Overnight Branched-Chain Amino Acid Infusion Causes Sustained Suppression of Muscle Proteolysis

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Short-term (3 to 4 hours) infusion of branched-chain amino acids (BCAA) has been shown to suppress muscle protein breakdown. Whether these effects are sustained with chronic elevations of BCAA is not known. In the present study, we examined the effect of an overnight (16-hour) systemic BCAA infusion on whole-body and skeletal muscle amino acid metabolism, as assessed by simultaneously measured ^{3}H-phenylalanine and ^{14}C-leucine kinetics in eight normal volunteers; 10 overnight-fasted subjects studied during a 4-hour saline infusion served as controls. Overnight BCAA infusion increased plasma BCAA concentrations by fivefold to eightfold, and this was associated with a 20% to 60% decline in arterial concentrations of other amino acids. For Phe, this decline was mediated by a reduction in the systemic rate of appearance ([R_a] 0.38 ± 0.03 *v* 0.60 ± 0.01 μmol/kg/min for BCAA and saline, respectively, $P < .001$). Endogenous Leu R_a, calculated more indirectly as the difference between the total Leu R_a and the unlabeled Leu infusion rate, did not differ between groups. In the forearm, overnight BCAA infusion resulted in a diminished net release of Phe (−3 ± 2 *v* −18 ± 4 [saline] nmol/min/100 mL, $P < .02$), and BCAA balance became markedly positive (751 ± 93 *v* −75 ± 30, $P < .001$). The diminished net forearm Phe release was accounted for by a decrease in local Phe R_a ($P < .02$). As with the systemic endogenous Leu R_a, forearm Leu R_a was not reproducibly affected by infused BCAA. These findings suggest a need for caution in the application of amino acid tracer kinetic methods when tracer and unlabeled tracee are infused simultaneously. In conclusion, overnight BCAA infusion caused a sustained decline in most plasma amino acids and in net forearm release of Phe, effects attributable to a sustained suppression of whole-body and muscle proteolysis.

SHORT-TERM (3 to 4 hours) infusion of branched-chain amino acids (BCAA) or leucine alone decreases plasma concentrations of other essential and nonessential amino acids.[1-6] This decline stabilizes after 1 to 2 hours. The decrease in plasma phenylalanine in this setting involves a general suppression of its rate of appearance (R_a) into the systemic circulation and a specific inhibition of its release from forearm muscle.[3] These results suggest that acute BCAA infusion suppresses whole-body and skeletal muscle proteolysis, at least in the short term.

Insulin, epinephrine, and insulin-like growth factor I infused for 2 to 4 hours have also been shown to decrease plasma amino acid concentrations, and for each the decline has been attributed to a decrease in body tissue protein breakdown.[7-10] Epinephrine's effect on plasma amino acid levels develops over 2 hours and can persist for periods of 8 to 9 hours.[10] However, its effects on whole-body proteolysis are seen within the first 2 hours and quickly wane beyond that period. Whether the antiproteolytic action of BCAA demonstrated in response to short-term infusions can be sustained with long-term infusions is not known.

Acute changes in amino acid pool size can complicate the interpretation of amino acid tracer kinetics, especially when one attempts to relate such kinetic observations to the metabolism of body protein. For example, in previous short-term BCAA infusion studies, proteolysis was inhibited and net protein anabolism (synthesis > breakdown) was observed, a condition that should require provision of all essential amino acids, not just the BCAA. One possible explanation for these results is that endogenous free amino acid pools can, in the short term, provide the other essential amino acids necessary for net accretion of tissue protein. This interpretation is consistent with the observed acute declines in plasma amino acid concentrations. However, with more-prolonged BCAA infusion, once free amino acid concentrations stabilize, whether inhibition of proteolysis and a state of net anabolism can persist is not known.

In the present investigation, we administered an overnight (16-hour) infusion of BCAA to determine whether the antiproteolytic and net anabolic effects observed in the short term are sustained under conditions where free amino acid pools have been stable for a long period.

SUBJECTS AND METHODS

Subjects

Eighteen healthy volunteers (aged 18 to 34 years; mean ± SEM, 23 ± 1) were studied. Eight subjects (five women, three men) received a 16-hour overnight infusion of BCAA. Ten subjects (two women, eight men) were studied as outpatients with a 4-hour saline infusion and served as statistical controls. Except for the fact that saline-control subjects arrived at the research center on the morning of the tracer study and the BCAA group were hospitalized overnight, all study procedures for the two groups were the same. The 12-hour duration of overnight fasting was verified by interview in the saline-control subjects. All subjects were within 15% of their ideal body weight (Metropolitan Life Insurance Tables, 1983). None had a history of endocrine or other major organ system disease, and none were taking any medication.

The purpose and potential risks of the study were explained to each subject, and informed written consent was obtained before the study. The experimental protocol was reviewed and approved

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by the Human Investigation Committee of the Yale University School of Medicine.

Experimental Design

All subjects were fasted beginning at 8:00 PM on the evening before tracer infusion and were studied in the postabsorptive state. The eight subjects who received BCAA infusions were admitted to the Yale Clinical Research Center the day before the tracer study. Starting at 8:00 PM, a 16-hour overnight infusion of an equimolar mixture of leucine, isoleucine, and valine (Branchamin, 4% in water; Clintec International, Deerfield, IL; kindly provided by Dr D. Madsen) was begun via an antecubital vein. The infusion rate of each amino acid was 1.66 μmol/kg/min. At 8:00 AM, catheters were inserted into a brachial artery and retrogradely into an ipsilateral deep forearm vein of the arm opposite to the one receiving the BCAA or saline infusion. Patency of the catheters was maintained by slow infusion of normal saline. Starting at 8:30 AM, subjects received a 3.5-hour primed, continuous infusion of L-[ring-2,6-^{3}H]phenylalanine and L-[1-^{14}C]leucine via the antecubital vein. The dose of [^{14}C]leucine in saline studies was approximately 8 μCi (prime) followed by 0.13 μCi/min, with a 2.5-μCi bolus of [^{14}C]sodium bicarbonate to prime the body bicarbonate pools. In BCAA infusion studies, the L-[1-^{14}C]leucine dosage was increased (~22 μCi, 0.37 μCi/min), as was the [^{14}C]sodium bicarbonate bolus (4 μCi), to enable α-ketoisocaproate (KIC) specific activity (SA) determinations. The L-[ring-2,6-^{3}H]phenylalanine dosage (~32 μCi prime followed by 0.52 μCi/min) was the same in both infusion groups. After a 150-minute tracer equilibration period, arterial and deep-venous blood samples were obtained during the last hour of BCAA infusion at 15-minute intervals (150, 165, 180, 195, and 210 minutes), and at 30-minute intervals (150, 180, and 210 minutes) for saline-infused subjects. For 2 minutes before and during withdrawal of each deep-venous blood sample, a pediatric sphygmomanometer cuff was inflated on the wrist to 200 mm Hg to exclude blood flow to the hand. Forearm plasma flow was measured immediately after each arteriovenous sampling interval by dilution of indocyanine green dye (Hynson, Wescott, and Dunning, Baltimore, MD), which was infused intraarterially for 5 minutes with the wrist cuff inflated; blood flow was derived by dividing plasma flow by (1-hematocrit). Forearm volume was measured by water displacement.

For measurement of whole-body leucine oxidation, expired CO_2 was collected by bubbling expired air through a hyamine hydroxide trapping solution that contained a phenolphthalein indicator titrated to change color when a known amount of CO_2 was trapped. The total rate of CO_2 production was measured with the ventilated-hood technique using a Deltatrac Metabolic Measurement Monitor (SensorMedics, Anaheim, CA).

Calculations

Net forearm balances and forearm and whole-body amino acid kinetics were calculated as previously described.[3,11] Briefly, net forearm balance for a given substrate can be calculated using the Fick principle by knowing the arterial [a] and venous [v] concentration of the substrate and the forearm blood flow (f). For a given amino acid, the net balance reflects the contribution of two simultaneous processes: the uptake or rate of disposal (R_d) of arterial substrate and the release or R_a. That is,

$$\text{net balance} = ([a] - [v]) \times f = R_d - R_a. \qquad \text{Eq 1}$$

For phenylalanine and leucine, tissue disposal can be calculated from the measured fractional extraction (E) of tracer, ie,

$$R_d = E \times [a] \times f, \qquad \text{Eq 2}$$

where E is the arteriovenous difference in tracer radioactivity divided by the arterial tracer radioactivity concentration (all in disintegrations per minute per milliliter). The release of phenylalanine or leucine can be calculated using the difference of the net balance and R_d, ie,

$$R_a = (E \times [a] \times f) - \{([a] - [v]) \times f\}.$$

which reduces to the expression

$$R_a = f \times [v] \times (1 - SA_v/SA_a) \qquad \text{Eq 3}$$

where SA_a and SA_v denote the SA (disintegrations per minute per nanomole) of the amino acid in artery and vein, respectively.

It should be noted from equations 1 and 3 that tissue kinetics (R_a and R_d) can be fully defined by measuring [a], [v], f, and the arteriovenous SA ratio. Hence, if the venous to arterial SA ratio can be determined, measurement of absolute radioactivity concentrations in artery and vein is not essential. This approach was used in calculating phenylalanine kinetics, using measurements of arterial and venous phenylalanine SA by a high-performance liquid chromatography technique.[3,11]

Whole-body leucine and phenylalanine flux rates (Q) were calculated from the rate of tracer infusion (disintegrations per minute infused per minute) divided by the mean steady-state amino acid SA in arterial blood:

$$Q = IR/SA_a. \qquad \text{Eq 4}$$

Using the stochastic model, leucine kinetics can be defined by the equation Q = S + C = B + I, where Q is the total leucine flux, S is the rate of leucine incorporation into protein (nonoxidative leucine disposal), C is the rate of leucine oxidation, B is the rate of leucine release from protein (endogenous leucine R_a), and I is the rate of exogenous leucine infusion. During BCAA infusion, B (endogenous leucine R_a) is equal to Q minus I.

Leucine oxidation (C) was calculated as

$$C = \dot{V}(^{14}CO_2)/(SA_a \text{ leucine} \times 0.8), \qquad \text{Eq 5}$$

where $\dot{V}(^{14}CO_2)$ is the rate of production of $^{14}CO_2$ (in disintegrations per minute per minute), calculated from the product of the steady-state SA of expired CO_2 (in disintegrations per minute per millimole) and the total CO_2 production rate determined using the ventilated hood. The factor 0.8 corrects for nonexpired $^{14}CO_2$ generated from L-[1-^{14}C]leucine oxidation but retained within body bicarbonate stores. Nonoxidative leucine disposal (S) was calculated as

$$S = Q - C. \qquad \text{Eq 6}$$

In subjects receiving BCAA infusion, leucine kinetics were also calculated by the reciprocal pool method, in which arterial SA of KIC, measured during the last hour of the infusion, was used in place of leucine SA in equations 4 and 5.

Analytical Methods

Blood glucose concentration, plasma insulin concentration, and leucine and phenylalanine SA were determined as previously described.[3,11] Concentrations of selected amino acids (acidic and neutral) were measured in sulfosalicylic acid extracts of whole blood using an automated ion-exchange chromatographic technique (Dionex D-500, Sunnyvale, CA). Basic amino acid levels were not measured.

Measurements of branched-chain ketoacids (BCKA), KIC, α-ketomethylvalerate, and α-ketoisovalerate levels and KIC SA were made by a modification of the method previously described by

Nissen et al.[12,13] KIC SA was measured from samples taken during the last hour of BCAA infusion.

Data Presentation and Statistical Analysis

All data are presented as the mean ± SEM. Results from samples taken during the last hour of saline or BCAA infusion were averaged for each subject. Statistical comparisons between BCAA- and saline-infused groups were performed using an unpaired *t* test.

RESULTS

Effect of Overnight BCAA Infusion on Substrates and Hormones

As shown in Table 1, overnight (16-hour) BCAA infusion caused a marked (fivefold to eightfold) increase in concentrations of leucine, isoleucine, and valine ($P < .001$ *v* control), whereas concentrations of most other measured essential and nonessential amino acids were decreased significantly. Concentrations of BCKA derivatives also increased (approximately twofold to threefold) during BCAA infusion, but by less than for BCAA. BCAA infusion was associated with a reduction in arterial glucose levels (3.8 ± 0.1 mmol/L) as compared with the saline controls (4.4 ± 0.1 mmol/L, $P < .05$). Insulin concentrations were not significantly different in the two groups (8.7 ± 1.0 for BCAA infusion *v* 6.3 ± 1.0 μU/mL for saline control).

Effect of Overnight BCAA Infusion on Forearm Muscle Metabolism

Net forearm balances of amino acids and BCKA are listed in Table 1. In contrast to the net release of amino acids in the postabsorptive state, during BCAA infusion net forearm balances for leucine, isoleucine, and valine were markedly positive ($P < .001$). Accompanying the marked increase in forearm BCAA uptake was a net release of BCKA from the forearm. Net release of phenylalanine and methionine decreased significantly during BCAA infusion ($P < .02$).

Figure 1 depicts arterial phenylalanine and leucine concentrations and SAs during the last hour of BCAA and saline infusion. Arterial concentrations and SAs of phenylalanine and leucine were constant during this period, as were the corresponding venous values (all varied by <5%). This permitted the application of steady-state calculations of amino acid kinetics.

Table 2 lists forearm phenylalanine and leucine kinetic responses to BCAA infusion. The reduced net forearm phenylalanine output during BCAA infusion was attributable to a marked (43%) reduction of forearm phenylalanine R_a ($P < .02$). As noted with short-term (3 to 4 hours) BCAA infusions,[3] forearm phenylalanine R_d tended to decline slightly (P = NS) (Table 2).

For leucine, the marked increase in net uptake was due to a threefold increase in leucine R_d (105 ± 10 *v* 401 ± 63 nmol/min/100 mL, $P < .001$). Leucine R_a was not demonstrably affected by BCAA infusion.

Effect of BCAA on Whole-Body Amino Acid Kinetics

Table 3 lists the effects of prolonged BCAA infusion on whole-body amino acid kinetics as compared with saline infusion. Consistent with the forearm phenylalanine kinetic data, whole-body phenylalanine flux was reduced by 37%

Table 1. Arterial Blood Concentrations and Net Forearm Balance of Amino Acids and BCKA During an Overnight BCAA Infusion as Compared With Values in Saline-Infused Controls

	Arterial Concentration (μmol/L)			Net Forearm Balance (nmol/min/100 mL)		
	BCAA	Control	*P*	BCAA	Control	*P*
Alanine	289 ± 39	264 ± 18		−178 ± 41	−164 ± 25	
Glycine	237 ± 9	292 ± 18	<.03	−1 ± 9	−24 ± 15	
Serine	117 ± 4	128 ± 8		−24 ± 9	14 ± 6	
Threonine	88 ± 7	134 ± 14	<.01	−17 ± 6	−26 ± 6	
Phenylalanine	29 ± 1	50 ± 3	<.001	−3 ± 2	−18 ± 4	<.02
Methionine	9 ± 0	12 ± 1		−4 ± 2	−15 ± 3	<.02
Tyrosine	20 ± 1	50 ± 3	<.001	−14 ± 5	−17 ± 2	
Valine	1,104 ± 50	196 ± 11	<.001	325 ± 43	−30 ± 17	<.001
Isoleucine	468 ± 27	54 ± 4	<.001	196 ± 25	−16 ± 7	<.001
Leucine	561 ± 29	111 ± 6	<.001	213 ± 26	−27 ± 7	<.001
Taurine	151 ± 10	216 ± 10	<.003	78 ± 33	38 ± 26	
Asparagine	46 ± 4	70 ± 2	<.001	−13 ± 4	−10 ± 4	
Cysteine	25 ± 0	32 ± 1	<.001	11 ± 1	18 ± 3	
Glutamate	141 ± 14	163 ± 12		87 ± 7	74 ± 11	
Glutamine	519 ± 14	417 ± 13	<.001	−168 ± 13	−140 ± 36	
Total BCAA	2,134 ± 106	361 ± 19	<.001	751 ± 93	−75 ± 30	<.001
Total amino acids	3,834 ± 81	2,189 ± 46	<.001	589 ± 136	−347 ± 113	<.001
KIC	72 ± 6	36 ± 2	<.005	−30 ± 3	2 ± 4	<.001
KIV	10 ± 1	12 ± 1		−7 ± 1	6 ± 3	<.001
KMV	95 ± 8	21 ± 2	<.005	−4 ± 4	3 ± 2	
Total BCKA	177 ± 14	69 ± 3	<.005	−41 ± 7	9 ± 5	<.001

Abbreviations: KIV, α = ketoisovalerate; KMV, α = ketomethylvalerate.

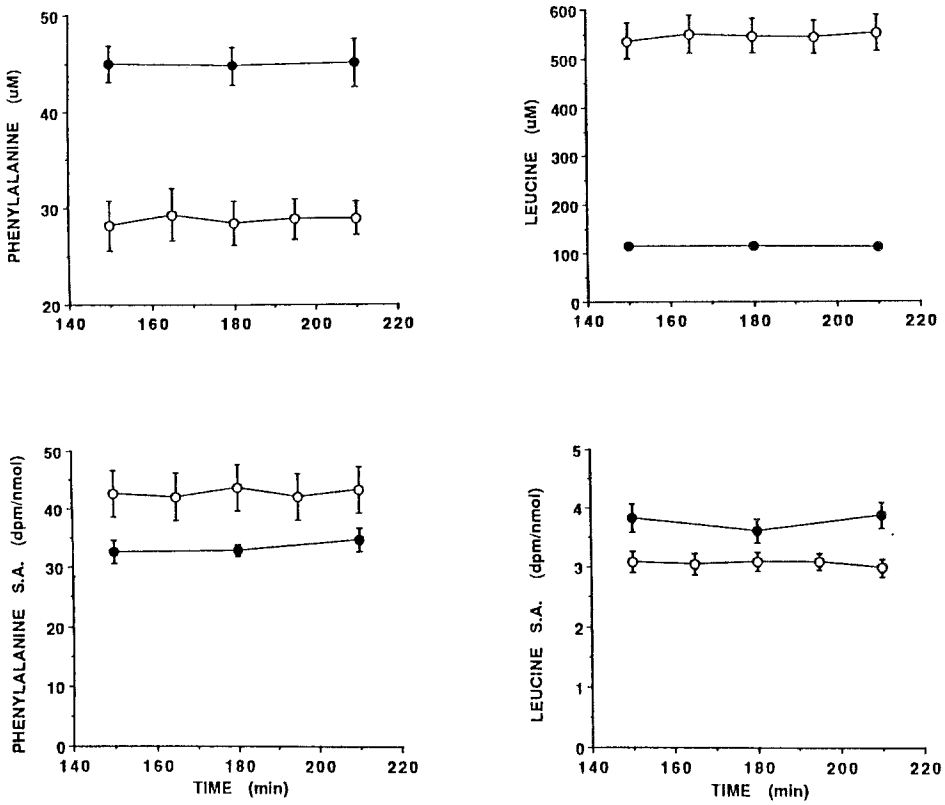

Fig 1. Arterial blood concentrations and SAs of phenylalanine and leucine during the last hour of BCAA (○) or saline (●) infusion. All measurements exhibited steady-state conditions.

($P < .001$) during BCAA as compared with saline infusion. BCAA infusion caused a marked 2.5-fold increase in total leucine flux as measured with the primary pool model. This was associated with significant increases in leucine oxidation (4.7-fold) and nonoxidative leucine disposal (2.2-fold). Endogenous leucine R_a, calculated as the total flux minus the exogenous leucine infusion rate, was not significantly affected.

Table 3 also lists the estimates of leucine flux obtained using the reciprocal pool model at the end of the 16-hour BCAA infusion. Our observed KIC to leucine SA ratio of 0.61 was consistent with previously published studies that reported ratios of 0.65 to 0.80 under a wide range of physiologic conditions.[14,15]

DISCUSSION

The present study demonstrates that overnight (16-hour) infusion of BCAA reduces circulating levels of other essential amino acids, and that this is associated with a suppression of net forearm release of acidic and neutral amino acids (basic amino acid levels were not measured). Quantitatively, the hypoaminoacidemia observed in the present study at 16 hours is similar to that found previously after only 2 to 3 hours of BCAA infusion at this dose.[3] This suggests that between 3 and 16 hours of BCAA infusion, plasma and tissue pools of free amino acids are at or close to steady state. Thus, results of forearm and whole-body amino acid kinetics under these circumstances are unlikely to be influenced by the acute changes in free amino acid pools that may complicate the interpretation of short-term observations.

Table 2. Forearm Amino Acid Kinetics During BCAA or Saline Infusion (nmol/min/100 mL)

	BCAA Infusion	Saline Infusion	*P*
Phenylalanine			
Net balance	−3 ± 2	−18 ± 4	<.02
R_a	31 ± 3	54 ± 7	<.02
R_d	27 ± 2	36 ± 5	NS
Leucine			
Net balance	213 ± 26	−27 ± 7	<.001
R_a	188 ± 42	132 ± 17	NS
R_d	401 ± 63	105 ± 10	<.001

Table 3. Whole-Body Leucine and Phenylalanine Kinetics During BCAA or Saline Infusion (μmol/kg/min)

	BCAA Infusion	Saline Infusion	*P*
Phenylalanine			
Flux (Q = B)	0.38 ± 0.03	0.60 ± 0.01	<.001
Leucine			
Primary pool			
Total flux (Q)	3.09 ± 0.10	1.26 ± 0.05	<.001
Oxidation	0.80 ± 0.04	0.17 ± 0.01	<.001
Nonoxidative disposal (S)*	2.28 ± 0.10	1.05 ± 0.05	<.001
Endogenous R_a†	1.43 ± 0.10	1.26 ± 0.05	NS
Reciprocal pool			
Total flux (Q)	5.20 ± 0.45		
Oxidation	1.33 ± 0.08		
Nonoxidative disposal (S)*	3.87 ± 0.40		
Endogenous R_a†	3.54 ± 0.45		

*Total flux minus oxidation.

†Total flux minus exogenous leucine infusion rate of 1.66 μmol/kg/min.

The phenylalanine kinetic results suggest that the reduction in circulating amino acid levels is mediated through a decrease in tissue proteolysis, rather than through an increase in tissue incorporation of amino acids into protein. In keeping with the results of previous short-term studies of BCAA infusion,[3] forearm phenylalanine R_a, a measure of skeletal muscle protein breakdown, was suppressed by 43% and whole-body flux of phenylalanine was similarly reduced (by 37%) in BCAA- as compared with saline-infused subjects (Table 3). Thus, in normal postabsorptive man, overnight BCAA infusion causes a sustained diminution in the rate of production of circulating phenylalanine in both skeletal muscle and the whole body. To our knowledge, this is the first demonstration that the inhibitory effect of BCAA on skeletal muscle and whole-body proteolysis in man is sustained during more prolonged infusion.

The molecular mechanism mediating the observed antiproteolytic effect of BCAA is unknown. The similarity of plasma insulin concentrations in BCAA-infused (8.7 μU/mL) and saline control (6.3 μU/mL, P = NS) groups argues against an indirect effect mediated through stimulation of insulin secretion. Although we cannot entirely exclude a small contribution of undetected mild hyperinsulinemia, the magnitude of the amino acid decrease (and suppression of phenylalanine R_a) far exceeds that which would be predicted for minimal changes in insulin within the normal postabsorptive range. A simple fuel effect also seems unlikely as the sole mechanism, since the infused BCAA (0.04 g/kg/h), even if entirely oxidized, would meet less than 20% of typical resting energy requirements (~1 kcal/kg/h). Our findings are consistent with a direct modulatory role of one or more BCAA (and/or their ketoacid derivatives) on skeletal muscle protein turnover.

Current understanding of the effects on amino acid and protein metabolism of other anabolic agents, such as insulin,[8,9,15] insulin-like growth factor I,[7] and growth hormone,[16] has been largely based on short-term infusion studies of several hours' duration. In addition to the difficulties of interpreting acute amino acid kinetic changes in the setting where free amino acid pool sizes may be changing,[17] it remains unknown whether short-term effects observed in such studies can be sustained for more-prolonged periods. The effects of epinephrine, for example, on body amino acid kinetics appear to be transient,[10] whereas other agents may evoke compensatory responses that alter their effects over time, such as the development of hyperinsulinemia with long-term glucocorticoid administration. These considerations underscore the importance of examining the chronicity of the influence of various factors known to regulate amino acid and protein metabolism in the short term.

Analysis of whole-body leucine kinetics showed an increase in nonoxidative leucine disposal, such that it significantly exceeded leucine R_a, using either the primary or reciprocal pool approach. This finding implies that prolonged BCAA infusion results in sustained net body protein anabolism (ie, synthesis > breakdown), analogous to that seen with a 3-hour BCAA infusion.[3] However, such a result is more perplexing in the face of the more-prolonged duration of the present infusion and the steady-state plasma concentrations of those essential amino acids that were not infused. During short-term BCAA or leucine infusion, net body protein anabolism could be supported by drawing from free essential amino acids in extracellular and intracellular pools, thus explaining the decline in plasma concentrations of these amino acids. However, with the more-prolonged BCAA infusion used here, only three of seven essential amino acids are provided and there is no apparent source for the other essential amino acids required to sustain net protein accretion over the period from 3 to 16 hours.

Indeed, a net anabolic effect of overnight BCAA infusion is not confirmed when one examines forearm skeletal muscle protein metabolism. Although overnight BCAA infusion did reduce the net release of most acidic and neutral amino acids from forearm muscle (Table 1), balances remained near zero or slightly negative; only BCAA themselves showed a net uptake. The leucine tracer kinetics suggest that net forearm uptake of leucine was due to a significant increase in local leucine R_d. In the absence of similar changes in net forearm balance of the other essential amino acids or similar changes in forearm phenylalanine R_d, this suggests that metabolism, via transamination with release of ketoanalogs (Table 1) and via oxidation, plays the major role in forearm BCAA disposal under these conditions. Local leucine R_a was not significantly affected by BCAA infusion. However, it should be noted that with elevated amino acid levels, the forearm tracer measurement becomes less sensitive for detecting changes in turnover, since at any given turnover rate the difference between arterial and venous SA will diminish as the tracee concentration increases (see equation 4).

Unlike leucine, phenylalanine is not catabolized in skeletal muscle, making it particularly suitable for the study of muscle protein turnover. In the present setting, moreover, phenylalanine kinetics offer the additional advantage of using as tracer an essential amino acid distinct from those being infused. This enables a more straightforward analysis of endogenous amino acid kinetics than is possible using leucine as a tracer. Thus, whole-body phenylalanine flux, or R_a, a measure of body protein breakdown, is directly calculated in this setting from the measured plasma phenylalanine SA, as compared with endogenous leucine R_a, which requires subtraction of the exogenous leucine infusion rate from the measured total leucine flux. In the present study, the exogenous leucine infusion rate (1.66 μmol/kg/min) was substantial and accounted for the major share of the total leucine flux (3.09 μmol/kg/min). Hence, changes in endogenous leucine turnover would be less sensitively detected by this technique, since they would have relatively less effect on total leucine flux. This inherent insensitivity may account for the inability to demonstrate any significant change in endogenous leucine flux during BCAA infusion, in the face of an unequivocal suppression of phenylalanine flux. Alternatively, amino acid kinetics may be prone to systematic errors of uncertain origin in the

setting of high rates of exogenous infusion of unlabeled tracee, much like the situation for certain glucose tracer kinetic methods.[18,19] Similar weaknesses in the interpretation of phenylalanine tracer kinetics would be expected in the setting of high-dose phenylalanine infusion. In any case, it should be underscored that phenylalanine kinetic results during BCAA infusion were consistent in the whole body and in skeletal muscle, indicating in both sites a sustained antiproteolytic effect of BCAA.

In summary, overnight BCAA infusion in normal man suppressed whole-body and forearm muscle proteolysis, as measured by phenylalanine kinetics, without stimulating muscle protein synthesis. In the setting of exogenous amino acid administration, endogenous amino acid and protein kinetics can be best assessed using a tracer amino acid distinct from those being infused.

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