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Antimicrobial Pattern of Microorganisms Cultured from the Oral Cavity of Students' from Owerri

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Abstract: Antimicrobial pattern of microorganisms cultured from the oral cavity of students' from Owerri was studied. A total of one hundred and five oral swab samples were collected and analyzed by standardized microbiological techniques. Identification of sub-cultured colonies was done using morphological and biochemical tests. The identified Isolates were also subjected to molecular characterization using the 16S rRNA for bacteria sequences and 18SrRNA for fungal sequences. Antimicrobial susceptibility testing was done by the method of disk diffusion method by Kirby–Bauer. The results of isolate characterization showed the following isolates namely *Streptococcus pyogenes* (34%), *Staphylococcus aureus* (19%), *Lactobacillus casei* (17%), *Pseudomonas aeruginosa* (10%), *Escherichia coli* (8%), *Candida albicans* (6%), *Aspergillus flavus* (4%) and *Aspergillus niger* (2%). Bacteria that were gram-positive were more prevalent (70%) than bacteria that were gram-negative (30%). Bacterial isolates have (88%) distribution while fungal isolates have a (12%) of total microorganism isolated and characterized. *Streptococcus pyogenes* (34%), was the predominant bacteria while *Candida albicans* (6%) was the predominant fungi. Antibiotic susceptibility testing revealed high sensitivity of bacterial isolates to ceftazidime, cefuroxime, augmentin and ciprofloxacin, whereas cefixime and ofloxacin showed the highest resistance. Antifungal susceptibility testing reveals high sensitivity of fungal isolates to ketoconazole, voriconazole amphotericin B while fluconazole showed the highest resistance. The order of sensitivity of antibiotics to bacterial were as follows, Ceftazidime, Cefuroxime, Augmentin and Ciprofloxacin, vancomycin, imipenem, Cefixime and Ofloxacin. For antifungal agents, the orders of sensitivity to fungal were as follows, ketoconazole, voriconazole amphotericin B and fluconazole. Multidrug resistance was detected in (27%) of the isolates suggesting the presence of resistant microorganisms within apparently healthy individuals. The study concludes that the oral cavity of healthy university students contains polymicrobial growth of microorganisms. The detection of multidrug-resistant isolates highlights the need for proper oral hygiene practices, regular access to dental care and rational antimicrobial use. There should be continuous monitoring of oral microbial flora to prevent emergence and spread of antimicrobial resistance that may lead to public health concern.

Keywords: *Antimicrobials, Microorganisms, Oral Cavity and Students.*

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I. INTRODUCTION

The oral cavity of human is complex and dynamic. The human oral cavity consists of microbial ecosystem that harbors diversity of oral microorganisms. Oral microorganisms include bacteria, fungi, viruses and protozoa. These oral microorganisms usually exist in a delicate balance, when the microbiome is stable, oral health is ensured but dysbiosis occurs when humans becomes immunocompromised. (Lamont et al.,2018). The human oral cavity contains over 700 bacterial genera such as Streptococcus, Actinomyces, Veillonella, Fusobacterium, Prevotella, Neisseria, and Lactobacillus species of, many of which colonize and form biofilms on the teeth, tongue, and mucosal surfaces The human mouth has an alkaline condition that is desirable and is an optimum habitat for most species of bacteria, thus making their involvement in various oral disease an established fact (Wade, 2013). There are nearly100 species of fungi found in the human oral cavity and it account for almost 0.004% of the overall oral microorganisms (Caselli et al., 2020). The common genera are Aspergillus, Penicillium, Candida, Aureobasidium, Cladosporium, Fusarium, Cryptococcus, Rhodotorula, Gibberella, Saccharomycetales, and Schizophyllum (Ghannoum et al., 2010; Peters et al., 2017). The oral organisms can be commensal and beneficial; others are opportunistic pathogens which are implicated in oral infections such as oral candidiasis, periodontitis, gingivitis, dental caries and even systemic diseases such as endocarditis and aspiration pneumonia (Wade, 2013).

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These oral microorganisms colonize various niches in the oral like the tongue, saliva, teeth, cheeks, gingival crevices, hard and soft palates. The structure of the oral microbiota is affected by various factors like as age, immune status, diet, hygiene practices, environmental exposures and genetics (Lamont *et al.,* 2018).

This microbial ecosystem exhibits a high degree of structural organization and stability, often forming complex biofilms on oral surfaces. These biofilms are not random assemblies but highly coordinated communities of oral microorganisms which communicate, share resources, and sometimes exchange genetic material, including antibiotic resistance genes (Kolenbrander *et al.,* 2010).

Nigeria that is among the developing countries, have factors like poor oral hygiene, minimum dental care, minimal dietary habits, and self-medication these also contributes to increase habitation of virulent microbes in the oral cavity, especially among young adults, students and elderly people (*Gomez.,* 2017). Imo State, Owerri has students that can be prone to oral diseases because of their unique lifestyles of irregular oral hygiene practices, limited access to fluoride, increase consumption of junk foods, sharing of personal items and utensils, all these can foster microbial colonization, microbial transmission and biofilm formation that may become a public health concern (Dewhirst *et al.,* 2010). Moreover, the oral cavity can also be conducive for multidrug-resistant organisms, and can pose as a significant threat to the health of the public through the potential spread of resistant genes that may lead to antibiotic resistance in clinical settings (Dewhirst *et al.,* 2010). The study aim of this work is to determine the antimicrobial pattern of microorganisms isolated from the oral cavity of man.

II. MATERIALS AND METHODS

➢ *Study Area*

The study area was in Owerri, Imo State. The site of the study is Imo State University, Owerri.

Owerri is one of the three geopolitical zones in Imo State. Imo State is in the eastern part of Nigeria. Neighboring states to Imo state are Abia State, Anambara State, Delta State and Rivers State and (Federal Republic of Nigeria official gazette, 2007).

➢ *Study Population*

The study population was from 105 students of Imo State University, Owerri. The study comprised of 45 males and 60 females which were chosen randomly.

➢ *Sample Collection Source*

Samples were collected from the oral cavities (buccal mucosa and tongue) of students.

- *Samples Collection:*

A total number of 105 sterile swab stick were used to collect specimens from the oral cavity from both male and female students in Owerri. Aseptical methods were used during the collection of samples to avoid contamination. The swabbed sticks were immediately taken to the microbiology laboratory for analysis.

➢ *Identification of Cultured Bacteria*

- *Sterilization Method*

Laboratory benches were disinfected before and after work with 75% alcohol. Glass wares used for the analysis were properly washed with detergent and dried with hot air oven. Then the glasswares were autoclaved to ensure sterilization. Culture media were autoclaved at 121^0 C, 15Psi. Bunsen burner was used to flame all loops to red-hot. Laboratory benches were cleaned before and after work with 75% alcohol (Ohazuruike *et al*., 2017).

- *Preparations of Culture Media*

The culture media namely, Nutrient Agar, MacConkey Agar, Mannitol Salt Agar, Muller Hinton agar and Sabouraud Dextrose agar used were prepared according to manufacturer's instructions. The appropriate quantity of agar was mixed with distilled water, plugged with cotton plug. Autoclaving was done at sterilization point at 121^0C for 15mins and cooled at 45^0C. Autoclaved media were poured unto petridishes and allowed to solidify. The plates were properly labeled after inoculation and then incubated at 37^0C for 24hrs and after which they were examined for growth. Sterility tests of the media were also done (Ohazuruike *et al*., 2017).

- *Sub-Culturing of Plates*

Discrete colonies observed from plates were subcultured to obtain pure cultures. The sub-cultured plates were then incubated for 24hrs at 37^{oC}. The pure culture obtained was used to perform morphological and biochemical tests like colony appearance, shapes, gram staining, catalase tests etc. Stock culture was also kept to perform further tests (Ohazuruike *et al*., 2017).

- *Identification of Bacteria*

Motility test, Gram staining, as well as biochemical tests like; Catalase, Oxidase tests, Coagulase, were carried out (Ohazuruike *et al*., 2017).

- *Gram Staining Tests*

Gram staining method described by (Cheesbrough, 2011) was adopted in this study. Red hot wire loop was used to smear the isolate on a clean grease free slide, which was air dried and heat fixed over a Bunsen burner. The smear was flooded with Crystal violet (primary stain) for 60 seconds, then washed off, flooded with lugol's iodine (Mordant) for 30 seconds, decolorized with 75% alcohol for 30seconds, washed off then counter stained with safranin for 30 seconds. The oil immersion objective (X100) was used to microscopically examine the smear. Gram positive stained purple while gram negative stained red (Cheesbrough, 2011).

- *Motility Test*

The stab method was used to perform this motility test. A semi solid motility medium was used. Inoculums were inoculated unto the semi-solid medium by stabbing a straight

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line, and then it was incubated at 370C for 24hours. Motile bacteria produced a growth away from the stab line into the medium and make the medium opaque (Cheesbrough, 2011).

➢ *Molecular Identification of Bacteria.*

The bacterial deoxyribonucleic acid (DNA) mini prep extraction kit was employed for the extraction of deoxyribonucleic acid. The agarose gel electrophoresis was done to estimate the qualitative genomic DNA. Amplification of the extracted DNA was performed using the Polymerase chain reaction amplification protocol. Molecular characterization using the 16S rRNA sequencing method was performed. ExoSAP Protocol was done on the polymerase chain reaction products. Nimagen brilliant dye terminator cycle sequencing kit was used to sequence fragments according to manufacturer's instructions (Platt *et al*., 2007). Basic Local Alignment Search Tool Nucleotide (BLASTn) was done on the sequence data to identify corresponding bacteria from National center for bioinformatics information (NCBI) as described by (Altschul *et al*., 1990).

➢ *Identification of Fungi*

The standard serial dilution technique and streaking technique methods were employed. The samples were diluted to 10^{-3} and 10^{-5} dilution factors. Bacteria growth was inhibited by the addition of chloramphenicol 50mg/l. Yeast growth was inhibited by the addition of 0.1ml of lactic acid (Valerie, *et al*., 2001). Aliquot 0.1ml of diluents were inoculated onto already prepared plates of Sabouraud dextrose agar (SDA).The plates were incubated at 28 ± 2°C for 7 days and fungal growth were later examined. Sub culture of Fungi colonies grown on media was done. (Larone, 2011). The results were expressed in colony forming unit per sample gram (CFU/g). Sensitivity test was performed using mueller hinton agar.

➢ *Morphological and Microscopic Identification of Fungi*

Colony morphology and examination of the fungi microscopically were used to identify isolated moulds. The fungal isolates were subcultured for identification. The fungi isolated were mounted on a clean grease free slide, then stained with lacto phenol-cotton blue to observe fungal structures and spores, and then the slide was covered with a cover slip, and examined under microscope. Fungi were identified based on the morphology and characteristics of their colony and spores respectively (Pitt *et al*., 1992).

- *Germ Tube Test:*

This test is done by incubating the yeast in serum at 37°C for 2-4 hours, which causes *C. albicans* to produce characteristic "germ tubes" (Pitt *et al*., 1992).

➢ *Molecular Identification of Fungi*

The fungi deoxyribonucleic acid (DNA) mini prep extraction kit was employed for the extraction of deoxyribonucleic acid. The agarose gel electrophoresis was done to estimate the qualitative genomic DNA. Amplification of the extracted DNA was performed using the Polymerase chain reaction amplification protocol. Molecular characterization using the 18S rRNA sequencing method was performed for fungi. ExoSAP Protocol was done on the polymerase chain reaction products. Nimagen brilliant dye terminator cycle sequencing kit was used to sequence fungi fragments according to manufacturer's instructions (Platt *et al*., 2007). The Basic Local Alignment Search Tool Nucleotide (BLASTn) was done on the sequence data to identify corresponding fungi from National center for bioinformatics information (NCBI) as described by (Altschul *et al*., 1990).

➢ *Determination of Susceptibility Test for Bacteria*

Susceptibility test for Antimicrobial was done using the disk diffusion method by Kirby-Bauer. Muller Hinton agar medium was used. The following antibiotics were employed for sensitivity was done using the following antibiotics namely, Ceftazidime (30 µg), Cefuroxime (30 µg), Augmentin (30µg), vancomycin (30 µg) imipenem (10 µg), Ciprofloxacin (5 µg), Cefixime (5 µg) and Ofloxacin (2 µg). The fungi growth were standardized to 1.0×10^{6} cfu/ml (0.5 McFarland standards). Then, 0.1ml of diluent was taken and spread on the surface of Muller Hinton agar using sterile glass rod. The antibiotic disc was asceptically placed on muller hinton plates with a sterile forceps. The agar plates were left on the bench for about 30mins to allow for diffusion of the antibiotics and were incubated at 37°C for 24hrs. The meter rule was used to measure the zones of inhibition after 24 hours incubation and then recorded in millimeter (mm). The results were interpreted based on break point reading set by the Clinical Laboratory Standards Institute. (CLSI, 2006).

➢ *Determination of Susceptibility Test for Fungi*

The approved standardized method for testing for the filamentous fungi moulds. (CLSI M38-A standard for moulds) has been set by the Clinical Laboratory and Standards Institute (CLSI) (Espinel-Ingroff, *et al*., 2010).

The oxoid disc diffusion method is the standard method. This method was set by the National Committee for Clinical Laboratory Standards, 2002 (Espinel-Ingroff, *et al*., 2010). Four oxoid antifungal agents were employed and are as follows: Amphotericin B (20 µg/ml), fluconazole (10 µg/ml), ketoconazole (25 µg/ml) and voriconazole (1 µg/ml).

- *Break Point end Reading:*

The Minimum Inhibition Concentration (MIC)

✓ The susceptible, ≥17 mm (azoles) and ≥15 mm (amphotericin B);
✓ The intermediate, 14 to 16 mm (azoles) and 13 to 14 mm (amphotericin B);
✓ The resistant, ≤13 mm (azoles) and ≤12 mm (amphotericin B) (CLSI, 2006).

➢ *Statistical Analysis*

Statistical analysis done using frequency distribution, percentage occurrence and the analysis of variance.

III. RESULTS

Total number of 105 oral swab samples were collected from the students. The study population, the gender distribution of students consisted of 45 males (43%) and 60

females (57%). This is as shown in Table 1. Preliminary identification of bacterial isolates was performed on the colony morphology and cellular arrangement observed after culture. The colonial and morphological characteristics of bacteria are presented in Table 2. Biochemical Characteristics of bacterial Isolates which are *Streptococcus* spp, *Staphylococcus aureus, Lactobacillus* spp, *Pseudomonas* spp, *Escherichia coli* are shown in table 3. Macroscopic and microscopic characteristics of fungi are in table 4. These fungi are *Candida albicans*, *Aspergillus flavus* and *Aspergillus niger.* Table 5 shows the molecular characterization using the sequence 16S rRNA identity of different bacteria which were *Streptococcus pyogenes, Staphylococcus aureus, Lactobacillus casei, Pseudomonas aeruginosa, Escherichia coli.*

The identification of fungi was also performed genetically by sequencing of 18S rRNA gene using ITS1 and ITS4 primers. The fungi seen were *Candida albicans*, *Aspergillus flavus* and *Aspergillus niger* as seen in table 6.

Table 1 Gender Distribution of Students

S/N	Gender	Frequency (n)	Percentage (%)
1	Male	45	43
2	Female	60	57
	Total	100	100

Table 2 Colonial and Morphological Characteristics of Cultured Bacteria

Samples	Colour	Shape	Surface	Arrangement	Probable Organism
1	Creamy white	Circular	Smooth	Cocci in chains	*Streptococcus spp.*
2	Yellow	Round	Glassy	Cocci in clusters	*Staphylococcus aureus*
3	White	Circular	Smooth and dry	Rods in singles	*Lactobacillus spp.*
4	Greenish	Irregular	Smooth	Rods	*Pseudomonas spp.*
5	Pink	Raised growth	Smooth and dry	Rods as singles	*Escherichia coli*

Isolates identified was based on colonial morphology, gram staining, and biochemical tests.

Table 3 Biochemical Characteristics of Isolates

Isolates	Bacteriological Tests			Biochemical Tests											Probable Organism
S/N	Gram reaction test	Cellular arrangement	Motility test	Catalase test	Citrate test	Indole test	Oxidase test	Coagulase test	Voges–Proskauer test	Methyl Red test	Glucose test	Lactose test	Sucrose test		
1	+	*Cocci in chains*	–	–	–	–	–	–	–	+	+	+	–		*Streptococcus spp.*
2	+	*Cocci*	–	+	–	–	–	+	+	+	+	+	+		*Staphylococcus aureus*
3	+	*Rods*	–	–	–	–	–	–	–	+	+	+	+		*Lactobacillus spp.*
4	–	*Rods*	+	+	+	–	+	–	–	–	–	–	–		*Pseudomonas spp.*
5	–	*Rods*	+	+	–	+	–	–	–	+	+	+	+		*Escherichia coli*

Table 4 Macroscopic and Microscopic Characteristics of Fungi

Isolate	Macroscopic Features	Microscopic Features	Probable Organism
F1	Creamy white colonies; smooth, soft, pasty texture; convex elevation; entire margin on Sabouraud Dextrose Agar (SDA)	Oval budding yeast cells observed; presence of pseudohyphae; Gram positive yeast cells under microscopy	*Candida albicans*
F2	Greenish colonies Reverse-Yellowish green	Hyphae are septate and hyaline.	*Aspergillus flavus*
F3	Black in colour Reverse: creamish-yellow	Septated hyphae, long smooth and colourless	*Aspergillus niger*

Table 5 Molecular Characterization using 16S rRNA Identity of Different Bacteria

S/N	Biochemical Isolates	Percentage (%)	Molecular Sequence
1	*Streptococcus* spp.	96	*Streptococcus pyogenes* NC018936
2	*Staphylococcus aureus*	97	*Staphylococcus aureus MH401415*
3	*Lactobacillus* spp	99	*Lactobacillus casei* AF158574
4	*Pseudomonas spp*	96	*Pseudomonas aeruginosa WE 41437*
5	*Escherichia coli*	99	*Escherichia coli M75029*

Table 6 Molecular Sequence 18S rRNA Identity of Different Fungi

S/N	Sequence ID	Percentage (%)	*NCBI Match*	Isolate
1	TXID237561.	99	*Candida albicans* TXID237561.	*Candida albicans*
2	NR111041.1	99	*Aspergillus flavus NR135325*	*Aspergillus flavus*
3	AY373852.1	91	*Aspergillus niger AY373852*	*Aspergillus niger*

Figure 1: Frequency Distribution of Isolated Microorganisms which were *Streptococcus pyogenes* (34%), *Staphylococcus aureus* (19%), *Lactobacillus casei* (17%), *Pseudomonas aeruginosa* (10%), *Escherichia coli* (8%), *Candida albicans* (6%), *Aspergillus flavus* (4%) and *Aspergillus niger* (2%). *Streptococcus pyogenes* (34%) *was* the most frequent isolated bacterial isolate while *Escherichia coli* (8%) was the least isolated bacterial. The most frequent isolated fungal was *Candida albicans* (6%) while *Aspergillus niger* (2%) was the least isolated fungi.

Table 7: Frequency Distribution of Isolated microorganisms. Bacterial isolates have 88% distribution while fungal isolates have a 12% of total microorganism isolated and characterized.

Table 8: showed the Gram Reaction of Bacterial Isolates. Gram staining was done to categorize bacterial isolates into Gram-positive and Gram-negative groups. Gram positive has a percentage of 70 while gram negative have a percentage of 30.

The antibiotic sensitivity and resistance pattern of the isolates to selected antibiotics are shown in Table 9. The bacteria identified showed various level of sensitivity to the following antibiotics which are Ceftazidime, Cefuroxime, Augmentin and Ciprofloxacin, vancomycin, imipenem, Cefixime and Ofloxacin. The highest sensitivity for all the isolates was observed with Ceftazidime. Sensitivity to bacterial isolates was seen with Ceftazidime, Cefuroxime, Augmentin, vancomycin and Ciprofloxacin while the bacterial isolates were resistant to imipenem, Cefixime and Ofloxacin.

The antifungal susceptibility profile of various fungal isolates was observed in Table 10.The fungal isolates showed different level of sensitivity to ketoconazole, voriconazole amphotericin B and fluconazole. The highest level of sensitivity for all the fungi was observed for Ketoconazole while the fungal isolates were resistant to fluconazole.

Figure 2 showed Isolates that were resistant to three or more antibiotic classes were classified as multidrug resistant (MDR) isolates. The prevalence of multidrug resistance (MDR) isolates were observed. Multidrug resistant isolates were 27% while non- multidrug resistant isolates were 73%.

Statistical analysis done using the antibiotic susceptibility tests using the analysis of variance showed that there was no significant difference ($P<0.05$) between Ceftazidime, Cefuroxime, Augmentin and Ciprofloxacin but there was a significant difference ($P<0.05$) between Ceftazidime ,Cefuroxime, Augmentin and Ciprofloxacin and vancomycin imipenem, Cefixime and Ofloxacin.

Statistical analysis done for antifungal susceptibility test for antifungal agents noted that the analysis of variance, showed no significant difference ($P<0.05$) between antifungal drugs of ketoconazole, voriconazole and amphotericin B but there was a significant difference ($P<0.05$) between, ketoconazole, voriconazole amphotericin B and fluconazole.

Table 11 showed the antimicrobial pattern for zone of inhibition for isolates in mm. *Staphylococcus aureus* was the most sensitive bacterial isolate while *Pseudomonas aeroginosa* was the resistant bacterial isolate. The level of sensitivity of bacterial isolates to antibiotics were in the following order*, Staphylococcus aureus, Escherichia coli, Streptococcus pyogenes, Lactobacillus casei and Pseudomonas aeruginosa.* The order of sensitivity of antibiotics to bacterial were in the following order

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Ceftazidime, Cefuroxime, Augmentin and Ciprofloxacin, vancomycin, imipenem, Cefixime and Ofloxacin. Table 12 showed the susceptibiity test using oxoid antifungal drugs in mm. For antifungal agents, the orders of sensitivity to fungal isolates are in the following order: ketoconazole, voriconazole amphotericin B and fluconazole.

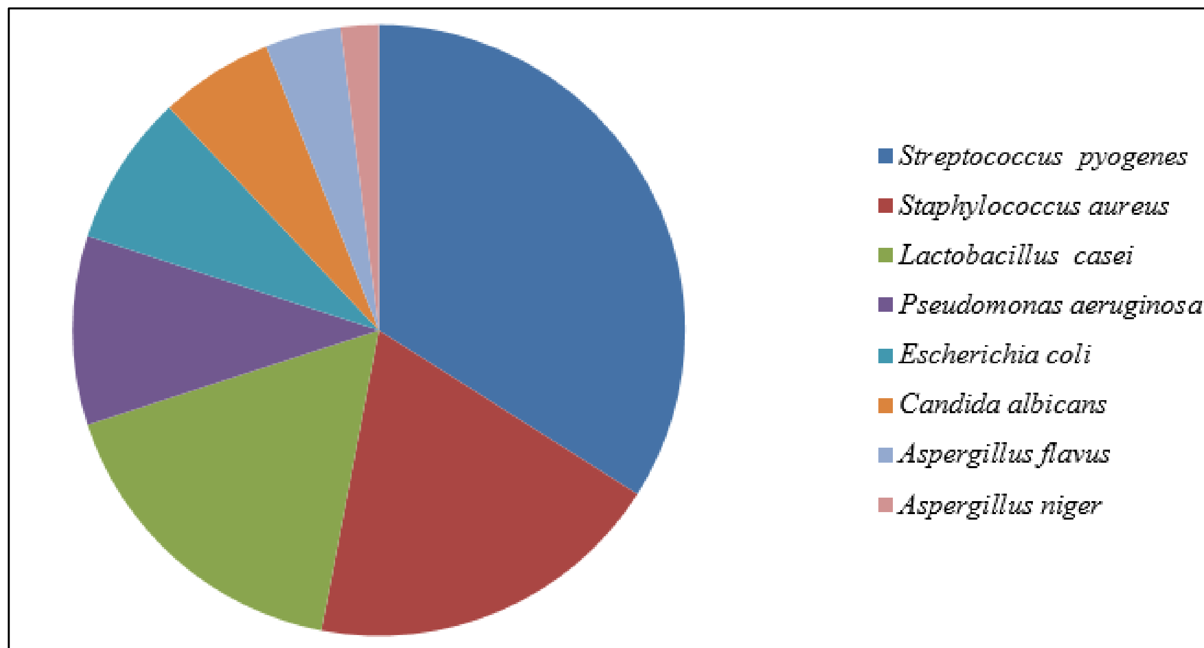

Fig 1 Frequency Distribution of Isolated Microorganisms.

Table 7 Frequency Distribution of Isolated Bacteria and Fungi.

Isolate	Frequency (n)	Percentage (%)
Bacteria	73	88
Fungi	10	12
Total	83	100

Table 8 Gram Reaction of Bacterial Isolates.

Gram Reaction	Frequency (n)	Percentage (%)
Gram positive	58	70
Gram negative	25	30
Total	83	100

Table 9 Antibiotic Susceptibility Pattern.

Antibiotic	Sensitive (%)	Resistant (%)
Ceftazidime (30µg)	80	20
Cefuroxime (30 µg)	80	20
Augmentin (30µg)	80	20
Vancomycin (30µg)	40	60
Imipenem (10 µg)	20	80
Ciprofloxacin (5 µg)	60	40
Cefixime (5 µg)	10	90
Ofloxacin (2 µg)	10	90

Table 10 Antifungal Susceptibility Pattern

Antifungal drugs	Sensitive (%)	Resistant (%)
Amphotericin B (20 µg/ml)	66	34
Flucanozole (25 µg/ml)	33	67
Ketoconazole (10 µg/ml)	100	0
Vorioconazole (1 µg/ml)	100	0

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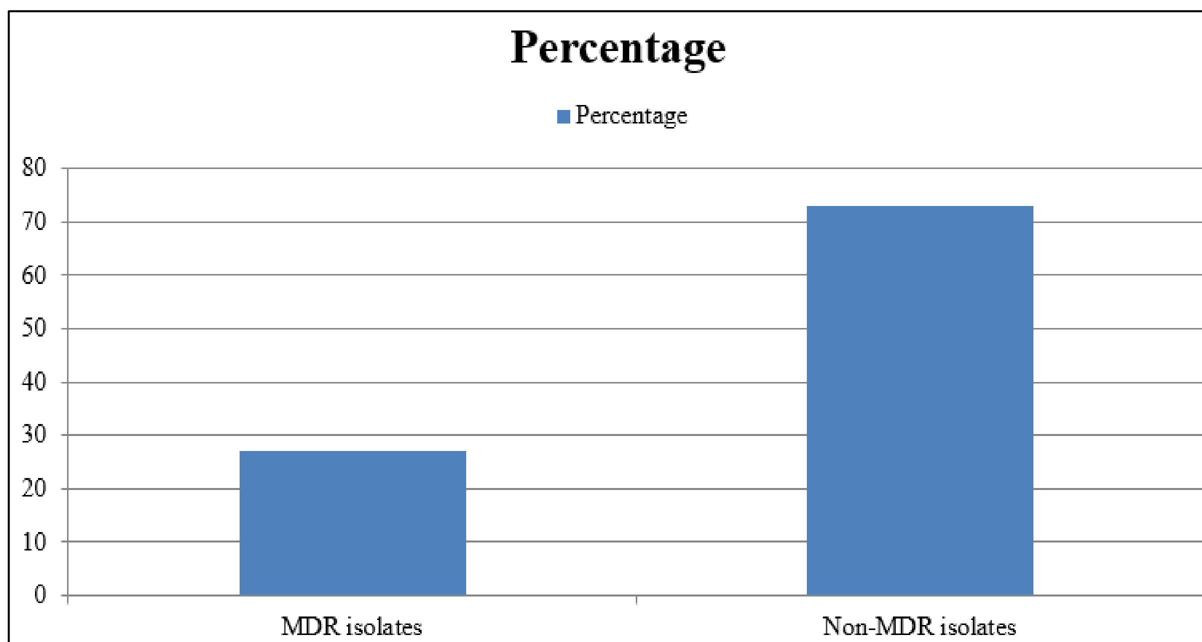

Fig 2 Prevalence of Multidrug Resistance (MDR) Isolates.

Table 11 Antimicrobial Pattern for zone of inhibition for isolates in mm.

Organism	CET (30 µg)	CEF (30 µg)	AUG (30 µg)	VAN (30 µg)	IMI (10 µg)	CIP (5 µg)	CEF (5 µg),	OFL (2 µg),
Streptococcus pyogenes (mm)	26 (S)	25(S)	23 (S)	21 (I)	19 (I)	23 (S)	12 (R)	12 (R)
Staphylococcus aureus (mm)	28 (S)	25 (S)	26 (S)	25 (S)	16 (R)	25 (S)	17 (R)	16 (R)
Lactobacillus casei (mm)	25(S)	19 (I)	21 (S)	18 (I)	14 (R)	16 (I)	13 (R)	12 (R)
Pseudomonas aeroginosa (mm)	19 (I)	24 (S)	19 (I)	14 (R)	13 (R)	14 (R)	13 (R)	14 (R)
Escherichia coli (mm)	25 (S)	24 (S)	26 (S)	21 (I)	17 (I)	24 (S)	16 (I)	15 (I)

- *KEY:*

CET: Ceftazidime (30 µg), CEF : Cefuroxime (30 µg), AUG : Augmentin (30µg), VAN : vancomycin (30 µg), IMI: imipenem (10 µg), CIP : Ciprofloxacin (5 µg), CEF : Cefixime (5 µg), OFL: Ofloxacin (2 µg).

- *Key:*

The Breakpoint table for *Staphylococcus aureus* Susceptible (S) ≥ 25 mm, Intermediate (I) 18-24 mm, The Resistance (R) ≤ 19mm. Family of Enterobacteriaceae Susceptible (S) ≥ 23mm, Intermediate (I) 15-22 mm, The Resistance (R) ≤ 14mm, Others Susceptible (S) ≥ 20 mm, Intermediate (I) 15-19 mm, Resistance (R) ≤ 14mm (CLSI, 2006) (CLSI, 2006).

Table 12 Susceptibiity Test Using Oxoid Antifungal Drugs in mm.

ISOLATES	Amphotericin B (20 µg/ml)	Flucanozole (25 µg/ml)	Ketoconazole (10 µg/ml)	Vorioconazole (1 µg/ml)
Candida albicans	23.00(S)	15.00(I)	35.00(S)	32.00(S)
Aspergillus flavus	17.00(S)	15.00(1)	30.00(S)	28.00(S)
Aspergillus niger	13.00(I)	13.00(R)	28.00(S)	21.00(S)

Endpoint reading for minimum inhibition concentration for oxoid antifungal drugs in mm.

- ✓ Susceptibility, ≥17 mm (azoles) and ≥15 mm (amphotericin B);
- ✓ Intermediate, 14 to 16 mm (azoles) and 13 to 14 mm (amphotericin B);
- ✓ Resistance, ≤13 mm (azoles) and ≤12 mm (amphotericin B) (CLSI, 2006).

IV. DISCUSSION

This study revealed that *Streptococcus pyogenes* (34%) was the predominant isolate which agrees with the findings of (Xu *et al.,* 2022) and that of that found out that in order of occurrence, *Streptococcus* spp., had the highest frequency of (42%), followed by *Prevotella* spp which was (34%), *Staphylococcus* and *Clostridium* spps. were least frequently observed and accounted for 15% and 9% respectively. *Streptococcus pyogenes* constitute major primary colonizers in dental biofilms. Their ability to adhere strongly to tooth

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surfaces and form extracellular polysaccharides contributes to their dominance in the oral cavity.

The isolation and identification of *Staphylococcus aureus* (19%) and Gram-negative bacteria such as and *Pseudomonas aeruginosa* (10%) and *Escherichia coli* (8%) suggests opportunistic colonization. Similar studies by (Lemberg *et al.,* 2022).

Have reported transient oral carriage of these organisms, particularly among young adults and healthcare-related populations, indicating that the oral cavity may become a habitat for virulent pathogens.

The isolation of *Candida albicans* from this study supports the established understanding that Candida species exist as commensals within the oral cavity but may become opportunistic under favorable conditions. The work of (Lemberg *et al.,* 2022) revealed that oral *Candida* was characterized, which is influenced by host immunity, oral hygiene, and microbial interactions within the oral biofilm

Comparatively, earlier studies by (Kashya, *et al*., 2024) described *Candida albicans* as the most common oral yeast which agrees with this study. *Candida albicans* is capable of transitioning from harmless colonizer to pathogen under favourable conditions. The predominance of Gram-positive bacteria in this study agreed with the findings of (Xu *et al.,* 2022) which describes Gram-positive cocci and rods as dominant early colonizers of oral surfaces. Thick peptidoglycan layers enhance adhesion and survival in fluctuating oral environments, explaining their higher prevalence

Overall, the findings of this study agrees with global reports describing the oral cavity as a complex microbial ecosystem dominated by Gram-positive organisms, especially *Streptococcus pyogenes*, with coexistence of opportunistic bacteria and fungi such as *Candida albicans*. This ecological balance may shift toward pathogenicity under favorable conditions, emphasizing the importance of oral hygiene and antimicrobial stewardship (Xu *et al.,* 2022).

This study observed that the bacteria showed various levels of sensitivity to Ceftazidime, Cefuroxime, Augmentin and Ciprofloxacin, vancomycin, imipenem, Cefixime and Ofloxacin. The highest sensitivity for the bacteria was observed with Ceftazidime. Sensitivity to bacteria was seen with Ceftazidime, Cefuroxime, Augmentin, vancomycin and Ciprofloxacin while the bacteria were resistant to imipenem, Cefixime and Ofloxacin. This finding disagreed with the work of (Udoh *et al*., 2025), that revealed that all antibiotic susceptibility pattern of isolates of *Streptococcus* spp, *Staphylococcus aureus*, *Escherichia coli* and *Bacillus* spp were resistant to antibiotics like ceftazidime, Vancomycin , Chloramphenico and Levofloxacin with different degrees. The highest antibiotic resistance value obtained for gam negative bacteria was 66.7% for Ceftazidime. The work of (Babaiwa, & Amarikwa, 2024) showed that Fluoroquinolones like ciprofloxacin was sensitive to both gram positive and gram negative bacteria like *Streptococcus pyogenes, Staphylococcus aureus, Pseudomonas aeruginosa* and *Escherichia coli.*

Fungal isolates showed different level of sensitivity to ketoconazole, voriconazole amphotericin B and fluconazole. The sensitivity with the highest value for all the bacteria was seen for Ketoconazole while the fungal isolates were resistant to fluconazole, this showed similarity to the work of (Krishnan, 2012) that found out that ketoconazole and nystatin showed high sensitivity to oral *Candida albicans*, also *Candida albicans* was resistant to fluconazole.

The detection of multidrug resistance of this study concurred with the work of (Udoh *et al*., 2025) which detected multidrug resistance in oral microorganisms. There is a current global concerns regarding increasing antimicrobial resistance among commensal microbiota, highlighting the need for surveillance and rational antibiotic use even in non-hospital settings (Udoh *et al*., 2025).

V. CONCLUSION

Human oral cavity can serve as reservoir for microorganisms some of which possess antibiotic resistance traits. The recovery of multiple isolates from oral cavity samples demonstrates the polymicrobial nature of the oral microbiome. The predominant occurrence of *Streptococcus pyogenes* indicates that Gram-positive bacteria constitute a major component of the normal oral flora. The presence of other microorganisms like *Staphylococcus aureus*, *Lactobacillus casei*, *Pseudomonas aeruginosa*, and *Escherichia coli* as well as the isolation of *Candida albicans, Aspergillus flavus* and *Aspergillus niger* suggests that opportunistic microorganisms may coexist within the oral cavity even among apparently healthy individuals. The sensitivity of bacteria with the highest value was seen with Ceftazidime. Sensitivity to bacterial isolates was seen with Ceftazidime, Cefuroxime, Augmentin, vancomycin and Ciprofloxacin while the bacterial isolates were resistant to imipenem, Cefixime and Ofloxacin. The highest sensitivity for fungi was seen for Ketoconazole, voriconazole and amphotericin B while the fungal isolates were resistant to fluconazole. The detection of multidrug-resistant isolates points to the emerging challenge of antimicrobial resistance within oral microbial communities. This work shows the importance of constant monitoring of oral microorganisms, regular dental check-up and prescribed use of antimicrobial agents to prevent development and spread of resistant microorganisms. It is also important to know etiological microorganisms of oral infection so as to provide adequate antimicrobial cure.

REFERENCES

[1]. Altschul, S. F., Warren, G., Miller, W., Myers, E.N., Luman, D. (1990). Basic Alignment Search Tool. *Journal of Molecular biology. 1990; 215(3):403-410.*

[2]. Babaiwa, U. F., & Amarikwa, N. A. (2024). Susceptibility profile of bacteria obtained from human oro-dental plaques at the dental clinic of the University

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of Benin Teaching Hospital, Benin City, Nigeria. *West African Journal of Pharmacy*, 35(1), 95-101.

[3]. Caselli, E.F., D'AccoltiM.S., Bassi, C., Mazzacane, S. (2020). Defining the oral microbiome by whole-genome sequencing and resistome analysis: the complexity of the healthy picture. BMC *Microbiol*.20:120. 10.

[4]. Cheesbrough, M. (2011). Distinct Laboratory Practice in *Tropical Countries.* Part 2: Cambridge University Press, United Kingdom, pp: 135—162.

[5]. Clinical Laboratory Standards Institute. (2006). Performance standards for antimicrobial disk susceptibility tests; Approved standard— 9th ed. CLSI document M2-A9. 26:1. Clinical Laboratory Standards Institute, Wayne, PA.

[6]. Dewhirst, F. E., Chen, T.I. Paster, B. J., Tanner, A. C., YuW. H. (2010). The human oral microbiome. J. *Bacteriol*.1925002–5017.

[7]. Espinel-Ingroff, A., White, T., & Pfaller, M. A. (2010). Antifungal agents and susceptibility tests. *Manual of clinical microbiology*. ASM Press, Washington D.C. 7th ed. *pp* 1640–1652.

[8]. Federal Republic of Nigeria official gazatte. (2007). Public Procurement Act. Government notice number 142. Lagos, Nigeria.

[9]. Ghannoum, M. A., Jurevic, R. J., Mukherjee, P. K.,Cui, F., Sikaroodi, M., Naqvi, A. (2010). Characterization of the oral fungal microbiome (mycobiome) in healthy individuals. PLoS *Pathog*.6:e1000713.

[10]. Gomez, A., Nelson, K. E. (2017). The oral microbiome of children: development, disease, and implications beyond oral health. Microb. *Ecol*.73492–503.

[11]. Kashya, B., Padala, S.R., Kaur, G., and Kullaa, A. (2024). Candida albicans Induces Oral Microbial Dysbiosis and Promotes Oral Diseases. *Microorganisms*, 12(11):2138.

[12]. Kolenbrander, P. E., Palmer, R. J.Jr., Periasamy, S., Jakubovics, N. S. (2010). Oral multispecies biofilm development and the key role of cell-cell distance. *Nat. Rev. Microbiol*.8471-480.

[13]. Krishnan, P. A. (2012). Fungal infections of the oral mucosa. *Indian J Dent Res*; 23:650-9.

[14]. Lamont, R. J., Koo, H., Hajishengallis, G. (2018). The oral microbiota: dynamic communities and host interactions. Nat. *Rev. Microbiol*.16745–759.

[15]. Larone, D.H. (2011). *Medically Important Fungi*. A Guide to Identification.ASM Press, Washington, D.C. p. 185.

[16]. Lemberg, C., De San Vicente, K.M., Fróis-Martins, R., Altmeier, S., Van Du T Tran, V. D., Mertens, S., Amorim-Vaz, S., Shanker Rai, L., D'Enfert, C., Marco., Pagni, M., Sanglard, D., Gut-Landmann, S. (2022). *Candida albicans* commensalism in the oral mucosa is favoured by limited virulence and metabolic adaptation. *Plos Pathogens*, 11; 18 (4):e1010012.

[17]. Ohazuruike, N.C., Ohalete, C.N., Njoku-Obi, T.N., Uwaezuoke, J.C., Obiukwu, C.E., Nwachukwu, M.I., Dike, S.K., Ohabughiro, N.B., Nnagbo, P.A. (2017). Seminar Presentation and Research Practicals. Henco Printing Press, Owerri Imo Nigeria, Pp.175-250.

[18]. Peters, B. A., Wu, J., Hayes, R. B., Ahn, J. (2017). The oral fungal mycobiome: characteristics and relation to periodontitis in a pilot study. BMC *Microbiol*.17:157.

[19]. Pitt, J.I., Hocking, A.D., Samson, R, A. & King, A.D. (1992). Recommended Methods for the Mycological Examination of Foods. *Modern Methods in Food Mycology*. Elsevier Science Ltd., Amsterdam, pp: 388-389.

[20]. Platt, A. R., Woodhall, R. W. and George, A.L. (2007). Improved DNA sequencing quality and efficiency using an optimized fast cycle sequencing protocol. *Bio Techniques* 43 (1): 58-62.

[21]. Valerie, T., Micheal, E.S., Philip, B. M., Herbert, A. K. & Ruth, B. (2001). *Bacteriological Analytical Manual*. Yeast, Molds and Mycotoxins. Cambridge University Press. pp74- 76.

[22]. Wade, W. G. (2013). The oral microbiome in health and disease. *Pharmacol.* Res.*69137–143.*

[23]. Xu T, Chen N, He X and Chen F (2023) Editorial: The relationship of oral and other body sites microbiome in human diseases. *Front. Cell. Infect. Microbiol*. 13:1276473.

[24]. Udoh, M. A. , Onwuezobe I. A. , Emanghe U. E. , Johnson E.A. , Ijezie A. E. , Olayemi A. G. (2025). Prevalence and antibiotic susceptibility pattern of agents of dental caries among patients in Uyo, *Nigeria Ibom Medical Journal 18, 3, 485-492.*