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Lowering Blood Urea Nitrogen with Amino Acid Supplementation

Keyphrases □ Amino acids—dietary supplementation, effects on blood urea nitrogen, rats □ Colorimetry—analysis of blood urea nitrogen in rat serum, effects of dietary supplementation of amino acids □ Dietary supplements—amino acids, effects on blood urea nitrogen in rats

To the Editor:

Currently we are studying the effect of dietary supplementation of amino acids on alcohol metabolism. The amino acid augmentation is based on the fasting plasma profile theory (1–4).

From past work on the metabolic effects of such diets, it was found that they produce a significant decrease in serum cholesterol in rats (5) and humans (6). This observed effect was theorized to be the result of an increase in net protein utilization.

While conducting the present study, we discovered that dietary supplementation of limiting amino acids to the third level (L-lysine, L-tryptophan, and L-threonine), based on the fasting plasma profile theory, produces a significant decrease in serum blood urea nitrogen.

Twenty male Sprague–Dawley rats, 250–275 g, were randomly divided into two groups of 10 each. Following a 2-week acclimation period during which both groups were fed a standard animal diet[1], the first group was fed a diet supplemented to the third level of limiting amino acid (Table I) for 2 additional weeks while the second group was maintained on the standard diet for the same period.

On the 15th day, blood collected by orbital sinus puncture was centrifuged, and the serum was retained. Blood urea nitrogen was measured colorimetrically by means of a standard kit[2].

The serum blood urea nitrogen for the treatment group was 8.45 ± 0.36 mg/100 ml (mean ± *SEM*) while that of the control group was 15.1 ± 0.49 mg/100 ml. A Student *t* test showed a significant difference to the $p < 0.01$ level.

It is theorized that the observed effects are caused by an increase in net protein utilization when the limiting amino acids are supplemented, based on the fasting plasma profile theory, and that this increase reduces the amount of underutilized amino acids that are otherwise available for energy metabolism or storage. The end result is a decrease in the amount of nitrogenous metabolic products (wastes) in serum.

[1] Purina Laboratory Chow.
[2] Sigma Chemical Co., St. Louis, MO 63178.

Table I—Amino Acid-Supplemented Animal Feed to the Third Level of Limiting Amino Acid

Ingredient	Amount, %
L-Tryptophan	0.309
L-Threonine	0.341
L-Lysine	1.667
Standard feed	97.683

This type of dietary supplementation can be of invaluable assistance to those who must reduce their blood urea nitrogen levels (dialysis patients, nephrotic patients, *etc.*).

The major advantage of the augmentation of the body's amino acids based on the fasting plasma profile theory is that it removes a lot of guesswork and establishes a hard mathematical formula for preparing the diet.

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Effect of Variation of Plasma Oleic Acid Concentration on Relative Concentration of Free and Protein Bound Warfarin

Keyphrases □ Warfarin—effect of plasma oleic acid concentration on free and protein bound warfarin concentration, protein binding, humans □ Protein binding—binding of warfarin to human serum albumin, effect of plasma oleic acid concentration on free and bound warfarin concentration □ Oleic acid—effect of concentration on free and protein bound warfarin concentration, humans

To the Editor:

Although the normal serum concentrations of free fatty acids are between 0.3 and 0.9 mmole/liter (1), pathological conditions such as diabetes, reduced renal function, cardiac infarction, and bacterial disease (2–12) can cause substantial increases in free fatty acid levels. In some cases, these levels can exceed 5 mmoles/liter. Moreover, the increasingly widespread use of intravenous fat emulsions probably is associated with significant variation in the serum concentrations of free fatty acids.

Schwartz *et al.* (13) recently demonstrated that variation in the concentration of oleic acid, a major component in the free fatty acids found in serum, can cause significant variations in the protein binding of salicylate. Similar findings concerning the effect of variation in free fatty acid concentrations on the protein binding of drugs were published previously (14–17).

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Table I—Scatchard Protein Binding Parameters for Warfarin in the Presence and Absence of Linoleic, Palmitic, and Oleic Acids

Fatty Acid Concentration, mmoles/liter	Number of Drug Molecules Bound at Binding Sites 1	2	Association Constants[a] at Binding Sites 1	2
0[b]	1.95 (0.42)	6.6 (1.6)	7.94 (3.57)	402 (134)
0	2	7	6.56 (2.2)	376 (53)
Linoleic acid, 2.0	2	7	2.54 (1.5)	171 (61)
Palmitic acid, 2.0	2	7	0.79 (0.5)	116 (30)
Oleic acid				
0.5	2	7	5.26 (0.2)	258 (18)
1.0	2	7	3.61 (0.4)	214 (9)
1.5	2	7	2.88 (0.3)	177 (6)
2.0	2	7	1.04 (0.2)	115 (13)

[a] $1\ M \times 10^{-4}$. [b] Unconstrained values; all other values reported are constrained. Values in parentheses are standard deviations.

Table II—Estimated Percentage of Nonprotein Bound Warfarin for Serum Containing a Total Warfarin Concentration of 10 μg/liter

Oleic Acid Concentration, mmoles/liter	Estimated Percentage of Nonprotein Bound Warfarin
0.0	1.2
0.5	1.6
1.0	2.2
1.5	3.0
2.0	7.3

The present paper reports the effect of variation in oleic acid concentrations on the protein binding of warfarin by human serum albumin. Warfarin was selected because it has a low therapeutic ratio for which individual patient titration is recommended (18).

The binding of warfarin[1] by human serum albumin in the presence of oleic acid concentrations of 0–2.0 mmoles/liter at pH 7.4 was examined at 37° using the continuous ultrafiltration method described previously (13). Warfarin concentrations were determined by UV spectroscopy at 308 nm. Raw experimental data were interpreted using the "constrained" Scatchard model (13).

Table I lists the Scatchard parameters for warfarin binding by human serum albumin in the presence and absence of linoleic, palmitic, and oleic acids. (The constrained values are those obtained when the maximum number of molecules bound at the different sites is restricted to integral values.) All three fatty acids caused a significant reduction in both Scatchard association constants; increasing the oleic acid concentration progressively reduced the values of the constants.

The clinical implications of the data shown in Table I are appreciated more readily from Table II in which the percentage of unbound warfarin (estimated using the results in Table I) is shown when the total serum concentration of warfarin is 10 μg/liter. The percentage of free, nonprotein bound warfarin increases almost sevenfold when the oleic acid concentration is increased from 0 to 2.0 mmoles/liter. Since there is good reason to believe that the intensity of pharmacological response is primarily a simple function of the free unbound warfarin serum concentration, these results are of considerable potential therapeutic relevance. Changes in the pathological status of patients, who have been carefully titrated for optimal warfarin therapy, are likely to result in substantial hazard if such changes are associated with variation in free fatty acid serum concentrations. Thus, patients being treated with warfarin should be retitrated whenever free fatty acid levels have changed significantly.

[1] Warfarin sodium racemic mixture, Endo Laboratories.

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