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# ASSESSMENT OF ANTIBIOTIC RESISTANCE IN BACTERIA ISOLATED FROM RADIOGRAPHY ROOMS IN MOSUL HOSPITALS

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**Abstract:** The increasing prevalence of antibiotic-resistant bacteria poses a significant threat to global public health. Hospitals and radiology rooms, as critical healthcare settings, have become potential reservoirs for the emergence and dissemination of such resistant strains. The rise of antibiotic resistance is primarily attributed to the overuse and misuse of antibiotics, creating a selective pressure that favors the survival and proliferation of resistant strains. Within hospital and radiology settings, this issue is exacerbated due to various factors, including the concentration of vulnerable patients, invasive medical procedures, prolonged hospital stays, and frequent contact with antimicrobial agents. Additionally, the nature of radiology rooms, which often house patients with severe infections or compromised immune systems, further contributes to the risk of bacterial transmission and subsequent resistance development. The aim of this study was to detect antibiotic-resistant bacteria found in hospital and radiology environments at Mosul hospitals. Four strains were isolated from communication surfaces in radiology rooms from three different hospitals. The isolated strains included two strains of E. coli and two strains of Salmonella enteritis. Chromogenic agar was used for strain differentiation, revealing a characteristic blue color for E. coli strains and a pink color for Salmonella enteritis strains. Subsequent sensitivity testing was performed to determine the antibiotic resistance and sensitivity profiles of the isolated strains. The results showed varying degrees of resistance among the strains. Antibiotic resistance was observed against commonly used antibiotics such as ampicillin, ciprofloxacin, and trimethoprim-sulfamethoxazole. However, the strains also exhibited sensitivity to certain antibiotics, including gentamicin and meropenem. The findings of this study provide valuable insights into the prevalence and resistance profiles of antibiotic-resistant bacteria in hospital and radiology environments. The detection of antibiotic-resistant strains, particularly E. coli and Salmonella enteritis, highlights the urgent need for stringent infection control measures and prudent antibiotic prescribing practices in these healthcare settings. Continued surveillance and monitoring of antibiotic resistance patterns are essential to effectively manage and mitigate the spread of resistant strains.

## Introduction

The term antibiotic was derived from the word “antibiosis” which literally means “against life”. An antibiotic was originally, broadly defined as a substance, produced by one microorganism, or of biological origin which at low concentrations can inhibit the growth of, or is lethal to other microorganisms (Russell, 2004, Denyer *et al*., 2008, Schlegel and Zaborosch, 1993). However, this definition has been modified in modern times, to include antimicrobials that are also produced partly or wholly through synthetic means.

Whilst some antibiotics are able to completely kill other bacteria, some are only able to inhibit their growth. Those that kill bacteria are termed bactericidal while those that inhibit bacterial growth are termed

bacteriostatic (Walsh, 2003). Although antibiotic generally refers to antibacterial, antibiotic compounds are differentiated as antibacterials, antifungals, and antivirals to reflect the group of microorganisms they antagonize (Brooks *et al*., 2007).

Penicillin was the first antibiotic discovered in september 1928 by an English Bacteriologist, late Sir Alexander Fleming who accidentally obtained the antibiotic from a soil inhabiting fungus *Penicillium notatum* but its discovery was first reported in 1929 (Aminov, 2010), and clinical trials were first conducted on humans in 1940 (Russell, 2004, Denyer *et al*., 2008, Schlegel and Zaborosch,1993)

**Classification of antibiotics**

There are several ways of classifying antibiotics but the most common classification schemes are based on their molecular structures, mode of action, and spectrum of activity. Others include route of administration (injectable, oral, and topical). Antibiotics within the same structural class generally show a similar pattern of effectiveness, toxicity, and allergic-potential side effects (Calderón and Sabundayo, 2007).

Some common classes of antibiotics based on chemical or molecular structures include Beta-lactams, Macrolides, Tetracyclines, Quinolones, Aminoglycosides, Sulphonamides, Glycopeptides, and Oxazolidinones (Van Hoek et al., 2011, Frank and Tacconelli, 2012, Adzitey, 2015).

**A Beta-lactams**

Members of this class of antibiotics contain a 3-carbon and 1-nitrogen ring that is highly reactive. They interfere with proteins essential for bacterial cell wall synthesis, and in the process either kill or inhibit their growth. More succinctly, certain bacterial enzymes termed penicillin-binding protein (PBP) are responsible for cross-linking peptide units during peptidoglycan synthesis. Members of beta-lactam antibiotics are able to bind themselves to these PBP enzymes. In the process, they interfere with the synthesis of peptidoglycan resulting in lysis and cell death. The most prominent representatives of the beta-lactam class include Penicillins, Cephalosporins, Monobactams, and Carbapenems (Heesemann, 1993, Alfei and Schito, 2022).

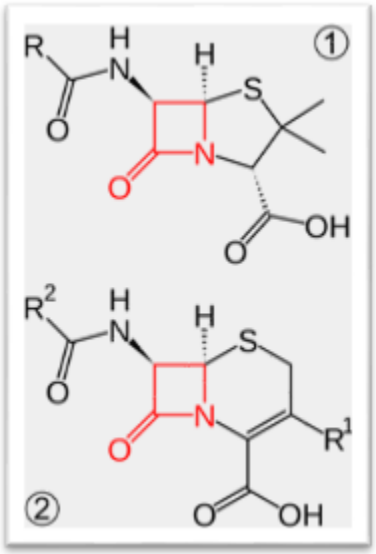



Figure.1 (structure of the beta-lactam)

**A1 Penicillins**

The first antibiotic, penicillin, which was first discovered and reported in 1929 by Alexander Fleming was later found to be among several other antibiotic compounds called the penicillins. Alexander observed that the fungus produced a substance that killed bacteria. Penicillin was first used clinically in the early 1940s and quickly became a widely used and effective treatment for bacterial infections (McGeer *et al*., 2001).

Penicillin works by interfering with the bacterial cell wall, which leads to the death of the bacteria. It is effective against a wide range of bacteria, including *streptococcus*, *staphylococcus*, and *pneumococcus*. However, it is not effective against all types of bacteria, and some bacteria have developed resistance to penicillin (Liras and Martín, 2009, Lima *et al*., 2020).

In view of this, some antibiotics such as ampicillin, carbenicillin and amoxicillin have been developed semi-synthetically with different side-chains. These side chains confer on the antibiotics the ability to evade the degradative capacity of certain enzymes produced by certain bacterial strains as well as facilitating the movement of antibiotics across the outer mmembrane of such bacterial cell walls. This double-pronged capability increases their spectrum of activity against gram-negative bacteria. In particular, some penicillins such as Augmentin are produced in combination with non-antibiotic compound that are able to inhibit the activity of bacterial penicillinase enzyme (Poirel *et al*., 2005, Etebu and Arikekpar, 2016a).

Penicillins are involved in a class of diverse group of compounds, Most of which end in the suffix -cillin. They are beta-Lactam compounds containing a nucleus of 6-Animopenicillanic acid (lactam plus thiazolidine) ring and other ring side chains (Zahner and Maas, 1972). Penicillin V, Oxacillin (dicloxacillin), Methicillin, Nafcillin, Ampicillin, Amoxicillin, Carbenicilin, Piperacillin, Mezlocillin and Ticarcillin (Boundless, 2016). As with every biological interaction systems where living systems seek to protect itself from attack, certain bacteria are able to counter the activity of antibiotics by encoding enzyme (Kardos and Demain, 2011, Etebu and Arikekpar, 2016a).

**A2 Cephalosporin**

Members of this group of antibiotics are similar to penicillin in their structure and mode of action. Cephalosporins are divided into several generations based on their spectrum of activity and their resistance to bacterial enzymes called beta-lactamases. First-generation cephalosporins are effective against gram-positive bacteria and some gram-negative bacteria, while later generations are increasingly effective against gram-negative bacteria and more resistant to beta-lactamases. They are subdivided into generations (1st-5th) in accordance to their target organism but later versions are increasingly more effective against Gram-negative pathogens (Mehta and Sharma, 2016, Erdem *et al*., 2005, Parsels *et al*., 2021).

**A3 Monobactams**

Monobactams are a class of antibiotics that contain a beta-lactam ring structure but have a unique monocyclic structure. They are bactericidal in nature, meaning they kill bacteria directly by disrupting their cell wall synthesis (Sykes and Bonner, 1985, Calderón and Sabundayo, 2007).

The mode of action of monobactams is similar to that of other beta-lactam antibiotics. They inhibit bacterial cell wall synthesis by binding to penicillin-binding proteins (PBPs) that are responsible for cross-linking the peptidoglycan chains in the bacterial cell wall. This binding leads to the activation of autolytic enzymes that break down the cell wall, resulting in bacterial death (Kapoor *et al*., 2017).

Aztreonam is active only against aerobic Gram-negative bacteria such as *Neisseria* and *Pseudomonas*; used for treating Pneumonia, septicemia and urinary tract infections caused by these groups of bacteria. The monobactams are not effective against Gram-positive bacteria or anaerobes. They are used as injectables and inhalers (Sykes *et al*., 1981).

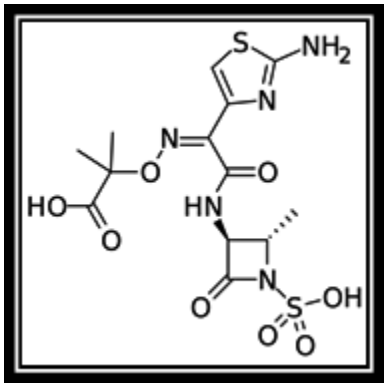


Figure 2. Monobactams structure

This class of antibiotics, was discovered out of necessity in 1976. Prior to this time in the late 1960‟s the effectiveness of penicillin was greatly threatened owing to the emergence of beta-lactamase in bacteria. Bacterial beta-lactamases conferred resistance on bacteria against penicillin (Papp-Wallace *et al*., 2011, Codjoe *et al*., 2019). Carbapenems occupy a very important place in our fight against bacterial infections. This is because they are able to resist the hydrolytic action of beta-lactamase enzyme. Among the several hundreds of known beta-lactams, carbapenems possess the broadest spectrum of activity and greatest potency against Gram-positive and gram-negative bacteria. As a result, they are often called "antibiotics of last resort" and are administered when patients with infections become gravely ill or are suspected of harboring resistant bacteria (Torres *et al*., 2007). Examples of carbapenem are: Imipenem, Meropenem and Ertapenem (Codjoe and Donkor, 2017, Aurilio *et al*., 2022, Young *et al*., 2019, Hsueh *et al*., 2019).

**B Macrolides**

The first antibiotic belonging to this class was first discovered and isolated in 1952 by J. M. McGuire as a metabolic product of a soil-inhabiting fungus Saccharopolyspora erythraea. This fungus was formerly known as Streptomyces erythraeus belonging to the Genus Saccharopolyspora of actinomycete bacteria (Jednačak *et al*., 2020).

Macrolides are characterized by 14-, 15-, or 16- membered macrocyclic lactose rings with unusual deoxy sugars L-cladinose and D-desosamine attached. They have a wider spectrum of antibiotic activity than Penicillins and are often administered to patients allergic to penicillin (Jednačak *et al*., 2020, Pichkur *et al*., 2020).

Macrolides either kill or inhibit microorganisms by effectively inhibiting bacterial protein synthesis. They do so by binding to bacterial ribosome, and in the process, prevent the addition of amino acid to polypeptide chains during protein synthesis. Macrolides tend to build up in the body because the liver is able to recycle them into bile. They also have the capacity to cause inflammation. As a result, clinicians usually recommend administering low doses. Although, Macrolides are generally broad spectrum, some bacterial species such as Streptococcus pneumoniae have resistance against antibiotics. Example of members includes Erythromycin, Azithromycin, and Clarithromycin (Hamilton-Miller, 1973) (Hamilton-Miller, 1973 (Jednačak *et al*., 2020)).

**C Tetracyclines**

Tetracycline was discovered in 1945 from a soil bacterium of the genus Streptomyces by Benjamin Duggar. The first member of this class was chlortetracycline (Aureomycin). Members of this class have four (4) hydrocarbon rings and they are known by name with the suffix )cycline(. Historically, members of this class of antibiotics are grouped into different generations based on the method of synthesis. Those obtained by

biosynthesis are said to be the First generation. Members include Tetracycline, Chlortetracycline, Oxytetracycline, and Demeclocycline (Etebu and Arikekpar, 2016c, Bradford and Jones, 2011).

Members such as Doxycycline, Lymecycline, Meclo Cycline, Methacycline, Minocycline, and Rolitetracycline are considered Second generation because they are derivatives of semi-synthesis. Those obtained from total synthesis such as Tigecycline are considered to be Third generation (Fuoco, 2012, UMOH, 2022).

Their target of antimicrobial activity in bacteria is the ribosome. They disrupt the addition of amino acids to polypeptide chains during protein synthesis in this bacterial organelle (Etebu and Arikekpar, 2016b). Patients are advised to take tetracyclines at least two hours before or after meals for better absorption. All tetracyclines are recommended for patients above eight (8) years because the drugs have been shown to cause teeth discoloration among patients below this age and can be used in treating malaria, elephantiasis, amoebic parasites, and rickettsia (Sánchez *et al*., 2004, Ahn *et al*., 2021)

In the past, antibiotics belonging to this class were very much the envy of numerous Clinicians owing to their wide antimicrobial spectrum but this is no longer the case because numerous bacteria are now able to resist them (Chopra and Roberts, 2001, Gasparrini et al., 2020).

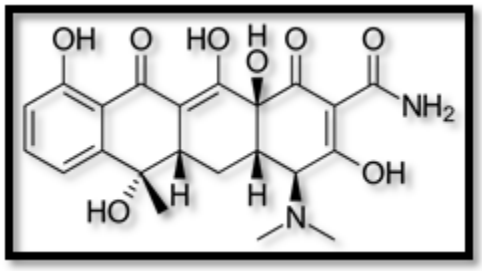



Figure 3. Tetracyclines structure

**D Quinolones**

This class of antibiotics was first discovered as nalidixic acid by scientists involved in the search for antimalarial drugs. They are able to interfere with DNA replication and transcription in bacteria (Bush *et al*., 2020). Two major groups of compounds have been developed from the basic molecule: Quinolones and naphthyridones which include cinoxacin, norfloxacin, ofloxacin, ciproxacin, temafloxacin,sparfloxacin, nalidixic acid, enoxacin, etc (Domagala, 1994). Their structure generally consists of two rings but recent generations of quinolones possess an added ring structure that enables them to extend their spectrum of antimicrobial activity to some bacteria, particularly anaerobic bacteria that were hitherto resistant to quinolone (Bush *et al*., 2020).

The nomenclature of members of this class of antibiotics is complex but members are often known by the suffix–oxacin, such as floxacin, ciprofloxacin, and levofloxacin (Domagala, 1994). Modifications in the basic structure of quinolones are reported to have improved their bioavailability and increased both their spectrum of activity and potency; enhancing their performance in the treatment of various forms of illnesses such as urinary, systemic, and respiratory tract infections. Notwithstanding these notable feats, there still exist safety concerns with some members of this class of antibiotics which has led to the withdrawal of grepafloxacin, sparfloxacin, temafloxacin, trovafloxacin, etc., all belonging to the class quinolones, from the market (Domagala, 1994, Chen and Jiang, 2023).

**Antibiotics mode of action**

The antimicrobial potency of most classes of antibiotics is directed at some unique feature of the bacterial structure or their metabolic processes. The most common targets of antibiotics are illustrated in Figure 1. The mechanisms of antibiotic actions are as follows:

- Inhibition of cell wall synthesis
- Breakdown of cell membrane structure or function
- Inhibition of the structure and function of nucleic acids
- Inhibition of protein synthesis
- Blockage of key metabolic pathways (Alkatheri *et al*., 2023).

**A Inhibition of cell wall synthesis**

Most bacterial cells are encased by a rigid layer of peptidoglycan (PG), also called murein in older sources) which both protect the cells in the face of prevailing osmotic pressure consistent with the often-harsh environment and conditions under which they exist. Peptidoglycan has a degree of cross-linking peptide bonds called β-(1-4) –N– acetyl Hexosamine (Etebu and Arikekpar, 2016b, Höltje, 1998).

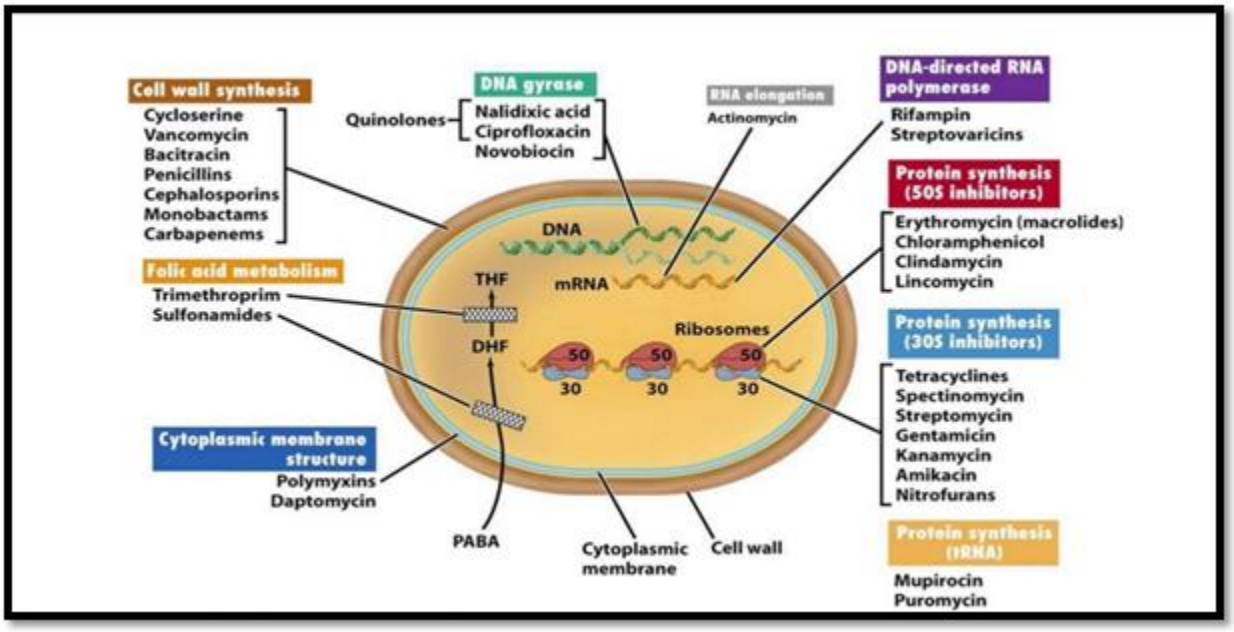



**Figure 1.** Antibiotic target sites (Madigan and Martinko, 2006).

To stay alive, bacteria must of necessity synthesize peptidoglycan; they do this through the activity of PBPs which are transglycosylases and transpeptidases. These two enzymes play very pivotal roles by adding disaccharide pentapeptides to extend the glycan strands of existing peptidoglycan molecules and also cross-link strands of immature peptidoglycan units (Olademehin, 2021, Park and Uehara, 2008). Drugs like penicillins, carbapenems, and cephalosporins are able to block the cross-linking of peptidoglycan units by inhibiting the peptide bond formation catalyzed by PBPs (Josephine *et al*., 2004, Voedts *et al*., 2022)

Most antibiotics belonging to the glycopeptide class of antibiotics (for example, vancomycin) are able to inhibit bacterial growth by inhibiting the synthesis of PG. They inhibit the synthesis of PG by binding themselves to PG units, as well as blocking transglycosylase and transpeptidase activity (Kumar *et al*., 2022, Kahne *et al*., 2005).

**B Breakdown of the cell membrane structure or function**

The classes of antibiotics that damage cell membranes of bacteria are specific in each microbial group based on the differences in the types of lipids in their cell membranes. For example, Daptomycin depolarizes calcium-dependent membrane, and that leads to the cessation of macromolecular synthesis and disruption of the cellular membrane in bacteria (Ledger *et al*., 2022, Alborn Jr *et al*., 1991).

The polymyxins cause disintegration of bacterial cell membrane by effectively binding to the lipid moiety of the lipopolysaccharide in the bacterial cell (Bhattacharjya *et al*., 2022, Falagas *et al*., 2010).

**C Inhibition of nuclei acid synthesis**

The metabolic pathways that result in synthesis of nucleic acids are very essential; disruption of nucleic acid synthesis is inimical to both the survival and posterity of bacterial cells. Antibiotics interfere with nuclei acid synthesis by blocking replication or stopping transcription. DNA replication involves the unwinding of the traditionaldouble helix structure, a process facilitated by the helicase enzymes (Fowler, 2021, Gale, 1981). The quinolones group of antibiotics, for example, do interfere with the functionality of the helicase enzyme thereby disrupts the enzyme from playing its function of unwinding DNA. This antibiotic action of the quinolones ultimately truncates the process of DNA replication and repair amongst susceptible bacteria (Ramos-Martín and D'Amelio, 2023, Chen *et al*., 1996).

Antibiotics whose mode of action is inhibition of nucleic acid synthesis also target topoisomerase II and topoisomerase IV of bacteria. Disrupting the activities of these enzymes in bacteria adversely affects RNA polymerase which in turn prevents RNA synthesis. Quinolones that inhibit bacterial nucleic acid synthesis in this way do not interact with mammalian RNA polymerase, making them specifically antagonistic to Gram-positive bacteria and some Gram-negative bacteria (Ramos-Martín and D'Amelio, 2023, Chen *et al*., 1996).

**D Inhibition of protein synthesis.**

Living things including bacteria are defined by the amount and type of proteins they are composed of, and continually produce. Proteins are responsible for the structural composition, metabolic and physiological processes, and response to adverse conditions, amongst other roles. However, the type and amount of proteins produced by a bacterium at any given time is dependent on information contained in yet another very important biomolecule – Deoxyribonucleic acid (DNA). DNA determines the type of protein a bacterial cell produces through certain information it harbors within itself. The information is a set of genetic codes called codons, handed down to an identical biomolecule – Ribonucleic Acid (RNA), specifically messenger RNA (mRNA). Transfer RNA (tRNA), a similar biomolecule is also formed under the directive of DNA. This biomolecule together with mRNA travels to the ribosomes – the factory for protein synthesis in a living cell. The tRNA then deciphers the codons contained in the mRNA and facilitates the translation of the sequence of codons to a sequence of amino acids which are the building blocks of proteins (Begum *et al*., 2021). The translation of mRNA into proteins occurs over three sequential phases (initiation, elongation and termination) involving the ribosome and a host of cytoplasmic accessory factors (Grimm *et al*., 2022). Ribosomes are made up of RNA and proteins, and are generally called ribonucleoproteins. The RNA component is what is referred to as Ribosomal RNA (rRNA), and comprises two subunits, one small subunit (SSU) and the other large subunit (LSU). These two subunits are usually described in terms of their sedimentation coefficients (that is, their rate of sedimentation is an ultracentrifuge), and are measured in svedberg units (symbols) termed the 30S and 50S, respectively (George *et al*., 2023, Nissen *et al*., 2000).

Bacterial possess 5S, 16S and 23S genes on their rRNA (Moore, 2001). The 16S rRNA gene resides as a single RNA gene in their SSU (16S) whilst the other two rRNA genes (23S and 5S) occur on the LSU of the bacterial ribosome (Naganathan and Culver, 2022). There is huge difference between prokaryotic and

eukaryotic rRNA, and this feat has greatly enabled scientists to develop antibiotics that would target rRNA of a wide spectrum of pathogenic bacteria (Uddin *et al*., 2021).

Given the importance of proteins in the metabolic and life processes of all living organisms, whatever disrupts the process of its synthesis in a bacterial cell would ultimately incapacitate the cell; inhibit its growth or even kill it completely. Drugs that inhibit protein synthesis are among the broadest classes of antibiotics and can be divided into two subclasses: the 50S inhibitors and 30S inhibitors.

Various antibiotics have been identified as inhibitors of ribosomes, specifically targeting either the 50S or 30S subunits. Antibiotics such as erythromycin, clindamycin, lincomycin, chloramphenicol, and linezolid are known to inhibit the 50S ribosome, interfering with either the initiation or elongation phase of protein translation. Oxazolidinones are examples of antibiotics that block the initiation phase, while macrolides like lincosamide and streptogramin inhibit the elongation phase of mRNA translation. However, these elongation inhibitors may lose their effectiveness once protein elongation surpasses a critical length. On the other hand, the 30S ribosome inhibitors, such as tetracycline, streptomycin, and spectinomycin, primarily work by blocking the access of aminoacyl-tRNAs to the ribosome. Understanding the mechanisms of action of these antibiotics is crucial for combating antimicrobial resistance and developing alternative strategies in the fight against infectious diseases (Mlynarczyk-Bonikowska *et al*., 2022, Lomakin *et al*., 2023).

**D Blockage of key metabolic pathways**

Some antibiotics like sulphonamides and trimethoprim have been shown to mimic a substrate needed for the cellular metabolism of bacteria. This deception cause bacterial enzymes to attach themselves to the antibiotic rather than the normal substrate. In particular, Sulphonamides act like tetrahydrofolate which is required to synthesize folic acid in bacterial cells (Begum *et al*., 2021, Park Talaro *et al*., 2002). Folic acid is vital in the metabolism of nucleic acid and amino acids; for this reason, Sulphonamides ultimately disrupt the production of nucleic acids (DNA and RNA) and amino acids, as they mimic substrates required for folic acid metabolism (Begum *et al*., 2021).

**Mechanisms of antibiotic resistance:**

Antibiotic resistance has become a global concern, posing significant challenges in the treatment of infectious diseases. Several bacterial species have emerged as notable multi-drug resistant pathogens, including Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa, Enterococcus spp., Acinetobacter spp., and Klebsiella pneumoniae. Understanding the mechanisms underlying their resistance is crucial for developing effective strategies to combat these infections.

Multi-drug resistant *Escherichia coli* strains often acquire resistance genes through horizontal gene transfer, which can confer resistance to multiple classes of antibiotics. Key mechanisms involve the production of beta-lactamases, such as extended-spectrum beta-lactamases (ESBLs) and AmpC enzymes, efflux pumps, and alterations in target sites, such as mutations in penicillin-binding proteins. Additionally, the formation of biofilms aids in antibiotic resistance by providing physical protection and reducing drug penetration.

In *Staphylococcus aureus*, resistance is primarily mediated by the acquisition of the mecA gene, which encodes the penicillin-binding protein PBP2a, conferring resistance to beta-lactam antibiotics. Other mechanisms include the production of various enzymes, such as beta-lactamases, and efflux pumps that actively extrude antibiotics from the bacterial cell.

*Pseudomonas aeruginosa* exhibits intrinsic resistance to multiple antibiotics due to its low outer membrane permeability, production of efflux pumps, and the formation of biofilms. Acquired resistance often involves the expression of beta-lactamases, such as metallo-beta-lactamases (MBLs), and alterations in the outer membrane porins and efflux systems, leading to decreased drug accumulation. Enterococcus spp. have developed resistance through several mechanisms, including the acquisition of genes encoding resistance to

aminoglycosides, glycopeptides (such as vancomycin), and beta-lactam antibiotics. These resistance determinants are often found on mobile genetic elements, facilitating their spread among Enterococcus strains. Acinetobacter spp. exhibit a remarkable ability to acquire and maintain resistance genes, making them formidable multi-drug resistant pathogens. Key mechanisms include the production of beta-lactamases (including carbapenemases), efflux pumps, alterations in target sites, and the ability to form biofilms.

*Klebsiella pneumoniae* is known for its carbapenem resistance, primarily mediated by the production of carbapenemases, such as *Klebsiella pneumoniae* carbapenemase (KPC) and New Delhi metallo-beta-lactamase (NDM). These enzymes hydrolyze carbapenems, rendering them ineffective. Additionally, *Klebsiella pneumoniae* can acquire genes conferring resistance to other antibiotics, including extended-spectrum beta-lactamases (ESBLs).

**Materials & Methods:**

**Samples**

Three swab specimens of contaminated inanimate surfaces, radiology equipment, and associated medical devices were collected from Ibn-Sina Hospital, Al-zahrawy Hospital, and Al-Salam Hospital located in Mosul, Iraq between February 1, 2023, and March 1, 2023.

Sterile cotton swabs were dipped which were supplied by Biotech containing Cary Blair transport medium in it, the samples were transferred to the laboratory for the purpose of conducting diagnostic tests.

**Culture Media**

The cultural media prepared by Difco, Himedia, and Alpha companies were prepared according to the instructions provided by the company. The media were sterilized in an autoclave at a temperature of 121°C and a pressure of 1 atmosphere for 15 minutes.

**Culture media used:**

1. Nutrient agar media
2. MacConkey Agar media.
3. Mannitol salt agar media.
4. Chromogenic Coliform agar (CCA Agar)
5. Mueller Hinton Agar

**Antibiotic Sensitivity Test:**

The bacterial isolates were tested against a board of antibiotics including Ceftriaxone, Ciprofloxacin, Clindamycin, Amoxicillin Acid, Cephalexin, Piperacillin, Levofloxacin, Imipenem, Azithromycin, Metronidazole and Trimethoprim (Table1) according to the Clinical and Laboratory Standards Institute (CLSI) guidelines.

Table 1. The antibiotics disk used in this study.

| Name of antibiotic | Ceftiraxone | Ciprofloxacin | Clindamycin | Amoxicillin | Cephalexin | peparacillin | levofloxacin | imipenem | Azithromycin | Metronidazole | Trimethoprim |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Antibiotic sample | CRO | CIP | DA | AMC | CL | PRL | LEV | IPM | AZM | MET | TMP |
| Concentrations of antibiotic | 10 | 10 | 10 | 30 | 30 | 100 | 5 | 10 | 15 | 30 | 10 |

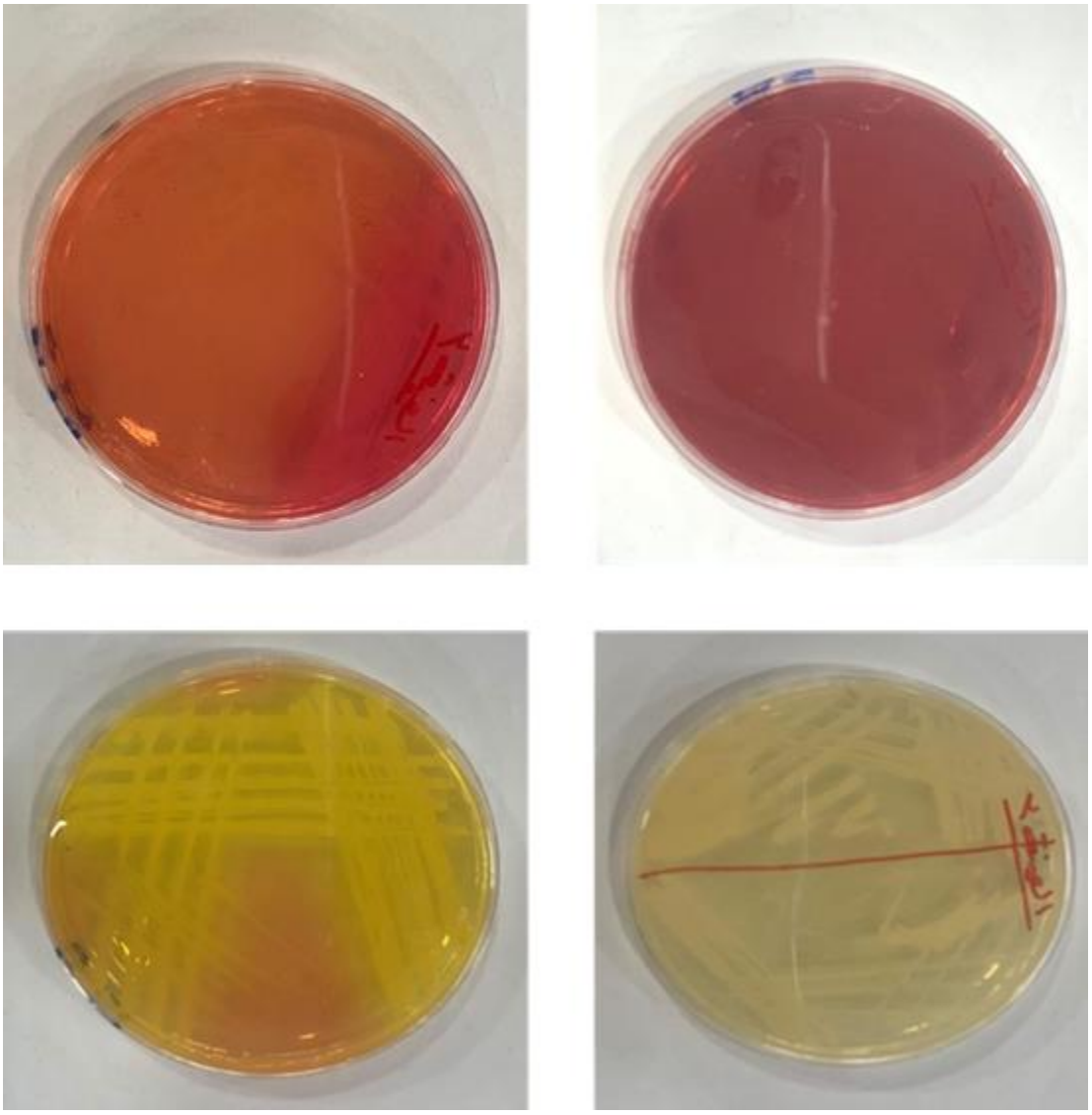
**Results & Discussion:**

The swab sticks collected from the hospital were streaked directly on the labeled agar plates such as MacConkey, nutrient, and Mannitol salt agar then incubated at 37 °C for 24 h and characterized. Inoculation on the MacConkey agar revealed a lack of growth in all samples. MacConkey Agar is a selective and differential medium primarily used to isolate and differentiate members of the Enterobacteriaceae family. MacConkey Agar also contains selective agents, such as bile salts and crystal violet, which inhibit the growth of certain bacteria. These agents specifically target and suppress the growth of Gram-positive bacteria, allowing for the selective isolation of Gram-negative bacteria. It is possible that the stick sample did not contain a sufficient number of viable bacteria for growth to occur on MacConkey Agar.

In this study, a sample was cultured on Mannitol Salts Agar (MSA) to assess its growth characteristics. MSA is a selective and differential medium commonly used in microbiology to isolate and identify bacteria, particularly those belonging to the Staphylococcus genus. The MSA contains mannitol as the sole carbon source and a high concentration of salt, making it inhibitory to many organisms except for halotolerant or halophilic bacteria.

Upon incubation, two distinct types of growth were observed on the MSA. The first type exhibited vigorous growth, characterized by abundant colonies that appeared as smooth, creamy, and yellow in color (Table 1). This type of growth indicates the presence of mannitol-fermenting bacteria. These organisms possess the enzyme mannitol dehydrogenase, which allows them to metabolize mannitol as a source of energy and produce acid as a byproduct. The acid production causes the pH indicator in the MSA to change from red to yellow, resulting in the characteristic yellow color of the colonies. The second type of colonies appeared as small, translucent or white, indicating the absence of mannitol fermentation. The lack of growth or minimal growth of these organisms suggests that they are unable to utilize mannitol as a carbon source or are inhibited by the high salt concentration in the medium.

| Samples | Yellow (mannitol-fermenting) | Pink (cannot ferment mannitol ) |
|---|---|---|
| Samples 1 | Yes | Yes |
| Samples 2 | Yes | No |
| Samples 3 | Yes | Yes |



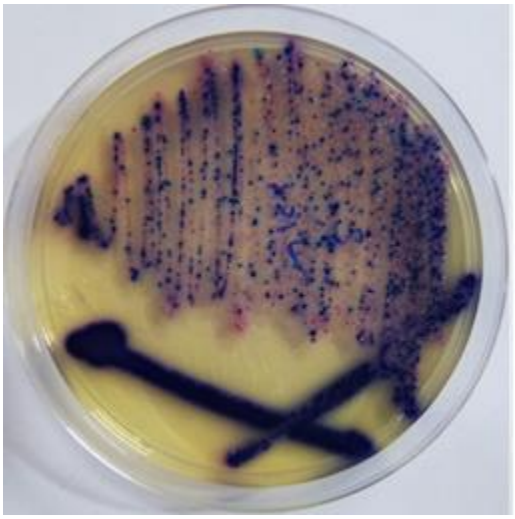
Four strains were isolated from the communication surfaces of radiology rooms in three different hospitals. Chromogenic agar was utilized to differentiate the bacterial strains, which included two strains of *E. coli* and two strains of *Salmonella enteritis*.

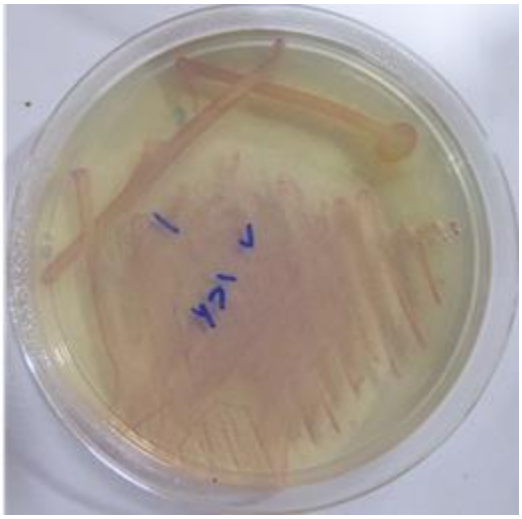
The chromogenic agar allowed for the identification and differentiation of the bacterial strains based on their distinct color reactions. This method enabled the detection of *E. coli* strains, which exhibited a characteristic blue color, and Salmonella enteritis strains, which displayed a pink coloration.

The presence of *E. coli* and *Salmonella* enteritis strains on communication surfaces within radiology rooms is a matter of concern. These bacteria are known to be associated with fecal contamination and are potential sources of nosocomial infections. The identification of these strains highlights the importance of maintaining proper hygiene and sanitation protocols in healthcare settings.

Further investigations, such as antimicrobial susceptibility testing and molecular typing, could provide valuable insights into the antibiotic resistance patterns and genetic relatedness of the isolated strains. These data would be crucial for implementing appropriate infection control measures and designing effective treatment strategies.

Taken together, the isolation of *E. coli* and *Salmonella* enteritis strains from communication surfaces in radiology rooms emphasizes the need for rigorous cleaning and disinfection protocols to minimize the risk of transmission within healthcare facilities. Continued surveillance and monitoring of bacterial contamination in high-traffic areas like radiology rooms are essential for maintaining patient safety and preventing the spread of infectious disease.





In this study, four bacterial strains were analyzed for their antibiotic susceptibility. The *Salmonella* strain demonstrated sensitivity to all antibiotics tested, indicating that it would likely respond well to a variety of antimicrobial treatments. This finding is significant as *Salmonella* is a common cause of enteric infections, and the availability of multiple effective antibiotics is crucial for successful treatment.

Among the two strains of *Escherichia coli* (*E. coli*) isolated and identified using chromogenic agar, one strain showed resistance to ceftriaxone while remaining sensitive to the other antibiotics tested. This suggests that alternative antibiotic options could be explored for treating infections caused by this particular strain of *E. coli*. Identifying the resistance pattern of this strain is important for tailoring appropriate treatment regimens and avoiding the use of ineffective antibiotics.

The second strain of *E. coli* exhibited resistance to multiple antibiotics, including Ceftriaxone, Clindamycin, Cephalexin, Pepraciline, Levofloxacin, Imipenem, Azithromycin, Metronidazole, and Trimethoprim. This multidrug resistance pattern is a cause for concern, as it limits the available treatment options and increases the risk of treatment failure. Monitoring the prevalence and spread of such multidrug-resistant strains is crucial to prevent their dissemination and preserve the effectiveness of antibiotics.

Furthermore, the last strain identified as *Salmonella enteritis* demonstrated resistance to Cephalexin, Amoxicillin, and Ceftriaxone. This resistance profile is significant, as Ceftriaxone is a commonly used antibiotic for the treatment of Salmonella infections. Identifying resistance to Ceftriaxone in *Salmonella* isolates highlights the emergence of antibiotic resistance, which poses challenges for effective treatment strategies.

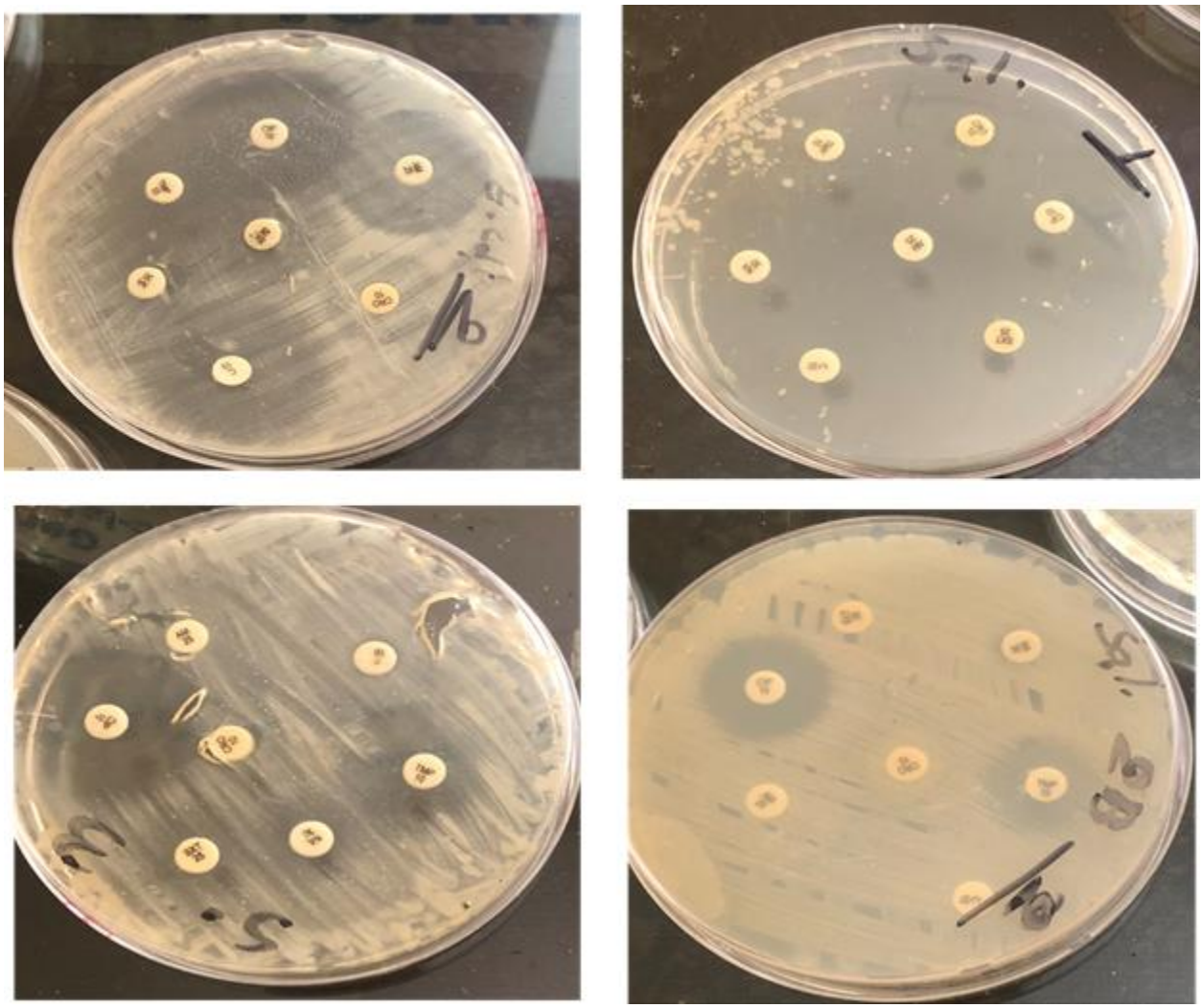
The results of this study emphasize the importance of continuous surveillance of antibiotic susceptibility patterns in bacterial strains. Monitoring antibiotic resistance trends is crucial for guiding empirical therapy and implementing appropriate infection control measures. The susceptibility profile of *Salmonella* strains isolated in this study indicates that a range of antibiotics can be effective in treating infections caused by this pathogen.

However, the presence of multidrug-resistant strains of *E. coli* is a concerning finding. The resistance to multiple antibiotics reduces treatment options and increases the risk of treatment failure. The identified

resistance to Ceftriaxone, a broad-spectrum antibiotic commonly used for various infections, underscores the need for judicious antibiotic use and the development of alternative treatment strategies.

Understanding the resistance patterns of bacterial strains is essential for implementing effective antibiotic stewardship programs. It allows healthcare providers to make informed decisions regarding appropriate antibiotic selection, dosage, and duration of therapy. Additionally, this knowledge assists in identifying emerging resistance trends and developing strategies to limit the spread of antibiotic-resistant bacteria.

In conclusion, this study highlights the variable antibiotic susceptibility profiles among different strains of bacteria. While Salmonella strains were generally susceptible to the antibiotics tested, the presence of multidrug-resistant *E. coli* strains raises concerns about treatment options. Continuous monitoring of antibiotic susceptibility patterns is crucial for informed decision-making in the management of bacterial infections and the preservation of antibiotic effectiveness.



Samples of the practical part

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