Influence of protein intake and training status on nitrogen balance and lean body mass

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TARNOPOLSKY, MARK A., J. DUNCAN MACDOUGALL, AND STEPHANIE A. ATKINSON. *Influence of protein intake and training status on nitrogen balance and lean body mass.* J. Appl. Physiol. 64(1): 187–193, 1988.—The present study examined the effects of training status (endurance exercise or body building) on nitrogen balance, body composition, and urea excretion during periods of habitual and altered protein intakes. Experiments were performed on six elite bodybuilders, six elite endurance athletes, and six sedentary controls during a 10-day period of normal protein intake followed by a 10-day period of altered protein intake. The nitrogen balance data revealed that bodybuilders required 1.12 times and endurance athletes required 1.67 times more daily protein than sedentary controls. Lean body mass (density) was maintained in bodybuilders consuming 1.05 g protein $\cdot$ kg$^{-1} \cdot$ day^{-1}. Endurance athletes excreted more total daily urea than either bodybuilders or controls. We conclude that bodybuilders during habitual training require a daily protein intake only slightly greater than that for sedentary individuals in the maintenance of lean body mass and that endurance athletes require daily protein intakes greater than either bodybuilders or sedentary individuals to meet the needs of protein catabolism during exercise.

resistance exercise; endurance exercise; urea excretion; lean body mass

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RECOMMENDED PROTEIN INTAKES for athletes undergoing daily training are a controversial issue. Suggested protein intakes range from those slightly above that for nonathletic individuals (12, 27), to values that are twice (20) or even four times (5, 16) that amount. A number of factors could have a significant influence on the protein requirement for an athlete, including the state of training (12), the type and volume of training (9, 16), the energy density of the diet (13, 26), and the carbohydrate content of the diet (25).

The question of increased protein requirements for athletes has been raised recently due to studies that have demonstrated that both endurance and resistance exercise may ultimately increase net protein utilization. The increase in contractile protein which results from heavy resistance training (22) suggests that protein intakes must exceed basal levels at some time to supply the amino acids for this process. Evidence to support this has been demonstrated in studies of elite weight lifters (16), subjects performing isometric exercises (8, 27), and power lifting (9). In contrast, it has also been demonstrated that a body building program did not increase

either urea or 3-methylhistidine excretion in young men (15).

An increase in protein oxidation during endurance exercise has been inferred from studies using a labeled leucine infusion technique (10, 18, 24, 28). In support of this, an increased urea excretion in association with endurance exercise has been observed in some (3, 9, 18) but not all (29) studies.

The recommended nutrient intake (RNI) for protein for Canadian males between 18 and 24 yr is 0.82 g $\cdot$ kg$^{-1} \cdot$ day^{-1} of mixed protein and in the United States the recommended daily allowance is 0.80 g kg$^{-1} \cdot$ day^{-1} of mixed protein (7, 23). At the present time no allowance is made in these recommended intakes for individuals undergoing either endurance or resistance training. The purpose of this study was to examine the effect of consuming habitual and altered protein intakes in excess of the RNI on urea excretion and nitrogen balance in endurance athletes, bodybuilders, and sedentary controls.

METHODS

Subjects. Three groups of six male volunteers gave their informed consent to serve as subjects for this study. *Group 1* was composed of sedentary controls (S); *group 2* was composed of elite endurance athletes (EA) who had been training for at least 5 yr as runners or nordic skiers; and *group 3* was composed of bodybuilders (BB) who had been training intensively for at least 3 yr and had not used anabolic steroids for the past 2 yr. At the time of the study both active groups were in a maintenance phase of training, with the EA running daily (>125 km/wk) and the BB training ~75 min/day.

Design. Each group participated in two different experiments. *Experiment A* investigated the nitrogen balance (NBAL) of subjects consuming their habitual dietary intakes. *Experiment B* investigated the NBAL of subjects on a diet of altered protein intake and immediately followed *experiment A*. The design of the experiments was identical, with dietary protein intake being the only parameter altered. Each experiment included a 10-day adaptation period followed by a 3-day NBAL period (Fig. 1).

For the adaptation period in *experiment A*, subjects followed a prescribed dietary and training regime which was representative of each groups' "habitual" mean intake and exercise program. Although the subjects were not randomized, it was felt that dietary compliance would

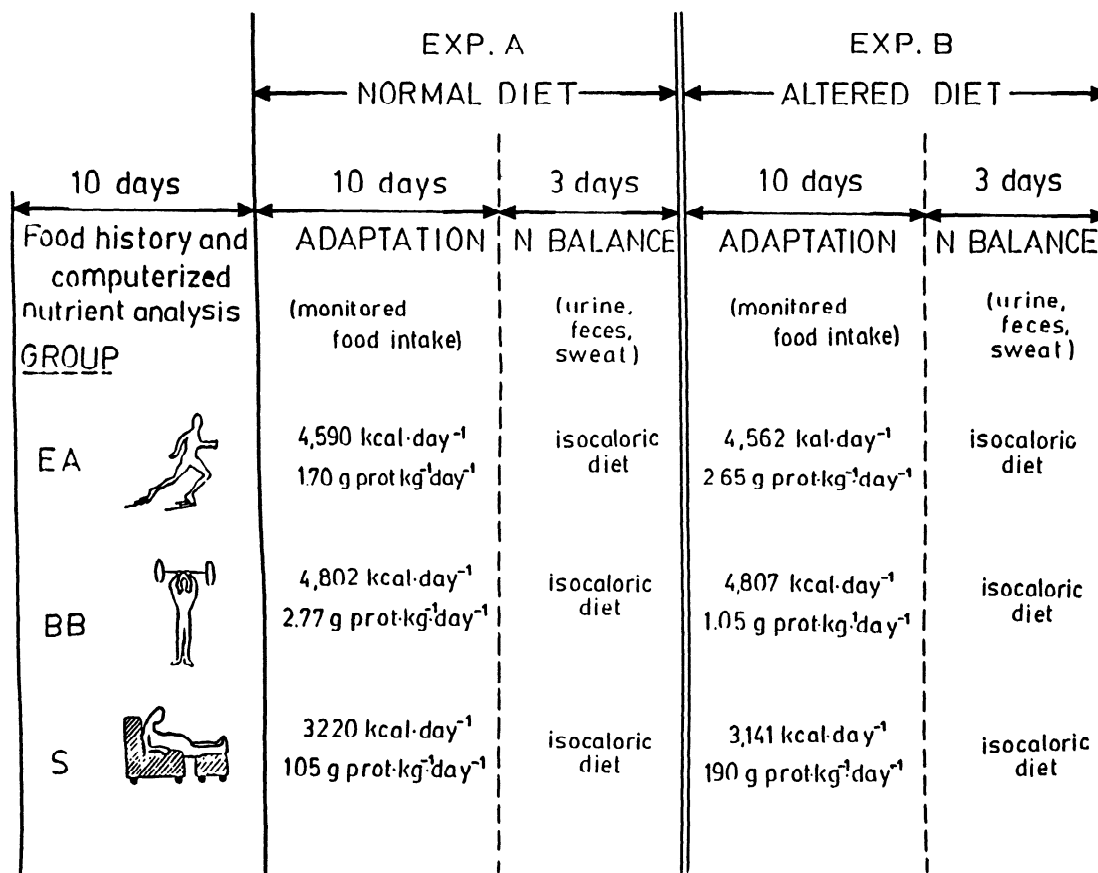| | EXP. A NORMAL DIET | | EXP. B ALTERED DIET | |
10 days	10 days	3 days	10 days	3 days
Food history and computerized nutrient analysis GROUP	ADAPTATION (monitored food intake)	N BALANCE (urine, feces, sweat)	ADAPTATION (monitored food intake)	N BALANCE (urine, feces, sweat)
EA	4,590 kcal·day^{-1} 1.70 g prot kg^{-1}day^{-1}	isocaloric diet	4,562 kal·day^{-1} 2.65 g prot·kg^{-1}day^{-1}	isocaloric diet
BB	4,802 kcal·day^{-1} 2.77 g prot·kg^{-1}day^{-1}	isocaloric diet	4,807 kcal·day^{-1} 1.05 g prot·kg·day^{-1}	isocaloric diet
S	3220 kcal·day^{-1} 105 g prot·kg^{-1}day^{-1}	isocaloric diet	3,141 kcal·day^{-1} 190 g prot·kg^{-1}day^{-1}	isocaloric diet

FIG. 1. Experimental design for all subject groups. EA, endurance athletes; BB, bodybuilders; S, sedentary individuals.

be maximized if all groups started on their respective habitual intake in *experiment A*. The diet prescription was based on the average nutrient composition of the habitual diet for each group, as derived from analysis of 7-day food records collected just before the initiation of the study. The average daily training workout was calculated from subjects' training logs.

For the 10-day adaptation in *experiment B*, subjects maintained their training as in *experiment A* while the protein content of their respective diets was altered. S and EA groups increased, whereas BB decreased their habitual protein intakes. The diets were isocaloric between experiments and were eucaloric (as determined from diet records) (Fig. 1). During the adaptation diet periods food was distributed to the subjects and self-prepared in strict accordance with a daily menu.

During the balance periods, food portions representative of each group's habitual intake (*experiment A*) or an altered protein intake (*experiment B*) were precooked (when necessary), weighed, and packaged before distribution to the subjects. Duplicate portions of these diets were kept for chemical analysis. Five percent of each duplicate sample was homogenized, lyophilized, and stored at −20°C for subsequent total nitrogen (TN) and energy analysis. The diets were lacto-ovo-vegetarian in composition and included a liquid supplement, Ensure Plus (Ross Laboratories, Montreal). During the high-protein intake periods, the diets were supplemented with

a soy protein powder (Shaklee Canada, Burlington, Ont), and the amount of high-carbohydrate foods (muffins and granola bars) was decreased to keep the diets isocaloric. All diets met the 1983 Canadian RNI standards for protein, energy, vitamins, and minerals (7). The detailed nutrient analysis of the diets is present in Table 1.

To determine TN excretion, three sequential 24-h urine collections, 72-h fecal collections, and representative resting and exercise sweat secretion samples were obtained. The daily urines were collected into 3-liter containers that had been acid washed, rinsed with deionized water, and treated with 5 ml of glacial acetic acid. The collected urine was kept at <5°C by the subjects and delivered to the testing center within 24 h. After volume determination, aliquots were taken and stored for subsequent TN and urea nitrogen (UN) analysis.

Fecal samples collected between carmine markers were kept frozen by the subjects and delivered to the testing center within 24 h of the end of each balance period. Each fecal collection was weighed, diluted with an equal weight of deionized water, and homogenized. Aliquots were taken and stored for subsequent TN analysis. The sweat collections were obtained by use of a whole-body washdown technique modified from that described by Lemon et al. (19) and recently validated by Lemon et al. (21).

During *days 1* and *2* of the balance periods, EA and

TABLE 1. *Diet summary*

Types of Intakes		Sedentary	Bodybuilders	Endurance Athletes
Maintenance energy, kcal/day	LP	3,222±39*	4,807±21	4,539±18*
	HP	3,141±41	4,802±22*	4,562±26
Protein, $g \cdot kg^{-1} \cdot day^{-1}$ (% energy)	LP	1.1±0.04* (11)	1.0±0.02 (7)	1.7±0.03* (12)
	HP	1.9±0.04 (19)	2.7±0.02* (19)	2.7±0.02 (18)
Fat, $g \cdot kg^{-1} \cdot day^{-1}$ (% energy)	LP	1.2±0.05* (26)	2.2±0.08 (33)	1.8±0.07* (26)
	HP	1.3±0.07 (28)	2.1±0.08* (32)	1.8±0.07 (26)
Carbohydrates, $g \cdot kg^{-1} \cdot day^{-1}$ (% energy)	LP	6.7±0.13* (64)	9.0±0.29 (60)	9.7±0.16* (62)
	HP	5.4±0.18 (53)	7.4±0.26* (49)	8.7±0.11 (56)

Values are means ± SE. All groups were lacto-ovo-vegetarians, and each diet met the 1983 Canadian recommended nutrient intake. LP, low protein; HP, high protein. * Habitual intake for each group.

BB maintained their respective training regimes, whereas the control group remained sedentary for both balance periods. The resting sweat collections were made on *day 2* of the balance period for all groups, and the exercise sweat samples for EA and BB were collected on *day 3* of the balance period. Exercise sweat samples for EA were taken after training on a treadmill in a climate-controlled chamber at 15°C and a low relative humidity (10–15%). The treadmill velocity and exercise duration was adjusted to match the total caloric cost of a typical training workout (~1,000 kcal). The exercise sweat sample from BB was obtained after they performed *day 3* of a typical 3-day split routine using free weights (3–5 sets of 8–12 repetitions to exhaustion for lower, upper, and midbody parts). To minimize sweat and dermal losses through contact with the equipment, BB dressed in freshly washed (and rinsed in deionized water) shorts, t-shirt, jock, and socks to absorb sweat and were periodically wiped with a fresh washed towel. The towel and clothes were included in the washdown procedure.

Although compliance with any testing protocol using free living subjects is difficult, subject adherence was maximized by supplying food during the adaptation period, the use of daily diet checklists, frequent subject contact, having living and exercise quarters in close proximity to the testing center, and using highly motivated subjects (remuneration). Dietary compliance was assessed using detailed daily diet records and urine collection compliance was assessed using the 24-h urinary creatinine excretion.

On *day 3* of each balance period anthropometric measurements were taken on all subjects. The measurements included height, weight, midarm circumference, midthigh circumference, and body density (by hydrostatic weighing).

Analytic methods. The total nitrogen content of the diets, feces, urine, and sweat was determined using the micro-Kjeldahl technique (1). Sweat and urine urea nitrogen values were determined using a colorimetric technique (6).

Accuracy of the urine and fecal collections was assured by measuring 24-h creatinine excretion and collection of stools between carmine labels. The energy content of the diets was determined using a computer program for nutrient analysis.

Statistical procedures. A two-way analysis of variance implementing a between/within split-plot design was used to determine whether significant differences existed between the factors (i.e., diet and group). When a significant F ratio was observed, Tukey's post hoc test was used to isolate the means that were significantly different. A confidence level of $P < 0.05$ was taken to indicate significance.

RESULTS

The descriptive characteristics of each group are presented in Table 2. The members of group EA ran for between 70 and 80 min on the treadmill at an O_2 uptake ($\dot{V}O_2$) equivalent to 75% of their respective maximal $\dot{V}O_2$. The members of group BB used free weights to perform their standard bodybuilding workout.

The results of the NBAL (intake and urinary, fecal, and sweat losses) on low- and high-protein intakes are summarized in Table 3. All groups were in positive NBAL while consuming their habitual intakes. On the high-protein (HP) diet, NBAL became significantly more positive for all groups ($P < 0.001$) (Table 3), and TN excretion in urine and feces was also significantly greater on the HP diets ($P < 0.001$). Negative NBAL values were found for two BB athletes (2, 4) on the low-protein (LP) intake diet. There was a significantly positive correlation ($r = 0.77$, $P < 0.01$) between daily dietary protein intake and daily urinary UN excretion (Fig. 2).

Estimation of daily losses of TN via sweat demonstrated that BB produced significantly greater daily sweat TN ($P < 0.05$) than S, even though nitrogen intakes and urinary and fecal nitrogen losses were similar during both experiments (Table 3). In Table 3 the exercise sweat values for BB and EA represent both TN and UN; UN values during exercise were 95 ± 5% of the TN values For the resting sweat values UN averaged 90 ± 7% of the TN values overall for all groups. EA tended to have exercise sweat TN and UN losses that averaged

TABLE 2. *Characteristics of study groups*

	Sedentary	Bodybuilders	Endurance Athletes
Age, yr	22±1	24±1	22±1
Wt, kg	76±2	80±2	73±1
Body fat, %*	15.4±1.3	9.6±1.7	7.1±0.8
Habitual exercise, h/wk	<2	≥10†	≥12‡

Values for age, weight, and body fat are means ± SE. * Calculated from body density measurements. † Exercise consisted of 3-day split routine using free weights for 75 min/day. ‡ Maximal O_2 uptake = 76.2 ± 2.7; total weekly mileage ≥ 125 km.

TABLE 3. *Nitrogen balance summary*

	N Intake, g/day	N Excretion, g/day						Balance
		Urine[a]	Feces[a]	Daily sweat		Misc[c]	Total	
				Exercise[b]	Rest			
SLP	12.69	8.63±0.58	1.53±0.20		0.50±0.05	0.3	10.94±0.72	1.73±0.72
SHP	22.91	13.48±0.98[d]	1.93±0.19[d]		1.05±0.08[f]	0.3	16.76±1.05[d]	6.15±1.05[d]
EALP	19.91	13.94±0.62	2.68±0.36	0.22±0.04	1.02±0.09	0.3	18.16±0.61	1.75±0.62
EAHP	30.65	18.09±0.67[d]	3.37±0.31[d]	0.30±0.06	1.50±0.16[f]	0.3	23.46±0.79[d]	7.09±0.76[d]
BBLP	12.66	8.72±0.50	1.37±0.23	0.14±0.02	1.07±0.14[e]	0.3	11.60±0.54	1.06±0.56
BBHP	34.47	17.55±1.1[d]	1.91±0.28[d]	0.20±0.02	1.16±0.11	0.3	21.12±1.37[d]	13.35±1.54[d]

Values are means ± SE. Balance was computed as intake − excretion. SLP, sedentary controls (low protein); SHP, sedentary controls (high protein); EALP, endurance athletes (low protein); EAHP, endurance athletes (high protein); BBLP, bodybuilders (low protein); BBHP, bodybuilders (high protein). [a] Average of 3-day pooled samples; [b] urea N was calculated as 95 ± 5% total N; [c] estimate of miscellaneous losses (from Ref. 4). [d] Significant difference compared with LP ($P \leq 0.01$). [e] Significant difference compared with SLP ($P < 0.05$). [f] Significant difference compared with LP ($P < 0.01$).
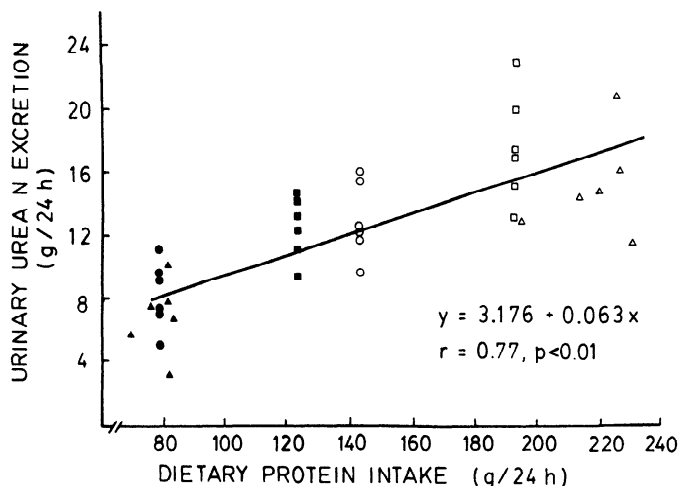

FIG. 2. Relationship between daily protein intake and daily urinary urea nitrogen excretion for all groups. *Open symbols*, high-protein diets; *filled symbols*, low-protein diets. △, ▲: bodybuilders; □, ■: endurance athletes; ○, ●: sedentary controls.

$$y = 3.176 + 0.063x$$
$$r = 0.77, p < 0.01$$

TABLE 4. *Total daily urea excretion*

	Total Daily Urea Excretion, g/day	Relative Urea Excretion, g urea·day⁻¹·g N intake⁻¹
SLP	8.66±0.9	0.68±0.07
SHP	13.55±0.7	0.59±0.03
EALP	13.73±0.8	0.69±0.04
EAHP	19.18±1.4	0.63±0.05†
BBLP	7.89±1.0	0.62±0.08
BBHP	16.12±1.3*	0.47±0.04*

Values are means ± SE. For definitions of abbreviations, see Table 3 footnote. * Significant difference compared with EAHP ($P < 0.05$). † Significant difference compared with SHP ($P < 0.05$).

65% more than that for BB, but this difference was not statistically significant. Consumption of the HP diet resulted in significantly greater ($P < 0.01$) sweat TN + UN excretion for the EA and BB groups during exercise. For all groups the total daily sweat TN losses were greater on the HP diet.

The total daily urea nitrogen (TUN) excretion for EAHP was significantly greater ($P < 0.05$) than for BBHP with the same protein intakes. Since the protein intakes of SLP vs. BBLP and SHP vs. EALP were slightly different, the TUN was expressed relative to protein intake. The significant relationship ($r = 0.77$, $P < 0.01$) observed between dietary protein intake and urinary urea excretion (Fig. 2) indicated that it was necessary to express the TUN values relative to dietary protein intake (Table 4). EALP excreted significantly ($P < 0.05$) more TUN ($P < 0.05$) than SHP, and EAHP excreted significantly more TUN ($P < 0.05$) than BBHP.

The regression lines calculated from the NBAL at two levels of protein intake for each group are presented in Fig. 3. The extrapolated protein intake for a zero NBAL was calculated to be 0.73 g·kg⁻¹·day⁻¹ for group S, 0.82 g·kg⁻¹·day⁻¹ for group BB, and 1.37 g·kg⁻¹·day⁻¹ for group EA.
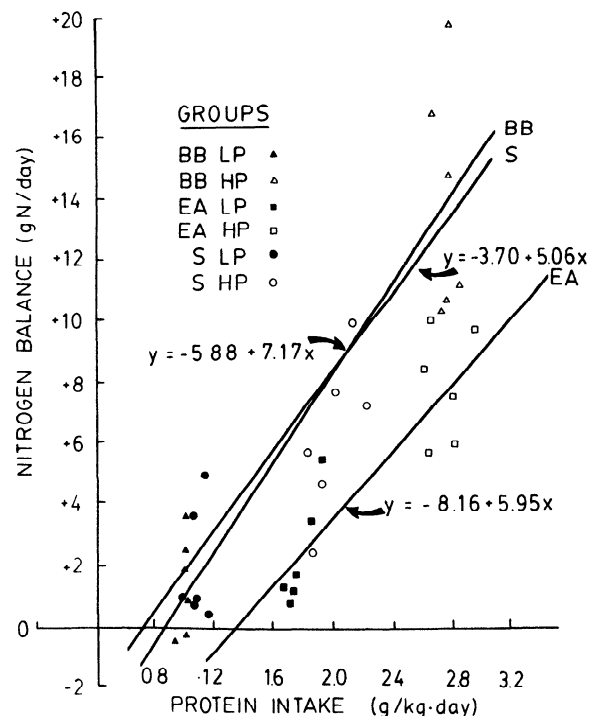
Diet had no effect on any of the anthropometric in-

FIG. 3. Predicted protein intake for a 0 nitrogen balance extrapolated from mean values for BB, EA, and S on their low-protein (LP) and high-protein (HP) diets. Extrapolation was based on regression equation calculated for each group. See legend of Fig. 1 for definitions of other abbreviations.

dexes measured. A higher protein intake was not associated with an increase in body density (lean body mass) (Fig. 4). No significant relationship was demonstrated
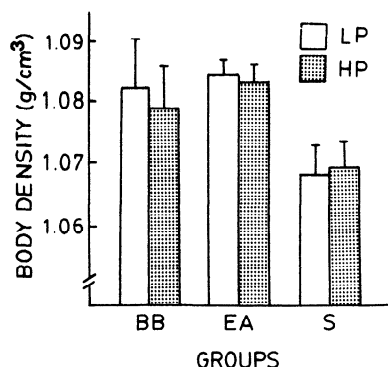
FIG. 4. Effect of low- (LP) and high-protein (HP) diets on body density for each group. See legend of Fig. 1 for definitions of other abbreviations.
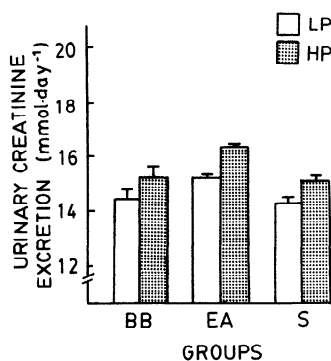

FIG. 5. Effect of low- (LP) and high protein (HP) diets on 24-h urinary creatinine excretion measured for each group. See legend of Fig. 1 for definitions of other abbreviations.

between protein intake and creatinine excretion (an indicator of lean body mass) (Fig. 5).

DISCUSSION

It is a common belief among weight lifters and bodybuilders that high protein intakes (3–4 times the RNI) are necessary to promote and maintain lean body mass. Intakes of this magnitude are supported by research on elite weight lifters which demonstrated that at least 2 (16) or 3 (5) g protein·kg^{-1}·day^{-1} were required for a positive N$_2$ balance. However, in these two studies the lack of sweat collection, diet adaptation periods, distribution of accurately analyzed diets, and a control group make these conclusions questionable. In properly controlled NBAL studies on young men performing resistance exercise, it has been the conclusion that protein intakes of between 0.8 and 1.4 g protein·kg^{-1}·day^{-1} were adequate to achieve a positive NBAL (8, 13, 26, 27).

It was evident from the positive NBAL observed for all study groups that their habitual protein intakes were more than adequate in meeting their dietary protein needs. It was surprising to find that four of the six bodybuilders were in positive NBAL on the LP diet that supplied only 1.0 g protein·kg^{-1} day^{-1}. In addition, the failure of the LP diet to decrease any of the anthropometric indexes measured over a 2-wk period in the bodybuilders supports the fact that for this group lean body mass can be maintained, at least over the short term, on a relatively low protein diet that is only slightly greater than the RNI for normal young men (18–24 yr).

Although the NBAL values were significantly greater while subjects were on the HP diets the failure of the anthropometric indexes to change suggests that lean body mass was not altered in response to higher nitrogen retention. In a review of balance studies, Hegsted (14) proposed that a number of factors may contribute to high NBAL values seen at high protein intakes. NBAL studies underestimate nitrogen excretion either by a lack of complete sample collection or through unmeasured losses (skin desquamation, hair, nails, mucous, semen). In the present study the completeness of collection of urine and feces was determined. The exercise sweat losses were measured and the resting sweat losses were extrapolated to 24 h from 90-min collections. It is possible that some error may have been inherent in the sweat collections; however, the magnitude of this error would not affect the NBAL values significantly (i.e., sweat nitrogen <5% of total nitrogen excretion). Miscellaneous losses as estimated by Calloway et al. (4) were so small that it is unlikely that unaccounted nitrogen losses in the present study would significantly affect the NBAL.

Hegsted (14) also suggested that the overestimation of NBAL may reflect insufficient diet adaptation periods. He claimed that adaptation periods of 10 days are likely adequate when protein intakes are lowered, yet ≥2 wk are needed for metabolic stability when protein intakes are increased. In the present study, the 10-day adaptation periods would have been of sufficient duration for the BB groups (for the decrease of their PRO intake) and were likely long enough for EA and S to achieve relatively stable conditions, since the magnitude of the increased protein intake was not large (<1 g protein·kg^{-1}·day^{-1}) and the addition of protein powder was the only significant dietary modification.

Although the present NBAL study was carried out under strict conditions, the review by Hegsted (14) indicates that an overestimation of apparent NBAL is inherent in all balance studies. As a consequence of this inherent problem with NBAL studies, the apparent NBAL at high protein intakes does not directly indicate an increased lean body mass, which also leads to an overestimation of extrapolated minimal protein requirements. These problems, however, would not be expected to affect the relative relationships between the groups in the present study.

Determination of the minimal intake of dietary mixed protein for bodybuilders and endurance athletes may be estimated using the regression lines in Fig. 2. Extrapolation of the regression line to a level of zero NBAL resulted in minimal intake estimates of 0.73, 0.82, and 1.37 g protein·kg^{-1}·day^{-1} for S, BB, and EA, respectively. Since the error of NBAL studies is greater at higher protein intakes (14), these extrapolated values may represent an overestimation of the true levels for 50% of each group. The exact minimal requirement can only be obtained with several NBAL studies carried out at protein intakes just below and just above that required to attain zero balance. Zackin et al. (30) performed a NBAL study using three levels of protein intake (2 below and 1 above) to determine the minimal protein intake in a group of endurance athletes that would achieve zero

NBAL. Their calculated minimal intake of 0.97 g protein $kg^{-1} \cdot day^{-1}$ for endurance athletes is less than that found in this study. The discrepancy of the findings between the two studies may be a reflection of the lower maximum $\dot{V}O_2$ values and training intensity of the subjects and the lower experimental protein intakes in the study by Zackin et al. (30).

From the minimal intakes estimated from our data, safe levels of intake were estimated. Individual differences necessitate that a safety margin be included in suggesting safe levels of protein intake. The finding that two of the body builders were in negative NBAL on the LP diet demonstrates that degree of interindividual differences in a group of athletes, hence the need for safety margins. The safe mixed protein intake for BB was found to be 1.2 g protein $\cdot kg^{-1} \cdot day^{-1}$ and for EA was found to be 1.6 g protein $\cdot kg^{-1} \cdot day^{-1}$. These calculations represent one SD from the extrapolated minimal intakes. Since the extrapolated minimal intakes were an overestimation, one SD was used rather than the usual two SD values used for setting safe levels for a population from actual minimal requirements.

The estimated safe level of protein intake for the BB group of 1.2 g protein $\cdot kg^{-1} \cdot day^{-1}$ is less than the 1.3–2 0 g protein $\cdot kg^{-1} \cdot day^{-1}$ suggested for strength athletes by some investigators (16), much less than the 3.0 g protein $\cdot kg^{-1} \cdot day^{-1}$ reported for zero balance in elite weight lifters (5) and much less than the 2.7 g protein $\cdot kg^{-1} \cdot day^{-1}$ habitually consumed by the BB group of this study. The chronic protein supplementation that is common among many bodybuilders is costly, both in terms of dollars and the possible ultimate contribution of high-protein intakes to kidney degeneration (2).

The safe protein intakes estimated for EA and BB can only be recommended for males consuming a high-energy and carbohydrate diet and who are in steady-state training intensity. Factors such as *1*) levels of very high training intensity (16), *2*) reduced energy intake (13, 26), and *3*) reduced dietary carbohydrate-to-fat ratio (25) would decrease the efficiency of protein utilization, altering the above recommendations for some athletes. In addition, pregnant or lactating females and adolescent athletes would likely require higher protein intakes to cover the effect of these particular physiological states on protein requirements (7).

The NBAL data in this study suggest that endurance athletes require more protein than either sedentary controls or BB and that BB require only slightly more protein than do sedentary controls. Since none of the anthropometric indexes changed significantly for any group, net protein accretion could not explain the increased protein need of EA. An increased protein catabolism during endurance exercise, as demonstrated by several studies (9, 10, 18), is the most likely reason for the increased protein requirements for EA. To examine this the TUN was used to determine the daily net protein catabolism in the three study groups.

The similar TUN excretion of BB and S while subjects were on the same protein intakes (BBLP and SLP) demonstrates that the maintenance bodybuilding routine was not associated with an increased protein catabolism or turnover. A recent study by Hickson et al. (15) supports our finding that a bodybuilding program has no acute effect on urinary urea nitrogen excretion. Contradictory results have been found by Dohm et al. (9) in a study where diet was not strictly controlled and the intensity of the weight-lifting program may have been higher than in the present study

The greater TUN excretion for EA relative to S on the LP diet and the greater TUN excretion for EA relative to BB on the HP diet demonstrated that endurance exercise was associated with a greater protein catabolism than either a bodybuilding program or a sedentary lifestyle. This increased protein catabolism is the likely cause of the increased protein need of EA. An increase in urea excretion following endurance exercise has been reported by Calles-Escandon et al. (3), who studied subjects exercising for 90 min at 45% $\dot{V}O_2$ and Dohm et al. (9), who studied men competing in a 10- to 12-mile run. On the other hand, Wolfe et al. (29), using an isotopic analysis of urea kinetics, found no increase in urea production in subjects exercising for 105 min at 30% maximum $\dot{V}O_2$. However, the low intensity of exercise employed in the study by Wolfe and co-workers may have contributed to the failure of endurance exercise to increase urea excretion. The significant relationship between protein intake and urinary nitrogen excretion (r = 0.79, $P < 0.01$) found in this study suggests that the discrepancies in the literature concerning urea excretion with exercise may derive from the failure to control for protein intake when measuring urea excretion.

Estimation of daily losses of TN via sweat demonstrate that BB produced significantly greater daily sweat TN ($P < 0.05$) than S, even though nitrogen intakes and urinary and fecal nitrogen losses were similar during both experiments for these groups (Table 3). The similar weight loss during a given exercise bout for both EA and BB indicated that sweat volume losses were the same; however, EA produced sweat TN and UN losses that averaged 65% more than in BB but probably due to intersubject variability were not significantly different. Consumption of the HP diet was associated with significantly greater ($P < 0.01$) sweat TN and UN excretion for all groups. This demonstrates the importance of strictly controlling for protein intake when using sweat UN to indicate net protein catabolism.

The exercise sweat TN and UN excretion values found in this study were lower than those reported by either Gontzea et al. (12) or Lemon and Mullin (17). This discrepancy may be due to the different sweat collection techniques used by these researchers and a reflection of the high dietary carbohydrate intake in the present study, which has been demonstrated to decrease the sweat UN excretion (17).

It may thus be concluded that endurance exercise increased net protein catabolism which resulted in an increased recommended protein intake for EA relative to BB or S. The bodybuilding routine used in this study may not increase net protein catabolism significantly. At the same time, lean body mass (as determined anthro-

pometrically and with NBAL) can be maintained at a dietary protein intake considerably less than that habitually consumed by the BB under study.

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