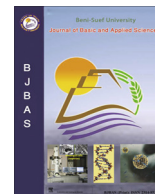


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Epidemiological determinants correlating hepatitis C and Schistosomiasis *mansoni* in one of Upper Egypt governorates

Samah S. Abdel Gawad^{a,*}, Enas Yahia Abu-Sarea^a, Lamiaa Saleh^b^a Departments of Parasitology, Faculty of Medicine, Beni-Suef University, Egypt^b Department of Public Health, Faculty of Medicine, Beni-Suef University, Egypt

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ABSTRACT

Schistosomiasis and hepatitis C virus (HCV) are endemic diseases with high prevalence in Africa especially in Egypt. Many sociodemographic and behavioral related determinants were implicated to be associated with schistosomiasis, HCV or coinfection. Recently, polymerase chain reaction (PCR) is used as a diagnostic tool as it is a sensitive and a specific method especially in early stage of infection. But, diagnosis of *S. mansoni* is depending on microscopic examination remains the most widely used direct diagnostic method in endemic area. The current study was carried out to determine the prevalence of schistosomiasis *mansoni*, HCV and coinfection if any among the studied population, to identify any associated factors for schistosomiasis *mansoni*, HCV or coinfection if any.

A descriptive cross sectional study was conducted on 400 participants, inhabitants in Beni-Suef Governorate, Upper Egypt. The studied population was screened for both schistosomiasis *mansoni* and HCV. They were subjected to fulfill a well-structured field tested interviewing questionnaire focusing on many suspected associated factors. Moreover, testing the performance characteristics of the used techniques was determined. The prevalence of schistosomiasis *mansoni* was (2.8%) and HCV was (42.5%) among the studied subjects. The study highlights on many behavioral and sociodemographic determinants significantly associated with HCV infection such as home crowding index, shaving at the community barber, sharing razors within the family, delivery at home and circumcision outside the health settings. Also, the study revealed that there are certain determinants associated with both schistosomiasis *mansoni* and HCV infection such as blood transfusion and liver cirrhosis. Studying the linked determinates with schistosomiasis and HCV is the cornerstone to plan and implement a preventive strategy in the Upper Egypt. Thus, further studying the associated environmental determinants in relation to schistosomiasis, HCV and coinfection is recommended in the Upper Egypt.

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1. Introduction

Schistosomiasis is an endemic disease with high prevalence and morbidity rates in many countries especially in Africa. It affects about 15–45% of Egyptian population (El-Khoby et al., 2000). Similarly, *S. mansoni* infected 60% of people in Northern and Eastern parts of Nile Delta and 6% in Southern part (Barakat, 2013). There are many risk factors associated with schistosomiasis such as age (<18 years), presence of infected family members, and contact with canal water (Dawaki et al., 2016). On the other hand, hepatitis C virus infection is a major health problem in the world of public health interest, with approximately 3% of people being infected with HCV (Marinho et al., 2014). In Egypt, the prevalence of HCV

in the Upper Egypt and Nile Delta was 28% and 26% respectively (Wanis, 2014). Karaca et al. (2006) reported that the most common risk factors associated with HCV were history of surgery, blood transfusion, dental procedure, abortion, long term hospitalization, hemodialysis, history of intravenous drug abuse, suspected sexual contact, history of occupational transmission, history of tattooing, percutaneous needle puncture and circumcision. Coinfection with schistosomiasis *mansoni* and hepatitis C is of medical importance as the patients with higher HCV RNA titers have, greater incidence of cirrhosis/hepatocellular carcinoma, histopathological activity and higher mortality rates than patients with single infections (Kamal et al., 2001). Diagnosis of *S. mansoni* is depending on microscopic detection of eggs in stool samples which remains the most widely used direct diagnostic method in endemic areas. There was a relation between eggs output, parasitic burden and morbidity parameters (WHO, 2002). Also, IHT is used for serological

* Corresponding author.

E-mail address: sayed.samah74@yahoo.com (S.S. Abdel Gawad).

diagnosis of schistosomiasis. These test show good sensitivity and specificity but high sensitivity may be due to cross reaction with other parasites (Yu et al., 2007). Recently, PCR is used as a diagnostic tool. It is a sensitive and a specific method especially in early stage of infection or in selection of the optimal treatment (Hussein et al., 2012). Diagnosis of *S. mansoni* is depending on microscopic detection of eggs in stool samples which remains the most widely used direct diagnostic method in endemic areas. There was a relation between eggs output, parasitic burden and morbidity parameters (WHO, 2002).

2. Material and methods

2.1. Study design, subjects and methods

A descriptive cross sectional study was conducted on 400 participants, inhabitants in Beni-Suef Governorate, Egypt, attending tropical medicine outpatients (Beni-Suef University Hospital, Beni-Suef General Hospital, and the Hospital of Health insurance) from – February 2016 to June 2017. The current study was carried out to detect and determine the prevalence of schistosomiasis (*mansoni*), hepatitis c virus (HCV) and HCV/schistosomiasis infection if any among the studied population, to identify any associated factors for both schistosomiasis and HCV. A well-structured field tested questionnaire form was prepared. All participants were interviewed to fulfill the designed well-structured interviewing questionnaire and their consents were obtained. The questionnaire was structured so that it was focusing on many suspected associated factors for both schistosomiasis and HCV. The studied determinants were supposed to be associated in schistosomiasis, HCV or both of them such as socio-demographic (age, gender, residence, housing characteristics, and educational level etc.). Certain behavioral factors associated with HCV infection were studied such as shaving at the community barber, sharing razors within the family, blood transfusion, and circumcision outside the health settings. Other studied behavioral factors were associated schistosomiasis such as exposure to contaminated water. A highlight on the medical history of the studied subjects was considered. All the studied subjects were subjected to laboratory investigations to detect the

presence of *Schistosoma mansoni*, HCV or schistosomiasis/HCV infection if any, and determine their prevalence in the studied population (Fig. 1).

2.2. Collection and processing of samples

Blood and stool samples were collected from all participants by tropical medicine outpatients. Blood samples were divided into two parts. The first part was examined for HCV by enzyme linked immunosorbant assay (ELISA) and positive cases were examined by PCR to detect active and inactive cases in clinical pathology department. The second part of blood samples and stool samples were sent to parasitology department, Beni-Suef University for parasitological examination. A portion of the collected stool sample was examined by microscopy and the remaining parts of the specimens were stored at -20°C for molecular studies. Blood samples were centrifuged to obtain serum for indirect haemagglutination test (IHT) and stored at -20°C .

2.3. Detection of *Schistosoma* by microscopy, IHT and PCR

S. mansoni eggs were detected in smear of stool specimens (2–10 mg) with suspension in saline. Sedimentation and concentration technique were used to detect patients with light infection. A commercial IHT kit sold by Fumouze Laboratories was used and the test was performed following procedures given by manufacturer. Genomic DNA of eggs was extracted from stool by using the FavorPrep Stool DNA Isolation Mini Kit (Favorgen Biotech Corporation, Ping-Tung 908, Taiwan), according to the manufacturer's instructions, after thermal shock of samples (five cycles of deep freezing and boiling in a water bath, each for 5 min), with prolongation of incubation for 1 h at 95°C after 56°C at 10 min. The extracted DNA of egg was preserved at -20°C until used for amplification. PCR was carried out by employing specific primers (forward: 5'-TTTTTGGTCATCTGA GGTGTAT3', and Sm.R: 5'-TGC AGATAAAGCCACCCCTGTG-3') previously designed by Webster and others, for specific amplification of COX 1 repeats of *S. mansoni* which amplify a fragment of 375 bp. The reaction volume of 25 μL consisted of 12.5 μL master mix, 0.1 μL Taq, 1 μM of universal forward primer, 1 μM Sm.R., 4 μL DNA and 6.4 μL of ddH₂O. The products were analyzed on agarose gel (1.5%) stained with ethidium bromide (10 mg/mL) and visualized with UV light.

2.4. Data analysis

The data had been coded to fit the program of statistical analysis (SPSS) Statistical Package for Special Sciences version 22 under windows 7. A random sample of 10% of cases was then selected and reviewed to ensure an adequate quality of data. Different variables have been summarized and housing crowding index has been calculated. Descriptive statistics were used such as frequency and percentage (For qualitative variables) and the mean and standard deviation (mean \pm SD) (for quantitative variables). A logistic regression analysis was done to detect risk factors associating with HCV and schistosomiasis infection and HCV by univariate analysis.

2.5. Ethical considerations

The studied groups were willing to participate in the study after explaining the purpose of the study. Permissions were officially obtained from the study settings. The filled data in the interviewing questionnaire was confidential. The respondents were assured that the socio demographic will be for identifying their characteristics not their identity. Regarding the laboratory investigations, all the participants were willing to be screened for schistosomiasis *mansoni* and HCV.

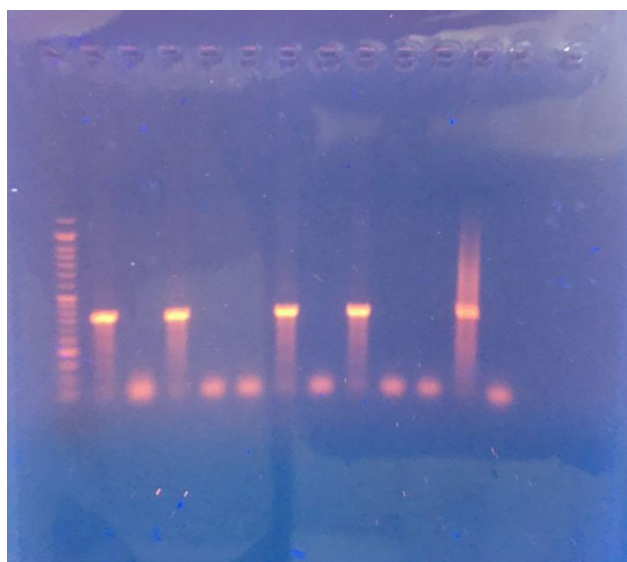


Fig. 1. Agarose gel electrophoresis and PCR results. L1: 50-bp DNA molecular weight marker. L2: positive control. L3: negative control. Lanes: 4, 7, 9 and 12: products of the polymerase chain reaction (PCR) targeting the COX1 gene of *Schistosoma mansoni* at 375 bp. Lanes: 5, 6, 8, 10, 11 and 13: negative samples of PCR.

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