Parasitic Contamination Of Vegetables In Three Selected Local Government Areas Of Adamawa State

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DOI: 10.29322/IJSRP.11.08.2021.p11632
http://dx.doi.org/10.29322/IJSRP.11.08.2021.p11632

Abstract- Fresh vegetables are important element of human food for healthy living. Yet the use of fresh vegetables plays an important epidemiological role in the transmission of food-borne parasitic diseases. This study was conducted to identify parasitic organisms whose cysts and ova are present in vegetables, determined the prevalence of pests on vegetables and to assess the distribution and the contamination of vegetables with cyst and parasitic eggs. Three local government areas were randomly selected by balloting in Adamawa State, North-eastern, Nigeria. A total of 525 fresh vegetable samples comprising of 35 grams each of Spinach (*Spinacia oleracea*), Sorrel (*Rumex acetate*), (*Allium cepa*), Radish (*Moringa oleifera*), and Sesame (*Sesamum orientale*), were purchased from vegetable farms of Loko, Dasin-hausa and Gerio in Song, Fufore and Yola-North Local Government Areas of Adamawa State. Sedimentation and flotation techniques were used by centrifugation at 2000 rpm for 3 minutes, twice, to extract parasitic stages using normal saline solutions and zinc sulphate. After microscopic examination under X40 and X100 results indicated 14 parasites, including 6 protozoa and 8 helminths were identified of which *Ascaris sp* 169 (32.2%) had the highest occurrence, followed by *Entamoeba sp* 146 (27.8%) while *Strongyloides sp* 118 (22.5%) was the third most occurring species whereas *Enterobius sp* 15(2.9%) and *Toxoplasma sp* 11(2.1%) were the least occurring parasites. Spinach 87 (49.7%) had the highest level of contamination, followed by sorrel 76 (43.4%), sesame 46 (26.3%) and onion 42 (24.0%) and radish 24 (13.7%). The differences between levels of contamination of individual vegetables in the study areas were statistically significant ($P < 0.05$). The study showed that vegetables under study were highly contaminated by parasitic cysts and ova.

I. INTRODUCTION

Vegetables are a vital part of human diet because of their nutritional value. They are rich sources of the small proportion of Proteins, Fat and high proportion of vitamins such as Vitamin A, B_6, C and K. Several studies have reported that vegetables also contain dietary minerals like Calcium, Magnesium, Potassium, Iron and a variety of phytochemicals that have antioxidants, anti-bacterial, anti-viral and carcinogenic properties (Ada *et al*., 2018). Food and Agricultural Organization (FAO) and WHO (2003), recommended the intake of 400g of vegetables and fruits daily for the prevention of chronic diseases such as heart diseases, cancer, diabetes and obesity as well as for the prevention and alleviation of several micronutrient deficiencies, especially in less developed countries. Several studies within and outside Nigeria have reported the presence of many protozoan cysts, helminth eggs, larvae and parasites like *A. lumbricoides, T. trichiura, S. stecoralis, Fasciola spp,* Hookworm, *G. lambia, E. histolytica, I. belli* and *E. vermicularis* in vegetables. Their presence is still a public health concern due to gastrointestinal infection (Simon-Oke *et al*., 2014; Tefera *et al*., 2014). Because vegetables are edible plants, whose leaves, roots, stems or succulent fruits are eaten raw or partially cooked they have a tendency to host protozoan cysts, cysts, helminth eggs and larvae. Hence, despite the significant role that vegetables play in human life, their consumption constitutes a major epidemiological means of transmission of parasitic food borne diseases. For example, the problem of gastrointestinal infection is becoming of increasing concern because of the expanding number of susceptible people, the elderly and the immune-compromised in the developing countries (Alhabal, 2015). The role of poor hygiene and sanitation in the epidemiology of parasitic disease transmission has been identified with the housefly (*Musca domestica*) being responsible for the transmission of intestinal parasites in Ibadan and Espuma in Nigeria (Gupta *et al*., 2009; Adeyeba and Okpala, 2000; Nmorsi *et al*., 2006). In addition, contamination could be through insects contaminated with infected feces (Istifanus and Panda, 2018) while in transportation. However, while the presence of intestinal parasites in vegetable samples is suggestive of fecal contamination (Chiodini, 2001), the conveyance of fruits and vegetables in the early hours of the morning from farms to the markets with either by motor-cycles, tri-cycles or in dirty rickety pick-up vans which sometimes are loaded with passengers is another cause of contamination indicating sociocultural practices in certain geographical areas as the main causes of different levels of contamination (Istifanus and Panda, 2018). Also, some reports from different parts of the world identified the use of untreated wastewater as a major cause of parasitic contamination on vegetables (Al- Binali *et al*., 2006). Consequently, owing to the diversity and universality of the problem of parasitic contamination more attention and further investigation highlighting mainly the role of foods especially vegetables in the transmission of parasitic diseases is required. Therefore, this study was conducted to determine the presence and prevalence of parasites and the extent of contamination of vegetables by parasites in the study area.

II. MATERIALS AND METHODS

Study Area

Three vegetable farms were selected randomly by balloting; one in each of the three Local Government Areas in Adamawa State, Nigeria. These are; Dasin- Hausa in Fufore LGA which lies on the coordinate $9^0$13'N $12^0$39'E with the total population of 209,460 (Wale, 2014), Gerio in Yola North LGA which lies on the coordinate $9^0$13'48" N $12^0$2736" E with the total population of 392,854 (FRN, 2016) and Loko in Song LGA which lies on the coordinate $9^0$49"28" N, $12^0$37'30" E with the population of 195,188 (Aliyu and Orisakwe, 2006).

Study Population

A total of 525 samples comprising of 35 samples each of Spinach"Aleho" (*Spinacia oleracea*), Onion "Albasa" (*Allium cepa*), Sesame leaves " Karkaashii" (*Sesamum orientale*), Sorrel "Yakuwa" (*Rumex acetosa*), Horseradish tree "Zoogale" (*Moringa oleifera*) were purchased from different farms in the selected farms across the three Local Government Areas in the early hours of the morning between 7:00Am and 9:00Am. Collected samples were stored in sterile polythene bags and labelled accordingly before transportation for laboratory analysis at the Zoology laboratory

Data Collection and Analysis

Vegetable samples were selected randomly from the vegetable farms in each of the selected Local Government Area. The vegetable samples were purchased from the farms in the selected local government areas, placed in a sterile polythene bag and label accordingly between the hours of 7:00 a.m. to 9:00 a.m. and transported to the laboratory for analysis. Laboratory analysis was conducted at the Zoology laboratory of the Department of Zoology, Modibbo Adama University of Technology, Yola - Nigeria. The samples were washed in 250 ml of 0.9 % Sodium chloride solution. Both the sedimentation (were centrifuged at 2000 rpm in Normal saline) and simple floatation (using Zinc sulphate solution of 1.18-1.20 specific gravity) techniques was performed on each sample to isolate and identify parasites as described by Elaigwu, 2015

Identification of Parasites Stages

Identification of the cysts, and eggs of encountered parasites were done on the basis of morphological characteristics with reference to keys. (Cheesbrough, 2006).

Statistical Data Analysis

Data were analyzed using the Statistical Package for Social Sciences (SPSS) software version 22. The data were analyzed by Chi- square test using SPSS to compare the levels of contamination of different vegetables across the study areas ($P < 0.05$).

III. RESULTS

Distribution of Parasite

A total of 14 parasites comprising of 6 protozoans namely; *Entaemoeba histolytica, Cryptosporidium parvum, Cyclospora cayetanensis, Isospora belli, Giardia lamblia* and *Toxoplasma gondii* and 8 heminths including *Ascaris lumbricoides, Hymenolepsis nana, Strongyloides stercoralis, Toxocara canis, Taenia saginata, Enterobius vermicularis, Trichiuri trichiura* and hookworm ere encountered (Table 4. 1)

Table 4. 2 showed, Gerio had the highest number of parasites identified with 391(45.7%), followed by Loko 275(32.2%), and Dasin-hausa 189(22.1%). Furthermore, out of the five vegetable samples collected for the study, Spinach 272(51.8%), had the highest number of infestation with parasites, followed by Sorrel 217(41.3%), while Radish 60(11.4%) had the least occurrence. There was no significant relationship between the levels of contamination ($P > 0.05$).

Samples from the study areas showed parasites cysts and eggs on vegetables. Out of the 175 vegetables samples, Spinach 87(49.7%), had most frequent occurrence, followed by Sorrel 76(43.4%) while 24(13.7%) infestation was found in Radish (Table 4. 3).

Among the parasites observed on the vegetables, *Ascaris lumbricoides* 55(31.4%) had the highest distribution, *Entamoeba histolytica* of 43(24.7%), over *Strongyloides stercoralis* with 40(22.9%) and *G. lamblia* was the least of the samples from different areas. There was no significant relationship between the levels of contamination of samples from different areas ($P > 0.05$). Out of a total 175 vegetable samples collected and examined in Dasin-hausa, Spinach 59(33.7%) had the most frequent number of the parasites cysts and eggs, followed by Sorrel 41(24.4%) and Radish had the least occurrence of 15(8.6%) (Table 4.4). Meanwhile, *Ascaris lumbricoides* 48(27.4%) had the highest distribution, followed by *Entamoeba histolytica* 33(18.6%) and *Strongyloides stercoralis* 32(18.3%) while *T. trichiura* was the least. There was significant relationship between the levels of contamination ($P< 0.05$).

Table 4.1 Parasitic Contamination of Vegetable in 3 Selected Farms LGAs of Adamawa State, Nigeria (N=525).

Parasitic Contaminants	Farms			Total	Chi-Square	Df	P-Value
	Gerio	Dasin Hausa	Loko				
Protozoa							
G. lamblia	3(1.7)	11(6.3)	2(1.1)	16(3.0)	8.42	2	0.368ns
C. parvum	22(12.6)	14(8.0)	17(9.7)	53(10.1)	13.1	2	0.368ns
E. histolytica	70(40)	33(18.9)	43(24.6)	146(27.8)	18.1	2	0.368ns

C. cayetanensis	40(22.9)	12(6.9)	35(20)	87(16.6)	10.4	2	0.217ns
I. belli	16(9.1)	4(2.3)	14(8)	34(6.5)	45.67	2	0.004**
T. trichiura	15(8.6)	1(0.6)	3(1.7)	19(3.6)	12.31	2	0.368ns
Helminths							
T. canis	37(21.1)	0(0)	29(16.6)	66(12.6)	24.12	2	0.112ns
A. lumbricoides	66(37.7)	48(27.4)	55(31.4)	169(32.2)	22.3	2	0.368ns
Toxoplasma sp	11(6.3)	0(0)	0(0)	11(2.1)	6.87	2	0.368ns
S. stercoralis	46(26.3)	32(18.3)	40(22.9)	118(22.5)	23.81	2	0.122ns
Hymennolepsis sp	18(10.3)	11(6.3)	6(3.4)	35(6.7)	23.21	2	0.101ns
T. saginata	39(22.3)	6(3.4)	24(13.7)	69(13.1)	25.59	2	0.000**
E. vermicularis	8(4.6)	0(0)	7(4)	15(2.9)	12.54	2	0.234ns
Hookworm sp	0(0)	17(9.7)	0(0)	17(3.2)	17.45	2	0.123ns
Total	391(45.7)	189 (22.1)	275(32.2)	855(100)	124.86	2	0.00012**

Table 4. 2 Prevalence of Vegetable Contamination among 3 Selected Farms in Adamawa State, (N=525)

Farms	Spinach	Sorrel	Vegetable Onion	type Radish	Sesame leaves	Total	Chi-Square	Df	P-value
Gerio	126(32.2)	100(25.6)	69(17.6)	21(5.4)	75(19.2)	391(45.7)	42.00	4	0.046ns
Dasin-Hausa	59(31.2)	41(21.7)	32(16.9)	15(7.9)	42(22.2)	189(22.1)	14.00	4	0.442ns
Loko	87(31.6)	76(27.6)	42(15.3)	24(8.7)	46(16.7)	275(32.2)	40.00	4	0.036**
Total	272(51.8)	217(41.3)	143(27.2)	60(11.4)	163(31)	855(100)	123.00	4	0.0021**

Table 4. 3 Contamination of Different Vegetables by Parasitic Cysts and Ova in Loko Farm, Song LGA (N=175).

Identified Parasite	Spinach	Sorrel	Vegetable Onion	types Radish	Sesame leaves	Total	Chi-Square	Df	P-Value
Protozoa									
G. lamblia	1(2.9)	1(2.9)	0(0)	0(0)	0(0)	2(1.1)	3.65	4	0.406ns
C. parvum	6(17.1)	7(20)	3(8.6)	1(2.9)	0(0)	17(9.7)	4.67	4	0.406ns
E. histolytica	13(37.1)	10(28.6)	6(17.1)	3(8.6)	11(31.4)	43(24.7)	20.67	4	0.023**
C. cayetanensis	11(31.4)	10(28.6)	3(8.6)	7(20)	4(11.4)	35(20)	21.99	4	0.033**
Toxoplasma spp	-	-	-	-	-	-	-	-	-
I. belli	5(14.3)	2(5.7)	4(11.4)	1(2.9)	2(5.7)	14(8)	3.67	4	0.406ns
Helminths									
S. stercoralis	10(28.6)	10(28.6)	9(25.7)	7(20)	4(11.4)	40(22.9)	19.45	4	0.223ns
T. trichiura	1(2.9)	0(0)	0(0)	0(0)	2(5.7)	3(1.7)	2.70	4	0.406ns
T. canis	8(22.9)	12(34.3)	3(8.6)	1(2.9)	5(14.3)	29(16.6)	12.8	4	0.076ns
Hymennolepsis spp	3(8.6)	1(2.9)	0(0)	0(0)	2(5.7)	6(3.4)	1.23	4	0.406ns
A. lumbricoides	17(48.6)	12(34.3)	9(25.7)	4(11.4)	13(37.1)	55(31.4)	23.87	4	0.002**
T. saginata	9(25.7)	7(20)	5(14.3)	0(0)	3(8.6)	24(13.7)	11.45	4	0.243ns
E. vermicularis	3(8.6)	4(11.4)	0(0)	0(0)	0(0)	7(4)	3.78	4	0.406ns
Total	87(49.7)	76(43.4)	42(24)	24(13.7)	46(26.3)	275(100)	234.87	4	0.00031**

Identified Parasite	Spinach	Sorrel	Vegetable Onion	type Radish	Sesame leaves	Total	Chi-Square	Df	P-Value
Protozoa									
I. belli	7(20)	0(0)	6(17.1)	0(0)	3(8.6)	16(9.1)	4.00	4	0.406ns
C. parvum	8(22.9)	9(25.7)	4(11.4)	0(0)	1(2.9)	22(12.6)	12.65	4	0.021**
E. histolytica	19(54.3)	16(45.7)	14(40)	4(11.4)	17(48.6)	70(40)	32.67	4	0.002**
Cyclospora spp	13(37.1)	11(45.7)	8(22.9)	3(8.6)	5(14.3)	40(22.9)	12.95	4	0.342ns
G. lamblia	2(5.7)	1(2.9)	0(0)	0(0)	0(0)	3(1.7)	4.00	4	0.406ns
Helminths									
T. gondii	1(2.9)	3(8.6)	5(14.3)	0(0)	2(5.7)	11(6.3)	4.00	4	0.406ns
A. lumbricoides	22(62.9)	14(40)	13(37.1)	2(5.7)	15(42.9)	66(37.7)	43.76	4	0.032**
Hymennolepsis spp	5(14.3)	4(11.4)	0(0)	0(0)	9(25.7)	18(10.3)	4.00	4	0.406ns
T. canis	11(31.4)	14(40)	0(0)	5(14.3)	7(20)	37(21.1)	13.77	4	0.221ns
S. stercoralis	14(40)	12(34.3)	11(31.4)	2(5.7)	7(20)	46(26.3)	13.67	4	0.221ns
T. trichiura	8(22.9)	4(11.4)	0(0)	0(0)	3(8.6)	15(8.6)	4.00	4	0.406ns
T. saginata	11(31.4)	9(25.7)	8(22.9)	5(14.3)	6(17.1)	39(22.5)	14.23	4	0.021**
E. vermicularis	5(14.3)	3(8.6)	0(0)	0(0)	0(0)	8(4.6)	4.00	4	0.406ns
Total	126(32.2)	100(25.6)	69(17.6)	21(5.4)	75(19.2)	391(45.7)	123.22	4	0.00010**

Table 4. 4 Cysts and Eggs of Parasitic Organisms on consumed vegetables in Dasin-Hausa Farm, Fufore LGA (N=175).

Identified Parasite	Spinach	Sorrel	Vegetables Onion	types Radish	Sesame leaves	Total	Chi-Square	Df	P-Value
Protozoa									
I. belli	3(8.6)	0(0)	1(2.9)	0(0)	0(0)	4(2.3)	4.00	4	0.406ns
C. parvum	6(17.1)	4(11.4)	0(0)	0(0)	4(11.4)	14(8)	12.12	4	0.012**
E. histolytica	11(31.4)	7(20)	1(2.9)	8(22.9)	6(17.1)	33(18.6)	14.66	4	0.021**
C. cayetanensis	0(0)	2(5.7)	9(25.7)	1(2.9)	0(0)	12(6.9)	13.55	4	0.003**
G. lamblia	1(2.9)	1(2.9)	6(17.1)	0(0)	3(8.6)	11(6.3)	4.00	4	0.406**
Helminths									
Hymennolepsis spp	0(0)	3(8.6)	0(0)	1(2.9)	7(20)	11(6.3)	4.00	4	0.406ns
A. lumbricoides	15(42.9)	13(37.1)	10(28.6)	1(2.9)	9(25.7)	48(27.4)	23.56	4	0.002**
S. stercoralis	11(31.4)	7(20)	3(8.6)	3(8.5)	8(22.9)	32(18.3)	21.36	4	0.011**
T. trichiura	0(0)	0(0)	0(0)	0(0)	1(2.9)	1(0.6)	4.00	4	0.406**
T. saginata	5(14.3)	0(0)	0(0)	0(0)	1(2.9)	6(3.4)	4.00	4	0.406**
Hookworm *spp*	7(20)	4(11.4)	2(5.7)	1(2.9)	3(8.6)	17(9.7)	14.23	4	0.052ns
Total	59(33.7)	41(23.4)	32(18.3)	15(8.6)	42(24)	189(36)	76.98	4	0.00011**

Table 4. 5 showed the distribution of parasite cysts and eggs on different vegetables in Gerio farm. Spinach were more contaminated with parasitic cyst and eggs with 126(32.2%), Sorrel 100(25.6%), while Radish 21(5.4%) was the least infested vegetable. According to this study, *Entamoeba histolytica* were more frequent with 70(40%), *Ascaris lumbricoides* 66(37.7%), *Strongyloides stercoralis* 46(26.3%). *Giardia lamblia* was the least most frequent parasite as shown above. There was significant relationship between the levels of contamination (P<0.05).

Table 4.5 Contamination of Different Vegetables with Cysts and Eggs of Parasites in Gerio Farm, Yola North LGA (N=175).

IV. DISCUSSION

The levels of prevalence of parasitic agents on vegetables in this present study were lower than in similar reports in Iran 32% (Daryani *et al*., 2008) and India 44% (Gupta *et al*., 2009). Sorrel 217 (25.4%) had the second level of infestation of parasites cysts and eggs. This could be due to the broad and large surface area of the leaves which are in close proximity with the sewage contaminated soil surface (Larkin *et al*., 1978). Sesame 163 (19.0%) had a contamination level next to sorrel in line with the

findings of Maikai *et al* (2012). Although the leaves of Sesame are not broad and have a small surface area, the large number of the leaves on the stalk could be the reason for this level of occurrence of cysts and eggs of parasites on the vegetable. Onion 143 (16.7%) was among the least contaminated vegetables with cysts and eggs of parasites. This agrees with the study of Uga *et al* (2009), which showed that contamination is usually higher in the leafy than the root and fruity vegetables. Radish 60 (7.0%) had the least contamination with cysts and eggs of the various parasitic organisms due perhaps to the shape and surface of the vegetables (El-Said, 2012). The overall prevalence of cysts and eggs of the different parasites identified on vegetables in the study areas differed. This largest occurrence was at Gerio, 391 (45.7%), followed by Loko 275 (32.2%) and Dasin-hausa 189 (22.1%). This difference in contamination levels among study areas could be attributed to varying environmental conditions and other hygiene factors ranging from indiscriminate open defecation habits on farm soils by children and adults, use of animal dungs as organic manure, poor personal and domestic hygiene, the kind of water used for irrigation as well as methods of transportation which constitutes favorable conditions for transmission unto vegetables (Pires *et al*., 2012: Auta *et al*., 2017). Among the parasites screened on various vegetables in the study areas, A. *lumbricoides* was more predominant. This finding is similar to those of (Uneke, 2007; Istifanus and Panda, 2018; Gupta *et al*., 2009; and Shafa-ul-Haq *et al*., 2014) in Nigeria, India and Pakistan respectively. This seems to be due to the epidemiological factors that cause the spread of pests, especially in poor health conditions and practices, poor housing and high population density, and illiteracy in the developing world. The relatively high occurrence of *Ascaris spp* and *Strongyloides spp* reported in this study agrees with Fagbero *et al*. (2016). However, detection of some parasites such as *Ascaris lumbricoides*, *Ancylostoma*, *Strongyloides* and *Giardia* in our samples suggests poor hygienic standard in the study areas. The parasites screened have mainly fecal origin implying fecal contamination of the samples. This kind of contamination is increased by poor sanitary conditions and hygienic habits (Damen *et al*., 2007; Daryani *et al*., 2008; Slifko *et al*., 2000). Thus, intestinal parasitic infections are expected in the area where there is low standard of personal hygiene as well as poor sanitary conditions. In this study, of the parasites detected, *Ascaris sp* was the most frequently detected, followed by *Entamoeba sp*, *Strongyloides sp*, *Cyclospora sp*, *Taenia sp* and *Toxocara sp*. This report is in contrast to the study of (Al- Kassr, 2010) which reported that *Entamoeba sp* had the most detected parasites followed by *Entamoeba sp*, but it was similarly reported in the case *Strongyloides sp* and *Taenia sp* which were the fourth and fifth according to the degrees of contamination of vegetables. The result of Table 4. 5 reported a similar result with (Al- Kassr, 2010) who studied the prevalence of different parasitic stages on commercial vegetables in Al-Nassriah City, Iran and found the highest contamination of *Entamoeba sp*, *Ascaris sp* was reported as the second most prevalent parasitic stage. This report is also similar to the previous study in Jimma town, Ethiopia by (Girmaye and Bent, 2014) who reported *Ascaris sp* as the second most prevalent (13%) of the vegetables studied in Casablanca, Morocco, but in a higher percentage in this present report 66(37.7%). *Entamoeba sp* was reported as the third most prevalent (16%) which is lower than the level of contamination discovered in this study 70(40%). Similar results were obtained in Southwestern Saudi Arabia (Al- Binali *et al*., 2006). Although *Entamoeba sp* a commercial intestinal parasite, its presence 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 areas. This results are in agreement with (Ishaku and Ishakeku, 2013) who reported (1.33%), (0.33%) and (0.33%) for the three species respectively, with the least prevalence of Taenia and *Trichiuri species*, while, *Taenia sp* was the fifth most prevalence parasite 69(13.15%). *Hookworm sp* 17(3.2%) occurred in only one study area. This disagrees with the reports of Ayaji and Agahlu (2016) and Ojemudia (2011), who reported *Hookworm sp* as the most prevalent helminths and the report of whose report revealed *Hookworm sp* as the most prevalent parasite.

V. CONCLUSION

This study found that vegetables consumed in study areas, were contaminated with cysts and eggs from parasitic organisms. The occurrence and prevalence of the infection varies among the vegetables under study and among the areas where the vegetables were grown. Overall infection with cysts and eggs from parasitic organisms were widely distributed in the vegetables and the areas where they were grown. With market gardeners, sellers and consumers completely unaware that vegetables can carry and transmit parasites, the risk of infection amongst the human population is high. This has consequences for the overall wellbeing of the people living in these areas and are consuming vegetables produced in these areas.

VI. RECOMMENDATIONS

i. There is a need for further research to isolate other microbes that harbours the edible vegetable.
ii. There is need to make further research on edible vegetables during wet and rainy season.
iii. The vegetable farmers, seller and consumers to wash the vegetables properly with iodized salt.

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