Lifeline
Technologies™

November 15, 2001

Dockets Management Branch
HFA-305
Food and Drug Administration
5630 Fishers Lane, Rm 1061
Rockville, MD 20852

Re: Docket Nos. 00P-1275 and 00P-1276
Food Labeling: Health Claims; Plant Sterol/Stanol Esters and Coronary Heart Disease

Dear Sir or Madam:

Lifeline Technologies, Inc. (Lifeline) would like to comment to the Food and Drug Administration on the Health Claims for plant sterol/stanol esters referenced above. Lifeline supports this Health Claim and the Company believes that it is consistent with data in the literature and also with the Company's own findings.

The purpose of this response is to make available new human clinical data that Lifeline has accumulated over the past three years and which we believe will assist the Agency in reaching a decision on the Health Claim. Specifically, we will address the following three issues:

1. Eligibility of Unesterified Plant Sterols and Plant Stanols for the Health Claim

2. Daily Intake Levels Necessary to Reduce the Risk of CHD

3. Issues Regarding Safe Use of Plant Sterol/Stanol Esters in Foods and Advisory Label Statements

We appreciate your consideration of these comments, and please contact me (636 537-1382) if you have any questions.

Sincerely,

Curtis A. Spilburg

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President

00P-1275

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LIFELINE TECHNOLOGIES, INC.
COMMENT ON PROPOSED HEALTH CLAIM FOR PLANT STEROLS AND STANOLS

I. Company/Technology Background

Lifeline Technologies is developing proprietary technology that is focused on the ability of lecithin formulated soy sterols and soy stanols to lower human LDL-cholesterol by blocking its absorption in the gut. These plant-derived compounds or their derivatives are ideal agents to lower serum cholesterol since they are already in the diet and, unlike cholesterol, they are absorbed little or not at all. However, for the past 40 years little progress has been made on their commercialization, largely because their inherent water insolubility renders them physiologically unavailable. To circumvent this problem, sterols and stanols can be esterified with fatty acid, which renders them soluble in oil (triacylglycerol) the major component of margarine and salad dressing. This delivery strategy produces a food product that has been clinically proven to lower LDL-cholesterol.

Lifeline's approach provides an attractive alternative to these oil-based systems. By combining soy lecithin and soy sterols/stanols in the correct proportions, a water dispersible, fat-free solid is generated that can be added to a wide variety of food products. Importantly, the average consumer can readily understand that this fat-free system is consistent with established fat recommendations (National Cholesterol Education Program), which have been shown over the last twenty years to contribute to population-wide cholesterol reduction. Over the past three years, Lifeline has performed a number of human clinical studies that demonstrate the efficacy and

safety of this approach for reduction of cholesterol absorption and reduction of LDL-cholesterol. These studies serve to support previous work with sterol/stanol esters and demonstrate the utility and applicability of both delivery approaches (oil for sterol/stanol esters, water for free sterol/lecithin or free stanol/lecithin) for cholesterol reduction. Lifeline would like to share some of these new findings with the Food & Drug Administration to provide the Agency with a broader framework for establishing a Health Claim for free sterol/stanols and their ester derivatives.

II. Eligibility of Free, Unesterified Plant Sterols and Plant Stanols for the Health Claim

A. Background

Two methods can be used to determine the effect of lecithin-formulated free soy sterols or lecithin-formulated free soy stanols on intestinal cholesterol metabolism. The first method is an acute (single dose) human clinical study in which deuterated cholesterol is consumed as part of a test meal that contains the active substance (lecithin/sterol or lecithin/stanol) or placebo (lecithin alone). The concentration of plasma deuterated cholesterol is then measured three or four days later to determine the effect of the active substance on the absorption of cholesterol. This method can be used to determine the effect of different dose concentrations, food matrices and other dosing variables on cholesterol absorption.

The second method is a double blind, placebo controlled study in which lecithin formulated soy sterols or stanols are given to subjects over a four-week interval to

determine their effect on plasma total cholesterol and plasma LDL-cholesterol. This method requires diet control (American Heart Association Step I diet) and more subjects than the acute study described above

B. The Effect of Various Food Matrices and Cooking on Potency

Lecithin-formulated soy sterols/stanols are compatible with different foods, and the acute cholesterol absorption test described above can be used to determine the effect of different food matrices on the activity of the formulated material. A dose of 630 mg soy stanols (450 mg sitostanol) was blended into a lemonade-flavored beverage or added to commercial egg white and cooked into a pudding at 60^0 C. When added to the beverage, dispersed soy stanols reduced cholesterol absorption by 32.1 ± 5.0% (n=10, p=0.0045), while its addition to pudding lowered cholesterol absorption by a similar amount, 38.2 ± 5.4% (n=11, p=0.0022) (Spilburg et al., 2001).

This work shows that soy stanols can be dispersed in very different food matrices to produce a similar clinical effect. Protein-rich egg white products provide an especially convenient and practical system for testing the ability of powdered stanol/lecithin to remain bioavailable after cooking. At elevated temperatures ($\geq 60^0$C), egg protein undergoes irreversible denaturation or coagulation, a property that is used in many cooked foods such as custards, cakes and pie fillings. Moreover, in many food products coagulated egg protein binds together other ingredients, providing an excellent test system for activity after entrapment

in a food matrix. The data presented here show that when formulated soy stanols are presented in this food product as the delivery vehicle there is a 38% reduction in cholesterol absorption, very similar to that found for the beverage product. Taken together, these two studies show that lecithin formulated soy stanols, delivered by very different methods, provide a consistent and reproducible means for the reduction of cholesterol absorption and that cooking and other manipulations required for the preparation of a food product do not diminish this potency.

C. Reduction of Cholesterol Absorption (Acute Studies)

The results found above with soy stanols are similar to those reported earlier using purified sitostanol in the lecithin formulation system (Ostlund et al., 1999). Results from that work are summarized below.

Dose Response of Sitostanol Using Two Delivery Methods

Delivery Method and Amount	Reduction of Cholesterol Absorption	p-Value
Sitostanol powder; 1,000mg	11.3 ± 7.4%	N.S.
Formulated Sitostanol; 100 mg	5.6 ± 7.2%	N.S.
Formulated Sitostanol; 300 mg	34.4 ± 5.8%	0.01
Formulated Sitostanol; 700 mg	36.7 ± 4.2%	0.003

These data show that for this formulation system the effective dose of purified sitostanol is 300 mg and little therapeutic benefit, if any, is derived by doubling the dose.

In addition, soy sterols were also shown to be effective in this formulation system. When lecithin formulated soy sterols (630 mg) were administered to subjects in the same lemonade flavored drink as that used for soy stanols (described in II B),

there was a 23.5% ± 8.2% (p=0.015) reduction in cholesterol absorption. There was no statistical difference between these two results (p = 0.56) indicating that lecithin formulated sterols and lecithin formulated stanols have similar effects on cholesterol absorption. Importantly, all these studies demonstrate that lecithin formulated sterols or stanols, whether purified or present as mixtures from native soy, provide a potent means to lower human cholesterol absorption.

D. Reduction of LDL-Cholesterol (Chronic Studies)

1. Soy Stanols. In a chronic study, the same lemonade flavored drink described above was administered (t.i.d.) to 24 normal to mildly hypercholesterolemic adults for a total daily dose of 1.8 grams of free soy stanols. For the first six weeks, all the subjects received placebo drink that contained only lecithin. At randomization, half the group continued to receive placebo while the other half received soy stanols/lecithin. After four weeks treatment, the plasma total cholesterol and plasma LDL-cholesterol were lowered relative to placebo control by 10.1% (p=0.0019) and 14.4% (p=0.0016), respectively. There were no significant changes in HDL and triglycerides (Spilburg et al., 2001).

2. Soy Sterols. A chronic study of similar design was performed with lecithin formulated soy sterols. Once again, a lemonade flavored drink was consumed t.i.d. by 24 normal adults to give a total daily dose of 0.9 grams. For this preparation, relative to the placebo, control plasma LDL-cholesterol was

lowered 11.4% (p=0.03) and plasma total cholesterol was reduced 6.7% (p=0.05).

III. Daily Intake Levels Necessary to Reduce the Risk of CHD

Based on these results from chronic and acute studies and from a review of the literature, Lifeline has concluded the following:

A. From single dose acute studies, lecithin formulated soy sterols and soy stanols have similar potencies. The maximum potency of a single dose occurs within a range of 300 – 600 mg of free sterol or free stanol. To achieve the maximum effect on LDL-cholesterol, it may require more than one dose per day, a possibility that is currently under investigation.

B. Lecithin formulated soy sterols and soy stanols can also significantly reduce LDL-cholesterol, demonstrating for the first time that a relatively small amount (1.0 – 1.8 gram) of properly formulated compound can have a significant impact on circulating lipid levels. This is in marked contrast to previous work with solid sterols (Cytellin) that required greater than ten grams of material to produce small erratic reductions in LDL-cholesterol. Separate dose response studies will determine the efficacy of formulated sterols and formulated stanols.

IV. Issues Regarding Safe Use of Plant Sterol/Stanol Esters in Foods and Advisory Label Statements

A. Absorption of Soy Sterols and Soy Stanols

Despite widespread therapeutic use and occurrence in natural foods, there is little quantitative information about the absorption of phytosterols in humans. Formal absorption measurements are important in assessing safety since plasma phytosterol levels alone may give an incomplete picture of their metabolism. To better understand this dynamic process, Lifeline measured the systemic absorption of lecithin-emulsified soy sterols and soy stanols during test meals using dual stable isotopic tracers. Ten healthy subjects underwent two single-meal absorption tests in random order two weeks apart, one with intravenous dideuterated soy sterols and oral pentadeuterated soy sterols and the other with the corresponding labeled stanols. The oral/intravenous tracer ratio in plasma, a reflection of absorption, was measured by a sensitive negative ion mass spectroscopic technique and became constant after 2 days. The results are summarized below for the absorption of a single dose of lecithin formulated soy sterols or soy stanols given with a standard test breakfast.

Absorption And Plasma Half-Life For Each Component In Soy Sterols and Soy Stanols*

Phytosterol	% Absorption	$T_{½}$ (days)
Sitosterol	0.512 ± 0.038	2.94 ± 0.15
Sitostanol	0.0441 ± 0.0041	1.84 ± 0.24
Campesterol	1.89 ± 0.27	4.06 ± 0.33
Campestanol	0.155 ± 0.017	1.69 ± 0.11

* Ostlund et al., 2001

The data presented here indicate that the gastrointestinal absorption of lecithin-formulated soy sterols and soy stanols measured by mass spectrometry is less than 2%, a value that is lower than that reported previously in studies using radioactive isotopes (Ling & Jones, 1995). Importantly, the corresponding soy stanols are absorbed about ten-fold less than that for the unreduced counterparts. Therefore,

these results for soy sterols and soy stanols indicate that, at least in this formulation system, there should be less of an issue with safety than previously thought.

B. EC and ANZFSC Comments

1. Two regulatory agencies (EC and ANZFSC) recently expressed concern about the childhood (under 5 years of age) exposure level to plant sterols and plant stanols. This concern is surprising in light of the compositions of various approved infant formulas shown in the Table below (Huisman et al., 1996).

Composition of Various Infant Formulas

Sterol	Infant Formula (Plant Sterol Content in mg/dL)									
	Nu	Fri	S/NL	Apt	Enf	SMA	S26	S/Ire	Soya	Alm
Sitosterol	54.1	34.8	34.8	30.9	69.6	30.9	30.9	77.3	77.3	65.7
Campesterol	30.9	11.6	11.6	11.6	27.1	11.6	11.6	23.2	38.7	23.2
Stigmasterol	3.9	7.7	7.7	7.7	11.6	7.7	7.7	11.6	7.7	7.7
Fucosterol	ND	ND	3.9	ND	3.9	ND	ND	3.9	ND	3.9
i-Fucosterol	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9	3.9
Total Plant Sterols	92.8	58.0	61.9	54.1	116.1	54.1	54.1	119.9	127.6	104.4

Abbreviations: Nu, Nutrilon (Nutricia, the Netherlands); Fri, Frisolac (Friesland Dairy Foods, Netherlands); S/NL, Similac (Abbot, the Netherlands); Apt, Aptamil (Milupa, Germany); Enf, Enfamil (Mead Johnson, Canada); SMA, SMA (Wyeth, USA), S26, S-26(Wyeth, USA); S/Ire, Similac (Abbot, Netherlands); Soya, Nutrilon-Soya (Nutricia, the Netherlands); Alm, Almiron (Nutricia, the Netherlands)

Using these published compositions, a 3.5 kg newborn who drinks 700 mL of liquid formula per day has an exposure to plant sterols ranging from 10.82 mg/kg (Apt, Enf, S26) to 25.52 mg/kg (Soya). For comparison, a 70 kg adult consumes about 300 mg of plant sterols per day or 4.28 mg/kg, indicating that bottle-fed newborns may currently experience the highest exposure to these compounds of any population subgroup.

2. The two regulatory agencies cited above also raised concerns that subjects currently using statin therapy or other cholesterol reduction strategies should consult a physician before self administration of sterol/stanol containing spreads. Since these naturally occurring compounds have an attractive safety profile and since they have proven efficacy, following this combination therapy would appear to add little, if any, risk. Moreover, there is now clinical evidence that shows that administration of sterols/stanols can permit a lower dose of statin (Vuorio et al., 2000; Blair et al., 2000).

It is not clear why this type of combination therapy has been identified as one which requires physician approval, especially since the Food & Drug Administration has taken a different position with other foods that have been approved to bear a claim for cholesterol reduction. For instance, Quaker Oats claims

> Oatmeal helps remove cholesterol. Three grams of soluble fiber from oatmeal daily in a diet low in saturated fat and cholesterol may reduce the risk of heart disease. This cereal has two grams per serving.

There is no mention of a need for consumers on statin therapy to consult a physician in order to change their breakfast cereal, and it is not clear why such a statement should be needed if this same group would like to change their spread.

In summary, the goal of all these therapies is to provide a "seamless" strategy for cholesterol reduction. In the past, borderline high cholesterol was untreated in younger subjects because of concerns about very long term use of potent statin drugs. The introduction of formulated sterols and stanols, along with the consumption of less fat and more fiber, provides a safe starting therapy for younger subjects, a strategy that is consistent with the Therapeutic Lifelstyle Changes recommended in the latest report of the National Cholesterol Education Program (Executive Panel, National Cholesterol Education Program, 2001). Following this regimen, statin drugs can be introduced later in life when the aging process raises serum cholesterol even in the presence of these natural compounds. Statin drugs should then be thought of as adjunct therapy to diet, exercise and sterols and stanols.

V. Conclusion

Based on all the considerations described above, we request that the Interim Final Rule be amended as follows:

1) Lecithin formulated free sterols and free stanols should be recognized as active agents for cholesterol reduction. This fat-free delivery system for the unesterified compounds lowers human cholesterol absorption regardless of the food matrix. Moreover, when properly formulated, these naturally-occurring compounds also lower LDL-cholesterol by 11-14%, as demonstrated in 4-week dosing studies.

2) For purposes of a health claim, the dosage level for both formulated sterols and formulated stanols should be 1000 mg/day.

3) Lecithin formulated soy sterols and soy stanols are absorbed less than previously thought, indicating that safety may be less of a concern. In addition, since bottle-fed infants are exposed to one of the highest levels of soy sterols (on a mg/kg basis), it would seem that the concerns relating to childhood exposure to these compounds in other foods are unwarranted. Therefore, the labeling of sterols and stanols should not be treated any differently than that used for other naturally-occurring compounds that have been shown to lower LDL-cholesterol.

Thank you for the opportunity to comment on the Interim Final Rule and if you have any further questions please contact Dr. Curtis A. Spilburg, President, Lifeline Technologies, Inc. at (636) 537-1382.

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