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International Journal of Current Research
Vol. 7, Issue, 11, pp.22591-22595, November, 2015

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

ISSN: 0975-833X

RESEARCH ARTICLE

SERO PREVALENCE OF HIV INFECTION AMONG KNOWN TUBERCULOSIS PATIENTS IN KHARTOUM STATE –SUDAN

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ARTICLE INFO

Article History:
Received 29th August, 2015
Received in revised form
21st September, 2015
Accepted 18th October, 2015
Published online 30th November, 2015

Key words:

Sero prevalence, Tuberculosis,
HIV, ELISA,
Khartoum –Sudan.

ABSTRACT

Co-infection with Human Immunodeficiency virus (HIV) and Mycobacterium tuberculosis the causative agent of Tuberculosis (TB), has been referred to as the "cursed duet" as a result of the attendant morbidity and mortality due to their synergistic actions. This study was carried out to detect Sero prevalence of HIV infection among known tuberculosis patients In Khartoum state – Sudan, and to detect relation between certain factors such as gender, age, residence, occupation, treatment and duration of TB. It was descriptive cross-sectional study conducted from April to June 2015. A total of 89 known Tuberculosis patients who attended hospitals (67 males and 22 females) were enrolled. Serum specimens were tested by ELISA for anti HIV 0, 1, 2. Data were analyzed by chi squared test in SPSS software. sero positive of HIV antibodies 0,1,2 was detected in 2(2.2%) of cases and negative was 87(97.8%).

***Citation*:** *Shiraz Awad Elkarim Elsayed Elhag, Nafisa Ahmed Ibrahim and Wafa Ibrahim Elhag*. 2015. "Sero prevalence of hiv infection among known tuberculosis patients in khartoum State–Sudan", *International Journal of Current Research*, 7, (11), 22591-22595.

INTRODUCTION

The human immunodeficiency virus (HIV) is a lentivirus (a subgroup of retrovirus) that causes the acquired immunodeficiency syndrome (AIDS), a condition in humans in which progressive failure of the immune system allows life-threatening opportunistic infections and cancers to thrive. Without treatment, average survival time after infection with HIV is estimated to be 9 to 11 years, depending on the HIV subtype. Infection with HIV occurs by the transfer of blood, semen, vaginal fluid, pre-ejaculate, or breast milk. Within these bodily fluids, HIV is present as both free virus particles and virus within infected immune cells (Weiss, 1993). HIV attacks the body's immune system. Normally, the immune system produces white blood cells and antibodies that attack viruses and bacteria. The infection fighting cells are called T-cell lymphocytes. Months to years after a person is infected with HIV, the virus destroys all the T-cell lymphocytes. This disables the immune system to defend the body against diseases and tumors. Various infections will be able to develop, these opportunistic infections take advantage of the body's weakened immune system. These infection which normally won't cause severe or fatal health problems will eventually cause the death of the HIV patient (Rombauts, 1997).

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One-third of the world's population is thought to have been infected with *M. tuberculosis*, (WHO, 2011) and new infections occur in about 1% of the population each year (WHO, 2002). In 2007, an estimated 13.7 million chronic cases were active globally, (WHO, 2009) while in 2013, an estimated 9 million new cases occurred (WHO, 2014). In 2013 there were between 1.3 and 1.5 million associated deaths, (WHO, 2014 and GBD, 2013) most of which occurred in developing countries (WHO, 2011). The total number of tuberculosis cases has been decreasing since 2006, and new cases have decreased since 2002 (WHO, 2011). The rate of tuberculosis in different areas varies across the globe; about 80% of the population in many Asian and African countries tests positive in tuberculin tests, while only 5–10% of the United States population tests positive, (Konstantinos, 2010). More people in the developing world contract tuberculosis because of a poor immune system, largely due to high rates of HIV infection and the corresponding development of AIDS, (Lawn, 2011). Immuno suppression as a result of HIV infection increases the frequency and speed of progression from latent tuberculosis (TB) infection to active TB (Barnes, 1991). According to the World Health Organization (WHO), TB is one of the major causes of death among HIV infected people, and TB/HIV co-infection has been found to reduce the effectiveness of directly observed therapy (DOT) treatment of TB. (Anti tuberculosis drug resistance in the world, 1997; Anti tuberculosis drug resistance in the world, 2000) Concern over growing HIV-driven epidemics of TB has galvanized global consensus on this issue;

however large scale implementation of public health strategy to jointly address HIV/TB co infection. (Narain and Lo, 2004; TB/HIV research priorities in resource-limited settings WHO Geneva, 2005) Manjareeka, *et al.* in Eastren India (2013), 50 (12.3%) of the consenting 406 TB patients were HIV positive. Of these 406 patients, 44% had pulmonary TB, and 56% had extrapulmonary TB (EPTB). (Manjareeka, 2013) However Jain, *et al* in Delhi (2000) obtained the 2,361 specimens, 36 (1.52%) were found to be reactive by the initial ELISA test. (Jam, ?)

The aim of this study was to detect Sero prevalence of HIV infection among known tuberculosis patients attended hospitals.

MATERIALS AND METHODS

This was descriptive- cross sectional study which had been conducted in Khartoum state during period from April to June 2015, 89 known Tuberculosis patients who attended hospital were enrolled, Data was collected by using direct interviewing questionnaire; ethical clearance was obtained from research ethical committee of Faculty of Graduate studies of Al-Neelain University and verbal consent also was obtained from each patients.

Experimental work

Specimen collection

Blood specimens were collected from 89 known tuberculosis patients, under direct medical supervision by vein puncture using 5 ml syringe into plain tube to obtain serum by centrifugation at5000 rpm for 10 min. serum was kept in -20°C till serological study was performed.

Specimens were analysed by using Enzyme linked immune sorbent assay (ELISA) (4th generation ELISA) (Weka-China) for detection of antigen and antibodies to Human Immunodeficiency Virus. All reagents and samples were allowed to reach room temperature for 15 minutes before use. Washing buffer was prepared 1:20 from buffer concentrate with distilled water. Set the strips needed in strip-holder and number sufficient number of wells including three Negative control, three positive control and Blank were set. 20µl of biotinylated anti HIV, P24Ag was added in to each well except in the blank. 100µl was added from Negative control and positive control and specimen according to respective wells. Plate was covered with the plate cover and was incubated 60 minutes at 37° C. At the end of incubation the plat cover was removed and discard.

Each well was washed with wash buffer 5 time with Soaked for 30-60 seconds (washing 1). After the final washing cycle the plate was turned down on to blotting paper and was removed any remaining liquids. 100µl from HRP-Conjugateswas added in to the each well except in the blank.the plate was covered by plate cover and was incubated 60 minutes at 37° C. After the end of incubation the plat cover was discard. Each well was washed by wash buffer (washing 2). 50µl of chromogen A and 50µl chromogen B soluation were dispensed into each well including the blank. The plate was covered with plat cover and mixed by tapping gently. Then it was incubated at 37 for 15 minute .light was avoided.

The enzymatic reaction between chromogen A and chromogen B soluation produced blue colour in positive control and HIV 0/1/2 positive for antigen /antibodies sample wells. plate cover was removed and discarded from the plate, 50µl from stop solution was added and was mixed genty. Intensive yellow color was developed in positive control of HIV 0/1/2 positive for antigen antibodies sample wells.

Measuring the absorbance

The plate reader was calibrated with blank well and the absorbance was read at 450nm. The results were calculated by relating each sample optical density (OD) value to the Cut off value of plate. Calculation of cut off (C.O) value.

$$C.O = *Nc*2.1$$

*Nc= the mean absorbance value for the three negative controls.

The absorbance was read with micro well reader at 450nm.

Interpretation of Results

Negative results: samples giving absorbance less than Cut-off value are negative for this assay.

Positive result: sample giving absorbance equal to or greater than Cut-off considered initially reactive.

Borderline: sample with absorbance to Cut-off value are considered borderline and retesting of these samples in duplicate is recommended.

Data analysis: Data was analyzed by SPSS (Statistical Package of Social Science) software program version 16.

RESULTS

A total of 89 known tuberculosis patients who attended Tropical Hospital during the period from April to June 2015, consented to the study were included, 67(75.3%) of them were males and 22(24.7%) females .The mean age of patients was 40.5 years (ranged from 11 to 70 years) , most of patients 39 (44.0%) were belonged to the age group (21-30) (Fig. 1). Among the total studied group 2(2.2%) showed positive results for HIV. (Fig. 2) which observed among males. Most of studied population were Married 47(52.8%) and lived in Omdurman, most of positive cases were observed among Omdurman patients (Fig. 3). Regarding occupations most of Tuberculosis patients were working as free trade (Table 1). regarding duration of TB most of Tuberculosis patients had the infection ranging from month to 6 month (Fig. 4).

Table 1. Frequency of HIV seropositivity among tuberculosis patients (n=89) according to their occupation

			HIV antibody		Total
			+ve	-ve	
Occupation	Student	Count	0	11	11
		% within HIV antibody	.0%	12.6%	12.4%
	free trade	Count	1	27	28
		% within HIV antibody	50.0%	31.0%	31.5%
	housewife	Count	0	13	13
		% within HIV antibody	.0%	14.9%	14.6%
	without job	Count	0	13	13
		% within HIV antibody	.0%	14.9%	14.6%
	Other	Count	1	13	14
		% within HIV antibody	50.0%	14.9%	15.7%
	employee	Count	0	10	10
		% within HIV antibody	.0%	11.5%	11.2%
Total		Count	2	87	89
		% within HIV antibody	100.0%	100.0%	100.0%

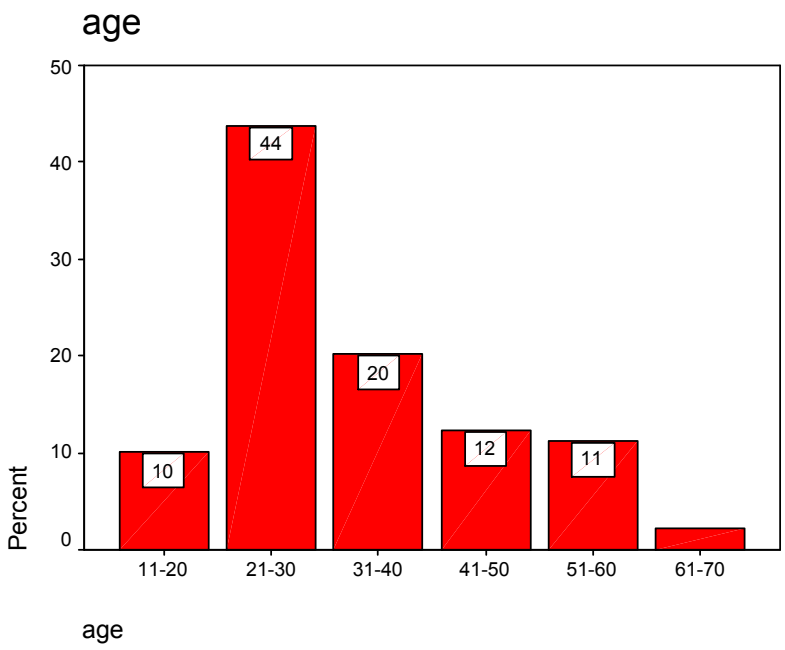

Figure 1. Age distribution of tuberculosis patients (n=89)

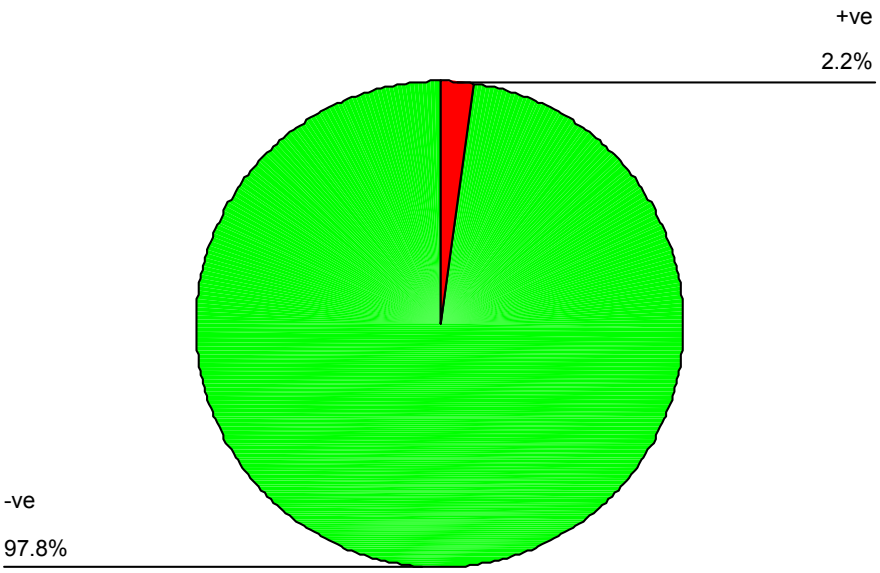

Figure 2. Seroprevalence of HIV among studied tuberculosis patients (n=89)

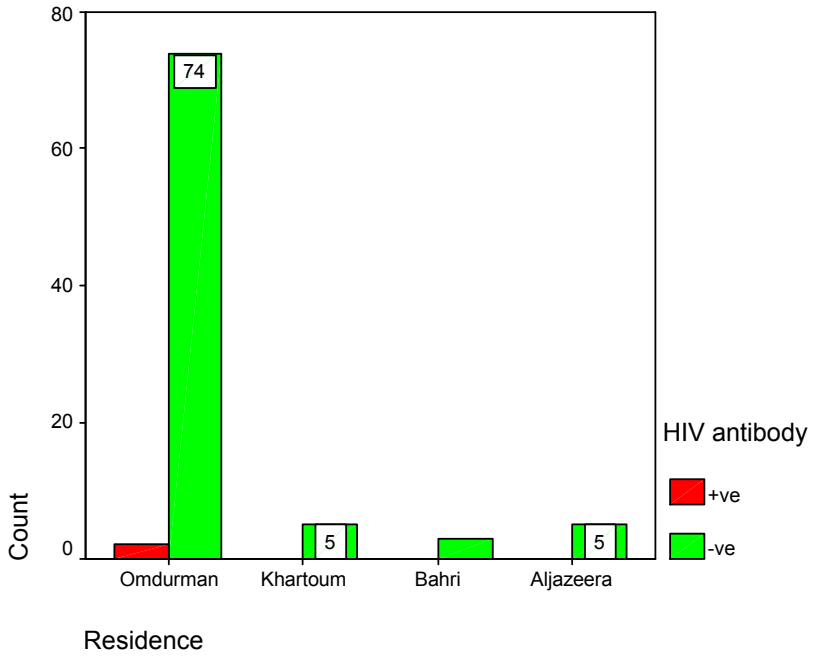

Figure 3. Frequency of HIV among tuberculosis patients (n=89) according to their residence

None of studied group have past history of organ transplantation, however 1(1.1%) has past history of haemodialysis, 10 (11.2%) has past history of blood transfusion, 14(15.7%) had history of surgical procedure, however positive HIV cases, had previous surgical procedure.

DISSCUSION

Immunocompetent individuals infected with *M tuberculosis* have approximately 10% lifetime risk of developing TB with half of the risk occurring in the first 1-2 years after infection. (Zwang *et al*., 2007) The present study result revealed 2.2% seropositivity of HIV among Tuberculosis patients. When compared with other studies in Sudan, there was similar published study conducted by Tajeldin Mohammedine *et al*. (2011) obtained 858 specimens, 18.3% of HIV seropositivty was reported among tuberculosis patients in Kassala, eastern sudan The present study give slightly higher than the study conducted by Jain *et al.* (2000) obtained the 2,361 specimens, 36 (1.52%) of HIV seropositive was reported among Tuberculosis in Delhi. However higher results was obtained by Manjareeka *et al.* (Eastren India) who reported 12.3% of the consenting 406 TB patients were HIV positive, also higher results was obtained by Enki *et al.* (1991) found that 66% newly diagnosed tuberculosis patients in Kampala (Uganda) were HIV seropositive. Eilhot *et al*. (1990) reported 60% seroprevalence among tuberculosis patients in Zambia, but lower results was repurted by Onorato and McCray (1992) had reported that 3.4% of the 3,077 tuberculosis patients had HIV co-infection in U.S.A.

Conclusion

The HIV seroprevalence in this TB patient population is of concern both in terms of patients management and public health prospective. It also underscores the need for routine HIV serology on all TB patients to avoid adverse drug reactions to antituberculosis drugs. There is also a need to develop aggressive public awareness, good health education and provide routine HIV screening for TB patients.

Acknowledgement

Our acknowledgments goes to Faculty of Medical Laboratory Sciences, AL Neelain University, also to the clerical staff of the Tropical Hospital and the technical staff of the TB and HIV laboratories, for their valuable assistance.

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