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SEROEPIDEMIOLOGY AND SERODIAGNOSIS OF SCHISTOSOMIASIS AT PASTEUR INSTITUTE OF CÔTE D'IVOIRE FROM 2006 TO 2014: A RETROSPECTIVE CASE STUDY

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Abstract- Objectives: Schistosomiasis is a major parasitic disease affecting more than 200 million people in the developing world, and 400 million people are at risk for infection. The main objective of the study was to assess the prevalence of schistosomiasis through a serological method in a reference laboratory in Côte d'Ivoire. This retrospective study was conducted using laboratory data obtained from 2006 to 2014 at the Parasitology Unit of Institute Pasteur of Côte d'Ivoire. The serological routine method was the indirect hemagglutination assays schistosomiasis kit from Fumouze Laboratories (Levallois-Perret, France). Sociodemographic and clinical indications for the diagnosis of schistosomiasis were collected using a database of laboratory records spanning 2006 to 2014. Any difference in the prevalence of infections among different sexes or age groups were interpreted statistically. A total of 502 patients were attended Parasitology Unit from 2006 to 2014 for a serological diagnosis of schistosomiasis. The sex ratio was 1.9 male to female with a mean age of 35.1 years [3-85 years]. Prevalence of schistosomiasis was 23.3%, however, no statistically significant difference was observed according to age or sex ($p > 0.05$). High seroprevalence was observed in 2012 (45.7%) following by 2007 (42.9%) and less prevalence was observed in 2009 (3.6%). The seroprevalence of *Schistosoma* infection was found to be relatively high among the study population suggesting major interventions to reduce the burden of this disease in the country.

Keywords- Schistosomiasis, Diagnosis, Antibody-detection, Institut Pasteur Côte d'Ivoire.

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Introduction

Schistosomiasis is an important cause of disease in many parts of the world affecting more than 200 million people [1,2]. Majority of cases occur in sub-Saharan Africa with high morbidity and mortality [3]. Among the five of *Schistosoma* that infect humans, important are *Schistosoma mansoni* and *Schistosoma. Haematobium* [3]. Due to its geographical distribution and the affected populations, schistosomiasis is a neglected tropical disease and a neglected infection of poverty [4,5].

In Côte d'Ivoire, where both *S. mansoni* and *S. haematobium* are endemic and many people suffer from intestinal or urogenital schistosomiasis [6-9] with prevalence generally higher in the south (Mé region), the center (Marahoue) and the west (Tonkpi region).

A study carried out in Agboville and Azaguié in the south of the country, among school-aged children showed a very high prevalence of 85.3% and 53.8% for *S. haematobium* and *S. mansoni* respectively and also high levels of coinfections [10, 11].

Diagnostic approach for schistosomiasis depends upon the epidemiological situation. The diagnosis of schistosomiasis is usually based on clinical data associated with the detection of eggs in stool, urine, and/or rectal and bladder biopsy specimens in endemic area.

Microscopic methods to diagnose schistosomiasis are cheap, but are time consuming and are not sensitive enough to detect low burden infections, which resulted in false-negatives, although they have excellent specificity. Indirect immunofluorescent-antibody tests (IFATs), indirect hemagglutination assays (IHAs), and enzyme-linked immunosorbent assays (ELISAs) using different antigens, such as crude or purified adult worm antigen (AWA), soluble egg antigen (SEA), and cercarial antigen (CA) preparations have been developed [12-14].

Serological examinations are of limited use for the diagnosis of active infection, as large parts of the population may carry antibodies due to past infections in endemic area.

The slow reduction in the antibodies after treatment and the existence of cross-reactivity that result in false-positives are the major disadvantages of the immunological methods [15,16].

Meanwhile sensitive serological tests have the potential to increase diagnostic yield, especially in those with light infection who excrete few eggs [12,17].

S. mansoni and *S. haematobium* are rarely fatal, but cause long-term chronic morbidity [18].

Independent of the initial presentation, untreated schistosomiasis might lead to complications such as obstructive uropathies, hepatic fibrosis, or granulomatous cerebral lesions [19,20].

To prevent those late manifestations, any case of schistosomiasis should be detected and treated. Consequently, it is important to understand the burden of the infection to facilitate appropriate interventions. Therefore, the first approach for prevention and control is to diagnose the disease by applying different laboratory

methods.
Most of patients referred to Institute Pasteur of Côte d'Ivoire for serology test have yet a negative microscopic test (detection of eggs in stool, urine) test negative
The main objective of the study was to assess the prevalence of schistosomiasis through a serological method in a reference laboratory in Côte d'Ivoire.

Materials and Methods

This was a descriptive cross-sectional study based on the results of serodiagnosis of schistosomiasis. Patients from various hospitals of Abidjan were referred to the Parsitology Unit of Institute Pasteur of Côte d'Ivoire as a reference laboratory from 2006 through 2014 for antibody testing against *Schistosoma* infection because of suspicion by physicians that patients may have had schistosomiasis.
Abidjan is the economical capital of Côte d'Ivoire located in the south of the country. Abidjan is a cosmopolitan city with an estimated population of about 5 million of people. Most of them come from other parts of the country or neighboring countries. However, bulks of the patients were coming from Abidjan.
Since a decade the diagnosis of schistosomiasis at the Parasitology Unit was based on the detection of eggs in stool, urine, and/or rectal and bladder biopsy specimens. The Parasitology Unit has also established the serological method as an alternative test to detect human *schistosoma* infection to meet the growing demand of serological test from physicians. The routine serological method used in the laboratory was the IHA schistosomiasis kit from Fumouze Laboratories (Levallois-Perret, France). The test was performed following manufacturer's instructions [12,21]. In these way infections with *S. mansoni, S. haematobiumand S. intercalatum* can be diagnosed.
The results were evaluated with a cutoff titer of 1:320 as a more sensitive range.

Data collection

The study examined lab records from an office access computer designed database. The data covered the period ranging from January 2006 to December 2014 and focused on the serological diagnosis of schistosomiasis.
All sera sample records were examined to record the positive results for schistosomiasis infections. Sex, age and clinical indications data were also collected. Few data were available regarding origin region and profession of patients and were not collected.

Statistical analysis

For the statistical analysis, we used the free software Quantitative Parasitology v.3 [22]. The prevalence of infections was reported in proportions. The relative frequencies of schistosoma-positive individuals, determined by serology, were explored to look for differences between groups (sex and age) (Chi□squaretest ($\chi 2$)). Fisher's exact test (at risk 5%) was used for comparison of proportions. Data analysis was conducted using SPSS version 11.5 (SPSS Inc, Chicago, Illinois).

Ethical considerations

The study was conducted using the ethical standards. Ethical clearance to conduct this study was obtained from the Comite National d'Ethique de la Recherche of Côte d'Ivoire.

Results

Baseline characteristic of patients

A total of 502 patients (males and females) attended the Parasitology laboratory of Institute Pasteur of Côte d'Ivoire for serological diagnosis of schistosomiasis from 2006 to 2014.
The highest rates of attendance were observed during 2013(105/502) following by 2014 (80/502) and 2012 (70/502) [Fig-1].
Sex ratio was 1.9 males to each female with a mean age of 35.1 years [3-85 years old]. Patients above 45 years were the most represented population with 28.5% following by the patients between 30 and 45 years of age (28.3%). Patients aged from 3 to 15 years represented 17.9% of patients attending at the Parasitology Unit during this period.
Most of the patients attending had hepatosplenomagaly (206/502), the remaining have both heamaturia (87/502) and splenomegaly (85/502).

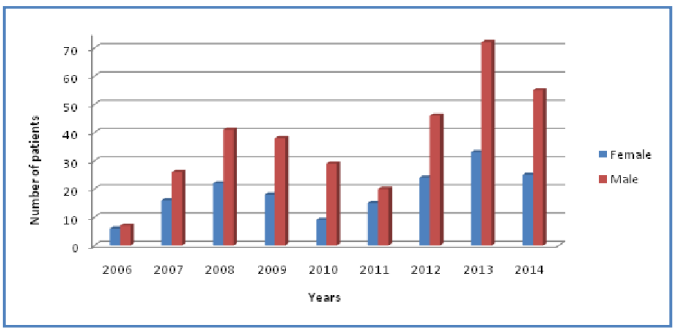

Fig-1 Number of patients attending at Parasitology Unit for serological Test

Serological data

Overall prevalence of schistosomiasis in Parasitology Unit of Institute Pasteur of Côte d'Ivoire was 23.3% with high seroprevalence in 2012 (45.7%) following by 2007 (42.9%) and less prevalence was observed in 2009 (3.6%) [Fig-2].

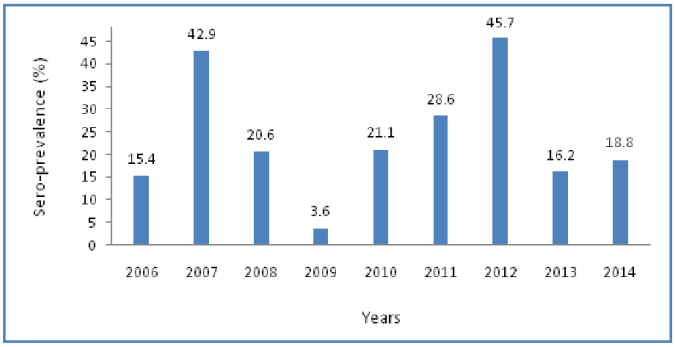

Fig-2 Seroprevalence of *Schistosoma* infection reported by year

More males patients (25.4%) patients presented positive result o comparison of the females patients (19%) but the difference was not statistically significant (p=0.12) [Fig-3].

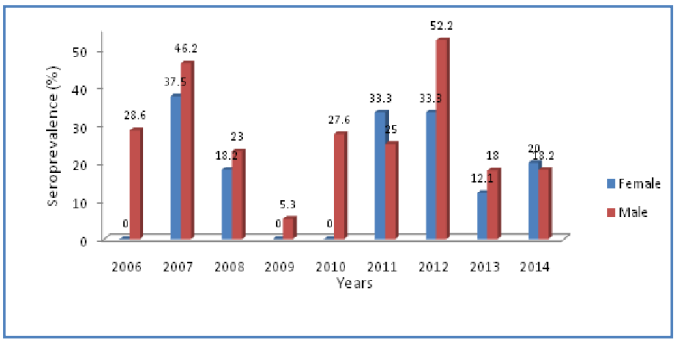

Fig-3 Distribution of *Schistosoma* infection among male and female patients

High seroprevalence was observed in the age range of 3-15 years (26.7%) following by age range of 30-45 years (24.6%). Others seroprevalence was 23.1 and 19.7 for the age group more than 45 years and from 15 to 30 years respectively. In each age group most infected patients were male but difference was not statistically significant (p>0.05) [Fig-4].

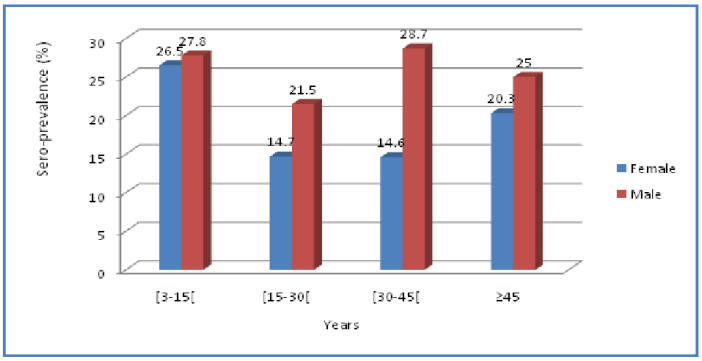

Fig-4 Seroprevalence of *Schistosoma* infection according to age group.

Positivity of IHA was found in 44, 21 and 20 patients with hepatosplenomegaly, heamaturia and splenomegaly respectively [Fig-5].

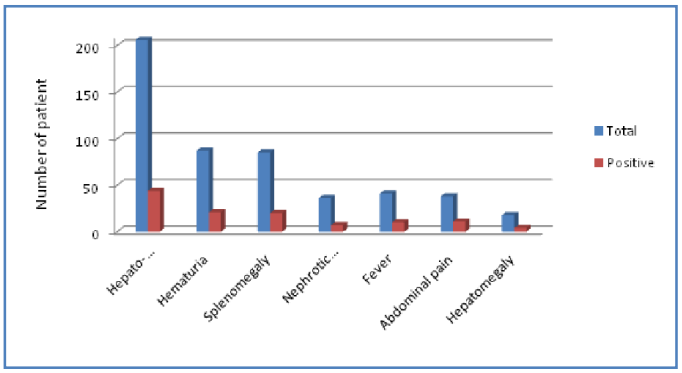

Fig-5 Positivity of IHA according to clinical indication

Discussion

Despite intensive efforts towards disease control, schistosomiasis is still highly prevalent in most endemic countries. Schistosomiasis is still one of the important chronic parasitic diseases in Africa, despite the continued implementation of control measures [23]. The currently recommended diagnostic method for schistosomiasis (coprological examination by Kato-Katz technique or urine filtration) failed to detect low burden infection. Consequently, there is an underestimation of infection cases with several false-negatives [24]. For this reason, alternative diagnostic methods (serological methods) have been developed. Although studies and national prevalence surveys of schistosomiasis have indicated that the disease is endemic in most parts of Côte d'Ivoire, studies on seroprevalence of the disease are very limited [11].

Overall, our study showed a seroprevalence of schistosomiasis of 23.3%. Similar rates have been reported in other African countries including Madagascar and Sudan [25,26].

Our results showed an increase in the seroprevalence between 2009 and 2012 with a peak in 2012 and a significant decline in 2014 despite more screening in 2013.

This improvement in prevalence in 2014 could be due to the increase of the population that has access to safe water and health education. In addition, construction and use of latrines have allowed better control of the disease [27].

In a study conducted by Coulibaly et al (2013) [28] seroprevalence of *S. mansoni* and *S. haematobium* infections was 66.9% in preschool-aged children using the SmCTF-RDT.

Diagnostic approach for schistosomiasis depends on the epidemiological situation. In areas of endemicity, where past *Schistosoma* infections, high *Schistosoma* loads, and polyparasitism (cross-reactions with others helminthes) are frequent, serological testing requires high specificity to avoid false-positive results.

Patients referred to the Parasitology Unit of Institute Pasteur of Côte d'Ivoire had already perform various tests including microscopic methods to diagnose schistosomiasis. These methods generally were negative. Results of the serological test is intended first to determine the *Schistosoma* past infection. In this case, it is an interesting alternative diagnosis despite its limitations.

The negative IHA results obtained in the study could be a possible consequence of the short period between infection and clinical presentation. A known obstacle to serodiagnosis of acute or recent *Schistosoma* infection is the prolonged seronegative window period. Newly infected individuals start producing antibody within 4 to 7 weeks after infection, and although the majority of patients exhibits seroconversion within 3 months [29-31] prolonged seronegative window periods of up to 6 months have been described [32, 33].

Despite their limitations, serological methods have to be resorted to in the absence of test to detect infection directly.

Our results showed that male patients were predominant at the testing centre. This could be due to the fact that male subjects are more involved in social and professional activities in rural and suburban areas that require contact with fresh waters that contain cercariae, infesting forms of *Schistosoma* [34-36]. Meanwhile difference between males and females patients was not statistically significant.

In a study conducted in Brazil, a higher proportion of males than females had detectable IgE against AW antigen [37]. Similar results were obtained by Nauss et al [38].

In contrast, higher prevalence were reported in females [39,40]. In a study carried out in Tanzania, no significant differences of infections were detected between boys and girls [41]

We observed no statistically significant difference between age groups in terms of the positivity of the IHA despite that children seem to be more affected following by patients aged from 30 to 45 years. In endemic area, children are intensely infected than adults. Infection intensities rise in early childhood, peak around the age of 12 years, and then rapidly decline. Around this age, the behavioral patterns of boys and girls diverge, giving rise to differences in exposure levels over many years. Occupations involving contact with infested water, such as fishermen, farmers, irrigation workers, or women in their domestic tasks are the second group at risk of schistosomiasis. The profession, ethnic and birth area of most patients were not available in the records as these factors is sometimes schistosomiasis risk factors.

Patients came for the serological examination had one or more clinical signs including hepatomegaly, splenomegaly, heamaturia, fever or hepatosplenomegaly. The signs and symptoms illicited are potentially attributable to multiple causes. But regarding high seroprevalence of *Schistosoma* antibodies and negativity of previous tests we assume that the majority of these late signs and symptoms could be attributable to *Schistosoma* infection.

Payne *et al* [42] found a strong correlation between majority of late signs and symptoms of hepatosplenic pathology and high seroprevalence of *Schistosoma* antibodies.

Our study has several limitations. First confirmation of schistosomiais infection as indicated by the manufacturer of IHA test in case of positivity was not done. Kato-katz and urine filtration were not done in our lab although there were done elsewhere with negative results.

IHA method used did not distinguish species of *schistosoma* (*S. mansoni, S. haematobium*and *S. intercalatum*).

Another limitation of this study includes patients being restricted to those referred at the Parasitology Unit of Institute Pasteur of Côte d'Ivoire. Therefore, we cannot be sure that our data are representative of the general population.

Despite the limitations, the study provided baseline information on seroprevalence in a reference laboratory. The seroprevalence of *Schistosoma* infection was found to be relatively high (23.3%) suggesting major interventions to reduce the burden of this disease in the country.

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Conflicts of interest: The authors have no conflicts of interest concerning the work reported in this paper.

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