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Cross Sectional Survey of Parasitic Contamination in Vegetables Consumed in Birjand City in 2024

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Abstract

Background: Vegetables are a staple in diets worldwide, providing essential vitamins for human health. However, throughout the processes of planting, harvesting, and distribution, they are often at risk of contamination from various parasites, increasing the potential for transmission.

Objectives: This study aimed to compare the parasitic contamination of different vegetables in the center of South Khorasan province.

Methods: In this cross-sectional study, 200 samples of vegetables, including leeks, basil, mint, scallions, radishes, parsley, lettuce, watercress, green almonds, coriander, spinach, tarragon, turnips, and purslane, which are consumed raw by the public, were collected from Birjand city from summer to autumn 2024. Samples of 250 g from each type of vegetable were collected and brought to the laboratory. These samples were dissolved in buckets containing 1 liter of physiological serum, and the sediments were subjected to centrifugation. The supernatant contents were carefully discarded, and the sediments were examined using the laboratory method of direct smear and staining with Lugol's iodine under a microscope with $10 \times$ and $40 \times$ magnification. Data analysis was performed using IBM SPSS software version 21, employing statistical tests, including Fisher's exact test, at a significance level of P < 0.05.

Results: Out of 200 vegetable samples examined, 39% were found to be infected with parasites. The highest rates of parasitic infection were observed in spinach (70%), green almonds (66%), leeks (66%), mint (53%), coriander (44%), lettuce (50%), and purslane (29%). The most frequent parasitic infection was associated with *Blastocystis* (23.5%), followed by *Giardia* (2%), nematodes (2%), *Dicrocoelium* eggs (1.5%), and amoeba cysts (1%). No significant relationship was observed between the frequency of parasitic infection and variables such as season, distribution areas, or breeding places (P > 0.05). However, a significant relationship was observed with the type of vegetables consumed (P < 0.05).

Conclusions: Based on the results and the high level of contamination in vegetables, it is essential to implement appropriate disinfection measures before consumption and provide community members in Birjand city with education on proper vegetable handling and hygiene practices.

Keywords: Vegetables, Birjand, Parasitic Contamination

1. Background

Vegetables are a vital component of the global diet, renowned for their high nutritional value. Humans have consumed these food items both raw and cooked for centuries. In Iran, consuming raw vegetables alongside meals is a common practice, providing essential nutrients, minerals, and vitamins necessary for the body (1). However, consuming contaminated vegetables poses a significant risk of parasitic infections if proper

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disinfection measures are not followed. Intestinal parasitic infections are a major public health concern worldwide (2). These infections are widespread, with over 10% of the global population at risk. The primary parasites responsible for these infections include *Giardia lamblia*, *Ascaris lumbricoides*, *Entamoeba histolytica*, and *Blastocystis*, which are increasingly becoming a global public health concern (3).

This issue is particularly pronounced in poor and developing countries due to inadequate hygiene practices, limited personal hygiene, and a lack of awareness regarding the sanitation of vegetables. Additional factors, such as environmental temperature, climate, geography, rainfall, and soil type, significantly influence the presence of parasites in vegetables (4). Raw and contaminated vegetables are key contributors to the spread of parasitic infections. During cultivation, harvesting, transportation, preparation, and market distribution, edible vegetables are at constant risk of contamination, potentially serving as a source of infection for humans (5). Contamination risks are heightened by irrigating vegetables with polluted water, using inappropriate fertilizers like human waste or untreated animal manure, cultivating crops on unsuitable land, inadequate washing, and improper storage practices (6).

Using wastewater for irrigation is an economical way to dispose of increasing amounts of wastewater generated in urban and rural areas (7). While wastewater contains nutrients that act as fertilizers for crops, it also harbors numerous organisms that may survive treatment processes and enter the soil and food products such as vegetables (8). Protozoa and helminth eggs are particularly resilient, capable of surviving in the environment for extended periods, and can transfer to humans, causing infections and parasitic diseases (9).

Various studies have documented the presence of different parasites in vegetables and the associated risk of parasitic infections. The extent of parasitic contamination varies across regions, and identifying contamination in specific areas can assist health authorities and healthcare providers in implementing effective measures to prevent and control parasitic diseases (10).

2. Objectives

The objective of this research was to investigate the prevalence and types of parasitic contamination in raw vegetables consumed in Birjand city in 2024. These

findings aim to enable health officials to implement preventive measures and effectively control parasitic infections in the region.

3. Methods

3.1. Study Design

According to the prevalence of parasites in vegetables reported in similar studies, this cross-sectional study collected a total of 200 samples from various vegetable shops and street vendors across Birjand city from early summer to autumn 2024.

3.2. Data Collection

The random samples were purchased from various locations across the city, including the north, south, west, east, and center. The vegetable samples included leeks, basil, mint, scallions, radishes, parsley, lettuce, watercress, green almonds, coriander, spinach, tarragon, turnips, and purslane, all of which are consumed raw by the public. Sampling was conducted during the summer and autumn seasons. These vegetables are typically sourced from local farms and sometimes imported from neighboring provinces and counties, particularly from Tabas, Torbat Heydarieh, and Nishabur. For each type of vegetable, 250-g samples were purchased, placed separately in plastic bags, and transported to the laboratory.

Upon arrival at the laboratory, the samples were placed in buckets containing 1 L of 0.95% saline (NaCl) solution with a few drops of detergent to facilitate the release of contaminants. The mixture was stirred vigorously for ten minutes and then allowed to settle for 12 hours to collect sediments containing protozoan cysts, helminth eggs, and larvae. The upper layer of the sediment was gently discarded, and the remaining contents of each container were transferred to Eppendorf tubes and centrifuged at 9000 RPM for five minutes. The upper contents were carefully discarded again, and one drop of the sediment was combined with one drop of Lugol's iodine solution. A wet mount was then prepared and observed under a microscope at magnifications of 10× and 40×.

3.3. Data Analysis

Data were analyzed using IBM SPSS software version 21. The chi-square test was applied to determine statistical significance, with a significance level set at (P

< 0.05). Additionally, a One-Way ANCOVA was conducted to evaluate the relationship between vegetable contamination and key predictor variables while controlling for potential confounding variables.

3.4. Ethical Considerations

This article is based on an approved research project with the code 6418 and ethics code IR.BUMS.REC.1402.444. It was financially and scientifically supported by the Research Department and the Student Research Committee of Birjand University of Medical Sciences, Birjand, Iran.

4. Results

The results, based on the relative frequency distribution of parasites in the studied vegetables, are presented in Tables 1 - 4. Of the collected vegetable samples, 78 (39%) were found to be contaminated with parasites. The highest frequency of parasitic contamination was associated with Blastocystis (23.5%), followed by Giardia lamblia cysts (2%), free-living nematode larvae (2%), and Dicrocoelium dendriticum eggs (1%). The identified helminths and protozoa showed varying prevalence rates. A total of 11 samples (5.5%) of the isolated parasites were related to different types of helminths, while 55 samples (27.5%) corresponded to protozoa, and 12 samples (6%) showed simultaneous contamination. The highest number of parasite eggs observed were from Fasciola hepatica (1%) and Dicrocoelium dendriticum (1%), with the highest number of cysts being associated with Giardia lamblia (2%).

Based on the results presented in Table 1, a significant relationship was observed using the chi-square test between the frequency distribution of parasitic contamination and the type of consumed vegetables (P-value < 0.05).

According to Table 2, the highest frequency of parasitic contamination in the studied vegetables, based on cultivation areas, was found in samples harvested from the western part of Birjand (20, 51.3%). A significant relationship was observed using the chisquare test between the frequency distribution of parasitic contamination in the studied vegetables and cultivation location (P-value < 0.05).

Furthermore, the highest frequency of parasitic contamination based on collection areas was found in Workers' Street (12, 60%), followed by Ghafari Street (6, 54.5%), and Pasdaran Street (14, 50%) (Table 3). Based on

the results presented, no significant relationship was observed using the chi-square test between the frequency distribution of parasitic contamination in the studied vegetables and collection areas (P-value > 0.05).

A comparison of the frequency distribution of parasitic contamination in the studied vegetables by season (Table 4) showed that the highest contamination occurred in summer (64, 40%), followed by autumn (14, 35%). Based on the results presented, no significant relationship was observed using the chi-square test between the frequency distribution of parasitic contamination and the variables of collection areas, cultivation sites, and seasons of the vegetables (P-value > 0.05). However, a significant relationship was observed using the chi-square test between the frequency distribution of parasitic contamination and the type of consumed vegetables (Table 3) (P-value < 0.05).

5. Discussion

Iran has a predominantly dry and semi-arid climate, and water scarcity is a major issue. With a growing population and increasing demand for agricultural products and vegetables, farmers in some regions resort to using polluted wastewater and surface water for irrigation, leading to contamination of crops. Additionally, the use of human and animal manure for fertilization, the presence of contaminated animals such as dogs and cats in fields, the vastness of farms, and the lack of sanitary toilets in agricultural areas contribute significantly to product contamination.

Extensive studies have been conducted on vegetable contamination. Based on the present study, the parasitic contamination rate of vegetables consumed in Birjand, Iran, was found to be 39%, a significant concern given the central role of vegetables in people's diets. Studies in other regions of Iran report varying contamination rates: 31.5% in Qom, 16.3% in Sanandaj, and 19.5% in Tabriz (11). In this study, 11 samples (5.5%) of the isolated parasites were related to various types of helminths, 55 samples (27.5%) corresponded to protozoa, and 12 samples (6%) were related to simultaneous contamination.

Results from a study conducted in Malayer indicated a contamination rate of 14.6% with pathogenic and non-pathogenic parasites, including protozoan cysts (3.7%), helminth eggs (3.9%), and free-living larvae (7%) (12). Similarly, a study in Rasht, Guilan province, reported helminths and pathogenic protozoa in 4.1% of samples

Table 1 Comparing the Frequency	Distribution of Parasitic Contamination in	n the Studied Vegetables by Type a

Type of Vegetables	Negative Samples	Positive Samples	P-Value
Leek	15 (7.5)	6(3)	
Basil	5 (2.5)	5 (2.5)	
Mint	8 (4)	9 (4.5)	
Green onion	12 (6)	3 (1.5)	
Radish	6(3)	3 (1.5)	
Parsley	12 (6)	4 (2)	
Lettuce	6 (3)	6(3)	
Watercress	3 (1.5)	3 (1.5)	
Chives	3 (1.5)	6(3)	0.040 b
Cilantro	9 (4.5)	7(3.5)	0.040
Spinach	3 (1.5)	7(3.5)	
Tarragon	2 (1)	2 (1)	
Turnip	2 (1)	4(2)	
Chickweed	1(0.5)	0 (0.00)	
Alfalfa	0 (0.00)	3 (1.5)	
Green basil	11 (5.5)	3 (1.5)	
Dill	13 (6.5)	2 (1)	
Fenugreek	11 (5.5)	5 (2.5)	
Iotal	122 (61)	78 (39)	-

^a Values are expressed as frequency (%).

 $\textbf{Table 2.} Comparing the Frequency Distribution of Parasitic Contamination in the Studied Vegetables Based on Cultivation Location {}^{a}$

Sampling Locations	Negative Samples	Positive Samples	P-Value
Central city	18 (9)	10 (5)	
Western city	19 (9.5)	20 (10)	
Eastern city	31 (15.5)	18 (9)	0.543 ^b
Southern city	31 (15.5)	17 (8.5)	
Northern city	23 (11.5)	13 (6.5)	
Total	122 (61)	78 (39)	-

^a Values are expressed as frequency (%).

(7). Studies in other parts of the world have also investigated vegetable contamination by parasitic agents. In Turkey and Zambia, the contamination rates of consumed vegetables were reported to be 26.2% and 35.8%, respectively (13,14).

In this study, the most commonly isolated parasite was *Blastocystis* (23.5%). Studies conducted in Ilam and Sanandaj reported *Blastocystis* contamination rates of 4.29% and 0.8%, respectively (2, 10). The findings of this study regarding the presence of *Blastocystis* align with a study carried out in Lebanon (9). *Giardia* cysts, with a 2% contamination rate, ranked second. The contamination

rate of *Giardia* cysts in Zanjan was 3.1% (15), while rates in Qom and Ilam were 7.95% and 35%, respectively (2, 11), indicating a higher prevalence compared to this study. Given the pathogenic nature of *Giardia* and its transmission route to humans, the necessity of consuming properly sanitized vegetables becomes evident.

The contamination rate of *Taenia saginata* eggs was the lowest (0.5%) among the observed parasites, consistent with a study conducted in Qazvin (1.3%) (16), but lower than in Zanjan (23%) (15). Overall, contamination levels and types of parasites in

^b Chi-square test

^b Chi-square test

Table 3. Comparing the Frequence	y Distribution of Parasitic Contamination in the Studied	Vegetables Based on Collection Areas a

Collection Areas	Negative Samples	Positive Samples	P-Value
Amirabad	19 (9.5)	10 (5)	
Hajjiabad	12 (6)	7(3)	
Terminal	23 (11.5)	13 (6)	
Pasdaran	14 (7)	14 (7)	
Ghaffari	5 (2.5)	6 (4)	0.182
Jomhouri	26 (13)	12 (6)	
kargar	8 (4)	12 (6)	
Aliyeh	15 (7.5)	4 (2)	
Total	122 (61)	78 (39)	-

 $^{^{\}rm a}$ Values are expressed as frequency (%).

Table 4. Comparing the Frequency Distribution of Parasitic Contamination in the Studied Vegetables Based on the Seasons ^a

Seasons	Negative Samples	Positive Samples
Autumn	26 (61)	14 (39)
Summer	96 (60)	64 (40)
Total	122 (26)	78 (14)

a Values are expressed as frequency (%).

vegetables vary depending on health status, social and economic conditions, livestock and agricultural practices, geographic location, and the methods used for parasite identification.

In this study, spinach was identified as the most contaminated vegetable, with a 70% contamination rate. The higher contamination in spinach compared to other vegetables can be attributed to its broad leaves and greater contact with soil, which facilitates the establishment of parasites within the leaves. These findings are consistent with studies conducted in Izmir, Turkey (13). According to the results of this research, the lowest contamination among the studied vegetables was found in dill (13.3%) and scallions (20%).

Studies conducted in Tabriz reported scallions (8.6%) as the least contaminated vegetable after radishes, consistent with the findings of the current study (5). The highest frequency of parasitic contamination in the studied vegetables occurred in summer (40%), followed by autumn (35%). A study conducted in Rasht, Guilan Province, reported contamination rates of 5.2% in summer and 2.8% in autumn. Similarly, a study conducted in Hamadan reported summer contamination at 7.6% and autumn at 6.7%. These

assessments indicated higher contamination levels in summer than in autumn, aligning with the findings of the current study (17).

The highest frequency of parasitic contamination based on cultivation and growing areas was observed in vegetables collected from the western part of Birjand, Iran (20, 51.3%). In this region, most vegetables are sourced from neighboring counties and provinces, increasing the potential for contamination not only in the fields but also during harvesting transportation. A study conducted in Rasht, Guilan Province, revealed that the highest contamination was associated with locally grown vegetables (7.9%), while non-local vegetables accounted for 1.9% of contamination (7). Additionally, a study conducted in Sanandaj reported the highest parasitic contamination in vegetables harvested from outside the province at 81.6%, compared to 18.4% for in-province vegetables (10).

This study aimed to determine the level and type of parasitic contamination in vegetables consumed in Birjand, Iran. Human infection by parasites can occur through various routes, with oral transmission being one of the most significant. Consuming raw vegetables and salads is a major pathway for parasitic infections

^b Chi-square test

and a source of foodborne outbreaks. A significant portion of the vegetables consumed in Birjand, Iran, is sourced from neighboring counties and provinces. Based on the results of this study, it is essential to implement fundamental measures at every stage of planting, harvesting, storage, transportation, and distribution of vegetables. Moreover, public awareness regarding the washing and sanitization of vegetables must be enhanced.

One important limitation of this study was the lack of funding for further testing using more accurate diagnostic methods, such as molecular techniques.

5.1. Conclusions

Through comprehensive public health education, promoting proper disinfection practices before consuming raw vegetables, and raising awareness about the importance of thoroughly washing and sanitizing vegetables, the risk of parasitic infections transmitted through vegetables and food can be significantly reduced.

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Footnotes

Authors' Contribution: A. T. K. and M. Kh.: Made substantial contributions to the conception or the design of the manuscript; A. R., F. M., F. Z., and M. N.: Contributed to the acquisition, analysis, and interpretation of the data; All the authors participated in drafting the manuscript; All the authors read and approved the final version of the manuscript.

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