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Original article

Co-contamination of Raw Vegetables with *Meloidogyne* spp. and Human Intestinal Parasites: An Emerging Insight in Public Health

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Abstract: This study was aimed at detecting the parasitic contamination in raw vegetables sold in the four public markets in the city of Manila, Philippines. This cross-sectional study was carried out from March to September 2022. Observations on the personal hygiene of the vendors and environmental sanitation were conducted. A total of 87 fresh vegetable samples were collected and processed for the detection of parasitic life forms using standard parasitological techniques for protozoa and helminth worms. The occurrence rate of at least one parasitic contamination of the vegetables was 39.1%, mostly with *Meloidogyne* spp. (26.4%). Other parasites detected are *Ascaris lumbricoides* (8.0%), hookworm (6.9%), *Fasciola* spp. (5.7%), *Giardia* spp. (2.3%), and *Cyclospora* spp. (1.1%). Other parasites are macroscopic (6.9%). Remarkably, a high level of contamination was detected in Chinese cabbage (*Brassica rapa*) 68.8%, cabbage (*Brassica oleracea*) 55.6%, lettuce (*Lactuca sativa*) 55.6%, ginger (*Zingiber officinale*) 44.4%, and carrots (*Daucus caruta*) 40%. The contamination rates of vegetables from the four public markets were analyzed and a significant difference was noted (x^2 = 11.852, p = 0.007908) at p-value <.05. This implies that public markets in the city of Manila could play a role in the transmission of parasitic infections in the area. The high occurrence rate of *Meloidogyne* spp., which is a spurious parasite of man, was included in the analysis and discussions. It is in response to the emerging discussions regarding the reported passage of *Meloidogyne* spp. in human stool samples.

Keywords: *Soil-transmitted helminths, protozoa, neglected tropical disease, food-borne parasitic infections*

1. INTRODUCTION

Diarrheal disease is one of the leading causes of death globally (Dadonaite et al., 2018), most of which can be attributed to contaminated food or water (WHO, 2017). In 1997, the Department of Health (DOH) in the Philippines issued AO No. 29-A, s. 1997 to create food and waterborne diseases (FWBDs) prevention and control programs (DOH, 2017). However, diarrhea remains a public health issue in the country affecting the general population in recent years (Statista, 2022). Thus, new foci and priorities were charted by the DOH through the 2019-2023 Food and Water-Borne Diseases Prevention and Control Program Strategic Plan (FWBD-PCP). It was introduced by the DOH to its stakeholders to successfully reduce the morbidity rate and eliminate deaths in the country due to diarrhea. Some of the strategies in the new program highlight the regulation and monitoring of food and water sanitation practices at the local level and the promotion of personal hygiene, food and water sanitation practices, and the principles of environmental health (DOH, 2019).

Intestinal parasites are among the common causes of diarrhea in the country alongside viruses and bacteria (Peligrino, 2021). The enteric parasites detected in humans by public health researchers across the country include *Ascaris lumbricoides*, *Trichuris trichiura* (Ross et al., 2017; Labana et al., 2021), and protozoa (Labana et al., 2018; Weerakoon et al., 2018), among others. In separate studies, *Ascaris lumbricoides*, *Trichuris trichiura,* and protozoa were also detected in the fresh vegetables sold in the public markets in the Philippines (Vizon et al., 2019). To provide inclusive and comprehensive practices toward meeting the shared goals of ensuring healthy people in the community, regular detection of parasitic contamination of food and water is recommended by the DOH (DOH, 2019).

This study was conducted to detect possible parasitic contamination of raw vegetables sold in the public markets in the city of Manila, the capital of the Philippines known for being the second most populous city in the country. It aims to contribute to the continuous review and upscaling of health programs in the country and to strengthen local legislation for the control of food-borne diseases such as persistent diarrhea.

2. METHODOLOGY

2.1. Study design and sampling techniques

A cross-sectional study was carried out from March to September 2022 in the city of Manila. Manila City has a land area of 42.34 km^2 with a population of ~1.8M based on the 2020 census. It is divided into six congressional districts. Four public markets across the city were purposively chosen in this study including the Divisoria Market in the 2nd district, the Quiapo Public Market in the 3rd district, a wet market in Sampaloc, Manila called *Talipapa in Barangay 407* situated in the 4th district, and the Altura Marketplace in the 4th district. The four public markets are all open-air markets with Divisoria Market considered by the locals as the “mother of all markets” due to the vast array of merchandise sold in the area (Domingo, 2012). Quiapo Public Market, on the other hand, is considered Manila’s central market where Quinta Market, a historical market built in 1851, is situated (Marpa, 2022). *Talipapa* in Barangay 407 represents the temporary makeshift market. Lastly, Altura Marketplace is a public market adjacent to the community flocked by informal human settlers.

Multistage cluster sampling was done in the collection of vegetable samples in each public market. The sampling frame was the stalls of raw vegetables. Each group of vegetable stalls was allocated with a unique number for a simple random sampling. The commonly consumed raw vegetables, either leafy or root vegetables, were collected by purchasing them from selected vegetable stalls. The collected vegetable samples varied from each vegetable stall depending on the availability of the vegetables being sold.

2.2 Observations of the Public Markets

Before the vegetable sample collection, observations on the personal hygiene of the vendors and the environmental sanitation of the public markets were conducted. A structured observational checklist answerable by a “yes” or a “no” was used. The two-part instrument was based on the study of Iwu et al. (2017). The first part was used to guide in observing the personal hygiene of the vegetable vendors, while the second part focused on the sanitation condition of the vegetable stalls. In this study, the number of observations were based on (1) the number of vegetables collected from the vegetable vendor and (2) the sanitation condition of the stall where the vegetables were sold. Inter-rater reliability was used wherein the three observers ensured agreement on the concluding observations. Figure 1 presents the map of the city of Manila and the four sampling sites. The inset images depict the stall sanitary conditions per sampling site.

Figure 1. Map of the City of Manila, the sampling site. Insert images: upper left – Divisoria public market; lower left – Quiapo public market, upper right – Talipapa in Barangay 407; lower right – Altura Marketplace.

2.3. Sample collection, preparation and washing

The sample collection, preparation, and washing were based on the methods conducted by Eraky et al. (2014) and Abougrain et al. (2010), with minor modifications. A total of 87 fresh vegetable samples were collected. Each vegetable sample was directly placed in a separate resealable clear cellophane bag with a measurement of 5×7 inches. Each bag was labeled with a unique code, the date of collection, and the place of collection. The samples were sent to the Microbiology-Parasitology Laboratory of the Engineering Science Research Center (ESRC), Polytechnic University of the Philippines, for processing. A portion of the vegetable sample was carefully picked by hand and discarded to retain an approximate measurement of 200 g for each sample. A 500-mL physiological saline solution was added to soak the vegetable for 15 minutes. It was followed by vigorous shaking with the aid of a mechanical shaker (Labtron Equipment Ltd, Surrey, United Kingdom) for 15 minutes. The vegetable sample was carefully removed from the bag after the vigorous shaking. The wash solution was placed in a beaker and left overnight to sediment. The top layer was removed. The remaining wash solution was filtered through a mesh with approximately 400 µm pore size. The large debris was discarded, and the filtered wash solution was centrifuged at 2000 rpm (447 *x*g) for 15 minutes. A few drops of the sediments were placed on a regular glass slide and examined microscopically for the detection of parasite life forms.

2.4 Examination of samples

The sediment was mixed before creating a stained and unstained smear. Two staining techniques were conducted. In Lugol's iodine staining, a small drop of Lugol's iodine solution was added to the smear of sediment. A cover slip was carefully placed on the glass slide before viewing it under the compound microscope (OMAX, China). In modified *Ziehl-Neelsen* staining, the other drops of sediment were stained with concentrated carbol fuchsin for 15 to 20 minutes then rinsed with tap water. It was decolorized with 1% HCL for 15 to 20 seconds then

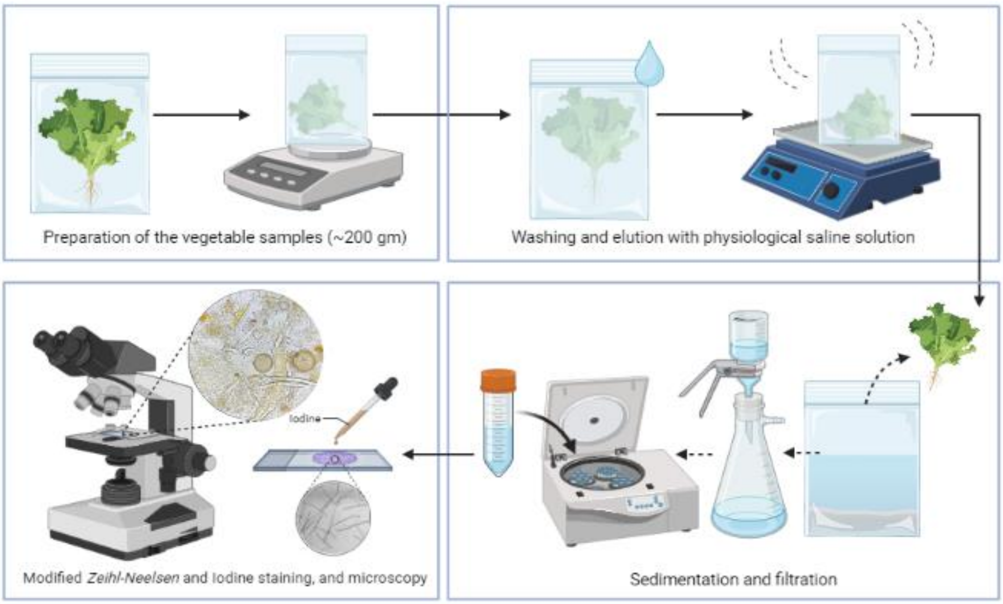


Figure 2. Workflow in detecting parasites in vegetable samples.

rinsed with tap water, counterstained with 0.4% malachite green for 30 to 60 seconds, rinsed thoroughly with tap water then air-dried before examining it under the microscope. The modified *Ziehl-Neelsen* staining was conducted to detect coccidian protozoal oocysts (Labana et al., 2017). Figure 2 presents the diagram of the step-by-step protocols for detecting parasites present in the vegetables.

2.5 Statistical analysis

All data were first encoded to MS Excel 365 (Microsoft Corp., Washington, USA) for the organization. Three major MS Excel sheets were created for the encoding of data from the observational checklist (personal hygiene and environmental sanitation), listing of vegetable samples and their characteristics (weight of the vegetable, date, and time of collection, among other parameters), and recording of the detected parasites in each sample. The findings were analyzed using Statistical Package for Social Science (SPSS) version 23.0 (IBM Corp., Armonk, NY, USA). Descriptive statistics were used to identify the total number of parasites detected in each sample. The occurrence rate was computed by dividing the number of parasite-positive vegetables by the total number of samples collected in each type of vegetable or each public market where the vegetables were collected. Logistic regression analysis evaluated the association between parasitic contamination of vegetables sold in the public markets with the personal hygiene of the vegetable vendor and the environmental sanitation of the public markets. The results were considered as crude odds ratio (cOR). Risk and protective factors at p-value <0.05 was considered significant.

3. RESULTS AND DISCUSSION

3.1. Hygiene and sanitation conditions in the four public markets

There was a total of 87 observations recorded on the personal hygiene of vegetable vendors in four public markets. The observations were ranked according to the number of positive observations, which means the observer noted “yes” to the items in the observational checklist. The upper interquartile range shows the five (5) commonly observed personal hygiene conditions or practices of the vegetable vendors in the four public markets. These include having no observable skin infection (87/87; 100%), being neatly dressed (83/87; 95.4%), not eating in the store (73/87; 83.9%), wearing hair tie (51/87; 58.6%), and having a well-kept fingernail (46/87; 52.9%).

The lower interquartile range shows that there were vegetable vendors who had their hair coloring during the time of vegetable collection (53/87; 60.9%). This is a reversed scoring in the observational checklist. Having hair coloring can be unhygienic to some people caused by the possible adverse effects of hair dye components, which may include skin rashes (He et al., 2022). This case is the same with the wearing of hand jewelry wherein 62.1% (54/87) of the observations were noted “yes” for having hand jewelry during the time of vegetable collection. Other observations included in the lower interquartile range are wearing an apron (14/87; 16.1%), wearing a hairnet (1/87; 1.1%), and wearing gloves (1/87; 1.1%).

Regarding the environmental sanitation of the vegetable stalls, most of the vegetable vendors ensured that no vegetables were placed on the bare floor (70/87; 80.5%). Vegetables were usually stacked on the vegetable rack or put inside a straw basket. There was the presence of enough garbage bins in most of the stalls (69/87; 79.3%), although part of the upper interquartile range, there was a low percentage observed. The observers did not see disease vectors like rats, cockroaches, and flies in the premise of the vegetable stalls (37/87; 42.5%). The tables used in the stalls, as well as the surroundings, are generally clean (37/87; 42.5%). There were also adequate accessible water supplies (19/87; 21.8%).

There are five aspects of the environmental hygiene observed in vegetable stalls that need improvement. They are part of the lower interquartile range, and these included the presence of towel (14/87; 16.1%), the presence of protection against vectors (9/87; 10.3%), the presence of a clean, wash-hand basin (9/87; 10.3%), adequate sanitary condition of the environment (2/87; 2.3%), and presence of soap (1/87; 1.1%). The sanitary condition of the environment was scored low by the observers due to lack of the organization of the stalls, absence of plastic coverings for the vegetables, presence of smoke from the vehicles, and unregulated contacts of unhygienic people which could be associated with being open-air public markets.

3.2 List of raw vegetables collected for the detection of parasites.

A total of 87 raw vegetables including 64 leafy vegetables and 23 root vegetables were collected in the four public markets. The most collected vegetables are Chinese cabbage *(Brassica rapa)* (*n*=16), lettuce *(Lactuca sativa)* (*n*=10), cabbage *(Brassica oleracea)* (*n*=9), ginger (*Zingiber officinale*) (*n*=9), and potato *(Solanum tuberosum)* (*n*=9). These are the most sold vegetables in the public markets during the time of sample collection. The full list of the collected vegetables with their local names and their scientific names is shown in Table 1.

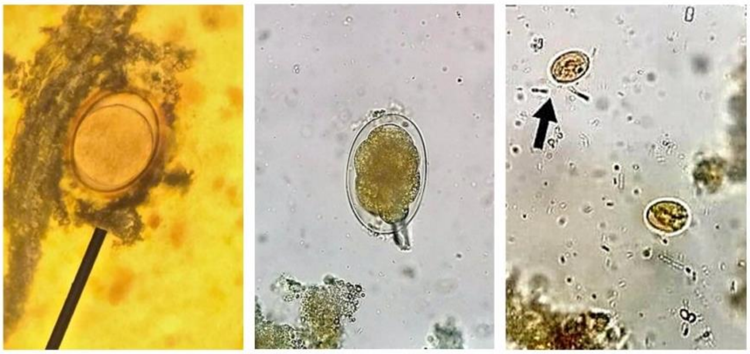
3.3. Parasites detected in raw vegetables sold in the four public markets

Of the 87 raw vegetable samples, 34 (39.1%) were contaminated with at least one parasite. Intestinal parasites were detected in 21 (24.1%) vegetable samples. The parasites detected include *Cyclospora* spp. (1/87; 1.1%), *Giardia* spp. (2/87; 2.3%), *Fasciola* spp. (5/87; 5.7%), hookworm (6/87; 6.9%), and *Ascaris lumbricoides* (7/87; 8.0%). There are other parasites that are grouped together in this study. These are macroscopic parasites or pests including slugs, root maggot fly, and other unidentified moving parasites (6/87; 6.9%). A big portion of the vegetable samples were contaminated with *Meloidogyne* spp. (23/87; 26.4%), or commonly called root-knot nematode. Figure 3 shows the morphology of the parasites detected in vegetable samples.

The detected parasites in each of the vegetable samples are presented in Table 2. *Ascaris lumbricoides*, a common intestinal parasite in the Philippines was detected in Chinese cabbage (*Brassica rapa*) (6.3%), cabbage (*Brassica oleracea*) (11.1%), ginger (*Zingiber officinale*) (11.1%), potato (*Solanum tuberosum*) (11.1%), Malavar spinach (*Basella alba*) (16.7%), and lettuce (*Lactuca sativa*) (20.0%). Hookworms were detected in Chinese cabbage (*Brassica rapa*) (6.3%), lettuce (*Lactuca sativa*) (10.0%), cabbage (*Brassica oleracea*) (11.1%), and ginger (*Zingiber officinale*) (22.2%). *Giardia* spp., a causative agent of watery diarrhea, was detected in lettuce (*Lactuca sativa*) (10.0%), and cabbage (*Brassica oleracea*) (11.1%).

Table 1. List of raw vegetable samples collected in the four public markets in the city of Manila.

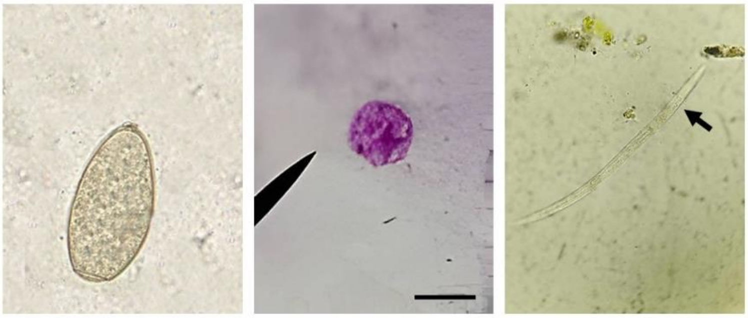
Vegetables	Type of vegetables	Altura Marketplace	Divisoria Market	Talipapa in Barangay 407	Quiapo Public Market	Total number of vegetables
Chinese cabbage / pechay *(Brassica rapa)*	Leafy	6	5	2	3	**16**
Cabbage / repolyo *(Brassica oleracea)*	Leafy	3	2	3	1	**9**
Mustard / mustasa *(Brassica juncea)*	Leafy	0	1	2	0	**3**
Lettuce / litsugas *(Lactuca sativa)*	Leafy	3	5	2	0	**10**
Iceberg lettuce *(Lactuca sativa var. capitata)*	Leafy	2	3	0	1	**6**
Alugbati / Malabar spinach *(Basella alba)*	Leafy	3	1	2	0	**6**
Ampalaya tops / talbos ng ampalaya *(Momordica charantia)*	Leafy	0	1	1	0	**2**
Swamp cabbage / kangkong *(Ipomoea aquatica)*	Leafy	2	0	0	1	**3**
Kamote tops / talbos ng kamote *(Ipomoea batatas)*	Leafy	1	0	1	0	**2**
Celery / kintsay *(Apium graveolens)*	Leafy	3	2	2	0	**7**
Carrots / karots *(Daucus carota)*	Root crop	3	0	1	1	**5**
Ginger / luya *(Zingiber officinale)*	Root crop	3	4	2	0	**9**
Potato / patatas *(Solanum tuberosum)*	Root crop	3	2	2	2	**9**
Total number of vegetables		**32**	**26**	**20**	**9**	**87**



Ascaris lumbricoides egg
Magnification: 400x

Hookworm egg
Magnification:400x

Giardia spp. cyst
Magnification:100x



Fasciola spp. egg
Magnification:400x

Cyclospora spp. oocyst
Scale bar = 10µm

Meloidogyne spp.
Magnification:100x

Figure 3. Morphology of the parasites detected in the vegetable samples sold in the selected public markets in the city of Manila.

The intensity of parasitic contamination was not included in the analysis due to the low number of detected parasites in most of the raw vegetables. This is contrary to the high-intensity contamination with *Meloidogyne* spp. in most of the parasite-positive vegetables. Instead of the intensity of contamination, the degree of parasitic contamination was noted. It is defined in this study as either contaminated with only one species of parasite or contaminated with two or more species of parasites. Of the 87 vegetable samples, 21 (24.1%) were contaminated with only one species of parasite and 13 (14.9%) were contaminated with two or more species of parasites. The vegetables poly-contaminated with four to five parasites include ginger (*Zingiber officinale*), lettuce (*Lactuca sativa*), cabbage (*Brassica oleracea*), and Chinese cabbage (*Brassica rapa pekinensis*). A chi-square test with Yates correction (x^2 = 1.7912, *p*-value = 0.180783) was computed to compare the two degrees of contamination and the value is not significant at $p < .05$.

Table 2. Detected parasites in raw vegetables sold in the four public markets.

Vegetables	Type of vegetable	Total Number of Vegetables	*Cyclospora* spp. n (%)	*Giardia* spp. n (%)	*Ascaris lumbricoides* n (%)	Hookworm n (%)	*Fasciola* spp. n (%)	*Meloidogyne* spp. n (%)	Others n (%)
Chinese cabbage/peachy *(Brassica rapa)*	Leafy	16	1 (6.3%)	0 (0%)	1 (6.3%)	1 (6.3%)	2 (12.5%)	6 (37.5%)	3 (18.8%)
Cabbage/repolyo *(Brassica oleracea)*	Leafy	9	0 (0%)	1 (11.1%)	1 (11.1%)	1 (11.1%)	0 (0%)	3 (33.3%)	1 (11.1%)
Mustard/mustasa *(Brassica juncea)*	Leafy	3	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Lettuce/ litsugas *(Lactuca sativa)*	Leafy	10	0 (0%)	1 (10.0%)	2 (20.0%)	1 (10.0%)	1 (10.0%)	5 (50%)	0 (0%)
Iceberg lettuce *(Lactuca sativa var. capitata)*	Leafy	6	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Malabar spinach/alugbati *(Basella alba)*	Leafy	6	0 (0%)	0 (0%)	1 (16.7%)	1 (16.7%)	0 (0%)	1 (16.7%)	0 (0%)
Ampalaya tops *(Momordica charantia)*	Leafy	2	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Swamp cabbage/kangkong *(Ipomoea aquatica)*	Leafy	3	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (33.3%)	0 (0%)	0 (0%)
Kamote tops/talbos ng kamote *(Ipomoea batatas)*	Leafy	2	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
Celery/Kintsay *(Apium graveolens)*	Leafy	7	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (14.3%)	0 (0%)
Carrots/karots *(Daucus carota)*	Leafy	5	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (20.0%)	1 (20.0%)
Ginger/luya (*Zingiber officinale*)	Root crop	9	0 (0%)	0 (0%)	1 (11.1)	2 (22.2%)	1 (11.1%)	4 (44.4%)	0 (0%)
Potato *(Solanum tuberosum)*	Root crop	9	0 (0%)	0 (0%)	1 (11.1)	0 (0%)	0 (0%)	2 (22.2%)	1 (11.1%)
TOTAL		**87**	**1 (1.1%)**	**2 (2.3%)**	**7 (8.0%)**	**6 (6.9%)**	**5 (5.7%)**	**23 (26.4%)**	**6 (6.9%)**

Others **=** slugs, root maggot fly, other macroscopic parasites

This study shows that the leafy and root vegetable samples collected in Altura Marketplace, Divisoria Market, *Talipapa in Baranggay 407*, and Quiapo Public Market, in the city of Manila, Philippines are contaminated with different types of parasites. Of the parasites detected, four species are known to cause diarrhea when ingested. These parasites include *Cyclospora* spp., *Giardia* spp., *Ascaris lumbricoides*, and hookworm, which can be transmitted to humans through the consumption of contaminated food (Minetti et al., 2016; Li et al., 2020; Mationg et al., 2021). Despite the low occurrence rates of these parasites, ranging from 1.1% to 8.0%, public education on proper preparation of vegetables before consumption is still recommended to avoid the spread of infection (Slifko et al., 2000).

This is not the first record of intestinal parasite contamination in raw vegetables sold in the public markets in the Philippines. In a study conducted in 2012, 45% of the vegetables sold in the selected markets in Metro Manila are contaminated with pathogenic parasites. Of the 80 samples examined, 36 (45.00%) were contaminated with *Ascaris*, four (5.00%) with hookworm, and one (1.25%) with *Giardia* spp. (Su et al., 2012). These are the same species of parasites detected in the current study.

In a separate study, *Ascaris lumbricoides* was also detected in the raw vegetables sold in the Divisoria public market (Malison et al., 2009). This market has a reputation of having poor sanitation due to overcrowding of people caused by the availability of low-priced goods and diverse manufacturing activities in the area (Domingo, 2012). Despite the improved sanitation in the area in the past few years (Madarang, 2019), the current study still detected parasites in the raw vegetables sold in the public markets. Further studies must be done to assess the risk of parasitic contamination in the vegetables sold in urban places like the city of Manila.

Rural farms in the Philippines are the usual sources of the vegetables sold in Metro Manila. These places were also reported to have parasitic contamination in the environment. In a recent study, rural farms in Laguna, Philippines were surveyed for environmental contamination with parasites. Parasites were detected in vegetables (17.3%), soil (47.3%), and water (73.3%). Among the parasites detected are *Ascaris* spp. and *Giardia* spp. (Paller et al., 2022). In a separate study, green-leafy vegetables sold in the public markets in the province of Nueva Ecija, Philippines were contaminated with *Ascaris* spp., and hookworm, among others (Vizon et al., 2019). Another study conducted with the soil samples from selected organic and conventional farms in the Philippines revealed that around four in every 10 soil samples are contaminated with parasites. *Ascaris spp.* was also detected in this study (Paller et al., 2022). These study findings provide a picture of an unexplored possible spread of parasites from the rural provinces to the urban cities in the Philippines. Further studies about this possible agricultural phenomenon in the country must be done. It is not only important to the farming industry of the Philippines but also to public health.

3.4. Risk and protective factors on the parasitic contamination of the raw vegetables in the four public markets

The occurrence rate of parasitic contamination of vegetables per public market was identified with Quiapo Public Market with the highest occurrence rate of 55.6% (5/9). It was followed by the *Talipapa in Baranggay 407* with an occurrence rate of 55.0% (11/20) and the Divisoria Market with an occurrence rate of 50.0% (13/26). The lowest occurrence rate of vegetable parasitic contamination was detected in Altura Marketplace (5/32; 15.6%). A chi-square test was computed to compare the parasitic contamination of raw vegetables sold in the four public markets. The result (x^2 = 11.852, *p*-value < 0.007908) showed significant values at $p < 0.05$. Table 3 shows the parasitic contamination of raw vegetables sold in the four public markets.

Univariate logistic regression was computed to analyze the risk and protective factors of the parasitic contamination of the raw vegetables sold in four public markets (Table 4). The binary outcomes are either the vegetables are contaminated (yes) or not contaminated (no). The independent variables are the number of observations on the personal hygiene of the vendors and the environmental sanitation of the vegetable stalls in the four public markets. The protective factors identified in this study include wearing gloves (cOR= 0.247; 95% CI=0.066-0.918), protecting vegetables from flies and/or rodents (cOR= 0.075; 95%

CI = 0.022, 0.259), presence of clean wash hand basin (cOR= 0.156; 95% CI = 0.061-0.401), presence of soap (cOR=0.025; 95% CI=0.003-0.185), presence of towel (cOR=0.128; 95% CI=0.047-0.352), avoiding contact of vegetables on the bare floor (cOR= 0.156; 95% CI = 0.061-0.401), adequate supply of water (cOR=0.184; 95% CI= 0.075-0.451), and adequate sanitary condition (cOR=0.050; 95% CI=0.011-0.216). The only risk factor of vegetable parasitic contamination, as observed in this study, is being neatly dressed (cOR=2.945; 95% CI=1.348-6.433).

Table 3. Parasitic contamination of raw vegetables sold in four public markets.

Public Markets	**N**	**Negative**	**Positive**	***p-value***
Altura Marketplace	32	27 (84.4%)	5 (15.6%)	0.007908
Divisoria Market	26	13 (50.0%)	13 (50.0%)	
Talipapa in Baranggay 407	20	9 (45.0%)	11 (55.0%)	
Quiapo Public Market	9	4 (44.4%)	5 (55.6%)	

Significant *p*-value = <0.05

3.5. Emerging public health discussions on Meloidogyne spp.

Of the six parasites and other parasites grouped as one in this study, *Meloidogyne* spp. was the most prevalent parasite in the raw vegetables sold in the four public markets. It also had the highest intensity of contamination among all the parasites detected. Therefore, it merited discussion on its possible public health implications. *Meloidogyne* spp. is an obligate parasite of plants with a worldwide distribution. It is one of the top 10 plant-parasitic nematodes based on a 2013 survey for the journal *Molecular Plant Pathology* (Jones et al., 2013). Generally, this parasite causes significant damage to crops including potatoes, carrots, cabbage, and lettuce, among others. It is therefore reported as a threat to many crops in various regions of the world including Mexico (Tapia-Vasquez et al., 2022) and South Africa (Onkendi et al., 2014). In the Philippines, this plant-parasitic nematode has already been impacting the country's agricultural produce for many years (Davide, 1988; Pedroche et al., 2013).

Recently, a topic on *Meloidogyne* spp. appeared in a letter to the editor of the *Journal of Clinical Microbiology*. The focus of the letter is not on the threat of this nematode in agriculture but on its presence in the human stool specimens. The nematode eggs were characterized by the author as similar in morphology to *Trichostrongylus* (Bradbury et al., 2015), a parasite that can cause anorexia, abdominal pain, and diarrhea in humans (CDC, 2017). Another report of the detection of *Meloidogyne* eggs in human stool was traced in a private laboratory network in Salvador, Brazil. The egg of *Meloidogyne* was described as elongate-ovoid with rounded ends and thin hyaline shells. It resembles hookworm, an intestinal parasite that completes the "unholy trinity" alongside *Ascaris lumbricoides* and *Trichuris trichiura* or the soil-transmitted helminths that can cause diarrhea and anemia in human (Roach et al., 2021).

The reports on the detection of *Meloidogyne* eggs in human stools do not make it a true parasite of man. It is a spurious parasite that can invade humans through incidental intake, but cannot cause disease (Bradbury et al., 2015). Although not yet an important public health issue, the passage of *Meloidogyne* eggs in human stool opens other public health challenges. Concerns about the wrong diagnosis of human infections with the false *Trichostrongylus,* hookworm, or other intestinal parasites were raised (Bradbury et al., 2015; Bradbury, 2022). Misdiagnosis of intestinal parasitic contamination can result in unnecessary drug use, debilitating quality of life, and other social damages (Bonini et al., 2002; Petti et al., 2006).

Table 4. Variables associated with parasite contamination of the vegetable samples sold in the public markets in the city of Manila

Factors	Positive	Negative	cOR	Lower 95% CI	Upper 95% CI
Personal Hygiene					
1. Neatly dressed	26	61	2.945	1.348	6.433*
2. Well-kept fingernails	14	73	1.325	0.565	3.108
3. Use of hair coloring	16	71	1.557	0.677	3.582
4. Use of apron	9	78	0.797	0.313	2.032
5. Wears hairnet	5	82	0.421	0.14	1.268
6. Wears hair tie	19	68	1.93	0.858	4.346
7. Wears hand jewelry	20	67	2.062	0.921	4.616
8. Wears gloves	3	84	0.247	0.066	0.918*
9. With observable skin infection	0	87	MV		
10. Eating in the store	11	76	Reference		
Environmental Hygiene					
1. Stored vegetables protected from flies and/or rodents	3	84	0.075	0.022	0.259*
2. Presence of a clean wash hand basin	6	81	0.156	0.061	0.401*
3. Presence of soap	1	86	0.025	0.003	0.185*
4. Presence of towel	5	82	0.128	0.047	0.352*
5. Evidence of disease vectors in premises	23	64	0.757	0.393	1.458
6. Clean surface table and surrounding	19	68	0.589	0.299	1.161
7. Vegetable items placed on the bare floor	6	81	0.156	0.061	0.401*
8. Adequate supply of water	7	80	0.184	0.075	0.451*
9. Adequate sanitary condition	2	85	0.050	0.011	0.216*
10. Presence of garbage bin	28	59	Reference		

cOR = crude odds ratio; * significant p <0.05; MV = missing values

Another discussion that surfaced is the cause of the possible continued invasion of *Meloidogyne* spp. to humans. Aside from the issues of hygiene and sanitation, food preparation can also be an important factor. The latter can even be more jeopardized with the recent popularity of “Paleo” diets among health-conscious individuals (Bradbury, 2015). “Paleo” diet is a dietary pattern based on the food consumption of the people during the Paleolithic era. It is also called the Stone Age or caveman diet which includes consumption of whole meat, whole fruits, and whole vegetables (Jönsson et al., 2009). The people practicing the “Paleo” diet avoid processed foods and they believe that the human body is genetically not compatible with modern food (Turner, 2013). It was made known in the Philippines with the “country’s first Paleo door-to-door meal delivery service” (Golangco, 2014), and with the influence of social media on the “very ancestral way of preparing Filipino dishes”, which may include grazing the vegetables without cooking it (Oriel, 2020).

Parasitology and public health researchers in the Philippines must ensure accurate morphological detection of parasites in the stool samples of diarrhea patients. Recent clinical findings in other countries show passage of *Meloidogyne* spp. in human stool samples. It can also be possible in the Philippines due to issues on hygiene and sanitation, and the recent interest of the Filipinos on the “Paleo” diet. This spurious parasite must not be mistaken with *Trichostrongylus,* hookworm, or other intestinal parasites to avoid misdiagnosis and to ensure good quality of life.

3.6. Limitations of the study

This study is a small-scale surveillance of the parasites in the vegetables sold in the four public markets in the city of Manila. A larger sample size can provide a comprehensive analysis of the possible food-borne transmission of pathogenic parasites through the vegetables sold in wet markets. In another aspect, the study is limited to a one-time data collection only. Longitudinal surveillance of parasitic transmission in the public markets provides robust data for health risk analysis and disease transmission forecasting.

4. CONCLUSIONS

The raw vegetables, either leafy or roots, sold in the four public markets in the city of Manila were detected with parasites. Some of the detected parasites are considered a public health threat that causes diarrhea, vomiting, and abdominal pain, among others. A high intensity and occurrence rate of *Meloidogyne* spp. were also detected. Although a spurious parasite, it merits discussion in the aspect of public health. Among the public health implications of *Meloidogyne* spp. contamination of raw vegetables include possible passage in human feces, misdiagnosis of parasitic infection, and provision of wrong medical treatments.

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