https://doi.org/10.48047/AFJBS.6.15.2024.7065-7078

African Journal of Biological Sciences

Journal homepage: http://www.afjbs.com

Research Paper Open Access

Co-Infection of Mycobacterium Tuberculosis and Non-Tubercular Bacteria among Tuberculosis Patients attending Mile Four Hospital, Abakaliki, Ebonyi State Nigeria.

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Volume 6, Issue 15, Sep 2024

Received: 15 July 2024

Accepted: 25 Aug 2024

Published: 05 Sep 2024

doi: 10.48047/AFJBS.6.15.2024.7065-7078

ABSTRACT

Background and Objective: This study aimed at determining the prevalence of *Mycobacterium tuberculosis* and non-tubercular bacteria co-infection among tuberculosis patients attending Mile four hospital, Abakaliki, Ebonyi State, Nigeria. The study was carried out in Microbiology laboratory of Alex Ekwueme Federal University Ndufu-Alike Ikwo, Ebonyi State. Methods: The samples were analyzed macroscopically for the presence of pus and blood and were cultured on chocolate agar using the streaking technique. Cultures were incubated for 24hours at 37. Bacterial growth after incubation were identified based on colonial characteristics, Acid Fast staining and Gram staining techniques and other biochemical tests. Results: The study revealed that the prevalence of co-infection between M tuberculosis and other bacteria pathogens is 4%. Out of the eight bacterial isolated from the different samples evaluated, Escherichia coli had the highest frequency and percentage occurrence of 5(25%), followed by Staphylococcus pyogens 3(15%), *Haemophilus influenzae* 2(15%), *proteus* 1(15%), *Streptococcus pneumoniae* 3(10%), *Pseudomonas spp* 2(10%), while *Klebsiella spp* has the lowest number of occurrence with frequency and percentage of 1(5%) and *Staphylococcus aureus* also had the lowest number of occurrence with frequency and percentage of 1(5%). It was observed that *Mycobacterium tuberculosis* and *Escherichia coli* co-infection had the highest occurrence from the study. This present study has revealed the prevalence of co-infection among *M. tuberculosis* and other bacteria pathogens among TB patients. The result of the sensitivity shows that oflaxacin, pelfloxacin and cephalosporin have an inhibition zone diameter of 18mm respectively why penicillin, pelfloxacin, septrin have the highest zone of inhibition diameter of 20mm. Conclusion: However, findings from this study will assist the concern health authority on the needs to intensity preventive and control measure against the treatment and management of tuberculosis.

Keywords: *Mycobacterium, tuberculosis*, preventive, control measure, pathogens

INTRODUCTION

Mycobacterium tuberculosis is the causative agent of tuberculosis, an air borne disease causing significant worldwide morbidity and mortality which was discovered by Robert Koch in 1882[1].The genus "*Mycobacterium*" comprises a number of aerobic bacteria and is the only member of the family Mycobacteriaceae closely related to the genera Norcadia" and "Corynebacterium" within the order Actinomycetales[2]. Mycobacteria are obligately aerobic non-motile rod-shaped bacilli non-spore forming.catalase-negative and facultative intracellular bacteria. Members of the genus Mycobacterium have several unique characteristics as compared to other genera of bacteria, largely due to structural differences in cell wall composition[3].Tuberculosis develops in only 10% of humans exposed to M. tuberculosis and generally develops within 1-2years of M[4]. tuberculosis infection in 5% of those infected[3]. There are two (2) stages of tuberculosis infection, the latent tuberculosis infection (LTBI) and the active tuberculosis infection (ATBI) with the active TB having a greater burden of TB bacilli than latent TB and acts as an infection source for contacts[5]. According to the 2018 World Health Organization (WHO) global tuberculosis (TB) report, about 10 million new cases of TB were recorded worldwide in 2017[6]. Nigeria ranked first in Africa with an estimated prevalence of 616 cases per 100,000[7].

Co-infection of tuberculosis with Human Immunodeficiency Virus (HIV) and several bacteria has been widely reported, and tubercular-bacterial co-infection needs to be considered especially if TB occurs in a typical pulmonary or extra pulmonary location[8]. Some of this tubercular-bacterial co-infection includes TB-streptococcus pneumonia co- infection, TB-klebsiella pneumonia co-infection, TB-haemophilus influenza co-infection, TB-staphylococcus aureus co-infection, TB-Norcadia asteroid co-infection, TB-Mycobacterium leprae co-infection and TB-Salmonella co-infection[9]. Also, some cases of tubercular-viruses co-infection have also been recorded. These include tuberculosis (TB)-HIV co-infection, TB-Influenza Virus co-infection etc[10]. TB bacteria most commonly grows in the lungs and can cause symptoms such as a bad cough that lasts 3 weeks or longer, pain in the chest, coughing up blood or sputum (mucus from deep inside the lungs), weakness weight loss, chills, sweating at nights etc. Underlying infections with HIV, diabetes mellitus, leukemia, head or neck cancer, and extracts with infected sputum samples or bronchial aspirates are some risk factors to contracting TB[11]. TB can be diagnosed by series of laboratory and diagnosed test which includes Acid Fast Bacilli (AFB) or Ziehl-Neelsen staining techniques, Mycobacterium culture using Lowenstein Jensen (LJ) media and other media that can support the growth of Mycobacteria, the use of serological /immunological techniques such as TB-Interferon Gamma Release Assays (IGRAs), Mantoux tuberculin skin test and the use of molecular techniques such as Genexpert and True Nat tests[12]. Individuals diagnosed with a pulmonary form of tuberculosis, not exposed to anti-tuberculosis drugs (i.e new cases of tuberculosis), have to be treated for 6 months[13]. During the 2 months intensive phase, patients should be administered a combined regimen including ethambutol, isoniazid, pyrazinamide and rifampicin are prescribed during the 4 months continuation phase[14]. Although superadded bacterial infection can occur in TB patients, the simultaneous occurrence of both

infections leads to delayed diagnosis and inadequate treatment[15]. Tuberculosis-bacterial co-infection needs to be considered, especially if TB occurs in a typical pulmonary and extra pulmonary location. This research is aimed at ascertaining the microbiological assessment of co-infection of bacterial pathogen and Mycobacteria tuberculosis from patients attending Mile Four hospital Abakiliki, Ebonyi State Nigeria, and to isolate and identify bacteria species and other microorganisms causing disease co-infection in tuberculosis patients, to conduct an antimicrobial susceptibility test in order to ascertain if an organism is resistant or susceptible to a particular TB drug and to identify multi-drug resistant M[16]. tuberculosis (MDR-TB) and other bacteria which complicates clinical presentation leading to inadequate treatment and unsatisfactory outcomes.

MATERIALS AND METHODS

Study Area

This study was carried out in mile four hospital located in Abakaliki, Ebonyi State, Nigeria.

Ethical Consideration

Ethical consideration was sought for and obtained from the chief medical staff and the head of the medical unit of mile four hospital. Prior to sample collection, participants were informed clearly about the objectives and procedures of the study. Potential participants were told that participation was totally voluntary and any information obtained would remain confidential.

Inclusion and Exclusion criteria

The inclusion criteria for the study included those who are confirmed TB patients while the excluded criteria excluded those who does not have TB infection

Sample Collection and Processing**:** Samples were collected at the medical laboratory unit, transported in an ice pack to microbiology laboratory of Alex Ekwueme Federal University Ndufu Alike Ikwo, Ebonyi state, for analysis which was processed within twenty-four hours of collection.

Quality Control

A standard bacteriological procedure was followed to keep quality of all laboratory tests. The quality control was performed in every required step. The blood samples were collected using sterile leak proof containers aseptically to avoid contamination. The sterility of each batch of the test medium was confirmed by incubating an un-inoculated plate and tube overnight at 37℃ and was not used if these plates and tubes showed any evidence of bacterial growth and other visual reactions after incubation. The various samples were examined macroscopically with the naked eyes. The colour, odour, turbidity/cloudiness were noted and recorded appropriately.

Isolation, Characterization and Identification of Microbial Isolates

A wire loop was sterilized by placing it on an already lit spirit lamp to red hot and allowed to cool before using to pick a loopful of the sample and then streaked on the prepared media plate. This procedure was repeated until the last sample was inoculated and they were inoculated on

both PDA media and nutrient broth agar. The contents were allowed to set before incubation for 37c for 24 hours.

After 24hours, the growth was on the plates were subculture in a fresh media plate and incubated for another 24hours to obtain a pure culture.

Acid Fast Bacilli Staining: The Acid fast bacilli staining are to differentiate bacteria into acid fast group and non-acid fast groups. This method is used for those microorganisms which are not stained by simple or Gram staining method, particularly the member of genus Mycobacterium, are resistant and can only be visualized by acid-fast staining

The bacterial sample was smeared on clean and grease free slide using sterile technique, and allowed to air dry and then heat fix. The smear was flooded with carbol fuchsin stain and heated until the stain began to rise. The heated stain was allowed to remain on the slide for 5minutes and stained was washed off with clean water. The smear was flooded with 3% acid alcohol for 5 minutes until the smear is sufficiently decolorized and then washed with clean water. The smear was stained with malachite green stain for 2 mins and washed off with clean water. The slide was cleaned and placed on a staining rack for the smear to air dry and examined under oil immersion (X 100) and result recorded.

Gram Staining

The structure of bacteria cell wall determines whether a bacterium is gram-positive or gram-negative.Gram-positive cell wall contains thick layer of peptidoglycan which retain the crystal violet-iodine complex and resistant decolourization. In contrast, gram-negative cell wall contains thin layer of peptidolglycan which are easily decolorized. After decolorization the gram-positive cell wall remains purple in colour whereas gram-negative bacteria loses the purple colour and will retain the pink or red color of the counter stain (safranin). These characteristics changes permit microbiologist to categorized bacteria into two broad groups,gram positive and gram-negative. A sterile pipette was used to pick a drop of distilled water and placed on a grease free microscope slide. The loop was sterilized and then used to pick a bacteria colony from a 24hour culture and placed on the water on the slide and emulsified to form a smear. The smear was allowed to air-dry and then heat fixed by passing the slide over a Bunsen flame. Crystal violet (primary stain) was poured over the fixed smear and left to stand for one minute then rinsed gently with running water. The iodine (mordant) was poured on the smear and allowed for one minute before rinsing with gentle stream of running water. The smear was decolorized with acetone alcohol for few seconds and rinsed with water,and safranin (counter stain) was applied for 30 seconds, and then washed blotted and examined under oil immersion (X 100) and result recorded.

Catalase Test: Some bacteria produce an enzyme catalase to neutralize toxic forms of oxygen metabolites like hydrogen peroxide (H2O2). It breaks down hydrogen peroxide into water and oxygen. About 2drops of 3% hydrogen peroxide was placed on a sterile slide. A sterile wooden applicator stick was used to pick a discrete colony from a 24hours culture and smear in hydrogen peroxide solution. The solution was observed for the presence of bubbles as an indication for positive results.

Oxidase Test

The oxidase reagent was prepared by dissolving 0.1g of tetra methyl-p-phenyloncdiaminedihydrochloride in 10ml of distilled water and used immediately. A Whattman filter paper was impregnated with oxidase reagent using a sterile pasture pipette to place a drop each of several spots on the filter paper. A sterile wooden applicator stick was used to pick a colony from a 24hours culture on a nutrient agar plate and smeared on the impregnated filter paper and the result was recorded within 5-10seconds.

Indole Test

Approximately, 15g of peptone water was weighed and dissolved in 1000mI of distilled water and heated over a Bunsen flame to dissolve completely. A 6ml volume of the broth was dispensed into test tubes and sealed immediately with aluminum foil and sterilized at 121℃ for 15minutes in an autoclave. After sterilization, the broth was allowed to cool in the working bench. A sterilized applicator stick was used to pick the test organisms from a 24hour culture and inoculated in the peptone water. The tubes were again sealed with aluminum foil and incubated at 37°C for 24-48hours. After the incubation, a sterile syringe was used to add 0.5m of kovac's reagents into the tubes and shake gently, the tubes were observed for a red surface later within 10 minutes which indicated a positive result.

Vogues Proskauer Test

A 6ml volume of the broth was dispensed into test tubes and sealed immediately with aluminum foil tube and sterilized at 121C for 15minutes in an autoclave. After sterilization, the broth was allowed to cool. A sterilized wire loop was used to pick the test organism from a 24 hours culture and inoculated in the broth. The tubes were again sealed with aluminum foil and incubated at 37°°C for 48hours. After the incubation, a sterile syringe was used to add1ml of 40%KOH followed by a 3ml of 5% solution of a-naphthol, the tubes were observed for a red surface layer within 2-5minutes and the result recorded. used to pick a colony from a 24hours culture on a nutrient agar plate, and smeared on the impregnated filter paper and the result was recorded within 5-10seconds.

Indole Test

Approximately, 15g of peptone water was weighed and dissolved in1000ml of distilled water and heated over a Bunsen flame to dissolve completely. A 6ml volume of the broth was dispensed into test tubes and sealed immediately with aluminum foil and sterilized at 121C for 15minutes in an autoclave. After sterilization, the broth was allowed to cool in the working bench. A sterilized applicator stick was used to pick the test organisms from a 24hour culture and inoculated in the peptone water. The tubes were again sealed with aluminum foil and incubated at 37°°C for 24-48hours. After the incubation, a sterile syringe was used to add 0.5m of kovac's reagents into the tubes and shake gently, the tubes were observed for a red surface later within 10 minutes which indicated a positive result.

Vogues Proskauer Test

A 6ml volume of the broth was dispensed into test tubes and sealed immediately with aluminum foil tube and sterilized at 121C for 15minutes in an autoclave. After sterilization, the broth was allowed to cool. A sterilized wire loop was used to pick the test organism from a 24 hours culture and inoculated in the broth. The tubes were again sealed with aluminum foil and incubated at 37°C for 48hours. After the incubation, a sterile syringe was used to add 1ml of 40%KOH followed by a 3ml of 5% solution of α-naphthol, the tubes were observed for a red surface layer' within 2-5minutes and the result recorded.

Sugar Fermentation Test

The carbohydrate/sugar fermentation test is used to determine whether or not bacteria can ferment a specitic sugar. It tests for the presence of acid and/or gas produced from sugars fermentation. Approximately, 15g of peptone water was weighed and dissolved in 1000ml of distilled water. About 10g of the carbohydrate source (e.g glucose, lactose, sucrose) was weighed and dispensed into the mixture. 0.1g of phenol red indicator was also weighed and added. Approximately 6ml of broth was dispensed into sterile test tubes, and inverted Durham tubes were inserted to detect gas production. The test tubes were then sealed with aluminum foil, autoclaved at 121℃ for 15minutes and allowed to cool. A sterile wire loop was used to pick the test organisms from a 24hours culture and inoculated into the sterilized medium.The test tubes were sealed with aluminum foil and then incubated at 37℃ for 18-24hours,thereafter,the test tubes were observed for colour changes (acid production and gas production.

RESULTS

Sputum Macroscopy

In this study, the macroscopic examinations of the various sputum samples were observed. The various macroscopic characteristics of the samples are presented in Table 1.The result shows that approximately 50% of the sputum samples appeared yellow while the other samples varied from pale yellow to Amber incolour. About one-third of the total samples were observed to have a

turbid/thick appearance with one-quarter having a slimy and watery texture. Some of the sputum samples were observed to be purulent while others contain blood.

Physiochemical Characteristics of Isolates

The cultural, morphological and biochemical characteristics of bacteria isolates from the samples are presented in Table 2. On Chocolate agar, the colonies appeared milky, creamy, pink, white and light brown in colour with distinct elevations (raised or flat), texture (moist or dry). and edges (rough, smooth or serrated) respectively.

The result from this study showed the isolation of *Mycobacterium tuberculosis, Streptococcus pneumoniae, Staphylococcus aureus, Klebsiella* spp, and *Haemophilus* spp, from the various sputum samples analyzed Results from Gram staining reaction showed that 31 different bacteria isolates were positive for Gram reaction and retained the blue/black colour of the primary dye, crystal violet while 8were Gram negative and appeared pink when viewed under the microscope.

Results from Acid fast stain showed that 4bacteria isolates were acid fast positive by retaining the carbon fushin dye and appeared red when viewed under the microscope.

Results from catalase test showed that 15 bacteria isolates were catalase positive and reacted positively when reacted with hydrogen peroxide. 5 bacteria species were catalase negative.

Results from oxidase test showed that only 5 bacteria isolates reacted positively t oxidase test.15 bacteria isolate appeared oxidase negative. Voges-Proskauer (VP) results showed that 15 bacteria isolates were Voges-Proskauer (VP)negative while 5 bacteria isolates were Voges-Proskauer (VP) positive.

Total Frequencies and Percentage occurrence of all Isolates from the samples

From the different samples evaluated, 20 isolates were obtained. *Escherichia coli* had the highest frequency and percentage occurrence of 5(25%) followed by Staphylococcus pyogens 3(15%), *Haemophilus influenzae* 2(15%) *Proteus 1(15%), Streptococcus pyogens* 3(15%). *Pseudomonas* spp 2(10%) while *Klebsiella* had the least frequency and percentage occurrence of 1(5%) (Table 3). Out of the 20 isolates obtained,4 were acid fast bacteria, 8 were Gram positive bacteria, while 12 were Gram negative bacteria.

Total Frequencies and Percentage occurrence of all Isolates from the Samples Based on Co-Infectivity.

From this study, it was observed that *Mycobacterium tuberculosis* and *Escherichia coli co*-infection had the highest frequency and percentage occurrence of 5(25%), followed by *Mycobacterium 'tuberculosis* and Proteus co-infection 1(15%), while Mycobacterium *tuberculosis, Proteus* and *Haemophilus* co-infection had the least frequency and percentage occurrence of 1(5%). (Table 4). From this study, it was observed that the prevalence of co-

infection between *Mycobacterium tuberculosis* and other bacteria pathogens isolated from sputum samples is 4%.

The result of the biochemical characterization (table 1 below) showed that 20 suspected bacterial isolates were isolated and identified. Out of the 20 bacterial isolates, 4 were streptococuss spp including 2 suspected S. pneumoniae, 5 were Escherichia coli, 3 proteus spp, 2 pseudomonas spp and one S. aureus, 1 klebsiella spp and 1 S. pyrogens were identified including 3 H. influenza. Out of the 20 suspected bacterial isolates, 12 were gram negative (-ve) while the rest are gram positive(+ve).

S/N	Macroscopy	Microscopy	Gram staining	AFB	oxidase	catalase	indole	Voges prosk.	glucose	Lactose	Suspected organism
1	Creamy, smooth	Rod in chains	-	-	-	+	+	+	ag	ag	*Escherichia coli*
2	creamy	Cocci in chains	+	-	-	-	+	-	ag	a	*Streptococcus pyogens*
3	Mucoid, smooth	Rod in chains	-	-	-	+	+	-	ag	a	*Escherichia coli*
4	Creamy, irregular	Long rods in chains	-	+	-	+	+	+	ag	ag	*Escherichia coli*
5	Light brown creamy	Rod in chain	-	-	-	+	+	-	ag	a	*Escherichia coli*
6	Smooth, and irregular mucoid	Long rods in chains	+	+	-	+	-	-	ag	a	Proteus spp
7	Filamentous, wrinkled	Cocci in short chains	+	-	-	-	-	-	a	ag	*Streptococcus pyogens*
8	Irregular, mucoid	Small rods	-	-	+	+	-	-	ag	a	*Pseudomonas spp*
9	Creamy and clustered	Rod in chains	-	-	-	+	-	-	a	a	*Proteus spp*
10	Brownish, mucoid	Small rods in clusters	-	-	-	+	-	-	a	a	*Pseudomonas spp*
11	Filamentous,	Cocci in chains	+	-	-	-	-	-	ag	ag	*Streptococcus*

	dull surface										*pneumonia*
12	mucoid	Long rods	-	-	-	+	-	-	ag	ag	*Proteus spp*
13	Creamy, smooth	Rod	-	+	-	+	+	+	a	ag	*Escherichia coli*
14	Creamy, smooth	Cocci in cluster	+	-	-	+	-	+	ag	ag	*Staphylococcus aureus*
15	Mucoid, smooth	Rod	-	-	-	+	+	+	ag	ag	*Klebsiella spp*
16	Mucoid, rough	Long rod with terminal spores	-	-	+	+	-	-	ag	a	Haemophilus influenza
17	Creamy	Cocci in pairs	+	-	-	-	-	-	ag	ag	*Streptococcus pneumonia*
18	Brown	Distinct colony with rods shape in chains	+	-	+	-	-	-	ag	a	*Streptococuss pyrogenes*
19	Creamy, rough	Rod with terminal spores	-	-	+	+	+	-	ag	ag	*Haemophilus influenza*
20	Creamy and smooth	Rod with terminal spores	-	-	+	+	+	-	a	ag	*Haemophilus influenza*

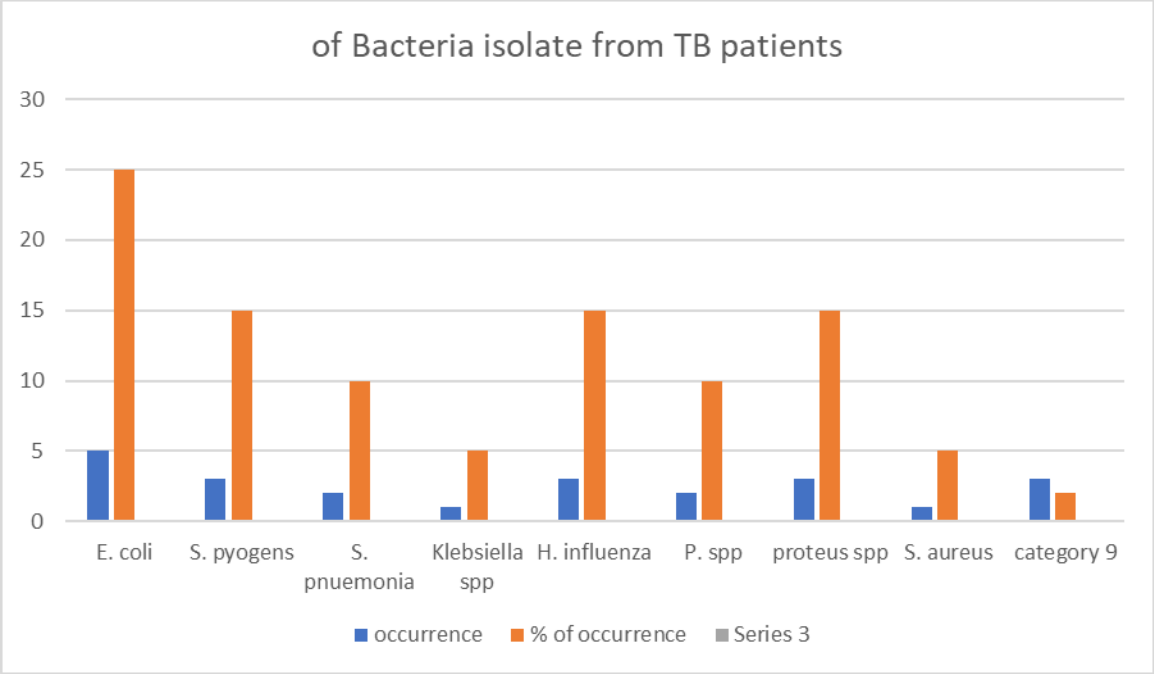
Table 1: macroscopic, microscopy, gram staining and Biochemical tests results

The result in table 2 below shows Escherichia coli has the highest percentage of bacteria present, followed by S. pyogens (15%), haemophilus influenza (15%), proteus spp (15%), S. peumoniae (10%), pseudomonas spp (10%), Klebsiella spp (5%) and S. aureus (5%)

S/N	**ISOLATES**	**OCCURENCE**	**% OF OCCURENCE**
1	Escherichia coli	5	25%
2	Streptococcus pyogens	3	15%
3	Streptococcus pneumonia	2	10%
4	Klebsiella spp	1	5%
5	Haemophilus influenza	3	15%

6	Pseudomonas spp	2	10%
7	Proteus spp	3	15%
8	Staphylococcus aureus	1	5%
TOTAL		**20**	**100%**

Table 2: occurrence of bacteria isolates from suspected tuberculosis patients.



SUSPECTED BACTERIA ISOLATE	**FREQUENCY**	**AFB FREQUENCY**	**AFB COINFECTION FREQUENCY**
Escherichia coli	2	2	50
Proteus	1	1	25
Haemophilus spp	1	1	25

Table 3: frequency and percentage occurrence of isolates from the samples based on co-infection with AFB.

DISCUSSION

The prevalence of co-infection between *Mycobacterium tuberculosis* and other bacteria pathogens isolated from sputum samples in this study is 4%. Co-infection with TB and bacterial

pathogens has been described, in particularly in populations with a high TB prevalence. Differentiating TB from other Lower Respiratory Infections (LRIs) such as Bacteria-pneumonia is an important clinical challenge in these settings, and inability to differentiate TB from other LRIs may result in poorer health outcomes (Whittaker et al.,2019). From this *study, Mycobacterium tuberculosis* and *Escherichia coli* co-infection was reported to be the highest with a frequency and percentage occurrence of 5(25%). Streptococcus *pneumoniae* and *Mycobacterium tuberculosis* in particular are the most important and dangerous bacterial pathogens of the respiratory tract infection. CAP caused by co-infection of Streptococcus *pneumonia and Mycobacterium tuberculosis* has been previously reported only in areas of high MTB prevalence and in the setting of active HIV infection. Garcia in 2019 reported the first case of CAP caused by co-infection with pneumococcus and active pulmonary TB in the absence of HIV co-infection. According to his report, it is likely that the Streptococcus *pneumoniae* may have provided fertile ground for latent TB to reactivate. This study is in line with that of Ihongbe et al., in 2020 who also reported that 44.1% of their subjects were co-infected with *Streptococcus pneumoniae and Mycobacterium tuberculosis.*

Purulent pericarditis is a rare and life threatening condition corresponding approximately with 1% of all cases of pericarditis. In the antibiotic era, the most common causative organism is *Staphylococcus aureus.* Tuberculosis is a common cause of pericarditis,which is mostly common in HIV-endemic areas. Lamas *et* al., in 2019 reported a first case of primary purulent pericarditis with co-infection with *M.tuberculosis* and *S.aureus* as first manifestation of HIV infection with a high prevalence of 24.4% which is in line with this study having a prevalence of 20%.

Lower Respiratory Infections (LRIs) are a major cause of morbidity and mortality worldwide, especially in resource-limited settings. *Streptococcus pneumoniae and Haemophilus influenzae* are the most common respiratory pathogens, *Pseudomonas aeruginosa, Klebsiella pneumonia* and *Burkholderia pseudomallei* are also common etiologies of community-acquired pneumonia (CAP). Attia *et al.,* 2019 reported a case of *Mycobacterium tuberculosis* and *Haemophilus influenza* co-infection with a prevalence of 11.6%. This is in line with our findings which suggest a *MTB-Haemophilus influenza* co-infection prevalence of 5%.

Other bacteria species responsible for causing extrapulmonary TB in tuberculosis patients includes *Chlamydia trachomatis, Shigella spp, Yersinia spp, Listeria monocytogenes etc.* This research shows a high prevalence of *Mycobacterium tuberculosis* and non-tubercular bacteria especially in tuberculosis patients. This may be attributed to a weakened immune system and the environment may play a pivotal role in the spread of this infections.

CONCLUSION

Co-infection with tuberculosis (TB) and bacteria has not been widely reported. Although superadded bacterial infection can occur in TB patients, the simultaneous occurrence of both infections leads to delayed diagnosis and adequate treatment. Tubercular-bacterial co-infection

needs to be considered, especially if TB occurs in atypical pulmonary or extra-pulmonary locations. Hence, bacterial co-infection with *Mycobacterium tuberculosis* has been reported in immuno-compromised asymptomatic patients, their co-occurrence in individuals with low immunity may complicate the clinical presentation, leading to inadequate treatment and unsatisfactory outcomes. It is therefore recommended that with patients with pulmonary tuberculosis, needs additional clinical diagnosis and treatment of the second infection. Also,it is advised that the spread of tuberculosis should be curtailed and well prevented. This can be achieved by proper quarantine techniques and adequate treatment of patients suffering from pulmonary tuberculosis.

Declarations

Ethical approval and informed consent

Ethical Consideration: Ethical consideration was sought for and obtained from the chief medical staff and the head of the medical unit of mile four hospital (MH/578/62/2022. Prior to sample collection, participants were informed clearly about the objectives and procedures of the study. Potential participants were told that participation was totally voluntary and any information obtained would remain confidential.

Authors contribution

OEN and IDC conceptualized the study, OOJ designed the study, NOL participated in the field work, K-MOO participated in data collection. OEN prepared the initial draft of the manuscript. All authors contributed to the development of the final manuscripts and approved its submission.

Conflict of Interest
Authors declared that there was no conflict of interest.

Funding
None

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