

Pre harvest and post-harvest assessment of geohelminth and protozoan parasites contamination of vegetables in Ibadan, Southwest, Nigeria

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Abstract

Consumption of fresh vegetables is one of the major routes of transmission of intestinal parasites of public health importance which have been incriminated as one of the main causes of foodborne outbreaks. In an attempt to bridge the knowledge gap associated with the pre-harvest and post-harvest parasitic contamination of vegetables in Nigeria, this study was conducted to assess geohelminths and protozoa contamination of vegetables in farms and markets in Ibadan, southwest, Nigeria. Vegetable samples were screened using simple floatation and sedimentation methods for identification of geohelminths ova, larva and protozoan oocysts. Of the total 200 vegetable samples collected, 70 (35%) were positive for geohelminth and protozoan parasites. The results show that the highest level of contamination was detected in African spinach 20 (35.1%), followed by White jute 15 (45.5%), Waterleaf 14 (51.9%), Quill grass 12 (24.5%), Garden eggplant leaf 4 (57.1%), while the least number of contaminated samples was detected in Pepper 1 (%). However, Fluted pumpkin leaves and English Spinach had no parasitic contamination. Waterleaf 3 (11.11%), African spinach 2 (3.5%), Quill grass 2 (4.1%) and White jute 2 (6.1%) had mixed helminths and protozoa infections. Of the total intestinal parasites observed, *Strongyloides stercoralis* had the highest prevalence 67 (33.5%), followed by *Entamoeba coli* 27 (13.5%), *Ascaris* spp. 24 (12%), *Trichuris* spp. 10 (5%), *Filariform* 9 (4.5%), *Dicrocoelium* spp. 9 (4.5%), *Oesophagostomum dentatum* 7 (3.5%), *Fasciola* spp 7 (3.5%), *Entamoeba histolytica* 6 (3%) while *Eimeria* spp 6 (3%) had the lowest prevalence. Most farmers 44 (80%) used stream and waste water as their source of irrigation while 11 (20%) of the farmers used well and stream. The relatively high rate of parasitic contamination observed in this study might be due to poor hygienic and sanitary handling of vegetables, poor transportation, packaging of vegetables and contaminated water used for irrigation and washing of vegetables in farms and markets. There is therefore a need for education of farmers and market women on proper handling and hygienic management of vegetables to safe guard the health of the populace.

Keywords: Intestinal parasites, vegetables, farms, markets, Nigeria.

INTRODUCTION

Vegetables contain valuable food components which have been strongly associated with maintenance of good health and prevention of diseases (Hanif et al., 2006; Keatinge et al., 2010). Vegetables can become contaminated with enteric parasitic pathogens throughout the process of planting to consumption and parasites from contaminated vegetables can pose potential health risk to humans; therefore food safety irrespective of the specific food item should be of utmost concern to everybody involved in the food chain (Amoah et al., 2006; Kays, 2011). Vegetables are sometimes eaten raw or parboiled to retain the natural taste and preserve heat labile nutrients (Said, 2012; Slifko et al.,

2009). This practice favours the likelihood of food-borne parasitic infections in humans (Sunil et al., 2014). The level of contamination depends on numerous factors which include the use of untreated waste water and water supplies contaminated with sewage for irrigation (Simoes et al., 2011; Beuchat, 2002; Amoah et al., 2006). The contamination of vegetables with geohelminths and food borne protozoa can occur at any time during the food chain, this can be pre-harvest: occurring on farms which include soil, faeces (human and animal origin) and water (irrigation, cleaning) (Halablab et al., 2011; Amoah et al., 2009). Also, contamination may be post-harvest, occurring during transportation and in market places which include rinsing and sprinkling with contaminated water (Mensah et al., 2002; Alade et al., 2013; Olyaei &

Hajivandi, 2013). Over the years, very few studies have been conducted on parasitic contamination of vegetables in Nigeria (Obetta et al., 2011; Elom et al., 2012; Alade et al., 2013). All these studies focused on vegetables from open markets (during post-harvest periods). However, none has considered the assessment of pre and post-harvest parasitic contamination of vegetables from farms and markets in Nigeria. This present study was therefore conducted to assess parasitic contamination on vegetables from selected farms and markets in Ibadan. Information obtained from this assessment may help in the development of preventive and control strategies that will raise people's awareness and safeguard the health and welfare of the community. This information is also required to assist the local health authorities to be able to take the proper actions to improve the quality of vegetables being sold to the populace for consumption.

MATERIALS AND METHODOLOGY

Study Design and Sample Collection

200 vegetables were sampled from three major farms from which 55 farmers practice oleiculture and from five main commercial markets from six Local Government areas in Ibadan, Southwestern, Nigeria. Eight common vegetables eaten by the people in the area were used for this study. The vegetables used are Tomatoes (*Lycopersicum sativus*), Water leaf (*Talinium triangulare*), Quill grass (*Celosia argenta*), African spinach (*Amaranthus hybridus*), White jute (*Corchorus olitorius*), Fluted pumpkin leaf (*Telfaria occidentalis*), Garden eggplant leaf (*Solanum macrocarpon*) and Pepper (*Capsicum Chines*e). These were chosen because they are the major vegetables eaten by the residents of Ibadan. A total of 1100 vegetables strands pooled into 20 strands per sample making 55 samples were collected from farms while a total of 2900 vegetables strands pooled into 20 strands per sample making up to 145 samples were bought from sellers in each of the open markets selected for the study making a total of 200 samples of vegetables.

Parasitological Analysis

Floatation Method

100-200 g samples of each vegetable were washed in 0.85% saline solution in a plastic container for 10-24 h for the removal of parasitic ova, larva or cysts. The suspension was strained through a sterile sieve to remove debris and then centrifuged at 4000 rpm for 5 min and the supernatant discarded into the disinfectant jar. The sediment obtained was re-suspended in sodium chloride floatation fluid and re-centrifuged. The floatation fluid was added to fill to the brim and a cover slip was superimposed on it. The cover slip was lifted, placed on the glass slide and examined under microscope using X100 and

X400 objectives (Nyarango et al., 2008). The parasite eggs was identified on the basis of their morphological characteristics shape and size, and compared with standard eggs on charts using The Bench Aid for the Diagnosis of Intestinal Parasites.

Sedimentation method

100-200 g samples of vegetable was washed in 0.85% saline solution in a 15 ml test tube for 5 h and then centrifuged at 3500 rpm for 5 min (Damen et al., 2008). A drop of the sediment was mixed with two drops of lugol's iodine solution. The process was systematically repeated until the mixture in each test tube was exhausted and examined under a light microscope using X100 and X400 objectives (Garcia, 2007). Ova, oocysts and larvae of parasites found under the light microscope were identified as previously described by (Downes & Ito, 2001).

Statistical Analysis

Data was entered into a Microsoft excel spread sheet and analyzed using descriptive analysis. Graphs and tables were used to calculate distribution frequency of geohelminths with regards to vegetable species, locations sampled and method of irrigation.

RESULT

Of the total 200 vegetable samples examined, 70 (35%) were positive for geohelminths and protozoan parasite. The parasites detected were *Strongyloides stercoralis*, *Ascaris* spp., *Trichuris* spp., *Oesophagostomum dentatum*, *Fasciola* spp., *Dicrocoelium* spp., *Entamoeba histolytica*, *Entamoeba coli* and *Eimeria* spp. *Strongyloides stercoralis* has the highest prevalence 67 (33.5%), followed by *Entamoeba coli* 27 (13.5%), *Ascaris* spp. 24 (12%), *Trichuris* spp. 10 (5%), *Filariform* 9 (4.5%), *Dicrocoelium* spp. 9 (4.5%), *Oesophagostomum dentatum* 7 (3.5%), *Fasciola* spp 7 (3.5%), *Entamoeba histolytica* 6 (3%) and *Eimeria* spp. 6 (3%) had the lowest prevalence (Figures 1 and 2). Out of the six local Government area investigated, Lagelu has the highest level of parasitic contamination 13 (100%) of the total 13 vegetables collected with *Strongyloides stercoralis* being the most prevalent 8 (61.5%). This is followed by Ibadan southwest 23 (57.5%), Ibadan north 22 (32.4%), Akinyele local government 11 (29%), while Ido local government has the lowest level of parasitic contamination 3 (8.8%) (Table 1). Of the six markets investigated, Oja Oba market has the highest level of parasitic contamination with 23 (57.5%), followed by Ojo market 10 (37.04%), Bodija market 13 (35.14%), Oje market 2 (28.6%) and Eleyele market 3 (8.82%) (Table 1). The highest level of contaminated was detected in African spinach 20 (35.1%), followed by White jute 15 (45.5%), Waterleaf 14 (51.9%), Quill grass 12 (24.5%), Garden eggplant leaves 4 (57.1%), while the least contamination was detected in Pepper 1 (50

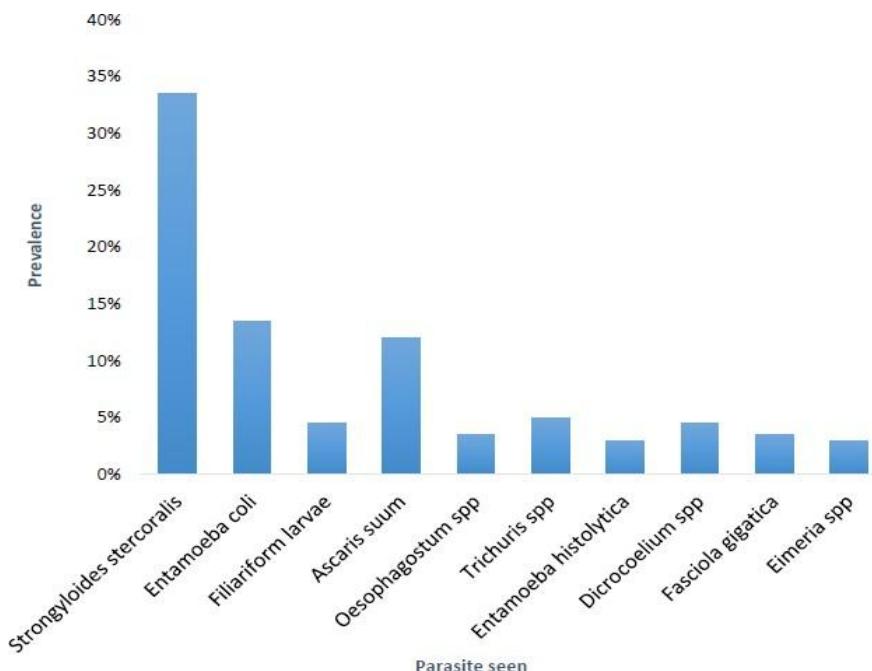


Figure 1: Percentage Prevalence of Parasites found in farms and markets.

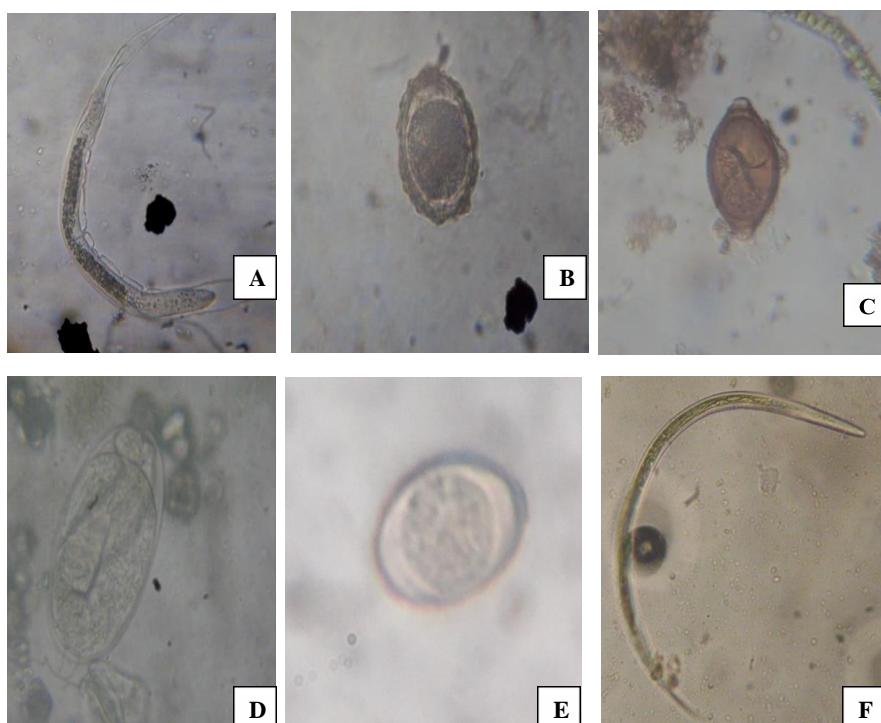


Figure 2: showing some of the geohelminths egg and larva with protozoa oocyst detected in different vegetable species collected in Ibadan metropolis.

A: *Oesophagostomum dentatum* larvae, **B:** *Ascaris* spp, **C:** *Trichuris* spp.

D: *Strongyloides stercoralis* egg **E:** *Eimeria* spp. **F:** *Strongyloides stercoralis* larva (first stage)

%), however, fluted pumpkin leaves and English spinach had no parasitic contamination. The result showed that Waterleaf 3 (11.1%), African spinach 2 (3.5%), Quill grass 2 (4.1%) and White jute 2 (6.1%) had mixed helminths and protozoa infections (Table 3). Out of all the farmers interviewed, 11 (20%) used animal faeces and chemical as fertilizers, 31 (56.4%) used animal faeces only as their fertilizer source while 13 (23.6%) used chemical as their source of fertilizer. Most farmers 44 (80%) used stream and waste water as their source of irrigation while 11 (20%) of the farmers used well and stream (Tables 1-3).

DISCUSSION

Consumption of fresh vegetables is an essential route of transmission of intestinal parasites of public health importance which have been incriminated as one of the main causes of foodborne outbreaks (Daryani et al., 2008; Pires et al., 2012). From this present study, the overall prevalence of geohelminths and protozoan parasites observed is similar to the findings of other authors in Vietnam (Uga et al., 2009) and in Nigeria (Damen et al., 2007). However, it is higher than that reported in Iran (Daryani et al., 2008), in Pakistan (ul-Haq

et al., 2014) and in Yemen (Alsubaie et al., 2014). It is however lower than the observation of (Alade et al., 2013) in Nigeria and (Hajjami et al., 2013) in Morocco. The high level of contamination of African spinach observed in this present study might be due to their broad leaves and their development near ground level which provide large contact area with contaminated water and soil (ul-Haq et al., 2014). African spinach and other green leafy vegetables have 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 in market (Damen et al., 2007). *Strongyloides* spp, *Entamoeba coli*, *Ascaris suum* and *Trichuris* spp. were among the intestinal parasites of public health importance detected in vegetables from farms and markets, as this is similar to the observations of previous authors in Ethiopia (Gimaye & Fedaku, 2014), in Pakistan (Anwar & Mckenry, 2012) and in Nigeria (Uneke, 2004; Baker, 2007). *Strongyloides stercoralis* is a common cause of morbidity and mortality particularly in developing countries, as this parasite has the ability to auto-infect the host which makes it a significant public health problem (Roxby et al., 2009). The risk of acquiring strongyloidiasis

Table 1: Distribution of geohelminths and protozoa in vegetables sampled from different Local Government areas in Ibadan metropolis.

LGA	Number Examined	No. (%)	Parasite observed		
			Positive	Helminths	Protozoa
Ibadan North	68	22 (32.4)		<i>Strongyloides stercoralis</i> , <i>Filariform larvae</i> , <i>Ascaris</i> spp., <i>Fasciola</i> spp.	<i>Entamoeba coli</i> , <i>Eimeria</i> spp.
Akinyele	38	11 (28.5)		<i>Strongyloides stercoralis</i> , <i>Dicrocoelium</i> spp. <i>Ascaris</i> spp.	—
Ido	34	3 (8.82)		<i>Strongyloides stercoralis</i> .	—
Lagelu	13	13 (100)		<i>Strongyloides stercoralis</i> , <i>Filariform larvae</i> .	<i>Entamoeba coli</i> ,
Ibadan southwest	40	23 (57.5)		<i>Ascaris</i> spp., <i>Oesophagotum dentatum</i> , <i>Dicrocoelium</i> spp., <i>Trichuris</i> spp., <i>Strongyloides stercoralis</i> .	<i>Entamoeba coli</i> , <i>Entamoeba histolytica</i> , <i>Eimeria</i> spp.
Ibadan northeast	7	2 (28.6)		<i>Strongyloides stercoralis</i> .	—

Table 2: Prevalence of geohelminths and protozoa in vegetables from sampled markets in Ibadan metropolis.

Market	No.(%) examined	No. (%) positive
Bodija	37 (18.5)	13 (35.14)
Oja oba	40 (20)	23 (57.5)
Oojo	27 (13.5)	10 (37.04)
Oje	7 (3.5)	2 (28.6)
Eleyele	34 (17)	3 (8.82)

Table 3: Prevalence of geohelminths and protozoa in different vegetables species collected in Ibadan metropolis

Vegetables	Common name	No. (%) examined	No. (%) positive	Parasite observed	
				Helminths	Protozoa
<i>Amaranthus hybridus</i>	African spinach	57 (28.5)	20 (35.1)	<i>Strongyloides stercoralis</i> , <i>Trichuris</i> spp., <i>Oesophagotum dentatum</i> , <i>Dicrocoelium</i> spp., <i>Filariform larvae</i> .	<i>Entamoeba coli</i> , <i>Entamoeba histolytica</i> .
<i>Corchorus olitorius</i>	Jute	33 (16.5)	15 (45.5)	<i>Strongyloides stercoralis</i> , <i>Trichuris</i> spp., <i>Fasciola</i> spp., <i>Ascaris</i> spp.	<i>Entamoeba coli</i> , <i>Entamoeba histolytica</i> , <i>Eimeria</i> spp.
<i>Celosia argentia</i>	Quill grass	49 (24.5)	12 (24.5)	<i>Strongyloides stercoralis</i> , <i>Trichuris</i> spp., <i>Fasciola</i> spp., <i>Ascaris</i> spp.	<i>Entamoeba coli</i> , <i>Eimeria</i> spp.
<i>Telfairia occidentalis</i>	Fluted pumpkin	14 (7)	0 (0)	—	—
<i>Solanum macrocarpon</i>	Garden egleaf	7 (3.5)	4 (57.1)	<i>Strongyloides stercoralis</i>	<i>Entamoeba coli</i>
<i>Talinum triangulare</i>	waterleaf	27 (13.5)	14 (51.9)	<i>Strongyloides stercoralis</i> , <i>Dicrocoelium</i> spp <i>Ascaris</i> spp., <i>Oesophagotum</i> spp.	<i>Eimeria</i> spp.
<i>Solanum lycopersicum</i>	Tomato	10 (5)	3 (30)	<i>Strongyloides stercoralis</i> , <i>Ascaris</i> spp.	—
<i>Capsicum chinense</i>	Pepper	2 (1)	1 (50)	<i>Ascaris</i> spp.	—
<i>Basella alba</i>	English spinach	1 (0.5)	0 (0)	—	—

is higher in rural areas, among people who work with soil and among lower socio-economic groups (Vadlamudi et al., 2006; Viney & Lok, 2007). It is important to note that the presence of these parasites may be due to lack of modern toilet facilities, poor sewage disposal and inadequate public health enlightenment which make people defecate indiscriminately in the environment resulting in pollution of water and soil (Tsado et al., 2013). The species of *Ascaris* detected in this study may probably be *Ascaris suum*, this is due to the usage of pig faeces as a major fertilizer as stated by one of the farmers. *Ascaris suum* has been known to be a zoonotic parasite causing viscera larva migrant in humans (Baker, 2007; Dutto & Petrosillo, 2013). The threat of infection with intestinal parasites in the community is increased because these contaminated vegetables are sometimes eaten raw or parboiled to retain the natural taste and preserve heat labile nutrients (Slifko et al., 2009). These findings may have an essential implication for food safety and emphasize the importance of raw vegetables in threatening public health by transmission of zoonotic parasites to humans in Ibadan. A greater percentage of the farms investigated in this study applied untreated organic manure (animal faeces) for the cultivation of their vegetables. Choice of fertilizer was dependent mainly on cost and availability, as the expensive nature of inorganic fertilizers makes farmers opt for untreated organic fertilizer. However, the use of untreated organic fertilizer pre-dispose vegetables to faecal helminthes contamination as humans can consequently be infected when these vegetables are consumed undercooked or raw (Daryani et al., 2008). Although vegetable farmers using wastewater as their source of irrigation in this study are aware of the fact that the use of wastewater can pose a major hazard to their health, however, inability to have access to potable water for irrigation, which is also expensive has restrained them to using wastewater. (Obuobie et al., 2006) noted that farmers practicing olericulture lack better options hence end up using wastewater sources for irrigation which in most cases are not paid for and are readily available. There is therefore a need for education of farmers and market women on proper handling and hygienic management of vegetables to safe guard the health of the populace.

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