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# THE FATE OF SODIUM GLUCURONATE AND GLUCURONOLACTONE IN MAN<sup>1</sup>

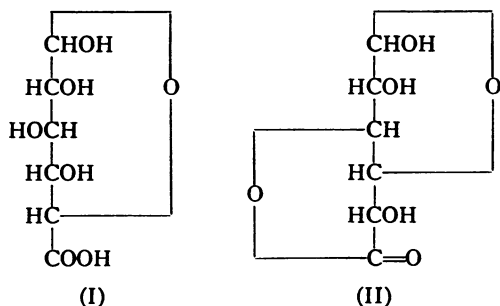
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Glucuronic acid occupies an important place in the body economy. A large number of toxic substances as well as many normal metabolites are excreted as conjugates with glucuronic acid (1). The glucuronic acid moiety occurs in the mucopolysaccharides including hyaluronic acid, chondroitinsulfuric acid, mucoitinsulfuric acid, heparin and several antigenic polysaccharides (2). The use of glucuronolactone has been suggested in the treatment of arthritis (3, 4) and in the management of recurrent nephrolithiasis (5).

Glucuronic acid occurs both as the free acid (I) and as a lactone (II). These two forms differ chemically in another way; the hemiacetal ring as



well as the lactone ring are five-membered in glucuronolactone, while the free acid has a six-membered pyranose structure (6, 7). It would not be unreasonable, therefore, to expect glucuronolactone and free glucuronic acid, or its salts, to be metabolized in different fashion. In the feeding experiments which have been reported (8-12), substantial amounts of reducing substances have been found in the urine, and naphthoresorcinol reactive substances have appeared in the blood and urine after the ingestion of glucuronolactone. Quick (13), however, was not able to demonstrate any urinary reducing substances after the ingestion of 3 g. of sodium glucuronate and only small

amounts of reducing substances were found after the ingestion of 10 g. or more. The present study was undertaken to determine the comparative fates of sodium glucuronate and glucuronolactone in man.

## METHODS

Commercial sodium *d*-glucuronate monohydrate and *d*-glucuronolactone<sup>3</sup> were given to apparently healthy young adults in two series of studies. Some glucuronolactone and sodium glucuronate was recrystallized, the lactone from absolute ethanol and the sodium salt from 40 per cent ethanol in water, for use as standards. For intravenous administration, recrystallized sodium glucuronate or glucuronolactone was given as a 2½ per cent solution in 2½ per cent dextrose in water over a 30-minute period.

In the first group of observations, 6 g. of either the sodium salt or lactone, divided into three equal doses, was ingested daily. The total 24-hour urinary glucuronic acid was determined prior to the period of administration and again during the fourth day of administration. In the second set of studies, urine and plasma glucuronic acid and urine pentose were determined prior to, and at intervals after the administration of a single dose of either the lactone or the sodium salt. In addition, the total amount of glucuronic acid excreted above the basal excretion level during the 12 hours after the administration of the test dose was calculated.

Total glucuronic acid was determined photometrically by means of the color reaction with carbazole (14). One ml. of diluted urine or a protein free filtrate of plasma containing less than 100  $\gamma$  glucuronic acid was added to 6 ml. concentrated sulfuric acid and heated in a boiling water bath for 20 minutes. The mixture was quickly cooled to room temperature and 0.2 ml. of a 0.1 per cent solution of carbazole in ethanol was added. After several minutes, the absorption was measured at 530 m $\mu$ . in a Beckman model DU spectrophotometer using a blank similarly prepared except that 0.2 ml. ethanol was substituted for the carbazole solution. In recovery experiments in which 50.0 mg. glucuronolactone was added to each of ten random urine specimens, the increase in glucuronic acid determined by this method corresponded to 93.6  $\pm$  3.1 per cent of the added glucuronolactone. In a similar experiment using sodium glucuronate, the recovery was 98.6  $\pm$  4.2 per cent. The color reaction with

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TABLE I  
Effect of oral sodium glucuronate and glucuronolactone on 24-hour urinary glucuronic acid excretion

Subject	Sodium glucuronate		Glucuronolactone	
	Baseline	6 g./day	Baseline	6 g./day
	mg.	mg.	mg.	mg.
B. S.	433	542	421	1,339
J. R.	444	656	462	1,573
G. W.	492	607	472	1,347
R. D.	460	638	487	1,290

naphthoresorcinol (15, 16) was used in preliminary studies, but it was found to be less specific and reproducible than the carbazole method of Dische.

Pentoses were determined by the procedure of Roe and Rice (17).

Glucuronolactone and the ester glucuronides react with hydroxylamine at pH = 4.5 to form hydroxamic acids, while glucuronic acid and its salts and the acetal ("ether") glucuronisides do not react with hydroxylamine. The hydroxamic acids are determined photometrically after the addition of ferric chloride (18). This method was used to assess the amount of glucuronic acid excreted in the lactone form.

#### RESULTS

The excretion of glucuronic acid in the urine before and after the daily ingestion of 6 g. of sodium glucuronate or glucuronolactone is given in Table I. The average increment in the total urinary glucuronic acid was 156 mg. per 24 hours on the fourth day of ingestion of sodium glucuronate, and 927 mg. per 24 hours after ingestion of the lactone. In the case of the lactone, the additional urinary glucuronic acid represents an average of 15.4 per cent of the amount ingested, while an average of only 2.6 per cent of the ingested sodium salt appears in the urine.

After the ingestion of a single dose of 15 g. glucuronolactone, a prompt rise in the plasma glucuronic acid and in the urinary glucuronic acid and pentose excretion was observed. Only slight increases were observed after similar oral doses of sodium glucuronate. Typical results obtained in one subject are illustrated in Figure 1. It was found that the bulk of the urinary glucuronic acid recovered after the ingestion of the lactone did not react with hydroxylamine, and appears, therefore, to be in the free acid form (Table II). The results of the intravenous administration of 5 g. of sodium glucuronate and of glucuronolactone in one subject are shown in Figure 2. In contrast to the

small amount of an oral dose which appeared in the urine, an intravenous dose of sodium glucuronate was almost quantitatively excreted. The urinary glucuronic acid recoveries above basal excretion during the 12 hours after a single dose of either sodium glucuronate or the lactone are given in Table III.

#### DISCUSSION

The observation of but slight increments in the plasma and urine glucuronic acid levels after ingestion of sodium glucuronate in contrast to the prompt and substantial increases found after the ingestion of glucuronolactone is consistent with the concept that the free acid form and salts of glucuronic acid are poorly absorbed from the gastrointestinal tract.

Almost all of the glucuronic acid which appears in the urine after ingestion of glucuronolactone is unreactive with hydroxylamine. The excreted glucuronic acid is presumed to be that portion of the ingested lactone which is hydrolyzed within the organism to the free acid (19). Parenterally

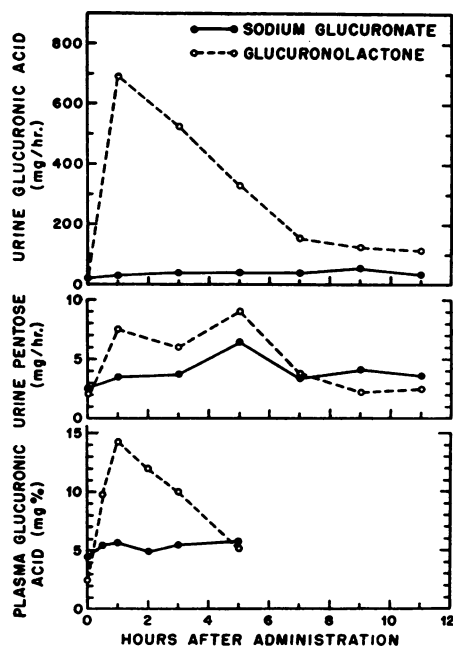


FIG. 1. A COMPARISON OF THE URINE GLUCURONIC ACID AND PENTOSE AND THE PLASMA GLUCURONIC ACID AFTER THE ORAL ADMINISTRATION OF 15.0 G. SODIUM *D*-GLUCURONATE AND AFTER A SIMILAR DOSE OF *D*-GLUCURONOLACTONE (L. W.)

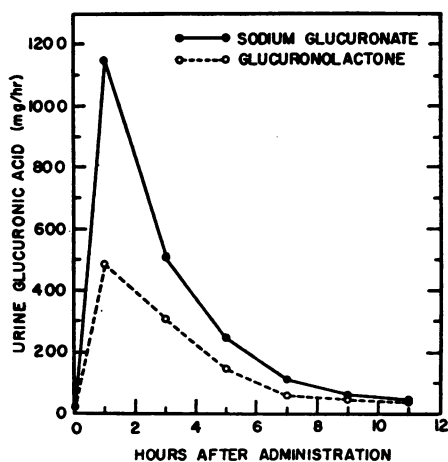


FIG. 2. A COMPARISON OF THE URINE GLUCURONIC ACID AFTER THE INTRAVENOUS ADMINISTRATION OF 5.0 G. SODIUM *d*-GLUCURONATE AND AFTER A SIMILAR DOSE OF *d*-GLUCURONOLACTONE (R. D.)

administered sodium glucuronate is promptly excreted in the urine.

The observations of this study are in agreement with the findings of recent tracer studies in guinea pigs and rats. Douglas and King (20, 21), and Packham and Butler (22, 23) found about one-half of parenterally administered glucuronolactone labeled with radiocarbon in the urine, and about one-third in the expired carbon dioxide of their animals. A parenteral dose of labeled sodium glucuronate was almost quantitatively excreted in the urine and no label was found in the expired carbon dioxide.

Enklewitz and Lasker (24) found in cases of essential pentosuria that ingestion of *d*-glucuronolactone resulted in an increase in the urine pentose. Touster, Hutcheson, and Reynolds (25) discovered that normal as well as pentosuric subjects excrete pentose after ingestion of glucuronolactone. In normal subjects, however, only a small amount

TABLE II  
Total glucuronic acid and hydroxylamine reactive substances excreted after ingestion of 15 g. glucuronolactone (L. W.)

	Total urinary glucuronic acid ( $\mu$ Eq./hr.)	Hydroxylamine reactive cmpds. ( $\mu$ Eq./hr.)
Control	119	7.3
1 hr.	3,543	90.5
3 hr.	2,102	23.8
5 hr.	553	17.8

of pentose appears in the urine. In both reports, the pentose was identified as *l*-xylulose. Indeed, in these studies a slight pentosuria was observed after ingestion of glucuronolactone.

It appears that *d*-glucuronolactone is absorbed from the gastro-intestinal tract and is metabolically active. Sodium *d*-glucuronate is poorly absorbed and does not enter into the metabolic reactions of the organism but is promptly excreted in the urine.

#### SUMMARY

An average of 23.4 per cent of an orally ingested dose of 15 g. *d*-glucuronolactone was excreted in the urine within 12 hours while an average of only 1.9 per cent of a similar dose of sodium *d*-glu-

TABLE III

Urinary glucuronic acid recovered above baseline during 12 hours after administration of sodium glucuronate or glucuronolactone

Subject	Amt. given	Route	After sodium glucuronate		After glucuronolactone	
			g.	%	g.	%
L. W., 24M	15.0	Oral	0.260	(1.7)	3.536	(23.6)
J. W., 33M	15.0	Oral	0.312	(2.1)	3.730	(24.9)
R. D., 29M	15.0	Oral	0.382	(2.5)	3.906	(26.0)
C. D., 24F	15.0	Oral	0.163	(1.1)	2.846	(19.0)
J. T., 24M	15.0	Oral	0.350	(2.3)	—	—
R. D., 29M	5.0	I.V.	4.014	(80.3)	2.102	(42.1)

curonate was excreted. The excreted glucuronic acid was largely in the free acid form. A prompt rise in the plasma glucuronic acid was observed after ingestion of the lactone but not after sodium glucuronate. After intravenous administration, 42 per cent of a 5 g. dose of the lactone and 80 per cent of a similar dose of sodium glucuronate was recovered from the urine. After feeding 6 g. glucuronolactone daily in divided doses for four days, the total urinary glucuronic acid increased an average of 927 mg. per 24 hours. In the case of sodium glucuronate, the increase was 156 mg.

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