BEHAVIORAL AND HYGIENIC CHARACTERISTICS OF PRIMARY SCHOOLCHILDREN WHICH CAN BE MODIFIED TO REDUCE THE PREVALENCE OF GEOHELMINTH INFECTIONS: A STUDY IN CENTRAL JAVA, INDONESIA

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Abstract. Five schools in central Java that enroll more than 500 students in grades one through six were chosen for a study of the prevalence of parasitic geohelminths and selected protozoan infections. The schools are located in regions that differ in geological features, density of vegetation and cultural and economic attributes. The prevalence of soil-transmitted helminths among children in the five schools ranged from 8.7% to 76.1%, and protozoan infections from 2.8% to 32.1%. The principal objective of the study was to identify physical, hygienic and behavioral characteristics of the children that increase the likelihood of becoming infected. Although most of the characteristics studied are considered to be contributing factors, few previous attempts have been made to rank them in order of importance in causing infection. The results of this study suggest that a systematic and sustained effort to teach children to (a) avoid certain types of behavior that favor infection, and (b) practice good personal hygiene, are the best approaches to significant and enduring reduction of the scourge of intestinal parasitism.

INTRODUCTION

Soil-transmitted geohelminths together with parasitic intestinal protozoa are responsible for a vast amount of morbidity, discomfort and, often, mortality in tropical and sub-tropical regions around the world. No segment of the population is more severely affected than pre-adolescent children (with the possible exception of the elderly, about which little is known). In many countries and regions, infections with intestinal parasites have been considered a normal phase of growing up and little has been done to control or eliminate them. However, during the last 10-15 years the problem of parasitic intestinal helminths among children in less-developed regions of the world has attracted considerable attention. It is now clear that intestinal helminth infections in children of pre-school and primary school age (roughly, 4-12 years of age) can affect growth and development as well as cognitive abilities (Stephenson *et al,* 1993; Adams *et al,* 1994; Simeon *et al,* 1994; Hadju *et al,* 1996; Kvalsvig, 2003). Infected children (especially those with higher intensities of infection) are often predisposed to infections with other pathogens (Actor *et al,* 1993; Pearlman *et al,* 1993; Liwski *et al,* 2000) as well as to other disorders (Rocken *et al,* 1992; Agersberg *et al,* 2001).

As a consequence of the belated, but now intense, concern about “this wormy world” (Stoll, 1947), three courses of action are under consideration. The first, centers on the development of vaccines to prevent and/or limit geohelminth infections. Given the complexity of metazoan parasite infections, and the empirical approaches to vaccine development, it may be some time before useful vaccines for geohelminths appear. The second course of action is mass de-worming by use of anthelminthic drugs. Thanks to the urging and the efforts of the World Health Organization, that course of action is being implemented in several regions where helminths are highly endemic. There are five drugs that are

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considered safe and moderately to highly efficacious, dependending on the type of helminth being treated (Albonico, 2003; Horton, 2003). Other drugs are being developed, an activity spurred by the growing evidence of emerging drug resistant helminths, both in livestock and humans (Albonico, 2003; Horton, 2003). There are several problems and potential problems associated with the use of drugs in mass-treatment programs. Here only the problems of selection for drug-resistant helminths and the variable efficacy of individual drugs for the three most troublesome helminths, *Ascaris lumbricoides, Trichuris trichiura* and hookworms, need be mentioned.

The third course of action, the one with which this report is concerned, is the prevention of helminth infections through closer attention to sanitation, the instillation in children of the desire to avoid infection and the knowledge of how to do so. This report deals with a fairly comprehensive study of the prevalence of infection with *Ascaris, Trichuris* and hookworm, as well as with four protozoan parasites, among children enrolled in five selected primary schools (grades 1 through 6) located in the Provinces of Jawa Tengah and Yogyakarta, on the island of Java, Indonesia. Each child was interviewed, measured (height) and weighed, and asked to respond to a simple questionnaire designed to provide insight into her/his personal hygiene and behavior when playing or doing chores. In addition, the homes of many of the students were visited by members of the team who conducted the study and discussions held with many of the parents. The study of behavioral and hygienic characteristics provided results which were combined into a composite score for each student then compared with the prevalence of infection at each school. In this manner, it was possible to identify characteristics that predispose to infection. Although ours is not the first attempt to draw attention to the importance of teaching children to revise certain behavioral and hygienic habits as an approach to prevent helminth infections, there have been few such efforts. As we will show here and as others have shown (Siharath *et al,* 2000; Hohmann *et al,* 2001; Wachidanijah, 2002), it is an approach that merits serious consideration.

MATERIALS AND METHODS

Selection of schools to be studied

Although there have been surveys of the prevalence of helminthiasis at a number of locales in Indonesia (Hadju *et al,* 2003; Margono, 2003; Sasongko *et al,* 2003), the central region of Java has received relatively little attention. For the present study, five primary schools were selected on the basis of their location in different political districts and in different environments. A brief description of the environs of the five schools and the students' homes is provided in Table 1.

To begin the study, members of our team visited a number of primary schools in order to select schools to be involved. Several members of the team are Indonesian and are familiar with the language, cultures and geography of Java. The only advance criterion for selecting a school was that the school be involved in the Student Health Improvement Program (SHIP) of International Relief and Development (IRD). IRD is a non-governmental, not-for-profit organization devoted to humanitarian aid in many lesser-developed countries. One major activity of SHIP is the daily distribution of packages of fortified "snack noodles" to children in many primary schools in the Provinces of Jawa Tengah and Yogyakarta.

Once the schools were selected, further discussions were held with school and local health officials to secure their approval and cooperation. A statement was sent to the parents or guardian of each student to explain the study and provide assurance that participation in the study would be entirely voluntary.

Student interviews and questionnaires

The study embraced all children enrolled in grades one through six in each of the five schools. At the beginning of the study, each student was weighed and height measured. Each student was assigned an identification number. A brief, oral interview followed, for the purpose of ascertaining the student's attitude toward helminth infection and knowledge about how to avoid infection. At the same time, the children were observed for any overt signs of illness, such as pallor, severe rash, or distended abdomen.

Table 1
Some features of the environments of the five schools included in the study.

School code	District	Location	Approx. altitude (m)	Soil	Fresh water nearby	Sanitary facility (frequency)
A	Kebumen	Rural	1,000	Clay	River	Sparse
B	Kulon Progo	Rural	200	Sandy	River	Moderate
C	Bantul	Semi-urban	200	Coarse sand	Stream	Moderate
D	Gunung kidul	Semi-urban	1,000	Coarse, rocky	None	Common
E	Wonogiri	Rural	800	Clay, rocky	None	Sparse

Three sets of questionnaires were prepared: one for the teachers, one for the parents, and one for the students. The questionnaires for the teachers and parents were intended to reveal attitudes toward helminth infections and knowledge about the consequences of such infections and how to avoid them. The simpler questionnaire for students was designed to reveal the behavior patterns of students that would increase or reduce the risk of acquiring infections: *ie*, the students' awareness of "risk elements" (RE).

Collection and analysis of stool specimens

All children were provided with kits for collection of stool samples and carefully instructed in their use to avoid contamination of the samples. They were asked to bring the specimens to school on the morning they were obtained. Specimens were accepted each morning for several days until all the children had contributed. Each morning the specimens were assembled, carefully identified, and delivered to the laboratory (in a cold ice chest) of the Department of Medical Parasitology of Gadjah Mada University, located in Yogyakarta (Prof Dr Sugeng Juwono Mardihusodo, Head).

The examinations of stool samples were performed immediately by experienced personnel in the laboratory of the Department of Medical Parasitology under a contract with IRD. The color and texture of each sample were recorded. Helminth eggs were detected and quantified by a combination of standard wet mounts stained with iodine and the Kato-Katz thick smear. These analyses included *Ascaris lumbricoides, Trichuris trichiura* and hookworm (*Necator americanus* is the predominant form in Indonesia). In addition, four species of protozoa were included in the study: *Entamoeba histolytica, Entamoeba coli, Giardia lamblia*, and *Iodamoeba buetschlii*. Protozoan cysts were evaluated by Mallory's brine flotation technique.

Statistical analyses

The Student's *t* test and ANOVA were used for analysis and comparisons of prevalence and intensities of infection (IOI) (after log transformation of the data). Chi-square was employed for tests of association. Weight and height data were used to calculate weight-height ratios and body mass indices (BMI). Median and quartile values for the latter were compared with quartiles of US girls and boys of the same age (US National Center for Health Statistics).

RESULTS

Locations and environments of the schools included in the study

Table 1 provides an abbreviated characterization of the environments in which the schools and most of the homes of the corresponding students are situated. There was some variation in the types of soil. Vegetation was moderate in the vicinity of the schools and homes in all five districts. The most important variations, as will be seen, were the proximity of bodies of fresh water and the frequency and accessibility of latrines/toilets.

Prevalence and intensities of parasitic infections

Table 2 shows the prevalence of infection among children in the five schools. The frequency of infection with each helminth is shown separately. No distinction was made whether a given helminth occurred alone or with one or more

Table 2
Frequency of infection by intestinal helminths and/or intestinal protozoan parasites among children enrolled in five primary schools in Java.

School	No. of students	Frequency of infection (%)			
		Ascaris	*Trichuris*	Hookworm	Protozoa[a]
A	109	36[b]	73	6	35
B	116	35	61	21	14
C	108	1	6	8	3
D	92	0	3	4	7
E	92	8	19	19	7

[a]Frequencies of infections with *Entamoeba histolytica, Entamoeba coli, Giardia lamblia* and *Iodamoeba buetschlii* are combined in one overall number.
[b]Percent of all students, whether infected with one or more helminths.

other helminths. The data pertaining to the frequency of infection of the four intestinal protozoa were combined into one number. Several details were apparent. First, students enrolled in the schools in Districts A and B were more frequently infected with helminths and protozoa than were those in the other three schools. Infections with *T. trichuris* were much more prevalent than those with *A. lumbricoides* or hookworm. Approximately one-third of the students of school A (*ie*, in District A) were infected with one or more intestinal protozoa and about one-eighth of the students in school B were similarly infected. The children enrolled in schools C and D displayed surprisingly low prevalences of infection either with helminths or protozoa. There was, however, a significant frequency of infection with hookworm among the students in school C. The prevalence of infection among students in school E was intermediate for *Ascaris* and *Trichuris* but one of the two highest for hookworm.

The data obtained from assessment of helminth infections were analyzed in various ways. Analysis of the frequencies of infections among girls in different grades and boys in different grades in the two schools having the high prevalence (schools in Districts A and B) revealed no differences between genders or grades (ANOVA; p=0.14). Similarly, there were no significant differences between prevalence of infections of children in different classes or between boys and girls in the school in District E (p=0.51) (data not shown).

In areas where helminth infections are highly endemic, it is common to find a significant proportion of subjects who are infected with two or more of the parasites. That was true of the children in schools A, B and E, as shown in Table 3. In schools A and B, *Ascaris* and *Trichuris* were present concurrently in roughly one-third of the infected children. In all five schools, a small percentage of the infected students were hosts to both *Trichuris* and hookworm or to all three helminths. One finding of interest was the complete absence of concurrent infections with *Ascaris* and hookworm, while *Trichuris* and hookworm co-infections were relatively common. Concurrent infections with all three parasites occurred in a portion of children of schools A, B and E.

An analysis of intensitiy of infection (IOI *ie,* number of eggs of a given species of helminth per gram of feces) was performed. The egg counts per gram of feces from each infected student were recorded. The color and consistency of the specimens from all students were noted, infected as well as non-infected. The latter observations were uninformative because the specimens from more than 98% of all students were recorded as being "brown" and "firm." The IOI data, however, were informative. As expected, the egg counts in the case of all three helminths (*Ascaris, Trichuris* and hookworm) were not normally distributed. Therefore, all counts were transformed to logarithms (which were normally distributed) prior to statistical analysis. The most obvious feature of the data (not shown) is that the IOI in the case of all three helminths was substantially greater when any of the three occurred in combination with one or both of the others. For example, in schools A, B, and E the IOI of *Ascaris* in combination infections was approximately 3-fold, 9-fold and 13-fold greater, respectively, than in the case of single parasite infections.

Effects of helminth infections on student growth and development

In order to evaluate the effect of helminth infections on the growth and development of schoolchildren, the body weights and heights were measured. Weight (kg) and height (m or cm) data were analyzed separately as well as combined to compute weight/height ratios (W/H) and body mass index (kg/m^2) (BMI). One question to be answered was whether or not more severe infections (*ie*, multiple infections with two or three helminths) caused more pronounced effects on growth and development than did less-severe, single helminth infections. The weight and height data from the students in a given grade of all five schools were combined to form four separate groups: single-infected (mono-infected) girls and mono-infected boys; poly-infected (two or three helminths concurrently) girls and poly-infected boys. Comparison of weights or heights of mono- or poly-infected girls of a given grade/class revealed no statistically-significant differences. Similarly, there were no significant differences in weights and heights of boys of a given grade that resulted from mono- or poly-infections. Therefore, for further analyses, the data for girls of a given grade were combined into two groups: infected or non-infected. Likewise, the data for boys were combined into infected and non-infected groups. It should be noted that most of the data pertaining to infected children came from schools A and B, whereas schools C and D provided most of the data for non-infected children. This was legitimate because preliminary analyses revealed no statistically verifiable differences between non-infected children (either in weight or height) in schools A, B, and E *vs* C and D. Nor was there a significant difference between weights and heights of infected children based on comparing the same two groups of schools.

The W/H ratios were calculated for infected and non-infected girls at each grade/class level. The same ratios were calculated for boys. Graphs of those ratios relative to grade level (not shown) suggested that there was an effect of helminth infection on growth of the schoolchildren. To gain better insight into such an effect, the following analysis was performed using the data for students' heights. The data for such gender, and grade, and status of infection, were evaluated by linear regression analyses and generation of estimating equations. The lines obtained from regression plots representing data for non-infected and infected girls, and non-infected and infected boys, are presented in Fig 1 (A and B). Statistical evaluation (t test) of the differences between the regression coefficients of non-infected and infected girls and non-infected and infected boys revealed that for both genders helminth infections significantly retarded growth in height (in both cases, $p<0.001$).

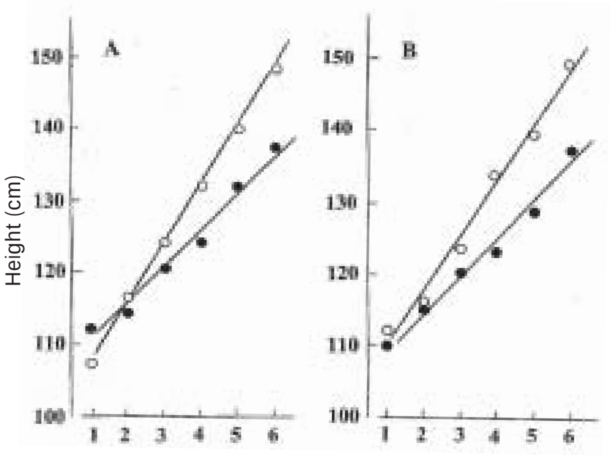

Fig 1–Regression lines of children's height (cm) on school grade (or childrens' ages). A: non-infected girls (○) (estimating equation, $Y = 8.21X + 99.5$); infected girls (●) (estimating equation, $Y = 4.91X + 106.5$). B: non-infected boys (○) (estimating equation, $Y = 7.63X + 101.6$); infected boys (●) (estimating equation, $Y = 5.3X + 103.4$).

Table 3
Numbers and proportions of infected students in the five schools and percentages who were concurrently infected with two or three helminths[a].

School district	Number of students		Percent of infected students harboring the following combinations			
	Infected[b]	Total	A+T[c]	A+H	T+H	A+T+H
A	83	109	36	0	2	7
B	86	116	29	0	10	9
C	17	108	0	0	12	0
D	8	92	0	0	13	0
E	30	92	3	0	17	10

[a]Two or three of: *A. lumbricoides, T. trichiura,* hookworm.
[b]Infected with one or more of the three helminths listed under footnote above.
[c]One-letter code: A = *Ascaris*, T = *Trichuris,* H = hookworm.

Table 4
Median BMI of non-infected students (girls and boys) enrolled in the five schools according to students' ages.

Statistic[a]	Median BMI (found) by student age					
	6	7	8	9	10	11
Girls						
Median	14.0	14.6	14.7	14.8	15.1	16.4
Upper CL	14.5	15.2	15.2	15.4	15.8	16.8
Lower CL	13.4	14.0	14.0	14.2	14.7	16.2
N	27	39	28	44	42	45
Boys						
Median	14.6	15.0	15.2	14.8	15.4	15.4
Upper CL	15.2	15.5	16.1	15.3	16.0	16.1
Lower CL	13.8	14.2	14.4	14.5	14.8	15.0
N	37	40	58	38	40	46

[a]CL = 95% confidence limit; N = number of samples (BMI values) in each rank.

Although the preceding analysis demonstrated that helminth infections retarded the growth of schoolchildren, it was possible that the infections merely added to an existing sub-optimum rate of growth attributable to other causes (*eg*, under-nutrition). That possibility was explored by analysis of body mass indices (BMIs) of non-infected students arranged by age/class and gender. The BMIs for girls and boys of each class were ranked separately and the medians, along with their 95% confidence limits were determined. The data are presented in Table 4. A slight upward trend in the median BMI with age is apparent both for girls and boys. The ranked BMI data were further analyzed by computing the quartiles of each rank. Those values were tabulated together with the quartiles of BMI data of US children of the same ages (Table 5). In that manner, it was possible to gain an impression of the status of the non-infected children who attended the five schools under study in comparison to US children. It is apparent that the children included in this study were significantly, but not severely, of less bulk than US children. The reason cannot be ascribed unequivocally to nutritional differences. We have not found data on height and weight of "adequately nourished" Indonesian children which might be used in a

Table 5
BMIs of non-infected students (girls and boys) enrolled in the five schools ranked by age (class); reported as quartiles and compared with US children of same ages.

Percentile	BMI by student age					
	6	7	8	9	10	11
Girls						
25	13.3	13.6	13.9	14.1	14.4	15.4
	(14.4)[a]	(14.5)	(14.7)	(15.1)	(15.5)	(16.0)
50	14.0	14.6	14.7	14.8	15.1	16.4
	(15.2)	(15.5)	(15.8)	(16.3)	(16.8)	(17.5)
75	14.5	15.2	15.2	15.6	16.0	17.2
	(16.3)	(16.7)	(17.3)	(17.9)	(18.2)	(19.5)
Boys						
25	13.2	14.1	14.2	14.2	14.5	14.5
	(14.6)[a]	(14.7)	(14.8)	(15.1)	(15.5)	(15.9)
50	14.6	15.0	15.2	14.8	15.4	15.4
	(15.4)	(15.5)	(15.7)	(16.2)	(16.6)	(17.2)
75	15.5	15.6	16.2	15.4	16.4	16.8
	(16.4)	(16.6)	(17.0)	(17.6)	(18.2)	(18.9)

[a]Numbers in parentheses are corresponding quartiles for adequately nourished US children (Source: US National Center for Health Statistics, 2000).

more-appropriate comparison (see Discussion).

Behavior and risk of acquiring helminth infection

The major reason for initiating this study was to identify aspects of childhood behavior that contribute to the risk of Indonesian children acquiring infections with *A. lumbricoides, T. trichiura* and hookworm. Therefore, every child was interviewed by a member of the research team and each was asked to complete a simple questionnaire concerned with behavior and personal hygiene. A listing of the questions that elicited the most informative and useful answers follows:

1. Where do you go to defecate at home? At school?
2. Do you wash hands after defecating?
3. Do you wash hands before eating? With soap?
4. Do you clean or cut your fingernails?
5. Do you wear sandals/shoes?
6. Do you sit on the ground when playing?
7. Where do you get water to drink?

The question about wearing shoes/sandals was asked twice: once orally and once on the written questionnaire. More than 10% of the students gave two different answers. We conclude that those students did not always wear shoes/sandals. The results of the interview and questionnaire were matched with the parasitological data for each student. The combined results enabled a search for behavioral characteristics that most likely placed a child at risk of becoming infected (those characteristics are termed "risk elements" or RE).

The most evident RE was the disregard for latrines. Table 6 reports that in schools A, B, and E, where the prevalence of infections with helminths was high (Table 3), between 30% and 85% (approximately) of the students did not routinely defecate in latrines. In sharp contrast, nearly all students in schools C and D routinely used latrines. Thus, there was a striking correlation between routine defecation in latrines and avoidance of helminth infections. It should be noted that most students of schools C and D reported that they had access to latrines at home, whereas many fewer in schools A and B, especially A, had latrines at home. What the students reported was, in many cases, verified by visits of members of the research team to the students' homes. It should be noted, also, that nearly all students reported that they never defecated at school. They gave various reasons for that. However, neither the condition nor the accessibility of the school latrine was given as one of the reasons.

From the list of questions above, there were five that provided the most useful responses. Questions 4 and 7 were not helpful because nearly all the students, infected or not, gave the same answers, as follows: mother or father cut their fingernails about once a week; and students brought water from home or purchased drinks at school. The remaining five questions elicited varying responses. Preliminary analyses indicated no clear correlation between any one of the five questions and the condition of infection or non-infection among the students. Therefore, we developed a rating system in which the response to each of the five questions was rated (+) or (-). A rating of five (+) was assigned a grade of "A". Four (+) and one (-) received a grade of "B". All other rating [>2(-)] were graded F. The grades ("behavior ratings") for all infected and non-infected students of each school were compiled; examples for school B and school D are presented in Table 7. As shown, there appeared to be a strong correlation between the behavior rating (grade) of "F" and the prevalence of helminth infection in both schools. To determine whether or not that apparent correlation was statistically reliable, the chi-square test was employed. Contingency tables were constructed to test the null hypothesis that the frequency of students having a behavior grade of F was unrelated to the prevalence of helminth infection. The data from each of the five schools were analyzed separately. The probability the null hypothesis was correct was found to be: for school B, $p<0.001$; for school D, $p<0.005$. Similarly, analyses of the results from the other schools gave the following probabilities: school A, $p<0.05$; school C, $p>0.1$; and school E, $p<0.01$. Only in the case of school C was the null hypothesis acceptable. In the other four schools, the prevalence of infection was strongly correlated with behavior ratings of "F".

Presumably, the data pertaining to student behavior are suitable for multivariate analysis in order to estimate which characteristics are the most important RE. We have not attempted to carry the analysis that far. In future studies, the questions posed to the students should be revised and sharpened so as to obtain even more useful responses. For example, question no. 6 was intended to gain some insight into the frequency of geophagia (soil eating) without asking a direct question that might be considered offensive. It seems unlikely, in retrospect, that we learned much about that probable RE.

Are some schoolchildren immune or innately resistant to helminths?

Composite behavior ratings (A, B or F) and the parasitological data were employed to estimate the frequency of non-infected students in schools A, B and E whose behavior grades were "F". There is the possibility that some students whose behavior characteristics place them at high risk for acquiring helminth infections, but who remain uninfected, are resistant to infection. They could be immune, as a result of having been exposed to some type of cross-reactive antigen; or they could be naturally/innately resistant. Table 8 shows the frequency of students in schools A, B and E who were not infected but whose grade of "F" for behavior suggested that they were at high risk for becoming infected. The data show that 6 of 14 non-infected students in school A, and 5 of 26 in school B, were at relatively high risk for acquiring helminth infections as a consequence of poor behavioral characteristics. Thus, in those two schools, approximately 5% of all the students might possess pre-determined resistance to infections with *Ascaris, Trichuris* or hookworm. The four-fold higher frequency of non-infected, "F"-rated students in school E suggests that the environment in which they live and attend school may provide exposure to some cross-reactive, immunogenic substances. Other explanations are possible.

DISCUSSION

The five schools that were selected for this study illustrate the wide range in prevalence of helminth infection in Java. In schools A and B,

Table 6
Percent of students in each school who routinely defecated in latrines or elsewhere.

School district	Percent of students (%)		
	Latrine	River	Elsewhere[a]
A	13	84	3
B	66	28	6
C	99	0	1
D	98	0	2
E	64	14	23

[a]Frequently in family gardens.

Table 7
Composite behavior rating ("grade") of infected and non-infected students of schools B and D[a].

School district	Students' status	Percent of students graded		
		"A"	"B"	"F"
B	Non-infected	23	54	23
	Infected	7	15	78
D	Non-infected	24	47	29
	Infected	0	17	83

[a]All non-infected students of all six classes combined; similarly, all infected students combined.

Table 8
Frequency (%) of students in schools A, B and E who were not infected but at high risk for acquiring helminth infection.

School	Students rated "F" for risky behavior	
	% of non-infected	% of total students
A	43 (6/14)[a]	5.5 (6/109)[b]
B	19.2 (5/26)	4.8 (5/105)
E	32.8 (19/58)	20.7 (19/92)

[a]Numbers in parentheses are numbers of non-infected students rated "F" divided by the total of non-infected students.
[b]Numbers in parentheses are numbers of non-infected students rated "F" divided by the total of students at that school.

76% and 74%, respectively, of the students were infected, whereas in schools C and D, approximately 16% and 9%, respectively, were infected. In school E, an intermediate number of about about 33% harbored intestinal helminths. In schools A and B, *Trichuris* was the most common helminth and *Ascaris* the second-most common. Hookworm infections were found in about one-fifth of the infected students in schools B and E. As expected, the frequency of children with multiple worm infections in a given school correlated with the overall prevalence of infection in that school. Thus, about one-third of the infected students in schools A and B were infected concurrently with *Ascaris* and *Trichuris*. Also, as expected, the intensity of infection (as judged by egg counts) with *Ascaris, Trichuris* or hookworm was substantially greater in students harboring concurrent infections. This result agrees with a previous report (Brooker *et al,* 2000) that children with two or more concurrent infections have higher egg counts for both helminths than do children singly infected. It has been reported (Booth *et al,* 1998) that children harboring two or more soil-transmitted helminths are at higher risk for morbidity than those infected singly.

As shown in Table 2, protozoan parasites were common among students in schools A and B. The two found most often were *G. lamblia* and *E. coli*.

In the effort to try to understand why the frequency of helminth infection varied so greatly between schools A and B, and C and D, attention was focused on two likely possibilities. First, the physical features of the local environments. The most distinctive feature of the environments of schools A and B was the proximity of the rivers and the sandy soil in District B. The flow of water in those rivers is intermittent. Running or dry, they are sites where many children go to defecate as well as to play. There are relatively few latrines in the homes where the children reside. The family garden is often the substitute latrine. School C is located near a large city. Neither the soil nor the small streams are attractive to the students. The children are likely to use latrines. School D is located in an area without fresh water streams and close to the coast. A characteristic of that community is the common availability of suitable latrines in the students' homes. School E is located in an inland, rural district. Although there is a large lake in the area, the homes where the children reside are not close. The soil is coarse, reflecting its ancient volcanic origin. Suitable latrines are not common in the homes of the students. Thus, the variations in characteristics of the schools and homes in the five Districts explain some of the marked variations in the prevalence of soil transmitted helminth infections.

A second likely possibility to explain the differences in frequency of helminth infections among children in the five schools is the variation in sanitary conditions as reflected by the availability and accessibility of suitable latrines (last column, Table 1). The correlation between the prevalences of helminth infections in schools A, B and E (Tables 2 and 3) and the failure of many students routinely to use latrines (Table 6) is striking. Similarly, the correlation between routine use of latrines by students in schools C and D and the very low prevalence of helminth infections is impressive. The same differential correlations are apparent in the prevalence of intestinal protozoan parasite infections (Table 2).

Obviously, if children have not become permeated with the importance of latrines and their hygienic significance, their toilet habits become part of a larger complex of behavioral characteristics. Certain of those characteristics, including random defecation, must be changed and the children imbued with the attitude and knowledge necessary to avoid intestinal parasite infection. Health education as an approach to preventing primary helminth infection and preventing re-infection following de-worming has been advocated by others (Siharath *et al,* 2000; Hohmann *et al,* 2001; Wachidanijah, 2002) and the limited studies to date have shown that it can be effective. For example, studies conducted in Lao PDR demonstrated that modification of personal hygiene, as a result of teaching how helminths enter the human body to residents of a small village, resulted in a substantial decrease in the rate of re-infection compared to the rate in a control village (Siharath *et al,* 2000; Hohmann *et al,* 2001).

Behavioral characteristics, in addition to failure to use latrines, have been proposed as RE affecting acquisition of helminths in a recent study of children attending school in Central Java (Wachidanijah, 2002). It was concluded that many children have a lackadaisical attitude toward behavioral traits that are, in actuality, RE. For example: defecating at random locations or in the river near their home, forgetting to wear sandals, or failing to wash hands after defecating or before eating. That study was conducted from a behavioral perspective. The findings led to a recommendation that improving the knowledge and attitude of parents (especially mothers) and children through health education would greatly facilitate the reduction of soil transmitted helminth (and other) infections in Central Java. The results reported here echo and extend that recommendation.

Because the correlations between individual behavioral characteristics and prevalence of infection in schools A, B and E were not highly significant, the data were combined to give a composite score (*ie*, behavior rating or grade). A poor composite rating of "F" was then found to correlate strongly with prevalence of infection among the children in schools A, B, and E. Among non-infected children, especially those in schools D and E, the majority of the composite grades were "A" or "B" with a low proportion of "Fs". In the case of school C, the frequency of "F" ratings did not correlate with the prevalence of infection. The reason for this apparent exception is uncertain. Perhaps it is the proximity of school C to a major urban center. Several studies have suggested that the prevalence of *Ascaris* and *Trichuris* infections may be extraordinarily high in urban slum areas (Margono, 2003). The eggs of helminths may be readily transported on urban waste from foci of infection to proximate areas.

Several previous studies have provided evidence that geohelminth infections retard the growth of young children (Crompton, 1986; Stephenson *et al,* 1993; Adams *et al,* 1994; Hadju *et al,* 1996; Hlaing *et al,* 2003). A gain in both height and weight following de-worming has been the principal observation. The enhanced growth has been accompanied by improved appetites and physical activity in the drug-treated children relative to controls. It has been concluded that helminth infection exacerbates malnutrition among undernourished children. Results obtained in the present study concur with previous findings that helminth infections retard growth and development of primary school students. However, no relationship between IOI or multi-species infections and the extent of retardation of growth was apparent; merely being infected was enough to affect growth. The interaction between undernutrition and geohelminth infection was not addressed directly in the present study. However, comparison of the BMI computations using the data from this study with the BMIs of US children of the same age suggested that the Indonesian children might be undernourished. What is not clear, however, is whether or not that is a valid comparison because there are a number of variable environmental conditions that may affect the two groups of children. Furthermore, there are significant genetically-determined differences in the physical characteristics of the two groups of children. A study of the effects of de-worming on the growth of children in the Philippines (Hlaing *et al,* 2003) included an estimate of malnutrition among those children. Height and weight data were compared with similar data of children of the same ages from the US National Center for Health Statistics leading to the conclusion that some 45% to 66% of the children were malnourished. Comparison of the data from the children involved in this study with those from the Philippine children suggests that the children of this study are somewhat better nourished. The difference could lie in the fact that all students of the five schools included in the present study receive a dietary supplement each day (5 days per week) consisting of a package of noodles prepared from a blend of wheat flour fortified with vitamins and minerals. It would appear to be important to study in considerable detail how effective the combination of adequate nutrition and attention to correcting/removing the RE associated with childhood behavior could be in preventing initial helminth infections and re-infection after anthelminthic treatment.

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REFERENCES

Actor JK, Shirai M, Kullberg MC, Buller RM, Sher A, Berzofsky JA. Helminth infection results in decreased virus-specific CD8+ cytotoxic T-cell and Th1 cytokine responses as well as delayed virus clearance. *Proc Natl Acad Sci USA* 1993; 90: 948-52.

Adams EJ, Stephenson LS, Latham MC, Kinoti SN. Physical activity and growth of Kenyan schoolchildren with hookworm, *Trichuris trichiura* and *Ascaris lumbricoides* infection are improved after treatment with albendazole. *J Nutr* 1994; 124: 1199-206.

Agersborg SS, Garza KM, Tung KS. Intestinal parasitism terminates self tolerance and enhances neonatal induction of autoimmune disease and memory. *Eur J Immunol* 2001; 31: 851-9.

Albonico M. Treatment of soil-transmitted helminth infection:prescribing information for disease control. In: Compton DWT, Montresor A, Nesheim MC, Savioli L, eds. Controlling disease due to helminth infections. Geneva: World Health Organization, 2003: 109-26.

Booth M, Bundy D, Albonico M, Chwaya H, Alawi K. Associations among multiple geohelminth species infections in schoolchildren from Pemba Island. *Parasitology* 1998; 116:85-93.

Brooker S, Miguel E, Moulin S, Luoba A, Bundy D, Kremer M. Epidemiology of single and multiple species of helminth infections among schoolchildren in Busia District, Kenya. *East Afr Med J* 2000; 77:157-61.

Crompton DWT. Nutritional aspects of infection. *Trans R Soc Trop Med Hyg* 1986; 80:697-705.

Hadju V, Stephenson LS, Abadi K, Mohammed HO, Browman DD, Parker RS. Improvements in appetite and growth in helminth-infected schoolboys three and seven weeks after a single dose of pyrantel pamoate. *Parasitology* 1996; 113: 497-

504.

Hadju V, Thaha AR, Agus Z, Jalal F. Effectiveness of deworming in schoolchildren through school feeding in Indonesia. In: Crompton DWT, Montresor A, Nesheim MC, Savioli L, eds. Controlling diseases due to helminth infections. Geneva: World Health Organization, 2003: 211-23.

Hlaing T, Khine WW, Than TT, *et al.* Impact of deworming on the growth of schoolchildren in Yangon. In: Crompton DWT, Montresor A, Nesheim MC, Savioli L, eds. Controlling disease due to helminth infections. Geneva: World Health Organization, 2003: 43-54.

Hohmann H, Panzer S, Phimpachan C, Southivong C, Schelp FP. Relationship of intestinal parasites to the environment and to behavioral factors in children in the Bolikhamxay Province of Lao PDR. *Southeast Asian J Trop Med Pub Health* 2001; 32: 4-13.

Horton J. The efficacy of anthelminthics: past, present, and future. In: Crompton DWT, Montresor A, Nesheim MC, Savioli L, eds. Controlling disease due to helminth infections. Geneva: World Health Organization, 2003: 143-55.

Kvalsvig JD. Parasites, nutrition, child development, and public policy. In: Crompton DWT, Montresor A, Nesheim MC, Savioli L, eds. Controlling disease due to helminth infections. Geneva: World Health Organization. 2003: 55-65.

Liwski R, Zhou J, McAlister V, Lee TD. Prolongation of allograft survival by *Nippostrongylus brasiliensis* is associated with decreased allospecific cytotoxic T lymphocyte activity and development of T cytotoxic cell type 2 cells. *Transplantation* 2000; 69: 1912-22.

Margono SS. Important human helminthiasis in Indonesia. In: Crompton DWT, Montresor A, Nesheim MC, Savioli L, eds. Controlling disease due to helminth infections. Geneva: World Health Organization. 2003:3-14.

Pearlman E, Kazura JW, Hazlett FE Jr, Boom WH. Modulation of murine cytokine responses to mycobacterial antigens by helminth-induced T helper 2 cell responses. *J Immunol* 1993; 151: 4857-64.

Rocken M, Urban JF, Shevach EM. Infection breaks T cell tolerance. *Nature* 1992; 359:79-82.

Sasongko A, Lubis F, Mahaswiati M, Ratnawulan D. Practical experience from the primary school-based soil-transmitted helminth control programme in Jakarta (1987-1999). In: Cromton DWT, Montresor A,Nesheim MC, Savioli L, eds. Controlling disease due to helminth infections. Geneva: World Health Organization, 2003: 225-38.

Siharath K, Soukphathag S, Tanyavong K, *et al*. Control of intestinal parasitic infection–a pilot project in Lao PDR. *Southeast Asian J Trop Med Public Health* 2000; 31 (suppl 2):41-50.

Simeon T, Callender J, Wong M, Grantham-McGregor S, Ramdath DD. School performance, nutritional status and trichuriasis. *Acta Pediatr* 1994; 83: 1188-93.

Stephenson LS, Latham MC, Adams EJ, Kinoti SN, Pertet A. Physical fitness, growth and appetite of Kenyan schoolboys with hookworm. *Trichuris trichiura* and *Ascaris lumbricoides* infections are improved four months after a single dose of albendazole. *J Nutr* 1993; 123: 1036-46.

Stoll NR. This wormy world. *J Parasitol* 1947; 33: 1-18.

Wachidanijah. Childrens' knowledge, attitude and behavior, and home and school environments relative to occurrence of worm disease infection in elementary school pupils. Yogyakarta, Indonesia: Gadjah Mada University, 2002. MS thesis.