Increased Rates of Whole Body Protein Synthesis and Breakdown in Children Recovering from Burns

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The rates of whole body protein synthesis and breakdown were determined, with the aid of a constant administration of [^{15}N]glycine, during recovery in 11 acutely burned children, involving a total of 24 studies. Eleven studies were also conducted in seven healthy children before and after reconstructive surgery. Rates of whole body protein synthesis and breakdown, expressed as g protein/kg body weight/day, were significantly ($p < 0.05$) and positively correlated with per cent body surface area total burn, per cent third-degree burn, and per cent open wound. These rates (synthesis, 7.1 ± 2.1 g protein/kg/day; breakdown, 6.3 ± 1.8 g protein/kg/day) were 80 to 100% greater ($p < 0.05$) in patients with total burns ≥60%, as compared to patients with <25% total burns or to the surgical patients. Because of the high energy cost of protein synthesis, it is proposed that an increased whole body protein turnover is partly responsible for the reported elevations in rates of heat production occurring in patients recovering from thermal injury.

MARKED CHANGES IN ORGAN and whole body protein metabolism accompany severe thermal injury. Persistent body nitrogen (N) losses may exceed 30 g N per day in an initially well-nourished adult and thus result in debilitating protein wasting [18–20], unless adequate nutritional therapy is provided. Although significant amounts of protein, amino acids, and urea N may be lost via the exudate and transudate from large open wounds, the major route of the total N loss occurs via an increased urinary urea output.[20] Correlated with the urinary N loss is an elevated resting energy expenditure, which may reach a level twice normal intensity.[21] Because the energy required for total body protein synthesis accounts for a major proportion of the total basal energy expenditure,[1,12,17,24] alterations in the rates of whole body and organ protein turnover, as a consequence of thermal injury and treatment, may account, in part, for the changes in the resting energy expenditure.

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Much of our present knowledge of the nature of whole body protein metabolism after trauma has been obtained from N balance studies. N balance determinations only indicate, however, whether there are changes in body N content, without revealing the ways in which these changes are brought about. An understanding of the nature of such changes is necessary for developing rational strategies for controlling the mobilization of body protein under conditions of burn trauma.

In this study we have examined dynamic aspects of N metabolism in burned children by utilizing a model for measuring rates of whole body protein synthesis and catabolism. We have previously used this model in studies with healthy subjects of various ages.[14,17,23,25] Our observations reveal that rates of protein synthesis and breakdown are increased during recovery in severely burned children and that these increases may account for the increased energy expenditure and heat production under these conditions.

Methods

Patients and Ethical Considerations

Children, aged four to 12 years, admitted to the Shriners Burns Institute, Boston, were the subjects for this study. Some of the patients were receiving treatment for acute burns and others were admitted for reconstructive surgery. A total of 24 studies were conducted in 11 burned children, at various times after their injuries occurred and at varying protein and calorie intakes. In addition, 11 studies were conducted in children undergoing reconstructive surgery (all

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Submitted for publication: July 7, 1977.

0003-4932-78-0400-0383-0105

burned more than one year prior to study). Nine of these latter studies were conducted between four and 11 days after elective surgery, and two were performed immediately before the operations. The experimental protocol received the administrative approval of the Human Studies Subcommittee of the Massachusetts General Hospital and the M.I.T. Committee on the Use of Humans as Experimental Subjects. Informed written consent was obtained for each study with the help of a pediatrician who served as the patient's advocate. None of the experimental procedures interfered with the normal medical care of the patient.

Experimental Design

Burned patients. The experimental protocol was designed to include every patient admitted with an acute thermal injury during the period of study. We had to exclude some patients, however, for reasons such as urinary incontinence or inability to obtain parental consent. Each patient was studied within two weeks after the initial burn and, whenever possible, the study was repeated at about two- or three-week intervals until discharge from the hospital. Intravenous and oral calorie and protein intakes were provided according to the patient's estimated needs.

The individual studies usually lasted six days. Complete daily urine collections were made on days 1–5, in order to assess the stability of urea N output. The coefficient of variation in urinary urea N excretion during the first five days of each study was compared to data previously obtained in healthy young adults on a diet which had been constant for eight days.[8] Although the variation in urea N excretion was greater than in healthy persons on a constant diet, the mean urea N excretion in our patients during the first three days was not significantly different from that of the last two days (paired t-test). Stool collections were made on days 3–6. On days 4 and 5, [^{15}N]glycine was administered either orally or intravenously every three hours, and urine was collected every four hours throughout the 48 hours. Dressings from the burn wounds were collected on days 3–5 for estimation of wound N loss. Total N intake was determined for the two days of [^{15}N]glycine administration. A team of research nurses was assigned to each patient during his/her stay in the intensive care unit where each study was performed. This individual care assured accurate sample collections and monitoring of the nutrient and isotope intakes.

The acutely burned patients were usually studied after the burn wounds had been excised and closed by grafting. Excision and closure were carried out during the first week after the injuy, when burns covered up to 60% of the body surface area (BSA); for burns greater than 60% BSA, excision and closure were performed within the first two weeks. The surgical efficiency of excising a deep second-degree or third-degree burn up to 60% BSA, with immediate closure of the wound, approaches 100%. For patients with burns of greater than 60% BSA, wound closure is not as efficient, and significant open wounds will persist beyond the second week. Therefore, for the patients with burns up to 60% BSA, the initial studies were carried out after the burn wound had been closed. In studies with patients whose burn size exceeded 60% BSA, the initial study was carried out while there was a significant open burn wound.

For evalution of the metabolic data, we used information from the attending surgeons regarding the initial burn size (per cent total and per cent third-degree BSA). In addition, the per cent open wound at the time of study was estimated from descriptions provided by the attending surgeons and from photographs. "Per cent open wound" is defined as the per cent full thickness open wound (excised burn) plus one-half the sum of the per cent BSA due to the following: burn wounds that are eschar covered, second-degree burns, recently grafted wounds (less than one week), and new donor sites (less than one week).

Reconstructive surgery patients. Each study was carried out over a six-day period in a manner similar to that with the burned patients, except that wound dressings were not collected. For some patients, we were able to perform studies on one or more occasions after the surgery.

Experimental Model

To determine the rates of whole body protein synthesis and breakdown, we applied the Picou and Taylor-Roberts model,[15,17] which involves a constant infusion of [^{15}N]glycine given intragastrically or intravenously for about 30 hours (Fig. 1). Several minor modifications of the original model were necessary for application to our study with burned patients. The isotope was administered every three hours orally or intravenously. The [^{15}N]glycine (Stohler Isotope Chemical Corp., Waltham, MA) was administered at an average rate of 0.5 mg ^{15}N per kg per day. The ^{15}N enrichment of the [^{15}N]glycine was approximately 83 atoms per cent excess when measured on our mass spectrometer. Sterile pyrogen-free [^{15}N]glycine (Leberco Laboratories, Roselle Park, New Jersey) was prepared for intravenous use as an aqueous solution by the pharmacy of the Massachusetts General Hospital. Previous studies have indicated that the route of administration does not alter the estimates of protein synthesis and breakdown rates.[15,17] Furthermore, we did not find any sig-

nificant difference in the ^{15}N enrichment of fecal N following oral and intravenous isotope administration. A relatively constant level of ^{15}N enrichment of urinary urea was achieved in all of the studies.

Total N intake (including blood products) was used to calculate N balance and to assess the relationships between protein intake and protein turnover. However, only orally administered N or intravenously administered nonprotein N was used to calculate the amount of N entering the metabolic pool from sources other than breakdown of tissue and organ protein. Urinary N losses were considered in the calculation of the whole body synthesis rate.

The intake of N by the acutely burned patient was generally through intravenous protein hydrolysates or a defined formula diet. We determined the amount of ingested solid food by analyzing a duplicate preparation of all food offered to the patient. The N content of food refused was also determined.

During the ^{15}N-tracer studies, urines were usually collected by the use of existing catheters in the acutely burned children. Small urine losses occasionally occurred, but in none of the cases reported here did such loss exceed an estimated 5% of the total. Measurements of urinary creatinine were used to help judge the completeness of collections.[26]

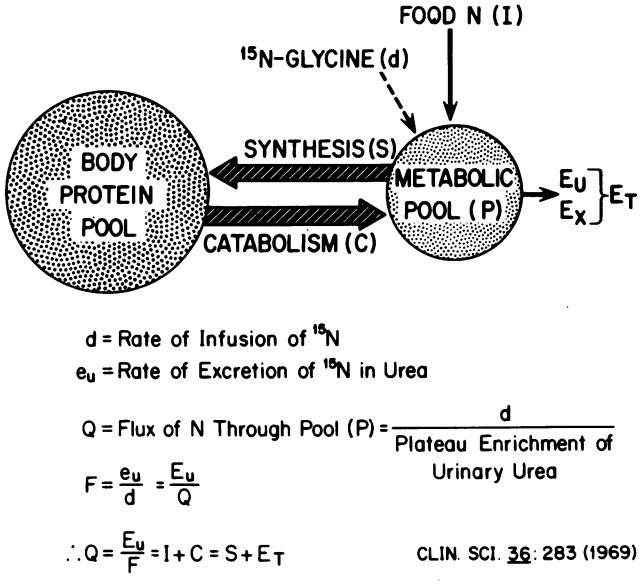

FIG. 1. The Picou and Taylor-Roberts model for studying dynamic aspects of whole body N metabolism by continuous infusion of [^{15}N]glycine. I, C, and S are intake, protein breakdown, and protein synthesis, respectively (mg N/kg/day); E_u, E_x, and E_t are urinary urea, urinary nonurea, and urinary total N excretions, respectively (mg N/kg/day); and Q is the flux (mg N/kg/day) of N for the metabolic pool, P. The rate of administration of ^{15}N is d (mg ^{15}N/kg/day). F is the fraction of the administered dose (d) that is excreted as ^{15}N urea or the fraction of total N entering the pool that is excreted as urea N. Q is equal to d divided by the average plateau enrichment (Sd), which is obtained from the isotope enrichment curve (see Fig. 2) (Q = d/Sd).

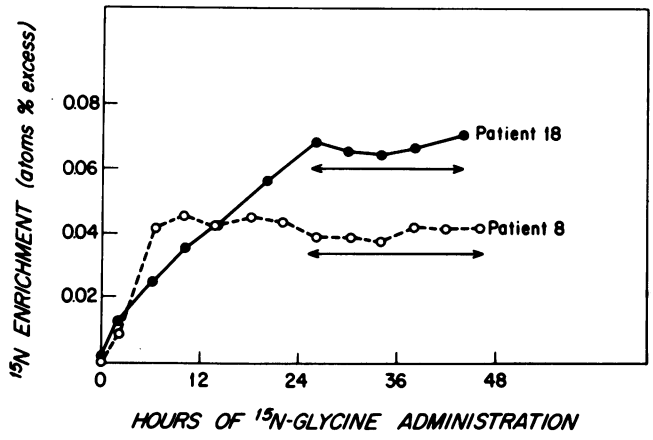

FIG. 2. ^{15}N enrichment of urinary urea with continuous administration of [^{15}N]glycine in two representative patients.

Samples and Analyses

The N concentrations of urine, stools, diet, and burn exudate, and the urea N concentration of urine, were determined by methods previously described.[26] Aliquots from washings of burn dressings were pooled and concentrated by freeze drying. Recovery experiments, in which we applied known amounts of N as whole blood to clean dressings, indicated that wound N losses may be somewhat underestimated.

Urinary urea N was isolated as ammonia by use of the Conway diffusion method[9] after pretreatment of urine with permutit.[6] The ammonia was then reacted with hypobromite to produce N_2 gas[22] and ^{15}N enrichment was determined[16] using a double-collector, isotope ratio mass spectrometer (Model 3-60 RMS, Nuclide Corp., State College, Pennsylvania). Over a one-year period, frequent determinations of a [^{15}N]glycine standard gave a value of 0.0512 ± 0.0006 (S.D.) atoms per cent excess (n = 44). Replicate urine samples were also prepared for mass spectrometry and analyzed with a coefficient of variation of one per cent.

Interpretation of Urinary Urea ^{15}N

Figure 2 shows an example of the change in ^{15}N enrichment of urinary urea N with time following administration of [^{15}N]glycine. Plateau values of urinary urea ^{15}N enrichment, to be used for calculation of body N flux, were obtained by inspection of the curves. For each study the coefficient of variation in "plateau values" was calculated and the mean coefficient for the 35 studies was 7.2%.

Statistical Evaluation

Before evaluating the results, we grouped the various studies according to per cent initial burn and also, in some cases, according to per cent open wound. An a priori decision was made to group the studies according to the two periods after injury, as discussed in the results. We analyzed variance and covariance by the use of BMD computer program packages.[4] Covariate analysis was used to explore interrelationships among burn size, energy intake, and protein intake on rates of whole body protein synthesis and breakdown. Paired t-tests, two sample tests, Mann-Whitney test, and Scheffe's method for multiple comparisons were also applied.[3]

Results

Table 1 provides a summary of protein turnover data for each burned patient (all of whom survived), as well as additional information used in evaluation of the results. Table 2 summarizes similar data for the patients undergoing elective surgery. A diagnosis of septicemia was made if the patient showed clinical evidence of sepsis and positive blood cultures (bacterial or fungal). Wound infection was defined by the isolation of a moderate to heavy growth of bacteria or fungi from more than one culture. Table 1 shows that all cases of septicemia and seven of nine cases of wound infection were present in patients with burns of ≥60% BSA. For this reason, we did not analyze the data for the effects of infection *per se*.

In the burned patients whole body protein synthesis (S) was significantly ($p < 0.05$) correlated with per cent total burn ($r = +0.49$) and per cent third-degree burn ($r = +0.42$). Similarly, whole body protein catabolism (C) was correlated ($p < 0.05$) with per cent total burn ($r = +0.55$) and per cent third-degree burn ($r = +0.46$).

Because of differences in surgical treatment, results with burned patients were grouped according to initial per cent total burn. Reconstructive surgery patients were considered in this analysis as an unburned group. Rates of S and C were 80–100% greater in the group with burns ≥60%, as compared with the group with burns <25% or with the unburned group (Fig. 3, Table 3). There were no statistically significant differences between the groups with respect to age, time after burn, protein intake (g protein per kg per day), or calorie intake (kcal per kg per day). The unburned children had a mean age of 14 years, whereas those with burns ≥60% or with burns <25% had mean ages

TABLE 1. *Characteristics of Burn Patients*

Patient and study number	Age and sex	Weight	Height	Total burn	Third-degree burn	Open wound	Day post burn	Calorie intake	Carbohydrate	Fat	Total protein intake	Whole body N metabolism*		
												S	C	Q
	(yr)	(kg)	(cm)	(% BSA)	(% BSA)	(% BSA)		(kcal/ kg/day)	(% total calories)	(% total calories)	(g protein/ kg/day)	(g protein/ kg/day)		
1a	5F	18.8	106	36	30	5	33	106	—	—	2.7	4.3	2.9	5.6
2a	12M	34.6	155	15	7	5	15	76	55	34	2.6	3.5	3.3	6.0
4a	11M	44.0	148	55	39	27	7	47	62	14	1.7	3.8	3.9	5.5
4b	11M	39.3	148	55	39	5	30	73	48	35	1.9	3.6	2.8	4.7
5a†	9M	36.4	134	36	30	30	9	92	73	19	5.3	7.9	7.2	10.9
5b†	9M	31.2	134	36	30	8	24	85	42	40	2.9	5.4	4.2	7.2
6a‡	12M	37.7	153	65	45	39	12	111	57	30	5.4	6.9	6.0	10.0
6b	12M	38.2	153	65	45	40	31	142	59	31	4.2	7.1	5.0	9.2
6c†	12M	44.2	153	65	45	2	88	68	55	37	1.7	7.8	7.4	9.1
7a‡	12M	33.9	152	83	74	40	15	210	66	22	7.7	12.2	10.7	17.0
7b‡	12M	35.2	152	83	74	40	32	120	79	14	3.7	7.0	6.2	8.9
7c†	12M	34.1	152	83	74	18	63	72	85	4	4.5	5.4	5.2	7.0
7d	12M	32.9	152	83	74	9	86	46	44	29	1.8	4.2	4.0	5.7
8a	9F	31.6	130	75	50	55	14	62	93	0	7.8	6.3	6.9	8.8
8b‡	9F	33.7	130	75	50	52	26	119	66	7	5.0	6.5	5.2	8.2
8c†	9F	31.0	130	75	50	28	52	84	54	32	2.7	7.6	6.5	9.2
9a	4M	14.8	106	25	0	12	13	155	44	38	5.2	6.3	4.9	10.1
9b	4M	15.3	106	25	0	5	23	108	38	39	5.3	4.9	2.9	8.2
10a	5M	20.0	114	50	50	30	14	100	70	26	5.6	4.2	1.5	6.3
10b	5M	17.8	114	50	50	18	28	101	43	38	4.0	6.0	3.4	7.4
10c	5M	20.4	114	50	50	2	57	116	45	38	3.6	6.1	5.0	8.6
11a	11F	47.4	153	8	8	5	24	32	48	35	0.9	2.4	2.7	3.6
11b	11F	48.6	153	8	8	1	32	33	41	39	1.4	3.2	2.6	3.9
21a	5M	17.2	112	10	8	11	11	116	45	33	6.1	6.9	4.7	10.8

* S = protein synthesis, C = protein breakdown, Q = N flux. † Wound infection only. ‡ Septicemia.

TABLE 2. *Characteristics of Reconstructive Surgery Patients*

Patient and study number	Age and sex	Weight	Height	Days post surgery	Calorie intake	Carbohydrate	Fat	Total protein intake	Whole body N metabolism*		
									S	C	Q
	(yr)	(kg)	(cm)		(kcal/kg/day)	(% total calories)	(% total calories)	(g protein/kg/day)	(g protein/kg/day)		
12a	18M	61.0	174	5	60	39	33	2.2	2.6	2.1	4.3
14a	15M	48.0	162	—	80	41	44	2.8	3.8	2.9	5.7
14b	15M	45.4	162	5	56	41	42	2.1	2.9	3.0	5.2
14c	15M	45.6	162	7	68	55	33	2.1	4.1	3.6	5.6
15a	18M	63.8	175	4	75	34	46	3.2	4.0	3.6	6.8
15b	18M	61.8	175	7	73	63	29	1.9	3.6	3.3	5.2
16a	13F	45.6	146	—	68	46	42	1.7	4.1	3.4	5.1
17a	4M	18.4	103	11	99	44	42	3.3	4.6	3.1	6.4
17b	4M	18.8	103	4	92	56	31	1.5	2.4	3.7	5.2
18a	15M	57.2	160	6	82	59	30	2.0	3.4	2.9	4.9
19a	16M	61.3	163	6	72	51	36	2.4	3.2	2.1	4.6

* S = protein synthesis, C = protein breakdown, Q = N flux.

of 11 and ten years, respectively. The significance of these age differences is considered in the discussion. Analysis of covariance was used to assess further the effects of these factors on the relationship between protein turnover and burn size. Adjustment of the means of S or C for each covariate effect did not alter the above conclusions.

Although the burn groups did not show statistically significant differences in the per cent of calories provided by carbohydrate, a 38% increase ($p < 0.01$) in the percentage of carbohydrate calories was evident when the ≥60% group was compared to the unburned patients. When S was corrected for fecal and wound N loss ("corrected S"; Table 3), it remained 91% greater in the ≥60% group, as compared with the unburned patients. All groups were in positive N balance, and, with respect to this parameter, no differences existed among the burn patient groups (Table 3). When compared to the nonburned patients, however, the ≥60% group had a significantly higher N balance.

The per cent open wound at the time of study was positively correlated ($p < 0.05$) with S ($r = +0.51$) and C ($r = +0.51$). In Table 4 the studies have been grouped and analyzed according to per cent open wound; this grouping shows a 56% increase in "corrected S," a 71% increase in S, and a 76% increase in C in the ≥31% open wound group compared to the ≤10% group. However, caloric intake was 72% higher in the ≥31% group than in the ≤10% group ($p < 0.05$, Scheffe's test), and each group differed significantly from the

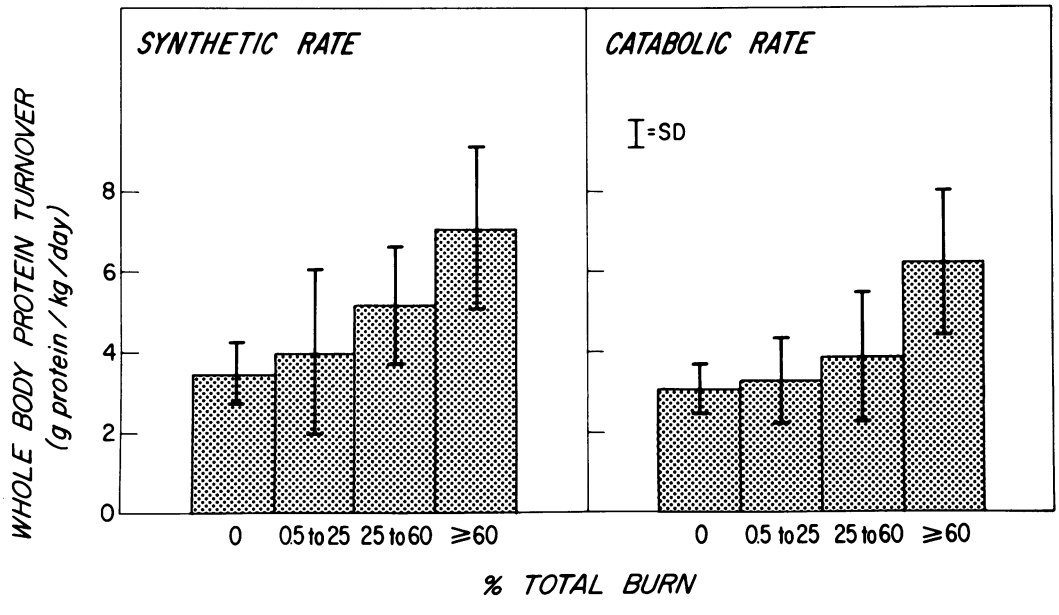

FIG. 3. Relationship between burn size and whole body protein turnover. The number of observations were 11, 4, 10, and 10 for the 0%, 0.5 to 25%, 25 to 60%, and ≥60% total burn groups, respectively.

TABLE 3. *Relationship between Extent of Thermal Injury and Aspects of Whole Protein Turnover*

Variable*	Patients Grouped by Per Cent Total Burn			
	I (0%) (n = 11)	II (0.5–24%) (n = 4)	III (25–59%) (n = 10)	IV (≥60%) (n = 10)
Synthesis (g protein/kg(day)	3.5 ± 0.7	4.0 ± 2.0	5.2 ± 1.4	7.1 ± 2.1
Breakdown (g protein/kg/day)	3.1 ± 0.6	3.3 ± 1.0	3.9 ± 1.6	6.3 ± 1.8
Corrected S† (g protein/kg/day)	3.4 ± 0.4	3.8 ± 1.6	4.8 ± 1.3	6.5 ± 1.8
N balance‡ (g N/kg/day)	0.05 ± 0.06	0.08 ± 0.16	0.21 ± 0.13	0.26 ± 0.19

* Results represent the means ± S.D. F-Test findings were as follows: Synthesis, $p < 0.01$; Breakdown, $p < 0.01$; Corrected S, $p < 0.05$; N balance, $p < 0.01$. Scheffe's test showed the following significant differences between groups: Synthesis—I vs. IV ($p < 0.05$), II vs. IV ($p < 0.05$); Breakdown—I vs. IV ($p < 0.05$), II vs. IV ($p < 0.05$), III vs. IV ($p < 0.05$); Corrected S—I vs. III ($p < 0.05$), I vs. IV ($p < 0.01$), II vs. IV ($p < 0.05$); N balance—I vs. IV ($p < 0.01$).

† Corrected S refers to the whole body protein synthetic rates corrected for fecal and wound N loss.

‡ For this calculation, intake represents the actual measured N intake including whole protein. Output represents the estimated N loss via urine, wound and stool.

others with respect to total protein intake. Therefore, the specific effect of size of the open wound, if any, cannot be determined because of concurrent changes in protein and energy intake. However, a reduction in open wound size, together with the lowered intakes of protein and energy that accompany clinical improvement, is apparently associated with a diminished protein turnover. Table 4 also indicates that N balance was significantly higher in the ≥30% group compared with the ≤10% group.

An initial evaluation by analysis of variance of the entire results, grouped according to three time periods after burn, did not reveal a significant effect of the postburn period on protein synthesis and breakdown. Therefore, two time periods were chosen for evaluation of the time factor for those seven patients who were studied at least twice (Table 5). This analysis removes the potentially confounding effect of initial burn injury that may be introduced by analyzing all of the burn patients together.

Table 5 indicates that a statistically significant decrease in C occurred with time in these seven patients, but that S was not significantly reduced during the period lasting three to six weeks after the injury. Coinciding with the decline in C was a mean 27% decrease ($p < 0.025$) in the size of the open wound during the second 20-day period of study. For patients 6, 8, and 10, the third study at 88, 52, and 57 days postburn, respectively, indicated rates of C that were higher than the value obtained for the second study. With the limited number of cases available for analysis, the significance of these observations cannot be fully assessed. The late recovery period may, however, be associated with an enhanced protein turnover, but this possibility requires further study.

The rate of S was significantly ($p < 0.05$) and positively correlated with caloric intake ($r = +0.74$) and protein intake ($r = +0.62$). The rate of C was also positively correlated with calorie intake ($r = +0.50$) and protein intake ($r = +0.49$). Age was not significantly correlated with either S or C, and there were no significant relationships between the parameters of protein turnover and the number of days since the previous surgical procedure.

Wound N Loss

A significant ($p < 0.05$) correlation ($r = +0.55$) occurred between wound N loss (g per kg per day) and per cent total burn, as well as between wound N loss and protein intake ($p < 0.01$; $r = +0.68$). Wound N loss in the ≥60% group was significantly ($p < 0.05$) greater than in the other groups. Wound N loss in the largest open wound group also differed from the other two groups (Table 6); a significant ($p < 0.05$) correlation ($r = +0.73$) existed between wound N loss and per cent open wound.

TABLE 4. *Effect of the Extent of Open Wound on Protein Turnover*

Variable*	Patients Grouped by Per Cent Open Wound		
	I (≤10%) (n = 10)	II (11–30%) (n = 8)	III (≥31%) (n = 6)
Synthesis (g protein/kg/day)	4.5 ± 1.6	6.0 ± 1.5	7.7 ± 2.2
Breakdown (g protein/kg/day)	3.8 ± 1.5	4.7 ± 1.8	6.7 ± 2.1
Corrected S† (g protein/kg/day)	4.3 ± 1.6	5.6 ± 1.6	6.7 ± 2.0
N balance‡ (g N/kg/day)	0.08 ± 0.10	0.27 ± 0.13	0.32 ± 0.19

* Results represent the means ± S.D. F-Test findings were as follows: Synthesis, $p < 0.01$; Breakdown, $p < 0.05$; Corrected S, $p < 0.05$; N balance, $p < 0.01$. Scheffe's test showed the following significant differences between groups: Synthesis—I vs. III ($p < 0.01$); Breakdown—I vs. III ($p < 0.01$); Corrected S—I vs. III ($p < 0.05$); N balance—I vs. III ($p < 0.01$).

† Corrected S refers to the whole body protein synthetic rates corrected for fecal and wound N loss.

‡ For this calculation, intake represents the actual measured N intake including whole protein. Output represents the estimated N loss via urine, wound and stool.

Discussion

Metabolic studies in the burned child are difficult to conduct. However, by careful handling of each patient and with meticulous attention to methodology, we have succeeded in exploring dynamic aspects of N metabolism under these conditions. We had originally hoped that we would be able to regulate the child's energy and protein intakes for two or three days, in order to achieve a relatively "steady state" of N metabolism in each patient before conducting the [^{15}N]glycine infusion studies. This procedure, however, was neither practical nor medically appropriate. However, because the mean coefficient of variation in ^{15}N-isotopic enrichment of urinary urea N during plateau was only 7%, it appears that a "steady state" in the metabolic N pool, satisfactory for our purposes, had been achieved. The relatively small variation in urea N enrichment at plateau, as compared to that previously found in healthy adults,[17] indicates that with short study periods of two days, there is an achievement of an isotopic steady state within the metabolic N pool.

The greater variation in urea N excretion in the present studies (compared with that observed in healthy men given a constant protein intake[8]) suggests some uncertainty regarding the precision of the estimated balance between S and C. However, we should emphasize that the focus of our studies was not the level of N balance *per se*, but rather how S and C change as a function of the severity and duration of the thermal injury.

In burned patients a portion of tissue protein breakdown occurs at sites not in equilibrium with the administered tracer, such as the necrotic wound. Thus the whole body N flux and S and C would be underestimated. Within this context, differences between actual and estimated rates of S and C are not likely to be great; hence, these results provide a satisfactory approximation of the relative magnitude of the differences in S and C among the large and small burn size groups.

The markedly higher rates of S that we observed in children recovering from severe burns (≥60% BSA) may provide a partial explanation for the raised basal metabolic rate noted in burned patients.[21] It is also worth noting that seven of our patients, including all in the ≥60% group, had their calorie intakes assessed on a daily basis during the second to fifth weeks after injury. The average caloric intake of these patients was 153% of the mean energy requirements proposed by FAO/WHO[5] for healthy, normal children. Despite the relatively high mean intake of calories in our patients, body weight did not change during the period of study.

There are a number of possible factors, which are associated with the response to and recovery from severe thermal injury, that could account for the changes in whole body protein turnover in the burned children. In this series of experiments, we have attempted to evaluate the effects of some of these factors through a critical statistical analysis of the data, because direct experimental manipulation of these factors was not feasible under our conditions. Each study within an individual occurred at a different stage of recovery and, frequently, at different levels of nutrient intake. Thus, we treated each study as an independent observation. We feel that this approach offers a more reliable method than that of evaluating each patient on an individual basis, because only a limited number of observations were possible in any one patient.

Because of the relationships between age and rates of whole body protein synthesis and breakdown,[25] we needed to evaluate the age factor before comparing the rates in the severely burned group and the reconstructive surgery patients. In a recent series of studies with five healthy children, age four to 13 years, rates of S and C were 4.1 ± 1.0 and 3.5 ± 0.6 g protein/kg/day, respectively.[11] These rates are considerably less than those found in the ≥60% burn group. It seems

TABLE 5. *Effect of Time after Injury on Mean Values of Whole Body Protein Synthesis and Breakdown**

Variable	0–20 Days postburn (A)	20–40 days postburn (B)	Difference (A–B)
Synthesis (g protein/kg/day)	6.8	5.8	1.01†
Breakdown (g protein/kg/day)	5.9	4.2	1.63‡

* Seven patients studied twice during the course of their recovery.
† $p > 0.05$.
‡ $p < 0.05$ by paired t test.

TABLE 6. *Estimates of Wound N Loss in Patients with Varying Burn Size*

Patients grouped by burn size	Wound N loss*
	(g N/kg/day)
Per cent total burn:	
I (0–24%) (n = 4)	0.01 ± 0.01
II (25–59%) (n = 9)	0.04 ± 0.02
III(≥60%) (n = 10)	0.09 ± 0.07
Per cent open wound:	
I (≤10%) (n = 8)	0.02 ± 0.02
II (11–30%) (n = 6)	0.05 ± 0.02
III (≥31%) (n = 6)	0.12 ± 0.06

* Results represent means ± S.D. (F-test, $p < 0.01$). Scheffe's test showed the following significant differences between groups: Per cent total burn—I vs. III ($p < 0.05$), II vs. III ($p < 0.05$); Per cent open wound—I vs. III ($p < 0.01$), II vs. III ($p < 0.01$).

unlikely that the 100% difference in whole body protein synthesis between these two groups is due to age-related differences. This conclusion is further supported when one considers that the changes occurring in whole body protein synthesis during growth and development parallel those in basal energy expenditure over the same period.[25] Because the energy needs of children between ages 11 and 14 decrease about 21%, one would expect the rates of whole body protein synthesis to show only a small decline during this phase of growth.

We observed a markedly elevated rate of whole body turnover in children recovering from extensive body surface burns. These patients were receiving generous protein and energy intakes, and they were in positive nitrogen balance at the time of each study. Several plausible hypotheses could be used to explain these results. It might be speculated that high protein-energy intakes stimulate increased rates of protein synthesis and breakdown. This concept has been explored by Garrow[7] in relation to possible mechanisms for preserving energy balance in health. However, covariate analysis of our data suggested that increases in synthesis and breakdown were related to the size of the burn injury and that this relationship was independent of the level of protein and energy intakes. A second explanation is that burn injury *per se* and/or the presence of an open wound that may be infected stimulates an increase in whole body protein turnover. There are no published data to support or refute this hypothesis. Because our patients were in positive nitrogen balance and they were receiving high energy and protein intakes, it is reasonable to speculate that the increased rates of whole body protein turnover are causally related to the physiologic processes of wound repair and tissue repletion following a period of net body N depletion. Indeed, the refeeding of malnourished infants is associated with increased rates of resting energy expenditure[13] as well as with increased rates of protein synthesis and breakdown.[2,15] However, nutritional status is difficult to evaluate objectively in these patients by the use of available methods, and we do not have unequivocal evidence that the patients with >60% burns had suffered severe body N depletion.

Although measurements of heat production were not undertaken in the present studies, it is probable that the increased protein turnover resulted in an increased rate of heat production, particularly in the severely burned children. If this were the case, a portion of the high energy intake would have been utilized for meeting the increased energy costs associated with enhanced body protein turnover.

We should emphasize that the responses observed in the present studies apply specifically to burned children receiving nutritional support. Thus, the applicability of our results to the thermally injured, nongrowing adult and to patients who may not be receiving adequate energy and protein intakes during treatment is unclear. Nevertheless, it seems quite possible that nutritional therapy will profoundly affect the protein metabolic response to burn injury.

Finally, an alteration in catecholamine balance has been suggested to be a mediator of the elevated rates of energy expenditure in burn injury.[21] Ion pump activity, stimulated by catecholamines,[10] may contribute to the alterations in energy balance and energy utilization that occur during recovery from thermal trauma. From our results, it is apparent that an increased body protein turnover would also contribute to an increased heat production in children who are recovering from thermal injury.

Acknowledgments

This work was supported by N.I.H. Grant 5-P5O-6M21700 and in part by N.I.H. Grant AM 15856. Dr. Kien was supported by N.I.H. Training Grant No. 5-TO1-AM-05371-13. We are indebted to the physicians and nursing staff of the Shriners Burns Institute for their cooperation with the protocol, to Drs. A. Antoon and J. Crawford and their colleagues for acting as patient advocates, to Dr. W. Rand for mathematical advice, to Misses M. Doran, P. Hegarty, K. McGinn, and S. Zolin, R.D., for their assistance with the clinical aspects of the N balance studies, to Miss C. Bilmazes, Miss M. Marcucci, Mr. R. Leighton, and Dr. H. Zayas-Bazan and their staffs for the N concentration determinations, and to Misses Flint, Hudgens, and Wing for typing the manuscript.

References

1. Blaxter, K. L.: Methods of Measuring the Energy Metabolism of Animals and Interpretation of Results Obtained. Fed. Proc., 30:1436, 1971.
2. Brooke, O. G. and Ashworth, A.: The Influence of Malnutrition on the Post-Prandial Metabolic Rate and Respiratory Quotient. Br. J. Nutr., 27:407, 1972.
3. Colquhoun, D.: Lectures on Biostatistics. Oxford, Clarendon Press, 1971.
4. Dixon, W. J.: Biomedical Computer Programs, 3rd ed. Los Angeles, University of California Press, 1973.
5. FAO/WHO Ad Hoc Expert Committee: Energy and Protein Requirements. WHO Technical Report Series, No. 522. Geneva, WHO, 1973.
6. Folin, D. and Bell, R. D.: Applications of a New Reagent for the Separation of Ammonia. J. Biol. Chem., 29:329, 1917.
7. Garrow, J. S.: Energy Balance and Obesity in Man. New York, American Elsevier Publishing, 1974.
8. Garza, C.: Human Protein Requirement: An Evaluation of the 1973 FAO/WHO Safe Level of Protein Intake in Young Men. Ph.D. thesis, Department of Nutrition and Food Science, Massachusetts Institute of Technology, Cambridge, Mass., 1975.
9. Hawk, P. B., Oser, B. L. and Summerson, N. H.: Practical Physiological Chemistry, 13th ed. New York, McGraw-Hill, 1954.
10. Himms-Hagen, J.: Cellular Thermogenesis. Ann. Rev. Physiol., 38:315, 1976.
11. Kien, C. L., Young, V. R., Rohrbaugh, D. K. and Burke, J. F.: Whole Body Protein Synthesis and Breakdown Rates in

Children Before and After Reconstruction Surgery of the Skin. Metabolism, in press.
12. Milligan, L. P.: Energetic Efficiency and Metabolic Transformations. Fed. Proc., 30:1454, 1971.
13. Montgomery, R. D.: Changes in the Metabolic Rate of the Malnourished Infant and Their Relation to Body Composition. J. Clin. Invest., 41:1653, 1962.
14. Pencharz, P. B., Steffee, W. P., Cochran, W., et al.: Protein Metabolism in Human Neonates: Nitrogen Balance Studies, Estimated Obligatory Nitrogen Losses and Whole Body Nitrogen Turnover. Clin. Sci. Mol. Med., 52:485, 1977.
15. Picou, D. and Taylor-Roberts, T.: The Measurement of Total Protein Synthesis and Catabolism and Nitrogen Turnover in Infants in Different Nutritional Status and Receiving Different Amounts of Dietary Protein. Clin. Sci., 35:283, 1969.
16. Rittenberg, D., Keston, A. S., Rosebury, F. and Schoenheimer, R.: Studies in Protein Metabolism. II. The Determination of Nitrogen Isotopes in Organic Compounds. J. Biol. Chem., 127:291, 1939.
17. Steffee, W. P., Goldsmith, R. S., Pencharz, P. B., et al.: Dietary Protein Intake and Dynamic Aspects of Whole Body Nitrogen Metabolism in Adult Humans. Metabolism, 25:281, 1976.
18. Taylor, F. H. L., Davidson, C. S. and Levenson, S. M.: The Problem of Nutrition in the Presence of Excessive Nitrogen Requirement in Seriously Ill Patients with Particular Reference to Thermal Burns. Conn. State Med. J., 8:141, 1944.
19. Taylor, F. H. L., Levenson, S. M., Davidson, C. S. and Adams, M. A.: Abnormal Nitrogen Metabolism in Patients with Thermal Burns. N. Engl. J. Med., 229:855, 1943.
20. Wilmore, D. W.: Nutrition and Metabolism following Thermal Injury. Clin. Plast. Surg., 1:603, 1974.
21. Wilmore, D. W., Long, J. M., Mason, A. D., et al.: Catecholamines: Mediator of the Hypermetabolic Response to Thermal Injury. Ann Surg., 180:653, 1974.
22. Wilson, D. E., Nier, A. O. C. and Reimann, S. P.: Preparation of Isotopic Tracers. Ann Arbor, Edward Brothers, 1946.
23. Winterer, J. C., Steffee, W. P., Perera, W. D., et al.: Whole Body Protein Turnover in Aging Man. Exp. Gerontol., 11:79, 1976.
24. Young, V. R.: Energy Metabolism and Requirements in the Cancer Patient. Cancer Res., 37:2336, 1977.
25. Young, V. R., Steffee, W. P., Pencharz, P. G., et al.: Total Human Body Protein Synthesis in Relation to Protein Requirements at Various Ages. Nature, 253:192, 1975.
26. Young, V. R., Taylor, Y. S. M., Rand, W. M. and Scrimshaw, N. S.: Protein Requirements of Man: Efficiency of Egg Protein Utilization at Maintenance and Submaintenance Levels in Young Men. J. Nutr. 103:1164, 1973.

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