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Parasitological Contamination of Fresh Vegetables and Its Prevalence in Dessie Town, Northeast Ethiopia

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Abstract
Background: Consumption of vegetables and fruits is vital for health as they are good sources of carbohydrate, vitamins, minerals and fiber contents. On the other hand, vegetables and fruits are potential vehicles for the transmission of pathogenic parasites and protozoa that are transmitted through fecal-oral route.
Objectives: The objective of this study was to assess the parasitic contamination of vegetable samples collected from farms, home gardens and street vendors in Dessie town.
Material and Methods: A Cross-sectional study was conducted from April to June, 2014 to determine the level of parasitic contamination of vegetables and fruits sold in Dessie town. A total of 150 samples of different type of fruits and vegetables were washed with physiological saline, followed by vigorous hand shaking in a clean plastic bag for 15 minutes and then examined using sedimentation concentration technique. Parasites stages were identified based on WHO guideline.
Results: Out of 150 vegetable samples collected, 95(63.4%) of the vegetables were contaminated with at least one parasite. *E.histolytica*/*dispar* (24%) was the most frequently detected parasite with*G.lamblia* cyst (1.33%) the least frequently detected one.The result also shows that therewas positive relation between types of vegetable and sites or sample area with p-value<0.000.
Conclusions: The present study showed that there is a risk of acquiring parasitic infections from consumption of raw vegetables and fruits in Dessie town. Thus efforts should be made to aware the public and restaurant owners in particular to properly wash or disinfect raw vegetables before consumption.
Keywords: Parasite, Contamination, vegetables, Dessie, Ethiopia

1. Background

Consumption of vegetables and fruits is vital for health as they are good sources of carbohydrate, vitamins, minerals and fiber contents. WHO consultation on diet, nutrition and the prevention of chronic diseases has recommended a daily intake of 400 grams of vegetables and fruits for prevention of chronic diseases such as heart diseases, cancer, diabetes and obesity [1]. On the other hand, vegetables and fruits are potential vehicles for the transmission of pathogenic parasites and protozoa that are transmitted through fecal-oral route [6, 7].

The consumption of enteric parasite contaminated food leads to morbidity and mortality with a negative socio-economic impact as a result of the cost of treatment and subsequent hospitalization [6, 10].Globally, intestinal parasitic infections (IPIs), including helminthes and protozoa, remain endemic. About one-third of the world's populations, more than 2 billion people, are infected with the largest majority being children. *A. lumbricoides* is the largest and most common helminthes, infecting about 819 million people globally; this is followed by *T. trichiura*, which infects about 464.6 million, and hookworm infecting about 438.9 million people globally. Giardiasis and Amebiasis, caused by *Giardia duodenalis* and the pathogenic *Entamoeba spp*., respectively, are the two most prevalent protozoan infections, infecting about 200 million and 500 million people, respectively [2].

Food borne illness caused by contaminated vegetables and fruits are still a public health problem in the developing countries, those which are sold on streets in the markets and directly on garden areas. Vegetables become a source of infection through contamination during production, collection, transport and preparation and the sources of contamination are usually feces and fecal contaminated soil or water [14]. Particularly, vegetables which are eaten rawand without peeling can be agents of intestinal protozoan [8, 9] and helminthes infection [9, 23].

Developing countries like Ethiopia have high risk of acquiring parasitic infections due to poor sanitation, and substandard and crowded living condition. Some studies have reported the prevalence of parasitic contamination of vegetables indifferent parts of Ethiopia (16, 19, 22, 28).To the best of our knowledge, there is no published document about the level of parasitological contamination of vegetables in Dessie town. Therefore, the objective of this study was to determine the contamination levelof intestinal parasites from fresh vegetables sold in Desse town, northeast Ethiopia.

2. Material and methods

2.1 Description of the study area

This cross-sectional survey study was conducted in Dessie town, the zonal town of South Wollo, Northeast Ethiopia from April to June, 2014. The town is located on Addis Ababa-Mekelle road at an altitude and longitude of 11°8'N 39°38'E with an elevation between 2,470 and 2,550 meters above sea level.According to the town has a total population of 136,056. Borkona River in to which sewage and feces is released passed through the middle of the town. Farmers living along the length of the wastewater drainage canal adjacent to the river cultivate vegetables for both household consumption and income generation.

2.2 Sample collection

Five types of vegetables which are common in the study area were selected from home gardens following Borkena River, Dose River, Memhir Akalewold, Ehil Sebil and main vegetable vendors in the town. A total of 150 vegetable samples were collected, comprising 30 samples for each vegetables; tomatoes, cabbage, carrot, lettuce andSpinach. The vegetable samples were collected using sterile, labeled polythene bags and transported to the laboratory of biology department, college of Natural Sciences, Wollo University. Samples were prepared for examination within 3 hours of collection. Samples collected from home gardens of farmers were based on the verbal consent and agreement of owners. Samples with excessive dirt and spoilage were not included in the study.

2.3 Parasitological processing and analysis of samples

Each fresh vegetable sample was chopped into pieces and weighted to 200 grams; it then washed in physiological saline solution (0.85% NaCl) in sterile plastic bags. After removing the chopped fragments using clean sterile forceps, the washing water was then allowed to sediment for 10 hours at room temperature. The top water was removed leaving about 5 ml of the sediment. The sediment was then centrifuged (centrifuge model.no AVI 5580, India) at 3000 rpmfor 5minutes.After the supernatant was discarded, three lugol stained slides were prepared from each sample residue and examined using light microscope for helminthes ova and protozoan cysts [11, 12]. Parasites were identified based on WHO guideline [13].

2.4 Data analysis

Data analysis was made using simple descriptive statistics and frequency using SPSS version 20. Chi-square test was used to determine p-value for significance relationship of sample area and parasite positivity of vegetables. .

3. Result

Out of 150 vegetable samples collected from two river basin urban gardens (Dose and Borkena river) and 3 vendors in Dessie town, 95(63.4%) of the vegetables were contaminated with at least one intestinal parasite that is indicative of potential of transmission to susceptible hosts.Of these five vegetables examined, major contamination for different parasites was recorded in Spinach (80%) followed by Lettuce (73.3%), cabbage (66.7%), carrot (63.3%) and tomato (46.7%) (**Table1**)

Table1**.** Distribution of intestinal parasites among five vegetables consumed in Dessie town from April to June,2014

Sampled Vegetable	**Sampled areas**					**Any parasite Positives** (%)
	Dose river	MemhrAkalewold	Ehilsable	Borkena river	Arada vendors	
Tomato (*Solanumlycpersican)* *n=30*	0	0	0	7	3	10 (33.4)
Cabbage (*Brassiaolaracea)* *n=30*	8	4	1	6	1	20(66.7)
Lettuce (*Latuca SPP)* *n=30*	10	4	3	4	1	22(73.3)
Carrot (*Daucuscarrota)* n=30	0	0	10	5	4	19(63.3)
Spinach (*Spinaceaoleracea)*	11	4	2	5	2	24(80.0)
Total (n= 150)	29	12	16	27	11	95(63.4)

Two intestinal protozoan parasites, *Entamoebahistolytica/dispar, Giardia lamblia* and a comonsal *Entamoeba coli* were detected in the sampled vegetables. Besides, five intestinal helminths; *Ascarislumbricoides*, Hook worms, *Trichuristrichiura*, *Taenia spp.* and *Strongloidesstercoralis*larvae were also detected (**Table**

2).The percentages of vegetables parasite distribution in the five regions of Dessie town are different. For a sample of 30 for each vegetable, the percentage of parasites distribution was higher (88% and 73.3%) in Spanish (*Spinaceaoleracea*) and Lettuce (*Lactucaspp*),respectively (**Figure1**).

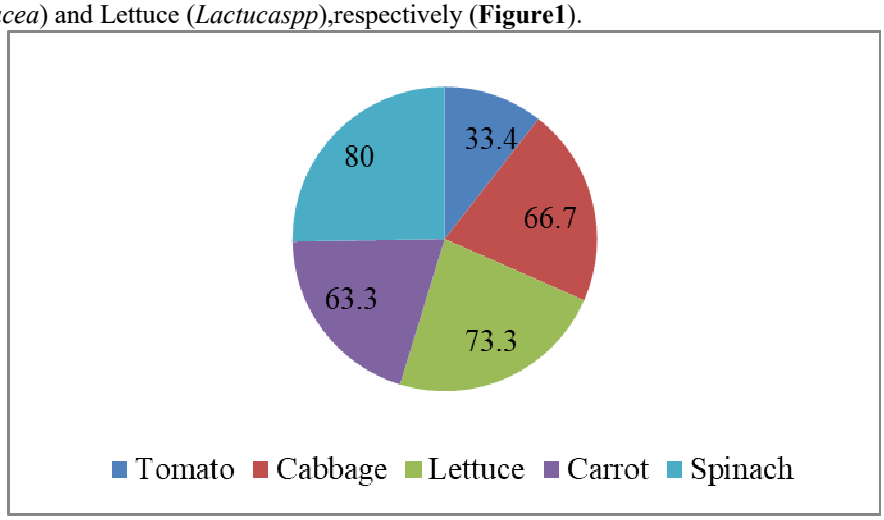

Figure 1: The percentage of intestinal parasites positivity distribution among five vegetables consumed in Dessie town from April to June, 2014.

The results also showed that samples collected from Borkona (66.7%) had the highest contamination rate, followed by samples collected from Dose (55.6%), Memhre Akalewold (53.3%), Ehilsebil(42.2%) and Arada vendors (11.5%). In general parasite stages werefrequently detected in Dose and Borkena River than in the other places(**figure 2**), in which of the total 30 samples,29 and 27 parasite stages were detected respectively.

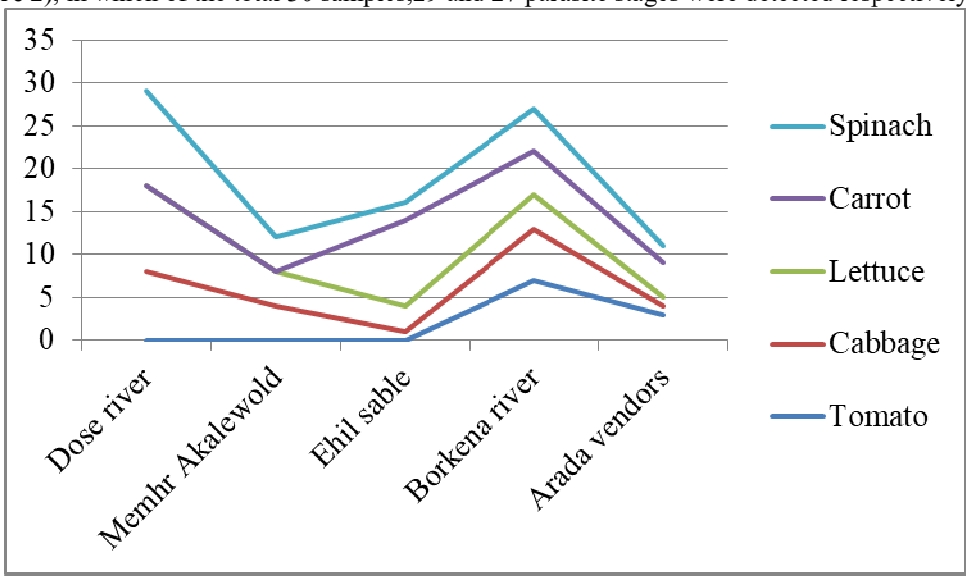

Figure 2**:** Intestinal parasites positivity distribution among five vegetables consumed areas in Dessie town from April to June, 2014.

In this study, *E.histolytica*/*dispar*cyct was the most frequently detected (24%) parasite stage followed by *Entamoeba coli*cyst and ova of *Ascarislumbricoides*(9.34 % each),*Hook worm*(6%), *Trichuristrichuria*(4%), *Taenia spp. (*2.67%) and *G.lamblia* cyst(1.33%) the least detected (Table2.)

Table 2.The prevalence of the different stages of helminthes and protozoan parasite in sampled vegetables.

parasites detected	Number positives (%)					
	Tomato *(n=30)*	Lettuce *(n=30)*	Cabbage *(n=30)*	Carrot *(n=30)*	Spinach *(n=30)*	Total (n=150)
Ascarislumbricoides	0	5	2	3	4	14 (9.34)
Hook worm	2	0	0	4	3	9 (6)
Trichuristrichuria	0	0	4	0	2	6 (4.00)
Taenia spp.	2	0	0	0	2	4 (2.67)
Strongloidesstercoralis larvae	4	2	2	2	0	10 (6.67)
Entamoebahistolytica/dispar	0	13	6	8	9	36 (24.00)
Giardia lamblia cyst	0	0	2	0	0	2 (1.33)
*Entamoeba coli**	2	2	4	2	4	14 (9.34)

Where *is commensal not parasite; it can predict the level of fecal contamination.

The result given in table 3 indicates that there was a relation between positivity of types of vegetable and sites or sample area with p-value<0.000. The chi-square test indicates rejection of the null hypothesis which states there was no relationship between positivity types of vegetables and sample area. Therefore, the sample area has an effect on the positivity of parasites of the four vegetables at 1% level of significance.

Table 3: Result of chi-square test of independence between positivity of parasites in the vegetables and sample area

vegetable conaminated * Sample area Crosstabulation

			Sample area					
			dose river	memhir akalewolde	ehil sable	Borkena river	Arada vendor	Total
vegetable conaminated	Tomato	Count	0	0	0	7	3	10
		Expected Count	3.1	1.3	1.7	2.8	1.2	10.0
	Cabbage	Count	8	4	1	6	1	20
		Expected Count	6.1	2.5	3.4	5.7	2.3	20.0
	Lettuce	Count	10	4	3	4	1	22
		Expected Count	6.7	2.8	3.7	6.3	2.5	22.0
	Carrot	Count	0	0	10	5	4	19
		Expected Count	5.8	2.4	3.2	5.4	2.2	19.0
	Spinach	Count	11	4	2	5	2	24
		Expected Count	7.3	3.0	4.0	6.8	2.8	24.0
Total		Count	29	12	16	27	11	95
		Expected Count	29.0	12.0	16.0	27.0	11.0	95.0

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	50.960	16	.000
Likelihood Ratio	57.763	16	.000
Linear-by-Linear Association	2.436	1	.119
N of Valid Cases	95		

4. Discussion

The present studyshowed that vegetable samples collected from the rivers, home gardens and street vendors of Arada in Dessie town werecontaminated with helminthes and protozoan parasites.Of the samples, 63.4%were found to be contaminated with at least one egg, cyst or larva of helminthes and protozoan parasites.The high rate of parasitic contamination observed in this study might be attributed to waste and contaminated water irrigation, poor hygienic and sanitary handling, poor transport and packaging of the vegetables. Contamination of soil with animal wastes and increased application of improperly composted manure to soil in which vegetables are grown might also be the cause for this contamination[23]

Table1 shows the frequency distribution of contamination for each vegetable. Spinach was highly contaminated with a frequency of 24 (80 %), followed by Lettuce 22 (73 %), Cabbage 20 (66.7 %), Carrot 19 (63 %) and Tomato 10 (33.4%). The results are in accordance to [19] who found high contamination profile of leafy vegetables in Eastern Showa, Ethiopia and with [11,12] who reported high parasitic egg contamination of

leafy vegetables followed by root and fruit vegetables in Hanoi, Vietnam. This may be due to the fact that Cabbage, lettuce and other green leafy vegetables had uneven surfaces that make parasitic eggs, cysts and larvae attached to their surface more easily, when washed with contaminated water either in the farm or market [22, 27].

Besides, 19 (63.3 %) of carrot samples were found contaminated in the present study. Similar result was obtained by [18]. This might be attributing to the specialized rough skin, the pits and crevices in this vegetable retain some dirt which may not be easily removed by slight washing in the field or retail outlet [15, 7].The least number of parasites contaminations were found on Tomato 10 (33.4%) in this study. This result is in accordance with [15] who reported that fruit vegetables including egg plant, cucumber,tomato and chili were least contaminated (3%) as compared with leafy (31%) and root vegetables (17%). This is due to the fact that these vegetables have smooth skin which enhances the removal of parasite eggs by slight washing.

In the present study, of the parasites detected, *Entoamoebahistolytica/dispar*cyst was the most frequently detected, followed by, *Ascaris Lambricoides, E. coli, Strongloid Stercoralis* larvae, Hook worm,*Trichuristrichuria*, *TaeniaSpp* and *Giardia lamblia* cyst, respectively(**table 2**). These results are similar to [17] who studied the prevalence of different parasitic stages in commercial vegetables in Al-Nassriah city, Iraq and found the highest contamination of *E. histolytica. Ascarislambricoides* egg was reported as the second most prevalent parasitic stage. This report is similar with the previous study in Jimma town, Ethiopia by [16] who reported *A. Lambricoides* as the most prevalent parasitic stage. This result also corroborate with [18] who reported *Ascaris Lambricoides* as the second most prevalent(13%) of the vegetables studiedin Casablanca,Morocco.

Entamoebacoli cyst was reported as the third most prevalent (16%). Similar result was obtained in south western Saudi Arabia[21]. Although *E.coli*is a commensal intestinal parasite,itspresence in high level in the sampled vegetables indicates the presence of high prevalence of contamination of the pathogenic intestinal and protozoan parasites in the study area.

Strongloid stercorlis larvae were detected as the fourth most prevalent (6.6%) of the vegetable samples In line with this [19] reported *Strongloide sercoralis* larvae as the most encountered parasite (91.2%) of the studied vegetables in Mekelle city, Ethiopia. This high prevalence might be due to the life cycle of the parasite which has both parasitic and free living state enhances the proliferation of larvae without the host [23]. Hook worm ova was also detected as a fifth prevalent parasite stage (6.6%) of the vegetables studied. In contorary to this result different studies do not detect the ova of hook worm [22, 24]. The differences might be attributed to differences in geographical locations, climate conditions, and the type of soil.The rest of the parasites detected in the present study including: *Trichuristrichuria*, *Giardia lamblia* and *Taenia* species was reported with a prevalence of 4%, 1.3% and 1.3% respectively, with *Giardia* and *Taenia* species the least detected. These results are in agreement with [26] who reported 1.33 %, 0.33% and 0.33% for the three species respectively with the least prevalence of *Taeniaspp* and *Trichuristrichuria.*

Several studies on recovery of parasites from vegetables have been conducted in Ethiopia and the prevalence was high in all examined vegetables and parasites such as *Entamoebahistolytica*, *E. coli*, *Jiardialamblia* and *Strongloidesstercoralis* have been reported. A study by [16] on contamination of sewage irrigated vegetables in eastern Showa reported high contamination of *Ascarislambricoides,* followed by *Entoamoebahistolytica* (18.18%) and *Jiardialamblia.* A study by [19] on the other hand reported high contamination of waste water irrigated vegetables in Mekelle city with *Strongloide stercoralis* (63.63%) the most encountered contaminant followed by *Taenia* (18.8%) and *Entamoeba spp* (18.8%).These observed differences in prevalence rates of the different pathogenic intestinal parasites from fresh vegetables is expected. Several factors may contribute to such differences. These may include, among other factors, geographical location, type and number of samples examined, methods used for detection of the intestinal parasites, type of water used for irrigation, and post harvesting handling methods of such vegetables [25].Similarly, the chi-square test in the present study showed that each sample sites or area contributes for positivity of parasites of vegetable, and there is a relationship on sample area and parasite positivity of vegetables.

5. Conclusion and recommendation

The present study clearly showed that raw vegetable samples from river, home gardens and market in Dessie town are contaminated with parasitic stages, indicating risk of acquiring infection from consumption of raw vegetables in the town.Therefore; farmers need to be educated on health risks of using waste water to irrigate vegetables. In addition, awaring the public in general and restaurant owners in particular about the importance of properly washing or disinfecting raw vegetables before consumption is needed. Furthermore, health authorities should take measures to improve the sanitary condition of the area where vegetables are growing by constructing latrines and educating the community to avoid contamination of water with feces.

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Conflict of interest
The authors declare that there is no conflict of interests regarding the publication of this article

Authors Contribution
Mr. Gebrehiwot Kiros collected and processed the samples and also involved in microscopic detection of parasites. Mr.Elifaged Hailemeskel designed and involved in the laboratory activities, Mr. Tilaye Matebe organized and analyzed the data obtained from the Laboratory results.

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