Parasitic Contamination on Fresh Vegetable Consumed in Casablanca City (Morocco) and Risk for Consumer

K. Hajjami1,2, M.M. Ennaji2, H. Amdouri3, S. Fouad1, N. Cohen1
1Laboratory of Microbiology and Food Safety, Pasteur Institute of Morocco, Casablanca, 20360, Morocco
2Laboratory of Virology, Hygiene & Microbiology, Faculty of Sciences & Technics, University Hassan-II-Mohammedia, 20650, Morocco
3Laboratory of Physiology and Molecular Genetics, University Hassan-II- Faculty of Sciences Ain Chock Casablanca, Morocco

ABSTRACT

Parasites are commonly associated with vegetable or fruit borne outbreaks of gastroenteritis. Among all pathogenic intestinal parasites, helminthes cause the most number of parasitic infections in humans and animals, especially in developing countries. We conducted a study to determine the extent of parasitic contamination of raw vegetables sold in some markets of Casablanca (Morocco). Vegetables collected for the study were lettuce, radish, beet, cabbage, parsley, coriander, and mint. Of the 128 samples of vegetables, 80 (62.5%) were found to be contaminated with parasites, of which 11 (8.6%) were contaminated with infestive strongyle larvae. Helminth eggs detected included those of Ascaris sp. (10.2%), Toxocara sp. (8.6%), Taeniidae eggs (1.6%) and strongyle eggs (50.8%). The highest mean charges of parasitic contamination were registered in radish (11.86 eggs/200g) and lettuce (7.9 eggs/200g). Results of this study show that parasitological contamination of vegetables sold in Casablanca markets may pose a health risk to consumers of such products.

Keywords: Vegetables, markets, helminthes, Casablanca.

1. INTRODUCTION

Parasitic food-borne diseases are generally underrecognized, however they are becoming more common (Dorny, 2009). One of the major factors influencing the prevalence of parasitic infections in the population is the habit, and traditional popularity of eating raw or inadequately cooked foods (Anantaphruti et al., 2001). Albeit it is estimated that as much as 60% of the world’s population is infected with gut parasites (pathogen and nonpathogen), which may be transmitted through direct and indirect contact, food, water, soil, vertebrate and arthropod vectors and, rarely from mother to offspring (Brown and Neva, 1987; Kang et al., 1998), intestinal parasites are widely prevalent in developing countries, it is probably due to poor sanitation and inadequate personal hygiene (Kang et al., 1998). There is a wide variety of food products that may be contaminated with one or more parasites and consequently enabling transmission to human beings. The prevalence of specific parasites in food supplies varies between countries and regions (Anantaphruti et al., 2001).

In recent years there has been an increase in the number of reported cases of food-borne illness linked to fresh vegetables (Daryani et al., 2008). In fact, several surveys in different parts of the world show that the vegetables can be agent of transmission of helmint eggs and larvae (Hymenolepis, Taenia, Fasciola, Whipworm, Trichostrongylus, Strongyloides and Hookworms) (Choi, 1972; Choi et al., 1982; Coelho et al., 2001; Ergodrul and Sener, 2005). Other studies conducted in Morocco and some countries with widespread helminthic infections (Amahmid et al., 1999; Da Silva et al., 1995; De Oliveira et al., 1992; Ogunba et al., 1986; Rude et al., 1984) have shown that consumption of raw vegetables and fruits without proper washing or peeling represents an important potential for the spread of related infections.

The extent of contamination depends on several factors that include, among others, use of untreated wastewater and water supplies contaminated with sewage for irrigation, post-harvest handling, and hygienic conditions of preparation in food service or home settings (Amoah et al., 2007; Beuchat 2002; Simoes et al., 2001).

The environmental route of transmission is important for many helminth parasites. Both the potential for producing large numbers of transmissive stages and their environmental robustness, being able to survive in moist microclimates for prolonged periods of time, pose a persistent threat to public and veterinary health. The increased demands on natural resources increase the likelihood of encountering environments and produce contaminated with parasites (Slifko et al., 2000). The helminth parasites that may be acquired by eating these foods are nematodes, trematodes, cestodes and protozoa (Anantaphruti et al., 2001).

The objective of the present study was to assess the degree of parasitic contamination of vegetables sold in Casablanca city markets, in order to have a global view
about potential health risk that human are exposed, if those vegetables are eaten raw.

2. MATERIALS AND METHODS

This study was carried out on vegetables consumed in Casablanca city between April 2011 and Mai 2012. The city of Casablanca (Latitude: 33 34’N, Longitude: -7 40’W) (Anonymous 2012) is located north-west of Morocco, in the Chaouia plain and had an estimated population of 3 672 900 inhabitant in 2009 (Anonymous 2012). The annual rainfall is around 425 mm (Anonymous 2012). A total of (n=128) vegetable samples, including lettuce, radish, beet, cabbage, parsley, coriander, and mint were collected randomly from some Casablanca city markets. The markets were also selected randomly and sampling was done according to the availability of such crops. Vegetables selected are generally eaten uncooked and without peeling. Specific information regarding water used to irrigate fields from which the vegetables were grown could not be obtained. The samples were transported to the laboratory in sterile plastic bags for parasitological analysis.

Portion vegetables are weighted (200 g for all items) into plastic bags and prewashed with tap water then with physiological saline solution (0.95% NaCl). The washing water was left for about 24 h for sedimentation to take place. The top layer was discarded and the remaining washing water centrifuged at 1200 rd.min-1 for 15 min. The supernatant was discarded and the residue carefully collected. The supernatant was discarded, the residue carefully collected and examined by following the technique of Arther Fitzgerald (Arther Fitzgerald et al., 1981; Hajjami et al., 2012). Microscopic observation was performed in a Mac Master counting cell at 100-fold magnification.

3. RESULTS

Results of helminth eggs contamination are presented in (Tables 1). About 60% of samples analyzed were contaminated with different intestinal parasites. The parasites recovered were Ascaris sp., Toxocara sp., Taeniidae eggs, strongyle eggs and some infestive strongyle larvae. This study showed that high percentages of examined samples were positive for embryonated helminth eggs.

Table 1: Prevalence of intestinal parasites in vegetables consumed in Casablanca, Morocco

<table>
<thead>
<tr>
<th>Infection position</th>
<th>No and (%) of samples analyzed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non contaminated</td>
<td>48 (37.5)</td>
</tr>
<tr>
<td>Contaminated</td>
<td>80 (62.5)</td>
</tr>
</tbody>
</table>

**Polyparasitism**

| One helminth     | 58 (72.5) |
| Two helminths    | 14 (17.5) |
| Three helminths  | 4 (5)     |

**Name of parasites**

| Toxocara sp.     | 11 (8.6) |
| Ascaris sp.      | 13 (10.2) |
| Taeniidae eggs   | 2 (1.6) |
| Strongyle eggs   | 65 (50.8) |
| Infestive strongyle larvae | 11 (8.6) |
| Free-living larvae | 1 (0.8) |

Table 2 shows parasitic contaminations in vegetables sold in Casablanca markets. Of the 33 lettuce, 25 radish, 27 mint and 31 parsley and coriander samples examined, parasites were found respectively in 69.7%, 76%, 70.4% and 51.6%.
Eggs of *Ascaris* sp. were detected in lettuce (12.1%), radish (16%), mint (3.7%), Parsley & coriander (9.7%) and cabbage (20%); eggs of *Toxocara* sp. in lettuce (9.1%), mint (11.1%) and Parsley & coriander (16.1%). It is noted that 7.4% mint samples contain *Taeniidae* eggs. For the infestive strongyle larva, they are found in radish (16%), beets (14%), Parsley & Coriander (9.7%) and lettuce (9.1%).

The number of helminth eggs detected per 200 g of the vegetables is presented in (Table 3). Globally, lettuce and radish samples noted the higher mean charges of helminth eggs, *Ascaris* sp. and strongyle eggs were found respectively with an average concentration of 0.32 eggs/200g and 11.54 eggs/200g in radish and 0.27 eggs/200g and 7.06 eggs/200g in lettuce. Also, the highest charge of infestive strongyle larvae was recovered in radish samples (0.6 larvae/200g) followed by beets samples (0.43 larvae/200g).

### Table 2: Distribution of intestinal parasites in different vegetable samples

<table>
<thead>
<tr>
<th>Vegetables</th>
<th>No and (%) of positive samples</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><em>Ascaris</em> sp.</td>
</tr>
<tr>
<td>Mint (n=27)</td>
<td>1(3.7)</td>
</tr>
<tr>
<td>Parsley &amp; Coriander (n=31)</td>
<td>3(9.7)</td>
</tr>
<tr>
<td>Cabbage (n=5)</td>
<td>1(20)</td>
</tr>
<tr>
<td>Lettuce (n=33)</td>
<td>4(12.1)</td>
</tr>
<tr>
<td>Radish (n=25)</td>
<td>4(16)</td>
</tr>
<tr>
<td>Beets (n=7)</td>
<td>0(0)</td>
</tr>
<tr>
<td>Total (n=128)</td>
<td>13(10.2)</td>
</tr>
</tbody>
</table>

### Table 3: Determined egg counts on infected samples

<table>
<thead>
<tr>
<th>Vegetables</th>
<th>No. of eggs*[(Mean and (range)) /200g]</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td><em>Ascaris</em> sp</td>
</tr>
<tr>
<td>Mint (n=27)</td>
<td>0.04 (0-1)</td>
</tr>
<tr>
<td>Parsley &amp; Coriander (n=31)</td>
<td>0.13 (0-2)</td>
</tr>
<tr>
<td>Cabbage (n=5)</td>
<td>0.20 (0-1)</td>
</tr>
<tr>
<td>Lettuce (n=33)</td>
<td>0.27 (0-5)</td>
</tr>
<tr>
<td>Radish (n=25)</td>
<td>0.32 (0-3)</td>
</tr>
<tr>
<td>Beets (n=7)</td>
<td>0.00</td>
</tr>
<tr>
<td>Total (n=128)</td>
<td>0.16</td>
</tr>
</tbody>
</table>

* No. of larvae/200g for infestive strongyle larva counting.

### 4. DISCUSSION

In developing countries, such as Morocco, intestinal parasites infestations are very common. Consumption of fresh vegetables is a major way in the transmission of parasitic contaminations (Daryani 2008). Our study revealed presence of helminths in 62% of samples analyzed with an average concentration of 4.57 eggs/200g and 0.2 larvae/200g. Several studies around the world, into the recovery of parasites from vegetables sold in markets or collected from wholesale have been conducted and showed their presence with a high prevalence (Akhlafi et al., 2000; Davami et al., 2000; Zohour et al., 2001; Dayrani 2008; Abougrain et al., 2010). In Morocco, a few studies has been conducted in this sens, Idrissa et al., (2010) identified parasites in mint and turnips; obtained from market traders of the city of Sidi Yahia (Morocco) with an average concentration of 4.71 eggs/100g and 4.3 eggs/100g respectively. But more works about parasitological contamination of vegetables irrigated by wastewater was conducted; Rhallabi et al., (1990) registered a presence of helminth eggs with an average concentration of 2eggs/kg in tomatoes, Bouhoum et al., (2002) found helminth eggs in Coriander, Carrots, Radish and Mint with a mean a average of 7.25 eggs/100g in lettuce and potatoes respectively. It has been proven that the water used in irrigation was implicated as a major route of direct parasitological contamination of the fruits and vegetables (Guilherme et al., 1999; Takayanagui et al., 2000; Ulukanigil et al., 2001; Kozan et al., 2005; Idrissa et al., 2010). And
studies conducted show that helminth eggs loads in vegetables crops are highly dependent on the contamination level of irrigation water (Bouhoum et al., 2002; Dssouli, 2001; Dssouli et al., 2001). Bryan (1977) observed that field vegetables were directly contaminated with irrigation water or indirectly by contact with contaminated soil.

The present study shows that detected helminth numbers varied according to the type of vegetables. Radish samples have noted the higher percentage of contamination (76%) and mean charge (11.86 eggs/200g) followed by lettuce then mint, parsley and coriander. This result corroborate with those reported by several authors who have found that the roots are the most contaminated by helminth eggs. It can be explained by their direct contact with contaminated water during irrigation cycles. (Rosas et al., 1984; Shuval et al., 1986; Firadi 1996; Naour et al., 1996; Dssouli et al., 1997; El Hamouri et al., 1998). Other studies show that vegetables with dense foliage (lettuce) was most contaminated that those growing on surface (Turnip) (Bouhoum et al., 2002; Idrissa et al., 2010). The dense foliage would protect the helminth eggs against unfavorable conditions to their survival and persistence, such as sunlight, drying, win (Dssouli 2001; Dssouli et al., 2006). In an other hand, the survival of parasites in the area of crop will be much shorter because the parasites are less well protected against light and drying, but may survive long enough to risk contaminating people who handle or consume products especially when time survival is longer than the growth cycle, as is often the case with those vegetables (WHO 1989).

In this survey we note contamination of vegetables with Ascaris eggs, Toxocara eggs, Taeniidae eggs, strongyle eggs and Infestive strongyle larvae. These eggs are very resistant in the environment and they can survive and remain viable in soil and crops for several months/years (Feachem et al., 1983; Olson et al., 1997; Jenkins et al., 1999; Roepstorff et al., 2001; Jiang et al., 2002; Ogden et al., 2002).

Considering the morphology, eggs of Ascaris lumbricoides are very difficult to distinguish from Ascaris suum. However, swine production, with which A. suum is associated, is extremely limited and pork consumption rarely occasion in Morocco. Therefore, it is suggested that the eggs are most likely those of A. lumbricoides. This roundworm is found worldwide and the WHO (1996) reported that it infect 1.4 billion people each year. It has a direct life cycle. Ova are found in insufficiently treated sewage fertilizer and in soils, where they embryonate upon exposure to air or oxygen in order to become infective. Under ideal conditions, this usually requires about 3 weeks. The ova may contaminate crops grown in soil or fertilized with sewage that has received non-lethal treatment. People can acquire the infection through consuming such raw produce (Cotruvo et al., 2004).

Ascaris ova are sticky and can adhere to items such as cooking utensils, furniture, money, door handles, and fingers. Presence of Ascaris eggs in the samples threat Moroccan health diet especially children (Cotruvo et al., 2004; Sakey, 2001). It has been reported to be of high prevalence in several regions in Morocco (Bouhoum, 1998; Amahmid et al., 2005; Kettani, 2006; Kettani, 2008; El Guamri, 2009).

A part from Strongyle eggs, it is true that the parasite concentrations are very low but should in no way obscure the fact that it risk contaminate the consumer. In fact, the consumption of just one embryonated helminth egg may be sufficient to cause infection (Cotruvo et al., 2004).

5. CONCLUSION

Taking account of results obtained, it is clear that irrigation process of these vegetables fields has been performed using contaminated water sources (wastewater). This work shows that viable-stage ova recovered constitute a potential risk for consumer especially children. Therefore, washing and disinfecting procedures of the raw eaten vegetables must be investigated, if not these ones can be important vehicles of transmission human pathogen helminthes to Casablanca’s citizen.

For having more clear vision about this danger, a quantitative analysis then estimation of risk will be investigated; the present study will be of considerable interest in the evaluation of this parasitological risk.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge all collaborators in this study.

REFERENCES


