

An intestinal parasitological survey among the Jehai Orang Aslis (aborigines) of the Temenggor forest, Perak state, Malaysia

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IeJSME 2014 8(2): 18-23

Background: In Malaysia, the most common soil-transmitted helminth infections are *A. lumbricoides*, *T. trichiura* and hookworms. However, as there have been no extensive surveys on these infections, it is difficult to estimate with certainty the current overall incidence of infection with soil-transmitted helminths (STHs) among the Malaysian population including the Orang Aslis.

Materials and Methods: A study was conducted to determine the infection rate of soil-transmitted helminths and intestinal protozoa among the Jehai Orang Aslis (Aborigines). The study was conducted between December 2005 and August 2006, in four Jehai villages of Perak State, Malaysia. A total of 175 stool samples was collected and personal identification such as name, age, household identification, and date of collection were recorded on the spot during collection. Faecal smears were stained with Trichrome for protozoa cysts and trophozoites and the modified Ziehl-Neelsen acid-fast method for the oocyst of *Cryptosporidium* and *Isospora*. Wet mounts with tincture of iodine of both stool samples (10% formalin and PVA) were also examined to detect cysts, ova and larva of intestinal helminths.

Results: The prevalence rates of *Trichuris trichiura*, *Ascaris lumbricoides* and hookworm among the Jehai were 70.8%, 24.0%, and 10.9% respectively. The prevalence of *Entamoeba coli*, *Entamoeba histolytica*, *Giardia intestinalis*, *Blastocystis hominis*, and microsporidium was 40.6%, 33.7%, 25.7%, 91.4%, and 27.4% respectively. The difference in prevalence rates among the different age-groups and sex were found not significant. Children aged 0-9 years old had the highest prevalence rate of intestinal parasites and only 2 (1.1%) were free of any intestinal parasites.

Conclusion: Intestinal parasitic infections were therefore still common among these people. Children aged 0-9 years old were found to have the highest infection rate of all the intestinal parasites examined. Further investigations are needed to determine more specific transmission of these infections, so that an attempt to control these infections can be made.

Keywords: Soil-transmitted helminths and intestinal protozoa, intestinal parasitic infections, Orang Asli

Introduction

The Orang Aslis are indigenous minority peoples of Peninsular Malaysia, numbering 147,412 in 2003.¹ Currently, the Orang Asli are divided into four language groupings namely the Northern Aslian, the Central Aslian, and the Southern Aslian groups, all of whom speak Austroasiatic languages; a fourth group in the South of Peninsular Malaysia speak a Malay dialect belonging to the Austronesian group of languages.² This research was carried out on only one of the Northern Aslian group, the Jehai, who are also named Negritos based on their physical features. The Jehai live in the Belum and the Temenggor Forest that straddles Upper Perak and West Kelantan and until recently, were nomadic and lived by hunting-gathering.²

The World Health Organization (WHO) estimates that approximately two billion people worldwide are infected with the soil-transmitted nematode helminths, *Ascaris lumbricoides*, *Trichuris trichiura* and the hookworms *Necator americanus* and *Ancylostoma duodenale*, with 400 million of these infected being children of school age.³ Global numbers of *A. lumbricoides* infection have been estimated at about 1.5 billion cases.⁴ (Crompton, 2001). *T. trichiura* infection affects approximately 1,049 million people worldwide⁵ and an estimated 1.2 billion people are infected by hookworms.⁶ In Malaysia, the most common soil-transmitted helminth infections are *A. lumbricoides*, *T. trichiura* and hookworms.⁷ However, as there have been no extensive surveys on these infections, it is difficult to estimate with certainty the current overall incidence of infection with soil-transmitted helminths (STHs) among the Malaysian population.

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Intestinal protozoa have a worldwide distribution, with higher prevalence in developing countries, especially those with low socio-economy.⁸ *Entamoeba* species, *histolytica* and *dispar*, infect up to 10% of the world's population, with *E. histolytica* causing an estimated 50 million cases of amoebiasis and 40,000 to 100,000 deaths annually, placing it second only to malaria as a cause of death resulting from parasitic protozoa.⁹ *Giardia intestinalis* has been estimated to infect 200 million people worldwide.¹⁰ It infects approximately 2% of adults and 6 to 8% of children in developed countries worldwide and is currently responsible for the largest number of waterborne outbreaks of diarrhea in the United States.¹¹ *Blastocystis hominis* has a worldwide distribution, mainly in developing countries where the prevalence is higher (approximately 30 to 50%) than those observed in developed countries⁸ and has been described as probably the most common intestinal parasite in humans.¹² We carried out surveys among these aborigines whose villages were along Lake Temenggor to obtain baseline data on their health status and the associated potential environmental health impact. This paper documents the parasitological findings among these Jehai aborigines whose health status has seldom been studied.

Materials and Methods

Study areas and study population

The study was conducted between December 2005 and August 2006, in four Jehai villages of Perak State, Malaysia. One Jehai village located along Sungai Tekam river, had 86 people and the other three Jehai villages located along the lakeside of Lake Temenggor, had 183 people (Fig. 1). Lake Temenggor, the second largest lake in Peninsular Malaysia, created in 1977 for the construction of the Temenggor Dam to generate electricity, is situated in Hulu Perak near the Thai-Malaysia border at 5.51°N 101.36°E. All the villages have limited access to the nearest city,

which is about 50 km away by road after a boat journey of up to 50 minutes to the jetty near the main East West Highway. A complete list of members of each household was compiled through information provided by the head of each household. Several meetings were held between the researchers and the head man of each village to obtain permission for the study. Informed consent for the research was obtained through signature or thumb print of the head man of each village and the head of household.

Stool samples collection and examination

Two specimen containers with 10% formalin and PVA (Polyvinyl Alcohol) preservatives for each person were given to the head of each household. The heads of the household were asked to help the researcher to fill each container with one family member's stool and to specially mark each container for identification. Two or three days later the researcher returned to each house to collect the containers. Those who did not provide stool samples were asked to fill the containers later and the researcher would come to collect the containers during the next trip. Personal identification (name, age, household identification, and date of collection) were recorded on the spot during collection. Stool samples were transported to the Research Laboratory in the International Medical University, Kuala Lumpur within one to three days after the collection. A total of 175 (65.6%) stool samples was successfully obtained. No anthelmintic treatment had been given to the Jehai at least three months prior to this study, and this having been confirmed verbally with the head man of all the four Jehai villages.

Faecal smears were stained with Trichrome for protozoa cysts and trophozoites and the modified Ziehl-Neelsen acid-fast method for the oocyst of *Cryptosporidium* and *Isospora*. Wet mounts with tincture of iodine of both stool samples (10% formalin and PVA) were also examined to detect cysts, ova and larva of intestinal helminths.

Statistical analysis

SPSS for windows (Release 11.5) (SPSS 2001) and Microsoft Excel (Excel, 2002) were used for data entry and statistical analyses. Comparison between age groups, gender and race were performed using chi-square. Comparisons were also made of relationship between parasite infection and categorical variables. The level of statistical significance was set at $p < 0.05$.

Ethical clearance

Approval for the study was obtained from the Research and Ethics Committee, International Medical University, Kuala Lumpur, Malaysia and from all the heads of the four villages, Sungai Tekam, Tebang, Desa Ria, and Chuweh.

Results

Infection with soil-transmitted helminths

A total of 175 stool samples from 87 (49.7%) males and 88 (50.3%) females, aged 0.4-60 years, with the median age 15.14 + 15.09 years were obtained. The combined positive rate in all the 175 subjects for *T. trichiura*, *A. lumbricoides*, and hookworm were 70.8%, 24.0%, and 11.0% respectively. The positive rates for *T. trichiura*, *A. lumbricoides*, and hookworm in male were 35.4%, 12.0%, and 5.2% respectively. The corresponding rates for female were 35.4%, 12.0%, and 5.7% respectively. There was no statistical significant difference in positive rates between males and females for these parasites ($p > 0.05$). The positive rates for *T. trichiura*, *A. lumbricoides*, and hookworm by age groups are as shown in Table 1. The difference in percentage was not statistically significant, *T. trichiura* ($\chi^2 = 2.732$ and p -value = 0.604); *A. lumbricoides* ($\chi^2 = 4.748$ and p -value = 0.314); hookworm ($\chi^2 = 4.606$ and p -value = 0.330).

Table 1: The prevalence of intestinal helminths by age-groups, in all four Jehai villages.

Age group (years)	<i>Trichuris trichiura</i>	<i>Ascaris lumbricoides</i>	Hookworm
0 – 9	71 (40.6%)	25 (14.3%)	8 (4.6%)
10 – 19	19 (10.9%)	4 (2.3%)	2 (1.2%)
20 – 29	14 (8.0%)	7 (4.0%)	4 (2.3%)
30 – 39	12 (6.8%)	5 (2.8%)	2 (1.2%)
≥ 40	8 (4.6%)	1 (0.6%)	3 (1.7%)

Of the 175 stool samples examined, 44 (25.2%), 67 (38.2%), and 64 (36.6%) had none, single, and multiple helminth infections. Of those with mixed intestinal helminthic infections, mixed infection with *T. trichiura* and *A. lumbricoides* was 36 (20.6%), with *T. trichiura* and hookworm 15 (8.6%), *A. lumbricoides* and hookworm 8 (4.6%), and with all three helminths 5 (2.9%).

Intestinal protozoan infections

Among the 175 (87 males and 88 females) examined, the overall positive rates for *E. coli*, *E. histolytica*, *G. intestinalis*, *B. hominis*, and microsporidium were 40.6%, 33.7%, 25.7%, 91.4%, and 27.4% respectively. Of the 87 males, those positive for *E. coli*, *E. histolytica*, *G. intestinalis*, *B. hominis*, and microsporidium were 32 (18.3%), 29 (16.6%), 26 (14.9%), 76 (43.5%), and 22 (12.6%) respectively. The corresponding positive rates for the 88 females examined were 39 (22.3%), 30 (17.2%), 19 (10.9%), 84 (48.0%), and 26 (14.9%) respectively. There was no significant difference in the positive rates of these protozoan infections between gender ($p > 0.05$).

The positive rates for the various intestinal protozoan infections by age- groups in the four Jehai villages are as shown in Table 2. The highest rate was seen in the 0-9 years age group, with 21.2%, 18.3%, 18.9%, 50.9%,

and 16.7% for *E. coli*, *E. histolytica*, *G. intestinalis*, *B. hominis*, and microsporidium respectively. The difference in percentage positive for the different protozoan infections was not statistically significant ($p>0.05$).

Table 2: Positive rates of intestinal protozoa by age-group in the Jehai aborigine villages

Age-group (years)	<i>Entamoeba coli</i>	<i>Entamoeba histolytica</i>	<i>Giardia intestinalis</i>	<i>Blastocystis hominis</i>	Microsporidium
0 – 9	37 (21.2%)	32 (18.3%)	33 (18.9%)	89 (50.9%)	29 (16.7%)
10 – 19	11 (6.3%)	9 (5.1%)	3 (1.7%)	24 (13.8%)	6 (3.4%)
20 – 29	10 (5.7%)	7 (4.0%)	3 (1.7%)	19 (10.8%)	5 (2.8%)
30 – 39	7 (4.0%)	8 (4.6%)	3 (1.7%)	17 (9.7%)	6 (3.4%)
≥ 40	6 (3.4%)	3 (1.7%)	3 (1.7%)	11 (6.3%)	2 (1.1%)

Discussion

Soil-transmitted helminths results

The highest intestinal parasitic infection was with *T. trichiura* and this pattern which is similar in other studies has been attributed to the special mode of attachment of *T. trichiura* to the intestinal mucosa, the longer life span of this parasite, as well as its relative resistance to most anthelmintics.¹³ *A. lumbricoides* infection rate was second highest in this study and it has been postulated that the thick shells of the parasite eggs allow them to resist harsh environmental conditions, contributing to their relative abundance.¹⁴ On the other hand, the lowest infection rate was seen with hookworms which have thin-shelled eggs and larvae that are very sensitive to desiccation and cold temperatures.¹⁵ Others have demonstrated a strong negative association of hookworm infection with the clay content of the surface soil and a strong positive association with high vegetation and population density.¹⁶ Clay becomes sticky and plastic when wet and as hookworm larvae are obligatory aerobes, to survive they have to migrate through the soil in response to changes in temperatures

and desiccation.¹⁵ However, a more precise correlation between soil composition and infection rates is problematic because detailed information is lacking in most developing countries where hookworm is most prevalent.¹⁸ The present study area is hilly, and the soil is more clayey than sandy in composition. This may explain the relatively low positive rate of hookworm infection among the Jehai in the area.

The positive rates for *T. trichiura* and *A. lumbricoides* were the same between male and female; hookworm positive rate was slightly higher in female. However, there was no significant difference in the overall positive rate of intestinal helminths between male and female in this particular Orang Asli community, reflecting similar life-styles in both genders.

As expected, children aged 0-9 years old had the highest infection rate for all the intestinal helminths examined (*T. trichiura*, *A. lumbricoides* and hookworm). This was most likely due to common childhood behaviour such as poor personal habits like eating soil, not wearing shoes and not washing hands after defecation, thus increasing risks for infection with STHs (Soil transmitted helminths).¹⁷ Direct observation in the field showed that 100% Orang Asli children in this study ran around barefooted. Based on field surveys and interview with some Orang Asli adults, information on children's habits such as defecating at any convenient place, not washing their hands before eating food, and eating soil are common in the Orang Asli children. However, the specific number of individuals who had these habits was difficult to determine.

Intestinal protozoa infection

B. hominis infection was highest (91.4%). *B. hominis* transmission occurs by the fecal-oral route and a cystic stage has been described.¹⁸ The importance of zoonotic blastocystosis has not been defined; neither do we know whether human *Blastocystis* is a distinct species.¹⁸ Thus, the variables that influence the prevalence of the infection are still being defined.

The lowest infection rate of intestinal protozoa was *Giardia intestinalis* (25.7%). Interestingly, *G. intestinalis* which is mostly transmitted from contaminated water supplies¹⁹ had the lowest positive rate among the intestinal protozoan infections in these subjects who live along the riverside and the lakeside and where the source of drinking water was from the river and the lake. Further investigations are necessary, since the Jehai in this research study who have never been studied before genetically and immunologically, are most likely to be quite different from other indigenous people outside Malaysia. For a start, the Jehai are negrito type people with wooly hair, quite distinct from the Senoi tribes who have straighter hair. Genetic and immunological studies might give deeper understanding as to why they have low infection rates for *Giardia intestinalis* compared to other people despite their close proximity to large bodies of water.

The overall infection rates of *E. coli*, *E. histolytica*, *G. intestinalis*, *B. hominis*, and microsporidium in the Jehai showed no significant differences between males and females. The infection rate of intestinal parasites between male and female was found to be not significant. This is probably due to similarity in contributory human factors such as toilet behaviour, lack of awareness of environmental sanitation, poor knowledge of disease transmission, individual hygiene practices and contact with contaminated soil in both groups.

We found no significant differences on the prevalence of intestinal protozoa between the age groups. Similar living standards, environment, daily hygiene routines most likely contributed to these results. However, the Orang Asli children, aged 0-9 years old had the highest infection rate compared to other age groups. This is not surprising, because children in general are more likely to practise less hygienic life styles, for example, not washing hands before eating food and drinking unboiled water.

Conclusion

High prevalence of soil-transmitted helminths and intestinal protozoa were found in the Jehai aborigines

examined. We consider that poor sanitation and poor living conditions are factors related to the high infection rates. Children aged 0-9 years old were found to have the highest infection rate of all the intestinal parasites examined. Further investigations are needed to determine more specific transmission of these infections, so that an attempt to control these infections can be made, as these parasitic infections can contribute to the failure of growth among children in this subject population.

Acknowledgements

The authors would like to thank Ms Ling Hua Zen and Dr Lee Lai Kah for their kind assistance in this study. This study was funded by IMU Internal Grant No. IMU 101/2005.

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Fig. 1: Location of study village

