Effects of psyllium hydrophilic mucilloid on LDL-cholesterol and bile acid synthesis in hypercholesterolemic men

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Abstract The goal of the current study was to determine the mechanism of the hypocholesterolemic effect of psyllium using a randomized, double-blind, crossover design. Twenty males (age 44 ± 4 yr, weight 79 ± 10 kg) with moderate hypercholesterolemia (total 265 ± 17 mg/dl, low density lipoprotein (LDL) 184 ± 15 mg/dl) were studied at baseline (B) and after randomization to receive a 40-day course of 15 g/day of either psyllium (Ps) or placebo (Pl) (cellulose). After a washout period (11 ± 2 days), subjects were crossed over to the other fiber treatment for an additional 40 days and restudied. Intestinal cholesterol absorption, cholesterol synthesis in isolated peripheral blood mononuclear cells, bile acid kinetics, gallbladder motility, and intestinal transit were measured at each study period. Psyllium lowered LDL cholesterol (x:184 (B), 169 (Ps), and 179 (Pl) mg/dl; Ps vs. B,Pl: $P < 0.004$, $P < 0.02$), decreased relative cholesterol absorption ($\bar{x}$:51 (B), 45 (Ps), and 49 (Pl) %; Ps vs. B,Pl: $P < 0.03$, $P < 0.03$), did not alter absolute cholesterol absorption, and increased the fractional turnover of both chenodeoxycholic acid ($\bar{x}$:0.176 (B), 0.203 (Ps), and 0.170 (Pl) day^{-1}; Ps vs. B,Pl: $P < 0.0001$, $P < 0.01$) and cholic acid ($\bar{x}$: 0.303 (B), 0.411 (Ps), and 0.301 (Pl) d^{-1}; Ps vs. B, Pl: $P < 0.006$, $P < 0.002$). Bile acid synthesis increased in subjects whose LDL cholesterol was lowered by more than 10% (Ps vs. B: 1304 ± 489 vs 992 ± 307 μmol/day, $P < 0.006$; Ps vs. PI: 1304 ± 489 vs. 914 ± 321 μmol/day, $P < 0.0002$). We conclude that psyllium lowers LDL cholesterol primarily via stimulation of bile acid synthesis.—**Everson, G. T., B. P. Daggy, C. McKinley, and J. A. Story.** Effects of psyllium hydrophilic mucilloid on LDL-cholesterol and bile acid synthesis in hypercholesterolemic men. *J. Lipid Res.* 1992. **33:** 1183–1192.

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Certain dietary fibers lower total serum cholesterol and LDL cholesterol in both laboratory animals and humans (1). Human trials have demonstrated that psyllium consistently reduces total cholesterol from 5 to 20% (2–11) and LDL cholesterol by 8 to 20% (10–13). Despite the known cholesterol-lowering effect of this source of soluble dietary fiber and the potential impact on atherosclerosis, the mechanisms by which fiber reduces serum cholesterol are incompletely defined. Some studies suggest that fiber increases the fecal excretion of bile acids (14, 15) in a manner analogous to that of bile acid-binding resins, such as cholestyramine (16, 17). However, other studies demonstrate no change or a reduction in bile acid excretion, and still others suggest that dietary fiber may lower LDL cholesterol by inhibiting cholesterol synthesis via absorption of products of fermentation (18) or by alteration of the composition of the bile acid pool (19).

The goal of our study was to characterize the mechanism of the hypocholesterolemic effect of psyllium in free-living human subjects using a double-blind, placebo-controlled, crossover study design. We investigated the effects of psyllium on the intestinal absorption of cholesterol, cholesterol synthesis by peripheral blood mononuclear cells, and the kinetics of bile acids. Mechanisms of changes in the enterohepatic circulation of bile acids were assessed by measuring gallbladder motility and intestinal transit. Two groups of subjects were defined: those who lowered LDL cholesterol by > 10% in response to psyllium (responders) and those who did not (nonresponders). Examination of the differences in cholesterol and bile acid metabolism between responders and nonresponders provided further insight into the mechanism of lowering of LDL cholesterol by psyllium.

METHODS

Subjects

The study protocol was reviewed and approved by the Human Research Committee of the University of

Abbreviations: LDL, low density lipoprotein; CDCA, chenodeoxycholic acid; CA, cholic acid; LDL-C, LDL-cholesterol.

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Colorado School of Medicine. Male subjects were recruited via notices placed in newspapers and local publications throughout the University of Colorado Health Sciences Center. All subjects were paid volunteers who gave informed consent to participate in the study and had the following characteristics: age 30 to 50 years inclusive, body weight of 100 ± 30% of ideal (20) according to the 1983 Metropolitan Height and Weight Tables (Metropolitan Life Insurance Company, Health and Safety Education Division), and hypercholesterolemia not related to known disease or medications. All subjects had to exhibit the following fasting lipid profiles[2] during initial screening prior to entry into the trial: total serum cholesterol > 240 mg/dl; LDL cholesterol > 75th and < 95th percentile (21) adjusted for age and sex; and triglycerides < 300 mg/dl. LDL cholesterol (mg/dl) was calculated (22) from the equation:

$$\text{LDL cholesterol} = \text{total cholesterol} - \text{HDL cholesterol} - \left[\frac{\text{triglyceride level}}{5}\right]$$

Exclusion criteria included: inability to give informed consent or follow instructions; use of any lipid-lowering medication, antibiotic, barbiturate, or drug known to effect gastric or biliary motility within 3 months of the start of the study; history of prior cholecystectomy, biliary tract or intestinal surgery, concurrent gallbladder or intestinal disease, symptomatic coronary artery disease, allergic reaction to psyllium, or a current average daily fiber intake > 30 g/day.

Study design

Once enrolled in the trial, subjects received an identification number and were randomly assigned to a treatment sequence by subject number via computerized randomization. Each pair of subjects entering the study was randomly assigned so that one subject per pair was on one sequence and the other on the reverse sequence. The study fiber (psyllium hydrophilic mucilloid) and placebo fiber (cellulose) were provided by The Procter & Gamble Company (Cincinnati, OH). Psyllium was supplied in the form of orange-flavored Metamucil® powder with each dose containing 5.1 g of psyllium. Placebo consisted of the excipients of orange-flavored Metamucil® powder and 5.1 g of microcrystalline cellulose (Avicel PH-101, FMC Corp., Newark, DE).

The baseline evaluation included a medical history, physical examination, and blood chemistries. Subjects were instructed by a trained nutritionist on maintaining dietary records, and body weight and other demographic data were recorded. All underwent screening ultrasonography to exclude cholelithiasis and were then studied according to the following schedule of procedures. Blood was obtained at baseline for the measurement of bile acid isotope ratios prior to the administration of isotopes. On day 1, deuterium-labeled bile acids were administered orally and blood was obtained for measurement of plasma lipids. On day 2, blood was drawn for measurement of bile acid isotope ratios and for isolation of monocytes for determination of cholesterol synthesis. In addition, ^{14}C- and tritium-labeled cholesterols were administered orally and intravenously, respectively, for determination of intestinal absorption of cholesterol. Gallbladder volume after an overnight fast and contraction in response to a standard liquid meal was measured by realtime ultrasonography. Intestinal transit was measured by lactulose-breath hydrogen technique. On day 3, blood was drawn for measurement of bile acid isotopes. On day 4, blood was drawn for measurement of bile acid and cholesterol isotopes. On day 5, blood was drawn for measurement of bile acid and cholesterol isotopes and plasma lipids. After completion of baseline studies, subjects were randomized to receive 15.3 g/day of either psyllium or placebo for 40 days. No supplemental fiber was taken during a washout period of 11 ± 2 days. After washout, subjects were crossed-over to receive the other fiber for another 40 days. Study procedures were repeated during the last week of each period of fiber treatment.

PROCEDURES

Diet records

The Clinical Research Center dietitian assessed each subject's current diet and instructed subjects to maintain their weight and usual caloric, cholesterol, fat, and fiber intake throughout the period of the study (23, 24). Dietary and weight records were periodically reviewed by the dietitian who used a PC-based program (Food Processor II, ESHA Research, Salem, OR) to calculate daily cholesterol intake, fat intake, fiber intake, and other lipid parameters. Final calculations of nutrient intake were performed using the Minnesota Nutrition Data System (NDS) software (Food Database version 2.2; Nutrient Database version 4a), developed by the Nutrition Coordinating Center, University of Minnesota (Minneapolis, MN). Compliance with test medications was determined by packet counts of returned medication.

Bowel habit

Bowel habit was defined by frequency of bowel movements (number per day) and estimated volume (cups) passed per day. Subjects were instructed in keeping bowel habit diaries for 1 week at baseline and at the end of each

[2]Serum lipid concentrations were all determined by the same lipid laboratory (Central Laboratory, University Hospital, Denver, CO), which is certified by the College of American Pathologists and the Center for Disease Control.

fiber treatment period. At the end of each period, the diaries were reviewed and summarized without knowledge of treatment fiber.

Gallbladder motility

A validated realtime ultrasonographic technique was used (25). Fasting volume (FV) was defined as the volume retained in the gallbladder after an overnight fast, and residual volume (RV) was the minimal volume achieved in response to the meal. The rate constant of emptying (k) was calculated by ln/linear regression of volume versus time, and the percent emptied (%E) from [1 − (RV/FV)] × 100%.

Intestinal transit

Transit was measured by breath hydrogen analysis after oral administration of the nonabsorbable carbohydrate, lactulose (26–28). Breath samples for measurement of hydrogen concentration were collected at baseline and every 15 min for 5 h after ingestion of lactulose (15 g). Hydrogen concentration was measured by gas chromatography using a molecular sieve column and a thermal conductivity detector (Quintron model CM2 microanalyzer, Quintron Instrument Company, Milwaukee, WI) and calculated by reference to standards. Initial transit time (TT_I) was defined as the time to the initial sustained rise in breath hydrogen, and peak transit time (TT_P) as the time to the peak in breath hydrogen concentration.

Bile acid kinetics

Bile acid kinetics were measured using stable isotopes, serum sampling, and isotope dilution–mass spectrometry (29). Fifty milligrams of stable isotopes of chenodeoxycholate ([11,12-^{2}H]CDCA) and cholate ([2,2,4,4-^{2}H]CA) were taken orally; plasma was collected prior to isotope administration and each day for 4 days after administration. The isotope ratios of bile acids isolated from serum collected prior to administration of isotopes (natural abundance) equalled the isotope ratios of purified standards (Sera vs. Standards, N_{pairs} = 60: CDCA *m/z* 372/370, 0.0495 ± 0.0017 vs. 0.0494 ± 0.0012; CA *m/z* 462/458, 0.0033 ± 0.0010 vs. 0.0036 ± 0.0004). The disappearance of labeled bile acids from the bile acid pool obeyed a single monoexponential function, and correlation coefficients for the plots of the logarithm of molar enrichment versus time after isotope administration were 0.98 ± 0.03 for CDCA (n = 59) and 0.97 ± 0.03 for CA (n = 57).

Cholesterol absorption and synthesis

Cholesterol absorption was measured by the isotope ratio method (30) using 2 μCi each of [4-^{14}C]cholesterol (57.7 mCi/mmol) orally and [1,2-^{3}H]cholesterol (60.0 mCi/mmol) intravenously (obtained from New England Nuclear Corporation, Boston, MA). The mean coefficient of variation in duplicate measurements of cholesterol absorption was 2.6%.

Cholesterol synthesis was measured in peripheral blood mononuclear cells, as described by McNamara, Davidson, and Fernandez (31) and used by us (32, 33), with [2-^{14}C]acetate (51 mCi/mmol) (New England Nuclear) as substrate. Mononuclear cells (monocytes and lymphocytes) were separated by centrifugation of 15 ml of blood in medium containing sodium diatrizoate and polyester gel (LeucoPREP, Nygaard and Company, Oslo, Norway). The mean coefficient of variation in duplicate measurements of cholesterol synthesis was 5.4%.

Statistical analysis

Data were presented as mean and standard deviation of the mean in all tables, results, and discussion. The study was analyzed as a two-treatment two-period crossover design (34, 35). Treatment comparisons (psyllium vs. placebo) were made by paired *t*-test when the carry-over effect was nonsignificant. However, if the carry-over effect was significant as measured by *t*-test (only found to be so for LDL cholesterol), then the treatment effect was analyzed by unpaired *t*-test using responses in the first time period only. Pearson and Spearman's correlations were computed between LDL cholesterol and all mechanistic variables of interest including bile acid kinetics, intestinal cholesterol absorption, cholesterol synthesis by peripheral blood mononuclear cells, gallbladder motility, and small intestine transit. All correlations were tested for significance using a two-sided test with a = 0.05.

RESULTS

Subjects and compliance

Two hundred fifty-six subjects responded to our recruitment notices; of these, 151 were rejected after a phone interview primarily due to intercurrent disease or concurrent use of medications. One hundred five subjects were formally screened, and the final 20 were selected from this group. The main reason for rejecting the remaining 85 was failure to document sustained hypercholesterolemia. Subjects selected for study had ages from 36 to 50 years (mean ± SD: 44 ± 4 years), weight from 65 to 100 kg (mean ± SD: 79 ± 10 kg), and body surface area from 1.7 to 2.2 m² (mean ± SD: 1.94 ± .13 m²). Eighteen of 20 subjects were Caucasian, the remaining two were Hispanic. All were highly motivated and compliant with treatment: 97 ± 5% of administered psyllium and 96 ± 6% of administered placebo were taken by study subjects.

Weight, diet, and bowel habit

The effects of fiber treatment on total body weight, bowel habit, and dietary habit are shown in **Table 1**.

TABLE 1. Effects of fiber treatment on body weight, dietary intake, and bowel habit

	Basal	Psyllium	Placebo	P^a B vs. Ps	B vs. Pl	Ps vs. Pl
Wt (kg)	78.6 ± 9.5	79.0 ± 9.5	79.0 ± 9.5	NS	NS	NS
Dietary intake						
Energy (calories/day)	1997 ± 431	2135 ± 605	2141 ± 607	NS	NS	NS
% Calories from fat	37.4 ± 8.7	36.6 ± 8.5	38.7 ± 6.1	NS	NS	NS
Monounsaturated	14.4 ± 3.7	14.1 ± 3.7	14.7 ± 2.3	NS	NS	NS
Polyunsaturated	7.9 ± 2.6	7.5 ± 2.2	7.9 ± 2.4	NS	NS	NS
Saturated	12.3 ± 3.5	12.0 ± 3.5	13.2 ± 3.2	NS	NS	NS
% Calories from carbo	43.1 ± 9.4	43.6 ± 8.9	42.3 ± 5.1	NS	NS	NS
% Calories from protein	16.1 ± 3.0	16.4 ± 3.0	15.6 ± 3.7	NS	NS	NS
% Calories from alcohol	4.6 ± 5.9	4.2 ± 5.1	4.1 ± 5.4	NS	NS	NS
Cholesterol (mg/day)	242 ± 140	296 ± 141	274 ± 105	NS	NS	NS
Dietary fiber (g/day)	16.4 ± 5.4	16.7 ± 6.7	16.1 ± 5.5	NS	NS	NS
Insoluble	10.7 ± 4.0	10.9 ± 5.1	10.6 ± 3.6	NS	NS	NS
Soluble	5.6 ± 2.1	5.7 ± 1.9	5.5 ± 2.3	NS	NS	NS
Bowel habit						
Freq (#/day)	1.48 ± 0.69	1.84 ± 0.91	1.51 ± 0.51	< 0.04	NS	<0.01
Volume (cups/day)	3.23 ± 1.75	4.32 ± 2.48	3.38 ± 1.41	<0.03	NS	<0.01

Abbreviations: B, basal; Ps, psyllium treatment; Pl, placebo treatment; NS, not significant; carbo, carbohydrate.
[a] *P* values were determined by paired *t*-tests. Each subject's values on fiber treatment were compared to baseline values.

There was no change in body weight or dietary habit during treatment with either psyllium or placebo. Psyllium increased both the frequency and volume of the bowel habit compared to both baseline ($P < 0.04$, $P < 0.03$, respectively) and placebo ($P < 0.01$, $P < 0.01$, respectively).

Serum lipid levels (Table 2)

Psyllium reduced LDL cholesterol by 8% versus baseline ($P < 0.004$) and by 6% versus placebo ($P < 0.02$). A significant ($P = 0.047$) carryover effect was detected for LDL-C, suggesting that the treatments given in period 1 influenced the LDL-C response in period 2. This is shown graphically in **Fig. 1**. Psyllium lowered LDL-C, compared to placebo, in those subjects randomized to receive psyllium as the initial fiber (sequence 1), but not in those randomized to receive placebo first (sequence 2). The apparent inability of psyllium to lower LDL cholesterol in those randomized to treatment sequence 2 was largely influenced by one subject (---, sequence 2, Fig. 1) whose LDL cholesterol increased progressively throughout the period of study. However, the mean reduction in LDL cholesterol during the use of psyllium in the other nine sequence 2 subjects (7%) was less than the mean reduction in the ten subjects randomized to treatment sequence 1 (13%). Given the relatively small number of subjects and the observed general lack of effect of the placebo fiber on lipid metabolism, the significant carryover could be due simply to the chance allocation of less responsive subjects into treatment sequence 2. There may also be a small

TABLE 2. Effects of fiber treatment on plasma lipids, intestinal cholesterol absorption, and cholesterol synthesis by peripheral blood mononuclear cells

	Basal	Psyllium	Placebo	P B vs. Ps	B vs. Pl	Ps vs. Pl
Plasma lipids (mg/dl)						
Total CH	265 ± 17	251 ± 32	260 ± 24	<0.03	NS	NS (0.18)
HDL	48 ± 15	49 ± 15	48 ± 14	NS	NS	NS
LDL	184 ± 15	169 ± 26	179 ± 19	<0.004	NS	<0.02[a]
Triglyceride	166 ± 57	175 ± 81	166 ± 61	NS	NS	NS
Cholesterol absorption and synthesis						
Absorption (%)	51 ± 15	45 ± 10	49 ± 12	<0.01	NS	<0.03
Synthesis (pmol/10^7 cell/h)	46 ± 16	48 ± 11	48 ± 11	NS	NS	NS

Abbreviations: Total CH, total serum cholesterol; HDL, high density lipoprotein; LDL, low density lipoprotein; B, basal; Ps, psyllium; Pl, placebo.
[a] This *P* value is not corrected for the observed carryover effect of the first administered fiber on the effects of the second fiber. Allowing for carryover, the difference between Ps and Pl is even more significant ($P < 0.003$) (see text for discussion).

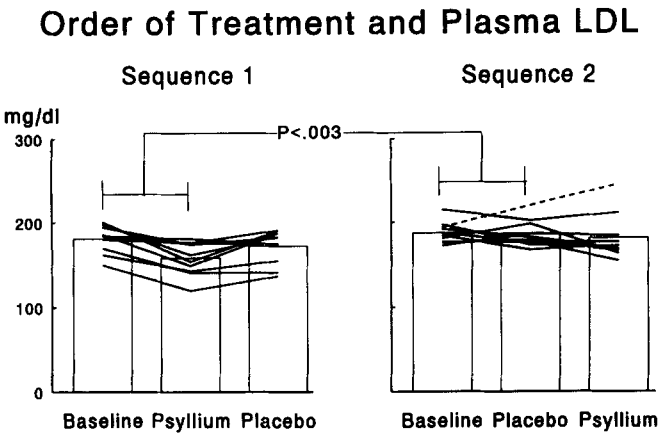

Fig. 1. In the first treatment period, psyllium (sequence 1) significantly lowered LDL from baseline compared to placebo (sequence 2) ($P < 0.003$). This was not true during the second treatment period due to a dramatic rise in LDL in a single subject (---) and an apparent carryover effect of placebo on the response to psyllium.

genuine carryover effect of psyllium, as the subjects randomized to psyllium first did not quite return to their baseline at the end of placebo treatment. If one accepts the carryover effect as real and therefore analyzes only the LDL-C results from period 1 by unpaired *t*-test, psyllium lowered LDL cholesterol by 12% versus placebo ($P < 0.003$).

Psyllium significantly reduced total cholesterol relative to baseline by 6% ($P < 0.03$) but not relative to placebo (3.5% reduction, $P = 0.18$). Neither test fiber altered levels of triglyceride or HDL cholesterol.

On further examination of LDL cholesterol levels, it was apparent that two groups of subjects were identified, those who lowered LDL cholesterol by > 10% in response to psyllium (responders) and those who did not (nonresponders) (**Fig. 2**). The eleven responders lowered LDL cholesterol by 16 ± 8% (baseline vs. psyllium: 183 ± 17 to 154 ± 16 mg/dl). In contrast, the nine nonresponders had no change in LDL cholesterol (baseline vs. psyllium: 186 ± 13 to 187 ± 25 mg/dl).

Gallbladder motility (Table 3)

Psyllium reduced the postprandial residual volume ($P < 0.05$) and increased the percent of gallbladder volume emptied ($P < 0.05$) but did not alter emptying rate or fasting volume of the gallbladder. Placebo fiber did not alter any parameter of gallbladder motility.

Transit time (Table 3)

Transit time could be determined from breath hydrogen curves in all subjects during baseline studies but could not be measured in one subject during use of psyllium and four subjects during use of placebo fiber because no definite peak in breath hydrogen occurred. Each of these five subjects exhibited hydrogen peaks basally. Psyllium did not alter intestinal transit. In contrast, placebo shortened initial transit time $P < 0.03$) without changing other parameters of the breath hydrogen curve. The significance of the latter finding is questionable because four studies were not analyzable.

Cholesterol absorption and synthesis (Table 2)

Psyllium reduced intestinal cholesterol absorption (%) versus baseline and placebo, but this effect was limited to nonresponders (responders: 44 ± 6 vs. 43 ± 6%, P = NS; nonresponders: 59 ± 19 vs. 48 ± 14%, $P < 0.002$) (**Fig. 3**). However, psyllium did not alter absolute intestinal cholesterol absorption because dietary intake of cholesterol was slightly greater during psyllium treatment (Table 2, P = NS). Cholesterol synthesis by peripheral blood monocytes was not altered by either placebo or psyllium. The response of cholesterol synthesis to therapy with either placebo or psyllium was similar in responders and nonresponders.

Bile acid kinetics (Table 4)

Psyllium exerted very significant and consistent effects on the turnover rates of bile acids. Fractional turnover rates of both chenodeoxycholate and cholate were significantly increased by psyllium versus both baseline and placebo. The pool size of CDCA increased, but CA and total pool did not change. The synthesis of chenodeoxycholate, cholate, and total bile acids significantly increased. These responses were specific to psyllium since placebo exerted no effect on any parameter of bile acid metabolism.

Alterations in bile acid kinetics induced by psyllium were compared between responders and nonresponders (**Table 5**). The fractional turnover rate of bile acids increased during psyllium therapy in both responders and

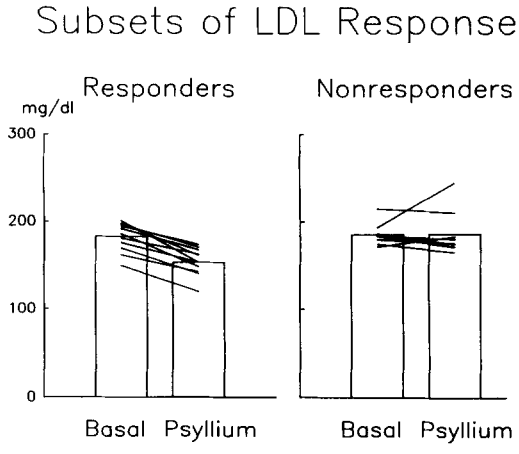

Fig. 2. The group of subjects was subdivided into those who lowered their LDL cholesterol by greater than 10% during use of psyllium (responders) and those who did not (nonresponders). The two groups had similar basal levels of LDL cholesterol.

TABLE 3. Effects of fiber treatment on gallbladder motility and small intestinal transit

	Basal	Psyllium	Placebo	P		
				B vs. Ps	B vs. Pl	Ps vs. Pl
Gallbladder motility						
FV (ml)	23 ± 9	22 ± 8	22 ± 6	NS	NS	NS
RV (ml)	5 ± 3	4 ± 2	5 ± 3	<0.05	NS	0.06
%E (%)	75 ± 13	80 ± 8	78 ± 11	<0.05	NS	NS
k (min^{-1})	0.016 ± 0.007	0.019 ± 0.010	0.020 ± 0.013	NS	NS	NS
Small intestinal transit						
TT_I (min)	102 ± 33	92 ± 58	79 ± 33	NS	<0.03	NS
H_2F (ppm)	24 ± 24	21 ± 20	17 ± 12	NS	NS	NS
H^2P (ppm)	126 ± 68	105 ± 67	130 ± 62	NS	NS	NS
TT_P (min)	181 ± 58	185 ± 45	190 ± 66	NS	NS	NS

Abbreviations: FV, fasting volume; RV, residual volume; %E, percent of FV emptied; k, rate constant of emptying; TT_I, initial transit time; H_2F, concentration of breath hydrogen prior to administration of lactulose; H_2P, concentration of breath hydrogen at the peak of the hydrogen curve after administration of lactulose; TT_P, time to the peak in breath hydrogen curve.

nonresponders, but total pool size did not significantly change in either group. The only feature that distinguished responders from nonresponders was the finding that the synthesis of CDCA, CA, and total bile acid was significantly stimulated by psyllium only in responders (**Fig. 4**). In addition, the decrease in LDL-C during use of psyllium correlated directly with the increase in total bile acid synthesis ($r = 0.46$, $P < 0.01$).

Baseline differences between responders and nonresponders

Responders had significantly lower basal rates of intestinal absorption of cholesterol (44 ± 6 vs. 59 ± 19%, $P < 0.02$); produced less hydrogen after administration of lactulose (peak [H_2]: 155 ± 65 vs. 91 ± 57 ppm, $P < 0.04$); tended to have higher basal fractional turnover rates of both CDCA (0.183 ± 0.057 vs. 0.166 ± 0.043 day^{-1}, P = NS) and CA (0.337 ± 0.120 vs. 0.261 ± 0.083 day^{-1} P = NS); and had smaller CA pools (2049 ± 490 vs. 2964 ± 1147 μmol, $P < 0.03$).

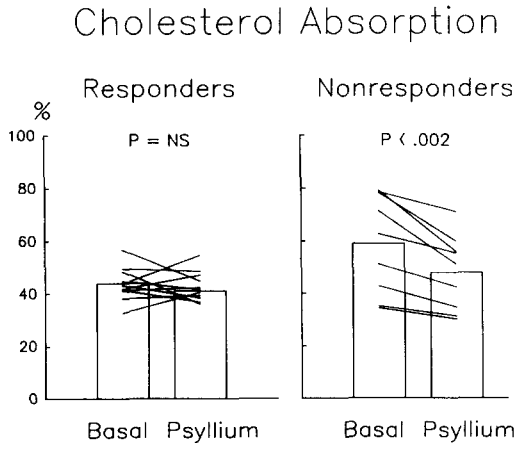

Fig. 3. The basal relative absorption (%) of cholesterol of responders was lower compared to nonresponders and did not decrease with psyllium. Nonresponders had higher basal rates of cholesterol absorption, and psyllium significantly decreased absorption.

DISCUSSION

We examined the effects of psyllium on two inputs to the hepatic cholesterol pool: cholesterol synthesis (peripheral blood mononuclear cells) and intestinal absorption of dietary cholesterol. Although rates of cholesterol synthesis by mononuclear cells are lower than those of hepatocytes (36), the relative change in synthesis after in vivo treatment with cholesterol, clofibrate, cholestyramine, and mevinolin is similar (31, 37–40). For this reason we and others have considered the monocyte an acceptable model for studying the regulation of cholesterol synthesis in intact human subjects. However, the method may lack sufficient sensitivity to detect small changes in cholesterol synthesis. For example, Miettinen and Tarpila (41) used a higher dose of psyllium and monitored serum levels of biosynthetic precursors of cholesterol to detect an increase in cholesterol synthesis on psyllium. We quantitated a second input to the hepatic cholesterol pool, dietary cholesterol, by determining cholesterol intake from dietary records (23, 24) and the percent absorption by isotope ratio technique (30). The accuracy of these methods is well validated.

The only measured output from the hepatic cholesterol pool was bile acid synthesis. The serum method for measuring bile acid kinetics that was used in this study has been validated by our laboratory (29). With this technique, isotope enrichments of bile acids in serum are identical to those in bile. Fiber treatment was withheld for 12 h before and 12 h after the administration of labeled bile acids to avoid entrapment and fecal elimination of labeled compounds prior to their complete mixing with the

TABLE 4. Effects of fiber treatment on bile acid kinetics

	Basal	Psyllium	Placebo	P		
				B vs. B	B vs. Pl	Ps vs. Pl
CDCA kinetics						
FTR (d^{-1})	0.176 ± 0.051	0.203 ± 0.053	0.170 ± 0.054	<0.0001	NS	<0.01
Pool (μmol)	1798 ± 594	1998 ± 703	1782 ± 677	<0.04	NS	<0.04
Syn (μmol/day)	298 ± 92	390 ± 137	286 ± 106	<0.0001	NS	<0.0002
CA kinetics						
FTR (d^{-1})	0.303 ± 0.110	0.411 ± 0.149	0.301 ± 0.111	<0.006	NS	< 0.002
Pool (μmol)	2461 ± 948	2425 ± 1300	2153 ± 593	NS	NS	NS
Syn (μmol/day)	694 ± 252	914 ± 445	630 ± 249	<0.04	NS	< 0.002
DCA pool (μmol)	1358 ± 633	1548 ± 927	1243 ± 604	NS	NS	NS
Total pool (μmol)	5614 ± 1483	5852 ± 1632	5108 ± 1205	NS	NS	NS
Total Syn (μmol/day)	992 ± 307	1304 ± 489	914 ± 321	<0.006	NS	< 0.0001
[CA + DCA:CDCA]	2.26 ± 0.80	2.25 ± 0.96	2.13 ± 0.65	NS	NS	NS

Abbreviations: CDCA, chenodeoxycholate; CA, cholate; DCA, deoxycholate; FTR, fractional turnover rate; Syn, synthesis.

bile acid pool. Although we did not measure the other main output from the hepatic cholesterol pool, biliary cholesterol secretion, recent evidence in the hamster suggests that psyllium may increase the output of biliary cholesterol (42).

The lowering of LDL cholesterol by psyllium observed in this study is consistent with previous studies (10–13). However, previous studies have not examined the mechanism of this hypocholesterolemic effect in man. Some, but not all, dietary fibers that exert a hypocholesterolemic effect increase the fecal elimination of bile acids (14, 15). Our study has demonstrated that psyllium increases the turnover of both primary bile acids. Although direct binding of bile acid to psyllium does not occur (42), the increased turnover may be due to entrapment of bile acids within hydrated psyllium and reduction in intestinal absorption of bile acid. Given a constant rate of enterohepatic cycling (9 cycles/day) and the pool sizes and synthetic rates (= fecal loss in the steady state) observed in our study, a less than 1% decrease in absorption of bile acid could account for the observed increase in turnover during use of psyllium. If absorption of bile acids was not affected by psyllium then a 30–40% increase in number of enterohepatic cycles would be required to cause the observed increase in bile acid turnover. Since gallbladder emptying only increased by 5% and intestinal transit was not altered, we contend that the latter explanation is unlikely. Thus, it is most likely that the increase in turnover is due to impairment of intestinal absorption of bile acids by psyllium, consistent with other studies showing increased fecal bile acid excretion (14–17).

Fiber may differentially affect the turnover rates of the various bile acids. For example, several dietary fibers increase the fecal elimination of CDCA but decrease that of CA (19). By this action, dietary fiber may alter the composition of the bile acid pool, an effect that could influence the intestinal absorption of cholesterol (43) or hepatic synthesis of cholesterol and bile acid (44). We found that psyllium did not significantly alter the composition of the bile acid pool; [CA+DCA]:CDCA did not change. Thus, the

TABLE 5. Bile acid kinetics of nonresponders and responders

	Nonresponders (n = 9)			Responders (n = 11)		
	Basal	Psyllium	P	Basal	Psyllium	P
CDCA kinetics						
FTR (d^{-1})	0.166 ± 0.043	0.190 ± 0.056	<0.003	0.183 ± 0.057	0.213 ± 0.052	<0.002
Pool (μmol)	1663 ± 361	1779 ± 479	NS	1908 ± 733	2177 ± 823	NS
Syn (μmol/day)	268 ± 65	335 ± 143	NS	323 ± 107	434 ± 120	<0.0005
CA kinetics						
FTR (d^{-1})	0.261 ± 0.083	0.386 ± 0.199	<0.04	0.337 ± 0.120	0.433 ± 0.0950	= 0.10
Pool (μmol)	2964 ± 1147	2401 ± 1013	NS	2049 ± 490	2446 ± 1571	NS
Syn (μmol/day)	729 ± 302	805 ± 330	NS	665 ± 214	1012 ± 527	<0.03
DCA pool (μmol)	1268 ± 766	1661 ± 1300	NS	1428 ± 529	1455 ± 510	NS
Total pool (μmol)	5894 ± 1864	5841 ± 1477	NS	5386 ± 1128	5856 ± 1946	NS
Total Syn (μmol/day)	997 ± 357	1140 ± 384	NS	987 ± 277	1354 ± 608	<0.03
[CA + DCA:CDCA]	2.56 ± 0.91	2.43 ± 0.93	NS	2.01 ± 0.63	1.95 ± 1.05	NS

Abbreviations: See Table 4.

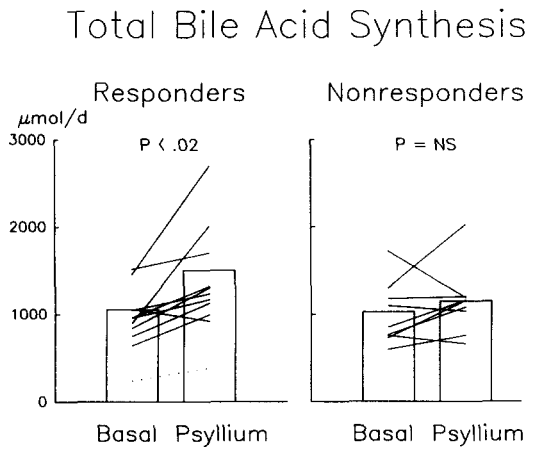

Fig. 4. Total bile acid synthesis of responders and nonresponders was similar under basal conditions. Responders increased synthesis on psyllium, but nonresponders did not. The dotted line (····) is the change in CDCA synthesis in the one subject in whom cholate synthesis could not be measured. CDCA synthesis in this subject obeyed the same trend as total synthesis in the group of responders as a whole.

effects of psyllium on bile acid turnover, bile acid synthesis, and cholesterol absorption measured in our study were not mediated by changes in the composition of the bile acid pool.

It has been suggested that the hypocholesterolemic effect of dietary fiber may be due to impairment of intestinal absorption of fats and sterols (45–47), although no previous study has directly measured intestinal absorption of cholesterol. In our study, we found that psyllium impaired intestinal cholesterol absorption, but this effect was limited to subjects with high baseline absorption of cholesterol who did not lower LDL cholesterol in response to psyllium (nonresponders). Thus, inhibition of cholesterol absorption did not correlate with the hypocholesterolemic effect of psyllium.

One suggested mechanism of the LDL-lowering effect of soluble fiber is inhibition of cholesterol synthesis through production of short-chain fatty acids (acetate, butyrate, propionate) from fermentation (18). Some studies have shown that administration of propionate to both intact rat (48, 49) and isolated rat hepatocytes (50) significantly inhibits cholesterol synthesis. In contrast, others have demonstrated that physiologic concentrations of propionate do not alter cholesterol synthesis (51). In our study, psyllium did not alter cholesterol synthesis by peripheral blood mononuclear cells. Thus, it seems unlikely that the hypocholesterolemic effect of psyllium measured in our study is related to inhibition of cholesterol synthesis by products of fermentation of psyllium.

Although individual variation in the response to dietary cholesterol is well described (52, 53), this is the only study that has characterized subjects as responders or nonresponders, based on the ability of psyllium to lower LDL cholesterol. Kies (54) summarized the results from 28 studies including 285 total subjects in terms of the cholesterol-lowering effect of dietary fiber. She concluded that subjects who lowered plasma cholesterol in response to fiber (responders) were those in whom fiber increased fecal wet weight. Of the fibers tested, psyllium was most effective in increasing fecal weight and water-holding ability, slowing fecal transit, increasing fecal fat loss, and producing the greatest reduction in serum cholesterol levels. Entrapment of bile acid by psyllium would deliver bile acid to the intestine where it could stimulate intestinal secretion of water and thereby increase fecal weight and water. Indeed, in our study, psyllium increased the frequency and volume of bowel movements and increased the fractional turnover of both CDCA and CA. However, these effects were similar in both responders and nonresponders. Thus, the increase in fecal weight and bile acid turnover alone are not sufficient to lower LDL cholesterol.

Psyllium increases the fractional turnover of bile acids, probably by increasing their fecal elimination. This effect would tend to reduce the hepatic flux of bile acids. If, as suggested by certain (55–63), but not all (64, 65), studies, bile acids inhibit their own synthesis, reducing hepatic bile acid flux would tend to stimulate bile acid synthesis. Since turnover was increased in both responders and nonresponders, we would have anticipated that the hepatic flux of bile acid would have been reduced similarly and synthesis would have been stimulated in both groups. The finding that bile acid synthesis increased in only one group (responders) suggests that mechanisms other than increased elimination of bile acid are important in the regulation of hepatic bile acid synthesis. Our results suggest that responders and nonresponders may differ at the level of hepatic regulation of bile acid synthesis. The hepatic synthesis of bile acid is determined by both the availability of cholesterol as substrate and the intrinsic activity of the rate-limiting enzyme, cholesterol-7α-hydroxylase (66). Since cholesterol synthesis did not increase and LDL cholesterol decreased during use of psyllium, it is unlikely that the stimulation of bile acid synthesis in responders was related to expansion of the cholesterol substrate pool. Thus, we propose that the ability of psyllium to lower LDL cholesterol may be due to stimulation of cholesterol-7α-hydroxylase.

In conclusion, psyllium hydrophilic mucilloid lowered LDL cholesterol in 50% of males with moderate hypercholesterolemia. Although psyllium reduced cholesterol absorption and increased bile acid synthesis, the hypocholesterolemic effect was associated only with stimulation of bile acid synthesis. Use of psyllium to lower LDL-C may identify subgroups of patients (responders and nonresponders) who have identifiable differences in bile acid metabolism. Subsequent studies will be required to determine the reproducibility and significance of these observations.

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