

# Acute Effects of Peritoneal Dialysis with Dialysates Containing Dextrose or Dextrose and Amino Acids on Muscle Protein Turnover in Patients with Chronic Renal Failure

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**Abstract.** Whether changes in substrate and insulin levels that occur during peritoneal dialysis (PD) have effects on muscle protein dynamics was evaluated by studying muscle protein synthesis (PS), breakdown (PB), and net protein balance (NB) by the forearm perfusion method associated with the kinetics of 3H-phenylalanine in acute, crossover studies in which PD patients served as their own controls. Studies were performed (1) in the basal state and during PD with dialysates that contained dextrose alone in different concentrations (protocol 1: eight patients), (2) during PD with dialysates that contained dextrose alone or dextrose and amino acids (AA) (protocol 2: five patients), and (3) in time controls (five patients). PD with dextrose alone induced (1) a two- to threefold increase in insulin, as well as a 20 to 25% decrease in AA, mainly BCAA, levels; (2) an insulin-related decline (−18%) in forearm PB ( $P < 0.002$ ); (3) a 20% decrease in muscle PS ( $P < 0.04$ ), which was related to arterial BCAA and K<sup>+</sup> ( $P < 0.02$  to  $0.05$ ); (4) a persistent negative NB; and (5) a decrease in the efficiency of muscle protein turnover, expressed as the ratio NB/PB. PD

with dextrose+AA versus PD with dextrose induced (1) similarly high insulin levels but with a significant increase in total arterial AA (+30 to 110%), mainly valine; (2) a reduced release of AA from muscle ( $P < 0.05$ ); and (3) a decrease in the negative NB observed during PD with dextrose, owing to an increase (approximately 20%) in muscle PS, without any further effect on muscle PB. This study indicates that in PD patients in the fasting state, the moderate hyperinsulinemia that occurs during PD with dextrose alone causes an antiproteolytic action that is obscured by a parallel decrease in AA availability for PS. Conversely, the combined use of dextrose and AA results in a cumulative effect, because of the suppression of endogenous muscle PB (induced by insulin) and the stimulation of muscle PS (induced by AA availability). The hypothesis, therefore, is that in patients who are treated with PD, when fasting or when nutrient intake is reduced, muscle mass could be maintained better by the combined use of dextrose and AA.

Several studies have documented that malnutrition and wasting are frequently observed in patients who have chronic renal failure (CRF) with end-stage renal disease (ESRD) (1,2). There is evidence that loss of lean body mass is associated with increased mortality, and that also contributes to poor quality of life and scarce rehabilitation (3–5). In the pathogenesis of malnutrition, superimposed on conditions that result from uremia (1,2,6), nutritional problems caused by dialysis, such as hemodialysis or peritoneal dialysis (PD), can play a major role. PD is a widely practiced ESRD treatment modality; more than 100,000 patients throughout the world are currently being

treated (7). PD is based on the exchanges between blood and a glucose-containing peritoneal solution, thus exploiting the fluid and solute transport characteristics of the peritoneum. Some major metabolic effects of PD can be caused by the absorption of glucose. As much as 300 to 600 kcal are provided daily by different PD regimens and are responsible for the development of sustained hyperinsulinemia and the increase in triglycerides, as well as for the occurrence or aggravation of atherosclerosis, which are observed after the initiation of treatment (8).

Insulin is also an important anabolic hormone (9,10). In fasting PD patients, the whole-body antiproteolytic response to insulin has been shown to be unaltered (11) and hyperinsulinemia could contribute to preservation of muscle mass in these patients. Despite substantial glucose absorption, loss of lean body mass, with preservation or increase in fat stores, may be even more prevalent in PD than in hemodialysis patients (8,12). On one hand, the amount of glucose absorbed through the peritoneum, with its attendant hyperinsulinemia, is thought to have favorable effects on energy balance. On the other hand,

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catabolism induced by PD may stem from the amino acids (AA) and protein losses via the peritoneal route. Despite their potential repercussions, the effects of PD on muscle protein turnover have been unexplored so far. It has been shown that PD patients who ingest low amounts of proteins experience decreased rates of whole body synthesis and degradation, as estimated by  $^{14}\text{C}$ -leucine kinetics (13). Recently, it was demonstrated that increasing AA availability by giving AA intraperitoneally is followed by a stimulation of whole-body protein synthesis (PS) and that this effect is enhanced by the inhibition of protein breakdown (PB) induced by a meal (14).

Both insulin and AA are widely considered as necessary for the stimulation of muscle protein anabolism. However, neither the effects of hyperinsulinemia nor the quantitative importance of AA availability on muscle protein metabolism has been established during PD. To provide such data, we measured the kinetics of 3H-phenylalanine as well as the exchange of AA across the forearm (which is mainly made of muscle) in the basal postabsorptive state and during PD with dialysates that contained dextrose (during which systemic hyperinsulinemia associated with decreased AA availability occur) or PD performed with a combination of dextrose and AA (during which systemic hyperinsulinemia and increased AA availability take place) in two crossover studies in continuous ambulatory PD (CAPD) patients.

## Materials and Methods

### Study Participants

Eighteen CAPD patients were studied. Eight of these patients participated in protocol 1, five participated in protocol 2, and five served as time controls. All patients were in stable clinical conditions and ingested a diet that contained 1.2 to 1.3 g/kg proteins and 30 to 35 kcal/kg. None of the patients had any history of diabetes or major organ disease with the exception of CRF.

Patients had been free of peritonitis for at least 2 mo before the study. All patients received vitamin supplements that contained B vitamins and folic acid. None of the patients were receiving insulin, androgenic steroids, or lipid-lowering agents. Before the patients' participation, the nature, purpose, and risks of the study were reviewed with all of the patients and their voluntary consent was obtained. The protocols described here were approved by the Ethical Committee of the Department of Internal Medicine of the University of Genoa. Procedures were in accordance with the Helsinki declaration.

### Experimental Protocols

Two different protocols were used. Both consisted in crossover studies with patients serving as their own controls. Protocol 1 was designed to study the acute effects of dialysates that contain dextrose alone on muscle protein turnover. In this protocol, the same patients were studied twice (in the postabsorptive state and during PD with dextrose). Protocol 2 was designed to study the acute effects of dialysates that contain dextrose alone or dextrose+AA on muscle protein turnover. In this protocol, the same patients were studied twice (when they were receiving dextrose alone and during dextrose+AA). Five patients in protocol 1 and five in protocol 2 were treated with the same dextrose concentration (2.27% dextrose). Because both studies were long and fasting could influence protein turnover, a group of patients who received only saline (time controls) were studied as a

control group for a period of time similar to that used in protocols 1 and 2.

In both protocols as well as in time control studies, the study of muscle protein turnover was performed in the postabsorptive, overnight fasted state. At 7.00 a.m., a forearm vein was cannulated with a 18-gauge polyethylene catheter and used for a primed-continuous infusion of L-(ring 2,6)-3H-phenylalanine (approximately 32  $\mu\text{C}$ , 0.35  $\mu\text{Ci}/\text{min}$ ). The tracer infusion was maintained until the end of the study. Catheters were inserted into a brachial artery and in a retrograde manner into the ipsilateral, deep forearm vein. Blood samples were taken every 30 min after the start of PD to assess the achievement of steady-state conditions of plasma substrate concentrations and specific activities (data not shown). Both phenylalanine and other AA were fairly stable in blood (coefficient of variation < 5%) during the study periods. During dextrose+AA administration, AA levels increased acutely in blood after 1 h, then progressively declined to a plateau (plateau was defined by the absence of significant slope) assessed by linear regression analysis. For determination of steady-state forearm fluxes, arterial and deep venous samples were obtained at 10-min intervals over a 30-min period in the basal state and the study periods.

**Study Protocol 1: Effects of PD with Dialysate that Contained Dextrose Alone.** Eight male CRF patients (age  $57 \pm 5$ ; range, 40 to 73 yr; creatinine clearance,  $3 \pm 2$  ml/min) treated with CAPD for 19 mo (range, 7 to 48 mo) were studied. Their mean percentage of ideal body weight, based on medium-framed individuals (15), was  $107 \pm 2$ , body mass index =  $26 \pm 0.5$ . On the basis of their peritoneal membrane characteristics, two of them were classified as average-high and six as average-low transporters (16). Their dialysis dose, expressed as Kt/V, was  $2 \pm 0.1$ , and their protein intake, as expressed as nPCR (6), was  $1.2 \pm 0.1$  g/kg per d. Blood urea nitrogen (BUN) was  $71 \pm 5$  mg/dl, and albumin was  $3.8 \pm 0.2$  g/dl.

Patients had the abdomen left dry and consumed their last meal at approximately 10 p.m. on the day before the study. After obtaining baseline samples (150 to 180 min from the start of tracer infusion), PD with a dextrose-containing dialysate was started. PD was performed with four 1-h, 2-L exchanges (dwell time, 30 min the first three, 60 min the fourth). Dialysis fluid contained 132 mmol/L sodium, 3.5 mmol/L calcium, 1.5 mmol/L magnesium, 102 mmol/L chloride, and 35 mmol/L lactate. To explore the effects of different amounts of dextrose administration on muscle protein turnover, solutions that contained different dextrose concentrations (13.6 g/L [three patients] or 22.7 g/L [five patients]) were used.

The forearm protein turnover study was repeated after approximately 200 min, after approximately 20 min from the beginning of the fourth exchange, *i.e.*, at approximately 380 to 420 min from the beginning of the infusion. Patients were not allowed to eat during the study.

**Study Protocol 2: Effects of PD with Dialysate that Contained Dextrose+AA Versus Dextrose Alone.** Five CRF patients (one woman/4 men; age,  $55 \pm 8$  yr; range, 27 to 73 yr; body mass index,  $24 \pm 2$ ; creatinine clearance,  $2 \pm 1$  ml/min; nPCR,  $1.2 \pm 0.1$ ; Kt/V,  $2 \pm 0.1$ ; BUN,  $73 \pm 7$  mg/dl; albumin,  $3.8 \pm 0.15$  g/dl;  $\text{K}^+$ ,  $4.2 \pm 0.3$ ,  $\text{HCO}_3^-$ ,  $24 \pm 1$  mmol/L) treated with CAPD for 18 mo (range, 12 to 30 mo) were studied. On the basis of their peritoneal membrane characteristics, four patients were classified as average-low and one as average-high transporters. Patients had the abdomen left dry from and had consumed their last meal at approximately 10 p.m. of the previous night. On the day of the study, from approximately 6 to 10 a.m., patients received four 1-h, 2-L PD exchanges (dextrose 2.27%; dwell time, 30 min) by a cyclor (Baxter HomeChoice, McGaw Park, IL). Subsequently, their dialysate was substituted for one that resulted

from the combination of one 1.1% Baxter 2-L Nutrineal A bag (AA 1.1%) to one 5-L 2.27% and one 2-L 3.86% dextrose solution. The final dialysate dextrose and AA concentrations were 2.12 and 0.24%, respectively. The composition of the AA solution was as follows: 0.3 g of tyrosine, 0.27 g of tryptophan, 0.57 g of phenylalanine, 0.64 g of threonine, 0.51 g of serine, 0.595 g of proline, 0.51 g of glycine, 0.95 g of alanine, 1.393 g of valine, 0.85 g of methionine, 0.85 g of isoleucine, 1.02 g of leucine, 0.955 g of lysine, 0.714 g of histidine, 1.071 g/L arginine; 0.184 g of calcium chloride (2H<sub>2</sub>O), 0.0508 g of MgCl<sub>2</sub> (×6H<sub>2</sub>O), 4.48 g of sodium lactate, and 5.38 g/L NaCl. Four 1-h cyclo-assisted PD exchanges were performed from 10 a.m. to 2 p.m., *i.e.*, from 240 to 480 min from the beginning of the study. The forearm protein turnover study was performed from 210 to 240 min from the start of dialysis and during the last 30 min of the dextrose+AA period, *i.e.*, from 450 to 480 min from the beginning of the tracer infusion.

Patients were not allowed to eat during the study.

**Control Subjects.** Five CAPD patients (age,  $51 \pm 5$  yr; range, 39 to 69 yr; 4 men, 1 woman; creatinine clearance,  $4 \pm 3$  ml/min; body weight,  $102 \pm 3\%$  of their ideal body weight; nPCR,  $1.2 \pm 0.4$ ; BUN,  $76 \pm 4$  mg/dl; K<sup>+</sup>,  $4.3 \pm 0.2$  mEq/L; albumin,  $3.8 \pm 0.4$  g/dl; HCO<sub>3</sub><sup>-</sup>,  $25 \pm 1$  mmol/L) served as time controls. After a 180-min basal period, normal saline (0.23 ml/min) was infused for 300 min. Arterial and deep venous samples were obtained at the baseline, *i.e.*, from 150 to 180 min from the start of the tracer infusion, and at 210 to 220, 220 to 240, and 450 to 480 min.

### Blood Flow Measurement

Blood flow across the forearm was determined immediately after each arteriovenous sampling by a indium-gallium strain gauge plethysmograph (Angiomed Instruments, Padua, Italy). The arm that contained the deep vein was raised approximately 20°, and a small supporting pillow was placed under the wrist to avoid interference with venous return. The strain gauge was wrapped around the forearm at the level of maximum circumference. The relative change in the volume of the forearm segment under the strain gauge was registered and was used to calculate plethysmographic blood flow. An occlusion cuff was placed around the arm. The pressure was 60 mmHg. Each set of measurements involved 10 separate measurements (coefficient of variation = 4%). For 1 min before and during withdrawal of each blood sample, blood flow to the hand was interrupted by means of a sphygmomanometer cuff inflated around the wrist to 200 mmHg. Flow was expressed per 100 ml of forearm volume. Room temperature was 20 to 22°C.

### Assays

AA were determined in whole blood and in the peritoneal effluent. Perchloric acid (0.75 mol/L) was used for protein precipitation. An aliquot of the supernatant was neutralized with a buffered solution, stored at -25°C, and used for the assay of glutamine and glutamate. Another aliquot was stored at -25°C and used for the determination of other AA. Blood samples were processed immediately after withdrawal. AA were measured in triplicate by an AA analyzer (Mod. 3A30, Fisons Instruments, Milan, Italy), with the use of lithium buffers. Phenylalanine-specific activity was determined in blood, as described previously (17,18).

Plasma insulin was determined by RIA (Diagnostic Products, Los Angeles, CA). Arterial blood pH and Pco<sub>2</sub> were estimated at 37°C with an ABL 505 apparatus (Radiometer Co., Copenhagen, Denmark). Blood bicarbonate concentrations were calculated by using the Henderson-Hasselbalch equation.

Hematocrit was measured by a microcapillary procedure. Albumin levels in serum and peritoneal fluid were determined by bromocresol purple dye (Sigma Diagnostics, St. Louis, MO). All other serum chemical measurements were determined by routine clinical chemistry laboratory procedures.

### Calculations

The net forearm balance for AA was calculated according to the Fick principle. Forearm phenylalanine kinetics were calculated using the A-V model described previously (17,18). Briefly, the net forearm balance (NB) for substrates was calculated as follows:

$$NB = ([A] - [V]) \times \text{blood flow}$$

where [A] and [V] are the arterial and venous concentrations, respectively. Phenylalanine is neither synthesized nor metabolized in muscle (14). Therefore, the rates of disposal of phenylalanine across the forearm in the steady state reflect PS, whereas tissue rates of appearance of phenylalanine reflect its release from tissue PB (19). Data are presented using arterial specific activity as an expression of the phenylalanine precursor pool.

Muscle phenylalanine rate of appearance (an expression of PB) was calculated as follows (17):

$$\text{Phenylalanine rate of appearance} = \text{blood flow} \times [V] \times (1 - SA_v/SA_a)$$

where [V] is the venous blood level of phenylalanine and SA<sub>a</sub> and SA<sub>v</sub> indicate the specific activity (dpm/nmol) of phenylalanine in the artery and the vein, respectively. The term  $(1 - SA_v/SA_a)$  expresses the dilution of tracer-specific activity across the forearm caused by unlabeled phenylalanine released by muscle.

Muscle disposal of unlabeled phenylalanine (an expression of muscle PS) was calculated as follows:

$$\text{Phenylalanine rate of disposal} = \text{Phenylalanine rate of appearance} + \text{net balance}$$

Using phenylalanine data to obtain a parameter relevant to the efficiency of muscle protein turnover, we calculated the fraction of the phenylalanine rate of appearance that escapes into the forearm vein as follows:

$$\text{Efficiency} = 1 - (NB/PB)$$

Whole-body phenylalanine flux was calculated from the rate of tracer infusion (dpm/min) divided by the 3H phenylalanine-specific activity in arterial blood (17).

The peritoneal glucose and AA absorption was calculated as the difference between the amounts initially present in the peritoneal fluid bags and the amounts finally recovered. The volume of fluid delivered to the peritoneal cavity was obtained by weighing the dialysis bag before and after it was emptied. The AA losses were calculated by multiplying the AA concentration in final fluid by the volume of fluid recovered.

The total decrease in extracellular AA concentration was estimated as the product of total body water (kg) × 0.33 (extracellular fluid volume) × decrease in plasma AA levels over basal levels. Total body water was calculated according to Watson *et al.* (20).

Extracellular volume was assumed to be one third of total body water. The percentage of the decrease in extracellular AA accounted for by dialysis removal was calculated as the ratio of removed amounts/total decrease in extracellular fluids. These calculations assume also that changes in plasma AA concentrations reflect changes in extracellular AA concentrations (21).

## Statistical Analyses

All data are presented as the mean  $\pm$  SEM. Statistical analysis was performed using the two-tailed *t* test to compare arterial with venous data. When the arteriovenous difference was different from 0 ( $P < 0.05$ ) or when intragroup statistical significances were to be evaluated, a repeated-measure ANOVA was used to compare the overall changes during the phases of the study. When ANOVA indicated statistical significance ( $P < 0.05$ ), a *post hoc* F-based test was performed between phases. To establish the differences in AA concentrations between patients and controls, we used the unpaired *t* test and one-way ANOVA followed by *post hoc* analysis. Linear regression and correlation were used to evaluate the relationship between two variables. Stepwise multiple regression analysis was used to discover which independent variables were significantly related to the dependent variables when the contribution of other independent variables was factored out and to discover what proportion of variations in dependent variables was accounted for by each independent variable and by the model. Statistical analysis was performed with the Statview Statistical Package (Abacus, Berkeley, CA).

## Results

### Serum Insulin, Forearm Blood Flow, Phenylalanine Concentrations, and Specific Activities

During protocol 1 (PD with dextrose alone), serum insulin was increased from 8 to approximately 19  $\mu$ U/ml (on the average from  $9 \pm 2$  to  $24 \pm 3$  and  $7 \pm 1$  to  $13 \pm 2$   $\mu$ U/ml in patients who received 2.27 or 1.36% dextrose, respectively) (Table 1). Arterial blood glucose rose slightly (basal,  $4.0 \pm 0.2$ ; 200 to 215 min,  $5.3 \pm 0.3$ ; 215 to 230 min,  $5.1 \pm 0.2$ ; 225 to 240 min,  $5.2 \pm 0.2$  mmol/L;  $P < 0.07$ ). Arterial potassium levels declined from  $4.8 \pm 0.3$  to  $3.9 \pm 0.2$  (200 to 215 min),  $3.9 \pm 0.2$  (215 to 230 min), and  $3.8 \pm 0.2$  (230 to 240 min;  $P < 0.005$ ). Arterial bicarbonate levels rose slightly, although not significantly (basal,  $25 \pm 1$ ; 200 to 215 min,  $25 \pm 1$ ; 215 to 230 min,  $26 \pm 1$ ; 230 to 240 min,  $26 \pm 1$  mmol/L;  $P = \text{NS}$ ). Serum albumin declined slightly ( $4.2 \pm 0.31$  g/dl at baseline versus  $3.9 \pm 0.23$  g/dl at 240 min;  $P < 0.08$ ). During protocol 2, when patients received dextrose+AA, insulin levels were elevated by approximately threefold in comparison with normal basal values and were not statistically different from those observed in patients who received dextrose alone in similar concentrations in protocol 1 and protocol 2. There was no change in arterial bicarbonate level (dextrose,  $24 \pm 1$ ; dextrose+AA,  $24.4 \pm 1$  mEq/L;  $P = \text{NS}$ ). Potassium levels were not significantly affected by the use of dextrose+AA versus dextrose alone (on the average,  $3.8 \pm 0.3$  versus  $3.9 \pm 0.3$  mEq/L, respectively;  $P = \text{NS}$ ). Albumin levels were similar at the end of both studies (dextrose,  $3.9 \pm 0.4$ ; dextrose+AA,  $3.8 \pm 0.3$  g/dl;  $P = \text{NS}$ ). During both protocols, baseline forearm blood flow was not significantly changed by treatments or saline. Phenylalanine arterial concentrations were slightly but significantly reduced (by approximately 9%) during PD with dextrose alone, but they increased (by approximately 37%) during PD with dextrose+AA. Basal venous concentrations of phenylalanine exceeded the arterial ones ( $P < 0.05$  to 0.01) in all studies. However, venous phenylalanine levels approached the arterial ones during PD with

Table 1. Serum insulin, forearm blood flow, phenylalanine concentrations, and specific activities in protocols 1 and 2 and in control subjects<sup>a</sup>

	Protocol 1: PD with Dextrose Alone (min)						Protocol 2: PD with Dextrose Versus Dextrose + AA (min)					
	Dextrose			Dextrose + AA			Dextrose			Dextrose + AA		
	Baseline	200 to 215	215 to 230	230 to 240	200 to 240	450 to 460	460 to 470	470 to 480	Baseline	210 to 220	220 to 240	450 to 480
Insulin ( $\mu$ U/ml)	$8.0 \pm 4.0$	$18.0 \pm 3.0^b$	$19.0 \pm 3.0^b$	$19.0 \pm 3.0^b$	$27.0 \pm 4.0$	$25.0 \pm 4.0$	$26.0 \pm 4.0$	$25.0 \pm 3.0$	$8.0 \pm 1.0$	$9.0 \pm 2.0$	$7.0 \pm 1.0$	$7.0 \pm 1.0$
Forearm blood flow (ml/min per 100 ml)	$3.6 \pm 0.3$	$3.5 \pm 0.3$	$3.6 \pm 0.4$	$3.8 \pm 0.4$	$2.9 \pm 0.2$	$3.1 \pm 0.4$	$2.8 \pm 0.5$	$3.1 \pm 0.5$	$4.0 \pm 0.9$	$3.9 \pm 1.0$	$4.4 \pm 1.2$	$4.1 \pm 1.0$
Arterial phenylalanine (nmol/ml)	$49.0 \pm 2.0$	$45.0 \pm 3.0^b$	$46.0 \pm 2.0^b$	$45.0 \pm 2.0^b$	$45.0 \pm 3.0$	$63.0 \pm 4.0^c$	$62.0 \pm 4.0^c$	$61.0 \pm 4.0^c$	$41.0 \pm 4.0$	$43.0 \pm 3.0$	$42.0 \pm 3.0$	$41.0 \pm 3.0$
Venous phenylalanine (nmol/ml)	$51.0 \pm 2.0^f$	$48.0 \pm 3.0^{b,e}$	$49.0 \pm 3.0^f$	$49.0 \pm 3.0^f$	$49.0 \pm 3.0^{e,e}$	$65.0 \pm 4.0^{e,e}$	$64.0 \pm 4.0^f$	$63.0 \pm 4.0^{e,e}$	$44.0 \pm 3.0^f$	$46.0 \pm 3.0^f$	$45.0 \pm 3.0^f$	$45.0 \pm 3.0^f$
Arterial phenylalanine specific activity (dpm/nmol)	$20.0 \pm 1.0$	$24.0 \pm 2.0^f$	$24.0 \pm 2.0^f$	$23.0 \pm 2.0^f$	$23.0 \pm 4.0$	$21.0 \pm 3.0^b$	$21.0 \pm 3.0^b$	$22.0 \pm 3.0^b$	$21.0 \pm 2.0$	$22.0 \pm 1.0$	$21.0 \pm 1.0$	$22.0 \pm 1.0$
Venous phenylalanine specific activity (dpm/nmol)	$15.0 \pm 1.0^f$	$19.0 \pm 1.0^{b,f}$	$19.0 \pm 1.0^{b,f}$	$19.0 \pm 1.0^{b,f}$	$18.0 \pm 3.0^f$	$17.0 \pm 2.0^f$	$16.0 \pm 2.0^f$	$17.0 \pm 2.0^f$	$16.0 \pm 1.0^f$	$17.0 \pm 1.0^f$	$16.0 \pm 1.0^f$	$17.0 \pm 1.0^f$

<sup>a</sup> AA, amino acids.

Significance of difference from the corresponding value at the baseline or peritoneal dialysis with dextrose period: <sup>b</sup>  $P < 0.05$ ; <sup>c</sup>  $P < 0.01$ ; <sup>d</sup>  $P < 0.001$ . Probability that arteriovenous difference does not differ from 0: <sup>e</sup>  $P < 0.05$ ; <sup>f</sup>  $P < 0.01$ .

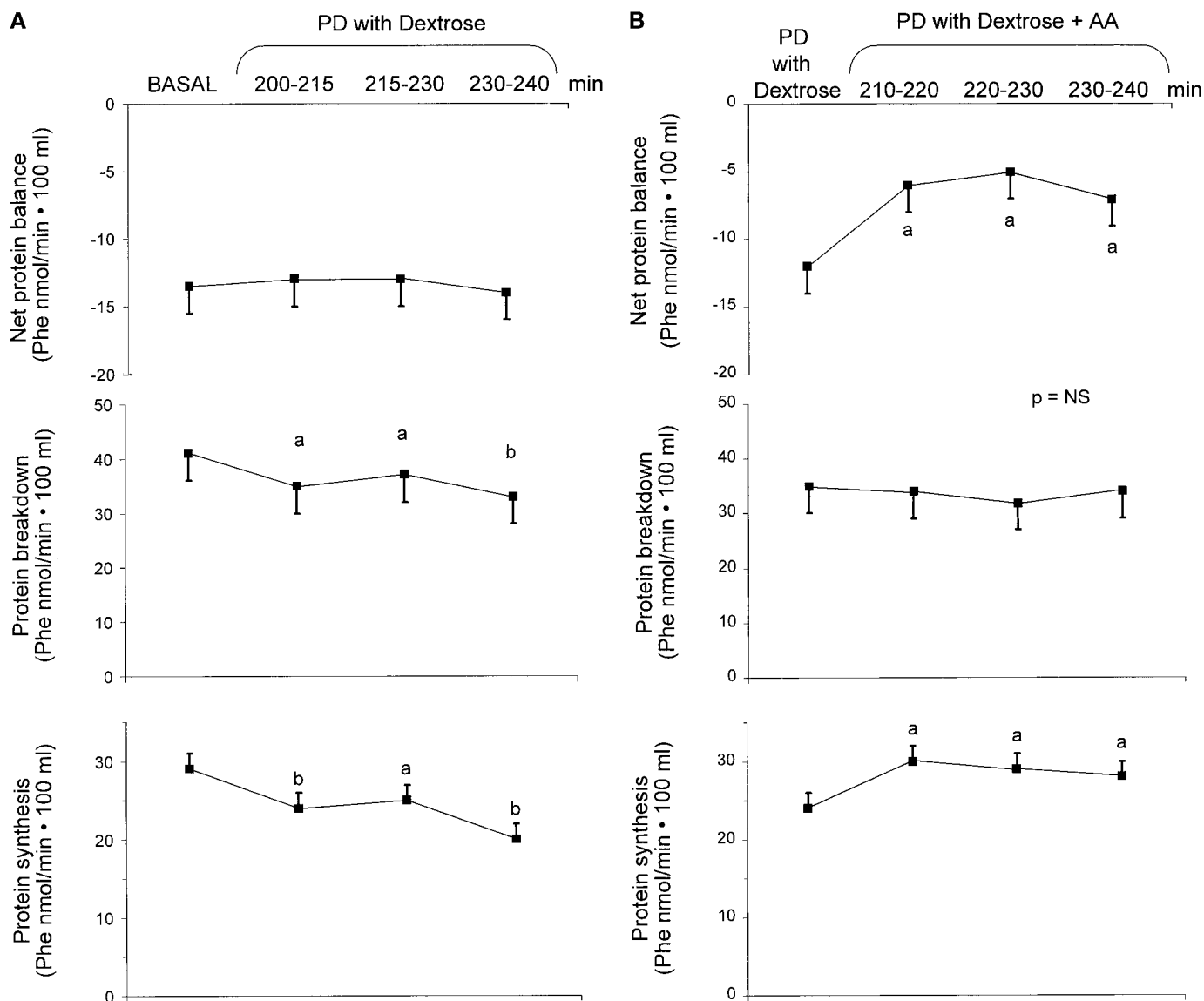


Figure 1. Net protein balance, protein breakdown, and protein synthesis across the forearm in the basal state and during peritoneal dialysis (PD) with dextrose (protocol 1) (A) or with dextrose+amino acids (AA; protocol 2) (B). Significance of the difference from basal values: a =  $P < 0.05$ ; b =  $P < 0.01$ .

dextrose+AA but not during PD with dextrose alone or in controls. Phenylalanine-specific activities in the artery and the vein were in steady-state conditions in the last period of the baseline and the PD with dextrose period, *i.e.*, from 200 to 240 min after the end of the basal period, as well as during the last periods of PD with dextrose alone, *i.e.*, from 200 to 240 min, or with AA, *i.e.*, from 450 to 480 min. Specific activities were significantly lower in the vein with respect to the artery in both protocols 1 and 2 and in controls. The average specific activity of phenylalanine in the artery and the deep forearm vein increased significantly during PD with dextrose ( $P < 0.02$ ). During the PD with dextrose+AA *versus* PD with dextrose study, a decrease in arterial-specific activity ( $P < 0.05$ ) was observed, indicating entry of unlabeled phenylalanine into the extracellular pool (Table 1).

### AA Balance across the Forearm

**Protocol 1: PD with Dextrose Alone.** In the postabsorptive state, the CRF patients studied exhibited lower arterial blood levels of serine, valine, leucine, and tyrosine and higher levels of proline, citrulline, glutamate, and cyst(e)ine as compared with those obtained in our laboratory in 35 normal subjects (22). As shown in Table 2, arterial concentrations of several AA decreased during PD. A significant decrease was observed for valine and leucine (-21 and 23%, respectively), tyrosine (-20%), and cyst(e)ine and threonine (11 to 18%). For some other AA, such as taurine, lysine, citrulline, and serine, a small, not statistically significant decrease was observed in whole blood. Alanine was the only AA that tended to increase (+10%). Despite the increase in insulin levels, forearm AA balance remained on the average unchanged. A ten-

dency toward an increased release of valine, threonine, and alanine was observed but did not achieve statistical significance. The release in plasma was similar in both quantitative and qualitative terms to what was observed in whole blood; therefore, it is not reported in the table. Changes in arterial plasma AA followed a similar trend to that observed in whole blood. Only for taurine and serine, the decrease was greater in plasma than in whole blood, suggesting that these AA are predominantly transported by way of plasma across the peritoneal membrane.

As a general trend, the amount of AA removed via PD was related to their arterial concentration. According to our estimate, removal with the peritoneal fluid accounted for 50 to 70% (valine, isoleucine, leucine, tyrosine, serine) or 70 to 100% (taurine, threonine, proline, lysine) of the decrease in extracellular AA levels (Table 2).

**Protocol II: PD with Dextrose+AA.** During PD with dextrose+AA, a rise in arterial blood was observed for several AA as compared with treatment with dextrose alone (Table 2). Most marked increments were found for valine and isoleucine (approximately 70%), leucine (approximately 60%), threonine and arginine (approximately 40%), histidine and lysine (approximately 20 to 26%), and serine and glycine (approximately 16 to 20%). Alanine levels did not change. The relationship

between the percentage increase in blood AA (excluding alanine) and the molar ratio of AA contained in the dialysate gave a correlation coefficient close to unity ( $r = 0.90$ ,  $P < 0.0005$ ).

As compared with the basal period in which patients received PD with dextrose alone, forearm NB from AA became less negative (from  $-640 \pm 223$  to  $-277 \pm 206$  nmol/min per 100 ml;  $P < 0.05$ ), owing to the occurrence of a net uptake of valine and a decreased release of serine, glycine, alanine, lysine, histidine, and arginine (Table 2). For several AA (valine, threonine, isoleucine, glycine, leucine, serine, tyrosine, histidine), variations in their exchange across the forearm were directly related to changes in their arterial concentration ( $P < 0.04$  to  $0.01$ ).

### Phenylalanine Kinetics across the Forearm

**Protocol I: PD with Dextrose.** In the basal, postabsorptive state, as well as during PD with dextrose, protein degradation exceeded PS ( $P < 0.05$  to  $0.003$ ) and the forearm muscle was in a negative protein balance (Figure 1A). However, during PD, muscle protein degradation decreased on the average by 17% ( $P < 0.01$ ). Protein degradation was inversely related to insulin levels ( $r = -0.578$ ;  $P < 0.002$ ; Figure 2), suggesting the occurrence of an insulin-induced inhibition of muscle PB. However, no net muscle anabolism ensued in

Table 2. Whole-blood arterial concentration (nmol/ml) and forearm exchange (nmol/min per 100 ml) of AA at the baseline, during peritoneal dialysis with dextrose (protocol 1), or during dextrose + AA (protocol 2)<sup>a</sup>

	Protocol 1				Protocol 2			
	Baseline		Peritoneal Dialysis (Dextrose)		Peritoneal Dialysis (Dextrose)		Peritoneal Dialysis (Dextrose + AA)	
	Arterial Concentration	Forearm Exchange	Arterial Concentration	Forearm Exchange	Arterial Concentration	Forearm Exchange	Arterial Concentration	Forearm Exchange
Taurine	216 ± 18	4 ± 9	199 ± 17	-14 ± 6	155 ± 30	6 ± 9	164 ± 4	-3 ± 6
Threonine	128 ± 11	-28 ± 11 <sup>e</sup>	105 ± 8 <sup>f</sup>	-41 ± 12 <sup>e</sup>	102 ± 11	-39 ± 14 <sup>e</sup>	151 ± 18 <sup>f</sup>	-24 ± 18
Serine	88 ± 9 <sup>b</sup>	-10 ± 5	78 ± 7	-3 ± 4	70 ± 4	-12 ± 11	81 ± 7 <sup>f</sup>	10 ± 15 <sup>f</sup>
Glutamate	209 ± 30 <sup>b</sup>	58 ± 10 <sup>e</sup>	219 ± 32	70 ± 11 <sup>e</sup>	177 ± 38	62 ± 20 <sup>e</sup>	205 ± 25	78 ± 32 <sup>e</sup>
Glutamine	457 ± 24	-77 ± 10 <sup>e</sup>	420 ± 27	-65 ± 13 <sup>e</sup>	490 ± 27	-88 ± 13 <sup>e</sup>	460 ± 23	-50 ± 14 <sup>e</sup>
Proline	290 ± 31 <sup>c</sup>	-8 ± 3	270 ± 30	4 ± 6	290 ± 21	-11 ± 6	—	—
Glycine	261 ± 16	-21 ± 8 <sup>d</sup>	255 ± 16	-14 ± 9 <sup>d</sup>	240 ± 24	-35 ± 18 <sup>e</sup>	283 ± 29	-18 ± 15 <sup>f</sup>
Alanine	260 ± 14	-110 ± 19 <sup>e</sup>	287 ± 10	-140 ± 20 <sup>e</sup>	279 ± 5	-215 ± 80 <sup>e</sup>	280 ± 15	-185 ± 83 <sup>d,g</sup>
Citrulline	107 ± 12 <sup>c</sup>	10 ± 4 <sup>d</sup>	90 ± 11	3 ± 4	89 ± 11	3 ± 4	89 ± 11	1 ± 3
Valine	154 ± 8 <sup>c</sup>	-14 ± 7 <sup>e</sup>	122 ± 10 <sup>g</sup>	-19 ± 7 <sup>e</sup>	119 ± 5	-20 ± 9 <sup>d</sup>	204 ± 12 <sup>g</sup>	28 ± 8 <sup>d,f</sup>
Cyst(e)ine	119 ± 7 <sup>c</sup>	11 ± 7	106 ± 6 <sup>c</sup>	13 ± 6	74 ± 9	3 ± 5	73 ± 11	12 ± 5 <sup>c</sup>
Isoleucine	58 ± 4	-9 ± 5 <sup>d</sup>	42 ± 5 <sup>g</sup>	-8 ± 5 <sup>d</sup>	40 ± 5	-6 ± 5	69 ± 4 <sup>g</sup>	10 ± 1 <sup>d</sup>
Leucine	86 ± 6 <sup>b</sup>	-21 ± 8 <sup>d</sup>	66 ± 5 <sup>g</sup>	-19 ± 8 <sup>e</sup>	61 ± 4	-16 ± 5 <sup>d</sup>	97 ± 9 <sup>g</sup>	-4 ± 2 <sup>f</sup>
Tyrosine	36 ± 4 <sup>b</sup>	-11 ± 3 <sup>e</sup>	29 ± 4 <sup>f</sup>	-12 ± 2 <sup>e</sup>	29 ± 4	-14 ± 5 <sup>e</sup>	36 ± 8	-10 ± 4 <sup>d</sup>
Ornithine	83 ± 6	-5 ± 4	78 ± 5	-3 ± 2	74 ± 13	-4 ± 1	80 ± 12	4 ± 3
Lysine	128 ± 8	-28 ± 6 <sup>e</sup>	120 ± 5	-29 ± 7 <sup>e</sup>	120 ± 11	-40 ± 18 <sup>e</sup>	145 ± 16 <sup>f</sup>	-28 ± 17 <sup>d,g</sup>
Histidine	83 ± 5	-10 ± 6	78 ± 3	-7 ± 2	75 ± 6	-17 ± 10	95 ± 11 <sup>f</sup>	-7 ± 6 <sup>g</sup>
Arginine	68 ± 5	-19 ± 3 <sup>e</sup>	63 ± 3	-17 ± 4 <sup>d</sup>	68 ± 9	-30 ± 10 <sup>d</sup>	95 ± 11 <sup>f</sup>	-9 ± 16 <sup>g</sup>

<sup>a</sup> Values before and after peritoneal dialysis are the combined pre- and postevent measurements.

Significance of difference from the arterial concentrations of amino acids in 36 normal subjects (see Materials and Methods section): <sup>b</sup>  $P < 0.05$ ; <sup>c</sup>  $P < 0.025$  or less. Probability that the A-V difference does not differ from 0: <sup>d</sup>  $P < 0.05$ ; <sup>e</sup>  $P < 0.025$  or less. Statistically different from baseline or from the peritoneal dialysis with dextrose period: <sup>f</sup>  $P < 0.05$ ; <sup>g</sup>  $P < 0.025$  or less.

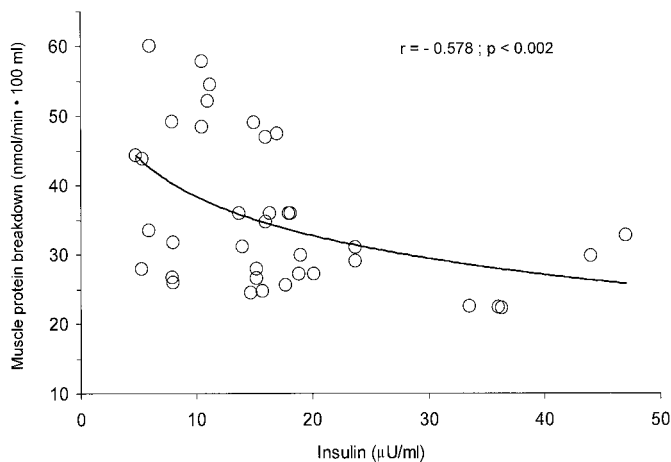


Figure 2. Relationship between muscle protein breakdown and plasma insulin during PD with dextrose in continuous ambulatory PD patients.

response to the increase in insulin levels. Muscle phenylalanine net balance (NB) was on the average unchanged, owing to the occurrence of an average 26% decrease in muscle PS ( $P < 0.04$ ). Furthermore, during the PD study, the NB of phenylalanine across the forearm was only marginally related ( $r = -0.28$ ;  $P < 0.064$ ) to insulin levels.

Changes in muscle PS during PD were related directly to arterial BCAA ( $r = 0.32$ ;  $P < 0.04$ ) and potassium concentrations ( $r = 0.682$ ;  $P < 0.001$ ). Moreover, basal arterial potassium predicted the dialysis-induced averaged changes in PS ( $r = 0.797$ ;  $P < 0.004$ ). In a multiple linear regression model, BCAA and potassium levels accounted for 52% of the observed variability (multiple  $r^2 = 0.522$ ;  $P < 0.001$ ), with a greater weight for potassium (partial  $r^2 = 0.46$ ;  $P < 0.002$ ) than for BCAA (partial  $r^2 = 0.059$ ;  $P < 0.06$ ). There was no significant relationship between the total amount of potassium removed or changes in arterial potassium levels and PS. When two patients with the lowest basal plasma potassium levels (3.4 and 3.7 mEq/L, respectively) were excluded, BCAA were the only predictors of changes in muscle PS.

In the basal period, as much as  $70 \pm 6\%$  of phenylalanine that appeared from PB was cycled back into PS. During PD with dextrose, this value declined to  $63 \pm 5\%$  ( $P < 0.05$ ) in the last period of the study (200 to 215 min,  $66 \pm 5\%$ ; 215 to 230 min,  $66 \pm 5\%$ ; 230 to 240 min,  $63 \pm 5\%$ ), owing to a significant decrease in PB not counterbalanced by a decrease in phenylalanine NB. During PD with dextrose, whole-body phenylalanine flux declined by approximately 14% (basal,  $0.573 \pm 0.06$ ; PD with dextrose,  $0.478 \pm 0.05$ ,  $0.480 \pm 0.06$ ,  $0.486 \pm 0.06$   $\mu\text{mol/kg}$  per min;  $P < 0.02$ ).

**Protocol II: PD with Dextrose+AA Versus PD with Dextrose Alone.** At the end of the PD with dextrose study, net phenylalanine balance across the forearm was negative, as already observed during protocol 1 (Figure 1B). This occurred even in the presence of low absolute values for protein degradation, which, however, were greater than the low absolute rates of PS. In response to PD with dextrose+AA, no change

in muscle protein degradation was observed. However, the overall response to the increased AA availability was a decrease in the negative muscle protein balance observed with respect to when dextrose alone was used. This occurred because of a 20% increase in muscle PS. We could not find a significant relationship between protein degradation and insulin when dextrose+AA have been studied. This may have occurred because all patients showed a sizable increase over baseline insulin levels, which suppressed protein degradation. However, when all of the experimental conditions (baseline, dextrose, and dextrose+AA) were considered, the correlation between protein degradation and insulin levels continued to be significant ( $r = -0.325$ ;  $P < 0.02$ ).

Whole-body phenylalanine flux increased by approximately 9% (PD with dextrose,  $0.55 \pm 0.12$ ; PD with dextrose+AA,  $0.60 \pm 0.11$ ,  $0.62 \pm 0.12$ ,  $0.60 \pm 0.12$   $\mu\text{mol/kg}$  per min;  $P < 0.05$  to  $0.02$ ) during PD with dextrose and AA as compared with the baseline period when patients received dextrose alone.

### Time Controls

In time-control subjects in the baseline period, forearm was in a negative balance because of a PB greater than PS. There was no change over time with regard to net phenylalanine balance or rates of phenylalanine uptake or release across the forearm (protein breakdown: baseline,  $40 \pm 5$ ; 200 to 220 min,  $38 \pm 8$ ; 220 to 240,  $36 \pm 7$ ; 440 to 460 min,  $41 \pm 6$  nmol/min per 100 ml; PS baseline,  $27 \pm 5$ ; 210 to 220 min,  $23 \pm 6$ ; 220 to 240,  $25 \pm 7$ ; 450 to 480 min,  $27 \pm 6$  nmol/min per 100 ml; NB baseline:  $-13 \pm 3$ ; 200 to 220 min,  $-13 \pm 4$ ; 220 to 240,  $-13 \pm 3$ ; 440 to 460 min,  $-14 \pm 3$  nmol/min per 100 ml;  $P = \text{NS}$ ). No change over time in either arterial or forearm exchange of AA was observed (data not shown).

### Discussion

Our study demonstrates that PD with dextrose alone or with the addition of AA significantly affects forearm protein dynamics in CAPD patients. Because the forearm is mainly represented by muscle, our data show that muscle is an immediate target for these metabolic effects. These effects, however, are divergent with regard to both the individual components of muscle protein turnover, *i.e.*, PS and degradation, and the final consequences on NB. The potential anabolic action of the moderate, systemic hyperinsulinemia that occurs during PD with dextrose is obscured by a parallel antianabolic effect induced by the decreased availability of AA for PS. In the present study, the increase in systemic insulin levels induced by glucose absorption was followed by a decrease in the rate of production of circulating phenylalanine measured across the forearm, reflecting a decrease in forearm proteolysis. However, no net forearm anabolic effect ensued, owing to a contemporary decrease in forearm PS. Data from the present study indicate that PD with dextrose induces acutely a new state in muscle protein dynamics, which is characterized by decreased turnover rates and a reduced efficiency. According to our results, the major acute effect of PD with dextrose is to restrain, rather than stimulate, muscle protein turnover. After the use of dextrose and AA, an increase in arterial AA availability was

associated with similar low values of muscle PB but with increased muscle PS. Therefore, our data indicate that the combined use of dextrose and AA results in a cumulative effect as a result of the suppression of endogenous PB (induced by insulin) and the stimulation of PS (induced by AA availability).

Insulin has been shown both *in vitro* (9) and *in vivo* (10) to be a powerful anabolic agent. Insulin suppresses BCAA metabolism (23) as well as protein degradation through specific pathways in muscle (24). Whereas uremia impairs insulin sensitivity with regard to glucose (25), insulin sensitivity regarding AA and whole-body protein metabolism seems to be unaltered in patients with ESRD (11,26). It is noteworthy that human muscle is extremely sensitive to insulin's antiproteolytic effect. For this effect, a dose-response curve has been demonstrated: infusions as low as to obtain insulin concentrations of approximately 30  $\mu\text{U/ml}$  maximally suppress ( $-40\%$ ) protein degradation (27). The effects of insulin on phenylalanine muscle balance are half-maximal at insulin concentrations of only 14  $\mu\text{U/ml}$ . Also in PD patients studied here, when dextrose alone was used in the peritoneal dialysate, rates of muscle protein degradation were inversely related to arterial insulin levels (Figure 2). Similar to the normal condition (25), the best fitting of the relation shows a curvilinear relationship with no decrease in protein degradation for increases in insulin levels above 30 to 35  $\mu\text{U/ml}$ . Therefore, PD, with its attendant hyperinsulinemia, theoretically could be followed by an anticatabolic effect. However, in the present study, the negative phenylalanine balance across the forearm observed in the basal, postabsorptive state remained unchanged despite a doubling of the insulin levels. In addition, the tracer data disclosed a marked decrement in phenylalanine kinetics. It is noteworthy that in previous studies in which insulin levels had been raised systemically in the high, physiologic range (60 to 90  $\mu\text{U/ml}$ ), a decrease in the estimate of PS (28,29) prevented the occurrence of a net anabolic effect in muscle tissue if the concurrent hypoaminoacidemia had not been avoided. In this setting, the decrease in AA availability derived from an insulin-dependent increase in AA uptake by extramuscle (29) or a decrease in the release from muscle (30) tissues. In our study, the decrease in AA levels observed with the use of dextrose was not associated with a diminished muscle release. According to our estimate, a large portion of this decrement could be accounted for by the removal by the peritoneal fluid. However, it cannot be excluded that an inhibition of AA release occurred in extramuscle tissues, such as splanchnic organs.

Patients with CRF display several abnormalities in postabsorptive blood and muscle AA profile, with a decrease in essential AA. In our study, PD with dextrose caused a further decrease in the abnormal arterial concentration of several AA. Besides leucine and valine, we observed a decrease also in arterial serine (in plasma) and tyrosine. Circulating and intracellular pools of these AA are low in uremia owing to decreased production by the kidney (31,32), and it is possible that these AA are limiting muscle PS in conditions of increased demand. We also observed a significant decrease in plasma arterial taurine levels. PD patients have a decrease in muscle

intracellular taurine, which correlates with plasma taurine levels (33). Taurine does not participate in PS; however, taurine depletion may cause a number of abnormalities at the cardiovascular and neurologic levels (34). Therefore, according to our findings, PD acts to restrain the already diminished circulating pools of these AA in uremia.

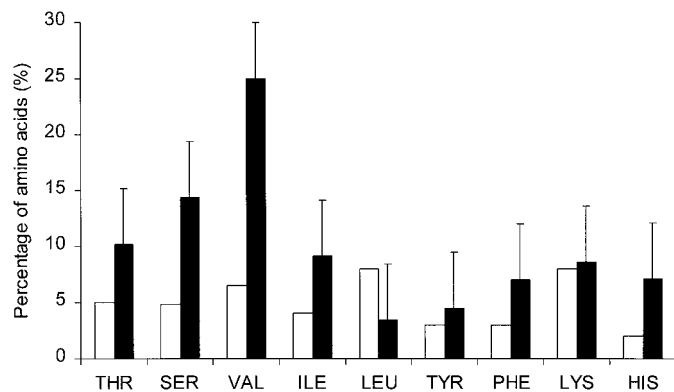
We have not been able to show any relationship between changes in blood glucose and protein turnover in our patients. In previous studies in which glucose was administered intravenously, an increase in insulin levels, with a decrease in protein degradation and unchanged PS, were observed (35). However, in a study in which endogenous insulin was controlled, neither parenteral glucose infusion rates nor plasma glucose concentrations affected whole-body proteolysis (36). Our data and those from the literature suggest that it is the insulin response rather than glucose *per se* that affects protein turnover.

Complementing the decrease in muscle PS and AA availability observed with dextrose alone, an increase in muscle PS with no significant variation in protein degradation occurred when arterial AA levels were raised by the use of dextrose combined with AA. This finding is in accordance with previous studies in which PS was shown to increase in the whole body (37) or in muscle (38) when AA levels were raised. The increase in AA levels during dextrose+AA was markedly dependent on their individual concentration in the dialysate. This is in keeping with previous observations obtained in children (38,39). Moreover, our data suggest that the amount of individual AA administered is a strong determinant of its arterial level and uptake by muscle. Alanine was an exception because this AA increased in blood only slightly even when it was present in sizable amounts in the dialysate. It is likely that this AA did not rise in blood because its release from the forearm decreased with the use of dextrose+AA. It is also of note that splanchnic organs are effective in removing alanine from the circulation (40). As also observed in our laboratory in previous studies in patients with CRF (40), variations in AA exchange across the forearm were related directly to changes in their arterial concentration. However, it is noteworthy that we have not been able to demonstrate a fully anabolic action by the use of dextrose+AA. Why was the stimulation of both mechanisms for muscle protein accretion, *i.e.*, a submaximal inhibition of muscle protein degradation and a stimulation of PS, not followed by a net anabolic response? One possible explanation is that the amount or the proportions of AA administered to our patients were too low to cause a sizable and appropriate increase in arterial supply to peripheral tissue and stimulate PS. Several reports have underscored the major role of AA availability in conditioning their transport and PS in muscle, both during hyperinsulinemia and independent of insulin concentration (37,39,41). In fact, the amount of AA given intraperitoneally was only 22 g, *i.e.*, the amount of AA provided by a small steak, and their arterial concentrations therefore were only slightly increased, in comparison to what is observed in CRF patients who received protein meals (40,42). If the total amount administered was low, then it is also possible that the formula of AA given was limited as to some AA. Available



data also indicate that PS requires a full complement of essential AA (43). Studies in healthy subjects indicate that limiting the availability of one of more essential AA, including BCAA, adversely affects whole-body PS but not proteolysis (44). Moreover, it has been shown recently that BCAA, mainly leucine, influence PS by modulating the availability of protein translation factors, an effect that is independent from insulin (45). We calculated the percentage variations in AA exchange across the forearm during PD with dextrose+AA *versus* dextrose alone, *i.e.*, the percentage of AA retained in muscle by the use of intraperitoneal AA, and put them in relation with the percentage of AA composition of muscle protein (Figure 3). Some AA (valine, serine, threonine, and isoleucine) were retained by muscle in large proportions relative to the muscle AA composition, whereas other AA (leucine, tyrosine, and lysine) were taken up by the forearm in more limited amounts. It is of note that the AA formula used in the present study contained relatively high amounts of valine and relatively low amounts of tyrosine and lysine. Our results show that even if incomplete, this solution may stimulate muscle PS and prevent the decrease in muscle PS induced by PD with dextrose alone. It would be of interest, however, to compare the effects with those of complete AA mixtures. It is possible that a more complete solution is more efficient in causing anabolism, therefore reducing other negative effects, such as azotemia and acidosis, and the use of AA like a caloric source, which have been observed with intraperitoneal AA (46).

It is of note that this study was performed during an overnight fast, a condition in which peripheral tissues are in a negative nitrogen balance owing to a continuous release of AA by muscle. The negative AA balance across the forearm in patients studied therefore must be considered as a physiologic condition and is not inherent to the uremic condition or to the treatment with PD. Basal rates of muscle protein turnover in patients studied were in the lower range of values observed in



**Figure 3.** The relation between changes in the AA balance across the forearm during PD with dextrose+AA *versus* dextrose alone. □, the composition of actin and myosin in skeletal muscle (49); ■, the percentage of AA retained in muscle during dextrose+AA *versus* dextrose alone. Some AA (valine, serine, threonine, and isoleucine) were retained by muscle in large proportions relative to the muscle AA composition, whereas some AA (leucine, tyrosine, and lysine) were taken up by the forearm in more limited amounts.

the normal condition both in the literature (17,26) and in our laboratory. In addition to being reduced, basal rates of muscle protein turnover in patients studied proved to be efficient. According to the model of forearm phenylalanine kinetics studied, as much as 60 to 65% of phenylalanine deriving from proteolysis is cycled back into PS in the normal condition (17,25,47). A figure even higher than this is found in PD patients observed in the present study, in which as much as 70% of phenylalanine deriving from PB was cycled back into phenylalanine disposal. Therefore, PD patients studied in the postabsorptive state seem to be minimally catabolic. This observation is in keeping with their well-preserved nutritional conditions. Our data indicate that in fasted patients, PD with dextrose induces acutely a change in the efficiency by which AA are cycled back into PS, as expressed by the net muscle protein balance unchanged in front of a decrease in protein turnover. One could speculate that a decrease in protein turnover and of its efficiency, as demonstrated by the present study during PD with dextrose, may result in a limited potential for net skeletal muscle protein accretion in uremia or predisposes to reduced anabolism when nutrient intake is diminished or during superimposed catabolic illnesses. However, the negative effects of dextrose reported here may not be manifested in regular PD patients who eat at least three times a day. Moreover, it is possible that the PD regimen that we used, which is similar to regimens used in patients who are treated with automated devices but is uncommon for the majority of PD patients, emphasized the metabolic effect of PD.

An estimate of the effects on muscle mass during the use of dextrose+AA in comparison with dextrose alone may be offered by the changes in forearm phenylalanine NB induced by the use of dextrose+AA. Considering that the mean fraction of forearm made up of muscle tissue is approximately 0.6, that forearm blood flow in forearm muscle is approximately 60 to 70% of total flow, that muscle is on the average 40% of body weight, and that phenylalanine content in proteins is approximately 280  $\mu\text{mol/g}$  protein (18,48), the change in net phenylalanine release from the forearm during the use of dextrose+AA *versus* dextrose alone would account for a less negative muscle balance of approximately 8 to 9 g/d.

There is evidence that muscle protein accretion is achieved through the combined effect of an energy-induced, insulin-mediated inhibition of PB and an AA-dependent stimulation of PS. Given the action in muscle induced by PD described in the present study, we hypothesize that in fasting PD patients, muscle mass could be maintained better by preventing, through continued AA supplementation, the dialysis-induced decrease in AA availability. Findings observed in the present study may also be relevant as regard to both the understanding of the pathophysiology of chronic renal failure and its treatment and the design of nutritional regimens that include the use of insulin and AA.

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