Human chronic fascioliasis: a possible cause of unexplained abnormal liver tests

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ABSTRACT

Fascioliasis is a cause of hepatic disease. Hepatitis B and C viruses are important causative factors in chronic liver disease. In this study, the frequency of hepatitis B (HBV) and/or hepatitis C (HCV) in cases of chronic human fascioliasis is studied. Egg count, indirect haemagglutination test (IHAT), haemoglobin level, total leucocyte and eosinophil counts, serum bilirubin, serum aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase (ALP) and acid phosphatase (ACP) are performed. Serum γ-glutamyltransferase (GGT), arylsulphatase (ASA) and lipid peroxide levels are determined. Results showed that levels of the latter group of enzymes were increased significantly in cases of chronic fascioliasis. Therefore, determination of GGT, ASA and lipid peroxide should be added to the list of liver function test used to diagnose this disease. Hepatitis B was not detected in any of the 27 chronic fascioliasis patients studied, while HCV was found in only two (7%) cases. However, greater disturbance of biochemical parameters was seen in patients with combined fascioliasis and HCV infection.

KEY WORDS: Enzymes. Fascioliasis. Hepatitis B. Hepatitis C. Parasites.

Introduction

The incidence of human infection with *Fasciola* species is on the increase across Egypt, and fascioliasis is now designated as an important emerging diseases in both Egypt and the Mediterranean region.[1,2] In Egypt, it is one of the causes of hepatic disease,[3,4] and clearly it must be distinguished from hepatitis of other aetiology.[5]

Fasciola hepatica is a zoonotic liver fluke that can cause disease in humans but fascioliasis is uncommon. The disease has two stages: the hepatic and biliary stages. Several drugs are used during the hepatic stage but endoscopic retrograde cholangiopancreatography (ERCP) is particularly effective in the biliary stage.[6]

Human infection has been reported in Mexico, Cuba, Puerto Rico, Chile, Peru, Uruguay, Brazil, Argentina, the USA, Europe, Eastern Africa, Japan and Australia. The parasite's miracidium invades one of the various water snail hosts and infection results from ingestion of encysted metacercariae attached to raw watercress (*Nasturtium officinale*). Symptoms recorded from human cases included irregular fever, epigastric pain and abdominal tenderness, obstructive jaundice and leucocytosis with eosinophilia (up to 60%).

Specific diagnosis is based on recovery of eggs from stool samples or from biliary tract drainage. Treatment is with emetine hydrochloride (given intramuscularly). Bithionol is given orally at a dosage of 30–50 mg/kg on alternate days for 10–15 doses.[7] The detection of anti-fasciola IgG_4 isotype is a more sensitive and accurate immunodiagnostic tool for fascioliasis, but is not a useful marker for assessment of cure.[8]

Preventive measures include education of the public on the mode of transmission of the parasite and its life cycle. Also, advice on the proper preparation of fresh watercress prior to consumption is valuable.[7]

Hepatitis B (HBV) and hepatitis C (HCV) are important causative agents in the aetiology of chronic liver disease.[3,9] Infection with HBV is associated with a wide spectrum of liver damage that ranges from fulminant acute hepatitis to chronic HBV carriage with minimal hepatic change. This variation is thought to be due to differences in the intensity of the host immune response to the viruses.[3]

Hepatitis C virus is a single-strand RNA virus comprising at least 12 genotypes.[10] There is a high prevalence of HCV infection among patients with chronic liver disease and HCV-RNA correlates with liver pathology.[11] Overall HCV seropositivity is approximately 15.6% among working Egyptians aged 15–65 years.[12] In 1995, the World Health Organization assessed the prevalence of hepatitis B surface antigen (HBsAg) in Egypt at between 3–11%.[13]

The relationship between enzyme activity and disease has attracted a great deal of interest. Aminotransferases (ALT or AST), γ-glutamyltransferase, alkaline phosphatase (ALP) and acid phosphatase (ACP) show a high sensitivity for liver disease.[14–16] Furthermore, pronounced changes in arylsulphatase activity have been observed in pathological cases,[17] and the investigation of experimental fascioliasis has demonstrated malondialdehyde (MDA, lipid peroxides) generation and the formation of conjugated dienes as a result of peroxidative damage to liver microsomal membrane lipids.[18,19]

These findings, together with the reported link between schistosomiasis and HCV infection,[20–22] has stimulated study of the frequency of HBV and HCV infection in cases of

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Table 1. Statistical analyses of results of the parasitological and haematological findings

	Egg count per g stool	Reciprocal IHAT titre	Hb level g/dL	Leucocyte count/cm m	Eosinophil count/cm m	Bilirubin mg/dL
Control group 1 (n=27)						
Range	–ve	–ve (10)	12-14.8	5800-10100	40-330	0.1-10
Mean	–ve	–ve (1 in 80)	12.8	7741.7	281.9	0.43
± SD	-	-	1.7	1763.6	0.2	0.1
± SE	-	-	0.33	734.8	0.04	0.02
Fascioliasis group 2 (n=27)						
Range	24-552	– ve – 1280	8-13.5	4000-130000	346-13600	0.12-1.98
Mean	166.2	393.8	10.3	12785.2	1440.7	0.51
± SD	162.1	275.2	1.7	23645.5	2504.7	0.47
± SE	31.2	52.9	0.33	4547.2	481.7	0.09
t value	5.33	7.4	5.3	1.09	2.41	0.89
P value	< 0.005	< 0.005	< 0.005	< 0.05	< 0.025	> 0.1

t: Student's t-test value for the comparison between the fascioliasis patients and controls.
P <0.05 was considered significant.

Table 2. Statistical analysis of the results of the biochemical findings.

	AST (U/L)	ALT (U/L)	ALP (U/L)	ACP (U/L)	GGT (U/L)	ASA (nM/mL per h)	Serum lipid peroxide (nmol MDA/mL	HBsAg	HCV
Control group 1 (n=27)									
Range	4-12	6-11.5	75-110	4.5-11	9-20	5.29-8.21	1.99-4.2	–ve	–ve
Mean	7.11	8.3	87.9	8.4	14.2	6.08	2.95	-	-
± SD	2.5	2.0	13.8	2.0	3.7	1.7	0.46	-	-
± SE	0.48	0.38	2.65	0.38	0.71	0.33	0.09	-	-
Fascioliasis group 2 (n=27)									
Range	110-1000	140-1150	185-690	11-21	29-65	13-41	3.95-20.20	–ve	–ve/+ve
Mean	430	395.6	294.4	14.3	40.6	18.9	5.5	-	2 (+ve)
± SD	238.5	263.6	120.9	2.02	8.2	5.5	4.0	-	-
± SE	45.87	50.69	23.25	0.39	1.58	1.06	0.77	-	-
t value	9.22	7.64	8.82	10.93	15.3	11.55	3.27	-	-
P value	< 0.005	< 0.005	< 0.005	< 0.005	< 0.005	< 0.005	< 0.005	-	-

t: Student's t-test value for the comparison between the fascioliasis patients and controls.
P <0.05 was considered significant

chronic human fascioliasis. In the present study, serum bilirubin, serum AST, ALT, ALP and ACP are determined. In addition, GGT, arylsulphatase (ASA) and lipid peroxide level are included in order to assess whether or not these enzymes should be added to the list of liver function tests.

Materials and methods

The study was conducted on 54 adult males (age range: 18–40 years) divided into two groups: Group 1 comprised 27 normal healthy persons (the control group); and Group 2 comprised 27 patients with chronic fascioliasis. None of the participants in the study had received blood transfusion, antibiotics or medication in the three months preceding the study.

Urine and stool samples were examined by simple sedimentation to detect other parasitic infections and to count Fasciola eggs by the modified Kato Katz technique.[23] An indirect haemagglutination test (IHAT) was used to detect specific anti-Fasciola antibodies (Fumouse kit), and haemoglobin (Hb) level, total leucocyte and absolute eosinophil count were determined.[24]

Venous blood (8–10 mL) was collected from each individual after overnight fast and serum was separated. Assays were performed for bilirubin,[25] AST and ALT,[25] ALP,[26] ACP,[27] GGT,[28] ASA[29] activity and lipid peroxide.[30] Detection of HBsAg and HCV was by enzyme-linked immunosorbent assay (ELISA) techniques.[31,32]

Data obtained were analysed using a method reported previously[33] and values were expressed as mean ± standard deviation (±SD). Significance was P <0.05.

Results

Table I shows the parasitology and haematology findings and Table 2 shows the biochemical findings in Groups 1 and 2. Level of infection range from £96 eggs/g stool in 15 patients (55.6%), 120–504 eggs/g stool in nine patients (33.3%) and over >504 eggs/g stool in three patients (11%).

The IHAT for fascioliasis gave positive results (≥ 1/320) in 17 of those in Group 2 (62.96%). Haemoglobin level was lower than normal. Eight cases showed leucocytosis and a high eosinophil count was found in 22 cases.

All biochemical and some parasitology and haematology parameters were increased significantly in the two cases of combined fascioliasis and HCV infection compared to the results obtained with fascioliasis alone.

Discussion

This study of 27 patients with confirmed fascioliasis showed that the majority had low or moderate egg counts, while a high egg count (>500 eggs/g stool) was seen in only three cases. This supports the work of others.[34] A positive IHAT was found in 70% of the cases, which supports the work of Salem *et al*.[35] and Allam *et al*.,[36] who reported similar findings in cases of chronic fascioliasis.

Anaemia in fascioliasis cases was reported by El Aggan *et al*.[37] and was attributed to haemorrhage and blood loss in bile (haemobilia), the effect of parasite toxins on bone marrow, or parasites feeding on blood. Leucocytosis and eosinophilia – well known findings in fascioliasis – were noted in the present work, although others have shown them to be more pronounced during the acute phase.[38]

Bilirubin was within the normal range in all but the three high-count cases. This could have been due to obstruction of the bile ducts by adult worms, as these three cases harboured a large number of worms. Similar findings in fascioliasis with obstructive jaundice have been reported.[39]

Elevated ALT and AST were probably due to necrosis of the hepatic parenchyma that occurs during migration of immature worms.[40] Elevation of ALP in all cases may have been due to a narrowing of the biliary tract, including hyperplasia of the biliary epithelium, in response to the presence of worms. A regenerative process in the liver, following recovery from the acute phase of infection, may participate in this rise in ALP.[41] The microsomal enzyme GGT was elevated in all cases and this supports the different factors related to cholestasis, including biliary obstruction and injury to the bile duct epithelium.[14] Elevated ASA indicates the presence of an inflammatory process leading to increased lysosomal enzyme activities.[38,40,42]

Lipid peroxide level showed a significant increase when compared to the control group (Group 1). This points to a degree of liver cell damage that occurred during migration of the immature flukes that had yet to be corrected. Peroxidative damage to liver microsomal membrane lipids may be another explanation.[43]

Two cases of fascioliasis were found to have concomitant HCV infection. This would not indicate a causal association between fascioliasis and HCV infection, and is in contrast to reported cases of schistosomiasis in which HCV infection was present in 60% of cases.[44]

In summary, in the present study almost all cases of chronic fascioliasis showed significant increase in liver function tests (AST, ALT, ALP and ACP). In addition, GGT, ASA and lipid peroxide levels were increased significantly and therefore should be added to the list of liver function tests performed. Finally, there was no association between fascioliasis and HBV, and only a weak association with HCV, although greater disturbance of biochemical parameters was observed when fascioliasis and HCV infection were present in the same individual.

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