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Renal Handling of Acetylsalicylic Acid in Female Volunteers

Bushra Majeed, Haq Nawaz Bhatti and Kouser Fatima
Department of Chemistry, University of Agriculture, Faisalabad, Pakistan

Abstract: Acetylsalicylic acid is unique among the Non-steroidal Anti-Inflammatory Drugs (NSAIDs) in irreversibly acetylated cyclooxygenase. Renal clearance of endogenous creatinine and salicylate (as free salicylic acid) was investigated in twelve healthy female volunteers after oral dose of 600 mg soluble aspirin. Blood and urine samples were collected at predetermined time intervals. The concentration of free salicylic acid and creatinine in plasma and urine was determined colorimetrically. The average \pm SE values, for free salicylic acid in plasma was 25.569 ± 0.60 and in urine was $37.892 \pm 4.537 \mu\text{g mL}^{-1}$, for diuresis $0.034 \pm 0.005 \text{ mL min}^{-1} \text{ kg}^{-1}$, concentration of creatinine in plasma was 14.16 ± 0.55 and in urine was $493.22 \pm 62.5 \mu\text{g mL}^{-1}$, renal clearance of creatinine was 1.438 ± 0.30 and free salicylic acid was $0.082 \pm 0.016 \text{ mL min}^{-1} \text{ kg}^{-1}$ and clearance ratio was 0.092 ± 0.023 . It was found that renal clearance of aspirin was low under indigenous conditions.

Key words: Aspirin, salicylate, creatinine, plasma, urine, renal handling and clearance ratio

Introduction

Acetylsalicylic acid most commonly known as "Aspirin" is one of the most popular drug in the world. Although it is xenobiotic but no doubt "Aspirin a wonder drug that works wonder".

Despite the introduction of many new drugs, aspirin (ASA) is the most widely prescribed analgesic, antipyretic and anti-inflammatory agent and is standard for the comparison of others. Aspirin is the common household analgesic (Insel, 1996). In addition to its role as an analgesic, aspirin is being used in the prophylaxis of ischemic heart disease and strokes. Aspirin desensitization has a role in the management of aspirin-induced asthma (AIA) (Babu and Salvi, 2000). Aspirin inhibits the synthesis of a class of hormones called prostaglandins, molecules that cause pain, fever and inflammation when present in the blood stream in higher than normal levels. Salicylic acid's mode of action is irreversible inhibition of cyclooxygenase, an enzyme necessary for the production of prostaglandins (Stoker, 2001).

Renal clearance of a drug results in its appearance as such in the urine changes in the pharmacokinetic properties of drugs due to renal disease may also be explained in terms of clearance concepts (Leslie *et al.*, 1996). Impairment of renal function, decreased gastric mucosal cytoprotection and inhibition of platelet aggregation can also result from inhibiting prostaglandin synthesis (Gold Standard Multimedia, 2001).

A series of indigenous studies in human beings and animals have clearly indicated that the kinetic behaviour, metabolism and urinary excretion of investigated drugs were different under indigenous condition when compared with values given in literature (Nawaz, 1994; Rashid *et al.*, 2001).

In view of therapeutic views and toxicity problems studies dealing with renal handling of the salicylates are exceedingly desirable. Therefore the present project was designed to study the renal handling of aspirin in human female volunteers under indigenous conditions.

Materials and Methods

Subject: Twelve healthy young female volunteers having age 20-22 years and weight 50-63 kg were selected for this study. Each volunteer was informed with the aims and course of study and written concern was obtained from each participant.

Study protocol: Each volunteer after overnight fastening received orally 600 mg Disprin (soluble aspirin). Volunteers did not receive any medication seven days prior to and during course of study. A sterilized plastic branula was inserted intravenously. Blood samples (5 ml) were drawn at 0, 15, 30, 45, 60, 90, 120, 150, 180, 240, 360, 480 and 600 min. Blood samples were rapidly transferred to plastic vials containing few drops of heparinized solution. After immediate centrifugation plasma was separated and stored at -20°C for analysis. Similarly urine

samples were collected at 30, 60, 120, 180, 240 and 600 minutes. The total volume of urine voided during this time was noted. The pH of all blood and urine samples was recorded. Known volume of urine sample was preserved in plastic bottles in freezer at -20°C for analysis.

Blood analysis: The plasma salicylate levels were determined by a validated colorimetric method of analysis (Martens and Meyer, 1995).

Urine analysis: Quantitative determination of an acetylsalicylic acid as free salicylic acid was carried out by a validated colorimetric method of Levy and Procknal as modified by Farid *et al.* (1975). It was based on selective extraction of urine sample with carbon tetrachloride (to extract SA) and reextraction into ferric nitrate reagent.

Renal clearance: The renal clearance of endogenous creatinine was used for the estimation of glomerular filtration rate (GFR). The endogenous creatinine renal clearance was measured in plasma and urine samples colorimetrically by Bonsnes and Tausky method (1945).

$$\text{Diuresis (D)} = \frac{\text{Volume of urine in a collection period (mL)}}{\text{Time (min)} \times \text{Body weight (kg)}}$$

$$\text{Drug concentration } (\mu\text{g mL}^{-1}) = \text{Absorbance} \times \text{standard factor}$$

$$\text{Creatinine concentration} = \text{Absorbance} \times \text{standard factor} \times \text{dilution factor} \quad (\mu\text{g mL}^{-1})$$

$$\text{Renal clearance} = \frac{\text{Urine conc. (Uc)} \times \text{Diuresis (D)}}{\text{Plasma conc. (Pc)}} \quad (\text{ml min}^{-1}\text{kg}^{-1})$$

$$\text{Clearance ratio} = \frac{\text{Renal clearance of drug (Cl}_d\text{)}}{\text{Renal clearance of creatinine (Cl}_{cr}\text{)}}$$

Statistical analysis: The results obtained were presented at mean \pm SE. The data was tabulated and subjected to regression/correlation analysis (Steel and Torrie, 1992).

Results and Discussion

The results showing average \pm SE values for average body weight, pH of blood and urine, diuresis, plasma and urine concentration of creatinine and salicylic acid, renal clearance and clearance ratio of creatinine and drug are given in Table 1 and graphically shown in Fig. 1, 2 and 3. Average \pm SE values for the pH of blood and urine in twelve female volunteers were 7.60 ± 0.008 and 5.98 ± 0.13

respectively and comparable to blood pH 7.6 ± 0.01 and urine pH 5.8 ± 0.01 (Ghaffar, 1999).

Beside seasonal influence, the urine pH markedly influenced by the type of food which is different in summer and winter season leading to a lower urinary pH. Present study showed an average \pm SE diuresis value $0.034 \pm 0.005 \text{ ml min}^{-1}\text{kg}^{-1}$. The average \pm SE diuresis value was 0.037 ± 0.0189 in male volunteers which is higher than the present value (Fouzia, 1998). The difference is due to environmental difference because in hot climate, the evaporation reduces the urine flow during summer while environmental temperature increases the rate of urine during winter (Nawaz and Shah, 1984). The average \pm SE value for the SA concentration in plasma and urine was 25.69 ± 0.60 and $37.892 \pm 4.537 \mu\text{g mL}^{-1}$, respectively. In earlier studies the average \pm SE value excreted in urine is $4.122 \pm 0.873 \mu\text{g mL}^{-1}$ in male volunteers (Fouzia, 1998). It is due to environmental difference. The major difference is due to non-fasting volunteers. The excretion was increased when pH of urine was >8.0 (Wesley, 1990).

The average \pm SE value for the concentration of creatinine in plasma was 14.16 ± 0.55 and urine was $493.22 \pm 62.5 \mu\text{g mL}^{-1}$. The average \pm SE values for creatinine in plasma was $9.468 \pm 0.430 \mu\text{g mL}^{-1}$ for female volunteers (Ayesha, 2001). The average \pm SE value of creatinine concentration in urine was $537 \pm 14.2 \mu\text{g mL}^{-1}$ reported in female volunteers (Uzma, 1997). The difference is due to the change in environmental conditions, age, weight and genome. The studies indicate that GFR is lower in local species than given in international literature. Renal clearance of creatinine and salicylic acid was 1.438 ± 0.30 and $0.082 \pm 0.016 \text{ ml min}^{-1}\text{kg}^{-1}$ respectively in present study. The present value is less than $2 \text{ ml min}^{-1}\text{kg}^{-1}$ given in literature (Ganong, 1997). The difference is due to environmental influence on the genetics.

The average \pm SE value of clearance ratio was 0.093 ± 0.023 . The clearance of salicylic acid is lower than the endogenous creatinine. The clearance ratio is less than 1, it indicates that reabsorption of salicylic acid takes place. There is non-significant positive correlation between diuresis and salicylic acid clearance which shows that back diffusion of drug takes place (Hardman *et al.*, 1996). There is non-significant negative correlation between urine pH and SA clearance. Acid drug can excrete only in basic urine while according to data, urine is also acidic, so drug does not excrete out easily. It remains in unionized form and reabsorb again. There is also non-significant positive correlation between plasma concentration of SA and clearance ratio, correlation between diuresis and

Table 1: Average data of renal clearance of endogenous creatinine and aspirin in female volunteers after oral dose of 2 x 300 mg tablets

Volunteers	Body weight (kg)	Diuresis (ml/min/kg)	pH		Concentration ($\mu\text{g ml}^{-1}$)				Renal Clearance ml/min/kg		Clearance ratio $\text{Cl}_{\text{drug}}/\text{Cl}_{\text{cr}}$
			Urine	Blood	Creatinine		Drug		Creatinine	Drug	
					Urine	Plasma	Urine	Plasma			
1	53	0.04	5.54	7.57	354.77	14.15	23.55	23.26	1.50	0.06	0.06
2	57	0.06	5.42	7.59	204.09	10.40	24.66	24.04	1.39	0.14	0.07
3	52	0.05	6.91	7.56	410.55	12.64	34.38	26.40	2.92	0.11	0.10
4	57	0.05	6.71	7.57	494.47	15.72	43.05	28.75	1.71	0.09	0.05
5	61	0.01	6.11	7.59	260.93	12.96	36.01	25.09	0.37	0.03	0.09
6	62	0.03	5.67	7.60	446.01	11.46	31.03	27.31	1.04	0.05	0.07
7	60	0.01	5.79	7.61	617.79	14.14	67.19	28.62	0.68	0.04	0.09
8	60	0.02	5.69	7.67	457.43	15.95	28.64	26.92	0.47	0.02	0.04
9	63	0.03	6.10	7.57	498.41	15.32	29.26	24.31	1.05	0.05	0.07
10	57	0.02	6.06	7.56	396.12	15.56	70.79	24.57	0.86	0.07	0.34
11	50	0.07	5.69	7.57	956.63	15.48	40.14	25.48	4.03	0.21	0.07
12	60	0.04	6.03	7.61	821.50	16.19	25.99	22.08	1.23	0.11	0.06
Mean	57.667	0.0348	5.977	7.590	493.2257	14.165	37.8922	25.5695	1.4383	0.0822	0.0916
\pm SE	0.1696	0.0054	0.130	0.009	62.5094	0.5513	4.5429	0.6013	0.3062	0.0158	0.0229

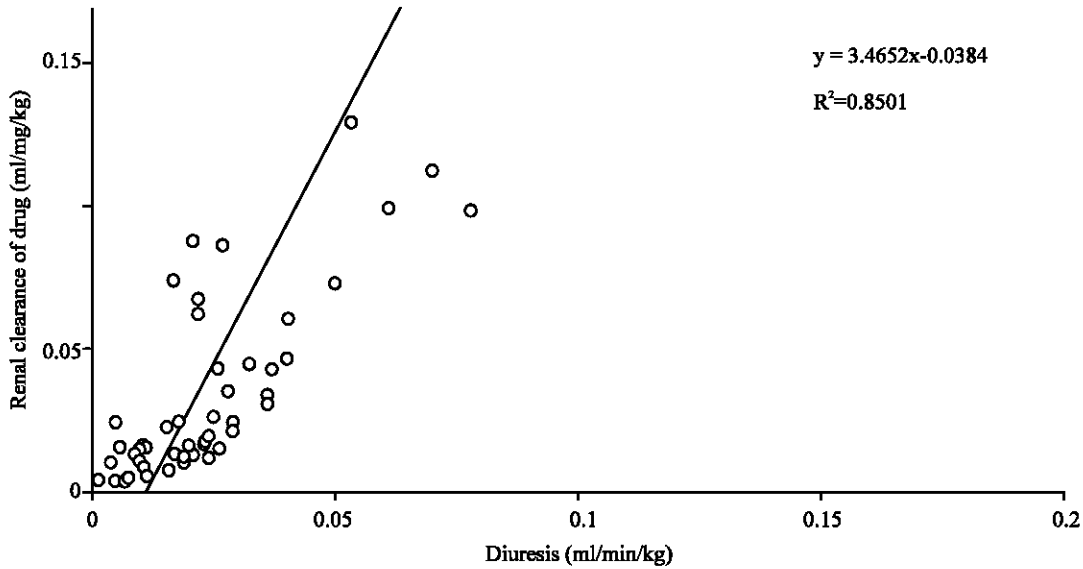


Fig. 1: Relationship of diuresis with renal clearance of Salicylic acid in 12 healthy female volunteers

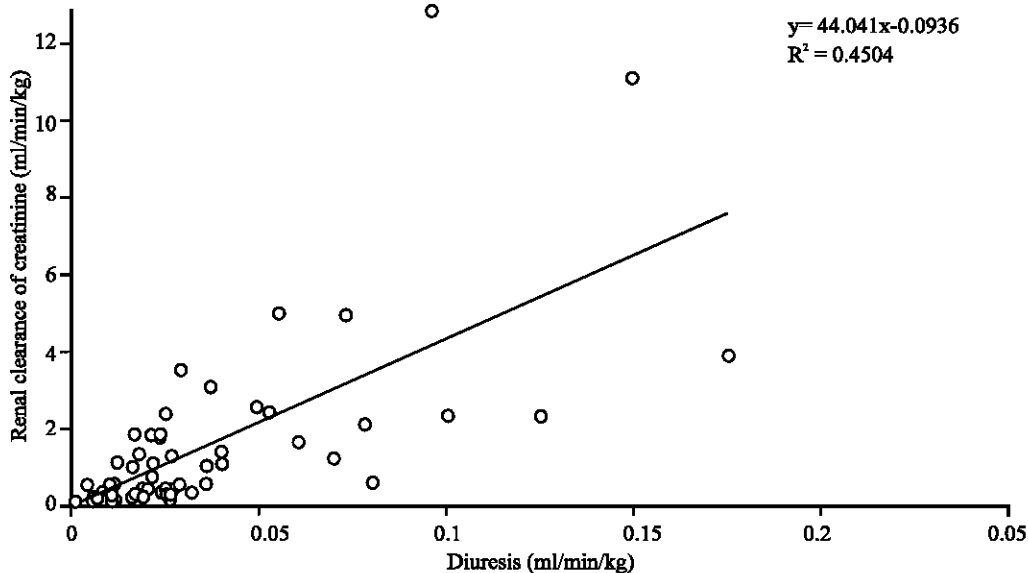


Fig. 2: Relationship between diureses and creatinine renal clearance in 12 healthy female volunteers

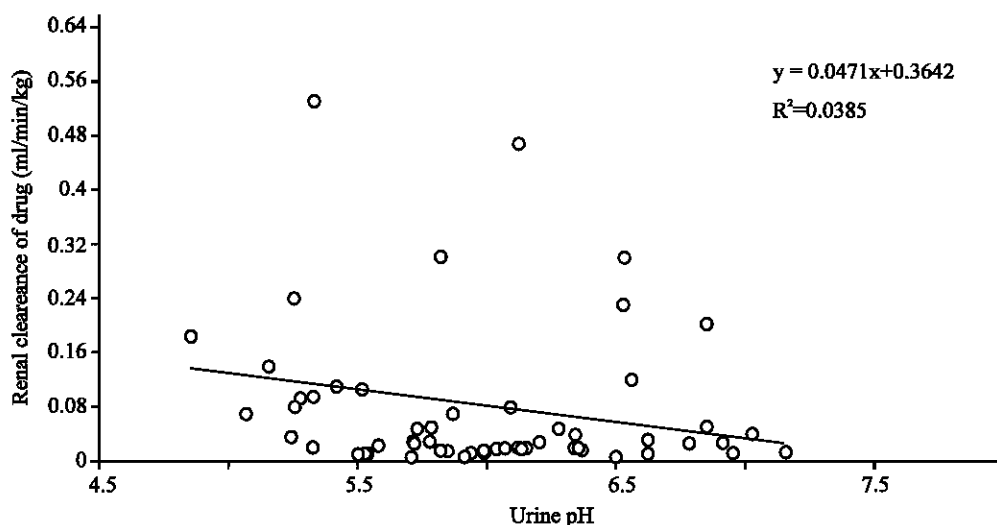


Fig. 3: Relationship between pH of urine and renal clearance of Salicylic acid in 12 healthy female volunteers

clearance ratio. It indicates that there is direct relationship between these two parameters. It is concluded that besides glomerular filtration mechanism, reabsorption was also involved in salicylic acid excretion.

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