Prevalence of Soil Transmitted Helminths in Fresh Vegetables Sold at Several **Traditional Markets in Surabaya City**

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Abstract

Soil-transmitted helminths infections are major public health problems in developing countries that require fecal contamination of the environment for transmission. The consumption of raw vegetables without proper washing is one of the main routes of parasite transmission. Therefore, this study conducted to detect the kind and prevalence of soiltransmitted helminths contamination in commonly consumed fresh vegetables sold at several traditional markets in Surabaya. 100 fresh vegetable samples consisting of yardlong bean, cabbage, lettuce, and lemon basil were purchased from five traditional markets in Surabaya. Each sample was washed with tap water, water washing examined using flotation method to find soil-transmitted helminths. The overall prevalence of soil-transmitted helminths contamination was 41% (41/100). The most predominant parasite was hookworms (20%), followed by Strongyloides stercoralis larvae (14%) and Ascaris lumbricoides eggs (7%). The highest prevalence of contamination found in lettuce, with 56% prevalence (14/25), while the lowest prevalence of contamination found in yardlong bean with 24% prevalence (6/25). Sequentially, the prevalence of soil-transmitted helminths contamination of fresh vegetable samples from Pabean traditional market (55%), Keputran traditional market (50%), Wonokromo traditional market (50%), Lakarsantri traditional market (35%) and Mulyosari traditional market (15%), respectively. The present study demonstrated that consumption of vegetables with parasite contamination in this area represents a potential route for the transmission of soil-transmitted helminths infection. Therefore, it is necessary for health authorities to educate consumers about the proper washing of vegetables prior to consumption. Preventive methods such as wearing gloves and washing hands after handling vegetables should advocated to sellers who are at risk of acquiring soil-transmitted helminths infection.

Keywords: Soil transmitted, helminths contamination, Fresh vegetables, traditional markets

Introduction

Soil-transmitted helminths or intestinal nematodes are nematodes which in their life cycle require soil to reach the infective stage. This group of nematode worms often causes infections in humans. Worm larvae can thrive in warm conditions in moist soil in tropical and subtropical countries. More than one billion people worldwide infected by this type of worm by at least one species (Bethony et al., 2006). Approximately 270 million sufferers are preschool children and around 600 million are school-age children living in areas with this parasitic infection so that effective treatment and prevention is needed (WHO, 2017). Meanwhile, the prevalence of worms in Indonesia is generally still very high, ranging from 10-85.9%, especially among underprivileged people with poor sanitation (Ditjen PPPL, 2012). The results of

research in Surabaya show that the prevalence rate of soil-transmitted helminths infection in elementary school age children is 13.04% (Amalia and Prasetya, 2017). The types of soil-transmitted helminths that infect humans are Ascaris lumbricoides (A. lumbricoides), Trichuris trichiura (T. trichiura) and hookworms [Ancylostoma duodenale (A. duodenale) and Necator americanus, (N. americanus)].

Soil-transmitted helminths infection can reduce the health condition, nutrition and productivity of sufferers so that economically it causes a lot of losses, because of the loss of carbohydrates, protein and blood which in turn can reduce the quality of human resources. In children, this infection can cause growth and development disorders and decreased concentration in learning, thereby affecting children's intelligence (Irianto, 2009).

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The low level of personal sanitation affects the incidence of this disease, such as not washing hands before eating and after defecating, not maintaining hygiene and cutting nails. Another factor that influences the incidence of worms is a supportive environment for the development of soil-transmitted helminths, that is flabby and moist soil conditions. In addition, farmers often use organic fertilizers in the form of humus or livestock manure (even human dung) to increase soil fertility (Sofiana, 2010) providing an opportunity for soil-transmitted helminths contamination. Worm contaminants can occur especially on vegetables that spread on the ground or at a height close to the ground from several agricultural production centers in East Java Province to the shipping and trade chain patterns in several traditional markets in big cities such as Surabaya. The habit of eating raw vegetables, which is a tradition in many Indonesians who less attention to hygiene aspects, also contributes to the incidence of worms (Widjaja et al., 2014).

Some vegetables such as cabbage, lettuce, yardlong beans and basil are some of the vegetables commonly consumed by Indonesians in their raw condition. These vegetables have the potential to transmit and spread soil-transmitted helminths because of the frequent contamination of soil-transmitted helminths. Several studies that have been conducted on several vegetables report that 61.2% of the prevalence rate of soiltransmitted helminths contamination of raw vegetables occurs in fields (Karuppiah, 2017). The results of another study by Mutiara (2015) showed that there was still contamination of worm eggs in foods made from raw vegetables in the Unila canteen by 21.1% of the sample studied. The contaminants were A. lumbricoides eggs (50%), N. americanus (25%), and a combination of *T.trichiura* and *A. lumbricoides* (25%). The results of previous studies indicate that there is intestinal worm contamination quite high in cabbage vegetables, that is 71.67% with the types of worm eggs found were A. lumbricoides (6.67%), T. trichiura (3.33%), and A. duodenale (80%) (Maemunah, 1993).

Several traditional markets in Surabaya are some of the destinations for the delivery of vegetable production centers in East Java Province, which are thought to have contributed to the transmission and spread of soil-transmitted helminths. Based on surveys and field observations, several traditional markets in Surabaya is less attention to environmental sanitation, both the placement and the stalls where the vegetables are sold.

A study conducted by Adrianto (2018) on lettuce sold in several traditional markets in Surabaya found that 61.90% of lettuce sold positively contained soil-transmitted helminths eggs from the Ascaris spp. The aim of present study is to detect the kind and prevalence of soil-transmitted helminths contamination in commonly consumed fresh vegetables sold at several traditional markets in Surabaya.

Methods Study area

A cross-sectional study was conducted from March to June 2020 in Surabaya (capital city of East Java province), which is located on northeastern border of Java island, 7°15′1.6020″S latitude and 112° 46′7.84206″E longitude. The temperature averages 27.1 °C | 80.8 °F. About 1679 mm | 66.1 inch of precipitation falls annually (Climatological Center, Indonesia Meteorological Department, Annual report 2019). According to official statistic registration systems, the total population in this area is approximately 2,843,144. Fresh vegetable samples were purchased from five central open-air traditional markets located in Keputran (Surabaya Center), Mulyosari (Eastern Surabaya), Lakarsantri (Western Surabaya), Pabean (Northern Surabaya) and Wonokromo (Southern Surabaya) (Fig. 1). The fresh vegetables sold in those markets were brought from different farms and agricultural areas in different parts of East Java province.

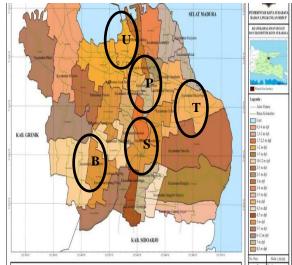


Figure 1. Several locations of traditional market at Surabaya City area.

Sample collection

A total of 100 fresh vegetable samples including 4 different types that are frequently consumed without cooking were randomly purchased from sellers in five central open-air

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traditional markets. The fresh raw vegetable samples used in this study included lettuce (*Lactuca sativa*), cabbage (*Brassica oleracea*), lemon basil (*Ocimum africanum*), and yardlong bean (*Vigna unguiculata*). The vegetable samples demonstrated in Fig. 2. The fresh vegetable samples collected in clean, labeled plastic bags and transported immediately to the parasitology laboratory at the Faculty of Medicine, Airlangga University for parasitological examination.

Detection of intestinal parasites

Fresh vegetable samples weighing 200 g washed with 1000 mL of physiological saline solution (0.9% sodium chloride) and shaken for 15 minutes in order to separate the parasites from vegetables. Then, the washing water collected and left overnight to allow sedimentation. Afterward, the supernatant was decanted, and the remaining washing water transferred to 12 mL conical tubes. To concentrate the parasitic stages, the sediment centrifuged at 2000×g for 15 minutes. After centrifugation, the supernatant carefully removed without shaking. Then, the sediment was agitated gently and examined under a light microscope using 10x and 40x objectives. To increase the chance of parasite detection, three slides were prepared from each sample by two independent investigators.

Statistical analysis

Data analysis performed with IBM SPSS Statistics for Windows, Version 23.0. Armonk, NY: IBM Corp. Qualitative variables described by frequency (percentage). The results served and narrated descriptively the rate of parasitic contamination among different types of vegetable and among different markets.

Results and Discussion

A total of 100 fresh vegetable samples were the presence of examined for parasite The results of contamination. parasite contamination in vegetable samples shown in Table 1. The parasites detected in vegetable samples were hookworm, S. stercoralis and A. lumbricoides. Pictures of some of the parasites found in this survey demonstrated in Fig. 3. The overall rate of intestinal parasite detection was 41% (41/100) in all vegetable samples. The highest rate of contamination was found in Lactuca sativa [56% (14/25)] while the lowest was found in Vigna unquiculata [24% (6/25)] (**Table 1**).

The highest prevalence of hookworm contamination found in cabbage, while the lowest was in yardlong beans. Larvae of *S. stercoralis* most frequently contaminated lettuce, while the lowest contamination occurred in yardlong beans. Eggs of *A. lumbricoides* often contaminate lettuce, followed by second highest contamination in cabbage and lowest in lemon basil and yardlong beans. Meanwhile, in this study, no contamination found, either eggs or larvae of *T. trichiura* worms in all types of vegetables studied and taken in several traditional markets in Surabaya.

Table 2 shows the prevalence rates of soil transmitted helminths (STH) parasite contamination in vegetable samples that varied among five traditional markets in Surabaya. Some of the prevalence of STH parasites that contaminate vegetable samples taken from several traditional markets, including Keputran market (50%), Mulyosari market (15%), Lakarsantri market (35%), Pabean market (55%) and Wonokromo market (50%). The highest prevalence of STH parasite contamination in vegetables found in the Pabean market, while the lowest prevalence found in the Mulyosari market.

Table 1. The distribution of the soil transmitted helminths parasites contaminated the fresh vegetables taken at several traditional markets in Surabava.

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Vegetables	Σ sample	Hookworm	Strongyloides	Ascaris	Σ positive	
		larva/eggs	stercoralis	lumbricoides	(%)	
		(%)	larva (%)	egg (%)		
Vigna unguiculata	25	3 (12)	2 (8)	1 (4)	6 (24)	
Brassica oleracea	25	7 (28)	4 (16)	2 (8)	13 (52)	
Lactuca sativa	25	6 (24)	5 (20)	3 (12)	14 (56)	
Ocimum africanum	25	4 (16)	3 (12)	1 (4)	8 (32)	
				()		
Total	100	20 (20)	14 (14)	7 (7)	41 (41)	

vegetables from several traditional markets in Surabaya												
Vegetables	es Σ sample Keputran Market		n Market	Mulyosari Market		Lakarsantri		Pabean Market		Wonokromo		
	(test)						Ma	rket			Ma	rket
	test	Σ	Σ	Σ	Σ	Σ	Σ	Σ	Σ	Σ	Σ	Σ
		positive	sample	positive	sample	positive	sample	positive	sample	positive	sample	positive
		(%)	test	(%)	test	(%)	test	(%)	test	(%)	test	(%)
Vigna	25	6 (24)	5	1 (20)	5	0	5	1 (20)	5	1 (20)	5	2 (40)
unguiculata												
Brassica	25	13 (52)	5	3 (6o)	5	1 (20)	5	2 (40)	5	4 (8o)	5	3 (6o)
oleracea												
Lactuca	25	14 (56)	5	4 (8o)	5	2 (40)	5	3 (6o)	5	4 (8o)	5	3 (6o)
sativa												

3 (15)

20

Table 2. The parasite distribution of soil transmitted helminths which contaminates different fresh vegetables from several traditional markets in Surabaya

Table 2 shows the prevalence of parasite contamination in different vegetable samples among the five markets. The parasites were detected in 55%, 50%, 50%, 35% and 15% of samples obtained from Pabean Market, Wonokromo market, Keputran market, Lakarsantri market and Mulyosari market, respectively. The highest rate of parasite contamination in vegetables found in Pabean market, whereas the lowest rate observed in Mulyosari market. Statistical analysis revealed that the rate of contamination in vegetables obtained from Pabean market was significantly higher than Lakarsantri and Mulyosari markets.

2 (40)

10 (50)

8 (32)

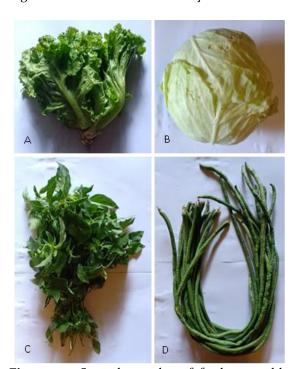
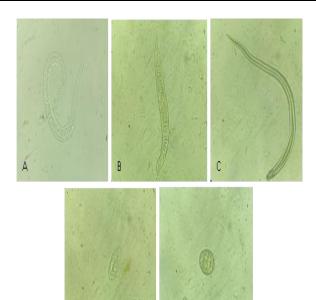


Figure 2. Several samples of fresh vegetables were tested for parasite contamination by soil transmitted helminths. A. *Lactuca sativa*; B. *Brassica oleracea*; C. *Ocimum africanum*; D. *Vigna unguiculata*.



2 (40)

11 (55)

2 (40)

10 (50)

1 (20)

7 (35)

Figure 3. Representation of soil transmitted helminths parasites, larvae and eggs that contaminate fresh vegetables were observed in the study. Several larvae [A, rhabditiform *A. duodenale* (Hookworm); B, rhabditiform *S. stercoralis*; C, filariform *S. stercoralis* and Egg (D, *A. duodenale* (Hookworm); E, *A. lumbricoides*]. Magnification: 400X (based on the key of nematode worm identification by Mkandawire *et al.*, 2022).

Discussion

Consumption of fresh raw vegetables has an important role in parasite transmission to humans. The control of parasites of vegetable origin is very helpful in understanding the potential sources of parasitic pathogens described in the scope of this study. This study shows that the prevalence of STH parasite contamination in

Ocimum

Total

africanum

vegetable samples in several traditional markets in Surabaya is 41%, which is higher than the study conducted by Mutiara (2015) which reported that contamination of STH eggs in food made from raw vegetables in the Unila canteen was equal to 21.1% of the sample studied. The results of several studies on the prevalence of STH parasite contamination in several countries show variations, ranging from 45% to 58%, recorded in Libya, Iran, Egypt, Brazil, and the Philippines (Bekele et al., 2017). The differences in the prevalence rates in this study with previous studies may be due to variations in climatic conditions, soil types, types of water used for agriculture and poor hygienic practices during transportation and marketing of vegetables. Annisa et al. (2018) reports that several factors influence the high and low prevalence of STH parasite contamination in raw vegetables, one of which is climate, which closely related to temperature. The influence of climate / season on parasite prevalence, according to the study, reports that the prevalence rate of STH parasite contamination is higher in the rainy season than in the summer. Rozendal (1997) states that parasites and disease vectors are very sensitive to climatic factors, especially temperature, rainfall, humidity, water level, and wind speed. The findings in this study suggest that parasite transmission and prevalence rates should be linked to climate and temperature.

Lettuce was found to be the most frequently contaminated vegetable sample (56%) followed by cabbage (52%), basil (32%), and long beans (24%). The results of this study are consistent with previous research, which reported that the highest STH lettuce had parasite contamination among several types of vegetables studied (Adrianto, 2018). In Indonesia, especially in Surabaya, lettuce is generally served in the form of several dishes known as " gado gado " in raw condition. The highest contamination in lettuce is probably because lettuce consists of a number of stems connected at the base (ground) with leaves near the top of the stem. The stem structure is Ushaped in a slit pattern, which allows parasites to adhere more easily to the surface of this vegetable and makes it more difficult to remove. Typically, lettuce samples obtained from five markets in Surabaya are often sold at the base of the stem, which increases the likelihood of contamination near the stem and then to the leaves. In almost the same conditions, cabbage vegetables are in contact with the soil, which allows STH parasite contamination, but because of the habit, the outer stems and leaves always discarded when they sold and taken clean, thus

reducing the possibility of contamination. The lowest STH parasite contamination observed in long beans, this happened because the condition of the long bean vegetable was hanging on the stems and stalks, thus minimizing the possibility of contact with the soil surface and reducing the possibility of STH parasite contamination, besides the smooth surface of long beans compared to the types of vegetable samples investigated in this study may reduce the likelihood of parasite adherence.

In this study, the prevalence of hookworm larvae and eggs recorded at 20% (20/100) in the examined vegetables and was the highest STH parasite found. This finding is higher than that of other studies conducted in Ghana (13%) (Duedu et al., 2014), Sudan (5.7%) (Mohamed et al., 2016), and northern Iran (4.40%). (Rostami et al., 2016). Report Previously in 2017, hookworm was the most common parasitic infection in Indonesia (Punsawad et al., 2017). The high prevalence of contamination of hookworm larvae and eggs on vegetables in this study could be attributed to poor sanitation and use of water contaminated by human waste for irrigation in agricultural areas. The high prevalence of hookworms caused by differences in geographic location, climatic conditions, and soil types (Silver et al., 2018). The second highest contamination found in this study was larvae of *S. stercoralis*. This finding is lower than previous studies in Ghana and Ethiopia, which reported the prevalence of *S. stercoralis* 43% and 21.9%, respectively (Tefera et al., 2014). The high prevalence of contamination with Strongyloides larvae may be due to Strongyloides spp. has a complex life cycle with a free life period in an environment that does not require a host for proliferation (Schar et al., 2013).

This study also found contamination of A. *lumbricoides* egg. The prevalence of A. lumbricoides egg was 7% (7/100) of all vegetable samples. The prevalence rate of A. lumbricoides egg in vegetables almost close to the results of a study conducted in Shahrekord, Iran at 8.17% (Fallah et al., 2016). While the absence of contaminant eggs or larvae of T. trichiura in all types of vegetables studied related to the life cycle of *T. trichiura* worms, where the eggs hatch in the small intestine, the larvae will come out, develop in the small intestinal mucosa and become adults in the cecum, eventually attaching to the mucosa. Large intestine (Garcia and Bruckner, 1997), so that the possibility of contamination of vegetables is also low.

Contamination of vegetables by STH parasites can occur at any point along the contamination chain; during the planting,

harvesting, transportation, or marketing of vegetables. Difference in the prevalence of contamination caused by soil type, quality of water used for planting and irrigation, and hygienic practices during the marketing process. The prevalence of STH parasite contamination is different for each vegetable taken from different markets; samples taken from the Pabean traditional market showed the highest prevalence of contamination. The differences between the five markets may be due to the different sources the vegetables come from as well as the hygienic practices of handling and washing by different vendors. The results of this study emphasize that raw vegetables from the market in the study area can be a risk factor for transmitting parasites to humans. The results of previous studies revealed that standard vegetable washing procedures are an effective method of preventing worm contamination in raw vegetables (Avcioglu et al., 2011; Fallah et al., 2012). Furthermore, to emphasize the proper vegetable washing procedure, research in Iran shows that prewashing procedures using tap water or underground water cannot completely remove parasites from vegetables (Fallah et al., 2016). The results of these studies recommend that health authorities should provide good knowledge about the proper and correct washing methods for the public to prevent parasite transmission. This research or study has several limitations which are important to note. Among them, this study does not involve the effect of seasonal variations on parasite contamination. Moreover, this study did not discuss the intensity of washing vegetables before they were put on display for sale or the water source used by each seller.

Conclusions

The results of the present study indicate that the contamination of raw vegetables with pathogenic parasites in several traditional markets in Surabaya city might represent a transmission vector for intestinal parasites to consumers. Prevention methods such as proper washing or cooking of vegetables before consumption should conveyed to consumers. In addition, comprehensive health education and hygienic practices, including wearing gloves and washing hands after handling vegetables, should be provided to sellers and farmer. This study suggests that the contamination of soiltransmitted helminths in some vegetables sold and bought in several traditional markets in Surabaya may occur from vegetable production centers to traditional markets in Surabaya. A more comprehensive study of soil-transmitted

helminths contamination in several types of vegetables sold and bought in several traditional markets in Surabaya needs to be done more thoroughly and is still important to be studied in order to provide information to the public about the dangers of soil-transmitted helminths contamination through several types of vegetables.

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Conflict of Interest

The authors declare no conflict of interest.

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