

Incidence of Streptococcus Pneumoniae Infections Among Patients Attending

Tuberculosis Clinics in Ekpoma, Nigeria.

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Abstract: Pulmonary pneumonia and tuberculosis are both lower respiratory tract infections that affect almost all class of people mostly in developing countries. Few current and up to date data are available on the trend of the above disease conditions in Ekpoma and its environs. To assess the incidence of Streptococcus pneumoniae infection among patients attending tuberculosis clinics in Ekpoma Nigeria. Alpha hemolytic organisms on chocolate and Blood agar from 330 sputum samples randomly selected from patients attending tuberculosis clinics in Ekpoma, suspected to be Streptococcus pneumoniae were identified using bile solubility and optochin sensitivity test while Mycobacterium tuberculosis were identified by the Ziel Nelson technique for acid and alcohol fast bacilli.

Out of the 330 samples examined, 21 (6.4%) and 81 (84.6%) of Streptococcus pneumoniae and Mycobacterium tuberculosis respectively were detected. Out of 144/186 male/female examined, 15 (10.4%), 6 (3.2%), and 48 (33.3%), 33 (17.7%) of Streptococcus pneumoniae and Mycobacterium tuberculosis from male/ female samples respectively. Age group 1-9 years was mostly affected by Streptococcus pneumoniae, age group 50 -59 years which had high incidence of Streptococcus pneumoniae and Mycobacterium tuberculosis respectively. The occupational group identified as 'others' which include mostly children, had the highest incidence of both Streptococcus pneumoniae and Mycobacterium tuberculosis followed by farmers. The relevance of accurate and current data on the infection trend of Streptococcus pneumoniae and Mycobacterium tuberculosis respectively.

Key Words: Incidence, Streptococcus pneumoniae, and Mycobacterium tuberculosis.

Introduction:

Despite the availability of effective chemotherapy pneumonia and tuberculosis are still major health problem in most countries of the world. There are many different causes of pneumonia but the most common are produced by Streptococcus pneumoniae, Klebsiella pneumoniae, Mycoplasma pneumoniae, Legionella pneumophila, Staphylococcus aureus, Haemophilus influenzae, and in rare cases Neisseira meningitides. Streptococcus pneumoniae accounts for over 60% of bacterial pneumonia in adults who require hospitalization.

A high incidence of pneumococcal bacteremia usually occur in infants under 2 years of age, low in teenage and young adults and high in people above 60 years when immunity is low due to age. Certain occupational groups including military recruits usually have the highest annual rate of infection except in recent report of South African gold miners which was higher ³ in normal individual with intact mucociliary clearance there is much lower risk of pneumococcal disease once colonization is established compared to those with preexisting pulmonary disease or immunosuppression. Those who are at risk of pneumococcal infection include with: tuberculosis, people viral respiratory infection especially due to influenza virus, malnutrition, liver and kidney disease, cancer and diabetes mellitus and persons with underlying conditions medical such as HIV infections sickle cell disease and others⁴. Streptococcus pneumoniae is a major cause of bacterial pneumoniae and meningitis in some developed countries even the United States. Outbreaks of S pneumoniae (antibiotic resistance and non resistant) have been reported from child care centers, nursing homes, hospitals, military camps, homeless institutions⁵. shelters, and panel Tuberculosis of man and of animals is caused by Mycobacterium tuberculosis and every year around two million people die of this disease despite the availability of inexpensive treatment that are effective in up to 95% of cases(a) Surveillance data on S. pneumoniae infections are few if available for healthcare providers in Ekpoma and its environs.

Accurate and up to date data is vital for successful management of lower respiratory tract infection due to tuberculosis and pulmonary pneumonia.

This study was therefore designed to determine possible incidence of S. pneumoniae among patients attending tuberculosis clinics in Ekpoma with hope of providing promising information which could help in its prevention and control. This investigation was also expected to reveal the current age and sex specific prevalence rate of M. tuberculosis among patients clinically diagnosed with pulmonary tuberculosis in Ekpoma.

Materials and Methods:

Ekpoma and its environs in Esan West Local Government Area of Edo State were the main study areas, while University Health Center and General Hospital all in Ekpoma served as the major sites for sample collection. Early morning randomly selected sputum samples were collected into a clean sterile dry wide-necked leak-proof universal container from patients.

patients who previously tested All positive for tuberculin or Mantoux test and required to undergo the routine Acid Bacilli and Alcohol fast test for pulmonary tuberculosis test were the group included in our random sampling. Direct examination of sputum samples the Grams staining by technique was adopted in selecting ideal samples for culture^{6, 7}. Three hundred and thirty sputum samples with at least 15-25 white blood cells and less than 10 epithelial cells in a microscopic field were finally selected at random as ideal for the $survey^7$. Sample collection in children was by nasal aspiration of sputum from the lungs.

All samples collected for processing were inoculated into blood and chocolate agar and smear of the purulent part of the specimens were made on a glass slide for Grams staining before transportation to Search-Light Medical Diagnostic Center Ekpoma for further processing. The Ziehl Neelsen technique of staining for M. tuberculosis was adopted in processing samples for acid and alcohol fast bacilli. Sheep blood which does not contain any streptococcal inhibitor was used for blood and chocolate agar preparation8 and 5 μ g/ml of gentamicin was added as selective inhibitor and duplicate non-selective was used as control⁹.

Sputum specimens for culture were homogenized gently with 2 ml sterile normal saline by refluxing the sputum saline mixture in a small syringe without а needle attached. Homogenized samples were streak-stabbed onto freshly prepared media the for of determination streptococcal hemolysis. Streak-stabbed plates were incubated in a candle extinction jar. Control strain of Streptococcus pneumoniae supplied by the National Veterinary Research Institute, Vom, Plateau State, Nigeria was added to all sets of culture. Alpha hemolytic isolates cultural with other characteristics consistent with S. pneumoniae were picked and identified using standard technique including bile solubility and optochin sensitivity test^{10, 11}.

The chi-square test was used in the statistical analysis of results obtained. The population compared includes: S pneumoniae prevalence among age groups 1-9 years and 20-29 years; and 1-9 years and 70-79 years. The prevalence rate of S. pneumoniae and

M. tuberculosis among males and females and also among the age group1-9 were compared.

(24.6%) respectively. The prevalence of S. pneumoniae among male/female patients were 15 (10.4%) and 6 (3.2%), while M. tuberculosis prevalence among male/female patients were 48 (33.3%) and 33 (17.7%) (Table 2).

Results:

S. pneumoniae and M. tuberculosis showed prevalence of 21 (6.4%) and 81

able1: Distribution of S	. pneumoniae and M.	tuberculosis in sputum	samples from 330	patients.
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Bacteria	NO (%) Positive
S. pneumoniae	21 (6.4)
M. tuberculosis	81 (24.6)

Table 2: Sex-specific prevalence of S. pneumoniae and M. tuberculosis in sputum specimens.

Sex	Total	Number (%) of sample positive for	
		S. pneumoniae	M.tuberculosis
Male	144	15(10.4)	48(33.3)
Female	186	6(3.2)	33(17.7)

The observed percentage prevalence of S. pneumoniae among the age groups1-9, 20-29 and 50-59 years 100%, 9.1% were and 50% respectively. Other age groups that falls between 10 and 79 years including other adults whose ages were not specified all had 0.0% prevalence. While the prevalence of M. tuberculosis among age groups 1-9 and 10-19 years were 0.0%, its prevalence among age groups 20-29, 30-39, 40-49, 50-59, 60-69, 70-79, and other adults whose ages were not specified , were 18.2%, 16.7%, 50.0%, 33.3%, 25.0%, 20.0%, 27.8% respectively. There was a significant statistical difference in S. pneumoniae

and M. tuberculosis prevalence among the age group 1-9 years and there was no significant statistical difference in S. pneumoniae and Μ. tuberculosis prevalence among male and females subjects. There was a significant statistical difference in S. pneumoniae prevalence between the age groups 1-9 years and 20-29 years, and also between 1-9 years and 70-79 years respectively. Farmers and the group identified as 'others' which included mostly the children and people who had no job had higher incidence of S. pneumoniae pneumonia compared to other occupation, while farmers and

pulmonary tuberculosis (Table 4).

Occupation	Number sampled	Number/ (%) positive samples for	
		S. pneumoniae	M. tuberculosis
Farmers	160	13 (8.1)	41 (25.6)
Students	43	0 (0.0)	8 (18.6)
Traders	91	0 (0.0)	22 (24.2)
Others	36	8 (22.2)	10 (27.8)

Table 4. Distribution of infectious agents according to occupation.

Discussion

From our result in (Table 1), the of frequency occurrence of Streptococcus pneumoniae was (6.4%). This falls within the range (5 - 75%)reported by Murray et al., ⁵, that its rate of occurrence is significantly affected by the method used in detection of the organism and the population studied. This result is similar to (8.5%) reported from a recent study carried out in Gambia and slightly different from (21%) from older United States studies ¹². This might be due to difference in geographical location of the areas where the researches were carried out and the fact that facilities for antimicrobial agents testing used may not be as advanced as those being used in developed countries.

When (6.4%) prevalence obtained from our investigation carried out in the dry season is compared with (10%) obtained within 1 year in Malaysia ¹³, it seams therefore that a higher/similar prevalence may be obtained if the number of months for the research is extended. According to Katz and Morrens, ¹⁴, incidence of carriage and associated disease is highest during the cold months and might also have contributed to low prevalence obtained from this investigation, since the period of this research was more of dry season than wet season (February to May). We observed that males had higher incidence of both S. pneumoniae and M. tuberculosis (10.4% and 33.3%), than females, but the difference was not statistically significant at (P < 0.05), (Table 2). The predisposing factors for both organisms seem to favor the males more than the females especially in most developing countries in Africa where overpopulation in military camps, prison, construction sites, and factories are dominated by males.

While this finding agrees with recent reports19 (53% / 47%) male / females ratio, it is different from (39% / 61%) reported by Ronald et al. ²⁰ in which male had lower incidence than females. Other factors like HIV infections, sickle cell disease and degree of exposure to persons with active infection makes it appear difficult to determine the trend of both pneumococcal and tuberculosis infection, both in Ekpoma and other developing countries.

The observed 100% rate of occurrence of S. pneumoniae infection showed by age group 1-9 seems to be pointing towards an entirely different trend of S. pneumoniae infection; compared to Fauci et al ²¹ report that rate of occurrence is not very high children. High susceptibility of age groups 1-9 years to pneumococcal infection may be due to inadequate amount of opsonins usually common among children who may also be deficient in naturally occurring antibodies and serum complement. There might also be inherent immunoglobulinin disorder especially hypogamaglobulinaemia, and multiple myeloma. Another factor that may contribute to high susceptibility of

this group to pneumococcus infection is pulmonary disease where mechanisms for clearance of pneumococci from the lungs are disturbed.

Table 3 shows (0.0%) prevalence of M. tuberculosis among the age group 1-9 years and 10 - 19 years. It could be that the children of this age group were given BCG vaccine at birth which might have protected them until other factors that can wear down immunity sets in. This also may mean that these groups might not have been exposed to tuberculosis There infection. was а significant statistical difference in S. pneumoniae prevalence at (P < 0.05) between the age groups 1-9 and 20 - 29 years. The difference may be illustrated when percentage prevalence is analyzed with respect to the number of samples examined (Table 3)

Age (Years)	Number of	Number (%) of sample positive for:	
	samples	S. pneumoniae	M.tuberculosis
1 - 9	6	6 (100)	0 (0.0)
10 – 19	9	0 (0.0)	0 (0.0)
20 – 29	66	6 (9.1)	12 (18.2)
30 – 39	54	0 (0.0)	9(16.7)
40 – 49	30	0 (0.0)	15 (50.0)
50 – 59	18	9 (50)	6 (33.3)
60 – 69	24	0 (0.0)	6 (25.0)
70 – 79	15	0 (0.0)	3 (20.0)
Others	108	0 (0.0)	30 (27.8)

Table 3. Prevalence of S. pneumoniae and M. tuberculosis in relation to age of patients.

The decrease in prevalence of S. pneumoniae from 100% of age group 1-9 years, to 9.1% of age group 20 – 29 may be due to the latter age group possessing adequate amount of: opsonins, naturally occurring antibodies, and serum complement. This might also mean absence of underlying infection like HIV, and inherent immunoglobulin disorders especially hypogamaglobulinaemia4. This decrease in prevalence is in line with the report of Fauci et al., ²¹.

Age groups 1-9 and 70-79 showed significant statistical difference in S. pneumoniae prevalence at (P < 0.05). Zero (0.0%) percent prevalence recorded among this age group does not agree with recent reports 4. However, incidence among age group 70-79 years could be zero from this survey probably due to improved HIV therapy (if HIV positive); expanded 23 use of the polyvalent polysaccharide vaccine and good management of other underlying disease in adults like sickle cell malnutrition disease, glucocoticoid, and defective antibody formation⁴.

When the prevalence rates of S. pneumoniae and M. tuberculosis among the age group 1-9, were compared, it was seen that S. pneumoniae showed higher prevalence.

There was also a significant statistical difference at (P < 0.05), in prevalence rates of S. pneumoniae and M. tuberculosis among the age group 1-9. This agrees with what other researchers reported elsewhere $^{5, 4}$.

This decreasing trend of prevalence among the age groups above 50, as observed in this survey did not agree with reports made by CDCP⁴.

Finally, in (table 4) the study showed that farmers and 'others' had the highest incidences of S. pneumonia infection with 8.1% and 22. 2%rates of occurrence respectively. Ekpoma is a small university town in Edo State, of Nigeria with about 60% natives as incidence of farmers. High both pneumonia and tuberculosis among farmers could becomparable to similar higher incidence reported among South African gold manners ³. Children were included in the occupational identified as 'others' and group children are known to be highly vulnerable to pneumonia infection due to low immune response. This could be why this group had slightly high result of both pulmonary pneumonia and tuberculosis.

Our observation in this study shows that S. pneumoniae is significantly present in patients attending tuberculosis clinics in Ekpoma and the prevalence was higher in males than female patients both S. for pneumoniae and M. tuberculosis. S. pneumoniae tends to be highly prevalent in children and low in adults above 60 years, while M. tuberculosis prevalence was observed to be low in children, slightly raised at age group 40-49 years and decreases slowly at age group 50 years and above within Ekpoma community and its environs.

References:

1. Nester EW, Roberts CE, Personal NN, Anderson DAI and Nesterr ML, Microbiology. A Human Perspective. 2nd E.D, W.C.B - Mcgraw –Hill, New York; 1998 525 –527.

2, Musher, DM, Watson DA and Baughn RE. Does naturally acquired IgG to cell wall polysaccharides protect human subjects against streptococcal infection? J Infect Dis, 1990; 161, 736-740.

3., Far BM, Mandell GL. Gramm-positrive pneumonia. Respiratory infections, Diagnosis and management. 3rd edn,ed. Pennington JE. Raven press, NY, 1994; 340-67.

4. Centers for Disease Control and Prevention. National Center for

Infections Disease Division of Bacteria And Mycotic Diseases – Streptococcus pneumoniae disease. Technical information; 2002, 2.

5. Musher D, Groover J, Reichler M, Riedo F, Schwartz B and Watson D. Emergence of antibody to capsular polysaccharides of Streptococcus pneumoniae during outbreak of pneumonia: association with nasopharyngeal colonization. Clin infect Dis; 1997; 24, 44-6.

6. Lentino JR. The non-value of unscreened sputum in the diagnosis of pneumonia. Clin Microbiol Newsletter. 1987; 9, 70.

7. Wetteran LM, Zeimis RT and Hollick GE. Direct examination of Unstained smear for the evaluation of sputum specimens. J Clin Microbiol. 1986; 24: 143.

8. LeBien TW and Browel MC. Determination of haemolysis by Streptococcus spp. Can J Microbiol. 1975; 21:101.

9. Raphael SS. Lynch's Medical Laboratory Technology. 4th Ed. WB Saunders Company London. 1983; 423-429. 10. Barrow GI and Feltham RKA. Cowan and Steel's Manual for the identification of medical bacteria. 3rd ed. Cambridge University Press, 1993; 60-66

11. Hadie JM. Genus Streptococcus. Rosenbach 1984, Bergey's manual of systemic Bacteriology eds Snealth PHS, Maris NS, Sharpe ME, Williams and Wilkins, Baltimore. 1986; 1043-47.

12. Lloyd–Evans NO, Dempsey TJ, Baldeth I, Socka O, Demba E and

Todd JE. Nasopharyngeal carriage of pneumococci in Gambian children and their families. Pediatr infect Dis J. 1996; 15:866-71

13. Malik AS, Ismail A, Pennie RA and Naidu JV. Susceptibility Pattern among Pre-school children in Kota Bharu Malaysia. Journal of Troipical Pediatrics.1998; 44, 1: 10 – 4.

14. Katz AR and Morens DM. Severe Streptococcal infections in historical perspective. Clin infect. Dis. 1992; 14: 298 – 307.

15. Texas Department of Health, Epidemiology Annual Report – Antibiotic Resistant Pathogen Isolate Surveillance. 1999; 1-2

16. Ronald LM, Renda P, John RH, Anthony L and James MC. High Prevalence of Penicillin Non Susceptible Streptococcus pneumoniae at a Community Hospital in Oklahoma. 2000; 1-3 http://www.cdc.gov/ncidod/eid/vol 6 no 3/Moolenacar.htm.

17. Fauci AS, Braunwald E, Issibacher KJ, Wilson JD, Mantin JB, Kasper DL, Hauser SL and Longo DL. Harrison's Principle of Internal Medicine. Melaraw – Hill Companies, Inc. New York USA. ed. 1998; 14.

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