

## SIMULTANEOUS CAECAL AND LIVER INFECTIONS IN HAMSTER, MOUSE AND GUINEA-PIG BY ORAL FEEDING OF *ENTAMOEBIA HISTOLYTICA* CYSTS

M. S. CHAUHAN and S. R. DAS

Division of Microbiology, Central Drug Research Institute, Lucknow 226 001, India.

### ABSTRACT

Both caecal ulceration and liver abscess were simultaneously produced in cholesterol-treated hamster, mouse and guinea-pig by oral feeding of *Entamoeba histolytica* cysts. On an average 43% hamsters, 35% mice and 50% guinea-pigs developed dual infections whereas cholesterol-untreated but amoebae-inoculated control animals developed only caecal infection by repeated oral feeding of *E. histolytica* cysts.

### INTRODUCTION

NATURAL and experimental *E. histolytica* infection using cysts have been studied by many workers. Experimental intestinal amoebiasis has been produced successfully in rats by the oral feeding of *E. histolytica* cysts<sup>1,2</sup>. Singh<sup>3</sup> reported autoinfection in rats with *E. histolytica* cysts.

The present studies are aimed at the development of a suitable animal model especially for the production of hepatic amoebiasis by oral feeding of *E. histolytica* cysts to cholesterol-treated laboratory animals.

### MATERIALS AND METHODS

Guinea-pigs weighing 100–150 g, hamsters 40–45 g and mice 19–20 g maintained in CDRI, animal house and kept on uniform diet and water throughout the experiment, were used. Before starting the experiment stool samples of these animals were checked microscopically in normal saline and Lugol's iodine solution.

*E. histolytica* cysts were collected from human and monkey stool samples following formalin-ether sedimentation technique<sup>4</sup>. Mature *E. histolytica* cysts were confirmed by the presence of 4 nuclei, bar-shaped chromatid body, glycogen mass and double wall in a cyst under Lugol's iodine wet preparation.

Cholesterol was given to each animal at a dose of 1.4 g/kg body weight. Cholesterol was fed orally suspended in water and injected intramuscularly dissolved in sesame oil, daily for two days before oral feeding of *E. histolytica* cysts and continued till the termination of the experiment (25–40 days).

Cholesterol was introduced to each animal through a rubber catheter attached to a 1 ml syringe. After two days of cholesterol treatment, *E. histolytica* cysts numbering approximately 2,00,000

cysts/ml/animal were fed orally to each animal for five to eight days and the feeding of cholesterol and normal diet was continued till the termination of the experiment. The control group of animals was fed with normal diet alone. *E. histolytica* cysts were fed orally to the control animals without cholesterol. Animals looked sickly and stopped eating food by 25–40 days and these were sacrificed to detect the dual infection. The scoring of liver lesion and caecal ulceration was done as described earlier<sup>5,6</sup>. Infection in animals was confirmed by the tissue smear preparation in normal saline, culturing in Robinson's medium<sup>7</sup> and by histopathological examination.

### RESULTS

Cholesterol was useful in the production of dual infection in hamster, guinea-pig and mouse introduced orally with *E. histolytica* cysts. Hamsters treated with cholesterol, both orally and intramuscularly, produced 33.3–50.0% liver lesion and 100% caecal lesions (dual infection) with an average caecal score of 3 and an average liver lesion of grades 2.5 to 3.0 between two batches of hamsters. Hamsters fed orally with cholesterol produced 33.3 to 40% infection with an average caecal score of 2 and liver score 1 to 2 between two other batches of animals. All cholesterol-treated hamsters showed caecal infection but in cholesterol-untreated controls caecal lesion was observed in 80% animals but none of them showed liver lesion.

In the case of cholesterol-treated guinea-pigs 50% had developed dual infection with an average caecal score of 3 and a liver score of 2.5. Caecal lesion was observed in all treated animals but only 80% untreated control guinea-pigs developed 2 grade caecal lesion only without liver infection. Similarly, 20–50% cholesterol-treated mice had developed dual infection with an average caecal score of 2 and

a liver score of 1.5–2. None of the control mouse had developed liver lesion; however, caecal lesion of 2 grade was found in 80% animals. The cultures of infected liver and caecal tissues were positive for *E. histolytica* trophozoites.

### DISCUSSION

*E. histolytica* infections both in liver as well as in caecum have been reported in hamsters<sup>8</sup>, rats<sup>9</sup> and guinea-pigs<sup>10,11</sup>, through intracaecal and intrahepatic inoculation of virulent *E. histolytica* trophozoites. These are not the natural routes of human amoebic infection. Amoebiasis in man generally occurs by the intake of water and food contaminated with *E. histolytica* cysts; as such the oral route and sometimes the rectal route are considered to be the main routes of amoebic infection. Quadri *et al*<sup>12</sup> produced experimental amoebiasis in mice which were treated with azathioprine, an immuno-suppressive agent, fed orally with *E. histolytica* cysts. Owen<sup>13</sup> reported caecal infection of rats and mice by oral inoculation of trophozoites of *E. histolytica*. There is no report where dual infection has been produced simultaneously in the same animal by oral feeding of *E. histolytica* cysts. Cholesterol has been found by several workers to induce invasiveness to non-invasive amoebae<sup>14,15</sup>. The present communication reports for the first time simultaneous caecal and liver infection in hamsters, mice and guinea-pigs by treating them with cholesterol and by oral feeding of *E. histolytica* cysts.

### ACKNOWLEDGEMENT

The authors thank Dr M. M. Dhar for his keen interest. Financial help from ICMR, New Delhi is thankfully acknowledged.

24 April 1987; Revised 20 July 1987

1. Tuchiya, H., *Am. J. Trop. Med. Hyg.*, 1939, **19**, 151.
2. McCowen, M. C. and Lawlis, J. F., *J. Parasitol.*, 1950, **36**, 25.
3. Singh, B. N., *Curr. Sci.*, 1973, **42**, 227.
4. Ritchie, L. S., *Bull. U.S. Army Med. Dept.*, 1948, **8**, 285.
5. Dutta, G. P., *Proc. Indian Nat. Sci. Acad.*, 1970, **B36**, 99.
6. Neal, R. A., *Trans. R. Soc. Trop. Med. Hyg.*, 1951, **51**, 313.
7. Robinson, G. L., *Trans. R. Soc. Trop. Med. Hyg.*, 1968, **62**, 285.
8. Jarumilinta, R. and Maegraith, B. G., *Ann. Trop. Med. Parasitol.*, 1961, **55**, 505.
9. Ishaq, M., Padama, M. C. and Habibullah, C. M., *Trans. R. Soc. Trop. Med. Hyg.*, 1980, **74**, 140.
10. Gill, N. J., Ganguly, N. K., Mahajan, R. C., Bhusnurmath, R. S. and Dilewari, J. B., *Indian J. Med. Res.*, 1983, **78**, 489.
11. Vinayak, V. K., Sawhney, S., Jain, P., Chuch, S. and Chakravorty, R. N., *Ann. Trop. Med. Parasitol.*, 1982, **76**, 309.
12. Quadri, G. S. A., Saleem, Y., Ishaq, M. and Habibullah, C. M., *IRCS. Med. Sci.*, 1985, **13**, 590.
13. Owen, D. G., *Trans. R. Soc. Trop. Med. Hyg.*, 1984, **78**, 160.
14. Biagi, F. F., Robledo, E., Servin, H. and Martuscelli, Q. A., *Am. J. Trop. Med. Hyg.*, 1962, **11**, 333.
15. Das, S. R. and Ghoshal, S., *Ann. Trop. Med. Parasitol.*, 1976, **70**, 439.

---

## ANNOUNCEMENT

---

### INDIAN INSTITUTE OF METALS

During the Silver Jubilee year of the Indian Institute of Metals, the National Metallurgists Day Awards were announced by the President, Dr V. S. Arunachalam.

The recipients were Dr Dipanker Banerjee, Scientist, DMRL, Hyderabad; Dr Chaitanyamoy Ganguly, Head, Radiometallurgy Division, BARC, Bombay; Dr R. V. Krishnan, Scientist, NAL, Bangalore; Mr P. Sriram, Director, Rapsi Engineer-

ing Industries, Bangalore; Mr S. Bhattacharya and Mr D. N. Gorai of the Bhilai Steel Plant; Mr V. K. Lakshmanan of TISCO, Jamshedpur and Dr Lokeshsinghal and Mr Basudev Roy, RDCIS, SAIL, Ranchi.

Each award carries a cash prize of Rs. 7,500 and scroll of honour. The awards were presented to the scientists by Dr S. Varadarajan, Chief Consultant, Planning Commission, New Delhi.

---