

Introduction

Antibiotic comes from the word 'antibiosis' which means 'against life' (Etebu & Ariekpar, 2016, p. 90). Antimicrobials are produced either partly or as a whole by use of synthetic methods (Etebu & Ariekpar, 2016, p. 90). Both antimicrobials and antibiotics have been used over the years to cure diseases caused by microorganisms such as *Staphylococcus*, *Mycobacterium*, *Pseudomonas* and *Streptococcus*. Antibiotics are antimicrobials of low molecular weight which are produced by microorganisms that kill or inhibit other microorganisms. Some of these antibiotics include; penicillin, tetracycline, and cephalosporin. However, there has been a setback in the field of medicine in creating antibiotic-resistant strains of some bacteria for instance, *Neisseria gonorrhoeae*, *Mycobacterium tuberculosis*, *Pseudomonas aeruginosa*, and *Staphylococcus aureus*. (Tertera, 2010) (Citation)

It has been observed that some antimicrobial agents are more sensitive to a certain type of bacteria compared to others. Some antimicrobial agents which are narrow in spectrum are more effective against gram-positive bacteria while others are more effective against gram-negative bacteria. However, broad-spectrum antimicrobials are effective against both gram-positive and gram-negative microorganisms. The broadness or narrowness of spectrum of a certain antimicrobial agent depends on its mode of action and its ability to be transported into the cell. (Ref.)

These mechanisms include, inhibition of protein synthesis, inhibition of cell synthesis, breakdown of cell membrane structure or function, and inhibition of the structure and function of nucleic acids (Wright, as cited in Etebu & Ariekpar, 2016, p. 96). The Kirby-Bauer method is used to determine the sensitivity of pathogenic bacteria to an antibiotic or an antimicrobial in order to help a physician select the best option for their patients (Hudzicki, 2009, para. 5). This method is simple, reliable and takes the shortest time possible to yield results.

Are antibiotics effective in the treatment of *Staphylococcus epidermidis* and *Bacillus megaterium* (*B. meg bacteria*)? Which antibiotic is the most effective? The main objective of this experiment is to find out the sensitivity of *Staphylococcus epidermidis* and *B. meg bacteria* to various antibiotics using the Kirby-Bauer method. After determining the sensitivity of these bacteria, then the most effective antibiotic will be determined.

Bacillus spp. have been used in many applications in the biotechnological field due to their ability to produce antimicrobial agents which inhibit or kill pathogenic bacteria (Barbosa et al., as cited in Adimpong et al., para. 2). There are few articles on *Bacillus spp.* and it was established that a few isolated strains are resistant erythromycin and lincomycin (Barbosa et al., as cited in Adimpong et al., para. 2) while other strains have shown resistance to tetracycline and streptomycin (Senesi et al., as cited in Adimpong et al., para. 2). These few cases show the *B. meg* belonging to *Bacillus spp.* might have a few antibiotics which can cure it because it is a rare bacterium which has not been investigated on a lot.

Staphylococcus epidermidis is a natural pathogen found on skin and this makes them the most common cause of infections in burn populations (Gallagher, Williams-Bouyer, Villarreal, Heggors & Herndon, 2007, para. 1). Antibiotics like nafcillin and methicillin were able to treat those infections (Gallagher et al., 2007, para. 2). These studies among others show that *Staphylococcus epidermidis* can be treated by various antibiotics. The antibiotics of choice for the two bacteria might be hard to predict theoretically and thus the need for this experiment.

You need to follow the notes 2.0 - 2.7

Objective

Hypothesis

Materials and Procedure

The materials required for this experiment include; 1 petri plate of Mueller-Hinton II agar, nutrient broth cultures (with swab cotton), disk dispenser, and cartridges of disks, forceps, Bunsen burner, zone interpretation charts and metric ruler.

} no list

The following antibiotics were used for this experiment; bacitracin 10 IU, amoxicillin/clavulanic acid 30 μ g, novobiocin 30 μ g, tetracycline 30 μ g, erythromycin 2 μ g, and penicillin 10 IU. The pathogenic bacteria considered for this case are *Staphylococcus epidermidis* and *B. meg.*

Kirby-Bauer test is performed by uniformly streaking a standardized inoculum of the test organism on the Mueller-Hinton II agar whose pH should be between 7.2-7.4 and should be poured to a uniform thickness of 4 mm in the petri plate. Then paper disks containing specific concentrations of the antibiotics are deposited on the agar surface. The antibiotic diffuses out from the disk into agar, forming a concentration gradient. If the antibiotic inhibits or kills the test organism, there will be a zone of inhibition around the disk where no growth takes place. This zone varies depending on the type of medium, size of inoculum, diffusibility of the agent among other factors. Inoculation of the surface of the medium is achieved by use of cotton swab after expressing excess fluid from the swab by pressing and rotating the swab against the inside walls of the tube above the fluid level. The agar surface is given around five minutes to dry before applying disks. The disks are dispensed in two ways; in the case of an automatic dispenser, the lid from the plate is removed, the dispenser is then placed over the plate and pushed down firmly on the plunger. The disks are then tapped by use of sterile forceps to secure them firmly to the medium. The other way involves use of forceps which must be sterilized first. The disks are kept

THE ROLE OF ANTIBIOTICS

at a distance of about 15 mm from the edge of the plate and secured to the medium by use of minimum pressure using the sterile forceps. After 16-18 hours of incubation at a temperature of 37 degrees Celsius, the plates are observed and diameters of the zones of inhibition measured to the nearest millimeter. The obtained diameters are then compared to those in a table which are based on values obtained for American Type Culture Collection (ATCC). The cultures are classified as resistant, sensitive, or intermediate in their response to the antibiotic. These classifications are gotten from the comparison to the response of the reference culture ("Antimicrobial sensitivity testing," n.d., p. 121-129).

check our exp. 7 days 25°C

Check 3.0 - 3.10

Results

The different antibiotics were tested against the two bacteria and the diameters of zones of inhibition recorded as shown below. The second and third row of Table 1 each have three sets of data taken at different times. In order to analyze this data well, then the average of the three sets of data is found and recorded in Table 2 shown below also.

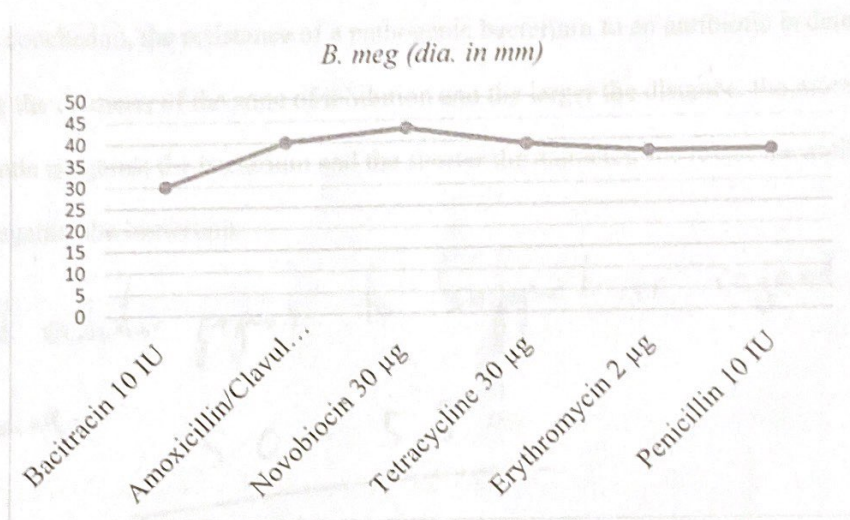
Table 1: Diameters of zone of inhibition for the antibiotics against the bacteria.

Antibiotics	Staphylococcus epidermidis	B. meg (diameters in mm)
	(diameters in mm)	(diameters in mm)
Bacitracin 10 IU	22, 26, 30	30, 30, 30
Amoxicillin/Clavulanic Ac. 30 µg	64, 70, 70	42, 38, 40
Novobiocin 30 µg	82, 98, 88	44, 42, 42
Tetracycline 30 µg	98, 102, 86	40, 40, 38
Erythromycin 2 µg	86, 94, 90	36, 40, 36
Penicillin 10 IU	62, 66, 60	40, 36, 36

Follow 4.0 - 4.7

Not necessary.

Graph 2: Effects of antibiotics on *B. meg* bacterium.



Conclusion and Discussion

From Graph 1 above, it is observed that tetracycline antibiotic had the largest diameter of zone of inhibition (95 mm) while the bacitracin antibiotic had the smallest diameter of zone of inhibition (26 mm). From Graph 2, novobiocin had the largest diameter of zone of inhibition (43 mm) while bacitracin had the smallest diameter of zone of inhibition (30mm).

This should be in results.

Thus, the most preferable antibiotic for *Staphylococcus epidermidis* is tetracycline with bacitracin being the least favorite. For *B. meg* bacterium, the most effective antibiotic against it is novobiocin and the least effective antibiotic is bacitracin.

From the two pathogens provided, *Staphylococcus epidermidis* is more sensitive to antibiotics as compared to *B. meg*. This is easily seen with the difference in diameters of the zone of inhibition. These findings contradict Bukhari's work which stated that *Staphylococcus epidermidis* has a high resistance to the antibiotics as compared to as other bacteria (2004, para. 5)

In conclusion, the resistance of a pathogenic bacterium to an antibiotic is determined by measuring the diameter of the zone of inhibition and the larger the distance, the more effective the antibiotic is against the bacterium and the shorter the diameter, the lesser the antibiotic is effective against the bacterium.

You need scholar papers to support or reject your findings

5.0 - 5.8

Add Tortora textbook.

References

- Adimpong, D.B., Sorensen, K.I., Thorsen, L., Stuer-Lauridsen, B., Abdelgadir, W.S., Nielsen, D.S.,...Jespersen, L. (2012). Applied and environmental microbiology: *Antimicrobial Susceptibility of Bacillus Strains Isolated from Primary Starters for African Traditional Bread Production and Characterization of the Bacitracin and Bacitracin Biosynthesis*, 78(22), 7903-7914. <https://doi.org/10.1128/AEM.00730-12>
- Benson's Microbiological Applications: Laboratory in General Microbiology. (n.d.).
Antimicrobial Sensitivity Testing: The Kirby-Bauer Method (13th ed., pp. 231-241)
- Bukhari, M. (2004, September 27). *Staphylococcus epidermidis*. Retrieved from:
<http://web.uconn.edu/mcbstaff/graf/Student%20presentations/S%20epidermidis/sepidermidis.html>
- Etebu, A., Arikekpar, I. (2016). Antibiotics: Classification and mechanism of action with emphasis on molecular perspectives. *International Journal of Applied Microbiology and Biotechnology Research*, 4, 90-101. www.bluepenjournals.org/ijamr
- Gallagher, J.J., Williams-Bouyer, N., Villarreal, C., Hegggers J.P., & Herndon, D.N. (2007). *Treatment of infection in burns*, 12, 136-176. <https://doi.org/10.1016/B978-1-4160-3274-8.50015-5>
- Hudzicki, J. (2009). *Kirby-Bauer disk diffusion susceptibility test protocol*. Retrieved from:
<https://www.amscience.org/content/education/protocol/protocol.3189>