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High thiamine diphosphate concentrations in erythrocytes can be achieved in dialysis patients by oral administration of benfotiamine

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Abstract *Objective*: The influence of either orally administered *S*-benzoylthiamine-*O*-monophosphate (benfotiamine) or thiamine nitrate on the thiamine status was tested in a randomised, two-group comparison study in 20 end-stage renal disease (ESRD) patients. Main outcome measures were the pharmacokinetics of thiamine diphosphate (TDP) in blood, the in vitro erythrocyte transketolase activity, its activation coefficient (α-ETK) and the TDP concentration in erythrocytes.
Methods: After ingestion of a single dose of either 100 mg thiamine nitrate (corresponding to 305 μmol thiamine) or 100 mg benfotiamine (corresponding to 214 μmol thiamine), the blood levels of thiamine phosphate esters were analysed by means of high-performance liquid chromatography for a 24-h period. The TDP concentration in erythrocytes was calculated using the haematocrit and TDP concentration in blood. Erythrocyte transketolase activity and α-ETK were measured before and 10 h after administration. The pharmacokinetics of TDP in blood were compared with healthy subjects of other studies retrieved from database query.
Results: Regarding the blood concentrations of TDP, the patients with ESRD had a 4.3 times higher area under the concentration–time curve after benfotiamine administration than after thiamine nitrate. After benfotiamine administration, the peak plasma concentration of TDP exceeded that in healthy subjects by 51%. In the ESRD patients, after 24 h, the mean TDP concentration in erythrocytes increased from 158.7 ± 30.9 ng/ml initially to 325.8 ± 50.9 ng/ml after administration of benfotiamine and from 166.2 ± 51.9 ng/ml to 200.5 ± 50.0 ng/ml after thiamine nitrate administration. The ratio between the maximum erythrocyte TDP concentration and basal concentration was 2.66 ± 0.6 in the benfotiamine group and 1.44 ± 0.2 in the group receiving thiamine nitrate ($P < 0.001$). After 24 h, it was 2.11 ± 0.4 and 1.23 ± 0.2, respectively. The transketolase activity increased from 3.54 ± 0.7 μkat/l initially to 3.84 ± 0.6 μkat/l after benfotiamine intake ($P = 0.02$) and from 3.71 ± 0.8 μkat/l to 4.02 ± 0.7 μkat/l after thiamine nitrate intake ($P = 0.08$). Likewise, α-ETK decreased from initially 1.10 ± 0.07 to 1.04 ± 0.04 ($P = 0.04$) and from 1.12 ± 0.05 to 1.08 ± 0.06 ($P = 0.09$). After 24 h, the phosphorylation ratio in whole blood decreased from 12.9 ± 6.9 initially to 5.6 ± 3.2 after benfotiamine administration ($P = 0.02$) and from 13.5 ± 7.3 to 9.0 ± 4.8 ($P = 0.03$) after administration of thiamine nitrate. No correlation between erythrocyte TDP concentration and transketolase activity and/or α-ETK was observed in ESRD patients, either before or 10 h after administration.
Conclusion: Compared with thiamine nitrate, the oral administration of benfotiamine leads to higher TDP concentrations in erythrocytes accompanied with a significant improvement of the erythrocyte transketolase activity in ESRD patients.

Key words Thiamine diphosphate · Transketolase · Benfotiamine

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Introduction

The total metabolic pool of thiamine (vitamin B_1) in the human body is about 30 mg, existing mainly as thiamine diphosphate (TDP) in addition to small amounts of free thiamine, thiamine monophosphate (TMP) and thiamine triphosphate (TTP). Eighty percent of the total thiamine content of blood is present in the erythrocytes, predominantly as TDP. Thus, it acts as an enzymatic

cofactor in oxidative decarboxylation reactions and as coenzyme in the vital transketolase reaction in the pentose phosphate shunt. In recent years, several studies showed that erythrocyte transketolase activity in patients with chronic renal insufficiency deviates from healthy subjects [1–4]. These investigations were of special interest when regarding possible connections between changes of the transketolase activity and the neuropathies often observed in uraemia [5, 6]. About 25% of all uraemic patients suffer from neuropathies [7].

Thiamine deficiency is measured by a decreased transketolase activity, which leads to a reduced glucose oxidising capacity of the pentose phosphate pathway. It may cause degeneration of the myelin sheaths (demyelinisation syndrome), which is accompanied by uraemic neuropathies. Clinical and electrophysiological aberrations in haemodialysis patients with peripheral neuropathies can be improved by the intravenous application of TDP, increasing the thiamine levels in plasma and erythrocytes as well as transketolase activity [8].

Mostly, active substances of commercially distributed neuropathy drugs contain either the water-soluble thiamine hydrochloride and thiamine mononitrate or the lipophilic thiamine prodrug *S*-benzoylthiamine-*O*-monophosphate (benfotiamine) [9]. In healthy people, the better bioavailability of oral benfotiamine ingestion was shown to lead to an elevation of blood TDP concentration [10–12]. Sparse information exists to date on the contribution of an oral administration of thiamine preparations to the formation of TDP, especially in patients with end-stage renal disease (ESRD) undergoing regular haemodialysis treatment.

The aim of this study was to investigate the pharmacokinetics of TDP after oral administration of hydrophilic and lipophilic thiamine preparations in healthy subjects and patients with ESRD. Additionally, the in vitro erythrocyte transketolase activity, its activation coefficient (α-ETK) and the TDP concentration in erythrocytes were determined in ESRD patients.

Material and methods

Study design and subjects

Twenty patients with ESRD (17 males, 3 females; 7 (35%) with anuria, see Table 1) undergoing haemodialysis three times a week were randomly separated into two groups. Two commercial thiamine preparations containing 100 mg benfotiamine (corresponding to 214 μmol thiamine) per capsule (“Milgamma 100”, Woerwag Pharma, Germany) or 100 mg thiamine nitrate (corresponding to 305 μmol thiamine) per tablet (“Neurotrat S forte”, Knoll, Germany) were administered in a blinded fashion. After an overnight fasting period, each group of these volunteers received a single oral dose of either 100 mg benfotiamine or 100 mg thiamine nitrate with 100 ml water. Blood samples were taken by venipuncture into heparinised tubes immediately before administration at 0800 hours and 0.5, 0.75, 1, 1.5, 2, 3, 4, 6, 8, 10 and 24 h afterwards. The patients were not subjected to dialysis during this time. The plasma was separated by centrifugation, and the erythrocytes were washed three times with an equal volume of normal saline. The samples were stored at −80 °C until analysed.

Table 1 Anthropometric characteristics, clinical and laboratory data of the patients with end-stage renal disease (ESRD)

	Median	Range
Age (years)	56.5	38.0–64.0
Body mass index (kg/m^2)	25.8	22.0–30.3
Duration of ESRD (months)	32.0	6.0–126.0
Duration of dialysis therapy (months)	24.0	6.0–124.0
Serum urea (mmol/l)	28.2	17.9–39.5
Serum creatinine (μmol/l)	919.1	459.7–1116.6
Serum protein (g/l)	70.6	61.2–81.0
Albumin (g/l)	42.8	40.6–44.8
Protein excretion (g/24 h)	0.13	0.0–2.6
Haematocrit (%)	31.0	27.0–42.0
Haemoglobin (mmol/l)	6.8	5.9–8.8
Systolic blood pressure (mmHg)	135.0	97.0–191.0
Diastolic blood pressure (mmHg)	82.0	49.0–99.0

The in vitro transketolase activity and its activation coefficient (α-ETK) were measured immediately before and 10 h after administration of the test preparations. Analytical procedures for the determination of TDP and the assessment of the transketolase activity and α-ETK are described in detail elsewhere [13].

For comparison purposes, a database query (Medline and Dissertation Abstracts Online), using the key words transketolase, thiamine diphosphate, thiamine, HPLC and benfotiamine, was performed to find studies carried out in healthy subjects, which reported of detailed TDP concentrations in blood.

All patients gave their written informed consent to participate in the study which was approved by the university’s ethics committee.

Pharmacokinetic parameters and statistics

From the individual profile of blood TDP concentration, the following pharmacokinetic parameters were assessed using non-compartmental analysis: relative peak blood concentration (C_{max}), i.e. using subtracted base levels, and the time to reach peak concentration (t_{max}) were derived directly from the blood concentration profiles; the area under the concentration–time curve (AUC_{0-24h}) was calculated using the linear trapezoidal rule from t_0 to t_{24h}. All pharmacokinetic parameters were calculated using the Topfit 2.0 program (Gustav Fischer Verlag, Stuttgart, 1993).

In order to evaluate the effectiveness of the two preparations on the thiamine status in the blood of ESRD patients, the erythrocyte TDP concentration, transketolase activity, α-ETK and the phosphorylation ratio of total thiamine were chosen as response criteria. The phosphorylation ratio expresses the proportional amount of phosphorylated thiamine in whole blood and was calculated from the ratio between the phosphate esters and the free thiamine (TTP + TDP + TMP/T). Erythrocyte TDP concentration was determined using the haematocrit before administration, at t_{max} and 10 h and 24 h after administration according to Eqn 1:

$$y_E = \frac{x_B - x_P\left(\frac{1-Hc}{100}\right)}{\left(\frac{Hc}{100}\right)} \quad (1)$$

where y_E = TDP concentration in erythrocytes, x_B = TDP concentration in whole blood, x_P = TDP concentration in plasma (ng/ml) and Hc = haematocrit (%) [14].

Differences of means between the two groups were tested using analysis of variance (one-way ANOVA). Period effects were tested using two-way ANOVA followed by pairwise comparison, using the non-parametric Wilcoxon signed-rank test. The comparison of data between the different studies was made using the Kruskal-Wallis-H test. Bivariate correlations were computed using the Pearson correlation coefficient. Complete analysis of data was performed with the aid of the statistical software package SPSS 8.0

Table 2 Pharmacokinetic parameters of thiamine diphosphate (TDP) in whole blood after administration of various preparations and doses of thiamine in patients with end-stage renal disease (bold letters) and healthy subjects (mean and 95% confidence interval)

Study reference	*n*	Preparation	Dose	AUC (h · ng/ml)	C_{max} (ng/ml)[d]	t_{max} (h)
This study	10	Benfotiamine	100 mg p.o.[f]	**1491.2** (1141, 1840)[a]	**81.9** (66, 97)[a]	**7.6** (6, 9)[a]
This study	10	Thiamine nitrate	100 mg p.o.[g]	**355.1** (260, 450)[b]	**21.3** (17, 26)[b]	**8.3** (6, 10)[a]
[15]	10	Benfotiamine	250 mg p.o.	974.3 (709, 1239)[c]	54.2 (38, 70)[c]	5.2 (4, 6)[a]
[16][e]	6	Thiamine hydrochloride	50 mg i.v.	460.8 (262, 660)[b]	57.4 (31, 84)[a,c]	7.7 (5, 10)[b]
[16][e]	6	Thiamine hydrochloride	50 mg p.o.	355.8 (10, 702)[b]	26.8 (6, 47)[b,c]	18.2 (8, 28)[c]

AUC: [a] versus [b]: $P < 0.001$, [a] versus [c]: $P < 0.05$, [b] versus [c]: $P < 0.01$; C_{max}: [a] versus [b]: $P < 0.001$, [a] versus [c]: $P < 0.05$, [b] versus [c]: $P < 0.01$; t_{max}: [a] versus [b]: $P < 0.05$; [a] versus [c]: $P < 0.05$; [b] versus [c]: $P < 0.05$; as calculated using pairwise Mann-Whitney U-test

[d] Basal concentrations were subtracted
[e] Individual concentrations were obtained from graphics
[f] Corresponding to 214 μmol thiamine
[g] Corresponding to 305 μmol thiamine

(SPSS Inc., Chicago, Ill., 1997), using the general linear model (GLM) procedure for two-way ANOVA. Statistical significance was considered if the probability associated with F was < 0.05.

Results

Pharmacokinetics of TDP in the blood of patients with ESRD and in healthy subjects

The patients with ESRD had a 4.3 times higher mean AUC after benfotiamine administration than after the same dose of thiamine nitrate (Table 2). However, if the healthy subjects in the study by Ziems [15] received a 2.5-fold higher benfotiamine dose, the AUC reached only 65% of the values observed in ESRD patients. Identical AUCs were observed both after oral administration of thiamine nitrate in ESRD patients and after administration of half the dose of thiamine hydrochloride in healthy subjects [16]. After administration of benfotiamine, C_{max} of TDP in ESRD patients exceeded that in healthy subjects by 51%. After oral administration of benfotiamine or thiamine nitrate, t_{max} of TDP in whole blood was not significantly different between ESRD patients and healthy subjects.

Erythrocyte TDP concentration, phosphorylation ratio and transketolase activity in patients with ESRD

The results of the high-performance liquid chromatography (HPLC) measurements and the transketolase assay are summarised in Table 3. The initial mean erythrocyte TDP concentration in the patients with ESRD was 162.4 ± 41.7 ng/ml (range 107.7–235.5 ng/ml). After t_{max}, 10 h and 24 h, significant differences were found in the mean erythrocyte TDP concentration between the groups. Both preparations resulted in maximum erythrocyte TDP concentrations which were

Table 3 Concentration of thiamine diphosphate (TDP) in erythrocytes, transketolase activity, activity coefficient of erythrocyte transketolase activity (α-ETK), and phosphorylation ratio (ratio between thiamine phosphate esters and free thiamine in whole blood) in patients with end-stage renal failure ($n = 20$) after oral administration of 100 mg benfotiamine (corresponding to 214 μmol thiamine) or 100 mg (305 μmol thiamine) thiamine nitrate (mean and 95% confidence interval)

	Preparation		Group comparison (ANOVA)	
	Benfotiamine	Thiamine nitrate	*F*	Two-tailed *P* value
Before administration				
Erythrocyte TDP (ng/ml)	158.7 (136.6, 180.8)[a]	166.2 (129.1, 203.3)[a]	0.153	0.700
Transketolase activity (μkat/l)	3.54 (3.06, 4.03)[a]	3.71 (3.13, 4.29)[a]	0.25	0.623
α-ETK	1.10 (1.05, 1.15)[a]	1.12 (1.08, 1.15)[a]	0.314	0.582
Phosphorylation ratio	12.9 (8.1, 17.9)[a]	13.5 (8.3, 18.7)[a]	0.034	0.856
10 h after administration				
Erythrocyte TDP (ng/ml)	383.1 (341.8, 424.3)[b]	227.1 (188.0, 266.2)[b]	38.57	<0.001
Transketolase activity (μkat/l)	3.84 (3.38, 4.29)[b]	4.02 (3.54, 4.50)[a]	0.394	0.538
α-ETK	1.04 (1.01, 1.07)[b]	1.08 (1.03, 1.12)[a]	2.885	0.107
Phosphorylation ratio	3.2 (2.0, 4.3)[b]	6.9 (5.3, 8.5)[b]	18.43	<0.001
t_{max} of Erythrocyte TDP (h)	7.6 (6.0, 9.2)	8.3 (6.4, 10.2)	0.399	0.535
Erythrocyte TDP (ng/ml)	408.8 (377.7, 439.9)[c]	231.8 (195.7, 267.9)[b]	70.6	<0.001
24 h after administration				
Erythrocyte TDP (ng/ml)	325.8 (289.3, 362.2)[d]	200.5 (164.4, 236.6)[c]	30.51	<0.001
Phosphorylation ratio	5.6 (3.3, 7.8)[c]	9.0 (5.6, 12.4)[b]	3.6	0.074

[a] versus [b]: $P < 0.001$, [a] versus [c]: $P < 0.05$, [b] versus [c]: $P < 0.01$; as calculated using two-way ANOVA with pairwise comparison (Wilcoxon test)

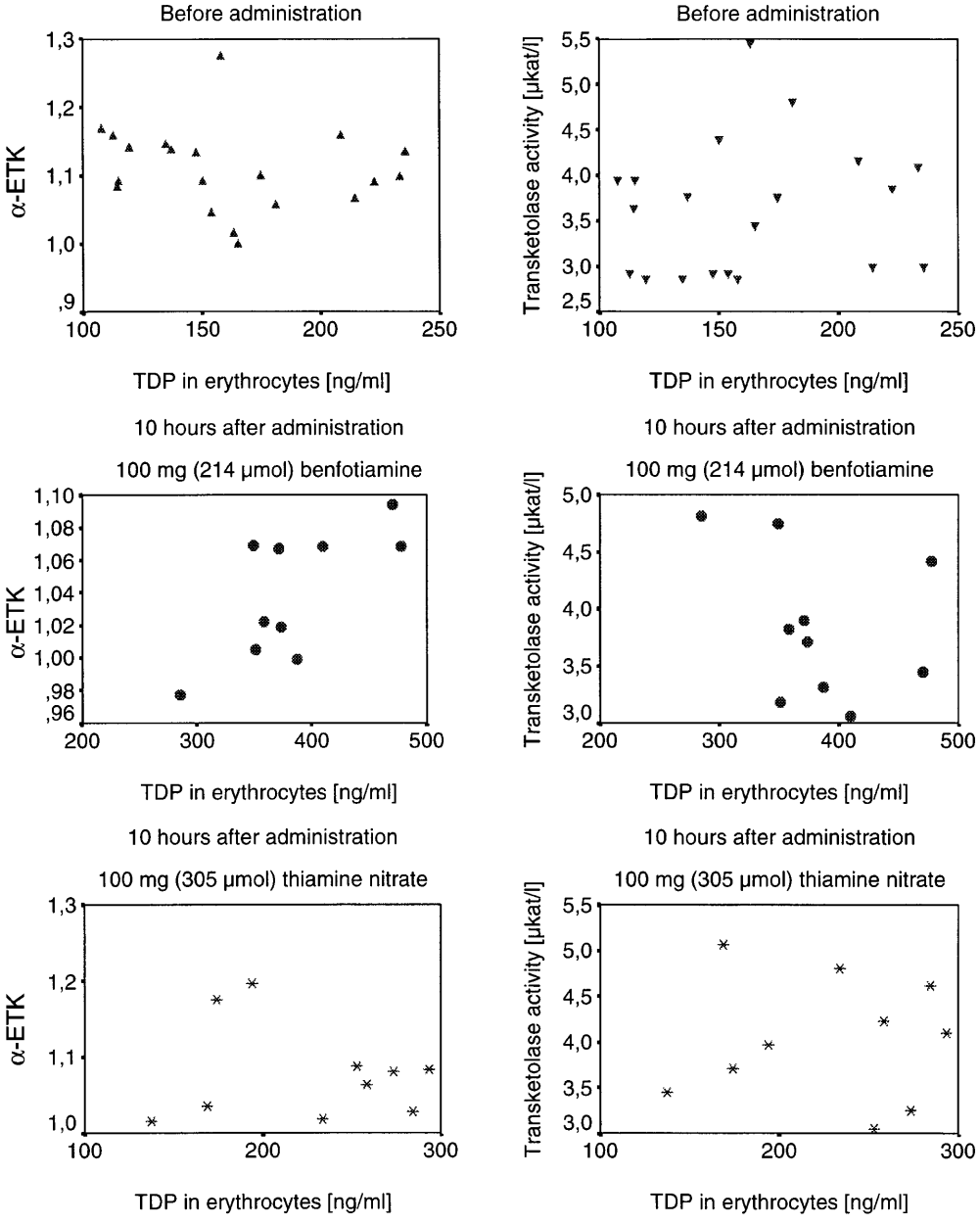

Fig. 1 No correlation of thiamine diphosphate (TDP) in erythrocytes with erythrocyte transketolase activity and/or its activation coefficient (α-ETK) before ($n = 20$) and 10 h after ($n = 10$) oral administration of various thiamine preparations in patients with end-stage renal disease

observed 7.9 h after administration. The ratio between the maximum erythrocyte TDP concentration and base level was 2.66 ± 0.6 in the benfotiamine group and 1.44 ± 0.2 in the group receiving thiamine nitrate (mean ± SD, $P < 0.001$). After 24 h, the ratio between erythrocyte TDP concentration and base level had slightly diminished to 2.11 ± 0.4 and 1.23 ± 0.2, respectively ($P < 0.001$). However, benfotiamine administration resulted in 77% higher maximum erythrocyte TDP concentrations than thiamine nitrate. The phosphorylation ratio decreased from 13.2 ± 6.9 initially to 7.3 ± 4.3 after 24 h and was identical in both groups before and after administration.

Before administration of the test preparations, only 1 of the 20 patients was in a thiamine-deficient state as indicated by an α-ETK >1.25 [17]. Nineteen of twenty patients had an α-ETK <1.20. Only after benfotiamine administration was transketolase activity increased significantly connected with a decrease in α-ETK. Neither the duration of renal insufficiency, the duration of haemodialysis treatment nor the age correlated with the transketolase activity and/or α-ETK. Correlations between erythrocyte TDP concentration and transketolase activity and/or α-ETK could not be observed, either initially or 10 h after administration (Fig. 1). Moreover, there was no correlation between the phosphorylation ratio and transketolase activity and/or α-ETK, either before or 10 h after benfotiamine administration. Only in the group receiving thiamine nitrate was a correlation between the phosphorylation ratio and transketolase activity observed 10 h after administration ($r = -0.64$, $P = 0.048$). Before administration, a positive correla-

tion of transketolase activity with the total serum protein ($r = 0.67$, $P = 0.001$) and an inverse correlation with the albumin concentration ($r = -0.58$, $P = 0.007$) was observed. Furthermore, α-ETK correlated positively with the albumin concentration ($r = 0.5$, $P = 0.02$). There was no correlation between the haemoglobin concentration and transketolase activity and/or α-ETK.

Discussion

Obviously, the TDP formation after application of lipid- and water-soluble thiamine derivatives occurred to a much higher extent in ESRD patients than in healthy subjects. Remarkably, the ingestion of smaller oral doses of benfotiamine lead to a much higher AUC and C_{max} in the patients than in healthy volunteers. With half the oral dose of thiamine hydrochloride in healthy people, identical AUC and C_{max} values of TDP were achieved as in the patients who received thiamine nitrate. Besides impaired renal excretion, differences in the oral bioavailability and inter-individual differences in the thiamine status may be the main reasons.

The chronic renal insufficiency is characterised by complex hormonal, biochemical and metabolic alterations. Therefore, the results should be interpreted with caution when comparing patients with healthy controls, because corresponding studies are rare, and mode of application and dose range as well as the basic thiamine status varied. A previous publication showed that, in the case of a severely diminished renal excretion, the TDP formation serves as an alternative way of achieving a new steady state of the thiamine blood level. This may partly explain the higher TDP levels [13].

Regarding the initial erythrocyte TDP concentrations, the patients with ESRD under investigation showed an adequate thiamine status before the thiamine preparations were given. Normal values of erythrocyte TDP concentrations in healthy subjects are within a range of 92–224 ng/ml [18, 19]. A marginal thiamine deficiency state is suggested with erythrocyte TDP concentrations ranging from 29.5 ng/ml to 38.5 ng/ml; an erythrocyte TDP concentration below 29.5 ng/ml is regarded as thiamine deficient [17]. The initial erythrocyte TDP concentration in patients with ESRD ranged between 107.7 ng/ml and 235.5 ng/ml and complies with the reference range. Several cross sectional investigations could demonstrate that patients with chronic renal insufficiency are able to maintain normal plasma- and erythrocyte thiamine concentrations without any thiamine supplementation [3, 4, 20, 21]. This was also confirmed in long-term studies. The mean erythrocyte TDP concentration in 15 dialysis patients was even 350 ng/ml and did not decrease below the reference range after a 6-month dialysis treatment without thiamine supplementation [22, 23]. In 13 anaemic haemodialysis patients, however, erythrocyte thiamine concentrations reached only 52.8 ng/ml, but did not significantly differ from those of healthy controls [4]. In patients with signs of peripheral neuropathy receiving 5 mg/kg TDP i.v. three times per week during 6 months of haemodialysis, the mean erythrocyte TDP concentration rose from 42.5 ng/ml to 153 ng/ml, equivalent to an increase by 260% [8]. In this study, erythrocyte TDP concentrations increased by 170% after administration of benfotiamine and by 40% after thiamine nitrate.

The phosphorylation ratio in whole blood may serve as an additional criterion of thiamine status which is independent of base levels as shown in alcoholics [24]. Accordingly, a decreased phosphorylation ratio expresses a dysfunction of thiamine-dependent enzymes. However, a decreased P phosphorylation ratio is a first consequence of either elevated free thiamine, decreased TDP, or both. In this investigation, the initial phosphorylation ratio was three times higher than that in alcoholics studied by Tallaksen et al. [24]. This may be caused by lower concentrations of free thiamine concentrations in our patients. After 24 h, the phosphorylation ratio after benfotiamine was as high as that after administration of thiamine nitrate. The lacking correlation between the phosphorylation ratio and the erythrocyte TDP concentration, transketolase activity and/or α-ETK does not support a causal relationship of the function of thiamine-dependent enzymes and the phosphorylation ratio. However, the negative correlation between transketolase activity and the phosphorylation ratio 10 h after administration of thiamine nitrate is a surprising finding.

In accordance with the measurements of erythrocyte TDP, the initial transketolase activity and/or α-ETK in patients with ESRD indicate an adequate thiamine status. As pointed out in Fig. 1, the transketolase is saturated with its cofactor but shows individual differences. However, some discrepancies appeared between transketolase activity and α-ETK when comparing them with other studies. While the mean α-ETK of 1.11 ± 0.06 in this investigation resembles the mean normal value of 1.10 obtained in a German sample of 1953 healthy persons, the transketolase activity exceeded the mean normal value more than twice (3.62 μkat/l vs 1.5 μkat/l) [25].

The transketolase activity under conditions of chronic renal insufficiency has been discussed controversially. On the one hand, a low activity was explained as a result of an accumulation of low-molecular-weight uraemic toxins in plasma [5, 26]. Pietrzak and Baczyk [4], for instance, found a 35% lowered activity in patients with chronic renal insufficiency and a 18% lowered activity in patients with haemodialysis treatment compared with healthy persons. In contrast to this, a normal or slightly enhanced activity obtained from other investigations is discussed as a result of a rejuvenated erythrocyte cell population following recombinant human erythropoietin therapy as well as due to a formation of non-specific metabolic factors induced by renal insufficiency [1, 2, 27]. Nevertheless, absolute values of

transketolase activity are of minor evidence owing to the considerable variations of given reference values by several authors. It is also noteworthy, that the transketolase activity is often taken as an arbitrary unit and is highly dependent on technical details. We calculated our values referring to a haematocrit of 1.0. However, the values in the German VERA-sample are related to a more diluted erythrocyte suspension (Hc < 1.0). In addition, there is a high-affinity binding between TDP and apo-transketolase. Thus, transketolase activity is not significantly decreased until the levels of erythrocyte TDP have dropped to less than 20% of the initial values [28]. Therefore, transketolase activity and its activation coefficient are thought to be a less sensitive index of thiamine status than HPLC measurements of the erythrocyte TDP concentration [29].

It is generally agreed that the basal in vitro transketolase activity has to be interpreted at least together with α-ETK. Furthermore, it has to be considered that the synthesis of the apoenzyme may be impaired, and α-ETK, therefore, does not reflect correctly the thiamine status. Kopple et al. [27] found a mean α-ETK of 1.08 in 16 uraemic patients. In the case of 14 patients with chronic renal insufficiency, Mestyan et al. [2] found a mean activation coefficient of 1.20 compared with 1.15 in 16 healthy controls. In a sample of 73 chronic renal-insufficiency patients with and without neuropathies, however, a significantly lowered mean α-ETK of 1.06 compared with 1.13 in 67 healthy controls was detected [6]. In former investigations with dialysis patients of the university hospital in Jena, an oral supplementation with either 1.5 mg or 8.0 mg thiamine hydrochloride three times per week had no influence on the mean α-ETK which was 1.11 ± 0.1 ($n = 15$) and/or 1.19 ± 0.1 ($n = 9$) (unpublished observations). Changes of transketolase activity and α-ETK in healthy persons after supplementation with either benfotiamine or thiamine nitrate were not consistent, as was demonstrated in previous studies [10, 29]. While water-soluble thiamine salts never did have any effect, α-ETK was either improved and transketolase activity not effected or vice versa after benfotiamine ingestion. Although the effect of a thiamine supplementation on transketolase activity shows conflicting results, our findings are in agreement with a previous report showing a significant influence on transketolase activity only in the case of benfotiamine administration [10].

The correlation of transketolase activity and/or α-ETK with erythrocyte TDP as well as other parameters differs between various studies. Warnock et al. [28] found a correlation between erythrocyte TDP and transketolase activity of $r = 0.6$ in 11 healthy men. Takeuchi et al. [30] found that the TDP concentration in haemolysate correlated highly positively with transketolase activity ($r = 0.88$). A better correlation between erythrocyte TDP and transketolase activity than between erythrocyte TDP and α-ETK could be observed by Baines and Davies [31]. In addition, Saito et al. [32] observed a correlation between the total thiamine concentration in whole blood and transketolase activity ($r = 0.97$) and/or α-ETK ($r = -0.525$) in outpatient diabetics. Pietrzak and Baczyk [4] found even a correlation between the total protein in serum and erythrocyte transketolase activity as well as between the haemoglobin concentration and transketolase activity in 25 patients with ESRD, suggesting a strong influence of nutritional status on apo-transketolase. We could not confirm these results in the present investigation. Due to a lacking healthy control group in our study, a possible relation of optimal transketolase activities to the severity of the renal symptoms could not be accomplished. However, we found that the only influencing factors of transketolase activity in 60 healthy subjects were the thiamine concentration in blood ($r = 0.41$, $P = 0.001$) and the albumin concentration ($r = 0.33$, $P = 0.013$) (unpublished results).

Our findings have some importance regarding the protective effects of TDP in preventing the formation of toxic and immunogenic advanced glycation end products (AGEs). AGEs may be a major contributor to the pathological manifestations of diabetes mellitus, which is the most important precursor of chronic renal failure. According to in vitro studies, TDP may have a novel therapeutic potential in preventing vascular complications of diabetes with its consequences for kidney function [33, 34]. Thus, the present investigation examined for the first time the influence of an oral application of high-dose thiamine preparations on the enhancement of erythrocyte TDP as well as transketolase activity. In this respect, the administration of benfotiamine was much more effective than thiamine nitrate, although there was obviously no correlation between erythrocyte TDP concentrations and transketolase activity and/or α-ETK. These results are in agreement with our former studies of healthy volunteers showing a significant improvement in the thiamine status only after benfotiamine administration. Further investigations should clarify whether the improvement of clinical signs of neuropathies accompanied with chronic renal insufficiency could be achieved by oral supplementation with thiamine preparations.

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