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Antimicrobial Efficiency of Multipurpose Lens Rinsing Solutions

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The sterility and antimicrobial activity of commonly used multipurpose lens rinsing solutions(MPS) ReNu (Baush & Lomb), Fresh Look (Ciba Vision) , Splash (Intra Ocular care) was tested against the common eye infection causing pathogens like *Candida albicans* ATCC 10231, *Klebsiella pneumoniae* ATCC13883, *Pseudomonas aeruginosa* ATCC 15442 , and *Staphylococcus aureus* ATCC 6538. Antimicrobial efficiency of lens rinsing solution was checked by Disc Diffusion Method, Agar Cup Method and Turbidometric Method. The exposure time needed by the MPS, as mentioned in the manufactures instructions, to show their antimicrobial activity was also assessed using the turbidometric method. On carrying out the sterility testing of the lens rinsing solution it was found out that all the samples used were sterile. The lens rinsing solution did not show any antimicrobial activity by disc diffusion & agar cup method against the selected organisms. Turbidometric Method using glass beads showed that ReNu & Fresh Look completely inhibited *K.pneumoniae* at 24 hours exposure time. ReNu had maximum anti fungal activity. From the selected test organisms *C.albicans* was found to be most sensitive & *Pseudomonas aeruginosa* was found out to be most resistant to the MPS used. Although the three MPS were efficient enough to show their antimicrobial activity in the given prescribed time, they did not show inhibition of all the test organisms.

Bioactivity /Microbiology

Contact lenses were introduced over 50 years ago and it is estimated that there are now over 75 million contact lens wearers worldwide. Contact lens wear has been associated with a variety of adverse ocular reactions (Morlet, 1997) and contact lens solution is attributed as a major factor in many of these events (Cheng, 1999). Efficient contact lens disinfecting systems are essential for safe contact lens wear, and multipurpose solutions (MPS) are currently the most widely prescribed regimen (Stiegemeier, 2006).

These products are single-bottle solutions which can be used to clean, disinfect, and rinse contact lenses. Such MPS must be potent enough to kill harmful microorganisms, which may be present on the contact lenses, while at the same time should not be damaging to the cornea, as some solution may come into contact with the cornea when the lens is inserted in the eye (Papas,2007).

Author contributions: Shifa Lala and Sumaiya Jalgaonkar- both did Data collection and Experimental workSejal Rathod – carried out the Design of the Experiment and Scientific analysis of the data.Pratibha Shah- Literature survey and data analysis

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Fear of infection and subsequent vision loss remains a primary concern with contact lens wear, perhaps out of proportion to the actual incidence of risk. Reports during the past decade associating microbial keratitis and overnight lens wear are prime examples. Micro-organisms from the environment, the normal flora of the eye and organisms from ocular infections may contaminate lenses and

lens cases. Environmental sources of micro-organisms include water, air, soil, animals and plants. Lens cases are frequently the source of contamination. Even handling of the lens may add potential contaminants (Mondino, 1982)
Contact lens contamination can be significantly reduced when lens cleaning, rinsing, disinfection and the storage instructions are carefully followed. This study was designed to address the need for comparative data exploring performance of currently available contact lens disinfecting products.

Materials and Methods

Cultures used: *Candida albicans* ATCC 10231, *Klebsiella pneumoniae* ATCC13883, *Pseudomonas aeruginosa* ATCC 15442, and *Staphylococcus aureus* ATCC 6538 were maintained by growing in Nutrient agar medium and stored as Glycerol stock at 4° C.
MPS used: ReNu (Baush& Lomb), Fresh Look (Ciba Vision), Splash (Intra Ocular care).

Sterility Testing

Sterility testing of the antimicrobials was carried out by using method mentioned in the British Pharmacopeia (1958).

Disc diffusion technique

This test employs the use of dried filter paper discs impregnated with 50µl of MPS against the test organisms seeded on Mueller and Hinton medium (Hi Media ltd), the zone of inhibition is then measured as mentioned by Bauer et al. 1966. A control was set up by dipping the disc in sterile saline and checking against same pathogens.

Agar Cup Diffusion

The test employs the bulk seeding of the organisms in Mueller and Hinton medium (Hi Media ltd), and punching holes with a sterile cork borer (internal diameter 8mm) and adding various MPS in the wells, as described by Collins et al. (1995). A control was set up using sterile saline.

Turbidometric method

Glass beads are added in culture suspension of each test organism (0.1 optical density at 540nm) used. These glass beads are allowed to remain in the culture suspension overnight. Each of these glass beads are then transferred to different tubes containing the antimicrobial solutions as well as saline which acts as the control. Beads in different tubes are allowed to stand in the solution & saline (control) for different interval of times like ½ minute, 1 minute, 2 minutes and 24 hours and mixed. These beads are then transferred in tubes containing sterile nutrient broth and incubated at 37°c for 24 hour. Efficacy of antimicrobials would be determined by measuring the optical density at 540 nm.

Data Analysis

The optical density obtained by the turbidometric analysis for the three multipurpose lens rinsing solutions were converted into percentage growth, considering the growth in control as 100%.The percentage inhibition was then calculated by subtracting the percentage growth from 100.

Results and Discussion

On carrying out the sterility testing, all the three MPS were found sterile (Table -1).

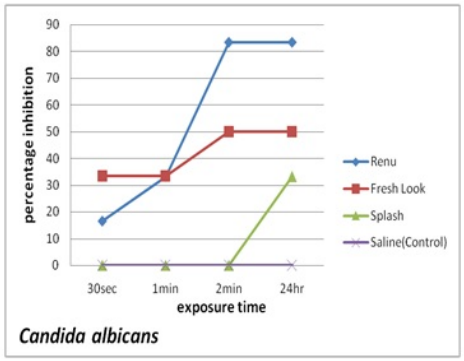
Table- 1: Properties and Sterility of the MPS

MPS / Properties	Renu	Fresh Look	Splash
Appearance	clear	clear	clear
pH	6.8	7.2	6.9
Active ingredient	Polyamino propyl biguanide (0.0001%)	Polyamino propyl biguanide (0.0001%)	Poly Hexamethylene Biguanide (0.0001%)
Sterility	sterile	sterile	sterile

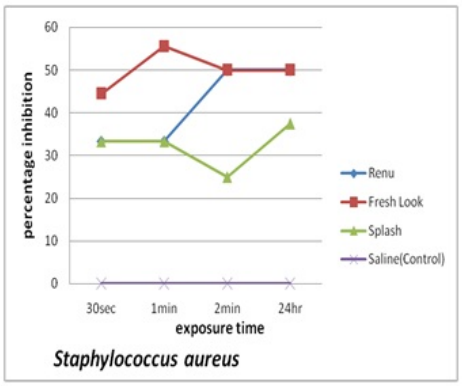
Disc diffusion and agar cup method against the selected organisms did not show any zone of inhibition against all the test organisms used. This could probably be because the amount of antimicrobial (active) ingredient present in the MPS is less which indicates that the exposure time to the lens rinsing solution has to be increased.

Antimicrobial efficiency of lens rinsing solution was checked by turbidometric method for different exposure time like 30sec, 1min, 2min, and 24hours. It was found out that ReNu and Fresh Look completely inhibited *K. pneumoniae*. *Candida albicans* was inhibited maximum (83.33%) by Renu and least inhibited by Splash (33.34%). *Staphylococcus aureus* was inhibited maximum by Freshlook (55.56%). *Pseudomonas aeruginosa* was inhibited maximum by Splash (57.14 %). From the selected organisms

K. pneumoniae and *C. albicans* was found out to be the most sensitive and *Pseudomonas* was found out to be the most resistant one. It was observed that percentage inhibition shown by the MPS used, increased as the exposure time was increased and the maximum inhibition was observed at 24 hours exposure time (Fig.1).



(a)



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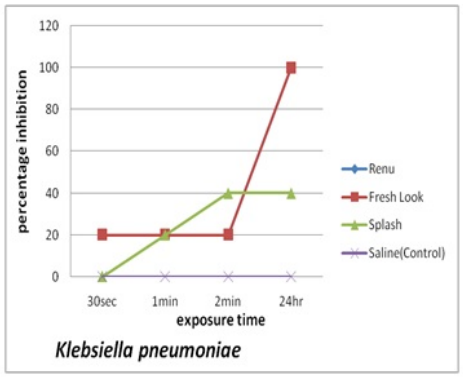

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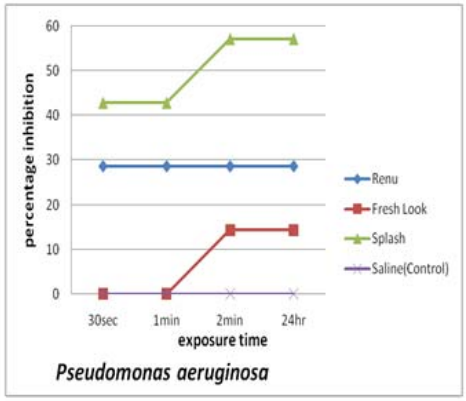

(d)

Fig.1: Percentage inhibition of (a) *Candida albicans* (b) *Staphylococcus aureus* (c) *Klebsiella pneumonia* (d) *Pseudomonas aeruginosa* by MPS at various exposure time

Turbidometric Method was also used for verifying the claims given by the manufacturers for the lens rinsing solutions, regarding the exposure time needed by the solutions to show their antimicrobial activity. The exposure time mentioned was Four hours for ReNu, ten minutes for Freshlook and six hours for Splash. None of the three antimicrobial solutions showed complete inhibition of the test organisms used. (Fig.2). This may be due to low concentration of active ingredient. Hence studies can be done to find optimum concentration of active ingredient against microbes that are most prevalent in the infected eye condition. Also similar activity can be checked *in vivo*.

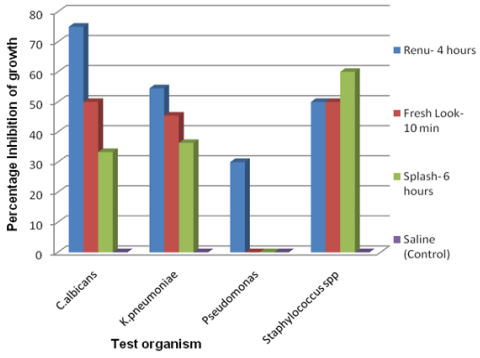

Fig.2: Percentage of inhibition of the test organisms at the manufacturers suggested time exposures

Conclusion

Infective keratitis is caused by invasion of the offending pathogens into the cornea. If not properly treated the infection can progress and possibly lead to corneal perforations, scarring and permanent loss of vision. Therefore steps must be taken to minimize the risk of infective keratitis in contact lens wearers. None of the three antimicrobial solutions showed complete inhibition of the test organisms used. Hence Lens Users should maintain good hand hygiene and take adequate lens care to prevent eye infections.

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