



## RESEARCH ARTICLE

# Prevalence and risk factors for infection with *Entamoeba histolytica/dispar/moshkovskii* complex in people living in the slightest and outermost islands of Indonesia

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## ABSTRACT

*Entamoeba histolytica* (*E. histolytica*), the causative agent of amoebiasis, is still a global public health problem that cannot be controlled, especially in tropical and subtropical countries. This study was conducted to obtain information about the incidence of *Entamoeba histolytica/dispar/moshkovskii* complex infection and the factors that influence it. The prevalence of infection with the *Entamoeba histolytica/dispar/moshkovskii* complex and the factors that influence it in people living on the smallest and outermost island of Indonesia, Sabang Island, Aceh Province. This study involved 335 respondents aged  $\geq 10$  years. Respondents were selected by non-probability sampling technique. Interviews and observations were conducted to identify risk factors. The *Entamoeba histolytica/dispar/moshkovskii* complex was identified by direct examination, concentration, and Whitley's trichrome staining techniques. A Chi-Square test was performed to analyze the correlation of risk factors with the incidence of infection. The prevalence of infection with the *Entamoeba histolytica/dispar/moshkovskii* complex in the people of Sabang Island was 26.6% (89/335). Source and adequacy of clean water correlated with the incidence of *Entamoeba histolytica/dispar/moshkovskii* complex infection. Demographic variables are not correlated with the incidence of infection. However, the group of women aged  $\geq 61$  years, unemployed, unmarried, and earning less than the regional minimum wage tend to be more likely to be found with *Entamoeba histolytica/dispar/moshkovskii* complex infections. Thus it can be concluded that the prevalence of infection with the *Entamoeba histolytica/dispar/moshkovskii* complex on Sabang Island is in the high category. The prevalence of *E. histolytica* as the causative agent of amoebiasis cannot be explained with certainty because the two identical non-pathogenic *Entamoeba* species cannot be distinguished by microscopic identification. Sources and adequacy of clean water correlate with the incidence of *Entamoeba histolytica/dispar/moshkovskii* complex infection in the people of Sabang Island.

**Keywords:** Epidemiology; *Entamoeba histolytica/dispar/moshkovskii* complex; risk Tractors.

## INTRODUCTION

Amoebiasis disease caused by the parasite *Entamoeba histolytica* (*E. histolytica*) is classified as Neglected Tropical Diseases (NTDs) and can cause death (Junaidi *et al.*, 2020; Gunther *et al.*, 2011). This parasitic intestinal disease has the third highest death rate after malaria and schistosomiasis (Kantor *et al.*, 2018). The virulence of this parasite can cause sores and suppuration in the large intestine and colon (Nowak *et al.*, 2015; Tharmaratnam *et al.*, 2020). In some cases, parasites invade other organs, especially the liver and lungs (Wuerz *et al.*, 2012; Liu *et al.*, 2018). Amoebiasis is endemic in tropical and subtropical regions (Zibaei *et al.*, 2012). Developed countries with adequate levels of hygiene and sanitation have a lower proportion than poor and developing countries (Shirley *et al.*, 2018).

National Institute of Allergy and Infectious Diseases (2003) categorized *E. histolytica* as one of the priority biodefense pathogens for category B, namely agents that can infect many people through water and food and potentially be used as biological weapons. In addition, mechanical vectors such as flies and cockroaches (Ishartadiati, 2015; Kawano *et al.*, 2017), direct contact during anogenital or oro-anal sexual activity (Hung *et al.*, 2012; Valdoleiros *et al.*, 2019), become other routes of transmission and transmission that must be watched out for.

Monitoring amoebiasis in the community is not easy because the intensity and distribution of the disease are highly influenced by sociocultural, economic, geographic, and climatic factors (van Hal *et al.*, 2007). Furthermore, Ali (2015) stated that the actual frequency of amoebiasis is challenging to find because there is a tendency to exaggerate in endemic areas where dysentery or bloody diarrhea

is often misdiagnosed. In contrast, in non-endemic areas with low disease incidence, there is a tendency to ignore the symptoms of *E. histolytica* infection in clinical cases. Besides that, the prevalence of *E. histolytica* is too high because of its microscopic epidemiological overlap with other species that are identical to it, namely *E. dispar* and *E. moshkovskii* (Soares et al., 2019). To anticipate this confusion, experts agreed to call the results of the microscopic identification of the three amoebae the *E. histolytica* /*dispar*/*moshkovskii* complex (Bahrami et al., 2019).

Sabang Island, as one of the 92 outermost small islands of Indonesia, is vulnerable to changes caused by nature and or humans. One of the natural influences on the population is the uneven distribution pattern. This area, mostly made up of rocky mountains, is one of the causes for the concentration of Sabang islanders' settlements around the coast and several other strategic areas. Even though the proportion of land to the population of Sabang Island is very sparse, most of the population settlements are in dense locations. This condition indirectly affects the quality of sanitation, one of which is the insufficient availability of clean water (Maifira, 2020). This condition may be a predisposing factor to the attack of various infectious disease agents, and one of them is *Entamoeba histolytica* (Shahdoust et al., 2016; Costa et al., 2018).

This study was conducted to obtain information about the incidence of *Entamoeba histolytica*/*dispar*/*moshkovskii* complex infection and the factors that influence it.

## MATERIALS AND METHODS

### Research permit

This research has received ethical approval from the Health Research Ethics Committee of the Faculty of Medicine, Syiah Kuala University, Banda Aceh number: 149/EA/FK-RSUZA/2019. Informed consent forms are given to individuals willing to become research respondents to be signed or affixed with a thumbprint.

### Research design and sample

This research is an epidemiological study using a cross-sectional study design. The population in this study are people who live on the island of Sabang and are  $\geq 10$  years old. The research sample was selected by non-probability sampling, namely, 335 respondents. The sample inclusion criteria were natives of Sabang Island, able to communicate well and willing to become research subjects. Exclusion criteria were the sample moved address, died, or did not complete the entire data collection process.

### Types and methods of data collection

The data used in this research is primary data. Data on the dependent variable, *Entamoeba histolytica*/*dispar*/*moshkovskii* complex infection, were obtained through microscopic examination of stool samples. Independent variable data, namely demographic characteristics, sources of clean water, and adequacy of clean water obtained through structured interviews.

### Questionnaire and observation techniques

Sociodemographic data tracking includes age, gender, education level, occupation, marital status, and family income. Searching for information about the source and adequacy of clean water is obtained from the answers of respondents or family members.

### Identification of the *Entamoeba histolytica*/*dispar*/*moshkovskii* complex

Stool sampling begins with providing a unique pot and spoon as a tool to collect the stool sample. The next day, the researchers collected the pots filled with stool samples and put them in a cool box for examination in the laboratory. Samples taken and not yet examined on that day are stored in the refrigerator at 4°C to continue the examination the next day. Identification of the *Entamoeba histolytica*/*dispar*/*moshkovskii* complex was carried out microscopically. This identification begins with screening for the presence of intestinal amoeba using direct examination techniques and concentration with formol ether or zinc sulfate solution.

A positive sample was found in one of the screening techniques. A dry preparation was made and stained with Wheatley's trichrome staining technique. Then the cell characteristics were identified to distinguish the *Entamoeba histolytica*/*dispar*/*moshkovskii* complex from other intestinal amoeba using an Olympus Binocular CX 23 Microscope and a digital camera microscope Brand Dino-Lite Series AM4025X.

### Data management and analytics

The research data were processed with the help of the Microsoft Excel 2013 application software. Data that had gone through editing, coding, entry, and cleaning were analyzed univariately and bivariate. The univariate analysis aims to describe the frequency distribution of each research variable. The bivariate analysis aimed to see the correlation of transmission of the *Entamoeba histolytica*/*dispar*/*moshkovskii* complex based on the characteristics of the independent variables at a significance level of 5% ( $p < 0.05$ ).

## RESULTS

### Prevalence of *Entamoeba histolytica*/*dispar*/*moshkovskii* complex infection

The microscopic identification of 335 stool samples of respondents showed that the prevalence of *Entamoeba histolytica*/*dispar*/*moshkovskii* complex infection was 26.6% (89/335). This prevalence is lower than the same research in Indonesia conducted by Kesetyaningsih et al. (2010). However, several other studies, such as those conducted by Sungkar et al. (2015) in rural communities in East Nusa Tenggara and Maria et al. (2016) in Central Maluku, have a lower prevalence compared to the results of this study.

Based on Table 1, it can be seen that demographic factors are not statistically correlated with the incidence of *Entamoeba histolytica*/*dispar*/*moshkovskii* complex infection. However, descriptively, this infection was more common in the female respondent group, aged <60 years, secondary education, unemployed, single, and the family income is less than the Regional Minimum Wage (RMW).

**Table 1.** The influence of sociodemographic factors on the incidence of *Entamoeba histolytica/dispar/moshkovskii* complex infections in the people of Sabang Island

Variable	N = 335 n (%)	Infected with Eh/Ed/Em complex*		P value
		n negative (%)	n positive (%)	
<b>Gender</b>				
Male	178 (53.1)	136 (76.4)	42 (23.6)	0.118
Female	157 (46.9)	110 (70.1)	47 (29.9)	
<b>Age</b>				
10-19	71 (21.2)	52 (73.2)	19 (26.8)	0.892
20-60	227 (67.8)	168 (74.0)	59 (26.0)	
≤ 61	37 (11.0)	26 (70.3)	11 (29.7)	
<b>Education</b>				
Higher Education	44 (13.1)	32 (72.7)	12 (27.3)	0.883
Secondary Education	167 (49.9)	121 (72.5)	46 (27.5)	
Elementary Education	124 (37.0)	93 (75.0)	31 (25.0)	
<b>Work</b>				
Formal	72 (21.5)	52 (72.2)	20 (27.8)	0.799
Informal	138 (41.2)	104 (75.4)	34 (24.6)	
Not working	125 (37.3)	90 (72.0)	35 (28.0)	
<b>Marital status</b>				
Married/Widowed	247 (73.7)	182 (73.7)	65 (26.3)	0.889
Never married	88 (26.3)	64 (72.7)	24 (27.3)	
<b>Family income</b>				
Less than the RMW**	119 (35.5)	85 (71.4)	34 (28.6)	0.312
More than The RMW**	216 (64.5)	161 (74.5)	55 (25.5)	

\* Infected with *Entamoeba histolytica/dispar/moshkovskii* complex.

\*\* Regional Minimum Wage.

**Table 2.** Effect of sources and adequacy of clean water on the incidence of *Entamoeba histolytica/dispar/moshkovskii* complex infections in the people of Sabang Island

Source and adequacy of clean water	N = 335 n (%)	Infected with Eh/Ed/Em complex		P value
		n No (%)	n Yes (%)	
<b>Clean water source</b>				
RDWC	100 (29.9)	83 (33.9)	17 (19.1)	0.002*
Well	111 (33.1)	85 (34.6)	26 (29.2)	
Other sources	124 (37.0)	78 (31.7)	46 (51.7)	
<b>Adequacy of clean water</b>				
Enough	60 (17.9)	33 (55.0)	27 (45.0)	0.001*
Insufficient	275 (82.1)	213 (77.5)	62 (22.5)	

\* p value &lt; 0.05.

The people of Sabang Island generally obtain clean water from the Regional Drinking Water Company (RDWC), dug wells, drilled wells, rivers, and rainwater. Table 2 shows that the sources and adequacy of clean water variables correlate with the incidence of *Entamoeba histolytica/dispar/moshkovskii* complex infection. This can be explained by the fact that, out of 100 respondents who obtained clean water from the RDWC, 17 (19.1%) were infected with the *Entamoeba histolytica/dispar/moshkovskii* complex, and of the 111 respondents who obtained clean water from wells, 26 (29.2%) of respondents infected with *Entamoeba histolytica/dispar/*

*moshkovskii* complex. Meanwhile, of the 124 respondents who obtained clean water from other sources (RDWC with wells, RDWC with collected rainwater, RDWC with rivers, or wells with stored rainwater, or wells with rivers), as many as 46 (51.7%) respondents infected with *Entamoeba histolytica/dispar/moshkovskii* complex. This analysis shows a positive correlation between water sources and the incidence of infection. Respondents who obtain clean water from the RDWC tend to have a lower incidence of infection with the *Entamoeba histolytica/dispar/moshkovskii* complex.

## DISCUSSION

Diseases caused by *E. histolytica* infection tend to affect people with low levels of hygiene and sanitation. Jayaram Paniker (2018) classifies the prevalence of this infectious disease into three statuses: high prevalence states > 30%, moderate prevalence states 10-30%, and low prevalence states <10%. Based on this classification, the prevalence of *Entamoeba histolytica/dispar/moshkovskii* complex infection on Sabang Island is high, requiring special studies to treat it. Quoting the classic epidemiological theory put forward by H. L. Bloom, namely the four factors that affect the degree of public health, namely environment, behavior, health services, and heredity, may be used as a foundation in the study and management of the problem of complex infection of *Entamoeba histolytica/dispar/moshkovskii* on Sabang Island, Aceh Province.

Demographic factors, namely gender, age group, occupation, marital status, and family income, did not correlate with the incidence of *Entamoeba histolytica/dispar/moshkovskii* complex infection. This study's results align with two previous studies, which stated that there was no correlation between demographic factors and the incidence of *E. histolytica* infection (Orji & Okpala, 2014; Samie et al., 2020). The similarity of the results of this study can be explained by the fact that demographic factors are not a direct factor that causes the incidence of infection. However, its role is more to the enabling factor, which in certain conditions such as a weak immune system and inadequate health care, will allow infectious agents to infect people or groups. This condition is different from the report's statement of Shahrul Anuar et al. (2012). His study mentions that demographic factors are correlated with the incidence of this parasitic infection in indigenous Malaysian communities, which may be related to other aspects and are not yet known.

Water is a basic human need. Deficiencies and low quality will directly impact the health of individuals and society. This is in line with the results of this study, namely, the availability and adequacy of clean water affect the incidence of *Entamoeba histolytica/dispar/moshkovskii* complex infection. The correlation between the adequacy of clean water and the incidence of *Entamoeba histolytica/dispar/moshkovskii* complex infection is known as a negative correlation. This explains that of the 60 respondents who got enough clean water, 27 (45.0%) were infected with the *Entamoeba histolytica/dispar/moshkovskii* complex.

Meanwhile, of the 275 respondents who received enough clean water, 62 (22.5%) were infected with the *Entamoeba histolytica/dispar/moshkovskii* complex. The occurrence of this negative correlation cannot be explained with certainty. However, water quality may be a factor in the high proportion of infections in the respondents with sufficient access to clean water, as where Grilc et al. (2015) in their study, stated that microbiologically contaminated clean water has the potential to cause widespread disease outbreaks.

## CONCLUSION

The prevalence of infection with the *Entamoeba histolytica/dispar/moshkovskii* complex on Sabang Island is in the high category. The prevalence of *E. histolytica* as the causative agent of amoebiasis cannot be explained with certainty because the two identical non-pathogenic *Entamoeba* species cannot be distinguished by microscopic identification. Sources and adequacy of clean water correlate with the incidence of *Entamoeba histolytica/dispar/moshkovskii* complex infection in the people of Sabang Island.

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### Conflict of interest

The author declares that there is no conflict of interest.

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