Synthesis of ^{14}C-Meglumine Salicylate and Its Disposition in Humans after Oral Administration

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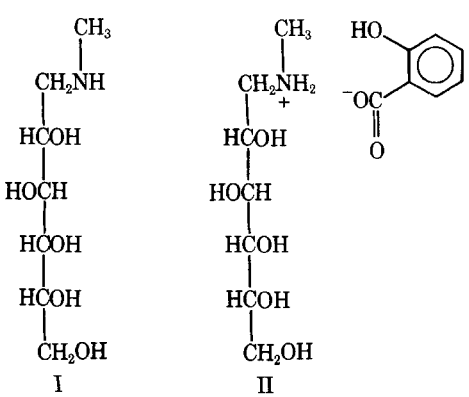
Abstract □ The synthesis of ^{14}C-meglumine salicylate was accomplished by heating ^{14}C-meglumine with salicylic acid, in equimolar ratios, in 2-propanol. The average radiochemical yield was 97.5%. Ten healthy adult male volunteers were given 1.2 g of the compound orally. Five took 1.2 g of 1-deoxy-1-[^{14}C]-methylamino-D-glucitol salicylate (containing about 47 μCi), and five others took 1.2 g of 1-deoxy-1-methylamino-D-[U-^{14}C]-glucitol salicylate (containing about 45 μCi). Urine and feces were collected for 5 days, and blood was sampled for 24 hr. The peak urinary excretion of meglumine and/or its metabolites occurred between 4 and 8 hr after administration (about 7.2% of the administered dose). Meglumine was excreted primarily in the feces (72.4% over 5 days) and, to a smaller extent, in urine (21.3% over 5 days). No activity was detected in blood. The excretion rate and percentage excreted were the same for both groups of subjects, suggesting that meglumine was not metabolized by *N*-demethylation or conversion to carbon dioxide. The highest blood salicylate level, 44.4 ± 1.9 μg/ml, was observed 1 hr after administration. Urinary levels of salicylic acid and its metabolites were observed to be at a maximum at 8 hr. Total salicylate recovery was 94.7 ± 1.5% in 48 hr. Salicyluric acid was the major metabolite, accounting for 69.5 ± 3.6% of the dose. Salicylic acid accounted for 6.8 ± 1.2%.

Keyphrases □ ^{14}C-Meglumine salicylate—synthesized, administered orally, biological distribution and excretion measured, humans □ Distribution, biological—^{14}C-meglumine salicylate, oral administration, humans □ Excretion—^{14}C-meglumine salicylate, oral administration, humans □ Radiochemistry—^{14}C-meglumine salicylate synthesized, administered orally, biological distribution and excretion measured, humans

Meglumine[1] (I) was first synthesized in 1935 (1). The use of I in pharmaceuticals began with the preparation of I antimonate for the treatment of leishmaniasis. Presently, several pharmaceutical preparations contain I. Some X-ray contrast media, for example, contain I as a solubilizing agent, and I was found to decrease the toxicity of such contrast media (2–4). Also, it was reported that I–coumermycin mixtures (4:1) elevated the blood antibiotic levels five- to 15-fold over those for the antibiotic alone (5, 6). The disposition of I in rats and dogs was reported recently (7).

Meglumine salicylate (II) is a promising new water-soluble salicylate under investigation. The suggested indications for the use of II include all conditions requiring systemic salicylate therapy. Since the compound is relatively new, its blood and urinary salicylate levels have not been reported. The literature also does not contain information about the disposition of I in humans.



The purpose of this study was to gain such information and to show the effect, if any, I had on the absorption and urinary excretion of the salicylate moiety. To this end, 1-deoxy-1-[^{14}C]-methylamino-D-glucitol salicylate (II-M) and 1-deoxy-1-methylamino-D-[U-^{14}C]-glucitol salicylate (II-G) were synthesized and administered orally to humans. Two different positions of ^{14}C-labeling were used to gain preliminary information on the metabolism of I in humans.

EXPERIMENTAL

Radiosyntheses—The syntheses of I-M and I-G were described previously (8). Several preliminary reactions for the synthesis of II were conducted with ethanol or 2-propanol as a reaction medium. When ethanol was used, as previously suggested (9), a syrup of II was formed. By using a small volume of 2-propanol and conducting the synthesis in low humidity (50%), II could be synthesized in high yields.

Compound II-M—A solution of 2.91 g of I-M (14.9 mmoles, 0.067 mCi/mmole) and 2.06 g of salicylic acid in 35 ml of 2-propanol was warmed in a water bath at 50° to allow complete dissolution of I-M. After the solution was allowed to cool to room temperature, it was placed in a refrigerator overnight. The resulting compound was filtered in a low humidity (50%) atmosphere, since it was slightly hygroscopic, and dried under vacuum. The yield was 4.90 g (first crop, 98.6% of theoretical) of II-M, mp 98–101°. The specific activity was 0.19 ± 0.002 μCi/mg or 0.063 mCi/mmole.

Anal.[2]—Calc. for $C_{14}H_{23}NO_8$: C, 50.45; H, 6.90; N, 4.20. Found: C, 50.52; H, 7.08; N, 4.09.

Compound II-G—A solution of 849 mg of I-G (4.35 mmoles, 0.204 mCi/mmole) and 601 mg of salicylic acid in 10 ml of 2-propanol was treated as described for II-M. The yield was 1.4 g (first crop, 96.5% of theoretical) of II-G, mp 98–100°. The specific activity was 0.56 ± 0.005 μCi/mg or 0.19 mCi/mmole.

Anal.—Found: C, 50.23; H, 7.30; N, 4.00.

A mixed melting point of each labeled compound gave no melting-point depression. The IR spectrum of II (Fig. 1) was compared with spectra of the labeled compounds. From these analyses, the two ^{14}C-labeled compounds were determined to be chemically pure.

TLC analysis of II for radiochemical purity determination was not successful. Solvent systems of varying polarities and compositions failed to give a single spot for II. Two spots, one corresponding to I and the other to salicylic acid, were obtained, apparently due to the dissociation of II when dissolved in water or methanol. The two labeled compounds were assumed to be radiochemically pure, since it was predetermined that the ^{14}C-labeled I compounds had a radiochemical purity of 98.4%.

Procedure—Two volunteer groups of five healthy male subjects were used. Their weight range was 66–90 kg (average 77.6 kg), and their age range was 21–29 years. Blood and urine samples were collected from each volunteer at 7:30 am to serve as controls. At 8 am following an overnight fast, each subject received orally a capsule (assigned by random number drawing) containing an exactly weighed amount of either II-M (referred to as the M group) or II-G (referred to as the G group). The range of

[1] 1-Deoxy-1-methylamino-D-glucitol or *N*-methylglucamine.

[2] Midwest Microlab, Ltd., Indianapolis, Ind.

Table I—Excretion of ^{14}C-Meglumine and/or Its Metabolites in Urine and Feces[a]

	Time Interval, hr												
	Urine								Feces				
Subject[b]	0–4	4–8	8–12	12–24	24–48	48–72	72–96	96–120	0–24	24–48	48–72	72–96	96–120
M-1	2.6	1.8	0.75	0.87	1.4	5.1	0.62	0.25	41.4	31.1	NS[c]	6.0	0.05
M-2	5.4	2.5	1.7	1.8	6.7	2.6	3.2	2.1	NS	37.0	8.0	7.7	NS
M-3	3.8	2.1	0.87	1.9	2.7	2.0	0.87	0.15	11.4	62.1	NS	12.9	0.07
M-4	9.8	19.8	6.5	2.9	2.6	2.0	0.55	0.11	29.7	7.4	11.3	2.8	0.67
M-5	5.9	8.9	2.4	1.9	4.0	5.2	1.6	0.28	NS	1.0	57.3	10.6	0.89
$\bar{X}_M$	5.5	7.0	2.4	1.9	3.5	3.4	1.3	0.58	16.5	27.7	15.3	8.0	0.34
SEM	1.2	3.4	1.0	0.32	0.90	0.73	0.49	0.38	8.2	10.9	10.7	1.7	0.18
G-1	8.2	18.3	4.1	1.8	0.98	0.25	0.10	0.12	0.04	59.3	6.0	NS	1.0
G-2	2.4	2.3	0.85	0.75	0.56	0.20	0.12	0.05	4.0	64.4	1.5	0.33	0.71
G-3	6.4	2.2	0.66	0.96	1.0	1.4	1.9	1.2	0.04	39.6	28.7	0.25	8.5
G-4	1.6	0.96	0.38	0.50	0.58	0.15	0.00	0.00	15.9	72.3	2.5	0.11	0.14
G-5	5.5	13.4	2.9	0.46	1.2	0.39	0.14	0.07	0.09	29.1	44.0	5.9	0.35
$\bar{X}_G$	4.8	7.4	1.8	0.89	0.86	0.48	0.45	0.29	4.0	52.9	16.5	1.3	2.1
SEM	1.2	3.5	0.73	0.24	0.28	0.23	0.36	0.23	3.0	8.0	8.4	1.1	1.6
$\bar{X}_{M+G}$	5.2	7.2	2.1	1.4	2.2	1.9	0.91	0.43	10.3	40.3	15.9	4.7	1.2
SEM	0.83	2.3	0.62	0.25	0.61	0.60	0.33	0.22	4.6	7.6	6.4	1.5	0.82

[a] Expressed as percentage of administered dose. [b] Prefixes M and G refer to the position of the ^{14}C-label. See text. [c] No sample was supplied; subject was constipated. Result was entered as zero in average calculations.

^{14}C-activity ingested was 42.2–49.3 μCi, based on the determined specific activities. Each subject also received two more capsules containing II to make the total ingested amount of II equal to 1200 mg. Food, but not water, was withheld until noon, when the same lunch was served to all subjects.

Urine and feces were collected for 5 days. Urine was collected at the intervals shown in Table I. Feces were collected during each 24-hr period and were pooled for each subject before analysis when there were more than one. All urine and feces samples were frozen until analyzed. Blood was collected at 1, 2, 4, 8, 12, and 24 hr after drug administration in tubes containing edetic acid. All blood samples were refrigerated until analyzed.

Salicylate Analysis—The levels of salicylic acid, salicyluric acid, and total salicylate in urine were determined (at least in duplicate) using the method of Farid *et al.* (10). Blood salicylate was determined using the method of Trinder (11), with at least three determinations per sample. Blood and urine salicylate data from the 10 volunteers were pooled for the determination of the mean and standard error of the mean at each time interval.

Sample Preparation and Radioanalysis—The volume of urine of each subject for a given time interval was measured. Aliquots of 0.5 ml, or 1 ml when the activity was expected to be low, of urine in triplicate were placed in low potassium vials, and 0.1 ml of acetic acid was added. Fifteen milliliters of scintillator[3] was added, and the radioactivity was determined.

A preliminary study compared three agents for digesting feces: a quaternary ammonium hydroxide[4], perchloric acid–hydrogen peroxide (12), and a tissue digestant[5]. The use of perchloric acid–hydrogen peroxide led to a loss of about 15% of the activity in the case of I-G. The quaternary ammonium hydroxide caused a loss of about 12%. These losses were attributed to the long digestion times required for feces. The use of the tissue digestant did not cause a loss of activity and, moreover, gave the highest counting efficiency (77.9%).

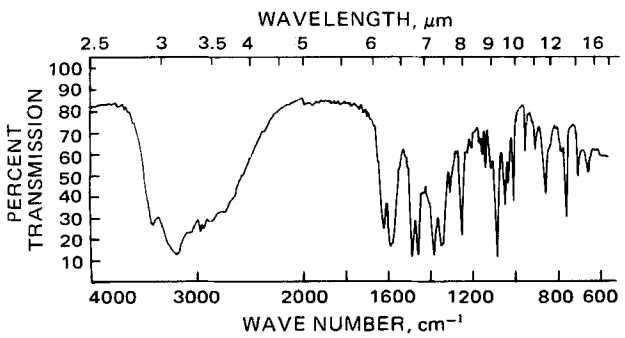


Figure 1—*IR spectrum of meglumine salicylate.*

The tissue digestant procedure was as follows. Feces were weighed and placed in a blender. Water, two to three times the sample weight, was added, and the mix was blended for 5–7 min. Quadruplicate samples of the resulting slurry, weighing 150–250 mg, were accurately weighed in tared counting vials. Two milliliters of tissue digestant and 0.3 ml of hydrogen peroxide were added, and the samples were left at room temperature. Six hours later, 0.3 ml of hydrogen peroxide was added, and the samples were left at room temperature for 24–36 hr. Then 0.1 ml of acetic acid was added followed by 15 ml of scintillator, and the radioactivity in the sample was determined.

Triplicate blood samples were prepared for counting by heating, in a counting vial, 0.1 ml of blood and 2 ml of tissue digestant at 50° for 4 hr. The vial was cooled to room temperature, and 0.3 ml of hydrogen peroxide was added dropwise. Four hours later, 0.3 ml of hydrogen peroxide was added. The samples were counted 3–4 hr later after the addition of 15 ml of scintillator.

All counting data were converted to disintegrations per minute with corrections for background and counter efficiency by internal standardization. Disintegrations per minute for the total volume or weight were calculated, and the result was reported as the percentage of the administered dose for each subject.

RESULTS AND DISCUSSION

Blood analysis showed no detectable activity above background. With the method used, the detection limit was 10 μg/ml of blood. Table I shows the concentration of ^{14}C-I and/or its radioactive metabolites in the urine and feces, expressed as the percentage of the administered dose. As can

Table II—Blood Salicylate Levels

	Salicylate Level, μg/ml	
Hours	Mean[a] ± *SEM*	Range
Control	16.6 ± 0.6	13.2[b]–19.5
1	44.4 ± 1.9	35.6–53.9
2	43.0 ± 1.6	37.5–53.2
4	35.6 ± 1.2	31.1–40.9
8	26.7 ± 1.4	20.0–32.2
12	20.6 ± 1.0	15.6–25.3
24	16.9 ± 0.7	13.2–20.3

[a] All values are the average of at least three determinations. [b] Ten subjects. See text for details.

[3] Phase combining system, Amersham-Searle, Arlington Heights, Ill.
[4] Hydroxide of Hyamine 10-X, a 1 *M* solution in methanol, Packard Instrument Co., Downers Grove, Ill.
[5] Soluene 100, Packard Instrument Co., Downers Grove, Ill.

Table III—Recovery of Salicylic Acid, Salicyluric Acid, and Total Salicylate in Urine

	Amount, mg						Cumulative Amount as Percentage of Administered Dose of Salicylic Acid				
	Control	0–4 hr	4–8 hr	8–12 hr	12–24 hr	24–48 hr	0–4 hr	0–8 hr	0–12 hr	0–24 hr	0–48 hr
Salicylic acid											
Mean ± *SEM*	0.12 ± 0.03	9.1 ± 3.1	9.8 ± 2.5	6.5 ± 1.2	2.3 ± 0.5	5.9 ± 1.7	1.8 ± 0.63	3.8 ± 0.88	5.1 ± 0.92	5.6 ± 0.94	6.8 ± 1.2
Range	0.00–0.30[a]	1.7–34.4	3.0–26.9	2.7–15.6	0.48–6.2	0.0–17.7	0.35–6.9	1.4–8.8	2.1–9.7	2.3–10.4	2.3–13.6
Salicyluric acid											
Mean ± *SEM*	1.8 ± 0.8	165.3 ± 11.1	177.9 ± 13.5	97.0 ± 6.8	40.5 ± 4.8	7.4 ± 1.4	23.6 ± 1.6	48.9 ± 3.0	62.7 ± 3.6	68.5 ± 3.5	69.5 ± 3.6
Range	0.23–8.4[b]	127.8–227.0	111.4–237.2	70.3–147.4	14.5–65.2	2.6–17.8	18.2–32.3	34.4–64.3	44.6–85.3	50.1–87.3	50.5–89.9
Total salicylate											
Mean ± *SEM*	2.0 ± 0.7	152.1 ± 8.2	167.5 ± 5.4	90.8 ± 4.9	47.2 ± 5.1	13.2 ± 1.3	30.6 ± 1.7	64.3 ± 1.9	82.6 ± 2.1	92.1 ± 1.6	94.7 ± 1.5
Range	0.27–6.7	119.4–209.2	143.6–190.2	66.0–114.8	13.3–79.8	4.9–19.8	24.0–42.1	55.5–71.6	73.8–93.4	84.3–101.5	87.5–102.5

[a] All values are the average of at least two determinations per sample. Values listed were not corrected for background salicylic acid levels because of the daily variability of the background levels. [b] For salicyluric acid, the amount in milligrams is listed as salicyluric acid; percentage of dose, however, is based on salicylic acid equivalent.

be seen, the excretion of I and/or its metabolites in the urine peaked 4–8 hr after drug administration. The average of the excreted I-M and/or its metabolites in urine over 5 days was 25.6 ± 5.7% of the administered dose. For the G group, the average was 17.0 ± 5.5% over the same period. This difference was attributed to biological variations between subjects. A t-test indicated that there was no significant difference in the excretion rate and the percentage excreted of I and/or its metabolites in urine over time between the M and G groups ($p > 0.1$).

About 68% of I-M and 77% of I-G and/or their metabolites were excreted in the feces (Table I) over 5 days. Again, a t-test indicated there was no significant difference between the excretion rate and the percentage of the administered activity excreted in the feces between the two groups ($p > 0.1$).

The 10 data points for each time interval were averaged (Table I), and the cumulative percentage of activity excreted in both the urine and feces was calculated. The resulting data showed that 21.3 ± 4.0% of the administered activity was excreted in the urine and 72.4 ± 4.1% was excreted in the feces over 5 days. Thus, 93.7 ± 2.8% of the administered dose was excreted in 5 days. Further examination of the data indicated that 50% of the administered dose was excreted during the first 35 hr.

After a single oral dose of 1.2 g of II, equivalent to 0.65 g of aspirin, blood salicylate levels (Table II) were 44.4 ± 1.9 and 43.0 ± 1.6 μg/ml in the 1st and 2nd hr, respectively. These values are comparable to the published blood salicylate levels after equivalent single doses of aspirin (13, 14), buffered aspirin (15), and sodium salicylate (16).

Table III shows the recovery of salicylic acid, salicyluric acid, and total salicylate in urine over 48 hr. Total salicylate concentration in urine samples collected beyond the 48-hr postdrug period were within the same concentration range as in the control samples and, therefore, were not included in Table III.

A semilogarithmic plot of the percentage of salicylate remaining in the body *versus* time showed that the half-life of the salicylate moiety in II was 6 hr. This value is in agreement with the previously reported half-life of a single 1-g dose of aspirin of 6 hr (17).

As with other salicylates (18), the major metabolite excreted in the urine was salicyluric acid, which accounted for 69.5 ± 3.6% of the administered dose. Salicylic acid excretion accounted for 6.8 ± 1.2% of the administered dose. Total salicylate excretion was 92.1 ± 1.6 and 94.7 ± 1.5% of the administered dose 24 and 48 hr after dosing, respectively.

These results show that the blood salicylate levels and the excretion of salicylic acid and its metabolites in urine followed the well-documented pattern of other salicylates. Thus, the salicylate moiety in II is absorbed and metabolized in humans like other salicylates.

The fact that, regardless of the position of the ^{14}C-label on I, the same percentage of the administered dose was excreted indicates that I is not metabolized through N-demethylation or through the formation of carbon dioxide. Whether I is excreted unchanged in the urine or metabolized needs further investigation.

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