Training Report

On

Anti-Microbial Ligand Testing

Carried out at

Atmiya University of technology – rajkot

Under (the trainer):

DR. NutanParkash Vishwakarma

(Head of department of biotechnology – **atmiya University**)

By

MR. Kishan C. Aghara

- **ACKNOWLEDGEMENT**

It’s great to hear that you had supportive mentors and friends during your internship! Here’s an example of an acknowledgement you could use:

“I would like to express my sincere gratitude to DR. Nutanprakash Vishwakarma sir my mentor during this internship, for their guidance, support, and encouragement throughout the entire process. Their expertise and insights were invaluable, and I learned so much from them.

I would also like to thank my friends Bhavya Parmar , Shreya Kathiriya, Anjaliba Jadeja ,Aashta Patel , Faizan Noida , who were always there to listen, share advice, and offer a helping hand when I needed it. Without their support, this experience would not have been as meaningful or enjoyable. Thank you all for your contributions to my growth and development.”

- **Preface**

Clinical Laboratory practice/internship is an integral part of the final year program and is designed to provide interns with an opportunity to integrate and apply Previously acquired. Knowledge and technical skills in actual clinical settings

This internship booklet is prepared with the intention to provide orientation to interns about various tasks to be performed and/or observed in different disciplines during 120 hours internship at the lab. The main goal of the internship is to acquire necessary practical skills in performing various laboratory tests in different disciplines at laboratory medicine department in lab that will contribute directly to efficient laboratory diagnosis and improve health care services.

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- Intern introduction

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Introduction:

Anti-Microbial Ligand Testing

Anti-microbial testing of synthesized ligands is an important aspect of drug discovery and development. It involves the screening of synthesized ligands for their ability to inhibit the growth of microorganisms, such as bacteria and fungi, that cause infectious diseases. The aim of this testing is to identify ligands with potent anti-microbial activity that can be further optimized to develop new drugs.

The process of anti-microbial testing typically involves exposing the synthesized ligands to various strains of microorganisms and measuring their ability to inhibit or kill the microorganisms. The concentration of the ligands required to achieve these effects is also determined. This information is used to generate a dose-response curve, which can be used to evaluate the potency of the ligand.

Several methods can be used to test the anti-microbial activity of synthesized ligands, including agar diffusion, broth dilution, and micro dilution assays. These methods vary in their complexity and sensitivity, but they all aim to determine the minimum inhibitory concentration (MIC) of the ligand, which is the lowest concentration that completely inhibits the growth of the microorganism.

Anti-microbial testing of synthesized ligands is an important step in the drug discovery and development process, as it allows researchers to identify promising candidates for further development. It also helps to guide the optimization of the ligand's structure and properties to improve its anti-microbial activity and minimize its toxicity. Ultimately, this testing can lead to the development of new drugs that are effective in treating infectious diseases and improving public health.

Equipment tour

- Shaker incubator

The incubation process involves maintaining a stable temperature, humidity, and other environmental conditions within the shaker incubator. This is achieved through the use of a heating system, a thermostat, and a humidifying system that work together to maintain the desired environmental conditions

- Auto clave

Used for sterilizing equipment and media by subjecting them to high pressure and temperature. The principle of an autoclave is based on the combination of two basic processes: moist heat and high pressure.

The moist heat process involves subjecting the material to be sterilized to high temperatures in the presence of water vapor. This is typically achieved by filling the autoclave chamber with water and heating it to a temperature of 121°C or higher. The water vapor generated under pressure by the heating process penetrates the material being sterilized and kills microorganisms by denaturing their proteins, nucleic acids, and other cellular components.

The high-pressure process involves pressurizing the autoclave chamber to create an environment in which the boiling point of water is elevated above 100°C. This is achieved by using a pressure vessel, which allows the pressure to build up inside chamber. The high pressure ensures that the water vapor is distributed evenly throughout the material being sterilized, thereby promoting effective sterilization

- Microtiter plate reader:

The basic principle of absorbance measurement involves the absorption of light by molecules in the sample. The microplate reader shines a beam of light through the samples in the microplate, and the amount of light absorbed by the sample is measured by a detector. The amount of light absorbed is proportional to the concentration of the molecules in the sample, allowing the microplate reader to measure the concentration of the sample.

The basic principle of fluorescence measurement Involves the emission of light by molecules in the sample. The microplate reader excites the fluorescent molecules in the sample by shining a specific wavelength of light on them. The fluorescent molecules absorb the light and then emit light at a longer wavelength. The microplate reader detects the emitted light and measures its intensity, allowing the microplate reader to measure the concentration of the fluorescent molecules in the sample

Eppendorf's tube :

Micropipette :

- Media & culture preparation

- MH Broth Agar Plate (2%) Preparation for Microbe Isolation

MH Media add 2% weight of agar for required amount of media depending on sub culture requirements

- Culture Preparation

Take isolated bacteria from plate and inoculate it in MH Broth

Incubate it at optimal temperature37°c rotation at ~ 110 RPM for 24 hrs for microbes and for fungus 72 hrs at 32° c ~110 RPM

- Drug preparation

Weigh amount of drug and add DMSO in such measurement that there is 100 mg Drug per 1000 µl of DMSO.

- Procedure

- Broth Serial Dilution: in this step we perform serial dilution of sample drug in broth inside Eppendorf's.

1. In first tube we add 1000 µl broth and next 4 tubes 500 µl.

2. Next we add 10µl drug dissolved in DMSO to tube 1 and mix it well with pipette

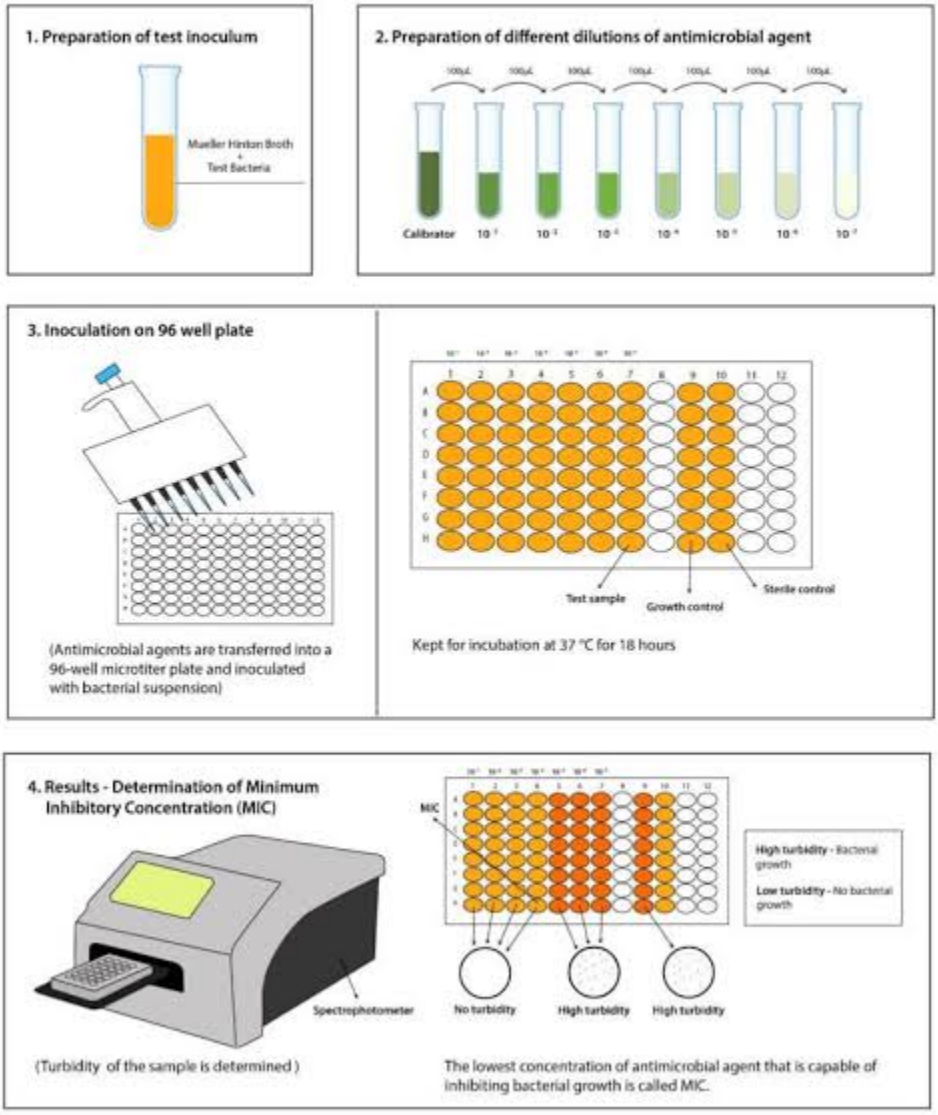
3. We collect 500 µl of broth from tube 1 and add it to next tube and repeat the step till last of broth tube which will serially dilute the drug concentration.

4. From the last tube we remove the excess 500 µl broth and discard it.

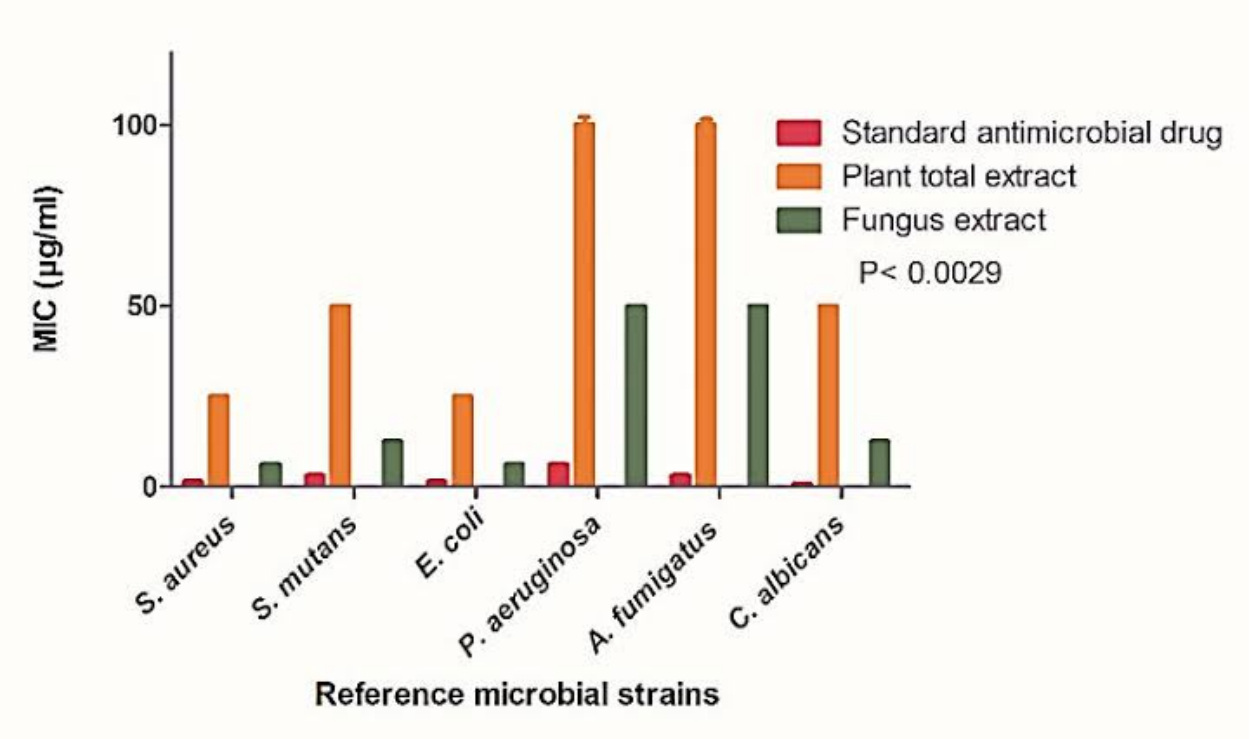
- Microbial Inoculation: in this step we inoculate our desired species of microbe in broth to check its growth inhibition from our sample drugs activity.

Used bacteria

Minimum Inhibitory Concentration Test



Result


MIC (µg/ml)
100
50
0
Standard antimicrobial drug
Plant total extract
Fungus extract
P< 0.0029
S. aureus
S. mutans
E. coli
P. aeruginosa
A. fumigatus
C. albicans
Reference microbial strains

Conclusion:

Overall, the drug testing internship program was a highly beneficial experience for me . The program provided me with hands-on experience in various drug testing methodologies, regulatory compliance, and legal aspects of drug testing. Additionally, the program allowed me to work with industry experts, which enhanced my knowledge and skills.

I am submitting this report to Department Of Biotechnology in my six semester (2020-23),All the work and information I leaned during this internship program is submitted in this training report

Thank you,