MICROBIOLOGICAL ANALYSIS OF BARBER'S TOOLS FROM SALONS AROUND UNIVERSITY OF BENIN

BY

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A PROJECT REPORT SUBMITTED TO THE DEPARTMENT OF MICROBIOLOGY, FACULTY OF LIFE SCIENCES UNIVERSITY OF BENIN, BENIN CITY, IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF BACHELOR OF SCIENCE (B.Sc HONS) DEGREE

AUGUST, 2023.

CERTIFICATION

This is to certify that this project work was carried out by Priscillia ISIMENMEH of the Department of Microbiology, Faculty of Life Science, University of Benin, Benin City.

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Prof. (Mrs.) F. I. Akinnibosun Date
(Project Supervisor)

……... ……..

Prof. (Mrs.) F. I. Akinnibosun Date
(Head of Department)

DEDICATION

This project work is dedicated to the Almighty God for his grace and mercies throughout my period of study and also to my late mom, Mrs Helen Abu, Rest in Power Mom.

ACKNOWLEDGEMENTS

My sincere appreciation goes to the Almighty God for his grace and mercies throughout my period of study.

I wish to acknowledge my project supervisor and also my head of department Prof. Mrs. F. I. Akinnibosun for her patience and understanding towards me and the success of this project and also for her motherly and inspiring role in the administration of the Department. May God Almighty richly bless you ma for your efforts.

I want to also thank my Lecturers for their academic Mentorships and assistance throughout my stay in this school especially.

With deep sense of honor, I will not fail to appreciate my wonderful Family, My Aunt, Mrs Maris Ogbodu and My Elder Brother, Mr Isimenmeh Precious for their financial support, Prayers and words of wisdom. To my amazing Friend, Mr Victor Ibukun Ghaoise for his Emotional, Financial and academic support all the way. I love you all.

I will not fail to appreciate my course mates and friends who have contributed in one way or the other to the success of the work, God bless you all.

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ABSTRACT

The presence of microorganisms on barber's tools has garnered significant attention due to its potential impact on hygiene and public health. This project was aimed to investigate and analyze the diversity, abundance, and pathogenic potential of microorganisms found on barbering tools, focusing on shaving sticks, clippers, and combs, within a range of barbershops. The proliferation of microorganisms on these tools can potentially lead to the transmission of various bacterial, fungal, and viral agents and also hair and skin infections including; impetigo, head lice, barbers' itch, and tinea capitis, raising concerns about skin and respiratory infections among clients. This study aimed at isolation and identification of pathogenic microorganisms associated with barber's equipment, three (3) clippers sample were collected from each of the three (3) barbing salons, three (3) comb samples were collected from the three (3) salon each and three (3) used personal shaving sticks were collected from three (3) individuals each making a total of nine samples (9) on the Combs, Clippers and Shaving sticks. Identification of the microorganisms after culturing was done using Gram staining techniques and biochemical test. Antibiotic sensitivity test was also carried out using Kirby Bauer disc diffusion method. The results showed that bacteria and fungi were present on the barber's tools. The probable identified bacterial isolates were *Clostridium* sp., *Pseudomonas* sp., *Bacillus* sp., *Klebsiella* sp., *Staphylococcus* sp. *Klebsiella* sp. was the most prevalent bacterial isolate while *Clostridium* sp. was the least prevalent bacterial isolate. *Saccharomyces* sp. was the most prevalent fungal isolate while *Fusarium* sp. was the least prevalent fungal isolate. This study reveals that barber's tools without treatment would pose a possible hazardous health effect. Therefore, barbers should ensure compliance with relevant prevention and control options to avoid risks to human health.

CHAPTER ONE

INTRODUCTION

1.1 Background of the study

In our daily grooming and beauty practices, we often rely on a variety of tools and equipment to enhance our appearance and personal hygiene. Items such as clippers, shaving sticks, and hair combs play a significant role in these routines. However, it is important to recognize that these tools, if not properly cleaned and sterilized, come into direct contact with the skin can serve as potential reservoirs for microorganisms (Stout *et al*., 2011). Microorganisms are ubiquitous in nature as they can colonize various surfaces, existing even on skin surfaces and are continually introduced into the environment which could easily spread between client and operators via contact with unwashed hands, soiled equipment or contact with blood and body substances (De Souza and Shibu, 2004). The presence of bacteria, fungi, and even viruses on these items raises concerns about potential health risks associated with their use. Pathogenic microorganisms can cause a range of infections, including skin infections, folliculitis, fungal infections, skin irritations, rashes, blood-borne viral diseases, and even more severe infections in individuals with compromised immune systems (Enemuor *et al.,* 2013). Therefore, it is crucial to assess the microbial contamination of these tools implement appropriate measures to ensure their safe usage and also for developing effective strategies to mitigate the risk of transmission and infection (Moore *et al.,* 2007). By investigating the microbial contamination of clippers, shaving sticks, and hair combs we can shed light on the potential hazards associated with these tools and emphasize the importance of proper cleaning and sterilization practices. Healthcare is one of the most important aspects of all human endeavors aimed at improving the quality of life, since

sound health is absolutely necessary for strength and prosperity of a nation. Health has been declared as the fundamental human right. Despite this recognition, there is a denial of this right to millions of people especially in the developing countries (Enemour *et al.,* 2012). In the developing countries it is known that infectious disease cause about 25% of all human death and account for 11 million deaths yearly (Kumar and Clark, 2005). Many of the infectious diseases are preventable or treatable but have continued to be successful due to lack of personal and environmental hygiene, ignorance and poor political commitment from the government. In developing countries, infections remain the main cause of death in humans and are mostly associated with poverty and overcrowding while in developed countries the prevalence of infection are reduced by increasing prosperity, universal immunization and appropriate antibiotics usage. Important routes of transmission of bacteria, fungal or viral infection include airborne, feacal-oral spread, vector borne and direct spread either through person to person contact or by direct inoculation. Different microbiological reports have supported the view that barbers are contributing to the spread of infectious diseases and allergic conditions including scabies, ringworm infection and dermatitis (Zahraoui *et al*., 2004).

Hair is one of the structures with a protein filament that grows from follicles found in the dermis, or skin in a body of human being. Hair is one among the defining characteristics of mammals that they possess. Hair strand is anatomically made up of the medulla, cortex, and cuticle, which appear to be different from every organism by having its own specific characteristics that determine the length of the hair. The hair found on the head serves as primary sources of heat insulation and cooling as well as protection from ultra-violet radiation exposure from the atmosphere or any other heat source (Evbuomwan *et al*., 2019). Barbershops are used by both men and women for dressing and trimming of hair in order to make them look neat and

beautiful. For men almost each week they attend barbershops for hair and beard trimming as hygienic measure. There is no enough data for those who attend barbershops per week/month as clients because it depends on the areas and nature of those clients per time. The human hair shaft is keratinized fibrous tissue that grows from follicles beyond the surface of the epidermis. Hair plays a key role in body temperature regulation, defense, protection from the environment, and aesthetics, as well as acting as a sensory organ. Human skin nurtures an estimated one million bacteria per square centimeter of skin, and the scalp houses miscellaneous common scalp commensal microflora (Chambers, 2001). Scalp disorders including folliculitis types, fungal diseases, dandruff, and folliculitis decal-vans, among others, are caused by or linked to microbes, which play a key role in disease predisposition and pathogenesis (Cosgrove *et al.,* 2009).

Barbers are important professionals in the society and in most cases, barbershops are owned, cared and financed by individual members of the society. Barbers' shops are classified as personal services establishments and such services may pose potential health concerns to their clients including the risk of infection and sometimes injury (Croxen *et al*., 2013). It is a demand on their profession to utilize instruments such as knife, blades, clippers which makes it necessary to evaluate health hazards relating to their profession and practices and to identify professional practices linked with infection transmission (Yvonne *et al*., 2005). Microorganisms are everywhere including skin surfaces and are continually introduced into the environment and could therefore easily spread between clients and operators and transferred by contact with unwashed hands, soiled equipment or contact with blood and other body substances (Eckburg *et al*., 2005). These health risks vary depending on the nature of the service, the tools and equipment that are used, the health status of the clients and service providers as well as the

infection control procedures. Therefore, the equipment used may clearly be associated with bacterial, viral and fungal organisms posing infection risks (Okonkwo *et al*.,2019).

A significant proportion of population is enjoying the services of barbers in the society and their shops and professional practices may be a transmission of various infections-directly or indirectly and some bacterial infections can occur without breaking the skin and for this reason equipment must be cleaned between each client (Mobley *et al*., 2004). The person at risk may be the next client on whom the contaminated instrument is used. Organisms that can cause potentially serious infections may be transmitted where appropriate precautions are not taken. It is believed that any service with the potential to break the skin's surface can be associated with infections which can then be transmitted to and between clients if proper infection control procedures are not implemented. Unfortunately, there are no established regulations, guidelines and best practices for many of these salons in our environment, Factors that led to microbial contamination in various studies have been revealed to be source of microbial contamination of hair dressing tools like limited knowledge on using sterilizing machines as most of them do not know, time of using sterilizers before and after attending the client as many of them sterilize only when they attend a client.

Hygiene and sanitation amplify an achievable pre-requisite for worker's safety in any occupational setting including barbershops. Personal and client hygiene practices should be taken as an important step in prevention of health and safety problems, which may be due to contamination in equipment used. However, ongoing unsafe or unhygienic practices as observed in daily attending barbershops in other studies may also be occurring in barbershops and hairdressing salons in Ugbowo, which may affect the health of both the customers and the

workers due to contamination (Aliye *et al*., 2009). For instance, for a procedure which might incidentally lead to piercing of the skin in processes such as cutting, manicure, pedicure, and shaving if not well managed properly may transmit bacterial, fungal and viral infections including HIV, Hepatitis B and Hepatitis C to workers, but also to their customers (Amodio *et al*., 2010). There is little information regarding microbial contamination of barbershop implements as limited studies have been done in Ugbowo and hence paucity of documented information on microbial contamination. There is a need to establish the microbial contamination, prevalence in barbershop routinely used tools like combs, brushes, and shaving sticks due to the fact that the findings obtained will help to improve public health. This study was therefore undertaken to shed more light on the extent of the problem, on factors that perpetuate bacterial and fungal contamination of the implements, as well as antimicrobial sensitivity pattern of these isolates.

Numerous factors have been associated with microbial contamination in barbering equipment, the factors affecting their presence, and the potential health risk they pose. These includes lack of practical knowledge on machine use, use of single tool for all clients attending the barbershop and poor hygiene and sanitation skills of the equipment (Richard *et al*., 2014). Among the hygienic practices they are supposed to adhere to include hand washing before and after attending the client, cleaning and sterilization of the machine and its accessories, use of single hair trimmer per customer, proper razor blade disposal, use of appropriate sterilization methods such as antiseptic solution, ultraviolet radiation, and use of 70% alcohol. Adequate and effective killing of infective microorganisms is reported to be achieved by allowing the tools to be exposed for a long time in a set temperature or medium of sterilization as failure to adhere to the

operational manual predisposes the barbershop customers to the risks of microbial contamination through equipment (Stanley *et al.,* 2019).

The isolation and identification of bacteria and fungi from barbing equipment is essential for several reasons. Firstly, it helps to understand the types and prevalence of microorganisms present on these tools. This knowledge can assist in the development of proper cleaning and sterilization protocols to eliminate or reduce microbial contamination.

Secondly, identifying specific bacterial and fungal species can provide insights into their potential pathogenicity and the associated health risks. Some microorganisms are known to be opportunistic pathogens, meaning they can cause infections in susceptible individuals. By determining the presence of these organisms, appropriate preventive measures can be taken to protect both the barbers and their clients.

Furthermore, the study can help in identifying any antibiotic resistance patterns exhibited by the isolated bacteria. Antibiotic resistance is a growing concern globally, and understanding the resistance profiles of microorganisms found on barbing equipment can aid in the selection of appropriate antimicrobial agents for treatment purposes. Since there is worldwide establishment of barbering industry controlled by persons with little or no knowledge on infection control practices, this has threatened the prevention and management of these fungal infections. Therefore, it has become imperative to explore alternative, cheap and effective means in the management and prevention of these infections. This work aimed at isolating the possible fungal species responsible for causing contamination of barbing tools in Ugbowo and also highlights the possible diseases which the masses are exposed to and to reveal a standard sterilization/disinfection technique, as well as to make necessary recommendations based on the

findings. However, further research is needed to optimize the relevance of sterilization and disinfection of barbering tools.

1.1. AIM OF STUDY

This project aimed to explore the microorganisms frequently associated with grooming tools and their potential impact on public health.

1.2 OBJECTIVES

The specific objectives of this research were to:

a. isolate and identify bacteria and fungi from clippers, combs and shaving sticks.

b. determine the total bacterial and fungal load.

c. carry out antibiotic sensitivity testing on the bacterial isolate

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 ORIGIN OF BARBING

The term barber is derived from the Latin word 'barba' meaning beard. A person, whose occupation is hair cutting, shaving and trimming of beards is called Barber (kondo *et al*., 2006). A barber differs from a hairdresser whose business is generally restricted to cutting and styling hair. Barbering is an ancient profession. The earliest records of barbers show that they were the foremost men of their tribe. They were the medicine men and the priests. They were also providing services for bloodletting, tooth drawing, cauterization, and the tonsorial operations. 5 With the advancement of medicine, surgery and dentistry, the barbers became less and less capable of performing the triple functions of barber-surgeon-dentist. In Pakistan, barbering operations include hair cutting, face and scalp massaging, nail trimming, pedicure, manicure and shampooing/dying of hair. In addition, barbers are also providing facilities for circumcision and incision /drainage of abscess especially in rural areas and urban slums. A Salon is a place where professional hairstylists provide services such as haircuts, hair styling, coloring and treatments. These salons cater to both men and women and are common in many cities and towns (Barn and Chen, 2011).

A significant population of the community enjoys the service of barbers, their shops and professional practices serve as sources of transmission of various infections directly or indirectly with some bacterial infection occurring without breaking the skin (Salami *et al*., 2006). Although the barbering industry is known for its aesthetic activity, researchers have shown the possibility of customers returning home with one infection or the other despite the aesthetics in

the industry. All individual (male and female) have approximately 300,000 hairs on their scalp with a growth rate of approximately half an inch per month (Elewski, 2000). Depending on the type of hairstyle people keep, some visit the barber's monthly, some weekly, some twice a week and some daily to have a haircut. Salons are personal service establishments that provide services which may present potential health concerns to their client including the risk of infection (caused by microorganisms) and sometimes injury (Adeleye and Osidipo, 2004). Although the barber's is supposed to be a safe place to cut and treat your hair, it could also be a nasty, secret germ factory. You may go there germ-free and return with an infection or infections. These health risks may vary depending on the kind of the service, the tool and equipment that are used, physiological state of the client and the kind of service provided (Stout *et al.,* 2011) which can be transmitted between clients if proper infection control procedures are not implemented. Since barbering practices involve the use of equipment such as clipper and razor, they may pierce the skin accidentally and transmit pathogenic microorganisms as well as causing injury. The blood and other body fluids do not have to be visible on instruments, equipment or working surfaces before infections can be transmitted.

2.2 ROLE OF BARBERSHOPS IN TRANSMISSION OF HAIR AND SKIN INFECTIONS

Infection can be transmitted accidentally by any means through blood contact during haircut and shaving in barbershops and therefore, it is postulated that those who work in barbershops may play an important role in the spread or control of infections (Omar *et al.,* 2014). Microorganisms including bacteria and fungi have the potential to colonize surfaces of any material including

equipment and skin surface and sharing of such contaminated implements in barbershops can spread the microorganisms between clients. Infection can occur during hair dressing or trimming since implements like razors, scissors, combs and clippers can accidentally penetrate and pierce the skin. Fluids from parts of the body including blood and sweats may not be visible or recognizable on instruments, equipment or working surfaces (Coulibaly, 2015). There are several infections that can be spread in barbershops and they include infections on the scalp, face and neck such as impetigo and fungal infections. There is a possibility of skin to be burnt during hair dressing or trimming by accidental use of hot tongs, clippers, driers and steamers. If the skin is burnt the protective effect of its surface is destroyed become easily infected by contaminants such as bacteria and fungi and this may result into health complications like skin diseases. For that reason, any client who will come to shave in that barbershop and get in contact with any shared equipment will be at risk of being contaminated with microbes (Uslu *et al.,* 2008)).

It has been discovered that some local barbers' shop has no autoclave or oven or UV sterilizers, and 70% alcohol is the main disinfectant used. However, since most users are not aware of how to use these substances, in which provision of continuing trainings on this issue seems essential. The same studies have been done regionally including countries like Nigeria which came with results of microbial contamination of hair dressing tools which was due to unhygienic procedures in barbershops (Spengane, 2018).) Hairdressing salon and barbershops are in higher risk of microbial contamination because of procedures undertaken by barbers, like hair shampooing, waving or chemical application and the rinsing of these chemicals. Human hairs may function as an air-collecting agent for microbial contaminants, because the hairs are constantly exposed to air and can readily adsorb a variety of airborne particles via electrostatic attraction, grooved surfaces, thin and long structures, and biochemical affinity. It is often that, most of practices

undertaken by barbers' results in their repeated exposure to microbial contamination. To be contaminated by microbes sometimes cannot be seen by our eyes as it need body reactions to reveal it. Also, contamination can be by visible organisms like lice or any parasites (Evbuomwan, 2019).

2.3 SALON HYGIENE

Salons tend to use chemical and physical methods to sterilize tools and equipment, such as chemical reagents, boiling water, autoclaving, chemical reagents, quartz bead sterilization, and UV light (Sources of Salon Hygiene, Proper Practices for Sanitation, Disinfection, and Sterilization). The method of using hot water sterilization involves placing the tools in boiling water for a period of no less than 3–5 min. While this method is relatively fast, it has been shown to be insufficient for killing all microorganisms. Autoclaving is the most reliable way to kill all microbes, although it is not ideal for sterilizing electrical equipment, and the process of sterilization requires a relatively long time. Chemical disinfectants are effective in killing microbes or at least slowing their growth; however, most of these chemicals are hazardous and need to be used with caution (Sources of Proper Sanitation Protocols Can Make or Break Your Salon). Currently, most salons use herbicide solution as a disinfectant, which is highly toxic to humans (Sources of Salon Hygiene, Proper Practices for Sanitation, Disinfection, and Sterilization).

Some Salons tend to use chemical and physical methods to sterilize tools and equipment, such as chemical reagents, boiling water, autoclaving, quartz bead sterilization, and UV light. The method of using hot water sterilization involves placing the tools in boiling water for a period of no less than 3–5 min. While this method is relatively fast, it has been shown to be insufficient for killing all microorganisms (Behravan *et al.,* 2005). Auto-claving is the most reliable way to kill

all microbes, although it is not ideal for sterilizing electrical equipment, and the process of sterilization requires a relatively long time. Chemical disinfectants are effective in killing microbes or at least slowing their growth; however, most of these chemicals are hazardous and need to be used with caution.

Currently, most salons use herbicide solution as a disinfectant, which is highly toxic to humans (Giacomel *et al*., 2015). Some salons use a quartz sterilizer or a UV sterilizer. The quartz sterilizer uses a process that consists of heating air through quartz beads for a period of 5–15 s at 250 °C (482 °F). This type of sterilizer accommodates a large number of tools, but the shape and structure of the tools may change after a period of time. The UV sterilizer is the safest method for tools and brushes that cannot be sterilized at high temperatures; the only disadvantage is that it accommodates very few tools (Draelos, 2001). After cleaning and sterilizing, the tools must be stored properly in cool, dry places to reduce the risk of any growth of bacteria and fungi remaining on tools and therefore, the spread of disease. Brushes should be stored in makeup bags and metal and wooden tools should be stored in closed plastic containers (Tharmila *et al*., 2012). About 35% of service providers use UV sterilization and about 20% use quartz beads to sanitize their tools. However, the use of an ultrasonic cleaner was reported by only 1% of providers. None of these described methods have been proven adequate in reaching a satisfactory level of sterilization by themselves, and a combination of approaches should be used (Anelich and Korsten, 1996).

2.4 SHAVING AND THE RISK OF MICROBIAL INFECTIONS

Shaving is a common grooming practice for many individuals, helping them maintain a clean and polished appearance. Whether shaving facial hair, underarms, legs, or any other body part, it is essential to pay attention to hygiene and safety to prevent potential infections (Buffoli *et al*., 2014). While shaving sticks, razors, and other tools are convenient and widely used, improper shaving techniques can lead to skin irritation, cuts, and an increased risk of infections. Shaving sticks, also known as disposable razors, are a popular choice due to their convenience and ease of use. However, they can pose certain risks if not handled correctly. The sharp blades of shaving sticks can cause micro-cuts and scrapes on the skin's surface, creating entry points for bacteria and other pathogens. This, in turn, increases the likelihood of infections in the shaved areas, especially if proper hygiene is not maintained (Marks *et al*., 2020).

2.4.1. Common Infections from Shaving Stick.

Folliculitis: One of the most common infections associated with shaving is folliculitis. It occurs when hair follicles become inflamed due to bacterial or fungal infection. Folliculitis can cause red bumps, itchiness, and discomfort in the shaved areas.

Cellulitis: If bacteria enter the small cuts or nicks on the skin while shaving, it can lead to cellulitis. This infection affects the deeper layers of the skin and can cause redness, swelling, warmth, and pain.

Boils: Boils are painful, pus-filled bumps that can develop at the site of infected hair follicles. They can be caused by Staphylococcus bacteria entering the skin through shaving cuts.

Herpes Simplex Virus (HSV) Infection: Shaving around the mouth area can potentially lead to HSV infection if there is an active herpes sore present. The virus can enter the tiny cuts during shaving, resulting in painful and recurrent cold sores.

Fungal Infections: Fungi, such as Candida, can thrive in warm and moist environments, like the shaved areas of the body. Improper shaving practices may lead to an increased risk of fungal infections (Bridgeman, 2004).

2.4.2. Preventing Infections from Shaving Sticks

To reduce the risk of infections associated with shaving, consider the following preventive measures:

Preparation: Before shaving, cleanse the area with mild soap and warm water to remove dirt and bacteria. Soften the hair with warm water or a warm towel to make shaving easier and minimize skin irritation.

Use a Clean and Sharp Razor: Always use a clean and sharp shaving stick or razor. Discard disposable razors after a few uses to prevent the accumulation of bacteria on the blades.

Shaving Cream or Gel: Apply a quality shaving cream or gel to create a protective barrier between the razor and the skin. This will help the razor glide smoothly and reduce the likelihood of cuts.

Shaving Technique: Shave in the direction of hair growth to minimize irritation and reduce the risk of ingrown hairs. Avoid applying excessive pressure, as this can lead to skin abrasions.

Aftercare: After shaving, rinse the area with cool water to close the pores, and pat the skin dry with a clean towel. Apply a soothing and moisturizing lotion to keep the skin hydrated and aid in the healing process.

2.5 MICRO ORGANISMS ASSOCIATED/ISOLATED FROM SALON TOOLS.

Many bacterial and fungi species live as normal flora of the skin and mucous membranes of humans as the human skin nurtures an estimated one million per square centimeter of skin (Grimalt, 2007) and the scalp houses miscellaneous common scalp commensal microflora. The microbial pathogens of focus in this study are those bacteria and fungi that are associated with the skin conditions. This are pathogens that exist when the barber fails to disinfect or sterilize the machine prior to and after shaving can cause skin infections. Microorganisms associated/ isolated, include *staphylococcus aureus, Streptococcus pyrogene, Escherichia coli, Staphylococus epidermis, Klebsiella pneumonia, Pseudomonas aeruginosa, Aspergillus* sp., *Micrococcus* sp., *Candida* sp., *Penicillum* sp. and *Trichophyton spp* (Janmohammadi *et al*., 2015).

2.5.1 *Staphylococcus aureus*

Staphylococcus aureus is a Gram-positive spherically shaped type of bacterium and a member of the 'Bacillota' or 'The Bacillus Family'. *Staphylococcus aureus* is a commonly found on the skin and mucous membranes of humans. It is often positive for catalase and nitrate reduction and is a facultative anaerobe that can grow without the need for oxygen (Masalha *et al*.,2001). Although *Staphylococcus aureus* usually acts as a commensal of the human microbiota, it can also become an opportunistic pathogen, being a common cause of various infections, including skin and hair infections.(Tong *et al*., 2015).Pathogenic strains often promote infections by producing virulence factors such as potent protein toxins, and the expression of a cell-surface protein that binds and inactivates antibodies. When the skin's natural defense barrier is

compromised, *Staphylococcus aureus* can enter through cuts, scrapes, or hair follicles, leading to infection (Schlecht *et al*., 2015).

Skin infections caused by *Staphylococcus aureus* can range from minor conditions like folliculitis (inflammation of hair follicles) and impetigo (a contagious skin infection), boils to more serious ones such as cellulitis (infection of deeper skin layers) and abscesses (collections of pus). The bacterium's ability to produce toxins and enzymes contributes to its pathogenicity, causing tissue damage and inflammation. Methicillin-resistant *Staphylococcus aureus* (MRSA) strains are particularly concerning due to their resistance to many common antibiotics, making treatment more challenging (Lindsay, 2010). *Staphylococcus aureus* relating to barbing tools in the context of infections that may occur when clippers, combs and other barbing equipment are not properly cleaned and sanitized between uses. Clipping or trimming hair with unclean or contaminated clippers can transfer Staphylococcus aureus bacteria from one person to another, potentially leading to skin infections or other health issues. To prevent such infections, it is essential to maintain good hygiene practices with clippers, especially in professional settings like barbershops or salons. Proper cleaning and disinfection of clippers after each use can help reduce the risk of bacterial transmission (Ajuzie and Osaghae, 2011).

2.5.2 *Staphylococcus epidermitis*

Staphylococcus epidermidis is a coagulase-negative, gram-positive cocci bacteria that form clusters. It is also a catalase-positive and facultative anaerobe. They are the most common coagulase-negative *Staphylococcus* species that live on the human skin. In its natural environments such as the human skin or mucosa, they are usually harmless (Cheung and Otto, 2010). The belief is that *Staphylococcus epidermidis* is one of the most common causes of

nosocomial infection, with infection rates as high as those of *Staphylococcus aureus (*Lax and Gilbert, 2015). In relations to barbing practices, when it enters the body through contaminated barbering tools, can potentially cause skin and hair infections. Barbering tools such as razors, scissors, and clippers can harbor bacteria if not properly cleaned and sterilized between uses. When these tools come into contact with the skin, they may introduce Staphylococcus epidermidis into the hair follicles or small cuts and nicks on the skin's surface. Once the bacteria enter the body, they can multiply and cause infections (Kleinschmidt *et al*., 2015). Staphylococcus epidermidis is generally considered less virulent than other staphylococcal species, such as Staphylococcus aureus, which is more commonly associated with skin infections. However, in certain circumstances, *Staphylococcus epidermidis* can still cause infections. Common skin/ scalp infections cause by *Staphylococcus epidermis* include; folliculitis, impetigo, cellulitis, abscesses and furuncles.

2.6. INFECTIONS TRANSMITTED THROUGH BARBER'S TOOLS

Contagious infectious diseases of the skin and blood have been found to be associated with barbering (Ryan and Ray, 2004).

2.6.0. Barber's itch (Folliculitis barbae*)*

Barber's itch, also known as sycosis barbae or folliculitis barbae, is a common and bothersome skin condition that affects the hair follicles in the beard and neck area. It is primarily caused by a bacterial infection, with Staphylococcus aureus being the most common culprit, although other bacteria like Staphylococcus epidermidis or fungi can also be responsible. The condition typically arises in men who have dense, coarse facial hair and regularly shave. The act of shaving can create tiny nicks and cuts in the skin, providing an entry point for bacteria to invade the hair

follicles, leading to inflammation and infection. Pseudo folliculitis Barbae is a related condition caused by irritation from shaving and ingrown hairs, it occurs when hair curls back into the skin after shaving, causing inflammation, redness and bumps. This can lead to scarring and skin discoloration. *Pseudo folliculitis nuchae*, is also a related condition, occurs on the back of the neck, often along the posterior hairline, when curved hairs are cut short and allowed to grow back into the skin. Left untreated, this can develop into ‘acne keloidalis nuchae’ a condition in which hard, dark keloid-like bumps form on the neck (Alexander, 1974). Additionally, the use of unclean or dull razors can exacerbate the problem by introducing more bacteria to the already vulnerable skin. Symptoms of barber's itch are not only uncomfortable but can also be embarrassing and confidence-shattering for those affected. The most common symptoms include; Red, inflamed, or swollen bumps in the beard and neck area: These bumps, called papules, may be small or larger and can be itchy and painful, Pustules or pus-filled lesions: As the infection progresses, some of the papules may develop into pustules, which can look unsightly and may discharge pus when squeezed, Itching and discomfort: The affected area may feel itchy and uncomfortable, which can be distracting and irritating, Pain or tenderness in the affected area: The inflamed hair follicles can be tender to touch and may cause discomfort, especially during shaving (McLean, 2004).

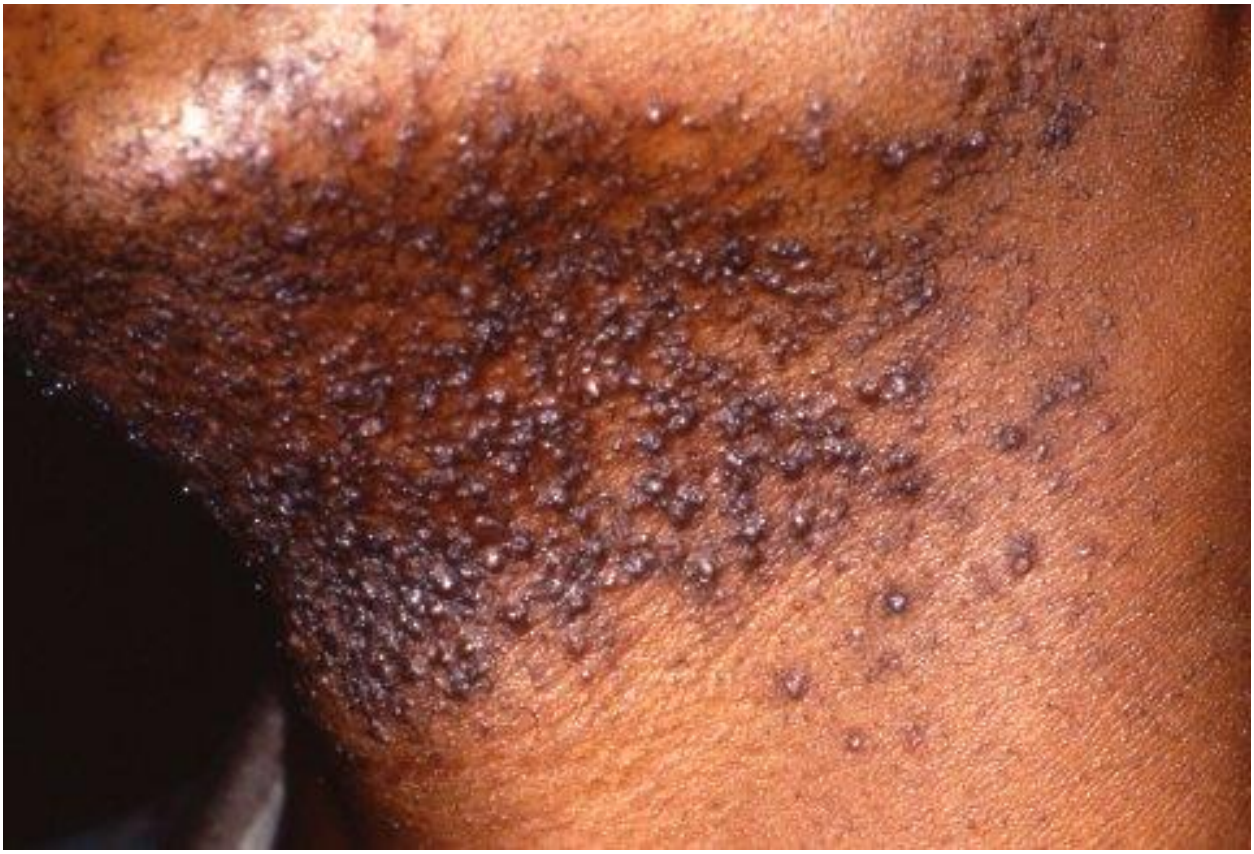


Fig 1: A severe case of Pseudo Folliculitis (Adebola, 2019*).*

2.6.1. Treatment of barber's itch (FOLLICULITIS BARBAE)

The simplest treatment for PFB is to let the beard grow. Complete removal of the hair from its follicle (epilation) is not recommended. Severe or transfollicular hairs may require removal by a dermatologist. For most cases, completely avoiding shaving for three to four weeks allows all lesions to subside, and most extrafollicular hairs will resolve themselves within at least ten days. Medications may also be prescribed to speed healing of the skin. Clinical trials have shown glycolic acid-based peels to be an effective and well-tolerated therapy which resulted in significantly fewer PFB lesions on the face and neck (Daniel *et al*., 2013).

The mechanism of action of glycolic acid is unknown, but it is hypothesized that straighter hair growth is caused by the reduction of sulfyldryl-bond in the hair shaft by glycolic acid, which results in reduced re-entry of the hair shaft into the follicular wall or epidermis. Salicylic acid peels are also effective. Prescription antibiotic gels (Benzamycin, Cleocin-T) or oral antibiotics are also used. Benzoyl peroxide may be used topically, combined or not with prescription antibiotics. Tretinoin is a potent treatment that helps even out any scarring after a few months. It is added as a nightly application of tretinoin cream 0.05–0.1% to the beard skin while beard is growing out. Tea tree oil, witch hazel, and hydrocortisone are also noted as possible treatments and remedies for razor bumps (Gray *et al*., 2016).

2.6.2. Impetigo

Impetigo is a contagious bacterial skin infection that primarily affects young children but can occur in people of all ages. It is characterized by red sores or blisters that can break open, ooze fluid, and develop a yellow-brown crust. Impetigo is caused by bacteria, usually *Staphylococcus*

aureus (staph) or *Streptococcus pyogenes* (strep) (Breyre and Frazee, 2018). It is commonly known as 'school sores' because a majority of cases are in school-aged children. *Staphylococcus* bacteria live on the skin and are mostly harmless, but they can cause an infection if they enter damaged skin. These bacteria can get into your body through a break in the skin from a cut, scratch, insect bite, or rash. Then they can invade and colonize. Impetigo is more common in areas where the skin or scalp have been cut or are abraded. It is also more common in younger children but you can get it at any age and it is most commonly spread via skin-to-skin contact, clothing, or towels, something to keep in mind if you're getting your hair washed at a barber's or hairdressers (Castro and Ramos, 2018). There are two main types of impetigo:

2.6.2.1. Non-bullous impetigo.

This is the most common form. It starts as red sores around the nose and mouth that quickly rupture and form honey-colored crusts. The sores can also appear on other areas of the body. Non-bullous impetigo often starts as a vesicle or a pustule. Multiple vesicles often coalesce and rupture after which the purulent exudate forms the characteristic honey-colored crust. An erythematous base is also present (Benedette, 1993). There are often multiple lesions on the face and extremities, especially in areas in which disruption of the skin barrier has occurred. The rapid spread and satellite lesion formation follow self-inoculation, often in areas with no apparent break in the skin barrier. Mild regional lymphadenopathy is a common associated finding. Systemic symptoms such as fever are typically absent in non-bullous impetigo (Dayrit *et al*., 2018).

2.6.2.2. Bullous impetigo

This form is less common but more severe. It involves the appearance of larger fluid-filled blisters, which are usually painless and occur on the trunk, arms, and legs. Bullous impetigo begins with small vesicles that become flaccid bullae. The exfoliative toxin A produced by *S. aureus* causes loss of cell adhesion in the superficial epidermis. The bullae contain a clear or yellow fluid which eventually progresses to become purulent or dark. Surrounding erythema and edema are typically absent. Once the bullae rupture, an erythematous base with a rim of scale remains. Bullous impetigo does not form a honey-colored crust. Lesions most commonly form in the intertriginous regions and on the trunk and, unlike non-bullous impetigo, may occur in the buccal membranes. There are typically fewer lesions present than in non-bullous impetigo. Regional lymphadenopathy is absent. Systemic symptoms, such as fever, are more common than in non-bullous impetigo (Sahu *et al*., 2019).

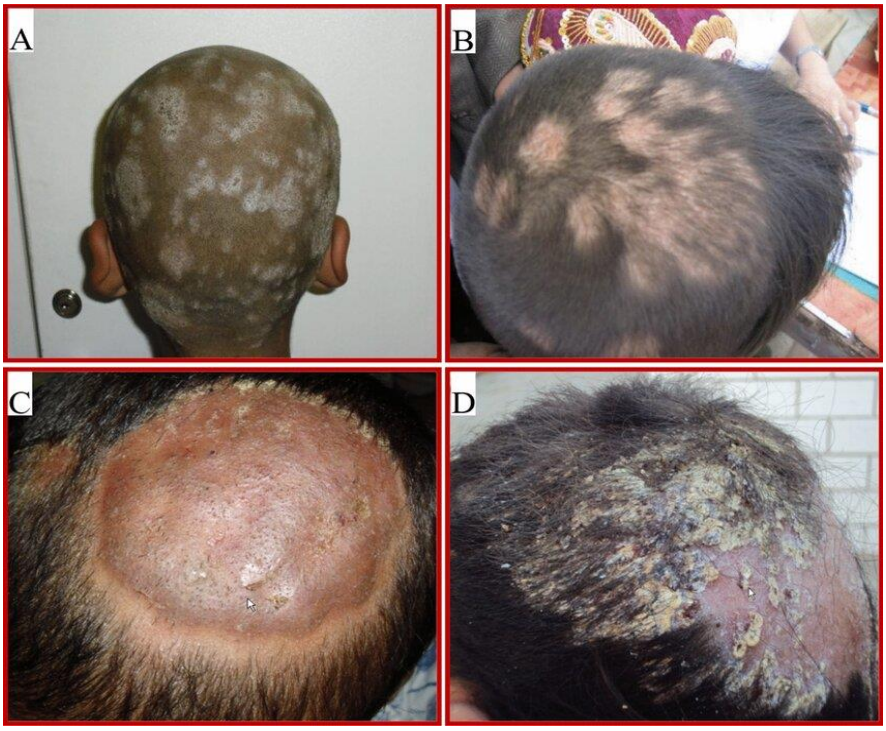
Impetigo is an infection that can potentially be transmitted through unclean barbing tools. Barbershops and hair salons use a variety of tools, such as razors, scissors, clippers, and combs, to provide haircuts and grooming services. If proper hygiene and sanitation practices are not followed, these tools can become contaminated with bacteria, including the ones responsible for impetigo.

2.6.3. Tinea capitis

Tinea capitis is a fungal infection that affects the scalp and hair. It can take the shape of ringworm (red patch with scale around the perimeter) or it can look like a red flakey itchy patch. The infection is caused by *Trichophyton, Microsporum* and *Epidermophyton* species *of* dermatophytes (Moto *et al.*,2015). Dermatophytes are also known as ‘ringworm’, fungi. The

name ringworm has been in use at least from the sixteenth century and it was carried to describe the circular lesion produced by dermatophyte on the skin or scalp. Ringworm is clinically referred to as Tinea and locations involved are usually the surface of the body (Tinea Copons), of the groin (Tinea Gruris) of the bearded area (Tinea Barbae), of the scalp (Tinea capitis) The dermatophytes invade hair follicles and keratinized layer of hairy skin leading to scaling, kerion, hair loss, folliculitis, favus and black dot. The infection could be anthropophilic, zoophilic or geophilic (Oguzkaya *et al*.,2013). The infection occurs in both sexes with high infection rates among males, it is predominant in children. Tinea capitis is of public health importance but not a notifiable disease therefore little is known on its prevalence in many endemic areas. Tinea capitis can be transmitted via infected persons, animal vectors and fallen infected hairs. Spread of tinea capitis by formites (contaminated barbershop instruments, hairbrushes, combs and shared hats or towels) is common (Elewski 2000). Factors affecting disease transmission are personal hygiene, overcrowding and low socioeconomic status. Shed hairs may harbour infectious fungal agents up to a year. Eradication is difficult due to common asymptomatic carriers (Pai *et al.,*2013). Barbering operations are still under little or no scrutiny despite the possible risk of spreading infectious diseases The barbers use clippers, blades, hair brushes etc. which are unsterilized and when use it on barbing the hair of individuals many develop different types of problems on their hair which Tinea Capitis is one of them many children go with their head white as if they have grey hair due to ringworm actions. Sometimes after barbing individual hair without sterilizing or changing the blade they use it on another person thereby spreading the infection from one person to another. Therefore, the problem lies at the unhygienic practice by the barbers before and while doing barbing work (Ayanbimpe *et al*., 2008). Tinea capitis often needs to be treated with an oral

anti-fungal medication. The fungus penetrates deep into the hair follicles, so it may be difficult to treat with topical medications alone.



(Xiadong *et al*., 2023)

Fig 2: A severe case of Tinea Capitis.

A; Tinea Capitis, “White tinea”, localized patch, multiple scaly lesions (“gray-patch”), stubs of broken hair, caused by *Trichophyton tonsurans.* B; Tinea capitis. “Black dot”ringworm caused by *Trichophyton tonsurans*, presents as multiple areas of alopecia studded with black dots. C; Kerion celsii, Inflammatory reaction of tinea capitis caused by *Microsporum canis*. Kerion may be followed by scarring and permanent alopecia in the areas of inflammation and suppuration. D; Favus. Favus of scalp, showing scutulae, caused by *Trichophyton schoenleinii.*

2.6.4. Head lice

Lice are small, parasitic insects that infest the hair and skin of humans and some animals. There are different types of lice, such as head lice, body lice, and pubic lice. Head lice are tiny parasitic insects that live on the human scalp and hair (Reed, 2007). Human head and body lice occupy distinct ecological zones: head lice live and feed on the scalp, while body lice live on clothing and feed on the body. Because body lice require clothing to survive, the divergence of head and body lice from their common ancestor provides an estimate of the date of introduction of clothing in human evolutionary history (Kittler, 2003). They feed on blood and attach their eggs (nits) near the base of the hair shafts, close to the scalp. It is common in the scalp, but it also can occur in the beard. Lice and nits are usually challenging to see with the naked eye, as they are very small and move quickly. Head lice spread primarily through direct head-to-head contact. It's common in places where people are in close proximity, such as schools, daycare centers, camps, and households with multiple family members. Lice can't jump or fly; they crawl from one person's hair to another when their heads are close together or when they share combs and clippers. Common signs of head lice infestation include; persistent itching of the scalp, tickling or crawling sensations on the head, Small red bumps or sores on the scalps, necks or shoulders, presence of nits, which look like tiny, oval-shaped, yellowish-specks attached to the hair shafts near the scalp, live lice moving on the scalp or hair (Light *et al.,* 2008).

Lice are common in the scalp, but they can also occur in the beard. The most common symptom is significant itching in the affected area. In addition to the adult louse, it is common to find nits, which are eggs, in the hair as well. A mediaeval treatment for lice was an ointment made from pork grease, incense, lead and aloe (Kowalski and Agger, 2009).

2.7. PREVENTION AND CONTROL

Preventing and controlling microbial infections related to shaving sticks, clippers, and combs in barbershops or personal grooming settings is crucial to ensure the safety and well-being of clients (Mbajiuka *et al.,* 2014). Here are some essential preventive measures and control strategies:

2.7.1. Regular cleaning and disinfection:

Ensure that all shaving sticks, clippers, and combs are thoroughly cleaned and disinfected between each use. Use soap and water to properly clean tools to remove debris, hair, and other contaminant and also use hospital-grade disinfectants to eliminate bacteria, viruses, and fungi effectively.

2.7.2. Single-use items:

Whenever possible, use disposable items like single-use razors or blades to avoid cross-contamination. These should be discarded after each use. This measure is particularly useful for minimizing the risk of infections from one client to another.

2.7.3. Sterilization of tools:

Certain tools, like scissors or metal combs, can be sterilized using heat or chemical methods to kill all microbial organisms effectively. Regularly perform sterilization of reusable tools according to industry standards. Sterilization is a more advanced cleaning measure and is especially important for tools that cannot be disposed of after use

2.7.4. Personal protective equipment (PPE):

Barbers and hairstylists should wear disposable gloves when providing services to clients to prevent direct contact with potentially infectious materials. Replace gloves between clients to

avoid cross-contamination. PPE ensures that barbers and stylists are protected from potential infections.

2.7.5. Hand hygiene:

Hand hygiene is a simple yet crucial measure to prevent the transfer of microbes. Barbers should practice regular handwashing with soap and water or use alcohol-based hand sanitizers before and after each client interaction.

2.7.6. Proper storage:

Store clean and disinfected tools in a clean, dry, and covered container or drawer to protect them from contamination. Proper storage ensures that the tools remain sanitized and ready for use.

2.7.7. Client exclusion:

If a client has visible signs of an infectious condition or skin infection, it is best to avoid providing services until the condition has cleared to prevent spreading the infection to other clients. Client exclusion protects other clients from potential exposure to infections.

2.7.8. Regular training:

Ensure that barbers and salon staff receive proper training in infection control practices, including cleaning, disinfection, and sterilization procedures. Regular training reinforces best practices and ensures consistent application.

2.7.9. Disinfection of work surfaces:

Regularly clean and disinfect workstations, chairs, and other surfaces that come into contact with clients. Disinfecting work surfaces helps minimize the risk of surface-to-person transmission.

2.7.10. Maintaining clean capes and towels:

Clean and sanitize capes and towels after each use to prevent the spread of infections. Clean capes and towels are essential for maintaining a hygienic environment.

2.7.11. Monitoring and record keeping:

Monitoring and record-keeping help ensure that proper practices are consistently followed. Maintain records of cleaning, disinfection, and sterilization procedures to ensure accountability and track compliance.

By implementing these preventive measures and control strategies, barbershops and grooming establishments can create a safe and hygienic environment for their clients, reducing the risk of microbial infections and maintaining a high standard of care and professionalism (Ibrahim *et al.,* 2007).

CHAPTER THREE

MATERIALS AND METHODS

3.1 **STUDY AREA**

The study area for this research was Ekososdin. Ekosodin community is situated in the east of Isihor in Ovia North East Local Government Area (LGA) of Edo state. Ovia North-East LGA has its headquarters in Okada town; it has an area of 2,301 square kilometres (Akinbo and Okaka, 2010). It is located along the longitude 5^{o} 45^{1} and 6^{o} 15^{1} east and latitude 5^{o} 15^{1} and 6^{o} 45^{1} north of the central province of Edo state. Ekosodin community has an estimated population of 7000 [21] as estimated by 2006 census by National Population Commission (Ogeah and Ajalaye, 2011). This population was projected by 543.2% using geometric method to year 2022 to be 45,000 people. The Community is very close to the University of Benin (343 metres away). Three barbing salons, three hairdressing salons and three Individuals were randomly selected from different areas in Ekosodin for the sample collection.

3.2 SAMPLE COLLECTION

Three clippers sample was collected from each of the three barbing salons, three comb samples was collected from the three salon each and three used personal shaving sticks was collected from three individuals each. A total of nine samples (9) were collected by rotating a moist swab over the surface of the cutting edge of the clippers, the surfaces of the combs and the razor end of the shaving sticks. Samples were collected in the morning and were then put in sterile tubes

containing I milliliter of sterile distilled water to avoid drying and transported to the laboratory. The name, source, and location were noted on the swab sticks and brought to the laboratory under sterile aseptic conditions for microbiological analysis.

3.3 STERILIZATION OF MATERIALS

Glass wares like petri dishes, glass rods, beakers, and test tubes required for this research were soaked and washed with detergent and rinsed with distilled water. They were covered with cotton wool and wrapped with aluminum foil paper and sterilized in an autoclave in an inverted position at 170^{o}C for 5-10 mins.

3.4 PROCEDURE

3.4.1 Preparation of culture media

The media used were prepared according to the manufacturer's instructions. The media used were Macconkey agar (MAC), potato dextrose agar (PDA) and Nutrient Agar.

3.4.2 Preparation of Macconkey agar

An amount of 51.55 grams of Macconkey agar (MAC) was dissolved in 1000 ml distilled water in a conical flask covered with cotton wool and aluminum foil paper. It was mixed thoroughly and sterilized by autoclaving at 121°C for 15 minutes. The medium was cooled to 45-50°C and then dispensed aseptically into sterile Petri dishes in the laminar flow.

3.4.3 Preparation of Nutrient Agar

Twenty-eight grams (28 g) of nutrient agar (NA) powder was dissolved in 1 liter of distilled water in a conical flask covered with cotton wool and aluminum foil paper. It was mixed thoroughly and sterilized by autoclaving at 121°C for 15 minutes. The medium was cooled to 45-50°C and then dispensed aseptically into sterile Petri dishes in the laminar flow.

3.4.4 Isolation of bacteria

The water sample was transferred to sterile Petri plate. The prepared nutrient agar (for bacteria growth) was poured in aseptically and incubated at 37°C for 24 hrs. After successful growth of microorganisms, the colonies were counted with a colony counter and the results per dilution count were recorded. The number of colony-forming unit per milliliter was calculated with the formula:

$$\text{Cfu/cm}^3 = \frac{number\ of\ colonies}{volume\ plated\ x\ dilution\ factor} \quad \_ \quad \_ \quad \_ \quad \_ \quad \_ \quad \_ \quad \_ \quad \_ \quad \_ \quad (1)$$

3.4.5 Pure culture

One single colony was identified and re-streaked as a primary inoculant on the surface of a nutrient agar plate medium. Pure cultures were checked from nutrient agar plates. After achieving a pure culture, the same colony was streaked onto a nutrient agar slant. These cultures were incubated at 37°C for 24h hours.

3.4.6 Cultural characteristics

Each colony morphology e.g., size, shape, margin, elevation, consistency, color, transparency was determined

3.5 MORPHOLOGICAL TEST

3.5.1 Gram staining

Smears of the bacterial isolates were prepared and heat fixed on clean grease free slides. The smears were stained for one minute with crystal violet. This was washed out with distilled water. The slides were flooded with dilute Grams' iodine solution for one minute. This was washed off with distilled water and the smears were decolorized with 95% alcohol for 30 seconds and rinsed off with distilled water. The smears were then counter stained with safranin solution for one minute. Finally, the slides were washed off with distilled water, air dried and observed under oil immersion objective.

3.5.2 Catalase Test

This is a test to detect the presence or absence of catalase enzyme. The catalase enzyme catalyses the breakdown of hydrogen peroxide to release free oxygen gas and the formation of water. A few drops of freshly prepared 3% hydrogen peroxide were added onto the bacterial isolates smeared on a slide. The production of gas bubble indicated catalase enzyme positive.

$2\ H_2O_2 \rightarrow 2\ H_2O + O_2$

3.5.3 Oxidase Test

A piece of filter paper was wet with a few drops of the dilute (1%) solution of oxidase reagent (tetramethyl-phenylenediamine-dihydrochloride) which was prepared by standard procedure. A bit of growth from the nutrient agar slant was obtained using sterilized platinum wire loop and smeared on the wet piece of paper. Development of an intense purple color by the cells within 30 seconds indicates a positive oxidase test.

3.5.4 Urease Test

The urease test is used to determine the ability of an organism to split urea in the presence of the enzyme urease. The bacterial isolates were inoculated into slants of urea medium and incubated at 370C for 24-48 hours. Urease positive cultures produced a red-pink colour due to changes in the colour of the indicator

$NH_2CO.NH_2 + H_2O \rightarrow 2NH_3 + CO_2$

3.5.5 Citrate Utilization Test

This test is based on the ability of some organisms to utilize citrate as a sole source of carbon. This was carried out by inoculating the test organism in test tube containing Simon's citrate medium and this was incubated at 370C for 24 - 48 hours. The development of deep blue colour after incubation indicates a positive result.

3.5.6 Hydrogen Sulphide (H_2S) Test

Hydrogen sulphide production can be detected by incorporating a heavy metal salt containing (Fe2+) or lead (Pb2+) ion as H_2S indicator to a nutrient culture medium containing cysteine and sodium thiosulfate as the sulphur substrates. Hydrogen sulphide, a colourless gas, when produced reacts with sulphur metal salt (ferrous sulphate) forming a visible insoluble black sulphide precipitate.

3.5.7 Indole Test

Indole test is performed to determine the ability of the organism to split tryptophan molecule into indole. This test is performed to help differentiate species of the family enterobacteriaceae.

Kovac's reagent which contains hydrochloric acid, dimethylaminobenzaldehyde and amyl alcohol is used. Inoculate broth with the test organism and incubate for 18 – 24 hours at 370C. Add 5ml of Kovac's reagent down the inner wall of the tube. Development of bright red colour at the interface of the reagent and the broth within seconds after adding the reagent is indicative of the presence of indole and is a positive test wile absence is negative.

3.5.8 Sugar Fermentation Test

Each of the isolates was tested for its ability to ferment a given sugar with the production of acid and gas or acid only. The growth medium comprised of peptone water, sugar (1%) and the indicator (bromocresol purple). The mixture was dispensed into test tubes and sterilized by autoclaving at 121°C for 15 minutes. After sterilizing, tubes were allowed to cool and then inoculated with the isolates and incubated at 37°C for 24hrs. Acid and gas production or acid only were observed after about 24 hours of incubation. Acid production was indicated by the change of the medium from purple to yellow colour indicated a positive test.

Sugars used are: lactose, sucrose, glucose, fructose, maltose, starch and sorbitol

3.5.9 Pathogenicity

Preparation of Blood Agar

1. Suspend 28 g of nutrient agar powder in 1 litre of distilled water.

2. Autoclave mixture at 121 degrees Celsius for 15 minutes.
3. Once the nutrient agar has been autoclaved, allow it to cool but not solidify.
4. When the agar has cooled to about 45-50 °C, add 5% (vol/vol) sterile defibrinated blood that has been warmed to room temperature and mix gently but well.
5. Avoid Air bubbles while dispensing into sterile plates while it's in molten form.
6. Allow to solidify then streak isolates on agar. Incubate for 24hrs at 37°C.

Interpretation

- Alpha-haemolysis (α) - the media is translucent with a greenish tinge around the colonies. This means is slight damage of the RBCs but absence of lysis.
- Beta-haemolysis (β) - the RBCs have been lysed and the media looks completely transparent around the colonies.
- Gamma-haemolysis (γ) - bacteria exhibit neither lysis nor clearing of any kind on agar plates. It is often referred to as non-haemolytic.

3.5.10 Antibiotic susceptibility test

Test organisms will be subjected to antibiotics sensitivity test using the Kirby Bauer disc diffusion on prepared media. Ten (10) different commercial antibiotic discs will be used. The antibiotic discs will be carefully and firmly placed on the inoculated plates using a sterile pair of forceps. The plates will be inverted and incubated for 37°C for 24 h. The diameter of the zone of inhibition will be measured in millimeters (mm) using a meter rule. The experiments will be carried out in triplicates to minimize probability of error

3.6 FUNGI PROCEDURE

Preparation of potato dextrose agar

An amount of 39.5(g) of potato dextrose agar powder was dissolved in 1 liter of distilled water in a conical flask covered with cotton wool and aluminum foil paper. It was mixed thoroughly and sterilized by autoclaving at 121°C for 15 minutes. The medium was cooled to 45-50°C and before dispensing aseptically into Petri dishes. Plates were then incubated for 72h at 5°C.

Cultural Characteristics

Each colony morphology e.g., size, texture, color, reverse colour, was determined by physical examination.

Pure culture

One single colony was identified and re-streaked as a primary inoculum on the surface of a potato Dextrose agar plate medium to make a pure culture. After achieving a pure culture, the same colony was streaked onto potato dextrose agar slant. These cultures were incubated at 25°C for 72h.

Isolation of Fungi

The sample was diluted serially using sterile distilled water as diluents. The aliquot was transferred aseptically to sterile petri plates.

The prepared agar (for fungi growth) was poured in aseptically and incubated at 28°C for 72 hrs. After successful growth of microorganisms, the colonies were counted with a colony counter and the results per dilution count were recorded. The number of colony forming unit per milliliter was calculated with the formula:

$$\text{Cfu/cm}^3 = \frac{number\ of\ colonies}{volume\ plated\ x\ dilution\ factor} \quad \_ \quad \_ \quad \_ \quad \_ \quad \_ \quad \_ \quad \_ \quad \_ \quad \_ \quad (2)$$

LACTOPHENOL COTTON BLUE MOUNTING OF FUNGI

Lactophenol cotton blue is a stain commonly used for making semi-permanent microscopic preparation of fungi. It stains the fungi cytoplasm and provides a light blue background, against which the wall of hyphae can readily be seen. It contains four constituents: phenol, which serves as fungicides; lactic acid, which act as a clearing agent; cotton blue, which stains the cytoplasm of the fungus; and glycerine, which gives semi- permanent preparation. A permanent preparation may be by incorporating polyvinyl alcohol in place of glycerine into the mounting medium.

For rapid and routine examination of almost all type of fungi, spores and spore bearing structures are tested out on a clean slide in a drop of mounting fluid (lactophenol cotton blue) and a cover-glass placed over the preparation which is then ready for microscopic examination.

PROCEDURE

Place a drop of lactophenol cotton on a clean slide

Transfer a small tuff of fungus, preferably with spore or spore boring structures into the drop on the slide using a flamed cooled needle. Gently tease using an inoculating needle. Mix gently the stains with the mold structures. Place a cover-glass over the preparation taking care to avoid trapping air bubbles in the stain.

OBSERVATION

Examine the preparation under low and high-power objectives.

Describe the type of hyphae, conidiophore cell, conidia and their arrangement on the conidiophore/conidiogenous cells.

Draw a representative microscopic field under low power and high-power magnification. Identify the mold on the basics of characteristics features produced.

The fungal cytoplasm is seen as a lightly stained blue region forming a layer inside the unstained cell wall of hyphae, conidiophores, phialides and conidia, that is conidia, that is surrounded by a light blue background on the slide.

CHAPTER FOUR

RESULTS

4.1 Results

The total heterotrophic bacterial and fungal counts along with growth on MacConkey agar from the clipper and combs and shaving sticks samples are represented in Table 1, 2 and 3 respectively. A dilution factor of 10^{-3} was used for the inoculation and the volume of inoculum plated was 0.5 ml. However, a dilution factor of 10^{-2} was used for bacterial count on MacConkey agar.

Table 4 shows the cultural characteristics of the bacteria isolated from the clipper, comb and shaving stick samples. Morphological characteristics such as colony shape, size, elevation, optical activity, margination and pigmentation on nutrient agar and MacConkey agar were taken into account. The bacteria that were isolated are *Bacillus* sp., *Klebsiella* sp., *Pseudomonas* sp., *Staphylococcus* sp. and *Clostridium* sp. The distribution pattern of the bacterial isolates is represented in Table 5.

Table 6 shows the morphological and biochemical characteristics of the bacteria isolated from the clipper, comb and shaving stick samples. Biochemical tests were carried out to further identify the bacterial isolates and Gram staining was carried out to observe the shape of the bacteria cells and their Gram reaction. The biochemical tests carried out were; Urease test, Indole test, Citrate test, Catalase test, Coagulase test, Oxidase test and Haemolysis test. Sugar fermentation tests, such as lactose, fructose, sucrose and starch hydrolysis tests were carried out on the bacterial isolates.

Table 7 shows the cultural characteristics of the fungi isolated from the clipper, comb and shaving stick samples. Morphological characteristics such as nature of colony, type of spores produced and hyphae structure were observed. The bacteria that were isolated are *Saccharomyces* spp. and *Fusarium* spp. The distribution pattern of the fungal isolates is represented in Table 8.

Antibiotic susceptibility test was carried out to observe the susceptibility of the bacterial isolates to antibiotics. The in vitro antimicrobial assay was carried out using the Kirby Bauer disk diffusion technique. The zones of inhibition were measured in millimeters and compared with standard tables to determine the susceptibility and resistance of the bacterial isolates. The antibiotics used were; Pefloxacin, Gentamycin, Ampiclox, Zinnacef, Amoxacillin, Rocephin, Ciprofloxacin, Streptomycin, Septrin, Erythromycin, Saprifloxacin, Chloramphenicol, Tarivid and Augmentin. The results are represented in Tables 9 and 10.

Table 4.1: Colony count for the clipper samples in CFU/cm^3.

SAMPLE	Heterotrophic bacteria count (NA)	Heterotrophic fungal count (PDA)	Bacteria count (MCA)
Clipper 1	$8.67x10^4 \pm 1.76$	$7.27x10^4 \pm 1.20$	$4.40x10^3 \pm 0.58$
Clipper 2	$8.00x10^3 \pm 0$	$1.4x10^4 \pm 0$	$8.33x10^3 \pm 0.89$
Clipper 3	$1.27x10^4 \pm 0.67$	$1.73x10^4 \pm 0.67$	$1.01x10^4 \pm 0.89$

Values represented as mean ± standard error of mean

KEY:

NA: Nutrient agar

PDA: Potato dextrose agar

MAC: MacConkey agar

Table 4.2: Colony count for the comb samples in CFU/cm^3.

SAMPLE	Heterotrophic bacteria count (NA)	Heterotrophic fungal count (PDA)	Bacteria count (MCA)
Comb 1	$2.00x10^3 \pm 0$	$2.13x10^4 \pm 0.33$	-
Comb 2	$2.00x10^3 \pm 0$	$2.87x10^4 \pm 0.67$	$8.67x10^2 \pm 0.33$
Comb 3	$6.07x10^4 \pm 0.67$	$4.87x10^4 \pm 1.45$	$5.47x10^3 \pm 1.45$

Values represented as mean ± standard error of mean

KEY:

NA: Nutrient agar

PDA: Potato dextrose agar

MAC: MacConkey agar

Table 4.3: Colony count for the shaving stick samples in CFU/cm^3.

SAMPLE	Heterotrophic bacteria count (NA)	Heterotrophic fungal count (PDA)	Bacteria count (MCA)
Shaving stick 1	$2.00x10^3 \pm 0$	$2.00x10^4 \pm 0.58$	-
Shaving stick 2	$8.00x10^3 \pm 0$	$8.67x10^3 \pm 0.33$	$1.04x10^4 \pm 15.68$
Shaving stick 3	$8.00x10^3 \pm 0$	$2.93x10^4 \pm 0.33$	$2.00x10^2 \pm 8.00$

Values represented as mean ± standard error of mean

KEY:

NA: Nutrient agar

PDA: Potato dextrose agar

MAC: MacConkey agar

Table 4.4: Cultural characteristics of the bacterial isolates

Organism	Shape	Size	Elevation	Transparency	Margin	Colour (NA)	Colour (MCA)
Bacillus sp.[1]	Circular	Small	Flat	Opaque	Entire	Cream	-
Bacillus sp.[2]	Irregular	Large	Flat	Opaque	Lobate	Cream	-
Klebsiella sp.[1]	Rhizoid	Medium	Flat	Opaque	Undulate	Cream	-
Bacillus sp.[3]	Irregular	Large	Flat	Opaque	Entire	Cream	-
Pseudomonas sp.	Rhizoid	Large	Flat	Opaque	Lobate	Cream	-
Klebsiella sp.[2]	Circular	Small	Flat	Opaque	Entire	-	Cream
Staphylococcus sp.	Circular	Small	Flat	Opaque	Entire	-	Dark cream
Clostridium sp.	Rhizoid	Medium	Flat	Translucent	Lobate	-	Cream

Table 4.5: Distribution pattern of the bacterial isolates

Source	Clipper 1	Clipper 2	Clipper 3	Comb 1	Comb 2	Comb 3	Shaving stick 1	Shaving stick 2	Shaving stick 2
Bacillus sp.[1]	+	+	-	-	-	-	-	-	-
Bacillus sp.[2]	-	-	+	-	+	-	-	-	-
Klebsiella sp.[1]	-	-	-	-	-	-	-	+	-
Bacillus sp.[3]	-	-	-	-	-	+	-	-	-
Pseudomonas sp.	-	+	-	-	-	-	-	-	+
Klebsiella sp.[2]	-	-	+	-	-	-	+	-	-
Staphylococcus sp.	-	-	-	-	-	+	-	+	-
Clostridium sp.	-	+	-	-	-	-	-	-	-

KEY:

+: Present

-: Absent

Table 4.6: Morphological, biochemical, and sugar tests of the bacterial isolates

	***Bacillus* sp.[1]**	***Bacillus* sp.[2]**	***Klebsiella* sp.[1]**	***Bacillus* sp.[3]**	***Pseudomonas* sp.**	***Klebsiella* sp.[2]**	***Staphylococcus* sp.**	***Clostridium* sp.**
Gram	+	+	-	+	-	-	+	+
Cell type	Rod	Rod	Rod	Rod	Rod	Rod	Cocci	Rod
Arrangement	Cluster	Chains	Single	Chains	Cluster	Cluster	Chains	Single
Urease	-	+	+	-	-	-	+	-
Indole	-	-	-	-	-	-	-	-
Citrate	+	-	+	-	+	+	-	+
Catalase	+	+	-	+	+	+	+	-
H_2S	-	-	-	-	-	-	+	+
Coagulase	-	-	-	-	-	-	-	-
Oxidase	-	-	-	-	+	+	-	-
Lactose	-	-	+	-	-	-	+	-
Sucrose	+	+	+	+	-	-	+	-
Glucose	+	+	+	+	-	+	+	+
Fructose	+	+	+	+	-	+	+	+
Maltose	+	+	+	+	-	-	+	-
Starch	+	+	-	-	-	-	-	-
Sorbitol	+	-	+	+	-	-	-	-
Haemolysis	beta	beta	gamma	beta	beta	gamma	gamma	gamma

KEY:

Gamma: no haemolysis

Beta: full haemolysis

+: Presence

-: Absence

Table 4.7: Fungal cultural characteristics

Isolate	*Saccharomyces* spp.	*Fusarium* spp.
Nature of colony	Cream coloured colonies with entire margins	Wooly white colonies with spores
Nature of hyphae	Pseudohyphae	Septate
Spore type	Chlamydospore	Chlamydospore

Table 4.8: Distribution pattern of the fungi isolates

Source	*Saccharomyces* spp.	*Fusarium* spp.
Clipper 1	-	-
Clipper 2	-	-
Clipper 3	+	-
Comb 1	-	+
Comb 2	+	-
Comb 3		-
Shaving stick 1	+	-
Shaving stick 2	+	-
Shaving stick 3	-	-

KEY:

+: Present

-: Abscent

Table 4.9: Antibiotic sensitivity test on Gram positive bacteria isolates

Isolates	PEF	CN	APX	Z	AM	R	CPX	S	SXT	E	RESISTANCE INDEX
***Bacillus* sp.[1]**	20(S)	20(S)	12(I)	0(R)	0(R)	10(S)	20(S)	16(I)	12(I)	8(R)	4
***Bacillus* sp.[2]**	10(R)	4(R)	0(R)	0(R)	0(R)	6(R)	10(R)	0(R)	16(I)	12(I)	8
***Bacillus* sp.[3]**	16(I)	10(R)	0(R)	0(R)	8(R)	6(R)	14(I)	18(S)	8(R)	8(R)	7
***Staphylococcus* sp.**	0(R)	10(R)	0(R)	4(R)	0(R)	12(I)	10(R)	14(I)	12(I)	0(R)	7
***Clostridium* sp.**	0(R)	14(I)	0(R)	6(R)	0(R)	0(R)	14(I)	12(I)	14(I)	16(I)	5

KEY:

Resistance (R) = 0-10mm

Intermediate (I) = 11-16mm

Sensitive (S) = 17mm and above

PEF: Pefloxacin

CN: Gentamycin

APX: Ampiclox

Z: Zinnacef

AM: Amoxacillin

R: Rocephin

CPX: Ciprofloxacin

S: Streptomycin

SXT: Septrin

E: Erythromycin

Table 4.10: Antibiotic sensitivity test on Gram negative bacteria isolates

Isolates	SXT	CH	SP	CPX	AM	AU	CN	PEF	OFX	S	RESISTANCE INDEX
***Klebsiella* sp.[1]**	8(R)	12(I)	16(I)	18(S)	0(R)	10(R)	12(I)	12(I)	14(I)	12(I)	3
***Pseudomonas* sp.**	0(R)	6(R)	16(I)	0(R)	0(R)	6(R)	10(R)	6(R)	12(I)	0(R)	8
***Klebsiella* sp.[2]**	0(R)	12(I)	10(R)	0(R)	0(R)	0(R)	0(R)	14(I)	12(I)	0(R)	7

KEY:

Resistance (R) = 0-10mm

Intermediate (I) = 11-16mm

Sensitive (S) = 17mm and above

SXT: Septrin

CH: Chloramphenicol

SP: Saprifloxacin

CPX: Ciprofloxacin

AM: Amoxacillin

AU: Augmentin

CN: Gentamycin

PEF: Pefloxacin

OFX: Tarivid

S: Streptomycin

CHAPTER FIVE

5.0 DICUSSION AND CONCLUSION

5.1 Discussion

Data from this study revealed that barbershops in Ekosodin, Ugbowo harbor microbiological hazards namely *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Klebsiella* sp. This present study is unique in that a metropolitan setting was used as the study area while educational settings were used by (Enemour *et al.,* 2012) and (Mbajiuka *et al*., 2014) isolates, *Staphylococcus* sp. and *bacillus* sp. obtained in this present study were also reportedly isolated in Kogi State university from instruments used by barbers (Enemour *et al*.,2012) and Michael Okpara University of Agriculture, Umudike, Abia state (Mbajiuka *et al.,* 2014). The only difference in both studies is the isolation of *Enterobacter* sp., and not *Bacillus* sp., by (Enemour *et al*. 2012) while *Bacillus* sp. was isolated from this present study, (Mbajiuka *et al*., (2014) isolated *Staphylococcus* s*p., Streptococcus* sp. and *Micrococcus* sp. *Bacillus* sp. was the most abundant of all bacterial isolates while *Staphylococcus* sp. was the least abundant because *Staphylococcus* sp. exist as normal flora on the skin while *Bacillus sp.* is found in soil which are normally found on combs and clippers (which was the materials that was sampled and cultured). Among the fungal species, *Saccharomyces* sp. was the most abundant while *Fusarium* sp. was least abundant because *Saccharomyces* sp. Are a type of yeast that can thrive on various environments including on surfaces like the barbing tools while *Fusariun* sp. on the other hand, are molds that typically require different conditions to grow and might not be as commonly found on barbing tools. The antibiotic sensitivity test as performed in this study was

to ascertain the effectiveness of antibiotics in the treatment of the infections transmitted by barbers' equipment.

The bacterial isolates demonstrated varied patterns of susceptibility to antibiotics which should be one of the major reasons to maintain sanitary and hygienic conditions in barbing equipment. Bacillus sp., isolated from this study was the most resistant bacterial isolate. It showed resistance to five different antibiotics out of the eight used. With this poor sensitivity to antibiotics, infections with this organism could be lethal since treatment required will basically be futile. This bacterial species has been shown to cause various persisting disease in humans such as Head lice, folliculitis and impetigo. In terms of the distribution of the isolates on various equipment, this study showed that the most contaminated of all the equipment was clippers followed by shaving sticks, while the least contaminated is comb. This is so because Clippers and shaving sticks have moving or piercing parts that can trap hair, skin cells, and other debris, creatin a conducive environment for microbial growth unlike combs that have simpler designs and fewer spaces where debris can accumulate, they also tend to have less direct contact to the skin reducing the transfer of contaminants.

The presence of specific microorganism can depend on factors like hygiene, cleaning practices and the handling of the tools. According to the study most of the barbers in Kenyatta Market place their materials and equipment such as combs, brushes and shaving machines and clippers on the benches and shelves after shaving or attending to a client which exposes them to more contamination with pathogenic bacteria from within and without the barber shop. Therefore, the use of these barber shops materials and equipment that already have been infested with pathogenic bacteria expose the barbers as well as the public in general to pathogenic bacteria

which in the long run pose a high risk of infection to the clients and barbers. This agrees with a similar study by (Rebekar, 2010) who indicated that most of the barber shops shelves are used for a similar purpose and little or no attention has been given to these useful materials. Due to lack of attention, materials are exposed to dust and other form of dirt and more severely infestation with pathogenic bacteria which pose a risk of infection to the clients and the barbers. Combs, shaving sticks machines and clippers were all found to be infested with pathogenic bacteria which in general pose a risk of causing and transmitting infection to clients and the barbers. This finding agreed fully with (Hollund *et al*., 2003) who reported in his study that contamination of towels, brush, apron, clippers, combs and shelves if used on an infected customer would certainly spread pathogenic bacteria to other clients. This risk of exposure to pathogenic bacteria was as a result of poor sanitary practices which according to (Ibrahim *et al.,* 2007) is true in that if the instruments and equipment used in barber shops are not sterilized appropriately expose the clients and the barbers at risk of infection. Disease such as Ringworm head lice, herpes, staphylococcus and streptococcus according to (Anderson, 2009) have been found to be transmitted from one person to another via direct contact which is a similar case in barber shops.

Presence of these organisms in almost all the materials sampled indicated that unsafe or unhygienic practices in barber shops in Ekosodin, Ugbowo poses a potential risk of infection by bacterial pathogens to both the barber and the clients. This agrees with the studies done in Ethiopia, Pakistan, and Bangladesh that found out that the sanitary practices in barbershop, however, are largely underestimated and unaddressed as one of a route of blood-borne disease transmission (Arulogun *et al*., 2009). According to (Keene *et al*., 2004) other countries have reported that barbers used sharp instruments which may facilitate the transmission of HBV and

HCV. Barbering procedures according (Nkrumah, *et al*,.2011) could create opportunities for HIV as well as other blood borne and skin diseases transmission. This has been associated with barbering operations such as cutting, face and scalp massaging, nail trimming, pedicure, manicure and shampooing/dying of hair and various health hazards including communicable diseases and skin conditions are associated with barbers' profession to which their clients are exposed to Isolation and the prevalence of these pathogens from the surfaces of these materials concurs with a similar study which found out that the barbering profession/practices pose a risk of infection with *Staphylococcus spp, Klebsiella, Pseudomonas, E. coli* and scabies as well as hepatitis B, C and HIV through contaminated razor blades, clippers and shaving machines (Ibrahim *et al*,. 2007). Use of chemicals such as dettol, Methylated spirit, Bleach (jik), Ethanol, savlon and surgical spirit in barber shops to disinfect the equipment and materials during shaving have been found to expose the barbers and the customers to allergy (Aliye *et al*., 2009). People who are allergic to these chemicals become ill of dermatitis which is as a result of direct contact. This agrees with the study where disinfectants use in barbershops within Ekosodin, Ugbowo is a common practice. According to (Hollund *et al*., 2003) the increased use disinfectants such as shampoo and alcohols containing chemicals pose as an occupational health risk to barbers', hairdressers and the clients which according to him could be attribute to lack of appropriate measures such as general window ventilation and indoor ventilation to prevent this exposure. As a result, these chemicals lead to allergies that cause irritation to the nostrils leading to flu and skin rashes (Hollund *et al*., 2003). This is in agreement with a similar study by (Aliye *et al*., 2009) whose study revealed that exposure to cosmetic disinfectants in barber shops causes allergy and this according to the study is a result of in appropriate measure to prevent it. (Aliye *et al*., 2009) recommended that areas of dye preparation and chemicals including the whole work

place should be well ventilated. dermatitis on their hands while wearing and shaving, but also dermatitis can be expressed on the face or neck of the client while shaving. This does not occur willingly but tiny amounts of allergen fall off the glove by accident. Ringworm an infection caused by dermatophytes are easily transmitted by direct contact or by contact with contaminated equipment and towels is of primary importance. If towels, brush, apron, clippers, combs and razors are used on an infected client successively without proper cleaning and disinfection, the likelihood of spreading an infectious diseases or infestation is almost certain (Hollund *et al.,* 2003). The practice of barbering has continued to expose its practitioners and their customers to multiple infectious diseases. Different microbiological reports have supported this view that barbershops are contributing to the spread of infectious diseases and allergic conditions including scabies, ringworm infection and dermatitis.

5.2 Conclusion

In conclusion, understanding the types of microorganisms present on these tools, their potential risks, and effective preventive measures, we are able to contribute to the improvement of hygiene standards in the beauty and grooming industry. The evaluation of saloon equipment for bacterial and fungi contamination and antibiotics susceptibility have revealed that the microbial loads are very high and the sterilization methods used on this equipment may not be as effective in inhibiting the microorganisms. It also emphasizes the significance of regular sanitation practices to ensure the safety and well-being of both service providers and clients. More so, the bacterial types identified in the study could be normal or transient flora from the persons who utilize these materials. Thus, the transfer of bacterial types from one person to another could predispose people who go for haircuts especially those who are susceptible to skin or other forms of

infections. Although the level of resistance of the bacterial isolates to these antibiotics is alarming, ciprofloxacin, streptomycin, ofloxacin and septrin have shown to be the drug of choice.

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