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Excretion of Aspirin Through Urine of Female Volunteers

Fouzia Zahid, Rakhshanda Nawaz, Zahid Mahmood, Muhammad Shahid and [1]Khurram Shahzad

The present study was conducted for urinary excretion of aspirin in adult female volunteers. Aspirin (2×300 mg) was given to each volunteer orally and urine samples were collected at different time intervals and analyzed by spectrophotometric and thin layer chromatographic methods. The results indicated that the free aspirin excreted as salicylic acid at 720 min was 0.142 ± 0.073, 0.029 ± 0.015 and conjugated drug was 0.066 ± 0.022 mg, respectively. The value of dose excreted as total salicylic acid in the urine was 0.594 ± 0.108%. These values deviate from the literature values emphasizing that such kind of studies must be conducted in local populations so that an appropriate dosage can prescribed.

Key words: Aspirin, urinary excretion, female volunteers, acetylsalicylic acid, TLC, spectrophotometer

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For further information about this article or if you need reprints, please contact:

Dr. Muhammad Shahid
Department of Chemistry
University of Agriculture
Faisalabad, Pakistan

ANSInet
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Department of Chemistry, University of Agriculture, Faisalabad, Pakistan
[1]Pakistan Council of Scientific Industrial Research Laboratories Complex, Ferozepur Road, Lahore, Pakistan

Introduction

Acetyl salicylic acid (aspirin) is non-steroidal anti-inflammatory drugs (NSAIDs) of the most commonly used remedies over a century. Aspirin is most extensively employed as analgesic, antipyretic and anti-inflammatory drug in conditions such as rheumatoid arthritis. Aspirin is drug of choice for treating the majority of articular and musculoskeletal disorder due to its low cost, greater efficiency and long history of safety (Katzung, 2001). Aspirin is used as antibiotic primer, antihistaminic, decongestant, cold remedy and in minor injuries. It is also used for relief of fever, headache, pain and arthritic complaints. It is mild CNS depressant but does not lead to addiction (Anonymous, 1970).

Aspirin also increases the bleeding time, decrease platelet adhesions and in large doses, may cause hypoprothrombinaemia (Reynolds ***et al.***, 1989).

Aspirin is rapidly deacetylated to salicylic acid which is pharmacologically active metabolite. Salicyclic acid then oxidized its gentisic acid and conjugated with glycine to form salicyluric acid and with glucuronide to form ester and ether conjugates. Metabolism is affected by many factors such as age, species, disease conditions, strains, hormones, sex, high altitudes, route and climatic conditions (Pottage, 1979).

The fate of certain chemicals including drugs have been extensively investigated under indigenous environments. These investigations have revealed difference between the foreign and local species due to the difference in geographical and genetic conditions (Nawaz and Shah, 1985; Nawaz ***et al.***, 1988). The objective of this experiment was to study metabolism and urinary excretion of aspirin as free and conjugated under indigenous conditions.

Materials and Methods

Drug administration and sampling

The study was conducted in the Departments of Physiology and Pharmacology and Chemistry in six months duration. Urinary excretion of aspirin was investigated in ten healthy human female volunteers after oral administration of 2 × 300 mg of Reckitt and Colman Pakistan Ltd. tablets to each volunteer. The urine samples were collected at 30, 60, 90, 120, 150, 180, 360, 540 and 720 min and a control (before drug administration) pH and volume of urine were also recorded.

The standard solutions of aspirin concentrations 5, 10, 15, 20, 25, 30, 35, 40, 45 and 50 μg ml^{-1} were prepared in blank urine.

Analytical determination

Spectrophotometric method

Each urine sample was analyzed at least in triplicate by spectrophotometric method (Farid ***et al.***, 1975), taking 5 ml sample and 1 ml colouring reagent (1% ferric nitrate in 1:99 nitric acid:distilled water) violet colour was develop, centrifuged and noted the absorbance at 525 nm by spectrophotometer.

The amount of conjugates was estimated after hydrolysis of 5 ml sample by 1mL of 0.25M HCl then boiled for 1h added colouring reagent and recorded the absorbance. This give the total

amount of salicylic acid formed as a result of hydrolysis of conjugates. The difference in concentration gave the amount of free conjugates.

TLC method

Aspirin metabolites were determined by TLC method used by Chrastil and Wilson (1978). Random sample was alkalized by 0.2M NaOH (pH 8.5), then 14 ml ethyl acetate and 1 ml ether were added and shacked for 2 min. Ten ml of lower layer was transferors and acidified by 1ml concentrated HCl. Standard solution of aspirin (1 mg 100 ml^{-1}) was made in blank urine and treated as above. They hydrolyzed urine was treated in the same way (no standard was added) except that before extraction after addition of HCl, the urine was boiled for 2h; cooled and extracted by 14 ml ethyl acetate and 1 ml of ether.

Standard sample (unhydrolyzed and hydrolyzed) 10 μl of each was applied on the TLC (silica gel) plate developed with diethyl ether n-butyric acid (10:1). The plate was studied after spraying with coloring reagent. Plates were kept under iodine vapour and U.V. lamp. Different spots appeared and identified by calculating the Rf values and compared with standards and calculate the urinary excretion as

$$\text{Dose excreted (\%)} = \frac{\text{Amount of drug excreted}}{\text{Dose of drug}} \times 100$$

$$\text{Cumulative dose excreted (\%)} = \frac{\text{Cumulative amount excreted}}{\text{Dose of drug}} \times 100$$

Results and Discussion

The metabolism and excretion of aspirin in urine of 10 healthy female was investigated. The mean of volumeter was 22.7 ± 0.202 year and mean body weight was 48.1 ± 2.91 kg.

Roderick ***et al.*** (1985) studied the excretion of salicylate in man. He reported 10% free salicylic acid was excreted in urine was excreted in urine as free salicylic acid 10%. In this study excretion of free salicylate was 0.344%. Excretion of free salicylate is extremely variable and depends upon both the dose and urinary pH. In alkaline urine, more than 30% of ingested drug might be eliminated as free salicylate whereas in acidic urine it might be as low as 2%.

This study showed that 0.594% of total mean drug was excreted in urine in 12 h while Patel ***et al.*** (1990) reported the metabolism of aspirin in man and rat after (10-100 mg kg^{-1}) administration and resulted in excretion of 81.91% of dose in urine in 24 h.

Total urinary recovery was 22.4% of dose after 500 mg administration of aspirin in man (Brume ***et al.***, 1993) but total urinary recovery in present study is 0.594% with SEM 0.108 after an oral dose of 2 × 300 mg of aspirin. Excretion rate is effected by dose and pH of urine.

Excretion of free salicylate was extremely variable and depends upon pH. The excretion

was increased when pH of urine was > 8.0 (Wesley, 1990). But in this study, pH of collected samples was < 8.0 due to which low excretion of salicylate takes place.

The excretion of aspirin is effected by sex and environment to show the effect of sex. The cumulative amount of total aspirin excreted as total salicylic acid in female volunteers is 0.170 ± 0.077, 0.566 ± 0.195, 0.937 ± 0.278, 1.407 ± 0.349, 1.654 ± 0.431, 2.170 ± 0.518, 3.072 ± 0.629, 3.468 ± 0.639 and 3.565 ± 0.640 mg but in case of male volunteers (Table 2), the cumulative amount of total aspirin excreted as total salicylic acid was 0.315 ± 0.109, 0.902 ± 0.204, 0.749 ± 0.396, 2.34 ± 0.673, 3.154 ± 0.675, 3.822 ± 0.773, 5.007 ± 1.01 and 5.263 ± 1.062 mg. It is seen that total aspirin excretion in male volunteers was higher than in female volunteers.

It is noted that excretion of conjugated drug is higher in female volunteers than male volunteers. The mean cumulative of conjugated salicylic acid (in female) is 1.314 ± 0.232 mg and percentage is 0.280 ± 0.069 and in males cumulative of conjugate is 1.081 ± 0.327 mg and cumulative is 0.181 ± 0.054% (Bakar and Niazi, 1983).

The estimated metabolites in present study are salicylic, gentisic and salicyluric acid by TLC while Liu and Smith (1996) determined salicylic acid, salicyl acyl acid glucuronide, salicyluric acid and gentisic acid in urine by HPLC. Aspirin is quickly deacetylated to salicylic acid, thus no aspirin spot is found in urine. These differences are due to lack of experimental facilities.

Table 1: The mean amount of aspirin excreted as free, conjugated and total salicylic acid in urine of female volunteers

	Time (min)								
Drug form	30	60	90	120	150	180	360	540	720
Free	0.142	0.317	0.281	0.161	0.278	0.332	0.482	0.143	0.029
Conjugated	0.020	0.077	0.087	0.092	0.131	0.180	0.415	0.248	0.066
Total	0.170	0.396	0.371	0.306	0.411	0.669	0.901	0.396	0.097

Table 2: The mean cumulative amount of aspirin excreted as free, conjugated and total salicylic acid in urine of female volunteers

Drug form	Time (min)								
	30	60	90	120	150	180	360	540	720
Free	0.140	0.459	0.740	0.951	1.229	1.561	2.063	2.186	2.215
Conjugated	0.020	0.096	0.183	0.275	0.406	0.586	1.001	1.249	1.314
Total	0.170	0.566	0.937	1.407	1.654	2.170	3.072	3.468	3.565

Table 3: The mean %age dose of aspirin (mg) excreted as free, conjugated and total salicylic acid in urine of female volunteers

Drug form	Time (min)								
	30	60	90	120	150	180	360	540	720
Free	0.024	0.053	0.047	0.027	0.046	0.055	0.080	0.024	0.005
Conjugated	0.003	0.013	0.040	0.015	0.059	0.030	0.069	0.041	0.011
Total	0.028	0.066	0.120	0.051	0.072	0.086	0.150	0.066	0.016

Table 4: The mean cumulative %age dose of aspirin excreted as free, conjugated and total salicylic acid in urine of female volunteers

Drug form	Time (min)								
	30	60	90	120	150	180	360	540	720
Free	0.024	0.076	0.123	0.135	0.181	0.236	0.316	0.340	0.344
Conjugated	0.003	0.016	0.055	0.070	0.129	0.159	0.226	0.269	0.280
Total	0.028	0.094	0.155	0.206	0.278	0.364	0.513	0.579	0.594

According to present study, the mean cumulative amount of free salicylic acid is (Table 2) 2.215 ± 0.586 mg and percentage of cumulative free salicylic acid is 0.344 ± 0.094% (Table 4) which is less than male volunteers that as 6.74 ± 0.711 and 0.568 ± 0.126%. Almost same kind of deviation was also observed in mean %age dose of aspirin excreted as free conjugated and total salicylic acid in urine of female volunteers. The mean %age dose of aspirin (mg) excreted as free, conjugated and total salicylic acid in urine of female volunteer at time 30, 720 min were 0.024, 0.005, 0.003, 0.011, 0.28, 0.0016%. The total %age dose of aspirin (mg) as free 0.361%, conjugated 0.281% and total 0.655% were calculated in female volunteers (Table 3) that the values in male were different to present study. This difference may be due to sex variation, fluctuation in urine pH, environmental conditions and nutritional ingredients. Species variation may also affect the metabolism and urinary excretion (Nawaz ***et al.***, 1988).

References

Anonymous, 1970. Encyclopedia International. Grolier Incorporated, New York, pp: 188.

Bakar, S.K. and S. Niazi, 1983. High performance liquid chromatographic determination of aspirin and its metabolites in plasma and urine. J. Pharm. Sci., 72: 1020-1023.

Brume, K., B.Neurnberg and H.T. Schneider, 1993. Biliary elimination of aspirin after oral and intravenous administration in patient. Agents. Actions. Suppl., 44:51-57.

Chrastil, J. and J.T. Wilson, 1978. Quantitative estimation of salicylic acid and its metabolites by thin layer chromatography. J. Chromatogar., 152: 183-189.

Farid, N.A., G.S. Boron, W.V. Kessler, S.M. Shaw and W.E. Lange, 1975. Improved calorimetric determination of salicylic acid and its metabolites in urine. Clin. Chem., 21: 1167-1168.

Katzung, B.G., 2001. Basic and Clinical Pharmacology, 8th Ed. McGraw Hill Co. New York, pp: 599-603.

Liu, J.H. and P.C. Smith, 1996. Direct analysis of salicylic acids, salicylacyl glucuronide, salicyluric acid and gentisic acid in human plasma and urine by high performance liquid chromatography. J. Chromatogr. B. Biomed. Appl., 675: 61-70.

Nawaz, M. and B.H. Shah, 1985. Geonetical consideration in the quality assurance of pharmaceuticals. Proc. Int. Seminar on Policies Management and Quality Assurance of Pharmaceuticals. 21-25th April WHO and Ministry of Health, Govt. Pakistan.

Nawaz, M., T. Iqbal and R. Nawaz, 1988. Geonetical considerations in disposition kinetics evaluation of chemotherapeutic agents. Vet. Pharmacol. Toxicol and Therapy in Food Producing Animals. 2:260. Proc. 4th Cong. Europ. Assoc. Vet. Pharmacol Therap., 28th August-2nd September Budapest.

Patel, D.K., L.J. Notarianni and P.N. Bennett, 1990. Comparative metabolism of high doses of aspirin in man and rat. Xenobiotica, 20: 847-854.

Pottage, A., 1979. Mechanism involved in drug action. Clin. Pharmacology, 24th ed. MacMillan, New York.

Reynolds, J.E., F.K. Parfitt, A.U. Parson and S.C. Sweetman, 1989. Martindal, the extra pharmacopoeia. The Pharmaceutical Press, London, pp: 32.

Roderick, J., F.M. Salvandor and J.R. Vane, 1985. The Pharmacological Basis of Therapeutics, 7th ed. MacMillan, New York, pp: 686.

Wesley, G.C., D.Q. Craig, M.D. Brater and A.R. Johnson, 1990. Goth's medical pharmacology. Asian Student Edition. Galgotia Publication, Ltd.