Effects of feeding 4 levels of soy protein for 3 and 6 wk on blood lipids and apolipoproteins in moderately hypercholesterolemic men

Sandra R Teixeira, Susan M Potter, Ronald Weigel, Sandra Hannum, John W Erdman Jr, and Clare M Hasler

ABSTRACT
Background: Replacing animal protein with soy protein has been shown to reduce total and LDL-cholesterol concentrations in humans. However, the minimum amount of soy protein required for significant reduction of blood lipids is not known.

Objective: We evaluated the amount of soy protein needed to reduce blood lipids in moderately hypercholesterolemic men.

Design: Eighty-one men with moderate hypercholesterolemia (total cholesterol concentration between 5.70 and 7.70 mmol/L) were studied. After a 3-wk lead-in on a Step I diet, total cholesterol was measured and subjects were randomly divided into 5 groups. For 6 wk, each group received 50 g protein/d, which included isolated soy protein (ISP) and casein, respectively, in the following amounts: 50:0, 40:10, 30:20, 20:30, and 0:50 (control group) g. Blood was collected at baseline and weeks 3 and 6 of the intervention.

Results: At week 6, significant reductions ($P < 0.05$) from baseline compared with the control group were found for non-HDL cholesterol and total cholesterol and apolipoprotein (apo) B for all ISP groups (except total cholesterol with 40 g ISP). At week 3, significant reductions ($P < 0.05$) were found in apo B for the groups that consumed $\geq 30$ g ISP and in non-HDL cholesterol for the groups that consumed $\geq 40$ g ISP. HDL-cholesterol, apo A-I, lipoprotein(a), and triacylglycerol concentrations were not significantly affected by dietary treatment.

Conclusion: Our findings show that consuming as little as 20 g soy protein/d instead of animal protein for 6 wk reduces concentrations of non-HDL cholesterol and apo B by $\leq 2.6\%$ and 2.2%, respectively.

KEY WORDS Men, soy protein, diet, apolipoproteins, hypercholesterolemia, blood lipids, total cholesterol, apolipoprotein B, low-density-lipoprotein cholesterol, LDL cholesterol, non-HDL cholesterol, hyperlipidemia

INTRODUCTION
A large body of evidence indicates that increased blood cholesterol concentrations, specifically those of LDL cholesterol, increase the risk of coronary heart disease (CHD) (1). Furthermore, several clinical trials have shown that lowering serum cholesterol concentrations reduces new CHD events and mortality from CHD in patients who do not have established CHD (2, 3). Numerous studies have shown, in both animals and humans, that blood cholesterol concentrations may be reduced by consuming soy protein in place of animal protein (4, 5). Although several clinical trials have confirmed the hypocholesterolemic effects of high amounts of soy protein, the minimum amount required for a biologically significant reduction of blood cholesterol concentrations remains to be defined. Therefore, this dose-response study was designed to determine the effects of graded amounts of dietary soy protein on plasma cholesterol concentrations in moderately hypercholesterolemic men.

SUBJECTS AND METHODS
Subjects
A total of 92 free-living men aged 23–74 y were recruited from the Champaign-Urbana, IL, area by using fliers, letters, and physician referrals. The primary inclusion criteria were age $\geq 23$ y, total cholesterol (TC) concentration at screening between 5.69 and 7.76 mmol/L (220–300 mg/dL), and a body mass index (BMI; in kg/m$^2$) between 20 and 33. Exclusion criteria included the presence of diabetes mellitus, thyroid disease, or any other chronic illness that could affect blood lipid concentrations or limit the individual’s ability to participate in the study, and the use of any medication known to affect lipid concentrations. Subjects who failed to comply with the diet or had a weight variation $\leq 3$ kg during the study were excluded from the final analysis. Subject characteristics at baseline are shown in Table 1 and Table 2.
TABLE 1
Baseline characteristics of subjects

<table>
<thead>
<tr>
<th>Group</th>
<th>0 g ISP (control)</th>
<th>20 g ISP</th>
<th>30 g ISP</th>
<th>40 g ISP</th>
<th>50 g ISP</th>
</tr>
</thead>
<tbody>
<tr>
<td>(n = 16)</td>
<td>(n = 15)</td>
<td>(n = 18)</td>
<td>(n = 17)</td>
<td>(n = 15)</td>
<td></td>
</tr>
<tr>
<td>Age (y)</td>
<td>49.8 ± 12.9</td>
<td>41.9 ± 12.3</td>
<td>43.3 ± 11.4</td>
<td>47.4 ± 9.4</td>
<td>44.6 ± 10.3</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>87.3 ± 14.6</td>
<td>89.2 ± 17.6</td>
<td>86.0 ± 12.1</td>
<td>84.6 ± 9.9</td>
<td>87.2 ± 11.2</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>27.1 ± 3.9</td>
<td>27.8 ± 3.8</td>
<td>27.1 ± 3.6</td>
<td>26.4 ± 2.9</td>
<td>27.4 ± 3.6</td>
</tr>
</tbody>
</table>

Diets and study design

During a 3-wk lead-in period, all subjects followed a National Cholesterol Education Program Step I diet (<30% of energy from fat, <10% of energy from saturated fat, and <300 mg cholesterol/d). A registered dietitian instructed the subjects on this diet and counseled them about their individual needs for protein, fat, and energy. The subjects were also told to maintain a consistent level of activity throughout the study. After this 3-wk lead-in period, all subjects continued on the Step I diet and were randomly assigned to 1 of 5 experimental groups. For 6 wk, each group received 50 g protein/d, which included 1) isolated soy protein (ISP; Supro Plus 675HG; Protein Technologies International, St Louis) with 1.9 mg total isoflavone aglycone units/g protein, 2) casein in the form of calcium caseinate (Alanate 391; New Zealand Milk Products, Wellington, New Zealand), or 3) both ISP and casein. The 5 groups received 50, 40, 30, 20, and 0 g (control) ISP with 0, 10, 20, 30, and 50 g casein, respectively.

The test products were incorporated into a variety of baked products and ready-to-mix beverages (Protein Technologies International), which the subjects received at breakfast 5 d/wk on-site. Subjects were given enough study products for the remainder of the day and for weekends. Their consumption of study foods was monitored 5 times/wk to track compliance and acceptance of the foods. Subjects kept 3-d food records during alternate weeks. The registered dietitian used these food records to determine the average daily nutrient intake with a computerized nutrient database, NUTRITIONIST IV (version 4.1; N-Squared Computing, Salem Park, OR) as soon as the food records were received. When noncompliance was noted, the subjects were counseled on how to modify their diets to achieve the prescribed nutrient intake.

Blood lipid and apolipoprotein analyses

After the subjects fasted for 12 h, blood samples were collected on 2 consecutive days at 3 time points during the study: at the end of the 3-wk lead-in period (baseline) and at weeks 3 and 6 during the study. The results from the 2 consecutive days were averaged and that value was used for statistical analysis. The blood was collected in tubes with and without EDTA and was centrifuged at 800 × g for 15 min at 18°C to obtain plasma or serum, which was stored at −70°C until analyzed.

Concentrations of TC (6), HDL cholesterol (7), and triacylglycerol were analyzed enzymatically with a Hitachi 917 system (Hitachi Inc, Indianapolis) by the Core Laboratory for Clinical Studies, Washington University School of Medicine, St Louis. Intersay CVs were <1.5% for TC, <2.5% for HDL cholesterol, and <2.0% for triacylglycerol. Non-HDL cholesterol (VLDL cholesterol + LDL cholesterol) was calculated by subtracting HDL cholesterol from TC. Lipoprotein(a) concentrations were measured as described by Taddei-Peters et al (8) with a commercial, enzyme-linked immunosorbent assay kit (PerImmune Inc, Rockville, MD). Lipoprotein(a) concentrations [nmol/L apolipoprotein(a)] were obtained by nonlinear regression with a 4-parameter, logistic equation that approximated the shape of the standard curve. The intraassay CV was <10%, as recommended by the manufacturer of the assay kit. Apolipoprotein (apo) A-I and B concentrations were measured in the second-day blood samples by immunoturbidimetric assays, as described by Rifai and King (9), with commercial kits (Raichem, San Diego). Apo A-I and B concentrations were determined from a calibration curve constructed by a second-order polynomial curve fit to measurements of the assay standards; the intraassay CV was <4%.

Other measurements

Body weight was monitored twice weekly (Detecto Physician’s Scale; Detecto Scale Company, Webb City, MO) and BMI was calculated. The daily activity level was assessed by using 3-d activity records that were completed concurrently with the dietary intake records. An activity score was calculated from the information provided on the activity records, as described by Bouchard et al (10).

Blood isoflavone concentrations were measured in the second-day plasma samples collected at baseline and at weeks 3 and 6. The isoflavones daidzein, equol, dihydrodaidzein, α-desmethyl angolentoin, genistein, 4-ethyl phenol, and glycitin were measured by HPLC coulometric array detection (model 5600 CoulArray 8-channel detector; Ralston Analytic Laboratories, Ralston Purina Co, St Louis). Basic steps in this procedure were performed as described by Coward et al (11). The total isoflavone concentrations were determined by summing the concentrations of all the individual isoflavones.
Individual treatment effects were examined only if the multi-
represents the change from baseline in the control group (12).
was included in the model as a covariate, coded as the devia-
each group contrasted with the control group (dummy coding).
isoflavones. The outcome measure was the change from base-
cholesterol, triacylglycerol, apo B, apo A-I, lipoprotein(a), and
indexes: TC, HDL cholesterol, non-HDL cholesterol, TC:HDL

### Statistical analysis

We used multiple linear regression to analyze the effects of
different amounts of soy protein on the concentrations of 9 blood
indexes: TC, HDL cholesterol, non-HDL cholesterol, TC:HDL
cholesterol, triacylglycerol, apo B, apo A-I, lipoprotein(a), and
isoﬂavones. The outcome measure was the change from baseline
for each subject, with treatment effects represented as
each group contrasted with the control group (dummy coding).
For each subject, the baseline value of each outcome variable
was included in the model as a covariate, coded as the deviation
from the baseline mean. In this model, the intercept term
represents the change from baseline in the control group (12).
Individual treatment effects were examined only if the multi-
ple $R^2$ for the model was significant ($P < 0.05$). The treat-
ment effect for each outcome variable was tested first for the
week 6 results. Those variables with significant results were
further tested for the week 3 results. This was done to reduce
experiment-wise type-I error.

Between-group differences in nutrient intake, physical activity,
BMI (at baseline and weeks 3 and 6), and baseline subject
characteristics were identiﬁed by using the Kruskal-Wallis test.
Protein and energy intakes were analyzed separately for each
group to compare baseline and week 6 values by using the
Wilcoxon paired-sample test.

All statistical analyses were conducted with SAS (version 6;
SAS Institute, Cary, NC) with an $a$ level of 0.05. For concentra-
tions of TC, non-HDL cholesterol, and apo B, and TC:HDL
cholesterol, it was predicted that treatment with soy protein at all
doses would result in a decrease relative to the control group; for
isoﬂavone concentrations, an increase relative to the control
group was predicted. For these outcomes with directional pre-
dictions, one-tailed $P$ values were calculated in evaluating the
values were used.

### Table 2

Plasma lipid and serum apolipoprotein concentrations at baseline and weeks 3 and 6 of the intervention

<table>
<thead>
<tr>
<th>Group</th>
<th>TC (mmol/L)</th>
<th>HDL cholesterol (mmol/L)</th>
<th>Non-HDL cholesterol (mmol/L)</th>
<th>TC:HDL cholesterol</th>
<th>Triacylglycerol (mmol/L)</th>
<th>Apo B (g/L)</th>
<th>Apo A-I (g/L)</th>
<th>Lipoprotein(a) (nmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td>6.08 ± 0.22</td>
<td>1.10 ± 0.08</td>
<td>4.98 ± 0.24</td>
<td>2.14 ± 0.28</td>
<td>1.40 ± 0.08</td>
<td>1.38 ± 0.05</td>
<td>42.8^1</td>
<td>Baseline</td>
</tr>
<tr>
<td>Week 3</td>
<td>6.24 ± 0.24</td>
<td>1.08 ± 0.07</td>
<td>5.17 ± 0.25</td>
<td>2.61 ± 0.37</td>
<td>1.54 ± 0.09</td>
<td>1.37 ± 0.05</td>
<td>1.39 ± 0.06</td>
<td>Week 3</td>
</tr>
<tr>
<td>Week 6</td>
<td>6.29 ± 0.24</td>
<td>1.10 ± 0.07</td>
<td>5.19 ± 0.25</td>
<td>2.32 ± 0.25</td>
<td>1.50 ± 0.10</td>
<td>1.39 ± 0.05</td>
<td>1.39 ± 0.07</td>
<td>Week 6</td>
</tr>
<tr>
<td>0 g ISP (control)</td>
<td>(n = 16)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>20 g ISP</td>
<td>6.01 ± 0.18</td>
<td>1.13 ± 0.09</td>
<td>4.88 ± 0.19</td>
<td>2.52 ± 0.31</td>
<td>1.47 ± 0.08</td>
<td>1.41 ± 0.07</td>
<td>1.29 ± 0.05</td>
<td>Baseline ^1</td>
</tr>
<tr>
<td>30 g ISP</td>
<td>6.03 ± 0.18</td>
<td>1.12 ± 0.08</td>
<td>4.91 ± 0.18</td>
<td>2.59 ± 0.31</td>
<td>1.40 ± 0.07</td>
<td>1.29 ± 0.05</td>
<td>1.38 ± 0.10</td>
<td>Week 3</td>
</tr>
<tr>
<td>40 g ISP</td>
<td>5.96 ± 0.20</td>
<td>1.11 ± 0.08</td>
<td>4.86 ± 0.20</td>
<td>2.05 ± 0.22</td>
<td>1.41 ± 0.07</td>
<td>1.47 ± 0.07</td>
<td>1.40 ± 0.06</td>
<td>Week 6</td>
</tr>
<tr>
<td>50 g ISP</td>
<td>6.28 ± 0.18</td>
<td>1.13 ± 0.05</td>
<td>5.15 ± 0.19</td>
<td>2.18 ± 0.28</td>
<td>1.47 ± 0.07</td>
<td>1.47 ± 0.07</td>
<td>1.45 ± 0.07</td>
<td>Baseline ^1</td>
</tr>
<tr>
<td>(n = 15)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(n = 18)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(n = 17)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(n = 15)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

^1 ISP, isolated soy protein; TC, total cholesterol; apo, apolipoprotein.
^2 $\bar{x}$ ± SEM unless otherwise indicated.
^3 Median; range in parentheses.
RESULTS

Dietary intake, physical activity, body mass index, and blood isoflavone concentrations

Consumption of the study protein was well accepted by most of the subjects. However, 2 of the 8 subjects who dropped out described possible allergic reactions, such as skin rash and itching. No significant differences were found among the different groups for the nutrient intakes that we analyzed, except for cholesterol intake at week 3 (P = 0.049, Kruskal-Wallis test). Although the average intake of total dietary cholesterol was < 300 mg/d in all groups (230.9, 116.7, 136.1, 199.5, and 223.4 mg/d for the groups consuming 0, 20, 30, 40, and 50 g ISP, respectively), some subjects had higher intakes occasionally.

Protein intake increased in all the groups between baseline and both weeks 3 and 6, because most of the subjects did not completely substitute the 50 g study protein for their usual dietary protein. Differences between baseline and week 6 were statistically significant in all the groups (P < 0.01, Wilcoxon paired-sample test). Energy intake also increased in all the groups from baseline to weeks 3 and 6, but significant differences between baseline and week 6 were found in only the groups that consumed 0, 20, 30, 40, and 50 g ISP, respectively.

On average, subjects from all the groups consumed < 20% of total energy from protein and 57% from carbohydrates. Fat consumption was < 23% of total energy intake, with < 6% as saturated fat. No significant differences were found between groups for BMI or physical activity.

At baseline, no significant differences in plasma total isoflavone concentrations were found between groups (Kruskal-Wallis test) (Figure 1). As determined by multiple linear regression, plasma total isoflavone concentrations were significantly higher than those of the control group at week 6 for all groups that received ISP (P = 0.04 for 20 g ISP, P = 0.005 for 30 g ISP, and P < 0.0001 for 40 and 50 g ISP). These effects were also seen at week 3 for the groups that consumed 30, 40, and 50 g ISP (P = 0.0136, P < 0.0001, and P < 0.0001, respectively). At week 6, the average plasma total isoflavone concentrations ranged from 244 nmol/L (control group) to 1141 nmol/L (40-g-ISP group).

Plasma lipids and serum apolipoproteins

Blood concentrations of lipids and apolipoproteins in each group at baseline and at weeks 3 and 6 are shown in Table 2. Non-HDL-cholesterol concentrations were significantly reduced at week 6 in all groups that received ISP (Figure 2 and Table 3). Adjusted mean changes of −0.139, −0.163, −0.095, and −0.182 mmol/L were found for the groups that received 20, 30, 40, and 50 g ISP, respectively. At week 3, significant reductions were found only when 40 and 50 g ISP were consumed, with adjusted mean changes of −0.236 and −0.278 mmol/L, respectively.

The reduction in plasma TC concentration (Figure 3 and Table 3) was significant at week 6 for the groups that consumed 20, 30, and 50 g ISP (adjusted mean changes of −0.126, −0.115, and −0.167 mmol/L, respectively). For the 40-g-ISP group, a nonsignificant reduction of 0.053 mmol/L was found. At week 3, the multiple R^2 for the multiple linear regression was not significant (P = 0.059; Figure 3). However, there were identifiable trends when ≥40 g ISP was consumed. The adjusted mean changes were −0.230 and −0.240 mmol/L (P = 0.01 for both; Figure 3) for the 40- and 50-g-ISP groups, respectively. This suggests that the effects identified at week 6 were also apparent at week 3 for the 40- and 50-g-ISP groups.

Serum apo B concentrations (Figure 4 and Table 4) were significantly reduced at week 6 in all groups that consumed ISP. Adjusted mean changes of −0.055, −0.113, −0.055, and
20.063 g/L were found for the 20-, 30-, 40-, and 50-g-ISP groups, respectively. At week 3, significant reductions were found when 30 g ISP was consumed. The adjusted mean changes were 0.059, 0.116, and 0.087 g/L for the 30-, 40-, and 50-g-ISP groups, respectively. At week 6, no significant changes were found for concentrations of HDL cholesterol, triacylglycerol, apo A-I, or lipoprotein(a) or TC:HDL cholesterol in any group that received ISP (Tables 3 and 4).

**DISCUSSION**

Replacement of 20 g animal protein (casein) with soy protein in the context of a Step I diet for 6 wk produced significant reductions in plasma non-HDL-cholesterol (<2.6%) and TC (1.8%) concentrations in moderately hypercholesterolemic men. Replacement of animal protein with 30, 40, and 50 g soy protein resulted in significant reductions in non-HDL-cholesterol concentrations of 3.0%, 1.5%, and 4.5%, respectively. At week 3, non-HDL-cholesterol concentrations were significantly lower in the 30-, 40-, and 50-g-ISP groups compared to the control group.

**TABLE 3**

<table>
<thead>
<tr>
<th>Group</th>
<th>0 g ISP (control)</th>
<th>20 g ISP</th>
<th>30 g ISP</th>
<th>40 g ISP</th>
<th>50 g ISP</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(n = 16)</td>
<td>(n = 15)</td>
<td>(n = 18)</td>
<td>(n = 17)</td>
<td>(n = 15)</td>
</tr>
<tr>
<td>TC (mmol/L)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Week 6</td>
<td>0.222 NS</td>
<td>0.026 NS</td>
<td>0.024 NS</td>
<td>0.029 NS</td>
<td>0.030 NS</td>
</tr>
<tr>
<td>HDL cholesterol (mmol/L)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Week 6</td>
<td>0.224 NS</td>
<td>0.024 NS</td>
<td>0.025 NS</td>
<td>0.026 NS</td>
<td>0.027 NS</td>
</tr>
<tr>
<td>Non-HDL cholesterol (mmol/L)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3</td>
<td>0.194 NS</td>
<td>0.040 NS</td>
<td>0.020 NS</td>
<td>0.236 NS</td>
<td>0.278 NS</td>
</tr>
<tr>
<td>Week 6</td>
<td>0.224 NS</td>
<td>0.044 NS</td>
<td>0.021 NS</td>
<td>0.048 NS</td>
<td>0.031 NS</td>
</tr>
<tr>
<td>TC:HDL cholesterol</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Week 6</td>
<td>0.52 NS</td>
<td>0.51 NS</td>
<td>0.71 NS</td>
<td>0.11 NS</td>
<td>0.09 NS</td>
</tr>
<tr>
<td>Triacylglycerol (mmol/L)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Week 3</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>Week 6</td>
<td>0.664 NS</td>
<td>0.632 NS</td>
<td>0.782 NS</td>
<td>0.443 NS</td>
<td>0.872 NS</td>
</tr>
</tbody>
</table>

1 Values adjusted for baseline. ISP, isolated soy protein; TC, total cholesterol; ND, not determined.
2 For the comparison with baseline.
3 For the comparison with the control group.
4 Not determined because the $R^2$ values indicated nonsignificance.
5 Not determined because results at week 6 were not significant.
only 40 and 50 g soy protein produced significant reductions (4.64% and 5.84%, respectively). Most earlier studies used much higher amounts of soy protein, which is not a practical approach in many individuals. Twenty grams of soy protein can be incorporated quite easily into a typical American diet, thus enabling the use of this dietary means of lowering blood cholesterol. Further, our findings show that replacement of animal protein with soy protein improved the cholesterol-lowering effect of a Step I diet without lowering HDL-cholesterol concentrations or increasing triacylglycerol concentrations. Finally, this amount of ISP (20 g) is similar to the qualifying amount of soy protein (25 g) proposed in a health claim petition that was approved by the Food and Drug Administration (13).

Other studies have confirmed that similar amounts of soy protein produce significant decreases in blood lipid concentrations. Bakhit et al (14) reported decreases in TC concentrations in men (baseline TC $\geq 5.7$ mmol/L) who consumed 25 g soy protein/d. Furthermore, in a recent study by Crouse et al (15), the effects of 25 g soy protein with different amounts of isoflavones were examined in moderately hyperlipidemic men and women. The authors found that 25 g of the soy protein with the highest isoflavone content (62 mg aglycone units/d) significantly reduced TC and LDL-cholesterol concentrations. Among the subjects who received soy protein with 37 mg aglycone units/d, significant reductions were found only in subjects with the highest initial TC and LDL-cholesterol concentrations. In the present

FIGURE 3. Adjusted mean change from baseline in plasma total cholesterol (TC) concentration in each group [0, 20, 30, 40, and 50 g isolated soy protein (ISP)] at week 3 ($R^2 = 0.13$, $P = 0.0590$) and at week 6 ($R^2 = 0.15$, $P = 0.0339$) of the intervention. Values were adjusted for baseline and obtained by multiple linear regression. $R^2$ is the proportion of total variance in outcome accounted for by the amount of dietary ISP, adjusted for the baseline concentration of TC. * * Significantly different from control group (0 g ISP): * $0.01 < P < 0.05$, ** $P < 0.01$.

FIGURE 4. Adjusted mean change from baseline in serum apolipoprotein B (apo B) concentration in each group [0, 20, 30, 40, and 50 g isolated soy protein (ISP)] at week 3 ($R^2 = 0.28$, $P = 0.0001$) and at week 6 ($R^2 = 0.20$, $P = 0.0049$) of the intervention. Values were adjusted for baseline and obtained by multiple linear regression. $R^2$ is the proportion of total variance in outcome accounted for by the amount of dietary ISP, adjusted for the baseline concentration of apo B. * * * Significantly different from control group (0 g ISP): * $0.01 < P < 0.05$, ** $P < 0.01$. 
study, 20 g of the ISP that contained a similar amount of
isoflavones (37.5 mg aglycone units/d) significantly decreased
non-HDL-cholesterol and TC concentrations by week 6.

Apo B concentrations have been proposed as an indicator
of CHD risk (16). Previous studies conducted with soy protein at the
University of Illinois by Bakhit et al (14), Potter et al (17), and
Baum et al (18) showed varied effects of soy on apo B concentra-
tions. Bakhit et al (14) and Baum et al (18) did not observe
significant alterations in the concentrations of this apolipoprotein
in moderately hypercholesterolemic men and postmenopausal
women, respectively. However, Potter et al (17) observed signifi-
cant reductions in apo B concentrations in moderately hypercho-
lesterolemic men who consumed ISP. In the current study, we
also found significant decreases at week 6 in all the groups that
consumed ISP. The different results found in these studies may be
explained by different populations, sample sizes, or both.

No change in HDL-cholesterol concentration was detected in
any of the ISP treatment groups. This finding agrees with many
other studies in men (5), which also did not detect significant
increases in HDL-cholesterol concentrations when animal pro-
tein was replaced with soy protein. However, in women, Baum
et al (18) reported a significant increase in HDL-cholesterol
concentrations with consumption of soy.

In a meta-analysis by Anderson et al (5), triacylglycerol con-
centrations decreased with the substitution of soy protein for ani-
mal protein. However, there were no significant alterations in the
concentrations of triacylglycerol in the studies by Potter et al
(17), Bakhit et al (14), and Baum et al (18), or in the present
study. The intra- and interindividual variation in triacylglycerol
concentrations is large, and thus a greater number of subjects may
be needed to find statistically significant changes in this lipid.

Lipoprotein(a) concentrations have been shown to be a valu-
able indicator of CHD risk (19). Although no dietary factors have
been found to influence the concentration of this lipid so far, stud-
ies with hormones showed that estrogens may decrease lipopro-
tein(a) concentrations by as much as 35% (20). In the present
study, we found no significant changes in lipoprotein(a) concen-
trations. These results agree with those of Crouse et al (15), who
also did not report significant changes when ISP was consumed.

An unexpected finding in this study was that the 40-g-ISP
group did not show a significant reduction in TC at week 6,
whereas all the other ISP groups did. Dietary noncompliance
for the 40-g-ISP group was unlikely because dietary intake patterns
of subjects were carefully monitored for 3 d/wk every 2 wk, and
those few subjects with poor compliance were eliminated from
the final statistical analysis. Blood total isoflavone concentra-
tions were also measured and no obvious changes occurred in
this group between weeks 3 and 6 (Figure 1). In fact, although
the 0.053-mmol/L reduction in TC when 40 g ISP was con-
sumed was not significant at week 6, the P value of 0.07 indi-
cated a nearly significant change. Thus, the results for the
40-g-ISP group are consistent with those of the groups con-
suming different amounts of ISP in indicating a decrease in TC
concentrations relative to the control. We also noted increases
in non-HDL-cholesterol, TC, and apo B concentrations at week 3,
and these effects were maintained at week 6. Apparently, with lower amounts of soy protein,
a longer duration of dietary treatment was needed to achieve the
metabolic effects that resulted in altered plasma cholesterol. The
reductions in non-HDL-cholesterol, TC, and apo B concentra-
tions were observed in addition to the reductions achieved with a
National Cholesterol Education Program Step I diet. These reduc-
tions were not accompanied by significant changes in HDL-chole-
sterol, apo A-I, or triacylglycerol concentrations.

We thank Julie Hafermann and William Deverell for their technical assistance.

REFERENCES

1. Summary of the second report of the National Cholesterol Education
Program (NCEP) Expert Panel on Detection, Evaluation, and Treat-
2. West of Scotland Coronary Prevention Study: identification of high-
risk groups and comparison with other cardiovascular intervention


8. Taddei-Peters WC, Butman BT, Jone GR, Venetta TM, Macomber PF, Ranson JH. Quantification of lipoprotein(a) particles containing various apolipoprotein(a) isoforms by a monoclonal anti-apo(a) capture antibody and a polyclonal anti-apolipoprotein B detection antibody sandwich enzyme immunoassay. Clin Chem 1993;39:1382–9.


