ORIGINAL RESEARCH ARTICLE

Detection of Blastocystis hominis and Assessment of Personal Hygiene Practice in Elementary School Children in Kampa District, Kampar Regency, Riau, Indonesia

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DOI: 10.55489/njcm.160420254933

ABSTRACT

Background: *Blastocystis* is an opportunistic intestinal protozoan and the pathogenicity of the disease is still unclear. It can cause malnutrition and disrupt growth and development in children. This study aims to detect *Blastocystis* and determine characteristics and personal hygiene practices.

Methodology: A cross-sectional study, with a sample of 104 students from 8 elementary schools in Kampa District. Detection of *Blastocystis* using stool culture with Jones Medium. Characteristics of subjects and risk factors using a questionnaire through guided interviews.

Results: The prevalence of *Blastocystis* was 45.2% and all of them had no symptoms. Most of them had unclean nails (70.2%), did not wash their hands with soap after defecating (57.7%). All students had good behavior in defecating. Generally, students drink from refillable water purchased at drinking water depots, 53.8% of students keep and have contact with pets. There was no significant relationship between personal hygiene and *Blastocystis* (p>0.05).

Conclusions: The prevalence of *Blastocystis* is quite high, although no significant relationship was found in this study, teachers and parents still need to pay attention to students' personal hygiene, such as keeping their nails clean and washing their hands with soap after defecating.

Keywords: *Blastocystis,* children, feces, elementary school

ARTICLE INFO

Financial Support: None declared

Conflict of Interest: The authors have declared that no conflict of interests exists.

Received: 29-11-2024, **Accepted**: 08-03-2025, **Published**: 01-04-2025

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How to cite this article: Maryanti E, Wardany Y, Mislindawati, Fadhlie LA, Balqis AA, Rahmayeni A. Detection of Blastocystis hominis and Assessment of Personal Hygiene Practice in Elementary School Children in Kampa District, Kampar Regency, Riau, Indonesia. Natl J Community Med 2025;16(4):388-394. DOI: 10.55489/njcm.160420254933

www.njcmindia.com | pISSN: 0976-3325 | eISSN: 2229-6816 | Published by Medsci Publications

Introduction

Blastocystis sp is a single-celled protozoan that can infect the gastrointestinal tract of animals and humans. This protozoon has a non-specific host, spreads very widely and infects humans in all age groups with unclear clinical symptoms and its pathogenicity is still controversial.[1,2] *Blastocystis hominis* can be detected in the feces of healthy people, meaning it has no symptoms or asymptomatic and patients with varying symptoms, such as diarrhea, abdominal pain, flatulence, nausea, vomiting, anal itching and other gastrointestinal symptoms. There are four morphologies of *Blastocystis hominis*; granular, vacuolar, amoeboid and cyst forms. Vacuolar is the form that is often found on microscopic examination, while the amoeboid form is the form that causes damage to the host.[2–4]

The prevalence is very widespread in the world, ranging from 0.08% to 90%, this depends on the level of economic development of the region or country. Developing countries have a high prevalence due to low socio-economic factors, poor sanitation and inadequate personal hygiene, and community culture in an area that supports the transmission of *Blastocystis hominis*. Some studies state that the prevalence in industrialized countries is around 5% and in developing countries 30 – 60%.[4,5] Even in Asia and Africa the prevalence of *Blastocystis hominis* infection ranges from 37-100%.[6] Transmission of this parasite is mainly via fecal-oral route but The main source of transmission is still unclear,[5] contact with pets can also cause infection with this parasite, such as dogs and cats.[7] All age groups can be infected by this protozoa starting from children to the elderly. Based on research in Jakarta, 41% of *Blastocystis hominis* infections were reported in school-aged children.[8] *Blastocystis hominis* is an opportunistic intestinal protozoan whose infection depends on the patient's immune system. It is reported that chronic diarrhea often occurs in children under 5 years and in people with irritable bowel syndrome.[8,9]

There are currently 32 subtypes of *Blastocystis hominis*,[10] and there are some ways to detect it; microscopically, culture and molecular tests.[7] Direct microscopic examination requires special skills because *Blastocystis hominis* resembles other microorganisms in feces so it is very difficult to differentiate and only trained personnel can detect it.[8] Culture examination is the gold standard examination used for the detection of *Blastocystis hominis*, because this examination can directly detect the parasite. Molecular examination is an examination that has a high sensitivity value and with this examination subtypes of *Blastocystis hominis* can be detected, but this examination is expensive.[8,9] This study aims to detect *Blastocystis* in the feces of elementary school children and determine the characteristics and personal hygiene practices of students who are detected with *Blastocystis hominis*.

Methodology

This research was a cross-sectional descriptive study. The subjects were elementary school children in Kampa District, Kampar Regency, Riau Province. The elementary school that was used as the subject was a school on the riverside area and had poor hygiene in the surrounding area. The study was conducted from July to October 2024, fecal examination was carried out at the Parasitology Laboratory, Faculty of Medicine, Universitas Riau. This study has been declared to have passed ethical review by the Ethical Review Board for Medicine and Health Research, Faculty of Medicine Universitas Riau No: B/ 098/ UN19.5.1.1.8/ UEPKK/ 2024.

The total number of students in the public elementary schools located along the riverbanks in Kampa District (8 schools) was 593. The inclusion criteria were elementary school students in Kampa District who attended these 8 schools, resided in Kampa District, were present during data collection, obtained parental and teacher consent to participate in the study, and were willing to collect their feces in the provided stool containers. The exclusion criteria were students who did not return the stool containers and those whose stool samples were inadequate. The sample size was calculated using the single proportion formula: $n = (Z^2 \times P \times Q) / d^2$. Based on this formula, the minimum required sample size was 96. A total of 200 study subjects were randomly selected, of whom 183 students were present. Among them, only 122 students submitted stool samples, and only 104 provided adequate samples for examination.

Every child who is a subject and has received informed consent from their parents will be explained the research and distributed stool container. Apart from that, a questionnaire form was also provided which was filled in with a guided interview. The questionnaire questions were based on the PHBS questionnaire issued by the Ministry of Social Affairs of the Republic of Indonesia[11] and were also adapted from study of Sari et al.[12] Fill in the questionnaire in the form of the subject identity which includes name, gender, age, grade, and personal hygiene practice in the form of a) nail cleanliness: good if their nails are short and not dirty, poor: if the nails long or short nails that are not clean. b) The habit of washing hands before eating and after defecating: good is if the student washes their hands with soap, and it is said to be poor if they don't wash their hands or wash their hands without soap. c) place to defecate, good if they defecate in the toilet, poor if they defecate other than the toilet. d) Source of drinking water, whether it is bottled water/refillable water/boiled well water. e) Keeping or not keeping pets. After completing the questionnaire, it is followed by weighing and measuring height to determine nutritional status. Nutritional status was assessed using the AnthroPlus application from WHO. Nutritional status is divided into 2; normal/overweight and underweight. Body weight measurement was conduct-

ed by weighing the students using a digital scale, recorded in kilograms (kg). The scale was placed on a flat surface, and students were required to remove their footwear and wear minimal clothing, without jackets or thick clothes. Height measurement was performed using a microtoise attached to a wall. The students stood with their heels and back against the wall, keeping their head straight while looking forward, and removing their footwear. Height was recorded in centimeters (cm). The recorded weight, height, date of birth, and measurement date were then entered into the WHO AnthroPlus application. The nutritional status classification: normal (-2 to +1 SD), overweight (>+1 SD), or underweight (<-2 SD). After that, the subjects will be given a stool container. The stool container will be collected the next day and then a stool examination will be carried out at the FK UNRI Parasitology Laboratory. The examination carried out was a direct microscopic examination and a culture examination of *Blastocystis hominis* using Jones Media[13] with sheep serum. The culture results were examined with a microscope after 48 hours and 72 hours. The data were processed with the SPSS program and then served on frequency distribution tables. Bivariate analysis was done using the Chi-square test with a significance level of 5% (p =0.05).

RESULTS

In this study, 104 children were willing to be subjects from 8 elementary schools in Kampa District, Kampar Regency. The characteristics of the research subjects and the results of the *Blastocystis hominis* culture examination can be seen in Table 1 below.

In Table 1, it can be seen that the incidence of positive *Blastocystis* was found in 47 elementary school students (45.2%) and 55% were found in girls. The age group 9 years – 12 years and grades 4 – 6 had more *Blastocystis* detected, whereas based on nutritional status it was found that the proportion of underweight and normal/overweight was not much different in positive *Blastocystis*. In table 1, it can also be seen that there is no significant relationship between gender, age group, class and nutritional status of students, because all p values are > 0.05.

Table 1. Distribution of Blastocystis based on characteristics of the subject

Characteristics	*Blastocystis*		Total (%)	p value
	Positive (%)	Negative (%)		
Total cases	47 (45.2)	57 (54.8)	104	
Gender				
Boys	21 (40.4)	31 (59.6)	52	0.431
Girls	26 (50)	26 (50)	52	
Age Groups				
6 yrs - 8 yrs	11 (42.3)	15 (57.7)	26	0.909
9 yrs - 12 yrs	36 (46.2)	42 (53.8)	78	
Level of Education/ Grade				
1 – 3 grade	14 (43.8)	18 (56.2)	32	1.000
4 – 6 grade	33 (45.8)	39 (54.2)	72	
Nutritional Status				
Underweight	15 (48.4)	16 (51.6)	31	0.833
Normal/ Overweight	32 (43.8)	41 (56.2)	73	

Table 2: Distribution of *Blastocystis* Based on Personal Hygiene Aspects

Personal Hygiene	*Blastocystis*		Total	p value	Odds ratio	95% CI
	Positive (%)	Negative (%)				
Nail Cleanliness						
Poor	33 (45.2)	40 (54.8)	73	1.000	0.998	0.429 - 2.322
Good	14 (45.2)	17 (54.8)	31		Ref	
Handwashing before meal						
Poor	14 (35)	26 (65)	40	0.147	1.977	0.876 - 4.461
Good	33 (51.6)	31 (48.4)	64		Ref	
Handwashing after defecations						
Poor	22 (36.7)	38 (63.3)	60	0.066	2.273	1.027 - 5.031
Good	25 (56.8)	19 (43.2)	44		Ref	
Place to defecate						
Good (toilet)	47 (45.2)	57 (54.8)	104		-	
Poor	0	0	0			
Source of drinking water						
Refill Water	44 (45.4)	53 (54.6)	97	1.000*	0.903	0.192 - 4.254
Boiled water	3 (42.9)	4 (57.1)	7		Ref	
Keeping pets						
Yes	27 (48.2)	29 (51.8)	56	0.637	1.303	0.599 - 2.835
No	20 (41.7)	28 (58.3)	48		Ref	

*Fisher's exact test

The personal hygiene aspect for *Blastocystis* can be seen in table 2. In Table 2, there are 70.2% of students who have unclean nails, 63.4% of students wash their hands with soap before eating, and 57.7% of students do not wash their hands with soap after defecating. All students have good grades in defecating in the toilet. For drinking water sources, 96% of students drink from refillable water purchased at drinking water depots. For pets, 53.8% of students keep pets and have contact with these pets, such as cats, chickens, birds, goats and dogs. In Table 2 it can also be seen that there is no significant relationship between risk factors and the discovery of *Blastocystis* in students' feces because p>0.05. Odds ratio (OR) values >1 was observed for three personal hygiene habits: handwashing before meals, handwashing after defecation, and keeping pets. This suggests that poor handwashing habits before meals, poor handwashing after defecation, and keeping pets may increase the risk of *B. hominis infection.* However, based on the p-values (>0.05) and confidence intervals (CI), these three variables, similar to the other variables, were not statistically significant for *B. hominis* infection in this study.

Discussion

This research was conducted at 8 elementary schools located on the riverside in Kampa District, Kampar Regency, Riau Province. In this study, it was found that the incidence of positive *Blastocystis hominis* in the feces of elementary school students was 45.2%. This incidence rate is lower than expected because the school area is an area on the edge of a river with poor hygiene in the surrounding area. The results of this study are not much different from research reported in Jakarta in 2017 which found that the prevalence of *Blastocystis hominis* in primary school children was 41.1%.[8] These results are different from research conducted at the orphanage in the Pekanbaru, the results showed that 66.3% of children in 9 orphanages were positive for *Blastocystis hominis*.[14] This incidence rate is quite high because the sample was children aged 5 to 18 years and hygiene and sanitation in orphanages are less than the environment in this study. Some literature states that the prevalence of *Blastocystis hominis* in developing countries varies; 50 - 60%,[15] and in Africa and Asia 37 - 100%,[6] Based on these data, Indonesia, which is still a developing country, is thought to have a high risk of the incidence of *Blastocystis hominis*. It is reported that in Malaysia the incidence of *Blastocystis hominis* is around 17.8% and reaches 25% in patients with comorbidities.[16] The incidence of *Blastocystis hominis* varies in some countries and regions within one country, depending on several things, environmental pollution, poor sanitation, poor personal hygiene, as well as exposure to pets and food and drinks contaminated with *Blastocystis hominis* cysts.[17]

In this study, all children identified did not have clinical symptoms/ asymptomatic. *Blastocystis hominis* is an opportunistic intestinal protozoan that is commonly found in human feces. If it affects immunocompetent people, this parasite does not cause symptoms, whereas in immunocompromised people it will show symptoms such as diarrhea and other symptoms of gastrointestinal disorders due to its occurrence decreased CD4,[18,19] as well as patients who have accompanying diseases or have comorbidities will cause clinical symptoms that also vary. In several studies, *Blastocystis hominis* is associated with irritable bowel disease, ulcerative colitis and colorectal cancer. It is suspected that in ulcerative colitis, *Blastocystis* has a protective role against this disease, although this is still unclear and needs further study.[20] In other studies, it was stated that the presence of *Blastocystis* in the intestine influences intestinal health and may have a potential pathogenic role in the presence of the organism. This parasite is thought to inhibit certain intestinal pathogens and improve the intestinal health of malnourished patient.[21] In another study, it was found that the presence of *Blastocystis* worsened the condition of patients with colorectal cancer.[22] This makes it ambiguous as to the role of this protozoa in intestinal health.

In this study, it was found that *Blastocystis hominis* was more frequently detected in girls 55.3%, and were in grades 4 - 6 with age group being 9 - 12 years and from statistic no significant differences were found. This is different from research in Jakarta on intestinal parasitic infections in school children, it was found that more boys had *Blastocystis* detected than girls and the same as the results of this study, more were found in grades 4 - 5 of primary school.[12] It can be seen that this gender has no effect of *Blastocystis*. In this study region, boys and girls have the same opportunity to play outside and come into contact with pets, so the risk of being exposed to this parasite is also the same. This is different from the ancient custom where girls were not allowed to play outside the house. *Blastocystis* transmission is mainly via the fecal-oral route, or through contaminated food and drink and is a zoonotic disease.[12,14,23,24]

In this study, it was found that 29.8% of students were underweight, it was found that 48.4% had *Blastocystis* detected in their feces. When compared with students who had overweight/normal no significant results were obtained (p>0.05). Based on this research, there is no relationship between nutritional status and the presence of *Blastocystis* in feces. In a 2023 publication regarding the relationship between malnourished children and the incidence of *Blastocystis* in several countries, it was found that there was a significant relationship between the incidence of *Blastocystis* and stunting in children, and in this study, it was implied that *Blastocystis* is a pathogen that affects children's intestinal health and will disrupt children's growth and development.[25] Contrary to the results of research in Bangladesh that in adults with malnutrition, *Blastocystis* is thought to play a role in inhibiting other intestinal pathogens and improving intestinal conditions.[19] In research in Egypt,

it was found that *Blastocystis* can indirectly trigger allergic reactions in children with asthma.[26] Some research results indicate that some *Blastocystis* play a pathogenic role and some play a protective role, further research is needed regarding the ambiguous role of *Blastocystis.* There are many subtypes and currently 32 subtypes of *Blastocystis* have been reported, these subtypes may have their respective roles.[10,15,27,28]

In this study, there was no significant relationship between risk factors and the presence of *Blastocystis* in the feces of elementary school students. The risk factors in question are personal hygiene such as clean nails, the habit of washing hands before eating and after defecating with soap, contact habits with pets, and sanitation factors such as places to defecate and sources of drinking water. All risk factors in this study did not have a significant relationship with the presence of *Blastocystis* in students' feces. Although nail hygiene in these students was generally poor (70.2%), it was not related to the presence of *Blastocystis* in the feces. The main transmission of *Blastocystis* is via fecal-oral, where *Blastocystis* enters the mouth through contaminated food and drink or unclean hands contaminated with *Blastocystis*. However, there is another transmission, *Blastocystis* infection is a zoonotic disease that is often found in pet feces, and in this study, there was no significant relationship between contact with pets and the presence of *Blastocystis*. Based on the literature, pets where *Blastocystis* is often found in their feces are cats, dogs, birds, chickens, goats and others.[28–30]

Although there was no significant relationship between the presence of *Blastocystis* and nail cleanliness, the attention of teachers and parents to the cleanliness of children's nails is very important. Dirty nails are a means by which pathogens enter the mouth, such as soil-transmitted helminth.[31] In this study, it was also found that 57.7% of students did not wash their hands with soap after defecating. Washing hands after defecating with soap is very important for personal hygiene. Washing hands after defecation is not enough with water only but must be done with soap to kill pathogens that remain when cleaning the remains of defecation or kill other pathogens on hands.[32,33] All subjects in this study had a good place to defecate, namely the toilet, and the source of drinking water was also good, namely refillable water and boiled well water. This refillable drinking water is categorized as good because if the depot owner follows the procedures by the government, then the water is suitable for drinking, although several refillable drinking water depots do not follow procedures and bacteria are found in the water.[34] A study on *Blastocystis* in refill water depots has not been reported.

In this study, it was found that the presence of *Blastocystis* in students' feces was quite high, but no significant relationship was found with student characteristics and personal hygiene for the presence of *Blastocystis* in students' feces. However, further research is needed on the source of *Blastocystis* transmission and risk factors in this area and a larger sample size is needed. Research on subtypes of *Blastocystis* is also important to determine the pathogenicity of this intestinal protozoan. In rural and remote areas of Indonesia, people still perceive parasitic infections as relatively unimportant and consider them a common occurrence. It is also essential to conduct studies on other types of parasites and protozoa, given the persistence of poor personal hygiene practices and inadequate sanitation.

Conclusion

The presence of *Blastocystis* in students' feces is quite high; 45.2%, all students were asymptomatic, although there was no significant relationship between the presence of *Blastocystis* and all the variables studied in this study, teachers and parents still have to pay attention to students' personal hygiene, such as keeping their nails clean and washing their hands with soap after defecating.

Acknowledgement: This publication is supported by Hibah Penelitian Dosen Pemula Faculty of Medicine, Universitas Riau, Indonesia.

Availability of Data: Research data has been displayed in the article, and the main data can be accessed by the author (by email; esy.maryanti@lecturer.unri.ac.id), data is available at the Parasitology Laboratory, Faculty of Medicine, Universitas Riau.

No use of generative AI tools: This article was prepared without the use of generative AI tools for content creation, analysis, or data generation. All findings and interpretations are based solely on the authors' independent work and expertise

Author contribution: EM: writing proposals, drafting articles, collecting data, carrying out examinations in the laboratory, analyzing data; **YW:** writing proposals, collecting data, analyzing data; **M:** carrying out examinations in the laboratory, analyzing data; **LAF, AAB and AR:** collecting data, carrying out examinations in the laboratory

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