

**Factors associated with intestinal parasite infections in a resettled indigenous community in Malaysia**

**ABSTRACT**

In Malaysia, the Orang Asli community (the indigenous people of Malaysia) are prone to intestinal parasite infections (IPIs) due to their living environment, socio-cultural, and personal hygiene practices. The prevalence and potential risk factors for infection among the indigenous community in Kg. Serendah in Malaysia was investigated. Stool samples were collected from 110 participating villagers who were then interviewed to obtain information on socio-demographic, behavioural, and environmental sanitation. The stool samples were processed and the parasites were identified by microscopy. To identify risk factors associated with intestinal parasite infection univariate and multivariate analysis were done. The level of significance was set at  $p < 0.5$  and for each statistically significant factor, odd ratios and confidence interval was computed. The prevalence for IPIs in the community was 67% and the intestinal parasites detected were *Trichuris trichiura* (50%), *Ascaris lumbricoides* (39%), *Blastocystis hominis* (10%), *Cryptosporidium parvum* (7.2%), *Taenia* spp (2.7%) and Microsporidia (2.7%). Forty five percent among them had a single infection while 46% had double infections with triple infections constituting 9.5%. Overall the prevalence of IPIs was slightly higher for females (68%) compared to males (66%). Higher IPIs prevalence was observed among children below 7 years compared to the adults. Univariate analysis showed that not using sanitary latrine, source of water supply and not boiling water before drinking and not using footwear were potential risk factors for intestinal parasite infection. However multivariate analysis indicated that not using the toilet and drinking unboiled water was predictive of intestinal parasite infection in this community.

**Key words:** intestinal parasites, prevalence, indigenous community, Malaysia, helminths

1. Intestinal parasite infections (IPIs) often affect the poor community living in tropics and sub tropics. They live in interior areas, remote places, urban slums and conflict zones. This disease is categorized as “Neglected Tropical Diseases” (NTDs) [1]. In Malaysia, IPIs are often related to the Orang Asli community; the indigenous minority people of Peninsular Malaysia who are among the most impoverished and marginalized community in Malaysia. Despite the government’s effort to upgrade the living standards of the Orang Asli, they still remain poor and face a high risk for intestinal parasitic infections, predominated by soil transmitted helminths. Although studies have shown IPIs among the Orang Asli have reduced

33 from 91.1% in 1978 to 64.1% in subsequent years, infection due to soil transmitted helminths  
34 (STHs) still remain high [2]. The Government has tried to resettle the Orang Asli near semi  
35 urban areas to bring them closer to the mainstream of development by providing free housing  
36 and other basic amenities. However cultural and social barriers among the Orang Asli have  
37 prevented them from going up the social ladder.

38

39 1.2. Most of the studies on IPIs among the Orang Asli have been conducted in remote Orang  
40 Asli villagers with poor infrastructure and transport facilities. It will be useful to understand  
41 the impact of the Government programme of resettling the Orang Asli near urban areas on the  
42 prevalence of IPIs among these groups of Orang Asli. Thus, this study was conducted in a  
43 resettled Orang Asli village at Kg.Asli Serendah, Selangor in order to determine the current  
44 prevalence of intestinal parasite infection and behavioural predictors associated with this  
45 infection.

46

## 47 **2. Materials and Methods**

48

### 49 **2.1. Study Area**

50 A cross-sectional study was conducted among the Orang Asli population in Kampung  
51 Serendah on March 2013. Kampung Serendah (101.62° E longitude, 3.37° N latitude) is  
52 located 6 kilometres from the Serendah town, in the State of Selangor .The village is situated  
53 on top of an undulating hill, surrounded by secondary forest. There is a metalled road going  
54 up the hill five kilometres from the main road to the village square .The government has  
55 provided free concrete houses and pour flush toilets. Electricity and water supply are  
56 connected to most of the houses. But many houses have not been able to pay the utility bills  
57 and except for a few houses treated water supply has been disconnected. Most of the villagers  
58 are dependent on a gravity feed system of untreated water for their daily use. The population

59 is about 250 people predominantly from the Temuan group living in about 50 residential  
60 dwellings. Most of the residents are employed as unskilled workers in nearby small and  
61 medium industries.

62

63 Permission to do the study was obtained from the local village head as well as the  
64 Department of Aboriginal Affairs of Malaysia. Ethical approval was obtained from the  
65 Research and Ethics committee of the International Medical University.

66

67 Several meetings were held with the village head to explain the rationale of the study.

68 Informed consent was obtained from the adults either through signature or thumbprint.

69 Stool samples were collected from the consenting villagers and those who contributed the  
70 stools were interviewed and data was collected on socioeconomic, environmental variables,  
71 and personal hygiene practices in a pretested questionnaire. The questionnaire was designed  
72 in Bahasa Malaysia which is a language well understood by the participants. For the children  
73 the questionnaire was completed by interviewing the parent who had signed the consent form.

74 Children below one year old were excluded from the study.

75

## 76 **2.2. Faecal sample collection and analysis**

77 All the stool samples were collected in sterile specimen containers attached with a scoop to  
78 aid in faecal collection. Each participant was given a specimen container containing 70%  
79 alcohol as preservatives (30mL). Each specimen container was marked with the family  
80 member's name, age, house number and date of collection. The stools samples were  
81 transported to the research laboratory at the International Medical University in an ice box.  
82 As soon as the sample arrived at the laboratory, the samples were removed and broken up  
83 with disposable wooden sticks for the preservation to be effective. If needed, more 70%

84 alcohol is added to achieve a ratio of 3:1. The specimen information was recorded and they  
85 were kept in an ice box at 4°C till further processing. All the collected stool samples were  
86 concentrated using formalin ethyl acetate technique [3,4] and samples on slides stained  
87 permanently with Trichrome stain. A person was considered positive for intestinal parasite  
88 infection if the stool showed the presence of at least one protozoan or helminthic species.  
89 Slides were observed under the light microscope using appropriate magnification.

90

91 Quantification of the parasite eggs were done on samples positive for helminth infections,  
92 using the McMaster slide [5]. Prior to quantification, 3 grams of stool were suspended in  
93 50ml of sugar/salt floatation solution (specific gravity: 1.28) and filtered using two layers of  
94 dampened gauze. The filtered solution was poured into a test tube and placed on a rack. The  
95 test tube was filled with floatation fluid, leaving a convex meniscus at the top of the tube. A  
96 coverslip was placed on the top of the test tube, and allowed to stand for 30 minutes. The  
97 coverslip was carefully lifted and immediately placed on the microscope slide for helminth  
98 observation. For quantification, 0.15ml aliquots from the filtered solution were loaded to  
99 each of the 2 chambers of a McMaster slide. The chamber was examined under 10x10  
100 magnification. The numbers of eggs within the grid was counted and multiplied by 50 giving  
101 eggs per gram of faeces (e.p.g.). The intensity of helminth infections were classified into  
102 mild, moderate and heavy infection based on WHO guidelines [6].

103

### 104 **2.3. Statistical analysis**

105 Data analysis was performed using SPSS version 20. Demographic, socioeconomic and  
106 behavioral characteristics were treated as categorical variables and presented as frequencies  
107 and percentages. Pearson's Chi-square ( $X^2$ ) test was used to test the association of IPIs  
108 prevalence with demographic socioeconomic and behavioral factors. Odds ratio (OR) and

95% CI were computed. To identify risk factors associated with IPIs, univariate analysis was used. All variables that were significantly associated with the prevalence of IPIs were included in the multivariate analysis by logistic regression to identify risk factors for IPIs.

### 3. Results

#### 3.1. Socio-demographic characteristics of the sample population

A total of 110 villagers participated in this study. Among them 50 were males (45.5%) and 60 were females (54.5%). With regards to age groups, children 6 years and below participated the most (50.9%), followed by children 7-12 years of age (20%) and the others(29%). More than half of the households reported to have monthly income below RM500 and the rest had income between RM500-RM700. Around 61.8% of the participants do not utilise toilets for defecation and the majority of the participants use untreated water from the gravity feed system (62.7%) for drinking and other domestic purposes. The survey also indicated that only 38.2% villagers boiled the water before drinking. Only 23.6% of the villagers used footwear when going out (Table 1).

**Table 1: Socio-demographic characteristics of the sample at Kg. Serendah**

| Variables          | N  | %    |
|--------------------|----|------|
| Gender             |    |      |
| Male               | 50 | 45.5 |
| Female             | 60 | 54.5 |
| Age                |    |      |
| 6 years and less   | 56 | 50.9 |
| 7-12 years         | 22 | 20.0 |
| 13-19 years        | 5  | 4.5  |
| 20-30years         | 15 | 13.6 |
| More than 30 years | 12 | 10.9 |

|                               |    |         |
|-------------------------------|----|---------|
| House hold income             |    |         |
| < RM500                       | 30 | 60      |
| ≥ RM500                       | 20 | 40      |
| Place of defaecation          |    |         |
| Pour flush toilets            | 42 | 38.2    |
| Others(Bush, rivers)          | 68 | 61.8    |
| Water Sources                 |    |         |
| Treated water                 | 41 | 37.3    |
| Untreated water(Gravity feed) | 69 | 62.7    |
| Drinking Boiled Water         |    |         |
| Yes                           | 42 | 38.2    |
| No                            | 68 | 61.8    |
| Use of Footwear               |    |         |
| Yes                           | 26 | 23.6    |
| No                            | 84 | 76.3    |
|                               |    | (N=110) |

### 3.2. Prevalence of intestinal parasitic Infections (IPIs)

A total 74 villagers were found to be positive for at least one of the IPIs giving a prevalence of 67.2%. Soil transmitted helminths predominated with 89% and protozoan spp contributed 18.1 percent. Among the parasites detected were *Trichuris trichiura* (50%), *Ascaris lumbrioides* (38.25%), *Blastocystis hominis* (9%), *Cryptosporidium parvum* (7.2%), *Taenia* spp (2.7%) and *Microsporidia* spp. (1.8%) (Table 2). The number of females positive for IPIs was 68% and males were 66%. Children 6 years and below recorded highest infection rate (69.6%), while 7-12 years had a prevalence of 68.2% (Table 3). More than 50% of the infected villagers had multiple infections. (Table 4)

**Table 2: Prevalence of IPIs by species among the Orang Asli at Kg. Serendah**

| IPI species | No | % |
|-------------|----|---|
|-------------|----|---|

|                           |    |      |
|---------------------------|----|------|
| <i>T. trichiura</i>       | 55 | 50   |
| <i>A. lumbricoides</i>    | 43 | 39.1 |
| <i>B.hominis</i>          | 10 | 9    |
| <i>C. parvum</i>          | 8  | 7.2  |
| <i>Microsporidiun spp</i> | 2  | 1.8  |
| <i>Taenia spp.</i>        | 3  | 2.7  |

(N=110)

**Table 3: Prevalence of IPIs by age groups among the Orang Asli in Kg Serendah**

| Age groups        | No of samples positive for IPI | Total samples in specified age group | Percent |
|-------------------|--------------------------------|--------------------------------------|---------|
| 6 years and below | 39                             | 56                                   | 69.6    |
| 7 – 12 years      | 15                             | 22                                   | 68.2    |
| 13-19 years       | 4                              | 5                                    | 80.0    |
| 20- 30 years      | 9                              | 15                                   | 60.0    |
| 31years and above | 7                              | 12                                   | 58.3    |
| Total             | 74                             | 110                                  | 67.2    |

As for the intensity of infection based on eggs per gram of faeces, the majority of those infected with *A.lumbricoides* were of moderate intensity (84%) while those infected with *T.trichiura* had moderate (42%) to heavy infections (42%)

Single and double infections were almost similar with 45% and 46% respectively. Triple infections constituted 7.3% Single infections were mainly due to STH, *A.lumbricoides* and *T. trichiura*. While most double infections were due to the combination of above two helminths.

148 With regards to double infections, the most common co-infection was seen between  
 149 *A.lumbricoides* and *T. trichiura* (35 %.)

150

151 **Table 4: Species of intestinal parasites seen in single and multiple infections among the**  
 152 **infected Orang Asli in Kg .Serendah**

| Type of infection                                                                        | Number | Percent |
|------------------------------------------------------------------------------------------|--------|---------|
| <b>Single infection</b>                                                                  |        |         |
| <i>Ascaris lumbricoides</i>                                                              | 10     | 13.5    |
| <i>Blastocystis hominis</i>                                                              | 4      | 5.4     |
| <i>Cryptosporidium parvum</i>                                                            | 2      | 2.7     |
| <i>Trichuris trichiura</i>                                                               | 17     | 23      |
| Total for single infection                                                               | 33     | 44.6    |
| <b>Double infections</b>                                                                 |        |         |
| <i>Ascaris lumbricoides</i> + <i>Trichuris trichiura</i>                                 | 26     | 35      |
| <i>Trichuris trichiura</i> + <i>Cryptosporidium parvum</i>                               | 2      | 2.7     |
| <i>Cryptosporidium parvum</i> + <i>Blastocystis hominis</i>                              | 1      | 1.4     |
| <i>Trichuris trichiura</i> + <i>Blastocystis hominis</i>                                 | 2      | 2.7     |
| <i>Ascaris lumbricoides</i> + <i>Blastocystis hominis</i>                                | 1      | 1.4     |
| <i>Microsporidium</i> + <i>Trichuris trichiura</i>                                       | 1      | 1.4     |
| <i>Microsporidium</i> + <i>Blastocystis hominis</i>                                      | 1      | 1.4     |
| Total for double infections                                                              | 34     | 45.9    |
| <b>Triple infections</b>                                                                 |        |         |
| <i>Ascaris lumbricoides</i> + <i>Trichuris trichiura</i> + <i>Blastocystis hominis</i>   | 2      | 2.7     |
| <i>Trichuris trichiura</i> + <i>Cryptosporidium parvum</i> + <i>Acsaris lumbricoides</i> | 2      | 2.7     |
| <i>Trichuris trichiura</i> + <i>Ascaris lumbricoides</i> + <i>Taenia spp</i>             | 2      | 2.7     |
| <i>Trichuris trichiura</i> + <i>Taenia spp</i> + <i>Cryptosporidium parvum</i>           | 1      | 1.4     |
| Total for triple infections                                                              | 7      | 9.5     |

|                                                   |             |            |
|---------------------------------------------------|-------------|------------|
| Total positive for intestinal parasite infections | <b>n=74</b> | <b>100</b> |
|---------------------------------------------------|-------------|------------|

### 3.3. Risk factors associated with IPIs infections

The risk factors associated with IPIs in relation to socio-demographic factors among the villagers were examined by univariate analysis. Four risk factors were identified which included, non-use of toilets (OR=4.24; 95% CI= 1.83-9.86;  $p=0.001$ ), using untreated water (OR=2.64; 95% CI=1.16-6.01;  $p<0.017$ ), not drinking boiled water (OR=2.96; 95% CI= 1.30-6.7;  $p=0.008$ ) and not using footwear (OR=3.28; 95% CI=1.32-8.2;  $p=0.008$ ). (Table 5)

However on logistic regression, only those not using toilet (OR= 3.13; 95% CI=1.23-7.95;  $p=0.016$ ) and not drinking boiled water (OR=2.73; 95% CI  $p<0.029$ ) are more likely to suffer from IPIs

Table 5: Univariate analysis of potential risk factors associated with intestinal parasites infection among the Orang Asli in Kg.Serendah

| Variables                    | N  | Intestinal parasite infection |         | Odds Ratio(CI)  | P value            |
|------------------------------|----|-------------------------------|---------|-----------------|--------------------|
| <b>Gender</b>                |    | no.+ve                        | Percent |                 |                    |
| Male                         | 50 | 33                            | 66      | 0.9,(0.45-2.0)  | 0.79               |
| Female                       | 60 | 41                            | 68.3    | 1               |                    |
| <b>Age(Years)</b>            |    |                               |         |                 |                    |
| 1-6                          | 56 | 39                            | 69.6    | 1.25(0.56-2.77) | 0.59               |
| >7                           | 54 | 35                            | 64.8    | 1               |                    |
| <b>Household income(MR)*</b> |    |                               |         |                 |                    |
| <500                         | 40 | 29                            | 72.5    | 0.88(0.15-5.03) | 0.88               |
| 500 and more                 | 8  | 6                             | 75      | 1               |                    |
| <b>Place of defaecation</b>  |    |                               |         |                 |                    |
| Others (Bush and Rivers)     | 68 | 54                            | 79.4    | 4.24(1.83-9.86) | 0.001 <sup>a</sup> |
| Pour flush toilets           | 42 | 20                            | 47.6    | 1               |                    |
| <b>Source of water</b>       |    |                               |         |                 |                    |

|                              |    |    |      |                 |                    |
|------------------------------|----|----|------|-----------------|--------------------|
| Gravity feed                 | 69 | 52 | 75.4 | 2.64(1.16-6.04) | 0.017              |
| Treated water                | 41 | 22 | 53.6 | 1               |                    |
| <b>Drinking boiled water</b> |    |    |      |                 |                    |
| No                           | 68 | 52 | 76.4 | 2.64(1.16-6.01) | 0.008 <sup>a</sup> |
| Yes                          | 42 | 22 | 52.4 | 1               |                    |
| <b>Use of footwear</b>       |    |    |      |                 |                    |
| No                           | 84 | 61 | 74.4 | 3.28(1.32-8.2)  | 0.008              |
| Yes                          | 26 | 12 | 46.2 |                 |                    |

(N=110)

N: Number examined, CI= (confidence interval); Significant association ( $p < 0.05$ )

\*Malaysian Ringgit.

<sup>a</sup> Confirmed as significant predictors by logistic regression.

#### 4. Discussion

The overall prevalence of IPIs' found in this study was 67.2%. This prevalence rate is in agreement with other local studies in Malaysia where the rates ranged from 70% to 80% [7, 8].

The prevalence rate of intestinal protozoan parasites is 18.1% with *Blastocystis hominis* predominating (9 %,) followed by *Cryptosporidium parvum* 7%. The results are similar to Ngui *et al.*, 2011 who found an overall prevalence of 21.4% for protozoan parasites. However the predominant protozoan in the above study was *Giardia duodenalis* while in the current study *B.hominis* was predominant.

The study findings showed that STH infections (89%) were much higher compared to protozoan infection (18.1%). The finding is similar to local studies carried out after socioeconomic development programmes for Orang Asli after 1987 [7, 8, 9, 10, and 11]. It was reported that before 1987, great diversity of protozoan infection was found among Orang Asli community. During the socioeconomic development, natural habitats were modified by

human intervention and some intestinal parasite species were eliminated due to their inability to adapt to the new environment. This resulted in the diversity of parasites found in the Orang Asli to decline naturally, but the STH survived the changes and continued to thrive [12]. Despite the socioeconomic development, the defecation habits among Orang Asli have remained the same and this contributes to soil contamination with STH eggs. Thus, the figures for STH still remains high.

Previous studies have shown that the predominant species contributing to STH infections were to *A. lumbricoides*, *T. trichiura* and hookworms [8]. However in this study, hookworms were not detected. This could be due to the non-loamy nature of soil in Kg. Serendah which is not suitable for development of hookworm larvae. Similar findings in Malaysia have reported low prevalence of hookworm infection among Orang Asli [8, 13].

Studies from other parts of the world showed that *A. lumbricoides* predominated all the three helminths [14, 15, and 16]. But in the current study *T. trichiura* showed the highest prevalence among STH infections. Some studies carried out in Malaysia had similar findings [14, 13, and 17]. Here, we found *T. trichiura* infection was between moderate to heavy infections. No light infections were found.

The resettlement of the indigenous community near urban areas did not have an impact on the incidence of intestinal parasitic infections in this community [18]. The provision of infrastructure alone is inadequate to reduce of IPIs .Social and cultural barriers relevant to hygienic practices need to be reduced by focused health educational strategies. The National Deworming Programme of schoolchildren which was discontinued in 1983, needs to be reintroduced in schools bordering resettlement villages. [19, 20, 21].

213

214 The limitation of this study was the small sample size due to the reluctance of the adults to  
215 provide stool samples as many feel it is a cultural taboo to give away one's waste.

216

## 217 **5. Conclusion**

218 This study shows that prevalence of IPIs; mainly due to STH is high among the Orang Asli  
219 community at Kg.Serendah, Malaysia. Furthermore, improvement of socioeconomic status,  
220 providing sanitation and giving health education will help to significantly reduce the  
221 prevalence and intensity of infection in this area. In conclusion, long term interventions will  
222 improve living conditions of this community and they will have a greater opportunity for a  
223 better future in aspects of health, nutrition and education.

224

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229

## 230 **Ethical Approval**

231 Permission to do the study was obtained from the local village head as well as the  
232 Department of Aboriginal Affairs of Malaysia. Ethical approval was obtained from the  
233 Research and Ethics committee of the International Medical University.

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