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The Parasitic Contamination of Farm Vegetables in Asadabad City, West of Iran, in 2014

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Abstract

Background: Consumption of fresh fruits and vegetables is a basic component of a healthy diet. Thus, the consumption of vegetables can have an important role in public health.

Objectives: Because of this concern, a prevalence study of parasitic contamination was carried out on raw vegetables to estimate the human risk of parasitic infections in Asadabad city, west of Iran.

Methods: In a cross-sectional study, 383 samples of different vegetables were obtained randomly from 12 vegetable farms in and around Asadabad. These samples included 10 types of seasonal vegetables: coriander, radish, spring onion, leek, parsley, tarragon, savory, basil, mint, and cress. The samples were examined by two parasitological methods: sedimentation and floatation techniques.

Results: Parasitic contaminations were detected in 34 (8.4%) vegetable samples, including five pathogenic and two non-pathogenic parasites. The parasites identified were *Giardia intestinalis* (1.6%), *Entamoeba coli* (2.6%), *Toxocara* spp. (0.8%), *Fasciola* spp. (0.5%), *Taenia* spp. (0.3%), *Dicrocoelium dendriticum* (0.3%), and free-living larvae (2.3%). Among the infested samples, coriander was the most contaminated vegetable (15.8%). The relationship between contamination of vegetables and untreated water used on farms was significant ($P < 0.001$).

Conclusions: The results implicate the importance of consumption of vegetables in the spread of parasitic diseases in the studied region. Thus, some basic hygiene measures should be carried out to improve public health and reduce infectious disease rates.

Keywords: Food Contamination, Iran, Parasites, Prevalence, Vegetables

1. Background

Fresh fruits and vegetables are two of the main components of a healthy diet. These foodstuffs are a significant source of nutrient and non-nutrient bioactive substances, which provide vitamins, minerals, dietary fiber, and phytochemicals, particularly antioxidants (1). Investigations have demonstrated that the incidence of some chronic diseases such as cardiovascular disease, cancer, diabetes, Alzheimer's disease, and cataracts can be decreased in the community by a healthy diet. Therefore, dietitians recommend consuming fruits and vegetables daily because of their role in health promotion and disease prevention (2). In spite of the advantages mentioned, the risk of microbial infections can be raised by the consumption of raw vegetables (3, 4). In developing countries, parasitic infections are a primary concern of public health and can lead to some serious health problems, such as malnutrition and growth retardation of children, as well as a considerable annual rate of morbidity and mortality (5). Poor sanitation and hygiene spread infectious diseases, especially intestinal infections, which affect millions of people. Food and waterborne diseases are major health problems in communities of both the developing and the developed world (3, 6). The incidence of intestinal parasitic infections is related to the consumption of raw vegetables (7, 8), and it is a common form of intestinal parasite transmission (6, 9). Contamination of farm vegetables occurs primarily during irrigation of farms with untreated sewage water and fertilization with animal manure and/or by the feces of stray dogs or other wild animals. For instance, a parasitic survey of dogs in rural areas of Hamadan demonstrated that 20.4% of them were positive for zoonotic parasites such as *Toxocara canis*, *Taenia/Echinococcus* spp., and *Giardia lamblia*. Contamination of vegetables can also occur during unhygienic post-harvest handling (3, 8, 10-12).

2. Objectives

Because of the importance of food hygiene in public health and the lack of information about this issue in the Hamadan province, the aim of this study was to determine the potential parasitic hazard of agricultural products and vegetables that are commonly eaten in Asadabad, west of Iran.

3. Methods

3.1. The Study Area

This study was conducted from July to October, 2014 in Asadabad, a city of the Hamadan province, located west of Hamadan. This city has a population of about 105,799 inhabitants (estimated in 2011) and an area of 1,195 square kilometers.

3.2. Sampling and Parasitic Analysis Procedure

A total of 383 vegetable samples were collected randomly from 12 farm lands, including savory, basil, tarragon, coriander, leek, mint, parsley, radish, cress, and spring onion. A clean nylon bag was used for sample collection for each of the specimens. Each sample labeled and its specifications recorded, and samples were then moved to the parasitology research laboratory at the Hamadan University of Medical Sciences. Samples were prepared based on standard methods previously described by other researchers (3, 8, 9) with some modifications. The mud and large particles attached to the vegetables were removed, and 250 grams of each sample was weighted and immersed into a clean container containing one liter of normal saline solution supplemented with detergent (1% sodium dodecyl sulfate, 1% Tween 80). After one hour of continuous shaking, the remaining wash water was transferred to a clean graduated cylinder and left to sediment for about 12 hours. After discarding the supernatant, the collected sediment was passed through a No. 40 sieve (425 μm) in order to remove coarse particles and facilitate the identification of larvae and free-living nematodes. The final volume of 50 mL was centrifuged at 500 g for 15 minutes, and the sediment was subjected to direct wet mount and flotation techniques. The tests were performed as follows: several wet mount slides were prepared for each individual sample and scanned by light microscope using low (100×) and high (400×) magnifications. The zinc sulphate flotation method was performed on the residual sample; after resolving the sediment in saturated zinc sulphate solution with a specific gravity of 1.8, the sample was centrifuged at 250 g for five minutes, after which a convex meniscus was formed by the additional solution and a coverslip was placed at the top of the tube (13). After 30 minutes, the coverslip was carefully removed, and the sample was examined using the direct wet mount method. Lugol's iodine and trichrome staining and morphological criteria were applied for identification of cysts of protozoa, larvae, and eggs of helminths.

3.3. Data Analysis

Data analysis was carried out by using a chi-square test and SPSS statistical software, version 16 to determine any correlation between parasitic contamination and the considered variables. P values less than 0.05 were reported as statistically significant.

4. Results

In the present study, 32 of the 383 vegetable samples (8.4%) were infested with parasites, including five pathogenic (*Giardia intestinalis*, *Toxocara* spp., *Fasciola* spp., *Dicrocoelium dendriticum*, and *Taenia* spp.) and two non-pathogenic parasites (*Entamoeba coli* and free-living larvae), with frequencies of 3.5% and 4.9%, respectively. The samples infected with protozoan cysts, helminth eggs, and free-living larvae were 4.2%, 1.9%, and 2.3%, respectively.

E. coli was the most prevalent parasite, detected in 2.3% of the vegetable samples. The highest rate of contamination was detected in coriander (15.8%) and the lowest was detected in basil and cress (2.6%) (P = 0.398). Types and frequencies of contamination in different vegetables are shown in Table 1.

In this study, 10 out of 12 investigated farms were irrigated with well water and two other farms with surface or waste water. Most of the contaminated samples (78.1%) were obtained from vegetable farms irrigated with surface or waste water. The relationships between parasitic contamination of vegetables and water and fertilizer used in the farms were significant (P < 0.001) and non-significant (P = 0.166), respectively (Table 2).

5. Discussion

Because of their essential nutrient and non-nutrient components, consumption of fruits and vegetables improves public health. Thus, it is strongly suggested by nutritionists (2). However, contamination of vegetables with different microbial agents can occur at farms and/or during processing for market, which can threaten public health (3, 8, 10, 11). Detection of parasites and other microbial agents from vegetables helps us identify possible sources of intestinal infections and could help us estimate the risk of infectious diseases in the community.

The findings of this investigation indicate that 8.4% of the vegetable samples from the farms located in the suburb of Asadabad were contaminated with parasites. In this regard, a number of reports from different parts of Iran and the world are available. Some prevalence rates of parasitic contamination of vegetables are 3.5% in Nigeria (7), 6.3% in Turkey (14), 31.7% in Egypt (11), 34.7% and 18.9% (in conventional and organic farms, respectively) in south-eastern Poland (15), 49.7% in Iraq (16), 58% in Libya (8), and 65.5% in Kenya (17). Other reports from Iran show

Table 1. Distribution of Protozoan Cysts and Larvae and Eggs of Helminths in Vegetables Used for Raw Consumption in Asadabad, Iran[a]

Vegetable Type	Pathogenic Parasites						Non-Pathogenic Parasites		
	Number	*G. intestinalis*	*D. dendriticum*	*Toxocara* spp	*Taenia* spp.	*Fasciola* spp.	*E.coli*	*free-living larvae*	Total
Radish	38	0 (0)	0 (0)	0 (0)	1 (2.6)	0 (0)	1 (2.6)	0 (0)	2 (5.3)
Cress	38	0 (0)	0 (0)	0 (0)	0 (0)	1 (2.6)	0 (0)	0 (0)	1 (2.6)
Spring onion	38	1 (2.6)	0 (0)	1 (2.6)	0 (0)	0 (0)	1 (2.6)	2 (5.3)	5 (13.2)
Savory	38	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	2 (5.3)	1 (2.6)	3 (7.9)
Mint	38	1 (2.6)	0 (0)	1 (2.6)	0 (0)	0 (0)	2 (5.3)	1 (2.6)	5 (13.2)
Basil	38	1 (2.6)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (2.6)
Parsley	38	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	2 (5.3)	2 (5.3)
Tarragon	38	2 (5.3)	0 (0)	1 (2.6)	0 (0)	0 (0)	1 (2.6)	0 (0)	4 (10.5)
Coriander	38	1 (2.6)	1 (2.6)	0 (0)	0 (0)	1 (2.6)	2 (5.3)	1 (2.6)	6 (15.8)
Leek	41	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (2.6)	2 (5.3)	3 (7.3)
Total	383	6 (1.6)	1 (0.3)	3 (0.8)	1 (0.3)	2 (0.5)	10 (2.6)	9 (2.3)	32 (8.4)

[a] Values are expressed as No. (%).

Table 2. Distribution of Parasitic Contaminations in Vegetables Grown on Farms With Different Types of Water and Fertilizer Consumption in Asadabad, Iran[a]

Water and Fertilizer Consumption	Contamination[b]		Total
	Positive	Negative	
Well water	7 (2.1)	328 (97.9)	335 (100)
Surface and waste water	25 (52.1)	23 (47.9)	48 (100)
Animal manure	29 (9.5)	276 (90.5)	305 (100)
Chemical manure	3 (3.8)	75 (96.2)	78 (100)

[a] Values are expressed as No. (%).
[b] Parasitic contamination.

various rates of contamination, including 43.7% in southern Iran (18), 46.5% in Amol (6), 52.7% in Khorramabad (13), and 71% in Ardebil (19). Seasonal variation may be responsible for different prevalence rates of vegetable contamination. Moreover, some other influencing factors include methods used in research, different rates of intestinal parasitic infection in the areas, weather conditions, type of water and fertilizer utilized for cultivation of vegetables in farm lands, and other epidemiological factors.

E. coli was the most abundant parasite (2.6%), followed by the free-living larvae found in this survey. Detection of *E. coli*, a non-pathogenic intestinal parasite, indicates contamination of vegetables by human feces and the existence of a potential risk factor for intestinal infectious diseases in raw vegetable consumers. Previously reported rates of *E. coli* contamination from different parts of Iran are as follows: 2.8% in Qazvin (20), 2.9% and 8.4% in winter and spring, respectively in Khorramabad (13), 10 and 18% in imported and native vegetables, respectively in Ardebil (19), and 19.3% in Amol (6). In a recent study conducted by Jafari et al. in the rural areas of Hamadan city, *E. coli* was the most common parasite (18.9%) in stool samples collected from the rural population (21).

Free-living larvae were the second-most common parasite, with a contamination rate of 2.3%. The studies performed in Shahrekord, Khorramabad, and Amol reported a 3.3%, 22.9%, and 25.8% prevalence of free-living larvae, respectively (6, 10, 13). The remarkable prevalence of this parasite in the vegetable samples may be due to the high prevalence of free-living and plant-parasitic nematodes in the soil.

G. intestinalis cyst was the third most common parasite, which was detected in 1.6% of the samples. This finding is consistent with results reported by Shahnazi et al. from Qazvin (1.6%) (20), but it is lower than the results obtained from other parts of the country reported by Ezatpour et al. (1.1% in winter and 5.8% in spring) (13), Daryani et al. (10% in imported and 18% in native vegetables) (19), Siyadatpanah et al. (22.5%) (6), Ebrahimzadeh et al. (8.1%) (9), and Olyaei et al. (11%) (18).

In this study, eggs of *Ascaris lumbricoides*, *Trichuris trichiura*, and other common intestinal nematodes were not identified in the samples. Until a few years ago, these parasites, especially *A. lumbricoides*, were common parasites in the Hamadan province. In 2003, the prevalence rate of *A. lumbricoides* was 19.5% in Hamadan city (22). A number of local recent studies, such as those conducted by Jafari et al. in 2012 (21) and Fallah et al. in 2014 (unpublished data), show a considerable decrease in the rates of intestinal parasitic infections in the area. This success is due to measures in the past years, such as performance of health promotion programs and extensive chemotherapy plans in the community (23).

Toxocariasis is a zoonotic parasitic disease caused by the ingestion of embryonated eggs of *Toxocara* spp., which

is an ascarid of carnivores, particularly dogs and cats. This infection involves visceral and ocular toxocariasis, particularly in children. The eggs of *Toxocara* spp. are often widespread in the environment all over the world because of the presence of dogs, cats, and other wild carnivores. In this study, recovery of *Toxocara* spp. eggs from vegetable samples is consistent with some investigations (6, 8-10, 15, 18) and inconsistent with others (13, 16, 17, 19). This finding and the other report from the area (12) indicate the presence of a potential hazard for toxocariasis.

Taeniid eggs, including those of *Taenia* and *Echinococcus* spp., which are morphologically indistinguishable, were detected in 0.3% of the specimens. The presence of these eggs was reported previously by Shahnazi et al. (1.8%) (20), Ebrahimzadeh et al. (13.1%) (9), Fallah et al. (9.2%) (10), Olyaei et al. (45.7%) (18), and Daryani et al. (14% in imported vegetables and 16% in native vegetables) (19). However, eggs of *Taenia/Echinococcus* spp. were not identified in the two studies conducted by Siyadatpanah et al. and Ezatpour et al. in Amol and Khorramabad, respectively (6, 13).

Eggs of *Fasciola* spp. and *D. dendriticum*, two species of zoonotic trematodes, were found in this survey at rates of 0.5% and 0.3%, respectively. Principally, contamination of vegetables with zoonotic parasites is related to the prevalence of the parasites in farm animals and the use of animal manure on farms as fertilizer.

In this study, among the infested samples, coriander was the most contaminated vegetable (15.8%). This is inconsistent with some studies, which reported leek as the most commonly contaminated vegetable (13, 16, 20).

In conclusion, although the evidence indicates that the prevalence of intestinal parasitic infections is decreasing in the region, the results of this study emphasize the importance of consumption of raw vegetables in the spread of infectious diseases. Thus, it is necessary to take hygienic measures to improve public health and health standards in the area.

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Footnotes

Authors' Contribution: Tayebeh Shamsi-Ehsan contributed to sample collection and experiments; Amir Hossein Maghsood contributed to study design; Mohammad Matini contributed at all stages.

Conflicts of Interest: The authors have no conflicts of interest to declare.

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