# Synthesis and Characterization of Silver ferrite delafossite/MXene Nanocomposite for Enhanced Antibacterial Activity

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Supervisor: Dr. Usman Liaqat

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# DEDICATION

*“Dedicated to Almighty Allah, my supervisor and my beloved family. It could not have been possible without them.”*

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Aqsa Arbab

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| | |
|---|---|
| BET | Brunauer-Emmett-Teller |
| SEM | Scanning Electron Microscope |
| XRD | X-ray Powder Diffraction |
| FTIR | Fourier-Transform Infrared Spectroscopy |
| AP | Agar plates |
| Nm | Nanometers |
| AgFeO2 | Silver Ferrite Delafossite |
| E-Coli | Escherichia Coli |
| MXene | $Ti_3C_2$ |
| MAX | $Ti_3AlC_2$ |
| S.aureus | Staphylococcus aureus |

# ABSTRACT

The development of improved antimicrobial materials is imperative due to the growing threat posed to global health by antibiotic-resistant bacterial diseases. In order to improve antibacterial capabilities through a synergistic mechanism, this study presents a novel MXene/$AgFeO_2$ composite. MXenes are a novel class of two-dimensional transition metal carbides, nitrides, and carbonitrides that possess special characteristics such enormous surface area, variable surface chemistry, and high conductivity. Our objective is to produce a composite with enhanced bactericidal performance by combining MXene with silver ferrite ($AgFeO_2$), which is widely recognised for its strong antibacterial properties.

An inventive physical mixing technique was used to create the MXene/$AgFeO_2$ composite, guaranteeing a uniform dispersion of $AgFeO_2$ nanoparticles on the MXene nanosheets. To verify the effective synthesis and structural integrity of the composite, advanced characterisation techniques were used, such as scanning electron microscopy (SEM), and X-ray diffraction (XRD), Fourier Using quantitative antibacterial assays, antibacterial efficiency was thoroughly assessed against strains of both Gram-positive (Staphylococcus aureus) and Gram-negative (Escherichia coli) bacteria.

According to our research, the MXene/$AgFeO_2$ composite greatly outperforms the individual components and has antibacterial activity never seen before. The combined impacts of $AgFeO_2$'s potent antibacterial activities and MXene's superior electron transfer capabilities are responsible for this outstanding performance. Additionally, the composite showed great stability, highlighting its potential for a wide range of biomedical applications.

To sum up, the MXene/$AgFeO_2$ combination offers a fresh strategy for fighting bacterial infections and constitutes a revolutionary development in antimicrobial material science. Future investigations that focus on refining synthesis settings, clarifying the fundamental antibacterial mechanisms, and investigating useful applications in wound dressings, medical devices, and antimicrobial coatings are made possible by this work.

# CHAPTER 1 INTRODUCTION

This thesis focuses on enhancing antibacterial activiy, incorporating titanium carbide MXene ($Ti_3C_2T_x$) with silver ferrite delafossite nanoparticles(AgFeO2). This study has focused on properties of MXene and its composite with silver ferrite delafossite nanoparticles(AgFeO2) . The purpose of this study is to understand the mutually advantageous link between Mxene and silver ferrite delafossite nanoparticles(AgFeO2)for enhancing antibacterial activity.

## 1.1 Background

### *1.1.1 What is Bacteria?*

Bacteria: Small, single-celled microorganisms that can be spherical (cocci), spiral (spirilla) or rod-like. Being prokaryotes, they lack membrane-bound organelles and a nucleus. Bacteria ruling at various ecosystems of Earth like Soil, Water, Hot Springs and Human Body. Bacteria are very diverse.

### *1.1.2 Characteristics of Bacteria***:**

Bacteria possess a prokaryotic cell architecture—they lack nucleus The nucleiod is the region in which their DNA can be found.Reproduction: Binary fission is the primary asexual process of bacteria in which one cell divides into two identical daughter cells.either heterotrophs (obtaining their organic molecules) or autotrophs (synthesize its own food). There are several ways that they obtain energy including respiration, chemosynthesis and photosynthesis.Genetic Variation: Though bacteria reproduce in basic way, however, they share genetic material through transformation (uptake of foreign DNA), transduction and conjugation that leads towards the increase variation within their populations.

## *1.13 Types of Bacteria (on the basis of strain)*

1-Gram-Positive Bacteria

- Characteristics: These bacteria's cell walls include a thick peptidoglycan layer that maintains the crystal violet stain, making them appear purple under a microscope.Examples:Staphylococcus aureus is a common skin and nasal bacteria that can cause infections.Streptococcus pyogenes causes strep throat, scarlet fever, and other illnesses.Bacillus anthracis causes anthrax.

2-Gram-Negative Bacteria

- Characteristics: These bacteria have a weaker peptidoglycan layer and an outer membrane that does not retain the crystal violet stain but instead absorbs the counterstain (safranin), looking red or pink under microscope.Examples:Escherichia coli: Commonly found in the intestines; some strains can cause food poisoning.Pseudomonas aeruginosa: Known for causing infections in immunocompromised individuals.Neisseria gonorrhoeae: Causes gonorrhea.[1]

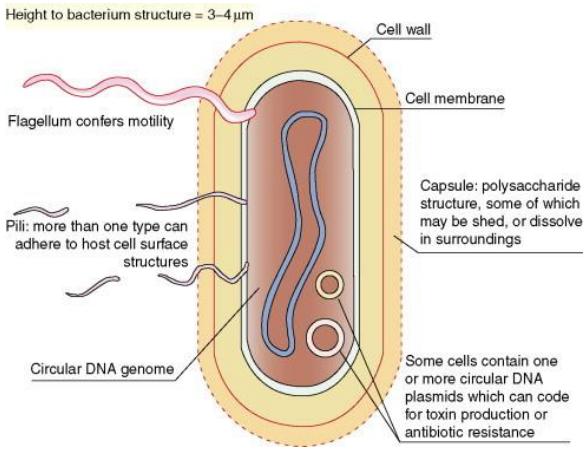



Figure 1.1 Structure of bacterium[2]

### *1.1.4 Bacterial Infections*

When pathogenic bacteria enter the body and grow ,it can lead to bacterial infections and sickness. These infections can affect the skin ,respiratory system,urinary tract and bloodstream, and they can range in severity from moderate to severe.

Common Bacterial Infections:
Skin Infections: Staphylococcus aureus and Streptococcus pyogenes are frequently responsible for infections such as impetigo, cellulitis, and abscesses. Respiratory Infections: Bacteria such as Mycobacterium tuberculosis and Streptococcus pneumoniae are the cause of TB, bronchitis, and pneumonia. 3-Urinary tract infections (UTIs): Escherichia coli is frequently the cause of UTIs. Infections of the Gastrointestinal system: Examples include diarrhoea and food poisoning brought on by germs like Shigella and Salmonella. Systemic Infections: When germs get into the bloodstream, they can cause bacteremia and sepsis. [3]

### *1.1.5 Need for Antibacterial Agents:*

In order to treat bacterial infections, antibacterial medicines, sometimes known as antibiotics, are essential. They function by either eliminating germs or stopping their growth. Antibiotics fall into several groups, and they are all designed to target different bacterial structures or functions, including the creation of cell walls, proteins, DNA replication, and metabolic pathways. [4]

### *1.1.6 Importance of Antibacterial Agents:*

In order to treat bacterial infections that would otherwise be fatal, antibiotics are necessary.They assist in stopping the transmission of bacteria to other areas of the body, one of the consequences that can result from infections.Antibiotics are occasionally used prophylactically to avoid infections, particularly in

immunocompromised patients or during surgical procedures.In order to keep bacterial illness outbreaks under control, antibiotics are essential. [5]

### *1.1.7 Challenges with Antibacterial Agents:*

As a result of the overuse and abuse of antibiotics, bacteria that are resistant to them have emerged, making infections more difficult to treat. Antibiotic side effects can range from minor allergic reactions to serious negative effects. Antibiotics have the potential to upset the delicate equilibrium of the normal microbiota, which can result in infections with Clostridium difficile. [6]

## 1.2 A Brief History

### *1.2.1 Ancient Practices:*

Ancient Civilizations: Ancient civilizations like the Greeks, Egyptians, and Chinese used a variety of natural remedies to treat illnesses. For example, plant extricates, rotten bread, and honey were regularly utilized in conventional medicine.Louis Pasteur and Robert Koch: In the last part of the 1800s, these researchers proposed the microbe speculation of sickness, which exhibited that microorganisms cause contaminations. This hypothesis gave the establishment to ensuing antimicrobial research.Alexander Fleming found penicillin, the primary genuine anti-infection, in 1928. It comes from the mold for Penicillium. By introducing a viable treatment for bacterial infections, this discovery changed medicine. In the early 20th century, Paul Ehrlich developed the first synthetic antibacterial, Salvarsan, and used it to treat syphilis in 1909. He made the expression "enchantment slug" to portray a medication that could kill illness causing organisms while not influencing the host[7]. Mid-century: In the 1940s and 1950s, antibiotics like streptomycin, tetracycline, and erythromycin were developed. The range of bacterial diseases that could be treated grew as a result of these drugs. During World War II, penicillin was produced in large quantities, saving a lot of lives and demonstrating its effectiveness against bacteria. The "golden age of antibiotics" encompasses the postwar period from the 1940s to the 1960s. Many new classes of antibiotics were developed during

this time, which led to a significant decrease in the mortality associated with bacterial infections. Late twentieth Hundred years to Introduce: The abuse and abuse of anti-infection agents has brought about the development of anti-toxin safe microscopic organisms, making a few diseases more hard to treat. To combat resistant bacteria, researchers are still looking for new antibiotics and alternative treatments. This includes researching bacteriophage therapy, creating synthetic chemicals, and natural sources.[8]

### *1.2.2 Modern Challenges and Innovations:*

In an effort to stop the development of resistance, antibiotics are being administered with greater caution. This includes improved diagnostic tools, patient education, stronger antibiotic laws in agriculture and medicine, and Developments like CRISPR innovation, bacteriophage treatment, and antimicrobial peptides are being utilized to battle anti-infection opposition. The creation of novel antibiotics has lagged behind the spread of resistant microorganisms. Pharmaceutical companies frequently focus on diets that result in higher profits. Adverse Effects: Due to the short duration of antibiotic courses in comparison to chronic treatments, adverse effects associated with antibiotics can range from mild (like gastrointestinal disturbances) to severe (like anaphylaxis, organ toxicity), with uncertain financial returns. Often, antibiotic susceptibilities and rapid and accurate detection of bacterial infections are postponed, leading to ineffective empirical treatment. Secondary infections like Clostridium difficile and other health problems can be brought on by antibiotics because they can disrupt the normal balance of the human microbiome. Absence of Fast Tests: to coordinate designated treatment, more quick analytic tests are required.[9]

## 1.3 Importance of studying antibacterial activity:

Antibacterial action is critical to read up for various reasons, including general wellbeing, clinical medication, biotechnology, and ecological examination. The following are a few crucial points that highlight its significance. Understanding antibacterial action is basic for making novel anti-microbials to treat bacterial contaminations, especially given the development of anti-microbial safe microorganisms. Procedures for controlling and

preventing bacterial outbreaks can be developed by examining how germs are inhibited or destroyed, safeguarding public health. As the predominance of anti-toxin safe microbes develops, examination into antibacterial movement supports the revelation of new drugs and opposition. The investigation of how microbes foster anti-microbial obstruction supports the improvement of measures to diminish or dispense with opposition. Understanding antibacterial action illuminates anti-toxin remedies, guaranteeing that patients get compelling and customized treatment. Patients can receive tailored treatment plans based on the pathogens they have by studying how various bacteria respond to various antibiotics. New antibacterial agents like natural products, synthetic compounds, and biotechnological advancements can only be developed through antibacterial activity research. Exploring antibacterial systems can prompt the improvement of existing anti-infection agents, expanding viability while limiting negative effects.Antibacterial research adds to the advancement of drugs that safeguard yields and domesticated animals against bacterial diseases, keeping up with food security and wellbeing. Understanding antibacterial action assists with planning systems for safeguarding food and forestalling bacterial tainting, thus limiting foodborne diseases.Research into antibacterial action can support the administration of bacterial microorganisms in different biological systems, subsequently keeping up with natural balance.Research into antibacterial action adds to the more extensive area of microbial science, improving comprehension we might interpret bacterial physiology, hereditary qualities, and interactions.Insights from antibacterial examinations can prompt imaginative arrangements in different fields, including nanotechnology, materials science, and manufactured science.[10]

### 1.4 Problem Statement

There are a number of major problems and shortcomings with current antibiotic treatments, such as the fact that many bacteria have developed resistance to commonly used antibiotics, making it more difficult to treat infections. Staphylococcus aureus that is resistant to methicillin (MRSA) and Mycobacterium tuberculosis that is resistant to multiple drugs are two examples. Overuse and misuse of antibiotics, as well as their use in livestock feed, accelerate resistance. The proliferation of resistant microorganisms has outpaced the

development of novel antibiotics. Drug organizations oftentimes focus on more beneficial constant sickness therapies. Due to the short duration of antibiotic courses in comparison to chronic treatments, the financial returns on new antibiotics' research and development are unclear. Antagonistic Responses: Incidental effects from anti-toxins can go from moderate (e.g., gastrointestinal unsettling influences) to serious (e.g., hypersensitivity, organ harmfulness). Secondary infections like Clostridium difficile and other health issues can result from antibiotics altering the normal balance of the human microbiome. Ineffective empirical treatment is caused by delayed detection, which delays the timely and accurate detection of bacterial infections and antibiotic susceptibilities. Absence of Quick Tests: More fast demonstrative tests are expected to direct designated treatment.

### 1.5 MXene

Two Dimensional (2D) nanomaterials have extraordinary physiochemical properties due to freestanding sheets and have a very high aspect ratio of lateral size (greater than 100nm) to thickness (less than 10nm).[11] These materials have the atomic scale thickness and exhibit unusual properties e.g., ultrahigh carrier mobilities, very high surface area, quantum hall effects, transparency, and have exceptional applications. Mxenes are one of the emerging 2D materials transition metal nitrides and carbides having general formula $M_{n+1}X_nT_x$ where M is transition metal i.e., Ti, V, Nb, and Mo, X is nitrogen and/or Carbon having atoms from 1 to 4, and $T_x$ is the terminal groups of MXenes F, OH, O, etc.[12] Mxenes have superior properties like electronic conductivity and high hydrophilicity i.e., 20–30° contact angles for films of $Ti_2CT_x$ and $Ti_3C_2T_x$ which can be utilized in biomedical applications like cancer therapy, biosensing, drug delivery medicine, bioimagingand antibacterial activity.The properties of MXenes can be varied by changing in surface terminations of MXenes with the help of different molecules or redox agents. Controllable drug delivery systems using MXene can be made due to their near-infrared (NIR)- and pH- responsive behavior. Due to the tunability of atomic structure and composition they have antimicrobial activities. Thin films, thick membranes or polymer composites of

Mxenes are used for antibacterial activities having good biocompatibility and biodegradability. [13]

### 1.6 Silver Ferrite Delafossite ($AgFeO_2$)

Delafossite-type oxides have attracted considerable attention and have been used in many technological applications . The general formula of the delafossite is $ABO_2$ where A is monovalent ion (Ag+1) and B is trivalent ion (Fe3+) which is located at the center of regular oxygen octahedral that are connected to (Ag+) [14].Silver nanoparticles have emerged as a new generation in medical applications .$AgFeO_2$ delafossite has been used as an antimicrobial agent due to its broad spectrum antibacterial activity and its stability. Moreover, it has been used in water treatment which infected by antibiotic-resistant bacteria . [15]Also, delafossite with hexagonal crystallite structure are typical absorptive materials used in the microwave frequency . The small size of silver nanoparticles should be used in many technological applications especially medical one due to the higher bactericidal ability of nanosilver than bulk one . The previous study showed the fabrication of $AgFeO_2$ delafossite by different methods and showed that Ag-nanoparticles can be employed as an effective bacteria inhibitor and can be applied in medical field .[16]

### 1.7 Significance of the Study

The research of antibacterial activity utilizing MXene and silver ferrite delafossite is important because of their distinct features, which have interesting applications in a variety of sectors, including medicine, environmental science, and materials engineering. Here are the major points that emphasise the importance of such studies:

MXene: The fact that many bacteria have developed resistance to commonly used antibiotics, making it more difficult to treat infections, is one of the major issues and shortcomings of current antibiotic treatments. Staphylococcus aureus that is impervious to methicillin (MRSA) and Mycobacterium tuberculosis that is impervious to various medications are two models. Abuse and abuse of anti-toxins, as well as their utilization in domesticated animals feed, speed up obstruction. The creation of novel antibiotics has

lagged behind the spread of resistant microorganisms. Drug companies frequently concentrate on more effective treatments for persistent illness. The financial returns on new antibiotic research and development are unknown due to the short duration of antibiotic courses in comparison to chronic treatments. Antagonistic Responses: The incidental effects of anti-toxins can range from mild (such as gastrointestinal agitation) to severe (such as hypersensitivity and organ harm). Antibiotics can disrupt the normal balance of the human microbiome, resulting in secondary infections like Clostridium difficile and other health problems. Ineffectual experimental treatment is brought about by deferred identification, which postpones the opportune and precise recognition of bacterial contaminations and anti-infection susceptibilities. Absence of Quick Tests: It is anticipated that designated treatment will be guided by faster demonstrative tests..[17] Synergistic antibacterial effects can be produced by combining MXene and metal ferrite nanoparticles. This means that the properties of the two materials work in conjunction with one another to kill or stop the growth of bacteria, making them more effective. Metal ferrites can transmit ROS, which are extremely responsive and can cause oxidative pressure in bacterial cells. ROS production can be increased when MXene is added, which can improve electron transfer and enhance antibacterial activity..[18] These materials can be utilized in injury dressings, clinical gadget coatings, and different applications that require antibacterial characteristics. MXene and metal ferrite composites can be used in water treatment to eliminate or neutralize bacterial pollutants, making drinking water safer and the environment cleaner. Their ability to target and destroy germs without harming human cells is a significant advantage.[19] Researching the antibacterial components of MXene and metal ferrite can uncover data on how these materials connect with bacterial cells, the job of ROS, layer burst, and DNA harm. Understanding these pathways can support the improvement of techniques to battle bacterial opposition, which is turning into an inexorably difficult issue with standard anti-toxins. [20] Many MXenes and metal ferrites are low-cost options for antibacterial treatments because they are composed of abundant and inexpensive elements. Sustainable development goals can be aided by synthesizing these materials using environmentally friendly methods.[21] MXene and metal ferrite nanoparticles can be tailored to specific antibacterial applications by altering their

structure, surface chemistry, and composition. These materials may also have magnetic properties that enable targeted distribution, imaging, or combination therapeutic techniques (such as hyperthermia treatment), in addition to their antibacterial activity.[22] Examination into MXene and metal ferrite nanoparticles is an imaginative way to deal with the mission for new antibacterial medications, going past standard anti-microbials and researching nanomaterials' remarkable properties. Materials scientists, microbiologists, chemists, and engineers can work together in this field to advance interdisciplinary research and innovation..[23].

### 1.8 Objectives

- Successful synthesis of the MXene .
- Successful synthesis of silver ferrite nanoparticles.
- Successful synthesis of Mxene Silver ferrite Nanocomposite
- Improvement in the stability of Mxene by incorporating silver ferrite nanparticles
- Improvement in antibacterial activity of Mxene

# CHAPTER 2: LITERATURE REVIEW

## 2.1 Overview of materials used in past for antibacterial activity:

### *2.1.1 Conventional materials :*

In the real world, bacterial adhesion and biofilm formation cause significant losses to a number of industries, including food packaging, water treatment, textile processing, marine transportation, and medicine in particular. A solution to slow down bacterial colonization has not yet been found, despite the extensive efforts of academics and manufacturers. As a result, nanomaterials are emerging as a potent alternative to new, efficient methods of controlling pathogens' functional behavior.[24]

Traditional substances with antibacterial properties in the past include:

Table 2.1 (summary of conventional materials used in past for antibacterial activity).

| Year | Material | Title | Authors | Summary | Reference |
|---|---|---|---|---|---|
| 2002 | Magnesium Oxide | Antibacterial effect of magnesium oxide nanoparticles | Stoimenov, P. K., Klinger, R. L. | Investigates the ability of magnesium oxide nanoparticles to inhibit the growth of several types of bacteria. | [25] |

| | | | | | |
|---|---|---|---|---|---|
| 2007 | Carbon Nanotubes | Antibacterial activity of carbon nanotubes | Kang, S., Pinault, M., Pfefferle, L. D. | Investigates the possible mechanisms of action and antibacterial qualities of carbon nanotubes. | [26] |
| 2007 | Gold Nanoparticles | Gold nanoparticles as potent antibacterial agents | Pan, Y., Neuss, S., Leifert, A. | Explains how gold nanoparticles are made and how well they work to combat different bacterial types. | [27] |
| 2008 | Titanium Dioxide | Antibacterial properties of titanium dioxide nanoparticles | Li, Q., Mahendra, S., Lyon, D. Y. | Examines the antibacterial properties and uses of titanium dioxide nanoparticles in the domains of medicine and the environment. | [28] |
| 2009 | Copper Nanoparticles | Antibacterial activity of | Ren, G., Hu, D., | Investigates the creation, characteristics, | [29] |

| | | | | | |
|---|---|---|---|---|---|
| | | copper nanoparticles | Cheng, E. W. C. | and antimicrobial effects of copper nanoparticles. | |
| 2009 | Silver Nanomaterials | Antibacterial applications of silver nanomaterials in medicine | Rai, M., Yadav, A., Gade, A. | Examines the safety and efficacy of employing silver nanoparticles in antibacterial applications. | [30] |
| 2010 | Chitosan-Based Polymers | Antibacterial activity of chitosan-based polymers | Kong, M., Chen, X. G., Xing, K. | Investigates the use of chitosan and its byproducts as antibacterial agents in a range of products, including food and medication. | [31] |
| 2011 | Zinc Oxide | Antibacterial and antifungal activities of zinc oxide nanoparticles | Raghupathi, K. R., Koodali, R. T., Manna, A. C. | Examines zinc oxide nanoparticle production, characterisation, and | [32] |

| | | | | | |
|---|---|---|---|---|---|
| | | | | antibacterial activity. | |
| 2013 | Silver Nanoparticles | Antibacterial activity of silver nanoparticles: a review | Tran, Q. H., Nguyen, V. Q., Le, A. T. | An overview of silver nanoparticle manufacturing, antibacterial processes, and medical uses. | [33] |
| 2014 | Graphene-Based Nanomaterials | Antibacterial properties of graphene-based nanomaterials | Hui, L., Piao, J. G., Auletta, J. | Explains the mechanisms and possible uses of graphene's and its derivatives' antibacterial properties. | [34] |

### *2.1.2 Challenges faced by Conventional materials*

Conventional materials do not have as much antibacterial activity like more advanced or tailored material. Here are just a few of the major challenges. And in English: The antibacterial potency of traditional materials is only limited to a number bacteria species. For this reason, they are not as useful against few types of infections. Eventually, germs get better at resisting them. The majority of these common antibacterials are toxic to human cells, which limits its use as therapeutic agents. These compounds may be slow to degrade and create low but potentially long-lived residues that accumulate in the environment, where their impact on non-target organisms cannot be ruled. Conventional antibacterial materials may lose their efficiency from degradation in vivo over time. Several environmental factors such as humidity, pH and temperature influences also. In the English words of Olson, "Traditional antibacterial have are costly to product which limits

access especially in low resource settings." Raising the production levels without losing efficiency and quality could pose a challenge. In biofilms, on surfaces, bacteria can resist normal antibacterial treatments. Most of the regular materials do not reach trapped microorganisms in tissues or complex surfaces, but also fail to pierce biofilm. Many traditional materials have only one mode of action, such as disrupting cell membranes or inhibiting the synthesis of new proteins. This could then promote their resistance. They may not be able to treat in synergy with other antibacterial medicines, so they could have an overall limited effect. Gaining regulatory approvals for new antibacterial agents can be expensive and take time.[35, 36]

### *2.1.3 Addressing the Challenges with Advanced Materials*

The research that is going in this direction can be synthesized by what has been entitled synthetic biology, biomaterials and nanotechnology. Zinc oxide, copper and silver nanoparticles have shown increased antibiotic activities due to larger surface area of the particles with different mechanism of action. To obviate the risk of resistance, materials that can respond to changes in pH and temperature within a microenvironment by releasing antibacterial chemicals only when needed will be beneficial. Blending of state-of-the-art substances or nanoparticles with conventional material for increasing efficacy and reducing resistance.Development of new biocompatible polymers encapsulating antibiotics with sustained effects.[36]

### *2.1.4 Research and Development*

Continuous research and innovation are very crucial in combating the challenges posed by conventional materials as far as antibacterial activity is concerned. innovative study of the new materials, processes and properties to develop the more effective and sustainable antibacterial agents it is important to perform research in using new materials, processes & application. The recent research of new materials with antibacterial activity, organized by year according to the below table:

Table 2.2 (summary of noval materials used now-a-days for antibacterial activity).

| **Year** | Material | Title | Authors | Journal | Reference |
|---|---|---|---|---|---|
| 2019 | MXene | MXene Nanosheets as Antibacterial Agents | Zhao et al. | Small | [37] |
| 2019 | MOFs | Design of Antibacterial MOFs for Biomedical Applications | Johnson et al. | Chemical Society Reviews | [38] |
| 2020 | Hydrogels | Antibacterial Hydrogels: Design and Applications | Patel et al. | International Journal of Biological Macromolecules | [39] |
| 2020 | MoS2 | MoS2-Based Nanocomposites for Antibacterial Activity | Liu et al. | Journal of Nanobiotechnology | [40] |
| 2021 | MXene | Antibacterial Properties of MXene-Polymer Composites | Sharma et al. | Biomaterials Science | [41] |
| 2021 | MOFs | Antimicrobial Metal-Organic Frameworks: | Wang et al. | Coordination Chemistry Reviews | [42] |

| | | | | | |
|---|---|---|---|---|---|
| | | Synthesis and Applications | | | |
| 2022 | Hydrogels | Injectable Antibacterial Hydrogels for Wound Healing | Chen et al. | Biomaterials Science | [43] |
| 2022 | MoS2 | Antibacterial Activity of MoS2 Nanosheets | Kumar et al. | Journal of Materials Chemistry B | [44] |
| 2023 | MXene | MXenes: Emerging 2D Materials for Antibacterial Applications | Li et al. | Advanced Functional Materials | [18] |
| 2023 | MOFs | Metal-Organic Frameworks for Antibacterial Applications: A Review | Zhang et al. | ACS Applied Materials & Interfaces | [45] |

## 2.2 2D Materials

In the new era of nanotechnology, graphene is what has initiated already abundant prying and generating numerous innovative two-dimensional materials. Two-dimensional materials possess new and unique physical, chemistry or applications because of their special properties compared to the conventional bulk materials that are widely used in industrial areas. This study is designed to summarize central material properties of these materials. Furthermore, discussions are given the synthesis and applications. Finally, an outlook for 2D materials is discussed aimed both at enhancing their performance and

fostering research in such a manner that it may provide more impactful applications benefiting society as well the electrical/electronics sectors. Extraordinarily discoveries are expected in 2D material research as well. 2D materials include:

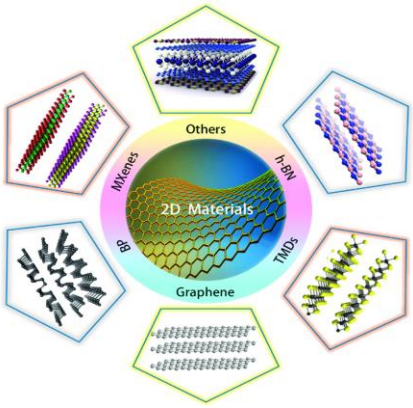



Figure 2.1 Schematics of 2D layered meterials[46]

### *2.2.1 Why 2D Materials for Antibacterial Activity?*

Owing to their unique properties, 2D materials (e.g., graphene, MXenes and transition metal dichalcogenides) such as MoS2 have raised great attention for antibacterial applications. The main reasons for the remarkable antibacterial properties of 2D materials are[46] High surface-to-volume ratio, which gives a large contacting area between bacterial cells and surfaces that enhances their antibacterial activity. Algorithm 2:Large surface area functionalizationextremely large number of antibacterial agents can be functioned with honey-suckle.extract[39] that drastically increase their efficacy. The sharp edges of 2D objects can physically breach bacterial cell membranes, killing off the cells in the process.

It has good mechanical robustness for the reusability of its antibacterial property[17,18]. The surface of two-dimensional materials is very unstable, which can generate nice ROS or do something good substratesrops stream if compared with the bulk. Some 2D materials can release metal ions with known antimicrobial properties, for example the Ag-MXenes silver ions. The researchers found, for example, that exposure to some wavelengths of light

can turn certain two-dimensional materials into heat - and when the surrounding temperature rises enough as a result of this process then bacteria around it is killed.In the presence of light, certain types of 2D material can generate reactive oxygen species (ROS), which destroy bacterial cells. Combining 2D materials with other antibacterial agents (e.g., nanoparticles) could result in positive synergistic effects leading to overall enhanced antibacterial properties. They work via multiple mechanisms (i.e. chemical toxicity and physical disruption) hence reducing to a greater extent, the likelihood of bacterial resistance.Addition of functionalizing groups to 2D materials that render them less toxic and more compatible with human tissues.They might be designed to target only certain strains of bacteria, or specific locations within an infection site to minimize side effects. 2D materials are not a single material but rather the name of multiple compounds such as graphene, MXenes and MoS2. Each of these have unique properties which are important for different antibacterial applications.The potential applications for 2D materials also seem to be as varied, relating from water purification systems to wound dressings and medical equipment coatings .[47]

### *2.2.2 Examples of 2D Materials in Antibacterial Applications*

By oxidative stress, photothermal and physical rupture of membranes are the mechanisms. Applications: Water filtration, antimicrobial fabrics and medical equipment coatings Application plausible mechanisms like photothermal effects, metal ion release and membrane disruption.[48, 49] Applications: Antimicrobial coatings, drug delivery system, wound dressings. Mechanisms, e.g., membrane rupture (developed in this study), photothermal effects [6], and ROS production.[49] IT has applications in the fields of water treatment, biomedical implants and antimicrobial coatings. Their unique characteristics make 2D materials an intriguing choice for next-generation antibacterial solutions. [50] Due to their diversity, functionalization capability and ability for interaction with bacterial

cells in different ways they have several advantages over traditional antibacterial medicines.

### *2.2.3 Synthesis of 2D materials*

There are two synthesis approaches for 2D materials :

**1-Top Down Approach :**

Processes referred to as "top-down" are those that break down large materials into smaller pieces or forms. On account of a 2D material, we can apply hierarchical technique to unstack or shed a few layers from a greater three-layered object. Several mechanical, chemical, or physical processes that precisely exfoliate the starting massive material to produce two-dimensional nanosheets are necessary to accomplish this feat. First Step: A bulk substance is used initially. Process: The matter is broken up or compressed. Their strategies incorporate ball processing, shear shedding, fluid stage peeling (the main technique to detach 2D materials from layered compounds), mechanical peeling and electrochemical delamination. Advantages: Some methods produce near-perfect high-quality monolayers at a lower cost and are simpler to perform on a smaller scale. Bottom-up methods typically have the following drawbacks: lower throughput and resolution, the possibility of process contamination due to synthesis flaws, and inadequate cross-linking efficiency.[51]

**Common Top-Down Methods:**

Top-down synthesis methods for 2D materials involve breaking down bulk materials into thinner layers. Here are the main top-down synthesis approaches:

Table 2.3 (Top down methods for the synthesis of 2D materials)[46]

| Method | Description | Advantages | Disadvantages |
|---|---|---|---|
| Mechanical Exfoliation | Using adhesive tape to remove layers from bulk material. | Easy to use and yields excellent monolayers. | Inadequate yield and unsuitable for industrial scaling. |
| Liquid Phase Exfoliation | Using ultrasonication and a solvent to disperse bulk material. | Scalable and capable of producing vast amounts. | Mixture of few-layer flakes and monolayers that needs to be separated via post-processing. |
| Electrochemical Exfoliation | Exfoliation of a bulk material in an electrolyte solution by applying an electric field. | Quick and yields big volumes. | May cause flaws and necessitate corrective action. |
| Ball Milling | Grinding bulk material into nanosheets in a ball mill by applying mechanical force. | Scalable and yields big volumes. | Potential for layer thickness variation and fault introduction. |
| Shear Exfoliation | Putting shear forces on a liquid-filled bulk material. | Scalable and possible in natural settings. | May produce flakes with a broad variety of sizes. |

**2-Bottom up approach:**

A technique for putting together materials from their atomic or molecular building blocks. When used in 2D material synthesis, the bottom-up strategy describes the process of constructing materials molecule by molecule or atom by atom to form two-dimensional structures. This method creates nanoscale materials similar to 2D materials by piecing together individual atoms or molecules into larger structures. Coming up next are the principal elements of the 2D union base up approach.To make the essential two-layered design, particles or particles are organized exactly.Guarantees an elevated degree of command over the piece and qualities of the material.Makes utilization of atoms' inborn capacity to orchestrate themselves into coordinated structures.Methods like sub-atomic pillar epitaxy (MBE) and synthetic fume testimony (CVD) are habitually utilized. A single atomic layer of material is developed at a time, ensuring layers of excellent quality and uniform thickness, and chemical reactions are used to create two-dimensional materials from more fundamental components. Large-area 2D materials can be produced with this method, which is essential for industrial applications. creates a means of producing high-quality 2D materials in large quantities. enables the production of certain two-dimensional materials, such as hexagonal boron nitride (h-BN), graphene, and transition metal dichalcogenides (TMDs). Both of these processes are widely used. The bottom-up approach can yield materials with fewer flaws and contaminants. Produces materials with improved performance attributes. Methods like liquid-phase exfoliation, electrochemical deposition, and solvothermal synthesis are used. Each method has special benefits with regard to cost, scalability, and material quality. All things considered, the bottom-up method for 2D synthesis provides exact control over material characteristics, scalability for industrial uses, and the capacity to produce a broad variety of 2D materials .[52]

**Common Bottom-Up Methods:**

Here are the main bottom-up synthesis methods for 2D materials:

Table 2.4 (Bottom-Up methods for the synthesis of 2D materials)[53]

| Method | Description | Advantages | Disadvantages |
|---|---|---|---|
| Chemical Vapor Deposition (CVD) | Gas phase precursors react with a substrate to produce a thin layer. produces large-area, industrially relevant films of superior quality. | Produces large-area, industrially relevant films of superior quality. | High cost and exact control are necessary. |
| Molecular Beam Epitaxy (MBE) | Chemical reactions in solution, often including precipitation or self-assembly, produce 2D materials. | Produces crystalline films of superior quality with atomic precision. | Very slow growth rate and pricey apparatus. |
| Solution-Based Synthesis | 2D materials are created by chemical reactions in solution, frequently involving | Scalable, economical, and capable of supporting intricate structures. | Possibly requiring several steps and varying in quality. |

| | | | |
|---|---|---|---|
| | precipitation or self-assembly. | | |
| Hydrothermal Synthesis | In a sealed vessel at high pressure and temperature, reactions take place. | Basic, able to generate 2D materials with high purity and homogeneity. | Only works with particular materials and needs high-pressure machinery. |
| Chemical Bath Deposition | The substrate is submerged in a precursor-containing solution. | Easy to make, inexpensive, and temperature-sensitive. | Restricted to small-scale uses and might result in fewer crystalline materials. |

## 2.3 MXenes As promising material

### *2.3.1 Structure:*

Two Dimensional (2D) nanomaterials have extraordinary physiochemical properties due to freestanding sheets and have a very high aspect ratio of lateral size (greater than 100nm) to thickness (less than 10nm)[54]. These materials have the atomic scale thickness and exhibit unusual properties e.g., ultrahigh carrier mobilities, very high surface area, quantum hall effects, transparency, and have exceptional applications.[55]. The scientific community has paid close attention to "MXenes" due to its unique structural and electrical properties, which open up a wide range of possible uses.

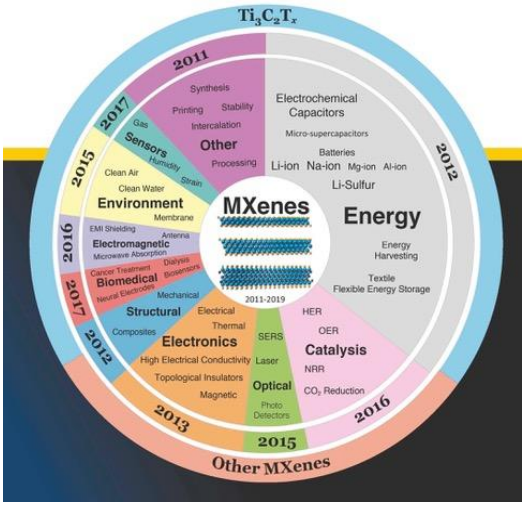



Figure 2.2 MXene Applications[56]

A group of transition metal carbides, nitrides, or carbonitrides known as "MXenes" are typically produced by chemically delaminating MAX phases, which are three-dimensional ternary (or quaternary) complexes. However, MXenes can also be produced by other layered compounds like Mo2Ga2C and Zr3Al3C5. The standard recipe for MXenes is Mn+1XnTx (n = 1-3), where M is a change metal (like Sc, Ti, Zr, Nb, and others), X is carbon or nitrogen, and Tx is the hydroxyl, oxygen, or fluorine terminations that are gotten through the blend processes. Thirty compounds have been synthesized since the Ti3C2Tx 2D molecule was discovered in 2011, with more to come.[57]. Because the MXene inherits the structure and chemistry of its parent MAX phase, research on the MAX phase has a significant impact on the development of MXenes. In addition to haphazard solid solutions on the M website. [58] MXene is another class of 2D nanomaterials with outstanding mechanical, optical, electrical, and electrochemical capacities as a result of its hydrophilicity, blended covalent/metallic/ionic person, and exceptional metal conductivity. Its construction is like that of graphene.[59] Accordingly, MXene materials stand out and have progressed rapidly during the past decade. [58] MXene is typically produced by selectively etching a layer in the MAX phase with hydrofluoric acid (HF). In an effort to improve the quality of MXene, simplify the experimental procedures, and lessen the negative effects of the chemicals, a

variety of preparation methods, such as thermal reduction, UV-induced etching, and alkali treatment, have been developed.[60]

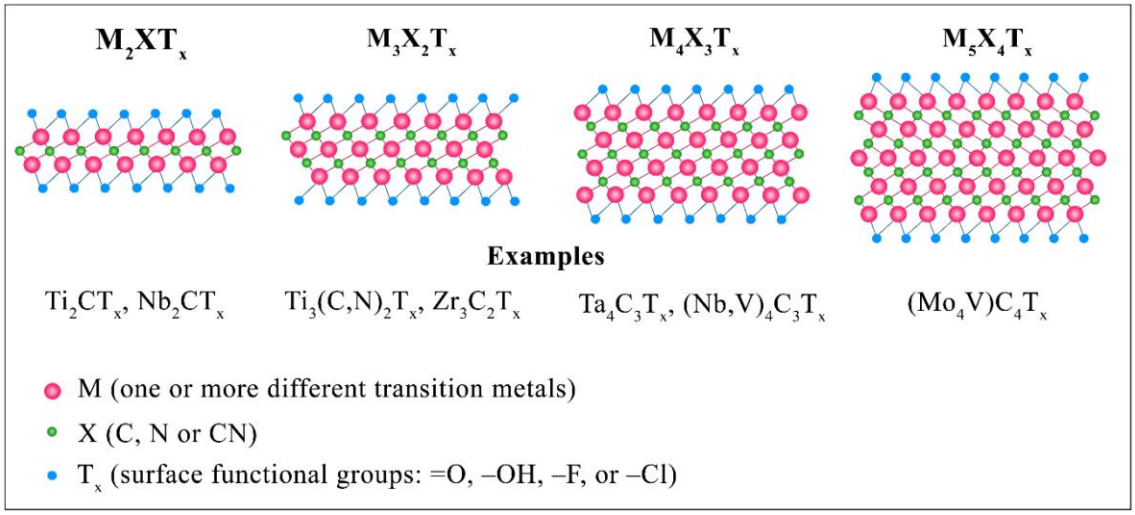



Figure 2.3 Structure of MXene [13]

### *2.3.2 Synthesis methods of Mxene*

Following the effective blend of the first layered Ti3C2Tx utilizing hydrofluoric corrosive, various different procedures were made and tried to create different MXenes with novel pieces. The "bottom-up" and "top-down" approaches to acquiring MXenes currently exist in two distinct ways (Figure 1). The top-down synthesis method separates bulk materials, like MAX phases, into many layers, whereas the bottom-up method forms the material from the bottom to the top using MXene deposition technology. This is the primary distinction between these two approaches. It is important to keep in mind that the synthesis procedure used can have an impact on how the MXenes behave. In addition, the properties of two-dimensional transition metal carbides, nitrides, and carbonitrides are influenced by the starting material.

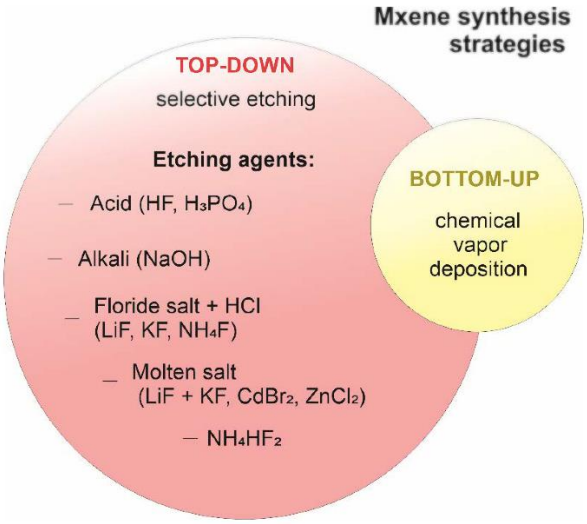



Figure 2.4 Methods for obtaining MXenes and examples of reagents used for selective etching of MAX phases or other precursor materials[61]

As depicted in figure 2.3, MXenes can be made by chemically etching specific atomic layers from stacked carbide, nitride, or carbonitride precursors. The etchants can be broadly divided into two categories: acidic solutions containing fluoride ions (HF, a combination of LiF and HCl, or NH4HF2) and salts containing fluorine ions (NH3F, KF, LiF, or NaF).

### *2.3.3 Properties of MXenes*

MXenes stand out in the world of 2D because they offer a unique combination of features and characteristics that make them appealing for a wide range of applications. It is interesting to note that Ti3C2Tx, the most researched MXene, continues to have its electrical conductivity increased by ongoing synthesis optimization. This is interesting because some MXenes can also be semiconductors or superconductors, depending on composition and surface terminations. [62] With adjustments to the etching process, it was recently claimed that Ti3C2Tx could have conductivity as high as 24,000 S cm1. MXenes are great contender for use as conductive added substances, electrical contacts, conductive straightforward anodes, and movies for protecting against electromagnetic impedance because of their high electrical conductivities and surface terminations, which empower the covalent or electrostatic mooring of different atoms and nanoparticles to make

solid points of interaction [63]. MXenes, like 3D transition metal carbides and nitrides, have high moduli of elasticity and strength. Lipatov et al. found that the Young's moduli of single layers for Ti3C2Tx and Nb4C3Tx were 330 and 390 MPa, respectively. [64] For other 2D materials (like graphene oxide or MoS2), both values are higher than those obtained through liquid exfoliation. MXenes are a promising electrode material for batteries and supercapacitors due to their capacity to house ions and allow quick ionic transport. This gives MXenes an advantage over other nanoscale reinforcements for polymers, ceramics, and metals to create strong functional composites. MXenes exhibit great electrical conductivity and are a promising electrode material for batteries and supercapacitors.[65] [66]

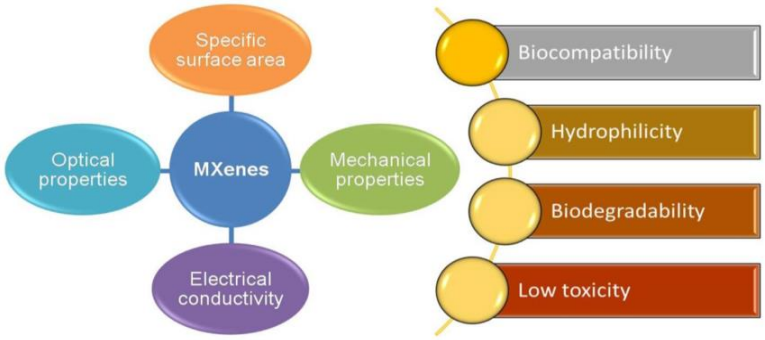



Figure 2.5 Properties of MXenes and MXenes based materials for biotechnology applications[56]

These properties of Mxenes make them extraordainary for use in biomedical field for different purposes.

### *2.3.4 Applications of MXenes*

MXenes' adaptability appeals to a wide range of applications. Because of their high Young Modulus, high electrical conductivity, and ability to alter surface chemistry, composites are popular. Chemical stability, ion intercalation, and changeable bang gaps demonstrate catalysis and energy. Lithium-ion batteries (LIBs), hydrogen storage, and fuel cells are examples of storage applications.[67] Promising results were gotten in a great many fields, including natural and therapeutic sciences, adaptable/wearable hardware,

opto-spintronics. [68] MXenes have demonstrated performance comparable to or superior to that of any other material currently in use in a number of applications, such as energy storage systems, surface-enhanced Raman scattering substrates, electromagnetic interference shielding in electronic and aerospace components, and so on. The majority of research has been focused on electric vehicles because they use it in electronic systems, including those that produce clean, renewable energy..[57]

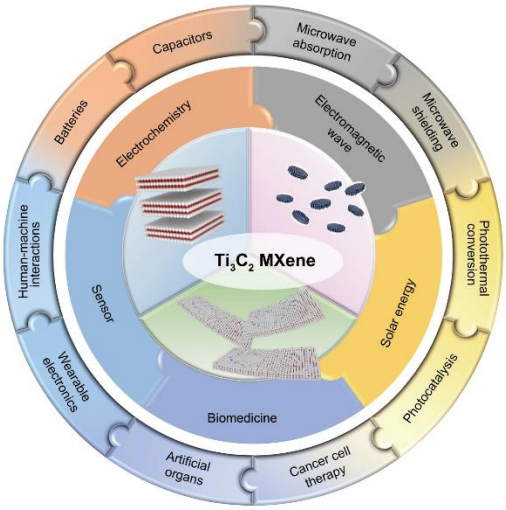



Figure 2.6 Applications of MXenes[69]

MXenes have large surface which makes them best for a lot of applications.[70] MXenes are a good choice for insulating electronic devices from interference because they have excellent EMI shielding properties.[71] MXenes' biocompatibility and practical surfaces are being explored for their true capacity in drug conveyance, biosensing, and antibacterial applications.[59] Due to their high selectivity and sensitivity, MXenes are utilized in a wide range of sensing applications, including biosensors and gas sensors..[72] MXenes are useful catalysts in chemical processes because they have active surface sites and variable electrical characteristics.[73] MXenes are promising for use in water purification and pollutant removal due to their strong adsorption capability and chemical stability..[74]

### *2.3.5 Antibacterial Activity of MXene*

Due to their distinct physiochemical properties and ultrathin lamellar structure, two-dimensional (2D) MXenes possess exceptional antibacterial properties. However, it is still unknown how the antibacterial process may be affected by the atomic structure or how the MXene sheet's size affects its antimicrobial activity.[75] MXenes demonstrate their antibacterial properties by rupturing bacterial cell membranes and producing reactive oxygen species (ROS)..[76] According to studies, MXenes are effective against a variety of bacterial species, including S. aureus and E. coli.[17] Research has shown that MXenes are biocompatible and can be used in biomedical applications, such as coating medical devices to prevent infections.[77] Combining MXenes' antibacterial properties with those of other compounds makes them suitable for advanced biomedical applications. [78].

### *2.3.6 MXenes-Modes of Antibacterial Action*

Because of their distinct physicochemical properties, MXenes can fight microorganisms in a variety of ways. MXenes are able to catalyze the formation of reactive oxygen species (ROS) such as hydrogen peroxide, superoxide anions, and hydroxyl radicals. MXenes can physically harm bacterial cell membranes, resulting in cell lysis and death. MXenes' large surface area and pointed tip make it easier for them to pierce bacterial cell walls, causing internal damage and allowing cell contents to escape. When metal nanoparticles (like Ag, Cu) are added to MXenes, metal ions (like Ag+) are released, which can disrupt bacterial biological processes. These ROS cause oxidative stress, which damages proteins, lipids, and DNA in bacterial cells and causes cell death. When exposed to near-infrared (NIR) light, xenes can function as photocatalysts, generating reactive oxygen species (ROS) and boosting antibacterial activity via oxidative stress mechanisms. MXenes have the ability to promote electron exchange between the bacterial cell membrane and the environment, upsetting the electrochemical equilibrium and energy metabolism of the bacteria and ultimately causing cell death. Metal ions can cause bacterial mortality by interfering with DNA replication, protein synthesis The large surface area of MXenes enables bacterial cells to adhere to their surface, isolating and preventing the growth of the bacteria. MXenes can also prevent the formation of biofilms, which are

bacterial colonies' protective coatings. This disturbance makes bacteria more susceptible to antimicrobial drugs.[37]

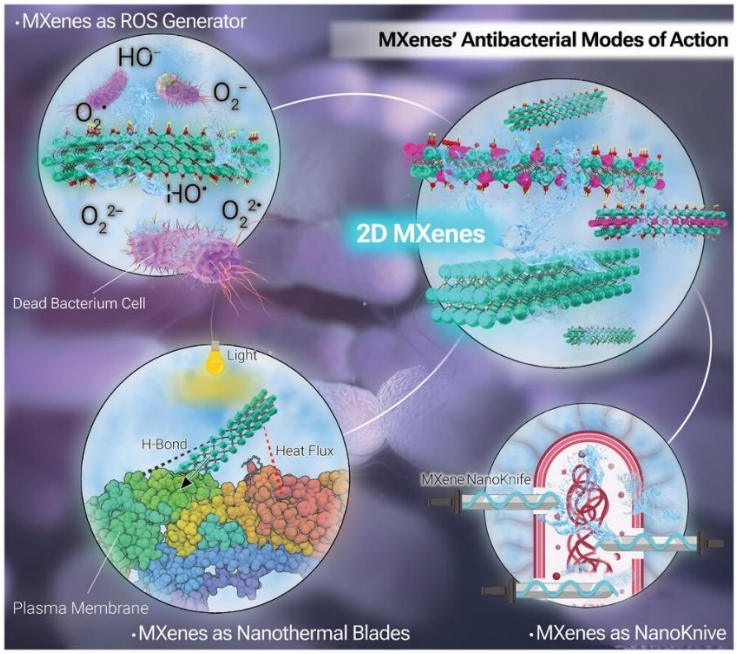



Figure 2.7 ( MXenes-Modes of antibacterial action)[18]

**Summary Table:**

Table 2.5 (MXenes-Modes of antibacterial action)

| **Mode of Action** | Description |
|---|---|
| Membrane Disruption | Bacterial cell membranes damaged physically cause cell lysis. |
| ROS Generation | Reactive oxygen species generation results in oxidative stress and cell damage. |
| Metal Ion Release | Toxic metal ion release impairs cellular processes |

| | |
|---|---|
| Photothermal Therapy | Under NIR light, localised heating kills microorganisms. |
| Photocatalysis | Under light irradiation, ROS production increases antibacterial action. |
| Electron Transfer | Disruption of electrochemical balance and energy metabolism |
| Physical Adsorption | Bacterial cell adsorption and isolation to stop proliferation |
| Biofilm Disruption | Biofilm development inhibition increasing the sensitivity of bacteria |

### 2.3.7 MXenes used for antibacterial activity:

MXenes are thin layers with a variety of early transitional metal atoms joined by carbon or nitrogen and several hydrophilic groups (-F, -OH, etc.) on their surface. O) with a high limit with respect to particle adsorption and hearty metallic conductivity[18]. They have a large surface area because they are planar, which makes it easy for them to become cargo carriers. every single one of their lateral dimensions, which range from nanometers to microns. Their hydrophilicity further qualifies them for biomedical applications, in addition to other equally unique properties like energy conversion and storage, light emission, light harvesting, and photothermal properties, as well as their use in functionalized materials, thermoacoustics, electromagnetic shields, and so on..[79]

Table 2.6 (Tabular representation of MXenes used for antibacterial activity against specific bacteria)

| Year | Title | Authors | MXene | Bacteria | Antibacterial Mechanism | Ref. |
|---|---|---|---|---|---|---|
| 2016 | Antibacterial Activity of Ti3C2Tx MXene | K Rasool, M Helal | Ti3C2Tx | E.coli, Staphylococcus aureus | Direct interaction and rupture of bacterial cells | [37] |
| 2017 | Efficient Antibacterial Membrane based on Two-Dimensional Ti3C2Tx (MXene) Nanosheets | K Rasool, KA Mahmoud | Ti3C2Tx | E.Coli | Direct interaction and rupture of bacterial cells | [37] |
| 2018 | Antimicrobial Mode-of-Action of Colloidal Ti3C2Tx MXene Nanosheets | A Arabi Shamsabadi, M Sharifian Gh | Ti3C2Tx | Escherichia coli, Staphylococcus aureus | Direct interaction and rupture of bacterial cells | [80] |
| 2019 | The Atomic Structure of Ti2C and Ti3C2 MXenes is Responsible for Their Antibacterial Activity Toward E. coli Bacteria | AM Jastrzębska, E Karwowska | Ti2C, Ti3C2 | Escherichia coli | Direct interaction and rupture of bacterial cells | [81] |
| 2021 | Rapid eradication of antibiotic-resistant bacteria and biofilms by MXene and near-infrared light through photothermal ablation | W Wang, J Guo | MXene | Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa, Klebsiella pneumoniae | Photothermal ablation with near-infrared light | [82] |

| | | | | | | |
|---|---|---|---|---|---|---|
| 2021 | Laser-mediated antibacterial effects of few-and multi-layer Ti3C2Tx MXenes | A Rosenkranz, G Perini | Ti3C2Tx | Escherichia coli, Staphylococcus aureus | NIR therapy | [77] |
| 2022 | MXene–laden bacteriophage: A new antibacterial candidate to control bacterial contamination in water | M Mansoorianfar, K Shahin | MXene | Escherichia coli, Staphylococcus aureus | MXene-bacteria interaction | [83] |
| 2022 | Size-dependent photothermal antibacterial activity of Ti3C2Tx MXene nanosheets against methicillin-resistant Staphylococcus aureus | Y Gao, Y Dong | Ti3C2Tx | Methicillin-resistant Staphylococcus aureus (MRSA) | Photothermal ablation with near-infrared light | [84] |

## 2.4 Metal Ferrites :

A class of chemical compounds known as metal ferrites has the formula MFe 2 O 4, where M is a divalent metal ion such as zinc (Zn), cobalt (Co), manganese (Mn), nickel (Ni), or zinc (Ni). These substances fall within the larger heading of ferrites, which are magnetic ceramic materials.

### *2.4.1 Structure and Properties:*

The crystal structure of metal ferrites typically resembles that of spinels. The general formula for spinel is AB 2 O 4, where the cations A and B represent distinct metals. The tetrahedral sites in the spinel lattice of metal ferrites are occupied by M 2 +, the divalent metal ion, while the octahedral sites in the spinel lattice are occupied by Fe 3+ ions. The communications between the iron and metal particles inside the precious stone construction give ferrites their attractive attributes. Soft ferrites, for example, are used in transformer

cores and inductors due to their low coercivity. Due to their high coercivity, hard ferrites are utilized in permanent magnets and magnetic recording media. Electrical Properties: Ferrites are by and large great electrical protectors, which makes them valuable in forestalling swirl flow misfortunes in attractive cores.Applications: Hard plate drives and attractive tapes are made utilizing attractive storage.Transformers and Inductors: Utilized in the centers of transformers and inductors to bring down energy misfortunes and increment efficiency.RF Circuits: Used to decrease electromagnetic obstruction (EMI) in radio recurrence circuits. Sensors: These are used in a wide variety of sensors, including biosensors and gas sensors. Chemical reactions utilize catalysts because of their special surface characteristics and resilience.[85] Examples: Nickel ferrite (NiFe2O4) is an alloy that is well-known for its high electrical resistivity and low losses at high frequencies. Cobalt ferrite (CoFe2O4) is a material with a low saturation magnetization and high coercivity. Manganese ferrite (MnFe2O4) is a material used in microwave devices that is well-known for having good magnetic characteristics. Zinc ferrite (ZnFe2O4) is:[86] A solid-state reaction occurs when iron oxides are combined with metal oxides and heated. A sol-gel technique is used to create ferrites from metal alkoxides or metal nitrates. Co-precipitation is the process of calcining metal ions after they have precipitated from a solution. Hydrothermal synthesis is the process of crystallizing ferrites from metal precursors at high temperature and pressure. The size, shape, and other characteristics of the ferrite particles can all be varied.

### *2.4.2 Types of ferrites:*

Ferrites can be classified into two primary categories according to their crystal structure:

Nickel ferrite (NiFe2O4) and magnesium ferrite (MgFe2O4) are examples of paramagnetic ferrites, which do not retain their magnetic properties when exposed to an external magnetic field. Spinel ferrites like zinc ferrite ($ZnFe_2O_4$) can likewise show paramagnetic way of behaving, particularly at higher temperatures. Moreover, blended ferrites like copper ferrite ($CuFe_2O_4$) may show paramagnetic properties under unambiguous circumstances. Unpaired electrons align with external magnetic fields to form these ferrites, resulting in a weak magnetic attraction.[87]

### *2.4.3 Delafossite:*

The general formula for delafossite is ABO2, Here A is a trivalent cation (such as Al, Fe,or Cr) and B is usually a monovalent cation (such as Cu,Ag, or Pd). Delafossite structures are differentiated by their special qualities and layered arrangement:Here in this figure A represents Ag .B represents Fe while O represents oxygen atoms.[88]

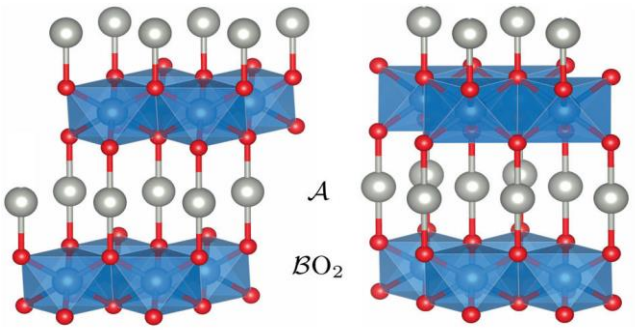



Figure 2.8 Structure of Metal Ferrite Delafossite

Due to their high electrical conductivity, transparent conductive oxides gravitate toward delafossite materials. Numerous delafossites are suitable for thermoelectric applications due to their capacity to convert temperature variations into electrical voltage. A delafossite structure with electrical and magnetic properties, such as CuFeO2, is made of copper and iron.[89]

### *2.4.4 Antibacterial Activity of Metal Ferrites:*

The possible antibacterial qualities of metal ferrites have attracted attention. Numerous processes, such as the production of reactive oxygen species (ROS), the rupture of bacterial cell membranes, and the release of metal ions that are poisonous to bacteria, can be linked to this activity. Here's a thorough examination of metal ferrites' antibacterial activity:

### *2.4.5 Mechanisms of Antibacterial Activity:*

Metal ferrites can catalyze the production of reactive oxygen species (ROS) like hydrogen peroxide (H2O2), hydroxyl radicals (OH2), and superoxide anions (O2O2). The attachment of ferrite nanoparticles can cause damage to the integrity of bacterial cell walls and membranes, resulting in oxidative stress that damages proteins, lipids, and DNA and ultimately causes cell death. This could lead to the death of the cell and/or the leakage of the cell's contents. Ferrites' ability to release metal ions, such as Fe2O, Fe3O, Zn2O, Ni2O, and Co2O, as well as their size and surface charge, are crucial to this process. These ions may interact with bacterial proteins and enzymes, preventing their activity and causing bacterial cell death. For example, nickel and cobalt particles can harm protein designs, and zinc particles can deter bacterial metabolic exercises.[89]

A lot of metal ferrites are already used in antibacterial activity. some are:

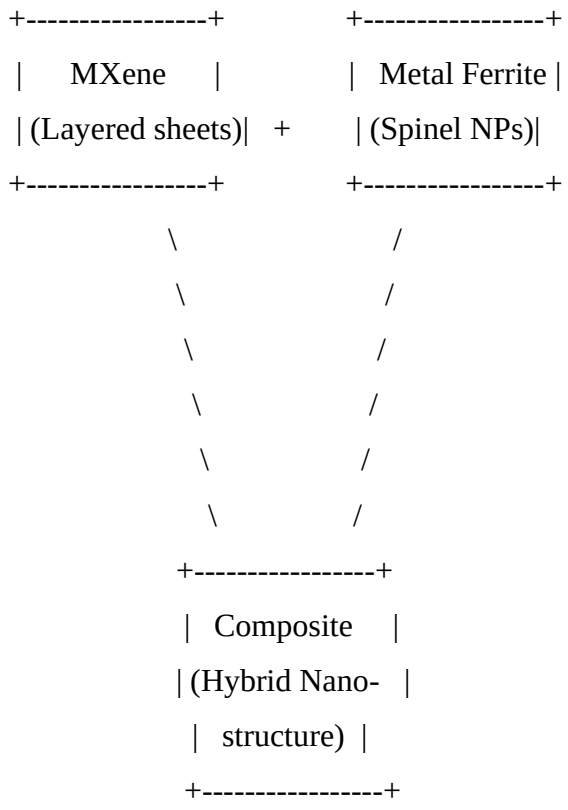
Table 2.7 (Tabular representation of Metal Ferrites used for antibacterial activity against specific bacteria:

| **Metal Ferrite** | Bacteria | Year | Reference |
|---|---|---|---|
| Fe3O4 (Magnetite) | Escherichia coli, Staphylococcus aureus | 2010 | [90] |
| ZnFe2O4 | Escherichia coli | 2013 | [91] |
| CuFe2O4 | Escherichia coli, Staphylococcus aureus | 2014 | [92] |
| CoFe2O4 | Escherichia coli, Bacillus subtilis | 2016 | [93] |
| NiFe2O4 | Escherichia coli, Staphylococcus aureus | 2017 | [94] |
| MnFe2O4 | Escherichia coli, Staphylococcus aureus | 2018 | [95] |
| MgFe2O4 | Escherichia coli, Staphylococcus aureus | 2019 | [96] |

### 2.4.6 Mxene/Metal Ferrite composite

Composites made of metal ferrites and MXenes have garnered a lot of interest and improved performance in a variety of applications due to their synergistic properties. The significance of MXene/metal ferrite composites is highlighted in the following main ideas::

**Schematic Representation:**



### *2.4.7 MXene/Metal Ferrite Composites-Structure:*

Construction of MXene/Metal Ferrite Composites:Transition metal carbides, nitrides, or carbonitrides are layered to make the two-layered materials known as MXenes. The overall recipe for them is M $n$ + 1 X $n$ T $x$ M n +1X n T x, where M is a change metal, X is carbon or potentially nitrogen, and T is surface terminations, for example, - Goodness, - O, and - F.MXenes are beneficial for applications that need to connect with a ton of dynamic locales due to their high surface area.MFe 2 O 4 MFe 2 O 4 is the overall equation for the spinel design of metal ferrites, where M is a divalent metal particle like Zn, Cu, Co, Ni, or Mg.Metal ferrites have attractive characteristics because of their spinel structure, which makes them reasonable for EMI safeguarding and attractive capacity, among other uses.When metal ferrites and MXenes are joined, a crossover nanostructure is made in which the metal ferrite nanoparticles are either intercalated into the MXene layers or scattered over the surface.A key calculate impacting the qualities of the composite is the point of interaction between metal ferrites and MXenes. The overall functionality and stability of the composite can be enhanced by robust interactions at the interface.[97]

### *2.4.8 Importance of MXene/Metal Ferrite Composites:*

**1-Enhanced Antibacterial Activity**:

Construction of MXene/Metal Ferrite Composites:Transition metal carbides, nitrides, or carbonitrides are layered to make the two-layered materials known as MXenes. The overall recipe for them is M $n$ + 1 X $n$ T $x$ M n +1X n T x, where M is a change metal, X is carbon or potentially nitrogen, and T is surface terminations, for example, - Goodness, - O, and - F.MXenes are beneficial for applications that need to connect with a ton of dynamic locales due to their high surface area.MFe 2 O 4 MFe 2 O 4 is the overall equation for the spinel design of metal ferrites, where M is a divalent metal particle like Zn, Cu, Co, Ni, or

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**MXene/Metal Ferrites in Antibacterial Activity:**

Table 2.8 (Tabular representation of MXenes/Metal Ferrites used for antibacterial activity against specific bacteria)

| **Material** | Bacteria | Year | **Ref.** |
|---|---|---|---|
| MXene/Fe3O4 | Escherichia coli, Staphylococcus aureus | 2019 | **[98]** |
| MXene/ZnFe2O4 | Escherichia coli, Staphylococcus aureus | 2020 | **[99]** |
| MXene/CuFe2O4 | Escherichia coli, Pseudomonas aeruginosa | 2021 | **[100]** |
| MXene/CoFe2O4 | Escherichia coli, Bacillus subtilis | 2022 | **[101]** |
| MXene/NiFe2O4 | Escherichia coli, Staphylococcus aureus | 2023 | **[102]** |

### *2.4.9 Novelty(AgFeO2/MXene)Composite:*

AgFeO2/MXene is the material I propose for increased antibacterial activity. The written word does not support that. Because MXene and silver ferrite delafossite (AgFeO2) complement one another, their combination is an excellent option for

enhancing antibacterial activity. While AgFeO2 has strong oxidative properties, production of (ROS) species, high photocatalytic activity, effective generation of free radicals, high surface area, stability under light exposure, biocompatibility and selective toxicity, ability to disrupt cell membrane, antibacterial activity under visible light, synergistic effects of Ag+ and Fe3+ ions, and negative surface charge, MXene has sharp edges for membrane disruption, photothermal properties, photocatalytic properties, high electrical conductivity, The composite's resistance to bacteria is enhanced by the antibacterial qualities of both materials. In addition, MXene provides stability through its layered structure, and AgFeO2's antibacterial properties are further enhanced by the presence of Ag. The AgFeO2/MXene composite is a better option for improving performance and effectiveness in antibacterial activity due to its improved mechanical strength and enhanced antimicrobial properties.

# CHAPTER 3: MATERIALS AND METHODS

## 3.1 MXene(Ti3AlC2):

MXene nanosheets (Ti3C2Tx-MXene) were produced from MAX by etching the Al layer using HF. In order to ensure homogeneous heating, an oil bath was first created and kept at 50°C. Next, 20 ml of HF were added to a Teflon cup and put inside the oil bath. Then, 1g of MAX phase was gradually added to HF. and agitated it for a full day at 600 rpm. After that, MXene was diluted with DI water and centrifuged six or seven times at 4500 rpm until its PH remained at ≥ 6. Next, put it in a drying oven set to 65°C for a whole day. The separation of MXene sheets by adding intercalating agent in next step. To do this, 45 ml of DMSO were added to the above-etched MXene, and the mixture was agitated on a hotplate for 20 hours at room temperature. After that, centrifuged it five or six more times to clean it and keep its pH at neutral. Following this, the MXene solution was subjected to a 3-hour, 45-minute probe sonicator. To obtain the sample, the fluid was ultimately centrifuged once at 3000 rpm. Ultimately, after being dried in a drying oven for 24 hours, MXene nanosheets were prodced.

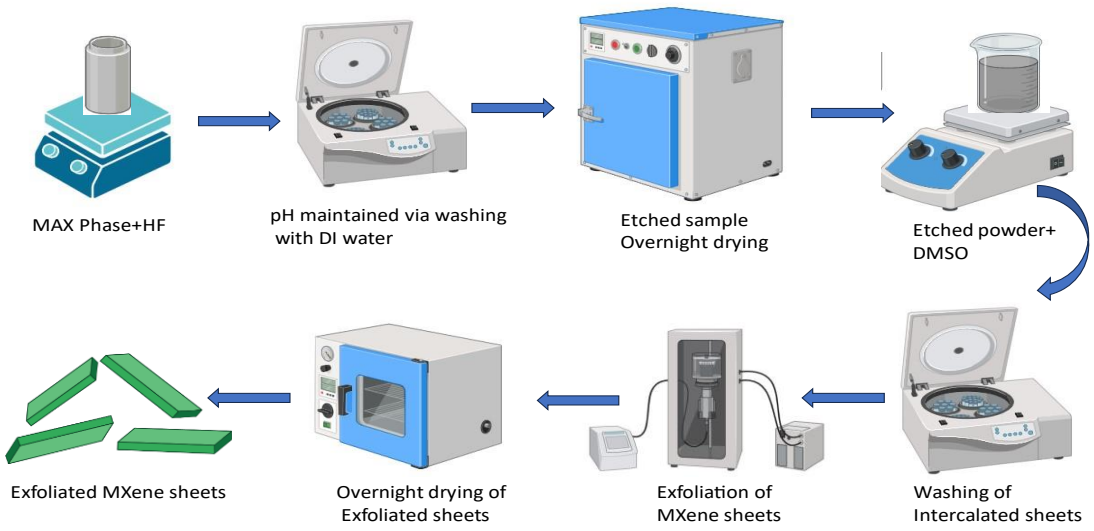
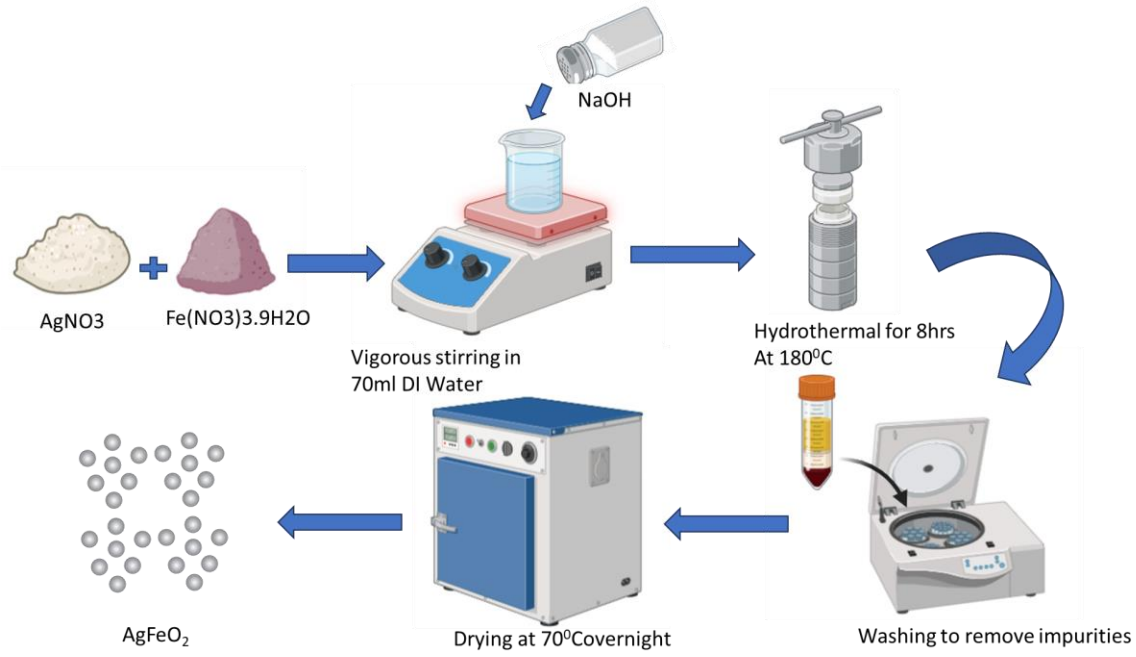



Figure 3.1 Schematics of MXene synthesis)[103]

### 3.2 Silver Ferrite Delafossite(AgFeO2):

An identical 5 mmol concentration of Ag(NO3) and Fe(NO3)3 · 9H2O were seized. The two salts were combined with 70 ml of deionized water to create a solution. The solution was supplemented with 50 mmol of NaOH pellets while being vigorously stirred continuously. The hydrothermal reaction was then carried out in a 100 cc Teflon-lined stainless-steel autoclave once a brown precipitate had begun to develop. For eight hours, the hydrothermal synthesis was carried out at 180 °C. Ultimately, the resultant ruby red-brown product was collected, centrifuged, and dried in a hot air oven for two hours at 70 °C to produce a powder. [104]




(Figure 3.2 Schematics of Silver Ferrite synthesis)

### 3.3 AgFeO2/MXene Composite:

AgFeO2 and MXene were individually distributed (2:1,4:1,6:1,8:1& 10;1) in 30 mL DI water and put in an ultrasonication bath to create AgFeO2/Ti3C2 nanohybrids. After mixing the two solutions, it was sonicated for 1h.After that, a composite was created and dried at 80 °C in a vacuum oven. Ultimately, a mortar and pestle were used to grind the product into a fine powder.[100]

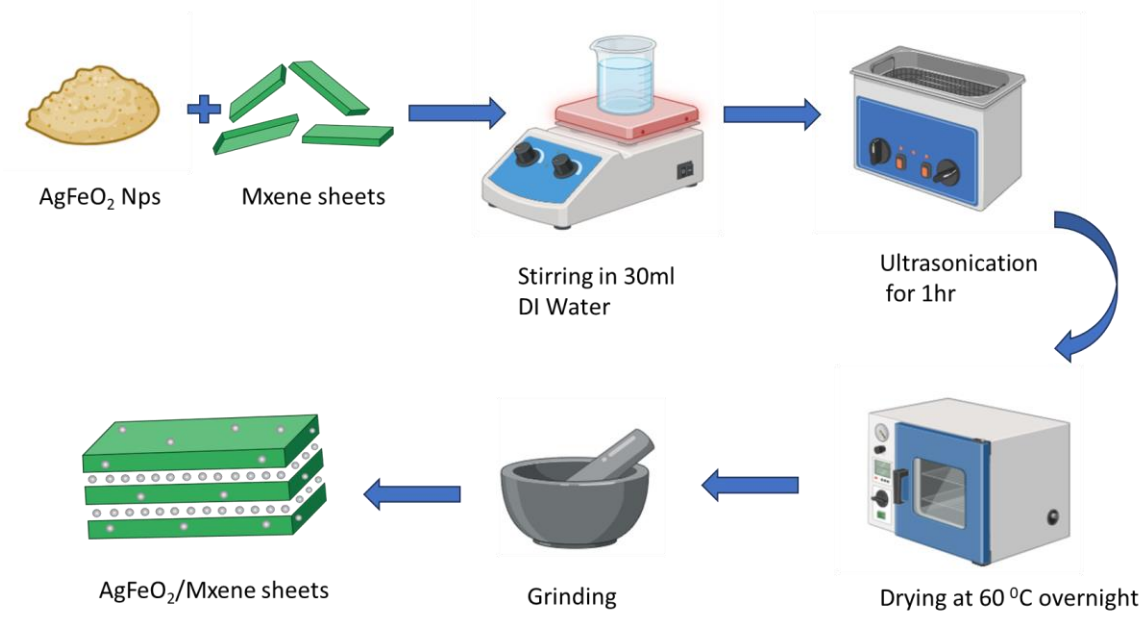



Figure 3.3 Schematics of AgFeO2/MXene synthesis)[100]

## 3.4 Testing Antibacterial Activity:

### *3.4.1 Culturing Bacteria*

Growing bacteria in a controlled environment for research purposes or other uses is known as culturing bacteria. Here's a detailed tutorial on cultivating bacteria:

**Materials Needed:**

Sterile petri dishes, Nutrient agar (or other appropriate growth media)

,Sterile swabs or inoculation loops, Incubator (optional, for controlled temperature),Sterile water or saline solution, Bunsen burner or alcohol lamp (for sterilizationParafilm or tape (to seal petri dishes), Lab coat and gloves (for safety)

**Procedure:**

The melted agar was then poured into sterilized petri dishes until it reached the bottom (approximately 15–20 mL) if you were using pre-made agar. Take care not to overheat the agar. With the covers somewhat partially open to stay away from buildup, let the agar cool

and solidify at room temperature.On the underside of the petri dishes, compose the date, the sort of test, and some other relevant details.If a swab was utilized, it was clean. dipped the loop or brush into the bacterial sample (a surface, a solution, or a colony of bacteria from a different plate) to sterilize it in the event that an inoculation loop is going to be utilized. Streaked the Plate: To completely cover the agar's surface with solid medium, streak the sample across the plate in a zigzag pattern. For liquid cultures, use a sterile spreader to evenly distribute the liquid over the agar surface. To prevent contamination, cover the edges of the petri dish with tape or parafilm. Place the plates upside down in an incubator or at room temperature to prevent moisture from getting onto the agar. The plates were incubated at the right temperature (usually 37°C for human diseases) for the particular bacteria being cultivated. After 24 to 48 hours, the bacterial growth on the plates was checked. These methods were successful in cultivating bacteria for a variety of microbiological applications or study, and isolated colonies were selected and respread onto fresh plates as needed to obtain pure cultures. Spots that stood out on the agar surface were colonies.If you were using pre-made agar, heat it in a water bath or microwave to liquefy it. took care not to become too hot.[105]

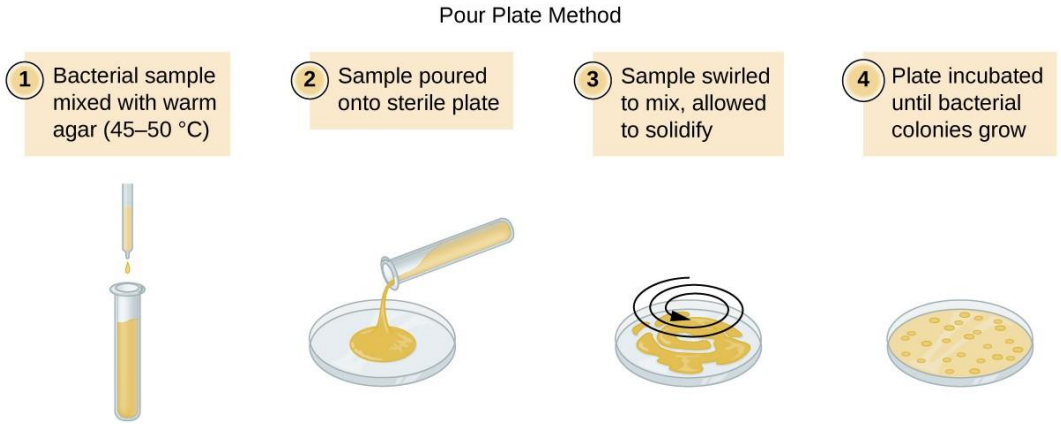



Figure 3.3 Shematics of culturing bacteria[105]

### *3.4.2 Disc Diffusion Method:*

The Kirby-Bauer test, also known as the disc diffusion method, works by introducing an antimicrobial agent in a uniformly infected agar medium into a gradient. When antibacterial discs are placed on the surface of the agar, the agent spreads outward. The zone of restraint is a particular roundabout region that structures around the plate because of microbes that are delicate to the antimicrobial specialist being kept from multiplying. The size of the zone has an impact on the effectiveness of the antimicrobial agent against the microorganisms that were tested. Variables such as the agent's diffusion rate, the concentration of bacteria, and the incubation conditions are standardized in order to guarantee reliable results when evaluating bacterial susceptibility to the tested antimicrobial agents.[106]

### *3.4.3 Testing Prepared Composites on Bacteria via Disc diffusion Method:*

**Materials Needed:**

Prepared antibacterial material**,**Sterile petri dishes with nutrient agar**,**Bacterial culture (e.g., **E. coli, S. aureus),**Sterile swabs or inoculation loops**,** Sterile forceps or tweezers**,** Incubator**,**Sterile water or saline solution**,** Bunsen burner or alcohol lamp (for sterilization)**,** Paper discs (e.g., filter paper)**,** Control disc (e.g., disc with no antibacterial material)**,** Lab coat and gloves (for safety)

**Procedure:**

The inoculation loop was sterilized by heating it up with an alcohol lamp or Bunsen burner until it burned red, then letting it cool. After dipping the sterile brush or chilled inoculation loop into the bacterial culture, spread the bacteria uniformly across the surface of the nutrient agar in the petri dishes. Write the date, the kind of bacteria, and any other pertinent information on the bottom of the sterile petri dishes. Weakenings of every one of the seven examples were made in 1ml of DI water with centralization of 1mg,5mg and 10mg

respectively.Cut consistently estimated paper circles, (for example, those produced using channel paper) with sterile scissors.Soak the paper plates in the antibacterial arrangements (MXene,AgFeO2 and all composites named as C1,C2,C3,C4 &C5) that has been made.Soak the control circles in sterile water or comparable idle arrangement. Weakenings of every one of the seven examples were made in 1ml of DI water with convergence of 1mg,5mg and 10mg respectively.The drenched paper circles were put into the vaccination agar plates' surface utilizing sterile forceps. Make sure the discs were evenly spaced and gently pressed on the agar. The control discs were added and placed on the same plate for comparison. Seal the edges of the petri dish with tape or parafilm to prevent contamination. In a hatchery, place the plates topsy turvy to prevent dampness from pouring onto the agar. hatched at the legitimate temperature (typically 37°C for human illnesses) for the specific microorganisms under test.:We searched for clear districts encompassing the circles where microbes didn't develop, or zones of hindrance, on the plates following 24 to 48 hours. utilized a caliper or ruler to gauge the measurement of these zones. The data were examined to determine whether the prepared material was more successful than the control at preventing the growth of bacteria. This comparison was made between the zones of inhibition surrounding the antibacterial material discs and the control discs in order to evaluate the antibacterial activity.

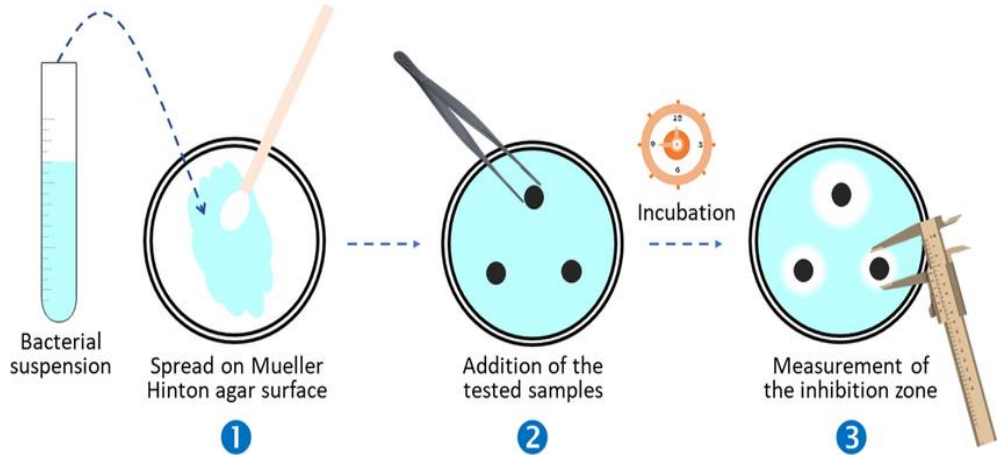



Figure 5.4 Schematics of Disc diffusion method)[106]

# CHAPTER 4: CHARACTERIZATIONS

## 4.1 Instruments

X-beam diffraction (XRD), Examining electron microscopy (SEM), Bright radiation(UV), Fourier change infrared spectroscopy (FTIR) were utilized to assess the examples. Other methods like Brunaure-Emmett-Teller (BET) and contact angle were used to get both qualitative and quantitative results. Atomic force microscopy (AFM) was also utilized for the purpose of structure analysis. In the end, the composite disc diffusion method's enhanced antibacterial activity was also checked.4.2 Scanning Electron Microscopy(SEM)

### *4.2.1 Working Principle*

A scanning electron microscope (SEM) is powered by a high-energy electron beam generated by an electron cannon. Then, utilizing electromagnetic focal points and openings, this shaft is packed into a little region. The focused electron beam is systematically moved across the sample's surface in a raster pattern by scan coils. When the beam interacts with the material, a variety of signals, such as secondary electrons, backscattered electrons, and distinctive X-rays, are produced. These signals are the result of the sample's atoms and the beam's interaction. Secondary electrons, which are released from the surface, provide extensive topographic information. Backscattered electrons, which scatter more electrons and are reflected back from heavier materials, can be used to infer the sample's composition. Characteristic X-rays are released when inner-shell electrons are displaced by the beam, allowing for compositional analysis of the sample's components. The SEM is able to produce high-resolution images as well as comprehensive compositional data on the sample thanks to the collection of these signals by a variety of detectors. A scanning electron microscope (SEM) works by concentrating a beam of high-energy electrons using an electron cannon. This beam is shaped and concentrated in a specific area using multiple electromagnetic lenses and apertures. The electron beam is then carefully moved across the sample surface in a raster pattern using scan coils. The beam generates a variety of signals when it interacts with the sample because of its

interactions with its atoms. The example's surface deliveries optional electrons, which furnish exact geological data with extraordinary goal and uncover fine surface subtleties. Backscattered electrons that are reflected back from the sample can provide compositional information because heavier elements scatter more electrons and provide contrast based on atomic number. In addition, the interaction of the electron beam with the sample can release inner-shell electrons from the atoms, resulting in the emission of distinct X-rays. These X-rays are element-specific, allowing for both quantitative and qualitative compositional analysis. There are numerous detectors in the SEM that can detect these distinct signals. Typically, a secondary electron detector can be used to capture high-resolution photographs of the sample's surface morphology. Backscattered electron detectors can produce compositional contrast images, and energy-dispersive X-ray spectroscopy (EDS) systems are used for elemental composition analysis. The SEM can offer itemized data at an exceptionally fine scale, much of the time down to the nanoscale level, on a superficial level design, piece, and geology of the example because of the mix of a few discovery procedures. Because of this, SEM is a very useful method in a lot of other fields, like electronics, biology, and materials science, that need to look at surfaces in depth.[107]

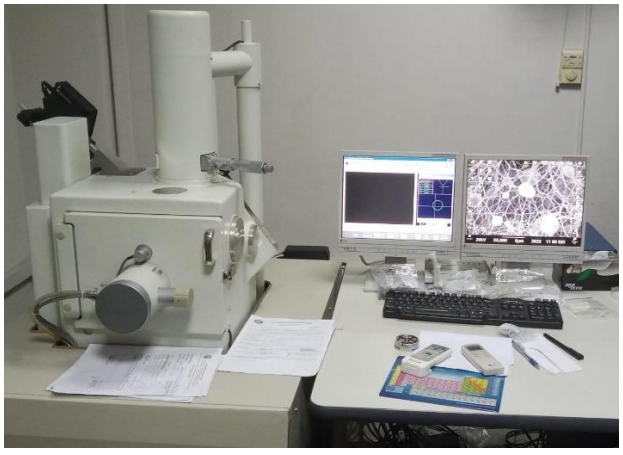


Figure 6.1 SEM (JEOL) at SCME ,NUST

### *4.2.2 Important Features*

Many aspects of a sample can be studied using scanning electron microscopy (SEM), such as its morphology, chemistry, crystallography, and plane orientation. The basic sample preparation procedures make it easy to study all of these features. The variable magnification range of the scanning electron microscope (SEM) is between 10 and 500,000 times. Any instrument can be used to examine the materials' morphology, but a FESEM will provide greater resolution. The essential piece can be found out by utilizing a Field Emanation Checking Electron Magnifying lens (FESEM) related to an Energy Dispersive X-beam Spectroscopy (EDS) indicator.[108]

### *4.2.3 Sample Preparation*

The example was either preheated or dried for three hours at 60°C in a drying stove. This procedure was carried out to get rid of any moisture that might have been in the material or got stuck between the layers. From that point onward, the dried examples were immediately moved to the SEM contraption's example holder or stub for assessment.

## 4.3 Xray Diffraction (XRD)

### *4.3.1 Working Principle*

X-ray diffraction (XRD) is a powerful analytical technique for examining materials' chemical composition, physical properties, and crystallographic structure. In the fundamental X-ray reflectance diagram (XRD), X-rays are directed toward a crystalline substance. When these X-rays come into contact with the crystal lattice, their atomic arrangement causes them to be diffracted in particular directions. The justification behind this diffraction is that the X-beams' frequency is like the distance between the molecules'

planes in the gem. The subsequent diffraction pattern, which consists of multiple peaks, can be recorded and examined.


Figure 4.2 XRD (BRUKER) at SCME, NUST

By measuring the angles and intensities of these diffracted beams, precise information on the crystal structure can be gleaned. Each crystal has a unique diffraction pattern that can be used to identify the substance, similar to a fingerprint. Peak intensities and peak positions in the diffraction pattern provide information about the unit cell's symmetry and dimensions, respectively.[109]

Graphs and interpreting XRD patterns requires a grasp of Bragg's Law, which is expressed as follows:

$$n\lambda=2d\sin\theta$$

where n is the order of the reflection, d is the distance between the crystal surfaces, n is the incidence angle, and n is the X-ray wavelength. XRD is frequently used in materials science, chemistry, geology, and biology to identify unknown crystalline materials, assess their purity, and comprehend their structural characteristics. At the School of Chemical and Materials Engineering (SCME-NUST) of the National University of Sciences and Technology, X-ray diffraction studies were conducted.

### *4.3.2 Sample Preparation*

For X-beam Diffraction (XRD) to deliver exact and dependable discoveries, test readiness is vital. The procedure typically begins with the collection and homogenization of the

sample to ensure that it is representative and uniform. Powdered samples, which are the most frequently used type in XRD, require particle sizes of approximately 10 micrometers for fine grinding. This can be done with a ball mill, mechanical grinder, or mortar and pestle. To avoid altering the crystal's structure, grinding must be performed with care. Sensitive materials can be prevented from degrading with cryogenic grinding. The powder is flattened and distributed evenly on a sample holder after being ground. This ensures predictable association between the X-beam pillar and the example. On account of dainty movies, the example is set on the right track onto a suitable substrate. Greater or bulkier translucent examples can be set on the right track into the example holder, ensuring that the X-beam pillar arrives at the area of interest. The example particles should be haphazardly situated to forestall issues with favored direction, in which precious stones might adjust such that changes diffraction designs. Proper sample preparation for XRD analysis ensures accurate information about the material's crystallographic structure, chemical composition, and physical characteristics.[110]

## 4.4 Fourier Transform Infra-Red (FT-IR) Spectroscopy

### *4.4.1 Working Principle*

An analytical method known as Fourier Transform Infrared Spectroscopy (FTIR) uses infrared absorption spectra to identify organic, polymeric, and occasionally inorganic materials. FTIR is based on the idea that an infrared (IR) beam should be directed toward a sample. The material's vibrational modes are represented by the sample's IR light absorption at specific frequencies. These retained frequencies structure an unmistakable phantom unique mark that can be utilized to recognize and evaluate various parts in the example.[111]


Figure 4.3 FTIR (PERKIN ELMER) at SCME, NUST

### *4.4.2 Important Features*

The IR beam in FTIR is split into two by passing through an interferometer. An interference pattern is created when the beams recombine, with one moving in a fixed direction while the other moves in a different direction. An interferogram is a complicated signal that simultaneously incorporates information from all infrared wavelengths. After that, a mathematical Fourier Transform that plots intensity against frequency is used to transform the interferogram into a typical IR spectrum. The IR spectrum that is produced shows the specific wavelengths at which the sample absorbs infrared radiation. This gives a lot of information about the sample's molecular structure and composition. Because each peak in the spectrum is associated with a distinct vibrational mode of a chemical bond, functional groups can be identified and the material can be characterized. For both qualitative and quantitative sample analysis, FTIR is widely used in numerous fields, such as pharmaceuticals, chemistry, materials science, and environmental research. Constructive interference occurs when the path difference between two light beams is an integer multiple of the wavelength (). Accordingly, the two bars are in stage, and the all out power is expanded by the amount of their amplitudes.The following specifies what constitutes constructive interference:

$$2d\cos\theta = m\lambda$$

where m is an integer (0, 1, 2,...), d is the route difference, θ is the angle of incidence, and λ is the wavelength of the light.

When the path difference between the two beams is an odd multiple of half the wavelength ($\lambda/2$ ), destructive interference occurs. Because of this, the beams are out of phase, and the amplitudes of the beams cancel each other out, lowering the intensity overall. The following represents the prerequisite for destructive interference:

$$2d\cos\theta = (\mathrm{m} + 1/2\ )\lambda$$

where d is the path difference, θ is the angle of incidence, λ is the wavelength of the light, and m is an integer (0, 1, 2, ...).[112]

### *4.4.3 Sample Preparation*

In order to guarantee reliable and accurate results from Fourier Transform Infrared (FTIR) spectroscopy, sample preparation involves several steps. The sample may be a solid, a liquid, or a gas, all of which necessitate different methods of preparation. For strong examples, they are many times ground into a fine powder and blended in with a non-engrossing framework like potassium bromide (KBr), then, at that point, squeezed into a slender, straightforward pellet. An attenuated total reflectance (ATR) crystal can also be used to directly place solid samples for analysis. Typically, liquid samples are applied as a thin film on an infrared-transparent substrate or placed in a calcium fluoride-based liquid cell with transparent windows. A gas cell with long path lengths is used to analyze gaseous samples to improve absorption signals. To avoid contamination and ensure that the characteristics of the sample are accurately captured in the FTIR spectrum, proper sample preparation is essential.[113]

## 4.5 Atomic Force Microscopy (AFM)

### *4.5.1 Working Principle*

A cantilever, or pointed probe, is used in atomic force microscopy (AFM) to measure the force exerted on a sample's surface. The AFM test filters the example surface very close with a nanometer-scale range on its tip. The interactions that the tip encounters

as it travels across the surface include chemical bonding forces, electrostatic forces, and van der Waals forces, to name just a few. A laser bar bounced away from the cantilever and onto a position-touchy photodetector identifies the diversions in the cantilever brought about by these collaborations. A high-resolution topographical map of the sample surface is created using an electrical signal derived from the deflections. There are a variety of ways that AFM can work, including contact and non-contact.

Figure 4.4 AFM (BRUKER) at SCME, NUST

### *4.5.2 Important Features*

Due to a number of significant aspects, atomic force microscopy, or AFM, is a versatile and efficient technique for imaging and analyzing at the nanoscale. AFM's extremely high-resolution imaging makes it possible to resolve features at the atomic level, which is crucial for studying nanostructures and surface topography. It measures the forces that interact between a sharp probe and a sample surface in order to produce precise maps of the morphology of the surface. AFM can be used to examine a wide range of materials, including hard metals and soft biological tissues. It has contact, non-contact, and tapping modes of operation. Moreover, AFM can be utilized in various applications since it doesn't need conductive coatings or extra example planning. Additionally, the method may be used to measure mechanical, electrical, and magnetic properties of surfaces. AFM's usefulness

expands across a wide range of scientific fields, including biology, materials science, and nanotechnology, thanks to its adaptability to ambient, liquid, and vacuum conditions.

### *4.5.3 Sample Preparation*

Atomic Force Microscopy (AFM) requires sample pretreatment in order to produce imaging that is both precise and high-resolution. The process begins with selecting a suitable substrate that is clean, level, and free of impurities because the surface quality of the substrate can have a significant impact on the outcome. The sample itself must be properly prepared for accurate AFM readings, which frequently requires cleaning to remove dust, oils, or other contaminants. In order to preserve the structure of biological samples, procedures such as fixation and dehydration may be necessary. After being cleaned using adhesives or another method, the sample is typically adhered to the substrate to ensure that it remains stable during scanning. Additionally, the sample must be sufficiently thin to permit the AFM probe to scan the surface unhindered. To further develop picture quality, further measures, for example, covering with a little layer of metal might be required in certain conditions, especially with delicate or delicate materials. The sample must be properly prepared to ensure low artifacts and an accurate depiction of the surface features in order to obtain reliable data for AFM analysis.

## 4.6 Contact Angle

### *4.6.1 Working Principle*

The contact angle is a fundamental concept in surface science and refers to the angle at which a liquid interface meets a solid surface. A measurement of the solid's wettability by the liquid is this angle, which is determined by the equilibrium of intermolecular interactions between the liquid and the solid. When a droplet of liquid is placed on a solid surface, the solid-liquid interface tension (SL), the solid-vapor interface tension (S V), and the liquid-vapor interface tension (L V) all come into play. These tensions determine the contact angle ($\theta$ ), which Young's equation describes:

$$\gamma SV=\gamma SL+\gamma LV\cos\theta$$

Practically, the contact angle reveals the interaction between the liquid and the surface. When the liquid spreads out to create a small contact angle (less than 90 degrees), a thin film forms on the surface. Hydrophilic surfaces, which have a strong attraction between the liquid molecules and the solid surface, exhibit this kind of behavior. However, as the liquid forms distinct droplets on the surface, a large contact angle (more than 90 degrees) indicates low wettability. These beads are commonplace of hydrophobic surfaces, which have powerless fluid strong association.


Figure 4.5 Contact Angle (XSZ 107BN) at SCME, NUST

### *4.6.2 Important Features*

The contact angle is influenced by the liquid's nature, impurities, chemical composition, and solid's surface roughness. Contingent upon the expected use, surface medicines like coatings and finishing can change the contact point and increment or diminishing wettability. Controlling the contact angle is crucial in the industrial setting for processes like coating, painting, printing, and creating surfaces that repel water. The sessile drop method, which involves putting a drop of liquid on a solid surface and measuring and observing the angle that forms at the contact line, is the most common method for measuring the contact angle. The tilting plate method and the capillary rise method are two additional methods that are appropriate for a specific purpose and offer varying degrees of

precision and usability. Surface contacts need to be made as good as possible in many scientific and technical applications. Understanding and being able to control contact angles are required for this.

### *4.6.3 Sample Preparation*

For contact angle measurements, sample preparation is necessary to ensure accurate and reliable results. To begin, the surface must be thoroughly cleaned to remove any oils, impurities, or residues that might obstruct the measurement. To eliminate natural contaminations, the example is typically washed with solvents like CH3)2CO, isopropanol, or ethanol. Then, particles and deposits are eliminated involving ultrasonic cleaning in a dissolvable filled shower. After that, the sample is cleaned with deionized or distilled water and dried with nitrogen gas or in a clean, dust-free environment. Anything that could introduce fibers, like paper towels or cloths, is avoided. Thusly, the surface could go through specific medicines to give the characteristics expected to the application. Plasma treatment, for instance, can increase surface energy and improve wettability, in contrast to chemical treatments that use compounds like silanes to alter surface chemistry. Additionally, polymeric materials benefit greatly from thermal treatment's ability to improve surface uniformity and eliminate absorbed impurities. After these treatments, the surface is characterized to ensure that it is smooth and error-free. Optical or scanning electron microscopy (SEM) is used to check for uniformity and any scratches or imperfections on the surface. Since roughness has a significant impact on the calculations of contact angles, it is also essential to quantify surface roughness using techniques like profilometry or atomic force microscopy (AFM). Last but not least, when taking measurements, it is essential to maintain constant conditions in the surroundings. Since humidity and temperature have an effect on how the liquid behaves and how accurate the measurement is, this requires controlling them. When measurements are made in a controlled environment with constant conditions, they are more reliable and reproducible.

Figure 4.6 Brunaur-Emmett-Teller (BET)

### *4.7.1 Working Principle*

The Brunauer-Emmett-Teller (BET) theory is one of the most important ideas for analyzing and measuring material surface area, especially that of porous materials. The BET method contributes to a deeper understanding of gas adsorption on solid surfaces by expanding Langmuir's concept of monolayer adsorption to multilayer adsorption. The BET hypothesis expresses that at consistent temperature, layers of gas particles truly adsorb onto a strong surface, and how much gas adsorbed might be communicated as a component of tension.

The BET equation, which is commonly expressed as follows, connects the amount of gas adsorbed to the gas's relative pressure:

$$1/\ v(P_0/P-1)=1/vmC+C-1P/VmCP_0$$

where v is the volume of gas adsorbed at pressure P, vm is the volume of gas required to form a monolayer, $P_0$ is the saturation pressure of the gas, and C is the BET constant, which is related to the energy of adsorption in the first layer.

In point of fact, nitrogen gas is frequently utilized for BET measurements due to its well-known adsorption properties and readily available nature. For the purpose of the BET study, nitrogen gas is adsorbed onto the surface of the sample at the temperature of liquid nitrogen (-196°C). At various relative pressures, the amount of gas adsorbed is then measured. After the BET equation has been plotted and a linear relationship has been

established, the slope and intercept of the linear plot can be used to determine the monolayer capacity (vmv m). The material's specific surface area is calculated after the monolayer capacity is determined, taking into account the cross-sectional area of the nitrogen molecule. The BET method is frequently used to characterize materials such as adsorbents, catalysts, and porous materials. It offers fundamental surface region data, which is vital for fathoming and boosting material characteristics for a scope of utilizations. Because of its dependability and accuracy, BET measurements are a common method in material science and engineering.

### *4.7.2 Important Features*

The Brunauer-Emmett-Teller (BET) theory is a fundamental idea for describing material porosity and surface area. By extending the Langmuir theory to multilayer adsorption, it provides a deeper comprehension of gas adsorption on solid surfaces. It was developed in 1938. The limit of Wagered to work out a material's particular surface region is one of its essential qualities. This is significant in regions like natural designing, materials science, and catalysis. The BET equation makes a connection between the amount of gas adsorbed and the pressure by calculating the surface area with a linear plot. The gas molecules are thought to form multiple layers on the adsorbent surface, with the same mechanism controlling the adsorption of each layer. Additionally, the pore size distribution of a material is revealed by BET theory, which is crucial for applications in filtration and adsorption. BET is an essential tool for both research and industry because it can be used to analyze a wide range of adsorbents, including polymers and metals.

### *4.7.3 Sample Preparation*

Sample preparation is necessary in order to obtain accurate Brunauer-Emmett-Teller (BET) surface area analysis results. To ensure that the surface is pure for the adsorption tests, the sample is first cleaned to get rid of any impurities like dust and grease. The sample is then dried to remove any moisture, usually by heating it for a certain amount of time in an oven or using a vacuum desiccator at a temperature that is determined by the material's thermal stability. During the crucial next step, known as degassing, the sample

is heated in a vacuum or in an inert gas flow to remove any pre-adsorbed gases and vapors. To avoid altering the sample's surface properties, the degassing settings must be adjusted carefully. Through grinding and sieving, a uniform particle size can be achieved for powdered materials to ensure accurate and repeatable measurements. Solid BET investigation relies upon appropriately pre-arranged examples since lingering toxins or irregularities can unfavorably influence the accuracy of surface region and porosity values.

## 4.8 Antibacterial Testing

Antibacterial testing is important in a lot of different industries, like consumer goods, medicine, and healthcare, to make sure that chemicals used to treat bacterial illnesses are safe and effective. In light of the serious threat that antibiotic resistance poses to global health, this testing is essential for the development of novel antibiotics and antimicrobial medications. Specialists can reveal and improve synthetic compounds that effectively restrain or kill unsafe microorganisms by evaluating a substance's antibacterial capacities. Additionally, this strategy aids in lowering the risk of infection and improving patient outcomes by ensuring that medical devices, surgical instruments, and other hygiene products adhere to stringent safety standards. Consumer goods like hand sanitizers, disinfectants, and personal hygiene products are subjected to antibacterial testing to ensure that they provide the advertised antibacterial protection. When everything is taken into account, antimicrobial testing is necessary for both improving scientific and technological advancements and preserving public health.

### *4.8.1 Disc Diffusion Method*

A popular method for determining a substance's antibacterial activity is the Kirby-Bauer test, also known as the disc diffusion method. A bacterial lawn is created by uniformly spreading a bacterial culture across an agar plate using this method. The surface of the agar is then coated with the test chemical or antibiotic in the form of tiny, circular discs. The material diffuses radially from the circle into the agar while the plate is hatched. Because the concentration of the antibacterial agent is high enough to stop the bacteria from growing there, there is a clear zone around the disc known as the zone of inhibition.

The size of this zone is measured and compared to standard values to determine whether the bacteria are susceptible to the chemical that is being tested or resistant to it. The disc diffusion method is highly regarded for its ease of use, cost-effectiveness, and capacity to produce qualitative data that help select effective antimicrobial therapies..

### *4.8.2 Sample Preparation*

A lot of preparation is required for the disc diffusion method to produce results that are accurate and reliable. A number of essential steps are included in the procedure to ensure consistency and repeatability. The test organism is initially grown as a pure culture, typically on a plate of nutrient agar. Selecting a few isolated colonies from this culture and suspending them in a suitable liquid medium, such as broth or saline, to produce a standard concentration is the next step. A 0.5 McFarland standard's turbidity is frequently adjusted to match this concentration. This standardization ensures that the bacterial inoculum will remain constant throughout testing. After that, the standard bacterial suspension is dipped into a clean cotton swab. The swab is used to evenly distribute the bacteria on a Mueller-Hinton agar plate by pushing the swab against the tube's side to squeeze out excess liquid. This ensures a uniform bacterial lawn, which is necessary for precise measurement of the inhibition zone. Following the preparation of the bacterial lawn, antibiotic-impregnated discs are carefully placed on the surface of the agar using sterile forceps. The discs should be spaced equally apart to prevent inhibition zones from overlapping and to enable precise measurement. In order to encourage bacterial growth and antibiotic diffusion from the discs, the plate is incubated for 16 to 18 hours at a suitable temperature, typically 35 to 37°C. The plates are examined after the incubation period has ended, and the diameter of the inhibition zones that surround each disc is measured in milliliters. To determine the microorganisms' awareness or protection from the tried medications, these estimations are contrasted with normalized tables. Proper sample preparation is required in order to obtain accurate and understandable results from the disc diffusion method, which are essential for directing the development of successful antimicrobial therapies.

# CHAPTER 5: RESULTS AND DISCUSSION

## 5.1 Analysis of MXene, AgFeO2 and AgFeO2/MXene Composite

Material analysis of MXene and its composite with AgFeO2 is performed by Xray diffraction (XRD), Scanning Electron Microscopy (SEM), Fourier Transform Infrared Spectroscopy (FTIR), Ultraviolet Specctroscopy(UV), Brunauer-Emmett-Teller(BET), Atomic force microscopy(AFM), Contact angle.

### *5.1.1 Xray Diffraction Analysis(XRD)*

X-ray diffraction (XRD) was employed to investigate the materials' structural formation. The measurement range for the diffracted intensities was 10° to 80°. The morphological characteristics of the AgFeO2/MXene composite, MXene, AgFeO2 and MAX phase are displayed in Figure 5.1. According to the Bragg equation $2d \sin\theta = n\lambda$, the diffraction peak (002) of Ti3C2 MXene at $2\theta = 8.98$ ¸ is moved to a lower angle and shows a significant d spacing of approximately 0.98 nm. This information has been documented in literature.

The removal of the sharp Ti3AlC2 MAX phase peak (104) upon etching with hydrofluoric (HF) acid is also confirmed by the MXene pattern. Following exfoliation, XRD unmistakably demonstrates a considerable loss in crystallinity and structural order. In other cases, the original powders had trace amounts of MAX phases. Following the HF treatment, these do not react, and the full width at half-maximum (FWHM) of their peaks stays unchanged.The AgFeO2 exhibits peaks at 2θ equals $14^0$,$29^0$,$34^0$,$36^0$,$38^0$,$44^0$ and $61^0$ . XRD pattern of AgFeO2/MXene composite has clearly shown the reinforcement of both material’s peaks with each other at these specific 2θ angles.

This research demonstrates that the degree of crystallinity of the MXene as well as AgFeO2 has a significant impact on the structural features of the composite. Thus, this study has verified the successful development of the AgFeO2/MXene composite, revealing

a well-integrated hybrid structure in which the MXene matrix and AgFeO2 influences the composite's overall crystallinity.

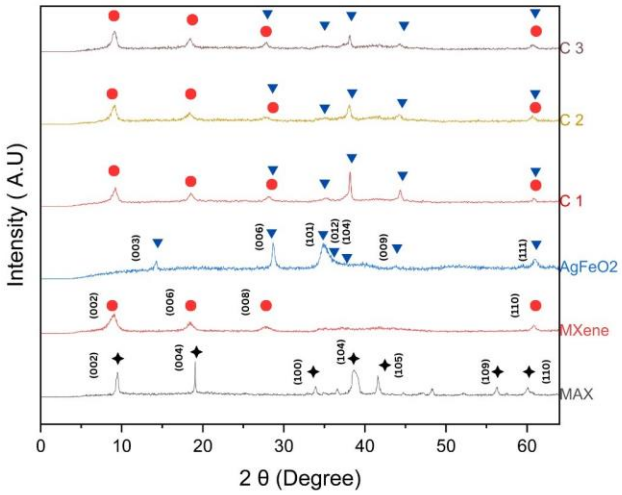



**Figure 5.1** XRD plot of MAX,MXene,AgFeO2 and AgFeO2/MXene Composites

### *5.1.2 Scanning Electron Microscope Analysis(SEM)*

Utilising scanning electron microscopy (SEM), the surface morphology of the generated samples was investigated. The morphological characteristics of MXene,AgFeO2 and AgFeO2/MXene Composites are displayed in Fig. 5.2. The multilayer crystalline structure of the MAX phase can be seen in its SEM image.(fig 5.3a)

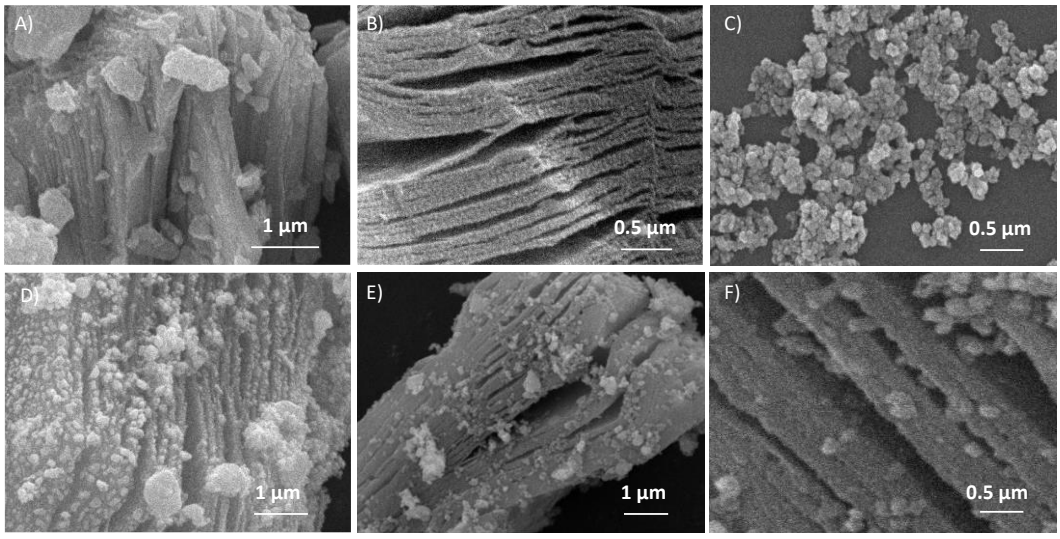



Figure 5.2 SEM images of (A)MAX phase (B)MXene (C)AgFeO2 (D,E,F)AgFeO2/MXene Composite

In (fig 5.2b), the layers are separated, generating an accordion-like shape, which provides evidence of selective etching of Al after the acidic treatment. Fig 5.2c shows AgFeO2 nanoparticles in a size range of 40-60nm.In fig 5.2c the material included two-dimensional AgFeo2/MXene and MXene nanosheets to improve stability of nanosheets and provide sticky spots for microbial attachment.Additionally , fig 5.2d shows intercalation of AgFeO2 nanoparticles inside the layers of MXene sheets .

MXene has various charged functional groups on the its surface. When MXene sheets are exfoliated, ferrite particles penetrate between them through electrostatic interactions and increase its c-lattice parameter. Fig. 5.2 d,e,f shows that the addition of nanonuts of silver ferrite reduced the restacking of MXene layers and agglomeration of copper ferrite. Nanonuts not only prevent the restacking of MXene layers but also provide sheet to sheet inter-links to boost the antibacterial activity.

### *5.1.3 Fourier Transform Infrared Spectroscopy*

The FTIR spectra of all manufactured samples were compared in the wavenumber range of 4000 cm-1 to 500 cm-1, as illustrated in Figure 5.3. Absorption peaks at 3424 cm-1 confirmed the presence of hydroxyl groups in both MXene and AgFeO2/MXene samples, implying external water, a strongly hydrogen-bonded OH, or extremely strong coordinated H2O.

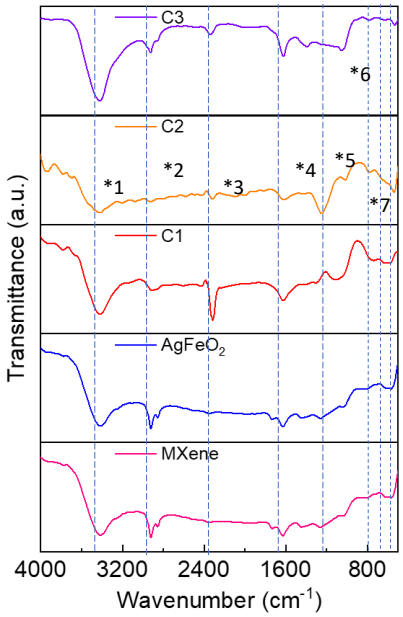



**Figure 5.3** shows a FTIR spectra of MXene ,AgFeO2 and AgFeO2/MXene in the wavenumber range of 4000 cm-1 to 500 cm-1

FTIR spectroscopy of MXenes reveals various unique peaks that provide information about their surface chemistry. An O-H stretching signal around 3200-3600 $cm^{-1}$ indicates the presence of hydroxyl groups, which are commonly supplied during synthesis or functionalization. The signal at 1650-1750 $cm^{-1}$ corresponds to C=O stretching from carbonyl groups. Peaks in the 500-800 $cm^{-1}$ range correspond to M-X bonds (where M is

a transition metal and X is carbon or nitrogen), which are essential for the MXene structure. These peaks aid in identifying the exact functional groups and bonding conditions present on the MXene surface, which are critical for understanding and customising their properties to various applications.

While FTIR spectroscopy of $AgFeO_2$ (silver ferrite) shows distinct peaks that disclose its chemical structure and functional groups. Typically, the spectrum includes large peaks about 3400 $cm^{-1}$, corresponding to O-H stretching vibrations. This indicates the presence of hydroxyl groups, possibly due to moisture absorption. Peaks near 1630 $cm^{-1}$ correspond to the bending vibrations of H-O-H bonds, indicating adsorbed water molecules. In the lower frequency band, significant peaks around 500-700 $cm^{-1}$ are attributed to the metal-oxygen stretching vibrations (Ag-O and Fe-O), which are critical for recognising the metal-oxide framework of $AgFeO_2$. These peaks provide insight into the bonding environment and structural aspects of $AgFeO_2$, facilitating research of its properties and applications.

Finally if we observe the AgFeO2/MXene composite spectra we can see all peaks .

### 5.1.4 Brunauer-Emmett-Teller

**Brunauer-Emett-Teller(BET)** The specific surface area of MXene and its composite with AgFeo2 was largely determined through theory, which is essential for comprehending their reactivity, catalysis, and adsorption capacity. Additionally, it provides information about pore volume and distribution, which is useful for applications like filtration, catalysis, and gas storage, and it aids in the analysis of material porosity. In addition, BET is a well-known method for characterizing materials, assisting researchers and engineers in maximizing the qualities of materials for a variety of industrial applications, specifically enhanced antibacterial activity in my case.

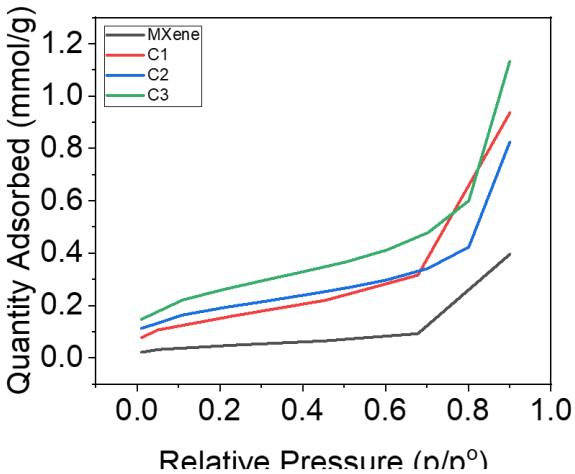



**Figure5.4** Isotherm of physical nitrogen sorption on the surface of MXene and AgFeO2/MXene

If we pay attention to the shapes of Isotherms in Fig 5.4 ,it can be clearly observed that adsorption of MXene is much more less then that of AgFeO2/MXene composite.The measured single point in surface area of AgFeO2/MXene composite is 12.0237m/g ,while the single point in pore volume is calculated less than 20.032499cm/g and pore size for adsorption is calculated to be 104.7968nm. While if this all is compared to MXene ,the single point in surface area is 3.6236m/g, while single point in pore volume has been calculated is less than 20.0137cm/g and pore size for adsorption is calculated to be 146.3932nm. The specific surface area rose from 3.62 to 20.0($m^2$/g) with the addition of AgFeO2 in MXene layers and on the surface. With a composition of 2:1,the surface area rose to 11.0146($m^2$/g), with composition of 6:1 ,it became 14.5786($m^2$/g) and then finally at 10:1 ,it reached to 20.0404($m^2$/g) .Thats highest area achieved in our research .It may be responsible for the increased surface area, which leads to greater number of active sites and enhanced surface area.That is key to enhanced antibacterial activity.

Hence surface area of AgFeO2/MXene composite has been increased compared to MXene and site of attachements are increased which in turn increases surface activity of material and caused enhanced antibacterial activity in our case.

### 5.1.5 Contact Angle

Contact angle of water on bare, MXene ,AgFeO2/MXene is taken to demonstrate hydrophilicity of materials(Figure 5). Mxene generally have hydrophililic nature.[23]Pure Ni-foam has a hydrophobic surface due to the absence of oxygen-rich groups and hinders bacterial attacments.The -O and -F groups on the surface of MXene@ Ni-foam make it hydrophilic and angle decreass from 119.97 $^{0}$ to 56.88$^{0}$. The water contact angle of AgFeO2/MXene@Ni-foam again showed a decline from 56.88$^{0}$C to 50.08$^{0}$ by addition of AgFeO2 nanoparticles that can be clearly seen in (figure 5). This means surface properties are more enhanced by adding AgFeO2 nanoparticles in MXene ,as more surface groups are added that facilitates more bacterial attachment which in turn increases in enhanced antibacterial activity. Thus, AgFeO2/MXene composite is improving hydrophilicity.

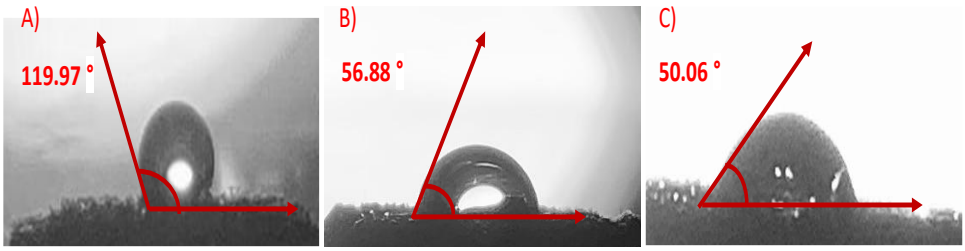



**Figure 5.6**:Contact angle measurement of A)Bare GF B)MXene@GF C)AgFeO2/MXene@GF

### 5.1.6 Zeta Potential

For surface charge analysis , zeta potential was done at 25$^{0}$C using DI water as solvent ,at pH 5,that actually influence the material's interaction with bacteria . As bacterial cell membrane are negatively charged(ranging from (-60 – 150) ,due to potential difference they are attracted towards material. From zeta potential analysis ,it was analyzed that Mxene had surface charge of 1.3 mV while AgFeo2 carried 2.07 mV charge ,but when they were combined together, $AgFeO_2$/MXene carried 2.34 mV .So, surface charge has

increased ,more potential difference was produced ,more bacteria were attracted towards composite material and it resulted in enhanced antibacterial activity .

### 5.1.7 Antibacterial Activity

Two tests were performed to check the antibacterial activity of our powder material i.e, MXene ,AgFeO2 and their five composites with different ratios against bacterial strains of E.Coli and S.aureus .

### 5.1.7(a) Disc Diffusion Method

Before everything was autoclaved, these steps were taken to apply the disc diffusion method to E. coli and S. aureus: First, make a lawn culture by uniformly swabbing a standardised bacterial suspension of E. coli or S. aureus across the surface of Mueller-Hinton agar plates. The plates need a few minutes to dry. Then, utilizing sterile forceps, embed anti-infection impregnated circles on the outer layer of the agar, it are all around dispersed separated to make that they. Delicately press the plates to guarantee they adhere to the agar. For 18–24 hours, incubate the plates at 37°C. Use a ruler or callipers to measure the diameter of the inhibition zones surrounding each disc following incubation. Contrasted these actions with traditional reference values to decide whether the microorganisms are defenseless or impervious to anti-infection agents.Dilutions were made in 5mg,10mg and 15mg/ml of all seven samples in autoclaved glass voils to avoid contamination.The composite powder with ratio 10:1 of MXene and AgFeO2 showd maximum antibacterial activity.The figures below shows the inhibition zones for powdered samples in dilution forms against E.Coli and S.aureus.

| Sr. No. | Strain | Zone of Inhibition with Augmentin (mm) | Conc. mg/ml | Zone of Inhibition with MXene (mm) | Zone of Inhibition with $AgFeO_2$ (mm) | Zone of Inhibition with C1 (mm) | Zone of Inhibition with C2 (mm) | Zone of Inhibition with C3 (mm) | Zone of Inhibition with C4 (mm) | Zone of Inhibition with C5 (mm) |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | E.Coli | 1.6 | 5 | 0.5 | 0.6 | 0.5 | 0.5 | 0.8 | 0.9 | 1 |
| | | | 10 | 0.7 | 0.8 | 1 | 0.9 | 1.1 | 1.1 | 1.2 |
| | | | 15 | 0.8 | 0.7 | 1.1 | 1.2 | 1.5 | 1.4 | 1.4 |
| 2 | S.aureus | 2 | 5 | 0.3 | 0.9 | 0.4 | 0.3 | 0.3 | 0.5 | 1.5 |
| | | | 10 | 1.4 | 1.5 | 1.8 | 1.8 | 1.8 | 1.8 | 1.7 |
| | | | 15 | 1.5 | 1.7 | 1.9 | 1.9 | 1.9 | 2.1 | 2.5 |

Table 5.1 Shows comparison of inhibition zones with prepared samples).

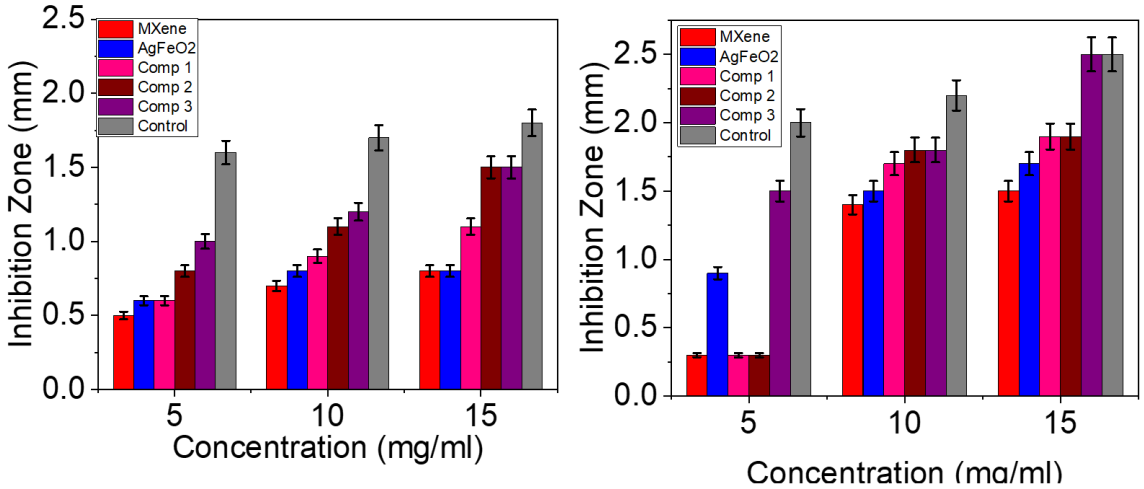



**Figure 5.7** Effect of MXene ,AgFeO2 And AgFeO2/MXene composites on the growth of E.Coli & S.aureus

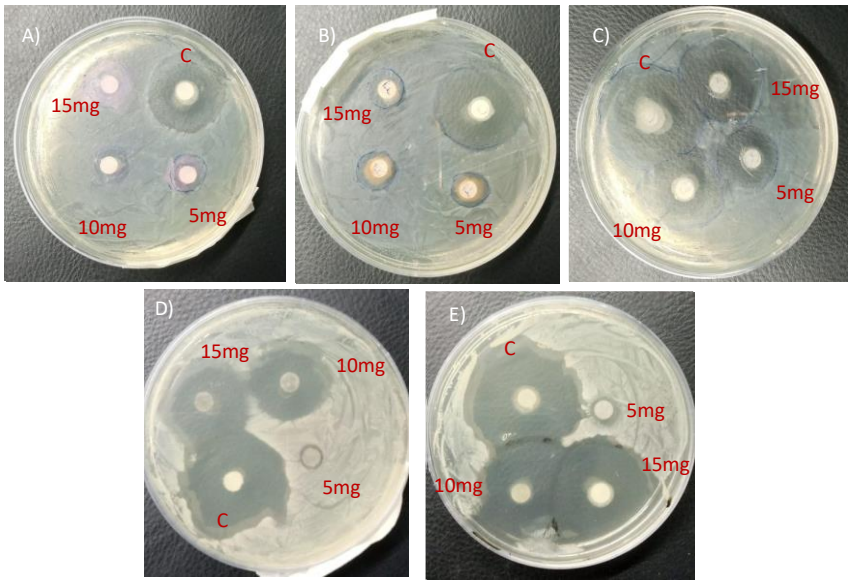



**Figure 5.6** Inhibition zones by MXene ,AgFeO2 And AgFeO2/MXene composites on the growth of S.aureus

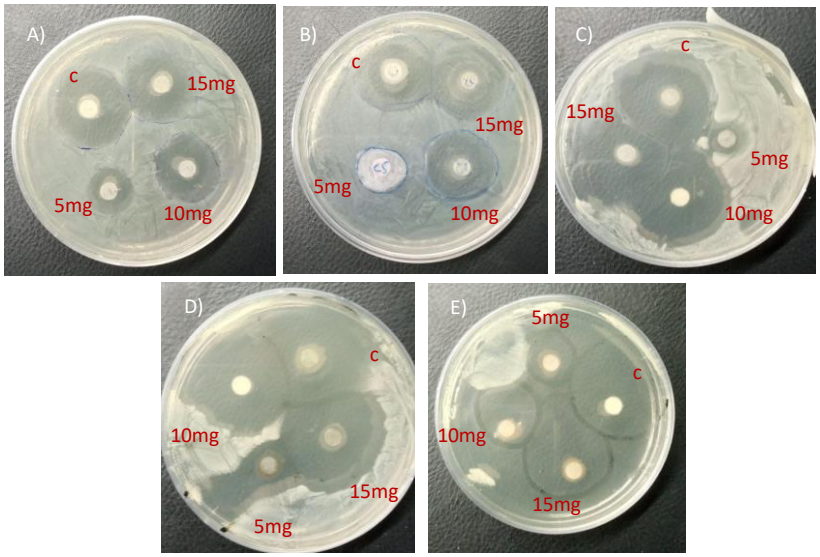



**Figure 5.7** Inhibition zones by MXene ,AgFeO2 And AgFeO2/MXene composites on the growth of E.Co

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