Intestinal Amebiasis in Children with Bloody Diarrhea

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ABSTRACT

Background: *Amebiasis affects more than 50 million people each year, resulting in 100,000 deaths. Entamoeba histolytica (E. histolytica) is clearly pathogenic causing intestinal infection that varies from asymptomatic to fulminant colitis. Appropriate diagnosis and treatment of amebiasis in children is important to avoid serious complication, such as hepatic abscesses. Bloody diarrhea is a classic symptom, which is often used as screening for intestinal amebiasis. This study aimed to know the prevalence of intestinal amebiasis and other clinical and laboratory characteristics in children with bloody diarrhea who visited Harapan Kita Women and Children Hospital, Jakarta.*

Method: *This was a retrospective descriptive study. Data was obtained by evaluating medical records at Harapan Kita Women and Children Hospital Jakarta, from January 2009 to December 2010. The data included age, sex, morphology of E. histolytica, blood hemoglobin level and leukocyte count, as well as fecal leukocytes and erythrocytes. Diagnosis was confirmed by finding trophozoites in 3 consecutive stool specimens. Statistical analysis was performed using SPSS program.*

Results: *Trophozoite forms were found in 58/889 (6.5%) children with bloody diarrhea. There were 40 (58.8%) boys, and 27 (39.7%) children were ≤ 1 years old. Fourteen (20.6%) children had anemia, 49 (72.1%) children had fecal leukocytes > 10 and 51 (75%) children had fecal erythrocytes > 5; however, only 21 (30.9%) children with blood leukocytes count > 14,000 u/L.*

Conclusion: *Intestinal amebiasis is common in children with bloody diarrhea, especially infants. Increased fecal leukocytes and erythrocytes are often found in intestinal amebiasis; however, blood leukocyte count may not increase.*

Keywords: *children, intestinal tract, amebiasis*

ABSTRAK

Latar belakang: *Amebiasis diderita lebih dari 50 juta orang setiap tahunnya, dan menyebabkan 100.000 kematian. Entamoeba histolytica (E. histolytica) merupakan penyebab infeksi usus yang bervariasi dari asimtomatik hingga kolitis fulminan dan bersifat sangat patogen. Diagnosis dan tata laksana yang tepat untuk amebiasis pada anak sangat penting untuk mencegah komplikasi berat seperti abses hepar. Diare berdarah merupakan gejala klasik yang sering digunakan sebagai skrining amebiasis usus. Penelitian ini bertujuan untuk mengetahui prevalensi amebiasis usus serta karakteristik klinis dan hasil laboratorium pada anak dengan diare berdarah yang datang ke Rumah Sakit Anak dan Bunda (RSAB) Harapan Kita, Jakarta.*

Metode: *Penelitian ini bersifat retrospektif deskriptif. Data diperoleh dari hasil rekam medik di RSAB Harapan Kita Jakarta, antara bulan Januari 2009 hingga Desember 2010. Data mencakup usia, jenis kelamin, morfologi E. histolytica, kadar hemoglobin dan hitung lekosit dalam darah, serta hitung lekosit dan eritrosit dalam feses. Diagnosis dikonfirmasi dengan penemuan trofozoit dalam 3 spesimen feses yang berurutan. Analisis statistik dilakukan dengan program SPSS.*

Hasil: *Bentuk trofozoit E. hystolytica ditemukan sebanyak 58/889 (6,5%) pada anak dengan diare berdarah. Terdapat 40 (58,8%) anak laki-laki, dan 27 (39,7%) anak berusia ≤ 1 tahun. Anemia ditemukan pada 14 (20,6%) anak, 49 (72,1%) anak dengan lekosit feses > 10 dan 51 (75%) anak dengan eritrosit feses > 5; namun, hanya 21 (30,9%) anak dengan lekosit darah > 14.000 u/L.*

Kesimpulan: *Amebiasis intestinal sering ditemukan pada anak dengan diare berdarah, terutama bayi. Peningkatan lekosit dan eritrosit dalam feses sering ditemukan pada amebiasis intestinal; namun jumlah hitung lekosit tidak selalu meningkat.*

Kata kunci: *anak, saluran cerna, amebiasis*

INTRODUCTION

Amebiasis occurs worldwide and affects more than 50 million people each year. The disease is highly endemic, especially in developing countries.[1,2,3] Over 100,000 deaths caused by amebiasis have been annually reported.[2,4] Among the four species of *Entamoeba* found in the human gastrointestinal tract (*Entamoeba hartmanni, Escherichia coli, Entamoeba disper,* and *Entamoeba histolytica), Entamoeba histolytica* (*E. histolytica*) is clearly pathogenic.[5] Virulence factors are related to a number of proteins produced by the parasite and interaction between genetic capabilities of the strain and host factors (bacterial flora of the gut).[6] The incubation period of intestinal amebiasis can vary ranging from a few days to months or years, but it is generally about 1 to 4 weeks. The wide spectrum of intestinal infection ranges from asymptomatic to transient intestinal inflammation (few loose stools) and fulminant colitis with an array of manifestations.[7] Extra-intestinal amebiasis is a sub-type that involves liver, brain, spleen as well as other organs of the human body.[2]

Amebiasis in children must be diagnosed and treated properly to avoid further serious complication, such as hepatic abscesses. Bloody diarrhea can be caused by various bacterial pathogens, as well as by parasites, such as in amebiasis. Bloody diarrhea is a classic symptom, which is often used as screening for intestinal amebiasis. The prevalence of intestinal amebiasis in children with bloody diarrhea varies from one report to another, either hospital data or community data, ranging from 11-14.9%.[8,9]

The accuracy of diagnosis of intestinal amebiasis is largely determined by the experience of laboratory analyst to find trophozoites or cyst forms of *E. histolytica*. Due to the limitations of trained laboratory personnel in some primary health care facilities, therefore, the characteristics of other clinical symptoms and laboratory tests may help physicians to confirm the suspected intestinal amebiasis. The aim of this study was to know the prevalence of intestinal amebiasis and other clinical and laboratory characteristics in children with bloody diarrhea who visited Harapan Kita Women and Children Hospital, Jakarta.

METHOD

A retrospective descriptive study was performed in children with bloody diarrhea who visited Harapan Kita Children and Women Hospital Jakarta between January 2009 and December 2010. Data was obtained from the computerized medical records database system. The data included age, sex, morphology of *E. histolytica*, blood hemoglobin level and leukocyte count, as well as fecal leukocytes and erythrocytes. Diagnosis was confirmed by finding trophozoites in 3 consecutive stool specimens. Statistical analysis was performed using SPSS.

RESULTS

Of 889 children with bloody diarrhea, 58 children (6.5%) had intestinal amoeba based on the findings of trophozoites form in stool specimens; while specimens with cyst only, without trophozoites were found in 10 children (1.1%). About 40 (58.8%) subjects were boys and approximately 39.7% of patients were under 12 months old. Patient characteristics show in Table 1.

Table 1. Patient characteristics

Characteristics	n	%
Bloody diarrhea	889	
Entamoeba histolytica	68	7.7
Trophozoite	4	5.9
Cyst	10	14.7
Trophozoite + cyst	54	79.4
Sex		
Boy	40	58.8
Girl	28	41.2
Age		
≤ 1 years	27	39.7
> 1-2 years	21	30.9
> 2 years	20	29.4
Other clinical manifestations		
Tenemus/abdominal pain	10	14.7
Fever	15	22.0
Dehydration	8	11.8
Mucous in stool	45	66.2
Serum hemoglobin level (g/dL)		
< 11	14	20.6
≥ 11	54	79.4
Serum leukocyte count (u/L)		
< 14,000	47	69.1
≥ 14,000	21	30.9
Stool leukocyte count (/µL)		
≤ 10	19	27.9
> 10	49	72.1
Stool erythrocyte count (/µL)		
≤ 5	17	25
> 5	51	75

Tenesmus was found in 10 children (14.7%) and anemia was observed in 20.6% of children with blood hemoglobin level ≤ 11 g/dL. Serum leukocyte count > 10,000/μL was found in 30.9% of children, while fecal leukocytes of more than 10/μL and fecal erythrocytes more than 5/μL were found in 72.1% and 75% of children, respectively. No complications were found in our subjects.

DISCUSSION

Amebiasis is highly endemic in developing countries. There are genetically distinct strains of *E. histolytica* that cause intestinal amebiasis.[10] Virulence factors are related to a number of proteins produced by parasite including lectin that mediates adherence to epithelial cells and peptide that lyses cells by creating a pore and matrix digesting proteases.[6] The interaction of genetic capabilities of the strain and host factors such as the bacterial flora of the gut, may determine the virulence of *E. histolytica*.[1,2,3] Virulence may also be related to trophozoites ability to cause apoptosis of inflammatory cells and subsequently phagocytose the cells; thus, limiting further inflammatory response.[1,2]

The infective cyst form of the parasite survives passage through the stomach and small intestine. Excystation occurs in the bowel lumen, where motile and potentially invasive trophozoites are formed. In most infections, the trophozoites aggregate in the intestinal mucin layer and form new cysts, resulting in a self-limited and asymptomatic infection. Invading trophozoites destroy epithelial target cells by releasing substances such as hemolysins, which disrupt cell membranes by creating an amoebapore. In vitro, the trophozoites have a powerful ability to kill T lymphocytes, neutrophils, and macrophages.

Based on examinations of stools which found trophozoite form of *E. histolytica*, this study revealed 58/889 (6.5%) of prevalence for intestinal amebiasis in children with bloody diarrhea; while specimens with cyst only were found in 10 children (1.1%). Our data showed a quite high number of patients, i.e. 27 (39.7%) patients who were less than 1 years old. This could be explained since during infancy, the immune system has not been optimal and when the children started the oral phase of their normal developmental milestones, they put almost everything into their mouth, which became the port d'entree for parasites getting into the intestines.[11]

Patients with acute amebic colitis have typical symptoms of watery stools containing blood and mucus, abdominal pain, or tenesmus. A minority of patients are febrile or dehydrated. Abdominal examination may reveal pain over the lower abdominal quadrants.[5] Symptoms of intestinal amebiasis vary with the location and extent of the infection. Patients with extensive involvement have symptoms similar to ulcerative colitis. In patients with non-fulminant cases, fever is uncommon; while in patients with fulminant colitis, fever can be prominent.[5]

In this study, there were 15 (22%) of patients presented with fever and 10 (14.7%) of patients with abdominal pain or tenesmus. No dehydration was found in the study. There were 14 (20.6%) children with anemia, which might occur due to dissolving intestinal tissue caused by *E. histolytica* or other causes.[2]

Anemia should be observed after the infection has been resolved. When there is subsequently no improvement in serum hemoglobin level, further examination for other possible causes should be conducted, especially on iron-deficiency anemia that has a relatively high incidence in developing countries, including Indonesia. There were 48 (60.3%) patients below 3 years of age, who were at the period of accelerated brain development. Therefore, iron-deficiency anemia should be treated as soon as possible. Prolonged deficiency could affect the children's quality of life.[12] Anemia may cause oxygen transfer disorder that affects brain cells metabolism. It also affects lipid metabolism in myelin membrane resulting in myelin formation disorder and disruption of impulse conduction. It may also inhibit dopamine receptor causing alteration of behavior and concentration.[13]

The diagnosis of intestinal amebiasis is established by finding and/or identifying trophozoite and cysts in stool specimens.[2] Cyst carriers, may be environmentally dangerous or may develop colitis after a period of months.[3] Stool examination is sufficient as diagnostic test for ameba infection. However, stool specimens must be prepared properly. Stool should be fresh with blood and mucus and it should be examined three times. Ideally a wet mount of stool should be examined no longer than 20 minutes after collection to detect motile trophozoites. Three separate stool examinations have 90% sensitivity for the diagnosis of amebic dysentery.[5] If the patients no longer have diarrhea, we should find *Entamoeba spp.* cyst using lugol solution to see the core. The concentration method using zinc sulfate solution or ether formalin solution is used when the sample was too little.

The presence of Charcot-Leyden crystals, the lack of stool leukocytes, and the presence of blood are the most common stool findings in acute stage.[3] This

study showed that there were 49 (72.1%) patients with fecal leukocytes more than 10/μL and 21 (30.9%) patients had leukocytes count above 14,000/μL. Such results confirmed colon infection. Injury to epithelial cells triggers release of cytokines leading to chemotaxis of leukocytes, which also contribute to the local inflammatory response.

Fecal erythrocytes in stool analysis confirms the damage of intestinal mucosa which was reflected by fecal erythrocytes above 10/μL in 32 (47.1%) patients of this study. Charcot-Leyden crystal was not found in this study since *E. histolytica* may invade any portion of the colon; however, the caecum and ascending colon are most commonly affected. Serologic testing is particularly useful for suspected amebic liver abscess. Most patients do not show overt intestinal symptoms nor detectable cysts or trophozoites in their stool. Newer serum antigen tests are being developed that can distinguish *E. histolytica* from *E. dispar*. The results may become negative with successful treatment. Antigen test can also be performed on material aspirated from liver abscess.[14]

All patients were treated with metronidazole at the dose of 50 mg/kgBW. Three (4.41%) patients had relapse.[10] Tinadazole, a more potent nitroimidazole against amebic infection, can be used for shorter treatment courses and it is well-tolerated in children. In patients who cannot tolerate metronidazole or tinidazole, erythromycin and tetracycline are effective against intestinal trophozoites but are ineffective against trophozoites in liver abscesses.[5,15,16] It is very important to re-evaluate trophozoite form in the stool specimen after completing treatment with three consecutive examinations. Patients who only present with cyst forms still require treatment since the commensal amoeba may develop into pathogens. Treatment of amoebic infection is complicated because different agents are necessary for eradicating the parasites from the bowel or tissue. Whether asymptomatic cyst passers should be treated remains controversial.[5] Follow-up of amoeba (trophozoites and cysts) in the stool specimens should be continued until three consecutive evaluations show negative results.[5]

A variety of excreted cysteine proteases disrupt the extra cellular matrix. Injury to epithelial cells triggers release of cytokines leading to chemotaxis of leukocytes, which also contribute to the local inflammatory response. Subsequently, ulceration of the mucosa occurs and invading amoeba may enter the portal circulation and eventually the liver. Once the intestinal epithelium is invaded, extra-intestinal spread to the peritoneum, liver, and other sites may follow. Complications to the liver cause abcess, anemia due to blood loss in stools, septicemia, perforation of intestines, and peritonitis. Neutrophils responding to the invasion contribute to cellular damage at the site of invasion. *In vitro,* the trophozoites have a powerful ability to kill T-lymphocytes, neutrophils and macrophages.[17] However, this study did not reveal such complications, except for anemia (hemoglobin level ≤ 11 g/dL) in 20% of children.

CONCLUSION

The prevalence of intestinal amebiasis in children with bloody diarrhea is 6.5% and is more common in boys than girls. Related with the lower immunity system, infants require special attention for they have higher risk of amebic infection. Most cases showed increased fecal leukocytes and erythrocytes, but no increase in serum leukocyte count was found.

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