Spontaneous Dietary Protein Intake During Progression of Chronic Renal Failure[1,2]

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ABSTRACT

Malnutrition at the initiation of dialysis is a strong predictor of subsequent increased mortality on dialysis. Few studies have documented the relationship between the progression of renal failure and spontaneous dietary protein intake (DPI) and other indices of malnutrition. In this prospective study, renal function was sequentially measured by creatinine clearance (CrCl) and DPI by 24-h urine collection; simultaneously, multiple sequential biochemical nutritional indices, including serum albumin, transferrin, prealbumin, and insulin-like growth factor-I (IGF-I) concentrations, were measured. The study involved 90 patients (46 men and 44 women) with chronic renal failure (CRF) of various causes monitored in an outpatient clinic. Dietary interventions were minimal. The mean duration of follow-up was 16.5 ± 11.8 months. The results show that the mean (± SD) DPI was 1.01 ± 0.21 g/kg per day for patients with CrCl over 50 mL/min and decreased to 0.85 ± 0.23 g/kg per day for patients with CrCl between 25 and 50 mL/min. The DPI further decreased to a level of 0.70 ± 0.17 g/kg per day for patients with CrCl between 10 and 25 mL/min and was 0.54 ± 0.16 g/kg per day for patients with CrCl below 10 mL/min. This trend was statistically significant ($P < 0.001$). A similar statistically significant trend was observed for serum cholesterol, transferrin, and total creatinine excretion (all $P < 0.01$). A mixed model analysis indicated that for each 10 mL/min decrease in CrCl, DPI decreased by 0.064 ± 0.007 g/kg per day, transferrin decreased by 16.7 ± 4.1 mg/dL, weight decreased by 0.38 ± 0.13% of initial weight, and IGF-I decreased by 6.2 ± 1.9 ng/mL. It was concluded that the progression of renal failure is associated with a spontaneous decrease in DPI, especially below a CrCl of 25 mL/min, and that most nutritional indices in CRF patients worsen as CrCl and DPI decrease. Dietary protein restriction should be used cautiously in CRF patients when CrCl falls below 25 mL/min.

Key Words: *Malnutrition, insulin-like growth factor I, albumin, mortality, weight*

Protein and calorie malnutrition is prevalent in ESRD patients (1,2). It is also associated with an increased risk of morbidity and mortality in this patient population. Specifically, low serum albumin levels have been shown to be associated with an increased risk of death in both chronic hemodialysis and peritoneal dialysis patients, as well as in chronic renal failure (CRF) patients (3–5). However, most studies on CRF patients are cross-sectional studies (6). Thus, the relationship between the progression of renal failure and the development of indices of malnutrition has not been clearly identified in a prospective study of CRF patients up to the time they are initiated on maintenance dialysis.

Anorexia, which eventuates in a decrease in dietary protein and caloric intake, is a well-known manifestation of CRF and is recognized as a contributing factor to the malnutrition in this patient population. The level of renal function at which this anorexia begins and subsequently results in a decrease in dietary protein intake (DPI) and indices of malnutrition is not well established. Such information is important because recent data from the U.S. Renal Data System (6) and other large patient population studies (E.G. Lowrie, personal communication) demonstrate that CRF patients with signs of malnutrition at the time of the initiation of dialysis (defined by a serum albumin less than 4.0 g/dL) had a higher mortality rate in the subsequent years compared with patients with a serum albumin >4.0 g/dL; the lower the serum albumin at the initiation, the higher was the risk of mortality. Other markers of malnutrition that may precede the decrease in serum albumin have not been clearly defined in this patient population.

In this prospective study, we attempted to identify the relationship between the progression of renal disease and spontaneous DPI in CRF patients with various causes of renal failure. We also monitored several commonly measured nutritional indices, including weight, serum albumin, transferrin, prealbumin, and insulin-like growth factor-I (IGF-I) in an attempt to determine early markers of malnutrition.

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MATERIALS AND METHODS

Patients

The study included 90 CRF patients monitored in the Renal Clinic at Vanderbilt University Medical Center over 4 yr (August 1990 to August 1994). Patient demographics are listed in Table 1. There were 46 male and 44 female patients. The mean (± SD) age was 53 ± 15 yr. The causes of renal failure were as follows: diabetic nephropathy (N = 35; 39%), chronic glomerulonephritis (N = 23; 26%), hypertension (N = 8; 9%), adult polycystic kidney disease (N = 7; 8%), and other (N = 17; 18%). The mean (± SD) follow-up time was 16.5 ± 11.8 months (range, 2 to 48 months). Patients who were treated with corticosteroids and/or immunosuppressive agents were not included in the study. There were no other restrictions for inclusion into the study.

Study Design

The study was a prospective observational design. Patients were monitored on a regular basis in the Renal Clinic by their primary nephrologists, and there were no specific interventions for study purposes. All patients were evaluated by the same renal dietitian during the entire study period. No restrictions were placed on the DPI of the patients throughout the study period. If anything, the patients were encouraged to eat to satiety. Patients who formerly participated in a project where dietary interventions were applied or who had received prior restrictions in DPI were excluded from the study. The only attempted dietary management was to decrease potassium intake if serum levels were over 6.0 mEq/L. This occurred in a limited number of patients, particularly those recently initiated on angiotensin-converting enzyme inhibitors, who were treated predominantly by the discontinuation of the drug. Increased phosphorus levels were managed by phosphate binders.

At each clinic visit, vital signs and weight were recorded and routine blood chemistries, including BUN, serum creatinine, total bicarbonate, glucose, serum albumin, and cholesterol, were obtained. Additional blood samples were collected at less frequent intervals for serum transferrin, prealbumin, and IGF-I. Patients were also instructed to collect 24-h urine at intervals ranging from every 4 wk to 6 months, depending on the rate of progression of their renal disease. Total daily urinary creatinine, total protein, and urea nitrogen were measured from these collections, and patients' creatinine clearance (CrCl), total creatinine excretion, and DPI were calculated according to the formulas mentioned below. A number of patients also underwent [^{125}I]iothalamate clearance to determine their true GFR at intervals of 12 months, but these data are not included here because of the limited number of observations.

TABLE 1. Baseline demographics of study patients and mean baseline values for multiple study parameters

Age	53 ± 15
Gender (M/F)	46/44
Mean Follow-Up (months)	16.5 ± 11.8
Serum Creatinine (mg/dL)	3.4 ± 2.2
BUN (mg/dL)	46 ± 24
CrCl (mL/min)	35.1 ± 26.1
DPI (g/kg per day)	0.82 ± 0.28
Serum Albumin (g/dL)	3.8 ± 0.45
Cholesterol (mg/dL)	240 ± 73
Transferrin (mg/dL)	282 ± 34
Total CO_2 (mEq/L)	22.5 ± 4.41

Methods

Blood Chemistries. BUN, serum creatinine, sodium, potassium, chloride, total bicarbonate, glucose, cholesterol, transferrin and complete blood count were measured with usual methods (Olympus 5223 Autoanalyzer). Serum albumin was measured by use of the bromocresol green method. IGF-I assays were performed by use of a radioimmunoassay technique after extraction by the acid-ethanol method (Nichols Institute Diagnostics, San Juan Capistrano, CA). Prealbumin was measured by a nephelometric assay (Behring BN 100 Nephelometer).

Urea and Creatinine Kinetics. CrCl was measured from 24-h urine creatinine according to the following formula:

$$\text{CrCl (mL/min)} = [\text{urine creatinine (mg/dL)} \times \text{urine volume (mL/min)}]/\text{serum creatinine (mg/dL)}.$$

DPI was calculated from 24-h urine urea nitrogen by the following formula (7):

$$\text{DPI} = 6.25 \times [\text{24-h urine urea nitrogen (g/day)} + (0.031 \text{ g of N/kg per day}) \times \text{desirable body weight (kg)}] + \text{urine protein (if} > 5 \text{ g/day)}.$$

Statistical Methods. Several statistical analyses, with increasing complexity, were performed on this prospective longitudinal study.

The first approach consisted of averaging the nutritional parameters (dependent variables) of all measurements classified by four broad ranges of CrCl (CrCl - independent variable) and analyzing for a trend across these four levels. These ranges include CrCl values over 50 mL/min, between 50 and 25 mL/min, between 24 and 10 mL/min, and finally, below 10 mL/min.

As a second approach to develop some insight into the relationship between variables, we calculated the correlations between DPI and CrCl (represented as a continuous function) using the Pearson correlation coefficient. Similar analyses were done with either CrCl or DPI as the independent (or predictor) variable and the other nutritional indices (serum albumin, transferrin, cholesterol, creatinine excretion, prealbumin, and IGF-I) as the dependent (or outcome) variable.

Both of these initial approaches use all data points from each subject in the analysis. Because observations are not independent within a subject, these results might be biased by using multiple measurements within a subject. In addition, because some subjects contributed only a few measurements, whereas others provided up to 15 measurements, we used a mixed model approach for the definitive analysis (8). In this model, the association between variables is assessed for each individual alone and is then averaged appropriately over all subjects. The model specifically incorporates the repeated measurements in a subject, allowing for one measurement to affect the next measurement, but with the effect decreasing over time. Thus, if the correlation for a measurement from 1 month to the next was ρ, then the correlation over 2 months would be ρ^2, the correlation over 3 months would be ρ^3, and so forth. Because ρ is inclusive between -1 and $+1$, the correlation decreases with increasing time. In

addition, we allowed each individual to have his or her own initial level of the outcome variable, as a random effect in the model. This model also allows adjustment for other variables, such as age, gender, or presence of diabetes, in the prediction of the outcome variable when appropriate. Following standard practice, we included these potential confounding variables in the final model only when they are statistically significant or have a significant effect (*i.e.*, change the regression parameter substantially) on the primary relationship under study. The results of the study presented below are based on this mixed model approach; where appropriate, and to provide a basic summary of specific relationships, we have demonstrated some associations by providing simple correlations between specific variables.

RESULTS

The baseline levels for several measured parameters are shown in Table 1. The average serum creatinine was 3.4 ± 2.2 mg/dL, and the mean serum albumin was 3.8 ± 0.45 g/dL. The means (± SD) of selected nutritional parameters for different ranges of CrCl are shown in Table 2 to provide an overview of how these variables change with a change in CrCl. As can be seen, the mean DPI was 1.01 ± 0.21 g/kg per day for patients with CrCl over 50 mL/min and decreased to 0.85 ± 0.23 g/kg per day for patients with CrCl between 25 and 50 mL/min. The DPI further decreased to a level of 0.70 ± 0.17 g/kg per day for patients with CrCl between 10 and 24 mL/min. Most important, the mean DPI was 0.54 ± 0.17 g/kg per day for patients with CrCl below 10 mL/min. The progressive decrease in mean DPI was statistically significant ($P < 0.05$). There was also a progressive decrease in daily creatinine excretion (Figure 1), serum cholesterol, and transferrin (Figure 2), as well as in total CO_2, as CrCl decreases.

To provide further insight into the relationship between CrCl, DPI, and other nutritional parameters, Tables 3 and 4 provide the correlations of these variables with each other and with other nutritional parameters. The results show a strong positive correlation between DPI and CrCl ($r = 0.46$, $P < 0.0001$). In addition, the decreases in CrCl and DPI were also significantly associated with a decrease in total creatinine excretion, a marker of total body muscle mass ($r = 0.44$ and $P < 0.0001$, for both variables). Serum cholesterol, CO_2, and serum transferrin also decreased as both CrCl (Table 3) and DPI (Table 4) decreased. For each of these variables, the analysis demonstrated a stronger association of these parameters with CrCl levels than with DPI.

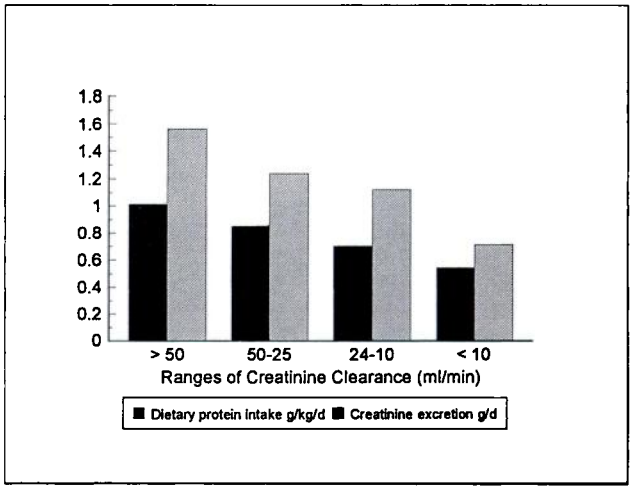

Figure 1. Average values of dietary DPI and total daily creatinine excretion of patients monitored during the study, classified according to different levels of CrCl.

The results of the definitive analysis, using the mixed model approach, are shown in Table 5. Although we assessed the importance of age (in decades), gender, race (white *versus* nonwhite), and diabetes, these variables were not important predictors in any relationship, except where specifically noted, so the adjustment for these variables was not included in the results shown. The rate of decrease in DPI, calculated from the model, was 0.0638 ± 0.0065 g/kg per day per 10 mL/min decrease in CrCl. This rate of decline was not substantially changed by consideration of gender, race, and diabetic status (change was less than 5% of the estimate for all models).

TABLE 2. Mean ± SD values of several nutritional parameters monitored during the study, classified according to different levels of renal function[a]

Parameter	CrCl			
	>50 mL/min	50 to 25 mL/min	24 to 10 mL/min	<10 mL/min
DPI[b] (g/kg per day)	1.01 ± 0.21	0.85 ± 0.23	0.70 ± 0.17	0.54 ± 0.16
Transferrin[b] (mg/dL)	313 ± 90	295 ± 80	260 ± 65	237 ± 52
Cholesterol[b] (mg/dL)	241 ± 70	235 ± 70	213 ± 47	213 ± 59
tCO_2[b] (mEq/L)	25.2 ± 3.7	24.3 ± 4.1	21.3 ± 3.6	20.1 ± 3.9
Prealbumin (mg/dL)	35.6 ± 7.5	37.4 ± 13.2	34.8 ± 10.0	37.9 ± 7.5
Crea-excr[b] (g/day)	1.56 ± 0.47	1.24 ± 0.46	1.12 ± 0.37	0.71 ± 0.23

[a] tCO_2, total bicarbonate; Crea-excr, total daily creatinine excretion.
[b] $P < 0.05$ with standard regression.

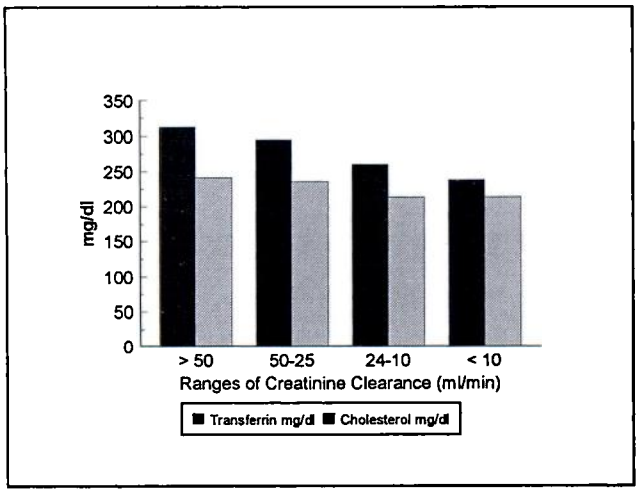

Figure 2. Average values of serum transferrin and cholesterol of patients monitored during the study, classified according to different levels of CrCl.

The mixed model analysis allowed us to further define the association of CrCl and other measured parameters (Table 5). There was an association between the percent decrease in each patient's initial weight with CrCl ($P < 0.01$). Further, the percent ideal body weight decreased by 0.38 ± 0.13% for each 10 mL/min decrease in CrCl. CrCl was also significantly associated with serum transferrin levels ($P < 0.01$), and the serum transferrin level decreased 16.7 ± 4.1 mg/dL for each 10 mL/min decrease in CrCl. In addition, there was evidence for an association of IGF-I with CrCl. On the basis of only 25 observations, IGF-I decreased 6.22 ± 1.94 ($P < 0.05$) for each 10 mL/min decrease in CrCl. No association was found between CrCl and the remaining parameters, namely, serum albumin, cholesterol, and prealbumin. Serum albumin, however, was associated with diabetes ($P = 0.016$). Thus, patients with diabetes had a lower predicted albumin level than did patients without diabetes (with a predicted difference of 0.162 ± 0.067 g/dL lower in diabetic patients).

DISCUSSION

The results of our study of CRF patients demonstrate that spontaneous DPI decreases significantly as renal failure progresses. This finding is consistent with the anorexia that is a well-known manifestation of uremia, especially toward food products that contain high-biologic-value proteins (9,10). The level of measured protein intake is particularly low in patients with CrCl between 24 and 10 mL/min (0.70 ± 0.17 g/kg per day) and decreases to 0.54 ± 0.16 g/kg per day at levels of CrCl less than 10 mL/min. This level of protein intake is below the suggested "safe" limits of DPI for uremic patients.

The cause of anorexia seen in uremia is not well defined. Several possible factors, generally described as "uremic toxins," have been proposed. These include several hormones such as parathyroid hormone and insulin, proteins like β-2 microglobulin, and metabolites such as middle molecules, guanidines, and polyamines (11). None of these substances have been definitely proved to be the specific cause or to have a dose-response relationship to anorexia. More recently, a low molecular-weight substance closely associated with anorexia has been proposed in an animal model, although its identity and specificity are not yet well defined (12).

A relationship between renal function and dietary protein and energy intake was earlier suggested by a report from the Modification of Diet in Renal Disease (MDRD) Study Group on the basis of the feasibility phase of the trial (14). In that pilot study, the protein and energy intakes of patients at baseline (before they were initiated on any dietary intervention) were also found to be significantly associated with their level of renal function. More recently, the same group reported their findings (in abstract form) regarding the 1,687 patients eligible for participation in the complete MDRD study (14). They suggested that there was a significant positive correlation between GFR and dietary protein as well as energy intake at the initial baseline visit. It is of note that both of these observations were cross-sectional and before any dietary intervention. Our study has confirmed the findings between renal function and DPI in a prospective design, without specific dietary intervention. We have not assessed the relationship between the energy intake and renal function because dietary recalls were not done as a part of our study.

The "minimal" daily protein requirement is one that maintains a neutral nitrogen balance and prevents malnutrition. This has been estimated to be a DPI of approximately 0.6 g/kg in healthy individuals (15). However, CRF patients have several metabolic abnormalities as renal failure progress, such as hormonal derangements and altered amino acid metabolism (16). Thus, the suggested intake of proteins for normal individuals may not necessarily apply to outpatient CRF patients, who may require higher levels because of concurrent metabolic abnormalities. Nevertheless, a few short-term studies in uremic subjects suggested that a neutral or positive nitrogen balance can be achieved in such patients with a minimum daily intake of 0.6 g/kg protein and 35 kcal/kg energy (17,18). It should be noted, however, that diets used in such studies contained "high-biologic-value" proteins and may not be typical of the "spontaneous" intake of CRF patients. The patients participating in such studies are also more stable and did not include diabetic patients. Finally, it should also be recalled that the estimated value of "minimal" daily protein requirements in healthy individuals and uremic patients are means of populations; a "safe level" of protein intake has been suggested to be at least the minimal requirement plus 2 SD, or approximately 0.75 g/kg per day (15).

TABLE 3. Pearson correlation analysis between CrCl and nutritional parameters[a]

Parameter	DPI	Creatinine Excretion	Chol	S. Alb	CO_2	Trans	Prealb	IGF-1
N	331	327	363	363	375	57	51	25
Correlation Coefficient	0.46	0.44	0.14	0.06	0.35	0.42	0.04	0.06
P Value	<0.0001	<0.0001	<0.01	>0.25	<0.0001	<0.01	>0.25	>0.25

[a] *N*, number of observations; S. Alb, serum albumin; Chol, serum cholesterol; Trans, serum transferrin; Prealb, serum prealbumin.

TABLE 4. Pearson correlation analysis between DPI and nutritional parameters[a]

Parameter	CrCl	Creatinine Excretion	Chol	S. Alb	CO_2	Trans	Prealb	IGF-I
N	331	293	307	307	314	47	41	19
Correlation Coefficient	0.46	0.44	0.12	0.11	0.19	0.33	0.38	0.44
P Value	<0.0001	<0.0001	<0.05	<0.05	<0.001	<0.05	<0.01	>0.05

[a] *N*, number of observations; S. Alb, serum albumin; Chol, serum cholesterol; Trans, serum transferrin; Prealb, serum prealbumin.

TABLE 5. The predicted changes (by mixed model analysis) in relation to the progression of renal failure in several nutritional parameters[a]

Parameter	Predicted Change	*P* Value
DPI (g/kg per day)	−0.0638 ± 0.0065	<0.0001
Weight (% initial wt)	−0.38 ± 0.13	<0.01
Transferrin (mg/dL)	−16.7 ± 4.1	<0.01
IGF-I (ng/mL)	−6.2 ± 1.9	<0.05
BUN (mg/dL)	4.5 ± 0.4	<0.0001
Total bicarbonate (mEq/L)	−0.53 ± 0.11	<0.0001

[a] All changes per 10 mL/min decrease in CrCl.

Several other caveats need to be mentioned with regard to the calculated DPI of our patients. Animal studies have shown that metabolic acidosis is a strong catabolic factor (19,20). Because the measurement of DPI from urinary urea nitrogen assumes that patients are in a neutral nitrogen balance (7), it is possible that our measurement of urea nitrogen output actually overestimated the protein intake, especially when renal failure was associated with total bicarbonate levels below 21 mEq/L. In addition, because we did not measure calorie intake, it is possible that a reduced calorie intake may contribute to a further catabolic stress and loss of endogenous proteins (21).

An important aim of our study was to establish possible associations between several indices of malnutrition and renal function and/or DPI. Several easily measurable nutritional indices worsened as renal function decreased. These include patients' weight, serum transferrin, serum cholesterol, and 24-h total creatinine excretion. These finding are consistent with the reports of the MDRD Study Group, both during the pilot study and during the comprehensive study (13,14). Specifically, those reports suggest that baseline GFR values are positively correlated with percent standard weight, arm muscle area, creatinine:height ratio, and serum transferrin and, in contrast to our study, with serum albumin. Our study documents that these changes occur in a progressive manner but, most important, that these parameters of malnutrition are substantially worsened when CrCl is below 10 mL/min.

In spite of the small number of observations, the mixed model analysis suggested a significant association between IGF-I levels and CrCl. This observation is important because IGF-I levels may be an early marker for worsening nutritional status, as compared with serum albumin. Using similar analysis, we were unable to find an association between prealbumin levels and CrCl. Although prealbumin has been proposed to be a reliable marker for nutrition in patients with steady-state renal function (22), such as chronic dialysis patients, this is probably not the case for patients with continuously changing renal function because the clearance and accumulation of prealbumin depends on the level of renal function. Note also that both of these parameters (IGF-1 and prealbumin) are strongly associated with the level of protein intake in our study patients as well as in other patient groups (23,24).

Although serum albumin is the most well-established marker for increased morbidity and mortality in the ESRD population (3), in our study, we observed no statistically significant relationship between serum albumin and the changes in CrCl; this may simply mean that the serum albumin of our patients did not decrease with decreasing CrCl or, conversely, may imply that serum albumin is not an early marker for CRF patients because of its long half-life and redistribution from extravascular into intravascular space (24). Because several other parameters of nutrition, such as patients' weights, serum transferrin, and cholesterol levels worsened as CrCl decreased, it is likely that the absence of a statistically significant relationship between serum albumin and CrCl is because of the long half-life of serum albumin. It is also

likely that our sample size was too small to detect such a relationship because studies with larger cohorts, such as MDRD, reported a significant positive correlation between serum albumin and GFR (14).

In summary, our study indicates that the spontaneous DPI of CRF patients decreases significantly as renal function declines. This decrease is notable particularly after CrCl is below 25 mL/min and is reduced to levels below the minimum daily protein requirements (determined in normals) when CrCl is below 10 mL/min. These low levels of protein intake are associated with the development of other indices of malnutrition. These findings imply that dietary protein restriction should be used cautiously in patients with CrCl below 25 mL/min; also, because of the large effect of malnutrition on subsequent survival, the early initiation of chronic dialysis therapy should be considered if the patient's DPI falls below 0.7 g/kg per day in spite of adequate nutritional counseling (25).

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REFERENCES

1. **Marckmann P:** Nutritional status and mortality of patients in regular dialysis therapy. J Intern Med 1989; 226:429–432.
2. **Schoenfeld PY, Henry RR, Laird NM, Roxe DM:** Assessment of nutritional status of the national cooperative dialysis study population. Kidney Int 1983;23:80–88.
3. **Lowrie EG, Lew NL:** Death risk in hemodialysis patients: The predictive value of commonly measured variables and an evaluation of death rate differences between facilities. Am J Kidney Dis 1990;15:458–482.
4. **Avram MM, Goldwasser P, Erroa M, Fein PA:** Predictors of survival in continuous ambulatory peritoneal dialysis pateints: The importance of prealbumin and other nutritional and metabolic markers. Am J Kidney Dis 1994;23: 91–98.
5. **Goldwasser P, Mittman M, Antignani A, *et al.*:** Predictors of mortality on hemodialysis. J Am Soc Nephrol 1993;3: 1613–1622.
6. **Hakim RM:** Initiation of dialysis. Adv Nephrol 1994;23: 295–309.
7. **Maroni B, Steinman TI, Mitch WE:** A method for estimating nitrogen intake of patients with chronic renal failure. Kidney Int 1985;27:58–61.
8. **Jennrich RI, Schluchter MD:** Unbalanced repeated-measures models with structured covariance matrices. Biometrics 1986;42:805–820.
9. **Hakim RM, Lazarus JM:** Progression of chronic renal failure. Am J Kidney Dis 1989;14:396–401.
10. **el-Lakany S, Eagon PK, Gavaler JS, Schade RR, Whiteside T, Van Thiel DH:** Gastrointestinal function, morphology, and immune status in uremia. Nutrition 1990; 6:461–468.
11. **Knochel JP:** Biochemical alterations in advanced uremic failure. In: Jacobson HR, Striker GE, Klahr S, Eds. The Principles and Practice of Nephrology. Philadelphia: BC Decker; 1991:682–689.
12. **Bergstrom J, Mamoun H, Anderstram B, Sodersten P:** Middle molecules (MM) isolated from uremic ultrafiltrate (UF) and normal urine induce dose-dependent inhibition of appetite in the rat [Abstract]. J Am Soc Nephrol 1994;5:488.
13. **Modification of Diet in Renal Disease Study Group (prepared by Kopple JD, *et al.*):** Nutritional status of patients with different levels of chronic renal failure. Kidney Int 1989;36:S184–S194.
14. **Modification of Diet in Renal Disease Study Group (prepared by Kopple JD, *et al.*):** Relationship between GFR and nutritional status-results from the MDRD study [Abstract]. J Am Soc Nephrol 1994;5:335.
15. **Young VR:** Nutritional requirements of normal adults. In: Mitch WE, Klahr S, Eds. Nutrition and the Kidney. Boston: Little, Brown and Company; 1993:1–34.
16. **Eknoyan G:** Effects of renal insufficiency on nutrient metabolism and endocrine function. In: Mitch WE, Klahr S, Eds. Nutrition and the Kidney. Boston: Little, Brown and Company; 1988:29–58.
17. **Kopple JD, Monteon FJ, Shaib JK:** Effect of energy intake on nitrogen metabolism in nondialyzed patients with chronic renal failure. Kidney Int 1986;29:734–742.
18. **Goodship THJ, Mitch WE, Hoerr RA, Wagner DA, Steinman TI, Young VR:** Adaptation to low-protein diets in renal failure: Leucine turnover and nitrogen balance. J Am Soc Nephrol 1990;1:66–75.
19. **Mitch WE, May RC, Maroni BJ:** Mechanisms for abnormal protein metabolism in uremia. J Am Coll Nutr 1989;8:305–309.
20. **Mitch WE, Medina R, Grieber S, *et al.*:** Metabolic acidosis stimulates muscle protein degradation by activating the adenosine triphosphate-dependent pathway involving ubiquitin and proteasomes. J Clin Invest 1994;93: 2127–2133.
21. **Slomowitz LA, Monteon FJ, Grosvenor M, Laidlaw SA, Kopple JD:** Effect of energy intake on nutritional status in maintenance hemodialysis patients. Kidney Int 1989; 35:704–711.
22. **Cano N, Feinandez JP, Lacombe P, *et al.*:** Statistical selection of nutritional parameters in hemodialysis patients. Kidney Int 1987;32 [Suppl 22]:S178–S180.
23. **Clemmons DR, Van Wyk JJ:** Factors influencing blood concentrations of somatomedins. J Clin Endocrinol Metab 1984;63:113–143.
24. **Waterlow JC:** Metabolic changes. In: Waterlow JC, Ed. Protein-energy Malnutrition. London: Edward Arnold; 1992:83–103.
25. **Hakim RM, Lazarus JM:** Initiation of dialysis. J Am Soc Nephrol 1995;6:1319–1328.