GALACTITOL EXCRETION IN THE URINE OF A GALACTOKINASE-DEFICIENT MAN *

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Galactitol does not seem to be of great physiological importance in mammals (Touster and Shaw, 1962). This polyol has been shown to accumulate in the lens and in a number of tissues (van Heyningen, 1959; Quan-Ma and Wells, 1965) of galactose-intoxicated rats and was isolated from the brains of galactosemic patients (Wells et al., 1965). In both conditions, significant amounts of galactitol were excreted in the urine (Wells et al., 1964; Chin, 1964). This communication reports the occurrence in mammals of a previously unrecognized abnormal metabolic situation in which galactitol is produced and excreted in the urine.

The individual described in this report (Gitzelmann, 1965) has galactokinase-deficient erythrocytes and may have a generalized galactokinase deficiency. He excretes large amounts of

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galactose in urine, is suffering from neurofibromatosis and has been operated on for cataracts. Erythrocyte galactose-1-phosphate uridyl transferase activity is normal and uridine diphosphogalactose-4-epimerase activity was also demonstrable (Table 1)

Table 1

ENZYME ACTIVITIES IN ERYTHROCYTES AND WHOLE BLOOD HEMOLYSATES (GALACTOKINASE), AND IN LYSED ERYTHROCYTES (TRANSFERASE, EPIMERASE) OF THE PATIENT.

Galactokinase was assayed with galactose-1-^{14}C as the substrate using paper chromatography (Robinson, 1963) or collection of $^{14}CO_2$ (Weinberg, 1961). Transferase was assayed by a modification (Beutler et al., 1965) of the UDPG-consumption procedure (Anderson et al., 1957).
Epimerase in 0.1 ml of erythrocyte lysate was assayed with 1 µC UDP-Gal-^{14}C (uniformly labeled; New England Nuclear Corp.; 120 mC/mmole) in the presence of pyruvate (5 µmoles) and DPN (1.25 µmoles) in a final volume of 0.25 ml at p_H 8.7 (0.5 M glycine buffer). Before and after incubation at 37°C., aliquots were hydrolyzed in 1 N HCl for 15 min., the supernatants were chromatographed on paper which was scanned for radioactivity.

Galactokinase	Virtually absent
Transferase	23 µmoles UDPG consumed/hr./g Hbg.
Epimerase	Active (Percent UDP-glucose-^{14}C present at zero time was less than 1 %; after 30 min. incubation with the enzyme preparation approx. 75 % of the label was in UDP-glucose).

Since a number of common features exist in human galactosemia, in galactose-intoxication of the rat, and in this patient (e.g. unusual high blood-galactose levels, galactosuria, cataract formation) it was thought that galactitol might be found in the urine of this patient.

Urine samples were kept frozen until chromatographed on paper (n-butanol 10, pyridine 4, H_2O 3, vol./vol.); the galac-

tose glucose regions which also contain the galactitol were eluted with H_2O in the cold, and the eluates dried in vacuo. Gas chromatography of the trimethylsilylation products (Bentley et al., 1963) was performed using two different systems (Fig. 1).

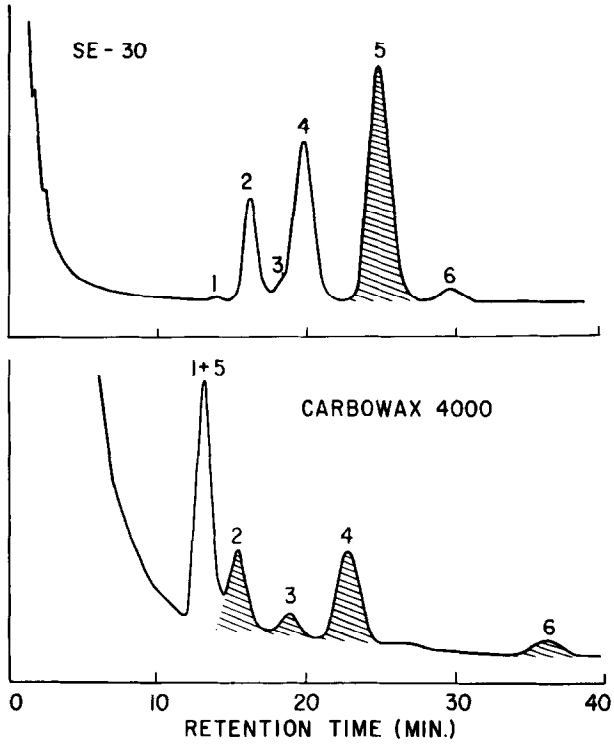

Figure 1

TRACINGS OF GAS CHROMATOGRAMS OF A URINE ELUATE

Chromatography conditions: SE-30, 3% on Gas-Chrom P; 2 m glass tube, i.d. 2.7 mm; T_c 170°, T_j 220°; N_2, 40 ml/min. Carbowax 4000, 40% on Gas-Chrom P; 2 m glass tube, i.d. 2.7 mm; T_c 160°, T_j 210°; N_2, 30 ml/min.

1 = β-Galactose, 2 = α-Galactose, 3 = α-Glucose
4 = β-Galactose, 5 = Galactitol, 6 = β-Glucose

Considerable concentrations of galactitol were found (Table 2). In one night, the patient excreted 2.65 g of galactitol together with 7.26 g of galactose and 0.45 g of glucose (Table 2, sample 2).

Table 2

CONCENTRATIONS (g/100 ml) OF GALACTOSE, GLUCOSE AND GALACTITOL IN THE URINE FROM THE PATIENT.

Sample 1 was collected one hour after a breakfast containing milk. Sample 2 is a one night's portion. Sample 3 was collected during the last hour preceding a milk breakfast, and samples 4 trough 8 were obtained thereafter at ½ hour intervals. Analysis by gas-liquid chromatography.

Urine sample	Galactose	Glucose	Galactitol
1	0.401	0.284	0.030
2	1.211	0.075	0.446
3	0.449	0.044	0.774
4	0.579	0.401	0.596
5	0.798	0.321	0.385
6	0.768	0.287	0.206
7	0.924	0.285	0.240
8	1.403	0.370	0.591

The site of galactitol formation in this patient is not yet known. Hers (1960) was unable to find aldose reductase in rat liver, kidney or brain, and Quan-Ma and Wells (1965) found only traces of galactitol in kidney and brain and none in the liver of galactose-fed rats. However, ocular lenses are known to contain an aldose reductase (Hayman and Kinoshita, 1965). Since the lenses of this patient were removed a number of years ago because of cataracts, this tissue could not be a source of the galactitol. The galactitol may be related to the formation of these cataracts, and polyol accumulation may be the common mechanism in the pathogenesis of the various forms of sugar cataracts (Kinoshita, 1963).

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