Research, Society and Development, v. 9, n. 8, e196985592, 2020
(CC BY 4.0) | ISSN 2525-3409 | DOI: http://dx.doi.org/10.33448/rsd-v9i8.5592

Contaminação microbiológica e parasitológica em alface (*Lactuca sativa* L.) comercializada em município do semiárido brasileiro

Microbiological and parasitological contamination of lettuces (*Lactuca sativa* L.) marketed in semiarid region of Brazil

Contaminación microbiológica y parasitológica en lechuga (*Lactuca sativa* L.) comercializada en un municipio de la región semiárida brasileña

Recebido: 13/06/2020 | Revisado: 14/06/2020 | Aceito: 17/06/2020 | Publicado: 29/06/2020

João Inácio Lopes Batista
ORCID: https://orcid.org/0000-0002-4984-2298
Universidade Federal Rural do Semi-árido, Brasil
E-mail: joaoinacio.batista@ufersa.edu.br
Tallyson Nogueira Barbosa
ORCID: https://orcid.org/0000-0001-8626-520X
Universidade Federal de Pelotas, Brasil
E-mail: tallyson_n_b@hotmail.com
Juliana Rafaela Granjeiro Rêgo
ORCID: https://orcid.org/0000-0002-8188-704X
Secretaria de Saúde Pública do Rio Grande do Norte, Brasil
E-mail: julia.aninha@hotmail.com
Cristina Karine de Oliveira Rebouças
ORCID: https://orcid.org/0000-0001-7072-1733
Universidade Federal Rural do Semi-árido, Brasil
E-mail: cristina.reboucas@hotmail.com
Taffarel Melo Torres
ORCID: https://orcid.org/0000-0001-8626-520X
Universidade Federal Rural do Semi-árido, Brasil
E-mail: taffarel.torres@ufersa.edu.br
Karoline Mikaelle de Paiva Soares
ORCID: https://orcid.org/0000-0003-1578-3733
Universidade Federal Rural do Semi-árido, Brasil
E-mail: karolinesoares@ufersa.edu.br

Ana Carla Diógenes Suassuna Bezerra
ORCID: https://orcid.org/0000-0002-1039-5187
Universidade Federal Rural do Semi-árido, Brasil
E-mail: anacarla@ufersa.edu.br

Resumo

O objetivo desta pesquisa foi avaliar a contaminação microbiana e parasitária em alface comercializada em feiras livres e supermercados do município de Mossoró. Para a avaliação parasitológica os métodos de sedimentação espontânea e flutuação foram aplicados. Para a parte microbiológica foi estimado o Número Mais Provável (NMP) de coliformes totais e termotolerantes, bem como verificada a positividade de *Salmonella* sp. Quanto à análise parasitária verificou-se que as amostras apresentaram um índice total de 62,5% (45/72) de contaminação, sendo 40,3% (29/72) com Ancilostomídeos, 29,2% (21/72) *Strongyloides* sp., 1,4% (1/72) *Ascaris* sp., 1,4% (1/72) *Entamoeba histolytica*/*díspar*, 1,4% (1/72) *Endolemax nana* e 2,8% (2/72) *Entamoeba coli*. Para as análises microbiológicas um índice de 100% (12/12) das amostras apresentaram coliformes totais e 25% (3/12) coliformes termotolerantes, acima do valor máximo permitido pela Legislação Brasileira, e ausência de *Salmonella* sp. Contudo, não houve diferença na contaminação microbiana e parasitária entre os estabelecimentos de comercialização desta pesquisa. Por fim, fica evidenciada a negligência para com as condições higiênico-sanitária das alfaces, por parte dos estabelecimentos comerciais aqui retratados, e o potencial risco a saúde na qual a população se encontra exposta.

Palavras-chave: Contaminação alimentar; Hortaliça; Enteroparasito; Coliformes.

Abstract

The objective of this research was to evaluate the microbial and parasitic contamination in lettuce sold in open markets and supermarkets in the city of Mossoró. For parasitological evaluation, spontaneous sedimentation and flotation methods were applied. For the microbiological part, the Most Probable Number (MPM) of total and thermotolerant coliforms was estimated, as well as the positivity of *Salmonella* sp. Regarding parasitic analysis it was found that the samples had a total index of 62.5% (45/72) of contamination, 40.3% (29/72) with Ancylostomatidae sp., 29.2% (21/72) with *Strongyloides* sp., 1.4% (1/72) *Ascaris* sp., 1.4% (1/72) *Entamoeba histolytica*/*dispar*, 1.4% (1/72) *Endolemax nana*, and 2.8% (2/72) *Entamoeba coli*. For microbiological analyzes, an index of 100% (12/12) of the samples

showed contamination by total coliforms and 25% (3/12) of thermotolerant coliforms, above the maximum value allowed by Brazilian legislation, and absence of *Salmonella* sp. However, there was no difference in microbial and parasitic contamination between the commercial establishments in this research. Finally, the neglect of the lettuce's hygienic-sanitary conditions by the commercial establishments depicted here is evident, as well as the potential health risk to which the population is exposed.

Keywords: Food contamination; Vegetable; Enteroparasite; Coliforms.

Resumen

El objetivo de esta investigación fue evaluar la contaminación microbiana y parasitaria en la lechuga vendida en mercados abiertos y supermercados en la ciudad de Mossoró. Para la evaluación parasitológica, se aplicaron métodos de sedimentación espontánea y flotación. Para la parte microbiológica, se estimó el número más probable (NMP) de coliformes totales y termotolerantes, así como la positividad de *Salmonella* sp. Con respecto al análisis parasitario, se encontró que las muestras tenían un índice total de 62.5% (45/72) de contaminación, 40.3% (29/72) con anquilostomas, 29.2% (21/72) *Strongyloides* sp., 1.4% (1/72) *Ascaris* sp., 1.4% (1/72) *Entamoeba histolytica*/disparate, 1.4% (1/72) *Endolemax nana* y 2.8% (2/72) *Entamoeba coli*. Para los análisis microbiológicos, un índice del 100% (12/12) de las muestras mostró coliformes totales y 25% (3/12) de coliformes termotolerantes, por encima del valor máximo permitido por la legislación brasileña, y ausencia de *Salmonella* sp. Sin embargo, no hubo diferencia en la contaminación microbiana y parasitaria entre los establecimientos comerciales en esta investigación. Finalmente, el descuido de las condiciones higiénico-sanitarias de la lechuga por parte de los establecimientos comerciales representados aquí es evidente, así como el riesgo potencial para la salud al que está expuesta la población.

Palabras clave: Contaminación alimentaria; Vegetales; Enteroparásitos; Coliformes.

1. Introduction

Vegetables and fruits are important components in daily meals because of their nutritional contribution to a balanced, healthy diet Lettuce (*Lactuca sativa* L.) is one of the most popular and consumed leaf vegetables in brazil (Dantas et al., 2020).

However, it is important for public health because it is consumed fresh. Thus, lettuce can be a disseminator of enteroparasitosis and gastroenteritis; these diseases can be

transmitted through the consumption of contaminated food with eggs, larvae, and cysts of parasites (Rodrigues et al., 2020) , or pathogenic bacteria.

Considering infectious pathogens, bacteria represented 95.9% of the known agents that cause foodborne diseases in Brazil in 2016-2017; *Escherichia coli* and *Salmonella* sp. are the main infectious agents found in food products (Ministério da Saúde, 2017; Souza et al., 2020). It is estimated that 107 species of parasites are transmitted through food and cause pathologies in humans (Orlandi et al., 2002).

Errors during handling, and environmental contamination caused by lack of basic sanitation can contribute to the dissemination of foodborne diseases. This denotes the social importance of studies on parasites and microorganisms; they can provide information on hygienic-sanitary conditions for production, storage, transport, and handling of foods. Thus, the objective of this work was to evaluate the microbiological and parasitological quality of lettuces marketed in Mossoró, state of Rio Grande do Norte, Brazil.

2. Methodology

Sampling

This work was carried out at the Laboratório de Biotecnologia Aplicada a Doenças Infecto-parasitárias and Laboratório de Biotecnologia de Alimentos, at the Universidade Federal Rural do Semi-árido, being a qualitative-quantitative laboratory research. Seventy-two lettuce samples were collected, 36 in supermarkets and 36 in a street market in Mossoró, Rio Grande do Norte, semiarid region of Brazil. The sampling was carried out weekly from December 2017 to April 2018, in six randomly supermarkets in the most populous neighborhoods (Ibge, 2013) in different points of the city, and in three stalls of a street market. Six samples were randomly collected in each market site, packaged in identified sterile plastic bags, placed in isothermal boxes and taken to a laboratory (Pereira et al, 2018).

Parasitological analysis

Leaves of the samples were manually defoliated, placed in distilled water, and left for sedimentation in a 24 hour period. The resulting sediment was analyzed in triplicate under an optical microscope (Takayanagui et al. 2001). Part of the sediment was analyzed according to the centrifuge-fluctuation method of faust, with adaptations. It was centrifuged, the

supernatant was discarded, and resuspended in a 33% zinc sulfate solution (Nascimento & Alencar, 2014). This material was centrifuged again and the resulting supernatant was analyzed under an optical microscope.

Microbiological analysis

One sample per market site was analyzed to determine the most probable number (MPN) of total coliforms and thermotolerant coliforms, and presence of *Salmonella* sp. Twenty-five grams of each lettuce sample were homogenized in 225 mL of 0.1% peptone water (first dilution).

Aliquots of 1 mL of 10^{-1} to 10^{-3} dilutions of each sample were inoculated in a lauryl sulfate tryptose (LST) broth for presumptive testing, and incubated in water bath at 36±0.5 °C for 48 hours to determine the MPN of total coliforms and thermotolerant coliforms. The test tubes that showed gas production inside the Duran tubes were taken as positive and subsampled in brilliant green bile broth 2%, using the same temperature and time to confirm the presence of coliforms. Subsamples of these positive samples were incubated in an *Escherichia coli* broth at 45.5±0.2 °C for 24 to 48 hours in a water bath to quantify the thermotolerant coliforms. The sequence of positive tubes of each dilution was recorded and the total and thermotolerant coliforms were determined using MPN tables (Silva et al, 2007).

The occurrence of *Salmonella* sp. was tested using the samples of the first dilution, which were incubated at 36 °C for 24 hours in a bacteriological oven in the pre-enrichment stage in a peptone saline solution. Subsequently, selective enrichment was performed—aliquots of 1 mL were transferred to three different broths (Rappaport Vassiliadis, Selenite Cystine & Tetrathionate) and incubated at 41° C for 24 hours. Then, differential plating was performed—samples were inoculated in eosin methylene blue agar, and Rambach agar in Petri dishes and incubated upside down at 36 °C for 24 hours. Formations of typical colonies were analyzed using confirmatory biochemical tests, such as lysine decarboxylation, lactose and sucrose fermentation, and H_2S production (Silva et al. 2007).

Statistical analysis

The experimental design is a randomized type. The data were analyzed using the χ^2 test, and the Fisher's exact test, considering the significance level of 5%. The data were analyzed in spreadsheets and the tests were carried out using the program PAST v2.17.

3. Results

According to the results, 62.5% (45/72) of the lettuce samples were contaminated with parasites, 61.1% (22/36) from supermarkets, and 63.9% (23/36) from the street market.

Most contaminated samples (73.3%; 33/45) presented only one enteroparasite species; however, 26.7% (12/45) of the samples presented multiple parasitism, 30.4% from the street market, and 22.7% from supermarkets (p = 0.559). As for the pathogenic parasites found, the frequencies are shown in Table 1.

Table 1 – Frequency of parasites and other contaminants in lettuce samples from supermarkets and a street market of Mossoró, Rio Grande do Norte, Brazil.

Parasite or contaminant	Supermarkets	Street market	Total		p
			n	%	
Ancylostomatidae sp.	15	14	29	40.3	
Strongyloides sp.	8	13	21	29.2	
Ascaris sp.	1	0	1	1.4	
Endolemax nana	1	0	1	1.4	
Entamoeba histolytica/dispar	1	0	1	1.4	
Entamoeba coli	0	2	2	2.8	
Free-living larvae	4	14	18	25.0	*
Oesophagostomum sp.	1	11	12	16.7	*
Insect	1	13	14	19.4	*
Ciliate protozoan	1	2	3	4.2	

* = $p < 0.05$. Source: Author.

Table 1 shows the pathogenic parasites diagnosed, with the highest frequencies referring to Ancylostomatidae sp with 40.3% (n = 29) and *Strongyloides* sp. with 29.2% (n = 21), followed by *Ascaris* sp. 1.4% (n = 1) and *Entamoeba histolytica* /dispar 1.4% (n = 1).

The analyses of parasites, larvae, and dirt showed that 73.6% (53/72) of the samples had some contamination, 63.9% (23/36) from supermarkets, and 83.3% (30/36) from the street market; however, the sites presented no significant difference ($p = 0.061$).

The microbiological analyses showed that 100% (12/12) of the samples presented contamination by total coliforms. Thermotolerant coliforms above 10^2 MPN g^{-1}, which is the maximum MPN allowed by the Brazilian Legislation (Brasil, 2001), were found in 25% (3/12) of the samples, 16.7% (1/6) from the street market, and 33.3% (2/6) from supermarkets; however, the sites presented no significant difference ($p = 0.505$) (Table 2).

Table 1 – Microbiological analysis of lettuces from supermarkets and a street market of Mossoró, Rio Grande do Norte, Brazil, considering the most probable number (MPN g^{-1}).

	Total coliforms		Thermotolerant coliforms		*Salmonella* sp.	
Sample	SM	Sup	SM	Sup	SM	Sup
1	>1100	>1100	20	120	Abs	Abs
2	>1100	21	23	<3.0	Abs	Abs
3	>1100	>1100	120	150	Abs	Abs
4	>1100	>1100	20	9.2	Abs	Abs
5	>1100	93	20	15	Abs	Abs
6	>1100	>1100	23	35	Abs	Abs

SM = street market; Sup = Supermarkets; Abs = Absence
Source: Author.

Table 2 also shows that the lettuce samples analyzed were negative for the presence of the microorganism *Salmonella* sp.

4. Discussion

The parasitological analysis showed 62.5% (45/72) of the analyzed lettuce samples contaminated with parasites, i.e., in disagreement with the brazilian resolution n°. 12/1978 (CNNPA), which requires the absence of these contaminants (Brasil, 1978).

Structural and hygienic flaws were found in the sampled street market. This street market is in a consolidated market site in the urban area. However, the lack of investments and inspections contribute to inappropriate practices by the traders. According to the Resolution n°. 216/2004, several nonconformities regarding good practices in food services were found in this street market (Brasil, 2004).

Most sampled street market stalls had a fixed structure, but some were improvised with removable wood structure, lined with tarpaulins or not impermeable materials that are difficult to sanitize. Full dumpsters, and litter on the ground were found; these conditions favor the appearance of disease vectors and pests (Ferreira et al., 2015). Moreover, traders handle money and food concomitantly, without antisepsis of the hands.

Precarious structure, inadequate handling of the products, and exposure of products to sunlight, insects, and dust favor the proliferation of pathogenic microorganisms, and contamination and dissemination of foodborne diseases (Ferreira et al., 2015).

The lettuces from supermarkets presented no significant differences (p = 0.808) regarding the presence of parasites. Contrastingly, Nascimento & Alencar (2014), found that 30% of lettuces from supermarkets and 70% from the street market were not suitable for consumption, and lettuce was the vegetable that had the highest parasite load.

The cultural practices are another factor that contribute to the contamination of vegetables. Lettuce is grown in conventional, organic, and hydroponic systems; the first is carried out directly in the soil, with more exposure to contaminants, microorganisms and parasites (Silva et al., 2020). Rainfall also affect the contamination of lettuces. Droplets of water falling on the soil can carry soil particles and parasitic structures to plants, and they can be lodged between lettuce leaves (Tefera et al., 2018).

The most found pathogenic parasites were Ancylostomidae species and *Strongyloides* sp., probably due to organic fertilization (Fernandes et al., 2015). Husbandry of animals that have access to agricultural areas can also cause contamination of crops and contribute to disseminating pathogens (Silva et al., 2020). *Ascaris* was another helminth genus found in the samples. Despite it was found in only 1.4% of the analyzed samples, ascariasis is one of the most common helminthiasis in the world (Corvino & Bhimji, 2017). Leafy vegetables are easily contaminated due to their large contact surface, juxtaposed and flexible leaves that favor accumulation of residues and water, and their direct contact with the soil, which also facilitates contamination by microorganisms (Tefera et al., 2018).

The analyses also showed presence of soil particles, earthworms, slugs, small insects, mites, larvae of *Oesophagostomum* sp., and larvae of unidentified free-living nematodes, which suggest fecal contamination from animals, possibly due to the use of manure-based fertilizers.

The frequencies of insects, larvae of *Oesophagostomum* sp., and free-living larvae were significantly higher in samples from the street market (p<0.05), probably due to inadequate use of organic fertilization, structural flaws, and inadequate exposure of the product in this street market (Ferreira et al., 2015). The supermarket samples appeared to be cleaner because they were possibly washed before being put up for sale.

Two ciliate protozoa were found, which are similar to *Balantidium* sp. These protozoa can be found in several vertebrate hosts, and the species that parasitize pigs is morphologically indistinguishable from that that contaminates humans (Rey, 2017). *Balantidium coli* is the largest protozoan that parasites humans, but medical records are rare. Its symptoms may vary from asymptomatic to chronic with dysentery conditions causing necrotic lesions similar to amebiasis (Rey, 2017). The microbiological analyzes showed that

the coliform counts at 35 ºC in the lettuce samples ranged from 21 to >1,100 MPN g^{-1}, with maximum counts (> 1,100 MPN g^{-1}) in all samples from the street market. Although expressive, finding these coliforms in water and food does not necessarily represent fecal contamination, since these microorganisms have environmental colonization capacity, and are found, for example, in vegetation, water, soil, and insects (Silva et al. 2020).

The presence of total coliforms indicates flaws in hygienic conditions of food production, whereas thermotolerant coliforms are used as indicators of sanitary quality of the food, indicating presence of *Escherichia coli*, which is the only reliable indicator of fecal contamination (Martin et al., 2016).

However, no parameters or limits have been established for the detection of total coliforms by the Brazilian legislation. The Resolution RDC n°. 12/2001 of the Brazilian Sanitary Surveillance Agency (ANVISA) establishes microbiological limits of up to 10^2 MPN g^{-1} for thermotolerant coliforms, and absence of *Salmonella* sp. for fresh vegetables (Brasil, 2001).

Preharvest factors include contaminations from soil, irrigation water, inadequately composted animal manure, presence of domestic or wild animals in the plantations, dust, insects, and human handling (Ssemanda et al., 2018). Post-harvest factors include contaminations from harvesting, processing, packaging, equipment used, transport vehicles, rinsing water, inadequate storage, cross-contamination with other foods, and human handling (farmers, traders, and consumers) (Tefera et al., 2018).

These results show the possibility of risks of transmission of diseases to humans by consumption of lettuce and the necessity of properly preparation of this vegetable for consumption, either by disinfection or cooking. . The application of sanitizers, such as acetic acid and sodium hypochlorite in lettuce leaves can reduce thermotolerant coliform counts to the limits recommended by Brazilian legislation, with sodium hypochlorite 1% being more efficient than acetic acid 6.6% for the removal of parasites, and total coliforms and thermotolerant coliforms (Nascimento & Alencar, 2014).

Thus, the high occurrence rate of pathogens found denotes that this is an important public health problem, mainly because lettuce is commonly consumed fresh. Therefore, greater attention on the inspection of the markets by the regulatory agencies and good production practices are necessary to a greater hygienic control in production, distribution, and trade processes of lettuce.

5. Conclusion

The lettuces (*Lactuca sativa* L.) marketed in Mossoró, semiarid region of Brazil, presented high biological contamination. They had high contamination by parasites and thermotolerant coliforms, disagreeing with the parameters recommended by the Brazilian legislation, showing that the fresh consumption of this vegetable had a potential risk to public health.

Due to this scenario, it is essential to further studies focused on product quality control techniques by commercial establishments, so that they adopt good manufacturing and distribution practices. As well as studies related to adequate and safe home hygiene offering the consumer sanitation methods to obtain a product suitable for consumption.

References

Brasil. (1978) Agência Nacional de Vigilância Sanitária. *A CNNPA do Ministério da Saúde aprova 47 padrões de identidade e qualidade relativos a alimentos e bebidas para serem seguidos em todo território brasileiro*. Resolução – RDC n°12, 24 julho de 1978. Diário Oficial da União. Disponível em: <http://portal.anvisa.gov.br/documents/33916/394219/Resolucao_CNNPA_n_12_de_1978.pdf/4f93730f-65b8-4d3c-a362-eae311de5547> Acesso em: 10 de janeiro de 2020.

Brasil. (2001). Agência Nacional de Vigilância Sanitária. *Aprova o Regulamento Técnico sobre padrões microbiológicos para alimentos*. Resolução – RDC n°12, 2 jan. 2001. Diário Oficial da União. Disponível em:<http://portal.anvisa.gov.br/documents/33880/2568070/RDC_12_2001.pdf/15ffddf6-3767-4527-bfac-740a0400829b>. Acesso em: 20 de Jun. 2019.

Brasil. (2004). Agência Nacional de Vigilância Sanitária (ANVISA). *Dispõe sobre Regulamento Técnico de Boas Práticas para Serviços de Alimentação*. Resolução RDC nº 216, de 15 de setembro de 2004. Diário Oficial da União. Disponível em: < http://portal.anvisa.gov.br/documents/33916/388704/RESOLU%25C3%2587%25C3%2583O RDC%2BN%2B216%2BDE%2B15%2BDE%2BSETEMBRO%2BDE%2B2004.pdf/23701496-925d-4d4d-99aa-9d479b316c4b>. Acesso em: 10 de Jan. 2020.

Corvino, D. de L., & Bhimji, S. (2017). Ascariasis. StatPearls [Internet]. Treasure Island (FL):

StatPearls Publishing. Acesso em 25 de janeiro de 2019 em: <http://www.ncbi.nlm.nih.gov/pubmed/28613547>.

Dantas, M. M., Lima, E. Q., & Queiroga Filho, E. (2020). Parasitic evaluation in vegetable commercialized in the semiarid region of Paraíba, Brazil. *Brazilian Journal of Development*. 6(1), 2667-2684.

Fernandes, N. D. S., Guimarães, H. R., Amorim, A. C. D. S., Reis, M. B., Trindade, R. A., & Melo, A. C. F. L. (2015). Avaliação parasitológica de hortaliças: da horta ao consumidor final. *Saúde e Pesquisa*, 8(2), 255-265.

Ferreira, A. B., Alvarenga, S. H. F., & São José, J. F. B. (2015). Qualidade de frutas e hortaliças orgânicas comercializadas em feiras livres. *Revista do Instituto Adolfo Lutz*, 74(4), 410-419.

IBGE. (2013). Atlas do censo demográfico 2010. Rio de Janeiro: IBGE.

Martin, N. H., Trmčić, A., Hsieh, T. H., Boor, K. J. & Wiedmann, M. (2016). The evolving role of coliforms as indicators of unhygienic processing conditions in dairy foods. *Frontiers in microbiology*, 7, 1-8.

Ministério da Saúde. (2017). Surtos de doenças transmitidas por alimentos no Brasil. Acesso em 13 de janeiro de 2019 em: <http://portalarquivos.saude.gov.br/images/pdf/2017/maio/29/Apresentacao-Surtos-DTA-2017.pdf>.

Nascimento, E. D., & Alencar, F. L. S. (2014). Eficiência antimicrobiana e antiparasitária de desinfetantes na higienização de hortaliças na cidade de Natal - RN. *Ciência e Natura*, 36(2), 92-106.

Orlandi, P. A., Chu, D. M. T., Bier, J. W., & Jackson, G. J. (2002). Parasites and the Food Supply. *Food technology*, 56(4), 71-81.

Pereira A. S. et al (2018). *Methodology of cientific research*. [*e-Book*]. Santa Maria City. UAB / NTE / UFSM Editors.

Rey, L. (2017). *Bases da parasitologia médica*. 3 ed. Rio de Janeiro: Guanabara Koogan, 391p.

Rodrigues, A. C., Silva, M. D. C., Pereira, R. A. S. & Pinto, L. C. (2020). Prevalence of contamination by intestinal parasites in vegetables (*Lactuca sativa* L. and *Coriandrum sativum* L.) sold in markets in Belém, northern Brazil. *Journal of the Science of Food and Agriculture*, 100(7), 2859-2865.

Silva, N., Junqueira, V. C. A., Silveira, N. F. de A., Taniwaki, M. H., Gomes, R. A. R. & Okazaki, M. M. (2007). Manual de Métodos de Análise Microbiológica de Alimentos e Água. 5ed. São Paulo: Blucher.

Silva, J. J. B., Miranda, F. S., Almeida, J. S., Amor, A. L. M. & Silva, I. M. M. (2020). Microbiological and parasitological contamination of vegetables, water and soil in rural communities. *African Journal of Microbiology Research*. 14(1), 8-15.

Souza, A. C. F., Viana, D. C., Souza, J. F. & Costa, A. L. P. (2020). Análise microbiológica de frutas e hortaliças minimamente processadas comercializadas em supermercados da cidade de Macapá – Amapá. *Research, Society and Development*, 9(6), 1-19.

Ssemanda, J. N., Reij, M. W., Van Middendorp, G. Bouw, E., Plaats, R., Franz, E., Muvunyi, C. M., Bagabe, M. C., Zwietering, M. H. & Joosten, H. (2018). Foodborne pathogens and their risk exposure factors associated with farm vegetables in Rwanda. *Food Control*, 89, 86-96.

Takayanagui, O. M.; Oliveira, C. D., Bergamini, A. M., Capuano, D. M., Okino, M. H. T., Febrônio, L. H. P., Silva, A. A. M. C. C., Oliveira, M. A., Ribeiro, E. G. A. & Takayanagui, A. M. M. (2001). Fiscalização de verduras comercializadas no município de Ribeirão Preto, SP. *Revista da Sociedade Brasileira de Medicina Tropical*, 34(1), 37-41, 2001.

Tefera, T., Tysnes, K. R., Utaaker, K. S. & Robertson, L. J. (2018). Parasite contamination of

berries: Risk, occurrence, and approaches for mitigation. *Food and Waterborne Parasitology*, 10, 23-38.

Percentage of contribution of each author in the manuscript

João Inácio Lopes Batista – 30%

Tallyson Nogueira Barbosa – 20%

Juliana Rafaela Granjeiro Rêgo – 10%

Cristina Karine de Oliveira Rebouças – 10%

Taffarel Melo Torres – 10%

Karoline Mikaelle de Paiva Soares – 10%

Ana Carla Diógenes Suassuna Bezerra – 10%