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Cefetamet pivoxil treatment causes loss of carnitine reserves that can be prevented by exogenous carnitine administration

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Two groups of pediatric patients receiving cefetamet pivoxil treatment (3 × 500 mg daily) for 7 days were studied. In the first group (Group A) the drug was administered alone; in the second group (Group B) the drug was given in combination with a molar excess of carnitine (3 × 1 g). Medication with cefetamet pivoxil alone was associated with a large urinary excretion of pivaloylcarnitine: Approximately 71% of the daily pivalate intake could be eliminated as carnitine ester in the urine. In this group, the plasma level and the urinary output of free carnitine decreased. By contrast, in Group B, the administration of molar excess of carnitine aided stochiometric elimination of pivalate as carnitine ester, and the plasma levels and carnitine-free urinary output were unchanged. The data show that medication of cefetamet pivoxil results in the formation of pivaloylcarnitine in children; the sustained loss of carnitine esters can ultimately lead to carnitine deficiency. Molar excess of exogenous carnitine aids in the elimination of pivalate derived from cefetamet pivoxil therapy and helps to maintain the carnitine reserves. (J. Nutr. Biochem. 10:670–673, 1999)

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Introduction

Carnitine forms esters with several short-, medium-, and long-chain fatty acids by enzymatic transfer of the acyl moiety from coenzyme A to the hydroxyl group of carnitine.[1,2] In metabolism the esterification of long-chain fatty acids for transport across the mitochondrial membrane is essential for the β-oxidation of long-chain fatty acids by mitochondria.[1,3]

In addition to physiologic fatty acids, carnitine can form esters with several fatty acids that are derived from nonmetabolizable drugs and xenobiotic acids,[2,4–7] which are excreted in the urine. An example is pivalic acid (trimethylacetic acid). It is widely used in therapeutics because its nonpolar character can enhance the enteral absorption of a parent drug. In several drugs pivalate is attached to the prodrugs as pivaloxyl-oxymethylesters (pivampicillin, pivmecillinam). In humans, pivalate released from the drug readily forms an ester with carnitine.[4,5,8] The sustained loss of carnitine as pivaloylcarnitine can deplete carnitine reserves.[7,10] This loss causes carnitine insufficiency or deficiency, which can be associated with serious metabolic consequences.[4,5,7–12]

We studied the possible carnitine ester-forming effect of cefetamet pivoxyl, a third generation oral cephalosporin that is effective in the therapy of several types of infections.[13–15] Because the structure of the parent drug differs from drugs

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Table 1 Plasma levels and urinary output of free carnitine and acid soluble carnitine esters in the two groups of cefetamet pivoxil-treated patients

	Group A		Group B	
	Day 0	Day 7	Day 0	Day 7
Plasma (μmol/L)				
Acid soluble total	42.6 ± 3.11	27.7 ± 1.78[a]	41.7 ± 2.92	54.6 ± 3.17[ab]
Free	32.7 ± 2.87	11.9 ± 1.36[a]	33.6 ± 3.02	32.2 ± 2.90[b]
Acid soluble acyl	9.90 ± 1.27	15.8 ± 1.55	8.10 ± 0.98	22.4 ± 3.79[ab]
Urine (μmol/day)				
Total	295.7 ± 34.4	2131 ± 326[a]	324.4 ± 68.1	5058 ± 487[ab]
Free	123.8 ± 26.6	30.2 ± 6.33[a]	103.7 ± 22.3	940.1 ± 211[ab]
Total acyl	171.9 ± 26.0	2101 ± 318[a]	220.7 ± 50.8	4118 ± 412[ab]

Values are means ± SEM; $n = 7$ subjects (4 males, 3 females) in both groups. values with superscript letters are significantly different ($P < 0.05$) from: [a] day 0 in each corresponding group and [b] from day 7 in Group A.

studied previously, the extent of pivalate liberation could not be predicted. Previous results showed that the exogenous carnitine was readily used as a substrate for pivaloylcarnitine synthesis in the case of pivampicillin.[5,12] Therefore, we studied the influence of the exogenous carnitine on the elimination of pivalate in a group of children treated with cefetamet pivoxil.

Patients and methods

Patients and treatments

A total of 14 pediatric patients (8 males, 6 females) participated in this study. Clinical indications for the cefetamet pivoxil treatment included bacteriuria ($n = 6$) or pharyngeal colonization of drug-sensitive bacteria ($n = 8$). Generally, patients were in good condition and had no history of severe disease or metabolic disorders. The patients were studied in two groups. In Group A ($n = 7$; 4 males, 3 females), the average age of the patients was 15.2 years (range 14–17 years); in Group B ($n = 7$; 4 males, 3 females), the average age was 14.8 years (range 13–17 years). Average body weight was 59.3 kg (range 28–60 kg) in Group A and 55.6 kg (37–68 kg) in Group B. The study period lasted 8 days: After a control day (day 0) the patients received cefetamet pivoxil (Roche, Basel, Switzerland) treatment for 7 days. In Group A the drug was administered alone and in Group B it was administered in combination with oral L-carnitine solution (Sigma-Tau Pharmaceuticals, Rome/Pomezia, Italy). The daily dose of cefetamet pivoxil was 3 × 500 mg (each 500 mg tablet contains 0.98 mmoles of pivalate) and the dose of administered carnitine was 3 × 1 g (in molar terms 1 g oral solution of carnitine corresponds to 6.2 mmoles of L-carnitine).

Procedures

Between 8:00 and 9:00 AM, 24-hour urine samples were collected for both groups before the introduction of the treatment (day 0) and on the seventh (day 7) day of the administration of the antibiotics. The completeness of the urine collections were assessed by urinary creatinine determinations. Blood samples also were taken between 8:00 and 9:00 AM after an overnight fast. The plasma and urine samples were frozen immediately and stored at −70°C. The study design was approved by the local ethics committee, and informed consent was obtained from the parents of all study participants.

Methods

Urinary creatinine levels were measured by routine picric acid method.[12] Plasma and urinary acid soluble carnitine levels were measured by a radiochemical method.[12] Pivaloylcarnitine was identified in the urine by fast-atom bombardment mass spectrometry as previously described.[11,16] Essentially, after the addition of the internal standard (homocarnitine), 5 mL aliquots of the urine samples were purified using ion exchange resins (Dowex 50 W X or AG 501 X 8 from Biorad, Richmond, CA USA), and the effluents were dried under nitrogen flow as described.[11] The dried materials were suspended in 10 μL water and were analyzed with a Jeol JMS HX 110 (Jeol USA Ltd., Peabody, MA USA) apparatus as described.[11] Pivalate in the urinary pivaloylcarnitine was also quantitated by gas chromatography.[5,12] For within-group comparisons the Student's paired *t*-test was used; for group-group statistics the unpaired Student's *t*-test was employed. A *P*-value of less than 0.05 was considered statistically significant.

Results

In Group A the plasma levels of acid soluble total and free carnitine decreased by day 7 of cefetamet pivoxil administration (*Table 1*). The level of short-chain carnitine esters in the plasma increased the effect of treatment, probably due to the presence of synthesized pivaloylcarnitine.[5] The volume of the urine was not different prior to and on the last day the drug administration; the daily creatinine output did not differ on these days (not shown). The urinary output of carnitine esters increased 12-fold on day 7 of the cefetamet pivoxil administration; this was accompanied by a 4-fold decrease in the urinary output of free carnitine.

In Group B administration of combined cefetamet pivoxil and molar excess of carnitine led to a 18.6-fold increase in the urinary carnitine ester output, which is an excretion rate approximately 2-fold higher than that of Group A (*Table 1*). The plasma level of carnitine esters also increased in Group B compared with the level found in Group A on day 7. The plasma level of free carnitine did not change, and the output of free carnitine increased considerably (9-fold) compared with the pretreatment value in the same group.

In urine the presence of the pivaloylcarnitine was identified by fast atom bombardment mass spectrometry (see the signal for the protonated molecule at m/z 246 in *Figure 1*). In Group A approximately 97% of total carnitine esters was pivaloylcarnitine, as determined by gas chromatography, whereas in Group B, approximately 74% of carnitine esters was pivaloylcarnitine.

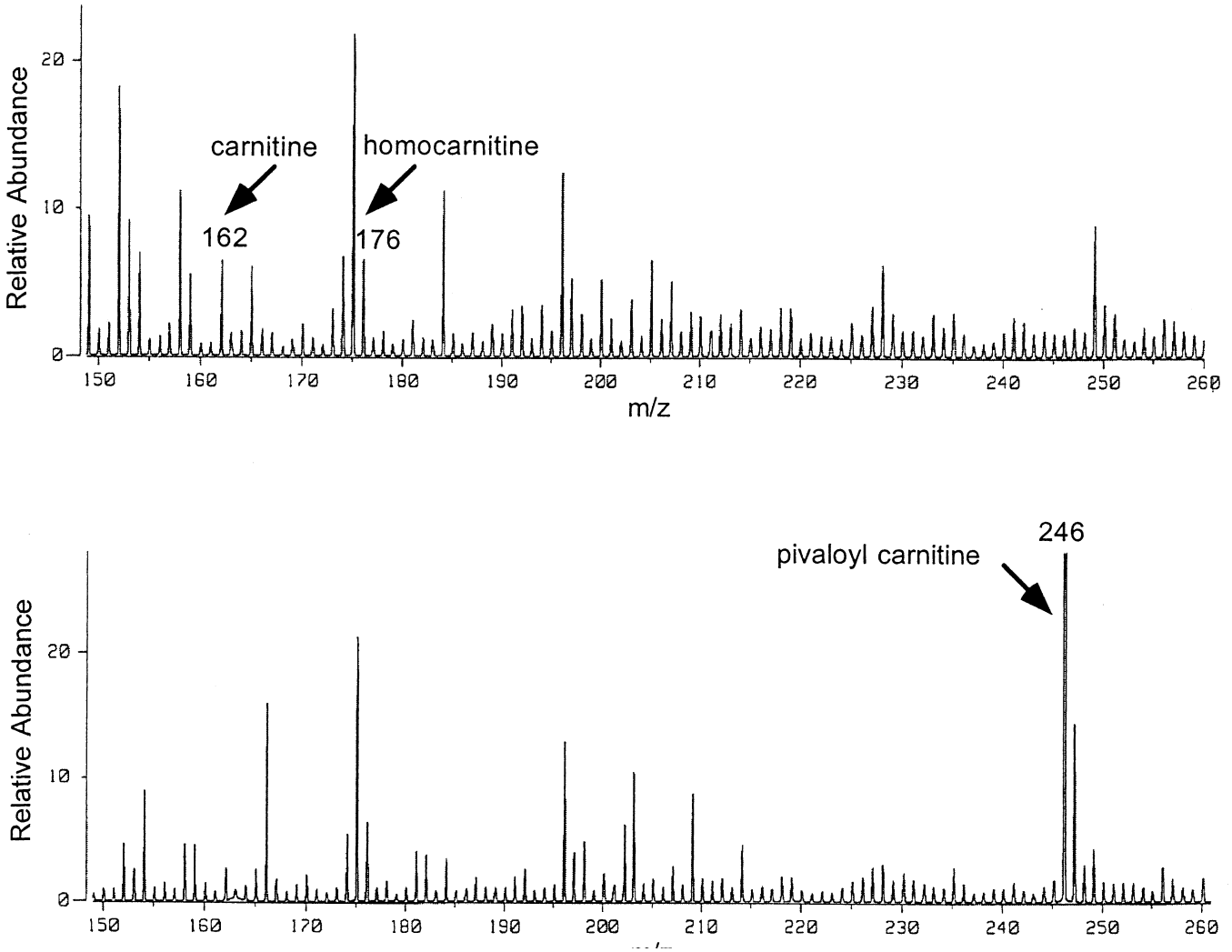

Figure 1 Representative fast atom bombardment mass spectrometry spectra of urine samples from a patient in Group A. Compared with the control (upper spectrum), pivaloylcarnitine in the sample gave a marked signal at 246 m/z from day 7 (lower spectrum). The signal of free carnitine was m/z 162; homocarnitine was used as internal standard (m/z 176).

Discussion

Pivalate or other ω-carbon trimethyl-containing moieties can be covalently attached to a parent drug with different binding methods. When pivalate is released from drugs that contain it as pivaloyloxymethyl ester (pivampicillin, pivmecillinam), it readily esterifies with carnitine.[4,7,17] The rate of release of pivalate from the prodrugs can be influenced by the structure of the parent drug, because the molecular geometry can seriously affect the binding characteristics to the hydrolases. Therefore, one cannot predict a priori whether a pivalate-containing drug will alter carnitine status in humans.

Such considerations prompted us to study the pivaloylcarnitine-generating capacity of cefetamet pivoxyl, which is the pivalate-containing third generation cephalosporine, in pediatric patients. Previously, an antibiotic agent (S 1108) that has a similar chemical structure to cefetamet pivoxil was studied in adults.[18,19] Compound S 1108 differs from cefetamet pivoxyl in two side chains; moreover, the dose range used in adults was only 300 to 600 mg. In pediatric patients the recommended dose range of cefetamet pivoxil is significantly higher, 10 to 40 mg/kg/day range.[13,15] It should be emphasized that this third generation oral cephalosporin is effective in the treatment of many types of pediatric infectious diseases.[13,15] In an internationally conducted multicenter study involving eight European countries, the drug was shown to be very effective against Gram-negative community-acquired pathogenic bacteria.[14]

In the present study the administration of cefetamet pivoxil caused excessive pivaloylcarnitine formation: Approximately 71% of the pivalate ingested was excreted as pivaloylcarnitine. The remainder of pivalate was either eliminated from the body by other detoxification processes or retained at least in part in the tissues by a mechanism similar to that for pivampicillin.[4,17] Because previous studies with pivampicillin showed that equal molar carnitine is insufficient to eliminate all of the pivalate, probably due to the bacterial degradation of carnitine in the gastrointestinal

tract,[5,12] a sixfold molar excess of carnitine was administered in the present study. The molar excess carnitine facilitated the stochiometric elimination of the pivaloyl moiety as carnitine ester from the organism in Group B. Moreover, the plasma free carnitine level and the urinary free carnitine output demonstrated that the carnitine administration helped to maintain the carnitine reserves.

The data from this study demonstrate that, despite the fundamental structural differences between the parent drugs, cefetamet pivoxil also promotes the excretion of pivaloylcarnitine in humans. This process can ultimately result in decreases in body carnitine reserves similar to other pivalate containing drugs.[4,9,17] A metabolic consequence of the administration of the pivalate-containing drug pivampicillin is altered metabolic fuel consumption[8] and lipid deposition in human quadriceps muscle.[9] The decrease in fat utilization and the compensatory increase of carbohydrate oxidation is considered a short-term consequence of treatment with pivampicillin.[8] These effects could be caused by the pivaloyl moiety that occurs because of carnitine depletion.[8,20] Thus, caution should be exercised with use of the cefetamet pivoxil, even if it is clinically well tolerated, especially in children with metabolic diseases or heart complications associated with decreased carnitine reserves.[21–23] Administration of carnitine should be considered to prevent the above mentioned adverse effects and to promote the elimination of pivalate from the body.

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