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Metabolism of galactose in the brain and liver of rats and its conversion into glutamate and other amino acids

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Abstract Time- and dose-dependent measurements of metabolites of galactose (with glucose as control) in various organs of rats are discussed. Not only the liver but especially the brain and to a lesser extent the muscles also have the capacity to take up and metabolize galactose. Primarily, the concentrations of UDP-galactose, a pivotal compound in the metabolism of galactose, and UDP-glucose are measured. An important feature lies in the demonstration that galactose and glucose are metabolized to amino acids and that the only increases observed in the brain appear in the concentrations of glutamate, glutamine, GABA measured after acute galactose loads. In addition the increase in the amino acid concentrations after galactose has been administered persists for longer periods of time than after glucose administration. This conversion of hexoses, especially galactose, to amino acids requires the consumption of ammonia equivalents in the brain; this finding might stimulate the use of galactose as a new means of removal of this neurotoxic compound from the brain in patients suffering from hepatic encephalopathy or Alzheimer's disease.

Keywords Galactose · UDP-galactose · UDP-glucose · Glutamic acid · Brain · Liver

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Introduction

Free galactose does not occur in appreciable amounts in eukaryotic systems. However, in glycosidic linkage galactose is an essential component of many complex oligosaccharides in which it plays a crucial role in many biological processes. In particular, it is involved in cell–cell adhesion and recognition (Varki 1983). It is a central component of the Lewis-structures of many cell adhesion glycoproteins, e.g., the selectins, which mediate leukocyte-endothelial cell interactions, or CEACAMs, which are involved in homophilic cell–cell interactions (Varki 1983; Geyer et al. 2000; Lucka et al. 2005). Terminal galactose in glycoproteins acts as a recognition signal for the asialoglycoprotein receptor which initiates the uptake and degradation of desialylated serum glycoproteins and of aged erythrocytes (Ashwell and Harford 1982). Moreover, galactose is specifically characteristic of the blood group substance B (Hakomori and Kobata 1974).

Two sources of galactose are available to the organism. First, during the suckling period it is obtained by means of the cleavage of lactose by the infant's intestinal lactase. After cleavage, galactose enters the blood stream by specific transport systems and can be taken up by every organ in an insulin-dependent (with the exception of the liver) or

insulin-independent manner by use of the hexose transporter systems (Mueckler 1994; Olson and Pessin 1996). In the adult organism the newborn lactase is replaced by the expression of the adult lactase. However, one-fourth of the population worldwide is deficient in this adult lactase, leading to the harmful lactose intolerance (Weijers et al. 1961). The first step in the following Leloir-pathway (Leloir 1951; Kalckar and de Robichon-Szulmajster 1959) is the conversion of ß-D-galactose to a-D-galactose by mutarotase, first described in *Escherichia coli* (Wallenfels et al. 1965) and characterized structurally in *Lactococcus lactis* (Thoden and Holden 2002). After its phosphorylation to galactose-1-phosphate (Kosterlitz 1937; Neufeld et al. 1960) it is activated to UDP-galactose by UDP-glucose-galactose-1-phosphate uridylyltransferase and UDP-glucose as coenzyme Kalckar et al. 1953; Cohn and Segal 1973). This enzyme is absent in hereditary galactosemia (Isselbacher et al. 1956). Second, in addition to the nutritional supply, galactose is formed within cells from UDP-galactose which is synthesized by 4′-epimerization of UDP-glucose (Leloir 1951). UDP-galactose is an essential substrate for the biosynthesis of all soluble and membrane-bound glycoproteins and of glycolipids. With respect to the microdynamics of membrane glycoproteins it should be noted that the turnover rate of galactose is much higher than that of the respective protein moiety (Tauber and Reutter 1983).

For brain cells, glucose is the only natural substrate for either the generation of energy by glycolysis or for the formation of precursors used in the biosynthesis of structural components (Pellerin and Magistretti 2003). Pathological consequences of a decreased cellular supply of glucose in the brain have been clearly shown by Hoyer and co-workers (Hoyer et al. 1988; Hellweg et al. 1992; Blum-Degen et al. 1995; Hoyer 2004; Hoyer and Lannert 2007). They pointed to the nosological role of the severely decreased glucose supply for brain cells in the onset of Alzheimer's disease. It seems logical that in brain cells this deficit leads to a decreased concentration of UDP-glucose and consequently of its 4′-epimer UDP-galactose as available precursors of structural components. As a further consequence of impaired glucose uptake/metabolism, the concentrations of other sugar nucleotides such as UDP-*N*-acetylglucosamine may be decreased. This compound is needed not only for the biosynthesis of structural components of the cell, but also for the attachment of *N*-acetyl-glucosamine to regulatory proteins, their *O*-GlcNAcylation (Zachara and Hart 2004). A typical finding in Alzheimer disease is the decreased *O*-GlcNAcylation of tau protein in favor to its hyperphosphorylation. This disturbance leads to structural failure and dysfunction, a significant characteristic in the pathophysiological sequence leading to this disease Liu et al. 2008).

The uptake of glucose and galactose in brain cells and many other cells is mediated by the insulin-dependent GLUT 4. However, galactose is also transported into brain cells by GLUT 3, which works in an insulin-independent manner (Mueckler 1994; Olson and Pessin 1996). Many metabolic studies have been performed with trace amounts of labeled galactose (Bauer et al. 1976). Therefore, we have focused in the present study on the effect of an acute loads of galactose on the concentrations of the primary metabolites UDP-galactose and UDP-glucose and in addition the secondary metabolites, the amino acid under these loads, since metabolization of galactose to amino acids has been shown earlier in cell culture experiments (Patschinsky et al. 1980).

Materials and methods

Animals

Female Wistar rats, weighing about 150 g, were fed on a commercial diet (Altromin R, Altromin GmbH, Lage/Lippe, Germany) and water ad libitum. The rats were anesthesized by intraperitoneal injection of nembutal (40 mg/kg).

Materials

The chemicals and solutes were of analytical grade and were obtained from Merck (Darmstadt, Germany), Sigma (München, Germany) and Roth (Karlsruhe, Germany). Enzymes, coenzymes and fine chemicals were from Hoffmann-La Roche/Boehringer (Mannheim, Germany) and Sigma. Radioactive hexoses and L-leucine were from NEN (Boston, USA) and American Radiolabeled Chemicals (St Louis, USA). The specific radioactivity of the hexoses was 15 Ci/mmol. The scintillation cocktails Quickszint 2000 and 501 were obtained from Zinsser Analytics (Frankfurt/Main, Germany).

Preparation of samples of rat organs

The rats were bred in our own laboratory and were fed ad libitum. They had free access to tap water and food. The room was kept at constant humidity and temperature (20°C). Under nembutal anesthesia (40 mg/kg) liver and brain were obtained. In brief, after opening the abdominal cavity the suspended liver was fixed by deep frozen (liquid nitrogen) clamp. With respect to brain, the skullcap was opened by pair of scissors and the removed brain was transferred to the patch-clamp as described by Wollenberger et al. (1960) and modified by Keppler et al. (1970). The frozen tissue (about 1 g, 0.7 mm thick) was weighed and immediately homogenized with 5 vol frozen 0.6 N perchloric acid and centrifuged. Sediments were washed with 0.6 mol/l perchloric acid and centrifuged again at

12,000×*g* at 4°C. The combined supernatants were neutralized with solid sodium carbonate and centrifuged again. The supernatants were used for amino acid analysis and enzymatic measurements of sugar nucleotides. In labeling experiments, rats were labeled by intraperitoneal injection of 2.7 mCi/kg of 6-^{3}H-D-galactose or ^{3}H-glucose. The organs were removed as described above.

Blood was drawn from the vena cava with a syringe, allowed to clot and centrifuged at 12,000×*g* for 10 min. Four parts of the resulting serum were mixed with one part of 10% sulfosalicylic acid, mixed for 30 min, and then centrifuged for 10 min at 12,000×*g*.

Analytical procedures

Chromatographic methods

Monosaccharides and amino acids were determined according to the method of automatic amino acid and sugar analysis on the basis of cation-exchange chromatography for amino acids, phosphamino acids and amino sugars with ninhydrin as reagent, and anion-exchange chromatography for neutral sugars with copper bicinchoninate as reagent (Josic et al. 1984). For separation and quantification the liquid chromatographic system of Biotronik (Munich, Germany) was used: LC 6000 for the hexoses and LC 5000 for the amino acids, according to the method of Hamilton (Hamilton 1963). For both procedures a polystyrene support cross-linked with 4–8% divinylbenzol was used; the particle size ranged between 20 and 8 µm, depending on the type of the resin. The counter-ion was lithium. The pH value of the sample before its application to the column was adjusted to 2.2 with concentrated HCl or solid LiOH. The elution was performed with Li citrate with increasing molarity and increasing pH. The amino acids were detected with ninhydrin and the extinction was measured at 570 nm. The extinctions of proline and hydroxyproline were measured at 440 nm. Labeled amino acids were quantified after the ninhydrin reaction by collecting the samples in a fraction collector (Pharmacia, Freiburg, Germany) followed by liquid scintillation counting. To avoid quenching by ninhydrin two runs were performed, an analytical one with ninhydrin detection and a second one without ninhydrin. Each fraction (volume 300 µl) was placed in 5 ml triton–toluene scintillation fluid. Using a L-[1-^{14}C]-leucine standard, the running time difference between the first and the second run was calculated to be 2 min.

Radioactivity was determined by liquid scintillation counting using a liquid scintillation analyzer (Canberra Packard, Zürich, Switzerland). Counting efficiency was monitored with an external standard and by the channel ratio method. Protein was determined by the method of Lowry et al. (1951) with bovine serum albumin as a standard.

Paper chromatography

To separate galactose from its metabolites, ^{3}H-galactose-labeled tissue extracts were chromatographed on Whatman No. 3 MM paper with 1-propanol: Na-acetate: H_20 [7:1:2] as solvent according to Lewin and Wei (1966).

Enzymatic method for quantification of monosaccharide nucleotides

The spectrophotometric measurements were performed with an Eppendorf photometer at 334 nm (extinction NADH = 6.0 cm^2/µmol, light path 1.0 cm). UDP-Glucose and UDP-galactose were determined according to the method of Keppler et al. (1970). In brief, 1 mol UDP-glucose was irreversibly oxidized to 1 mol of UDP-glucuronic acid, using UDP-glucose-dehydrogenase and 2 mol of NAD^+ as coenzyme; the resulting NADH was measured at 334 nm in an Eppendorf photometer. UDP-Galactose-4-epimerase was then added to convert UDP-galactose to UDP-glucose, which was measured as above. The reaction was performed in a buffer containing 0.5 mol/l glycine, 8 mmol/l EDTA and 6 mmol/l NAD^+.

Statistics

Results are presented as the mean ± standard deviation. Each experiment was performed independently three to four times. Statistical analysis was generally performed with the unpaired Student's *t*-test using PRISM software (Graph Pad Software, San Diego, CA, USA). The level of significance was set at $*P < 0.05$ and $**P < 0.01$.

Results

The paper chromatographic separation of the labeled supernatants from the liver, kidney and brain revealed the highest ratio of galactose metabolites (metabolites of hexoses and amino acids) to galactose in the brain where it was 50% higher than in the kidney and 20% higher than in liver (data not shown). The liver shows the highest initial capacity for the uptake and metabolization of galactose (Fig. 1). One hour after the administration of galactose, the concentration of total metabolites in liver was 1.5 times higher than in the brain or kidney and three times higher than in skeletal muscle tissue. Twenty-four hours after injection, the concentration of galactose metabolites was slightly higher in brain (about 20%). The rate of decrease of galactose was slower in the brain than in the liver. The rapid decrease of galactose in the liver is explained by its high need for the glycosylation of serum glycoproteins. To our surprise, in muscle the concentration of the

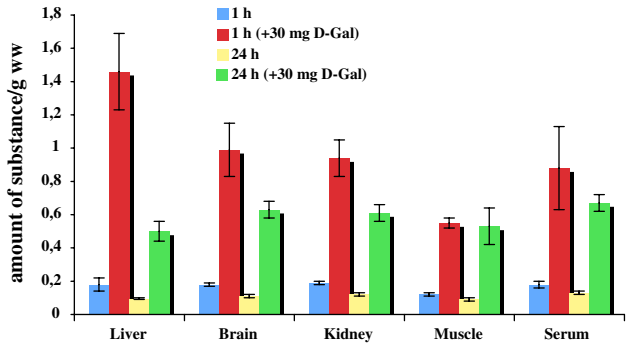

Fig. 1 Concentrations of total acid soluble metabolites in different organs. Two groups of female rats (average weight 150 g) were administered with ^{3}H-galactose of two different specific radioactivities. *Group 1* 300 µCi ^{3}H-galactose/kg b.w. with a specific radioactivity of 10.4 Ci/mmol, *group 2* 200 mg/kg b.w. together with 300 µCi 6-^{3}H-galactose/kg bw with resulting the specific radioactivity of 1.8 mCi/mmol. At 1 and 24 h, respectively, after galactose loads three rats of each group were anesthesized to remove the organs by freeze clamping. The acid soluble samples were obtained as described in the Sect. "Methods." The amounts of the metabolites were calculated from counts measured in each sample based on the specific radioactivities indicated above. Values are given for total acid soluble metabolites (in untreated rats) in nmole/g ww and after galactose loads in µmole/g ww. Statistics according to Student's *t*-test

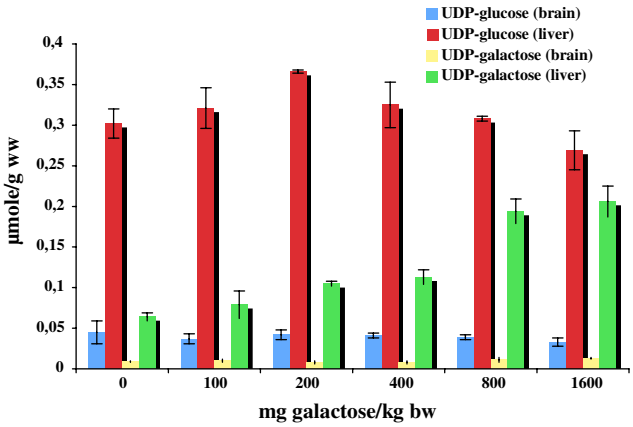

Fig. 2 Dose dependent increases of the concentrations of UDP-hexoses in the brain and in the liver after galactose loads. Female Wistar rats (three rats per dose) were injected intraperitoneally with increasing loads of galactose, dissolved in 0.9% NaCl. After 2 h the rats were anesthesized to remove the organs by freeze clamping and treated as described in the legend to Table 1. In the acid soluble supernatants the UDP-hexoses were separately measured enzymatically. Values of three independent experiments are given. Statistics according to Student's *t*-test

acid-soluble metabolites was maintained for the longest period of time, probably due to the low secretory capacity of this organ.

In the following the main metabolites of galactose, UDP-galactose and UDP-glucose, were measured in the brain and in the liver at increasing loads (Fig. 2) and at different times after the injection of one load of galactose (80 mg/kg b.w.; Fig. 3), respectively. In untreated rats, the concentrations of UDP-glucose and UDP-galactose are in the brain much lower than in the liver (Fig. 2) supporting earlier findings (Wollenberger et al. 1960; Lewin and Wei 1966); however, the concentrations of UDP-galactose measured in the present study are about 25% lower. After giving loads of galactose (Fig. 2), the concentrations of UDP-glucose remained nearly unchanged in the brain, whereas UDP-galactose showed a moderate increase leading to a decrease of the ratio UDP-glucose/UDP-galactose from 5.1 to 2.6 (Table 1). In the liver, the range of this ratio (normally between 3 and 5) decreased to 1.3, due to the increased biosynthesis of UDP-galactose at the high loads. The slightly decreased concentration of UDP-glucose at the high loads is explained by the increased utilization of its UMP-moiety during the biosynthesis of UDP-galactose.

The time dependent measurements showed 4 h after the galactose loads an increase of the ratio to nearly 2.83 (brain) and 4.14 (liver) (Table 2), respectively, due to the decrease of the concentration of UDP-galactose, which serves as precursor for the biosynthesis of glycoconjugates.

As expected the highest concentration of UDP-galactose was measured at the highest dose of galactose given to the rats. In contrast, the concentration of UDP-glucose decreased for doses of galactose higher than 200 mg/kg bw (Table 3). This may be explained by the inhibition of UDP-glucose-4-epimerase by the increased concentrations of galactose-1-phosphate which also inhibits galactokinase and uridylyltransferase (Cuatrecasas and Segal 1965; Bertoli and Segal 1966). After the application of high loads of galactose UDP-galactose increases in the brain, whereas the concentrations of UDP-glucose did not change. In liver, UDP-glucose also remained unchanged (Fig. 2). The concentration of UDP-galactose decreased significantly in the brain as well as in liver 4 h after the galactose loads (Fig. 3a). This decrease may be due to the inhibition of UDP-galactose-4-epimerase by galactose metabolites or by a decrease of the NAD^{+}/NADH-ratio (Hellerstein and Munro 1988).

Whereas the concentrations of UDP-glucose remained in a range between 0.237 and 0.280 µmol/g wet weight, the concentration of UDP-galactose increased significantly in liver as well as in the brain as early as 0.5 h after the administration of galactose,. Under the experimental conditions the administration of glucose did not alter the concentration of either UDP-galactose or UDP-glucose (Fig. 3b).

After the measurements of the hexose nucleotides as the primary metabolites, the secondary metabolites, the amino acids were measured (Fig. 4). Glutamate + glutamine (Fig. 4a) and GABA (Fig. 4b) showed the most pronounced increases in concentration, continuously increasing up

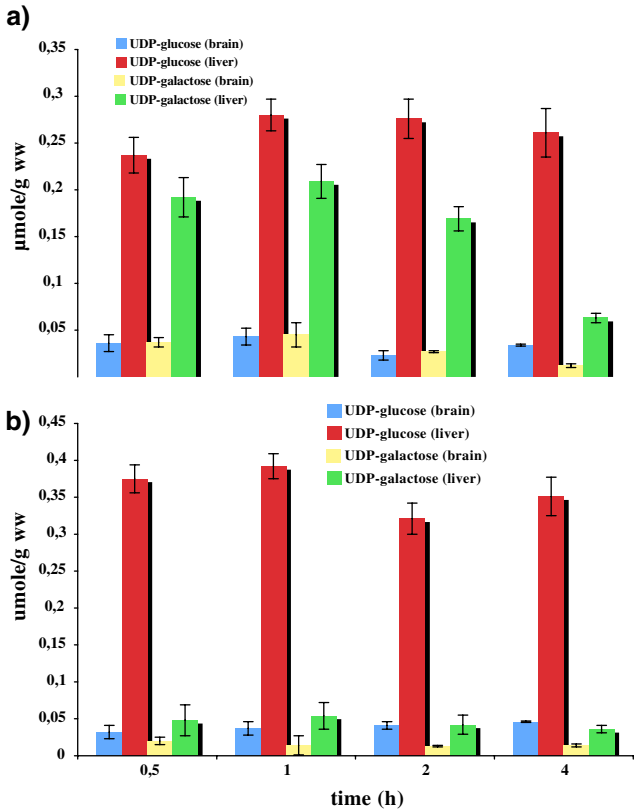

Fig. 3 Time dependent increases of the UDP-hexoses concentrations in the brain and in liver after galactose (**a**) or glucose (**b**) loads. Two groups of female rats received intraperitoneally galactose or glucose. At the times indicated the rats were anesthesized to remove the organs by freeze clamping. Three rats were used per time point. The enzymatic measurements of UDP-hexoses were performed as described in the legends to Fig. 2. At each time point three rats were used leading to three independent measurements. Statistics according to Student's *t*-test

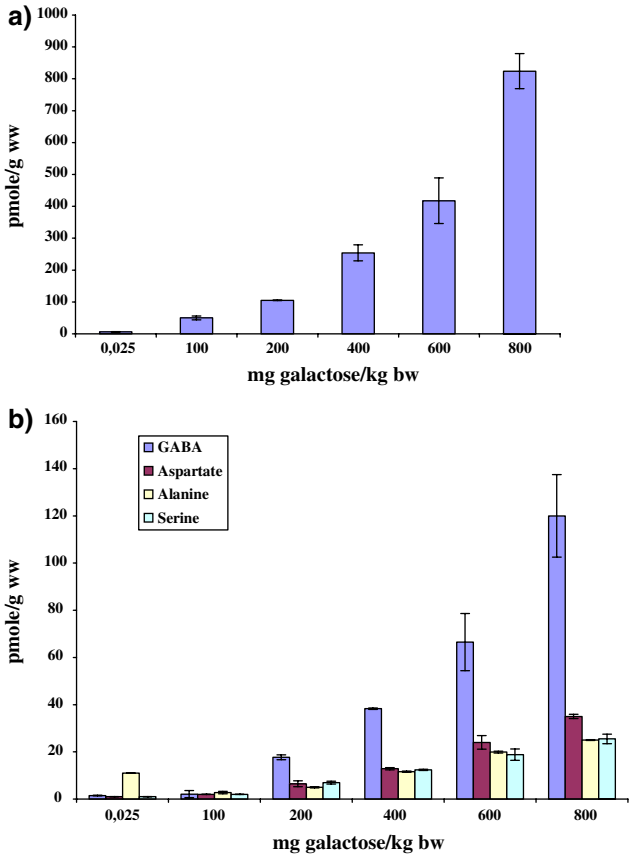

Fig. 4 Dose dependent increases of the concentrations of glutamate + glutamine (**a**) and other amino acids (**b**) in brain after galactose loads. Female Wistar rats (three in each group) were injected intraperitoneally with increasing loads of 6-^{3}H-galactose. The values were calculated from the counts measured and the specific radioactivity of 2.77 mCi/mmol. Two hours after the administration the organs were removed as described in Fig. 1. The acid soluble supernatants were analyzed using an amino acid analyzer. The amounts of amino acids were determined by ninhydrin staining and in the eluents the radioactivities were measured. The increments are calculated on the basis of the counts measured and the specific radioactivity. Statistics according to Student's *t*-test

Table 1 The ratio of UDP-hexoses in the brain and in the liver after galactose loads (dose dependency)

Galactose (mg/kg bw)	Brain (UDP-glucose/ UDP-galactose)	Liver (UDP-glucose/ UDP-galactose)
0	5.1	4.7
100	3.8	4.0
200	5.5	3.5
400	5.2	2.9
800	3.5	1.6
1600	2.6	1.3

Table 2 The ratio of UDP-hexoses in the brain and in the liver after galactose loads (time dependency)

Time (h)	0.5	1	2	4
Q (brain)	0.97	0.95	0.85	2.83
Q (liver)	1.23	1.34	1.63	4.14

Q UDP-glucose/UDP-galactose

to 824.0 nmol/g fresh weight at 800 mg galactose/kg b.w. At 200 mg galactose/kg b.w., aspartate, serine and alanine (Fig. 4b) also showed a significant and continuous increase in concentration, but to a much lesser extent than the glutamate family. It is surprising that in liver the concentrations of the members of the glutamate family do not increase, and a slight increase only is seen for aspartate and serine (17.7 and 11.7 nmole/g fresh weight, respectively). In serum, only serine, glutamate + glutamine and alanine were increased. These values were obtained after giving 600 mg galactose/kg b.w. (data not shown).

The time dependent increment of glutamate and GABA could be measured after loads of galactose or glucose. However, the increments persist longer after galactose loads (Fig. 5).

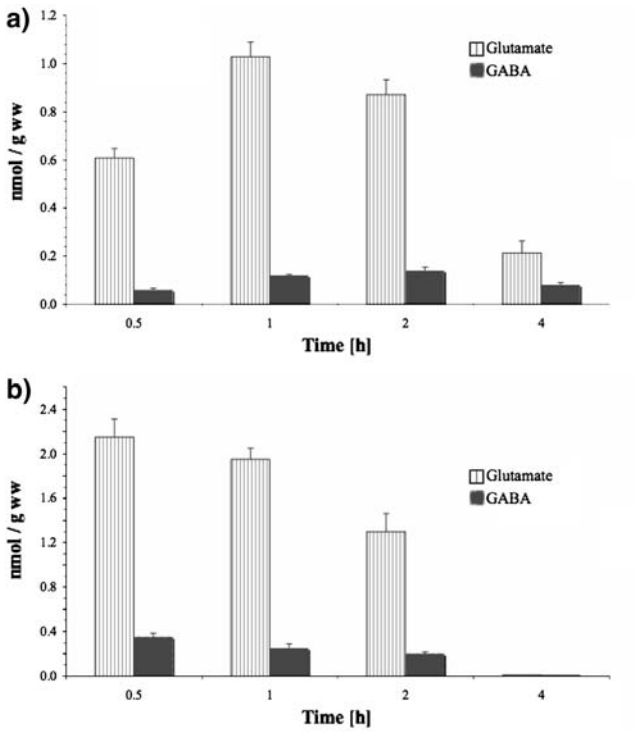

Fig. 5 Time dependency of the increments of glutamate and GABA after loads of galactose (**a**) and glucose (**b**), respectively. At each time point three rats were used. Measurement of the increments at different time after loads. The specific radioactivities: 6-^{3}H-galactose: 0 595 mCi/mmol and 1-^{3}H-glucose: 0.54 mCi/mmol, respectively. For experimental details see legend to Fig. 4. Please note, that 4 h after the loads of galactose the increments of glutamate and GABA are still present, unlike glucose loads

Discussion

The metabolism of galactose by the liver is well documented as reviewed by Cohn and Segal (1973), as is the metabolism of heavy loads in blood cells (Gibson et al. 1996); but little is known about galactose metabolism in the brain. In the present study acute loads of labeled galactose or glucose as control were given to rats followed by the determination of its metabolites. In every organ examined appreciable amounts of labeled acid-soluble hexose metabolites were detected, especially in the brain and liver. In the liver, the total metabolite concentration reached its maximum as early as 30 min after administering galactose, due to the high activities of hepatic galactokinase and uridylyltransferase (Kalckar et al. 1953; Cuatrecasas and Segal 1965; Bertoli and Segal 1966); this increase was followed by a steady decrease of about 60% in the following 4 h. In the brain, however, the ratio of the concentration of total galactose metabolites to the galactose loads applied was higher than in the liver, despite the lower activities of galactokinase and uridyltransferase in the brain (Cuatrecasas and Segal 1965; Bertoli and Segal 1966). The rapid decrease in the liver is primarily explained by its high secretory output of serum glycoproteins. Conversely, in the brain, which has a much lower secretory capacity, the decrease is only about 30% during the same period. Galactose is therefore available for metabolism by other pathways, such as the formation of amino acids. The increase in the concentrations of galactose metabolites, especially of the glutamate family was still significant 4 h after its administration, whereas the increase of metabolites after glucose administration was no longer detectable at that time (Fig. 5). This difference does not correlate with the concentrations of UDP-glucose and UDP-galactose, which decrease concomitantly. The concentration of UDP-glucose remains nearly constant except for a small increase 1 h after galactose administration (Fig. 3). This demonstrates the high demand for both UDP-hexoses for the synthesis of macromolecules, especially for the glycan part of secretory and membrane glycoproteins. Moreover, the relatively high concentration of UDP-galactose may also serve in the biosynthesis of glycogen, as described for the liver (Schlamowitz 1951; Nordin and Hanson 1963; Kunst et al. 1989). In the brain, glycogen, which is present in only low concentrations, is the only endogenous fuel store that can deliver significant amounts of energy in metabolic situations during which the supply of glucose is diminished (McKenna et al. 2006). More recently it has been shown that after intravenous administration of loads of galactose to human volunteers an increase in glycogen was measured (Fried et al. 1996). Moreover, in newborn rats the conversion of galactose into glycogen exceeds that of glucose, suggesting a preference for the use of galactose for neonatal glycogen synthesis (Kunst et al. 1989).

Analysis of the total acid-soluble fraction showed that appreciable amounts of galactose or glucose were converted to amino acids especially in the brain but only in trace amounts in the liver. In the brain where glutamate + glutamine and to a lesser extent GABA were the predominant amino acids formed after galactose. Compared with the base level, the increase was 12% for glutamate + glutamine and 6% for GABA. These could be detected at the lowest dose of galactose used and they displayed a steady dose-dependent increase. After a galactose load aspartate is also formed, but to a much lesser extent than the members of the glutamate family. The sources for these amino acids are pyruvate from glycolysis or α-ketoglutarate and oxaloacetate from the Krebs cycle (Garfinkel 1966; Meister 1979). Glutamate is synthesized from α-ketoglutarate, either by transamination (catalyzed by alanine aminotransferase) or by ammonia fixation, catalyzed by glutamate dehydrogenase (Fig. 6). It is noteworthy that the rate of increase of glutamate concentrations is most marked in the brain, where it may serve as a neurotransmitter. The recorded increases are average

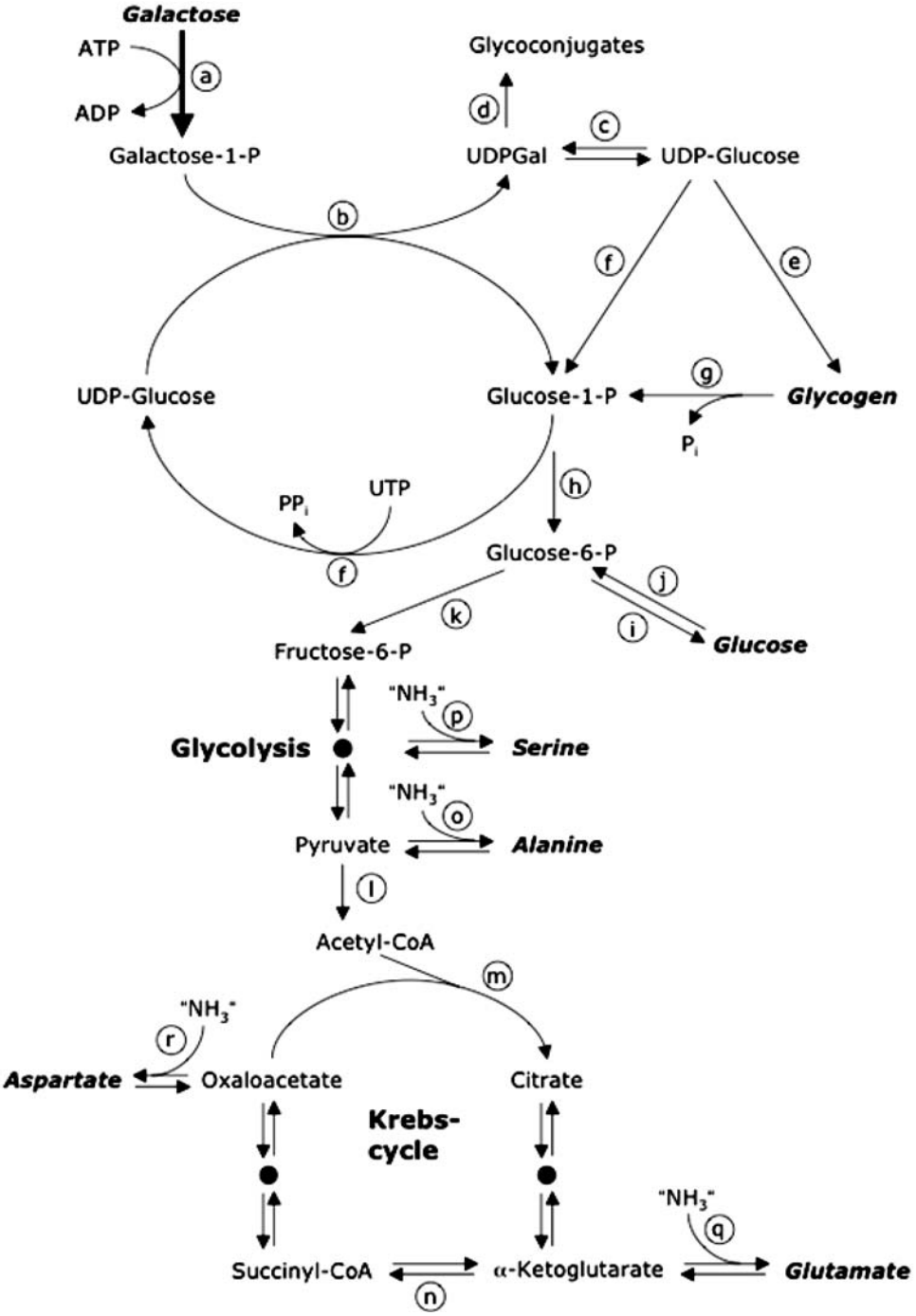

Fig. 6 Metabolism of galactose to amino acids. Enzymes: *a* Galactokinase, *b* UDP-galactose/glucose-1-phosphate-uridyltransferase (uridylyltrans-ferase), *c* UDP-glucose/UDP-galactose-4′-epimerase, *d* galactosyltrans-ferases, *e* glycogensynthase, *f* UDP-glucose-pyrophosphorylase, *g* glycogenphosphorylase, *h* glucose-6-phosphate mutase, *i* glucose-6-phosphatase, *j* glucokinase (in liver only), hexokinase, *k* phosphoglucomutase, *l* pyruvate dehydrogenase, *m* citrate synthase, *n* α-ketoglutarate dehydrogenase, *o* alanine aminotransferase (ALAT, GPT), *p* 3-phosphoglycerate dehydrogenase, *q* glutamate-oxaloacetate-aminotransferase (GOT) or glutamate-pyruvate-aminotransferase (GPT)—NH_3, ammonium equivalent

values for the whole brain; this study did not distinguish between the different regions of the brain. Normally, the highest concentrations of glutamate are in the presynaptic vesicles of neurons of the putamen and the nucleus caudatus, the cerebellar cortex, the thalamus and the frontal and occipital cortex; GABA is concentrated in the globus pallidus, substantia nigra, nucleus dentatus and to a lesser extent in the nucleus accumbens. Concentrations of these amino acids in specific areas of the brain will be the subject of a subsequent study.

An inverse situation is seen in the liver, where aspartate and serine show significant increases, while no increase was detected in the formation of glutamate + glutamine. Moreover, in the liver the increase of these amino acids is <5% of that in the brain. This demonstrates that hexoses are the most efficient resource for the biosynthesis of glutamate and its derivatives in the brain.

In order to be useful for the formation of these amino acids, galactose must be metabolized to glucose traversing the Leloir pathway with its final UDP-hexose epimerization

step. As a resource for amino acid synthesis, galactose has an advantage over glucose because the soluble galactose metabolites persist for longer time in the brain. It may be hypothesized that this longer persistence is due to the increased amount of galactose in glycogen combined with the fact that glycogen phosphorylase prefers glucose to galactose in the terminal positions of glycogen. Furthermore, the compartmentalization as shown for glutamate may exist also for glucose and galactose metabolism. An extended neuronal compartment of glutamate is formed, while glutamine is synthesized in a small astroglial compartment (Garfinkel 1966; Berl and Clarke 1983). Similarly, a compartment storing galactose separately from glucose may be assumed, based on the longer persistence of the increased concentration of galactose metabolites compared to glucose in the brain (Fig. 5). This assumption is also supported by the different rates of decrease of galactose- and glucose metabolites. Additionally it should be taken into account, that in the brain compartmentalization also occurs with regard to the energy source glycogen between astrocytes and neurons or axons (Brown and Ransom 2007).

The biosynthesis of amino acids from hexoses as shown here consumes equivalents of ammonia. Therefore, the increased supply of hexoses, followed by amino acid biosynthesis may serve as a means of ammonia detoxification in the brain. Progressive liver diseases are accompanied by a decreased capacity to remove the ammonia that arises from amino acid breakdown; this ammonia enters easily the brain, where it acts as a toxic agent, leading to hepatic encephalopathy. In a preliminary study it was shown that six patients suffering from state III/IV of hepatic encephalopathy recovered much faster after intravenous infusion with galactose (1.0 g/kg b.w. Heuckenkamp and Zöllner 1975) than those treated with glucose only (R. Büchsel, H. Elsbernd, W. Reutter, unpublished). Moreover, ammonia is increased in Alzheimer's disease and is suspected to be a factor able to produce symptoms of Alzheimer's disease and to affect the progression of the disease (Seiler 2002). Therefore, it may be suggested to include galactose in the treatment schedule of patients with impaired brain functions.

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