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Assessment of beneficial effect of Badri cow urine distillate on serum biochemical profile in Wistar rats

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Abstract
Present study was carried out to find the effect of Badri cow urine distillate on serum biochemical parameters in Wistar rats. Badri cow urine distillate was given to rats orally with drinking water at 0. 25ml/day/rat for 90 days. The blood sample were collected from retro orbital sinus of rats at 0th, 30th, 60th, 90th day post treatment in clot activator vials. Then serum was separated from blood by standard protocol for assessment of serum biochemical parameters. There was significant increase in serum total protein, serum globulin while other parameters like serum glucose, cholesterol values were significantly decrease in treated group as compare to control group. Some parameters like aspartate amino transaminase (AST), alanine amino transaminase (ALT) were decrease in value in treated group as compared to control group. Results of present study revealed that Badri cow urine distillate show ameliorative effect on serum biochemical parameters.

Keywords: Cow urine distillate, biochemical, Wistar rats

Introduction
Cows were considered as wealth and were the base of the ancient Indian economy. Most ancient medical systems including traditional Indian, Chinese, and Western systems, have mentioned indigenous cow products and secretions as healthcare remedies. One of the most important components of 'Panchagavya,' indigenous cow urine, has the potential to treat a wide range of illnesses, both curable and incurable, and is widely used in Ayurvedic medicines (Pathak and Kumar (2003a) [12]. Cow urine consumption has been a long-held belief in India for thousands of years. Cow urine has been identified in sacred texts as "Amrita" (immortality liquids) or "water of life," the God's nectar (Bartnett, 1988; Chauhan *et al.,* 2001; Chauhan, 2003a) [1, 5, 3].
Now days Panchgavya chikitsa is known as "Cowpathy," which refers to the treatment of human illnesses with five cow products: milk, ghee, curd, dung, and urine, as well as cow cuddling, cow back rub, Gaugrass, Gauseva, and living with cows (Chauhan, 2022) [4]. Modern therapy destroys the patient's immunity, whereas Cowpathy medicines strengthen it. Cowpathy improves an individual's aura, resulting in a happy and healthy life (Dhama *et al.,* 2008) [6]. There are no much more studies available on the effect of Badri cow urine distillate on biochemical parameters, thus the current study was designed to assess the beneficial effect of Badri cow urine distillate on serum biochemical parameters.

Material and Method
Preparation of cow urine distillate
For present study Badri cow urine was collected from the instructional dairy farm (IDF) in Pantnagar, Uttrakhand, India. Cow urine was distillated in a distillation apparatus at the veterinary pathology department and stored in an airtight brown colour bottle for later use. In group II rats, cow urine distillate was administered orally at 0.25ml/day/rat (Chauhan *et al.,* 2001) [5] along with drinking water.

Experimental Design
For the present experiment 35 rats were randomly divided into two groups. Group I (GI) and Group II (GII). Group I contains 20 rats and serves as the control group. Group II consists of 15 rats who were given cow urine distillate. Throughout the experiment, GI group rats were given standard feed and water ad libitum. From the 0th to the 90th day after treatment, the rats in the GII group were given standard feed and Badri cow urine distillate at a dose of 0.25ml/day/rat orally with drinking water.

Throughout the experiment, rats were observed twice a day for any unusual clinical signs or behavioural changes. The use of experimental rats in the present study was duly permitted by the IAEC vide no. IAEC/CVASc/VPP/456.

Collection of blood sample

Blood samples were collected in clot activator vials from 5 rats from each group (control and test) at the 0th, 30th, 60th, and 90th day post treatment. Blood samples were collected during the morning time and immediately transported to laboratory under sterile conditions in ice packs.

Separation of serum

Blood samples from 5 rats from both the groups were collected at 0, 30th, 60th, 90th DPT without anticoagulant in a clot activator vial and kept in slanting position for 20 to 30 minutes at room temperature followed by overnight refrigeration. The samples were then subjected to centrifugation at 3000 rpm for 30 to 60 minutes. Centrifugation of the sample resulted in separation of serum. The serum was then collected in a separate Eppendorf tube of 1ml size. The serum samples were stored at -20°C in deep freezer till further use in the future (Tuck *et al.,* 2009) [16]. The biochemical parameters viz. total serum protein, serum albumin, serum globulin, serum glucose, serum total cholesterol, serum calcium, serum phosphorus, serum aspartate aminotransferase (AST) and serum alanine transaminase (ALT) were estimated by using commercial kits with trade name Erba.

Statistical analysis

The experimental data was statistically analysed using standard statistical protocols and procedures (Snedecor and Cochran, 1980). The values are expressed as the mean and standard error of the mean (SEM). ANOVA (Analysis of one Variance) was used to compare mean differences between treated and control groups by using SPSS software. $P < 0.05$ was considered as statistically significant.

Results and Discussion

In the present study, there was decrease in mean serum ALT (2.5%) and AST (3.18%) values in cow urine distillate treated rats as compared to control rats, which indicates the hepatoprotective role of Badri cow urine (Sankhala *et al.,* 2022; Panicker *et al.,* 2012) [14, 11]. There was a significance increase in mean total serum protein values (19.24%) at 90th day post treatment in group II rats as compared to group I rats. The increase in the total serum protein might be due to anabolic effects of cow urine distillate on protein metabolism (Panicker *et al.,* 2012) [11]. There was non-significant changes was observed in serum albumin in group II rats as compared to group 1 rats. The albumin is synthesized in the liver and has been useful in the measurement of the synthetic functions of the liver. Mean serum globulin values were also significantly increase (33.68%) at 90th day post treatment in group II rats as compared to group I rats. The serum globulin has important immunological and nutritional implications. Most of these are synthesized in liver, although immunoglobulins are synthesized by plasma cells. Increases in the globulin fraction usually result from increase in immunoglobulins and might indicate liver functioned properly. The gamma globulin fraction is the source of almost all the immunological active protein of the blood. Gamma globulins are essential for maintaining a healthy immune system.

There was significant decrease in mean serum glucose values (18.51%) in group II rats as compared to group I rats. Results of the present study are in accordance with Sachdev *et al.* (2012) [13] and Gururaja *et al.* (2011) [8]. The decrease in the serum glucose level might be due to sulfur present in Badri cow urine which might have some action like sulfonylureas, or it might be increasing sensitivity of insulin receptors, or causing more release of insulin. Hence, the presence of antioxidant, free radical scavengers in cow urine distillate might be responsible for its anti-diabetic action (Gururaja *et al.,* 2011) [8]. There was significant decrease in mean serum cholesterol values (18.07%) in group II rats as compared to group I rats. These results are in accordance with that of Manubhai *et al.* (2014) and Shankhala *et al.* (2022). Shankhala *et al.* 2022 study revealed that serum cholesterol level significantly decrease in birds that received cow urine distillate along with imidacloprid in comparison to their respective groups that received only imidacloprid. Dietary and serum copper inversely associated with fasting glucose, total cholesterol and LDL-C (BO *et al.,* 2008) [2]. Previous study also indicated that the supplementation of moderate dietary copper inhibits atherogenesis in the cholesterol fed rabbit (Lamb *et al.,* 2001) [9]. Cow urine contains copper which might be responsible for its lipid lowering activity. Garg *et al.* 2004 [7] also reported that supplementation of cow urine to white leghorn layer showed significant amelioration in serum total cholesterol.

Effect of Badri cow urine distillate on biochemical parameters in Wistar rats is described in table 1.

Table 1: Mean values of serum biochemical parameters in control and Badri cow urine distillate treated Wistar rats.

Parameters	0th DPT		30th DPT		60th DPT		90th DPT	
	Group I	Group II	Group I	Group II	Group I	Group II	Group I	Group II
TSP (g/dl)	6.42 ± 0.05^{Aa}	6.42 ± 0.05^{Aa}	6.10 ± 0.23^{Ba}	6.26 ± 0.14^{Ba}	5.97 ± 0.10^{Ca}	6.57 ± 0.08^{Ca}	6.08 ± 0.04^{Da}	7.25 ± 0.12^{Ea}
Albumin (g/dl)	3.80 ± 0.04^{Aa}	3.80 ± 0.04^{Aa}	3.98 ± 0.04^{Ba}	3.97 ± 0.11^{Ba}	4.19 ± 0.04^{Ca}	3.93 ± 0.14^{Ca}	4.29 ± 0.09^{Da}	3.50 ± 0.10^{Ea}
Globulin(g/dl)	2.38 ± 0.03^{Aa}	2.38 ± 0.03^{Aa}	2.35 ± 0.03^{Ba}	2.36 ± 0.04^{Ba}	2.10 ± 0.06^{Ca}	2.25 ± 0.04^{Ca}	1.90 ± 0.16^{Da}	2.54 ± 0.09^{Eb}
ALT (IU/L)	30.25 ± 0.24^{Aa}	30.25 ± 0.24^{Aa}	30.26 ± 0.5^{Ba}	30.07 ± 0.13^{Cb}	30.37 ± 0.19^{Da}	29.84 ± 0.05^{Ec}	30.41 ± 0.08^{Fa}	29.62 ± 0.15^{Gd}
AST (IU/L)	68.81 ± 0.3^{Aa}	68.81 ± 0.3^{Aa}	70.52 ± 0.17^{Ba}	69.58 ± 0.2^{Ba}	71.21 ± 0.27^{Ca}	69.64 ± 0.44^{Ca}	71.63 ± 0.09^{Da}	69.35 ± 0.38^{Da}
Glucose (mg/dl)	93.76 ± 0.47^{Aa}	93.76 ± 0.47^{Aa}	94.95 ± 0.46^{Ba}	89.62 ± 1.09^{Ba}	96.29 ± 0.61^{Ca}	87.95 ± 1.18^{Ca}	99.32 ± 0.24^{Da}	80.93 ± 0.67^{Eb}
Cholesterol (mg/dl)	58.49 ± 0.33^{Aa}	58.49 ± 0.33^{Aa}	57.81 ± 0.44^{Ba}	56.84 ± 0.45^{Ba}	60.29 ± 1.03^{Ca}	55.28 ± 0.31^{Ca}	63.83 ± 0.19^{Da}	52.29 ± 0.45^{Ea}

Values Indicate Mean ± S.E. Means having different superscripts differs significantly ($P\leq0.05$) when compared horizontally within the same raw.

Conclusion

Based on the current findings, it is reasonable to conclude that Badri cow urine distillate has a beneficial effect on serum biochemical parameters such as serum total protein, serum globulin, serum glucose, and serum cholesterol. As a result, Badri cow urine distillate can be used to treat a variety of

human ailments with have improvement impact on the immunity by increasing serum globulin concentration because the serum globulin has important immunological and nutritional implications and increases in the globulin fraction usually result from increase in immunoglobulins and might indicate that Badri cow urine also upregulate the immunity by increasing globulin concentration.

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