

# Practical Biotechnology

## Lab 4

### Well diffusion method

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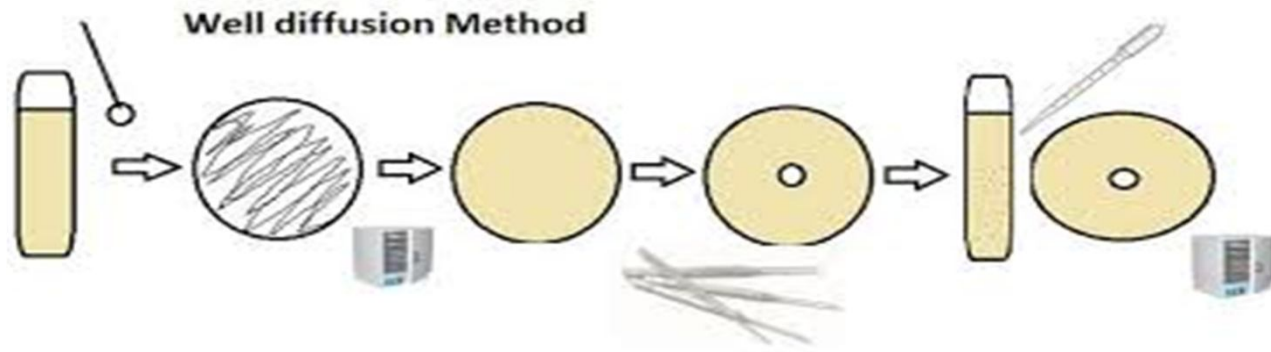
MSc. In molecular Biology



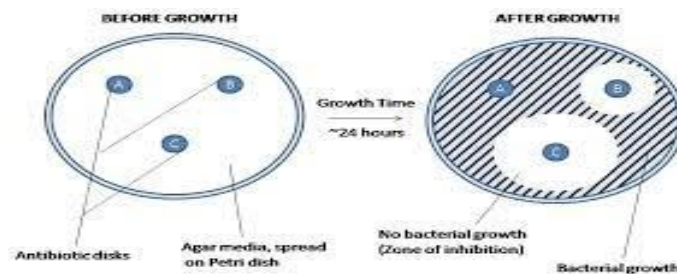
## Well diffusion method

Agar diffusion test is the primary method to determine the antimicrobial activity of the nanofibrous scaffolds. It is important to mention here that it is only suitable for the diffusive test materials.

The agar diffusion test is qualitative, easy to perform, and simple.



The methodology includes the inoculation of bacterial cells on nutrient agar Petri dishes and test samples are laid over these dishes. Afterward, the dishes are incubated for 18–24 h at 37°C and thereafter, The presence of antimicrobial activity is indicated by the absence of bacterial growth directly below the test sample.



## Procedure:

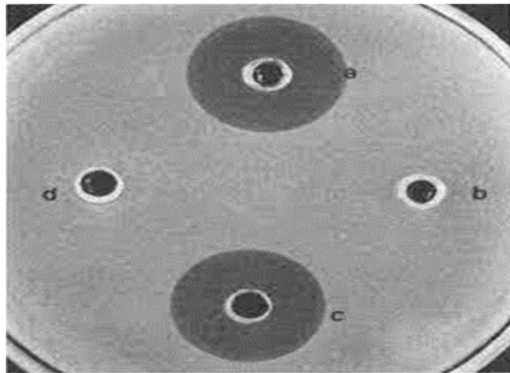
the well diffusion assay is performing in **Muller Hinton agar** . Similarly to the procedure used in disk-diffusion method, the agar surface plate is inoculated by spreading a volume of the microbial inoculum over the entire agar surface (lawn culture) and make the wells in the **MHA** with around 6-8 mm in diameter.



The wells are puncture with a sterile cork borer or with the sterile blue micropipette tip.

Then add the bacteria which isolated from soil to the wells ,then incubate at 37 C for 24 -48 hours.

Then we look for the appearance or absence of the growth inhibition zone.



Thank you for  
listening



Q1/ ...Explain Artificial Competence Method?

Q2/...what is the Principle of Competent Cells?

Q3/... what are the Functions of Trypsin?

Q4/... what are the Applications of Plasmids?

Q5/ ... Define electropermeabilization?

Q6/ ...How do you prepare the Competent cells?

Q7/ ...Explain heat shock treatment method?

Q8/ ... How can you prepare the Competent cells?