RENAL CLEARANCE OF ENDOGENOUS CREATININE, UREA AND KANAMYCIN IN COWS

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ABSTRACT

Following a single intravenous dose of kanamycin 5 mg/kg body weight to 8 health cows, renal clearance of the drug and that of endogenous creatinine and urea were determined. Before drug administration control samples and after drug administration blood and urine samples were collected at predetermined time periods. The plasma and urine samples were assayed for endogenous creatinine and urea by spectrophotometric methods and kanamycin by microbiological assay. Mean ± SE (n=32) values for the blood and urine pH mean were 7.37 ± 0.04 and 7.91 ± 0.07, respectively. The rate of urine flow (diuresis) was 0.024 ± 0.003 $ml.min^{-1}.kg^{-1}$, renal clearance of endogenous creatinine was 0.60 ± 0.08, urea 0.17 ± 0.04 and kanamycin was 0.08 ± 0.01 $ml.min^{-1}.kg^{-1}$ body weight. A significant ($P < 0.001$) positive correlation ($r = 0.55$) between diuresis and renal clearance of kanamycin indicated that renal handling of kanamycin in cows, besides glomerular filtration also involved back diffusion.

INTRODUCTION

Kanamycin is a valuable broad-spectrum antibiotic extensively used in veterinary clinics against susceptible infections. The drug is mainly eliminated via renal route therefore, study of mechanisms involved in the renal handling are of practical importance. Renal function in the indigenous species of ruminants with reference to glomerular filtration rate (GFR) has been recorded lower than the corresponding values of GFR given in literature (Nawaz *et al.*, 1988). These differences were defined by an original term "geonetics", the geographical influences on genetics (Nawaz, 1988, 1994; Nawaz *et al.*, 1988). These "geonetical" differences in ruminants have been recorded for pH of blood and urine, plasma proteins, drug metabolism and renal functions ultimately affecting biodisposition and fate of drugs. In view of the fact that kanamycin is primarily excreted through the renal route, this study in cows was undertaken to assess renal handling of the drug.

MATERIALS AND METHODS

Renal clearance of endogenous creatinine, urea and kanamycin was determined in 8 healthy cows of local Sahiwal breed during month of October. Animals were maintained under similar conditions of feeding and management at the Livestock Production Research Institute, Bahadurnagar, Okara. The mean ± SE value for body weight of animals was 416 ± 8 kg. In all experiments, before drug administration, a control venous blood sample was drawn from the jugular vein in heparinized centrifuge tube. Each animal was given a single intravenous dose of kanamycin (Kanachron, 10% Batch No.29493, Star Laboratories, Lahore, Paskistan) at dose are of 5 mg/kg body weight into the right jugular vein. Blood samples were collected through plastic cannula aseptically inserted in the jugular vein. Following drug administration, four blood samples were drawn in sterilized heparinized centrifuge tubes at each 30 minutes interval.

A balloon catheter (Foly No.18, 30 ml) was aseptically placed in the urinary bladder through urethra. External opening of catheter was connected to a flask in which all voided urine was quantitatively collected. Blank urine sample in each experiment was collected at the time of catheterization. After washing of urinary bladder, urine samples were taken with an interval of 30 minutes up to 120 minutes after drug administration.

Analysis

The pH of all blood and urine samples was recorded by using Beckman H_5 (Germany) pH meter by using glass electrode at 37°C. The creatinine concentration in plasma and urine was determined by the method of Bonsnes and Taussky (1945) by Jaffe reaction.

The concentration of urea in plasma and urine was determined spectrophotometrically (Anonymous, 1982) using diacetylmonoxine method. Kanamycin concentration in plasma and urine was determined by microbiological assay according to the method of Arret *et al.* (1971) by using *Bacillus subtilis* as test organism.

Calculations

The concentration of kanamycin in plasma and urine sample, rate of urine flow in a time period of diuresis and renal clearance was determined as:

Concentration of Kanamycin = Size of zone x Standard factor

$$\text{Diuresis } (\text{ml.min}^{-1}\text{kg}^{-1}) = \frac{\text{Urine volume in a collection time period (ml)}}{\text{Time (min) x Body weight (kg)}}$$

Renal clearance ($\text{ml.min}^{-1}\text{kg}^{-1}$) = Uc x D/Pc

Uc = Concentration of substance in urine

D = Diuresis

Pc = Concentration of a substance in plasma

The statistical calculations were done according to the standard method and results have been presented as mean ± SE. Correlations between renal clearance and plasma concentration of urea and kanamycin and pH of urine and diuresis were determined by regression/correlation analysis by using Microsoft Excel 97 software.

RESULTS AND DISCUSSION

The results showing mean ± SE of four experimental periods for pH of blood and urine, diuresis, plasma and urine concentration of cretainine, urea and kanamycin and renal clearance of creatinine, urea and kanamycin have been presented in Table 1.

In this study cows showed mean ± SE value for blood pH 7.37 ± 0.04. These values are lower than the previously reported values of blood pH 7.69 and 8.06 in cows during summer and winter seasons, respectively (Akhtar, 1987) and 7.67 ± 0.02 (Iqbal, 1994). The blood pH in indigenous species of ruminants was reported between 7.30 to 8.40 (Nawaz and Shah, 1984; Nawaz, 1994). Like blood, urine pH 7.91 ± 0.07 in the present study showed larger variations when compared with 8.45 and 8.47 during summer and winter seasons, respectively (Akhtar, 1987) and 8.58 ± 0.04 (Iqbal, 1994).

Earlier studies on renal clearance of exogenous inulin and endogenous creatinine in indigenous buffaloes, cows, goats and sheep (Nawaz, 1994) indicated that the renal clearance of endogenous creatinine was a satisfactory and convenient measure of FGR in these species. Therefore, renal clearance of endogenous creatinine was used as an index of glomerular filtration rate (GFR) in cows. The average ± SE value for renal clearance of endogenous creatinine or GFR was 0.60 ± 0.08 $\text{ml.min}^{-1}\text{kg}^{-1}$ comparable to 0.57 ± 0.03 $\text{ml.min}^{-1}\text{kg}^{-1}$ in cow (Iqbal, 1994) and 0.54 $\text{ml.min}^{-1}\text{kg}^{-1}$ during summer (Akhtar, 1987). The value of GFR in the indigenous species of ruminants were always lower than literature values (Nawaz, 1994). Lower GFR under indigenous conditions warrants an exploration of role of kidneys in the maintenance of melieu interieur and in excretion of endogenous substances and xenobiotics including drugs.

Table 1: Mean ± SE values for body weight, pH of blood and urine, diuresis, plasma concentrations and renal clearance of creatinine, urea and kanamycin in 8 cows.

Cow No.	Body Weight (Kg)	pH		Diuresis	Concentration µg/mL						Renal Clearance		
		Blood	Urine	$\text{ml.min}^{-1}.\text{kg}^{-1}$	Creatinine		Urea		Kanamycin		$\text{ml.min}^{-1}.\text{kg}^{-1}$		
					Plasma	Urine	Plasma	Urine	Plasma	Urine	Creatinine	Urea	Kanamycin
1	400	7.36	7.70	0.031	9.80	221	136	772	2.93	10.1	0.71	0.18	0.11
2	410	7.33	7.70	0.030	8.63	272	130	1419	3.87	11.8	0.93	0.32	0.09
3	430	7.53	8.03	0.022	4.20	147	103	881	3.87	9.2	0.77	0.19	0.05
4	410	7.38	8.08	0.020	19.65	486	142	369	3.22	17.6	0.49	0.05	0.11
5	440	7.25	7.68	0.014	13.65	472	122	467	6.12	22.6	0.47	0.05	0.05
6	440	7.30	8.03	0.028	13.13	281	82	808	3.87	17.2	0.60	0.28	0.13
7	421	7.49	7.90	0.033	14.78	119	68	574	5.58	14.6	0.26	0.27	0.09
8	375	7.35	8.13	0.017	12.09	401	165	403	3.41	8.2	0.55	0.04	0.04
Mean	416	7.37	7.91	0.024	11.99	300	119	712	4.11	13.9	0.60	0.17	0.08
±SE	8	0.04	0.07	0.003	1.73	53.1	12	130	0.43	1.89	0.08	0.04	0.01

The renal clearance of endogenous urea 0.17 ± 0.04 ml.min^{-1}kg^{-1} showed a significant ($p<0.01$) positive correlation ($r=0.53$) with diuresis (Fig.1) indicating back diffusion or real tubular reabsorption. A significant ($p<0.01$) negative correlation ($r=-0.53$) between plasma concentration of urea and its renal clearance (Fig.2) indicated renal tubular active secretion of urea similar to the original observations made in the mammalian species sheep (Nawaz and Shah, 1984). Renal active tubular secretion of urea in cows appears to have not been documented earlier.

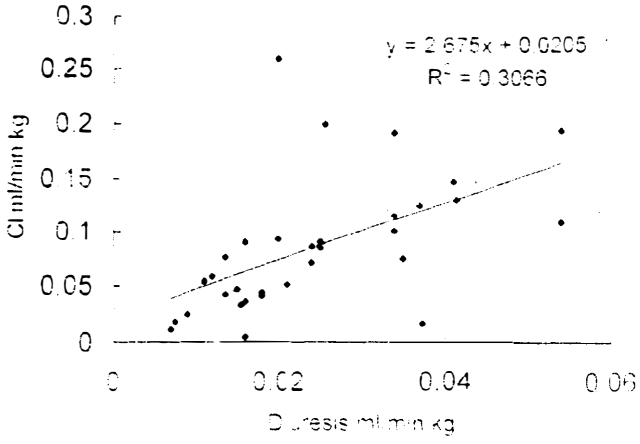

Fig.1: Showing highly significant ($P<0.01$) positive correlation between urine flow rate and renal clearance of urea in 8 cows intravenously injected kanamycin 5 mg/kg body weigth dose. Each data point is an average of 4 experimental periods in each animal

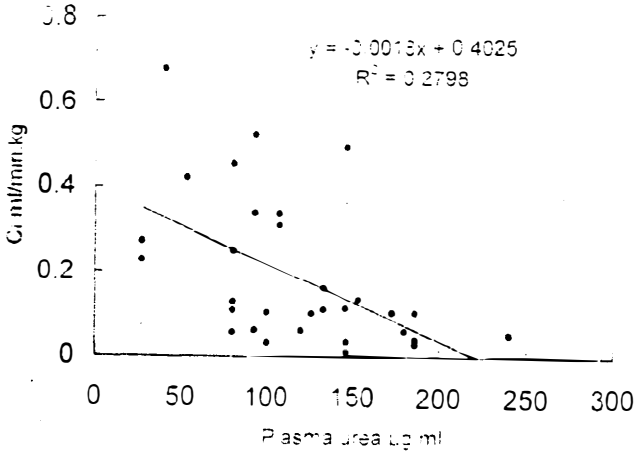

Fig.2: Showing highly significant ($P<0.01$) positive correlation between urine flow rate and renal clearance ofkanamycin in 8 cows intravenously injected kanamycin 5 mg/kg body weigth dose. Each data point is an average of 4 experimental periods in each animal.

The urine flow rate (diuresis) was 0.024 ± .0.003 ml.min^{-1}kg^{-1} being similar to 0.030 ± 0.004 ml.min^{-1}kg^{-1} in cows (Iqbal, 1994). Renal clearance of kanamycin 0.08 ± 0.01 ml.min^{-1}kg^{-1} was higher than 0.038 ml.min^{-1}kg^{-1} buffaloes (Saeed, 1994) and was lower than 0.33 ml. min^{-1} kg^{-1} in goats (Rasheed, 1993) and 0.263 ml.min^{-1}kg^{-1} in sheep (Aslam, 1993). A highly significant positive correlation observed between diuresis and kanamycin clearance (Fig. 1) indicated back diffusion or renal tubular reabsorption of kanamycin. The urine pH and plasma concentration of the drug did not reveal any correlation with the renal clearance of kanamycin. These results indicate that the renal handling of kanamycin in cows involved glomerular filtration and tubular reabsorption or back diffusion.

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