Prevalence of enteroparasites in vegetables marketed in the city of Jaguaré, Espírito Santo, Brazil

Prevalencia de enteroparásitos en hortalizas comercializadas en el municipio de Jaguaré, Espírito Santo, Brasil

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ABSTRACT

Objective To monitor the frequency of enteroparasites in lettuce (Lactuca sativa) and parsley (Petrosolium sativum) samples, marketed in commercial buildings in the municipality of Jaguaré, Espírito Santo, Brazil.

Materials and Methods Between August 2016 and July 2017, five different popular commercial buildings (markets, supermarkets and fairs) in the municipality of Jaguaré, considered the main local vegetable suppliers were selected. Throughout the research, 30 vegetable samples were randomly selected for parasitological analysis in 3 replicates for each commercial building under study. The obtained samples were analyzed by the modified spontaneous sedimentation method at the Laboratory of Parasitology and Hematology of Universidade Federal do Espírito Santo.

Results Of the 30 samples analyzed, 83.3% were positive for parasitic contamination and the most common parasites were Hookworm, Balantidium coli, Isospora belli, Ascaris sp. and Entamoeba sp.

Conclusions The results indicate a high degree of contamination of the vegetables, probably related to the precariousness of production, transportation and handling, making it necessary to adopt measures that ensure the best quality of food and, consequently, better population health.

Key Words: Parasitology; vegetables; food contamination; public health (source: MeSH, NLM).

RESUMEN

Objetivo Monitorear la frecuencia de enteroparásitos en muestras de lechuga (Lactuca sativa) y perejil (Petrosolium sativum), comercializadas en edificios comerciales en el municipio de Jaguaré, Espírito Santo, Brasil.

Materiales y Métodos Entre agosto de 2016 y julio de 2017, se seleccionaron cinco diferentes edificios comerciales populares (mercados, supermercados y ferias) en el municipio de Jaguaré, considerados los principales proveedores locales de vegetales. A lo largo de la investigación, se seleccionaron al azar 30 muestras de vegetales para el análisis parasitológico en 3 repeticiones para cada edificio comercial en estudio. Las muestras obtenidas fueron analizadas por el método modificado de sedimentación espontánea en el Laboratorio de Parasitología y Hematología de la Universidad Federal del Espírito Santo.

Resultados De las 30 muestras analizadas 83.3% fueron positivas para la contaminación parasitaria y los parásitos más comunes fueron Anquilostoma, Balantidium coli, Isospora belli, Ascaris sp. y Entamoeba sp.

Conclusiones Los resultados indican un alto grado de contaminación de las hortalizas, probablemente relacionado con la precariedad de la producción, el transporte y la manipulación, por lo que es necesario adoptar medidas que aseguren la mejor calidad.
Palabras Clave: Parasitología; verduras; contaminación de alimentos; salud pública (fuente: DeCS, BIREME).

MATERIALS AND METHODS

Study area
The municipality of Jaguaré is located in the far north of the state of Espírito Santo, and currently occupies an area of 659,751 km². According to the Census conducted by the Brazilian Institute of Geography and Statistics (IBGE) in 2010, the municipality had a population of 24,678 inhabitants, and may be considered small, when compared to the total population of Espírito Santo (3,514,952 inhabitants) (19).

Sample collection
Between August 2016 and July 2017, the vegetables were collected in five different popular commercial buildings (markets, supermarkets and fairs) in the municipality of Jaguaré, considered the main local vegetable suppliers. Throughout the research, 30 vegetable samples were randomly selected for parasitological analysis, in 3 replicates for each commercial building under study.

The selected vegetables were lettuce (L. sativa) and parsley (P. sativum), considering the head as the sample unit for lettuce and the package for parsley, regardless of the weight and size they had (9). The selected samples were isolated in individual plastic bags, identified and sent to the Laboratory of Parasitology...
and Hematology of Centro Universitário Norte do Espírito Santo (CEUNES), Universidade Federal do Espírito Santo, to be properly analyzed.

**Processing and analysis of vegetables**
The samples were processed according to the spontaneous sedimentation method (20) with some modifications, and the samples were submitted to two washing processes. In the first, by rinsing, 250 mL of distilled water were added to the plastic bag containing the sample, shaking it manually for 30 seconds, while in the second process, by defoliation, each leaf was carefully rubbed in a plastic container with 250 mL of distilled water (9,21,22).

The washing solution from each of the two processes was filtered into a conical cup using a surgical gauze folded into four, resting for 24 hours. After this period, this solution was centrifuged at 9000 rpm for 3 minutes, and the sediment was collected in test tubes (21,22).

The parasitic forms were identified using light microscopy, in which the sediment was deposited on a glass slide, covered by a coverslip, and stained with lugol solution. At least two slides were examined per test tube for each vegetable sample, using 10x and 40x lenses (23).

**RESULTS**

During the study, 25 (83.3%) of the 30 samples of vegetables analyzed during the 12-month period were positive for contamination with some parasitic species. It should be observed that 61.5% of the contaminated lettuce samples had more than two parasitic species (polyparasitism), while 38.5% of these samples were equivalent to those contaminated with only one parasitic species (monoparasitism). These values were similar for parsley samples, in which polyparasitism was observed in 66.7% of the samples and monoparasitism in 33.3% (Table 1).

| Samples | Monoparasitism (%) | Polyparasitism (%) |
|---------|--------------------|--------------------|
| Lettuce | 38.5               | 61.5               |
| Parsley | 33.3               | 66.7               |

Regarding the sampled buildings, vegetables from supermarkets presented a contamination index of 79.2%, while vegetables collected in fairs showed 100% contamination with parasitic species (Table 2).

| Buildings | Analyzed samples (number) | Positive samples (number) | Positive samples (%) |
|-----------|---------------------------|---------------------------|----------------------|
| Supermarkets | 24                        | 19                        | 79.2                 |
| Fairs     | 6                         | 6                         | 100                  |
| Total     | 30                        | 25                        | 83.3                 |

As for the distribution of parasites in the municipality of Jaguaré, it was observed that all five buildings in the study had lettuce and parsley samples positive for some parasitic species (Figure 1).

**Table 1. Degree of parasitism in lettuce and parsley samples marketed in the municipality of Jaguaré, Espírito Santo, Brazil, 2017**

**Table 2. Frequency of positive vegetable samples from supermarkets and fairs in Jaguaré, Espírito Santo, Brazil, 2017**

**Figure 1. Commercial buildings in the municipality of Jaguaré, Espírito Santo, Brazil, participating in the study, 2017**

Collection point 1. S 18° 54' 49.82"/040° 4' 41.47"; Collection point 2. S 18° 54' 19.57"/040° 4' 55.37"; Collection point 3. S 18° 54' 28.07"/040° 4' 32.31"; Collection point 4. S 18° 54' 22.03"/040° 4' 20.10"; Collection point 5. S 18° 54' 27.14"/040° 4' 5.83". Source: Google Earth, 2018.
The parasites with the highest frequency in parsley samples in the parasitological analyses were Hookworm (60.0%), Isospora belli (33.3%) and B. coli (33.3%) (Table 3).

Table 3. Frequency of enteroparasites in parsley (Petrosolium sativum) samples from commercial buildings in the municipality of Jaguaré, Espírito Santo, Brazil, 2017

| Parasitic species | Frequency (%) |
|-------------------|--------------|
| Toxocara sp.      | 6.7          |
| Entamoeba sp.     | 13.3         |
| Ascaris sp.       | 13.3         |
| Balantidium coli  | 33.3         |
| Isospora belli    | 33.3         |
| Hookworm          | 60           |
| Entamoeba coli    | 65           |

In relation to lettuce samples, the most frequent parasites were Hookworm (66.7%), B. coli (46.7%), Ascaris sp. (20.0%) and Entamoeba sp. (20.0%) (Table 4).

Table 4. Frequency of enteroparasites and commensals in lettuce (Lactuca sativa) samples from commercial buildings in the municipality of Jaguaré, Espírito Santo, Brazil, 2017

| Parasitic species | Frequency (%) |
|-------------------|--------------|
| Paramaecium sp.   | 6.7          |
| Giardia sp.       | 6.7          |
| Isospora belli    | 13.3         |
| Entamoeba sp.     | 20.0         |
| Ascaris sp.       | 20.0         |
| Balantidium coli  | 46.7         |
| Hookworm          | 66.7         |

**DISCUSSION**

The consumption of fruits and vegetables, especially fresh, provides important health benefits, with direct implications in life quality due to their nutritional attributes. However, the ingestion of these raw or minimally processed foods is directly linked to parasitic disease outbreaks, since these vegetables can be grown in areas contaminated with fecal waste or irrigated with polluted waters (6,9,11).

The results presented in the analyses carried out in Jaguaré showed 83.3% positive samples for some parasitic species, indicating a higher percentage, when compared to parasitological analyses carried out in Teresina, Piauí, and in the city of Rio de Janeiro which showed, respectively, 34.1% and 36.4% positive samples. However, they are considered inferior to the value found in samples collected in Campina Grande, Paraíba, where 100% were contaminated (14,17,24).

The variability in positive indices is probably related to the great territorial extension and its diversity regarding climatic and environmental characteristics, which may favor the occurrence of parasitic contamination in vegetables and fruits (25).

When the degree of polyparasitism was evaluated, a similar result was observed for lettuce and parsley (61.5% and 66.7%, respectively). Alves et al. (25) indicated 66.7% positive samples for the analyzed lettuce of supermarkets in the city of Cuiabá, Mato Grosso. Polyparasitism was the predominant type of contamination in the evaluated samples (70%).

Based on these data, the high parasitic contamination rates of the samples collected in Jaguaré, as well as the majority of them having presented multiple contamination, may be directly related to inadequate practices during the planting process, where soil and irrigation water may be contaminated by fecal waste. In addition, transport and storage under inadequate conditions, besides poor food sanitation before it is available to consumers, can directly influence its quality (11,13,14,25).

Freitas et al. (26) confirmed in their studies that samples of vegetables from supermarkets and fairs in the municipality of Campo Mourão, Paraná, had a percentage of contamination of 56% and 58.7%, respectively, while the samples collected in the municipality of Jaguaré showed much higher values (79.2% for supermarkets and 100% for fairs). These results may indicate that planting is carried out under inadequate conditions, possibly with contamination of soil or irrigation water with human or animal fecal waste. However, although the contamination rate is higher in fairs, the contamination index in vegetables from supermarkets is also worrisome, once they demonstrate that the hygiene practices for vegetables are frequently neglected by producers and distributors, as reported by Fernandes et al. (27).

The parasitic species most frequently found during the parasitological analyses of vegetables sold in Jaguaré were Hookworm, B. coli, I. belli, Ascaris sp. and Entamoeba sp. These parasites were also found in studies conducted in Londrina, Teresina, Rio de Janeiro and the Federal District, showing that environmental and climatic conditions throughout the Brazilian territory favor the spread of enteroparasites, in the same way that socioeconomic characteristics and hygienic-sanitary conditions influence these processes (9,17,28,29).

The main clinical manifestations of Hookworm and Ascaris sp. in the host, involve diarrhea, anemia, nutritional deficiency, intestinal obstruction and, in more severe conditions, can lead to death (17,23,30); the high incidence of Ascaris sp. (20%) in lettuce samples may be related to the morphology of the peel, which allows better adhesion to the plant foliage (26,31).

In general, B. coli is a commensal protozoa of pig intestinal lumen that can parasite humans when a lesion occurs in the mucosa of the colon and the cecum, occurring
a secondary infection, since it is not able to penetrate alone the intact mucosa. Its presence frequently indicates contamination of food and soil fertilized with swine, ruminal and human fecal waste (9,23).

Human isosporis is more frequent in hot regions where hygiene conditions are poor. Its infection occurs through the ingestion of food or water contaminated with sporulated *I. belli* oocysts, whose symptoms include fever, diarrhea, abdominal cramps and vomiting. More severe cases of this pathology commonly occur in children and individuals with some immunodeficiency, and may cause chronic diarrhea, leading to dehydration, weight loss and even hospitalization (23,33).

Not less important, the presence of *E. coli* in parsley samples, despite being a commensal, is considered an indicative factor of parasitic contamination with fecal waste, and its diagnosis is essential in order to propagate prophylactic measures to avoid other contaminations, for example, by pathogenic amoeba, such as *E. histolytica* (9,27,32). In addition, the observed *Toxocara* sp. eggs suggest the presence of dogs and cats, definitive hosts of the disease, in regions close to planting and in water sources used for irrigation (23,33).

The results obtained in Jauaré indicate a high degree of contamination of vegetables and are probably related to the precariousness of hygiene care, which directly influences the quality of these products. According to the National Sanitary Surveillance Agency (34), any type of food must be free from foreign matter indicating health risks such as dirt, insects and parasites, so that they are suitable for consumption. In this context, vegetables supplied in the municipality are in disagreement with the current legislation, have an important role in the dissemination of enteroparasites, given the high contamination rates observed during the parasitological analyses and may pose great risks to the health of the local population.

Therefore, in order to avoid parasitic disease outbreaks it is necessary to improve hygiene and sanitation during food production, processing, transportation and final handling as well as instructions to the population about prophylactic measures and the treatment of these diseases, emphasizing the need for Sanitary Surveillance in the inspection processes of vegetable gardens and food distributors, in order to ensure greater safety and food quality for the population.

**REFERENCES**

1. Capuano DM, Lazzarini MPT, Giacomelli Júnior E, Takayanagui OM. Enteroparasitoses em manipuladores de alimentos do município de Ribeirão Preto-SP, Brasil. Rev. bras. Epidemiol [Internet]. 2008; 11(4):687-95. DOI:10.1590/S1415-790X2008000400015.

2. Brasil. Ministério da Saúde, Secretaria de Vigilância em Saúde, Departamento de Vigilância Epidemiológica. Manual Integrado de Vigilância, Prevenção e Controle de Doenças Transmitidas por Alimentos [Internet]. Brasília: Ministério de Saúde; 2010 [cited 2018 May 23]. Available from: https://bit.ly/36zR8KO.

3. Oliveira ABA, Paula CMD, Capalonga R, Cardoso MRI, Tondo EC. Doenças transmitidas por alimentos, principais agentes etiológicos e aspectos gerais: uma revisão. Rev. HCPA & Fac. Med. Univ. Fed. Rio Gd. do Sul [Internet]. 2010 [cited 2018 Dec 18];30(3):279-85. https://bit.ly/3cymyph.

4. Angeluci CHG, Silva NSL, Rego LS, Santos EM. Avaliação da prevalência de parasitoses intestinais em escolares do município de formosa, GO. Rev. Sinergia [Internet]. 2013 [cited 2018 Dec 18]; 14(3):227-32.

5. Lacerda JS, Ardem CML. Estudo de prevalência de parasitoses intestinais em pacientes de um laboratório privado de Aracatuba-SP. Rev. saude. UniToledo [Internet]. 2017 [cited 2018 Dec 18]; 1(1):107-20. https://bit.ly/3tfzTr.

6. Soares B, Cantos GA. Qualidade parasitológica e condições higiênico-sanitárias de hortaliças comercializadas na cidade de Florianópolis, Santa Catarina, Brasil. Rev. Bras. [Internet]; epidemiol. 2005 [cited 2018 Dec 18]; 8(4):377-84. DOI:10.1905/S1415-790X2005000400006.

7. Montanher CC, Coradin DC, Fontoura-Da-Silva SE. Avaliação Parasitológica em Alfaces (Lactuca sativa) comercializadas em restaurantes Self-service por quilo, da cidade de Curitiba, Paraná, Brasil. Rev. Estud [Internet]. Biol. 2007 [cited 2018 Dec 18]; 29(66):63-71. https://bit.ly/3j7B7t6.

8. Neres AC, Nascimento AH, Lemes KRM, Ribeiro EL, Leitão VO, Pacheco JBP, Diniz DO, Aversi-Ferreira RAGMF, Aversi-Ferreira TA. Enteroparasitoses em amostras de alface (Lactuca sativa var. crispa) no município de Anápolis, Goiás, Brasil. Biosci. j. (Impr.) [Internet]. 2011 [cited 2018 Dec 18]; 27(2):336-41. https://bit.ly/2M2z2Uz.

9. Nomura PR, Ferreira ARM, Rafaeli RA, Augusto JG, Tatakahira VLH, Custódio LA, Murad VA. Estudo da incidência de parasitoses intestinais em verduras comercializadas em feira livre e supermercado de Londrina. Semina cienc. biol. saúde. 2015; 36(1):209-14. DOI:10.5433/1679-0367.2015v36n1supp209.

10. Andrade EC, Leite ICG, Vieira MT, Abramo C, Tibiriça SHC, Silva PL. Prevalência de parasitoses intestinais em comunidade quilombola no município de Bias Fortes, Estado de Minas Gerais, Brasil. Epidemiol. serv. saúde. 2011;20(3):337-44. DOI:10.5123/S1679-49742011000300008.

11. Santarém VA, Giuffrida R, Chesine PAF. Contaminação de hortaliças por endoparasitas e Salmonella spp. em Mercados Livres no município de Salvador/BA. Rev. Ciênc [Internet]. Méd. Biol. 2009;30(3):279-85. https://bit.ly/3tjvtjE.

12. Empresa Brasileira de Pesquisa Agropecuária. Serviço Brasileiro de Pesquisa à Micro e Pequenas Empresas. EMBRAPA/SEBRAE. Catálogo brasileiro de hortaliças: saiba como plantar e aproveitar 50 das espécies mais comercializadas no país [Internet]. Brasília: Embrapa; 2010 [cited 2018 Dec 18]; https://bit.ly/3tjyJvE.

13. Quadros RM, Marques SMT, Favaro DA, Pessoa VB, Arruda AAR, Santini J. Parasítoses em Alfaces (Lactuca sativa) de mercados e feiras de Lagos, Santa Catarina. Rev. ciênc. saúde [Internet]. 2008 [cited 2018 Dec 18]; 12(1):78-84. https://bit.ly/3ocqQPL.

14. Dufloth DB, Silva CM, Lacerda ASSPN, Silva SFV, Teixeira KTR, Monteiro TMR, Oliveira WS, Lessa CSS, Aguiar VM. Pesquisa sobre a contaminação de hortaliças por ovo de larvas de nematódeos e cistos de protozoários como método de estudo. Rev. patol. trop. 2013; 42(4):443-54.

15. Santos NM, Sales EM, Santos AB, Damasco NO, Thê S. Avaliação parasitológica de hortaliças comercializadas em supermercados e feiras livres no município de Salvador/BA. Rev. Ciênc [Internet]. Méd. Biol. (Impr.). 2009 [cited 2018 Dec 18]; 8(2):146-52. https://bit.ly/3ctrMBY.

16. Santos LM, Vieira JN, Pereira CP, Bastos CCG, Nagel AS, Villela MM. Pesquisa sobre a contaminação de hortaliças por ovos e larvas de nematódeos e cistos de protozoários como método de estudo. Rev. Estud [Internet]. Biol. 2007 [cited 2018 Dec 18]; 29(66):63-71. https://bit.ly/3j7B7t6.

17. Mesquita DR, Silva JP, Monte NDP, Sousa RLT, Silva RVS, Oliveira SS, Leal ARS, Freire SM. Ocorrência de parasitoses em alface-crespa (Lactuca sativa L.) em hortas comunitárias de Teresina, Piauí, Brasil: Ministério de Saúde; 2010 [cited 2018 May 23]. Available from: https://bit.ly/36zR8KO.
27. Fernandes NS, Guimarães HR, Amorim ACS, Reis MB, Trindade RA, Melo ACFL. Avaliação parasitológica de hortalícias: da horta ao consumidor final. Saúde. Pesq. 2015; 8(2):255-65.
28. Maciel DF, Gurgel-Gonçalves R, Machado ER. Ocorrência de parasitos intestinais em hortalícias comercializadas em feiras no Distrito Federal, Brasil. Rev. patol. trop [Internet]. 2014 [cited 2018 Dec 18]; 43(3):351-9. https://bit.ly/3pUeqmH.
29. Pires DR, Thomé SMG, Coelho PSJ, Santos HA, Azevedo LA, Frechette MF, Pires MS, Abboud LCS. Avaliação parasitológica de alfaces (Lactuca sativa) comercializadas no município do Rio de Janeiro (RJ). Semina cienc. biol. saude. 2014; 35(1):35-48. DOI:10.5433/1879-0367.2014v35n1p35.
30. Brauer AMNW, Silva JC, Souza MAA. Distribuição de enteroparasitas em verduras do comércio alimentício do município de São Mateus, Espírito Santo, Brasil. Nat. OnLine [Internet]. 2016 [cited 2018 Dec 18]; 14(1):55-60. https://bit.ly/2YdP3Ok.
31. Coelho LMPS, Oliveira SM, Milman MHSA, Karasawa KA, Santos RP. Detecção de formas transmissíveis de enteroparasitas na água e nas hortalícias consumidas em comunidades escolares de Sorocaba, São Paulo, Brasil. Rev. Soc. Bras. Bras. Med. Trop [Internet]. 2001 [cited 2018 Dec 18]; 34(5):479-82. https://bit.ly/3cxe7U.
32. Bellotto MVT, Santos Junior JE, Macedo EA, Ponce A, Galisteu KJ, Castro E, Taury LV, Rossit ARB, Machado RLD. Enteroparasitoses numa população de escolares da rede pública de ensino do Município de Mirassol, São Paulo, Brasil. RPAS. 2011; 2(1):37-44. DOI:10.5123/ S2176-62232011000100004.
33. Fontes AM, Gusson VP, Souza AA, Souza MAA. Identificação de enteroparasitas in recreations areas of elementary schools in Northern Espírito Santo, Brazil. Rev. salud pública (Bogotá). 2017; 19(6):795-9. DOI:10.15446/rsap.v19n6.57474.
34. Brasil. Ministério da Saúde. Agência Nacional de Vigilância Sanitária. Resolução de Diretoria Colegiada (RDC) no. 14, de 28 de março de 2014 [Internet]. Ministério da Saúde; 2014 [cited 2018 Dec 18]. https://bit.ly/3pIii9L.