Review

Efficacy and Safety of Plant Stanols and Sterols in the Management of Blood Cholesterol Levels

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Foods with plant stanol or sterol esters lower serum cholesterol levels. We summarize the deliberations of 32 experts on the efficacy and safety of sterols and stanols. A meta-analysis of 41 trials showed that intake of 2 g/d of stanols or sterols reduced low-density lipoprotein (LDL) by 10%; higher intakes added little. Efficacy is similar for sterols and stanols, but the food form may substantially affect LDL reduction. Effects are additive with diet or drug interventions: eating foods low in saturated fat and cholesterol and high in stanols or sterols can reduce LDL by 20%; adding sterols or stanols to statin medication is more effective than doubling the statin dose. A meta-analysis of 10 to 15 trials per vitamin showed that plasma levels of vitamins A and D are not affected by stanols or sterols. Alpha carotene, lycopene, and vitamin E levels remained stable relative to their carrier molecule, LDL. Beta carotene levels declined, but adverse health outcomes were not expected. Sterol-enriched foods increased plasma sterol levels, and workshop participants discussed whether this would increase risk, in view of the marked increase of atherosclerosis in patients with homozygous phytosterolemia. This risk is believed to be largely hypothetical, and any increase due to the small increase in plasma plant sterols may be more than offset by the decrease in plasma LDL. There are insufficient data to suggest that plant stanols or sterols either prevent or promote colon carcinogenesis. Safety of sterols and stanols is being monitored by follow-up of samples from the general population; however, the power of such studies to pick up infrequent increases in common diseases, if any exist, is limited. A trial with clinical outcomes probably would not answer remaining questions about infrequent adverse effects. Trials with surrogate end points such as intima-media thickness might corroborate the expected efficacy in reducing atherosclerosis. However, present evidence is sufficient to promote use of sterols and stanols for lowering LDL cholesterol levels in persons at increased risk for coronary heart disease.

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ABC = ATP (adenosine triphosphate)-binding cassette; ABCG5 = ABC subfamily G, member 5; CHD = coronary heart disease; LDL = low-density lipoprotein; NCEP = National Cholesterol Education Program

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Dietary therapy is the cornerstone of strategies to lower serum low-density lipoprotein (LDL) cholesterol levels and reduce the risk for coronary heart disease (CHD). Incorporating foods fortified with plant sterol and stanol esters into the daily diet can substantially enhance the cholesterol-lowering effect of diet, including in patients who are already taking statin drugs. Thus, the recent introduction of stanol- and sterol-enriched foods in many parts of the world[1-3] is an important development because CHD is the leading cause of morbidity and mortality worldwide. We summarize the deliberations of 32 experts on lipids, nutrition, and heart disease who met in Stresa, Italy, on March 7-9, 2001, under the auspices of the Nutrition Foundation of Italy to discuss the efficacy, safety, and future research required on plant sterols and stanols. (Note that throughout this article, quantities of sterol and stanol esters are expressed as the equivalent weights of free [ie, unesterified] sterols and stanols.)

BACKGROUND

Sterols have cellular functions in plants analogous to those of cholesterol in animals[4] (the structures of 2 major plant

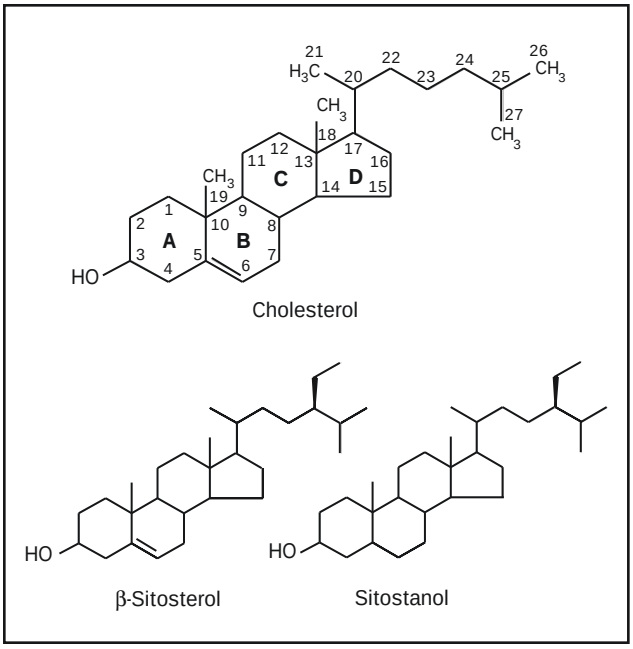

Figure 1. Structures of sterols. Cholesterol is the sterol of mammalian cells. β-Sitosterol is the most common sterol in plants; it differs from cholesterol by having an ethyl group attached at C-24. Hydrogenation of the 5,6 double bond of β-sitosterol converts it into sitostanol.[5] Campesterol and campestanol carry a methyl instead of ethyl group at C-24.

sterols and cholesterol are compared in Figure 1[5]). More than 40 plant sterols (or phytosterols) have been identified, but sitosterol, campesterol, and stigmasterol are the most abundant. Stanols are saturated sterols (they have no double bonds in the sterol ring structure). The major plant stanols are sitostanol and campestanol; they are much less abundant in nature than sterols. Whereas about 50% of cholesterol is absorbed in the intestinal tract, plant stanols and sterols are absorbed much less: absorption is about 10% to 15% for campesterol and campestanol, 4% to 7% for sitosterol,[6-8] and 1% for sitostanol.[9] Foods enriched with plant stanol or sterol esters lower serum cholesterol levels by reducing intestinal absorption of cholesterol.[1,10-12] Stanol (and presumably sterol) esters are hydrolyzed in the upper small gut.[13] Free stanols and sterols presumably displace cholesterol from mixed micelles and thereby reduce intestinal cholesterol absorption, but the exact mechanism is unknown. The effects of stanols and sterols probably involve the ATP (adenosine triphosphate)-binding cassette (ABC) subfamily G, member 5 (ABCG5) and ABCG8 transporter proteins. These membrane proteins selectively pump phytosterols from the enterocytes into the intestinal lumen, thus keeping their absorption low.[14,15]

The lowest amount of sterols and stanols tested in human trials is about 700 to 1000 mg/d.[16-18] Most recent studies used amounts of 1500 to 3300 mg/d of plant stanols or sterols in their esterified, fat-soluble forms. These amounts are much higher than the typical daily intake. In Western populations, the intake of sitosterol is about 150 to 350 mg/d and that of stanols is about 15 to 50 mg/d.[19-21] Vegetarians consume about 325 mg/d of sitosterol and about 60 mg/d of campesterol.[22] Thus, the usual daily intake of plant sterols and stanols ranges from 150 to 400 mg/d.

The cholesterol-lowering effect of extremely high intakes of plant sterols (10-20 g/d) was recognized in the 1950s through the 1970s[2,23,24] and reviewed by 2 pioneers in the field.[23] Cholesterol balance studies established that this effect was due to reduced cholesterol absorption.[25] However, widespread use of plant sterols as cholesterol-lowering agents was limited by the bulk and taste of large amounts of these agents and by the introduction of more effective cholesterol-lowering agents. Subsequent studies and demonstration of CHD risk reduction in cholesterol-lowering clinical trials renewed interest in plant sterols. An amount of 3 g/d was as effective as higher amounts in reducing cholesterol absorption if presented in appropriate physical form.[26] Mattson et al[27] suggested the esterification process to make plant sterols soluble in dietary fat. Esterification of sitostanol or sitosterol with fatty acids was found to enhance both their solubility in mayonnaise and margarines and their dispersion in the intestine, thereby promoting their efficacy.

Margarines containing either plant stanol or sterol esters are marketed in many countries. Other formulations, including yogurt, cream cheese spreads, and cereal bars, have been introduced in some countries, and cereals and fruit juice containing free (ie, unesterified) plant sterols and stanols are being test-marketed in the United States.[11,28]

Given the increased marketing of these products, a review of efficacy and safety is timely.

EFFICACY

What Is the Effect of Various Sterols and Stanols on Plasma Lipoproteins?

Data from published randomized trials that tested foods containing stanols or sterols are summarized in Table 1[5,11,16-18,28-63]; data from these trials updated a previous meta-analysis.[64] The trials were identified from MEDLINE and from review articles and by questioning the expert participants at our workshop. In total, 41 trials were identified, with 58 treatment arms or periods and 41 placebo arms or periods. The sterols and stanols were esterified except in 2 trials in which they were directly solubilized in fat-containing foods. The stanols and sterols were added to margarine (or to mayonnaise, olive oil, or butter in 7 trials), and

Table 1. **Randomized Double-Blind Trials Comparing Foods With and Without Added Plant Stanols or Sterols***

Reference	Country	No. of participants			Mean age (y)	Duration (wk)	Stanol or sterol	Dose (g/d)	Placebo-adjusted reduction in serum LDL cholesterol (mg/dL)
		Cross-over trials	Parallel trials						
			Treatment	Placebo					
Williams et al[29]	United States	19	NA	NA	4	4	Stanol	1.0	11
Tammi[30]	Finland	72	NA	NA	6	13	Stanol	1.6	8
Vissers et al[5]	The Netherlands	60	NA	NA	23	3	Sterol	2.1	8
Matvienko et al[31]	United States	NA	17	17	23	4	Sterol	2.7	17
Plat et al[32]	The Netherlands	39	NA	NA	31	4	Sterol	2.5	12
Plat & Mensink[33]	The Netherlands	NA	70	42	33	8	Stanol	4.0	14
Jones et al[34]	Canada	22	NA	NA	35	1.4	Sterol	1.6	12
Mensink et al[35]	The Netherlands	NA	30	30	36	4	Stanol	3.0	15
Hendriks et al[16]	The Netherlands	80	NA	NA	37	3.5	Sterol	0.8	8
							Sterol	1.6	10
							Sterol	3.2	12
Niinikoski et al[36]	Finland	NA	12	12	37	5	Stanol	3.0	19
Mussner et al[37]	Germany	63	NA	NA	42	3	Sterol	1.8	10
Hallikainen & Uusitupa[38]	Finland	NA	38	17	43	8	Stanol	2.3	18
Sierksma et al[39]	The Netherlands	76	NA	NA	44	3	Sterol	0.7	7
Weststrate & Meijer[40]	The Netherlands	80	NA	NA	45	3.5	Stanol	2.7	16
							Sterol	3.2	17
Miettinen & Vanhanen[41]	Finland	NA	[7]	8	45	9	Sterol	0.8	10
		NA	[9]	NA	NA	NA	Stanol	1.0	11
Davidson et al[42]	United States	NA	21	21	45	8	Sterol	3.0	5
Vanhanen et al[43]	Finland	NA	34	33	46	6	Stanol	3.4	13
Vanhanen et al[17]	Finland	NA	[7]	8	47	6	Stanol	0.8	11
		NA	[7]	NA	NA	NA	Stanol	2.0	21
Homma et al[44]	Japan	NA	[34]	34	47	4	Stanol	2.0	13
		NA	[36]	NA	NA	NA	Stanol	3.0	10
Vanstone et al[45]	Canada	15	NA	NA	48	3	Sterol	1.8	16
							Stanol	1.8	16
Hallikainen et al[46]	Finland	34	NA	NA	49	4	Sterol	2.1	17
							Stanol	2.0	21
Miettinen et al[47]	Finland	NA	[51]	51	50	52	Stanol	1.8	16
and Gylling et al[48]		NA	[51]	NA	NA	NA	Stanol	2.6	22
Jones et al[28]	Canada	NA	16	16	50	4	Stanol	1.7	25
Gylling et al[49]	Finland	22	NA	NA	51	7	Stanol	3.0	20
Christiansen et al[50]	Finland	NA	[47]	44	51	26	Sterol	1.5	18
		NA	[43]	NA	NA	NA	Sterol	3.0	19
Hallikainen et al[18]	Finland	22	NA	NA	51	4	Stanol	0.8	3
							Stanol	1.6	10
							Stanol	2.3	18
							Stanol	3.0	20
Volpe et al[51]	Italy	30	NA	NA	51	4	Sterol	1.0	13
Jones et al[11] and	Canada	15	NA	NA	52	3	Stanol	1.8	10
Raeini-Sarjaz et al[52]							Sterol	1.9	22
Neil et al[53]	Britain	29	NA	NA	52	8	Sterol	2.5	20
Gylling & Miettinen[54]	Finland	21	NA	NA	53	5	Stanol	2.4	17
Nguyen et al[55]	United States	NA	77	76	53	8	Stanol	2.0	8
							Stanol	3.0	17
Andersson et al[56]	Sweden	NA	19	21	55	8	Stanol	2.0	11
Blair et al[57]	United States	NA	71	77	56	8	Stanol	3.0	15
Tikkanen et al[58]	Finland	NA	36	35	56	5	Sterol	0.9	10
		NA	36	35	56	5	Sterol	1.9	14
		NA	36	35	56	5	Sterol	4.2	15
Noakes et al[59]	Australia	46	NA	NA	57	3	Sterol	2.3	13
							Stanol	2.5	16
		35	NA	NA	57	3	Sterol	2.0	15
Gylling & Miettinen[60]	Finland	11	NA	NA	58	6	Stanol	3.0	20
Nigon et al[61]	France	53	NA	NA	58	8	Sterol	1.6	9
Maki et al[62]	United States	NA	[90]	39	59	5	Sterol	1.1	17
		NA	[90]	NA	NA	NA	Sterol	2.2	20
Nestel et al[63]	Australia	15	NA	NA	60	4	Sterol	2.4	23

*Brackets indicate multiple treatment groups. LDL = low-density lipoprotein; NA = not applicable.

Table 2. **Summary Estimates From Randomized Placebo-Controlled Trials of the Absolute and Percentage Reductions in LDL Cholesterol According to Age***

Mean age of trial subjects (y)	No. of trial arms	Reductions in LDL cholesterol	
		Absolute (mg/dL) (95% CI)	% (95% CI)
4-6	2	8 (7-9)	8.0 (5.2-10.8)
20-29	2	11 (2-19)	10.0 (7.0-13.0)
30-39	7	12 (10-14)	10.5 (8.3-12.6)
40-49	13	15 (13-17)	10.3 (8.5-12.0)
50-60	26	16 (14-18)	9.6 (8.4-10.7)

*Data are from 50 trial arms (in adults) that tested daily doses of stanols/sterols of ≥1.5 g/d. CI = confidence interval; LDL = low-density lipoprotein.

placebo margarines or other foods were used to make the trials double-blind. In each trial, the average placebo-adjusted reduction in LDL cholesterol was calculated. Using STATA statistical software (Stata Corp, College Station, Tex), we determined the average reduction in LDL cholesterol across all the trials and across specified subgroups of trials. A random effects model was used; however, there was no statistically significant heterogeneity between trial results. Therefore, the summary estimates are equivalent to weighting the result of each trial by the inverse of its variance. The subjects in these trials often were selected for having relatively high cholesterol levels, but the effect of such selection proved slight because the average LDL cholesterol levels in the placebo groups were close to the age-specific average values for northern Europe where most of the trials were done (mean LDL values in the placebo groups across all trials were 137 mg/dL or 3.55 mmol/L at ages 45-54 years and 161 mg/dL or 4.17 mmol/L at ages 55-64 years). Trials in children with familial hypercholesterolemia were not included in the analysis and are discussed separately.

The absolute placebo-adjusted reduction in LDL cholesterol produced by sterols and stanols increases with age (Table 2); this may be due to the increase of the baseline values of LDL cholesterol with age. The percentage reduction in LDL cholesterol did not vary significantly with age.

The percentage reduction in LDL cholesterol as a function of dose is shown in Figure 2 and Table 3. The effect appeared to taper off at intakes of about 2 g/d or more, and there is little additional effect at doses higher than 2.5 g/d (Table 3): The maximum effect is an estimated 11.3%. In the trials, testing doses of 0.7 to 1.1 g/d suggested that approximately half the effect may be attained at this dose. In an analysis that excluded 2 trials in children and 8 low-dose (≤1.1 g/d) trials in adults, the mean reduction in the LDL cholesterol level was 10.1% (95% confidence interval, 8.9%-11.3%) in 27 trials testing stanols (mean dose, 2.5 g/d) and 9.7% (95% confidence interval, 8.5%-10.8%) in 21 trials testing sterols (mean dose, 2.3 g/d). The difference was not significant although the comparison lacked the statistical power to detect a moderate difference. Therefore, these trials cannot support a claim that either is better than the other. Although increases in high-density lipoprotein levels and reductions in triglyceride levels have been seen sporadically,[54,60] stanols and sterols in general produced little or no change in high-density lipoprotein or very low-density lipoprotein cholesterol, so the absolute reductions in LDL and total cholesterol levels were almost identical. Stanols lower LDL levels by inhibiting LDL apolipoprotein B production.[65] The response curve for apolipoprotein B was similar to that for LDL cholesterol, suggesting that stanols and sterols lower LDL by reducing the number of particles rather than their size or composition.[18,66]

The effect of stanols and sterols on plasma lipoproteins is primarily established within a few weeks, and it remained stable in studies lasting 1 year.[47] In these studies, treatment with 1.8 g/d of stanols lowered LDL cholesterol by 8.5% after 1 year,[47] and treatment with 1.6 g/d of sterols lowered LDL cholesterol by 5.9% after 1 year,[67] both relative to the change in the concurrent placebo group. The issue of whether this shows a difference in long-term efficacy between sterols and stanols was debated extensively by the experts, but firm conclusions are impossible because the data are limited to single studies, the doses were somewhat different, and there are no hard data on compliance. More information on these long-term effects obviously is needed.

What Is the Effect of Formulation?

The physical form of the sterols and stanols is important. Free (ie, unesterified) sterols and stanols can have the same effect on plasma lipoproteins as stanol and sterol esters[2,28]; however, the matrix and emulsification are important, and negative results are common.[68] Therefore, new food forms should be evaluated for efficacy if they differ greatly from previously tested forms. One study[69] showed that free stanols emulsified with lecithin reduced cholesterol absorption by 37% in single-meal tests. There is also evidence for efficacy of free plant sterols dissolved in diacylglycerol[70] and of plant sterol and stanol esters incorporated into low-fat products such as bread and cereals[63] or low-fat yogurt.

The effects of dimethylsterols from rice bran on lipoproteins are less than those of sterols derived from cholesterol, such as sitosterol (Figure 1).[5]

What Are the Effects of Intake Frequency?

In most of the trials, the total daily intake of plant stanols and sterols was divided into 2 or 3 portions over the day, but 1 study showed that 2.5 g of plant stanols taken at lunch

produced the same LDL-lowering effects as 2.5 g of plant stanols divided over the 3 meals.[32] Thus, the distribution of intake over the day may not be an important determinant of efficacy, and mechanisms other than replacing cholesterol from mixed micelles for inhibition of cholesterol absorption must be considered because this would presumably require ingestion of stanols and sterols with every meal. More trials to compare the efficacy and effectiveness of different dosing regimens are required. Studies of the effects of sterols and stanols on transporters of cholesterol may also shed light on this issue.[14,15]

What Are the Responses of Subgroups Including Children, Patients With Diabetes, and People With Defined Genotypes?

Two studies showed an LDL cholesterol reduction of about 15% in children with familial hypercholesterolemia treated with stanol esters, which compares favorably with the 8% reduction in children with average levels of LDL cholesterol (Table 2). A study with unesterified sitostanol showed an even more pronounced effect.[71] Sterol esters also produced a reduction in LDL cholesterol.[53] Therefore, sterols and stanols appear at least as effective in children with familial hypercholesterolemia as in children not so affected.

Stanol esters lowered LDL cholesterol levels by 9% in diabetic persons, similar to reductions in nondiabetic persons.[60] Absorption of cholesterol may be affected by apolipoprotein E genotype, but in most studies, the apolipoprotein E genotype has little effect on the response of LDL to sterols and stanols.[18,33,46,72] Other polymorphisms have not been studied widely.

Are the Effects of Stanols and Sterols Additive to Those of Diet?

The effects of stanols and sterols appear to be independent of the background diet.[62] Thus, addition of 2.3 g/d of stanol esters doubled the effect of a low-fat National Cholesterol Education Program (NCEP) step 1 diet, producing a total decrease in the LDL cholesterol level of 23%.[38] Similar effects were seen with free sterols and stanols.[28] Therefore, a few participants pointed out that sterols and stanols will lower cholesterol levels even in people eating otherwise unhealthy diets. However, diets recommended for the prevention of CHD provide more than just LDL lowering; they provide dietary fiber, essential fatty acids, vitamins, minerals, and beneficial substances from fruits and vegetables.[73] Therefore, most workshop participants believe that sterols and stanols should complement a healthy diet low in saturated fat and cholesterol and high in fruits, vegetables, and whole grains, with unhardened oils as the fat source.

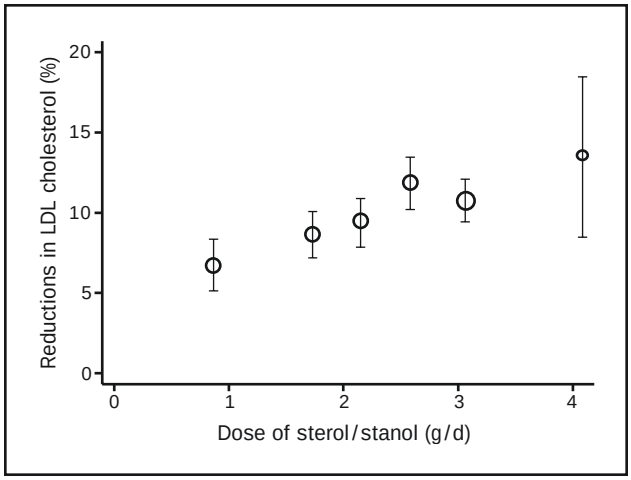

Figure 2. Summary estimates from randomized placebo-controlled trials of the percentage reductions in low-density lipoprotein (LDL) cholesterol with 95% confidence intervals according to dose. The area of each circle is proportional to the total number of persons in the trials in each dose range. The ranges of daily doses shown (number of trial arms in parentheses) are 0.7 to 1.1 g (8), 1.5 to 1.9 g (13), 2.0 to 2.4 g (14), 2.5 to 2.9 g (5), 3.0 to 3.4 g (13), and 4.0 to 4.2 g (2). (Two trials in children were omitted because doses are not equivalent in children and adults.)

Are the Effects of Sterols and Stanols Additive to Those of Cholesterol-Lowering Drugs?

In a trial of 167 adults receiving statin therapy, addition of stanol ester margarine lowered LDL cholesterol levels 10% more than addition of placebo.[57] Smaller studies in patients with CHD[49] and in adults with familial hypercholesterolemia treated with simvastatin[74] found additional LDL cholesterol level reductions of 16% and 20%, respectively. In a small study of patients with type 2 diabetes, addition of stanol esters to pravastatin therapy resulted in an LDL cholesterol reduction of 14%.[65] Sterols also reduced LDL levels in patients treated with statins.[53] Adding stanols and sterols appears somewhat more effective than doubling the statin dose, which usually produces an additional lowering of LDL cholesterol levels of only 5% to 7%.[57,75,76] Thus, for patients who are taking statins and need

Table 3. **Mean Percentage Reductions in LDL Cholesterol According to Dose***

Dose of sterol and stanol (g/d)	No. of trial arms	Reduction in LDL cholesterol (%) (95% CI)
0.7-1.1	8	6.7 (4.9-8.6)
1.5-1.9	13	8.5 (7.0-10.1)
2.0-2.4	14	8.9 (7.4-10.5)
≥2.5	21	11.3 (10.2-12.3)

*Data are from 56 trial arms that tested stanols/sterols (2 trials in children excluded). CI = confidence interval; LDL = low-density lipoprotein.

additional cholesterol lowering, it is more effective to add stanols and sterols to their diets than to increase their statin doses, and this option needs to be considered by physicians treating patients with hypercholesterolemia.

In patients with diabetes in whom cholesterol absorption was inhibited with neomycin, addition of stanol esters lowered LDL levels by 11%.[77] Cholestyramine also seems to produce an additive effect,[78] but more data are needed, as are data on the combined effects of sterols and stanols with niacin and ezetimibe.

What Are the Effects on Atherosclerosis in Animals?

Sterols and stanols lowered arterial lipid accumulation in mice, rabbits, and hamsters[79-82] but did not cause regression of established atherosclerosis.[83] In rabbits fed atherogenic diets, plant stanol consumption inhibited development of arterial plaque despite markedly elevated serum cholesterol levels, suggesting a process other than simple lowering of lipid levels.[80]

What Is the Potential Impact for CHD Risk Reduction?

Data from drug trials[84,85] indicate that the reduction in LDL cholesterol levels of about 10% could be expected to reduce the incidence of ischemic heart disease by about 12% to 20% over 5 years. An analysis of cohort studies indicates that the longer-term risk reduction would be about 20%.[86] No trials have directly tested the effects of sterols and stanols on CHD incidence; thus, there is a theoretical possibility that the expected beneficial effect on CHD will not be realized fully. However, most workshop participants believe that LDL cholesterol–lowering intervention rarely has failed to reduce the incidence of ischemic heart disease when tested in randomized trials.[86] Other participants would like the reassurance provided by a trial with hard end points.

How Should Plant Sterols and Stanols Be Incorporated Into the Clinical and Public Health Approaches to Cholesterol Lowering?

The lowering effect of stanols and sterols on LDL levels of 10% is similar to that of replacing 8% of energy as saturated fat by monounsaturated and polyunsaturated fats.[87] When sterols and stanols are combined with diets low in saturated fat and cholesterol, reductions of 20% or more can be achieved. The potential utility of plant stanols and sterols in clinical therapeutic lifestyle changes to lower LDL cholesterol levels has been recognized recently by the NCEP.[88] The NCEP's Therapeutic Lifestyle Changes diet thus recommends reduced intakes of saturated fat (<7% of total calories) and cholesterol (<200 mg/d) and encourages use of plant stanols/sterols (2 g/d) and soluble fiber (10-25 g/d) to enhance the LDL cholesterol–lowering effect of diet. Additional LDL lowering can result from weight reduction and increased physical activity, the other components of the Therapeutic Lifestyle Changes.

The optimal dose of sterol/stanol from margarines is consumed with 7 g of fat, of which 2 g is saturated; low-fat food forms also are available. Therefore, use of stanol- and sterol-containing foods is not expected to have adverse effects on weight or other risk factors. On the basis of current average LDL cholesterol levels in the United States and the recommended LDL cholesterol treatment goals, approximately 65 million US adults are candidates for these lifestyle changes. Also, the therapeutic diet should be an integral part of patient management, even when drug therapy is used, because patients treated with cholesterol-lowering drugs obtain additional significant LDL cholesterol reduction when plant stanols/sterols are added to their diets, enabling many of those not at their LDL goals to achieve those goals. Moreover, concomitant use of plant stanols/sterols may allow lower doses of drugs.

Plant stanols/sterols can have a role in the clinical management of high LDL cholesterol levels in children, particularly in those with familial hypercholesterolemia. They can facilitate use of lower doses of drugs or delay initiation of drug therapy. Long-term data on use of plant stanols and sterols in children are lacking, and such data are needed. However, data from studies in adults do not suggest any specific concerns regarding use by children. Specifically, levels of vitamin A (retinol) and D, which are particularly important for children, are not affected by the use of sterols and stanols in adults.

SAFETY

The safety of stanols and sterols has been reviewed by several regulatory agencies. The US Food and Drug Administration has accepted that plant sterol/stanol esters are asserted to be Generally Recognized as Safe (GRAS) by manufacturers. The Food and Drug Administration also has authorized a claim that foods containing plant sterol/stanol esters may reduce the risk of CHD (Federal Register of September 8, 2000 [65 FR 54686]). The Scientific Committee on Foods of the European Union concluded that phytosterol ester margarine was safe for human use (http://europa.eu.int/comm/food/fs/sc/scf/out56_en.pdf). In-use data of stanol and sterol esters are now available for up to 2 years in the United States and Europe and over 5 years in Finland, and no adverse effects have been reported. Thus, the balance of risk and benefit appears to be favorable. However, the lack of long-term (≥5 years) experience leaves a possibility of unforeseen effects.

What Do Toxicologic Tests in Animals Say About the Safety of Plant Stanols and Sterols?

Studies in rats confirm that stanols and sterols are absorbed poorly.[89] Plant stanols and sterols mainly were detected in the adrenal glands and ovaries. In humans, β-sitosterol can be metabolized to pregnenelone in adrenal glands, testes, and term placentas following the same pathways as cholesterol.[90] Whether plant stanols are metabolized in the same way is unknown.

In 2 studies, high doses of plant sterol esters and stanol esters were fed to rats.[91,92] There were no signs of toxicity or adverse effects except for some reduction of fat-soluble vitamins at the highest dose of 5%, equivalent to 4.1 g of plant stanols per kilogram of body weight per day (about 150 times higher than the recommended dose of 2 g/d in humans). In multigenerational rat studies,[93,94] plant sterol esters produced no effects on reproduction or development at up to 5% wt/wt of feed. For plant stanol esters, the No Observed Adverse Effect Level was the mid dose of 2.5% plant stanol because there were treatment-related effects on body weight of offspring in the high-dose group.

There is no evidence of a teratogenic effect for plant sterols and stanols.[95] Some reports say plant sterols and stanols have estrogenic effects[96-98]; however, plant sterols do not bind to the estrogen receptor,[99] and in various in vitro and in vivo assays there is no evidence of estrogenic activity for stanols.[100] Plant sterols and stanols are not genotoxic in vitro (bacterial mutation assay; metaphase chromosome analysis of human lymphocytes; mammalian cell gene mutation assay; mammalian cell chromosome aberration assay) or in rats in vivo (unscheduled DNA synthesis in rat liver; induction of micronuclei in rat bone marrow).[92,101,102]

Increased consumption of plant stanols and sterols elevated the amount of cholesterol in the large intestine. Cholesterol is metabolized by intestinal microflora into 4-cholesten-3-one, and a small increase in the fecal concentration of 4-cholesten-3-one was found after healthy volunteers consumed 8.6 g/d of phytosterols for 4 weeks.[103] 4-Cholesten-3-one has been reported to be mutagenic in some tests in vitro.[104,105] However, assessment of 4-cholesten-3-one in a bacterial mutation assay and in an in vitro chromosome aberration assay produced no evidence of mutagenic activity.[102,106]

One limitation of the animal studies is that the microflora of humans and rats are different. This might affect how plant sterols and stanols and cholesterol are metabolized in the gut. Also, the gastrointestinal systems of the rat and human are distinctly different. Finally, toxicologic studies have been confined to native stanols and sterols. If stanol- and sterol-enriched products were to be marketed for frying purposes, the stability of stanols and sterols on frying would need to be established first.

How Do Sterols and Stanols Affect the Absorption and Plasma Levels of Phytosterols and Stanols?

In normal subjects, intake of 2 g/d of plant sterols approximately doubles plasma campesterol levels; sitosterol levels increase also. Increased intake of stanols increases their plasma levels, but stanol concentrations are always a factor of 10 to 100 lower than sterols[46,55]; in addition, stanol consumption reduces plasma sterol concentrations.[8,11,46,107] Concentrations of plant sterols in plasma in subjects consuming sterol ester margarine are within the range of 0.6 to 2.0 mg/dL.[40,46] This is 20 to 100 times lower than in patients homozygous for phytosterolemia.

Phytosterolemia (also known as sitosterolemia) is an extremely rare recessive disease.[6,108] The homozygous state occurs in about 1 in 5 million people, with a wide margin of uncertainty. Patients are prone to premature atherosclerosis and CHD. Sitosterol absorption in homozygous individuals is typically 15% to 25% as opposed to 5% or less in normal persons, and excretion is reduced. Typical ranges for plasma concentrations are 14 to 65 mg/dL for sitosterol and 7 to 21 mg/dL for campesterol. Mutations in intestinal ABCG5 and ABCG8 transporters are responsible for at least some forms of phytosterolemia.[14,15,109]

In heterozygous persons (about 1 in 1100), absorption of phytosterols may be normal[110] or up to 15%,[6] but body pools are not greatly elevated because removal by the liver is rapid. Plasma levels of plant sterols in heterozygous people are normal or slightly elevated.[111] One study showed that consuming sterol margarine increased plasma campesterol concentrations 2- to 3-fold in people heterozygous for phytosterolemia, which is similar to the effect in people without the mutation.[112] This is reassuring because heterozygosity is much more common than homozygosity, and heterozygosity usually is undetected.

Does Absorption of Plant Sterols and Stanols Promote Atherosclerosis?

Sterols and stanols have been found in atheroma lesions. Mild hyperphytosterolemia is inherited[113] and is associated with an augmented risk of premature CHD[114,115]; it has been suggested that premature atherosclerosis in patients with homozygous phytosterolemia is due to an atherogenic effect of circulating sitosterol and campesterol. However, the high rate of atherosclerosis in patients with phytosterolemia could be due to other consequences of the genetic defect.

Some workshop participants believe that the slight increase in plasma sterols on consumption of foods enriched with plant sterol esters might detract from the beneficial effect on atherosclerosis produced by the decrease in LDL

Table 4. **Mean Change (95% CI) in Serum Concentrations of Vitamins in Randomized Placebo-Controlled Trials of Stanols/Sterols***

Vitamin	No. of trials	Mean change (%)	Mean change, adjusted for change in total serum cholesterol (%)	References
α-Tocopherol	15	–5.9 (–8.0 to –3.8) (*P*<.001)	2.1 (–0.3 to +4.5) (*P*=NS)	16,18,32,35,38,42,46,48,50,52,54,56,59,62,116
Alpha carotene	13	–8.7 (–13.8 to –3.5) (*P*<.001)	–0.3 (–5.7 to +5.2) (*P*=NS)	18,32,35,42,46,48,50,52,54,56,59,62,116
Beta carotene	15	–19.9 (–24.9 to –15.0) (*P*<.001)	–12.1 (–17.4 to –6.8) (*P*<.001)	18,32,35,38,42,46,48,50,52,54-56,59,62,116
Lycopene	13	–7.3 (–13.1 to –1.4) (*P*=.01)	–0.1 (–6.1 to +5.9) (*P*=NS)	16,18,32,35,39,40,42,46,52,56,59,62,116
Retinol	14	–0.1 (–1.6 to +1.5)	NA	18,32,38,42,46,48,50,52,54-56,59,62,116
Vitamin D	10	+0.5 (–2.6 to +3.6)	NA	16,38,42,46,48,52,54-56,62

*Only trials testing doses ≥1.5 g/d are included. CI = confidence interval; NA = not applicable; NS = not significant.

cholesterol. Others believe that the effect, if any, is completely compensated for by the decrease in plasma LDL. Further studies on the mechanism through which defects in ABC transporters produce atherosclerosis and CHD in patients with phytosterolemia might clarify this point. However, current data, although preliminary, suggest that not only normal subjects but also subjects heterozygous for phytosterolemia can consume plant sterols without adverse effects.[112]

How Do Stanols and Sterols Affect Plasma Levels of Fat-Soluble Vitamins?

Of the trials of sterols and stanols identified in the meta-analysis on efficacy (Table 1), 18 trials testing doses of 1.5 g/d or more reported plasma concentrations of fat-soluble vitamins. Table 4 summarizes the mean changes across trials. Statistically significant reductions occurred in the plasma concentrations of the hydrocarbon carotenes: alpha carotene by 9%, beta carotene by 28%, and lycopene by 7%. Part of this reduction probably is due to reduced absorption of carotenes and the rest to reduced concentrations of the lipoprotein carrier, LDL; after the decrease in total cholesterol induced by stanols and sterols was corrected for, a statistically significant reduction remained for beta carotene but not for alpha carotene or lycopene. The reduction in α-tocopherol was explained similarly by the reduction in cholesterol (Table 4). The decrease in beta carotene could be prevented by adding sufficient fruits and vegetables to the diet.[59] Vitamin D and vitamin A (retinol) concentrations are on average unaffected by sterols and stanols (Table 4). Vitamin K–dependent clotting factors did not change in subjects fed stanols.[33] When 8 patients taking coumarin were given stanol esters, no significant changes occurred in prothrombin time, and no major changes were needed in coumarin dose, suggesting that vitamin K status was unchanged.[117]

Do Reduced Absorption and/or Decreased Plasma Levels of Carotenes Constitute a Health Risk?

Substances that can reduce circulating carotenoids pose a theoretical public health concern because low levels of circulating carotenoids have been associated with increased risk of several chronic diseases including CHD, certain cancers, and macular degeneration.[118-120] In observational studies, lutein has been inversely associated with macular degeneration, beta carotene with lung cancer, lycopene with prostate cancer, and all the major circulating carotenoids with CHD.[120]

The amount of decrease in the serum carotenoid levels caused by plant stanols and sterols should be viewed in the context of other dietary factors that influence circulating levels. Coconsumption with wheat bran significantly inhibits lycopene and lutein absorption.[121] Volunteers who consumed olestra (8 g/d) had 38% lower levels of beta carotene than did controls after 8 weeks.[122] In addition, some lipid-lowering drugs (probucol and cholestyramine) cause decreases in beta carotene levels beyond what would be expected from the LDL-lowering effects of the drugs alone.[123]

Epidemiological studies cannot determine whether the associations between various carotenoids and disease outcomes are causal or due to confounding factors. Several randomized trials have tested the effects of supplemental beta carotene on cancer and CHD.[124-128] These trials show no benefit and in smokers show evidence of harm.[124,126] This weakens the case for beta carotene as a chemoprotective agent. Still, it is unclear whether supplement trials are relevant to predicting the effect of reductions in plasma carotenoid induced by stanols and sterols.

Part of these reductions are due to decreases in circulating LDL levels, which are the carriers for fat-soluble vitamins. In 2 large primary prevention trials using statins (each averaging about 5 years of follow-up), large reductions in LDL levels were not associated with an increase in any cancer.[84,85] The effects of these drugs on carotenoid levels have not been reported; thus, the interpretation of these trials with respect to carotenoid lowering is somewhat speculative.

Cholestyramine, a bile acid sequestering resin, reduces circulating carotenoid levels in excess of the decrease expected due to LDL lowering. In the Lipid Research Clinics Coronary Primary Prevention Trial,[129] total carotenoid levels decreased 26% in 3806 men after 1 year of intervention compared with a 19% decrease in LDL levels. Despite this large reduction in carotenoid levels, the reduction in coronary events was exactly that which would have been predicted by the changes in serum lipids.[130] Although the trial was underpowered for cancer end points, the on-trial (up to 10 years of follow-up) and posttrial (6 additional years of follow-up) experience showed a slight decrease in carotenoid-linked cancers in the intervention group relative to the placebo group.[131]

In summary, risks associated with decreased carotenoid levels secondary to sterol/stanol intake are theoretical at this point, but longer follow-up of studies would be desirable to fully address this issue.

Do Sterols and Stanols Interfere With the Absorption and Action of Therapeutic Drugs, Including Hormones?

Information is limited about sterol/stanol interference with the absorption and action of therapeutic drugs. An 8-week study of 318 subjects reported no adverse interactions of stanol intake with drugs.[55] In patients with diabetes, stanols had no effect on diabetic control.[60]

Stanols and sterols might theoretically interfere with cyclosporine absorption; further studies are needed. Few patients take cyclosporine, and levels are closely monitored. However, transplantation units should be aware of this potential interaction.

Could Stanols and Sterols or Their Metabolites Have Adverse Effects in the Colon?

Consumption of plant sterols and stanols increases the fecal excretion of cholesterol and its metabolites. Hypotheses were advanced in the 1960s and 1970s that bacterial metabolites of bile acids and cholesterol were involved in the genesis of colon cancer.[132] Toxicologic animal tests on this subject were discussed previously. Other experiments in vitro and in animals with chemically induced cancers suggested that dietary plant sterols may protect against colon, breast, and prostate cancer.[133,134] However, a prospective study in humans did not support any protective effect of plant sterols against colon or rectal cancer.[21] Thus, there are no convincing data to suggest that plant stanols and sterols either contribute to or prevent colon carcinogenesis.

What Is the Safe Upper Level of Intake of Stanols and Sterols? Is This Level Likely to Be Exceeded in Practice?

The evidence summarized previously suggests that intake of the recommended 2 g/d of sterols and stanols effectively lowers LDL cholesterol levels, produces no serious adverse effects, and poses no health risks of concern. Some consumers might reach higher intake levels if they consume a wide range of products enriched with stanols and sterols. There is no evidence that such higher intakes cause harm. However, increasing intake beyond 2 g/d of stanols and sterols produces little additional LDL lowering and therefore is not recommended.

RECOMMENDED FUTURE RESEARCH

Who Consumes Sterol/Stanol Products and What Is Their Effectiveness in the General Population?

In a Finnish survey,[135] 46% of people who used plant stanol ester margarine reported having cardiovascular disease. Users were mostly aged 55 years or older, were better educated, and had higher incomes than nonusers. They also had a healthier lifestyle. No data are available on the initial cholesterol levels of the users or on the extent of cholesterol lowering obtained when sterol- and stanol-rich foods were introduced into communities. Therefore, the effectiveness (efficacy multiplied by compliance) of sterols and stanols in the general population is still unclear.

Is Postmarketing Surveillance (Postlaunch Monitoring) Being Done, and Is It Useful for Detecting Unforeseen Effects?

About 2500 users of stanol margarines and matched controls are being followed up in Finland, and possible long-term health effects, both beneficial and adverse, will be determined by linking national disease and mortality registries with the cohort.[135] Although valuable, this study probably has limited power to detect rare adverse effects. The nonrandomized design also introduces confounding, difficult to avoid in observational studies. Telephone help lines have been installed and are monitored for self-reported adverse effects of stanol and sterol margarines.

Is a Trial With Hard End Points (Disease and Death) Required?

A randomized clinical trial might provide certainty about the efficacy of stanols and sterols in reducing CHD.

The costs and feasibility of a such a trial are prohibitive, as they have been for other dietary factors. The numbers of CHD patients needed to show the expected 12% to 20% reduction in CHD over 5 years would be 10,000 to 15,000. In healthy subjects, event rates are lower and required numbers proportionally higher. For example, with a 6% reduction in total cholesterol (corresponding to a 9% reduction in LDL cholesterol), close to 50,000 postmenopausal women might need to be randomized and followed up for 9 years to show the expected reduction of 14% in CHD at a power of 86%.[136,137] Moreover, such a trial would not provide total reassurance about the absence of long-term adverse effects because they are likely to be rare, and a clinical trial would be severely underpowered to detect them.

Trials with surrogate end points could be used to corroborate the expected efficacy for cardiovascular disease prevention. The most convincing surrogate outcome would be a decrease in the progression of intima-media thickness of the carotid artery, and some workshop participants believe that initiating such a trial would be useful. However, most agree that current evidence is sufficient to encourage use of sterols and stanols.

CONCLUSIONS

Abundant evidence shows that consuming 2 g/d of sterols and stanols lowers LDL levels by 10%, and based on epidemiological data and trials with cholesterol-lowering drugs, long-term use likely will lower CHD risk by 12% to 20% in the first 5 years and by 20% over a lifetime. Safety testing of sterols and stanols has exceeded that of ordinary foodstuffs that are eaten widely and generally recognized as safe. Adverse effects of the absorption of plant sterols into the circulation appear largely hypothetical in adults. Adverse health outcomes due to observed decreases in beta carotene levels in plasma are speculative and are of no major concern. Safety is being monitored by follow-up of samples from the general population eating these foods; however, the power of such studies to detect rare adverse effects, if any exist, is limited. A clinical trial would be extremely expensive and would probably not answer remaining questions about infrequent adverse effects. Trials with surrogate end points such as intima-media thickness might corroborate the expected efficacy of stanols and sterols in reducing the progression of atherosclerosis. However, current evidence is already sufficient to encourage use of sterols and stanols in persons with elevated cholesterol who are at increased risk for CHD.

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