



## IN VITRO SCREENING OF ANTIBACTERIAL ACTIVITY OF COW URINE AGAINST PATHOGENIC HUMAN BACTERIAL STRAINS

CHARMI P SHAH<sup>1\*</sup>, DEVAL M PATEL<sup>1</sup>, PARAS D DHAMI<sup>1</sup>, JANAK KAKADIA<sup>1</sup>, DHRUVESH BHAVSAR<sup>1</sup>, URMILA D VACHHANI<sup>1</sup>, MANISHA N TRIVEDI<sup>1</sup>, JOSHI V J<sup>2</sup>

<sup>1</sup>Rofel, Shri G M Bilakhia College of Pharmacy, Vapi, Gujarat, India, <sup>2</sup>NIMS University, Shobha nagar, Delhi highway, Jaipur, India  
Email: charmishah15@yahoo.co.in

Received: 22 Nov 2010, Revised and Accepted: 24 Dec 2010

### ABSTRACT

Indiscriminate uses of antibiotics have caused microbial resistance and also lead to many side effects. To overcome from such situation plants and animal materials are widely used for the treating various ailments due its antimicrobial properties. In Ayurveda, cow urine has been used to improve general health of an individual. Therefore, present study is undertaken to study in vitro antibacterial potential of cow urine against various pathogenic bacteria. The method employed to study antibacterial activity is agar well diffusion technique. A reference standard was also employed along with the test during the experimental study. The results showed good antibacterial activity of cow urine against most of the test bacterial strains by exhibiting zone of inhibition. These results thus proves that cow urine possess good inhibitory activities against various clinical bacterial strains and can be used to control infectious diseases.

### INTRODUCTION

Commonly, antibiotics are widely as conservative treatment in various microbial infections and diseases.<sup>1</sup> Considering the enormous quantity of antibiotics used, the situation should have been that there would be no infectious diseases. But, the fact is that the problems of infectious diseases are increasing day-by-day. Some of the major hindrances are that bacteria have genetic ability to transmit and acquire resistance towards the drugs<sup>2</sup> and there are also adverse effects of drugs on the host.<sup>3</sup> Therefore to combat such problems many natural products have been explored. The nature is an almost infinite resource for drug development and discovery. It has endowed with a complete repository of remedies to cure all ailments of mankind, as it has always been a first rate drug store with enormous range of plants, micro organisms and animals.<sup>4</sup>

In Veda, cow is considered the most valuable animal and is called Mother of all. Different products obtained from cow like urine, dung, milk, ghee and curd are used widely in number of Ayurvedic formulations.<sup>5</sup> As per Ayurvedic literatures cow urine possess many medicinal properties and is used in curing number of diseases like skin diseases, kidney problems, epilepsy, anemia, constipation, respiratory disease etc.<sup>6,7</sup> Due to its therapeutic values majority of rural population in India use cow urine as a folklore remedy to get rid of various diseases. Nowadays, different preparations of cow urine like urine distillate, photo-activated urine, fresh urine, sterile urine have been marketed with cheap and affordable prices.<sup>8</sup>

Thus the aim of the present work is to study antibacterial potential of photo-activated cow urine, fresh cow urine, cow's urine distillate against different pathogenic human bacterial strains.

### MATERIALS AND METHODS

#### Collection of Cow urine

Disease free cow was selected for urine collection in the study. Fresh cow urine was collected in a sterile container from local cow yard. For the experiment following methods were used for various preparation of cow urine.

1. Photo-activated urine was prepared by keeping fresh cow urine in sunlight for 72 h in sealed glass bottle. The urine was then filtered through Whatmann No. 1 filter paper to get rid of debris and precipitated material and was stored at 4°C before use. Before evaluation of antibacterial activity, cow urine was checked for the presence of microbial contamination.
2. Cow urine distillate was obtained by distilling cow urine at 100°C using distillation apparatus.<sup>8</sup>

#### Antibacterial activity

The test organisms used in the study were *E. coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, Coagulase negative staphylococci, *Streptococcus pyogenes* and *Bacillus subtilis*. All the test cultures were obtained from the Microbiology laboratory of the department of the college. The purity of all the cultures was checked before use. The cultures were maintained at 4°C on Nutrient agar (HiMedia) slants. The antibacterial of the selected cow urine preparations were performed by agar well diffusion method.<sup>9</sup> 20 ml of sterile Muller Hinton agar (Hi Media) was poured in sterile petri dishes. The plates were allowed to solidify and used. 10 ml of sterile, Muller Hinton agar medium (seed agar) was seeded with organisms (about 0.2 ml according to 0.5 Mc Farland's standard), in semi hot conditions and was poured uniformly on the base agar. 8mm bores were made each equidistant from one another on the medium using sterile borer and 100µl ml of the different urine preparation were added to respective bore. The plates were incubated at 37°C for 24 h and zone of inhibition was measured. A reference standard of streptomycin (100µg/ml) was also used to compare with the obtained results in the study. For each test, three replicates were performed.

### RESULTS AND DISCUSSION

The results of antibacterial potential of different cow urine preparation are shown in Table 1. The result of the present study shows that antibacterial activity of fresh cow urine is more active than phot-activataed urine. These observations are likely to be the result of the presence of certain volatile and non volatile components present in urine.<sup>10</sup> The antibacterial activity of photo-activated urine may be due to its acidic pH.<sup>11</sup> The antibacterial activity of the present study is also constituent with the reports of other authors which also show good antibacterial activity in fresh urine.<sup>8</sup> Urine distillate was not found to be active among all the cow urine preparations. These results are in contradictory to the report of others.<sup>12</sup> One of the possible reasons for resistant in antibacterial activity in urine distillate may be removal of toxic components from it or there no formation of biogenic volatile inorganic and organic compounds. It may also be due to presence of more cations and formation of nitrosoamines.<sup>13</sup> The activity of test urine sample preparations were comparable with that of standard, Streptomycin. It was observed that gram positive organisms were more sensitive than gram negative organisms. These observations are likely to be the result of the differences in cell wall structure between gram positive and gram negative bacteria, with gram negative outer membrane acting as a barrier to many environmental substances.<sup>14</sup>

Table 1: Antibacterial activity of different cow urine preparations against pathogenic bacterial strains\*

Name of the bacteria	Mean zone of inhibition in mm			
	1	2	3	4
<i>E. coli</i>	13	-	16	16
<i>Klebsiella pneumonia</i>	12	-	15	17
<i>Pseudomonas aeruginosa</i>	15	-	17	19
<i>Staphylococcus aureus</i>	17	-	18	26
Coagulase negative Staphylococci	15	-	18	29
<i>Bacillus subtilis</i>	15	-	20	29
<i>Streptococcus pyogenes</i>	14	-	20	26

\*1 = Photactivated urine, 2 = Urine distillate, 3 = Fresh urine, 4 = Streptomycin

## CONCLUSION

As cow urine showed remarkable antibacterial activity against the pathogenic bacteria, for which it can be selected for further studies to isolate bioactive natural constituents that may address to unmet therapeutic needs. Such screening of various natural compounds and identifying active agents is the needful, because successful prediction of lead molecule and drug like properties at the onset of drug discovery will pay off later in drug development. But *in vivo* studies on these cow urine preparations are necessary and should seek to justify its potential as antimicrobial agent.

## REFERENCES

- Daniel B, Alexander R, Ehud IA. Changing bacterial isolates and antibiotic sensitivities of purulent dacryocystitis. *Orbit*; 2005; 24 (2): 95-8.
- Cohen ML. Epidemiology of drug resistance: implications for a post-antimicrobial era, *Science* 1992; 257: 1050-1055.
- Ahmad I, Mehmood Z, Mohammad F. Screening of some Indian medicinal plants for their antimicrobial properties. *J Ethnopharmacol* 1998; 62: 183-193.
- Sujata MB, Charles HB. New agents for Gram-positive bacteria. *Current Opinion in Microbiology*. 2000; 3(5): 528-534.
- Shah E. Herbal composition in cow urine distillate. US5693327. 1997.
- Krishnamurthi K, Dutta D, Devi SS, Chakrabarti T. Protective effect of distillate and redistillate of cow's urine in human polymorphonuclear leucocytes challenged with established genotoxic chemicals. *Biomed Environ Sci* 2004; 17: 57-66.
- Chauhan RS, Singh BP, Singhal LK. Immunomodulation with kamdhenu Ark in mice. *J Immunol Immunopathol* 2001; 71: 89-92.
- Edwin J, Sheej E, Vaibhav T, Rajesh G, Emmanuel T. Antioxidant and antimicrobial activities of cow urine. *Global journal of pharmacology* 2008; 2(2): 20-22.
- Shah CP, Joshi BH. Antibacterial Activity of Some Medicinal Plants against the Pathogens Isolated from Dacryocystitis. *VNSGU Journal of Science and Technology* 2009; 1(2): 123-127.
- Shaw SL, Mitloehner FM, Jackson W, Depeters EJ, Fadel JG, Robinson PH, Holiizinger R, Goldstein AH. Volatile organic compound emissions from dairy cows and their wastes as measured by proton transfer-reaction mass spectrometry. *Environ Sci Technol* 2007; 14: 1310-1316.
- Hw W, Murphy MR, Constable PD, Block E. Dietary cation-anion difference effects on performance and acid base status of dairy cows postpartum. *J Dairy Sci* 2007; 90: 3367-3375.
- Arun Kumar S, Muthuselvam M, Rajasekran R. Antimicrobial activities of cow urine distillate against some clinical pathogens. *Global journal of pharmacology* 2010; 4(1): 41-44.
- Stephany RW, Schuller PL. The intake of nitrate, nitrite and volatile N-nitrosamines and occurrence of volatile N-nitrosamines in human urine and veal calves. *IARC Sci Publ* 1978; 443-460.
- Tortora GJ, Funke BR, Case CL. *Microbiology: An introduction*. San Francisco: Benjamin Cummings, 2001.