Acta Medica et Biologica
Vol. 45, No. 2, 89-94, 1997

Effects of One Week's Administration of Enalapril on the Renal Hemodynamic Response to the Ingestion of a Protein Meal in Healthy Subjects

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Received February 17 1997; accepted April 28 1997

Summary. **To determine whether the administration of an angiotensin-converting enzyme inhibitor (ACEI) affects the renal hemodynamic changes and the renal handling of urinary protein excretion in response to a protein meal ingestion, we examined the changes in creatinine clearance (CCr) and urinary albumin excretion rate (AER) in response to the ingestion of a meal of tuna fish before and after the administration of enalapril 2.5 mg/day for one week in 7 healthy male volunteers. Before treatment with enalapril, CCr 0, 1, 2, and 3 (before and 1, 2, and 3 h after protein loading; mean±SD) were 118.8±21.6, 136.8±21.7, 144.9*±23.2 and 135.9±15.3 ml/min/1.73 m^2 (*: $p<0.05$ vs CCr 0), respectively. Corresponding values after treatment with enalapril were 124.9±21.4, 146.6*±16.3, 139.3*±16.9 and 136.0*±22.7 ml/min/1.73 m^2 (*: $p<0.05$ vs CCr 0). Mean CCr 0, CCr 2 and individual maximal CCr after protein loading did not show any significant difference before and after enalapril administration (individual maximal CCr: before enalapril 150.3±13.7 vs after enalapril 153.4±13.6 ml/min/1.73 m^2, NS). Irrespective of the administration of enalapril, no significant differences were observed in AER values before and after the ingestion of the tuna fish meal. Thus, it might appear that in humans who have normal renal function and are receiving long-term treatment with ACEI, CCr 2 or individual maximal CCr after the ingestion of a tuna fish meal could be used as an index of "renal functional reserve". Furthermore, irrespective of the administration of enalapril, the ingestion of tuna fish meal did not affect the renal handling of urinary albumin excretion.**

Key words—renal functional reserve, angiotensin converting enzyme inhibitor, urinary albumin excretion rate.

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INTRODUCTION

Protein ingestion or amino acid (AA) infusion causes marked changes in renal hemodynamics such as increased renal plasma flow (RPF) and glomerular filtration rate (GFR).[1-7] The increments in GFR are often clinically used as an index of "renal functional reserve".[1,8,9]

With respect to the mechanism of protein-or amino acid-induced changes in renal hemodynamics, Woods et al. reported that such changes were mediated by tubuloglomerular feedback (TGF) because they were abolished during intravenous amino acid infusion when TGF was blocked.[10,11] Loop diuretics and a calcium channel antagonist were reported to blunt the renal response to a meat meal or amino acid infusion by blocking TGF.[12,13] These findngs suggest that the measurement of changes in renal hemodynamics after the ingestion of a protein meal in patients given these drugs leads to an underestimation of renal functional reserve. The results illustrate that it may be necessary to examine whether other drugs block TGF, when renal hemodynamic changes in response to protein meal ingestion are clinically used as an index of "renal functional reserve."

Angiotensin-converting enzyme inhibitors (ACEI) have been advocated as antihypertensive agents and also as agents that may possess specific advantages in decreasing proteinuria and slowing the progression of renal disease.[14,15] Well established specific effects of ACEI on the renal hemodynamics include falls in GFR, filtration fraction, and renal vascular resistance through a fall in intraglomerular capillary pressure by a predominantly post glomerular vasodilation.[16,17]

At least, the acute antiproteinuric effect of ACEI was thought to be mediated by these renal hemodynamic changes.[18,19] In view of the fact that ACEI are widely used in the treatment of renal disease and hypertension, it is important to elucidate whether or not these drugs block TGF and reduce the increments in GFR in response to protein meal ingestion or amino acid infusion when the evaluation of the renal functional reserve of patients treated wth ACEI is necessary, because the reduction of the increments in GFR in response to these stimuli leads to the underestimation of the renal functional reserve of patients. In contrast with loop diuretics and calcium antagonists, it is controversial whether the administration of ACEI influences renal hemodynamic changes in response to protein meal ingestion or amino acid infusion. Two conflicting reports exist: in one, long-term treatment with benazepril did not alter the renal hemodynamic changes in response to protein meal ingestion;[20] in the other, treatment with captopril for 3 days attenuated the increase in GFR in response to amino acids infusion.[21] It is also controversial whether or not the single-dose administration of ACEI influences the renal hemodynamic changes in response to protein meal ingestion[9,22,23] or intravenous amino acids infusion.[24-26]

This study was therefore undertaken to examine the effects of a one-week regimen of enalapril on changes in GFR after the ingestion of a tuna fish meal in healthy subjects. Ingestion of a tuna fish meal is more useful in out patients than amino acid infusion when volume overload is considered in elderly patinets.

Furthermore, as it is unknown whether ACEI affects the renal handling of urinary protein excretion in response to increased GFR by protein meal ingestion, the albumin excretion rate was studied before and after the administration of enalapril in response to the ingestion of a tuna fish meal in healthy subjects.

SUBJECTS AND METHODS

Subjects

Seven healthy male volunteers aged 20-36 years (mean age 25.7±6.2 years) were studied. All of the subjects shared the following characteristics: good nutritional status; no history of diabetes mellitus, renal systemic disease or other diseases; normal blood pressure; normal 24-hr urinary albumin excretion rate (7.3±3.6 μg/min); and normal 24-hr creatinine clearance (122.8±21.6 ml/min/1.73 m^2). No medication was allowed for two weeks before or during the study. The characteristics of the subjects are shown in Table 1.

Protocol

Protein loading tests were performed before and after treatment with oral enalapril 2.5 mg/day for one week. On the day before each protein loading test, 24-hr urine samples were collected from all of the subjects and the levels of albumin, urea nitrogen and creatinine in their urine samples were measured to calculate the 24-hr urinary albumin excretion rate (AER24), protein intake and 24hr-endogenous creatinine clearance (CCr24). Protein intake was calculated from urinary urea nitrogen (UUN) and estimated non-urea nitrogen (NUN) excretion of 31 mg $N \cdot kg^{-1} \cdot day^{-1}$ [27] as follows:

Table 1. Characteristics of 7 healthy subjects before and after administration of enalapril 2.5 mg/day for one week

	Enalapril		p-values
	Before	After	
Age (years)	25.7±6.2		
Body mass index (kg/m^2)	23.2±3.3		
Body surface area (m^2)	1.88±0.09		
Blood pressure (mmHg)	118.6±13.2/72.6±7.1	113.1±12.1/66.3±7.5	N.S.
Protein intake (g/day)	83.1±9.6	77.1±7.5	N.S.
CCr24 (ml/min/1.73 m^2)	122.8±21.6	121.3±9.9	N.S.
AER24 (μg/min)	7.3±3.6	6.6±2.6	N.S.

Values are expressed as mean±S.D.
CCr24, 24 hr endogenous creatinine clearance; AER24, 24 hr urinary albumin excretion rate.

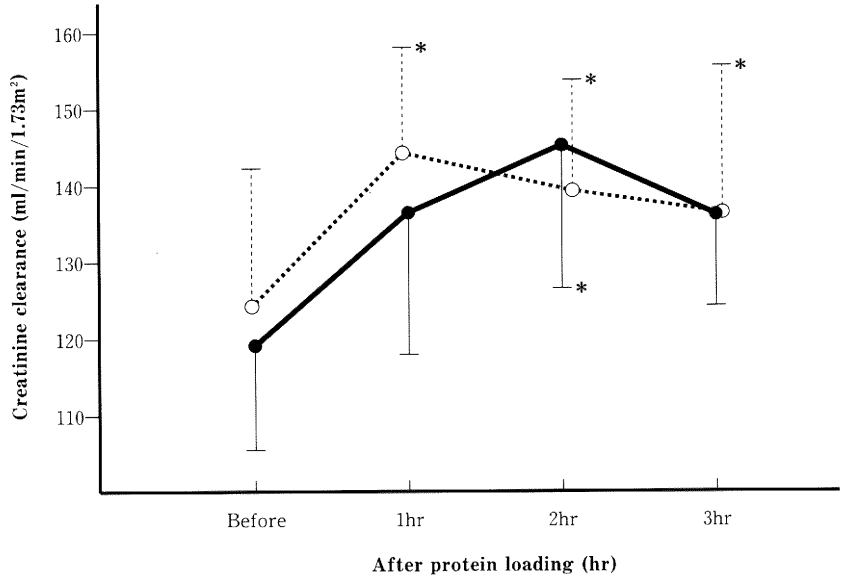

Fig. 1. Changes in glomerular filtration rate (creatinine clearance) before and after protein loading, ●, before enalapril administration; ○, after enalapril administration for one week. Vertical bars indicate S. D.; *, $p<0.05$ vs. before protein loadings.

$I_N = UUN + NUN$, $I_N \times 6.25 =$ protein intake (g/day)

In the fasting state after the collection of 24-hr urine samples, the subjects voided at 8 : 00 a.m. and ingested 500 ml of water. They were required to void every 60 min until 12 : 00 noon, and urine volume was replaced with water after each voiding for maintenance of adequate urine volume. Blood samples were drawn at the midpoint of each collection period. After the first collection period, the subjects ingested 0.8 g/kg body weight of protein in the form of a cooked tuna fish meal. During each collection period, urine volume was recorded and creatinine and albumin levels in both urine and serum were measured hourly. Before and 1, 2, and 3 h after protein loading, endogenous creatinine clearance (CCr 0, 1, 2, 3) and the urinary albumin excretion rate (AER 0, 1, 2, 3) were calculated. Thus, there were four periods of clearance determination.

Analytical techniques and statistics

Creatinine in urine and serum was measured by Follin's method, urinary urea nitrogen, by urease-UV method, and albumin levels in urine and serum, by radio immunoassay (RIA). CCrs were always corrected for body surface area and expressed as/1.73 m². Body surface area was calculated from height and body weight. All of the data are expressed as mean values±SD. Statistical significance was calculated using the Student's paired *t*-test.

RESULTS

Table 1 shows the characteristics of the 7 subjects before and after the administration of enalapril 2.5 mg/day for one week. Blood pressure levels did not decrease after treatment with enalapril. In addition, protein intake, CCr24, and AER24 on the day before the protein loading test were not significantly different between before and after the administration of enalapril.

Fig. 1 shows the serial CCr before and after protein loading. On the day before the initiation of treatment with enalapril, CCr 0 was 118.8±21.6 ml/min/1.73 m² and CCr 1, 2 and 3 were 136.8±21.7, 144.9*±23.2 and 135.9±15.3 ml/min/1.73 m² (mean ±SD, *: $p<0.05$ vs CCr 0). Thus, before the administration of enalapril, only CCr 2 increased significantly. On the day after treatment with enalapril for one week, CCr 0 was 124.9±21.4, and CCr 1, 2 and 3 were 146.6*±16.3, 139.3*±16.9, and 136.0*±22.7 ml/min/1.73 m². After the administration of enalapril, CCr 1, 2 and 3, therefore, showed a significant increase. There were no significant changes in CCr 0 and CCr 2 between the

Table 2. Changes in urinary albumin excretion rate following protein loading

	Enalapril	
	Before	After
AER 24	7.3±3.6	6.6±2.6
AER 0	8.3±4.7	4.9±1.3
AER 1	4.7±1.2	4.9±1.2
AER 2	5.0±2.6	4.2±1.1
AER 3	4.5±1.4	4.1±1.2

Values are expressed as mean ±S.D. (μg/min).
AER24, 24 hr urinary albumin excretion rate on the day before protein loading test; AER0, 1, 2, 3, urinary albumin excretion rate before 1, 2, and 3 hr after protein loading.

period before and after administration of enalapril. Although the maximal increase in individual CCr occurred in different collection periods, mean individual maximal CCr after protein loading, which increased significantly compared with CCr 0, did not differ significantly before and after enalapril administration (150.3±13.7 vs 153.4±13.6 ml/min/1.73 m^2, NS).

Table 2 shows the changes in AER before and after protein loading. Irrespective of the administration of enalapril, no significant differences were observed in AER values before and after protein loading.

DISCUSSION

The present study showed that the regular administration of enalapril did not affect mean CCr 0, CCr 2, or individual maximal CCr after ingestion of a tuna fish meal in healthy volunteers. In the present study, GFR was evaluated on the basis of endogenous creatinine clearance. Chan et al. reported that the use of creatinine clearance seemed to underestimate the renal functional reserve evaluated by measuring the increment in GFR after acute protein load, because the large contribution of tubular secretion to urinary creatinine clearance might obscure the magnitude of the post meal increment in GFR in patients with chronic glomerular disease and reduced baseline GFR[8]. On the basis of their report, the renal functional reserve of the subjects in our present study might be underestimated, but at least the significant rise in CCr after the ingestion of a tuna fish meal suggests that true GFR increased significantly in our study. Additionally, in healthy subjects, it was reported that changes in creatinine clearance in response to a protein meal ingestion were parallel to changes in inulin clearance[1] or iothalamate clearance.[28] For this reason, the results of the significant increase in CCr in response to a tuna fish meal in the present study were judged reliable. Although the evaluation of the renal functional reserve by measuring the increment in creatinine clearance after protein load seems to be especially useful in out patient clinics because of its simple and convenient method, it is recommended that the increment in true GFR should be masured by means of inulin clearance, iothalamate clearance or ^{54}Cr-EDTA clearance in case of a small response in creatinine clearance after protein load. In other words, the evaluation of renal functional reserve on the basis of the changes in creatinine clearance in response to a protein meal ingestion should be recognized as a screening test.

In patients with hypertension receiving long-term treatment with ACEI, Valvo et al.[20] reported that a 6-week regimen of benazepril 10 mg/day did not affect the response of GFR to a beef meal ingestion. Their results were consistent with our findings, although we evaluated healthy subjects and they studied patients with hypertension. In contrast, in healthy volunteers, Eisenhauer et al.[21] reported that the increase of GFR in response to amino acid infusion was attenuated after treatment with 25 mg of captopril three times daily for 3 days and a single-dose 50 mg of captopril on the day of amino acid infusion. Although the reason for the discrepancy between our results and those of Eisenhauer is unclear, the consistency of our results with those of Volvo et al. suggests that the discrepancy was due to differences in stimuli used in our and Eisenhauer's studies.

In contrast with the chronic administration of ACEI, Krishna et al.[9,22] reported that the single-dose administration of enalapril did not affect the increase in GFR after protein meal ingestion. However, Chagnac et al.[23] reported that a single-dose administration of enalapril attenuated the increase in GFR in response to the ingestion of a protein meal. In regard to amino acid infusion in healthy subjects after a single-dose administration of ACEI, Slomowitz et al.[24] and Heering et al.[25] independently reported that the increase in GFR in response to amino acid infusion was not reduced. Contrastively, Boehler et al.[26] reported an abolished GFR response to amino acid infusion after a single-dose administration of captopril. Thus, the renal hemodynamic response to protein meal ingestion or amino acid infusion is inconsistent after a single-dose administration of ACEI; the exact reason for this discrepancy remains unclear. At least, it dose not seem to be due to the variability of the individual absorption of a protein

meal since the inconsistency of the GFR response was reported in both protein loading tests and amino acid infusion tests. However, considering that treatment with ACEI tends to be long-term, it is clinically important to determine the chronic effect of these drugs on the renal hemodynamic response to the ingestion of a protein meal. The agreement of our results with those of Valvo et al.[20] suggests that the long-term administration of ACEI dose not affect the renal hemodynamic response to the ingestion of a protein meal. Thus, it could be that, at least in humans who have normal renal function and are receiving long-term treatment with ACEI, the serial increases in GFR after protein meal ingestion can be used as an index of "renal functional reserve."

Although only mean CCr 2 showed a statistically significant increase compared with mean CCr 0 before the administration of enalapril, the mean CCr 1, 2 and 3 showed statistically significant increases after treatment with enalapril. It remains to be elucidated why CCr 1 and 3 rose after the administration of enalapril. Neither the mean CCr 2 nor individual maximal CCr after protein loading were affected by enalapril. Therefore, it appears that, in humans receiving long-term treatment with ACEI, either the mean CCr 2 or individual maximal CCr after the ingestion of a protein meal could be used as an index of "renal functional reserve," although in cases of abnormal changes of creatinine clearance in response to a protein meal, the exact measurement of GFR is necessary.

In the present study, the urinary albumin excretion rate (AER) in response to a tuna fish meal showed no significant changes in healthy subjects. This finding is consistent with results from Nakamura et al..[29] In contrast, some authors reported that AER increased significantly after the ingestion of cooked beef.[30-33] Recently, Nakamura et al.[34] reported that a tuna fish meal did not increase AER whereas a cooked beef meal did, and that urinary thromboxane B_2 (TXB_2) excretion rose significantly following protein loading with beef but did not change after loading with tuna fish or beef with ethyl-icosapentate (EPA). Additionally, they reported that the tuna fish meal contained unsaturated-fatty acids such as EPA, which were not present in the beef meal. On the basis of these results, they speculated that the increase in urinary TXB_2 induced by loading with beef caused AER to increase, and changing the renal prostanoid profile by adding EPA to beef blocked the increase in AER. Thus, changes in AER after protein ingestion may differ corresponding with the difference of the type of protein ingested.

In the case of amino acid infusion which contains no fatty acids as a stimuli inducing the increase in GFR in normal subjects, one author reported that the increase in GFR did not combine with the increase in AER[28]; conversely, another reported an increase in the fractional clearance of albumin.[35] Although the reason for this discrepancy is unknown, the relative high age of the subjects in the latter report (44 ± 3: mean $\pm$SE, the former; 30.1 ± 5.5: mean$\pm$SD) might be responsible for this discrepancy.

REFERENCES

1) Bosch JP, Saccaggi A, Lauer A, Ronco C, Belledonne M, Glabman S: Renal functional reserve in humans. *Am J Med* **75:** 943-950, 1983.
2) Bosch JP, Saccaggi A, Lauer A, Glabman S: Short-term protein loading in assessment of patients with renal disease. *Am J Med* **77:** 873-879, 1984.
3) Bosch JP, Lew S, Glabman S, Lauer A: Renal hemodynamic change in humans. *Am J Med* **81:** 809-815, 1986.
4) Hostertter TH: Human renal response to a meat meal. (Renal Fluid Electrolyte Physiol 19) *Am J Physiol* **250**: F613-F618, 1986.
5) Graf H, Stummvoll KH, Lunger A, Prager R: Effect of amino acid infusion on glomerular filtration rate. *N Eng J Med* **308:** 159-160, 1983.
6) Ter Wee PM, Geerkings W, Rosman JB, Sluiter WJ, van der Geest S, Donker AJM: Testing renal reserve filtration capacity with an amino acid solution. *Nephron* **41:** 193-199, 1985.
7) Ruilope LM, Rodicio J, Robles RG, Sancho J, Miranda B, Granger JP, Romero JC: Influence of low sodium diet on the renal response to amino acid infusions in humans. *Kidney Int* **31:** 992-999, 1987.
8) Chan AYM, Cheng MLL, Keil LC, Meyers BD: Functional response of healthy and diseased glomeruli to a large, protein-rich meal. *J Clin Invest* **81:** 245-254, 1988.
9) Krishna GG, Kapoor SC: Preservation of renal reserve in chronic renal disease. *Am J Kidney Dis* **17:** 18-24, 1991.
10) Woods LL, Mizelle HL, Montani JP, Hall JE: Mechanisms controlling renal hemodynamics and electrolyte excretion during amino acids. (Renal Fluid Electrolyte Physiol 20) *Am J Physiol* **251**: F303-F312, 1986.
11) Woods LL, Young EW: Impaired renal hemodynamic response to protein feeding in dogs with experimental Fanconi syndrome. (Renal Fluid Electrolyte Physiol 30) *Am J Physiol* **261:** F303-F312, 1991.
12) Woods LL, Smith BE, De Young DR: Control of renal hemodynamics after protein feeding: role of calcium channels. (Renal Fluid Electrolyte Physiol 32) *Am J Physiol* **263**: F1044-F1050, 1992.
13) Woods LL, Smith BE, De Young DR: Regulation of

renal hemodynamics after protein feeding: effects of proximal and distal diuretics. (Regulatory Integrative Comp Physiol 33) *Am J Physiol* **264**: R337-R344, 1993.
14) De Zeeuw D, Heeg JE, De Jong PE: The antiproteinuric effect of angiotensin converting enzyme inhibitors in human renal disease. In: Andreucci VE, Fine LG (eds) Int Yearbook Nephrol. Springer Verlag, London 1992, p 99-113.
15) Lewis EM, Hunsicker LG, Bain RP, Rohde RD: The effect of angiotensin-converting-enzyme-inhibition on diabetic nephropathy. *N Eng J Med* **329:** 1456-1462, 1993.
16) Anderson S, Meyer TW, Rennke HG, Brenner BM: Control of glomerular hypertension limits glomerular injury in rats with reduced renal mass. *J Clin Invest* **76:** 612-619, 1985.
17) Pelayo JC, Quan AH, Shanley PF: Angiotensin II control of the renal microcirculation in rats with reduced renal mass. (Renal Fluid Electrolyte Pysiol 27) *Am J Physiol* **258**: F414-F422, 1990.
18) Heeg JE, De Jong DE, van der Hem GK, De Zeeuw D: Angiotensin II dose not acutely reverse the reduction of proteinuria by long-term ACE inhibition. *Kidney Int* **40:** 734-741, 1991.
19) Gansevoort RT, Heeg JE, De Jong DE: Dissociation between the course of the hemodynamic and antiproteinuric effects of angiotensin I converting enzyme inhibition. *Kidney Int* **44:** 579-584, 1993.
20) Valvo E, Casagrande P, Bedogna V, Dal Santo F, Alberti D, Fontanarosa C, Braggio P, Maschio G: Renal functional reserve in patients with essential hypertension: effect of inhbition of renin-angiotensin system. *Clin Sci* **78:** 585-590, 1990.
21) Eisenhauer T, Talartschik J, Scheler F: Angiotensin converting enzyme inhibition by captopril reduced amino acid-mediated increase in glomerular filtration rate. *J Hypertens* **4 (Suppl. 5):** S258-S260, 1986.
22) Krishna GG, Newell G, Miller E, Heeger P, Smith R, Polansky M, Kapoor S, Hoeldtke R: Protein-induced glomerular hyperfiltration: role of hormonal factors. *Kidney Int* **33:** 578-583, 1988.
23) Chagnac A, Gafter U, Zevin D, Hirsch I, Markovitz J, Levi J: Enalapril attenuated glomerular hyperfiltration following a meat meal. *Nephron* **51:** 466-469, 1989.
24) Slomowitz LA, Hirschberg R, Kopple JD: Captopril augments the renal response to an amino acid infusion in diabetic adults. (Renal Fluid Electrolyte Pysiol 24) *Am J Physiol* **255**: F755-F762, 1988.
25) Heering P, Wyes D, Plum J, Grabensee B: Influence of the renin-Angiotensin system and atrial natriuretic peptide on renal functional reserve. *Nephron* **66:** 14-20, 1994.
26) Boehler J, Woitas R, Keller E, Reetze-Bonorden P, Schollmeyer PJ: Effects of nifedipine and captopril on glomerular hyperfiltration in normotensive man. *Am J Kidney Dis* **20:** 132-139, 1992.
27) Maroni BJ, Steinman TI, Mitch WE: A method for estimating nitrogen intake of patients with chronic renal failure. *Kidney Int* **27:** 58-65, 1985.
28) Sølling K, Christensen CK, Sølling J, Christensen JS, Mogensen CE: Effect on renal haemodynamics, glomerular filtration rate and albumin excretion of high oral protein load. *Scand J Clin Lab Invest* **46:** 351-357, 1986.
29) Nakamura H, Takasawa M, Kasahara S, Tsuda A, Momotsu T, Ito S, Shibata A: Effects of acute protein loads of different sources on renal function of patients with diabetic nephropathy. *Touhoku J Exp Med* **159:** 153-162, 1989.
30) Viberti GC, Bongnetti E, Wiseman MJ, Dodds R, Gross JL, Keen H: Effect of protein-restricted diet on renal response to a meat meal in humans. (Renal Fluid Electrolyte Physiol 22) *Am J Physiol* **253**: F388-F393, 1987.
31) Amore A, Coppo R, Roccatello D, Martina G, Rollino C, Basolo B, Novelli F, Amprimo MC, Cavalli G, Piccoli G: Single kidney function: effect of acute protein and water loading on microalbuminuria. *Am J Med* **84:** 711-717, 1988.
32) Fioretto P, Trevisan R, Valerio A, Avogaro A, Borsato M, Doria A, Semplicini A, Sacerdoti D, Jones S, Bognetti E, Viberti GC, Nosadini R: Impaired renal response to a meat meal in insulin-dependent diabetes: role of glucagon and prostaglandins. (Renal Fluid Electrolyte Physiol 27) *Am J Physiol* **258:** F675-F683, 1990.
33) Kontessis P, Jones S, Dodds R, Trevisan R, Nosadini R, Fioretto P, Borsato M, Sacerdoti D, Viberti GC: Renal, metabolic and hormonal responses to ingestion of animal and vegetable proteins. *Kidney Int* **38:** 136-144, 1990.
34) Nakamura H, Nakamura T, Nakagawa O, Ohyama Y, Tani N, Ito S, Shibata A: Effect of acute protein loading of different sources on renal function in healthy volunteers and diabetic patients. *J Jpn Diab Soc* **39:** 613-618, 1996.
35) Nosadini R, Trevisan R, Fioretto P, Semplicini A, Sama B, Velussi M, Da Campo GL, Avogaro A, Vizzaccaro A, Donadon V, Mongillo A, Doria A: Kidney hemodynamics after ketone body and amino acid infusion in normal and IDDM subjects. *Diabetes* **38:** 75-83, 1989.