A comparative study on antimicrobial potency of amino glycoside antibiotics against skin infections

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Abstract: The present study aimed to investigate the comparative antimicrobial potential of aminoglycoside antibiotics against skin infecting pathogens. Five different antibiotics of the class namely gentamicin, kanamycin, amikacin, streptomycin and tobramycin were tested for their antimicrobial effectiveness against six severe dermatogens such as Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli, Micrococcus luteus, Staphylococcus epidermidis and Candida albicans. The results of the antimicrobial assay showed that among all the tested antibiotics streptomycin, kanamycin, gentamicin and amikacin revealed highest antimicrobial activity against all the tested pathogens at equivalent antibiotic concentration. The zone of inhibition (IZD) was 42.76 mm for Amikacin against staphylococcus aurues. The zone of inhibition against Escherichia coli was highest for Streptomycin (IZD: 36.66 mm), whereas for Staphylococcus epidermidis the higher zone of inhibition was observed for Kanamycin (IZD: 37.76 mm) than other tested amino glycosides. However, the antimicrobial activity of the antibiotics was much poor on the tested fungi. Among all tested amino glycosides, only Kanamycin was found to exhibit reasonable antifungal activity (IZD: 11.3 mm) than other antibiotics. In this study, we probably for the first time, have reported the comparative antimicrobial analysis of some selected aminoglycosides on the skin infecting pathogens, responsible for very severe human skin infections like furuncles, staphylococcal scalded skin syndrome, impetigo, ethyma, folliculitis, osteomyelitis, acne, cholecystitis, bacteremia, candidiasis etc. Thus, the study may be helpful to provide some essential information regarding antimicrobial efficacy of aminoglycoside class of antibiotics against severe human skin infecting pathogens.

Key words: Aminoglycosides, Zone of inhibition, Skin infecting pathogen.

INTRODUCTION

Aminoglycosides are highly potent, broad-spectrum antibiotics with many desirable properties for the treatment of life-threatening infections.¹ The term aminoglycoside is derived from the chemical structure of these compounds, which are made up of amino groups (−NH₂) attached to glycosides (derivatives of sugar). Amino glycosides have a long history of use as effective antimicrobial agents to combat different skin infections. Though many reports are available regarding the bactericidal activity of aminoglycoside antibiotics however, a comparative study of antimicrobial potency of different aminoglycosides against skin infecting pathogens is yet to be reported in a systematic way. Again, the comparative antifungal activity reports of them are further scarce. Thus, the present study was aimed to carry out the evaluation of antimicrobial activity of five different amino glycoside antibiotics against different strains of bacteria (both gm +ve and gm -ve) and fungi. The microorganisms used under study include Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli, Micrococcus luteus, Staphylococcus epidermidis and Candida albicans. These pathogens are the causative agents for severe human skin infections like furuncles, staphylococcal scalded skin syndrome, impetigo, ethyma, folliculitis, osteomyelitis, acne, cholecystitis, bacteremia, candidiasis etc.²³ The first line of treatment strategy usually includes different antimicrobials among which amino glycoside class of antibiotics such as gentamicin, kanamicin, amikacin etc. hold an important clinical status. In the study, we first determined the minimum inhibitory concentration (MIC) by broth dilution method. The MIC value helped to assess the minimum concentration at which microbial growth was inhibited. To determine the zone of inhibition for each antibiotic, agar well diffusion method was adopted. The concentration of each antibiotic was kept constant as 5g/mL. In this study we have evaluated the antimicrobial effectiveness of some selected aminoglycoside antibiotics against dermal pathogens. This comparative analysis results gave us a clear idea of their efficacy against the most prevalent dermal pathogens which in itself an unexplored interesting research finding.

MATERIAL AND METHODS

Materials

All the micro-organisms used in the study were purchased from the institute of microbial technology, Chandigarh, India. All the media were purchased from Hi-media.

Experimental

The stock culture for bacteria was maintained on nutrient agar slants at 4°C. The fungus under the study was maintained on Sabroad dextrose agar (SDA) slants at refrigerated temperature. For antimicrobial assay, bacterial and fungal cultures were grown from stock culture on...
nutrient agar media and on SDA media respectively. The fresh sub-cultures of micro-organisms were prepared by inoculation of each bacterial strain into 10ml of nutrient broth & fungal strain into the Sabroud dextrose broth (SDB). Incubation was performed at 37°C for 24h for bacteria and 30ºC for 48h for fungi.5,6 Cell densities of approximately 1×10⁷ CFU ml⁻¹ were prepared from the cultures for the analysis.

**Determination of minimum inhibitory concentration (MIC)**

At first, MIC was determined for the selected bacteria to have an overall idea on the effectiveness and toxicity of aminoglycosides on the tested microbes before disc diffusion analysis. In short, MIC is the minimum concentration at which no such visible turbidity observed in the test tubes. In this method, the broth dilution technique was utilized where antibiotic (Gentamicin) was prepared to the highest concentration of 50µg/ml (stock concentration) in distilled water and then serially diluted to different working concentrations. About 1ml suspension of the test organisms were inoculated in about 5 ml fresh nutrient broths. Then the test tubes were introduced with varying concentration of antibiotic. After 18 hours of incubation at 37°C, the test tubes were observed for turbidity.7,8 The concentration at which, no visual turbidity observed, was denoted as MIC.

**Assay of antimicrobial activity from zone of inhibition measurement**

The amino glycide antibiotics were tested for their antimicrobial activity by agar well disc diffusion method. The diameter of zone of inhibition obtained for different antibiotics on the tested microorganism was measured. In this method, the pre-sterilized petridish containing 15 ml of Muller Hilton Agar (MHA) for bacteria or SDA for fungi, were aseptically introduced with 0.2 ml of 24h broth culture of microbial strains and evenly spread using a bent sterile glass rod. A sterile cork borer was used to cut wells of 6mm diameter in each of plates. Suitable dilutions of antibiotics were made with distilled water and carefully placed in each well by micropipette. The plates were incubated at 37ºC for 24 h and at 30ºC for 48 h, for bacteria and fungi respectively. Antimicrobial activity was evaluated by measuring the inhibition zones. Inhibition zones were recorded as the diameter of no growth area, including the diameter of the well after the post incubation period. The dilution medium for the positive control was sterile water for injection. All the samples were tested in triplicate.

**RESULTS AND DISCUSSION**

<table>
<thead>
<tr>
<th>Aminoglycoside antibiotics</th>
<th>Inhibition Zone Diameter in mm</th>
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<tbody>
<tr>
<td></td>
<td>S. aureus</td>
</tr>
<tr>
<td>Gentamicin</td>
<td>23.66</td>
</tr>
<tr>
<td>Amikacin</td>
<td>42.76</td>
</tr>
<tr>
<td>Streptomycin</td>
<td>41.06</td>
</tr>
<tr>
<td>Kanamycin</td>
<td>26.5</td>
</tr>
<tr>
<td>Tobramycin</td>
<td>41.86</td>
</tr>
</tbody>
</table>

**Table 1: Comparative assay results of amino glycosides.**

![Fig-1: Zone of inhibition of Amikacin against S. aureus.](image1)

![Fig-2: Zone of inhibition of Kanamycin against S. epidermidis.](image2)
The antimicrobial assay results of all the tested antibiotics were given in the table 1. Corresponding bar diagrams of IZDs was also given for easier comparison (Figure 3). We have provided pictures of zone of inhibition of two antibiotics only (Amikacin, Kanamycin), having highest reported IZD value. Since in the study, we had to compare the antimicrobial potency of the amino glycosides against different tested pathogens,

From the test results, it has been observed that all tested antibiotics exhibited excellent activity against all most all the strains of bacteria under study. However result of antifungal activity was very poor. Among the tested amino glycosides, Gentamicin showed zone of inhibition of 23.66 mm against S. aureus followed by Tobramycin (IZD: 41.86 mm), Amikacin (IZD: 42.76mm), Streptomycin (IZD: 41.06mm), Kanamycin (IZD: 26.5mm). In case of P. aeruginosa the zone of inhibition was larger for Tobramycin (IZD: 39.83mm) than other antibiotics.

The zone of inhibition against E. coli was highest for Streptomycin (IZD: 36.66mm). Similarly for S. epidermidis the higher zone of inhibition was observed for Kanamycin (IZD: 37.76mm) than other tested amino glycosides. All the antibiotics were applied at equal concentration to facilitate the ease of comparison.

For C. albicans, the results obtained for Gentamicin, Streptomycin, Amikacin, Tobramycin were very poor indicating that the antibiotics exhibited very limited activity against above skin infecting pathogens. However Kanamycin showed zone of inhibition of 11.33mm. Thus it was confirmed that among all tested amino glycosides, Kanamycin was found to exhibit reasonable antifungal activity.

CONCLUSION

Amino glycosides are the antibiotics exhibiting profound antibacterial activity. However reports regarding comparative study of their antimicrobial potency against skin infecting pathogen are not available. In the present study, five amino glycosides were tested against most severe skin infecting pathogens. The results of zone of inhibition showed that among all tested antibiotics, Amikacin possesses highest activity in bacteria in comparison to other antibiotics. The antifungal activity showed by Kanamycin was highest (11.33 mm) when compared to other antibiotics. In the study we probably for the first time documented the comparative assay results of efficacy of selected amino glycosides against skin infecting pathogens. The study has its own uniqueness and the reports may be helpful in various research fields.

Acknowledgement

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