Absorption, Metabolism, and Excretion of Salicylic Phenolic Glucuronide in Rats

By GERHARD LEVY, LEONARD WEINTRAUB*, TAI MATSUZAWA, and STANLEY R. OLES*

Salicylic phenolic glucuronide is excreted rapidly by apparent first-order kinetics ($t_{1/2}$ about 0.7 hr.) after intravenous administration to adult rats. Only a small fraction of an injected dose is hydrolyzed *in vivo* to salicylic acid. The glucuronide is apparently not absorbed significantly as such from the gastrointestinal tract. Part of an orally administered dose is absorbed apparently from the lower region of the intestinal tract after hydrolysis to salicylic acid.

THE PHARMACOKINETICS of elimination of salicylic acid and its major metabolite, salicyluric acid, have been studied in some detail in man and rats (1–4). Much less is known about the kinetics of elimination of salicylic phenolic glucuronide, another important metabolite of salicylic acid.[1] This glucuronide has been prepared in pure form and (since technical problems did not permit its intravenous administration to man) its absorption, metabolism, and excretion have been studied in rats. A previous study has shown that the pharmacokinetics of salicylate elimination in man and rats are quite similar (4).

EXPERIMENTAL

Synthesis of Salicylic Phenolic Glucuronide.—Methyl (tri-*O*-acetyl-α-D-glucopyranosyl bromide) uronate was prepared from glucuronolactone by the procedure of Bollenback *et al.* (6). From it, *o*-carboxyphenyl-β-D-glucopyranosiduronic acid (salicylic phenolic glucuronide) was obtained by methods described by Lunsford and Murphey (7), m.p. 144.5–146°. (Lit. 145–146°.) $[\alpha]_D^{27} = -70.8°$ (C, 6,H_2O). [Lit. $[\alpha]_D^{23} = -75.9°$ (C,6,H_2O).]

Anal.—Calcd. for $C_{13}H_{14}O_9$: C, 49.69; H, 4.49. Found: C, 49.64, 49.73; H, 4.87, 4.72.

No free salicylic acid was detected by a thin-layer chromatographic method capable of detecting 0.5% of this compound in the glucuronide.

Animal Study.—Male Sprague-Dawley rats, weighing 250–400 Gm., were fasted for 24 hr. prior to drug administration but had unrestricted access to drinking water at all times. The drug was dissolved in 1.5 ml. distilled water and injected intravenously into the femoral vein, or administered orally by stomach tube as 5 ml. aqueous solution. The rats were confined in plastic animal holders for the first 24 hr. in order to permit frequent urine and feces collections. The animals were then transferred to individual metabolic cages for another 24 hr. Food and water were freely available to the rats throughout the 48-hr. period of urine and feces collection.

Assay Methods.—Salicylic acid and its metabolites were determined in the urine by a modification of the methods of Smith *et al.* (8). Feces was homogenized with distilled water and assayed similarly.

RESULTS

The urinary excretion kinetics of salicylic phenolic glucuronide (SPG) as a function of time after intravenous injection are shown in Fig. 1. Excretion followed apparent first-order kinetics, with a half-life of about 0.7 hr. The slow initial excretion of SPG in one of the animals appears to be due to accidental extravascular injection of part of the dose. About 82% of the injected amount was recovered in the urine; of this, about 85% was excreted as SPG (Table I). A much smaller fraction of the dose (about 36%) was recovered in the urine after oral administration and the fraction of urinary salicylate excreted as SPG was also much smaller than after intravenous injection (Table I). No measurable amounts of salicylic acid or its metabolites were present in the feces after intravenous administration of SPG. The time course of urinary excretion of total salicylate after oral administration of SPG indicates an appreciable delay in absorption (Table II).

DISCUSSION

Man excretes SPG relatively rapidly by a combination of glomerular filtration and tubular secretion (9). The rapid excretion of intravenously administered SPG by rats is consistent with these observations.[2] The renal excretion kinetics of SPG are not affected by urine pH and urine flow rate (9); this probably accounts for the lack of significant variation between animals in the excretion half-life of SPG. The excretion of SPG mainly as such after intravenous administration suggests that β-glucuronidase mediated hydrolysis in the tissues is too slow to reduce appreciably the net formation rate of SPG after salicylic acid administration. Inhibitors of β-glucuronidase, such as glucuronolactone, are therefore unlikely to enhance salicylate elimination. This conclusion is consistent with the results of a recent study (4). The lack of biliary excretion of SPG, suggested by the absence of detectable amounts of salicylates in the feces and by the absence of secondary maxima in the excretion rate *versus* time curve after intravenous injection of SPG, is in agreement with the observations of Williams *et al.* (10). These workers recovered just

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[1] Only a small fraction of salicylic acid is biotransformed to salicylic acyl glucuronide in man (5).

[2] The rapid excretion of SPG after intravenous injection indicates also that the SPG found in the urine is not the product of conjugation of free salicylic acid derived from the *in vivo* hydrolysis of injected SPG. Excretion of SPG by the latter mechanism is rate-limited by the formation process and would proceed at a considerably lower rate (4).

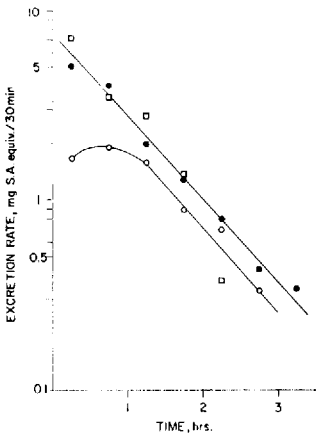

Fig. 1.—Urinary excretion rate of salicylic phenolic glucuronide in three rats as a function of time after intravenous injection of 120 mg./Kg. body weight. The slow initial excretion of SPG in one of the animals (○) appears to be due to accidental extravascular injection of part of the dose.

TABLE I.—URINARY RECOVERY OF SALICYLATE AFTER INTRAVENOUS AND ORAL ADMINISTRATION OF SALICYLIC PHENOLIC GLUCURONIDE TO RATS

Rt.	Animal	Dose, mg./Kg.	Total Urinary Recovery, % of Dose	% of Recovered Drug Excreted as Glucuronide
i.v.	*1*	120	86	86
i.v.	*2*	120	75	79
i.v.	*3*	120	84	89
Oral	*4*	600	41	22
Oral	*5*	600	28	6
Oral	*6*	600	39	29

1.5% of the dose (and only in the form of free salicylic acid) in the bile of rats within 24 hr. after i.p. injection of 50 mg. salicylic acid/Kg. body weight.

Vogt *et al.* (11) have shown that appreciable β-glucuronidase activity is located in the large intestine of the rat, but that no significant activity is present in the small intestine. They observed that desacetylbisacodyl, when administered as the glucuronide, is absorbed only in the large intestine, after enzymic hydrolysis. There is strong evidence

TABLE II.—TIME COURSE OF URINARY EXCRETION OF TOTAL SALICYLATE AFTER ORAL ADMINISTRATION OF SALICYLIC PHENOLIC GLUCURONIDE TO RATS[a]

Time, hr.	Amt. Excreted, mg. Salicylic Acid Equiv. Rat *4*	Rat *5*	Rat *6*
0–3	5.8	1.9	24.1[b]
3–5	2.0	1.2	
5–24	21.8	15.4	
24–48	2.8	4.9	12.1

[a] The administered amounts of SPG, expressed as mg. salicylic acid equivalent, were: rat *4*, 79.16 mg.; rat *5*, 83.55 mg.; rat *6*, 92.35 mg. [b] Only a single urine collection was made in rat *6* during the first 24 hr.

that other glucuronides also are not absorbed as such (10, 11). The approximately 4-hr. delay in absorption observed by Vogt *et al.* is similar to the delay in salicylate absorption after oral administration of SPG as judged by the time course of urinary excretion of total salicylates (Table II). The composition of urinary metabolites after oral administration of SPG (Table I) is similar to that which would be expected after administration of free salicylate (4). The very small SPG fraction in rat *5* appears to be due to the particularly slow absorption of salicylate in this animal and the resulting lack of saturation of the salicylurate formation process (4). The delayed and incomplete absorption of salicylate after oral administration of SPG, and the difference in the composition of urinary metabolites of SPG after oral and intravenous administration, respectively, suggest strongly that SPG is absorbed from the gastrointestinal tract of the rat only after hydrolysis to salicylic acid.

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