Original Research Article

Gastrointestinal Parasite Contamination ofReady-to-Eat Vegetables Soldin Selected Markets in Ashanti Region, Ghana.

Abstract

Introduction: Intestinal parasite infections pose a significant public health concern in many developing countries, including Ghana. Inadequate sanitation and poor hygiene practices contribute to the transmission of these parasites, with contaminated food being a common route of infection. This research article investigates the prevalence of gastrointestinal parasite infections in ready-to-eat vegetables sold in selected markets in Ashanti region, Ghana.

Method:A cross-sectional study was conducted, and vegetable samples were collected from various vendors in the markets. The samples were examined for the presence of intestinal parasites using standard laboratory techniques. The findings of this study shed light on the potential risks associated with consuming contaminated vegetables and highlight the importance of food safety measures in reducing the burden of intestinal parasite infections in the region.

Results: About22.7% (68/300) of the sampled vegetables were contaminated with intestinal parasites. The commonintestinal parasites wereIPI was *Entamoeba histolytica* (9.7%, 29/300), followed by both *Ascaris lumbricoides* and *Giardia lamblia* at 4.3% (13/300) and Hookworm infections at 3.0% (9/300). The prevalence of *Enterobium vermicularis* infections was 1.7% (5/300). However, *Trichuris trichiuria* and *Strongyloidesstercoralis* infections was 0.7% (2/300). The least common intestinal parasites IPI were *Isospora belli* and *Taenia sp*. infections (0.3%,

Comment [DA1]: Please be specific with numbers. Give the exact number of contaminated samples. Please I suggest that you delete the writing in red colour. Applicable to other words or letters.

Comment [DA2]: Intestinal parasites not intestinal parasites infections which is the disease.

Comment [DA3]: Which species of the Hookworms were encountered? Name the parasite species than the general name.

Comment [DA4]: Please delete letter i.

Comment [DA5]: Please correct as suggested above

1/300).*Entamoeba histolytica* was the only parasite detected in all vegetable type; and while *Ascaris lumbricoides* was detected in all vegetable types except cucumber.

Conclusion:our study highlights the need for heightened awareness, improved food safety practices, and tailored interventions to address the risks associated with the consumption of fresh vegetables.

Keyword: Gastrointestinal Parasite, Vegetables, consumption, contamination

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Introduction

Intestinal parasitic infections represent a significant global health problem, particularly in low and middle-income countries [1]. These infections are often associated with poor sanitation, limited access to clean water, and inadequate hygiene practices [2]. Ghana, like many other countries in Ssub-Saharan Africa, faces challenges in controlling and preventing intestinal parasite infections due to various socio-economic factors [3].

Comment [DA7]: Delete capital letter S.

Ready-to-eat vegetables play a crucial role in the diet of many Ghanaians as they are rich in essential nutrients [4]. However, their consumption may also pose health risks, especially when contaminated with intestinal parasites [2,5].Vegetables can serve as a source of intestinal parasite infection when contaminated with parasite eggs, cysts, or larvae [6]. The contamination can occur at various stages of the vegetable production and supply chain, making it crucial to implement proper food safety measures to minimize the risk of infection [7].The use of contaminated water for irrigation or the application of untreated sewage or animal manure as fertilizer can introduce parasite eggs or cysts into the soil, where they can contaminate the growing vegetables [8,9]. Improper hygiene practices among agricultural workers during harvesting can transfer parasites from their hands to the vegetables [10]. Additionally, contaminated equipment or surfaces can contribute to cross-contamination [4,11].If vegetables are not thoroughly washed before consumption, parasite eggs or cysts may remain on the surface and can be ingested, leading to infection [12].

Several types of parasites can contaminate vegetables and potentially cause intestinal infections [13].Parasites such as *Giardia lamblia* and *Entamoeba histolytica* are examples of protozoa that can contaminate vegetables [14]. Ingesting the cysts of these parasites can lead to gastrointestinal infections. Parasitic worms like *Ascaris lumbricoides* (roundworm), hookworms, and *Trichuris trichiura* (whipworm) are examples of helminths that can contaminate vegetables[10][15]. Ingesting the eggs of these worms can lead to parasitic infections [16].Depending on the parasite species and the amount ingested, individuals may experience a range of symptoms, from mild gastrointestinal discomfort to severe diseases [17]. Children, the elderly, pregnant women, and immunocompromised individuals are particularly vulnerable to the adverse effects of these infections [18].In addition to gastrointestinal symptoms, intestinal parasite infections can cause nutritional deficiencies, leading to malnutrition and its associated health consequences [19]. Chronic infections can lead to anaemia and impair growth and cognitive development in children, affecting their overall well-being and future potential [20].

Comment [DA8]: Reference 10 and 15 is the same authors. Please effect the change and correct all through the work. There are other repeated refences too.

This study aims to assess the prevalence of intestinal parasite contamination of ready-to-eat vegetables sold in selected markets in the Ashanti region, Ghana. By understanding the extent of contamination, appropriate public health interventions can be developed to mitigate the risks associated with consuming contaminated vegetables

Materials and methods

Study design and area

Comment [DA9]: Please delete design and since the subtopic describes only the study area without the design of collection of samples.

This study was a cross-sectional study carried out in the Ashanti region of Ghana from December 2022 to March 2023.Structured questionnaires were administered to vendors and consumers to obtain demographic information and gather data on food handling practices and hygiene behaviours.The study was conducted at three markets: Kejetia Market, Ejura Market and Ejusu Market. Kejetia market (6.6666° N, 1.6163°W) is one of the largest and busiest markets in Ghana, located in Kumasi, the capital city of the Ashanti Region of Ghana. The market serves as a significant trading hub, attracting traders from various regions and neighboring countries. Kejetia Market offers a wide range of goods, including fresh produce, clothing, textiles, crafts and household items.Kejetia Market plays a crucial role in the local economy and cultural life of Kumasi [21].Ejura Market (7.3856° N 1.3562° W) is a bustling trading center located in the town of Ejura, which is also situated in the Ashanti Region of Ghana. This market serves as a vital commercial hub for the local community and surrounding areas. Ejura Market is well-known for its agricultural produce, including fresh fruits, vegetables, grains, and livestock, making it an essential economic and social center in the town of Ejura [22].Ejusu Market(6.5388^0 N, 0.2601^0 W) is another significant trading center in the Ashanti Region, located in the town of Ejusu. Although smaller in size compared to Kejetia Market, it plays a crucial role in meeting the daily

Comment [DA10]: British English has letter u needs of the local community. Ejusu Market predominantly offers fresh produce, including fruits, vegetables, and grains, sourced from local farmers [23].

Figure 1: Map of the study area(source or reference)

Sample Collection

For this study, a total of 300 vegetables werecollectedgathered, including green pepper (*Capsicum* sp.), cucumber (*Cucumis sativus*), lettuce (*Lactuca sativa*), carrot (*Daucus carota*), green onions (*Allium* sp.), tomatoes (*Lycopersicon esculentum*), and cabbage (*Brassica oleracea*). The selection of these vegetables was based on their availability and the common methods of consumption, either raw or lightly cooked. Each week, these fresh vegetables and fruits were purchased in the smallest retail size available, typically 200 grams, from various retail vendors operating in the three different markets.

Comment [DA11]: Please cite the source of the map

Comment [DA12]: Please correct.

Comment [DA13]: Italicize all scientific names

Comment [DA14]: How many times were samples collected from December 2022 to March 2023 per each market? This should be stated clearly please.

Comment [DA15]: Why using weight not number of vegetables?

During the purchase, interviews were conducted with the vendors to gather information about the sources of these vegetables and the type of water used for their cleaning. However, it's important to note that the vendors couldn't provide specific details regarding the sources or types of water used for irrigating the fields where these vegetables were grown. Some of the leafy vegetables originated from nearby hinterlands and villages, while others were sourced from different towns within the region. All the vegetables were prewash before being put up for sale. The vegetables were collected separately in sterile polythene bags in which they were transported to the laboratory.

Comment [DA16]: How did you conduct the interviews? In written form or verbal. How many vendors and consumers per market? These should be stated clearly

Comment [DA17]: Which laboratory? Please give the name of the laboratory and the location.

Sample processing and parasitological examination

Each vegetable sample was cut into pieces, ensuring that each sample weighed 200 grams. These chopped samples were then individually washed in separate beakers containing 1 liter of physiological saline solution (0.9% NaCl). This washing process was carried out to detach any parasitic stages that might be present on the vegetables.After washing, the vegetable samples were removed from the saline solution. The remaining washing solution was allowed to settle for a duration of 8 hours to facilitate sedimentation. Subsequently, 15 milliliters of the sediment from each beaker were carefully collected and dispensed into a centrifuge tube. These tubes were then subjected to centrifugation at a speed of 3000 rotations per minute for a duration of five minutes, as described by Tefera et al. in 2014[24].

Comment [DA18]: This method of collection was carried out following who's method? or your own? Please cite reference.

Comment [DA19]: Please follow the Journals guide lines

To further analyze the samples and detect any parasitic stages, the supernatant (the liquid above the sediment) was carefully poured off. The sediment that remained in the tube was then gently agitated, placed onto a clean slide, and examined under a microscope, using both the 10x and 40x objective lenses. This examination was carried out following the formalin ether concentration technique, as outlined by the World Health Organization (WHO) in 1991 and 1994 [25,26].

Comment [DA20]: How did you identify the parasites? Using keys or your personal knowledge? Please indicate because there should be a key to compare the morphology of the parasites with. Formol ether is a technique used for the preparation of samples for identification while microscope or other means are use for examination of the slides. Please reconsider the sentence and correct it.

Data Analysis

Descriptive statistics (frequencies and percentages) were used to determine the prevalence of parasitic contamination. Association between parasitic contamination and vegetables type was determined using chi square test and univariate logistic regression analysis. Statistical significance threshold was set at p ≤ 0.05.

Results

Socio-demographic characteristics of participants and vegetable sampling

In the study, about 300 vegetable vendors and consumers were recruited as participants. Almost equal numbers were recruited from the Ejisu market (n=101), Kejetia (n=100), and Ejura markets (n=99) in Ghana's Ashanti Region. The majority of the participants were females (62.0%, n=186/300) (X^2 =17.280, df = 1, p < 0.001). It was observed that most of the participant were those aged between ages 36 to 50 years were represented the most (43.0%, n=129) (X^2 =58.987, df = 3, p < 0.001) and the majority of participants had a formal education (69.3%, n=208) (X^2 =44.853, df = 1, p < 0.001). Tomato (n= 58) and carrot (n= 50) were the most common vegetables in this study(**Table 1**).

Comment [DA21]: There was no consistency in age interval . This may be the reason why this group is having high infection.

Comment [DA22]: Weight in gram was used in the methodology while numbers are used here. Please check and make correction

Table 1: Socio-demographic characteristics of participants and vegetable sampling

		Participants = 300	
Characteristics		Frequency (*n*)	Percent (%)
Sex			
	Male	114	38.0
	Female	186	62.0
	Total		

Comment [DA23]: Pleaseinsert the total numbers in the table

Age Group			
	14-20	39	13.0
	21-35	71	23.7
	36-50	129	43.0
	>50	61	20.3
	Total		
Level of education			
	Formal	208	69.3
	Informal	92	30.7
	Total		
Study sites (market)			
	Ejura	99	33.0
	Kejetia	100	33.3
	Ejisu	101	33.7
	Total		
Vegetable samples	Green pepper	37	12.3
	Cabbage	40	13.5
	Cucumber	43	14.3
	Spring onion	46	15.2
	Carrot	50	16.6
	Tomato	58	19.4
	Lettuce	26	8.6
	Total		

n = frequency, % = percentage

Knowledge, safety and hygienic practices among participants at study sites

Generally, most participants indicated they adequately washed their vegetables (83.3%, n=250) (X2 =133.333, df = 1, p < 0.001). However, a significant high proportion of these participants were from the Kejetia market (95.0%, 95/100) (p < 0.001) (**Table 2**).Ahigher number of participants mentioned they adequately wash their hands (77.3%, n=232) (X2 =89.653, df = 1, p < 0.001), and the highest proportion of these participants were from Ejisu market (86.1%, 87/101), and the differences in response across the study sites were statistically significant (X2 =7.258, df = 2, p = 0.027) (**Table 2**). The commonest substances for method of washing vegetables was water with vinegar or salt (67.0%, n=201) (X2 =168.140, df = 2, p < 0.001).Most

Comment [DA24]: Why lumping the other age intervals in this group. Please follow the regular interval of (of 7) 14-20, 21-27, 28-34, 35-41, 42-48,

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of these participants were from the Ejisu market (80.2%, 81/101) (**Table 2**).At home, most participants stored vegetables in a refrigerator (79.3%, n=238) (X2 =103.253, df = 1, p < 0.001).All participants from the Kejetiamarket commonly refrigerated their vegetables at home (**Table 2**). Also, the majority of participants mentioned they were aware of IPI in vegetables (93.3%, n=280) (X2 =225.333, df = 1, p < 0.001), and most ofthese participants were from the Kejetia and Ejisu markets (**Table 2**).

Table 2: Knowledge, safety and hygienic practices among participants at study sites

		Participants at study sites [markets] *n* (%)			
Characteristic		Ejura	Kejetia	Ejisu	*p*-value
Adequate washing of vegetables					< 0.001*
	Yes	62 (62.6)	95 (95.0)	93 (92.1)	
	No	37 (37.4)	5 (5.0)	8 (7.9)	
Methods to wash vegetables					< 0.001*
	Running water	31 (31.3%)	38 (38.0)	8 (7.9)	
	Water with vinegar or salt	68 (68.7)	52 (52.0)	81 (80.2)	
	Vegetable wash solutions	0 (0.0)	10 (10.0)	12 (11.9)	
Methods of storing vegetables at home					< 0.001*
	Room temperature	24 (24.2)	0 (0.0)	38 (37.6)	
	Refrigerator	75 (75.8)	100 (100.0)	63 (62.4)	
Awareness of IPI in vegetables					< 0.0001*
	Yes	82 (82.8)	99 (99.0)	99 (98.0)	
	No	17 (17.2)	1 (1.0)	2 (2.0)	

n = frequency, % = percentage, * = statistically significant at 0.05 with Chi-square test

Comment [DA27]: Substances used to wash vegetables not methods. Suggest you delete the word methods

Prevalence of intestinal parasiteinfections in vegetables

Comment [DA28]: Please delete the word infection

Overall, about 22.7% (68/300) of the sampled vegetables were contaminated with intestinal parasites. Specifically, the most common IPI was *Entamoeba histolytica*infections (9.7%, 29/300), followed by both *Ascaris lumbricoides* and *Giardia lamblia*infections at 4.3% (13/300)

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and Hookworm infections at 3.0% (9/300). The prevalence of *Enterobium vermicularis* infections was 1.7% (5/300).However, *Trichuris trichiuria* and *Strongyloidesstercoralis* infections was 0.7% (2/300). The least common IPI were *Isospora belli* and *Taenia* sp. infections (0.3%, 1/300).

Common Intestinal parasite infectionsidentified

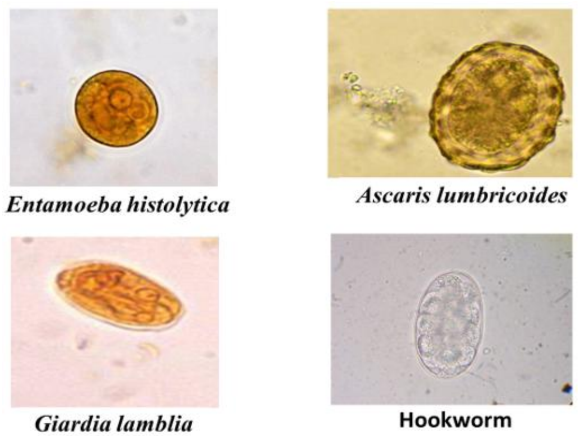


Figure 2. Plate 1. Common Intestinal parasite identified

Intestinal parasite infections stratified by study sites

Generally, it was observed that IPI most commonly occurred in the Ejura market (36.8%, 25/68) and occurred the least at the Kejetia market (30.9%, 21/68). Specifically, *Entamoeba histolytica* infections occurred most at the Ejura market (41.4%, 12/29). *Isospora belli* and *Taenia* sp. infections occurred only at the Kejetia market (n=1). *Giardia lamblia* infections occurred the most at both Ejura and Ejisu markets (46.2%, 6/13).While Ejisu market recorded the highest proportion of *Ascaris lumbricoides* infections (46.25, 6/13), the Ejura market recorded the

Comment [DA30]: *Enterobius*

Comment [DA31]: *trichiura*

Comment [DA32]: sp should not be in italic Where is the table showing these information?

Comment [DA33]: These are parasites but not diseases

Comment [DA34]: These are plates showing parasites encountered not figure. Please correct. There is no reference to this diagrams in the content of the work highest proportion of hookworm infections in the sampled vegetables (55.6%, 5/9). *Trichuris trichiuria*infections occurred only once at both the Ejura and Kejetia markets (n=1) with no occurrence at the Ejisu market[**Table 23**].

Comment [DA35]: The informationis in table 3 not 2. Please check

On another hand, *Strongyloidesstercoralis*infections occurred once at both the Ejura and Ejisu markets (n=1) with no occurrence at the Kejetia market. Finally, both Ejura and Kejetia markets had the highest proportions of*Enterobiums vermicularis* infections (40.0%, 2/5) [**Table 3**].

Comment [DA36]: This is not shown in the table. Please include it

Table 3: Specific Intestinal parasitic infections as stratified by study sites

Comment [DA37]: Please correct

	Prevalence in Study sites [markets], *n* (%)		
IPI§	Ejura	Kejetia	Ejisu
Entamoeba histolytica	12 (41.4)	10 (34.5)	7 (24.1)
Isospora belli	0 (0.0)	1 (100.0)	0 (0.0)
Giardia lamblia	6 (46.2)	1 (7.7)	6 (46.2)
Ascaris lumbricoides	3 (23.1)	4 (30.8)	6 (46.2)
Hookworm	5 (55.6)	2 (22.2)	2 (22.2)
Trichuris trichiuria	1 (50.0)	1 (50.0)	0 (0.0)
Taenia sp.	0 (0.0)	1 (100.0)	0 (0.0)
Strongyloidesstercoralis	1 (50.0)	0 (0.0)	1 (50.0)

n = frequency, % = percentage, IPI = Intestinal Parasitic Infections, § = positive proportions within specific parasitic infection shown

Comment [DA38]: Please correct the spelling

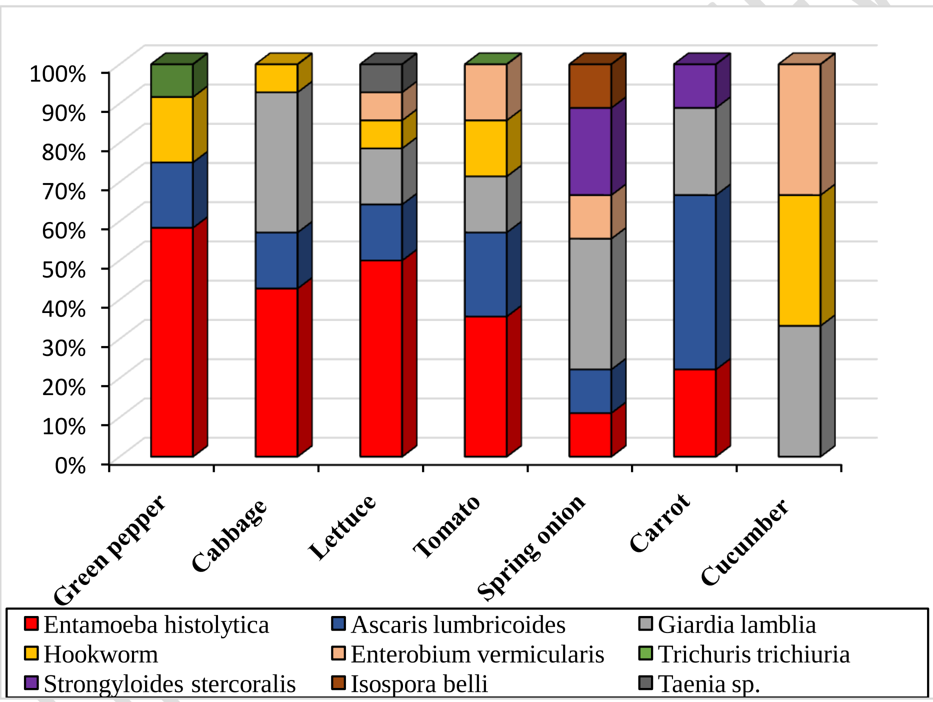
Distribution of IPIs by vegetable type

It was observed that parasitic infections occurred in all vegetable types. Specifically, *Entamoeba histolytica* was the only parasite detected in all vegetable type (**Figure 2**); and while *Ascaris lumbricoides* was detected in all vegetable types except cucumber (**Figure 2**), hookworm was

Comment [DA39]: *E. histolytica* was not found in cucumber .Pleasecompare this and other statements with the information in the figure

Comment [DA40]: Please correct

detected in all except spring onion (**Figure 2**). Similarly, to *Giardia lamblia* (Figure 2), *Enterobiums vermicularis* was not detected in green pepper and carrot, however, it was also not detected in cabbage (**Figure 2**). On the other hand, *Trichuris trichiuria* was rather detected in carrot and green pepper only (**Figure 2**).*Strongyloidesstercoralis*, *Isospora belli*, and *Taenia sp*., were detected in only one vegetable type (spring onion for *S. stercoralis*and tomato for both *I. belli* and *Taenia sp*. [**Figure 2**])



Comment [DA41]: Scientific names should be in italic in the key note. Correct the spellings of *Enterobius* and *trichiura* in the foot note

Figure 32:Distribution of IPIs by vegetable type

Comment [DA42]: Please correct

Discussion

The consumption of fresh vegetables is a vital component of a balanced diet, but growing concerns surround the potential transmission of parasites through contaminated produce [27]. In our research, we analyzed 300 vegetables and found that 22.7% of them were contaminated with intestinal parasites. To provide context, this percentage stands in contrast to a 31.7% contamination rate reported in a study conducted in Alexandria, Egypt [28]. Similarly, our findings deviate from higher prevalence rates reported in a study in Ghana, which documented an overall parasitic contamination rate of 36% [29], as well as a study in Nigeria, which also reported a 36% contamination rate [30]. Nevertheless, our study aligns with the 29% prevalence reported in Ardabil city, Iran [31]. This is an encouraging finding, as proper vegetable washing is a fundamental step in reducing the risk of foodborne illnesses associated with contaminated produce [32].

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The observed contamination rate carries significant implications for public health, indicating the potential health risks associated with the consumption of such vegetables [33].The study reveals that the occurrence of IPI varies significantly across different markets. The Ejura market exhibited the highest prevalence of IPI at 36.8%, while the Kejetia market had the lowest prevalence at 30.9%. This variation suggests that environmental and local factors may contribute to differences in contamination rates [34].

Comment [DA44]: What are these factors? Please mention them

An alarming discovery from our research is the presence of *Entamoeba histolytica* infections in approximately 9.7% of the sampled vegetables. This parasite was found across all types of vegetables, indicating widespread contamination. *Entamoeba histolytica* is a protozoan parasite known to cause amoebiasis, a gastrointestinal disease associated with a range of symptoms, including diarrhea, stomach pain, and fever [35]. The nearly 10% contamination rate suggests a relatively high prevalence of this parasite in the food supply, posing a considerable public health concern. Contaminated vegetables can be a source of amoebiasis, a severe illness. Identifying the source of infection is crucial in preventing transmission [36].

Comment [DA45]: Please check this number

In our study, *Ascaris lumbricoides* and *Giardia lamblia* infections were found in approximately 4.3% of the sampled vegetables, further raising concerns. It is noteworthy that *Ascaris lumbricoides* was not detected in cucumber, suggesting a potentially lower risk of contamination for this vegetable. Similarly, *Giardia lamblia* was absent from spring onions, green peppers, and carrots. These parasitic infections carry substantial public health risks, as their ingestion through contaminated food, including the sampled vegetables, can lead to various health issues[6][37]. The presence of *Ascaris lumbricoides* in contaminated vegetables implies fecal contamination, likely during cultivation and handling[14][38]. Once ingested, these worms can cause intestinal obstruction, a serious condition with severe symptoms. The presence of *Giardia* in vegetables suggests that water used in irrigation or washing may be contaminated [39]. Consumption of *Giardia*-contaminated vegetables can lead to giardiasis, characterized by gastrointestinal symptoms, particularly diarrhea. This discovery highlights the potential for cross-contamination during food processing or handling [40].

Comment [DA46]: Reference 6 and 37 is the same author

Comment [DA47]: Reference 14 and 38 is same authors. Please check all the refences and make the necessary corrections all through the work.

The presence of Hookworm infections in 3.0% of the sampled vegetables is another disconcerting finding in our research. Except for spring onions, hookworm was found in all types of vegetables, showing a widespread contamination risk. The presence of hookworm in multiple types of vegetables amplifies the potential health hazards for consumers. While susceptibility to contamination may vary among different vegetables, the presence of hookworm in any produce raises concerns due to the associated health risks [41].

In the study, *Enterobiums vermicularis* was detected in 1.7% of the vegetables, indicating a moderate prevalence. This parasite was notably absent in green peppers and carrots, as well as cabbage. This absence suggests that the distribution of parasites in vegetables is not uniform, possibly owing to differences in surface texture and other factors. *Trichuris trichiuria* and

Strongyloidesstercoralis were less common, each accounting for 0.7% of the contamination. *Trichuris trichiuria* was confined to carrots and green peppers, while *Strongyloidesstercoralis* was found in spring onions, indicating more limited distribution. Finally, *Isospora belli* and *Taenia sp*. infections were the least common IPI, each detected in only 0.3% of the sampled vegetables. These parasites were identified in only one specific type of vegetable (tomato), suggesting a localized risk associated with them [42].

The data shows that the types of parasites varied among the markets. For instance, the Ejura market had the highest proportion of hookworm infections, while the Ejisu market recorded the highest proportion of *Ascaris lumbricoides* infections. This variability suggests that specific environmental and agricultural practices in each region may influence the types of parasites present in vegetables [43].A substantial majority of the participants (83.3%) reported that they adequately washed their vegetables before consumption. It's notable that a significantly higher proportion of participants from the Kejetia market (95.0%) reported adequately washing their vegetables. This is an encouraging finding, as proper vegetable washing is a fundamental step in reducing the risk of foodborne illnesses associated with contaminated produce[33][44]. While a slightly lower percentage of participants (77.3%) mentioned they adequately washed their hands, it is still a considerable majority. Hand hygiene is essential in preventing the transfer of pathogens from hands to food, and the reported rates are generally positive [45].Participants from the Ejisu market (86.1%) had the highest proportion of individuals who reported proper handwashing practices. The variation across different market areas may be attributed to differences in public health initiatives or local education programs.The majority of participants (79.3%) reported storing vegetables in a refrigerator. Proper storage is crucial for maintaining the freshness and safety of vegetables, reducing the risk of contamination or spoilage [46].A

Comment [DA48]: Reference 33and 44 is the same aothours significant proportion of participants (93.3%) indicated awareness of intestinal parasites in vegetables. This awareness is crucial in promoting informed consumer choices and practices.

Conclusion

In conclusion, our study highlights the need for heightened awareness, improved food safety practices, and tailored interventions to address the risks associated with the consumption of fresh vegetables contaminated with intestinal parasites. Public health initiatives and education programs should continue to emphasize the importance of safe food handling and proper hygiene to reduce the risk of foodborne illnesses and safeguard public health.

Availability of data and materials

The datasets generated and/or analyzed during the current study are available from the corresponding author on reasonable request.

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