Difficulty Confirming the Diagnosis of Amoebic Enteritis

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Amoebic dysentery is designated a Category 5 disease under the Infectious Disease Control Law in Japan, with approximately 1,000 cases reported annually. About 10% of these are cases of invasive amoebic dysentery, 90% of which have an asymptomatic course and are often discovered incidentally, and there is concern that the number of undiagnosed cases is increasing since the reagent for that blood antibody test was discontinued in 2017. Invasive amoebic dysentery often causes ulcerative lesions that affect the cecum and rectum predominantly, but eradication of amoebic dysentery is possible with proper diagnosis and treatment. However, there have been cases in which delayed diagnosis and treatment have resulted in fulminant forms of colorectal ulceration, including perforated peritonitis and amoebic liver abscesses. In this report, the importance of the diagnosis and treatment of this disease is reiterated, and a case that was difficult to diagnose is presented.

Key words: amoebic colitis, Entamoeba histolytica, amoebic cystic forms, persistent infection, paromomycin

INTRODUCTION

Amoebic enteritis is caused by the protozoan *Entamoeba histolytica*, a member of the genus amoeba [1]. It is found worldwide, but it is particularly common in developing countries with inadequate sanitation and in tropical and subtropical countries [2]. Clinically, most cases are asymptomatic, but when colonic mucosal invasion and tissue destruction occur, symptoms such as diarrhea and bloody stools develop, and there have been reports of host immune response and immune evasion [3]. The diagnosis of *E. histolytica* infection is based on fecal examination for parasites, but these tests are less sensitive, and it is difficult to distinguish morphologically identical species from other nonpathogenic genera of amoeba [4, 5], which currently requires proficiency. In this report, a case of amoebic enteritis that was difficult to diagnose and in which paromomycin was effective is presented, and the clinical course and diagnosis of amoebic enteritis are discussed based on a review of the literature.

CASE REPORT

The patient, a 41-year-old, heterosexual man, came to our department with diarrhea and bloody stools as his chief complaints. His medical history included a history of overseas travel to Egypt and Libya when he was 22 years old and subsequent hospitalization for a colorectal ulcer during investigation of the cause of his diarrhea. Laboratory data are presented in Table 1. Colonoscopy (CS) showed multiple, irregularly shaped, ulcerative lesions in the cecum (Fig. 1), but no lesions elsewhere, including the rectum. Both biopsies and tests for amoebic antibodies were negative, and the patient was followed. At age 42 years, CS was performed for positive fecal occult blood and showed a residual cecal ulcer (Fig. 2a), and dysentery amoebotroph type was confirmed in biopsy tissue (Fig. 2b), so the patient was treated with oral metronidazole 1500 mg/day for 10 days. On follow-up CS, the ulcer in the cecum showed a tendency to heal (Fig. 3), but the biopsy tissue showed amoeboid trophotypes. Thereafter, the patient did not visit the outpatient clinic, and CS was performed at our hospital at the ages of 45 years (Fig. 4a), 46 years (Fig, 4b), and 48 years (Fig. 4c) with positive fecal occult blood, and the cecal ulcer repeatedly healed and relapsed, but the biopsy test was negative for amoebic dysentery, so the patient was followed. At the age of 49 years, CS was performed due to positive fecal occult blood, and a tendency toward increasing cecal ulceration was observed (Fig. 5a, b), but the biopsy tissue was negative for amoebic dysentery. Repeated cecal ulcers and their tendency to worsen led us to strongly suspect amoebic enteritis, and microscopic examination of the stools showed numerous amoebic cystic forms. The patient's wife was pregnant at the same time, and microscopic examination and PCR testing of the patient's and his wife's blood for amoebic dysentery antibodies and the stools of family members living with the patient were negative for all but the patient himself (Table 2). Oral metronidazole was started, but numerous amoeboid cystic forms were identified in the stool three days later, and paromomycin was also administered. Three months after treatment, the CS findings showed that the ulcer in the cecum had scarred (Fig. 6), and the amoebae in the stool had disappeared; the patient was therefore considered to be in remission, and stool occult blood was negative on the

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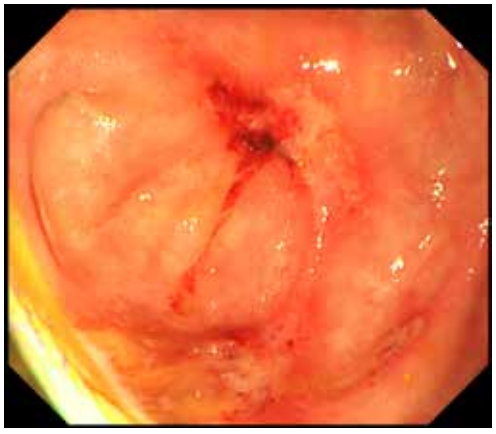

Fig. 1 Irregularly shaped ulcer near the appendix

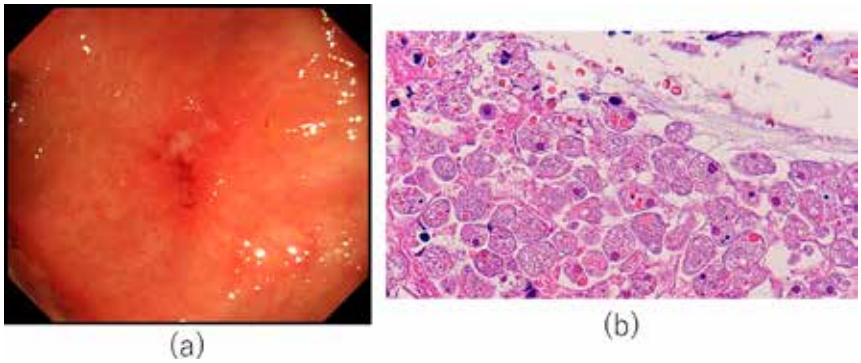

Fig. 2 Definitive diagnosis of amoebic dysentery
a) Residual ulceration near the appendix
b) Amoeba trophozoites present in biopsy tissue

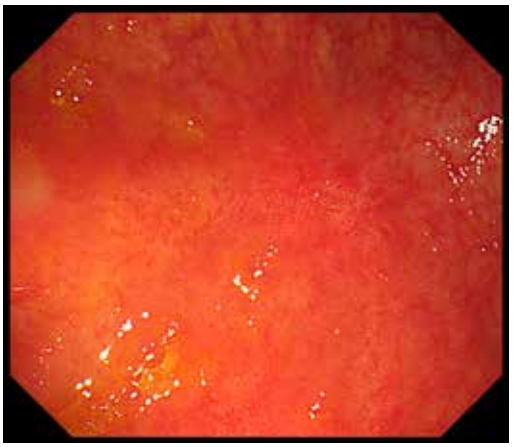

Fig. 3 Ulcer scarification

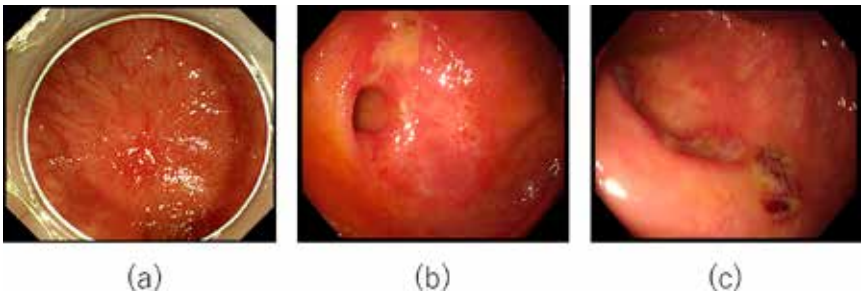

Fig. 4 Cecal ulcer with recurrent flare-ups and remissions
a) Ulcer scar (age 45 years)
b) Re-inflamed ulcer (age 46 years)
c) Residual or recurrent ulcer (age 48 years)

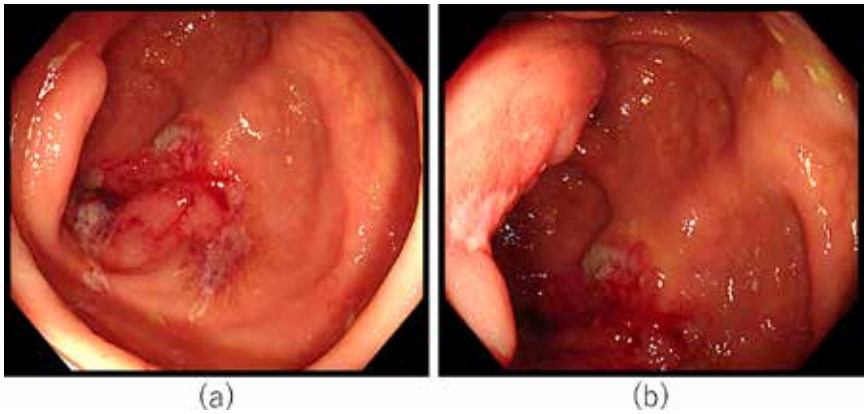

Fig. 5 CS findings at the time of another definitive diagnosis of amoebic colitis
a) Multiple ulcers on the appendix
b) Ulceration on Bauhin's valve

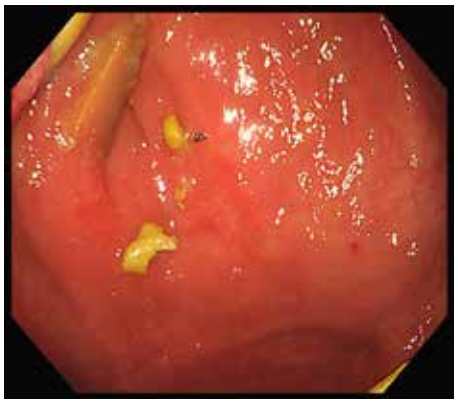

Fig. 6 Ulcer scarification

Table 1 Summary of laboratory data

Blood cell count			Biochemistry		
WBC	4,900	/μL	TP	7.3	g/dL
Neu	53	%	Alb	4.3	g/dL
Lym	36	%	AST	19	IU/L
Eosino	5.5	%	ALT	26	IU/L
RBC	5.52×10^6	/μL	LDH	123	IU/L
Hb	15.5	g/dL	ALP	239	IU/L
Plt	25.6×10^4	/μL	γ-GTP	111	IU/L
			T-Bil	1.1	mg/dL
Infectious disease test			Glu	98	mg/dL
Syphilis antibody	(-)		BUN	14	mg/dL
HBsAg	(-)		Cr	0.95	mg/dL
HCVAb	(-)		UA	3	mg/dL
Amoebic antibody	(-)		Na	142	mEq/L
			K	2.4	mEq/L
			Cl	97	mEq/L
			CRP	<0.09	mg/dL

Table 2 Results of parasitological examinations

	Microscopic examination (amoebic cyst)	PCR (*E. histolytica**)	Anti-amoebic dysentery antibody
Patient himself	(+)	(+)	1:1024
Wife	(-)	(-)	(-)
Daughter	(-)	(-)	
Mother	(-)	(-)	
Sister	(-)	(-)	

**E. histolytica: Entamoeba histolytica*

physical examination two years later.

DISCUSSION

E. histolytica causes infection by oral ingestion of infectious cystic forms in food or liquid contaminated by human feces. The cysts pass through the small intestine, unaffected by gastric acidity, and release their cysts near the terminal ileum or cecum to form trophozoites, which are pathogenic. Trophozoites begin to disrupt the colonic mucosal epithelial barrier, causing inflammation and amoebic colitis that lead to necrosis of host cells and formation of colonic ulcers [6]. It has been reported that more than 90% of such infections have an asymptomatic course, often self-limited over various periods of time [3], and it is possible that the present patient had repeated self-infections over a period of 8 years, but if the patient was first infected at the time of colitis at age 22 years, the possibility that it could have been 27 years cannot be excluded. However, considering that anti-amoeba antibodies were negative at the first visit at age 41 years, and that the same antibodies were positive at age 49 years, a false-negative initial antibody test or subsequent antibody production seems reasonable.

The reasons for this patient's repeated episodes of amoebic enteritis were: 1) amoeba antibody test and biopsy tissue of the ulcerative lesion at the initial visit were false-negative for amoeba; 2) biopsy tissue at repeat CS identified amoeba and the ulcer showed a tendency to heal with metronidazole, but the patient dropped out of outpatient follow-up; 3) repeated biopsies of recurrent/healing ulcerative lesions were false-negative, and the patient was asymptomatic; and 4) the patient was not treated with paromomycin to eradicate amoebic cystic forms in the gastrointestinal tract after metronidazole treatment. Identification of parasites in feces is the commonly used method for diagnosing amoebic infections, but it requires expertise in amoebic morphology and experience, and its accuracy is reported to be less than 80% [7]. Although false-negative results are possible, serum amoebic antibodies are also useful in differentiating *E. histolytica* from *E. dispar* [8], and the significant decrease in the number of reported cases of amebiasis due to the discontinuation of testing raises concerns about future transmission by asymptomatic infected individuals. Although PCR tests are highly accurate, and studies of amoebic strains have been reported [9, 10], they are not covered by health insurance in actual clinical practice. The patient's family requested a PCR test to search for intra-familial infection, but only the patient himself tested positive. Since all other family members were negative, the patient was considered to have repeatedly self-infected.

Treatment of amebiasis with nitroimidazole derivatives, including metronidazole, is expected to cure clinical symptoms and result in endoscopic cure, but treatment of cysts with tubular amoebic agents such as paromomycin is important for the eradication of *E. histolytica* [6]. In Japan, paromomycin sulfate has been imported by the Research Group on Drugs for Tropical Diseases since 1998, and a system has been established to make it available for the treatment of patients with dysentery amebiasis. The development of paromomycin by Pfizer Japan Inc. began in 2010, and in December 2012, it was approved for inclusion in the Japanese health insurance system for the treatment of intestinal amebiasis, and its administration is strongly recommended for the eradication of dysentery amebiasis. The experience of this patient, who had repeated relapses due to outpatient dropouts, re-affirmed that multiple negative tests with adequate explanation to the patient regarding amoebic dysentery during the course of treatment can lead to complete cure.

In conclusion, although confirmation of endoscopic findings after treatment is necessary for treating amoebic dysentery, frequent checks for disinfection of cysts in the lumen, as well as trophozoite disinfection, of *E. histolytica* are also considered essential to prevent persistent infection.

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