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Research Article

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DETECTION OF SOIL-TRANSMITTED HELMINTHS EGGS AND LARVAE IN VEGETABLE HERBS FROM ENVIRONS OF MEIKTILA, MYANMAR

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Abstract

Samples of common vegetable herbs; *Coriandrum sativum*, *Mentha arvensis* and *Eryngium foetidum* were collected from the two local markets (Paukchaung and Myothit) within Meiktila area and analyzed for helminths eggs/larvae using standard methodology. Incidence of larvae of *Strongyloides stercoralis*; eggs of *Ascaris lumbricoides*, *Taenia sp.*, *Trichuris trichiura*, and *Enterobius vermicularis* were recorded. The numbers of helminth eggs/larvae found on the roots of *C. sativum*, *M. arvensis*, and *E. foetidum* were 0 - 46, 0 - 18 and 0 - 27 per 100g wet weight, respectively. Total counts on helminthes eggs/larvae on the vegetable herbs from Myothit market were more abundant, especially *S. stercoralis* larvae (P<0.005) when compared with that of Paukchaung market. In the seasonal incidence rate of helminthes, August was found to be higher than in the rest of the months. Base on the results of this study, the importance of the transmission of intestinal parasites from vegetable herbs is stressed, and necessity to improve the sanitary conditions of these kinds of food is also suggested.

Keyword:- soil-transmitted helminths, prevalence, contamination vegetables herbs, Meiktila

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Introduction

Fresh vegetables are an important part of a healthy diet. In recent years there has been an increase in the number of reported cases of food-borne illness linked to fresh vegetables. The consumption of raw vegetables is a major way in the transmission of parasitic contaminations. Various epidemiological studies have indicated that the prevalence of intestinal parasites is high especially in developing countries. Intestinal parasites or soil-transmitted helminths (STHs) infections are endemic in communities where poor environmental sanitation and poor personal hygiene are prevalent as occurs in the majority of developing countries (Yodmani et al., 1982; Fallah et al., 2012).

Soil pollutions on vegetable productions (farms) with faecal materials are instrumental in the transmission of STHs infection. Fertilized eggs deposited in the soil, develop rapidly and depending on environmental conditions, may reach the infective stage within a matter of weeks (Klaas, 1987). Thereafter, eggs are transferred from soil to the vegetables then onto to the hands and finally to the mouth (Koyabashi, 1999). Both agricultural workers and vegetables or crops consumers, eating of raw or improperly washed vegetables, may cause parasitic infections through adhered helminthes eggs or larvae. Bad hygienic practice during production, transport, processing and preparation by handlers including consumers also contribute in vegetable contaminations (Gupta et al., 2010). Soil-transmitted helminths can produce a wide range of symptoms including intestinal manifestations (diarrhea, abdominal pain), general malaise and weakness, which may affect working and learning capacities and impair physical growth. Hookworms cause chronic intestinal blood loss that result in anaemia (WHO, 1989).

Meiktila is situated in the central part of Myanmar, with the suitable climate, vegetation and topography for the cultivation of several kinds of vegetables. They are grown throughout the year in Meiktila area using water derived from Meiktila Lake and hand dug wells. These cultivated as well as transported vegetable were sold in different markets of Meiktila environs. Some marketed vegetables are often contaminated by eggs and larvae of human intestinal nematodes that can infect human. The vegetable herbs, *Coriandrum sativum* (Coriander), *Mentha arvensis* (Podina) and *Eryngium foetidun* (Culantro or Shan-nannan) sold in different markets of Meiktila area are commonly used for preparation for Myanmar traditional recipe (Monk-hin-kha) and varieties of salad and used as parsley in cooking for fishes and meats. The roots of these green leafy vegetable herbs were usually contaminated with soil that may contain eggs/larvae of STHs. When these vegetable herbs are eaten in raw or without removing of soil-contaminated roots and unwashed, the eggs/larvae of STHs may be ingested and cause parasitic infections.

In Myanmar, the incidence of STHs in *Ipomea aquatic* and *Hibiscus sabdariffa* has been reported by Thiri Kyaw (2008) from the different markets of Yangon environs. However, the contamination of helminthes eggs/larvae on green leafy vegetable herbs have not been reported in Meiktila environs. The present

study was undertaken to assess the contamination levels of helminthes parasites on selected vegetable herbs and to examine the prevalence of helminths eggs/larvae in selected two study sites.

MATERIAL AND METHODS

Study Area and study period

The study area, Meiktila Township, is situated in the Central Dry Zone Myanmar and located between Latitude 20˚ 00 N and Longitude 95˚ 01' E. The samples were collected from the two local markets, Paukchaung and Myothit markets. This study was carried out from July 2009 to February 2010 on weekly basis.

Collection of vegetable herb samples

Vegetable herb samples (100g), *C. sativum* (Coriander), *M arvensis* (Podina) and *E. foetidum* (Culantro or Shan-nannan) were collected from two local markets. They were packed in separate transparent plastic bags and brought to the laboratory for examination. In this study, vegetable herb samples were collected from each study site per week. A total of 192 vegetable samples were used from the two study sites (64 samples for each type of vegetable herbs).

Preparation of samples for analyzing helminthes eggs/larvae

The roots from each type of collected samples were cut approximately one cm from the base of the stem. One hundred gram weight of root samples from each vegetable sample was taken and washed in 50 ml, normal saline solution in separate beakers, and let to settle in the beakers. The supernatant from each beaker was discarded until the sediments remain in the beaker for microscopic examination. Two ml of the sediment was taken with a syringe and only a drop was put onto the glass slide and mixed with a drop of 2% Lugol's Iodine solution. This preparation was examined under the microscope for the presence of parasite, using ×10 and ×40 objectives. Reagents and Lugol's Iodine solution was prepared based on the method of WHO (1991).

Identification of helminths eggs/larvae

Presumptive identification of the helminthes was done based on morphological characteristics described by Yamaguti (1961) and Brown (1964). The helminthes eggs were identified on the basis of their shape and size and compared with standard eggs on charts followed after instruction of WHO (1991). The counting was done under Olympus light microscope at ×40 magnification.

Statistical analysis

Statistical analysis (ANOVA test) was performed by Microsoft Excel (Version 2010). A *P*-value <0.05 was considered statistically significant.

RESULTS AND DISCUSSION

Examination on the three vegetable herbs, *C. sativum* (Coriander), *M. arvensis* (Podina) and *E. foetidum* (Culantro or Shan-nannan) revealed that the roots were contaminated larvae of *Strongyloides stercoralis* along with the eggs of *Ascaris lumbricoides*, *Taenia sp*., *Trichuris trichiura*, and *Enterobius vermicularis*.

Prevalence of helminthes eggs/larvae in the three kinds of vegetable herbs from the two markets

Total mean helminths eggs/larvae detected from *C. sativum*, *M. arvensis* and *E. foetidum* collected in Paukchaung and Myothit markets are shown in Figure1 and Figure 2. The total means number of larvae of *S. stercoralis* detected from the two markets showed significantly higher than other parasites eggs (P<0.05). Mean number of helminths eggs/larvae detected in vegetable herbs collected from Myothit market was found to be higher than that of Paukchaung market (Table 1).

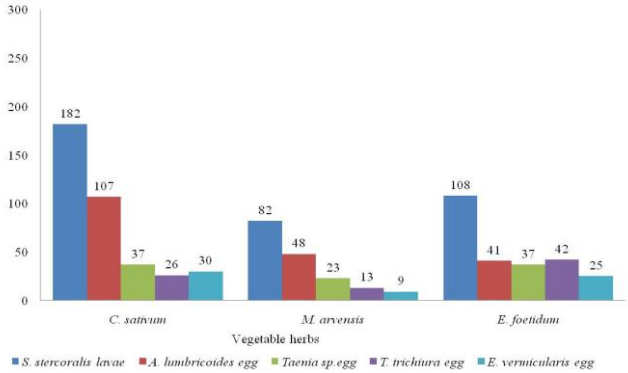

Figure 1. Total mean number of helminths eggs/larvae detected from *C. sativum*, *M. arvensis* and *E. foetidum* collected in Paukchaung market

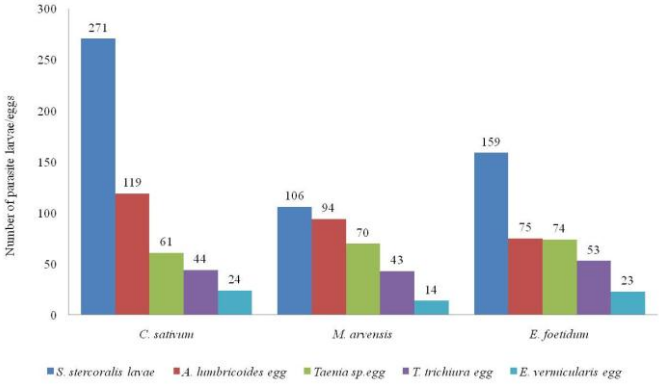

Figure 2. Total mean number of helminths eggs/larvae detected from *C. sativum*, *M. arvensis* and *E. foetidum* collected in Myothit Market

Helminths larvae and egg counts from all studied vegetable collected from the two markets are shown in Table 2. The numbers of helminth eggs/larvae found on the roots of *C. sativum*, *M. arvensis*, and *E. foetidum* were 0 - 46, 0 - 18 and 0 - 27 per 100g wet weight, respectively. Among the three vegetable herbs, *C. sativum* showed significantly higher number of *S. stercoralis* larvae and *A. lumbricoides* eggs (P<0.05) were contaminated than the other two vegetable herbs, *M. arvensis* and *E. foetidum*.

Monthly density of soil-transmitted helminthes on selected vegetable herb samples

Monthly densities of soil-transmitted helminthes larvae/egg are summarized in Table 3. Among the collected vegetable herbs from Paukchaung market, the highest numbers of helminths eggs/larvae were found in August. The prevalence rate of helminthes was found to be the highest in the vegetable samples of *C. sativum* and the second was that of *E. foetidum*. Among the samples of *M. arvensis*, the incidence of *T. trichiura* and *E. vermicularis* were revealed to be low.

Table 1 Total mean number of helminth eggs/larvae recorded from study sites

Sampling place	Mean number of helminthes				
	Larvae	Eggs			
	S. stercoralis	*A. lumbricoides*	*Taenia sp.*	*T. trichiura*	*E. vermicularis*
Paukchaung market	372	196	97	81	64
Myothit market	536	288	205	140	61
Mean total	908*	484	302	221	125

*P<0.05

Table 2 Relative abundance helminth eggs/larvae per 100 g of samples

Parasite	Vegetable			*P*
	C. sativum	*M. arvensis*	*E. foetidum*	
S. stercoralis lavae	453	188	267	0.002
A. lumbricoides egg	226	142	116	0.098
Taenia sp.egg	98	93	111	0.659
T. trichiura egg	70	56	95	0.219
E. vermicularis egg	54	23	48	0.169

The incidence rate of helminths eggs/larvae in *C. sativum* collected from Myothit market was found to be higher than in *E. foetidum* and *M. arvensis*. The highest prevalence rate was recorded in August and the lowest prevalence rate was found in November (Table 3 and Figure 3).

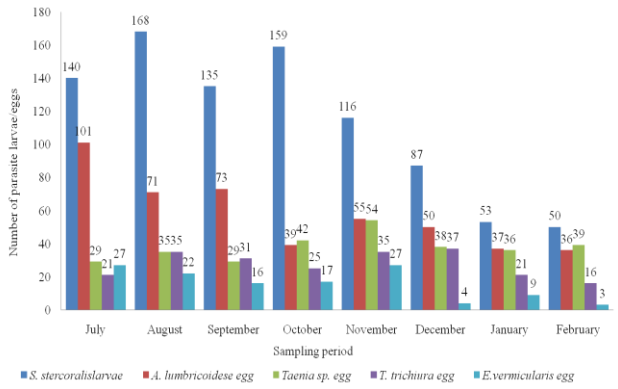

Figure 3. Monthly detected mean number of helminths eggs/larvae from vegetable herbs collected in Paukchaung and Myothit market

Taenia sp., *T. trichiura*., and *E. vermicularis*. Nearly all of the vegetable herb samples that collected and examined were contaminated with parasites.

The results of this study showed that *C. sativum* (Coriander) had the highest parasitic contamination (P<0.05) and *M. arvensis* (Podina) recorded as the least parasitic contamination; this indicated that the degree of contamination varies according to the nature of roots in vegetables. Since *C. sativum* (Coriander) and *E. foetidum* (Culantro or Shan-nannan) have many root hairs, parasitic eggs and larvae can easily attach to the roots of the vegetables during cultivation. On the other hand, *M. arvensis* (Podina) has the least prevalence. Therefore, it could be assumed that the quality of water used during plantation is important, since this herb prefers only clean environment during cultivation.

During study period, Myothit market recorded as a higher prevalence rate for helminthes compared with the vegetable herbs from Paukchaung market. The vegetable herbs sold in Myothit market are usually grown in urban agriculture sites in nearby villages of Myothit throughout the year. Most of urban agricultural farms obtained their water supply for plantation from a small irrigation canal (locally called Thae-chaung) running adjacent to these farms. Water flowing in this canal was also be used for carious purposes such as bathing, washing, drinking for live-stocked animals, and cleaning for domestic animals (pigs, cows and buffalos). Thus, the water from Thae-chaung could be contaminated with helminthes eggs and larvae from man as well as from other mammals. It was found that the numbers of helminths from Myothit market appeared higher than that of Paukchaung market. It is assumed that the water used in washing the vegetables might be contaminated because the environment surrounding the Myothit market was unhygienic with animals' excreta from domestic animals selling point which lies adjacent to the market.

Table 3 Monthly mean number of helminth eggs/larvae recorded from the vegetable roots of the two markets (P = Paukchaung; M = Myothit market)

Vetegable sample	Month	*S. stercoralis* lavae		Total	*A. lumbricoides* egg		Total	*Taenia sp.egg*		Total	*T. trichiura egg*		Total	*E. vermicularis egg*		Total
		P	M		P	M		P	M		P	M		P	M	
C. sativum	July	36	41	77	27	36	63	4	6	10	4	2	6	9	9	18
	August	43	46	89	28	28	56	3	5	8	5	3	8	4	5	9
	September	22	40	62	23	13	36	4	4	8	3	3	6	3	2	5
	October	34	41	75	5	6	11	9	5	14	5	2	7	2	4	6
	November	26	28	54	9	7	16	8	10	18	2	9	11	11	2	13
	December	13	32	45	8	10	18	5	8	13	3	12	15	0	1	1
	January	5	24	29	3	11	14	2	13	15	2	8	10	1	1	2
	February	3	19	22	4	8	12	2	10	12	2	5	7	1	0	1
M. arvensis	July	12	14	26	12	13	25	3	4	7	1	2	3	2	2	4
	August	14	15	29	8	9	17	4	5	9	2	2	4	1	2	3
	September	14	16	30	10	10	20	3	6	9	4	1	5	2	2	4
	October	19	15	34	5	7	12	5	6	11	1	1	2	0	3	3
	November	9	18	27	4	22	26	2	15	17	1	11	12	2	1	3
	December	9	12	21	5	12	17	3	10	13	2	14	16	0	0	0
	January	3	9	12	3	8	11	2	10	12	1	6	7	2	3	5
	February	2	7	9	1	13	14	1	14	15	1	6	7	0	1	1
E. foetidum	July	17	20	37	6	7	13	5	7	12	7	5	12	2	3	5
	August	23	27	50	10	10	20	7	11	18	10	13	23	5	5	10
	September	22	21	43	7	10	17	5	7	12	10	10	20	3	4	7
	October	25	25	50	5	11	16	10	7	17	8	8	16	6	3	9
	November	10	25	35	6	7	13	3	16	19	3	9	12	7	4	11
	December	4	17	21	3	12	15	4	8	12	3	3	6	2	1	3
	January	3	9	12	2	10	12	1	8	9	1	3	4	0	2	2
	February	4	15	19	2	8	10	2	10	12	0	2	2	0	1	1

The results of the present study indicated that vegetable herb samples of *C. sativum* (Coriander), *M. arvensis* (Podina) and *E. foetidum* (Culantro or Shan-nannan) collected from Paukchaung and Myothit markets were contaminated with five kinds of soil-transmitted helminths eggs/larvae, *S stercoralis*, *A. lumbricoides*,

The vegetable samples from Paukchaung market revealed as the less parasitic contamination; this might be due to the fact that the vegetables were washed by the water from Meiktila Lake before selling. It is assumed that there is minimal parasitic contamination of water in Meiktila Lake.

Among to the recorded parasites, the incidence rate of *S. stercoralis* larvae revealed to be the highest (908), and this is similar with the findings from Malaysia (Zeehaida et al., 2011). The highest number of *A. lumbricoides* eggs (484), was found to be contaminated parasites eggs in vegetable herbs and this finding is also similar to the reports from Ghana, Iran, Libya, Turkey, Egypt, Nigeria and Poland (Andoh et. al., 2009; Fallah et al., 2012; Abougrain et al., 2010; Avcioglu et al., 2011; Said, 2012; Adamu et al., 2012; Klapec and Borecka, 2012). The high prevalence of *S. stercoralis* may be accounted due to the life cycle of the parasite, where in reproduction of the rhabditiform larvae take place in the soil. Urban agriculture and the wet season keep the soil always sufficiently moist, so it is favorable for *S. stercoralis* larvae to direct optimally in soil. Moreover, prevalence of *S. stercoralis* infection in farmers might also high in this area.

In this study, higher incidence rates of helminthes eggs/larvae in vegetable samples were observed in wet season (July to September) and in cool season (November to February). Wet season enables them to survive better than the cool season, because of high moisture and humidity in the soil. *Ascaris* eggs are highly resistant to dryness and temperature fluctuation (Hunter et al., 1976), thus the eggs of *Ascaris* may remain as viable for a considerable period of time in the soil. The number of eggs of *E. vermicularis* and *T. trichiura* were found to be the least; it is appeared that their eggs are less resistant to the environmental stress. According to Rebecca et al. (2002), dogs can harbour one or more zoonotic species of gastrointestinal parasites and therefore the incidence of *Trichuris* eggs in the vegetable samples may also come from the dogs.

Aye Thant Zin (1984) and Thein Hlaing (1985) pointed out the majority of the helminthes cases admitted to hospitals were during the months of wet seasons. The results of the present study also revealed that the incidence of parasites was high during the wet season with a peak in August. During the cool months the incidence of parasites was found to be low.

The study site, Myothit market showed higher incidence rates than Paukchaung market, it may be due to the socio-economic factors and the poor sanitary conditions of the environment surrounding the Myothit market since the prevalence of STHs infections can be associated with type of soil, amount of rainfall, temperature, humidity, and other socio-behavioural variables (Belding, 1965).The irrigated water used during cultivation may be polluted with STHs because the domesticated animals, dogs and other mammals were passing to and fro in this irrigation canal. It may be one of the factors of infestation of vegetable samples. High soil contamination rate with parasitic helminths eggs in both the study sites in Meiktila may lead to various kinds of public health problems including the economic burden associated with STHs infections, so that urgent measures should be outlined to improve the environmental and sanitary conditions through a comprehensive community oriented health education programs.

Conclusions:-

The findings of this study is indicated that the importance of vegetable herbs in transmission of intestinal parasites. It is important to improve the sanitary conditions of these kinds of food at the plantation sites as well as personal hygiene of farmers in this study area.

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