

Original Research

Antimicrobial effect of cow urine on *Staphylococcus aureus* and *Streptococcus pyogenes*

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ABSTRACT

Background: Infectious diseases impose significant risk to the public health regardless of incredible growth in human medicine. Cow urine is an effective antibacterial substance against a variety of pathogenic microorganism. Hence; the present study was undertaken for assessing the antimicrobial effect of cow urine on *Staphylococcus aureus* and *Streptococcus pyogenes*. **Materials & methods:** For the antimicrobial assessment, disease free cow urine was collected. A sterile container was used for collecting free cow urine from local cattle yard. In a sealed glass bottle, fresh cow urine was kept in sunlight for a time period of three days (seventy two hours) before the starting of the experiment, with the aim of making cow urine photo activated. Experimental *Staphylococcus aureus* and *Streptococcus pyogenes* cultured were obtained from microbiology laboratory. Agar well diffusion method was used for assessing the antibacterial efficacy of cow urine. Also simultaneous pouring of bacterial strains in 2 vials each containing 1 ml of cow urine was done. Assessment of optical density (OD) was done after every one hour. **Results:** Mean zone of inhibition in relation to *Staphylococcus* test samples was 21.5 mm while in relation to *Streptococcus pyogenes*, it was found to be 20.9 mm respectively. The declines in the O.D values indicated that the cow urine is showing its antimicrobial activity. **Conclusion:** Cow urine possesses remarkable antibacterial activity.

Key words: Antimicrobial, *Staphylococcus aureus*, *Streptococcus pyogenes*

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INTRODUCTION

There has been drastic increase in the popularity of Complementary, and Alternative Medicine in the Asian subcontinent. They comprise of vaguely assembled disciplines that vary from Ayurveda, Unani, Irani etc. Ideas of religious beliefs lay the foundation of such products.¹⁻³

Infectious diseases impose significant risk to the public health regardless of incredible growth in human medicine. Appearance of extensive drug resistance to the presently accessible antimicrobials is a substance of profound worry. Significant proportion of nosocomial infections occur due to highly resistant bacteria such as methicillin-resistant *Staphylococcus aureus* and *streptococcus pyogenes*. Cow urine is an effective antibacterial substance against a variety of pathogenic

microorganism.⁴In relation to a spectrum of anti-microbial drugs; it acts as a bio-enhancer. Hence; under the light of above mentioned data, the present study was undertaken for assessing the antimicrobial effect of cow urine on *Staphylococcus aureus* and *Streptococcus pyogenes*.

MATERIALS & METHODS

The present study was conducted at Genesis Institute of Dental Sciences and Research in department of microbiology in Ferozepur, Punjab, India with the aim of assessing the antimicrobial effect of cow urine on *Staphylococcus aureus* and *Streptococcus pyogenes*. Ethical approval was obtained from the ethical committee of the institution after explaining in detail the entire research protocol. For the antimicrobial

assessment, disease free cow urine was collected. A sterile container was used for collecting free cow urine from local cattle yard. In a sealed glass bottle, fresh cow urine was kept in sunlight for a time period of three days (seventy two hours) before the starting of the experiment, with the aim of making cow urine photo activated. Whatmann No.1 filter paper was used for filtering the urine for eradicating debris and predicated materials. This was followed by storing of the cow urine at four degree celsius before utilization in the experiment. Thorough and strict checking of the cow urine was done for assessing the presence of any microbial contamination. Experimental *Staphylococcus aureus* and *Streptococcus pyogene* cultured were obtained from microbiology laboratory. Agar well diffusion method was used for assessing the antibacterial efficacy of cow urine. Pouring of 20 ml of sterile Muller Hinton agar was done in petri dishes. Plates were then left for solidification. Spreading of organism on Muller Hinton and wells were prepared with the help of puncture and cow urine was poured into the wells. This was followed by incubation of plates at 37°C for 24 hours. Assessment of zone of inhibition was done. Also simultaneous pouring of

bacterial strains in 2 vials each containing 1 ml of cow urine was done. Assessment of optical density (OD) was done after every one hour. Fall in OD values indicated antimicrobial activity of cow urine . All the results were compiled in Microsoft excel sheet and were analyzed by SPSS software.

RESULTS

Table 1 shows the mean zone of inhibition. In the present study, we observed that mean zone of inhibition in relation to *Staphylococcus aureus* test samples was 21.5 mm while in relation to *Streptococcus pyogenes*, it was found to be 20.9 mm respectively.

Table 2 shows the mean optical density at different time intervals. Mean OD at one hour, two hour, three hour and five hour in relation to *Staphylococcus aureus* specimen was 1.123, 0.986, 0.861 and 0.728 respectively. Mean OD at one hour, two hour, three hour and five hour in relation to *Streptococcus pyogenes* specimen was 1.213, 0.991, 0.892 and 0.759 respectively. The declines in the O.D values indicated that the cow urine is showing its antimicrobial activity.

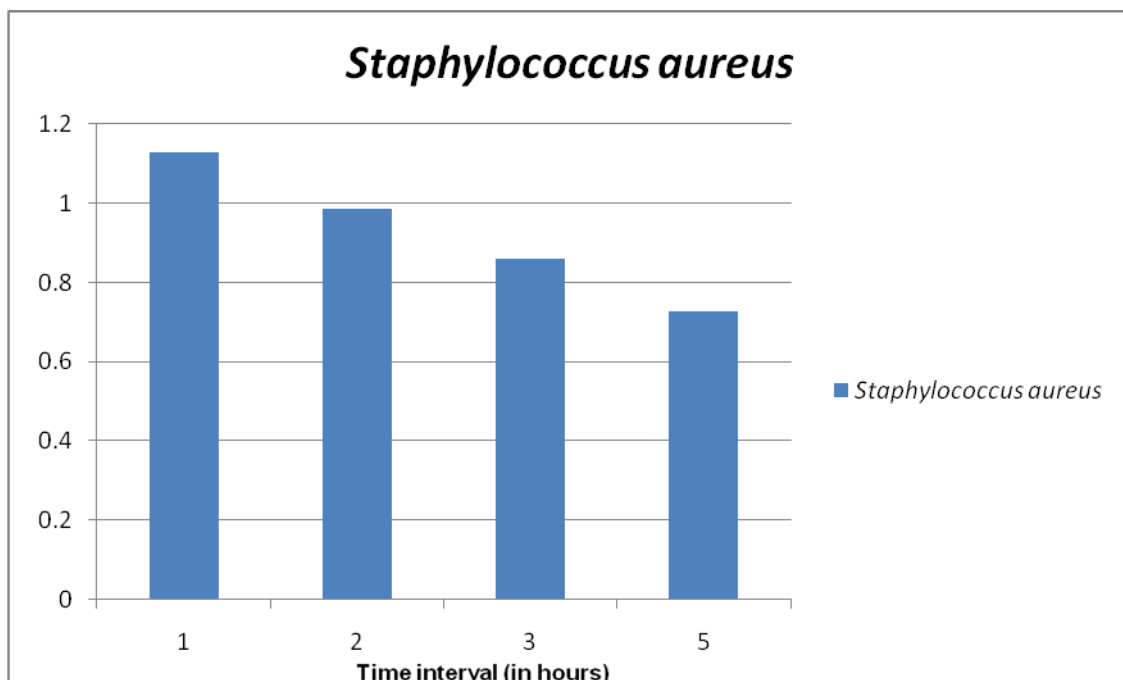
Table 1: Mean zone of inhibition

Parameter	<i>Staphylococcus aureus</i>	<i>Streptococcus pyogenes</i>
Mean zone of inhibition (mm)	21.5	20.9

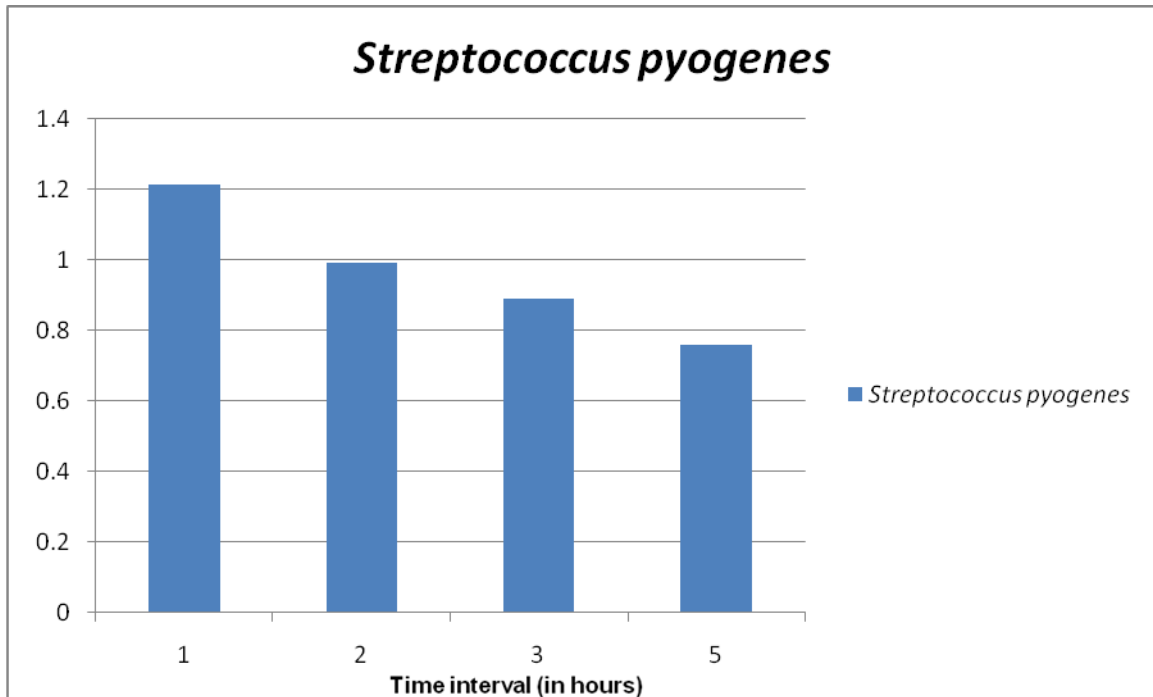
Table 2: Optical density

Hours	<i>Staphylococcus aureus</i>	<i>Streptococcus pyogenes</i>
1	1.128	1.213
2	0.986	0.991
3	0.861	0.892
5	0.728	0.759

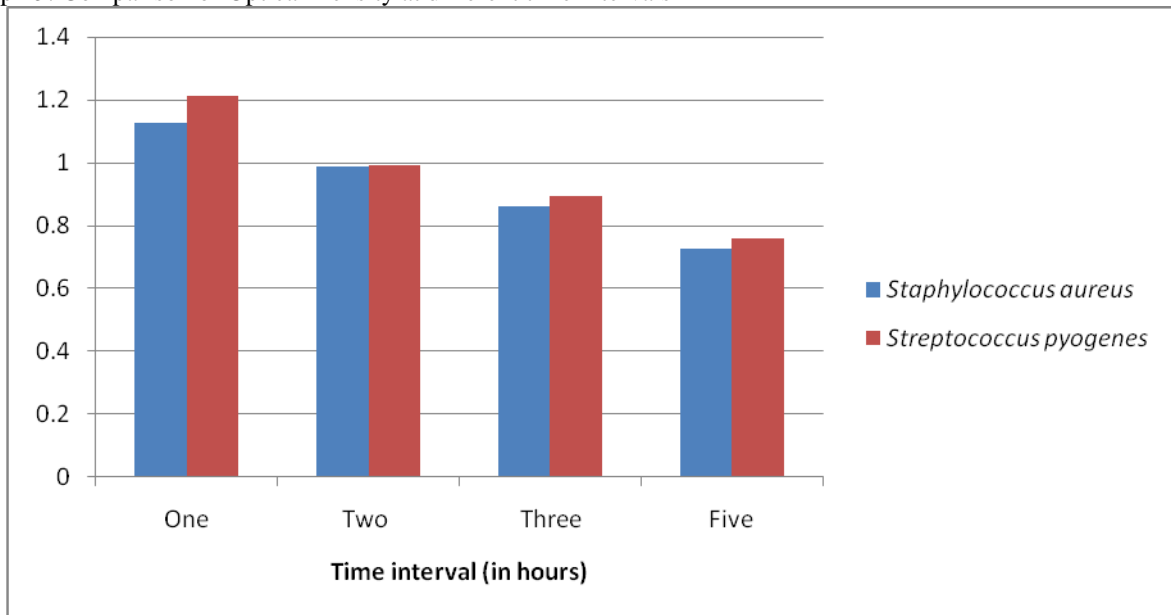
Graph 1 : Optical Density of *Streptococcus aureus* at different time intervals



Graph 2: Optical Density of *Streptococcus pyogenes* at different time intervals



Graph 3: Comparison of Optical Density at different time intervals



DISCUSSION

In routine practice, antibiotic therapy is widely used in treating various microbial infections. Despite the increasing use of antibiotics, a simultaneous increase in the problems caused by infectious disease with time. Certain significant interferences include genetic ability of bacteria to transmit and acquire resistance in relation to different drugs. Also significant association of adverse events is also associated with extensive use of these drugs. Data from past literature shows that cow urine has antifungal, anthelmintic, antineoplastic properties. Along with this, it is also useful in treating hypersensitivity reactions.⁵⁻⁷ Cow urine is often photo

activated before using. In contrast to fresh cow urine, Photo-activated cow urine (PhCU) becomes highly acidic. Elevated bactericidal action might be because of significantly fall in pH, occurrence of inorganic phosphorus, chloride and dimethylamine might also play a significant role, apart from elevated formation of reactive substances such as formaldehyde, sulfinol, ketones and some amines during photo-activation and long term storage. Along with providing antibacterial effect, it also retards the formation of antibacterial resistance by obstructing the R-factor, a part of plasmid genome of bacteria.⁸⁻¹⁰ Hence; under the light of above mentioned data, the present study was undertaken for

assessing the antimicrobial effect of cow urine on *Staphylococcus aureus* and *Streptococcus pyogenes*.

In the present study, we observed that mean zone of inhibition in relation to *Staphylococcus aureus* samples was 21.5 mm while in relation to *Streptococcus pyogenes*, it was found to be 20.9 mm respectively. A study assessed mixture of photo activated and its binary mixture against 7 bacterial strains. MIC value of PhACU was found to be 0.25 µl/ml minimum inhibitory concentration (MIC) value against *Staphylococcus aureus*, *Bacillus cereus*, *Lactobacillus acidophilus* and *Micrococcus luteus*, while it was found 0.125 µl/ml against *E. coli*. Binary combinations of cow urine with Neem and Bavchi oil has shown synergistic effect as the MIC value obtained was 0.125-0.25 µl/ml. They also observed that PhACU was found to be 32 to 36 mm inhibition zone diameter homogeneously against all bacterial strains. Their results proved that cow urine had very antimicrobial susceptibility.¹¹

In the present study, mean OD at one hour, two hour, three hour and five hour in relation to *Staphylococcus aureus* specimen was 1.123, 0.986, 0.861 and 0.728 respectively. Mean OD at one hour, two hour, three hour and five hour in relation to *Streptococcus pyogenes* specimen was 1.213, 0.991, 0.892 and 0.759 respectively. The declines in the O.D values indicated that the cow urine is showing its antimicrobial activity. Traditionally, cow urine is proved to be effective in fever. Also, cow urine in combination with pepper, curd and ghee is found to be useful in leprosy patients. Other combinations of cow urine are also useful in certain other pathologies. E.g. cow urine used in combination with dhruhardi is reported to be useful in treating Leprosy associated deformities.^{12, 13} A bioenhancer is referred to an agent having the potential of reinforcing the bioavailability and effectiveness of a drug with which it is co-administered, with absence of its own pharmacological activity at the therapeutic dose used. The use of bioenhancers lies in the fact that they can elevate the effectiveness of commonly used antibiotics. Activity of antibiotics is also potentially enhanced by use of Cow urine distillate (CUD). Therefore, this CUD can act as a potential therapeutic target. In combination with rifampicin, CUD increases the activity of drug by almost five to seven times against *Escherichia coli* and against other gram-positive bacteria.¹⁴ Council of Scientific and Industrial Research have patented CUD for its use against fungal and bacterial pathologies.¹⁵

CONCLUSION

From the above results, it is concluded that cow urine possesses remarkable antibacterial activity. Therefore; further large scale studies with broader parameters are advocated in future for establishing their role for therapeutic purposes.

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