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Phylloplane Parasites on Common Edible Plants of Eastern Nigeria: Easy Link to Intestinal Parasites Transmission

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Authors' contributions

All the authors contributed in carrying out this work. Author OCA designed the study and served as the principal supervisor who also coordinated the work. Author CVN identified the plant species. Author CJO wrote the protocol and served as the principal investigator. Authors EIN and PEM searched literatures and participated in the write up. All the authors read and approved the final manuscript.

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ABSTRACT

Aims: In recent years, particularly in developing countries, there has been an unprecedented number of reported cases of parasitic illnesses linked to the high demand for fast foods, majority of which may be contaminated, improperly cooked or washed. This study was carried out to determine the parasitic load on some selected plant leaves, identify the possible parasites which could be found on these leaves, and determine which particular plant leaves habour more parasites.

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Study Design: The study was laboratory-based observational investigation.
Place and Duration of the Study: The research was done in the laboratory of Biological Sciences Department of Ebonyi State University, Abakaliki, Ebonyi State, Nigeria between June 2014 and January 2015.
Methodology: The leaves of *Thaumatococcus daniellii, Alchornea cordifolia* and *Musa sapientum* were screened for parasites using standard microscopy methods.
Results: Out of the 30 samples of the plant leaves screened, 22(73.3%) were positive for intestinal parasites. The parasites encountered include some species of protozoa (*Entamoeba coli*), cestode (*Hymenolepis nana*), nematodes (*Ascaris lumbricoides*, *Trichuris trichiura*, hookworm and *Strongyloides stercularis*). *T. daniellii* leaves had the highest parasitic contamination of five species of parasites while *M. sapientum* and *A. cordifolia* showed the less parasitic contamination of three and two parasites respectively.
Conclusion: From the findings of this study, it is obvious that plant leaves consumed by people are quite often contaminated with intestinal parasites. This is an indication that humans are at risk of infection especially as some plant leaves are naturally present in diet of people of all classes. Hence, the findings underscore the public health implication of using improperly washed or undercooked leaves of these plants with vegetable famers, sellers and consumers, being at high risk of infection with strongyloidiasis, ascariasis, amoebiasis and others. The need to soak plant leaves for 10 minutes in saturated salt solution or to steam adequately before using them to prepare or wrap food is recommended. There is also need for government and non-governmental organizations to provide portable water and toilet facilities in order to discourage indiscriminate defecation in the environment.

Keywords: Phylloplane; parasites; contamination; Thaumatococcus daniellii; Alchornea cordifolia; Musa sapientum.

1. INTRODUCTION

In recent years, there has been an increase in the number of reported cases of food borne illness linked to consumption of fresh plant leaves [1,2]. Studies from different parts of the world implicate vegetables as agent for transmission of parasite ova, larvae, oocysts and cysts [3,4]. The consumption of raw plant leaves or their use in wrapping food items plays a major epidemiological role in the transmission of parasitic food-borne diseases [5]. Contamination may occur on the field during growth, harvesting, transporting, processing, distribution or marketing or in the home by food handlers. Once harvested, some of these leaves are not washed before use or are washed with contaminated water which serves as a source of contamination [6]. Intestinal parasites such as *Cryptosporidium spp., Giardia lamblia, Entamoeba histolytica, Ascaris lumbricoides, hookworms, Enterobius vermicularis, Trichuris trichiura, Toxocara spp., Hymenolepis spp., Taenia spp., Fasciola spp.,* and members of the family *Trichostrongylidae* could infect humans as a result of consumption of contaminated, uncooked or improperly washed plant leaves or vegetables. Lack of efficacious sanitizers that can remove or kill pathogens on raw plant leaves has contributed to their ability to gain access to locations within structures and tissues that may habour these pathogens [7]. Epidemiological studies have also indicated that parasitic diseases are endemic in areas where waste water is used to irrigate plants and vegetables [8]. In areas where helminth infections are prevalent in the population and where waste water is used for irrigation of farms, consumption of raw vegetables can serve as agent of transmission [9]. In rural communities of Nigeria, defecation on open fields is still practiced due to inadequacy of human waste disposal systems and the belief of uninformed residents that such practice help fertilize their farms [10]. In developing countries, due to inadequate and lack of systems for routine diagnosis, monitoring and reporting for many of the food borne pathogens, most outbreaks caused by contaminated plant leaves go undetected and the incidence of this occurrence in food is under-estimated. In recent years, in developing countries, there has been an increase in the number of reported cases of parasitic illnesses linked to a high demand for food wrapped in some plants' leaves. It therefore becomes imperative to investigate the phylloplane parasites assemblage of some of these leaves used in wrapping food in order to contribute to information required for the control of parasitic infections, especially those that make up part of the neglected tropical diseases.

2. MATERIALS AND METHODS

2.1 Study Area

The study was carried out in Ebonyi State, South-east Nigeria, with vegetation of the mosaic of the savanna and semi-tropical forest that is dense and remains evergreen throughout the rainy season. Majority of the inhabitants of this area are peasant farmers and petty traders of low economic status. It is a common practice that majority of the farmers use human and animal faeces as manure to augment and reduce the cost effective commercially processed fertilizer.

2.2 Sample Collection

Leaves of different species of plants were collected in three major zones of Ebonyi State, South-east of Nigeria. The plants screened were *Thaumatococcus daniellii, Alchonea cordifolia and Musa sapientum*. These plant species were selected for the study due to the extensive use of their leaves for wrapping food stuffs. They were randomly collected in batches of 10 per every location. A total of 30 leaves were investigated. The leaves were identified by the plant taxonomist in the Department of Applied Biology, Faculty of Biological Sciences, Ebonyi State University, Abakaliki, Nigeria and the voucher specimen are deposited in the Ebonyi State University Herbarium.

2.3 Analysis of the Sample

Thirty leaves of the plant species were examined for intestinal parasite profile as previously described [11]. Samples were washed with distilled water and the suspension was strained through a sterile sieve to remove undesirable materials. The filtrates were centrifuged at 3000 rpm for five minutes and the supernatant discarded while the deposits were placed on clean glass slides and covered with cover slips. This was followed by examination under a light microscope using 10x and 40x objectives fixed with digital camera to identify the cysts and eggs of these organisms as previously described [12]. Different parasite eggs, larvae or cysts present in the samples were counted and results documented as previously described [3].

2.4 Statistical Analysis

Data collected were analyzed in simple percentage prevalence.

3. RESULTS

Out of the 30 samples of the three plant leaves analyzed, 22(73.3%) were positive for intestinal parasites. The incidental parasites are some species of protozoa (*Entamoeba coli*), cestode (*Hymenolepis nana*) and nematodes (*Ascaris lumbricoides*, *Trichuris trichiura*, hookworm and *Strongyloides stercularis*).

Among the different locations surveyed, nine (90%) contamination rate occurred in plants from Izzi, 7 (70%) in those from Ezza North while the least 6 (60%) occurred in plants collected from Ebonyi (Table 1). *Thaumatococcus daniellii* had highest parasitic contamination with five species of parasites followed by *Alchonea cordifolia* which had three species, while *Musa sapientum* showed the least parasitic contamination with two parasite species (Table 2). The rates of contamination of the three plants were 90%, 60% and 70% respectively (Table 2).

4. DISCUSSION

The presence of intestinal parasites in plant leaves sampled is suggestive of faecal contamination of those leaves within their habitats. Routine diagnosis has implicated plant leaves as source of transmission of parasites. This result is in conformity with other previously reported works [13-17,10] which noted that factors such as hygiene of consumers and farmers, plant leaf anatomy cum nature of infective forms of the parasites have contributed to the prevalence of parasitic infection. Other factors such as the behavioural attitude of farmers in application of untreated human and animal dung as manure, and the use of irrigation source which receives raw affluent from human or animal wastes have also been implicated. These are also in tandem with previous reports work [18].

High levels of environmental sanitation, proper sewage disposal and personal hygiene tend to be more observed in Ebonyi Local Government area due to its proximity to the capital city of Ebonyi State compared to Ezza North and Ezzi that are far away. The prevalence of parasites according to different plant leaves showed that *T. daniellii* had the highest number of contamination of parasite of different species. This is due to the fact that *T. daniellii* are majorly found in humid areas were most of the contaminated faecal materials are dumped through erosion current [19]. It might also be due to its popular use as

Table 1. The intensity of parasitic contamination of plants at different locations

Area of collection (LGA)	Number of vegetables screened	Number contaminated	Percentage contamination
Ebonyi	10	6	60.0%
Ezza North	10	7	70.0%
Izzi	10	9	90.0%
Total	30	22	73.3%

Table 2. The contamination and infection rate of different plants' leaves

	Parasites	TH	MS	AL
1	Hookworm	-	+	+
2	*Ascaris lumbricoides*	+	-	+
3	*Strongyloides stercuralis*	+	+	+
4	*Trichuris trichiura*	+	-	-
5	*Hymenolepis nana*	+	-	-
6	*Entamoeba coli*	+	-	-
Types of plant leaves		**NE**	**NP**	**PS (%)**
Thaumatococcus daniellii		10	9	90
Musa sapientum		10	6	60
Alchornea cordifolia		10	7	70
Total		30	22	73.3

TH=Thaumatococcus daniellii; MS= Musa Sapientum; AL= Alchornea cordifolia; NE = Number Examined; NP = Number Positive; PS = Positive Specificity; + = Presence of a parasite; - = Absence of a parasite

sweetener since its aril contains a non-toxic, intensely sweet protein, thaumatin, which is at least 3000 times as sweet as sucrose [20]. The low prevalence of parasites on banana leaves might be due to the reason that plantation site are not majorly found in water logged areas.

5. CONCLUSION

Plant leaves consumed by people or used for wrapping foods are quite often contaminated with intestinal parasites. *Thaumatococcus daniellii* which is usually used in wrapping uncooked foods had the highest rate of contamination. This is an indication that humans are always at risk of infection especially as some plant leaves are naturally present in diet of people of all classes. These findings underscore the public health implication of vegetable famers, sellers and consumers, being at high risk of infection with strongyloidiasis, ascariasis, amoebiasis and others.

6. RECOMMENDATIONS

Plant leaves cannot be removed from human diet but can be excluded from the cycle of transmission and dispersion of parasites. This can be achieved by maintenance of simple personal and environmental hygiene by sellers and consumers. There is need to avoid the use of untreated human and animal wastes as fertilizer. There is also need to soak plant leaves for 10 minutes in vinegar or saturated salt solution for plasmolysis of parasites. Steaming leaves adequately before using them to prepare food (such as African salads) or wrapping of food should be practised. There is need to avoid indiscriminate defecation in the environment through provision of adequate sewage disposal facilities. The campaign to eradicate parasitic infections must also be intensified.

CONSENT

It is not applicable.

ETHICAL APPROVAL

It is not applicable.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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