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Comparative study of the effectiveness of Amikacin in serum and distilled water

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Abstract

A chemical compound acting against life is called antibiotic. Antibiotics are produced by submerged culture method. Antibiotics are the substance produced by various species of microorganisms that suppress the growth of other microorganisms and eventually may destroy them. Antibiotics are the antimicrobial agents.

Amikacin is an approved antibiotic used for treating severe bacterial infections. It is administered by intramuscular or intravenous injection. Side-effects can include hearing imbalance and kidney abnormalities.

Amikacin sulfate blocks protein synthesis in bacteria, resulting in death of the organism. Amikacin sulfate is effective against many gram–negative bacteria and some gram–positive bacteria.

Present study deals with comparative study of the effectivness of amikacin on Gram-negative bacteria *Escherichia coli* at different dilutions of drug in serum and distilled water and to check the interference of serum proteins with drug effect.

Key Words- Antimicrobial agent, Amikacin, Intramuscular, Intravenous, Abnormalities.

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Introduction: - An antibiotics (or antibacterial) are the compounds that kill bacteria or slow down the growth of bacteria. They are used as medicines to cure diseases caused by bacteria. They are low molecular weight compounds, having variety of chemical structures, elemental composition and physico-chemical properties[1]. Useful antibiotics are often discovered using a screening process. To conduct screening, isolates of many different microorganisms are cultured and then tested for production of diffusible products that inhibit the growth of test organisms. Hence must be tested for their selective toxicities and therapeutic activities and the best candidates can be examined and possibly modified.

Antimicrobial use is the major determinant in the development of resistance[2]. Many parameters of importance for optimal quality of antimicrobial therapy have already been defined. Maximal efficacy of the treatment should be combined with minimal toxicity at the lowest cost. Quality of antimicrobial drug use is dependent on the knowledge of many aspects of infectious diseases. Irrational use should be discouraged. Avoidance of the development of resistance is a quality parameter that will need increasing attention[3].

Amikacin is an antimicrobial drug comes under aminoglycosides, also known as amikacin sulphate[4]. Amikacin is active in vitro against *Pseudomonas species, Escherichia coli, Proteus species (indole-positive and indole-negative), Providencia species, Klebsiella-Enterobacter-Serratia species, Acinetobacter (formerly Mima-Herellea) species, Citrobacter freundii,* penicillinase and nonpenicillinase-producing *Staphylococcus species* including methicillin-resistant strains. However, aminoglycosides in general have a low

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order of activity against Streptococcus *pyogenes, enterococci, and Streptococcus pneumoniae (formerly Diplococcus pneumonia).*

Bacteriologic studies should be performed to identify causative organisms and their susceptibilities to Amikacin. Amikacin has also been shown to be effective in staphylococcal infections and may be considered as initial therapy under certain conditions in the treatment of known or suspected staphylococcal disease such as, severe infections where the causative organism may be either a Gram-negative bacterium or a staphylococcus, infections due to susceptible strains of staphylococci in patients allergic to other antibiotics, and in mixed staphylococcal/Gram-negative infections[5].

In certain severe infections such as neonatal sepsis, concomitant therapy with a penicillin-type drug may be indicated because of the possibility of infections due to Gram-positive organisms such as *streptococci or pneumococci.*

To reduce the development of drug-resistant bacteria and maintain the effectiveness of Amikacin and other antibacterial drugs, Amikacin should be used only to treat or prevent infections that are proven or strongly suspected to be caused by susceptible bacteria. When culture and susceptibility information are available, they should be considered in selecting or modifying antimicrobial therapy. In the absence of such data, local epidemiology and susceptibility patterns may contribute to the empiric selection of therapy.

Microbiological assay for amikacin can be very useful to check the interference of serum protein in drug effectiveness. The assay can be economical and simple to perform in a microbiology lab[6]. However, most of the time amikacin is not used as a single drug for

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therapy and other antibiotics like cipraflox or cephalosporins are frequently used. Thus, the basic principle should be non-interference of the other drugs. Obviously the test strain must be quite sensitive to amikacin but resistant to other antibiotics that are likely to be used simultaneously.

AIM:- Objective of the present study is to compare the drug effectiveness in serum and distilled water and to check the interference of serum protein in effectiveness of the drug.

Material and Methodology:-

E.Coli isolated from Pus, Urine and blood was selected for the development of Amikacin assay. The isolates were tested for antibiotic sensitivity using disc diffusion technique of Bauer et al.1966. The isolate which was sensitive to amikacin but resistant to ampicillin all generation of cephalosporins and ciprofloxacin were included as the candidate strains for the microbiological assay of amikacin[7].

Isolated colonies of the test organism were inoculated in peptone water and incubated for 2hrs at 37^{o}C to bring it to log phase. The turbidity was adjusted to 0.5 MacFarland units by dilution with sterile saline. The culture was then spread on Muller Hinton agar plate using sterile swab stick. The wells of 7mm were then punctured in the agar and different drug dilutions were added to the wells in duplicate plates[8]. The plates left at room temperature for ½ hr for the diffusion of the drug and then incubated overnight at 37^{o}C.

The zones of inhibition were measured in mm. Zones of inhibition for amikacin concentration 8.75ug/ml-140ug/ml for the test organisms. The numerical value of the mean

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and standard deviation from a particular sample than t-test is used to test the significance of difference between two mean samples and their correlations[9].

Observations:-

Table-1.1 Comparative study of the Effectiveness of the drug, dilution in serum and D/W

Dilution in serum	Sample I *E.coli*	Square	Dilution in D/W	Sample II *E.coli*	Square
140ug/ml	28	784	140ug/ml	24	576
112ug/ml	23	529	112ug/ml	22	484
93ug/ml	25	625	93ug/ml	23	529
70ug/ml	25	625	70ug/ml	20	400
47ug/ml	20	400	47ug/ml	17	289
35ug/ml	21	441	35ug/ml	18	324
17.5ug/ml	19	361	17.5ug/ml	16	256
8.75ug/ml	15	225	8.75ug/ml	15	225

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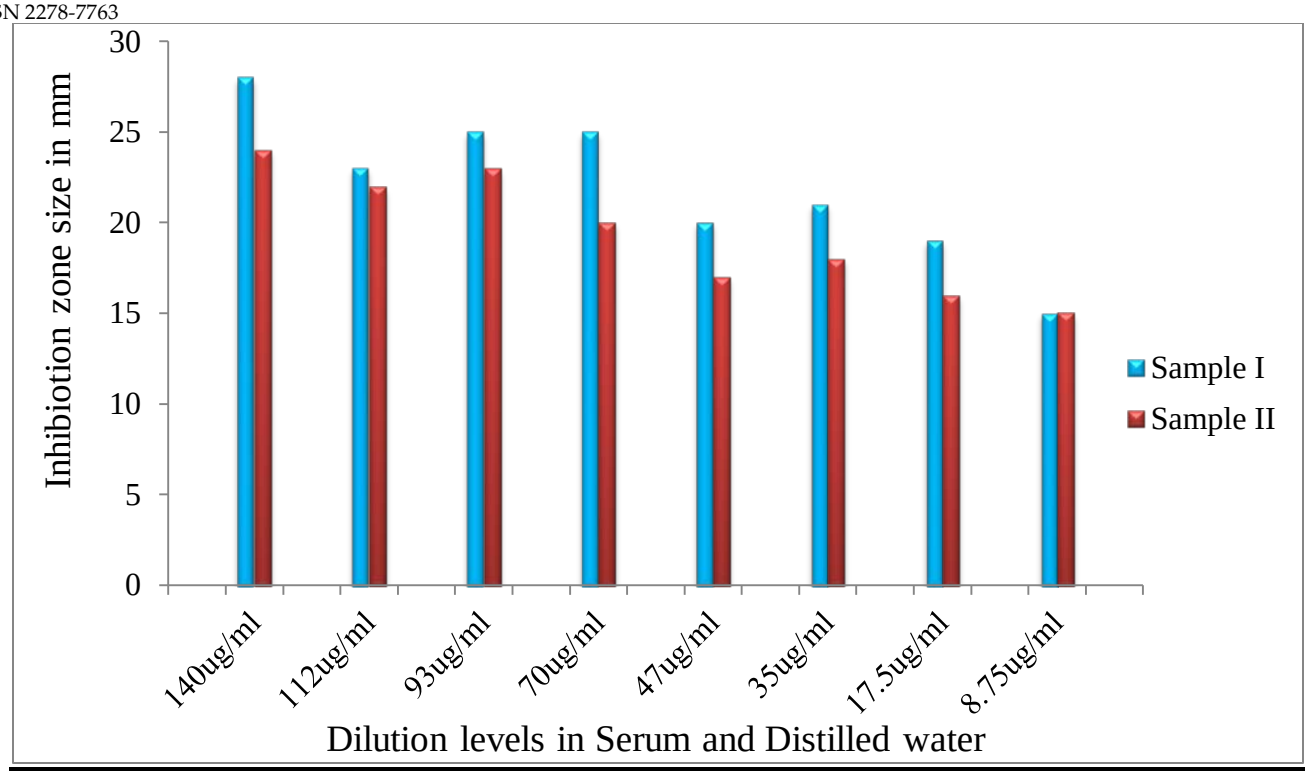

Result and Discussion- And then t-test (Statistical analysis) was performed on the above results to verified the significant difference and correlation on serum protein of two samples treated by amikacin was diluted in distilled water and serum. And the results obtained after performing t-test was highly significant that is 1.4064 above from the table value[10]. The t-value is significant that indicates the serum proteins are not interfering the effectiveness of the drug.

Conclusion- Range of amikacin assay was from 8.75ug/ml to 140ug/ml for microorganism. No protein interference was found for the different dilutions (prepared in blood, serum and distilled water). For pediatric patients the concentration of drug found in between <8.75ug/ml to 115ug/ml. The microbiological assay for amikacin level in blood has been well Standardized using *E.coli* as the test organism. This can be milestone step towards the study of in vivo drug effects of amikacin and it can also be useful in pharmaceutical

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industry. The proposed method allows cost-saving, precise and accurate determination of pharmaceutical equivalence of drugs in pharmaceutical dosage-form and may be used as a technique for testing generic antibiotics prior to their approval for human use[11].

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