# Amoebiasis among institutionalized psychiatric patients in Taiwan

### H.-S. CHENG1 AND L.-C. WANG2\*

- <sup>1</sup> Department of Internal Medicine, Chang-Gung Memorial Hospital, Kwei-San, Tao-Yuan, Taiwan
- <sup>2</sup> Department of Parasitology, College of Medicine, Chang-Gung University, Kwei-San, Tao-Yuan, Taiwan

(Accepted 2 December 1998)

#### **SUMMARY**

Although information on amoebiasis among institutionalized psychiatric patients is available, reports on the relationship between behaviour and this infection are not abundant. From July 1995 to June 1996, stool and blood samples were collected from 565 patients in three psychiatric hospitals of North Taiwan. Stool samples were examined using the direct smear and formalin-ethyl acetate sedimentation techniques as well as ProSpecT® *Entamoeba histolytica* Microplate Assay kit. Blood samples were examined by the Amebiasis Serology Microwell ELISA kit. Among these patients, 14 (2·5%) harboured one or two species of intestinal parasites. There were 6 (1·1%) *E. histolytica/E. dispar* cyst passers: 5 positive in stool ELISA test and 2 with antibodies against *E. histolytica*. Among demographic factors, type of psychiatric disorder and disability, only a significant sexual difference in seropositivity of *E. histolytica* was observed. These findings indicate that the infected patients acquired the infections before they entered the hospitals.

# INTRODUCTION

Amoebiasis is an important cosmopolitan health problem. It has been estimated that about 500 000 of the world population have this infection and invasive amoebiasis causes up to 100 000 deaths annually [1]. Although this infection is more prevalent in developing countries, outbreaks are usually associated with crowding and insanitary conditions. However, severe clinical manifestations only affect a small percentage of infected persons [2, 3].

Infections with *E. histolytica/E. dispar* are not uncommon in institutionalized psychiatric and mentally retarded patients. Epidemiological studies in the United States [4–6], England [7], France [8], Italy [9] and Japan [10, 11] revealed positive rates ranging from 7 % to over 30 %. The transmission of amoebiasis among these patients has been attributed not only to the direct faecal—oral route but also their abnormal behaviour [10, 12].

\* Author for correspondence.

In 1993, an outbreak of amoebiasis occurred among children at a primary school in Central Taiwan. After rapid treatment of the infected children with severe diarrhoea, the faculty of this school were examined serologically and a positive rate of 8.0 % was obtained [13]. Moreover, 42.6% of the aborigines on Lanyu, an offshore island in East Taiwan, had high titres ( $\geq 256$ ) of antibodies against E. histolytica [14]. A survey on the patients at a mental hospital in South Taiwan revealed that 10.9 % were cyst passers and 16.9 % were positive for serum antibodies [15]. These findings indicate that amoebiasis is prevalent among the isolated and institutionalized populations in the remote areas. However, no current information on the status of amoebiasis in the institutionalized groups in urban and suburban areas is available.

Although information on amoebiasis among institutionalized psychiatric patients is available [4–12], reports on the relationship between behaviour and the infection are not abundant [10, 12]. In the present study, we examined patients in psychiatric hospitals in

two suburban areas of North Taiwan using parasitological and immunodiagnostic methods. In addition, the demographic data and health status were obtained to determine the factors determining the transmission of this infection.

#### MATERIALS AND METHODS

### Psychiatric hospitals

This study was conducted from July 1995 to June 1996 in three psychiatric hospitals of Taipei County, North Taiwan. Two hospitals (Hospitals A and B) are located at Hsintein City in the central part of the county and the remaining one (Hospital C) at Pali District in the western part of the county. The project was approved by the Medical Ethics and the Human Clinical Trial Committee of Chang Gung Memorial Hospital, Taipei, Taiwan.

There were 408, 323 and 238 inpatients in Hospitals A, B and C respectively. These three hospitals are surrounded by a garden to which the patients have free access. The ratios of patients to the workers directly involved with daily care are 10:1, 10:1 and 3:1 respectively.

#### Collection of samples

Through the co-operation of the hospital faculty, three faecal specimens were obtained from each patient. These specimens were collected in 25-ml plastic containers and sent to our laboratory within 12 h. After taking a sample of 1 g from the container for parasitological examination, the remaining portion was then stored at -70 °C for examination by ELISA.

A whole blood sample was also collected from each patient into a centrifugation tube at the hospitals. These samples were placed in ice boxes and brought back to our laboratory immediately. After keeping at  $4\,^{\circ}\text{C}$  overnight, serum samples were prepared by centrifuging the clotted blood at 3000 r.p.m. for 30 min at the same temperature. The serum samples were then stored at  $-70\,^{\circ}\text{C}$  for ELISA examination.

#### **Examinations**

From each fresh stool specimen, two saline wet mounts were prepared and examined microscopically for trophozoites. Eggs, larvae and cysts of intestinal parasites were detected using the formalin-ethyl acetate sedimentation technique [14]. The ProSpecT® *Entamoeba histolytica* Microplate Assay kit (Alexon Inc., Sunnyvale, USA) was employed to detect *E*.

histolytica specific antigens in the aqueous extracts of the stool specimens. The Amebiasis Serology Microwell ELISA kit (LMD Laboratories Inc., Carlsbad, USA) was used to detect serum antibodies against *E. histolytica*. The procedures of the enzyme immunoassays were carried out according to the instructions of the manufacturers.

#### Demographic data and health status

Faculties of the hospitals were asked to complete questionnaires concerning patients in their care. Demographic data and health status of the patient were obtained from their medical records. Disability was determined from activities in their daily life and was evaluated using the Guttman index [15]. Moreover, recent occurrence of gastrointestinal distress was also assessed.

# Chemotherapy and follow-up

The tissue-acting agent tinidazole (2 g daily for 5 days) was administered to *E. histolytica/E. dispar* cyst passers because of the unavailability of luminal amebicides (paromomycin, iodoquinol or diloxanide furoate) in Taiwan. Infected patients with soil-transmitted helminths were treated with mebendazole (100 mg daily for 4 days) and those with *Clonorchis sinensis* with praziquantel (25 mg/kg t.i.d. for 2 days).

A stool specimen was obtained from each treated cyst passers on three separate days after the first, second and fourth weeks following treatment. A single specimen was collected from the treated infected patients with other helminth infections in the first and fourth weeks post-treatment. These specimens were also examined using the saline wet mount and formalin-ethyl acetate sedimentation techniques [14]. Another course of treatment was given to the treated cases with positive findings in the follow-up examinations.

## Statistical analysis

Rates were compared by the  $\chi^2$  test. P < 0.05 were considered to be statistically significant.

# RESULTS

#### **Subjects**

Of the 565 patients,  $61\cdot1\%$  were males and  $38\cdot9\%$  females. Most of them were between 17 and 65 years in age.  $72\cdot9\%$  had schizophrenia and the remaining

	Hospi $(n = 3)$		Hospital B $(n = 210)$		Hospital C $(n = 50)$	
Species*	No.	% pos.	No.	% pos.	No.	% pos.
Entamoeba histolytica	6	2.0	0	0	0	0
Trichuris trichiura	5	1.6	0	0	0	0
Hookworm	1	0.3	1	0.5	0	0
Strongyloides stercoralis	0	0	1	0.5	0	0
Clonorchis sinensis	1	0.3	0	0	0	0

Table 1. Prevalence of intestinal parasitic infections among 565 inpatients of three psychiatric hospitals in North Taiwan

Table 2. Entamoeba histolytica infections among inpatients in three psychiatric hospitals determined by parasitological examination, stool ELISA, and blood ELISA

	Institute A			Institute B			Institute C		
Examination	No. exam.	No.	% pos.	No. exam.	No.	% pos.	No. exam.	No.	% pos.
Detection of cysts in stool	305	6	2.0	201	0	0	50	0	0
Detection of specific antigens in stool	136	5	3.7	76	0	0	38	0	0
Detection of antibodies in serum	136	12	8.8	70	2	2.9	30	0	0

ones were hospitalized because of other types of psychiatric disorders. Although about 10% of the patients had hypertension and stroke, their general health was normal and only 20% needed help in their daily life.

# Prevalence of amoebiasis and other intestinal parasitic infections

Results of the parasitological examination are shown in Table 1. Of the 565 patients examined, 14 (2·5%) were found to be infected with one or two species of intestinal parasites: *E. histolytica/E. dispar* (6 cases), *Trichuris trichiura* (4 cases), hookworms (1 case), *Strongyloides stercoralis* (1 case), *Clonorchis sinensis* (1 case) and *T. trichiura* and hookworm (1 case). In addition, *Entamoeba coli* were found in 2 cases.

No trophozoites were detected in the fresh stool specimens using direct smear. By formalin-ethyl acetate sedimentation technique, the positive rate of *E. histolytica/E. dispar* infection was determined to be 1·1% and all cyst-passers were in Hospital A.

Moreover, five of them were also found to be positive for *E. histolytica* specific antigens in the stool. Blood ELISA examination revealed 14 patients from the hospitals in Hsintein City with serologic anti-*E. histolytica* antibodies (Table 2).

Among 235 patients examined with the three techniques, antibodies against *E. histolytica* were detected in 11 patients without positive findings in parasitological examination and stool ELISA. One cyst passer also had positive blood ELISA test and two patients showed positive results by the three techniques.

# Relationship between intestinal parasitic infections and various factors

The patients in Hospital A (11/305, 3.6%) had a higher prevalence of intestinal parasitic infections than Hospital B (2/210, 1.0%). However, the difference between these rates was not statistically significant (P > 0.05). Although no pathogenic parasites were found among patients in Hospital C, one infected person with  $E.\ coli$  was detected. Moreover,

<sup>\*</sup> One patient was infected with both *T. trichiura* and hookworm. In addition, one patient in Hospital A and another one in Hospital C harboured the non-pathogenic *Entamoeba coli*.

Table 3. Relationships between the prevalence of intestinal parasitic infections and demographic factors, type of
psychiatric disorder, and degree of disability of 565 patients in three psychiatric hospitals of North Taiwan

	Hospital A		Hospital B		Hospital C		Total	
Item	No. exam.	% pos.	No. exam.	% pos	No. exam.	% nos	No. exam.	% pos.
- Tem	CAAIII.	pos.	CAAIII.	pos.	CAAIII.	pos.	CAAIII.	pos.
Demographic factors								
Male	190	4.2	124	0.8	31	0	345	2.6
Age $\geq 40$ years	176	5.1	96	1.0	14	0	286	3.5
Taiwanese	219	4.1	143	1.4	41	0	403	2.7
Primary education or below	145	4.8	92	0	16	0	253	2.8
Single (marriage)	175	4.0	133	0.8	35	0	343	2.3
Types of psychiatric disorder								
Schizophrenia	199	3.0	163	1.2	40	0	402	2.2
Degree of disability								
Guttman index > 0 point	84	2.4	25	0	1	0	110	1.8

those who were male (2.6%), age  $\geq 40$  years (3.5%), Taiwanese (2.7%), primary education or below (2.8%), and single (2.3%) had a higher positive rates. The differences between these factors and their counterparts were not significant (P > 0.05) (Table 3).

Patients with certain degrees of disability (Guttman index > 0 point) had a slightly lower positive rate (1.8%) than those without disability (2.9%). The difference in positive rates of these two groups was not significant statistically (P > 0.05) (Table 3).

#### Relationship between amoebiasis and various factors

Among the patients, females (9.9%) were found to have a significantly higher positive rate of *E. histolytica* antibodies than the males (2.4%) (P = 0.0307). There were no significant associations between this infection with the remaining demographic factors as well as the type of psychiatric disorder and degree of disability (Table 4).

# Chemotherapy and follow-up

Although five treated *E. histolytica/E. dispar* cyst passers became negative after chemotherapy, positive findings were obtained in the stool specimens from the remaining one collected after treatment. This patient was administered an additional course and became negative in the second follow-up examination. The patients with infection of *T. trichiura*, hookworms, *S. stercoralis*, or *C. sinensis* were found to be negative in the follow-up stool examination.

Table 4. Relationships between the prevalence of seropositivity for Entamoeba histolytica infections and demographic factors, type of psychiatric diseases, and degree of disability of 565 patients in three psychiatric hospitals of North Taiwan

Item	No. exam.	No. pos.	% pos.
Demographic factors			
Female*	100	11	9.9
Age $\geq 40$ years	97	8	7.6
Taiwanese	167	10	5.6
Primary education or below	84	6	6.7
Single (marriage)	138	5	3.5
Types of psychiatric disorder Schizophrenia	186	12	6.1
Degree of disability Guttman index > 0 point	210	13	5.8

<sup>\*</sup>  $\chi^2$  test: P = 0.0307.

### **DISCUSSION**

According to a recent description of *E. histolytica*, this pathogenic parasite should be distinguished from its non-pathogenic counterpart, *E. dispar*, by appropriate biochemical, immunological, and/or genetic techniques [18]. These methods included specific DNA probes [19], PCR and restriction pattern analysis [20–23], monoclonal antibodies [24, 25] and isoenzyme typing of hexokinase and phosphoglucomutases [26, 27]. However, it is technically difficult to apply these techniques in large-scale epidemiological studies, since they require specialized instruments and cultivation of the isolated amoebae.

In the present study, we employed the parasitological method as well as stool (ProSpecT® Entamoeba histolytica Microplate Assay) and blood (Amebiasis Serology Microwell ELISA) ELISA kits to detect infections with E. histolytica. Our results indicate that one cyst passer had a positive result in blood ELISA and two patients were determined to be positive by the three techniques. In addition, 11 patients only had only positive findings in the blood ELISA test. It has been reported that persistent existence of serum antibodies indicates past infections of E. histolytica while negative results occur in those with E. dispar [3]. The patients with antibodies and positive findings in stool examination and/or stool ELISA should have current E. histolytica infections.

In Japan, an outbreak of amoebiasis was reported in an institute for mentally retarded patients in 1989. Although this institute had modern equipment, stool examination revealed cysts or trophozoites of *E. histolytica* in 20% of the patients and 38% were found to be positive serologically [11]. In the present study, we found infections of pathogenic intestinal parasites only in Hospitals A and B. These hospitals shelter more patients and the ratios of patients to workers directly involved with daily care are relatively low (10:1). These findings indicate that crowding and insufficient care may be important factors in the transmission of intestinal parasitic infections among institutionalized psychiatric patients.

Although the Guttman index [15] was designed for the study of household activities in elderly people, we employed this index to determine the degrees of disability in psychiatric patients through the observation of hospital faculties. It is relatively simple for the observer to record the daily life of the subject. There are six questions to be answered: (1) getting up and down stairs and steps, (2) getting around the house, (3) getting in and out of bed, (4) cutting the toenails by oneself, (5) bathing or washing all over, and (6) going out and walking down the road. The answers are scored 0 point for by oneself without difficulty, 1 point by oneself but with difficulty, and 2 points only with the help from someone else or not at all. The degree of disability is rated as none (0 point), slight (1–2 points), moderate (3–5 points), severe (6–8 points), very severe (9-12 points).

In Taiwan, patients with various psychiatric disorders and mental retardation were institutionalized in the same hospitals. However, patients with schizophrenia were in the majority. This situation was not only in the hospitals participating in the present study but also in most of the psychiatric institutes throughout Taiwan. We found most of our subjects without disability in their daily life. Moreover, no significant differences were observed in the prevalence of intestinal parasitic infections as well as seropositivity of *E. histolytica* according to the type of psychiatric disorder and the degree of disability. These negative findings indicate that the transmission of these infections may not be related to the inappropriate behaviour of the patients; transmission of *E. histolytica* in psychiatric patients has been attributed to their abnormal behaviour [10, 12]. Moreover, this infection may not be acquired in the hospital.

Of the patients participating in the present study, positive findings in the three techniques were aggregated to the hospitals in Hsintein City. It is possible that the infections may be related to environmental conditions. However, we also found significant sexual difference in the seropositivity of *E. histolytica*. Therefore, transmission of intestinal parasitic infections as well as amoebiasis may not occur in the hospitals. The patients may acquire these infections before they entered the hospitals and remained in the pre-clinical stage. This suggestion is inconsistent to the well-known fact that the abnormal behaviour of psychiatric patients plays an important role in the transmission of *E. histolytica* in psychiatric institutes [10, 12].

#### **ACKNOWLEDGEMENTS**

This study was supported in part by the Research and Development Program, Department of Health, Executive Yuan, R.O.C. (Grant No. DOH85-TD-016). The authors wish to thank the faculty of the three psychiatric hospitals for their co-operation in collecting the specimens and information from the patients. Special appreciation is also due to Miss Shi-Chih Lin, June-Wen Wang, Chin-Yi Wang and Mr Ing-Hou Loh and Boon-Huoi Ng for their valuable technical assistance.

#### REFERENCES

- Walsh JA. Problems in recognition and diagnosis of amebiasis: estimation of the global magnitude of morbidity and mortality. Rev Infect Dis 1986; 8: 228-38.
- 2. Petri WA Jr, Ravdin JI. Amebiasis in institutionalized populations. In: Ravdin JI, ed. Amebiasis. Human infection by *Entamoeba histolytica*. New York: Churchill Livingstone, 1988; 576–81.

- 3. Ravdin JI. Amebiasis. Clin Infect Dis 1995; **20**: 1453–66.
- Sexton DJ, Krogstad DJ, Spencer HC Jr, et al. Amebiasis in a mental institution: serologic and epidemiological studies. Am J Epidemiol 1974; 100: 414–23.
- Thacrer SB, Simpson S, Gordon TJ, Wolfe M, Kimball AM. Parasitic disease control in a residential facility for the mentally retarded. Am J Public Health 1979; 69: 1279–81.
- Braun TI, Fekete T, Lynch A. Strongyloidiasis in an institution for mentally retarded adults. Arch Intern Med 1988; 148: 634–6.
- Sargeaunt PG, Williams JE. A study of intestinal protozoa including non-pathogenic *Entamoeba histoly*tica from patients in a group of mental hospitals. Am J Public Health 1982; 72: 178–80.
- Chaker E, Keller F, Sommer A, Kremer M. Focus of amebiasis in a psychiatric hospital in the east of France. Arch 1 Instit Pasteur Tunis 1981; 58: 15–37.
- Gatti S, Cevini C, Marchi L, Novati S, Scaglia M. Entamoeba histolytica autochthonous isolates from mentally retarded Italian patients. Parasitol Res 1995; 81: 148–51.
- Nagarura R, Tachibana H, Tanara T, et al. An outbreak of amebiasis in an institution for the mentally retarded in Japan. Jpn J Med Sci Biol 1989; 42: 63-76.
- Nagarura R, Tachibana H, Kaneda Y, et al. Amebiasis in institutions for the mentally retarded in Kanagawa Prefecture, Japan. Jpn J Med Sci Biol 1990; 43: 123–31.
- 12. Scaglia M, Gatti S, Bruno A, Cevini C. Autochthonous amoebiasis in institutionalized mentally-retarded patients: preliminary evaluation of isoenzyme patterns in three isolates. Ann Trop Med Parasitol 1991; 85: 509–13.
- 13. Chiu JP, Wei SF, Chen KT, et al. Preliminary report of an outbreak of amebic dysentery in a primary school in Taichung City. Epidemiol Bull 1994; **10**: 74–88.
- Chou CH, Cheng MY, Liu KH, et al. Seroepidemiological survey on *Entamoeba histolytica* in Lanyu District. Epidemiol Bull 1994; 10: 23–7.
- 15. Ong SJ, Cheng MY, Liu KH, Hrong CB. Use of the ProSpecT<sup>®</sup> microplate enzyme immunoassay for the detection of pathogenic and non-pathogenic *Entamoeba histolytica* in faecal specimens. Trans R Soc Trop Med Hyg 1996; 90: 248−9.

- 16. Ash LR, Orihel TC. Parasites: A guide to laboratory procedures and identification. Chicago: ASCP Press, 1987; 99–116.
- 17. Arber S, Ginn J. Gender and later life. London: SAGE Inc., 1991; Appendix B.
- Diamond LS, Clark CG. A redescription of *Entamoeba histolytica* Schaudinn, 1903 (Amended Walker, 1911) separating from *Entamoeba dispar* Brumpt, 1925. J Eukaryot Microbiol 1993; 40: 340–4.
- Bracha R, Diamond LS, Ackers JP, Burchard GD, Mirelman D. Differentiation of clinical isolates of Entamoeba histolytica by using specific DNA probes. J Clin Microbiol 1990; 28: 680–4.
- Clark CG, Diamond LS. Ribosomal RNA genes of 'pathogenic' and 'nonpathogenic' Entamoeba histolytica are distinct. Mol Biochem Parasitol 1991; 49: 297–302.
- Cruz-Reyes JA, Spice WA, Rehman T, Gisborne E, Ackers JP. Ribosomal DNA sequences in the differentiation of pathogenic and non-pathogenic isolates of *Entamoeba histolytica*. Parasitology 1992; 104: 239–46.
- Tannich E, Horstmann RD, Knobloch J, Arnold H. Genomic DNA differences between pathogenic and non-pathogenic *Entamoeba histolytica*. Proc Natl Acad Sci USA 1989; 86: 5118–22.
- Tannich E, Burchbard GD. Differentiation of pathogenic from nonpathogenic *Entamoeba histolytica* by restriction fragment analysis of a single gene amplified in vitro. J Clin Microbiol 1991; 29: 250–5.
- Gonzalez-Ruiz A, Haque R, Rehman T, et al. A monoclonal antibody for distinction of invasive and noninvasive clinical isolates of *Entamoeba histolytica*. J Clin Microbiol 1992; 30: 2807–13.
- 25. Strachan WD, Chiodini PL, Spice WM, Moody AH, Ackers JP. Immunological differentiation of pathogenic and non-pathogenic isolates of *Entamoeba histolytica*. Lancet 1988; i: 561–3.
- Sargeaunt PG. The reliability of *Entamoeba histolytica* zymodemes in clinical diagnosis. Parasitol Today 1987;
   3: 40–3.
- 27. Martinez-Garcia C, Gutierrez-Trujillo G, Sanchez-Parez E, et al. Efficacy of zymodemes of *Entamoeba histolytica* technique in an epidemiologic study and report of new zymodemes in Mexico. Arch Invest Med 1990: 21: 203–8.