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THE TREND OF HEPATITIS B SURFACE ANTIGENEMIA AMONG TEACHING HOSPITAL PATIENTS IN KANO

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ABSTRACT

The prevalence and trend of Hepatitis B Virus infection in 2966 patients attending clinics of Aminu Kano Teaching Hospital, Kano Nigeria was determined over a 3 year period 2001 to 2003. The samples was initially screened by latex agglutination techniques while the positive samples repeated by Enzyme linked Immunosorbent Assay (ELISA) technique for confirmation. A seroprevalence of 23.3% units with coefficient variation of 22.5 to 24.1% were reported during the study period. More males (24.1%) than females 21.5% patients were observed to be seropositive for HBsAg. The differences was not however statistically significant. The overall trend in HBsAg seropositivity over the study period showed 21.7% in 2001, 24.7% in 2002 and 22.4% in 2003 respectively. Despite the availability of methods by which these viruses can be detected and surveillance activities to reduce the occurrence of the infection, the virus continues to constitute threat to health of the individuals. Our findings suggest that it is necessary to reappraise the need to intensify preventive measures in order to reduce the trend of HBV infections.

Key Words: Hepatitis B, HbsAg, Blood, Serum, Samples

INTRODUCTION

Hepatitis B virus (HBV) is one of the most important hepatotropic viruses known to be transmitted sexually, percutaneously, by blood, blood products and is known to be endemic in Africa (1&2). Despite the availability of methods by which this virus can be detected and surveillance activities to reduce the occurrence of the infection, the virus continues to constitute a threat to the health of patients. Hepatitis B surface antigen (HBsAg) in the blood is the most useful marker of active HBV infection which appears in the blood exclusively as a component of the virus and as incomplete viral forms (3&4).

It has been estimated that about one third of the world's population has been infected with HBV of which over 350 millions of them are said to chronic carriers (5).

WHO estimates suggest that HBV results in two million deaths each year worldwide and 230,000 of these occurring in Africa. Even though the incidence of acute hepatitis and death as a result of it is under reported, Nigeria appears to fall within the hyperendemic region of Sub-Saharan Africa. It has been reported that upto 25% of chronic carriers of HBV develop serious liver diseases including chronic hepatitis, cirrhosis and hepatocellular carcinoma (5).

Although the prevalence of HBsAg has been reported among blood donors in this centre (6), information on its prevalence and trend of infection among patients is not available. Therefore, the aim of this study is to present the carriage rate and the trend of HBV infection among patients in Aminu Kano Teaching Hospital, (AKTH) Kano.

METHODS

Serum samples were collected from 2966 consecutive patients presenting to Aminu Kano Teaching Hospital (AKTH) for a period of three years from January 2001 to December 2003. They consist of 2095 males and 871 females. The age ranged from one year to 61 years. In 2001, 406 specimens were collected while 1282 and 1278 were collected in 2002 and 2003 respectively. The samples were screened for HBsAg by latex

agglutination technique. All positive samples were repeated using Enzyme Linked Immunosorbent Assay (ELISA) technique (pathogyme Omega Diagnostics, UK) for confirmation.

RESULTS

Out of the 2966 patients tested over the three year period, 691 (23.1%) were HBsAg positive. This gives a period prevalence of 23.3% with a 95% coefficient variation of 22.5% to 24.1%. Of the 2095 male patients tested, 504 (24.1%) were HBsAg positive. Similarly, out of the 871 female subjects, 187 (21.5%) were positive. The differences was not statistically significant ($p=0.13$). An examination of the overall trend of HBsAg seropositivity over the three year study period showed 21.7% in 2001, 24.7% in 2002 and 22.4% in 2003 respectively. The stratification of HBsAg seropositivity by age and by sex are shown in table I and II respectively.

Table III Shows The Trend of Hepatitis B Virus Surface Antigenemia Among Teaching Hospital Patients.

Year	Total Screened	Total Pos	%
2001	406	88	(21.7)
2002	1282	317	(24.7)
2003	1278	286	(22.4)
Total	2,966	691	(23.1)

X²

X² (at 1dF and at $P_{0.001} = 13.81$) .Highly significant

Table 1 shows The Distribution of Hepatitis B surface Antigenemia according to Age –Groups.

Age Group (Years)	HBsAg Positive No. (%)	HBsAg Negative No. (%)	Total No.(%)
<10	49(25.3)	145(74.7)	194(100)
11 – 20	93(23.5)	303(76.5)	396(100)
21 – 30	230(24.7)	703(75.3)	933(100)
31 – 40	165(22.8)	559(77.2)	724(100)
41 – 50	87(23.6)	281(76.4)	368(100)
>50	59(17.2)	284(82.8)	343(100)
Not stated	8(100.0)		8(100.0)
Total	691(23.1)	2275(76.9)	2966(100)

χ^2 @1dF and at $P_{0.001} = (13.81)$.Highly significant

Table II shows The Gender Distribution of Hepatitis B surface Antigenemia among Teaching Hospital patients in Kano.

Sex	HBsAg Positive No. (%)	HBsAg Negative No.(%)	Total
Male	504(24.1)	1591(75.9)	2095(100.0)
Female	187(12.5)	684(78.5)	871(100.0)
Total	691(23.3)	2275(76.7)	2966(100.0)

χ^2 at 1dF and $P<0.05 = 2.3$. Not significant

DISCUSSION

Viral Hepatitis is a major problem in Nigeria and constitute a threat to life of infected patients both young and old. HBV can cause

persistent infection in chronic carriers and progressively cause terminal liver diseases (3&7). Studies have shown that the carrier rate of HBV in Nigeria is between 8 – 22% with an estimated

exposure rate of above 60% (8,9&10). This is consistent with this study, which showed a prevalence rate of 23.1% over a 3-year period. This relatively stable rate may reflect the adequacy of blood transfusion services with respect to screening for HBV infection in blood donors (3).

Although the seroprevalence rate observed in this study is consistent with other studies in Nigeria, it is however higher than 10% found in Pakistan (11&12) and 5% in India (13). But when compared with the rate observed in other African countries, the seroprevalence rate observed in this study is lower than 79% reported in Ethiopia, 56.2% in Kenya and 79.2% in Mozambique (5). Among the various viral Hepatitis, the HBV is more distributed and spreading at the rate which is higher than HIV within the individuals (13). Seroprevalence rate of HBV has been reported in patients with different ailments in Nigeria such as Sickle Cell disease (14), Acute Icteric Hepatitis (15), Diabetes Mellitus (16) and HepatoCellular Carcinoma (17). The prevalence in these various disease conditions are not different from that observed in the general population except for HepatoCellular Carcinoma and Acute Icteric Hepatitis.

There was no significant difference in the prevalence rate of infection in the various age groups except for those in the age of above 50 years. This may be due to the small number of patients in this age group (Table 1).

Conversely, the seropositivity in the males were higher than in the females patients (Table 1). This is also consistent with other authors¹ who reported a higher carriage rate of 77% in males than 50% in females in South Africa. The trend of infection within the study period is statistically significant ($P < 0.001$).

In Africa, transmission of HBV is usually horizontal in childhood. The prevalence of HBV

among pregnant women is said to be much lower than in Chinese women and so vertical transmission is less important.

Some risk factors identified among the study subjects include previous blood transfusion, traditional surgery, scarification, occupational exposure and exposure to infected sex partners. Social status appears to have no effect in carriage of HBV in this study population.

It is necessary to reappraise measures of prevention of HBV infection in order to reduce the trend of infection. Emphasis should be placed on immunization of those at risk, avoidance of sharp objects and contaminated infection, vigorous screening of all blood and blood products to be transfused, practice of safer sex, good personal hygiene and health workers must use the Universal Precaution for protection.

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SERUM FERRITIN AND HCV INFECTION IN NIGERIAN PATIENTS WITH PRIMARY LIVER CELL CARCINOMA¹Ola, S.O., ²Odaibo, G.NDepartments of ¹Medicine and ²Virology, College of Medicine, University of Ibadan, University College Hospital, Ibadan; Nigeria.

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Abstract

A prospective study aimed at determining the relationship between hepatitis C virus (HCV) infection and serum ferritin in Nigerian patients with primary liver cell carcinoma (PLCC) was carried out at the University College Hospital (UCH), Ibadan; Nigeria. The study involved 42 adult Nigerians made up of 14 healthy subjects as controls and 14 patients each with PLCC and liver cirrhosis (LC) who consented to participate in the study. The subjects were controlled for age and sex. The diagnosis of the diseases was made from relevant clinical features, ultrasonography and histology of liver biopsy specimen. Blood specimen collected from the subjects were analysed for ferritin by radio-immuno assay using Amersham Kits, hepatitis B virus (HBV) infection using HBsAg detection and anti-HCV by ELISA (Sanofi Pasteur, France). The study protocol was approved by the Joint UI/UCH Ethical Review Board. Data obtained was analysed with the SSPS software at a level of significance of $p < 0.05$. Serum ferritin $> 700\text{ng/ml}$ was detected only in 50% and 14% of the patients with PLCC and LC respectively with specificity of 93% as well as negative (78%) and positive (79%) predictive values. Serum anti-HCV and HBsAg were present in 14% and 71% of patients with PLCC respectively ($p < 0.005$). Similarly, 29% and 14% of the patients and the Controls respectively were sero-positive for anti HCV while serum HBsAg was detected in equal proportions of the patients with LC (50%) and the Controls (43%). There was correlation between elevated serum ferritin and HBsAg (χ^2 with Yates correction = 5.04, $p = 0.025$) but none with serum anti-HCV.

In conclusion, the study shows that serum ferritin level $\geq 700\text{ng/ml}$ is indicative of PLCC among Nigerians especially in the presence of HBV infection but may not be useful when there is associated HCV infection.

Introduction

Hepatitis C virus (HCV) infection occurs globally(1). It presents more with asymptomatic course resulting in chronic hepatitis and later progressing to primary liver cell carcinoma (PLCC). Although, PLCC has been associated more with hepatitis B virus infection in Nigeria(2).

may assume increasing trends once vaccine against HBV becomes efficient as in developed countries(3,4). Hence, HCV infection with PLCC and its sequelae remains a burden since there is presently no vaccine against the virus. Efforts directed at treatment of PLCC have been

met with poor success because of the late presentation of the patients at hospitals(5). Early diagnosis of the disease is therefore necessary. To ensure this, various tumour markers have been shown to be useful in the diagnosis of PLCC among different populations(6,7). Specifically, serum ferritin has been documented to be a useful marker in diagnosis of PLCC among Nigerians(8). The significance of serum ferritin in the presence of HCV infection remains unknown. This study was designed to determine the level of serum ferritin in Nigerian patients with primary liver cell

carcinoma (PLCC) especially when there is associated hepatitis C virus infection.

Materials and Methods

Forty-two adult Nigerian subjects were recruited into the study after obtaining informed consent. The subjects comprised 14 apparently healthy adults as control (group I) and 14 patients each with liver cirrhosis-LC (group II) and PLCC (group III). They were sex and age matched. There was no ingestion of multivitamin medication, obvious blood loss or blood transfusion in any of the subject within 12 months prior their inclusion in the study. In addition, no subject was anaemic at entry into the study. Relevant clinical features, ultrasonography and histology of liver biopsy specimen were utilised to make the diagnosis of both LC and PLCC. Blood specimens were collected from the subjects for packed cell volume (PCV) estimation, sero-analysis of ferritin, liver function tests as well as the serology of HBV and HCV infections. The assay of serum ferritin was carried out by radio-immuno assay using Amersham Kits (Ferritin RIA Kit IM 1051). The serum HBsAg (HBV infection) and anti-HCV were determined by ELISA. Liver function tests were measured in all patients by routine laboratory methods.

The study was carried out after obtaining clearance from Joint UIUCH Ethical Review Board. The SPSS statistical package was utilized for data entries and analysis on a micro-computer. The Kruskal Wallis statistics, a non parametric equivalent of the analysis of variance technique and the Mann Whitney U test, a non parametric equivalent of the student's t-test were used to compare the means of values of continuous variables. The Chi-square test and Fisher's exact test were used to determine the statistical significance of the association between two categorical variables. The validity of the diagnostic

value of ferritin for PLCC was also examined by calculating the sensitivity, specificity, positive and negative predictive values where the histological findings were the gold standard. All statistical analysis were carried out at 5% probability level.

Results

The forty-two 42 subjects studied were made up of controls (14) and patients with PLCC (14) and LC (14). They were 48 ± 14 , 49 ± 15 , and 55 ± 12 years of age respectively. Each group had male:female ratio of 11:3. The Child Pugh stages of the patients in both PLCC and LC were similar (4 and 10 patients in each group were scored A and B respectively). The PCV in the subjects were $43 \pm 4\%$, $38 \pm 9\%$, and $33 \pm 7\%$ for the control subjects and the patients with PLCC and LC respectively. The PCV among the control subjects was significantly higher than the value for either the patients with PLCC ($P < 0.05$) or LC ($P < 0.001$). Serum ferritin $> 700 \text{ ng/ml}$ was detected in 50% of the patients with PLCC (specificity, negative and positive predictive values were 93%, 78%, and 79% respectively, Table I) compared to only 14% of the patients with LC and none of the Controls who had values serum ferritin above the cut-off level, $p < 0.05$ each. Serum anti HCV and HBsAg were present in 14% and 71% of patients with PLCC respectively ($p < 0.005$, Table II). Similarly, serum anti-HCV was present in 29% and 14% of the patients with LC and controls respectively while serum HBsAg was detected in equal proportions of the patients with LC (50%) and the Controls (43%). Elevated serum ferritin correlated with serum HBsAg (X^2 with Yates correction = 5.04, $p = 0.025$) but not with serum anti-HCV. Among all the subjects studied, 2 patients each with PLCC and LC had co-infection of HBV and HCV with the former patients also having elevated serum ferritin levels.

Table I. The validity of serum ferritin in diagnosing PLCC among adult Nigerian patients at different cut-off points.

Ferritin (ug/L)	positive	negative	Validity(%)			
			Sen.	Spec.	PPV	NPV
≥ 700	7	2	50	93	78	79
< 700	7	26				
≥ 600	7	6	50	78	54	76
< 600	7	22				
≥ 800	6	2	43	93	75	76
< 800	8	26				

% - percentage Sen. - Sensitivity Spec. - Specificity
 PPV - Positive Predictive Value NPV - Negative Predictive Value
 PLCC - Primary Liver Cell carcinoma

Table II The association between serum ferritin, HBV and HCV in adult Nigerian patients with PLCC, LC and controls

Number of subjects	Serum ferritin ≥700ug/L				
	Total (42)		PHCC	LC	Control
	≥700ng/L	<700ng/L	14	14	14
@HBsAg ⁺	7	16	6	1	-
HBsAg ⁻	2	17	1	1	-
*Anti-HCV ⁺	2	6	1	1	-
Anti-HCV ⁻	7	27	6	1	-

@ X² with Yates correction = 5.04, p=0.025 for Ferritin & HBsAg in PLCC *P=0.64 for ferritin & HCV in PLCC
 PLCC - Primary liver cell carcinoma, LC - Liver Cirrhosis
 HBV - Hepatitis B Virus HCV - Hepatitis C Virus
 HbsAg - Hepatitis B surface Antigen

Discussion

Elevated serum ferritin level has been reported to be diagnostic of PLCC in different populations including Nigerians(6,8). This study has taken into consideration the role of blood loss, infectious anemia as well as ingestion of iron tablets on serum ferritin level hence the packed cell volume of the subjects studied are not unexpected. The serum ferritin level of $\geq 700\mu\text{g/L}$ as diagnostic level for PLCC is a better discriminatory level for the tumour from liver cirrhosis than the value of $400\mu\text{g/L}$ earlier reported among Nigerian(8). High serum ferritin levels in our patients with PLCC might be due to the production of ferritin by the tumour rather than to liver necrosis(9). Although, at serum ferritin level of $\geq 700\mu\text{g/L}$, two of our patients with LC were shown to have elevated levels of serum ferritin which may be suggestive of occult PLCC. This shows the limitation of histological diagnosis in absence of screening with a tumour marker. It is however comparable to the report among Europeans (10).

In view of the vary proportions of our patients with PLCC having HBV and HCV infections, both viruses could have contributed to the aetiology of the disease. Despite the use of ELISA for assay of HBsAg in our study, the correlation observed between elevated level of serum ferritin corroborates previous reports where heamagglutination method had been utilized for the detection of serum HBsAg(8). This also shows that serum level of ferritin $\geq 700\text{ng/ml}$ is diagnostic of PLCC especially in the presence of HBV infection. It suggests that serum level of ferritin in patients with PLCC may be influenced by HBV infection since the virus gets intercalated with the hepatocellular DNA in aetio-pathogenesis of the tumour. In view of the diverse aetiological factors for PLCC, there is the need to define the effects of the other causal factors like HCV on the expression of the markers

that may aid diagnosis of the tumour(11). In spite of the worldwide prevalence of HCV infection and its relationship to PLCC, the association between serum ferritin and HCV infection has hitherto remained unknown among Nigerians. However, our study has shown that there is absence of correlation between elevated serum levels of ferritin and HCV infection in Nigerian patients with PLCC. This observation might not be unconnected with the predominantly chronic clinical course and pathology of HCV infection(12). The presence of elevated level of serum ferritin among the two patients who had combined infection of HBV and HCV could be due to the effects of HBV. However, further study involving a larger sample size will be necessary to elucidate this observation.

In conclusion, this study shows that serum ferritin level $\geq 700\text{ng/ml}$ is diagnostic of PLCC among Nigerians. In addition, serum ferritin may not be useful in screening for PLCC among Nigerians with HCV infection but the converse holds for those infected with HBV.

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RISK FACTORS AND SEROPREVALENCE OF HEPATITIS C ANTIBODY IN MOTHERS AND THEIR PRE-SCHOOL AGE CHILDREN IN ILORIN.Agbede, O. O.¹, Iseniyi, J. O.², * Kolawole, O. M.³, Ojuawo, A.⁴¹Department of Medical Microbiology and Parasitology, University of Ilorin, Kwara State. ² Department of Haematology, University of Ilorin Teaching Hospital, Ilorin, Kwara State. ³ Department of Microbiology, University of Ilorin, P.M.B 1515, Ilorin, Kwara State, ⁴. Department of pediatrics and child health, University of Ilorin Teaching Hospital, Ilorin Kwara Nigeria.Correspondence to : Kolawole, O.M., Tel- 23408060088495 E-mail: tonak74@yahoo.com.**ABSTRACT**

In the tropics, hepatitis C virus (HCV) seroprevalence ranges from <0.2% in whole Africa. Mother-to-infant transmission of HCV though relatively low, have been reported worldwide and transmission may be intrauterine, intrapartum and post-natal. A descriptive seroepidemiologic study of hepatitis C virus and their associated risk factors have been conducted among pairs of mother and child of pre-school age attending the "well child" clinic of the University of Ilorin Teaching Hospital and the immunization clinic of the children specialist hospital, Ilorin. Sera of 70% mother/child pairs were subjected to Enzyme-Linked Immunoabsorbent Assay (ELISA) for the detection of antibodies directed against the core and structural proteins of hepatitis C virus (anti-HCV).

Anti-HCV prevalence of 1.4% was seen among mothers while none of the children was positive for anti-HCV. Scarification appeared to be the most significant risk factors that could possibly contribute to the transmission of HCV among the subjects. The only mother positive for anti-HCV antibodies had tribal mark scarification while her 5-year old baby who had no tribal mark was negative.

Vaccination has been effective in reducing the incidence of hepatitis B and attending complications of onset of hepatocellular carcinoma later in life; but preventive measures against hepatitis C virus are not yet available

INTRODUCTION

Over the past decade, hepatitis C virus (HCV) infection has risen from an obscure disease into a public health problem all around the world. At present, there are an estimated 170 million HCV carriers worldwide most of whom are thought to be in the developing countries (1). Like the HBV infection, HCV infection may be transmitted parenterally by transfusion of blood and blood products, intravenous - drug abuse and unsafe injections. However, in 40-45% of HCV infections, no obvious parenteral exposure have been identified (2). The acute HCV infection is commonly asymptomatic but leads to chronic

hepatitis in up to 80% of individuals. This is the highest rate of chronicity of the hepatotropic viruses occurring in adults and is striking when compared to HBV infections that becomes chronic in only 5% to 10% of infections occurring in adulthood (3).

Vertically transmitted infection becomes chronic in the majority of paediatric cases, as in adult cases. The ultimate outcome of neonatal infection remains largely unknown because of the lack of data from long term longitudinal studies of infected children (4). Mother-to-child transmission rate of hepatitis C virus (HCV) is low, less than 10% in women not co-infected with HIV. Due to

the fact that most published studies have included a small number of HCV-RNA positive children ($n < 10$), the transmission risk factors remain still unclear. Invasive procedures, such as aminocentesis or use of forceps, could increase the risk of transmission (5). In Nigeria, where hepatitis B virus infection is hyperendemic, the impact of chronic hepatitis C virus infection has not been adequately elucidated most especially in mothers and their pre-school age children. This study was therefore aimed at finding the seroprevalence of HCV and their associated risk factors among pairs of mother and child of pre-school age attending the "well child" clinic of the University of Ilorin Teaching Hospital and the immunization clinic of the Children Specialist Hospital, Ilorin.

MATERIALS AND METHODS

Subjects

Seventy (70) pre-school age children attending the "WELL CHILD" Clinic in the University of Ilorin Teaching Hospital and the Children Specialist Hospital (Center Igboro) in Ilorin and their mothers were randomly recruited for this study. Children attending these clinics in the company of other relatives and those above five years of age were exempted from this study.

Subjects were verbally informed of the study. A questionnaire was used to obtain personal data such as age and possible modes of transmission of HCV or risk factors such as history of previous blood transfusion, history of circumcision, history of jaundice, history of use of unsterile needles, history of HBV vaccination and history of scarification (Tribal marks).

Serum samples were obtained from mothers and also from their children by venepuncture and stored frozen in aliquots at -20°C . Anti-HCV was detected using third generation enzyme linked immunoabsorbent assay

(ELISA) kit (Ortho diagnostics Raritan, New Jersey USA). A repeat ELISA test was performed on each positive anti-HCV sample, in order to eliminate false positivity. Results were regarded as positive after a repeat positive ELISA test.

The results were subjected to statistical analysis using EPI-Info Version 6 Software package. Bar Chart and Cross tabulation tables were done. The critical level for statistical significance was set at 5% confidence level using the chi-square analysis.

RESULTS

The age distribution of children whose mothers participated in this study is shown in figure 1. Of the 70 respondents about 4.3% of the children are within the age of less than 1, 24.3% within the age of 1, 31.4% within the age 2, 21.4% within the age 3, 15.7% within age 4 and 2.9% within age 5. Of the respondents, 1 (1.43%) mother was positive for anti-HCV antibodies while 69 (98.57%) was negative. It is interesting to note in this study that none (0%) of the children was positive for anti-HCV antibodies (Table 1). The possible modes of transmission of risk factors and prevalence rates are shown in Tables 2 and 3. In the case of the only mother positive for anti-HCV antibodies, results indicated that the history of circumcision; history of scarification (tribal marks) and history of previous use of unsterile needles were the associated risk factors linked with her anti-HCV antibodies positivity (Table 2). This finding statistically were significant with P values of 0.0411, 0.0163 and 0.0425 respectively (Table 3).

However, no significant association statistically were found between HCV Seropositivity in the only positive mother with history of previous blood transfusion, history of jaundice and history of HBV vaccination [P values of 0.758, 0.758 and 0.717 respectively] (Table 3).

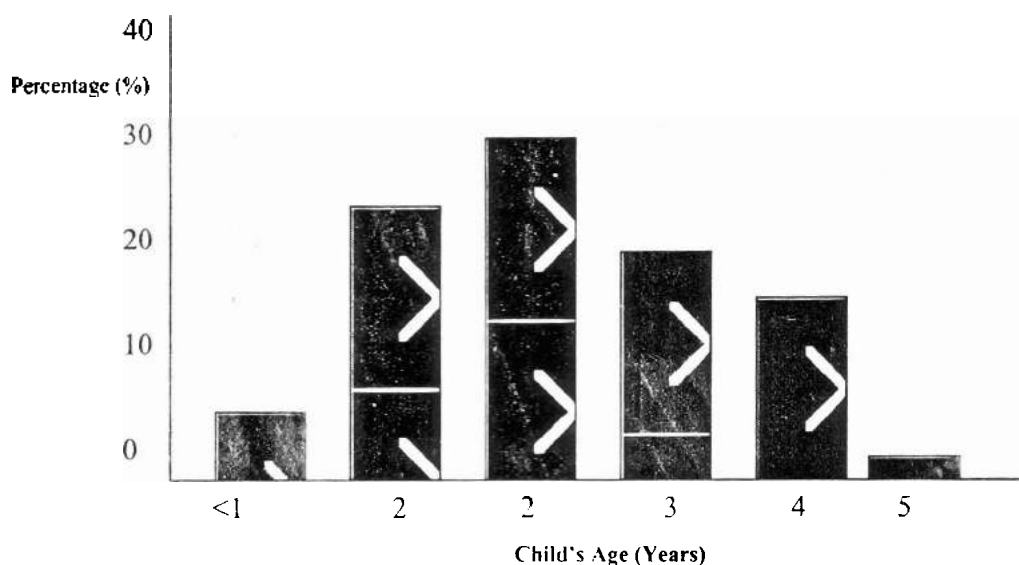


Figure 1. Age distribution of children of mothers used in this study.

This bar chart shows the age distribution of children whose mother participated in this study. About 4.3% of the children are within the age of less than 1, 24.3% within the age of 1, 31.4% within the age 2, 21.4% within age 3, 15.7% within age 4 and 2.9% within age 5.

Table 1 Prevalence of antibodies to HCV (anti-HCV) in relation to mother and child.

HCV Ab	Mother	Child
Positive	1 (1.43%)	0 (0%)
Negative	69 (98.57%)	70 (100%)
Total	70 (100%)	70 (100%)

Table 2 Description of the various modes of HCV transmission and prevalence rates in mothers .

Mode	anti-HCV Positive (n = 1)	Percentage %
History of previous blood transfusion	0	0
History of Circumcision	1	100
History of Jaundice	0	0
History of HBV vaccination	0	0
History of Tribal Marks (Scarification)	1	100
History of Use of unsterile needles	1	100

*where n is the total number anti-HCV positive mothers

Table 3 Risk factors, confidence intervals and P values in relation to mothers HCV transmission

Risk Factor	Chi-square 5% C.I & P values	Comments
History of blood transfusion	P = 0.758	P > 0.05 There is no statistically significant difference
History of Circumcision	P = 0.0411	P < 0.05 There is statistically significant difference
History of Jaundice	P = 0.758	P > 0.05 There is no statistically significant difference
History of HBV vaccination	P = 0.717	P > 0.05 There is no statistically significant difference
History of Tribal Marks	P = 0.0163	P > 0.05 There is no statistically significant difference
(Scarification)	P = 0.0425	P < 0.05 There is statistically significant difference
History of use of unsterile needles		P < 0.05 There is statistically significant difference

* C.I. means confidence interval.

DISCUSSION

Hepatitis C virus (HCV) belongs to the Flavi family and it is a RNA virus with prevalence rate in most countries of less than three (3) percent (6). Hepatitis C virus plays an important role in the causation of hepatocellular carcinoma (7). In this study, ages of children screened for hepatitis C falls between less than 1 and 5 years with children within age 2 having the highest percentage of 31.4% and children of age 5 having the lowest percentage of 2.9% (Figure 1). This age group is in consonance with the work done by (8) whose study determined the age wise prevalence of Hepatitis C virus in children under five years and analysed the relative importance of horizontal or vertical transmission.

In this study, of all the children screened for HCV anti-bodies, none was positive for anti-HCV. However, only 1 mother was found positive for anti-HCV (Table 1). It is interesting to note that of all the risk factors for the transmission of HCV infection investigated in this study, the only positive mother had history of circumcision, tribal

mark and history of usage of unsterile needle for injection in the past (Tables 2 and 3). This result revealed that the prevalence of hepatitis C infection in this environment is low. This finding is at variance with the result in the work done by (9) who reported 5% prevalence of antibodies to hepatitis C virus among normal blood donors and multi-transfused sickle-cell anaemia patients in the same environment.

The plausible reason for this differences could be in the sensitivity of the kits used in both studies. In this study, the third generation ELISA kit was used as against the second generation ELISA kits used in the work done by (9). Intravenous drug abuse, haemophilia and homosexuality have been identified as high risk factors for hepatitis C virus transmission (10). These same risk factors have been associated with HBV infection. Furthermore, it has been reported that rates of transmission varied from one study to another probably because the risk of transmission varies in different population of women studied. Socio-cultural factors such as tribal markings by

scarification and female genital circumcision are not common in the part of the world except in some African societies. These socio-cultural practices are often done by the use of scientifically unsterilized devices. It is plausible to suggest that engagement in these activities as verified in the singular case that was positive for HCV antibodies could have exposed this person to infection with Hepatitis C virus.

In summary, prevention remains the key for HCV transmissions. Older children and mothers require education about high-risk behaviours. Prevention of perinatal transmission should also be targeted. HCV infection occurs in children and is frequently less severe or more prolonged. Children might have a better response rate to therapy, but this is based upon very small and uncontrolled studies. Education is important to prevent transmission of HCV infection to adolescents and newborn (11).

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HIV-TB CO-INFECTION: PATHOGENESIS, DIAGNOSIS AND MANAGEMENT IN ADULTS

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ABSTRACT

There is a looming epidemic of HIV/TB co-infection in tandem to the prevailing wave of HIV infection in sub-Saharan Africa and South East Asia where TB is endemic. HIV positive patients become susceptible to Mycobacterial infection following depletion of the immune cells that usually resist mycobacterial infection i.e. CD4⁺, and macrophages. Host's TH1 immune response to TB produces several cytokines some of which further enhance local and systemic HIV replication. DOTS remain the best option for treating TB in HIV patients. This is sometimes co-administered with antiretroviral (ARV) drugs; however, this is fraught with complex bidirectional drug-drug interactions between rifamycin component of anti-TB regimen and the protease inhibitors (PI) and non-nucleoside reverse transcriptase inhibitors (NNRTI) components of ARV. Some patients could also develop paradoxical reactions. Cytokine inhibitors could have an adjunct role to Anti-TB therapy when it roles in the pathogenesis of TB in HIV/TB is fully understood.

KEYWORDS: HIV/TB co-infection, pathogenesis, DOTS, HAART, Drug interactions

INTRODUCTION

By estimate one third of the world population is infected with *Mycobacterium tuberculosis* (MTB) and every day 23,000 people develop active TB, i.e. about 8.7million cases per year (1). Averagely, TB kills 2million people every year (1). Also about 40million people are HIV infected worldwide with 16,000 people being infected every day (1). TB and HIV have formed a synergy and this is facilitating the spread of a co-epidemic of HIV-TB especially in sub-Saharan Africa and South-East Asia (2,3). HIV positive individuals are 50 times more susceptible to MTB infection, and once infected, are 800 times more likely to develop an active TB (4&5). About 70% of all TB cases in sub Saharan Africa are co-infected with HIV (1), the highest in the world.

This has impacted heavily on the regional TB control programme with a threat of MDR-TB especially in countries with ineffective DOTS implementation (1). The prevalence of HIV infection in Nigeria is about 5%, this is 4times lower than the estimated 20% mark of HIV prevalence at which the annual percentage increase in TB will be high at over 10% (6). However, Nigeria still has the highest annual estimate of new TB cases in Africa with about 27% of its adults infected with TB/HIV (7) while South Africa has TB as the leading cause of mortality amongst her HIV infected persons (8). According to WHO, TB accounts for 11% of all deaths in AIDS patients (9); this makes it one of the leading causes of mortality in HIV infected patients. To effectively control this growing epidemic of HIV/TB, both

HAART and DOTS should have a wider coverage. These medications will provide lifelines to millions of co-infected people. Clinicians have to be educated and updated on the complex interactions between TB and HIV as well as the dual drug-drug interactions that exist between anti-retroviral and anti-TB drugs.

PATHHOGENESIS

MTB is the infectious agent of TB and it is acquired through inhalation of aerosolized droplets nuclei produced by patients with pulmonary, bronchial or laryngeal TB when they cough, sneeze, speak, or sing (10). These are the main sources of mycobacterial transmission, but smear negative PTB and extra pulmonary TB (EPTB) could also transmit the bacilli (11), especially during cough induction (12&13), irrigation of TB abscesses (14) or changing of wound dressings (15). In most infected individuals the tubercle bacilli remain dormant for years after infection before entering a phase of exponential multiplication to give active disease (16). Development of active TB is often prevented by the host's intact immune system (17), specifically the cell-mediated type (18), but this is the target of HIV infection. In TB/HIV co-infected patients therefore, there will be a steady deterioration in this protective capacity of the cell-mediated immunity till a critical point at which tubercle bacilli begin to proliferate and cause clinical disease (17). That is the balance between quiescent TB focus and the host immunity has broken down and resulted in endogenous reactivation. This occurs with the CD4⁺ count around 500cells/ul or slightly higher. The ensuing reactivation often manifest as a localized pulmonary form, however, in advanced state of immunosuppression poor containment of the resulting infection could occur with resultant widespread of the bacilli causing extra pulmonary,

disseminated or miliary TB (18). When the CD4⁺ count is less than 500cells/ul (19) active TB could progress rapidly from primary mycobacterial infection (20&21) and some patients could have exogenous re-infection (22). The estimated annual risk of reactivation among HIV/TB patients is about 5 to 8% with a cumulative lifetime risk of 30% or more compared to a cumulative lifetime risk of 5 to 10% in HIV-negative patients (9). Early in the course of HIV infection reactivation is commoner in the upper lobes or the upper part of the lower lobes of the lungs where ventilation is greatest; Po₂ of 140 mmHg (23) and lymphatic drainage is relatively impaired (24&25) but as the disease progresses it could occur at any earlier seeded site in the body.

HIV and *MTB* influence each other in a synergistic and bidirectional ways (26). Host immune response to tubercle bacilli enhances both systemic and local HIV replication and tends to accelerate the course of progression of HIV infection (26&27). Mechanisms of these interactions are now being understood. The initial contact between the host immune system and *MTB* occurs in the alveolar macrophages that present mycobacterial antigens to antigen-specific CD4⁺ cells (18). This is via several cytokines, which are inflammatory mediators produced by macrophages, monocytes, and lymphocytes. When these cells are sensitized by prior exposure to *MTB*, and then re-exposed to the same antigens, they produce several cytokines such as interferon-gamma (IFN- γ), interleukin 6 (IL-6), IL-12, and IL-18 (28,29). This is a Th1cell-mediated immune response; it is typical for TB and other intracellular pathogens. This is in contrast to asthma and other atopic diseases whose response is Th2 cell-mediated with different cytokines, like IL-4 and IL-5 (30). Mycobacterial infected macrophage releases IL-12 and 18, these cytokines stimulate CD4⁺

lymphocytes to release IFN- γ (31&32), which in turn activate more macrophages to enhance their ability to contain mycobacterial infection. The activated macrophages also release tumor necrosis factor (TNF- α), IL-1 and IL-6, it is these set of cytokines that enhance viral replication (26,33-36). Tubercle bacilli and their products also enhance viral replication by inducing nuclear factor kappa-B (NF- κ B), this cellular factor binds to promoter regions of HIV (37&38) and TNF- α -induced-HIV replication is mediated predominantly through the increased activation of this factor (39&40). The long terminal repeat (LTR) of HIV contains 2 NF- κ B sites, and NF- κ B, either alone (41) or in concert with other transcription factors (42), is critical to the transcriptional activation of HIV. Activation of mitogen-activated protein (MAP) kinase pathway has also been implicated in the increased HIV replication (43). In particular, the p38 MAP kinase pathway has been found to be critical in HIV replication in both CD4+ cells (43) and macrophages (44). IL-1 β and TNF- α activate p38 MAP kinase and the HIV-1 LTR (45), and these cytokines are up-regulated by the *MTB* infection of mononuclear phagocytes (46&47). β -chemokine, monocyte chemotactic protein (MCP-1) are also known to play active roles in enhancing HIV replication (48). The recovered broncho-alveolar fluid from TB affected lung has demonstrated local increase in HIV replication by containing higher level of viral load compared to the unaffected segment of the lung and this correlated with TNF- α suggesting local production of the virus (49). The local immune activation against TB also favours the development of latent HIV infection in the macrophages and dendritic cells, thereby potentially enhancing dissemination of HIV (50&51). Thus in HIV-infected persons with active TB, the active sites of TB infection act as epifoci of increased HIV replication and evolution

independent of systemic HIV disease activity (50). The resulting HIV viraemia will deplete immune cells that play a central role in anti-mycobacterial defenses (52); such CD4⁺ lymphocytes, macrophages and monocytes. *MTB* is characterized by delayed-type hypersensitivity reaction and granuloma formation in the infected tissues. Resolution of this granuloma is controlled by both cell-mediated immunity and delayed-type hypersensitivity reaction, both of which are often accompanied by some level of tissue destruction (24). Cell-mediated immunity controls TB by activating macrophages to kill ingested bacilli while delayed-type hypersensitivity causes caseous necrosis that result in killing of bacilli-laden macrophages (53). Some of the granulomas may undergo necrosis and sloughs off forming cavities others may heal with fibrosis and some may calcify (54). The extent of necrosis and cavitation in HIV/TB patients is dependent on the relative efficacy of each of these two immunologic processes in inhibiting multiplication of *MTB*. However, both processes are reduced in this circumstance.

PATTERN OF TB IN HIV INFECTION: The location and pattern of distribution of TB in HIV/TB patients is a measure of their level of immunity (18). In the earlier stages of HIV disease the clinical features is typical and similar to that seen in HIV negative patients. The manifestation is often pulmonary with infiltrates and cavitations in the apical posterior segments of the upper lobe and the superior segment of the lower lobe of the lungs. As the level of immunosuppression increases the presentation becomes atypical resembling primary TB with interstitial non-cavitary lesions because of poor granuloma formation and these involve more of the lower lung fields (55). At the terminal stage of HIV/AIDS extra pulmonary presentation

involving single or multiple sites are commoner along with miliary and disseminated disease i.e., involvement of two or more non-contagious sites

DIAGNOSIS

Definitive diagnosis of TB in HIV infected patients requires the isolation and identification of *MTB* from the culture of the infected tissue or fluid; a presumptive diagnosis is often made from microscopic observation of acid-fast bacilli (AFB) in the stained smear of sputum (56). AFB are rod shaped organisms with large amount of lipid in their cell walls making them difficult to stain but once stained resist decolourization even when washed with 95% alcohol containing 3% hydrochloric acid, thus the characteristic acid-fast property (57&58). Laboratory diagnosis is by Ziehl-Neelson (ZN) or Kinyoun or Tan Thiam Hok staining procedures all of which utilize carbolfuchsin (57). The former is heat fixed while the latter two are cold staining methods that require increased concentration of phenol in the staining solution. ZN staining with light microscope is the most commonly used methods of the three, it is however, time consuming and have low sensitivity requiring at least 10 (5) of tubercle bacilli per ml of specimen for reliable routine diagnosis (58). However, auramine-rhodamine staining technique with fluorescence microscope is a much faster and sensitive alternative. Properly collected sputum smears that fail to demonstrate AFB do not exclude the diagnosis of TB; because post-primary TB, the main source of infection, or re-infection in a given population is smear positive in about 50% of cases (3&59) while primary and miliary TB are smear positive in less than 25% of the cases (60&61). These percentages decrease further in the HIV-seropositive population because of their lower propensity to develop cavitory disease. Invasive procedures such as bronchoscopy with

transbronchial biopsy may be necessary to establish the diagnosis of TB in them because of this high rate of smear negative PTB and increasing cases of extra pulmonary TB (EPTB) as well as other opportunistic diseases that may resemble TB in presentation (62-64). *MTB* can be differentiated from other mycobacterials that could equally infect HIV/AIDS patients by culture (65), but the commonly used Lowenstein-Jensen culture agar requires 4-8 weeks for adequate growth to allow identification (66). Recent improvement in methods of mycobacterial specification is with the use of radiometric technique (BACTEC method). The technique uses radio labeled palmitic acid, a substrate that is metabolized to released (14) CO₂, which is quantified to identify presence and growth of the mycobacteria (66&67). The BACTEC system allows detection of *MTB* growth with a mean detection time of 7-13 days for smear-positive and 14-22 days for smear-negative sputum specimens (66). Rapid diagnosis of TB is also possible with molecular amplification and identification of *MTB* specific DNA or ribosomal RNA sequences by polymerase chain reaction (68). For epidemiologic purposes, patterns of infection within a population could be studied, with identification of the points of transmission by restriction fragment length polymorphism also referred to as "DNA fingerprinting," this is a molecular biology technique that allows differentiation of unrelated strains of *MTB* by demonstration of nucleotide sequence differences at selected sites in their DNA genome (69).

WHO recently advocated the screening of all TB patients for HIV infection, but this has not been universally accepted by the clinicians especially in the developing countries for reasons that include; increase in the cost of patients care (70), regional variation in the prevalence of the TB/HIV co-infection (7) and the fact that DOTS if properly

implemented is effective in curing TB in most patients regardless of their HIV status (17). However, offering voluntary counseling and HIV testing (VCT) to TB patients is beneficial because early diagnosis of HIV infection in TB patients has been associated with a good prognosis in terms of TB cure and it minimizes the negative effect of TB on the course of HIV (71). Patients could as well plan for the future. HIV co-infection should therefore be suspected in TB patients with history of risky life style, or in a TB patient that does not show prompt sputum conversion while on standard anti-TB regimen. It should also be considered in TB patients with chronic diarrhea or mucocutaneous lesions such as oral thrush, multidermatomal herpes zoster, non specific generalized dermatitis or co-existing sexually transmitted disease. Patients with extra pulmonary or disseminated TB should also be evaluated for dual infection. These categories of TB patients should be offered VCT.

TREATMENT

Short course of anti-TB chemotherapy is as effective in HIV positive TB patients as it is in the HIV negative ones, sputum conversion is rapid and cure rate is good (72). However, drug compliance is often very poor (73) and this could encourage emergence of potentially incurable multidrug-resistant TB (MDR-TB). This is however, common in poorly managed TB control programmes, especially those that lack the basic elements of good control (74), (prevalence of MDR-TB in Nigeria is 1.7% (7)). Successful treatment of TB in HIV patients therefore, depends on early diagnosis and effective application of DOTS where anti-TB will be free and patients observed to swallow each pill. In order to increase patients' easy access to free ARV drugs the existing local DOTS infrastructure could be utilized to deliver both medications in a DOT-HAART programme (75).

However, where DOTS is not fully feasible, self-supervised patient-centered care should be encouraged to forestall non-compliance. Current guidelines (74&76) recommend that HIV infected patients with drug susceptible TB be started on the standard six-month regimen of four drugs and that treatment should be initiated with isoniazid (INH), rifampicin/ rifabutin, pyrazinamide and ethambutol, all in mg/kg body weight for the first 2 months followed by rifampicin and INH for the subsequent 4 months. Intermittent anti-TB drug administration is not advisable in the management of HIV/TB and if it is to be adopted at all, the thrice weekly regimen should only be tried during the continuation phase of therapy (77). However, daily drug intake is preferred to the intermittent regimen so as to prevent development of MDR-TB (74&76). Response of HIV/TB patients to this 6-month therapy has been found to be good with similar recurrent rate to that of HIV-negative patients (78&79). A higher recurrence rate was however, found in one report (80), but this was ascribed to re-infection rather than treatment failure. A multi-centered study may be needed to compare the outcome of short course anti-TB management in HIV patients with early and well advanced disease (81). At the interim, prolongation of the continuation phase to 7 months to make a total of 9 months of treatment has been suggested if there is evidence of a delayed clinical or bacteriological response to therapy (74&76). The practice of extended post-treatment INH therapy to prevent recurrence of TB in HIV patients has not been widely accepted though some have found it effective (82). The same principles of treatment for PTB in HIV infected adults also apply to EPTB. The drug regimens and treatment durations are the same (76&83). However, for certain forms of EPTB, such as TB meningitis, bone, or joint, using rifamycin-based regimens for at least 9 months is

generally recommended (76&83). A non-rifampicin based anti-TB regimen comprising INH, pyrazinamide and ethambutol is generally not recommended for treatment of HIV-related TB, but in cases of rifampicin intolerance, severe allergy or toxicity, this regimen should be administered daily for 18 months (83).

In order to limit mortality from HIV/TB co-infection anti-TB drugs may have to be co-administered with antiretroviral (ARV) drugs in those with advanced disease (84). However, treatment of TB should be given priority if possible to avoid drug induced hepatic reactions from the dual potential hepatotoxic combinations. Initiation of ARV should be based on CD4⁺ count and risk of disease progression (74&76). There is yet to be a consensus on the time to introduce ARV. Tables 1 and 2, show that commencement of ARV in HIV/TB should be individualized and balanced between potential overlapping drug toxicities, drug-drug interactions and increasing TB morbidity from immune reconstitution reactions, against the reciprocal beneficial effect each therapy have on each other, which is to the benefit of the patients (74&76). HAART improve immune responses to TB and reduces the risk of relapse and re-infection, while anti-TB drugs results in quick lowering of the viral load thereby reducing the rate of CD4⁺ cells loss (85&86). Good as this combination therapy may sound, it is associated with bi-directional pharmacokinetic interactions between the rifamycin components of anti-TB regimen and the protease inhibitors (PIs) and non-nucleoside reverse transcriptase inhibitors (NNRTIs) components of antiretroviral therapy (87). Therefore diligent consideration of choice of drugs becomes imperative to prevent or minimize these drug-drug interactions. PIs and NNRTIs may inhibit or induce hepatic cytochrome (CYP-450) isoenzymes and thus alter the serum concentration

of rifamycins (87). Rifamycins in turn can induce CYP-450 and therefore substantially decrease blood levels of the ARV drugs too. Rifampicin is the most potent CYP-450 inducer of all the rifamycins (88), followed by rifabutin, rifapentine has an intermediate activity (89&90). Only rifampicin is currently available in Nigeria. All the PIs inhibit CYP-450 (91&92) with ritonavir as the most potent and saquinavir as the least potent while indinavir, nelfinavir have intermediate inhibitory properties. The available NNRTIs have diverse effects on CYP-450: nevirapine is an inducer, delavirdine is an inhibitor, and efavirenz is both an inducer and an inhibitor (92&93). In contrast to the PIs and the NNRTIs, the other class of ARV drugs, NRTIs; zidovudine, didanosine, zalcitabine, stavudine, and lamivudine are not metabolized by CYP-450, therefore, concurrent use of NRTIs and rifamycins is not contraindicated. Also, no contraindication exists for the use of NRTIs, NNRTIs, and PIs with INH, pyrazinamide, ethambutol, or streptomycin. These first-line anti-TB medications, in contrast to the rifamycins, are not CYP-450 inducers or inhibitors. Commonly prescribed ARV for co-infected patients in Nigeria includes zidovudine or stavudine with lamivudine and efavirenz, other approved combinations are also possible as most of the HAART drugs are now available in the country. Efavirenz is contraindicated in pregnant women because of its teratogenicity. Squanvir / ritonovir or abacavir with stavudine or zidovudine and lamivudine are the alternative to it.

Adjunct therapy with cytokine inhibitors may have a role in the management of HIV/TB co-infection (94) to limit HIV replication before initiation of ARV. Thalidomide, a specific TNF- α inhibitor and pentoxifylline a nonspecific inhibitor have been tried (95). However, inhibition of TNF- α was associated with profound immune defects

akin to that of advanced AIDS and these predisposed patients to reactivation of old TB, which progressed to disseminated form (95). Full understanding of the role of cytokines inhibition in the pathogenesis of reactivation of TB in HIV/TB is therefore required. Good nutrition, including food supplements will serve as essential adjuncts to anti-TB chemotherapy.

PATIENTS MONITORING. All HIV/PTB patients should be monitored by sputum smear microscopy during treatment and if available, sputum culture and susceptibility testing. HIV-EPTB should also be monitored, but the frequency and types of evaluations will depend on the involved sites and the ease with which specimens can be obtained from these sites (81). In resource limited countries, a monthly clinical assessment has been recommended for three months followed by 3-6 monthly clinical evaluation (96) to monitor improvement, ARV adherence and to identify possible drug reaction (96). Clinical and laboratory assessment should be more frequent for patients with underline liver disease. In most developing countries viral load measurement is unavailable and monitoring of therapy is by regular clinical assessment for signs of disease regression, increasing body weight and rising CD4⁺ count (97). Laboratory monitoring is prioritized by observing the trimmed down WHO guidelines for HIV treatment in poor countries (98). These are inexpensive tests that have been divided into basic and desirable tests and include haemoglobin, white cells and total lymphocyte count, liver enzymes and CD4⁺ cells. These should be done every 3-6 months. Serum amylase, bilirubin and lipids are also desirable but optional (98). Some HIV TB patients could experience temporary exacerbation of symptoms and signs of TB, and some may show worsening of the radiographic features of TB at the beginning of anti-TB treatment. This phenomenon

is termed a paradoxical (or immune reconstitution) reaction (96). It could also occur among HIV negative TB patients, but it is commoner amongst HIV/TB co-infected while on HAART (99). Features of a paradoxical reaction include high fevers, increase in size and inflammation of involved lymph nodes, new lymphadenopathy, expanding central nervous system lesions, worsening of pulmonary parenchymal infiltrations, and increasing pleural effusions (99). A reaction that is not severe should be treated symptomatically with non-steroidal anti-inflammatory agents without a change in anti-TB or ARV therapy (100&101). Those with severe reactions (e.g. airway compromise from enlarging lymph nodes, enlarging serosal fluid collections, and sepsis syndrome) may benefit from the prednisone or methylprednisolone 1mg/kg body weight and gradually reduced after 1--2 weeks (100&101).

CHEMOPROPHYLAXIS.

Chemoprophylaxis against TB in HIV positive patients may not be strongly advocated in a developing country like Nigeria where TB is an endemic problem. The value of a positive tuberculin test may also be difficult to determine because BCG is routinely administered at birth. In addition INH prophylaxis may be abuse and this could encourage INH-resistance when decision is finally made to treat active TB.

Table 1. WHO. Suggested timing of HAART in HIV/TB co-infection (WHO- Dec 2003)

Cd4+ count	Recommended regimen	Comment
CD4+<200/mm ³	Start TB treatment as soon as possible	Start ARV as soon as anti-TB is tolerated. EFV is contraindicated in pregnant women.
CD4+ 200-350/mm ³	Start TB treatment.	Start ARV after intensive phase of anti-TB.
CD4+> 350/mm ³	Start TB treatment	Defer ARV*
CD4+ not available	Start TB treatment	Consider ARV**

*Unless non-TB stage IV conditions are present. Otherwise start ART upon completion of TB treatment.**If no other signs of immunodeficiency are present is improving on TB treatment, ART should be started upon completion of TB treatment.

Table 2. BHIVA. Suggested timing of HAART in HIV/TB co-infection

CD4+ count cells/uL	When to treat with HAART
<100	As soon as possible-dependent on physician assessment, [Some physicians delay up to 2 months]
100-200	After 2 months of TB treatment
>200	After completing 6 months TB treatment*

<http://www.bhiva.org> * BHIVA treatment guidelines for TB/HIV infection – February 2005

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POLIO ERADICATION IN NIGERIA: – CONTROVERSIES AND WAY FORWARD.

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ABSTRACT:

With the commitment to global goal of Polio eradication by the year 2002, Nigeria embarked on the Polio Eradication Initiative (PEI) campaign in 1996 using the fixed post and later house to house strategy on days designated as National Immunization days (NIDs). The National average coverage reached between 40-60million children by 2002-3.

The strength of these successes had always been in the un-relented political support of the various levels Government, as well as the continued financial and material support of the International agencies – WHO, UNICEF, Rotary Int., BASICS and EU.

But despite this achievement, Nigeria currently poses the highest risk to the global target on Polio eradication. In 2003, Nigeria had the highest number of polio cases anywhere in the world (cumulative total of 302 as at Jan 2004), and accounted for 45% of all confirmed cases globally. Intense transmission of Wild Polio Virus (WPV) continues in Northern Nigeria especially the mega Kano state accounting for 47% of all cases. Unfortunately also, there has been a 25% increase in the number of cases of wild polio virus with at least a confirmed case in 15 States.

Controversies based on unfounded rumours about alleged adverse health effects, vaccine safety, contamination, overdose as well as promotion of anti OPV sentiments by political and religious opinion leaders motivated by political sentiments have led to decline in service demand and acceptance. These have further resulted in the previous cancellation of polio-vaccination campaigns in several key Northern Nigeria states. The outcome of various governmental and religious committees on the said controversies at a point in time recommended outright suspension of the exercise, while the OPV testing was being carried out in International laboratories outside the country. Despite the assurances on the safety by both the WHO and the Federal Government after the result of the testing was released, controversies on the authenticity of the results, country of testing as well as court cases are now emerging particularly in the northern states.

The consequences of these are the enormous additional financial resources as well as re-infection from the North of the previously polio free areas within and outside Nigeria, thereby threatening the children in the southern Nigeria as well as across west and central Africa.

This situation therefore calls for urgent actions to attain the goal of Polio eradication. These includes restoration of public confidence on OPV safety, immediate restart of National immunization campaigns, active commitment of all stake holders at all levels, improved quality and coordination of campaigns, sincere commitment of traditional, opinion, religious leaders, community ownership, Integrated social mobilization and Information disseminations.

In conclusion, there is a need for a strong consensus among all stake holders, regaining public confidence on vaccine safety, building on previous experiences, high quality manpower, committing more resources, and improved quality and coordination of vaccination programme for Nigeria to overcome the current situation.

KEY WORDS: POLIO ERADICATION, CONTROVERSIES, WAY FORWARD.

INTRODUCTION.

Nigeria is a signatory to the declaration of the survival, protection and development of children, which was articulated by the 49th World Health Assembly in 1988 and reinforced by the World summit for children held in New York in 1990. (1,2) This declaration established the challenges for global immunization, which include eradication of Poliovirus from the world by the year 2002.

Poliomyelitis has been known for at least 3,000 years – ancient Egyptians engravings pictured the crippling effects of Polio. At its peak, Polio paralysed or killed about 500,000 children each year. It is highly infectious disease transmitted through oral faecal route usually in children less than five years (3).

Unfortunately, there has been no known cure and its paralytic effect is irreversible. Between 5-10% of the cases of Poliomyelitis die of breathing muscles paralysis while paralysis occurs in less than 1% of cases and 90% will have no distinctive symptom at all. About 200 children are at the risk of being infected before a paralytic case finally signals an outbreak (3).

With the development of an effective vaccine, prevention of Poliomyelitis became possible in majority of countries around the world. It was the development of an orally administered vaccine that made poliomyelitis the logical choice for a global eradication programme after the successful story of small pox (4).

This logic is particularly relevant in many developing countries where limited resources, poor existing infrastructure as well as constrained local health care services are very important limiting factors in childcare including immunization services. This logic was envisaged feasible because of availability, affordability and ease of administration.

The Global Polio Eradication (GPEI) spearheaded by National Governments, and many international agencies – WHO, UNICEF, CDC, Rotary International is the largest public health initiative the world has ever known. In the last decade, through improved immunization efforts and strategy, the global coverage has reached over two billion children around the world in more than 200 countries with over 20 million participating volunteers including Nigeria. There was also a corresponding significant decrease in polio cases from about 350,000 in 1988 to less than 4,000 by the end of the year 2000.⁵ This is with a view to eradicate the sufferings, paralysis and death associated with the disease and to save about 1.5 billion US dollars annually for other health issues (6).

During the same period, the repeated polio immunization campaign had interrupted polio transmission throughout the southern Nigeria and most especially the mega city of Lagos. The strength of these successes had always been in the un-relented political support of the various levels Government, as well as the continued financial and material support of the International agencies – WHO, UNICEF, Rotary Int., BASICS and EU (7).

Fifteen years has elapsed since the decision to eradicate polio was passed and we are now in the last phase. The end of any global eradication campaign is the most complex. At the end stage, there are usually very few cases of the target disease left and sometimes question the rationale for commitment of both enormous financial and human resources as it were in the beginning. This is coupled with personnel as well as public apathy due to programme fatigue, mistrust and dose safety. Several regions of the world among them the Americas, Western Pacific and Europe have already been certified polio free and are now awaiting the rest of the world for global

eradication. Polio free zones are also emerging in Africa particularly the Southern and the Eastern zones (4,7,8).

However, there are seven remaining polio-endemic countries in the world. The three critical ones are India, Nigeria and Pakistan. The eradication initiative has continued to be a success story in both India and Pakistan. However, this PEI success story has been difficult to sustain with the advent of the democratic dispensation. The recent cancellation of polio-vaccination campaigns in several key Northern Nigeria states where poliomyelitis is endemic (9) has focused attention of the whole world on the unfortunate reversals in the fight to eradicate polio.

Nigeria currently poses the highest risk to the interruption of poliovirus circulation and global target for eradication. In 2003, Nigeria had the highest number of polio cases anywhere in the world (302 as Jan 2004), and accounted for 45% of all confirmed cases globally. Intense transmission of Wild Polio Virus (WPV) continues in Kano state accounting for 47% of all cases. There has been also a 25% increase in the number of cases of wild poliovirus with at least a confirmed case in 15 States (10).

Polio Eradication Initiative in Nigeria

With the commitment to global goal of Polio eradication by the year 2004, one major step taken by Nigeria in achieving this was the restructuring of the Expanded Programme on Immunization (EPI) in 1997. This led to renaming the EPI to National Programme Immunization (NPI) and its establishment as a parastatal of the Federal Ministry of Health (11).

Nigeria implemented the PEI with days designated as National Immunization days using the fixed post strategy between 1996 and 1998. The objectives of this strategy were to immunize all children aged 0-59 months and active acute flaccid paralysis (AFP)

surveillance. The National average coverage reached 108% by 1998 (12). But despite these achievements, there were still identified cases of wild poliovirus. This obviously necessitated a change in the strategy in 1999 to the house to house.

The objectives of the new strategy included (12)

- Immunization of all children under the age of five with oral polio vaccine.
- Strengthening of routine immunization.
- Concept of revisiting all missed houses.
- Active surveillance to detect cases of AFP.
- Mop-up vaccinations campaigns in areas where WPV has been isolated.

This strategy led to remarkable achievements and progress in the Nigeria PEI and by the end of 2002, between 40 and 60 million Nigerian children under the age of five years were already immunized (13) using part of the over 10 billion doses of OPV in worldwide polio campaigns (14). This was also coupled with the reduction of AFP cases from over 100 to only 63 in 2001 (15). This is an affirmation of both potency and safety of the OPV used in the Nigeria campaign.

During the same period, the repeated polio immunization campaign had interrupted polio transmission for least 18 months throughout the whole of southern Nigeria and most especially the mega city of Lagos (16,17). The strength of these successes had always been in the un-relented political support of the various levels Government, commitment of the NPI Agency, as well as the continued financial and material support of the International agencies – WHO, UNICEF, Rotary Int., BASICS and EU.

CURRENT TREND IN NIGERIA

Between January and July 2003, a total of 75 wild polioviruses have been confirmed, with 45 cases being type 3 while 29 cases are type 1. About 72 % (54) of the cases were below the age of three, while 64% (48) had received less than 3 doses of OPV.¹⁰

There has been a 25% increase in the number and the spread of cases of wild poliovirus between Jan-July 2003, when compared to the same period in 2002. Also at least a case of confirmed wild poliovirus has been isolated in 15 states compared to 12 states of 2002 during the same period. Nigeria has now more than 302 cases of WPV and may be the largest last reservoir of polio virus on earth. Twenty states are infected with varying levels of virus load in 2004 (10).

Intense transmission of wild poliovirus continues in Kano State, accounting for 82 cases of wild poliovirus. Other states namely Kebbi (37 cases), Jigawa, Katsina, Zamfara, Kaduna, Sokoto, and Bauchi also continue to be reservoirs of wild poliovirus. Emerging cases are now being isolated in the once interrupted polio transmission southern states of Kwara, Lagos, Ogun and Edo to mention a few as a result of re-infection from the Northern part (10,16,17).

The performance of the surveillance system continues to be good; all the states in the federation have achieved the target rate of 80% stool adequacy and a non-polio rate greater than 1/100,000 children below 15 years of age (10).

Programme reviews have been conducted and new strategies have been developed and recommended to be implemented for the rest of the year 2004.

CONTROVERSIES

Unfolded rumours about alleged adverse health effects, vaccine safety, contamination, overdose as well as promotion of anti OPV sentiments by political and religious opinion leaders motivated by

political sentiments have led to rejection and or decline in service demand and acceptance (18,19, 20). This has led to free circulation of the wild poliovirus in the Northern Nigeria and re-emergence in the previously free southern part. Most unfortunate of all, the country is now an exporter of the poliovirus (21).

Ironically, some of the early countries to get rid of polio disease are front line Islamic states – Saudi Arabia, Syria, Iran, Jordan, Kuwait and Morocco. Countries facing rigours of war even made cease fire arrangement in-order to make progress in the eradication drive (8).

These controversies are thus always based on un-named "competent" sources mainly and usually subjects of religious sermons; editorials in both print and electronic media as well as journals. The controversies quoted in previous media reports are based on some of the under-listed issues (22, 23, 24,25).

- The result of the initial test that "the vaccines indeed found to contain a congenous steroid" on some of the vaccines at University teaching hospital and National Hospital, where there are more sophisticated equipment.
- Recommendation of further confirmatory vaccine test at International Laboratories in Lagos, which are considered multinational and seen as part of the agenda.
- WHO and UNICEF official rejections of the positive tests conducted in ABU/Abuja as inaccurate and their readiness to sponsor vaccine test abroad.
- The controversies on composition of investigating team.
- Test on the vaccine to include the SB 40, SIV and HIV.
- Citation of a previous similar experience with WHO in the Philippines in the 1990s

on the administration of later contaminated Tetanus vaccine.

- Desperation and aggressiveness of WHO to have children immunized despite the irresolution the controversies.
- The spending of billions of US dollars on a disease that recorded only 109 cases in the entire country, when devastating disease like measles killing two million children annually.
- Medical hypothesis reports by renowned scientist and research on the linkage between polio virus and the origin of HIV published in 1994 after the 8th annual Houston conference on AIDS.
- The motive behind introducing a potentially dangerous vaccine, proven, and ban for use in the United States and other European countries.
- American declassified document called national security memorandum 200 on the issue of population control in developing countries to ultimately serve the strategic, economic and military interest of the United States.

THE WAY FORWARD

Today, Nigeria still remains the number one polio reservoir in the world not because the country does not have the expertise, but because of misinformation and deliberate suppression of rights of the underprivileged.(8). This situation therefore calls for urgent consensus to regain the restoration of public confidence on the eradication initiative.

The consensus must be based on safety of the OPV and the principles should include the demonstration of the commitment of Government to work closely with all stakeholders. The strategies should include (13,15,26,27).

- Sustenance of the ongoing dialogue with both the political and traditional leaders of the North through the appointment of Polio Ambassador.
- A grand plan of action on monthly basis should be established and managed under the office of the Minister for Health addressing key issues of social mobilization, advocacies, political, community as well as private ownership.
- The continued broad base financial and technical support of the partner agencies to immediate restart of National immunization campaigns, improved quality and coordination of campaigns.
- Media should come fully on board and their organisational channels should be extended for dissemination of OPV safety information among communities and households.
- Active support of leaders particularly State, local and traditional leaders to take active part and own immunization campaigns, especially in the northern states.
- Doctors individually or as groups particularly those based in the northern part of the country need to join effort with health authorities and international agencies by studying the OPV test report as well as develop a strong advocacy programme in dispelling these rumours.

In conclusion, there is a need for a strong consensus among all stake holders, regaining public confidence on vaccine safety, building on previous experiences, high quality human power, committing more resources, and improved quality and coordination of vaccination programme for Nigeria to overcome the current situation.

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most frequently injured. The nature of work, the exposure rate to potentially infected blood and body fluids, and the exposure to contaminated materials at work place makes these groups especially vulnerable. Results of a study on the epidemiology of NSI among Health Care Workers (HCW) in two German hospitals indicate that 500,000 NSIs occur annually in Germany (4). NSI are also preventable sources of infection and stress of HCW in Africa. A study in Uganda found that needlestick injuries are common with an estimate of about 55% of HCW reporting at least one NSI in the previous year (5). In a study carried out in Benin City, Nigeria, amongst nurses, it was found that the nurses had a poor knowledge about universal precautions, as only 34.2% of had heard of universal precautions. There was also a poor observance of universal precautions. Knowledge of measures to be taken after the occurrence of occupational accidents and needlestick injuries was also poor (6). Similarly, 44.6% of surgical residents in Nigeria in a study conducted in 1997, had an idea of CDC guidelines for Universal Precaution against bloodborne infections, 42.2% knew it well, while 13.8% had no idea (7). This descriptive cross-sectional study was designed to look into the level of awareness and patterns of needlestick injuries among health care workers at the University of Ilorin Teaching Hospital Ilorin, Nigeria. The study covered workers who either use, carry or dispose needles and sharps. They include doctors, nurses, laboratory workers, attendants, ward aides and porters. It was carried out between January and June 2004.

METHODOLOGY

The study was conducted in the University of Ilorin Teaching Hospital. It is a tertiary and referral health institution located in the north central part of Nigeria. Majority of the HCWs in the teaching hospital are nurses and medical doctors, 51.1% and 35.4% respectively. Another group that is sizeable is the supporting staff of hospital attendants, aides and porters accounting for 13.5% of respondents. The 320 subjects for the study were selected out of the 1180 staff involved with

direct patient care. Proportionate simple random sampling method by balloting was used to select these study subjects based on their respective total numbers in their departments. The tool for data collection was a pre tested structured questionnaire that was completed by the respondents themselves. Epi-info version 6.0 computer software package was used for data analysis.

RESULTS

Three hundred and twenty copies of the questionnaire were distributed out of which 294 were found suitable for analysis after editing. Of the 294 respondents, 274 (93.2%) knew of the existence of needle stick injuries as a health hazard while 262 (89.1%) believed they were exposed to the hazard. The most commonly known methods of disposal of used needles were safety box (63.9%), incineration (60%) and dumping (24.3%). Others were burning 3.4% and recycling (6.8%). Most commonly known infections associated with needle-stick injuries were HIV/AIDS (96.6%), Hepatitis B (85.4%), Hepatitis C (37.1%), Hepatitis A (32.1%), Bacterial infections (27.2%), Hepatitis E (10.5%) and Malaria (4.4%). The commonly known methods of preventing needle-stick injuries were proper disposal (76.5%), recapping of needles (42.5%), gloving/double gloving (26.9%), use of special operating gloves (22.2%), health education of workers (21.3%), stop recapping (13.4%) and better concentration while performing procedures (6.7%). Only 13.4% mentioned no recapping of needles as a way of preventing NSI. Disposal methods employed for used needles and sharps were safety boxes (47.9%), baskets (46.9%), plastic cans (40.8%) and kick-about bowls (4.8%). Eighty-one point three percent (81.3%) of respondents usually recap used needles while (18.7%) did not. One hundred and eighty five (62.9%) of respondents has had at least one NSI before (table 1) with 57.8% of them having had one within the last 12 months preceding the study. All but one (99.5%) of those who had the NSI experience had their hands affected. One respondent had the needle puncture on the foot. Eighty (45.5%) experienced the injury during the day 38.2% during the

afternoon and 13.3% during the night. Fifty-nine point four percent (59.4%) of needle stick injuries occurred during injection procedures with 43.6% of them occurring during re-capping of needles after injections, 22.7% occurred during operations, 14.1% during intravenous manipulations while 6.5% was during other procedures such as packaging of linen and wastes. The main actions taken by those affected by NSI include

squeezing the site (63.2%), washing with soap (41.6%), washing with bleach (28.6%) and pressing the site to stop bleeding (20.5%). Other things done (7.6%) include using antibiotics, cleaning with spirit, bandaging and an individual was given post exposure prophylaxis with antiretroviral drugs. Only 22 people (7.5%) reported and these to their immediate boss, the compound master or staff clinic.

Table 1: Pattern of needle stick injuries among health care workers in various departments of UTHS Ikorin, Nigeria

Department	Yes	No	Total
Medical	26	17	43
Surgical	37	6	43
Laboratory	10	19	29
Nursing	101	57	158
Others	11	10	21
Total	185	109	294

Chi Square = 21.09 df = 4 P = 0.0003

The department to which a respondent belonged significantly influenced the occurrence of NSI (table 1). Respondents in the surgical departments (surgery, obstetrics and gynaecology, ophthalmology) and nursing were more likely to suffer from needle stick injuries than others. The medical departments constitute community medicine, paediatrics and internal medicine.

DISCUSSION

One of the most important occupational risks among health care workers is exposure to blood-borne pathogens. Needle-stick injuries (NSI) are recognized as posing this great risk to HCWs. The most important of these diseases are Human Immune-deficiency Virus (HIV), Hepatitis B (HBV) and Hepatitis C (HCV). The commonest is HBV, but HIV and HCV are of more

concern because there are no vaccines for them and no satisfactory treatment available yet. In this study, the level of awareness of needle-stick injuries can be described as high for 93.2% knew that it exists, 89.1% of respondents agreed they are exposed to the injury. The awareness of risk of contracting diseases from needle-stick injuries was also high as indicated by the response to the risk associated with the injuries. More than 90% of respondents knew that HIV/AIDS can be transmitted to health workers through needle stick injuries.

Ninety- six point six (96.6%) of respondents indicated HIV/AIDS which agrees with the study in Ilorin, Nigeria in which 97% of the health workers know that HIV can be transmitted by needle-stick injuries (8). Injection safety involves not exposing the patients, the health workers and

the community to dangers. The disposal of used needles safely is a very important aspect of prevention of needle stick injuries as advocated by the World Health Organisation (9). In the literature, recapping of used needles is a popular culprit in the cause of needle stick injuries. A significant 24.3% of respondents still picked dumping as an option. This is worrisome since dumping is not safe and it may be an indication that dumping is a major method of disposal system of most things in the study area 21. This common practice of waste disposal in the tropics, exposes scavengers in the community to the danger needle stick injuries (10&11). While almost all (94.6%) of respondents believed that NSI can be prevented, the methods suggested are not completely in agreement with international standards. Universal precaution recommends non-recapping of used needles and sharps. In this study 42.5% of respondents recommended recapping as a method of preventing NSI, while 13.4% suggested non-recapping. This showed a low level of knowledge concerning the issue of recapping as a cause of NSI. This evident in the practice of recapping by the majority (81.3%) of the health workers. In this study, majority of respondents, 81.3% usually recap needles after use. This was not unexpected because 42.5% mentioned recapping as a way of preventing NSI. Universal precaution standards prohibit recapping, but it continues to be an identified cause of NSI (12). The proportion of respondents (62.9%) that had experienced NSI is high. This shows a high exposure level to blood from patient and 36.4% of the respondents had experienced NSI in the preceding 12 months. This proportion is similar to rates found in studies in other teaching hospitals in Nigeria and Uganda (5&7). In the study in Ilorin, Nigeria, it was found to be 34% in the previous year (13). These figures are much less than 88.4% of reported NSI among surgical residents in Nigeria (7). This finding is in support of the higher risk to which surgeons were exposed when it involves exposure to blood and body fluids (14&15).

The main sites involved were the fingers for all but one (99.5%) respondent. It is the hand that is involved in the handling of needles and syringes, in packing linen, in suturing, in setting intravenous lines and in recapping of the needles. It is thus the most vulnerable and affected part of the body by needle stick injuries. Most actions taken by victims to manage the NSI were palliative. An individual who had NSI from a known HIV positive patient had to take post exposure prophylactic (PEP) antiretroviral drugs. This might be an indication that the action taken could be affected by the knowledge of the danger to which the health care worker is exposed. In environments of high prevalence of HIV positive individuals, PEP has been recommended (13&14). There was poor reporting of NSI in this study as only 11.9% of those who had the injuries reported. This observation had been made by other workers in other parts of the world (4 &16). The actions taken by those who reported were not different from those by the individuals who did not report. This might be an indication of the absence of a standardized protocol of managing such injuries as NSI in the hospital. Table 1 shows the effect that the department to which an individual belongs had on NSI experience. The health workers in the surgical departments (surgery, ophthalmology, obstetrics and gynaecology) had higher incidence of NSI. The nurses are also more likely to have NSI. Surgeons operate and use needles in suturing, sometimes doing blind suturing and thereby exposing themselves to the danger of NSI (15&17). It was also found in other surveys that suturing account for a significant proportion of NSI amongst surgical staff who were also relatively more injured than other groups (17).

CONCLUSION AND RECOMMENDATIONS

Needle stick injuries are an important occupational risk to which health workers are exposed. There was a high level of awareness of the risks associated with needle stick injuries. Most of the workers in the teaching hospital knew about the risks and that they are also exposed to these risks. The level of awareness may be very high (as indicated by the knowledge of the risks and the known

method of disposal), but the injection safety practice does not measure up to the level of awareness. There the practice of unsatisfactory disposal system of used sharps as a sizeable proportion still use methods that can expose the population to the risk of injuries from used needles (dumping, burning, etc). There was a high incidence of NSI and the workers in the surgical departments were at higher risk. The hand was the most commonly affected by NSI in this study. Injection administration, recapping of needles and surgical operations were the main procedures associated with NSI. The health care workers in the teaching hospital should have training and workshops on injection safety. The management of the teaching hospital should have a policy on injection safety, which should include needle stick injuries, their prevention and management. Specially designed needles for surgical procedures, especially the blunt needles should be provided for use in the operating theatres and emergency units.

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this has failed to bring relief that other differential diagnosis is considered. The next stage of the disease results in the development of the usual telltale signs and symptoms of a viral haemorrhagic fever such as reddish conjunctiva, bullneck, sorethroat, and later bleeding from the orifices (mouth, Giltract, urinary tract and into other internal organs).

Whole blood clotting time in 20 minutes (WBCT20mins) is an indirect crude test used to evaluate bleeding tendency. It was originally used in snakebite patients bitten by viperidae species that are prone to development of haemorrhagic systemic envenomation symptoms (5) In this situation, it was found very useful in initiating antsnake venom therapy once a value beyond 20minutes is obtained. This was found to reduce mortality significantly.

The high mortality associated with lassa fever in which the ultimate cause of death is bleeding can be reduced if there is an early diagnostic tool that can be used to detect bleeding tendency early enough while awaiting serological diagnosis. This laboratory confirmation often comes late to alter the course of the disease favourably if prompt treatment is not instituted before bleeding commences.

This small pilot observational study was therefore done to see if this simple test will be beneficial in improving the diagnosis of lassa fever.

METHODOLOGY

All febrile adult patients presenting in Irrua Specialist Teaching Hospital, Irrua (ISTH), to the medical team both during clinic sessions and at the accident and emergency (A&E) units were examined. Patients were assessed clinically and routine investigation such as blood film examination and full blood count were done.

Patients were then stratified into various groups according to their clinical diagnosis.

A sample of blood was taken from each patient for WBCT-20minutes.

Whole blood clotting time in 20 minutes (WBCT20minutes) was done by modifying the original method of Lee and white to the bedside. 2mls of venous blood is collected from the antecubital vein into a 5ml syringe. A free space is left above the blood column so that the entire blood in the syringe is visible before clotting commences. The syringe containing the blood is left standing and tilted slowly to the horizontal after four minutes and every minute subsequently. The time taken for clotting to occur is then noted. A normal WBCT is between 5 – 12 minutes (less than 20 minutes). Values beyond 20 minutes are considered abnormal and this forms a basis for prompt commencement of intravenous therapy, which is the only treatment available so far for lassa fever in these environment (6)

Samples were also taken from an equal number of non-febrile age and sex matches medical students and hospital workers as controls.

All patients were then stratified according to their WBCT- 20minutes.

All patients were followed up and re evaluated clinical at one week to evaluate those who developed classical symptoms and were then treated for lassa fever.

Patients were compared with controls. Chi Square was used to analyse discrete variables and ANOVA for continuous variables.

RESULTS.

A total of thirty eight febrile patients were examined- male twenty (52.6%) females eighteen (47.4%) Clinical diagnosis of Lassa fever were made in five patients (13%), others were malaria twenty two (58%), meningitis five (13%), Enteric fever four (11%), lobar pneumonia Two (5%).

Table 1 Frequency of clinical diagnosis

Diagnosis	Frequency	Percentage (%)
Lassa fever	5	13
Malaria	22	58
Meningitis	5	13
Enteric Fever	4	11
Lobar pneumonia	2	5
Total	38	100

Table 2. Age/Sex distribution of patients

Age (years)	Male	Female	Total
10 – 19	3	4	7
20 – 29	9	5	14
30 – 39	6	4	10
40 – 49	2	3	5
50 – 59	0	2	5
Total	20	18	2

WBCT was prolonged in all the five cases of clinically diagnoses lassa fever. (32%) of the patients diagnosed as malaria had highly elevated WBCT – 20minutes. Only one of those with meningitis had elevated WBCT>20minutes and none of those with pneumonia had elevates values. On evaluation of patients 1week later, All seven patients diagnosed as malaria but with prolonged WBCT 20minutes developed classical features of lassa fever such conjunctiva suffusion, swollen neck, blood stained sputum and had to commences on ribavirin.

The five patients whose WBCT were elevated were promptly commenced on I.V ribavirin and there was both clinical improvement and later reduction of the initial WBCT value to less than 20mins after one week of therapy. No fatalities were recorded in this group. However seven patients previously diagnosed with malaria and meningitis that had initial normal WBCT (less than 20 minutes) and who did not respond to

adequate doses of antimalarial and antibiotics were later found to have elevated WBCT on repeat one week later. two resulted in fatalities because of delay in initial of ribavirin therapy.

The WBCT in 20 minutes for all the control subjects were less than 20 minutes.

DISCUSSION

Lassa fever is a deadly disease (1). It is grouped among the viral haemorrhagic fevers and incidentally was discovered in Lassa near Jos in Nigeria (2) It is endemic in some parts of West Africa namely Sierra Leone, Guinea, and Nigeria (1) The Northern part of Edo state of Nigeria, especially the Esan speaking area is one of such endemic zones and epidemics occur annually during the dry season when bush burning displaces the rodents harbouring the virus from their normal habitat in the wild into the neighbouring villages where the disease is now spread by the ingestion of foodstuff contaminated by the rat body fluids such

as urine, saliva, and feces (2) Others also get infected when they ingest the meat of the rats considered a delicacy by some people.

There is a yearly epidemic because no effective preventive measure has been found to reduce the annual increased incidence rate during the dry season. Rats and other rodents (2) that spread the virus are highly sought out source of protein because of the harsh economic climate and local hunters will spare no effort to trap them. Burning is a way of forcing them into the open to do this but unfortunately it also serves to drive them into human houses where they urinate and stool on drying foodstuffs such as fermenting cassava and garri left outside the house to dry.

Lassa fever is a dreaded illness in these parts of the tropics because of the high mortality rate once bleeding commences. Disturbance of hemostasis is a sine qua non in all viral hemorrhagic fevers including lassa fever (6) and bleeding is ultimately a prelude to early mortality in most cases. Other effects of illness include lymphocytopenia and a moderate thrombocytopenia (7) The thrombocytopenia is associated with a serum inhibitor and with the occurrence of hemorrhage, depression of platelet aggregation thereby increasing the severity of lassa fever (8)

Until fairly recently, the prognosis was poor (3) and there was no effective therapy until the introduction of ribavirin (4) The use of ribavirin in a patient therefore depended on diagnosing lassa fever. The diagnosis of lassa fever in our environment is totally clinical since serological tests for it are unavailable and viral identification tests are absent. Prompt and early diagnosis is essential since there are a myriad of common diseases causing febrile illnesses, chief among them in this environment being malaria, pneumonia, enteric fever, meningitis among others (1,2,3)

Patients in this area with febrile illnesses present late, most having presented to a chemist, a traditional healer, a nursing home or a private clinic before presenting in this hospital usually on account of poor response to initial therapy taken or due to worsening symptoms and signs.

Ability to stratify presumed lassa fever patients into a group to enable immediate therapy with Ribavirin and also to avoid antiviral therapy for patients with other causes of febrile illnesses is absolutely important.

There is no lassa vaccination programme yet to protect the population of endemic regions.

The result of initial study indicates that this simple test can be quite invaluable in improving the diagnosis and institution of therapy for patients with suspected lassa fever. This has led to a drastic reduction in the mortality rate compared to when treatment was delayed while awaiting the late coming serological and other investigations that have to be done in far away centers equipped for this

WBCT, though non-specific test (in literature reviewed) was nonetheless sensitive in increasing diagnostic suspicion in lassa fever. This increased sensitivity may be because patients with illnesses generally present late to the hospital in this environment and prolongation of WBCT is also a late sign.

CONCLUSION.

It is recommended that even if WBCT-20minutes is non-specific, because of the sensitivity noted in this study, its use in the investigation of all febrile patients should be encouraged especially patients who are listed as resistant malaria and for fever presenting for more than a few days.

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SEROPREVALENCE OF *BACILLUS ANTHRACIS* IN JOS AND ENVIRONS

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ABSTRACT

The fears of a possible out break of infection following media reports of intentional release of anthrax spores has drawn attention to the need to establish a baseline information on the prevalence of *Bacillus anthracis* in the cattle, sheep and goat population in Jos. We conducted a survey to determine the prevalence of antibodies to *Bacillus anthracis* in cattle, sheep and goats slaughtered at the Jos Abattoir using the agar diffusion method. The survey involved two hundred animal subjects (160 cattle, 20 each of sheep and goats). Out of the total number of samples screened, 21(10.5%) were seropositive, with antibody titre levels of $\geq 1/80$. Of the 21 seropositive cases, 18 (9.0%) were cattle, 3(1.5%) were sheep and there was none for goats. These findings allay our fears of the possibility of an outbreak of anthrax infection following the consumption of meat from animals slaughtered at the Jos Abattoir.

INTRODUCTION:

Anthrax is an acute infectious zoonotic disease caused by spore-forming bacterium *Bacillus anthracis* and affects wild and domestic animals (cattle, sheep, goats, camels, antelopes and other herbivores). It affects humans when they are exposed to infected animals (1). Historically, human anthrax in its various forms has been a disease of those with close contact to animals or animal products contaminated with *Bacillus anthracis* spores, (2).

Anthrax is a disease of well-documented antiquity. For example, the accounts of the fifth and sixth plagues of Egypt given in the ninth Chapter of Exodus are seen by many as describing instances of systemic and cutaneous anthrax (3).

In humans, anthrax most commonly occurs as cutaneous lesions or boils which usually progresses to a black eschar despite antibiotic

treatment. Pulmonary anthrax (inhalation) and a third form gastrointestinal anthrax usually resulting from ingestion of meat or spore-contaminated food may be observed (4).

Anthrax in animals may occur in the following forms: peracute, acute, subacute and chronic. The peracute form generally found in cattles, sheep and goats at the beginning of an outbreak is characterised by a sudden onset and rapidly fatal course. Some are found dead without any previous signs of the disease (5). In the acute forms, animals exhibit signs up to two days before death (3). The first noticeable sign/symptom is a rise in temperature and both the acute and subacute conditions are characterised by excitement, simulating rabies followed by depression, respiratory distress, trembling, staggering, convulsion and death.

Anthrax has become a current issue because it is considered to be a potential agent for use in biological warfare. The identification of inhalational anthrax in a journalist in Florida on October 4, 2001, marked the beginning of the first confirmed outbreak associated with intentional release of anthrax in the United States (2).

In Nigeria, a member of Senate was reported to have received a suspected mail, which turned out to be a hoax after thorough laboratory investigation at the Bacteriology Laboratory at the National Veterinary Research Institute, Vom. There is therefore a need to establish the prevalence of this potential agent of biological warfare in our environment.

The aim of this study is to investigate the seroprevalence of *B. anthracis* responsible for infection in cattle, sheep and goats in Jos and its environs.

MATERIALS AND METHODS

Sample Collection

Two hundred (200) blood samples from slaughtered cattle, sheep and goats were collected from the Jos Abattoir in McCartney bottles. One hundred and sixty (160) of the blood samples were obtained from cattle while twenty of the blood samples each were collected from sheep and goats. The blood samples were allowed to clot after collection and sera from the clotted blood samples were separated into clean dry Bijou bottles using clean pipettes. The sera were kept at -20°C until they were ready for use.

Experimental Animals

The selected in-bred female Hartley guinea pigs, 350-400g were quarantined for one week. They were bled by cardiac puncture and screened to be sure they had no antibody to anthrax. The clean animals were then kept for the experiment.

Animal Inoculation

The harvested live attenuated anthrax spore suspension in saline was washed using normal saline. Viable count of the spore was determined using Miles & Misra (6) method. 0.2% formalin was added to the washed suspension ensuring that the final concentration of the spore suspension remained approximately 2.0×10^9 cfu/ml. Each guinea pig was given four daily inoculation intravenously of 1cc^3 of the suspension. After two weeks, post last inoculation, the guinea pigs were bled, serum separated and tested for antibody titre. The standard antisera was then stored in deep-freezer until when required as positive control antisera.

Standard Antigen

A large batch of attenuated Sterne strain culture filtrate which had a titre of 1×10^2 cfu/ml in the agar diffusion assay with the standard antiserum was used as a standard control for comparing line patterns of other antigen preparation.

Preparation of Medium for Agar Diffusion Plates.

One gram of agarose containing 0.015M phosphate buffer (pH 7.3), 0.01% thiomersolate and 0.09% NaCl was dissolved in 1000mls of distilled water and was then autoclaved at 121°C for 15 minutes. It was allowed to cool to 50°C and 15ml of the agar medium were dispensed into 90mm Petri dishes. Circular wells were made in the solidified agar with cork borers. The outer wells were 7mm in diameter and 5mm apart and 6mm from the inner well of 5mm in diameter. The plates were stored at 2°C and used as required.

Agar Diffusion Method

An agar diffusion method for titrating antibody against antigen as described by Thorne and Belton (7) was used.

The outer wells were filled with appropriate dilutions of test antisera in saline (1/20,

1/40, 1/80, 1/160) and the plates were held at 2°C for 18-24 hours. The inner well was then filled with a neat solution of antigen after which the plates were kept at room temperature. The plates were observed after 18-24 hours for lines of precipitation by holding over a light against a dark background. The final readings were taken on the second day after titration.

Antibody titre values of 1/80 and below were considered as negative.

Method of Antiserum Dilution

One millilitre (1ml) of the working serum was pipetted into the first test tube in the rack containing 9ml of 0.9% saline solution to give a dilution of 1:10. Double dilution process was carried out for the rest of the tubes (i.e. 1/20, 1/40, 1/80, 1/160, 1/320). One millilitre (1) ml from the last test tube was then discarded into a disinfectant jar. Equal amounts of the respective dilutions were then used to fill the outer wells on the agar medium as was done above.

RESULTS

Two hundred serum samples were titrated against filtrate of Sterne strain of *B. anthracis* in nutrient broth. The highest dilution of test antiserum that prevented formation of a visible precipitation line was taken as the end point. The results obtained showed that a total of 21(10.5%) serum samples were positive. Of the total number of positive cases 18(9.0%) were from cattle (Table 1) while 3(1.5%) were from sheep (Table 2). There were no positive cases from goats (Table 3).

Six (3.75%) of 160 cattle serum samples tested had titres of 1:80, 8(5.0%) had a titre of 1:40, 4(2.5%) had a titre of 1:20 while none had titre >1:160. Only 1(5%) out of the 20 serum samples tested for sheep had titre of 1:80 and 2(10%) had titre values of 1:20. All the goats had no detectable antibody to *B. anthracis*. Titre values of 1:80 and

below have been used as standards for evaluation that animals have been vaccinated while titre values of 1:160 and above are suggestive of infection.

Table 1: Antibody levels to *Bacillus anthracis* in Cattle.

N=160

Sera Sample Size	1/20	1/40	1/80	1/160	1/320	1/640
4(2.5%)	+ve	-ve	-ve	-ve	-ve	-ve
8(5.0%)	+ve	+ve	-ve	-ve	-ve	-ve
6(3.75)	+ve	+ve	+ve	-ve	-ve	-ve
142(88.75)	-ve	-ve	-ve	-ve	-ve	-ve

Table 2: Antibody levels to *Bacillus anthracis* in Sheep.

N=20

Sera Sample Size	1/20	1/40	1/80	1/160	1/320	1/640
2(10.0%)	+ve	-ve	-ve	-ve	-ve	-ve
1(5.0%)	+ve	+ve	+ve	-ve	-ve	-ve
17(85.0%)	-ve	-ve	-ve	-ve	-ve	-ve

Table 3: Antibody levels to *Bacillus anthracis* in Goats.

N=20

Sera Sample Size	1/20	1/40	1/80	1/160	1/320	1/640
20(100.0%)	-ve	-ve	-ve	-ve	-ve	-ve

DISCUSSION

Results obtained from this study showed that 10.5% of the study population (animals) were seropositive for antibodies to *B. anthracis* with antibody levels 1:80. The low titre values suggest that animals have been vaccinated against the infection. The percentage positive was higher in cattle (9.0%) than in sheep (1.5%). The local herdsmen could attribute this to the routine

vaccination of cattle with the Anthrax spore vaccine.

In a study in Zambia, 365 specimens of various tissues from animals and surface water over a 5 year period (1987-1991) were examined for anthrax, 85 animals were positive. Of this number, 35 were in domestic animals comprising of 33 cattle, 1 sheep and 1 pig (8). Over 100 human deaths from anthrax, usually associated with eating infected meat were recorded in the Western and North Western provinces of Zambia between 1990 and 1993.

Sheep are usually not vaccinated against Anthrax in our environment but the few positives may be due to grazing on contaminated feed/grasses resulting from spills from the vaccine during vaccination of cattle. This is possible since the sheep are reared alongside the cattle. The fact that no goats, which are reared separately, recorded a zero percent seropositivity gives credence to the earlier assertion.

It has been mentioned earlier that the disease can affect humans when they are exposed to infected animals or tissues from these infected animals. This study suggest that the animal population examined (cattle, sheep, and goats) may not be capable of causing anthrax infection in Jos and its environs when ingested. There also has been no record of any outbreaks among butcher in the Jos Abattoir.

Our findings in this study suggest that due to an effective vaccination scheme against anthrax in cattle in Jos, Nigeria, there has been no reported of anthrax infection. We therefore allay fears of a possible outbreak of anthrax in the human population from animal sources.

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ANTIBIOTIC RESISTANCE TREND OF *PSEUDOMONAS AERUGINOSA* IN PORT HARCOURTOBUNGE, O. K.¹, ONYEJEPU, N.²

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ABSTRACT

A study was carried out in a tertiary government teaching hospital in Nigeria to assess the resistance trend of *Pseudomonas aeruginosa* over a four year period, 1997-2002 and to compare the percentage resistance of isolates in in-patients and out-patients. The resistance pattern of five hundred and eighteen (518) non duplicate *P. aeruginosa* strains against gentamycin, ampicillin, augmentin, ciprofloxacin, pefloxacin, tetracycline, ceftazidime, cefuroxime, ceftriaxone, cefotaxime were investigated using the disc diffusion method. The results indicated that the resistance rates were higher among the beta-lactam antibiotics; ampicillin (96-98%), cefotaxime (90-95%) and cefuroxime (80-84%). Over the years, the clinical isolates were significantly more sensitive to fluoroquinolones (80-96%) with higher rates of resistance observed for isolates from in-patients (14.4-25.6%) compared to the out-patients (4.2-15.6%). A decreasing resistance trend was observed for some antibiotics such as ciprofloxacin with decreasing resistance from 19% in year 1997-1998 to 4% in 1999-2000. Establishing an antibiotic resistance surveillance system and continual review of antibiotic policies is an important factor in containing the resistance trend of *P. aeruginosa*.

INTRODUCTION

The emergence of multi-drug resistant *P. aeruginosa* remains a global public health burden and a current concern (1, 2). The increasing morbidity, mortality and higher health cost associated with multi-drug resistant *Pseudomonas* infection (1, 3) necessitate this concern.

Globally, multi-drug resistant *P. aeruginosa* are increasingly implicated in both community acquired and nosocomial infections affecting predominantly patients in intensive care units (ICU's) (1-9).

National surveillance data by National Nosocomial Infections Surveillance System (NNIS) for 1992-2003 reports from U.S. hospital has reported a 32% increase in imipenem, 37% to quinolones and 22% increase in resistance to 3rd generation cephalosporins (10). Furthermore, countries in Europe, Asia and some developing countries have reported similar trends (3, 6, 11). However, in the

UK, resistance rates have been reported as stable and low (12).

Various clinical studies in Nigeria reports that *P. aeruginosa* is the major gram negative pathogen involved in both nosocomial (5, 13) and community acquired infection (7). Furthermore, Kehinde *et al* (14) reported 80% resistance of *Pseudomonas* isolated from burn wounds.

P. aeruginosa is inherently resistant to many antimicrobial agents (15, 16, 17). Thus, the pattern of antibiotic use and prevalence of resistant strains differ widely in different settings (8, 18), impacting directly on the selection of antibiotic resistance. The above fact necessitates the need to establish the antibiotic resistant pattern of *P. aeruginosa* to commonly used antibiotics in Nigeria. This is with a view to obtaining baseline information on the drug resistant trend of *P. aeruginosa* for appropriate antibiotic therapy in the face of changing resistance trend.

The aim of this study is to describe the resistance pattern of *P. aeruginosa* in the University of Port Harcourt Teaching Hospital, Port Harcourt over a four-year period from 1997 to 2000 in order to contribute in the development of antibiotic policy for the hospital.

MATERIALS AND METHODS

Five hundred and eighteen (518) non duplicate *P. aeruginosa* isolates were collected over a four year period from various clinical specimens from patients presenting at the University of Port Harcourt Teaching Hospital, in Port Harcourt metropolis, Nigeria. Repeat isolates and samples from same patients were excluded. Bacterial isolates were identified as *P. aeruginosa* using standard techniques (19). Minimum inhibitory concentration (MIC) was determined by disc diffusion method according to modified Kirby-Bauer NCCLS modified technique (20). Commercially available antibiotics tested were; Ceftriaxone (CRO), Ciprofloxacin (CIP), Pefloxacin (PEF), Gentamycin (GEN), Cefuroxime (CXM), Augmentin (AUG), Ceftazidime (CAZ), Tetracycline (TET), Ampicillin (AMP). After overnight incubation at 35°C, zone sizes were measured and interpreted using NCCLS interpretive guidelines. The reference strain *P. aeruginosa* ATCC 27853 was used. Data obtained was analyzed using epi-info soft ware, version 6.04.

RESULT

Table 1 summarizes the overall resistant pattern of *P. aeruginosa* isolated from both in-patient and out patient. *P. aeruginosa* strains resistant to most antibiotics were from in-patients. However, strains from out-patients were significantly more sensitive to gentamycin (68.5%), ciprofloxacin (95.8%).

Frequency of resistance was highest among the β -lactam antibiotics; ampicillin (97%-98%), augmentin (80%-95%), cefotaxime (83.9%-90.2%) and ceftriaxone (58.9-70.4%). Resistance level for aminoglycosides, gentamycin were 53.3% and 31.5% for isolates from in-patients and out-patients respectively.

Resistance levels for the fluoroquinolones were 4.2%-14.4% for ciprofloxacin and 5.6% -25.6% for pefloxacin showing a decreasing trend over the four years.

Resistance trend for the other antibiotics did not differ remarkably over the period (Fig 1) except for ceftriaxone with resistance decreasing from 63% in 1997-1998 to 48% in 1999-2000.

76% (79) and 78% (118) of ceftazidime and ceftriaxone resistant strains respectively were sensitive to the ciprofloxacin resistant isolates. However, only 20% (70) of the cefuroxime resistant isolates were sensitive to ciprofloxacin. Furthermore of the cefuroxime resistant isolate 68% and 62.4% were susceptible to cefotaxime and ceftazidime respectively.

Table 1: Comparison of percentage resistance of *P. aeruginosa* isolates from in-patients and out-patients to various antibiotics

% Of Resistance

Antibiotics	In-patient Isolates % (n)	Out-Patient Isolates % (n)	P value
Gentamycin	53.5 (150)	31.5(127)	<0.001
Ciprofloxacin	14.4 (90)	4.2(95)	<0.05
Cefotaxime	90.2 (41)	83.9 (31)	NS
Pefloxacin	25.6 (78)	15.6 (90)	NS
Tetracycline	97.5 (122)	89.6 (106)	<0.05
Ceftazidime	26.3 (80)	24.2 (62)	NS
Augmentin	95.0 (46)	80.0 (20)	NS
Ampicillin	98.0 (113)	97.0 (88)	NS
Ceftriaxone	70.4 (71)	58.9 (56)	NS
Cefuroxime	80.0 (46)	83.0 (49)	NS

DISCUSSION

P. aeruginosa presents a particular challenge due to its inherent resistance to many antimicrobial agents. In Nigeria, many studies have described *P. aeruginosa* as a major gram-negative pathogen associated with both nosocomial and community acquired infection (5, 7, 14).

In our study, highest resistance was observed in general among isolates from in-patients especially shown to be resistant in particular to the β -lactam antibiotics. However, resistance was statistically significant ($p < 0.05$) for isolates resistant to a group of antibiotics; gentamycin, ciprofloxacin, and tetracycline. Most studies have reported similar trend (4, 10, 12) especially in the intensive care units (21). This trend is attributable to the pattern of antibiotic usage, which in most cases is higher in this setting (8). Over the years, various extended spectrum β -lactamases has been found in *P. aeruginosa* (15, 17, 22). Carmeli *et al.* (1) and Gencer *et al.* (6) reported that Increasing resistance to the β -lactam by *P. aeruginosa* during treatment occurs more frequently in imipenem than in ceftazidime and ciprofloxacin. Furthermore, Higgins *et al.* (20), in their own study reported that imipenem resistance does not necessarily mean blanket resistance to all available drugs. Overall, our study indicated lower resistance to ceftazidime and ciprofloxacin 26.3% and 14.2% for isolates from out-patients and 24.2% and 4.2% from in-patients respectively.

Resistance among the Beta-lactam antibiotics especially to ampicillin, augmentin, cefuroxime and cefotaxime was more frequent all year round. Furthermore, compared with earlier studies in Nigeria (14) and elsewhere (6,22), there is a clear tendency towards decreasing resistance for all groups of antibiotics over the years with significant decrease ($p < 0.05$) for ciprofloxacin (19% to 4%), gentamycin (48% to 30%), ceftriaxone (63% to

48%) and tetracycline (97% to 90%). The significant decrease in resistance for gentamycin may indicate the controlled use as the drug of choice for gram negative bacteremia in the hospital. It is generally cited that anti-pseudomonal drugs available for the clinicians include the aminoglycosides, ureidopenicillin, ceftazidime, carbapenem and ciprofloxacin (19, 23). In the local setting, ceftazidime is considered as one reserve drug for multi drug resistant organism including *Pseudomonas* species. From our study, ceftazidime resistant strains tend to be sensitive to the fluoroquinolones such as ciprofloxacin (77.7%) and pefloxacin (73.0%). Again, ciprofloxacin resistance decreased from 19% in (1997-1998) to 4% in (1999-2000) suggesting that ciprofloxacin may be of therapeutic value with ceftazidime resistant strains.

In summary, resistance of *P. aeruginosa* to β -lactams remains common in our environment. The fact that the high resistance of the organism to most antibiotic remains stable over the years is of grave concern. It adds to the global concern about the rising incidence in resistance (24) especially by *P. aeruginosa* (particularly in intensive care settings) which compromises therapy. The study suggests that a significant percentage of these multi-resistant organisms are hospital acquired. Nipping this problem in the bud requires a review of the hospital antibiotic and hygiene policies and establishing a robust antimicrobial surveillance system. Also, allocation of resources to infection control and raising awareness among health workers and patients on the rising threat of multi drug resistant *P. aeruginosa* may be useful particularly in the face of re-emerging and emerging infectious diseases. Further work to type these *P. aeruginosa* resistant phenotypes is ongoing.

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PREVALENCE OF METHICILLIN-RESISTANT *STAPHYLOCOCCUS AUREUS* WITH REDUCED SUSCEPTIBILITY TO VANCOMYCIN AMONG HEALTHY WOMEN IN TWO NIGERIAN METROPOLITAN CITIES.

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ABSTRACT

Methicillin-resistant *Staphylococcus aureus* (MRSA) infections, which had been uniformly susceptible to vancomycin, are increasingly reported to develop resistance to the same antibiotic worldwide and infections caused by *S. aureus* with reduced susceptibility to vancomycin are a new clinical and public health dilemma. This study investigated its prevalence in urine of healthy women in two cities of Nigeria and its resistance pattern to other antibiotics. "First catch" urine samples collected from healthy women volunteers in Abuja and Zaria cities in Nigeria were cultured and screened for *S. aureus* using standard microbiological procedures. The isolates were then subjected to antibiotic susceptibility testing using disc diffusion technique. A total of 114 *S. aureus* isolates and 57 coagulase-negative staphylococci (CoNS) were isolated from 300 samples of urine screened. Isolates from Abuja were made up of 60 (63.2%) *S. aureus* and 35 (36.8%) CoNS, while those from Zaria women were made up of 54 (71.7%) *S. aureus* and 22 (28.9%) CoNS. Of the *S. aureus* isolated, 46(76.7%) and 37(68.5%) were methicillin-resistant in Abuja and Zaria respectively while 37 (80.4%) and 33(89.2%) of the methicillin-resistant isolates in Abuja and Zaria respectively had reduced susceptibility to vancomycin. These strains had very low resistance to ofloxacin, ciprofloxacin, sparfloxacin and gentamicin but high resistance to cephalixin, ampicillin and clindamycin. The high prevalence of *S. aureus* with resistance to both methicillin and vancomycin in the urine of healthy women in these cities emphasizes the importance of the prudent use of antibiotics and the use of infection-control precautions to prevent their transmission.

Keywords: *Staphylococcus aureus*, prevalence, methicillin, vancomycin, resistance, urine, healthy women.

INTRODUCTION

Staphylococcus aureus has been recognized as one of the major cause of infection in human beings, occurring in both community and the hospital. (1,2) *S. aureus* was uniformly sensitive to Penicillin when it was introduced in the late 1940s but resistant strains producing β -lactamase enzyme soon emerged (3,4). The introduction of the β -lactamase stable penicillins (methicillin) marked an initial victory over β -lactamase producing

penicillin-resistant *S. aureus* infections until the 1980s when methicillin-resistant *S. aureus* (MRSA) became endemic in many hospitals, (5,6). This resistance was found to be as a result of the production of altered cell wall synthesis enzymes, the penicillin-binding proteins (PBPs) that are encoded on the *mecA* gene and function by preventing the binding of penicillins (4). Glycopeptide antibiotics (vancomycin, teicoplanin) introduced into clinical practice in 1958 for the

treatment of Gram-positive bacteria, (6) were found to be effective in the treatment of MRSA infections by virtue of their unique mechanism of actions (1, 7). The emergence of Vancomycin-resistant strains of coagulase-negative staphylococci (CoNS) caused concern that such observation might presage similar developments in *S. aureus* (6, 8, 9). The threat of vancomycin resistant in *S. aureus* has been the topic of intense research and concern in the scientific world (10). Various levels of vancomycin-resistance have since been encountered in MRSA infections worldwide (10). Problems created by the increasing level of antimicrobial resistance have focused attention on measures for fighting antimicrobial resistance, foremost of which is susceptibility surveillance (11).

MRSA are often multi-drug resistant leading to failure of empirical therapy; therefore, the knowledge of the prevalence of such pathogens and their antimicrobial susceptibility pattern will be essential for infectious disease managers in their routine work.

It is generally believed that effective antibiotic therapy in developing countries is being compromised by the large reservoir of antibiotic resistant bacteria that exists within their population. The healthy members of the community represent the largest reservoir of bacteria resistant to antimicrobial agents.

This study determined the prevalence of MRSA and MRSA that are resistant to vancomycin in urine samples from healthy women in Abuja and Zaria, Nigeria.

MATERIALS AND METHODS

Sample collection

First "clean catch" urine samples were collected randomly from 150 women each from Abuja and

Zaria communities after informed consent had been obtained from each woman. Samples were collected into sterile bottles, kept in an iced-bag and transported to the laboratory.

Bacteriology

Each urine sample was immediately inoculated into Mannitol Salt agar (Lab M, International Diagnostics, UK) plates and incubated aerobically at 37°C for 18hrs. The characteristic isolates were aseptically isolated and characterized using established microbiological methods, which included colonial morphology, Gram stain characteristics, ability to produce enzymes peroxidase and coagulase (12, 13).

Antimicrobial Susceptibility Testing

The antibiotic susceptibility pattern of *Staphylococcus aureus* isolates to ampicillin 10µg (Medreich, India), cephalexin 30µg (Fidson, India), clindamycin 2µg (Pharmacia, Belgium), gentamicin 10µg (Wuham, China), ofloxacin 5µg (Pathoteq, India), ciprofloxacin 5µg (Fidson, India), pefloxacin 5µg (Fidson, India), sparfloxacin 5µg (Pathoteq, India), vancomycin 30µg (Dumex, S. Denmark), and methicillin 10µg (Oxoid, UK) were determined by the modified Kirby-Bauer diffusion technique (12). Standardized overnight culture of each isolate (counting about 10⁶ cfu/ml) was used to flood the surface of Mueller Hinton agar (MHA) plates, excess drained off and dried. The standard antibiotic discs were then aseptically placed at reasonable equidistance on the inoculated MHA plates and allowed to stand for 1hr. The plates (prepared in duplicates for each isolate) were then inoculated at 37°C for 18hrs (14).

The diameter of the zone of inhibition produced by each antibiotic disc was measured, recorded and isolates classified as "resistant" "intermediate" or "sensitive" based on the standard interpretative chart updated according to the current NCCLS standard (12, 15).

Statistical Analysis

Frequencies were obtained and percentages were calculated for study variables. Chi-square and two-tailed Fisher's exact test were used to calculate probabilities and determine significance. A p-value of less than or equal to 0.05 is considered to be statistically significant ($p \leq 0.05$).

RESULTS

Bacteriology

A total of 114 *Staphylococcus aureus* isolates and 57 coagulase-negative staphylococci (CoNS) were isolated from 300 urine samples screened. Isolates from Abuja women were made up of 60 (63.2%) *Staphylococcus aureus* and 35 (36.8%) CoNS while those from Zaria women were made up of 54 (71.1%) *Staphylococcus aureus* and 22 (28.9%) CoNS.

Antimicrobial Susceptibility Testing

The Performance Standards for Antimicrobial Disc Susceptibility Test Approved Standard was described by NCCLS(15). By this an isolate was considered methicillin resistant when diameter of

zone of inhibition produced by methicillin 10 μ g disc was ≤ 17 mm and Vancomycin resistant when the diameter of zone of inhibition produced by Vancomycin 30 μ g disc was ≤ 15 mm. Table 1 shows the relative proportion of the *Staphylococcus aureus* isolates that are methicillin-resistant (MRSA) and Vancomycin-resistant (VRSA) in Abuja and Zaria. The resistance pattern of the MRSA and VRSA isolates from Zaria and Abuja to other antimicrobial agents is shown in Figures 1 and 2. The two-tailed fisher's exact test was conducted on the relative proportion of MRSA and VRSA. It showed that the observed difference between the two cities with respect to MRSA and VRSA isolates was not statistically significant ($p > 0.05$) while the difference observed between VRSA and MRSA isolates in Abuja and in Zaria was significant ($p < 0.05$). That is, there is a significant relationship between the isolates of VRSA and MRSA in each of the two localities.

Table 1: Distribution of Staphylococci, MRSA and VRSA among healthy women in the two cities

CITIES	TOTAL (URINE)	SAMPLE	No of Staphylococci		MRSA	VRSA	MRSA/VRSA
			$n_A = 95$	$n_Z = 76$	No (%)	No (%)	No (%)
			Co NS	<i>S. aureus</i>			
			No (%)	No (%)			
ABUJA	150		35 (36.8)	60 (63.2)	46 (76.7)	41 (68.3)	37 (61.7)
ZARIA	150		22 (28.9)	54 (71.1)	37 (68.5)	36 (66.7)	33 (61.1)

Key: n_A = Number in Abuja

n_Z = Number in Zaria

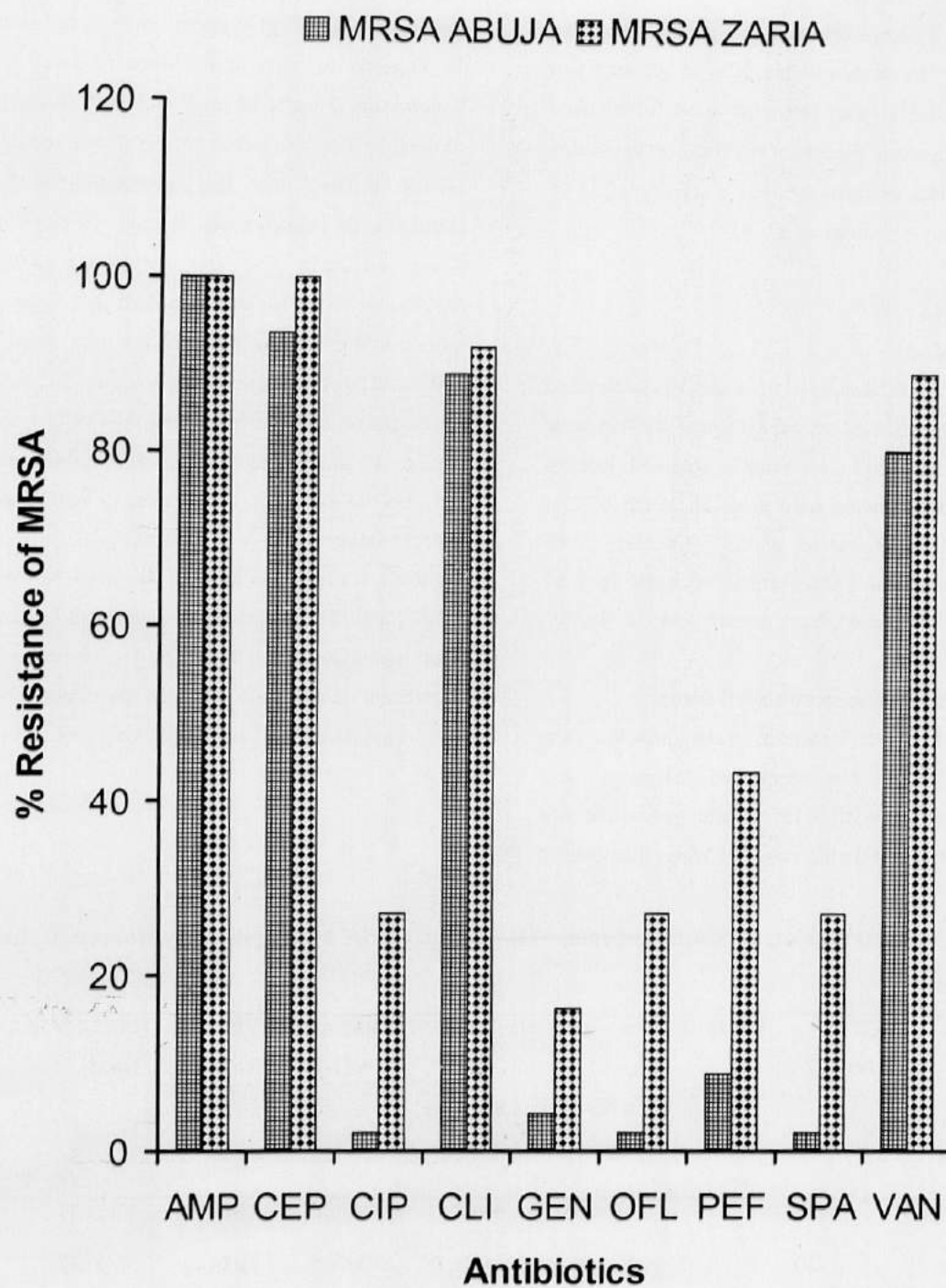


Figure 1: Resistance pattern of MRSA isolates from the two cities to other antibiotics•

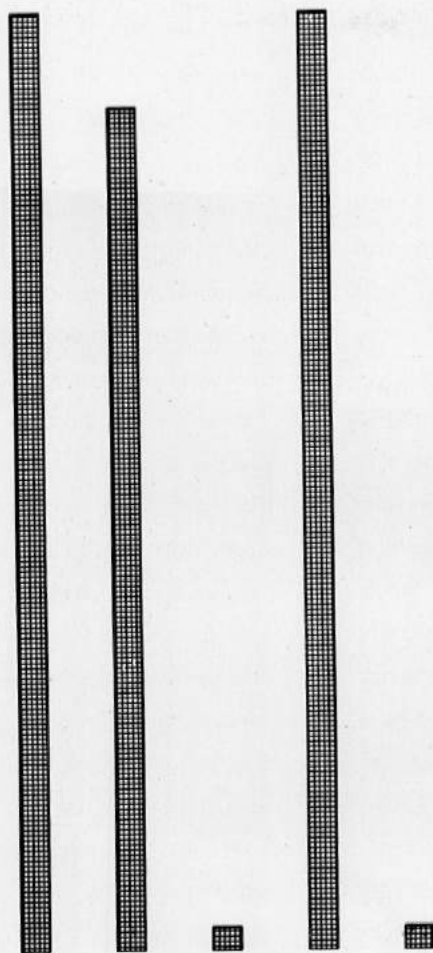


Figure 2: Resistance pattern of VRSA isolates from the two cities to other antibiotics

DISCUSSION

Infections caused by vancomycin-resistant *Staphylococcus aureus* (VRSA) are well documented and have been reported from Asia and the USA (2, 16-18). Our study reports the prevalence of MRSA isolates from healthy women that exhibited reduced susceptibility to vancomycin in Abuja and Zaria cities in Nigeria.

The result of our study showed a high prevalence rate of MRSA at 46 (76.7%) and 37 (68.5%) in Abuja and Zaria respectively, of which 37(80.4%) and 33 (89.2%) of the isolates from Abuja and Zaria respectively were resistant to vancomycin. Such a high prevalence of MRSA with high level of resistance in these environments where methicillin and vancomycin are rarely used for treatment of infections call for great concern as it is observed in the healthy (asymptomatic) women in these communities. This result is contrary to various reports of 100% susceptibility of MRSA infections to vancomycin (19,20). However our findings support the report by Olayinka *et al* (21) which had 57.7% VRSA and, the reports from Japan, France, United States, Korea and Germany of infections caused by MRSA strains with intermediate susceptibility to vancomycin (2,18,22,23).

All previous reports of patients who were infected or colonized with VRSA described a history of prolonged vancomycin exposure (24). However the asymptomatic women volunteers in our investigation lived in communities where vancomycin is rarely used for treatment of infections or diseases and they were not on any antibiotics at the point of investigation. Therefore, concurrent or recent vancomycin exposure is not a prerequisite for the development of VRSA. Instead, it can be concluded that frequent use of other commonly available antibiotics provided sufficient selective pressure to promote colonization and/or

infection with vancomycin resistant enterococci (25) and MRSA, eventually resulting in the emergence of VRSA (26).

These observed recent alarming increasing emergence of vancomycin resistance to MRSA strains might be due to acquisition of resistance determining genes like *van A, B, C* responsible for vancomycin resistance in enterococci (22,26,27) or a result of the thickening of the cell wall as reported by some authors (18,28).

The susceptibility pattern of the MRSA and VRSA isolates to other antibiotics revealed that they generally show high resistance to ampicillin, cephalexin and clindamycin but show low resistance to pefloxacin, ofloxacin, gentamicin, ciprofloxacin and sparfloxacin. This support the findings that MRSA isolates are generally resistant to other β -lactam antibiotics (29). The observed high resistance to clindamycin which is one of the antibiotics used in the treatment of MRSA infections (20) supports the report by Lu *et al* (30) which had 92.9% community-associated MRSA isolates showing resistance to clindamycin.

In this study, it was established that both the MRSA and VRSA isolates from both cities had over 70% susceptibility to ofloxacin, ciprofloxacin, sparfloxacin and gentamicin indicating that these antimicrobial agents could be used in the treatment of infections caused by MRSA and VRSA. This supports the previous reports where ciprofloxacin and gentamicin were used in the treatment of MRSA and VISA infections (10, 20).

The difference observed in the prevalence rates of MRSA and VRSA in two cities is highly significant ($p < 0.05$) which suggest that there is an association between MRSA and VRSA strains. However, the difference observed in these strains between the two cities is not significant which suggests that geographical location is not a determining factor for the prevalence of these strains in this country.

The findings in this study suggest the need for establishing standard infection control precautions that will be effective in preventing the spread of this resistant pathogen. Thus, systematic surveillance of VRSA will enhance the ability of the public health and health care systems to rapidly recognise and aggressively contain infection and colonization due to this antibiotic resistant-pathogen in the healthy population.

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COMPARISON OF SALINE WET PREPARATION, GIEMSA STAINING AND CULTURE METHODS FOR THE DETECTION OF *TRICHOMONAS VAGINALIS*.

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ABSTRACT

Objective of the study was to compare the performance of three diagnostic methods for *Trichomonas vaginalis* infection viz-Saline wet mount, Giemsa staining and Culture.

Methods

Vagina swabs from 2720 women comprising 1420 pregnant and 1300 non pregnant women were examined for *Trichomonas vaginalis* infection using the three methods above.

Results

Isolation of *Trichomonas vaginalis* was highest 246 (9.04%) using the culture method. This was followed by the Saline wet preparation 218 (6.01%) and lastly Giemsa staining method with 182 (6.7%) positive results. Using Chi square (χ^2) test at $p \leq 0.05$, it was found that there was statistical difference between the results of the three methods.

Conclusion

The culture method was the most sensitive method though costly and time consuming. It could be used to complement the wet preparation method so as to achieve a higher isolation rate in the patients with *Trichomonas vaginalis* infection in our environment and thereafter proper treatment which will also help in reducing HIV transmission.

Key words: *Trichomonas vaginalis*, Isolation, Saline wet preparation. Giemsa staining. Culture method.

INTRODUCTION

Trichomonas vaginalis infection is a sexually transmitted infection (STI) linked with reproduction health complications (1). It is the commonest curable sexually transmitted infection: the World Health Organization estimates that 170 million new infections occur each year (2). Prevalence is highest in underdeveloped countries and disadvantaged population in developed countries (3-5).

Infection with *T.vaginalis* facilitates the transmission of HIV (6) and treatment of *T.*

vaginalis infection significantly lowers the vaginal HIV viral load in dually infected subjects (7,8). Thus control of *T.vaginalis* will have a significant impact on the HIV epidemic in Africa and may reduce the incidence of adverse pregnancy outcome (9).

Major tools in the control of sexually infections in general are accurate diagnosis and treatment. If done properly, will reduce the reservoir of infection.

Diagnosis of *T.vaginalis* infection in most parts of the world is carried out by the Saline wet preparation (wet prep) method (10). However, this

technique has a low sensitivity of 30-80% (11), requires trained and experienced microscopists. With the epidemic of HIV/AIDS, it becomes imperative that more sensitive methods be used in our health care systems for the diagnosis of *T.vaginalis* infection.

Studies have been done as relate to the epidemiology, prevalence and transmission of *T.vaginalis* infection in Nigeria and beyond (12-15), but there is paucity of information on comparing the wet preparation, Giemsa Staining and Culture methods along side each

Other (16), and this study was therefore carried out for that.

MATERIALS AND METHODS

The study was a cross sectional study involving 2720 women attending clinic at Nnamdi Azikiwe University Teaching Hospital (NAUTH). Nnewi and Chimex Specialist Hospital Nnewi. The number of pregnant women was 1420 while 1300 were not.

Ethical clearance was obtained from the Research and Ethics committee of NAUTH.

gh Vaginal swab specimens were collected using 3 sterile, cotton-tipped applicators (swabs). Samples were processed immediately after collection.

The first swab was put in a tube containing 0.2ml of physiological saline solution for wet mount examination (15). The second swab stick was used to inoculate a plate containing 25mls of the Ewang's modification of Chocolate agar. The inoculated plate was incubated at 37°C for up to 4 days. A color change to yellow indicated a positive culture. Such cultures were further confirmed by the wet saline method.

The third swab was used to make a smear on a clean grease free slide. This was dried, fixed with methanol and stained with 3% Giemsa stain for 30minutes. It was then washed, dried and examined under oil immersion objective for characteristic morphological features.

RESULTS

Out of the 2720 patients screened *Trichomonas vaginalis* was isolated in 246 samples (9.04%) using the culture method, 218 (8.01%) with Saline wet mount and in 182 samples (6.7%) using the Giemsa Staining technique. Using Chi square (X^2) test at $P \leq 0.05$, there was a significant difference between the results if the three methods. This is shown in Table I

Table 1. Comparing the three diagnostic techniques for identification of *Trichomonas Vaginalis*

Diagnostic method	Number of positive samples	Number of negative samples
	Isolated	
Culture	246	2474
Wet mount	218	2502
Giemsa staining	182	2538

DISCUSSION

The study compared three techniques used in identification of *Trichomonas vaginalis* viz-Wet

mount, Giemsa staining and Culture. These methods were compared in terms of diagnostic yield, cost of performing the test and turn around time for results to be produced.

It was seen that in terms of diagnostic yield there was statistical difference (using the Chi square (χ^2) test at $P \leq 0.05$) between the results of the three methods with the culture method having the highest diagnostic yield of 218 and lastly Giemsa stain technique 182. It shows that the wet mount technique through relatively cheap and easier to perform is not a very sensitive diagnostic method, comparatively, the culture method which is slightly expensive and has about 4 days turn around time, is the most sensitive method.

The presence of sexually Transmitted Infections (STIs) increases the susceptibility of a person to HIV. It therefore becomes imperative that a more sensitive technique be used to complement the wet mount method especially in the negative samples so as to improve diagnostic yield and increase the treatment cure rate which thus will help reduce the spread of HIV /AIDS scourge in our society.

Thus the culture method is being recommended in symptomatic patients whose wet preparation examination do not show *Trichomonas Vaginalis*.

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INTESTINAL HELMINTHES INFESTATION AMONG PUPILS IN RURAL AND URBAN COMMUNITIES OF KWARA STATE, NIGERIA

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Abstract

Background: There are good theoretical reasons to think that urban and rural communities will have different risk factors for geohelminth infestation, and that the rural areas are more likely to suffer more helminthic infections than urban areas. This is because of the likely preponderance of factors that favour the continued existence of worms are mostly found in rural communities. These are low socioeconomic status of the people, and total absence of, or inadequate basic social amenities like good drainage network, pipe borne water supply and waste disposal facilities.

Objective: To compare the prevalence and intensity of intestinal helminth among pupils in rural and urban communities and to identify the risk factors associated with the infestation.

Methods: This cross-sectional study was carried out among 304 rural and urban pupils randomly selected from 2 communities in Kwara state, Nigeria. Semi-structured questionnaires were administered to the pupils and each of them had a stool sample collected and examined microscopically for ova of helminthes.

Results: The prevalence of helminthic infection in the rural and urban pupils was 17.6% and 18.5% respectively. The helminthes isolated in the stool samples were *Ascaris lumbricoides*, *Trichuri trichiuria* and hookworms. *Ascaris lumbricoides* constituted over three-quarters of the total helminthes isolated. The intensity of infestation with *Ascaris lumbricoides* and *Trichuri trichiuria* was moderate among the pupils in both rural and urban areas. However, Hookworm showed heavy intensity of infestation among pupils from the rural area as evident by high mean egg load of the parasite in the stool. Factors that were significantly associated with the risk of acquisition of the infestation included age of the pupils, educational status of the mother and type of toilet facilities used by the pupils at home.

Conclusion: The prevalence of intestinal helminth in the rural and urban pupils was similar and low compared to what was reported in other local studies carried out 5 years back. The risk factors for the infestation give the impression that all school pupils, regardless of where they stay or live in Nigeria are at risk of helminthiasis.

Recommendation: Children in both rural and urban areas must both be targeted in any anti helminth campaign since the dwelling place has not shown any reasonable risk factor among the 2 groups of pupils.

Key words: intestinal helminth, school children, mean egg load of helminthes.

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INTRODUCTION

The public health impact of helminthic infestation has consistently been under-estimated. This is based on the fact that disease states mostly manifest at a chronic stage. Diseased states have also been reported to manifest earlier, where there are factors in the host that compromise the ability of the host to fight infection.(1)

The population mostly at risk is school-aged children, who may suffer nutritional deficits, cognitive impairment, serious illnesses, and occasionally deaths rarely from complications of helminth infestation. Thus the risk of an individual suffering geohelminth related morbidity appears to be a joint function of the species harboured, the intensity of infestation and / or the virulence of the specy (1,2) The growing body of literature has identified different risk factors that perpetuates the existence of helminths in the communities, and these are individual, household, cultural and environmental factors. (3)

Generally, rural areas are expected to have higher worm load, than the urban area, because of the preponderance of those factors that perpetuate the continued existence of the worm, such as poverty, poor environmental hygiene, and complete absence of municipal services (1,4,5). Local studies have shown prevalence of helminthes in children in rural areas to be in range of 30% to 74% (4,6-8). while studies focusing on urban areas are mostly

hospital based with prevalence ranging from 15-30% (9-11)

The importance of risk factors in initiating and perpetuating the continued spread of geohelminth infestation cannot be overemphasized. Knowledge of these factors will guide and inform preventive activities at the primary care level, and in initiating governmental policies that will help in the control of helminthes infestation. A comparative study of rural and urban communities will therefore provide current epidemiological status of helminths, in terms of the prevalence, intensity, and transmission dynamics with a view to identifying needs or areas for better intervention.

METHODS

This descriptive cross-sectional survey was conducted between April and July 2003 in rural Ganmo community and urban community of Ilorin metropolis of Kwara state. The population surveyed was primary school pupils aged 6 - 12 years attending public primary schools in the study areas. From the list of public primary schools in Ilorin, simple random sampling technique was employed to select Baptist LGEA primary school in Ilorin metropolis by balloting. The only public school in Ganmo (Community LGEA) was used for the study. Systematic random sampling technique using the school registers was used to select 344 pupils (158 from the rural and 186 from the urban) for the study. Where the selected pupil was unwilling to participate in the study, the next student on the

register was chosen and the sampling interval maintained. Pupils with history of use of anti-helminthic drug in the last 6 months prior to the study were excluded.

Semi-structured questionnaire was administered to each pupil and a stool sample collected for microscopic examination. The interval between stool collection and laboratory processing was 8 – 12 hours. Saline and iodine preparations of the faecal specimens were made for microscopic examination, while formal ether sedimentation technique was used to concentrate the stool for ova counting. Stoll's method was used to determine the egg count per gram of the faeces(12)

The World health Organization (WHO) method of classification of intensity of ova of parasite in stool was used to classify the mean egg load (appendix 1) (13). The major limitation in the study was the use of formal ether sedimentation method to concentrate the ova of helminth in stool. This method is not suitable for identification of *Enterobius vermicularis* ova, hence some of the pupils with this worm might be missed which may make the prevalence of intestinal helminth to be lower than the actual.

RESULTS

In all, 304 pupils (153 rural and 151 urban) out of 344 were involved in the study giving a participatory rate of 97% and 81% for the rural and urban areas respectively. Majority of the pupils were female 166(54.6%) and predominantly in the age group 9 – 11 years in the 2 schools, Table. The

The helminthes isolated in the stool samples included *Ascaris lumbricoides*, *Trichuris trichiura* and hookworms. *Ascaris lumbricoides* constituted over three-quarters of the total helminthes isolated. Moderate infestation with *Ascaris* and *Trichuris* were mostly seen among the pupils in both areas. Only hookworm, showed heavy infestation and only among pupils from the

prevalence rate for helminthes infestation was similar among pupils in both the rural (17.6%) and the urban (18.5%) schools. The infestation cut across the whole school age groups, although pupils in age 12years and above showed slightly higher number of infestations but this was not significant. Table 2

rural area. The mean egg load for helminthes encountered was generally higher in rural than urban. The mean egg load for both communities statistical differences in number of cases of helminthes infestation among pupils in rural and urban areas. (Table 4)

Appendix 1: WHO classification of intensity for soil transmitted helminth infections in stool examination.²³

Helminth	Light intensity of infections	Moderate intensity of infections	Heavy intensity of infections		
<i>A. lumbricoides</i>	1-4999epg	5000-49999epg	≥50000epg		
<i>T. trichiura</i>	1-999epg	1000-9999epg	≥10000epg		
Hookworms	1-1999epg	2000-3999epg	≥4000epg		
<i>Epg=</i>	<i>eggs</i>	<i>per</i>	<i>gram</i>	<i>of</i>	<i>faeces</i>

TABLE 1: SOCIO DEMOGRAPHIC CHARACTERISTICS OF RESPONDENTS

Demographic Variables	Rural (N=153) n (%)	Urban (N=151) n (%)	Total (N=304) n (%)
Age groups (years)			
0-8	51 (33.3)	53 (35.1)	104 (34.2)
9-11	62 (40.5)	82 (54.3)	144 (47.4)
≥12	40 (26.1)	16 (10.6)	56 (18.4)
Mean & Standard deviation	9.2 ± 2.0yrs	9.2 ± 1.7yrs	9.2 ± 2.0yrs
Sex			
Male	65 (42.5)	73 (48.3)	138 (45.4)
Female	88 (57.5)	78 (51.7)	166 (54.6)
Mother's educational level			
primary and below	132 (86.3)	85 (56.3)	217 (71.4)
post primary	21 (13.7)	66 (43.7)	87 (28.6)
Mother's occupation			
civil servants	5 (3.3)	14 (9.3)	19 (6.3)
artisans	17 (11.1)	16 (10.6)	33 (10.9)
traders	119 (77.8)	114 (75.5)	233 (76.6)
farmers	2 (1.3)		2 (0.7)
unemployed	10 (6.5)	7 (4.6)	17 (5.6)
Father's educational level			
primary and below	126 (82.4)	98 (64.9)	224 (73.7)
post primary	27 (17.6)	53 (35.1)	80 (26.3)
Father's occupation			
civil servant	37 (24.2)	41 (27.2)	78 (25.7)
artisans	89 (58.2)	72 (47.7)	161 (53.0)
traders	11 (7.2)	24 (15.9)	35 (11.5)
farmers	16 (10.5)	14 (9.3)	30 (9.9)

Table 2a: Age specific Prevalence of infestation among the pupils

Age group (years)	RURAL COMMUNITY		URBAN COMMUNITY		TOTAL FOR 2 COMMUNITIES
	Total children in the age group n(%)	Age specific Prevalence of infestation n(%)	Total children in the age group n(%)	Age specific Prevalence of infestation n(%)	Age specific prevalence for both schools n(%)
6 – 8	51 (33.3)	62 (40.5)	53 (35.1)	11(20.8%)	17 (16.3)
9 – 11		11(17.7%)	82 (54.3)	15(18.3%)	26 (18.1%)
>12	40 (26.1)	10(25%)	16 (10.6)	2(12.5%)	12 (21.4%)
Total	153 (100%)	27(17.6%)	151(100%)	28(18.5%)	55(18.1%)

Table 2b: Relative proportion of helminthes isolated in faecal specimens

Helminthes	Rural n=153	Urban n=151	Total for 2 schools n=304
Ascaris	22 (14.4%)	22 (14.6%)	44 (14.5%)
Trichuiris	2 (1.3%)	4 (2.6%)	6 (2.0%)
Hookworm	3 (2.0%)	2 (1.3%)	5 (3.3%)
Total	27 (17.6%)	28 (18.5%)	55 (18.1%)

TABLE 3: INTENSITY OF INFESTATION AND MEAN EGG LOAD OF EACH HELMINTH

HELMINTH	INTENSITY OF INFESTATION (RURAL PUPILS)				INTENSITY OF INFESTATION (URBAN PUPILS)			
	Light n(%)	Moderate n(%)	Heavy n(%)	Total	Light n(%)	Moderate n(%)	Heavy n(%)	Total
Ascaris	5(23%)	17(77%)	0	22(100%)	8(36%)	14(64%)	0	22(100%)
Trichuris	0	2(100%)	0	2(100%)	0	4(100%)	0	4(100%)
Hookworm	0	0	3(100%)	3(100%)	1(50%)	1(50%)	0	2(100%)
TOTAL	5	19	3	27	9	19	0	28
HELMINTHES	Mean & Standard deviation (RURAL PUPILS)				Mean & Standard deviation (URBAN PUPILS)			
Ascaris	14,821 ± 11,705				9,587 ± 10,270			
Trichuris	3,900 ± 2404				4,275 ± 3,508			
Hookworm	7,100 ± 3536				1,700 ± 849			

TABLE 4: COMPARISON OF RISK FACTORS FOR HELMINTHES INFESTATION AMONG PUPILS FROM RURAL AND URBAN AREAS

INFESTED N ₁ =55				NOT INFESTED N ₂ =249		
Variable	Rural N=27 n (%)	Urban N=28 n (%)	P Value	Rural N=126 n (%)	Urban N=123 n (%)	P Value
Age group (yrs)						
6-8	6 (11.8)	11 (20.8)	0.025	45 (88.2)	42 (79.2)	0.005
9-11	11 (17.7)	15 (18.3)		51 (82.3)	67 (81.7)	
≥12	10 (25.0)	2 (12.5)		30 (75.0)	14 (87.5)	
Sex						
Male	10 (15.4)	10 (13.7)	0.919	55 (84.6)	63 (86.3)	0.232
Female	17 (19.3)	18 (23.1)		71 (80.7)	60 (76.9)	
Mother's educational level						
Primary and below	22 (16.7)	11 (12.9)	0.001	110 (83.3)	74 (87.1)	0.000
Post primary	5 (23.8)	17 (25.8)		16 (76.2)	49 (74.2)	
No of persons living in household						
1-2	0 (0.0)	0 (0.0)	0.324	9 (100.0)	1 (100.0)	0.023
3-4	3 (12.0)	7 (29.2)		22 (88.0)	17 (70.8)	
≥5	24 (20.2)	21 (16.7)		95 (79.8)	105 (83.3)	
Type of toilet						
pit latrine	11 (19.0)	10 (22.7)	0.018	47 (81.0)	34 (77.3)	0.000
water closet	3 (10.0)	12 (16.7)		27 (90.0)	60 (83.3)	
bush	13 (20.0)	6 (17.1)		52 (80.0)	29 (82.9)	
Eating in the school						
yes	20 (16.4)	26 (18.4)	0.129	102 (83.6)	115 (81.6)	0.003
no	7 (22.6)	2 (20.0)		24 (77.4)	8 (80.0)	

DISCUSSION

Intestinal helminth infestations persist and flourish wherever poverty, inadequate sanitation, improper health habits, and overcrowding are entrenched. In Nigeria, these factors are present in both rural and urban communities. Rural children may be considered to be at relatively higher risk than urban children, because these risk factors are more pronounced in rural than urban centres. The overall prevalence of helminths in this study was relatively low and similar for both rural and urban pupils. It was a surprise to see that the prevalence was about the same in children from the 2 communities. Similar findings was reported in Pemba Island, Zanzibar, Tanzania in East Africa (14).

There had been attempts to explain why urban children may be as much susceptible to helminthic infestation like their rural counterpart by Crompton *et al* (15). Their study revealed overcrowding and the technical problems in sewage disposal as probable factors. The meagre resources of the cities are often overstretched and their social services of water supply, sanitation, garbage disposal, health care are inadequate. These factors may be responsible for the insignificant differences in prevalence between the urban and rural pupils.

The relatively low prevalence observed in this study contradicts reports from previous local studies where higher prevalence ranging from 33.6% to 74.8% for rural (4,6-8) and 20 - 30% for urban were reported. (9-11).

The low prevalence does not appear to be due to good personal hygiene among the pupils, but may be related to routine use of anti-helminthic drugs. Mass de-worming campaign by Government as well as the incorporation of anti-helminthic drugs into the maternal and child health (MCH) programme by the Federal Government probably

contributed more than any factor to this low prevalence. The majority of infested cases seen probably represented re-infested cases or those who for one reason or the other had not used anti-helminthics.

The female pupils had a relatively high prevalence rate than males in both communities. (Table 4) Differences in infestation between male and female are occasionally reported, but more often than not, the prevalence of infestation is either similar in both sexes,^{16, 17} or males have an edge over the female.^{7,18} However, few local studies have reported female sex having a higher prevalence than their male counterpart^{9,11} The probable explanation for the female sex having higher prevalence in this study is that the female gender is more exposed to potential domestic sources of transmission of these helminths, like food preparation, fetching water, and disposal of waste than their male counterpart. However, the influence of gender on prevalence of helminthic infection is inconclusive as it may or may not play a role depending on the regional and environmental factors.

Of the three helminths detected in this study, *A. lumbricoides* was the most prevalent, accounting for over 80% of infestations among the pupils in the 2 communities. The other two parasites, *T. trichiura*, and hookworm were very low in prevalence. This pattern was in agreement with some studies (9,19) but at variance with others (7,20). The relatively high prevalence of *A. lumbricoides* in this study might be due to climatic and environmental conditions of the communities, such as poor water supply and poor sanitation facilities, which could be more favourable for *A. lumbricoides* than the other worms. It may also be due to the presence of other unknown risk factors. Multiple infestations were low in this study. This may be related to frequent mass campaigns against

helminth infestation via community based distribution of antihelminthic drugs in the state, in the last 3 years.

Infestation with intestinal helminth is associated with a wide range of variables, and the pattern is often determined by cultural, behavioural, household and individual factors. Factors like pupil's age, maternal education and type of toilet facility at home were found to be significantly associated with risk of acquisition of helminthic infestation. While other prevalence studies noted that the highest prevalence of intestinal helminthes occur in children aged 5-12 years (4,19). This study found that age specific prevalence rate was greater for age 12 years and above, although this is not statistically significant. This is particularly obvious among rural pupils. Other age groups showed uniform rate of infection as reported in other literatures (3,4). The reason why this age group is at the greatest risk of acquiring helminth might be due to frequency of host-parasite contact as children in that age group would be expected to be able to assist their parents in domestic work like cooking and fetching water.

Lack of or low formal educational status of mothers has consistently been associated with likelihood of worm infestation in children. For example, two separate studies in Malawi and Panama, reported that mothers with little or no education had significantly higher number of children infected with helminthes than mothers who are of high educational status and lived in the same area or community.²¹ Our finding is contrary as kids of urban mothers with at least post primary education had higher prevalence of worm infestation than kids of urban mothers with primary or no formal education. (Table 4) This contrasting finding may be due to the increasing number of women in the workforce, leaving their children no choice but to patronize food vendors.²²

The use of pit latrine strictly speaking is associated with a lower risk of acquisition of intestinal helminth compared to the use of open bush, as it reduces contact between persons and the infective larvae. But where the provision of pit latrine is not accompanied with adequate supply of water, the chances of faecal contamination becomes higher (23). Furthermore, the unacceptably high number of persons per toilet (over-crowding), improper usage and poor quality hygiene of the toilet have been shown to influence acquisition of intestinal helminth. (20,23) Against the background of polygamy (large family greater than five persons, living in single room of shared apartment), and poor water supply within the communities studied, our findings among those using pit latrine is not surprising. It may thus be summarized, that while the use of pit latrine protect against intestinal helminth, it must be provided alongside with adequate water supply to ensure personal cleanliness and the cleanliness of the latrine.(24). One may state that where water supply is in short supply, the construction of ventilated improved pit latrine could serve as an alternative.

CONCLUSION:

The prevalence of intestinal helminth in rural and urban communities studied were similar and within the national average of 14 -30%. The determinants of infestation in both rural and urban area include age of child, level of maternal education and type of toilet facilities at home.

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