

# Parasitological Assessment of Lettuce Quality in the Maxixe City Markets, Mozambique

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## Abstract

**Introduction:** The consumption of lettuce provides several benefits, such as high source of dietary fiber, minerals and vitamins. However, despite these benefits, it can be a vehicle for the transmission of parasitic diseases, taking into account that its consumption is done in a raw way. Thus, this study aimed to assess the parasitological quality of lettuce that is sold in the markets of Maxixe city in Inhambane Province, the southern of Mozambique.

**Materials and Methods:** In June and July 2018, 50 samples of lettuce were purchased in three markets from Maxixe city. The samples were placed in plastic bags, correctly identified and transported to the Laboratory of Microbiology and Parasitology at the Faculty of Health Sciences of Pedagogical University of Mozambique, where the parasites' eggs and larvae were investigated, using the method of Hoffmann and Pons- Janer (1934). The statistical analysis of data was performed by using SPSS program, version 24.

**Results and Discussion:** From 50 samples of lettuce analyzed, 35 of them were positive that corresponds to 70% of the contaminated samples, with the remaining 15 (30%) being negative, with significant differences ( $X^2, P=0.002$ ). The highest contamination rate was found in the Tshuhla market, with 100% of the contaminated samples, followed by Dumbanengue market with 64.7% and finally the Xicadjuanine market with 43.75% of contaminated samples. The parasites found in the lettuce samples were *Ascaris lumbricoides*, *Entamoeba histolytica* cysts, *Strongyloid stereoralis* larvae and *Hepatic Fasciola*.

**Conclusion:** Expressive contamination rates of lettuce sold in the three markets were obtained, with the need to adopt proper hygiene practices before its consumption, in addition to the need to strengthen the health surveillance system.

**Keywords:** Lettuce, Parasitological Assessment, Intestinal Parasites, Markets

## Introduction

Vegetables are important constituents of a healthy diet and

are a source of vitamins, minerals and biologically active compounds [1]. It is estimated that the daily intake of 400g of plant-based foods is sufficient to prevent cardiovascular disease, cancer, diabetes and obesity. Worldwide, it is estimated that up to 2.7 million lives can potentially be saved every year if the consumption of fruits and vegetables is increased [2].

The consumption of vegetables has increased not only because of the growing population, but also because of the consumer's changing eating habits, making an increase in their production inevitable [3].

Lettuce is a plant belonging to *Asteracea* family (formerly *Compositae*), with scientific name *Lactuca sativa L.* It is the most popular of leafy vegetables, being grown in almost all regions of the globe. It is an herbaceous plant, with an annual cycle, with a straight and undeveloped root, more or less branched according to the production method and soil type [4]. The consumption of this vegetable provides several benefits, including a high source of dietary fibre, low calorie levels, as well as being rich in mineral salts, vitamins (especially vitamin A and C), thiamine and riboflavin [5,6]. It also has several medicinal properties such as calming, diuretic, laxative, depurative and detoxifying effect [7]. For this reason, it is recommended by doctors and nutritionists as part of a healthy diet [8].

Human infection with protozoa and helminths from raw vegetables such as lettuce has increased considerably, due to the expansion of food trade, changes in eating habits and disorderly growth in large urban centers, affecting

individuals of all ages [9,10].

Lettuce contamination has several forms, including the use of water contaminated by human or other animal excrement during the irrigation process, or by contact of vegetables with animals such as flies and rats. This contamination by pathogenic agents occurs not only during the production chain, but also during harvesting, transportation or handling at the place of sale [11].

Lettuce sold in markets and open fairs in Maxixe city has a higher probability of suffering biological alterations and contamination by different pathogenic microorganisms, due to the low level of health in the markets, precarious sewage networks, inadequate handling in these places sales, in addition to factors related to the place of production. In addition, no study has been conducted in this province of Mozambique, with the intention of assessing the level of contamination of vegetables, especially edible raw. It was for these reasons that the present study aimed to assess the parasitological quality of lettuce sold in the markets of Maxixe city in Inhambane province, the southern part of Mozambique.

## Materials and Methods

### Collection and transport of samples

The study was conducted in June and July 2018, in Maxixe city, Inhambane province, the southern part of Mozambique. A total of 50 lettuce samples were purchased in 3 markets, namely the Tshuhla, Xicandjuanine and Dumbanengue. The selection of samples in these markets was based on the macroscopic characteristics of the lettuce.

The selected samples were placed in plastic bags, correctly identified and they were transported to the Laboratory of Medical Microbiology and Parasitology at the Faculty of Health Sciences of Pedagogical University of Mozambique, for further analysis.

### Sample processing and analysis

In the Laboratory of Medical Microbiology and Parasitology, lettuce samples were analyzed using the technique of Hoffmann and Pons-Janer (1934), also known as the spontaneous sedimentation technique [12,13]. Briefly, with the use of gloves, the individual samples of the lettuce were fragmented and placed in Elermeyers containing 100 ml of distilled water and stirred for a period of 30 minutes, with the aim of helping to remove dirt and possible parasites. After this stage, the leaves were removed and the water resulting from the washing was strained in gauze folded 4 times for specific sedimentation cups. The filtrate was left in the goblets for a period of 24 hours, so that the spontaneous sedimentation process could

take place. After the sedimentation step, using a Pasteur pipette, 0.05 ml of the sediment portion was transferred to a clean glass slide, then stained with a drop of lugol solution. Staining with the lugol solution was intended to facilitate the visualization of parasitic structures. Then, the preparation was covered with a coverslip and observed under a Motic binocular optical microscope. The sediment was analyzed in triplicate and in all fields of the slide, using 10x and 40x objectives to identify parasitic forms. Parasite identification and statistical analysis were performed with the aid of Parasitological Atlas: Atlas of Medical Protozoology and Helminthology and the SPSS program (24) was used to evaluate the contamination frequencies of the samples.

## Results and Discussion

In this work 50 samples of lettuce were analysed, coming from the lower areas of Maxixe city, Inhambane city and the district of Homóine. Macroscopically, the lettuce presented small amounts of sand characteristic of the growing areas, besides the presence of unidentified larvae in the laboratory. In general, the lettuce underwent a pre-washing process carried out by traders at the points of sale.

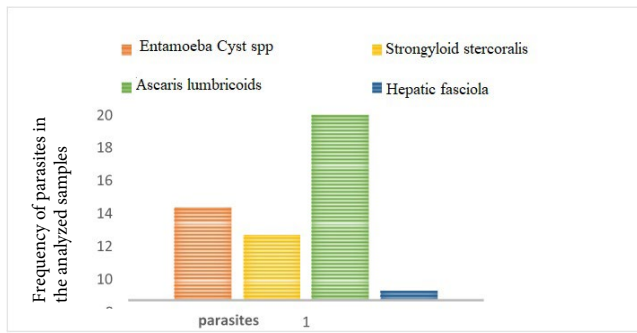
From 50 lettuce samples analysed in the Microbiology and Parasitology laboratory, 35 were positive, which corresponds to 70% of the contaminated samples, while the remaining 15 (30%) were negative, with statistically significant differences ( $X^2, P=0.002$ ) as shown in table 1. Among the markets, the highest rate of contamination by parasites was found in Tshuhla, where from 17 lettuce samples analyzed, all were positive, corresponding to a percentage of contamination equal to 100%, only in this market. On the other hand, Dumbanengue market was the second with the highest contamination rate, with 11 (64.7%) positive samples found in a universe of 17 samples analyzed, with the remaining 6 (35.3%) having a negative result. Finally, Xicandjuanine market recorded the lowest number of contaminated samples, with only 7 (43.75%) positive samples found, and the remaining 9 (56.25%) were found to be negative, as shown in Table 1 below.

**Table 1:** Number of positive and negative samples in the three markets in Maxixe city.

Place of purchase	N of samples	Positives (%)	Negatives (%)
Dumbanengue	17	11 (64.7)	6 (35.3)
Tshuhla	17	17 (100)	0 (0)
Xicandjuanine	16	7 (43.75)	9 (56.25)
Total	50	35 (70)	15 (30)

Among the parasites found in the 35 positive samples from the three markets, *Ascaris lumbricoides* was the

most frequent, followed by *Entamoeba histolytica* cysts, *Estrongiloides estercoralis* larvae and hepatic *Fasciola*, respectively, as shown in graph 1 below.



**Graph 1:** Frequency of parasites found in lettuce samples collected in Dumbanengue, Xicandjuanine and Tshuhla markets.

It was also found that most of the samples of lettuce 21 (60%) were contaminated by a single type of parasite, 10 (28.6%) were contaminated by two types of parasites and only 4 (11.4%) had polyparasitism, according to table number 2, shown below.

**Table 2:** Prevalence of monoparasitism, biparasitism and polyparasitism in lettuce samples sold in Maxixe’s markets.

Merket	Monoparasitism	Biparasitism	Polyparasitism	Total
Dumbanengue	6 (54.5)	2 (18.2)	3 (27.3)	11
Tshuhla	10 (58.8)	6 (35.3)	1 (5.9)	17
Xicandjuanine	5(71.4)	2 (28.6)	0(0)	7
Total	21 (60)	10 (28.6)	4 (11.4)	35

Lettuce is the vegetable with the highest rate of enteroparasitic contamination, having direct repercussions on human health, causing severe diarrhea, which can lead to dehydration, weight loss and anemia in more serious situations.

Mozambique is an endemic country for intestinal parasitic infections, with the highest prevalence of infection being seen in the central and northern provinces of the country, with southern provinces generally having the lowest infection rates [14]. According to data published by the aforementioned author, the prevalence of infection by intestinal parasites in school children in Maxixe city was moderate, varying between 20 and 47%. In this study, 70% (35/50) of the lettuce samples were contaminated by several types of intestinal parasites and 15 (30%) were negative for both eggs and larval forms. The high number of contaminated samples may be associated with poor hygiene conditions in the places where it is sold, inadequate handling and conservation, in addition

to the environmental conditions in the areas where it is cultivated, considering that sellers normally use water from some traditional wells existing in the low areas, to be able to wash the lettuce, which in a way favor the greater contamination of this vegetable.

These data corroborate the results obtained in other studies carried out in different parts of the world, with a high incidence of lettuce contamination, with emphasis on the research carried out by Cantos et al (2004), in Florianópolis, having obtained a prevalence of contamination lettuce samples in the order of 100% [15]. Similar results were also obtained in the study conducted by Rocha et al, having observed 98.5% of contamination of lettuce, sold at fairs in Recife, Brazil [16].

Most of the samples were contaminated by *Ascaris lumbricoides*, intestinal parasite of humans as illustrated in graph 1. The greater frequency of this parasite may be related to its greater adhesion to the leaves of the vegetable, considering its external morphology but also, this situation can be attributed to the fact that the large number of eggs released by females in the environment, facilitating the contamination of leafy vegetables. These results are in strict corroboration with the results found by Nascimento et al, in which there was also a greater contamination of lettuce samples by the parasite *Ascaris lumbricoides*, with a prevalence of 72.7% [17].

However, depending on the level of parasitism, nutritional and immunological status, *A. lumbricoides* can lead to death although in most cases, the infection is clinically less severe [18].

The second highest prevalence of parasitic contamination of lettuce analyzed in this study was by cysts of *Entamoeba histolytica*, also indicating possible contamination of human fecal origin. This parasite can cause acute amoebic colitis, severe dysentery, intestinal ulcerations and abscess (triggered by the body’s inflammatory response). Epidemiologically it occurs in about 10% of the world population, and with greater prevalence in tropical regions, such as Mozambique.

The Tshuhla market had the highest contamination rate of lettuce samples (100%). This market receives lettuce from several producers in Inhambane, mainly in Districts of Homoine, Morrumbene and several lower areas of Maxixe. In terms of sanitary hygienic characteristics, this market is in precarious hygiene conditions, with the presence of ponds with stagnant water, producing a bad smell, in addition to the occurrence of cockroaches and flies-vectors of parasites that cause diarrheal diseases. A different scenario was seen in Dumbanengue market, characterized by the existence of a fence wall, an efficient

and water drainage system in most stalls, however, despite the installation of the black water drainage system in this market, they were also found relevant prevalence of contamination of lettuce sold in the same market, as shown in table number 1.

### Conclusion

In this study high contamination rates of the lettuce marketed in the three markets of the city of Maxixe were observed, with the need to adopt adequate hygiene practices of the lettuce before its consumption, besides the need to strengthen the sanitary surveillance system, improve the conditions of the markets, through the adequate drainage of the black water and the installation of appropriate containers for the deposit of the solid residues generated there, thus avoiding the proliferation of vectors of diarrheal diseases.

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