

Amoebiasis in homosexual men

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SUMMARY The clinical, histopathological, and serological features of 35 homosexual men with infection with *Entamoeba histolytica* were studied and compared with a group of 35 non-infected homosexual men. Each isolate was of Zymodeme type I. Although there was no significant difference in the numbers of infected and non-infected men with gastrointestinal symptoms (48.6% and 28.6% respectively), the mean duration of symptoms was greater in men with amoebiasis ($p < 0.05$). The histology of the rectal mucosa was abnormal in 17 (63.0%) of the 27 men with amoebic infection only and in two (7.4%) of the 27 control subjects ($p < 0.001$). Serum antibodies reactive with *E. histolytica* were not shown in any patient.

Within the last decade the sexual transmission of organisms generally acquired by the faecal-oral route has become apparent. Reports from the United States of America and, less commonly, from other countries have detailed the occurrence of intestinal diseases in homosexual men. Enteric organisms which may be transmitted within this population, usually as the result of oro-anal and oro-genital contact include bacteria (*Shigella* spp, *Salmonella* spp, *Campylobacter* spp), viruses (hepatitis A, hepatitis B), protozoa (*Entamoeba histolytica*, *Giardia intestinalis*) and nematodes (*Enterobius vermicularis*).¹⁻⁴

Of particular interest is the transmission of *E. histolytica*. For many years there has been considerable debate regarding the pathogenicity of this protozoon.⁵ From a consideration of their isoenzyme patterns, Sargeant and his coworkers have classified *E. histolytica* isolates into 18 groups (zymodemes). Based on their association with dysentery or hepatic abscess formation these workers consider that only amoebae of zymodeme types II, VI, VII, and XI pathogenic.⁶

The present study was undertaken to determine if infection with *E. histolytica* in homosexual men was associated with intestinal pathology.

Methods

PATIENTS

Thirty five men with *E. histolytica* infection were

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studied. These men were identified during a survey undertaken to determine the prevalence of intestinal protozoa infection among a group of 350 homosexual men who attended consecutively the University Department of Genito-Urinary Medicine, Edinburgh Royal Infirmary during a 14 month period. As a comparison group, 35 homosexual men who had no evidence of gastrointestinal infection were investigated. These men were matched, as far as possible, with the infected men with respect to age, social class, number of years during which they had been sexually active, number of sexual partners within the preceding six months, the interval between their most recent sexual contact and examination, past history of sexually transmitted diseases, and history of travel to tropical and subtropical areas.

The study was approved by the Ethics Committee of the South Lothian District of the Lothian Health Board.

CLINICAL METHODS

A careful history was taken from each patient who was asked specifically about the occurrence and duration of diarrhoea (defined as the passage of at least three liquid stools for at least 24 hours) pruritus ani, ano-rectal bleeding and anal discharge. Patients were also asked about travel to tropical and subtropical areas.

After general physical examination and the taking of the appropriate specimens to confirm or exclude genital or pharyngeal sexually transmitted infection a plastic disposable proctoscope (Welch-Allen) lightly lubricated with KY jelly (Johnson &

Johnson, Slough UK) was passed and material for culture for *Neisseria gonorrhoeae*, *Chlamydia trachomatis* and *Herpesvirus hominis* taken as described elsewhere.⁷ The proctoscope was withdrawn and with the patient's consent sigmoidoscopy was undertaken. Rectal biopsies were taken from at least one site.

A single stool sample was obtained from each patient. In men with gastrointestinal symptoms and in those who acted as control subjects in the analysis of the clinical and pathological features of amoebiasis, three consecutive faecal specimens were examined.

Serum from each patient was examined for treponemal antibodies and for hepatitis B surface antigen. Sera from 20 men were tested for amoebic antibody.

In five patients with amoebiasis who had had troublesome diarrhoea for at least six months, radiological examination of the colon by double contrast barium enema was undertaken before treatment.

MICROBIOLOGICAL METHODS

Culture of rectal material for *N gonorrhoeae*, *C trachomatis*, and *Herpesvirus hominis* was as described.⁸⁻¹⁰ Faecal specimens from men with amoebiasis and from control subjects were cultured for *Shigella* spp, *Salmonella* spp and *Campylobacter* spp. Stool samples from eight and four men with amoebiasis were cultured for *Clostridium difficile*¹¹ and examined by electron microscopy for the presence of virus particles respectively.

Fresh specimens only from men with diarrhoea were examined microscopically for trophozoites; faeces from every patient were concentrated by the formal-ether method¹² and examined for protozoal cysts. The identity of cysts suspected of being *E histolytica* was confirmed by the staff of the Department of Medical Protozoology, London School of Hygiene and Tropical Medicine. Culture for *E histolytica* was undertaken as a routine using Robinson's medium.¹³ Isoenzyme studies¹⁴ were performed on *E histolytica* trophozoites which were cultured from cysts in stool samples sent from Edinburgh to the London School of Hygiene and Tropical Medicine.

HISTOPATHOLOGICAL METHODS

These methods are described elsewhere.¹⁵ Histological sections were examined by one of us (HMG) who was unaware of the clinical diagnosis. The grading system used was that of Dickinson *et al.*¹⁶ Briefly, grade A is normal, grade B is associated with a mild to moderate infiltration of chronic inflammatory cells in the lamina propria and

grade C refers to a predominantly acute inflammatory reaction in the lamina propria with degenerative or reactive changes in the epithelium of the superficial part of the crypts and mucosal surface.

INDIRECT IMMUNOFLUORESCENT ANTIBODY TEST FOR THE DETECTION OF SERUM ANTIBODIES AGAINST *E HISTOLYTICA*

The test system was that described by Jeanes.¹⁷ As antigen, trophozoites of *E histolytica* cultured in Robinson's medium from the faeces of three patients were used. FITC-labelled rabbit anti-human IgG and IgM sera were obtained from Wellcome Laboratories (Beckenham, Kent, UK). Sera which were not heat-inactivated, were used at dilutions of 1/16 to 1/128.

TREATMENT OF PATIENTS WITH AMOEBIASIS

Patients with symptomatic amoebiasis were treated with metronidazole 200 mg by mouth every eight hours for 10 days. These patients were also given diloxanide furoate in an oral dosage of 500 mg eight hourly for 10 days.

Asymptomatic cyst carriers were treated with diloxanide furoate only in the above dosage.

Stool specimens for parasitological examination were obtained from patients four, eight, and 12 weeks after treatment. Cure was assumed if cysts of *E histolytica* were not detected in these samples. Reinfection was considered to have occurred if *E histolytica* was later shown in faeces of men whose three months post-treatment specimens had not contained amoebae.

STATISTICAL METHODS

In the statistical analysis of the data the χ^2 test with Yates' correction, the Student's *t* test and the McNemar test were used.

Results

DEMOGRAPHIC DATA

The mean age of the men with amoebiasis was 30.3 years (range 18-55 years). All but one man, an Iranian, were resident in the Lothian Region of Scotland.

The social class distribution of these men was: Social Class I 5; II 7; III 14; IV 6; V 3.

With the exception of two divorced men, the patients had had exclusively homosexual contacts. These men had been sexually active for a mean of 10.4 years (range two to 23 years). The mean number (range) of sexual partners within the preceding six months was 3.7 men (one to 23 men). Twenty of the 35 men had been treated on at least

one occasion previously for sexually transmitted diseases (but not for amoebiasis). Nineteen men had travelled to tropical or subtropical areas within the preceding five years.

ISOENZYME CHARACTERISTICS OF THE ISOLATES OF *E. HISTOLYTICA*

Isolates from 18 men were available for isoenzyme characterisation. Each was of zymodeme type I.

CONCOMITANT ENTERIC INFECTIONS

Neisseria gonorrhoeae was isolated from the rectum of one man with amoebiasis; *C. trachomatis* from that of another man, and *Herpesvirus hominis* from a third man. No other enteric pathogens were isolated from the 35 men.

Concomitant enteric protozoal infection was found in 16 (45.7%) men (Table 1). Eight, five, and three men with amoebiasis also harboured one, two, and three other species of intestinal protozoa respectively.

CLINICAL AND HISTOPATHOLOGICAL FEATURES OF THE STUDY GROUP

Symptoms and signs

Seventeen (48.6%) men with amoebiasis and 10 (28.6%) non-infected men complained of diarrhoea ($\chi^2=2.17$; $p>0.1$). The mean duration of the diarrhoea in men with amoebiasis was 25.8 weeks (range four days to two years) and 2.7 weeks (range one day to three months) in non-infected men ($t=2.14$; <0.05). After treatment of the men with amoebiasis the diarrhoea resolved. Four and three men with and without amoebiasis respectively complained of anal discharge; one man had concomitant Herpes simplex infection. Of the 27 men who had amoebiasis only and who consented to sigmoidoscopy, the macroscopic appearance of the rectal mucosa was normal in 23 men and in each of the non-infected men. In six infected men, one of whom had had concomitant chlamydial infection, the rectal mucosa appeared abnormal in that the vascular pattern was not discernible and there was oedema and contact bleeding; mucosal ulceration was not seen.

Table 1 *Concomitant enteric protozoal infection in 35 men with amoebiasis*

Protozoon	Infected men (no %)
<i>Entamoeba hartmanni</i>	3 (8.6)
<i>Entamoeba coli</i>	12 (34.3)
<i>Endolimax nana</i>	6 (17.1)
<i>Iodamoeba butschlii</i>	4 (11.4)
<i>Giardia intestinalis</i>	1 (2.9)
<i>Retortamonas intestinalis</i>	1 (2.9)

Histopathological findings in the rectal mucosa

Rectal biopsies were taken from 27 men who had amoebiasis but in whom no other intestinal pathogen was identified. Table 2 shows the results of the histological examination of these biopsies and of those from the control subjects. Amoebae were not detected within the rectal mucosa.

The histology of the rectal mucosa was abnormal significantly more frequently in men with amoebiasis than in the controls ($p<0.001$).

Radiological examination of the colon and rectum

By double contrast barium enema examination, the colon and rectum appeared normal in the five men in whom this investigation was undertaken.

IMMUNOFLUORESCENT TEST FOR AMOEBIC ANTIBODIES

Antibodies reactive with *E. histolytica* were not detected in the 20 sera tested at dilutions of 1/16 to 1/128.

RESULTS OF TREATMENT

Thirty men were reviewed three months after treatment. Cysts were not shown in stool samples from any of these men. One patient presented 12 months after treatment when he had again developed diarrhoea. Trophozoites and cysts of *E. histolytica* were again shown in the faeces and he was successfully retreated with metronidazole and diloxanide furoate. His regular sexual partner was investigated for the first time and cysts of *E. histolytica* were found in the faeces.

Nine men whose pretreatment rectal histology was graded B (seven men) and C (two men) consented to repeat biopsy. The mean interval between completion of therapy and biopsy was 3.6 months (range five weeks to six months). In each case the histology was graded A.

Discussion

It has been known for many years that an individual

Table 2 *Histology of the rectal mucosa of men with amoebiasis and of non-infected men*

Histological* grading	Biopsies included in each grading (no)	
	From men with amoebiasis	From non-infected men
A	10	25
B	14	2
C	3	0
Total	27	27

* See text for details of grading system

may harbour *E histolytica* in the bowel without developing gastrointestinal symptoms. Indeed, Morton *et al*¹⁸ found cysts of *E histolytica* in the stools of 16 of 1000 symptomless air apprentices aged 17 to 18 who had never been outside the United Kingdom. Some authors suggest that amoebae survive in the lumen of the bowel without producing damage to the mucosa;¹⁹ others claim that careful examination of the intestine will always show some, albeit microscopic lesions.²⁰ Based on the organism's ability or inability to produce hepatic abscesses in hamsters, virulent and avirulent strains of *E histolytica* are known to exist.²¹ McGowan *et al*²² have recently isolated a heat labile cytotoxin from highly virulent strains of axenically cultivated *E histolytica* and shown that avirulent strains produced little or none. Sargeant *et al*⁶ claim that only a restricted number of strains with particular isoenzyme characteristics are associated with dysentery and hepatic abscess formation.

As amoebiasis (that is the condition of harbouring *E histolytica* with or without clinical manifestation) is common among homosexual men, it is important to assess the significance of this condition. Is the protozoan a commensal, its presence merely indicating a previous breakdown of hygienic conditions, or does it exert a pathogenic role?

The isolates from our patients were not tested for virulence in the hamster. Isoenzyme electrophoresis, however, was performed on isolates from 18 men and in each case the zymodeme was of type I; strains of this type are considered by Sargeant *et al*⁶ to be non-pathogenic. Indeed, there was no clinical radiological, (five cases) or histological evidence of intestinal invasion. The results of serological testing tend to confirm these findings. By immunofluorescence, amoebic antibodies can be found at a titre of >64 in the sera of 95% and 75% of patients with hepatic abscesses and acute intestinal amoebiasis respectively;¹⁵ sera from about 30% of asymptomatic cyst carriers contain antibodies at a titre of 16.¹⁵ In our series, antibodies were not detected in any serum at this dilution.

Gastrointestinal symptoms, especially diarrhoea, are common among homosexual men³ and there was no significant difference in the prevalence of diarrhoea between men with amoebiasis and the non-infected men. The duration of diarrhoea, however, was significantly greater in the former group, and the diarrhoea resolved after treatment.

The histopathology results were interesting. Although the histological appearances of intestinal amoebiasis vary considerably,²³ a common feature is non-specific proctocolitis.²⁴ Proctitis in homosexual men can be caused by a variety of micro-organisms.² In the analysis of the histological findings, we

selected patients in whom *E histolytica* was the only recognised pathogen (or potential pathogen) present. The control subjects were matched as carefully as possible with the infected men, although some parameters were not open to objective assessment. It was clear that proctitis was found more frequently in men with amoebiasis than in the non-infected group. Although the resolution of the proctitis after treatment might lend support to the hypothesis that these protozoa were exerting a pathological effect we have found that acute and chronic proctitis in homosexual men with no detectable infection may resolve spontaneously (HMG – personal observations).

Although 46% of men with amoebiasis had some concomitant enteric protozoal infection, it was thought unlikely that this played a part in the aetiology of the proctitis. With the exception of *G intestinalis* which was found in the faeces of only one patient, these protozoa are regarded as harmless commensals.²⁵ In a study of similar design to the present, we found no relationship between the presence of *E coli*, *E hartmanni*, *Endolimax nana* or *Iodamoeba butschlii* and proctitis (unpublished data).

In conclusion, we feel that homosexual men in whom *E histolytica* is found should be treated as described.

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References

- 1 Felman YM, Ricciardi NB. Sexually transmitted enteric diseases. *Bull NY Acad Med* 1979; **55**: 533–9.
- 2 Quinn TC, Corey L, Chaffee RG, Schuffler MD, Holmes KK. Campylobacter proctitis in a homosexual male. *Ann Intern Med* 1980; **93**: 458–9.
- 3 Phillips SC, Mildvan D, William DC, Gelb AM, White MC. Sexual transmission of enteric protozoa and helminths in a venereal disease clinic population. *N Engl J Med* 1981; **305**: 603–6.

- 4 Waugh MA. Threadworm infestation in homosexuals. *Trans St John's Hosp Dermatol Soc* 1972; **58**: 224-5.
- 5 Anonymous. Pathogenic *Entamoeba histolytica*. [Editorial] *Lancet* 1979; **1**: 303.
- 6 Sargeant PG, Jackson TFHG, Simjee A. Biochemical homogeneity of *Entamoeba histolytica* isolates especially those from liver abscess *Lancet* 1982; **1**: 1386-8.
- 7 McMillan A, Lee FD. Sigmoidoscopic and microscopic appearance of the rectal mucosa in homosexual men. *Gut* 1981; **22**: 1035-41.
- 8 Young H. Cultural diagnosis of gonorrhoea with modified New York City (MNYC) medium. *Br J Vener Dis* 1978; **54**: 36-40.
- 9 Hunter JM, Smith IW, Peutherer JF, MacAulay A, Tuach S, Young H. *Chlamydia trachomatis* and *Ureaplasma urealyticum* in men attending a sexually transmitted diseases clinic. *Br J Vener Dis* 1981; **57**: 130-3.
- 10 Smith IW, Peutherer JF, Robertson DHH. Characterisation of genital strains of *Herpesvirus hominis*. *Br J Vener Dis* 1973; **49**: 385-90.
- 11 Brettle RP, Poxton IR, Murdoch JMcC, Brown R, Byrne MD, Collee JG. *Clostridium difficile* in association with sporadic diarrhoea. *Br Med J* 1982; **284**: 230-3.
- 12 Ridley DS, Hawgood BC. The value of formol-ether concentration of faecal cysts and ova. *J Clin Pathol* 1956; **9**: 14-16.
- 13 Robinson GL. The laboratory diagnosis of human parasitic amoebae. *Trans Roy Soc Trop Med Hyg* 1968; **62**: 285-94.
- 14 Sargeant PG, Williams JE. Electrophoretic isoenzyme patterns of *Entamoeba histolytica* and *Entamoeba coli*. *Trans Roy Soc Trop Med Hyg* 1978; **72**: 164-6.
- 15 McMillan A, McNeillage G, Gilmour HM, Lee FD. The histology of rectal gonorrhoea in men, with a note on anorectal infection with *Neisseria meningitidis*. *J Clin Pathol* 1983; **36**: 511-4.
- 16 Dickinson RJ, Gilmour HM, McClelland DBL. Rectal biopsy in patients presenting to an infectious diseases unit with diarrhoeal disease. *Gut* 1979; **20**: 141-2.
- 17 Jeanes AL. Evaluation in clinical practice of the immunofluorescent amoebic antibody test. *J Clin Pathol* 1969; **22**: 427-9.
- 18 Morton TC, Neal RA, Sage M. Indigenous amoebiasis in Britain. *Lancet* 1951; **1**: 766-9.
- 19 Meloney HE. Some unsolved problems of amoebiasis. *Am J Trop Med Hyg* 1957; **6**: 487-98.
- 20 Craig CF. Pathology of amebiasis in carriers. *Am J Trop Med* 1932; **12**: 285-99.
- 21 Mattern CFT, Keister DB. Experimental amebiasis. II Hepatic amebiasis in the newborn hamster. *Am J Trop Med Hyg* 1977; **26**: 402-11.
- 22 McGowan K, Deneke CF, Thorne GM, Gorbach SL. *Entamoeba histolytica* cytotoxin; purification, characterization, strain virulence and protease activity. *J Infect Dis* 1982; **146**: 616-25.
- 23 Prathap K, Gilman R. The histopathology of acute intestinal amebiasis. *Am J Pathol* 1970; **60**: 229-39.
- 24 Pittman FE, Hennigar GR. Sigmoidoscopic and colonic mucosal biopsy findings in amebic colitis. *Arch Pathol* 1974; **97**: 155-8.
- 25 Albach RA, Booden T. Amoebae. In: Kreier JP, ed. *Parasitic protozoa*. Vol. II. New York, London: Academic Press, 1978: 455-506.