions. *International Journal of Food Microbiology*, 134: 37–45.



- Gil, M.I., Selma, M.V., Suslow, T., Jacxsens, L., Uyttendaele, M., and Allende, A. (2015). Pre-and post-harvest preventive measures and intervention strategies to control microbial food safety hazards of fresh leafy vegetables. *Critical Reviews in Food Science and Nutrition*, 55: 453–468.
- IBGE (2019). Pesquisa de orçamentos familiares 2017–2018: primeiros resultados. (Family Budget Survey 2017–2018: First Results). Rio de Janeiro, Instituto Brasileiro de Geografia e Estatística – IBGE, pp. 69. Available at: <https://biblioteca.ibge.gov.br/visualizacao/livros/liv101670.pdf>. (accessed 23 March 2020).
- ISO (2002) *Microbiology of food and animal feeding stuffs – Horizontal method for the detection of Salmonella spp.* 4th ed. The International Organization for Standardization, ISO 6579 (2002), Amendment 1: 15/07/2007.
- Jung, Y., Jang, H., Guo, M., Gao, J., and Matthews, K.R. (2017). Sanitizer efficacy in preventing cross-contamination of heads of lettuce during retail crisping. *Food Microbiology*, 64: 179–185.
- Kibret, M. and Abera, B. (2012). The sanitary conditions of food service establishments and food safety knowledge and practices of food handlers in Bahir Dar Town. *Ethiopian Journal of Health Sciences*, 22: 27–35.
- Kornacki, J.L., Gurtler, J.B., and Stawick, B.A. (2015). *Enterobacteriaceae*, coliforms, and *Escherichia coli* as quality and safety indicators. In: Salfinger, Y. and Tortorello, M.L. (Eds.), *Compendium of methods for the microbiological examination of foods*, 5th ed. American Public Health Association, Washington, D.C.
- Meireles, A., Giaouris, E., and Simões, M. (2016). Alternative disinfection methods to chlorine for use in the fresh-cut industry. *Food Research International*, 82: 71–85.
- Saksena, M.J., Okrent, A.M., Anekwe, T.D., Cho, C., Dicken, C., Effland, A., Elitzak, H., Guthrie, J., Hamrick, K., Hyman, J., Jo, Y., Lin, B.-H., Mancino, L., McLaughlin, D., Rahkovsky, I., Ralston, K., Smith, T.A., Stewart, H., and Tuttle, C. (2018). *America's eating habits: Food away from home*, EIB-196. Saksena, M., Okrent, A.M. and Hamrick, K.S. (Eds.) U.S. Department of Agriculture, Economic Research Service, 172 pages.
- Sao Paulo (2013). Centro de Vigilância Sanitária. Secretaria de Estado da Saúde. Portaria CVS 5, de 09 de abril de 2013. Aprova o regulamento técnico sobre boas práticas para estabelecimentos comerciais de alimentos e para serviços de alimentação, e o roteiro de inspeção, anexo. Available at: [http://www.cvs.saude.sp.gov.br/up/PORTARIA%20CVS-5\\_090413.pdf](http://www.cvs.saude.sp.gov.br/up/PORTARIA%20CVS-5_090413.pdf) (accessed on 10 November 2020).
- Wadamori, Y., Gooneratne, R., and Hussain, M.A. (2017). Outbreaks and factors influencing microbiological contamination of fresh produce. *Journal of the Science of Food and Agriculture*, 97: 396–1403.
- Yuan, M., Seale, J., Jr., Wahl, T., and Bai, J. (2019). The changing dietary patterns and health issues in China. *China Agricultural Economic Review*, 11: 143–159.

