

Prevalence and Reinfection of *Ascaris lumbricoides* and *Trichuris trichiura* among Elementary School Children in Rural Villages of Bali

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ABSTRACT

Background: A study was carried out on the prevalence of infection and reinfection of *A. lumbricoides* and *Trichuris trichiura* among elementary school children in Abiansemal Subdistrict, Bali.

Objective: To assess the prevalence of the two worm species and reinfection that occurred after treatment.

Method: The elementary schools of SD1 Taman, SD3 Mambal, and SD3 Sibang Kaja were selected as the study sites by random sampling. Faecal samples from school children of the three selected elementary schools were collected and examined by Kato-Katz thick smear technique. Interview was done to assess the children's habits and risk factors for reinfection. Data of prevalence were analyzed by chi-square test and the other data were analyzed descriptively.

Results: The study found an overall prevalence of intestinal worm infection of 72.8%, the highest was found at SD1 Taman (92.4%). The prevalence in male school children was higher (75.7%) than in female children (69.3%), but the difference was not statistically significant ($p > 0.05$). Infection intensity of *A. lumbricoides* was mostly (77.7%) light and that of *T. trichiura* were mostly (84.8%) very light. Treatment of ascariasis with Pyrantel 10 mg/kg BW in a single dose, and trichuriasis with Mebendazole 100 mg twice a day for three days, and of mixed infection of the two species with Pyrantel 125 mg and Mebendazole 100 mg in a single dose for three days gave an overall cure rate of 95.5%. Reinfection rates of *A. lumbricoides* at second and third months after treatment were 1.3% and 11.9%, respectively. Reinfection rates of *T. trichiura* at first, second, and third months after treatment were 4.7%, 7.6%, and 20.9%, respectively. School children whose families routinely covered their meals at home had a lower reinfection rate than those whose families did not cover their meals ($p < 0.05$).

Conclusions: The overall prevalence of intestinal helminth infection among elementary school children in the three rural villages surveyed was high. Reinfection rate of *Ascaris* and *Trichuris* occurred with the highest rates at three months after treatment.

Keywords: *A. lumbricoides*; *T. trichiura*; Prevalence; Risk factors; Reinfection

INTRODUCTION

Soil-Transmitted Helminths (STHs) are the most common cause of helminthic infections in humans. More than a quarter of the world's population is infected with one or more of these parasites. The highest infection prevalence of *Ascaris lumbricoides* and *Trichuris trichiura* is usually found in school-age children and that of hookworm in adults [1-3]. In Indonesia, the prevalence of STH infections especially *A. lumbricoides* and *T. trichiura* in school-age children in rural areas are still high hence imposes a significant public health problem [4,5]. A study done on school children at seven elementary schools in Badung district, Bali has shown 95.9% prevalence of *A. lumbricoides* and 60.5% prevalence of *T. trichiura* [6]. Mixed infections of two STH species were more prevalent than

single infections [7,8]. Another study done on school children of 26 elementary schools in Denpasar (the capital city of Bali) has found a prevalence of STH of 3-10% in the central area and 29%-33% in the periphery of the city [9].

The objective of our present study is to observe the infection prevalence of *A. lumbricoides* and *T. trichiura* and their reinfection rates after treatment among elementary school children in the Subdistrict of Abiansemal, Bali.

MATERIALS AND METHODS

Location, population, samples, and time of study

This study was carried out in the Subdistrict of Abiansemal, Badung

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District, Bali, Abiansemal is a rural sub-district of 69.01 km² in area (Figure 1). The nearest part of the Subdistrict is around 8 km north of Denpasar, the capital city of Bali Province, and the farthest part is around 30 km north of Denpasar. Its total population is 65,420 people who are mostly farmers [10]. Population of this study was all elementary schools located within the service areas of Health Center 1, Health Center 2, and Health Center 3 of Abiansemal Subdistrict. From each health center area, one elementary school was chosen as the study sample by simple random sampling technique. By this method, elementary schools of SD1 Taman, SD3 Mambal, and SD3 Sibang Kaja were selected as the study samples. The distance of the villages of Mambal is about 8 km, Sibang Kaja is 12 km and Taman is 15 km, respectively, north of Denpasar city, the capital of Bali Province. The total number of school children of the three selected elementary schools was 391 comprising 101 persons at SD1 Taman, 100 persons at SD3 Mambal, and 190 persons at SD3 Sibang Kaja. From the total 391 school children, 356 faecal samples could be collected and examined (91.05% coverage). Oral informed consent was obtained from each of the study samples. This study was carried out from March to June 2018.

Technique of examination

The 356 faecal samples collected from school children of the three selected elementary schools were examined by modified Kato-Katz thick smear technique to detect infection by finding worm eggs as well as to evaluate the intensity of infection, which was assessed based on the number of worm Eggs Per Gram of faeces (EPG), according to the criteria as follows: Intensity of *A. lumbricoides* infection: light (EPG<10,000), moderate (EPG 10,000-40,000) and heavy (EPG>40,000). Intensity of *T. trichiura*: very light (EPG<1000), light (EPG 1000-4000), moderate (EPG (>4000-10,000), and heavy (EPG>10,000) [11,12].

Treatment and reinfection rate assessment

Out of 356 school children who were found positive for worm eggs in their stools were subsequently treated. Those who had ascariasis were treated with pyrantel 10mg/kg body weight as a single dose; children who had trichuriasis were treated with mebendazole 100 mg twice a day for three consecutive days, and those with mixed infection of the two worms were treated with pyrantel tablet (125

mg single dose for one day) combined with mebendazole 1 tablet (100 mg) as a single dose for three consecutive days. Fourteen days after treatment, faecal samples were re-collected from the treated school children and reexamined. The treated school children who were found cured (no more worm eggs found in the stool) were followed-up by reexamining their stools each following month for three consecutive months, to see if reinfection with *A. lumbricoides* and *T. trichiura* occurred (indicated by finding of worm eggs of one or both of the two species). For *A. lumbricoides*, reinfection was assessed beginning at the second month after curing in accordance with the nature of its life cycle, so we must take into account that if *Ascaris* eggs are still found in the stool samples at the first month after cure, it should not be regarded as reinfection.

Interview on study subjects

An interview using a pre-designed questionnaire was done to all of the involved school children to study their knowledge about the two worms and their health-related habits such as the use of latrine for defecation, washing hands before eating, boiling drinking water, protecting or covering their prepared meal at home. Direct observation was done to evaluate the cleanliness of the children's fingernails.

Data analysis

The data relating to the prevalence of infection were analyzed statistically by chi-square test to assess any significant difference at $p<0.05$. Data related to reinfection and those obtained from interviews with the school children were analyzed descriptively.

RESULTS

Of 391 school children of the three selected elementary schools, 356 children submitted their faecal samples (91.05% coverage), consisted of 193 males and 163 females. Those who did not give their faecal samples were excluded from the study. Their reasons for not submitting faecal samples included embarrassment, not having defecated during the day of study, etc. Results of the assessment of prevalence rates according to elementary schools, gender, worm species, and infection intensity are shown in Tables 1-4. Examination of 356 faecal samples showed that 259 were

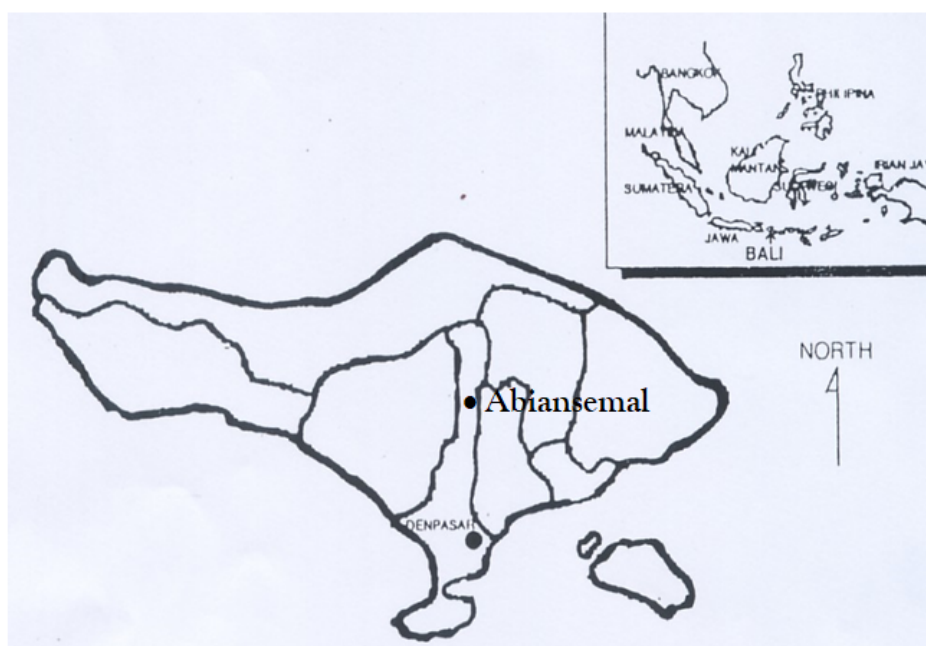


Figure 1: Map of Indonesia and Bali showing the approximate location of Abiansemal Subdistrict.

positive for worm eggs, giving an overall intestinal worm infection of 72.8% (Table 1).

The highest prevalence of infection was found among school children of SD1 Taman (92.4%), followed by school children of SD3 Mambal (80.9%), and school children of SD3 Sibang Kaja (58.3%) (Table 1). The prevalence rate of intestinal worm infection among male school children was 75.7%, which is higher than that in female children (69.3%), but the difference is not statistically significant (X^2 test, $p>0.05$) (Table 2).

Table 3 shows that the total prevalence of *T. trichiura* (88.8%) was higher than that of *A. lumbricoides* (60.6%), and in each of the three schools' prevalence of *T. trichiura* in the school children was consistently higher than *A. lumbricoides*. Hookworm infections were found only among school children of SD1 Taman, with a prevalence of 14.1%.

Table 4 shows that most (77.7%) of *A. lumbricoides* infection was the light intensity with an average Egg Per Gram of stool (EPG) of 2,107, and 7.6 % were heavy with an average EPG of 102,475. Most (84.8%) of *T. trichiura* infection was very light with an average EPG of 246. Infection intensity of hookworm and Enterobius was

Table 1: Prevalence of intestinal helminth infection in elementary schoolchildren in Abiansemal sub-district, by locations (Kato Katz thick smear technique).

| Location | No faecal samples examined | No positive | Prevalence % |
|-------------------------|----------------------------|-------------|--------------|
| SD1 Taman village | 92 | 85 | 92.40% |
| SD3 Mambal village | 89 | 72 | 80.90% |
| SD3 Sibang Kaja village | 175 | 102 | 58.30% |
| Total | 356 | 259 | 72.80% |

Table 2: The prevalence of intestinal helminth infection in elementary school children in Abiansemal sub-district, by gender (Kato-Katz thick smear technique).

| Gender | No of faecal samples examined | No positive | Prevalence % |
|--------|-------------------------------|-------------|--------------|
| Male | 193 | 146 | 75.70% |
| Female | 163 | 113 | 69.30% |
| Total | 356 | 259 | 72.80% |

Table 3: Prevalence of intestinal helminth infection in elementary school children in Abiansemal Subdistrict, by location and species (by Kato-Katz thick smear technique).

| Species | SD1 Taman (No infected=85) No + species (%) | SD3 Mambal (No infected=72) No + species (%) | SD3 Sibang Kaja (No infected=102) No + species (%) | Total (No infected=259) No + species (%) |
|---------|--|---|---|---|
| Al | 65/85 (76.5) | 40/72 (55.6) | 52/102 (51.0) | 157/259 (60.6) |
| Tt | 75/85 (88.2) | 68/72 (94.4) | 8/102 (8.3) | 230/259 (88.8) |
| Hw | 12/85 (14.1) | 0 | 0 | 12/259 (4.6) |
| Ev | 3/85 (3.5) | 0 | 1/102 (1.0) | 4 /259 (1.6) |

Al: *A. lumbricoides*; Tt: *T. trichiura*; Hw: hookworm; Ev: *E. vermicularis*

Table 4: Intensity of infection in 259 school children harboring Ascaris and Trichuris in Abiansemal Subdistrict, by Kato-Katz thick smear technique.

| Worm Species | Intensity | | | |
|------------------------|---------------------|----------------------|----------------------|----------------------|
| | Very light | Light | Moderate | Heavy |
| <i>A. lumbricoides</i> | - | 122 (77.7%) EPG=2107 | 23 (14.7%) EPG=20753 | 12 (7.6%) EPG=102475 |
| <i>T. trichiura</i> | 195 (84.8%) EPG=246 | 28 (12.2%) EPG=1883 | 6 (2.6%) EPG=5789 | 1 (0.4%) EPG=11180 |

Intensity of *A. lumbricoides* infection: light (EPG<10,000), moderate (EPG 10,000-40,000) and heavy (EPG>40,000). f *T.trichiura*: very light (EPG<1000), light (EPG 1000-4000), moderate (EPG (>4000-10,000) and heavy (EPG>10,000) [12].

not analyzed because the number of children infected was very small.

All of the 259 schoolchildren positive for worm eggs in their faecal samples were treated with anthelmintic drugs. Fourteen days after treatment, 246 faecal samples were recollected and examined by the same technique. Microscopic examination showed 236 of 246 faecal samples were negative for worm eggs, giving a cure rate: of 95.9%. Faecal samples of all the 236 cured school children were collected and reexamined by modified Kato-Katz smear technique each month for three consecutive months to see if reinfection had occurred by finding worm eggs in the stool. The results of the examination showed that at one month of follow-up reinfection rate of *T. trichiura* was 4.6% and no *A. lumbricoides* reinfection was noted. At the second month of observation, the *T. trichiura* reinfection rate became 7.6%, and *A. lumbricoides* reinfection rate was found to be 1.3%. At the third month of follow-up, reinfection rates of *A. lumbricoides* and *T. trichiura* increased to 10.9% and 20.9%, respectively.

Results of the interview done to 236 school children who were cured by anthelmintic treatment showed 76.0% and 98.1% had a low level of knowledge about *A. lumbricoides* and *T. trichiura*, respectively. Results of the interview related to main risk factors revealed that reinfection rates of *A. lumbricoides* and *T. trichiura* were evenly higher among school children who usually defecated outside latrine, did not wash hands before eating, had dirty fingernails, and did not boil their drinking water, but the differences were not statistically significant (X^2 test, $p>0.05$). However, there were significantly higher reinfection rates in school children whose families did not routinely cover their meal at home than in those whose families routinely covered their meal (X^2 test, $p<0.05$).

DISCUSSION

Three hundred fifty-six (356) faecal samples were collected from school children of three randomly selected elementary schools and examined by Kato-Katz thick smear method. Results of the examination found an overall prevalence of intestinal worms (*A. lumbricoides*, *T. trichiura*, hookworm, and *E. vermicularis*) infection of 72.8%. The highest prevalence was found in school children of SD1 Taman (92.4%), followed by SD3 Mambal (80.9%) and

SD3 Sibang Kaja (58.3%) (Table 1). SD1 in Taman village with the highest prevalence rate is located farthest from Denpasar, SD3 Sibang Kaja with the lowest prevalence rate is located nearest from Denpasar, and SD3 Mambal is located in between the two villages. A study done previously on 4858 faecal samples of school children of 24 elementary schools in Denpasar found a lower overall prevalence rate of 10.3% [9]. Another study done in 2003 on school children of five elementary schools in Punggul and Jagapati villages in Badung district found a lower prevalence rate in elementary schools in Jagapati village, which is located closer to Denpasar city than in Punggul village which is more distant from Denpasar [7]. Those lower prevalence rates may be associated with better hygiene -sanitation in the villages within the close vicinity of Denpasar city due to higher possession of latrine used for defecation in households. It may also be due to the provision of clean water (piped water) supply for drinking and sanitary use in the city and villages close to Denpasar as compared with the more remote villages.

The prevalence rate among male school children (75.7%) was higher than in females (69.5%), but this difference was statistically not significant (X^2 test, $p>0.05$) (Table 2). In a study carried out on elementary school children at Punggul and Jagapati villages, Bali, a similar result was found where the prevalence of intestinal worm infection in male elementary school children was higher (91.5%) than in female children (90.0%) [7]. Similar result was also found at Belok Sidan village, Badung district, Bali where the prevalence rate among male elementary school children was 80% and among female school children 77%, although the difference was not significant ($p>0.05$) [8]. In contrast, a study in China has found the prevalence of *A. lumbricoides* and *T. trichiura* infection was higher in females than in males [13]. Similar finding was also found in Iran where the prevalence of infection in females was significantly higher than in males [14].

The high prevalence of infection with *A. lumbricoides* and *T. trichiura* (Table 3) might be due to continuous infection transmission of the two species caused by persistent soil pollution with the worm eggs by infected people who defecate outside latrines because they do not have latrines at home. This may also relate to poor knowledge of the school children about intestinal worm infections, as shown by results of the interview that most (76.0% and 98.1%) of the school children had a low level of knowledge about *A. lumbricoides* and *T. trichiura*, respectively. Infection of hookworms (prevalence 14.1%) that was found only in the school children of SD1 Taman proved to be co-infection with *A. lumbricoides* and/or *T. trichiura*. Of the three villages, Taman is the most remote village from Denpasar with inadequate sanitation, as shown by the data from interview that 39.7% of the school children said they routinely defecated openly outside latrines.

Results of our present study showed that 45.2% of the intestinal worm infections were mixed infections with *A. lumbricoides* and *T. trichiura*. Another study done in 2003 on school children found 60% and 33.9% mixed infections with the two worm species at Punggul Jagapati and Belok Sidan village, respectively [7]. For the development of their eggs, the two worm species need the same moist and shaded soil condition, which is usually found on house yards where children often defecate and play with the earth [2]. A study in the province of Yang Tse, China has found that 62.6% of the populations were infected with one or more intestinal nematodes. *A. lumbricoides* and *T. trichiura* infections considered as reemerging diseases in this province were thought to be associated

with the use of untreated manure for fertilizing soil and accidental ingestion of *Ascaris* eggs due to people's habit of chewing sugar cane [13]. A somewhat similar situation was encountered in Iran where poor sanitation existed as a result of indiscriminate defecation and the use of human excreta as fertilizer for farming [14]. In Bali, however, there is no practice of using human excreta as fertilizer for farming, hence worm eggs pollution of the soil and water is solely associated with people's habit of indiscriminate defecation.

In our present study, we found the prevalence rate of *T. trichiura* was higher than *A. lumbricoides* among elementary school children studied. On interview many of them said that they had taken Combantrin tablet (an over-the-counter anthelmintic drug-containing pyrantel), given by their parents who had known the drug from TV commercials. As we know, pyrantel is effective for treating *Ascaris* but not as effective for Trichuriasis. Higher prevalence of *T. trichiura* than *A. lumbricoides* was also consistently found in some other studies done in Bali such as those at five elementary schools in Punggul and Jagapati villages, one elementary school in Penatih village, and 48 elementary schools in Denpasar municipality [7-9].

Regarding the intensity of infection, our present study found most (77.7%) of *A. lumbricoides* infection was light and 22.3% was moderate to heavy intensity. For *T. trichiura*, 97% was very light to light and 3% was moderate to heavy intensity (Table 4). Similarly, a study in China found that 77% of *Ascaris* infections and 94% of *Trichuris* were light in intensity [13]. In Punggul and Jagapati villages, Bali, it was also found that the overall intensity of worm infections was light, despite the prevalence rate being high [7]. A study in Pulau Panggang and Pramuka islands, offshore Jakarta, found that 69% of *A. lumbricoides* infection among school children of one elementary school was light in intensity, but among school children of two other elementary schools more severe infection intensity (moderate to heavy) was found, which was believed to be associated with poor personal and environmental sanitation [15]. Heavily infected persons (so-called "the wormy persons") may act as a source of environmental contamination with worm eggs in case they defecate indiscriminately.

STH infections with light intensity generally cause non-specific symptoms such as indigestion, nausea or vomiting, diarrhoea, abdominal discomfort, and loss of appetite [2,3]. In moderate to heavy STH infection, malnutrition may result in children, but even light infection can impair the growth of children who already have vulnerable nutrition status [16]. There is evidence that children chronically infected by STHs tend to become more vulnerable to other infectious diseases like tuberculosis, malaria, and HIV/AIDS infection [2]. One study in Jakarta has found that children with light to moderate STH infection intensity had mild to moderate malnutrition as compared to non-infected children [17]. A study in China has found that moderate to severe STH infection intensity can cause stunting in children [18]. Some studies have shown that severe *Trichuris* infection can cause growth impairment in children [2,16]. Prolonged and heavy STH infection can cause lowered cognitive capacity in children. In severe infection intensity of hookworm and occasionally *Trichuris* can cause iron-deficiency anaemia [3,16]. On occasions, *Ascaris* infection may cause serious complications that need urgent medical care, such as that due to erratic migration of the worm into the bile duct, appendix, Eustachian tube, nose, and intestinal obstruction or perforation [2,3]. In regard with the high prevalence in children and adults and the various bad impacts it may cause especially to children's

health, STH infection is considered as a public health problem in Indonesia in general and in Bali in particular [4,19].

Result of our present study shows that treatment of *Ascaris* with pyrantel and *Trichuris* with mebendazole and mixed infection of the two worms with pyrantel plus mebendazole as a single dose for three consecutive days had cured 95.5% of the infections. Mild side effects were encountered during treatment such as nausea, heartburn, and dizziness. A longitudinal study in Iran showed that a 2-year treatment program with the same drug regimens significantly decreased the prevalence rate of *Ascaris* from 53.3% to 6%, and 32% of *Ascaris* eggs expelled became unfertilized after treatment [14].

Reinfection of *A. lumbricoides* was assessed at the second month of observation, on the consideration that during treatment some *Ascaris* larvae might still be circulating in the pulmonary blood circulation which eventually will reach into the small intestine to become adults and produce eggs. Therefore, if *Ascaris* eggs were found in the faecal examination at the first month after cure, it should not be regarded as reinfection. This study showed at the second and third months of observation reinfection rates of *A. lumbricoides* were 1.3% and 11.9%, respectively. A study in 1990 at Jembatan Besi, Jakarta found that the reinfection rate of *A. lumbricoides* was 58.8% three months after treatment in individuals who got both treatment and health promotion, while the reinfection rate was 40% in those who got only treatment with no health promotion. Reinfection rates of *A. lumbricoides* and *T. trichiura* in those who had adequate knowledge about the two worm species did not differ significantly from the group who had inadequate knowledge about the two worm species. Health promotion could raise people's knowledge about worm infection but it did not have any influence on reinfection [20]. In Iran, a study has shown that children with parents having a higher level of education, especially of the mothers, have a lower infection rate of intestinal worms. This study has also found that there was a significant correlation between the number of siblings in the families and the increased rate of intestinal worm infection [21]. A study in rural Kwa Zulu Natal/South Africa showed those 16 weeks after cure, reinfection of hookworm was low (10%), reinfection of *A. lumbricoides* was moderate and reinfection of *T. trichiura* was low. A second treatment was done 6 months later and 18 weeks after treatment reinfection of hookworm was relatively high, reinfection of *A. lumbricoides* was moderate and that of *T. trichiura* was low. Differences of reinfection after the first and the second treatment may be due to slightly longer reinfection period and seasons (cool and dry during the first treatment and hot and humid summer during the second treatment) that influence the life cycles of the worms [22]. Reinfection with *A. lumbricoides* can occur when children play in a house yard densely contaminated with worm eggs by which they may accidentally ingest the embryonated eggs through their soiled hands. Reinfection of *Ascaris* and *Trichuris* after treatment is correlated with the level of infection intensity in children before treatment [23].

Considering the high prevalence of STH infection in elementary school children in the Abiansemal sub-district and several places in Bali with a portion of the children having heavy infection intensity, thus it is important to carry out prevention and control programs to reduce infection transmission. The recommended prevention and control strategies comprise drug treatment, health education, and sanitation improvement [4]. Regular drug treatment aims to reduce the prevalence and level of soil contamination with worm

eggs, especially in areas where sanitation and health facilities are inadequate. Health education aims mainly to reduce transmission of infection by promoting the school children's knowledge and health-related habits such as defecating in the latrine, washing hands before eating and after defecation, drinking only boiled or save water, always having clean fingernails, and covering or hygienic handling of food at home [2]. In our opinion, improvement of sanitation conditions particularly in rural villages and slum areas in the cities by providing latrines and water supply for households imposes the biggest challenge due to people's poverty and budget limitation on the part of the government, however, it is crucially worth implementing in the perspective towards having a healthy and bright future generation, free from helminth infection.

In conclusion, according to our present study, the overall prevalence rate of soil-transmitted helminths was high among elementary school children in the Abiansemal subdistrict, Badung district, Bali. In order to reduce or eliminate soil-transmitted helminth infection among elementary school children, it is recommended that a preventive and control program be implemented by means of regular anthelmintic treatment, health education, and improvement of hygiene-sanitation.

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ETHICAL CLEARANCE

Ethical clearance approval was obtained before the implementation of the study from the Ethical Clearance Committee of the Faculty of Medicine and Health Sciences, Warmadewa University.

CONFLICT OF INTEREST

We declare that no conflict of interest has occurred related to this study.

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