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Stimulation of renal amino acid reabsorption after treatment with triiodothyronine or dexamethasone in amino acid loaded rats

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Summary. In adult female anaestetized rats, the influence of triiodothyronine or dexamethasone on renal amino acid handling was investigated in leucine (20 mg/100 g b.wt.) or glutamine (45 mg/100 g b.wt.) loaded animals. Bolus injections of both amino acids were followed by temporary increase in fractional excretion of the administered amino acids as well of the endogenous amino acids which were not administered.

Under load conditions (leucine and glutamine), dexamethasone treatment (60 μg/100 g b.wt. for 3 days, i.p. once daily) was followed by a stimulation of renal amino acid reabsorption. The increase in fractional amino acid excretion after amino acid load was significantly lower than in untreated rats. The effect of triiodothyronine (20 μg/100 g b.wt. for 3 days, i.p. once daily) was different in leucine and glutamine loaded animals: after leucine bolus injection a comparable stimulatory effect as shown for dexamethasone could be demonstrated, but after glutamine administration the stimulatory action of T3 was masked. T3 even increases fractional amino acid excretion in glutamine loaded rats as a sign of enhanced "house-keeping" in the renal tubular cells. These results confirm previous findings and indicate different effects of both hormones on the renal handling of amino acids.

Keywords: Amino acid transport – Kidney – Triiodothyronine – Dexamethasone – Amino acid load – Rats

Introduction

Only little information exists in the literature describing hormonal control of renal amino acid handling. For example, testosterone was shown to stimulate amino acid uptake in murine renal cortical slices (Koenig et al., 1982), parathyroid hormone increases amino acid excretion by inhibition of amino acid reabsorption (Scriver and Bergeron, 1974), and dexamethasone stimulates taurine transport in flounder renal tubules (King et al., 1982). In own

experiments (Fleck, 1992a), repeated administration of triiodothyronine (T3) or dexamethasone caused significant changes of endogenous amino acid plasma concentrations in young (10 days old) and adult rats (2 months old). In the kidney, the reabsorbed fraction of endogenous amino acid was enhanced after both T3 and dexamethasone treatment in young rats, whereas in adult rats the two hormones were without influence on tubular reabsorption of amino acids. In young animals the fractional excretion of endogenous amino acids was reduced in 15 of 22 amino acids after dexamethasone and in 12 of 23 amino acids after T3 treatment, indicating stimulatory effects of both hormones on tubular amino acid carrier systems in immature animals. In adult rats the stimulatory effects of hormone treatment could not be found under physiological conditions. However, after administration of an amino acid mixture containing various amino acids in high doses to overload the renal amino acid reabsorption capacity the reabsorbed amounts of amino acids were higher in rats treated with dexamethasone, but not after treatment with T3. But there was no stimulatory hormone effect concerning fractional excretions of the amino acids, neither after dexamethasone nor after T3 (Fleck and Nußbaum, 1996).

On the other hand, in both immature and adult rats it is possible to stimulate the renal excretion of p-aminohippurate after treatment with the two hormones (Bräunlich, 1984; Bräunlich et al., 1986). This means, the tubular secretion of organic anions, located at the basolateral membrane of the renal tubuli (Ullrich, 1994), can also be stimulated in adult animals both after dexamethasone and T3. Therefore the lack of effect of T3 treatment on renal amino acid reabsorption should be investigated in this study. After loading the animals with high amounts of amino acids it should be possible to prove stimulating hormone effects, because amino acid reabsorption is employed to capacity under these conditions (Silbernagl, 1992). In our study, leucine and glutamine were chosen as bolus injections to overload amino acid reabsorption carriers. In accordance with Yao et al. (1994), 6 different transport systems for leucine transport seem to exist. Jorgensen et al. (1990) described leucine transport in vesicles of the tubular membrane in the nephron. This transport is pH-dependent, H^+-independent and electrogenic. Genetic characterization of the carrier (Matsubara et al., 1992), determination of leucine-specific binding proteins (Williamson and Oxender, 1990) and the analysis of the amino acid sequence of the leucine carrier (Adams et al., 1990; Hoshino et al., 1992) are described. Glutamine is transported via at least two different carrier systems in the kidney (Weiss, 1978). The transport is an active one, Na^+-dependent and electrogenic (Tanaka et al., 1989; Welbourne, 1989). Beside rBAT (Bertran et al., 1992) two further glutamine transporters were differentiated recently by Tate et al. (1992) and Nakanishi et al. (1994).

Both amino acids are nearly non toxic. Therefore they can be administered in relatively high doses. Furthermore, their renal reabsorption is quite effective indicating the high capacity of the respective reabsorption carriers. For these reasons it could be expected that after leucine and/or glutamine load stimulatory effects of dexamethasone or T3 could be proven for the

renal amino acid handling. Nevertheless, using amino acid loading, beside renal effects metabolic actions of both hormones must also be taken into consideration in *in vivo* experiments and the possible stimulation of renal amino acid reabsorption could be masked by extrarenal hormone mechanisms.

Material and methods

Animals

Investigations were performed on female Wistar rats (Han:Wist) of our institute's own out-bred stock. At the beginning of the experiments the animals were 2 months old and the average body weight was 162 ± 6g. The rats were kept under standardized conditions including standard Altromin 1316 diet and free access to tap water.

Experimental design

The rats were anaesthetized with ketamine (Ursotamin®, Serumwerk Bernburg, F.R.G., 7.5 mg/100 g b.wt.) and xylazine (Ursonarkon®, Serumwerk Bernburg, F.R.G., 1.2 mg/100 g b.wt.). Both substances were administered intramuscularly. A catheter was placed in a tail vein. The animals were then infused isotonic saline containing 4 g/l fluorescein isothiocyanate (FITC)-inulin (Bioflor, Uppsala, Sweden) at a priming rate of 3 ml/100 g b.wt. per 1 hour for 15 minutes and then at 2 ml/100 g b.wt. per hour for the remainder of the experiment. Thereafter a polyethylene catheter was inserted into the urinary bladder by opening the abdominal cavity. To minimize urine collecting periods for the determination of GFR and fractional amino acid excretion (FE), urine was collected in 30-minute periods over a period of 3 hours. In previous experiments it could be shown that under these experimental conditions both hematocrit (Fleck et al., 1992) and blood pressure (Fleck and Bräunlich, 1986) remain nearly constant during the clearance study. In the middle of each period and at the end of the experiment blood was collected from the retrobulbar plexus.

Glomerular filtration rate (GFR) was determined by inulin clearance. Inulin concentration was measured spectrofluorometrically using FITC-inulin (Sohtell et al., 1983) in blood and urine samples. Fluorescence was measured at 480 nm excitation and 520 nm emission wave lengths in a HITACHI F-2000 spectrofluorometer.

Amino acid load

Rats were loaded with leucine (20 mg/100 g b.wt.) or glutamine (45 mg/100 g b.wt.). The amino acids were dissolved in 2 ml normal saline per 100 g b wt. Leucine and glutamine were chosen for the following reasons: The physiological concentrations of these amino acids are relatively high and their FE is relatively low. Thus their renal reabsorption seems to be very effective. Amino acid doses were determined in previous experiments. It was the goal of the experimental schedule to enhance amino acid plasma concentration more than 10fold, whereas toxic effects of amino acids should be prevented. The injection solutions of leucine and glutamine had an osmolarity of 365 and 322 mosmol and a pH of 7.5 and 5.2, respectively. Both amino acids were administered as a bolus injection intravenously at the beginning of the clearance experiment. Control animals received the same volume of normal saline.

Hormone treatment

Triiodothyronine (T_3; SIGMA, St. Louis, U.S.A.) was administered intraperitoneally in doses of 20 μg/100 g b.m. once daily for 3 days. In this dose range, hormone receptor sites are completely saturated (Azimova et al., 1986).

Dexamethasone (Dexa; Fortecortin° Mono; E. Merck, Darmstadt, F.R.G.): 60 μg/100 g b.m. were given i.p. for 3 days, once daily. Following this dose, glucocorticoid receptor sites are completely saturated by dexamethasone (Rafestin-Oblin et al., 1986).

Both substances were dissolved in normal saline (1 ml/100 g b.m.). Controls received the solvent only.

Amino acid determination

The determination of amino acids by column chromatography with fluorescence detection is based on that developed by Roth and Hampai (1973) and has been described in detail elsewhere (Silbernagl, 1983). Briefly, proteins were removed from urine and plasma samples by administration of trichloroacetic acid. After centrifugation, the supernatant was neutralized by adding 0.4 N NaOH. Then the samples were diluted with citrate buffer and analyzed by HPLC on an amino acid analyzer (Knauer, Berlin, F.R.G.) with o-phthalaldehyde as a fluorescent amino ligand (Roth, 1971). Calibration runs were performed with freshly prepared amino acid solutions composed of analytical grade amino acids (Serva, Heidelberg, F.R.G.).

Mode of presentation and statistics

Clearances (CL) of amino acids (AA) were calculated as usual:

$$CL_{AA} = (\text{urine conc.}_{AA} * \text{urine volume}) : \text{plasma conc.}_{AA}$$

To calculate the amounts of amino acids reabsorbed in the renal tubules, the difference between filtered amount (plasma conc._{AA} * $\text{clearance}_{inulin}$) and amount excreted into urine (urine conc._{AA} * urine volume) was determined. The fractional excretion (FE) of amino acids is given in % of $\text{clearance}_{inulin}$:

$$FE_{AA} = (\text{clearance}_{AA} : \text{clearance}_{inulin}) * 100\ [\%]$$

The results are summarized as means ± S.E.M. with n = 6 in each group. The level of significance for differences between the observations was assessed with Student's t-test and considered statistically significant when $p \leq 0.05$ (FE) and ≤ 0.0001 (plasma concentrations), respectively.

Results

As shown in Table 1, physiological amino acid plasma concentrations and parameters for the renal handling of amino acids could be reproduced in our rat strain. The amino acids can be differentiated into those with low fractional excretion including leucine and glutamine and those with high FE indicating less effective renal tubular reabsorption of the respective amino acid (acidic amino acids: Asp, Glu; Phe; ß-Ala and Tau). There is obviously no correlation between physicochemical properties of the amino acids and their renal transport. In general, amino acid reabsorption is very effective and reaches about 96–99% of the filtered amino acid load.

Table 1. Normal plasma concentrations and various parameters characterizing the renal transport of amino acids (AA). Arithmetic means ± S.E.M.; n = 6–9

Amino acid	Plasma concentration	AA-clearance	FE	Amount		
				filtered	reabsorbed	excreted
	[μM]	[μl/min * 100 g]	[%]	[nmol/min * 100 g b.wt.]		
acidic AA						
Asp	45 ± 3	37.9 ± 3.9	4.14 ± 0.38	41 ± 4	40 ± 3	1.7 ± 0.2
Glu	112 ± 8	25.3 ± 9.1	2.61 ± 0.74	104 ± 4	101 ± 4	2.9 ± 0.9
basic AA						
Arg	138 ± 4	4.3 ± 0.4	0.51 ± 0.04	122 ± 9	122 ± 9	0.6 ± 0.1
Lys	200 ± 4	2.9 ± 0.3	0.32 ± 0.03	174 ± 9	174 ± 9	0.6 ± 0.1
neutral AA aliphatic						
Ala	202 ± 4	7.5 ± 1.0	0.85 ± 0.08	180 ± 12	179 ± 12	1.5 ± 0.2
Gly	136 ± 3	13.8 ± 2.1	1.51 ± 0.04	125 ± 4	123 ± 3	1.9 ± 0.3
Ile	139 ± 12	3.2 ± 0.4	0.36 ± 0.05	123 ± 7	123 ± 7	0.5 ± 0.1
Leu	*158 ± 7*	*3.7 ± 0.6*	*0.42 ± 0.06*	*141 ± 5*	*140 ± 5*	*0.6 ± 0.1*
Ser	170 ± 3	8.7 ± 1.7	0.97 ± 0.14	150 ± 9	148 ± 9	1.5 ± 0.3
Val	255 ± 18	2.6 ± 0.5	0.29 ± 0.04	226 ± 11	225 ± 11	0.6 ± 0.1
aromatic						
Phe	95 ± 8	60.3 ± 14.2	6.43 ± 1.02	88 ± 3	83 ± 3	5.4 ± 0.8
Tyr	76 ± 9	8.1 ± 1.3	0.90 ± 0.12	69 ± 5	69 ± 5	0.6 ± 0.1
ω-amides						
Asn	63 ± 3	10.0 ± 1.4	1.10 ± 0.12	60 ± 4	57 ± 4	0.7 ± 0.1
Gln	*307 ± 5*	*6.7 ± 0.6*	*0.77 ± 0.05*	*274 ± 11*	*272 ± 11*	*2.1 ± 0.2*
others						
β-Ala	11 ± 1	24.4 ± 7.6	2.45 ± 0.55	10 ± 1	10 ± 1	0.3 ± 0.1
Tau	106 ± 2	22.9 ± 4.6	2.46 ± 0.43	96 ± 3	94 ± 2	2.4 ± 0.5

After administration of both leucine and glutamine the plasma concentrations of these amino acids increased as expected more than tenfold (Fig. 1). But already one hour after administration of the two amino acids their plasma concentrations were no longer different from baseline values as a sign of rapid normalization of disturbances in amino acid homoeostasis. During the elevated amino acid plasma concentrations, the fractional excretions of leucine and glutamine are significantly enhanced as a consequence of amino acid carrier overloading. In the case of glutamine, increase in FE is in parallel with the rise in plasma concentration, whereas the FE of leucine remains enhanced after normalization of its plasma concentration up to the end of the experiment (Fig. 1).

The bolus injection of both leucine and glutamine has no distinct influence on plasma concentrations of the physiological amino acids if one considers a high level of significance ($p \leq 0.0001$) because of the well-known variations in normal values of plasma concentrations (Table 2). But in the first clearance period after amino acid administration the fractional excretion of 13 of 15 endogenous amino acids is enhanced after glutamine loading as a consequence of overloading of amino acid reabsorption capacity. After leucine

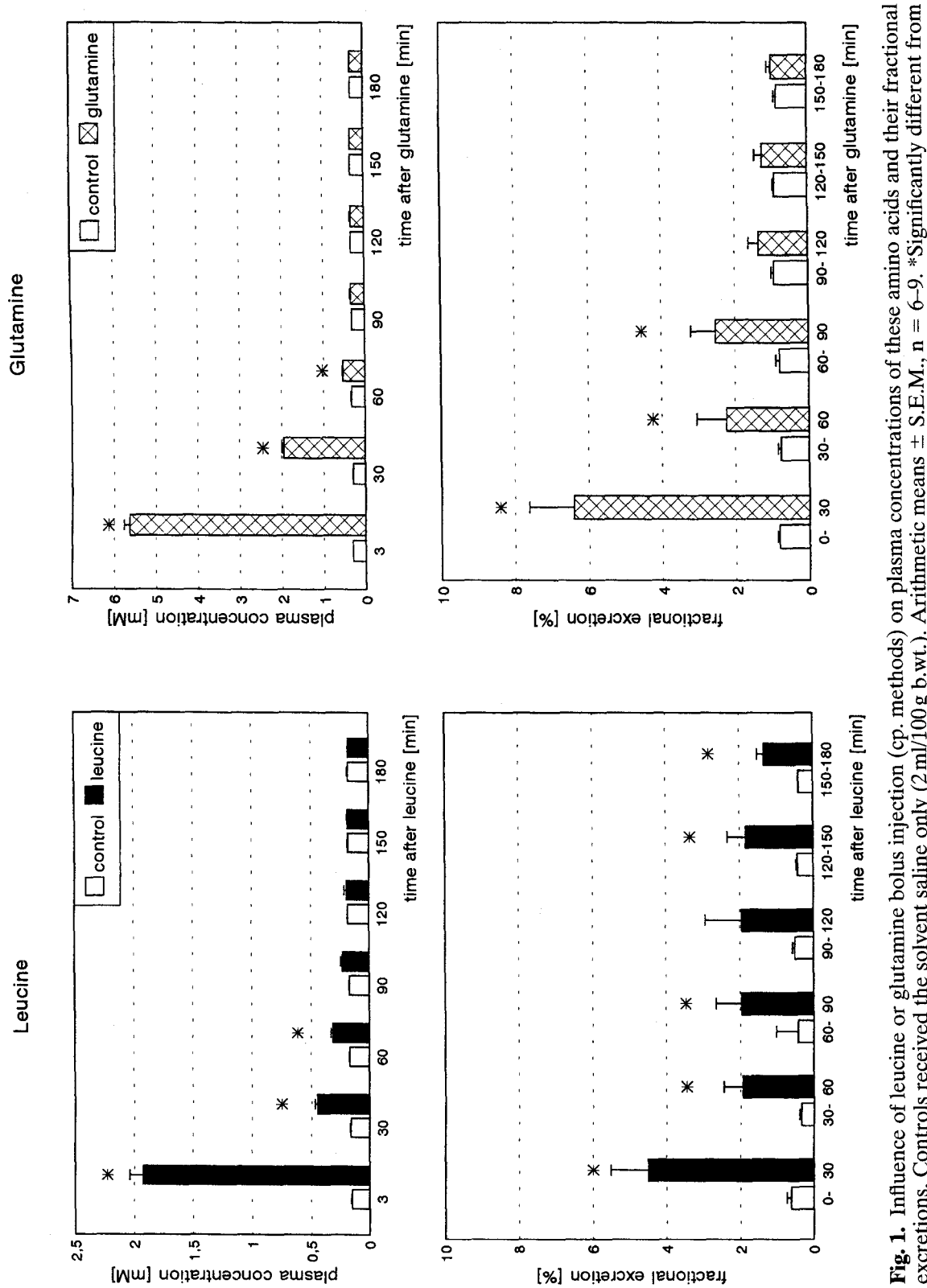

Fig. 1. Influence of leucine or glutamine bolus injection (cp. methods) on plasma concentrations of these amino acids and their fractional excretions. Controls received the solvent saline only (2 ml/100 g b.wt.). Arithmetic means ± S.E.M., n = 6–9. *Significantly different from saline group ($p \leq 0.05$)

Table 2. Amino acid plasma concentrations and fractional excretion (FE) of amino acids in rats immediately after bolus injection of leucine or glutamine in comparison with saline treated animals. Arithmetic means ± S.E.M.; n = 6–9

Amino acid	Plasma concentration [μM]			FE [%]		
	NaCl	leucine	glutamine	NaCl	leucine	glutamine
acidic AA						
Asp	45 ± 3	52 ± 2	61 ± 4	4.1 ± 0.4	2.3 ± 0.1*	29.6 ± 6.9*
Glu	112 ± 8	289 ± 9	208 ± 15	2.6 ± 0.7	4.4 ± 0.7	4.6 ± 1.3
basic AA						
Arg	138 ± 4	108 ± 5	175 ± 5	0.5 ± 0.1	0.9 ± 0.1*	0.8 ± 0.2
Lys	200 ± 4	423 ± 20	199 ± 9	0.3 ± 0.1	0.8 ± 0.1*	2.4 ± 0.9*
neutral AA						
aliphatic						
Ala	202 ± 4	307 ± 22	267 ± 18	0.9 ± 0.1	0.9 ± 0.1	4.5 ± 1.2*
Gly	136 ± 3	123 ± 5	116 ± 2	1.5 ± 0.1	7.2 ± 1.1*	4.8 ± 0.4*
Ile	139 ± 12	103 ± 9	65 ± 7	0.4 ± 0.1	n.d.	10.0 ± 2.7*
Leu	*158 ± 7*	*1,936 ± 87***	*117 ± 8*	*0.4 ± 0.1*	*6.9 ± 1.4**	*2.1 ± 0.3**
Ser	170 ± 3	155 ± 11	135 ± 6	1.0 ± 0.1	5.6 ± 0.9*	5.0 ± 0.9*
Val	255 ± 18	232 ± 12	233 ± 20	0.3 ± 0.1	1.6 ± 0.1*	1.9 ± 0.5*
aromatic						
Phe	95 ± 8	55 ± 8	64 ± 4	6.4 ± 1.0	4.6 ± 0.3*	n.d.
Tyr	76 ± 9	54 ± 1	73 ± 4	0.9 ± 0.1	5.4 ± 0.8*	4.3 ± 0.8*
ω-amides						
Asn	63 ± 3	48 ± 2	127 ± 9	1.1 ± 0.1	2.6 ± 0.5*	7.6 ± 2.1*
Gln	*307 ± 5*	*362 ± 21*	*5,615 ± 90***	*0.8 ± 0.1*	*4.2 ± 0.9**	*6.3 ± 1.4**
others						
β-Ala	11 ± 1	8 ± 1	41 ± 25	2.5 ± 0.6	23.1 ± 4.2*	15.6 ± 5.3*
Tau	106 ± 2	340 ± 2	92 ± 2	2.5 ± 0.4	15.0 ± 2.7*	9.7 ± 1.9*

* FE significantly different from saline bolus ($p \leq 0.05$). ** Increase in plasma concentration more than 10fold compared to saline bolus.

Table 3. Urine volume, GRF, and kidney wet weight after treatment with dexamethasone (Dexa) or triiodothyronine (T3) in rats loaded with leucine (Leu) or glutamine (Gln), respectively (cp. method). Comparison with non hormone treated rats (Co) and rats with saline loading (2 ml/100 g b.wt.) instead of amino acid load

Group	Urine volume [ml/100 g b.wt.]		GFR	Kidney weight [mg/100 g b.wt.]
	0–30 min	0–180 min	[ml/min * 100 g b.wt.]	
Co + NaCl	2.0 ± 0.4	6.4 ± 0.3	0.92 ± 0.06	834 ± 18
Dexa + NaCl	1.1 ± 0.5	4.1 ± 0.4**	0.96 ± 0.10	964 ± 25**
T3 + NaCl	0.7 ± 0.1**	4.2 ± 1.2	0.78 ± 0.07	881 ± 31
Co + Leu	2.5 ± 0.2	9.9 ± 1.0*	1.10 ± 0.06	n.d.
Dexa + Leu	2.4 ± 0.3*	7.4 ± 0.7*	1.15 ± 0.05	n.d.
T3 + Leu	1.1 ± 0.1**	7.0 ± 0.9	0.94 ± 0.04	n.d.
Co + Gln	2.7 ± 0.3	9.7 ± 1.2*	1.18 ± 0.06*	852 ± 26
Dexa + Gln	1.8 ± 0.3	4.9 ± 0.5**	1.02 ± 0.06	898 ± 24
T3 + Gln	1.2 ± 0.2**	5.4 ± 1.0**	1.30 ± 0.11*	1050 ± 25**

Arithmetic means ± S.E.M.; n = 6–9. * Significantly different from saline loading ($p \leq 0.05$); ** Significant effect of hormone treatment ($p \leq 0.05$).

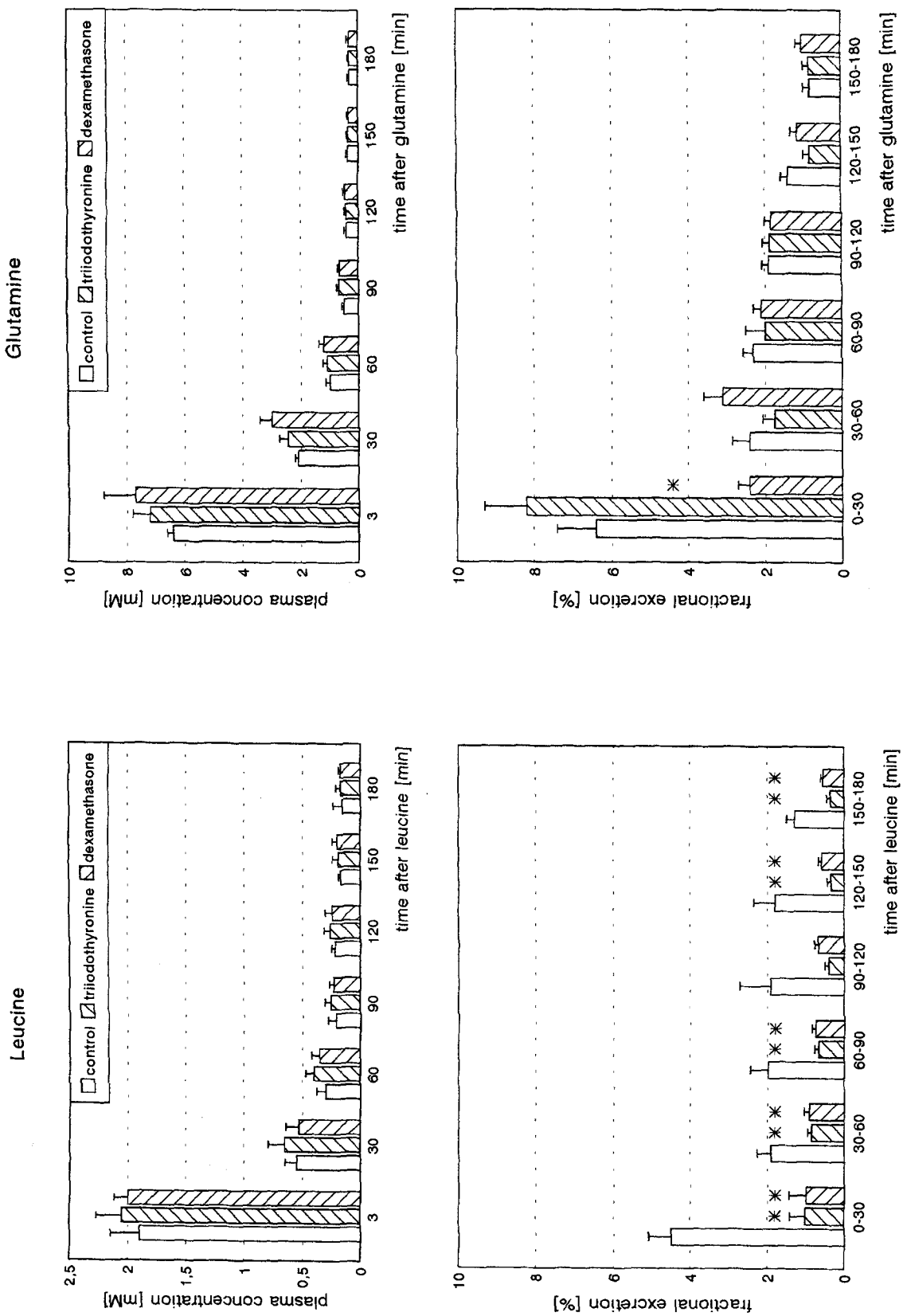

Fig. 2. Influence of a treatment with triiodothyronine or dexamethasone (cp. method) on leucine and glutamine plasma concentrations, respectively, and on the fractional excretion of these amino acids in leucine or glutamine loaded rats (cp. method). Arithmetic means ± S.E.M., n = 6–9. *Significantly different from non hormone treated rats ($p \leq 0.05$)

bolus 12 of 15 FE values are increased (Table 2), too; the reason for the decreased FE of aspartic acid remains open.

In the first clearance period after administration of leucine or glutamine, the urine volume is not different compared to that of rats injected only a bolus of normal saline. The urine output during the three hours of the experiment, however, is significantly higher in amino acid loaded animals (Table 3). On the other hand, the GFR is almost hardly influenced by the amino acid bolus injections.

Three days treatment with dexamethasone or T3 caused the well-known changes in renal function: urine flow is diminished although the GFR is not significantly changed. After dexamethasone the kidney net weight increased (Table 3). The hormone treatments are without significant effects on the plasma disappearance of leucine and glutamine after bolus injection of these amino acids: about one hour after administration of leucine or glutamine control values were reached (Fig. 2). The most impressive result of this study is given in the lower part of Fig. 2: The fractional excretions of leucine and glutamine are significantly reduced after dexamethasone treatment as a sign of stimulated renal tubular amino acid reabsorption. The effect is more pronounced after leucine bolus injection. The stimulatory effectiveness of T3 becomes visible only in the case of leucine loading: its FE is significantly lower in T3 treated animals indicating improved amino acid reabsorption. In glutamine loaded rats this stimulation phenomenon is missing.

Discussion

The physiological values of amino acid plasma concentrations and the normal parameters describing renal amino acid handling measured in this study are in good accordance with own previous findings (Fleck, 1992a,b; Fleck and Nuaßbaum, 1996) and data from the literature (Lingard et al., 1974). Differences between these three sources caused by differences in food intake, animal strain, sex, and in methodological difficulties, can be neglected for further interpretation of the results.

Immediately after administration of a volume load (2 ml/100 g b.wt.), urine flow is significantly enhanced. This polyuria should be kept in mind, because it shortens the contact time of the ultrafiltrate within the tubuli followed by a possibly reduced amino acid reabsorption. This effect, however, is quite similar in amino acid and saline loaded rats and, therefore, it is without consequence for the subsequent comparisons between the different experimental groups. The administered bolus volume is nearly completely excreted within the first clearance period and hyperhydratation is avoided. Furthermore, glomerular filtration is slightly enhanced after amino acid load. This well-known phenomenon is described to be caused e.g. by glucagon, the secretion of which is distinctly enhanced after amino acid administration (Brenner et al., 1982; Alvestrand and Bergström, 1984).

As expected, after amino acid load, plasma concentrations and fractional excretion of the administered amino acids (leucine, glutamine) increased

distinctly even if an unusually high level of confidence ($p \leq 0.0001$) is considered. This high level of confidence for the comparison of plasma concentrations was chosen because of the interindividual variations in amino acid plasma concentrations (Silbernagl, 1988). But the plasma concentrations of the administered amino acids reached baseline values already after 60 minutes, whereas their FE remained enhanced up to 90 min. (glutamine) or even up to the end of the experiment (leucine). This finding reflects the effective regulation of amino acid homoeostasis in the organism within a relatively short time. Nevertheless, the capacity of the transport carriers for both glutamine and leucine seems to be employed to capacity after administration of the respective amino acid.

The amino acid load, however, also interferes with the transport of endogenous amino acids which were not administered. This is in accordance e.g. with Tietze et al. (1992) and Fleck and Nußbaum (1996) describing similar findings for amino acid load in humans and rats, respectively. The reason for this phenomenon could be

- a direct competition at the amino acid carrier sites in the kidney between leucine or glutamine and endogenous amino acids using the same carriers (Collarini and Oxender, 1987; Christensen, 1990),
- a loss of ATP and, therefore, a reduction of the Na^+-gradient in the renal tubuli (Gutmann et al., 1993) as a consequence of enhanced GFR and urine flow,
- the reduced contact time in the nephron because of the increased urine flow,
- or it could be caused by displacement of endogenous amino acids at the carrier sites by the amino acid administered.

Transamination steps reported by Blade et al. (1988), who administered amino acids and found enhanced plasma levels of endogenous amino acids, are of minor importance because changes in plasma amino acid concentrations were observed already 3 to 5 minutes after amino acid load. Therefore the most probable reason for the general increase in endogenous amino acid plasma concentrations seems to be the interferences at the transport carrier systems, because the fractional excretion of many endogenous amino acids was significantly enhanced after amino acid loading. Altogether, the amino acids which were not administered can be divided into three groups:

- distinctly increased plasma concentrations after amino acid bolus injection
- moderate rise in plasma concentration after amino acid loading
- unchanged plasma concentration after amino acid load

First of all this differentiation is caused by different extents of interrelationship between endogenous and administered amino acids. At the end of the clearance experiments the amino acids administered were completely excreted and plasma concentrations of endogenous amino acids reached baseline values, too.

The treatment of the animals with T3 or dexamethasone, two catabolic hormones, caused different effects on renal amino acid handling. After administration of the two hormones, kidney weight is slightly increased, but this effect is significant after T3 only in saline loaded rats and after dexamethasone only in leucine loaded animals (cp. Table 3). In principle, urine flow is reduced after T3 and dexamethasone, especially in amino acid loaded rats. The glomerular filtration rate is not changed significantly, neither after T3 nor after dexamethasone administration and independently of amino acid load. This result confirms previous findings reporting only slight to moderate changes in urine flow and GFR after hormone pretreatment, too (Bräunlich, 1984; Bräunlich et al., 1986; Fleck and Nußbaum, 1996).

For the renal transport of p-aminohippurate (PAH) it could be shown that both T3 and dexamethasone are able to stimulate this transport at the basolateral site of the renal tubular cell (Bräunlich, 1988). This phenomenon should be proved in our study for transport steps located at the luminal membrane, e.g. for amino acid reabsorption. As mentioned in the introduction, a stimulation of amino acid transport can be expected only after employment of the transport capacity. In non amino acid loaded animals no effect of both T3 or dexamethasone on renal amino acid transport could be found in adult rats. Therefore, in this study hormone effects were investigated after amino acid loading as described above. Previous experiments have already indicated the principal possibility to stimulate the renal tubular handling of amino acids (Fleck, 1992b; Fleck and Nußbaum, 1996). However, in these studies the stimulatory hormone effect on the kidney was ambigiuous. In the present investigation it could be shown clearly that three days administration of both T3 and dexamethasone significantly reduces the fractional excretions of leucine (T3 and dexamethasone) and glutamine (dexamethasone only) in amino acid loaded rats. The latter finding is in good accordance with previous results: after administration of amino acid mixtures containing leucine or glutamine, an increase of the reabsorbed amino acid amount could be found only after dexamethasone (Fleck and Nußbaum, 1996). Therefore it can be concluded that the hormonal control of renal amino acid handling is not unique. Obviously the various amino acid carriers are differently influenced by the two hormones. The following reasons might be responsible for the hormonal stimulation of renal amino acid reabsorption:

- Dexamethasone induces the gene expression and might enhance the content of carrier molecules in the tubular cells (Yamamoto, 1985).
- Dexamethasone (Baum and Quigley, 1993) and T3 (Guernsey and Edelmann, 1984; Klein et al., 1984) enhances the activity of the Na^+/K^+-ATPase and increase the sodium gradient between tubular lumen and tubular cell. This might increase the highly sodium dependent amino acid reabsorption: e.g. the removal of leucine from the ultrafiltrate depends 80% on the sodium gradient (Yao et al., 1994).
- The potassium permeability at the basolateral membrane of the tubular cells is increased after T3 administration (De Nayer, 1987) and might cause an enhanced Na^+/K^+-ATPase activity, too.

- T3 is reported to increase the synthesis of proteins in the kidney (Capasso et al., 1987). This de-novo protein synthesis could include amino acid carrier proteins followed by an enhanced amino acid transport capacity in the tubular cell.
- Gordon et al. (1986) found an increased calcium uptake in heart cells after T3 treatment. If it also occurs in the kidney, it might activate adenylate cyclase activity via calmodulin, increase the cAMP content and enhance amino acid transport capacity. However, this is in contrast to Surks et al. (1984) postulating interactions between thyroid hormones and nuclear receptors as the reason for enhanced amino acid uptake.

One reason for the lack of effect of T3 in glutamine loaded animals might be the hormone effect on amino acid metabolism (Baxter, 1976). Possibly the actions of this catabolic hormone, resulting in distinct changes in amino acid plasma concentrations (not shown in detail), could mask the renal effects of T3 after glutamine, but not after leucine loading. Obviously the renal handling and the metabolism of these two amino acids is controlled by the two hormones in different ways. Because of enhanced metabolism within the renal tubular cells under hormone treatment the so-called house-keeping of the tubular cells requires higher amounts of amino acids followed by a nonspecifical increase in the cellular uptake of amino acids (Foulkes and Blanck, 1990). Therefore, the uptake of amino acids from the blood at the basolateral site could be enhanced after T3, too. This uptake is followed by an enhanced intracellular amino acid concentration and, therefore, by a reduced concentration gradient between tubular fluid and tubular cell. This might be the reason for a diminished driving force for amino acid reabsorption and might mask stimulatory T3 effects at the luminal site. This phenomenon reflects the overlapping between possible effects of T3 directly at the amino acid carrier sites and its catabolic action within the tubular cell.

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