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Agricultural practices and intestinal parasites: A study of socio-environmental risk factors associated with leafy vegetable production in La Plata horticultural area, Argentina

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A R T I C L E I N F O

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A B S T R A C T

Foodborne diseases now represent one of the most important public health problems. The objectives were to analyze the leafy vegetables and crop soil to detect parasitic species and evaluate the factors that increase the risk of parasitic contamination in the productive units in La Plata horticultural area. The study included 261 leafy vegetable and 87 crop soil samples that were processed using washing, sedimentation, and flotation techniques. Socio-environmental characteristics and agricultural practices were surveyed, and a generalized linear model was used to assess the change in parasitic prevalence with different predictor variable. The 58.6% of leafy vegetable and 31.0% of crop soil samples contained parasitic species, the most prevalent being *Blastocystis* sp. and oocysts of *Cryptosporidium* spp. Risk factors were the limited access to health, dirt roads, children and dogs circulating in crops, field cultivation, furrow irrigation and lettuce cultivation. The high prevalence of intestinal parasites in the vegetable crops was mainly associated with the conditions of structural precariousness in the production units. These results elaborated with the participation of the population, provide valuable knowledge for the planning of epidemiological programs that include environmental health as a fundamental nexus in health campaigns.

1. Introduction

Foodborne diseases are the leading cause of diarrheal diseases and over 125,000 million people die every year, mostly children (WHO, 2015); they are a major factor in reducing economic productivity in agro-livestock regions, especially for the family farming because they affect the safety of food products (FAO/WHO, 2016; Gilardi et al., 2018). The risks of intestinal parasite contamination are determined by the infecting stages of different parasitic species (i.e. hookworms) that survive in the environment for prolonged periods of time, even under adverse conditions for their development (Sedionoto et al., 2021). The distribution of intestinal parasites depends on geographic and socio-environmental factors, therefore climatic variables and soil characteristics determine the viability of parasitic forms (Camacho-Alvarez et al., 2021).

Global warming, with extreme fluctuations in temperature and changes in precipitation intensity and frequency, has increased

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geohelminthiases and the dissemination of waterborne parasitic protozoa (Ceraso et al., 2018; Kaabache and Badaoui, 2022). Also, the sanitary infrastructure, and the cultural and hygienic-food habits in rural regions promote the emergence and transmission of parasites (Ruiz-Taborda et al., 2018). *Cryptosporidium* spp., *Entamoeba histolytica* and *Ascaris lumbricoides* are the main parasites transmitted by fresh vegetables globally (FAO/WHO, 2014). In Latin America, studies on fruit and vegetables in markets in Colombia, Venezuela, Peru and Brazil found species such as *Cryptosporidium* spp., *Giardia* spp., *Blastocystis* sp., *Entamoeba coli*, *A. lumbricoides* and hookworms (Benites-salcedo et al., 2019; Devera et al., 2021; de Farias et al., 2021; Devera et al., 2006; Guerrero, 2015). In addition, *Toxoplasma gondii, Cystoisospora* spp., *Balantidium coli* and *A. lumbricoides*, species of sanitary risk, have also been detected in crops in these regions (López Ureña et al., 2022; Puig-Peña et al., 2013; Rodríguez et al., 2015). Food crop application of Good Agricultural Practices (GAP) and integrating the integration of community needs into local health care management are key aspects to limit the spread of regional zoonose (FAO/WHO, 2016; Casallas-Murillo, 2017; Falcone, 2021).

Family farming is a strategic and sustainable sector for hunger eradication in Latin America, the Caribbean and the world (Devera et al., 2021). In Argentina, it produces >80% of the food supply protected by law 27,118 on the historical reparations of family farming. In this context, La Plata horticultural area (Buenos Aires province) has grown significantly in "leafy vegetable" production (SENASA, 2020; Shoaie Baker and García, 2021). These agriculture was characterized by the figure of "mediería" (Law 13,246), an agricultural partnership contract in which the landowner contributes a rural farm to agricultural families who settle on the farm under precarious working and living conditions. The relationship between human and environmental health suggests the need for an integrated approach that supports food safety in vegetable production (Eandi et al., 2021; FAO/OIE/WHO, 2009).

In Argentina surveillance systems of foodborne and waterborne diseases have focused on the detection of bacteria and viruses and there are few controls for identifying parasitic species, a situation that requires special attention due to the increased globalization of trade in fresh products (Rivero et al., 2020). An analysis carried out on migrant horticultural families settled in the rural area of La Plata, south of Buenos Aires city (Argentina), showed a parasitic prevalence of 79.1% and these populations were exposed to risk factors of parasitic infection such as lack of piped water, and critical overcrowding (Falcone et al., 2020). Our study is based on the hypothesis that horticultural practices carried out under inadequate sanitary conditions increases the risk of parasitic contamination. The objectives were to analyze the leafy vegetables and crop soil to detect parasitic species and evaluate the factors that increase the risk of parasitic contamination in the productive units.

2. Materials and methods

2.1. Study area and population

La Plata horticultural area (34°56′S, 58°02′W) is located in a rural area south of Buenos Aires (Buenos Aires province, Argentina). The climate is temperate with an average temperature of 16 °C, 77% annual relative humidity and 1040 mm annual average rainfall (Imbellone and Mormeneo, 2011). The predominant soil type is argiudoll, silty-loam in texture with abundant organic matter making it optimal for vegetable cultivation (Hurtado et al., 2006). The production is carried out in greenhouses in areas between 2 and 15 ha using few high-yielding, high-input chemically-dependent varieties. La Plata horticultural area is located within the region surveyed with unmet basic needs (INDEC, 2010). The fieldwork was carried out in Abasto, Melchor Romero, Angel Etcheverry, and El Peligro neighborhoods in the southwest of La Plata horticultural area (Fig. 1), which has the highest production. The region is inhabited by families who emigrated from the north of the country or from neighboring countries, mostly from Bolivia, who worked the land with

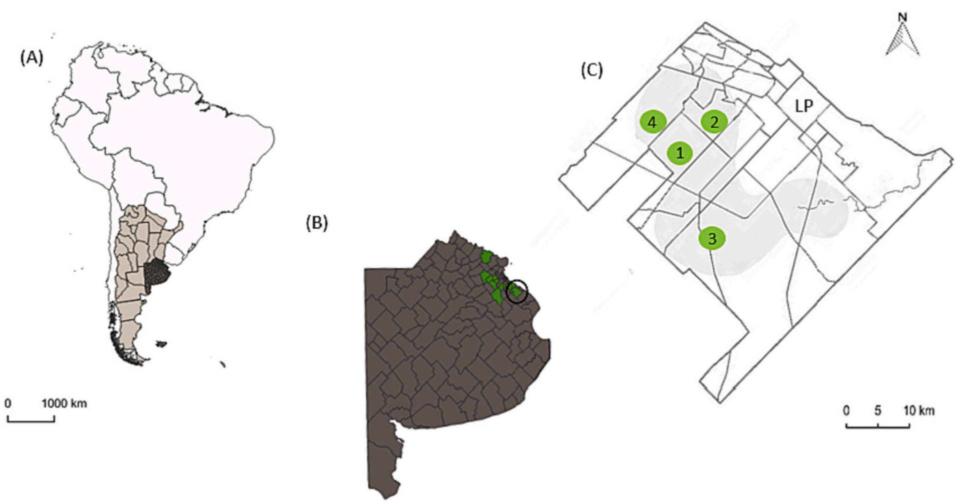

Fig. 1. Geographical location of Argentina in South America and Buenos Aires province in Argentina (A). The La Plata horticultural area in Buenos Aires province (B). District of the La Plata horticultural area sampling site (C). LP: La Plata city; 1: Abasto (sampling site 1), 2: Melchor Romero (sampling site 2), 3: Angel Etcheverry (sampling site 3), 4: El Peligro (sampling site 4).

peasant farming practices and currently carry out peri-urban-rural agriculture with greenhouse technology.

2.2. Study design

A cross-sectional and descriptive study was conducted consecutively in March–June and August–November in 2017–2020. The sample selection was non-probabilistic and largely determined by voluntary participation of horticultural families who gave written consent in parasitological workshops at educational and public health centers, and also in greenhouses. The collection of leafy vegetables and soil samples was undertaken randomly in greenhouses of families in which the presence of at least one intestinal parasite had been observed in a previous study (Falcone et al., 2020).

2.3. Sample collection

The study was carried out in four productive sites (Fig. 1), and included 261 leafy vegetable and 87 crop soil samples collected in greenhouses in a 7 × 10 m zigzag transect (Araújo e Silva Ferraz et al., 2016; FAO, 2013). Four to seven plants (approx. 800 g) of chard (54/261), spinach (24/261), lettuce (168/261), chicory (9/261) and arugula (6/261) were collected. These samples were taken along transect at equidistant points 2 m apart, depending on availability (plant size and type) at sampling time. Also, 20 crop soil subsamples (400 g) were taken with a soil sampler (up to 5 cm in depth) along transect at equidistant points 1 m apart. Each sample was placed in a plastic bag and labeled, separating the leaves from the roots, and incorporating the soil from the roots with the soil samples (Mehrnejat et al., 2018; Melín-Coloma et al., 2016), and then they were transported in a cold box for immediate analysis.

2.4. Parasitological analysis

To increase the possibilities of parasite detection in environmental samples, a bibliographic analysis on parasite recovery techniques in leafy vegetables was undertaken.

The vegetable leaves were weighed, divided into three parts (approx. 200 g c/u) and three washing techniques were performed: technique #1 (Devera et al., 2006); technique #2 (Pérez-Cordón et al., 2008) and technique #3 (Cazorla et al., 2009). According to the procedure of the technique #1, the vegetable leaves were divided into two subsamples (A and B). The A subsamples were placed in a plastic bag with distilled water and were agitated manually for 30 s. The B subsamples were washed manually in a beaker with distilled water. In the technique #2, the vegetable leaves were washed with saline solution in an orbital shaker (Model T5–1.000) in agitation for 2 h. In the technique #3 the leaves were cleaned with a brush (No. 16–18) for 2 min in distilled water.

Each resulting liquid was filtered separately through double gauze. In the technique #1, the filtrate was concentrated by centrifugation at 514*g*ravity (g), while in the technique #2 it was left to settle for 24 h. In the technique #3 the resulting filtrate was allowed to decant for 24 h and centrifuged at 289 *g* for 5 min. The sediments were concentrated using the modified Ritchie and Sheather coproparasitological techniques (Girard de Kaminsky, 2014; WHO, 2019).

In addition, the crop soil and the soil recovered from the roots were left to dry in the open air and two techniques were performed: technique #1 (Shurtleff and Averre, 2000) and technique #2 (Gnani Charitha et al., 2013). The crop soil and soil from the roots were left in the open air. In the technique #1 the soil was homogenized in distilled water. The supernatant was filtered with a pore diameter of 130 μm (A solution), and then with a 55 μm pore diameter (B solution). The A and B solutions were centrifuged for 10 min at 514*g*. The B solution concentrate was poured onto a filter of 25 μm pore diameter, which was taken from the filter surface (C solution) and centrifuged for 5 min at 289*g*. In the technique #2, the soil was added to Tween 80 0.1% solution and was manually agitated for two minutes. The solution was filtered with a filter of 1 mm pore diameter. Next, two series of washes with distilled water were performed and it was concentrated with sodium nitrate (δ: 1.4 g/ml).

Observations were made in triplicate with an optical microscope at 100×, 400× and 1000× magnifications. The modified Ziehl-Neelsen technique and non-permanent stains were used. Identification of the parasitic elements (eggs/larvae/cysts/oocysts) was based on their measurements and morphological characteristics (Girard de Kaminsky, 2014; WHO, 2019; Unzaga and Zonta, 2018).

2.5. Socio-environmental characteristics and agricultural practice analyses

Semi-structured, non-invasive questionnaires were developed to infer the degree of intestinal parasite risk to which the productive units were exposed. The surveys were developed using bibliographic information on living conditions and production in families living in rural areas and taking into account the characteristics of sanitary infrastructure observed in this region (FAO/WHO, 2016; Falcone et al., 2020; INDEC, 2010; Cursi and Sartor, 2016). Information observed in situ was also incorporated into the survey (e.g. nearness of crop to houses, drainage ditches, and animal production) [Eq. (A.1)]. The questionnaires were conducted by interviewing with producer families, which included questions about access to health and education, hygiene habits and domestic animals, as well as agricultural practices and other environmental characteristics of interest.

Workshops with farming families and collaborative mapping (Ortiz and Borjas, 2008) were conducted to understand crop management practices before, during and after production, and study the social determinants of local health related to vegetable production. Subsequently, information leaflets were produced with the community on achievable hygienic practices to mitigate parasitic contamination of crops. Several workshops were carried concerning the treatment to improve drinking water as well as the washing and hygiene of vegetables before fresh consumption according to the accesses of each participating family [Eq. (B.1)].

2.6. Statistical analyses

A sample was considered positive when any parasite species was observed by any methods. The association between species pairs was evaluated using the Chi square test ($\chi 2$) and Fisher's exact test was performed for expected values lower than five. To identify those variables related to parasite prevalence, we performed two analyses in subsequent steps: (1) a multiple correspondence analysis (MCA) to explore the association between the variables measured (SI) and identify a potential set of predictors of prevalence, and (2) generalized linear models (GLMs) of prevalence as a function of those variables selected in step (1). Multiple correspondence analysis is a multivariate technique for visualizing datasets of categorical variables. Like other multivariate methods, MCA is an exploratory dimension-reduction technique that aims to reduce the dimensionality of a dataset, while preserving as much information as possible (Palacio et al., 2020). To select those variables related to the prevalence to be used as predictors in the GLM, individual ANOVAs with one factor was carried out for each dimension (function dimdesc from the R package FactoMineR; i.e., the coordinates of the observations are explained by the categorical variable (Husson et al., 2011). Those factors that significantly ($p < 0.05$) related to a dimension for which prevalence also related to, were considered as potential predictors of prevalence (number of people parasitized / number of people in the family).

We then performed model selection based on the Akaike's Information Criterion corrected for small sample sizes (AICc), which combines the level of data support with the complexity of the model measured in parameter quantity (Anderson and Burnham, 2020). To this end, we developed a theoretically meaningful a priori set of GLMs by combining hypotheses that explain prevalence (binomial error distribution, logit link function). A total of 33 models were used as competing models, including a null model with no covariates. Models with less than two units of differences in their AICc values were considered likely explicative. We computed Akaike weights, which describe the probability that a particular model is the best model given the candidate set of models (Burnham et al., 2011). To assess the importance of predictors while limiting uncertainty from model selection, we used a model averaging approach (Anderson and Burnham, 2020). A full-model averaging approach was used, in which parameters that were not included in a model were set to zero and included when averaging the coefficient estimates (Grueber et al., 2011). The relative importance of each predictor was computed as the sum of Akaike weights over all models that contain the term (Anderson and Burnham, 2020). All statistical analyses were processed using R software (version 4.0.2) (Team Core, RA, 2020), using the packages FactoMineR (Barton, 2020) and MuMIn (Le et al., 2008).

2.7. Ethics approval and consent to participate

The study was carried out without affecting the participants' physical, psychic, or moral integrity and, protecting their identity. Informed consent was obtained from the participants and the study complied with all regulations. It was conducted according to the principles in the Universal Declaration of Human Rights (1948), the ethical standards established by the Nüremberg Code (1947), the Declaration of Helsinki (1964) and its successive amendments. The Working Protocol of the research group was endorsed by the Central Consultative Committee of Bioethics of Universidad Nacional de La Plata (Exp. No. 100–20,120/18) and approved by the institutional committee for the review of research protocols (PIO CONICET-UNLP). All participants signed the informed consent form before participating in the study and horticultural families have given written consent for the publication of the analyzed data in this research.

All families received their environmental parasitological results in written certificates. Also, workshops were held in the greenhouses to develop effective preventive strategies based on the species found and the living conditions of each family.

Table 1
Prevalence (%) of parasite species detected in vegetable and soil samples in La Plata horticultural area.

Microorganism	Vegetables $N = 261$ (Prevalence, %)[a]	Crop soil $N = 87$ (Prevalence, %)[b]
Protozoa		
Cryptosporidium spp.	60 (39.1)	0 (0.0)
Giardia spp.	6 (3.9)	2 (5.7)
Entamoeba coli	15 (9.8)	5 (6.8)
Endolimax nana	9 (5.8)	3 (3.4)
Blastocystis sp.	63 (41.1)	21 (17.2)
Helminths		
Oxyuridae	6 (3.9)	0 (0.0)
Ascaris lumbricoides	9 (5.8)	3 (1.1)
Hookworms	12 (7.8)	4 (5.6)
Samples parasitized	153 (58.6)	27 (31.0)

[ab]Prevalence estimated in relation to total number analyzed in the greenhouses (n = 87).

3. Results

3.1. Parasitological analysis

3.1.1. Vegetable samples

Of the total leafy vegetable samples analyzed, 58.6% (153/261) showed parasitic contamination. All five vegetables were contaminated by parasite species, and 64.7% ($N = 99$) of the positive samples were from lettuce, followed by chard (25.5%), spinach (3.9%), chicory (5.8%) and arugula (3.9%). Lettuce was the predominant crop in the greenhouses of farming families. The most prevalent species were *Blastocystis* sp. and oocysts of *Cryptosporidium* spp., detected by all three techniques. Other protozoa detected were *E. coli*, *Endolimax nana* and *Giardia* spp. Also, *A. lumbricoides*, hookworms, and eggs of Oxyuridae were found (Table 1).

Statistically significant association were observed between *Blastocystis* sp./*Cryptosporidium* spp., *Cryptosporidium* spp./hookworms and *Blastocystis* sp./ *A. lumbricoides* ($p < 0,05$).

3.2. Crop soil samples

At least one parasite species was found in 31.0% (27/87) of crop soil samples, the most frequent being *Blastocystis* sp. (17.2%). *E. coli*, *Giardia* spp., *E. nana* and geohelminths such as hookworms, and also *A. lumbricoides* were detected (Table 1). Other free-living nematodes, such as *Diploscapter coronata,* were found. Statistically significant associations were not observed between species.

3.3. Socio-environmental characteristics and agricultural practices analyses

The questionnaire results (87) showed that half of the horticulturists, 74.7% Bolivians and 25.3% Argentinians had received prior information about intestinal parasites (49.4%), in general they had access to health (85.1%). Most families did not own the domestic-production units (88.5%) and a percentage of the profits was part of the rental contract. Other factors of interest were that most horticultural families had their houses and septic tanks (wastewater disposal) <20 m from the crops (67.8% and 69.0%, respectively), and 33.3% of the families kept animals, such as chickens (29.9%) and pigs (4.6%), for family consumption (26.4%) which were housed <20 m from the crops. All the families had dogs; the children and dogs frequently circulated in the crops (88.5% and 82.8%, respectively). They incinerated their planting waste (74.0%) and their production units were frequently flooded (81.6%) (Table 2).

All horticulturists harvested in greenhouses, and more than half also did field cultivation. One third of the families hired third parties for crop cultivation and mainly lettuce was grown (64.7%). Most horticulturists reported hand washing before and after

Table 2
Socio-environmental characteristics of interest in the production of leafy vegetables.[a]

Variables	Frequency	
	n	%
Horticulturist		
Bolivian	65	74.7
Argentine	22	25.3
Access to health	74	85.1
Knowledge of the problems prior to intervention	43	49.4
Lodging or house tenure status		
House owner	10	11.5
Lease holder	77	88.5
Housing with walls of fired-brick masonry	20	23.0
Housing with walls of sheet and wood	67	77.0
Housing with floors of concrete or other	82	94.3
Housing with dirt floors	5	5.7
Paved road	19	21.8
Dirt road	68	78.2
Environmental surroundings of the production unit		
Outside tap for water supply	75	86.2
Bathroom installation outside the house	69	79.3
Bathroom with discharge	2	2.3
Flooding of the greenhouses	71	81.6
Incineration or burial of waste	30	74.0
Nearness of crop to house <20 m	59	67.8
Nearness of crop to septic tank <20	60	69.0
Nearness of crop to drainage ditches <20	60	69.0
Nearness of crop to animal production <20	29	33.3
Animal production	23	26.4
Chickens	26	29.9
Pigs	4	4.6
Circulation of dogs in the greenhouses	72	82.8

[a] Frequency was estimated in relation to the total population with horticulture practice data ($n = 87$).

growing vegetables (77.0%) and disinfected the soil before cultivation (75.9%). The chicken litter (80.4%) was the most frequently used fertilizer. The irrigation water came from protected wells, and in greenhouse crops families using drip (98.9%), while in the field they used furrow (57%) irrigation. At the post-harvest stage, most horticulturists stored their production (81.6%) and soaked leafy vegetables with non-chlorinated water (89.6%) (Table 3).

3.4. Statistical analyses

Based on the cumulative percentage criterion, 16 dimensions of the MCA were retained (71.5% of accumulated variability) to identify potential predictors to be included into the GLM candidate models (see below). A total of 16 variables were related to these dimensions for which prevalence also related to with the three first. The variables were sampling site, dirt roads, house building material (wall and floor), incineration or non-sanitary burial, septic tanks, flooding of greenhouses, distance of house and drainage ditch from the crop <20 m, limited access to health, prior knowledge on intestinal parasites, dog circulation in crops, soil disinfection, lettuce field cultivation, irrigation water from protected wells, guano fertilizer, storage of production, and non-chlorinated water for soaking leafy vegetables. To strengthen the study with the One Health paradigm, the variables of irrigation water type and child circulation through crops were considered.

The best GLM indicated that the risk factors for parasites in the soil and vegetable samples were the lack of access to health, dirt roads, children and dogs circulating in crops, field cultivation, furrow irrigation and lettuce cultivation. The odds ratio was reduced by 40% with access to health (OR = 0.6; 95% IC: 0.30–1.08) and by 10% for crops other than lettuce (OR = 0.9; 95% IC: 0.59–1.41). However, it was incremented by 10% in field cultivation and furrow irrigation (OR = 1.1; 95% IC: 0.74–1.75; OR = 1.3; 95% IC: 0.86–2.04, respectively) and also in dirt roads, and dogs and children in crops (OR = 1.1; 95% IC: 0.69–1.93; OR = 1.0; 95% IC: 0.60–1.71; OR = 1.1; 95% IC: 0.60–2.22, respectively) (Table 4).

In the multivariate GLM, the variables significantly associated with the species composition found were limited access to health, no prior information on intestinal parasites, dirt roads, lettuce crops, chicken litter fertilizer, production storage, incineration or burial of waste, and drainage ditches <20 m from crops. In the univariate analyses, *Cryptosporidium* spp. were significantly associated with basic education, *E. coli* with dirt roads and *Blastocystis* sp. with limited access to health and no prior information on intestinal parasites ($p < 0.05$).

4. Discussion

In Argentina, studies on agricultural production are scarce and analysis in leafy vegetable production units is of utmost importance for understanding parasitic transmission and the level of food contamination. This study provides knowledge on the quality of leafy vegetables and soil in the southwest of La Plata horticultural area, a highly competitive area at national and regional level. In addition, the present study provides community interventions that, in addition to providing knowledge for the control of parasitic infections in

Table 3
Variables of horticultural practices in horticultural populations.[a]

Variables	Frecuency	
	n	%
The family cultivate the soil	61	70.3
Third parties hired for cultivating the soil	26	29.7
Circulation of children in the greenhouses	77	88.5
Wearing gloves (fruits)	45	51.7
Washing hands before and after work	67	77.0
Greenhouse cultivation	87	100
Field cultivation	54	62.1
Lettuce crop	99	64.7
Other crops	54	35.3
Disinfection of the soil	66	75.9
Disinfection with methyl bromide (Ch_3Br)	60	69.0
Disinfection by other methods	7	0.8
Irrigation water from tanks	1	1.1
Irrigation water from protected wells	87	100
Furrow irrigation system	50	57.5
Drip irrigation system	86	98.9
Fertilized with chicken litter	70	80.4
Fertilized with guano	12	13.7
Fertilized with manure	20	23.0
Other fertilizers	3	3.4
Production storage	71	81.6
Soaking leafy vegetables	81	93.0
Soaking leafy vegetables with water	78	89.6
Soaking leafy vegetables with chlorinated water	3	3.4

[a] Frequency was estimated to the total population with horticulture practice data (n = 87).

Table 4
Risk factors associated with parasites in vegetable and soil samples in La Plata horticultural area[a].

Risk factors	β	SE	OR
Limited access to health	−0,6	0,3	0,6
Children circulating through the crop	0,1	0,3	1,1
Dogs circulating through the crop	0,0	0,3	1,0
Dirt roads	0,1	0,3	1,1
Furrow irrigation	0,3	0,2	1,3
Field cultivation	0,1	0,2	1,1
Lettuce crop	−0,1	0,2	0,9

β (beta): regression coefficient; SE: standard error; OR: odds ratio. [a]Other models were considered but only the ones selected are shown.

the leafy vegetable production chain, involve the community in the construction of local health by strengthening action plans for regional food security.

Parasitic contamination detected in leafy vegetables was high (58.6%) being double that in the soil (31.0%): the same species were identified, except that eggs of Oxyuridae and oocysts of *Cryptosporidium* spp. were only observed in the leafy vegetable samples. Studies carried out on vegetables in Argentine production units showed a prevalence between 23.8% and 47.7%, the lowest being in the northeast of La Plata horticultural area (Costamagna et al., 2002; Rea et al., 2004; Zonta et al., 2016). Studies on crops in Cuba and Brazil also showed a wide range in prevalence (6.0%–55.5%) (Puig-Peña et al., 2013; Ferreira-Pires et al., 2019). In those studies, the irrigation sources used for cultivation were exposed to parasitic contamination due to anthropogenic activities around the productive environments, and the prevalence observed could be related to the different forms of settlements of the populations analyzed. Moreover, a study carried out in Bolivia emphasized the implications that the vegetable leaf characteristics had on the presence of pathogenic microorganisms (Rodríguez et al., 2015). Most leafy vegetables have overlapping, flexible leaves that could contain irrigation water for longer than the soil and so protect the parasitic species detected.

In the present study, *Blastocystis* sp. was the most frequent species in the leafy vegetable samples and crop soil (41.1% vs. 17.2%, respectively). A high prevalence of oocysts of *Cryptosporidium* spp. was also observed in leafy vegetables, and similar percentages of these genres have been detected in countries from different regions, such as Bolivia and Syria (Ferreira-Pires et al., 2019; Al and Aboualchamat, 2020). *Cryptosporidium* spp., *Giardia* spp. and *Blastocystis* sp. are the most frequently detected species in agricultural production. *Cryptosporidium* spp. and *Giardia* spp. have been reported together in epidemiological outbreaks caused by water and food contamination due to their low infective dose and high resistance to chlorine (Hernández-Gallo et al., 2018). However, in the present study *Giardia* spp. was detected at a low frequency, with similar values in the soil and vegetable samples within the range reported for the region (Rivero et al., 2020; Juárez and Rajal, 2013). The high presence of *Blastocystis* sp. and *Cryptosporidium* spp. in leafy vegetables could be mainly due to the exposure of manure (chicken litter), and circulation dogs. Also, species such as *E. coli* were found at a low frequency in both soil and vegetable samples. *E. coli* were identified in vegetables in Bolivian and Venezuelan markets, whereas *Giardia* spp. and *Cryptosporidium parvum* were observed in Peru, but not all the species detected in this study were found (Benites-salcedo et al., 2019; Devera et al., 2021; Rodríguez et al., 2015). It is also important to highlight that in the coproparasitological analyses carried out on the families living and working in these productive units, *Blastocystis* sp., *Giardia* spp. and *E. coli* were the most prevalent species (58.9%, 24.0% and 26.3%, respectively) (Falcone et al., 2020). Therefore, the high frequency of protozoa found in greenhouses could mainly be related to the use of groundwater for irrigation contaminated with fecal matter, as most families had septic tanks near the crops and frequent flooding overwhelmed the drainage ditches (Ceraso et al., 2018).

Species, such as hookworms and *A. lumbricoides* were detected in low frequencies in the production units. Geohelmints are endemic to the north of Argentina being resistant to extreme heat and desiccation (Rivero et al., 2017). Moreover, La Plata horticultural area show alterations in soil infiltration and chemical composition due to the impact of production with greenhouse technology (Shoaie Baker and García, 2021). However, it was observed that organic fertilizer are located near the greenhouses, which were frequently left open, being exposed to potential vectors, such as rodents, dogs and cats. In relation to this, *Ancylostoma caninum* species and hookworms were identified in the pet dogs of these families, as well as in other studies on parasitosis in dogs in rural regions (Falcone et al., 2020; Enríquez et al., 2019; Hawdon and Wise, 2021). Therefore, the presence of geohelminths may be due the presence of dogs in crops, favoring parasitic dispersal.

Eggs of Oxyuridae have been detected at low frequency in both the soil and leafy vegetables, and it was also observed in another productive region of Argentina (Team Core, RA, 2020). This can be explained by the closeness of the domestic and productive units and values of prevalence of *Enterobius vermicularis* (26.0%) detected in the families analyzed in this region (Falcone et al., 2020). However, the possibility that these were Oxyuridae nematode eggs present in rodents was not excluded (Miño et al., 2012). Molecular studies are required for the correct identification of this species. On the other hand, it is worth mentioning that *D. coronata* was found in the crop soil and Salas et al. (2017) related its presence to hygiene practices during harvesting.

The GLMs determined that the risk of intestinal parasites was reduced considerably when horticultural families had access to health. Additionally, lack of previous information on intestinal parasites was significantly associated with the species composition found, particularly with the most prevalent species in this analysis (*Blastocystis* sp.). The health education and hygiene of the producers are among the most relevant issues for food safety during production (FAO, 2013; FAO, 2014). A study conducted by Zuta Arriola et al. (2019) showed that educational talks on health promotion focused on these infections are the best way to adopt the necessary hygienic-sanitary measures to reduce the prevalence and incidence of parasites and other pathogenic microorganisms.

It was observed that the circulation of dogs in crops increased the risk of parasite species in the production units. In addition, it is worth noting that incineration or burial of waste and drainage ditches near crops, were associated with the species detected. In this context, children are more exposed to the transmission of intestinal parasites due to their close relationship with the environment. The behavior of dogs and their relationship with children in a contaminated environment could probably explain the presence and dispersal of parasites in the crops (FAO/OIE/WHO, 2009).

The risks of parasitic contamination increased with furrow irrigation and chicken litter fertilizer, and these variables were associated with the species composition found. These results are consistent with other studies that indicate that untreated irrigation water and inadequately processed organic fertilizers are the practices that most influence a reduction in the sanitary quality of vegetables (Puig-Peña et al., 2013; Tenesaca et al., 2019). In this respect, deficiencies in septic tanks construction (i.e. casing, cementing) and groundwater boreholes might cause contamination of the water extracted. In addition, it was reported in the surveys that floods were frequent and that the surrounding road were dirt, a variable that was a risk factor for parasitoses. Lettuce cultivation and field cultivation also increased the risks of parasitic contamination. In this sense, the relationship between these conditions (dirt road, field cultivation) and the morphology and arrangement of lettuce leaves (*Lactuca sativa*), a typical species of the region, could explain the superficial contamination observed (Chávez-Ruvalcaba et al., 2021).

On the other hand, it is important to highlight that the storage of produce was significantly associated with the species composition detected. At this stage, the vegetables are often soaked in untreated water, and the produce is stored in open sheds exposing it to a greater possibility of contamination from the circulation of people and animals. Several studies showed that the application of GAP during harvest and postharvest is essential for limiting the transmission of geohelminths and species associated with water contamination (Rivero et al., 2017; Salas et al., 2017). Therefore, these results show the importance of access to drinking water, sanitation and hygiene (WASH), and interventions situated in the socio-cultural diversities to mitigate foodborne infections (Casallas-Murillo, 2017; FAO/OIE/WHO, 2009; Ortiz and Borjas, 2008).

Unhealthy environmental conditions and lack of knowledge about the sources of parasitic infection in the horticultural families were the determining factors of parasitic contamination in the leafy vegetable crops. Moreover, this scenario showed that groundwater may become contaminated when horticultural families live in conditions of structural precariousness, and the impact these conditions have on agricultural practices and food security. Further research is needed to identify the precise source of contamination, as well as sanitary education campaigns on intestinal parasites. Therefore, these results, elaborated with the participation of the population, provide valuable knowledge for the planning of epidemiological programs that include environmental health as a fundamental nexus in health campaigns for the control of parasitic diseases in the food, in the pursuit of a concept of One Health that contributes to food sovereignty.

4.1. Study limitations

The low frequencies of parasite species observed in vegetable and soil samples, the low number of samples obtained from chicory and arugula, and the increase in rents caused families to frequently change production units did not allow for a broader scope of analysis, as increase the sample size or performing other more specific detection techniques (PCR). Despite those limitations, the parasitological results of this study revealed important information on environmental health in leafy vegetable crops in the southwest of La Plata horticultural area.

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CRediT authorship contribution statement

Andrea Celina Falcone: Conceptualization, Methodology, Investigation, Formal analysis, Visualization, Writing – original draft, Writing – review & editing. **María Lorena Zonta:** Conceptualization, Visualization, Writing – review & editing. **Juan Manuel Unzaga:** Conceptualization, Methodology, Investigation, Writing – review & editing. **Graciela Teresa Navone:** Conceptualization, Investigation, Writing – review & editing, Supervision, Project administration, Funding acquisition.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data and materials are available at the La Plata environmental observatory: http://omlp.sedici.unlp.edu.ar/dataset/horticultores-platenses-parasitosis-y-saneamiento-ambiental.

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Appendix A. Supplementary data

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References

Al, Nahhas S., Aboualchamat, G., 2020. Investigation of parasitic contamination of salad vegetables sold by street vendors in city markets in Damascus, Syria. Food and Water. Paras. 21, e00090.
Anderson, D., Burnham, K., 2020. Model Selection and Multi-Model Inference, , Second63. Springer-Verlag, NY, p. 10.
Araújo e Silva Ferraz, G., Silva de Oliveira, M., Moreira da Silva, F., Carvalho Avelar, R., Castro da Silva, F., Ferreira Ponciano Ferraz, P., 2016. Methodology to determine the soil sampling grid for precision agriculture in a coffee field. DYNA 84, 316–325.
Barton, K., 2020. MuMIn: Multi-Model Inference. R package Version 1.43.17. Retrieved from. https://cran.r-project.org/package=MuMIn.
Benites-salcedo, D., Castillo-valdivieso, C., Jara-Campos, C., 2019. Contaminación parasítica de hortalizas de consumo humano expendidas en mercados de Trujillo. Perú. Revi. de Investig. Científica REBIOL 39 (1), 41–49.
Burnham, K.P., Anderson, D.R., Huyvaert, K.P., 2011. AIC model selection and multimodel inference in behavioral ecology: some background, observations, and comparisons. Behav. Ecology and Sociob. 65 (1), 23–35.
Camacho-Alvarez, I., Goyens, P., Luizaga-López, J.M., Jacobs, F., 2021. Geographic differences in the distribution of parasitic infections in children of Bolivia. Paras. Epidemiol. and Control 14, e00217.
Casallas-Murillo, A.L., 2017. La medicina social-salud colectiva latinoamericanas: una visión integradora frente a la salud pública tradicional. Revista Ciencias de la Salud 15 (3), 397–408.
Cazorla, D., Morales, P., Chirinos, M., Acosta, M.E., 2009. Evaluación parasitológica de hortalizas comercializadas en Coro estado Falcón Venezuela. Bolet. de Malariol. y Salud Ambient. 49 (1), 117–125.
Ceraso, C., Retola, G., Unazaga, J.M., 2018. La ciudad de las ranas. Construcción social del riesgo hidrico post inundación de la cuidad de La Plata. EDULP 262.
Chávez-Ruvalcaba, F., Chávez-Ruvalcaba, M.I., Santibañez, K.M., Muñoz-Carrillo, J.L., Coria, A.L., Martínez, R.R., 2021. Foodborne parasitic diseases in the Neotropics–a review. Helminthol. 58 (2), 119–133.
Costamagna, S.R., García, S., Viscicarelli, E., Casas, N., 2002. Epidemiología de las parasitosis en Bahía Blanca (Provincia de Buenos Aires) Argentina-1994/1999. Parasitol. Latinoam. 57, 103–110. Retrieved from. https://doi.org/10.4067/S0717-77122002000300004.
Cursi, N., Sartor, A., 2016. Guía de Formación en Buenas Prácticas Agrícolas Para Hortalizas: Un Maravilloso Viaje por las Buenas Prácticas Agrícolas en Hortalizas: Mis Amigos y yo, 112. Instituto Interamericano de Cooperación para la Agricultura (IICA).
de Farias, Santos F., Gomes, D.S., de Amorim Santos, I.G., 2021. Enteroparasitos em Alface (*Lactuca Sativa L.*) comercializada em uma feira livre de um Município Alagoano. Divers. Jour 6 (4), 3882–3889.
Devera, R., Blanco, Y., González, H., Lisdet, G., 2006. Parásitos intestinales en lechugas comercializadas en mercados populares y supermercados de Ciudad Bolívar, Estado Bolívar, Venezuela. Rev. La Soc. Venez. Microbiol.
Devera, R.A., Martínez, L.R.C., Sadek, M.A.Z., 2021. Formas parasitarias de interés médico en muestras de lechugas comercializada en el municipio Caroní, estado Bolívar, Venezuela. Rev. Venez. Salud Public 9 (1), 20–36.
Eandi, M.A., Dezzotti, L., Butinof, M., 2021. Exposición a plaguicidas y cuidados de la salud en la horticultura periurbana: el caso del Cinturón Verde de la Ciudad de Córdoba, Argentina. Ciência & Saúde Coletiva 26, 1575–1584.
Enríquez, G.F., Macchiaverna, N.P., Argibay, H.D., Arias, L.L., Farber, M., Gürtler, R.E., Garbossa, G., 2019. Poliparasitismo y parásitos zoonóticos en perros de una zona rural del Chaco argentino. Parasitol. Veterinaria: estudios e informes regionales 16, 100–287.
Falcone, A.C., 2021. Parasitosis Intestinales en Poblaciones del cinturón Hortícola Platense, Buenos Aires: Factores Socio-Económicos y Ambientales en la Evaluación de Estrategias de Control. Universidad Nacional de La Plata. Doctoral dissertation.
Falcone, A.C., Zonta, M.L., Unzaga, J.M., Navone, G.T., 2020. Parasitic risk factors in migrant horticultural families from Bolivia settled in the rural area of La Plata, Buenos Aires, Argentina. One Health 11, 100179. https://doi.org/10.1016/j.onehlt.2020.100179.
FAO, 2013. El manejo del suelo en la producción de hortalizas con buenas prácticas agrícolas.
FAO, 2014. Agricultura Familiar en América Latina y el Caribe.
FAO/OIE/WHO, 2009. Un Mundo, Una Salud.
FAO/WHO, 2014. Segunda Conferencia Internacional sobre Nutrición de la FAO y la WHO (CIN 2).
FAO/WHO, 2016. Manipuladores del Alimentos. Retrieved from. www.fao.org./3/a-i5896s.pdf.
Ferreira-Pires, E.P., Andrade Avelar, L., Leite Dias, J.V., Mota Queiroz, D.R., Gomes Murta, N.M., Bahia de Oliveira, G.H., Rodrigues, Martins H., 2019. Intestinal parasitosis and environmental contamination with helminths and protozoa in a Quilombola community of Southeast Brazil. Rev. Cubana Med. Trop. 71 (1), 1–13.
Gilardi, G., Garibaldi, A., Gullino, M.L., 2018. Emerging pathogens as a consequence of globalization and climate change: leafy vegetables as a case study. Phytopathol. Mediterr. 57 (2), 146–152. https://doi.org/10.14601/Phytopathol.
Girard de Kaminsky, R., 2014. Manual de parasitología: técnicas para laboratorio de atención primaria de la salud y para el diagnóstico de las enfermedades infecciosas desatendidas. https://doi.org/10.1017/CBO9781107415324.004. OPS 2012.
Gnani Charitha, V., Rayulu, V., Kondaiah, P., Srilatha, C., 2013. Comparative evaluation of flotation techniques for the detection of soil borne parasites. J. Parasitol. 37, 260–263. https://doi.org/10.1007/s12639-012-0176-2.
Grueber, C.E., Nakagawa, S., Laws, R.J., Jamieson, I.G., 2011. Multimodel inference in ecology and evolution: challenges and solutions. J. Evol. Biol. 24 (4), 699–711.
Guerrero, J.V., 2015. Enteroparásitos y factores de riesgo relacionados en frutas y hortalizas de los expendios públicos y privados de la ciudad de Cartagena, p. 10. https://doi.org/10.3923/ijss.2017.32.38.
Hawdon, J.M., Wise, K.A., 2021. Ancylostoma caninum and other canine hookworms. In: Dog Parasites Endangering Human Health. Springer, Cham, pp. 147–193.
Hernández-Gallo, N., Hernández-Flórez, L.J., Cortés-Vecino, J.A., 2018. Criptosporidiosis y «Una Salud». Rev. Salud Pública 20, 138–143.
Hurtado, M.A., Giménez, J.E., Cabral, M.G., Da Silva, M., Martínez, O.R., Camillón, M.F.C., Lucesoli, H., 2006. Análisis Ambiental del Partido de La Plata, Aportes al Ordenamiento Territorial. Instituto de Geomorfología y Suelos.
Husson, F., Lê, S., Pagès, J., 2011. Exploratory Multivariate Analysis by Example Using R. 15. CRC Press, Boca Raton.
Imbellone, P., Mormeneo, L., 2011. Vertisoles hidromórficos de la planicie costera del Río De La Plata, Argentina. Ciencia del Suelo 29.
INDEC, 2010. Encuesta Anual de Hogares 2011. Instituto Nacional de Estadística y Censos (INDEC).

Juárez, M.M., Rajal, V.B., 2013. Parasitosis intestinales en Argentina: Principales agentes causales encontrados en la población y en el ambiente. Rev. Argent. Microbiol. 45 (3), 191–204. https://doi.org/10.1016/S0325-7541(13)70024-5.

Kaabache, R., Badaoui, B., 2022. The impact of climate change on food security and health, case of Algeria. 86–106 ,(1) 5 مجلة الاقتصاد والتنمية المستدامة.

Le, S., Josse, J., Husson, F., 2008. FactoMineR: an R package for multivariate analysis. J. Stat. Softw. 25 (1), 1–18.

López Ureña, N.M., Chaudhry, U., Calero Bernal, R., Cano Alsua, S., Messina, D., Evangelista, F., Álvarez, García G., 2022. Contamination of soil, water, fresh produce, and bivalve mollusks with *toxoplasma gondii* oocysts: a systematic review. Microorganisms 10 (3), 517.

Mehrnejat, N., Kadkhodaie, S., Farrokhzadeh, H., Ali, H., Pourgheysari, H., Seif, S., 2018. Evaluation of parasitic contamination in consuming vegetables in a city of Iran in 2011. Envir. Health Eng. 4 (2), 96–132.

Melín-Coloma, M., Villaguala-Pacheco, C., Lisboa-Navarro, R., Landaeta-Aqueveque, C., 2016. Estudio de la presencia de huevos de Toxocara sp. en suelos de áreas públicas de la ciudad de Chillán, Chile. Rev. Chil. Infectol. 33 (4), 428–432.

Miño, M., Herrera, E., Notarnicola, J., del Robles, M.R., Navone, G., 2012. Diversity of the helminth community of the Pampean grassland mouse (*Akodon azarae*) on poultry farms in Central Argentina. J. Helminthol. 86 (1), 46.

Ortiz, M., Borjas, B., 2008. La Investigación Acción Participativa: aporte de Fals Borda a la educación popular. Espacio abierto 17 (4), 615–627.

Palacio, F.X., Apodaca, M.J., Crisci, J.V., 2020. Análisis multivariado para datos biológicos: teoría y su aplicación utilizando el lenguaje R.

Pérez-Cordón, G., Rosales, M.J., Valdez, R., Vargas-Vásquez, F., Cordova, O., 2008. Detección de parásitos intestinales en agua y alimentos de Trujillo, Perú. Revi. Peruana Med. Exp.Salud Pública 25 (1), 144–148.

Puig-Peña, Y., Leyva-Castillo, V., Rodríguez-Suárez, A., Carrera-Vara, J., Molejón, P.L., Pérez-Muñoz, Y., Dueñas-Moreira, O., 2013. Calidad microbiológica de las hortalizas y factores asociados a la contaminación en áreas de cultivo en La Habana. Rev. Habanera Ciencias Méd 13 (1), 111–119.

Rea, M.J., Fleitas, A., Borda, E.C., 2004. Existencia de parásitos intestinales en hortalizas que se comercializan en la ciudad de Corrientes, Argentina. Rev. Univer. Nac. Nordeste 6, 1–5. Retrieved from. https://www.researchgate.net/profile/Maria_Rea4/publication/237315958_Existencia_de_parsitos_intestinales_en_hortalizas_que_se_comercializan_en_la_ciudad_de_Corrientes_Argentina/links/0deec53c43b93be228000000.pdf.

Rivero, M.R., De Angelo, C., Nuñez, P., Salas, M., Motta, C.E., Chiaretta, A., Liang, S., 2017. Environmental and socio-demographic individual, family and neighborhood factors associated with children intestinal parasitoses at Iguazú, in the subtropical northern border of Argentina. PLoS Negl. Trop. Diseases. 11 (11), 1–26. https://doi.org/10.1371/journal.pntd.0006098.

Rivero, M.R., Feliziani, C., De Angelo, C., Tiranti, K., Salomon, O.D., Touz, M.C., 2020. *Giardia* spp., the most ubiquitous protozoan parasite in Argentina: human, animal and environmental surveys reported in the last 40 years. Parasitol. Res. 119, 3181–3201. https://doi.org/10.1007/s00436-020-06853-7.

Rodríguez, M., Zapata, M., Solano, M., Lozano, D., Torrico, F., Torrico, M., 2015. Evaluación de la contaminación microbiológica de la lechuga (*Lactuca sativa*) en la cadena alimentaria, provincia de Quillacollo, Cochabamba, Bolivia. Gaceta Médica Boliviana 38 (2), 31–36. Retrieved from. http://www.scielo.org.bo/scielo.php?pid=S1012-29662015000200006&script=sci_arttext.

Ruiz-Taborda, J.P., Casas-Valencia, A., Cardon-Arias, J.A., 2018. Análisis del parasitismo intestinal y la malnutrición en Suramérica desde sus determinantes sociales CES Salud Pública, 8, 25–33.

Salas, A., Rusconi, J.M., Camino, N., Eliceche, D., Achinelly, M.F., 2017. First record of *Diploscapter coronata* (Rhabditida). Rev. de la Facult. de Ciencias Agrarias 49 (1).

Sedionoto, B., Wasessombat, S., Punsawad, C., Anamnart, W., 2021. The quality of soil and high prevalence of hookworm infection in Muara Kaman and Marangkayu districts, Indonesia. Ann. Trop. Med. Public Health 24.

SENASA, 2020. Planilla de Exportaciones de Hortalizas 2018 Informe y estadísticas Servicio Nacional de Sanidad y calidad Agroalimentaria. http://www.senasa.gob.ar/cadena-vegetal/hortalizas/informacion/.

Shoaie Baker, S., García, M., 2021. Jóvenes De Familias Migrantes Y Transición Agroecológica En el Cinturón Hortícola de La Plata, Argentina. Eutopía. Revista De Desarrollo Económico Territorial 19, 97–118. https://doi.org/10.17141/eutopia.19.2021.4966.

Shurtleff, M., Averre, C., 2000. Diagnosing plant diseases caused by nematodes. Capítulo 2: methods: extracting nematodes from plant tissue or soil. Centrifugal flotat.

Team Core, RA, 2020. Language and Environment for Statistical Computing, Austria, 4.0.2. R Found. Stat. Comput, Vienna.

Tenesaca, J.B., Serrano, M.E.Á., Guzmán, R.C.Z., 2019. Parásitos en expendedores y hortalizas de los mercados públicos. Cuenca 2015. Rev. de la Facult. de Ciencias Médicas de la Univ. de Cuenca 37 (1), 21–30.

Unzaga, J.M., Zonta, M.L., 2018. Atlas Comentado de Protozoología: Protozoos parásitos de importancia sanitaria y epidemiológica.

WHO, 2015. Inocuidad de los alimentos. World Health Organization.

WHO (Ed.), 2019. Bench Aids for the Diagnosis of Intestinal Parasites, 2° ed. WHO, Geneva.

Zonta, M.L., Susevich, M.L., Gamboa, M.I., Navone, G.T., 2016. Parasitosis intestinales y factores socioambientales: Estudio preliminar en una población de horticultores. Salud (i) Ciencia 21 (8), 814–822. https://doi.org/10.21840/siic/147782.

Zuta Arriola, N., Rojas Salazar, A.O., Mori Paredes, M.A., Cajas Bravo, V., 2019. Impacto de la educación sanitaria escolar, hacinamiento y parasitosis intestinal en niños preescolares, 10 (1), 47–56.