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MICROBIAL QUALITY AND LABELING OF MINIMALLY PROCESSED FRUITS AND VEGETABLES

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Abstract
The consumption of minimally processed, or ready-to-eat (RTE), fruits and vegetables has been growing because people have shorter time to eat and due to the pursuit of convenience and health benefits. Minimal processing includes raw material selection and sorting, pre-washing, debarking, cutting, slicing, sanitizing, rinsing, centrifuging, and packing. Thus, natural protection barriers are affected during cutting methods and they release nutrients that enable microorganism growth. The aim of the current study was to evaluate the levels of aerobic mesophilic bacteria, molds and yeasts, coliforms at 35°C, *Escherichia coli* and *Salmonella* sp., as well as the labeling adequacy of minimally processed fruits and vegetables traded in Vitória, Espírito Santo, Brazil. Fruits and vegetables presented aerobic mesophilic bacteria, molds and yeasts, and coliforms at 35°C, above the safe limit for consumption purposes. The count of aerobic mesophilic bacteria in RTE fruits ranged from 4.00 to 6.30 Log CFU/g. The highest count of this microorganism group was recorded for fruit salads. *Salmonella* sp. was not identified in fruit or vegetable samples, whereas *Escherichia coli* was detected in four vegetable samples. None of the evaluated labels (n = 40) followed the Brazilian legislation. Minimally processed fruits and vegetables had poor microbiological quality and labeling was also unsatisfactory. The adoption of good manufacturing practices and quality control tools as strategies to produce safe food can help minimizing risks to consumers' health.

Keywords: *Escherichia coli*. Food Safety. Hygiene. Quality Control. *Salmonella* sp.

1. Introduction

The consumption of minimally processed, or ready-to-eat (RTE), fruits and vegetables has been increasing because people have shorter time to eat and due to the pursuit of convenience and health benefits. Minimally processed food associate's healthy food features with ready-to-eat products (Putnik et al. 2017; Cole and Singh 2018; Schuh et al. 2020). Besides that, this sector has presented important progress because of the similar attributes of RTE to those *in natura* (Schuh et al. 2020).

Minimal processing includes raw material selection and sorting, pre-washing, debarking, cutting, slicing, sanitizing, rinsing, centrifuging, and packing (Santos et al. 2010; Schuh et al. 2020). Fruits and vegetables have metabolically active plant tissues; therefore, they are more susceptible to contamination resulting from food stress during processing. For example, natural protection barriers are affected during cutting procedures and can increase respiratory activity and the release of ethylene, what can culminate in sensorial changes. Furthermore, the cutting might release nutrients that enable microorganism growth (Ranjitha et al. 2018; Schuh et al. 2020).

Data collected from 2008 to 2014 have shown that outbreaks associated with fruit and vegetable intake resulted in 2,926 disease cases and led to 347 hospitalizations in Brazil (Elias et al. 2018). *Salmonella* sp. and some *Escherichia coli* serotypes are the main pathogenic microorganisms associated with the contamination of fresh vegetables. These microorganisms play a significant role in several foodborne outbreaks recorded worldwide (Gurler et al. 2015). The microbiological safety of the aforementioned product type depends on the sanitization stage, which helps reducing the deteriorating microbiota, as well as eliminating pathogenic microorganisms. These products are often consumed without any subsequent preparation; thus, they may pose risks to consumers' health (Gurler et al. 2015). Therefore, extreme care must be taken from production to trading to reduce health-related risks (Jeddi et al. 2014). Another critical aspect that can help minimizing consumer risks is related to information available in the label of RTE products. These labels play the role of communication link between producers and consumers. Data made available in them should follow the current legislation and enable consumers to have easy access to product information such as ideal storage temperature and expiration date, as well as to information about producers (Prado et al. 2008). The microbiological quality of RTE food has been investigated in Brazilian states such as São Paulo, Ceará and Minas Gerais (Rosa 2002; Bruno et al. 2005; Pinheiro et al. 2005; Prado et al. 2008; Smanioto et al. 2009; Santos et al. 2010; Oliveira et al. 2011; Sant'ana et al. 2011; Maistro et al. 2012; Ferreira et al. 2016).

However, information on the microbiological quality and proper labeling of RTE products traded in Espírito Santo State remain incipient. Given the importance of such knowledge, the aim of the current study was to analyze the microbiological quality, as well as the proper labeling of minimally processed fruits and vegetables traded in Vitória, Espírito Santo State, Brazil.

2. Material and Methods

In total, 40 samples (10 fruits and 30 vegetables) of minimally processed products were purchased in four supermarkets in Vitória, Espírito Santo, Brazil. The selection was made according to product availability in the establishments (Table 1).

Table 1. Description of 40 samples of minimally processed food traded in Vitória, Espírito Santo, Brazil.

Sample	Scientific name	N
Fruits		10
Fruit salad*	-	2
Mango	*Mangifera indica*	3
Melon	*Cucumis melo*	1
Papaya	*Carica papaya*	3
Passion fruit	*Passiflora edulis*	1
Vegetables		30
Baby soup*	-	2
Barred potato	*Arracacia xanthorrhiza* Banc.	3
Sugar – Beet	*Beta vulgaris* L.	2
Cabbage	*Brassica oleracea* L. var. *capitata*	2
Cabbage and broccoli	*Brassica oleracea* L. var. *acephala* D.C. and *Brassica oleracea* L. var. *italica Plenck*	1
Carrot	*Daucus carota* L.	2
Carrot and sugar-beet	*Daucus carota* L. and *Beta vulgaris* L.	1
Carrot and chayote	*Daucus carota* L. and *Sechium edule* Sw.	1
Cauliflower	*Brassica oleracea* var. *botrytis*	1
Cauliflower and broccoli	*Brassica oleracea* var. *botritys* and *Brassica oleracea* L. var. *italica Plenck*	1
Mixed-leaf salad*	-	2
Potato and carrot	*Solanum tuberosum* and *Daucus carota* L.	2
Pumpkin	*Cucurbita moschata* Duch	4
Vegetables for mayonnaise salad*	-	1
Vegetables for yakisoba*	-	2
Yam	*Colocasia esculenta* (L.) Schott	3

* The composition of mixed samples is described in Tables 2 and 3.

All samples were transported in isothermal boxes, stored in refrigerators, and analyzed within 24 hours at the Microbiology Laboratory of the Pharmaceutical Sciences Department, at Federal University of Espírito Santo.

Microbiological analysis

This procedure was carried out in compliance with the American Public Health Association (APHA), as described in the Compendium of Methods for the Microbiological Examination of Foods (Downes and Ito 2001). First, 225 mL of previously sterilized peptone water (0.1%) was added to 25 g of each sample. Homogenization was performed in a stomacher (Seward Medical Co., London, United Kingdom), for 5 minutes. Next, serial dilutions were performed, and aliquots were transferred to culture media specific for each microbial group. Plating was done in duplicate and results were expressed in colony forming units per gram (CFU/g).

Aerobic mesophiles were determined based on the pour plate technique with standard plate count agar (PCA, Himedia®) on incubated plates inverted at 35 ± 1°C, for 48 h by using 1 mL of the previously prepared dilutions (Downes and Ito 2001). The spread plate technique was used to determine the incidence of molds and yeasts by inoculating 0.1 mL of the dilutions on the surface of Sabouraud Dextrose Agar (SDA, Kasvi). Plates were incubated without inversion at 25 ± 1°C, for 5 to 7 days.

Petrifilm® plates (3M Company, St. Paul, MN, USA) were used to count coliforms at 35°C and *Escherichia coli* (Downes and Ito 2001). Plates were incubated at 35 ± 1°C, for 48 h. Blue colonies with gas were classified as *E. coli,* whereas red and blue colonies with gas were classified as coliforms at 35°C.

To analyze *Salmonella* sp. were carried out according to the IN 62 methods (Brasil 2003). A pre-enriched was performed with 25 g of samples in 225 mL of peptone water (1 %). After 24 incubation hours at 36 ± 1°C, aliquots of this pre-enriched step were transferred to selective enrichment broths, Rappaport Vassiliadis (Acumedia®) and Selenite-cystine (Acumedia®). Sterile loops were streaked were used to carrying inoculums of these broths were transferred to plates filled with selective culture media for *Salmonella* sp. (Hektoen, *Salmonella Shigella*, and Brilliant Green). Two plates of each culture medium were obtained, one derived from the Rappaport Vassiliadis broth and the other from the Selenite-cystine broth. Inverted plates were incubated at 36 ± 1°C, for 18 to 24 h. Next, 3 to 5 typical colonies per sample were selected. Selected colonies were picked in non-selective agar and incubated at 36 ± 1°C, for 18 to 24 h, to check their purity in biochemical tests. Suspected colonies per sample were transferred to Triple Sugar Iron Agar (TSI), Lysine Iron Agar (LIA), urea agar, SIM medium and oxidase test. The positive result for *Salmonella* was based on cultures presenting typical reactions in biochemical tests and positive serological response to the polyvalent antiserum O. Results were expressed as presence or absence of *Salmonella* sp. in 25 g (Brasil 2001).

Labeling adequacy

The analysis of information available on the packaging was based on Brazilian RDC resolutions n. 259/2002 (Brasil 2002), n. 359/2003 (Brasil 2003c) and n. 360/2003 (Brasil 2003d), issued by the Brazilian Health Regulatory Agency (ANVISA), as well as on Law n. 10.674 (Brasil 2003a). The following information described as mandatory by ANVISA was analyzed: sales denomination, ingredients, net content, origin and lot, expiration date, storage mode, nutritional information, and information about the supplier such as phone number, website, and e-mail, in addition to terms such as "contains gluten" or "gluten-free".

Data analysis

Microbiological analyses results were compared to the Brazilian national standard for fruits and vegetables set by RDC n. 12/2001 (Brasil 2001). The value established for the indicative sample was taken into consideration for comparison purposes. Data about microorganisms whose standard is not stipulated in the Brazilian legislation were compared to data available in the literature.

Data available in the labels were tabulated in Microsoft Excel® spreadsheet and subjected to descriptive analysis.

3. Results and Discussion

Microbial quality of RTE products

Minimally processed fruits presented more than 40% of inadequacy, according to the Brazilian legislation and Santos et al. (2010), Arruda et al. (2004) and Rosa et al. (2002) (Figure 1). Food safety is especially important factors related to food quality. The microbiological parameters can help to verify the hygienic-sanitary conditions in food handling process (Schuh et al. 2020). For minimally processed vegetables, the level of inadequacy found was higher than 80% for aerobic mesophilic bacteria, molds and yeasts, and coliforms at 35°C. Molds and yeasts presented 96.7% of inadequacy compared to the references considered in the present study. In the study conducted by Schuh et al. (2020), were observed similar results and a significant contamination related to mesophilic aerobic, and molds and yeasts.

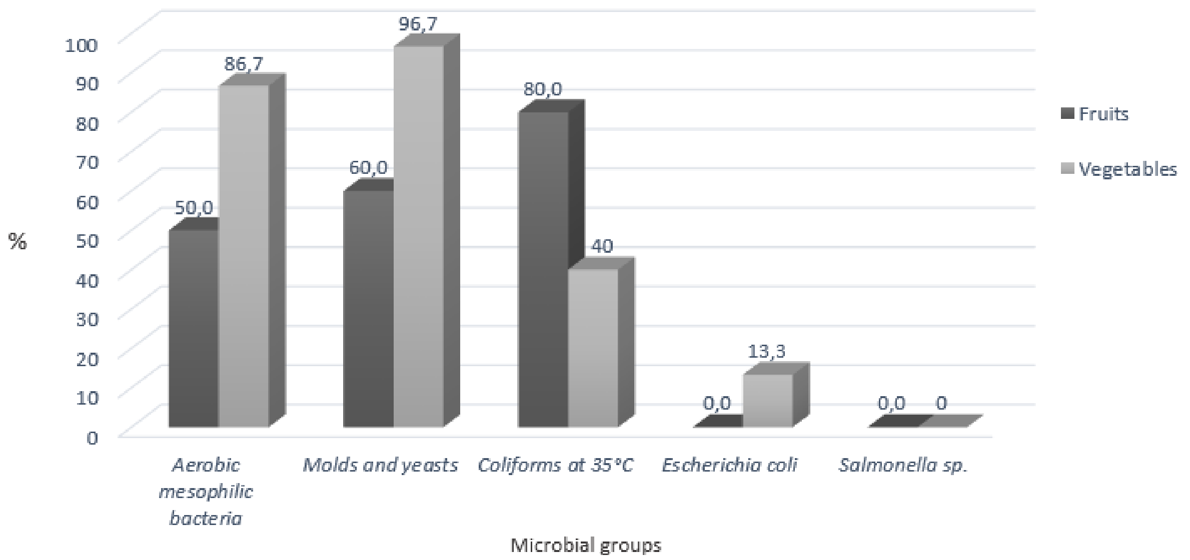

Figure 1. Summary of microbiological inadequacy (%) in minimally processed products traded in supermarkets in Vitória, Espírito Santo, Brazil - according to the Brazilian legislation for *Salmonella* sp. and *Escherichia coli*, and to Santos et al. (2010), Arruda et al. (2004) and Rosa et al. (2002), who investigated aerobic mesophilic bacteria, molds and yeasts, and coliforms at 35°C.

RTE fruits

The count of aerobic mesophilic bacteria in RTE fruits ranged from 4.00 to 6.30 Log CFU/g. The highest count of this microorganism group was recorded for fruit salads (Table 2). These results may be associated with the inclusion of different fruits in the analyzed product and with significant handling during preparation. Furthermore, it is important to ensure an efficiency of washing and sanitizing processes because they are related to the microbiological quality of minimally processed vegetables and fruits (Lepaus et al. 2020; Schuh et al. 2020).

The Brazilian legislation (Brasil 2001) does not set limits for aerobic mesophilic bacteria. Although tolerable values are not described for this microbial group, it is essential evaluating it. These microorganisms are seen as general indicators of contamination; thus, they allow evaluating the microbiological quality of food products based on their shelf life.

The French and German governments have set the limit of 7.70 log CFU/g for aerobic mesophilic bacteria in fresh vegetables (Legnani and Leoni 2004). According to this criterion, all RTE fruit samples meet the recommendations in France and Germany; however, according to Arruda et al. (2004) food products presenting mesophilic bacteria higher than 5 log CFU/g are overall unfit for human consumption due to loss of nutritional value, organoleptic changes, deterioration risk and to the possible incidence of pathogens. Schuh et al. (2020) points out that counts between 3.4×10^4 and 4.9×10^5 CFU/g for minimally processed vegetables indicated that they were more susceptible to deterioration, which reduce the shelf life. Consequently, in present study, 50% (n = 5) of fruit samples are unfit for consumption.

Table 2. Total counts (log CFU/g) of aerobic mesophilic bacteria, molds and yeasts, coliforms at 35°C, *E. coli* and *Salmonella* sp. in minimally processed fruit samples traded in supermarkets in Vitória, Espírito Santo, Brazil.

Fruit sample	Aerobic mesophilic bacteria	Molds and yeasts	Coliforms at 35°C	*E. coli*	*Salmonella* sp.
Papaya 1	5.20	3.30	4.65	ND	Absent
Papaya 2	5.45	5.20	5.10	ND	Absent
Papaya 3	4.71	4.85	3.90	ND	Absent
Mango 1	4.40	3.50	5.10	ND	Absent
Mango 2	4.00	4.25	4.10	ND	Absent
Mango 3	4.20	4.40	4.00	ND	Absent
Passion fruit	5.15	2.40	5.10	ND	Absent
Melon	4.25	3.00	5.15	ND	Absent
Fruit salad 1[a]	6.30	5.30	4.70	ND	Absent
Fruit salad 2[a]	5.50	5.10	4.30	ND	Absent

ND: Not detected at the lowest plated dilution; [a]: mango, grapefruit, papaya and apple.

Minimally processed food products present reduced shelf life due to the processing stages. Thus, it is necessary controlling the growth of aerobic mesophilic bacteria. Moreover, inadequate storage conditions contribute to the high counts observed in these products. Several foodborne pathogenic bacteria are mesophilic; therefore, high counts of this pathogenic group can predict favorable conditions for bacterial growth, which poses risks to consumers' health (Maistro et al. 2012).

The mean count recorded for molds and yeasts was 4.13 log CFU/g; the highest value (5.30 log CFU/g) was recorded for fruit salad samples, whereas the lowest one (2.40 log CFU/g) was found in passion fruit samples. The present study recorded 5.10 log CFU/g and 5.30 log CFU/g molds and yeast, respectively, for fruit salad samples. Research carried out in Fortaleza, Ceará, Brazil recorded similar value (5.25 log CFU/g) for the same microbial group in two fruit salad samples (Bruno et al. 2005). Furthermore, they found 6.40 log CFU/g count in a melon sample, which was higher than the value recorded in the present study (3.00 log CFU/g). Papaya samples presented similar values, which varied from 4.10 to 5.50 log CFU/g.

Coliform counts at 35°C ranged from 3.90 to 5.15 log CFU/g. Although there is no legal standard set to analyze coliforms at 35°C in fruits, the incidence of high coliform levels indicates unsatisfactory hygiene conditions during the production and handling processes. In other words, there may be inefficiency in the application of Good Manufacturing Practices.

The Brazilian legislation (Brasil 2001) sets the maximum limit of 2.6 log CFU/g for thermotolerant coliforms. The present study has only investigated *Escherichia coli*, which is the main pathogen belonging to this group, which, in its turn, was analyzed because it comprised microorganisms that indicate fecal contamination. Fecal contamination indicates the possible incidence of enteric pathogens in the food, fact that renders the product unfit for consumption. Some *Escherichia coli* serotypes can damage human health and lead to diseases such as gastroenteritis, urinary tract infection and cystitis (Kamala and Kumar 2018).

None of the RTE fruit samples showed *E. coli* and *Salmonella* sp. The Brazilian legislation (Brasil 2001) advocates the absence of *Salmonella* sp. in fresh fruits; according to this criterion, the herein analyzed samples were suitable for consumption. *Salmonella* sp. causes severe diseases in humans due to contaminated food intake. Although the natural habitat of this bacterium is the intestinal region of humans and animals, it can also be detected in the air, water, feces, and food (Shinohara et al. 2008). Smanioto et al. (2009) have found that all minimally processed fruit samples analyzed in their study followed the current Brazilian legislation. Based on Bruno et al. (2005), all RTE fruit samples presented counts of thermotolerant coliforms in compliance with the Brazilian legislation. However, 26.6% (n = 4) of the samples were unfit for consumption due to incidence of *Salmonella* sp. Pinheiro et al. (2005) have also found *Salmonella* spp. in 25% (n = 5) of the minimally processed fruit samples (guava, mango, melon, papaya, and pineapple) traded in Fortaleza, Ceará, Brazil.

Contamination with *Salmonella* sp. may happen during improper fruit processing and storage processes. The surface of fruits, water, equipment and utensils, the adopted packaging, and the handlers themselves can be contamination sources (Olaimat and Holley 2012; Kamala and Kumar, 2018). Also, it is necessary to highlight that contamination with *Salmonella* can occur by animal manure as a fertilizer, water utilized for irrigation and animals that cross vegetable fields (Cruz et al. 2019). In this context, it is essential the attention to the Good Agricultural Practices and Good Manufacturing Practices. Procedures should be established for an adequate sanitization process and conducted carefully the following steps avoid cross-contamination. The present study suggests that the processing and storage procedures adopted for minimally processed fruits traded in Espírito Santo State were sufficient to avoid product contamination with *Salmonella* sp.

RTE vegetables

With respect to RTE vegetables, 86.7% (n = 26) of the analyzed samples presented aerobic mesophilic bacteria count higher than 5 log CFU/g (Table 3). Based on recommendations by Arruda et al. (2004), these samples were unfit for human consumption.

Table 3. Total counts (log CFU/g) of aerobic mesophilic bacteria, molds and yeasts, coliforms at 35°C, *E. coli* and *Salmonella* sp. in minimally processed vegetable samples traded in supermarkets in Vitória, Espírito Santo, Brazil.

Vegetable sample	Aerobic mesophilic bacteria	Molds and yeasts	Coliforms at 35°C	*E. coli*	*Salmonella* sp.
Baby soup [a]	6.40	>6.3	>6.3	3.05	Absent
Baby soup [a]	6.20	5.90	5.10	ND	Absent
Barred potato 1	>6.3	>6.3	>6.3	ND	Absent
Barred potato 2	5.85	>6.3	5.35	3.20	Absent
Barred potato 3	5.35	4.25	4.95	ND	Absent
Beet	4.60	6.00	5.30	ND	Absent
Beet (grated)	4.55	4.95	4.90	ND	Absent
Cabbage 1	6.40	5.40	4.05	ND	Absent
Cabbage 2	6.30	6.20	5.30	ND	Absent
Cabbage and broccoli	5.20	5.30	5.15	ND	Absent
Carrot (cubes)	>6.3	>6.3	5.35	ND	Absent
Carrot (grated)	5.45	6.20	5.80	ND	Absent
Carrot and beet (grated)	5.40	6.20	>6.3	ND	Absent
Carrot and chayote (cubes)	5.20	>6.3	5.30	3.65	Absent
Cauliflower	6.10	3.40	>6.3	ND	Absent
Cauliflower and brocoli	5.35	4.90	5.25	ND	Absent
Mixed-leaf salad [b]	6.10	4.75	5.30	ND	Absent
Mixed-leaf salad [c]	5.45	5.05	5.10	ND	Absent
Potato and carrot (cubes)	4.35	4.70	4.55	3.30	Absent
Potato and carrot (grated)	>6.3	4.90	>6.3	ND	Absent
Pumpkin 1	6.00	>6.3	5.00	ND	Absent
Pumpkin 2	5.50	5.10	5.70	ND	Absent
Pumpkin 3	5.30	4.85	5.20	ND	Absent
Pumpkin 4	5.20	4.10	4.05	ND	Absent
Vegetables for mayonnaise salad [d]	5.00	4.55	5.45	ND	Absent
Vegetables for Yakisoba [e]	5.40	5.30	5.50	ND	Absent
Vegetables for yakisoba [e]	6.10	5.75	5.15	ND	Absent
Yam 1	5.50	4.90	4.55	ND	Absent
Yam 2	6.30	4.90	5.45	ND	Absent
Yam 3	>6.3	5.70	5.75	ND	Absent

ND: Not detected at the lowest plated dilution; [a]: potato, carrot and chayote; [b]: iceberg lettuce and cabbage; [c]: cabbage and carrot; [d]: potato, barred potato, chayote, carrot and yam; [e]: broccoli, cauliflower, carrot, cabbage and green pea.

However, 90% (n = 27) of the analyzed samples were in compliance with the French and German legislations, since they recorded values lower than 7.70 log CFU/g (Legnani and Leoni, 2004). In addition, 10% (n = 3) of the samples did not allow detecting the exact CFU value because of the dilution used in the analysis, which limited the count to > 6.30 log CFU/g. Santos et al. (2010) have also analyzed aerobic mesophilic bacteria in RTE fruits and vegetables; they found mean count of 7.30 log CFU/g, which was higher than the one found in the present study. A study conducted with 172 RTE vegetable samples in Campinas, São Paulo, Brazil has found aerobic mesophilic bacteria count higher than 4 log CFU/g in all samples. Of the total, 72% (n = 124) presented count higher than 5 log CFU/g (Maistro et al. 2012).

The count of molds and yeasts ranged from 3.40 to > 6.30 log CFU/g; mean count was 5.14 log CFU/g. The Brazilian legislation (Brasil 2001) does not set limits for these groups in fresh or prepared fruits and vegetables. However, it is important analyzing this microbial group due to the deteriorating action of these microorganisms and to health risks induced by some mycotoxin-producing species. Based on the analysis conducted in the present study, 96.7% (n = 29) of the samples recorded values higher than 4 log CFU/g for molds and yeasts. Rosa (2002) has analyzed 140 RTE vegetable samples in Campinas, São Paulo and Belo Horizonte, Minas Gerais in Brazil and recorded yeasts and mold counts higher than 4 log CFU/g in 58.8% of the analyses.

The mean coliform count at 35°C was 5.14 log CFU/g. Among the analyzed samples, 80% (n = 24) presented coliform count higher than 5 log CFU/g. According to Arruda et al. (2004), scores above this value characterize the food as unfit for human consumption. Santos et al. (2010) recorded values ranging from <2 to 8.30 log CFU/g for RTE vegetables traded in Campinas, SP. Another study has found total coliform values higher than 3 log MPN/g (most probable number) in 81.5% (n = 132) of the RTE vegetable samples traded in Ribeirão Preto, São Paulo, Brazil (Oliveira et al. 2011). Vegetable samples (n = 172) analyzed in Campinas (SP) have shown total coliform counts ranging from 1.02 to 3.74 log CFU/g (Maistro et al. 2012). It is worth emphasizing that the Brazilian legislation does not set limits for coliforms at 35°C. However, coliform analysis can be used to indicate food quality, since high values indicate flaws in the hygiene process (Ferreira et al. 2016).

It was possible seeing that 13.3% (n = 4) of the RTE vegetable samples were contaminated with *E. coli*. The incidence of this microorganism in processed products may suggest contamination after food processing, as well as indicate the use of inadequate techniques during food handling and hygiene procedures (Oliveira et al. 2011). In the case of *E. coli*-contaminated vegetables in the present study are subjected to cooking procedure, this step could help eliminate the contamination through heat application. Carrots, in their turn, present increased risk to consumers' health because they can be consumed raw. No post-purchase procedure would be performed to eliminate the pathogen. Food producers are responsible for providing safe microbiological processed food to consumers.

Santos et al. (2010) have found *E. coli* count higher than the one allowed by the legislation in 29% (n = 52) of minimally processed fruit and vegetable samples. Contaminated vegetable food comprised leafy vegetables, grated carrots, grated zucchini, and mixed salads. Of the 512 vegetable samples analyzed in the city of São Paulo, only 2.7% (n = 14) presented *E. coli* counts higher than the one allowed by the Brazilian legislation (Sant'ana et al. 2011). Oliveira et al. (2011) have found *E. coli* in 53.1% (n = 86) of the vegetable samples analyzed in Ribeirão Preto, São Paulo, Brazil, whereas Gurler et al. (2015) have detected *E. coli* in 4% (n = 10) of the analyzed salad samples.

None of the vegetable samples presented *Salmonella* sp. Similar to the present study, Smanioto et al. (2009) did not find *Salmonella* sp. in any of the 15 vegetable samples analyzed in Bauru, São Paulo, Brazil. Prado et al. (2008) did not find *Salmonella* sp. in RTE vegetables traded in Ribeirão Preto, SP. A study conducted with 512 RTE vegetable samples in São Paulo, Brazil has found less than 1% (n = 4) contamination with *Salmonella* sp. (Sant'ana et al. 2011). However, Ferreira et al. (2016) and Oliveira et al. (2011) have found *Salmonella* sp. in 50% (n = 6) and 1.2% (n = 2) of the analyzed vegetable samples, respectively. Gurler et al. (2015) have found the pathogen in 8% (n = 14) of the analyzed salad samples.

Schuh et al. (2020) emphasize that the quality and shelf-life of minimally processed vegetables precisely depend on the production chain, since acquiring the raw material, to processing and commercialization. According to Cruz et al. (2019), minimally processed vegetables are vulnerable to spoilage and then should be storage in 1-5°C. In this context, both those who process this type of food and the supermarkets that trade them need to guarantee the storage in refrigerated conditions. By controlling the cold chain, microbial growth and spoilage could be reduced in these products. An increase of 5°C in storage temperature could increase the growth rate of any foodborne pathogens in these products (Cruz et al. 2019).

RTE vegetables analyzed in the current study did not show contamination with *Salmonella* sp., but results have indicated failures in the whole fruit and vegetable production process. Raw material quality, growing conditions, quality of the water used for irrigation, time and temperature control during the production process, equipment cleaning, sanitation, storage, and training programs focused on food handlers are procedures that should be implemented and improved. These efforts help improving the quality of the final product since high counts of indicator microorganisms can represent unsatisfactory hygiene conditions.

Labeling adequacy

Label evaluations have shown that 100% (n = 40) of the samples were not in compliance with one, or more, evaluated parameters (Table 4). This result was similar to the one observed by Prado et al. (2008), who analyzed the labeling of minimally processed vegetables in Ribeirão Preto, São Paulo, Brazil.

Table 4. Labeling adequacy (%) of 40 minimally processed fruit and vegetable samples traded in supermarkets in Vitória, Espírito Santo, Brazil - according to the Brazilian legislation.

Item	Present (%)	n	Absent (%)	n
Sales denomination	100	40	0	0
Supplier information	27.5	11	72.5	29
Ready-to-eat information	40	16	60	24
Ingredients	62.5	25	37.5	15
Net content	100	40	0	0
Origin and lot	100	40	0	0
Expiration date	92.5	37	7.5	3
Storage mode	92.5	37	7.5	3
Nutritional information	42.5	17	57.5	23
Gluten information	42.5	17	57.5	23

Based on the herein evaluated parameters, 100% of the samples presented sales denomination, which is a mandatory information that presents the product to the consumers. However, only 27.5% (n = 11) of the products had information about the supplier. This outcome indicates lack of transparency as to the origin of the product since this information regards the producing company and relevant data about it. Of the total labels, 40% (n = 16) presented the ready-to-eat indication. This information is necessary because it informs whether it is necessary subjecting the product to some process (e.g., cooking or sanitization) before its consumption (Prado 2008).

All samples had information about net content and manufacturing date. This information concerns the food amount purchased by consumers and the time elapsed since it was produced. Time directly affects the microbiological quality and organoleptic aspects of food. Of the evaluated labels, 92.5% (n = 37) presented expiration date and conservation mode. Such information is essential because it is directly associated with the microbial load found in the product. Inadequate storage temperature may favor pathogen growth and product deterioration (Maistro et al. 2012).

With respect to nutritional information, 57.5% (n = 23) of the analyzed samples did not inform whether the food "contained gluten" or was "gluten-free". The Brazilian legislation requires this information to help consumers prevent and control celiac disease (Brasil 2003a).

Labeling is an important communication instrument between producers and consumers. The information available in them, such as the incidence of allergenic proteins, additives, and other substances, has direct impact on consumers' health.

4. Conclusions

Minimally processed vegetables and fruits sold in Vitória, Espírito Santo, Brazil, presented high counts for aerobic mesophilic bacteria, molds and yeasts, and coliforms at 35°C but *Salmonella* sp. was absent in the samples. Although no pathogen was found in the samples, it is important to emphasize the high counts of the others microbial groups. These data may be related to failures during minimally processed fruits and vegetables processing steps, as well in the inadequate storage in the supermarkets. Labeling was also unsatisfactory due to lack of relevant information, which is categorized as mandatory by the Brazilian legislation. Good manufacturing practices, Hazard Analysis and Critical Control Points (HACCP), and stricter labeling enforcement should be adopted as strategies to help improving product quality and minimizing risks to consumers' health.

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References

ARRUDA, M.C.D., et al. Conservação de melão rendilhado minimamente processado sob atmosfera modificada ativa. *Food Science and Technology*. 2004, **24**(1), 53-58. https://doi.org/10.1590/S0101-20612004000100011.

BRASIL. Resolução RDC nº 12, de 02 de janeiro de 2001. *Aprova o Regulamento Técnico sobre Padrões Microbiológicos para Alimentos*. Brasília, 2001. Available from: https://portal.anvisa.gov.br/documents/33880/2568070/RDC_12_2001.pdf/15ffddf6-3767-4527-bfac-740a0400829b.

BRASIL. Resolução RDC nº 259, de 20 de setembro de 2002. *Aprova o Regulamento Técnico sobre Rotulagem de Alimentos Embalados*. Brasília, 2002. Available from: https://www.gov.br/agricultura/pt-br/assuntos/inspecao/produtos-vegetal/legislacao-1/biblioteca-de-normas-vinhos-e-bebidas/resolucao-rdc-no-259-de-20-de-setembro-de-2002.pdf/view.

BRASIL. Lei n° 10.674, de 16 de maio de 2003. *Obriga a que os produtos alimentícios comercializados informem sobre a presença de glúten, como medida preventiva e de controle da doença celíaca.* Diário Oficial da União, Brasília, DF, 19 maio 2003. p.1. Brasília, 2003a. Available from: https://www.planalto.gov.br/CCIVil_03/LEIS/2003/L10.674.htm.

BRASIL. Resolução RDC n° 359, de 23 de dezembro 2003. *Regulamento técnico de porções de alimentos embalados para fins de rotulagem nutricional.* Brasília, 2003b. Available from: http://portal.anvisa.gov.br/documents/33880/2568070/res0359_23_12_2003.pdf/76676765-a107-40d9-bb34-5f05ae897bf3.

BRASIL. Resolução RDC n° 360, de 23 de dezembro 2003**.** *Regulamento técnico sobre rotulagem nutricional de alimentos embalados.* Brasília, 2003c. Available from: https://portal.anvisa.gov.br/documents/33880/2568070/res0360_23_12_2003.pdf/5d4fc713-9c66-4512-b3c1-afee57e7d9bc.

BRUNO, L.M., et al. Avaliação microbiológica de hortaliças e frutas minimamente processadas comercializadas em Fortaleza (CE). *Boletim do Centro de Pesquisa de Processamento de Alimentos*. 2005, **23**(1), 75-84. https://dx.doi.org/10.5380/cep.v23i1.1272.

COLE, M.L. and SINGH, O.V. Microbial occurrence and antibiotic resistance in ready-to-go food items. *Journal of Food Science and Technology*. 2018, **55**(7), 2600–2609. https://doi.org/10.1007/s13197-018-3180-4.

CRUZ, M.R.G. da et al. Microbiological quality of minimally processed vegetables commercialized in Brasilia, DF, Brazil. *Food Science and Technology*. 2019, **39**(2), 498-503. https://doi.org/10.1590/fst.16018.

DOWNES, F.P. and ITO, K. *Compendium of methods for the microbiological examination of foods*. 4th ed. Washington: American Public Health Association. 2001.

ELIAS, S.O., DECOL, L.T. and TONDO, E.C. Foodborne outbreaks in Brazil associated with fruits and vegetables: 2008 through 2014. *Food Quality and Safety.* 2018, **2**(4), 173-181. https://doi.org/10.1093/fqsafe/fyy022.

FERREIRA, C.C., et. al. Análise de coliformes termotolerantes e *Salmonella* sp. em hortaliças minimamente processadas comercializadas em Belo Horizonte-MG. *HU Revista*. 2017, **42**(4), 307-313.

GURLER, Z., et al. The microbiological quality of ready-to-eat salads in Turkey: A focus on *Salmonella* spp. and *Listeria monocytogenes*. *International Journal of Food Microbiology*. 2015. **196***,* 79-83. https://doi.org/10.1016/j.ijfoodmicro.2014.11.021.

JEDDI, M.Z., et al. Microbial evaluation of fresh, minimally-processed vegetables and bagged sprouts from chain supermarkets. *Journal of Health, Population, and Nutrition*. 2014, **32**(3), 391-399.

KAMALA, K. and KUMAR, V.P., 2018. Food Products and Food Contamination. In: GRUMEZESCU, A. and HOLBAN, A.M. *Microbial Contamination and Food Degradation.* London: Academic Press, pp. 1-19. Available from: https://doi.org/10.1016/B978-0-12-811515-2.00001-9.

LEGNANI, P.P. and LEONI, E. Effect of processing and storage conditions on the microbiological quality of minimally processed vegetables. *International Journal of Food Science and Technology*. 2004, **39**(10), 1061–1068. https://doi.org/10.1111/j.1365-2621.2004.00891.x.

LEPAUS, B.M., ROCHA, J.S. and SÃO JOSE, J.F.B. de. Organic acids and hydrogen peroxide can replace chlorinated compounds as sanitizers on strawberries, cucumbers and rocket leaves. *Food Science and Technology*. 2020, **40**, 242-249. https://doi.org/10.1590/fst.09519.

MAISTRO, L.C., et al. Microbiological quality and safety of minimally-processed vegetables marketed in Campinas, SP–Brazil, as assessed by traditional and alternative methods. *Food Control.* 2012, **28**(2), 258-264. https://doi.org/10.1016/j.foodcont.2012.05.021.

OLAIMAT, A.N. and HOLLEY, R.A. Factors influencing the microbial safety of fresh produce: a review. *Food Microbiology*. 2012, **32**(1), 1-19. https://doi.org/10.1016/j.fm.2012.04.016.

OLIVEIRA, M.A., et al. Microbiological quality of ready-to-eat minimally-processed vegetables consumed in Brazil. *Food Control*. 2011, **22**(8), 1400-1403. https://doi.org/10.1016/j.foodcont.2011.02.020.

PINHEIRO, N.M.S., et al. Avaliação da qualidade microbiológica de frutos minimamente processados comercializados em supermercados de Fortaleza. *Revista Brasileira de Fruticultura*. 2005. **27**(1), 153-156. https://doi.org/10.1590/S0100-29452005000100040.

PRADO, S.D.P.T., et al. Avaliação microbiológica, parasitológica e da rotulagem de hortaliças minimamente processadas comercializadas no município de Ribeirão Preto, SP/Brasil. *Revista do Instituto Adolfo Lutz*. 2008, **67**(3), 221-227.

PUTNIK, P., et al. Modelling the shelf-life of minimally-processed fresh-cut apples packaged in a modified atmosphere using food quality parameters. *Food Control*. 2017, **81**, 55-64. http://dx.doi.org/10.1016/j.foodcont.2017.05.026.

RANJITHA, K., et al. Integrating calcium chloride treatment with polypropylene packaging improved the shelf-life and retained the quality profile of minimally-processed cabbage. *Food Chemistry*. 2018, **256**, 1-10. http://dx.doi.org/10.1016/j.foodchem.2018.02.012

ROSA, O.O. *Microbiota associada a produtos hortícolas minimamente processados comercializados em supermercados***.** Lavras: Universidade Federal de Lavras, 2002, pp. 1-120. Tese de Doutorado em Ciências dos Alimentos.

SANT'ANA, A.S., et al. Prevalence and counts of *Salmonella* spp. in minimally-processed vegetables in São Paulo, Brazil. *Food Microbiology.* 2011, **28**(6), 1235-1237. http://dx.doi.org/10.1016/j.fm.2011.04.002.

SANTOS, T.B.A., et al. Microrganismos indicadores em frutas e hortaliças minimamente processadas. *Brazilian Journal of Food Technology*. 2010, **13**(2), 141-146.

SCHUH, V., et al. Evaluation of the microbiological quality of minimally processed vegetables. *Food Science and Technology*. 2020, **40**(2), 290-295. https://doi.org/10.1590/fst.38118.

SHINOHARA, N.K.S., et al. *Salmonella* spp., importante agente patogênico veiculado em alimentos. *Ciência & Saúde Coletiva*. 2008, **13**(5), 1675-1683. https://doi.org/10.1590/S1413-81232008000500031.

SMANIOTO, T.F., et al. Qualidade microbiológica de frutas e hortaliças minimamente processadas. *Revista do Instituto Adolfo Lutz*. 2009, **68**(1), 150-154.

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