Biochimica et Biophysica Acta, 455 (1976) 90–101

BBA 77487

METABOLISM AND TRANSPORT OF GALACTOSE BY RAT INTESTINE

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(Received April 6th, 1976)

SUMMARY

Intestinal uptake and metabolism of galactose were examined in everted jejunal rings from fasted adult rats using 0.2–28 mM sugar. After 60-min incubations, the total uptake (free tissue plus amount metabolized) of galactose ranged from 1.75 μmol/g at 0.2 mM to 21 μmol/g at 28 mM. Free tissue galactose was 17 % of the former and 73 % of the latter amount while that oxidized to $^{14}CO_2$ represented only 6–16 % of amount taken up. Compared to glucose, similar amounts of galactose are taken up at 0.2–2.0 mM, however, glucose removal from the media is about 60 % greater than galactose from 4 to 28 mM. Between 0.2 and 2 mM similar amounts of both sugars are metabolized, although a greater portion of the glucose is oxidized to $^{14}CO_2$. Above 2.0 mM, 2–3 times more glucose is metabolized than galactose. Both uptake and metabolism showed saturability and kinetic analysis revealed two limbed Lineweaver-Burk plots, suggesting operation of a high affinity low K_m and a low affinity high K_m system for sugar transport. In a series of in vivo studies, to assess the role of the intestine in the total body metabolism of galactose, ^{14}C-labeled galactose injected intraperitoneally at a dose of either 50 or 300 mg into fasted normal, sham operated and enterectomized rats, no observable difference in $^{14}CO_2$ production resulted in between the groups. It would thus appear that although extensive metabolism of galactose may take place in intestinal tissue in vitro, the intestine does not play a significant role in galactose disposition in vivo.

INTRODUCTION

The presence of the sugar nucleotide (Leloir) pathway enzymes in rat intestine has been known for some time. These enzymes, galactokinase, galactose-1-*P* uridyl transferase and uridine diphosphogalactose epimerase are responsible for the metabolism of galactose and its conversion to glucose, glycogen, or structural polysaccharides. The developmental pattern of these enzymes in rat intestine has been delineated and their kinetic characteristics determined in our laboratory [1–3].

Despite the presence of galactose-metabolizing enzymes in rat intestinal mucosa, studies in vitro of the utilization of very high concentrations of galactose by

intestinal sacs and measurement of maintenance of electrical potential gradients have failed to provide evidence of significant metabolism of this sugar [4, 5]. On the basis of these findings, galactose has been accepted as a poorly metabolized model sugar for intestinal monosaccharide transport experiments [6–8]. To explore whether galactose, indeed, is a poorly metabolized monosaccharide, we have examined the uptake and metabolic disposition of galactose at a wide range of substrate concentrations in vitro.

We have also assessed the role of the intestine in the overall handling of galactose by measuring galactose oxidation in enterectomized animals. Our results form the basis of this report.

MATERIALS AND METHODS

Animals. 45-day-old male rats of the Sprague-Dawley strain were obtained from Huntington Farms, Conshohocken, Pa. and were deprived of food but allowed free access to water for 18 h prior to use.

Chemicals. [1-^{14}C]Galactose (specific activity 2.7 Ci/M) was purchased from International Chemical and Nuclear Corp., Irvine, Calif. [1-^{14}C]Glucose (specific activity 7.08 Ci/M) was a product of New England Nuclear Corp., Boston, Mass. Galactose and glucose were obtained from Sigma, St. Louis, Mo. Each of these sugars was essentially free of the other sugar. Galactose oxidase was obtained as Galactostat Kit from Worthington Co., Freehold, N. J.

Experimental procedure

Sugar accumulation and phosphorylation by gut segments. The animals were sacrificed by decapitation and the mid-jejunum was quickly removed, everted over a glass rod, cut into small rings each weighing about 20 mg, and pooled in iced saline. Three rings were placed in 30-ml plastic bottles containing the incubation medium consisting of 2 ml of Krebs-Ringer bicarbonate buffer, pH 7.4, containing varying concentrations (0.2, 0.5, 1.0, 1.5, 2, 4, 10, 20, 28 mM) of substrate (galactose or glucose) and 0.2 μCi of the same labeled substrate. The bottles were gassed for 30 s with O_2 CO_2 (95 : 5, v/v) and incubated in a Dubnoff metabolic shaker at 37 °C. After 60 min incubation the gut segments were blotted, weighed, and placed in 2 ml of boiling water for 6 min to extract the neutral sugars and phosphorylated metabolites. A $Ba(OH)_2$ and $ZnSO_4$ precipitation was performed on 1 ml of the water extract to remove the protein and phosphorylated compounds [9, 10]. A 0.2 ml aliquot of the filtrate and water extract was each assayed for radioactivity by previously described techniques for scintillation counting [11]. The difference in radioactivity in the two fractions was used to determine the amount of free sugar and phosphorylated intermediates. In several experiments the free galactose in the tissue extract was assayed by gas-liquid chromatographic analysis of the trimethylsilyl derivative [12] and the values corresponded to those calculated from the radioisotope method. In each experiment, triplicate determinations were made for each datum.

Galactose oxidation to $^{14}CO_2$. Intestinal rings, incubation medium and incubation conditions were identical with those used above. The collection and measurement of evolved $^{14}CO_2$ were performed by the method of Weinstein et al. [11]. After incubation for 15, 30, 60, and 90 min, 1 ml of hyamine was injected into the center well and 0.3 ml of 3 M H_2SO_4 into the medium. $^{14}CO_2$ was absorbed by the hyamine

for a period of 45 min shaking at room temperature. The hyamine was added to 14 ml scintillation fluid (liquifluor), New England Nuclear Corp. and counted in a Packard liquid scintillation counter.

Total sugar uptake. Intestinal rings, incubation conditions and incubation medium without the labeled sugar were identical with those used in tissue accumulation studies. After 15 or 60 min incubation, the vials were opened and an aliquot of the media was taken and assayed using galactose oxidase (Galactostate Kit). The difference between the amount of sugar present at the start of the incubation and the amount recovered after each time period of incubation represents the amount which disappeared or was taken up. When glucose uptake was studied, glucose oxidase was used for assay.

Kinetic analysis. Double reciprocal plots were made according to Lineweaver-Burk for those processes which showed saturability. Since the metabolic events evaluated reflect the net flux through a multisystem pathway, the kinetic parameters described are resultant constants for the entire pathway. Calculations were made using steady-state data based on the mathematical model of Tenenhouse and Quastel [13] and as described by Scriver and Mohyuddin [14]. The velocity parameter (v) used in plotting the data is not a true velocity determination, but reflects steady-state accumulation. Therefore, calculation of maximum velocity (V) cannot be made. The determination of the apparent transport K_m, however, using these data closely approximates that derived using initial velocities of uptake.

$^{14}CO_2$ collection in vivo. Three groups of animals were employed for comparisons: (1) a normal control; (2) a sham operated with ether anesthesia; and (3) an enterectomized in which the gut was removed from duodenum to rectum under ether anesthesia. Subcutaneous injections of the substrate were given 30 min after the surgery when the animals were fully active. Two different amounts of galactose were administered. In one set of experiments 50 mg and in another 300 mg were given in 0.2 ml containing 0.5 μCi of [1-^{14}C]galactose. The method for collection of $^{14}CO_2$ was essentially that of Frederickson and Ono [15]. Rats were placed in airtight glass restraining cages. Room air, freed of CO_2 by prior passage through Dryrite containing towers, was drawn through the metabolic cages. The effluent air was passed through hyamine columns for trapping $^{14}CO_2$ for a duration of 10 min at 25-min intervals. 2-ml aliquots were placed in 14 ml of scintillator liquifluor and counted by liquid scintillation techniques [11]. The total duration of the study was 5 h for each animal. Each group consisted of four animals.

RESULTS

Tissue accumulation of galactose

The incubation of jejunal tissue with 10 and 0.25 mM galactose was carried out to determine the time-dependent accumulation of sugar at both high and low substrate concentrations. Results of this study are shown in Fig. 1. At both substrate concentrations free sugar accumulated rapidly in the tissue reaching a steady state after 15 min. Phosphorylated metabolites of galactose were formed with a similar time dependence as free sugar. At steady state the concentration of phosphorylated derivatives was about 10 % that of free sugar for high substrate concentration (Fig. 1A, 1 μmol/g

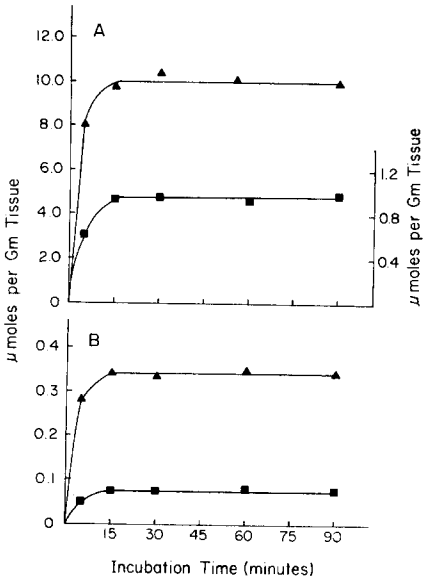

Fig. 1. Metabolism of galactose by intestinal rings as a function of time. Three 20-mg everted rings were pooled and incubated in 2.0 ml of Krebs-Ringer phosphate Buffer, pH 7.4, for times specified with either (A) 10 mM galactose or (B) 0.25 mM galactose. Free water extractable hexose (▲) was assayed enzymatically and phosphorylated compounds (■) were determined by $Ba(OH)_2$ – $ZnSO_4$ precipitation methods. Values were means of nine determinations. Right hand scale applies to lower line.

vs. 10 μmol/g). At the low substrate level phosphorylated derivatives were about 20 % of free sugar levels (Fig. 1B, 0.07 μmol/g vs. 0.33 μmol/g).

The data for tissue galactose levels shown in Fig. 1 may be analyzed to determine the galactose per ml of intracellular fluid and the concentration gradient, the ratio of galactose concentration in intracellular fluid to that in the medium [18]. This calculation has been performed in assessing substrate transport and depends on the fluid spaces of which the extracellular space has previously been determined to be 12 % of wet tissue weight using inulin and total tissue water to be 80 % using the difference between wet and dry tissue weight [17]. At 10 μmol/ml medium concentration, the steady-state intracellular free galactose is 12.94 μmol/ml giving a ratio of 1.29. At 0.25 μmol/ml of medium the intracellular fluid level is 0.46 μmol/ml with a ratio of 1.82. The higher concentration in intracellular fluid than in the medium establishes that galactose participated in an active transport process.

Disposition of cellular galactose

The total uptake of galactose from the medium, accumulation of the sugar in the tissue and its metabolic fate after 60 min incubation is shown in Table I and graphically in Fig. 2. The total uptake of galactose increases with concentration from 1.75 μmol/g at 0.2 mM to 7.80 μmol/g at 2.0 mM. A similar 10-fold increase in concentration from 2.0 to 20 mM raises the uptake to 21 μmol/g. The kinetics of the uptake process will be examined in more detail below. Free galactose in the tissue (column 4, Table I) increases as the substrate concentration increases from 0.29 μmol/

g at 0.2 mM to 19.19 μmol/g at 28 mM. The former value represents 17 % of the galactose which disappeared from the medium while the latter represents 73 % of the galactose taken up by the tissue. At low galactose concentrations between 0.2 mM the free galactose in the tissue does not exceed 29 % of the uptake.

The difference between total galactose uptake and free tissue galactose reveals the amount of sugar metabolized (column 6, Table I). The maximum amount metabolized appears to be about 8 μmol/g at 10 mM galactose. Below 2 mM greater than 70 % of that taken up is metabolized. This figure falls progressively such that at 28 mM only 26 % of galactose taken up by the tissue is metabolized.

Galactose may have several metabolic fates, two of which have been directly assessed, conversion to CO_2 and formation of phosphorylated intermediates. The latter include galactose 1-phosphate and nucleotide sugars. Only relatively small amounts of the sugar metabolized are oxidized to CO_2. Between 0.2 and 10 mM, only 6 % follow this route. This increases to the highest value observed, 16 % conversion to CO_2 at 28 mM. The amount of phosphorylated derivatives in the tissue is minute (4 % of total metabolized) and essentially constant between 0.2 and 2 mM. At higher sugar levels, the amount phosphorylated increases to 29 % at 28 mM. Up to 2 mM substrate concentration, the μmol/g of phosphorylated compounds do not exceed the amount oxidized to CO_2 whereas at higher substrate levels the amount of phosphorylated material in the tissue exceeds the amount oxidized.

When the latter two fates are summed, a major portion of the metabolized galactose is unaccounted for (column 12, Table I). Between 0.2 and 2.0 mM galactose, 90–94 % of the metabolized sugar has gone to fates other than CO_2 or phosphorylated compounds. This percent decreases progressively at higher substrate concentrations to 54 % at 28 mM. The nature of the unknown metabolites has not been determined. Likely candidates would be lactate, free glucose or glycoproteins.

TABLE I

GALACTOSE AND GLUCOSE UPTAKE AND METABOLISM BY RAT SMALL INTESTINAL MUCOSA

Values are means ±S.E. of nine determinations (three separate experiments) after 60 min incubation. Hexose "totally metabolized" was derived by subtracting the amount free in the tissue from the amount totally taken up. "Other metabolic fates" was calculated by subtracting the sum of the amount oxidized plus phosphorylated intermediates from "totally metabolized". The results are expressed in μmol/g tissue.

Substrate concentration (mM)	Uptake		Free in tissue		Totally metabolized	
	Gal	Glc	Gal	Glc	Gal	Glc
0.2	1.75±0.03	1.4 ±0.15	0.29±0.008	0.064±0.005	1.46	1.34
0.5	3.75±0.64	3.0 ±0.13	0.71±0.02	0.21 ±0.06	3.04	2.79
1.0	5.90±0.26	5.75±0.10	1.36±0.05	0.55 ±0.04	4.54	5.20
1.5	7.00±0.33	7.65±0.94	1.61±0.12	1.06 ±0.12	5.39	6.59
2.0	7.80±0.67	8.6 ±0.87	2.26±0.15	2.34 ±0.2	5.54	6.26
4.0	8.60±1.2	13.0 ±0.75	3.81±0.2	4.79 ±0.15	4.79	8.21
10.0	16.00±4.43	25.5 ±3.4	7.77±0.4	9.52 ±0.5	8.23	15.98
20.0	21.00±4.6	40.5 ±1.8	14.00±1.9	16.38 ±2.4	7.00	24.12
28.0	26.00±4.2	41.5 ±5.36	19.19±2.8	18.34 ±2.1	6.81	23.16

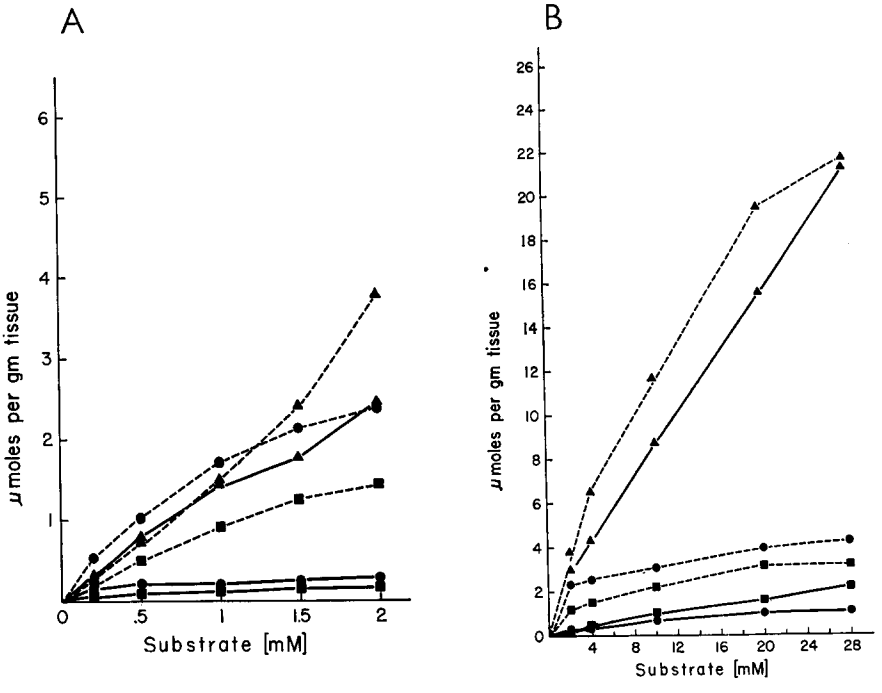

Fig. 2. Total metabolism of sugar by intestinal rings as a function of substrate concentration. Incubations were performed for 60 min as in Fig. 1 with either galactose (———) or glucose (– – – –) at concentrations indicated. Free hexose (▲), phosphorylated intermediates (■) were measured as in Materials and Methods. In separate experiments 0.2 μCi of the hexose used was added to the incubation flask and $^{14}CO_2$ was liberated with H_2SO_4 and trapped in hyamine (●). All points are means obtained from six determinations. A, concentration from 0.2 to 2.0 mM and B, from 2.0 to 28 mM.

Amount oxidized		Phosphorylated intermediates		Other metabolic fates	
Gal	Glc	Gal	Glc	Gal	Glc
0.09±0.001	0.55±0.02	0.06±0.004	0.22±0.02	1.31	0.57
0.14±0.005	0.98±0.05	0.10±0.02	0.53±0.02	2.8	1.28
0.16±0.004	1.60±0.08	0.12±0.07	0.98±0.03	4.26	2.62
0.19±0.008	2.17±0.10	0.14±0.08	1.35±0.1	5.06	3.07
0.20±0.02	2.22±0.08	0.14±0.10	1.41±0.1	5.20	2.63
0.27±0.03	2.46±0.11	0.41±0.2	1.61±0.15	4.11	4.14
0.56±0.03	2.93±0.12	0.98±0.2	2.14±0.15	6.69	10.91
0.95±0.08	3.88±0.10	1.51±0.2	3.01±0.25	4.54	17.23
1.10±0.07	4.18±0.12	2.00±0.3	3.19±0.3	3.71	15.79

Comparison of galactose and glucose metabolism

Galactose uptake and disposition by intestinal mucosa is compared with that of glucose in Table I and shown graphically in Fig. 2. The amount of sugar disappearing from the medium between 0.2 and 2 mM is about the same for glucose and galactose. Above 2 mM glucose removal is about 60 % greater than that of its epimer. Glucose accumulation in the tissue is less than that of galactose when the concentration is below 2 mM. At 2 mM and higher the amount of both sugars in the tissue is comparable (Table I, columns 4 and 5). The free glucose in the tissue, however, represents a much smaller fraction of the sugar taken out of the incubation medium (at 28 mM 44 % for glucose, 73 % for galactose).

Between 0.2 mM and 2 mM total metabolism of both sugars is similar, above 2.0 mM, 2–3 times more glucose is metabolized than galactose. Although similar amounts of both sugars are metabolized at low substrate concentrations, about 10

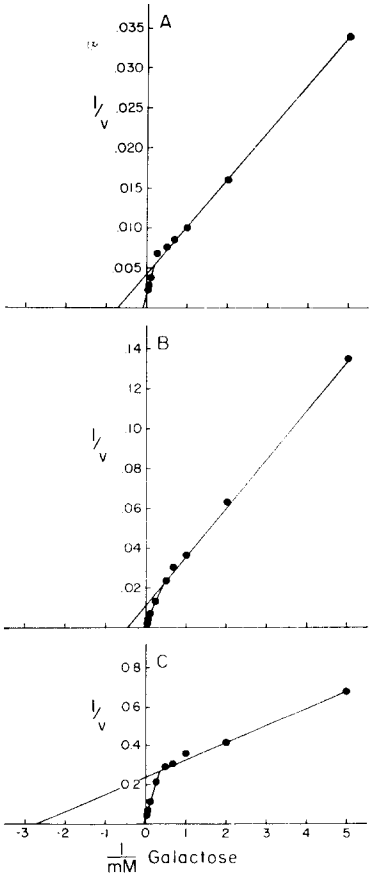

Fig. 3. Double reciprocal, Lineweaver-Burk, plots for tissue metabolism of galactose by intestinal rings. A, disappearance of galactose from medium; B, galactose and metabolites in tissue plus amount oxidized and C, oxidation to $^{14}CO_2$. Linear regression lines plotted as based on Monroe 1775 computer fit of the data. Calculated K_m data are listed in Table I. Each datum is the mean value of nine determinations. V represents an equilibrium concentration calculated from uptake after 60 min of incubation.

TABLE II

APPARENT K_m OF GALACTOSE UPTAKE AND METABOLISM

Values are derived from Lineweaver-Burk plots of data and computer fit linear regression lines.

Data analyzed		K_m (mM)
A Galactose disappearance from medium	Low	1.4
	High	14.3
B Galactose and metabolites plus amount oxidized	Low	1.8
	High	40
C Oxidized to $^{14}CO_2$	Low	0.40
	High	40

times as much glucose is oxidized to $^{14}CO_2$ (Table I, columns 8 and 9). This is reduced to a 4-fold difference at higher concentrations. Greater amounts of glucose also appear as phosphorylated derivatives. Between 0.2 and 2 mM substrate twice as much galactose as glucose is metabolized to fates other than CO_2 or phosphorylated compounds. Above 4 mM this is reversed and much greater amounts of metabolized glucose appears in this fraction. In terms, however, of the percent of total metabolism, these "other" fates are comparable for both sugars at the high substrate concentrations, at 28 mM 54 and 68 % for galactose and glucose, respectively (Table I, columns 12 and 13).

Kinetic parameters of galactose uptake

The disappearance of galactose from the incubation medium after both a 15 and 60 min incubation was analyzed by the double reciprocal Lineweaver-Burk plot of the data. The results of the 60 min incubation are shown in Fig. 3A. A two-limbed curve is obtained which indicates the uptake process is saturable and suggests that there may be two mechanisms involved. Calculation of the apparent transport K_m values for the two mechanisms are shown in Table II. There appears to be a high affinity low K_m system (K_m 1.4 mM) and a low affinity high K_m system (K_m 14.3 mM). Similar curves and K_m values were obtained when the incubation was shortened from the 60 min steady-state period to 15 min incubation. Fig. 3B is a double reciprocal plot of the data from Table I, where the amounts of free tissue galactose, tissue phosphorylated compounds, and quantity oxidized to CO_2 have been summed to represent the velocity of uptake. Again, a two-limbed concentration dependence curve is obtained with both an apparent low and high K_m system (Table II). The correspondence of K_m values obtained from the uptake data with that from the sum of tissue galactose, phosphorylated material and CO_2 is reasonable. Fig. 3C is a Lineweaver-Burk plot of the data of Table I showing the concentration dependence of the $^{14}CO_2$ production. This also shows the two-limbed curve seen with the uptake data.

In vivo oxidation of galactose to $^{14}CO_2$

The $^{14}CO_2$ excretion in expired air after injecting radioactive galactose subcutaneously to normal, sham-operated and enterectomized rats is shown in Fig. 4. There is no appreciable difference in the curves between the three groups either when 50 mg (0.35 g/k) or 300 mg (2.1 g/k) was administered. Integration of the area under

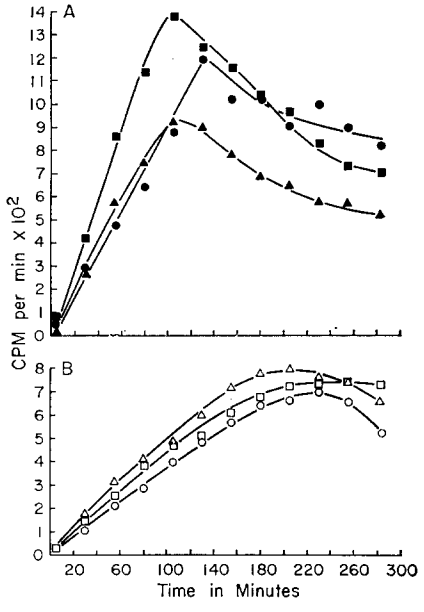

Fig. 4. In vivo $^{14}CO_2$ production by adult rats after intraperitoneal injection of 0.5 μCi of [1-^{14}C]-galactose with either (A) 50 mg galactose or (B) 300 mg galactose per animal. ■, normal control; ▲, ether anesthetized sham operated; ●, enterectomized. All animals were fully awake and active at the time of study. Values are the means of four separate experiments.

TABLE III

[1-^{14}C]GALACTOSE OXIDATION BY INTACT RAT

Values are the means of four separate experiments. Animals weighing 150 g were injected subcutaneously with 50 or 300 mg of galactose plus 0.5 μCi of tracer. $^{14}CO_2$ was measured as described in the text. The cumulative $^{14}CO_2$ was calculated by integrating the area under the $^{14}CO_2$ expiration curve. Values are expressed as percent: $^{14}CO_2$ collected versus [1-14 C] galactose injected.

Group of animals	Percent of dose expired as $^{14}CO_2$	
	50 mg dose	300 mg dose
Control	30.6	17.1
Sham	20.9	18.6
Enterectomized	27.5	15.2

the curves gives the total radioactivity excreted which when divided by the dose gives the fraction of the injected radioactivity expired as $^{14}CO_2$. These results expressed as percent of the dose are shown in Table III and indicate that the absence of the intestine does not impair the animal's ability to dispose of galactose via oxidation to $^{14}CO_2$.

DISCUSSION

Numerous studies of intestinal transport of galactose have been performed in vitro since Fisher and Parsons [18] in 1953 first examined the question in surviving rat intestine [6, 19, 8, 17]. In his paper on energetics of intestinal transfer in 1971, Smyth

[5] stated "galactose has been used as representative of a carbohydrate not significantly metabolized, but actively transported by rat intestine" [14]. This statement was based on the work of Barry et al. [4], who reported that everted sacks of rat jejunum incubated 1 h with 28 mM galactose or glucose metabolized 12 μmol/g wet weight compared to 88 μmol/g of glucose. These values were obtained by measurement of the difference in total reducing substances in the system at the beginning and end of 1 h incubation period. The "non-metabolism" of galactose by intestine has been assumed despite reports by Landau and Wilson [20], Saunders and Isselbacher [21] and Diedrich and Anderson [22] that galactose 1-phosphate is formed when intestine is incubated with galactose.

In the present studies with everted intestinal rings, we have employed specific enzymatic methods for galactose assay and measured galactose metabolites over a wide range of galactose concentration. Indeed, if the data for 28 mM galactose are examined (Table I), the conclusion would be that galactose metabolism is 34 % that of glucose metabolism (6.81 μmol galactose/g of tissue metabolized compared to 23.16 μmol of glucose/g). When the entire range of galactose is examined, however, especially between 0.2 and 2.0 mM the amounts of galactose metabolized are very similar to that of glucose. Therefore, in the range of galactose levels that might be observed under physiological circumstances, galactose cannot be considered a poorly metabolized sugar by rat intestine. Although Fisher and Parsons [18], Barry et al. [4] and Smyth [5] used 28 mM galactose in their transport studies with rat tissue, most other investigators using mainly hamster intestine have employed concentrations of about 5 mM [20, 23]. Landau and Wilson [20] have reported that hamster intestinal sacs incubated with 5 mM galactose transported 6 μmol/g in 1 h and metabolized 2 μmol/g. Glucose utilization was about eight times that of galactose. At 4 mM galactose, our ring preparation of rat transported 8.6 μmol/g in 1 h and metabolized 4.8 μmol/g. Our higher values may be due to a species difference or to the nature of the tissue preparation.

Using a 10–56 mM range and 30-min incubations of everted rat intestine, Fisher and Parsons [18] reported a transport K_m of 35 mM. With our everted rings of jejunum and a broad substrate range, we have observed two transport K_m values of 1.4 and 14 mM. Barry and Eggenton [6], using rat jejunal mucosa sacs and measuring electrical potentials from galactose concentrations above 2 mM report a single K_m of 14 mM. Most other galactose transport studies have been performed with preparations of hamster intestine [23–25]. When high levels of galactose were employed only one K_m was observed ranging from 5 to 20 mM. Honegger and Semenza [26] analyzed galactose transport across suspended hamster intestine after 2 min incubation and observed a transport K_m of 3.5 in the range of 0.1–3.0 mM and a second K_m of 12.3 mM at concentrations between 10 and 100 mM. Our data show clearly this same duality of transport systems in the rat. The nature of these systems has not yet been explored in this species. Honegger and Semenza [26] have shown the low K_m system has broad specificity for galactose, glucose, 6-deoxyglucose and 3-methylglucose, whereas the high K_m system is specific for glucose and galactose. Both systems were sodium dependent.

The existence of high and low K_m transport systems have been reported for amino acids in bacterial cells [27] and for galactose in brain synaptosomes [28] and renal tubule cells [12]. Indeed, McNamara and Segal [12] have reported two

transport systems for galactose in adult rat renal cortex slices while emphasizing the difficulty of studying transport of a rapidly metabolized sugar. Warfield and Segal [28] found high and low K_m galactose uptake processes in rat brain synaptosomes where considerable sugar phosphorylation occurred even with 3-min incubations. In the present studies the dual concentration dependence for uptake shown in Fig. 3A after 60-min incubations was also seen if the incubation time were reduced to 15 min. Honegger and Semenza [26] used 2-min incubations in their studies with hamster intestine. The two systems were also evident in our experiments if we measured primarily the galactose in the tissue (Fig. 3B), the $^{14}CO_2$ released (Fig. 3C) or examined the tissue phosphorylated fraction of Table I. Whether these data merely reflect the intrinsic membrane uptake process or metabolic influences on transport cannot be stated with certainty. Perhaps, some light is shed on this phenomenon by the finding that the binding characteristics for galactose by isolated and purified galactose binding protein from bacterial cell membranes are different in the high and low concentration ranges [29]. This would suggest that the two K_m transport systems observed in mammalian tissue may be due to substrate-induced conformational changes of the membrane proteins associated with galactose transport.

Our present results showing considerable metabolism of galactose by rat intestine conform to the past observations that the enzymes for galactose metabolism are present in this tissue. The fact, however, that enterectomized animals show no impairment of galactose oxidation would imply that intestinal galactose metabolism does not normally play a major role in the physiological disposition of galactose. One may speculate, however, that in conditions of impaired metabolism of galactose by liver, intestine metabolism may play a role in handling the sugar.

ACKNOWLEDGEMENT

This work was supported by grants 10894 and 1 Pol HD 08536 from the National Institutes of Health, Bethesda, Md.

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