Variable Effects of Soy Protein on Plasma Lipids in Hyperlipidemic and Normolipidemic Hemodialysis Patients

Shu-Tzu Chen, MS, Shyang-Hwa Ferng, MD, Chwei-Shiun Yang, MD, Sheng-Jeng Peng, MD, Huei-Rong Lee, MS, and Jiun-Rong Chen, PhD

**Background:** Hyperlipidemic factors contribute to the high cardiovascular risk in hemodialysis patients. Soy protein has decreased some atherogenic lipid concentrations in subjects with normal renal function. This study evaluates the effect of soy protein on serum lipid profiles in hyperlipidemic and normolipidemic hemodialysis patients.

**Methods:** Nineteen hyperlipidemic and 18 normolipidemic hemodialysis patients were enrolled in a randomized, double-blind, placebo-controlled, clinical trial. After a 4-week run-in phase, subjects in each category were randomly assigned to 2 groups. Thirty grams of isolated soy protein or milk protein was consumed daily as a beverage at breakfast or postdialysis for 12 weeks.

**Results:** In hyperlipidemic subjects, soy protein intake significantly decreased total cholesterol levels by 18.6% (95% confidence interval [CI], −11.4 to −25.8; P = 0.04), triglyceride levels by 43.1% (95% CI, −34.0 to −52.2; P = 0.02), non–high-density lipoprotein cholesterol levels by 23.6% (95% CI, −14.7 to −32.5; P < 0.01), apolipoprotein B levels by 15.4% (95% CI, −5.4 to −25.4; P = 0.01), and insulin levels by 49.8% (95% CI, −23.3 to −66.1; P < 0.01). Low-density lipoprotein cholesterol concentration was decreased significantly (−25.8%; 95% CI, −8.3 to −42.7; P = 0.01), and high-density lipoprotein cholesterol level was increased significantly (17%; 95% CI, 2 to 32.0; P = 0.03), but there was no significant difference compared with the milk protein group (−5.5% ± 16.9% and 7.0% ± 11.8%, respectively). There were no significant changes in serum lipid and lipoprotein concentrations in normolipidemic subjects.

**Conclusion:** These results indicate soy protein substitution has lipid-lowering effects in hyperlipidemic hemodialysis patients. However, soy protein intake had little effect on plasma lipid levels in normolipidemic hemodialysis patients.

**INDEX WORDS:** Soy protein; hemodialysis (HD); lipoprotein; apolipoprotein; triglyceride; cholesterol; lipid.

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**Patients on Maintenance Hemodialysis Treatment** are at elevated atherogenic risk, and hyperlipidemia appears to be one of the major risk factors. Management of hyperlipidemia is one of the important therapeutic goals for hemodialysis patients. The National Cholesterol Educational Program Step I diet, which restricts fat intake (<30% of total calories), is recommended as an initial treatment to decrease serum lipid levels. However, patients on hemodialysis therapy have a number of other nutritional concerns, and it is difficult to decrease fat intake to less than 30% of total calories in the usual hemodialysis diet. The diet usually prescribed for maintenance hemodialysis patients contains high-protein (>1.2 g/kg/d). Foods containing proteins are major sources of dietary fats and cholesterol. Research showed that soy protein intake improved lipid profiles and enhanced the hypocholesterolemic effect of the National Cholesterol Educational Program Step I diet in hypercholesterolemic individuals with normal renal function and patients with nephrotic syndrome and diabetic nephropathy. However, the effect of soy protein in normolipidemic subjects is conflicting. There are reports in the literature that hypocholesterolemia is associated with greater mortality in maintenance hemodialysis patients. Because of the limited data available, it is unclear whether soy protein has a clinically relevant and beneficial effect on plasma lipid level management in hemodialysis patients.

In this clinical trial, we examine the effects of soy protein consumption on lipid metabolism in hemodialysis patients and evaluate the lipemic re-
response between normolipidemic and hyperlipidemic subjects.

METHODS

Patients

Nondiabetic maintenance hemodialysis patients from the Hemodialysis Center of Cathay General Hospital, Taipei, Taiwan, were recruited for this randomized, double-blind, placebo-controlled, clinical trial. Criteria for enrollment as normolipidemic subjects included a fasting plasma total cholesterol (TC) concentration between 150 and 200 mg/dL (3.9 and 5.2 mmol/L) and a fasting plasma triglyceride (TG) concentration less than 200 mg/dL (<2.3 mmol/L). Hyperlipidemic subjects were screened for an initial fasting plasma TC concentration greater than 240 mg/dL (>6.2 mmol/L), fasting plasma TG concentration greater than 200 mg/dL (>2.3 mmol/L), and no lipid-lowering drug therapy within the past month. All subjects had been on regular thrice-weekly maintenance hemodialysis therapy for at least 1 year, with no intercurrent illness and a normal fasting plasma glucose concentration. Exclusion criteria were the presence of liver disease, thyroid disease, severe hypertension, or cancer and the use of any medication known to affect lipid concentrations. Primary end points of the study are changes in lipid, lipoprotein, and apolipoprotein proteins. The protocol was approved by the Human Investigation Review Committees of Cathay General Hospital and Taipei Medical University. All subjects were informed about the study and provided written informed consent before beginning the study. Subjects who failed to comply with the diet or had a weight variation of 3 kg or greater during the study were excluded from the final analysis.

Of 46 patients screened, 42 patients gave their informed consent, and 39 subjects completed the study. Three patients withdrew from the study because of gastrointestinal problems (e.g., constipation, diarrhea, nausea, and gastrointestinal upset). Two subjects who failed to consume the assigned protein dose (1.2 g/kg/d), as assessed by the dietitian, were excluded from statistical analyses. Nineteen hyperlipidemic and 18 normolipidemic subjects completed the study and were included in statistical analyses. We chose subjects with hyperlipidemia and normolipidemia to examine effects of soy protein in hemodialysis patients in accordance with following reasons.

1. Initial serum cholesterol concentrations had a powerful effect on changes in serum TC and low-density lipoprotein cholesterol (LDL-C) concentrations. Serum cholesterol level increases greater than 240 mg/dL (>6.2 mmol/L) have been implicated as a risk factor for cardiovascular heart disease. For these reasons, we chose subjects with serum cholesterol levels greater than 240 mg/dL (>6.2 mmol/L) to potentially maximize the effect of soy protein.

2. The hypocholesterolemic effect of soy protein also was shown in normocholesterolemic subjects with normal renal function. There are reports in the literature that maintenance hemodialysis patients with low-normal (<150 mg/dL [<3.9 mmol/L]) serum cholesterol levels have greater mortality than those with greater cholesterol levels.

3. Patients with a serum cholesterol concentration between 200 and 240 mg/dL are not the primary target population of cholesterol-lowering therapy. Diet modifications (low-fat, low-cholesterol) have been shown to decrease serum cholesterol levels by approximately 14%. In this population, a change in eating pattern could achieve a desirable lipid level, in our experiences. Thus, we did not include patients with serum cholesterol concentrations of 200 to 240 mg/dL (5.2 to 6.2 mmol/L) in the present study.

Study Design and Diet

During a 4-week run-in phase, all subjects followed the usual hemodialysis diet (35% fat, 1.2 g/kg/d of protein, and ~32 to 35 kcal/kg/d of energy). A registered dietitian instructed subjects on this diet and counseled them about their individual needs for protein, fat, and energy. After the run-in phase, baseline blood samples were drawn and subjects in each category (hyperlipidemic or normolipidemic) were randomly assigned to 1 of 2 dietary treatment subgroups that provided isolated soy protein (ISP) or milk protein (control group). For 12 weeks, all subjects continued to consume the usual hemodialysis diet and were asked to consume one 30-g packet of ISP (Supro 660; Protein Technologies Int, St Louis, MO) or one 30-g packet of milk protein (P93; Sensoto, BV, Holland) each day at breakfast (nondialysis day) or after dialysis (dialysis day). Individual dietary counseling was provided with the goal of making a dietary adjustment to incorporate the test protein into the diet without causing weight gain or changes in protein or energy intake. This requirement was reinforced at clinic visits and monitored during the study by reviewing dietary recalls. Clinic visits were conducted every dialysis day. At each clinic visit, the study protein was delivered, and any leftover packets from the previous visit were collected to determine compliance. A 24-hour dietary recall was performed for all subjects for 3 days every 4 weeks. The 3 days included a dialysis day, a nondialysis day, and a Sunday. Compliance with dietary protein intake was measured by using the 24-hour dietary recall and calculating the protein equivalent of nitrogen appearance (PNA) from urea nitrogen appearance determination. ISP was fortified with calcium (calcium lactate) to amounts similar to those found in milk protein. Test products were formulated in sealed packets. The test products were isonenergetic and contained equal amounts of carbohydrate and fat to provide similar calories to those found in 30 g of meat protein. It was suggested that the test product be mixed with 200 mL of water or soup, and shakers were provided for mixing purposes. Throughout the study, all subjects were asked to maintain their edema-free body weight and take no nutritional supplements for the duration of the investigation. No lipid-lowering drug therapy or medication known to affect lipid concentration was used during the trial period. Subjects, study staff, and laboratory
technicians were blinded to treatment assignments during the trial period.

**Blood Sampling and Laboratory Methods**

Blood samples were collected every 4 weeks after subjects had fasted for approximately 10 to 12 hours. Laboratory assessments included fasting serum lipids (TC, TG, high-density lipoprotein cholesterol [HDL-C], LDL-C, apolipoprotein A-I [apoA-I] and B [apoB]), insulin, glucose, urea nitrogen, uric acid, calcium, phosphate, and albumin. Body weight was measured every dialysis day; edema-free body weight was obtained postdialysis and used to calculate body mass index. Daily nutrient intake was estimated from the 24-hour dietary recall and analyzed by using standard food composition tables.

Serum TC and TG were measured enzymatically, whereas HDL-C and LDL-C were quantified by means of the homogeneous assay method using an automated analyzer (Olympus AU2700 automated chemistry analyzer; Tokyo, Japan). Serum apoA-I and apoB concentrations were measured by means of immunoturbidimetry with the use of an automated analyzer (Integra 800; Roche, Mannheim, Germany). All serum nutritional parameters were determined by using an automated analyzer (Olympus AU2700). Serum insulin concentration was measured by using the microparticle enzyme immunoassay using an Abbott AxSYM analyzer (Abbott Laboratories, Abbott Park, IL).

Normalized PNA was calculated by using a 2–blood urea nitrogen, single-pool, variable-volume model. Hemodialysis adequacy was monitored by measurement of a urea kinetic model (Kt/V, calculated from the natural log formula).

**Statistical Analysis**

Estimated sample size was calculated by means of the method of Cohen, using the large effect of Cohen’s d value (d = 0.9). Estimated minimum sample size was 9 to 13 subjects, with 80% to 90% statistical power at a significance level of α = 0.5.

Statistical analyses were performed using SAS software (version 8.2; SAS Institute, Cary, NC). All statistical tests were 2-tailed, and significance for all analyses was set at P less than 0.05. Changes from baseline within each group were evaluated by using paired t-test, and differences at each time were compared by using 1-way analysis of variance and Dunnett test. Between-group differences were tested by using Student t-test.

**RESULTS**

Subject characteristics at baseline are listed in Table 1. Subject body weights remained stable in each group throughout the study. No significant differences were found among different groups in nutrient intake, serum nutritional parameters, and hemodialysis adequacy, and no significant changes between baseline and study end were noted within any group (Table 2).

Plasma TC and TG concentrations measured at baseline and 4, 8, and 12 weeks throughout the study are listed in Table 3. Both TC and TG levels decreased significantly in hyperlipidemic subjects who consumed soy protein and were significantly less than levels in subjects who consumed milk protein at week 12. In addition, there were significant differences in time-by-treatment interactions in the ISP group of hyperlipidemic subjects. No significant changes in TC or TG concentrations were observed in the milk group of hyperlipidemic subjects or the 2 normolipidemic subjects.

Lipoprotein, apolipoprotein, and insulin concentrations are listed in Table 4. In hyperlipidemic subjects, non–HDL-C, apoB, TC/HDL-C ratio, and insulin values for the ISP group decreased significantly and were significantly less than values for the milk group at week 12. LDL-C concentration decreased significantly, and HDL-C concentration increased significantly in the ISP group of hyperlipidemic subjects at week 12, but no significant difference was observed between the 2 hyperlipidemic groups. Soy protein intake significantly de-

<table>
<thead>
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<th>Table 1. Baseline Characteristics of Subjects</th>
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<td>Variable</td>
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<tr>
<td></td>
</tr>
<tr>
<td>Women</td>
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<tr>
<td>Men</td>
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<tr>
<td>Age (y)</td>
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<tr>
<td>Body mass index (kg/m²)</td>
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<td>Duration of dialysis therapy (y)</td>
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NOTE. Data expressed as mean ± SD or number of patients. Body mass index was calculated from edema-free body weight and height. There were no significant differences between groups in age, body mass index, or years of dialysis therapy.
significant differences in changes between the groups. To convert albumin in g/dL to g/L, multiply by 10.

To convert cholesterol in mg/dL to mmol/L, multiply by 0.02586; TG in mg/dL to mmol/L, multiply by 0.01129.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Hyperlipidemic Subjects</th>
<th>Normallipidemic Subjects</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Milk Group (n = 9)</td>
<td>ISP Group (n = 10)</td>
</tr>
<tr>
<td>Energy (kcal/kg)</td>
<td>32.2 ± 6.3</td>
<td>32.8 ± 1.95</td>
</tr>
<tr>
<td>Protein (g/kg)</td>
<td>1.2 ± 0.2</td>
<td>1.2 ± 0.3</td>
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<tr>
<td>Fat (% of energy)</td>
<td>36.7 ± 0.7</td>
<td>36.8 ± 0.8</td>
</tr>
<tr>
<td>Carbohydrates (% of energy)</td>
<td>48.7 ± 2.7</td>
<td>48.3 ± 1.8</td>
</tr>
<tr>
<td>Normalized PNA* (g/kg/d)</td>
<td>1.3 ± 0.3</td>
<td>1.4 ± 0.4</td>
</tr>
<tr>
<td>Kt/V</td>
<td>1.9 ± 0.3</td>
<td>1.8 ± 0.2</td>
</tr>
<tr>
<td>Albumin (g/dL)</td>
<td>4.0 ± 0.3</td>
<td>4.1 ± 0.3</td>
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NOTE. Data expressed as mean ± SD. Nutrient intake was calculated from 3-day 24-hour dietary recall. There were no significant differences in changes between the groups. To convert albumin in g/dL to g/L, multiply by 10.

Abbreviation: Kt/V, urea kinetic model calculated from the natural log formula.

*Calculated by using the 2–blood urea nitrogen, single-pool, variable-volume model.

creased TC levels by 18.6% (95% confidence interval [CI], −11.4 to −25.8; P = 0.04), TG levels by 43.1% (95% CI, −34.0 to −52.2; P = 0.02), non–HDL-C levels by 23.6% (95% CI, −14.7 to −32.5; P < 0.01), apoB levels by 15.4% (95% CI, −5.4 to −25.4; P = 0.01),

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<tbody>
<tr>
<td></td>
<td>Milk Group (n = 9)</td>
<td>ISP Group (n = 10)</td>
</tr>
<tr>
<td>TC (mg/dL)</td>
<td>254.3 ± 16.7</td>
<td>265.7 ± 28.2</td>
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<tr>
<td>Week 4</td>
<td>232.8 ± 21.9</td>
<td>239.5 ± 24.3</td>
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<td>Week 8</td>
<td>238.6 ± 25.9</td>
<td>228.1 ± 30.9</td>
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<tr>
<td>Week 12</td>
<td>257.7 ± 23.7</td>
<td>216.7 ± 28.1</td>
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<tr>
<td>TG (mg/dL)</td>
<td>343.4 ± 137.6</td>
<td>333.2 ± 114.6</td>
</tr>
<tr>
<td>Week 4</td>
<td>275.3 ± 106.5</td>
<td>276.7 ± 107.2</td>
</tr>
<tr>
<td>Week 8</td>
<td>310.3 ± 163.3</td>
<td>227.6 ± 93.2</td>
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<tr>
<td>Week 12</td>
<td>307.9 ± 132.4</td>
<td>185.7 ± 62.6</td>
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NOTE. Data expressed as mean ± SD. Values in the same column with different superscript letters significantly differ, P < 0.05, by the significance of the time-by-treatment interaction in the general linear model. There were no significant changes in the milk group of hyperlipidemic subjects or in either group of normallipidemic subjects. To convert cholesterol in mg/dL to mmol/L, multiply by 0.02586; TG in mg/dL to mmol/L, multiply by 0.01129.

*Significantly different from the milk group.
LDL-C levels by 25.8% (95% CI, −8.3 to −42.7; P = 0.01), and insulin levels by 49.8% (95% CI, 23.3 to 66.1; P < 0.01) and significantly increased HDL-C levels by 17% (95% CI, 2 to 32.0; P = 0.03) in hyperlipidemic subjects. The 95% CI of mean changes in TC, TG, non–HDL-C, LDL-C, HDL-C, apoB, and insulin levels did not include zero, and statistical power of all tests was greater than 90%. There were no significant changes between the 2 hyperlipidemic groups in apoA-I concentrations. Serum insulin levels (P = 0.050) and TC/HDL-C ratios (P = 0.051) in normolipidemic subjects decreased more in the ISP group than the milk group, but there were no significant differences in lipoprotein and apolipoprotein concentrations between the 2 normolipidemic groups at study end.

**DISCUSSION**

Results from this study show that soy protein has a beneficial effect on serum lipid profiles in hyperlipidemic hemodialysis patients. Replacement of 30 g of daily protein intake with soy protein in the usual hemodialysis diet of hyperlipidemic subjects for 12 weeks produced significant decreases in TC, LDL-C, non–HDL-C, and apoB concentrations and significant increases in HDL-C levels at the end of the study, whereas milk protein had no such effect. In addition, time-by-treatment interaction of TC levels was significant in the soy protein group of hyperlipidemic subjects. TC concentration also decreased in the milk group of hyperlipidemic subjects in the first few weeks, but the decrease was not significant throughout the study (P = 0.286). This change may have been attributed to decreasing saturated fat and cholesterol contents in the diet during the study period, whereas the reversed increase in TC concentration after week 4 may have been caused by casein being used as the major protein component in this group. Casein is known to be more hypercholesterolemic than soy protein.21,22
Decreases in apoB and non–HDL-C concentrations with soy protein consumption were shown in several clinical trials. In the present study, the decrease in apoB levels paralleled that in LDL-C levels, consistent with findings from Potter et al. Significant decreases in apoB and non–HDL-C concentrations in the soy group may indicate a positive effect on antiatherogenesis in hyperlipidemic hemodialysis patients. Recent data from a cohort of 525 hemodialysis patients showed that non–HDL-C level was a significant and independent predictor of cardiovascular disease mortality in hemodialysis patients. ApoB level also is an independent factor associated with morbidity of myocardial infarction in hemodialysis patients.

Prominent characteristics of uremic dyslipidemia are an increase in TG-rich apoB-containing lipoprotein levels and a decrease in HDL-C concentrations. The effect of soy protein on TG levels in the general population is conflicting. Some studies reported no significant effect of soy protein on TG concentrations in subjects with normal renal function, although decreases were identified. The clinical significance of consuming soy protein on HDL-C concentration also remains controversial. In the present study, TG concentration significantly decreased and HDL-C level significantly increased in the soy protein group of hyperlipidemic subjects. These results suggest that lipid metabolism is affected by soy protein in hemodialysis patients, and soy protein may be effective in ameliorating uremic hyperlipidemia. Attman et al found insulin resistance and changes in insulin-mediated processes to be related significantly to decreased catabolism and clearance of TG-rich apoB-containing lipoproteins. In this study, serum insulin concentrations in the ISP group of hyperlipidemic subjects were significantly less than those in the control group. These data indicate that improved insulin resistance may have an important role on the effect of soy protein on lipoprotein metabolism. No significant changes were observed in the current study in lipid, lipoprotein, and apolipoprotein concentrations in the 2 normolipidemic groups, but there were decreases in insulin concentrations and TC/HDL-C ratios in the soy protein group. Results indicate that soy protein consumption may help normolipidemic hemodialysis patients increase HDL-C levels and attenuate insulin resistance.

Although the present observations suggest favorable effects on cholesterol-related risk factors for cardiovascular disease, a study reported that lipoprotein(a) concentration was twice as high in those consuming a soy protein diet (20% of total energy consumption) than those consuming a casein diet in normolipidemic healthy men. Thus, more information is needed on the effect of soy protein on levels of lipoprotein(a) and other atherogenic lipoproteins to identify whether soy protein could provide atherogenesis protection.

Results of the present study show that LDL-C level is significantly decreased by 25.6% and HDL-C level is significantly increased by 17.0% in the soy protein group of hyperlipidemic subjects compared with baseline; the literature reported that the best changes in LDL-C concentrations range from 10% to 13%, and HDL-C level improvement, in the range of 2% to 7%. Causes of variability in lipid responses to soy protein remain unknown. Our data are similar to findings from Nilausen and Meinertz, in which mean decrease in LDL-C levels with a soy protein diet was 26%, and increase in HDL-C levels, 17%. LDL-lowering and HDL-elevating effects of the soy protein diet in hyperlipidemic subjects in the present study were more effective than those reported in some studies, possibly because subjects in present study had much greater initial serum TG concentrations or that monounsaturated fat was contained in the test products. Decreases in serum cholesterol and LDL-C concentrations were related directly to initial serum cholesterol concentrations. Initial serum TG concentrations may affect responses to soy protein administered to hyperlipidemic hemodialysis patients. The combination of soy protein and monounsaturated fat may achieve a more effective decrease in LDL-C level and increase in HDL-C concentrations. A study was reported that enhance the effectiveness in reducing serum cholesterol of diet combination of cholesterol-lowering foods.

Estimation of dietary protein intake from the 24-hour dietary recall and normalized PNA confirmed that test proteins were consumed in substitution of other animal proteins. Body weights remained stable in each group throughout the study, suggesting good compliance with dietary...
prescriptions. There were no significant differences in nutritional parameters between groups at the end of the study, indicating that when substituting soy protein (30 g/d) for animal protein, nutritional status of hemodialysis patients can be maintained.

Despite these observations, our findings have the following limitations: (1) the number of subjects studied was small, (2) duration of dietary periods may have been too short to know long-term compliance and effects, and (3) lipoprotein and apolipoprotein were measured at the beginning and week 12 only, and it is not clear whether soy protein substitution had a time-by-treatment interaction effect on LDL-C and HDL-C levels.

However, few data are available for defining the effects of soy protein on lipid metabolism in hemodialysis patients. This study shows that TC, LDL-C, non–HDL-C, apoB, and TG concentrations of hyperlipidemic hemodialysis patients decreased significantly and there was a significant increase in HDL-C levels with soy protein administration. Soy protein intake had little effect on plasma lipid levels in normolipidemic hemodialysis patients. In conclusion, our findings show that replacing a part of the daily protein with soy protein could be used in the nutritional management of moderately hyperlipidemic hemodialysis patients. The 12-week efficacy of soy protein administration in 19 hyperlipidemic hemodialysis patients should encourage more extensive investigations into its long-term effect.31

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REFERENCES

9. Nilausen K, Meinertz H: Lipoprotein(a) and dietary proteins: Casein lowers lipoprotein(a) concentrations as compared with soy protein. Am J Clin Nutr 69:419-425, 1999