

Clinical and epidemiological data of patients with clonorchiasis

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Abstract

AIM: To study the clinical and epidemiological features of patients with clonorchiasis so as to provide scientific evidences for the diagnosis and prevention of clonorchiasis.

METHODS: Stools from 282 subjects suspected of having clonorchiasis were examined for helminth eggs with modified Kato's thick smear and sedimentation methods, and their sera were tested for HAV-DNA, HBV-DNA, HCV-RNA, HDV-RNA and HEV-RNA with polymerase chain reaction (PCR). Clinical symptoms of patients with clonorchiasis only were analyzed, and their blood samples were tested for circulating antigen (CAg) with Dot-ELISA, eosinophilic granulocyte count, and alanine aminotransferase (ALT). Meanwhile, they were asked to provide data of occupation, eating habit, hygienic habit and knowledge of clonorchiasis. In addition, the ecosystem of the environment in epidemic areas was surveyed.

RESULTS: Among the 282 patients, 61 (21.43%) were infected with *clonorchis sinensis* only, 97 (34.64%) were co-infected with *clonorchis sinensis* and other pathogens, 92 (32.86%) were infected with hepatitis virus only and 31 (11.07%) neither with *clonorchis sinensis* nor hepatitis virus. Among the 61 patients with clonorchiasis only, there were 14 (22.95%) subjects with discomfort over hepatic region or epigastrium, 12 (19.67%) with general malaise or discomfort and inertia in total body, 6 (9.84%) with anorexia, indigestion and nausea, 4 (6.56%) with fever, dizziness and headache (6.56%), and 25 (40.98%) without any symptoms; sixty one (100%) with CAg (+), 98.33% (59/60) with eosinophilic granulocytes increased and 65.00% (39/60) with ALT increased. B-mode ultrasonography revealed 61 cases with dilated and thickened walls of intrahepatic bile duct, and blurred patchy echo acoustic image in liver. Twenty-six cases had stones in the bile duct, 39 cases had slightly enlarged liver with diffuse coarse spots in liver parenchyma. Twenty cases had enlarged gallbladder with thickened coarse wall and image of floating plaques, 9 cases had slightly enlarged spleen. By analysis of epidemiological data, we found that the ecologic environment was favorable for the epidemiology of clonorchiasis. Most patients with clonorchiasis were lack of knowledge about the disease. Their living environment, hygienic habits, eating habits and their occupations were the related factors that caused the prevalence of the disease.

CONCLUSION: The clinical symptoms of clonorchiasis are non-specific, and the main evidences for diagnosis of clonorchiasis should be provided by etiologic examination,

B-mode ultrasonography and clinical history. The infection of *clonorchis sinensis* is related to occupations, bad eating habits and lack of knowledge about prevention of the disease.

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INTRODUCTION

Clonorchiasis, a food-borne infection by eating raw fish contaminated with *Clonorchis sinensis* metacercariae, is more frequently found in individuals along the great rivers and streams in areas of China, northern Vietnam, Korea and other Eastern Asia countries^[1-8]. After oral infection, *C. sinensis* often engulfs the biliary epithelial tissues and blood cells in the intestine, and causes cholangitis with marked eosinophilic infiltration, biliary adenomatous hyperplasia, bile duct obstruction, and subsequently cholangiofibrosis^[9-21]. In an effort to provide scientific evidences for diagnosis of this disease, clinical and epidemiological data of patients with clonorchiasis were analyzed in this study.

MATERIALS AND METHODS

Subjects

Two hundred and eighty-two individuals suspected of being infected with clonorchiasis were involved in this study, including 187 males and 95 females. Their age distribution was as follows: 29 aged 10-16 years, 182 aged 17-55 years and 71 aged 55-67 years.

Methods

Etiological examination Stools from 282 subjects were examined for helminth eggs with modified Kato's thick smear and sedimentation methods, and sera of them were tested for HAV-DNA, HBV-DNA, HCV-RNA, HDV-RNA and HEV-RNA with polymerase chain reaction (PCR).

Analysis of clinical symptoms Clinical symptoms of the patients only with clonorchiasis diagnosed by microscopic fecal examinations were expressed as percentages according to the formula (the number of patients with some symptoms/the total number of the patients with clonorchiasis)×100% and subsequently statistical analysis was conducted.

Laboratory examination Blood samples of patients only with clonorchiasis were detected for circulating antigen (CAg) with Dot-ELISA, counted for eosinophilic granulocytes, and tested for alanine aminotransferase (ALT). The number of eosinophilic granulocytes above $0.5 \times 10^9/L$ was regarded as increased, and the value of alanine aminotransferase (ALT) above 30 u/L was judged as abnormal.

Examination by B-mode ultrasonography Abdominal part of the patients only with *C. sinensis* eggs in their stools was examined by B-mode ultrasonography.

Analysis of epidemiological data A questionnaire was given to the patients only with clonorchiasis, including occupation, eating habits, hygienic habits and knowledge about prevention

of clonorchiasis. In addition, we surveyed the ecosystem environments in the epidemic areas.

RESULTS

Etiological examination

Of the 282 individuals investigated, 21.43% (61/282) were confirmed to be infected with *C. sinensis* only by etiological examination, 34.64% (97/282) were infected with *C. sinensis* and other pathogens, 32.86% (92/282) were infected with hepatitis virus only, and 11.07% (31/282) with neither *C. sinensis* nor hepatitis virus (Table 1).

Table 1 Results of etiological examination

Types of pathogens	Case numbers	Percentages (%)
Infection of <i>C. sinensis</i> only	61	21.63
Infection of <i>C. sinensis</i> and other pathogens	97	34.40
Infection of <i>C. sinensis</i> and other helminth	32	11.35
Infection of <i>C. sinensis</i> and hepatitis virus	24	8.51
Infection of <i>C. sinensis</i> , other helminth and hepatitis virus	41	14.54
Infection of hepatitis virus only	92	32.62
Neither with <i>C. sinensis</i> nor hepatitis virus	32	11.35
Totally	282	100

Analysis of clinical manifestations

Among the 61 patients with clonorchiasis only, 14 (22.95%) subjects had discomfort over hepatic region or epigastrium, 12 (19.67%) had general malaise or discomfort, 6 (9.84%) had anorexia, indigestion and nausea, 4 (6.56%) had fever, dizziness and headache, and 25 (40.98%) had not any symptoms.

Laboratory examination

The results of laboratory examination of the 61 patients with clonorchiasis only showed that the positive rates of CAg were 100%, eosinophilic granulocytes increased in 98.33% (59/60) patients and the values of ALT increased in 65.00% (39/60).

Examination by B-mode ultrasonography

In B-mode ultrasonography, 61 cases had dilated and thickened walls of intrahepatic bile ducts. Blurred patchy acoustic image was seen in liver. Twenty-six cases had calculi of intrahepatic ducts, 39 cases had slightly enlarged liver with diffuse coarse spots in liver parenchyma. Twenty cases had enlargement of gallbladder with thickened rough wall, floating plaques with postural variation were seen in the gallbladder. Nine cases had slightly enlarged spleen.

Analysis of epidemiological data

Occupational structure The constituent ratios of the occupation of 61 patients with clonorchiasis only were: fishermen 37.7%, fishmongers 9.84%, workers in processing fish products 16.39%, officials 19.67%, students 9.84% and others 6.56%.

Eating habits According to the results of questionnaires sent to 61 patients with clonorchiasis only, there were 26 subjects with habits of eating raw fishes and shrimps, 41 eating undercooked fish and shrimps, 48 with kitchen knives used in both raw and cooked food, 30 grasping food directly after contacting live fish and shrimps with unwashed hands, and 11 drinking unclean or unboiled crude water.

Knowledge about prevention of clonorchiasis All patients, did not have any knowledge about metacercaria of *C. sinensis* that might be in fish and shrimps of Huaihe River System. They had no idea about the possible infection with *C. sinensis* after ingestion of raw and undercook fish and shrimps. Even

if infected with *C. sinensis*, symptoms such as epigastric pain, diarrhea, decreased appetite, dizziness, *etc.*, would occur.

Ecologic survey Among the sixty-one patients confirmed to be infected with *C. sinensis* only, 18 lived in villages along the Huaihe River, 19 lived near fishponds. We found a crowd of domestic animals coming and going on both banks of the river, such as chickens, ducks, dogs, pigs, *etc.*, and murine animals, hares and wild cats running in brushes and weeds near the river. In addition, wild ducks and birds, frolic king were looking for food on the river. Some mollusca, like *viviparidae*, *unionidar*, *bithyniidae*, *lymnaeidae*, and some fish and shrimps, such as *Pseudorasbora parva*, *Ctenopharyngodon idellus*, *Mylopharyngodon aethops*, *Cyprinus carpio*, *Lobstergrow* were found in waterweeds in the river.

About two thousand kinds of mollusca were collected and examined for cercaria of *C. sinensis*, and the positive rate was 0.25% (5/2000). When *Ctenopharyngodon idellus* was infected with cercaria, we could separate encysted metacercaria from its body. Similarly, the adult worms could be found from cats that were infected by encysted metacercaria. In addition, the positive rate of encysted metacercaria in fish and shrimps was 9.50% (19/200).

DISCUSSION

Clonorchiasis is endemic in East Asian countries and about seven million subjects were estimated to be infected with the fluke^[22,23]. After oral infection with *C. sinensis* metacercariae, the organism excysts in the digestive tract and migrates up to the bile duct where it grows into adult worms. Infected humans excrete faeces containing *C. sinensis* eggs, and human infections could be diagnosed by microscopic fecal examination with demonstration of the characteristic eggs^[24-26]. In the present study, we examined 282 subjects suspected of being infected with *C. sinensis* with methods of modified Kato's thick smear and sedimentation, and 61 individuals were found to be infected with *C. sinensis* only, while 97 were co-infected with *C. sinensis* and other pathogens, 92 were infected with hepatitis virus only, 31 had neither *C. sinensis* nor hepatitis virus. Etiological examination is a major method in the differential diagnosis of clonorchiasis and other diseases of the liver and biliary tract.

The prevalence of clinical symptoms was determined by the fluke quantity, frequency of infection and the hosts immunities. Subjects with mild infection of *C. sinensis*, often had symptoms or only some epigastric discomfort, anorexia, dyspepsia, *etc.* Subjects with moderate infection often had inertia, lassitude, dyspepsia, abdominal pain and other mild complaints, all of which could also be obviously observed in the subjects with severe infection and cirrhosis of liver. Portal hypertension and ascitis might finally occur^[27-32]. The outcome of this study showed there were 40.98% of patients without any symptoms and others with symptoms of epigastric discomfort, general malaise, inertia, anorexia, dyspepsia, nausea, fever, dizziness, headache, *etc.* had no specificity. Differential diagnosis should include schistosomiasis Japonica, fascioliasis and fasciolopsiasis.

During the development of *C. sinensis* in human body, CAg was produced and released into circulation. Generally, CAg might be detected in sera of the subjects three days after infection^[33,34]. For this reason, detection of CAg may be used to examine whether live worms exist in human body or not, and for check up of treatment effects. By Dot-ELISA, all of the 61 *C. sinensis* egg-positive individuals were positive, although 40.98% of them had no symptoms. So detection of CAg may provide an evidence for diagnosis of clonorchiasis. In addition, results from this study showed that eosinophilic granulocytes were increased in 98.33% patients, ALT was increased in 65% patients and image changes were found in

liver with B-mode ultrasonography in all patients. These are all helpful in the diagnosis of clonorchiasis.

Both natural and social factors affect transmission of *C. sinensis*. We investigated the ecologic system in the Huaihe River on the spot and saw various kinds of mollusca crawling on water plants and some fish and shrimps swimming in the river, all of which are intermediate hosts of *C. sinensis*. Near the river, a large number of domestic animals, livestock, murine animals, hares and wild cats are reservoir hosts. Moreover, the villagers have got accustomed to feeding fish with their own stools and domestic animals with raw fish and shrimps, which help *C. sinensis* to be transmitted from one host to another. On both banks of the river, simple toilets and pigsties are arranged everywhere, and the villagers brush chamber pots in the river without scruple. Terribly, they live on water untreated from the river. All of these natural environments and human activities contribute to overflowing of *C. sinensis* in this area.

The incidence of clonorchiasis was shown in this study to vary greatly and was related to occupations, and was higher in fishermen, fishmongers, workers in processing fish products, officials than in those with other occupations. Because of bad working and eating habits, they have chances to be infected with encysted metacercaria. Therefore, knowledge about prevention of clonorchiasis should be strengthened in these populations to correct their bad eating and health habits and to reinforce their protection consciousness.

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