GR

ORIGINAL SUBMISSION

December 14, 2009

Office of Food Additive Safety (HFS-255)
Center for Food Safety and Applied Nutrition
Food and Drug Administration
5100 Paint Branch Parkway
College Park, MD 20740-3835

RE: Notification of GRAS Determination for Krill-based Phosphatidylserine in Food

Dear Sir/Madame:

In accordance with proposed 21 CFR § 170.36 (Notice of a claim for exemption based on a GRAS determination) published in the Federal Register (62 FR 18939-18964), I am submitting in triplicate, as the agent to the notifier, Enzymotec, a GRAS Notification for krill-based phosphatidylserine under the conditions of its intended use in food.

Please let me know if you have any questions.

Sincerely,

Edward A. Steele
President

Enclosures

EAS
CONSULTING GROUP

Specializing in FDA Regulatory Matters

GRAS NOTIFICATION

I. Claim of GRAS Status

A. Claim of Exemption from the Requirement for Premarket Approval Requirements Pursuant to Proposed 21 CFR § 170.36(c)(1)

Enzymotec Ltd. (the notifier) has determined that phosphatidylserine derived from krill is Generally Recognized As Safe, consistent with Section 201(s) of the *Federal Food, Drug, and Cosmetic Act*. This determination is based on scientific procedures as described in the following sections, under the conditions of its intended use in food as a nutrient. Therefore, the use of krill-based phosphatidylserine is exempt from the requirement of premarket approval.

Signed,

______________________ Date 12/14/09

Edward A. Steele

Agent for:

Enzymotec Ltd.
P.O.Box 6, Migdal HaEmeq
Israel 23106

EAS Consulting Group, LLC
1940 Duke Street, Suite 200, Alexandria, Virginia 22314
(877) 327-9808 Toll Free • (703) 684-4408 Local • (703) 684-4428 Fax
www.easconsultinggroup.com

B. Name and Address of Notifier:

Sigalit Zchut, Ph.D.
Regulatory Affairs Manager
Enzymotec Ltd.
P.O.Box 6, Migdal HaEmeq
Israel 23106

Telephone: 972-74-7177177
Facsimile: 972-74-7177001
Email: sigalit@enzymotec.com

C. Common or usual name of the notified substance:

The common name of the substance of this notification is phosphatidylserine. The substance is a complex mixture of primarily polar lipids, with phosphatidylserine as the primary phospholipid present. As described below, the composition of the commercially available phosphatidylserine may vary depending on the edible food source from which it is obtained. Because the fatty acid composition of marine-derived substances differs from that of plants from a nutritional perspective, a unique name is appropriate. In the case of krill-based phosphatidylserine, the fatty acids attached to the glycerophosphate backbone are primarily the long-chain polyunsaturated fatty acids eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA) and palmitic acid. In this notice, we are describing the product as krill-based phosphatidylserine.

D. Conditions of use:

Phosphatidylserine derived from krill is intended for use as a nutrient in the following food categories, when these foods do not contain other ingredients that are good sources of EPA or DHA: Breakfast Cereals, Dairy Product Analogs, Grain Products and Pastas, Milk Products, and Processed Fruits and Fruit Juices at use levels of 30 mg phosphatidylserine *per* serving (reference amounts customarily consumed, 21CFR 101.12). In addition to these categories, phosphatidylserine is also intended for use under the supervision of a physician in Medical Food at levels not to exceed 300 mg phosphatidylserine/person/day. Under Section 5(b) of the Orphan Drug Act (ODA), a Medical Food is defined as a food that is formulated to be consumed or administered enterally under the supervision of a physician and that is intended for the specific dietary management of a disease or condition for which distinctive nutritional requirements, based on recognized scientific principles, are established by medical evaluation. The intended use of krill-based phosphatidylserine in medical foods will be as per these and other applicable regulations.

Krill-based phosphatidylserine is intended for use in those foods that do not contain other significant sources of EPA and DHA. The intended use of krill-based phosphatidylserine in the above mentioned food categories results in an estimated daily intakes for "users only" at mean and 90th

percentile exposure estimates of 39 mg/person (0.83 mg/kg body weight/day) and 79 mg/person (1.84 mg/kg body weight/day), respectively. The resulting 90th percentile intake of EPA+DHA from the intended use of krill-based phosphatidylserine is determined to be 33 mg/person/day.

E. Basis for GRAS Determination:

In accordance with 21 CFR 170.30, the intended use of krill-based phosphatidylserine has been determined to be Generally Recognized As Safe (GRAS) based on scientific procedures. A comprehensive search of the scientific literature was also utilized for this review. There exists sufficient qualitative and quantitative scientific evidence, including human and animal data to determine safety-in-use for krill-based phosphatidylserine. Both phosphatidylserine and other sources of fatty acids (DHA and EPA) has been the subject of several previous GRAS notifications. In four separate GRAS notifications on phosphatidylserine (GRN 000186; GRN 000197; GRN 000223; GRN 000279), FDA responded that they had no questions regarding the conclusions that the use of phosphatidylserine is GRAS under the conditions described in the notices. The safety determination of phosphatidylserine is primarily based on human observations and a variety of animal as well as *in vitro* studies that further corroborate the human data. Results from over 35 human clinical trials demonstrated that phosphatidylserine is well-tolerated at doses from 200 to 600 mg/day.

The safety of two important fatty acids of krill-based phosphatidylserine, namely EPA and DHA, has been extensively evaluated by the FDA in the 1997 final rule on the approved use of menhaden oil as a direct food ingredient and subsequently, regarding the use of omega-3 fatty acids as a dietary supplement in 2005. In 21 CFR 184.1472 and as noted in the Final Rule FR March 23, 2005 (Volume 70, Number 55 PP14530-14532), FDA affirmed the GRAS status of menhaden oil for use in foods provided daily intakes of DHA and EPA did not exceed 3 g/person/day. In this Final Rule, the FDA reaffirmed that the intake of DHA and EPA must not exceed 3 g/day from all fish oil sources.

On the basis of both scientific procedures[1] corroborated by history of exposure from natural sources, Enzymotec considers the consumption of krill-based phosphatidylserine as an added food ingredient to be safe at levels up to 300 mg/day. Recently, FDA has agreed to exercise enforcement discretion for a Health Claim Petition[2] on phosphatidylserine, in which the petitioner demonstrated that phosphatidylserine is safe at levels up to 500 mg/day.[3] FDA permitted the use of a Qualified Health

[1] 21 CFR §170.3 Definitions. (h) Scientific procedures include those human, animal, analytical, and other scientific studies, whether published or unpublished, appropriate to establish the safety of a substance.

[2] Letter Regarding Dietary Supplement Health Claim for Phosphatidylserine and Cognitive Dysfunction and Dementia" (2003) Available at: http://www.cfsan.fda.gov/~dms/ds-ltr33.html

[3] Petition for Health Claim: Phosphatidylserine and Cognitive Dysfunction, Phosphatidylserine and Dementia (2002) Available at: http://www.fda.gov/ohrms/dockets/dailys/02/Sep02/091302/80027351.pdf

Claim on dietary supplements containing EPA and DHA on October 31, 2000 as well as for conventional foods on September 8, 2004. FDA concluded that the use of EPA and DHA omega-3 fatty acids as dietary supplements is safe, provided that daily intakes of EPA and DHA do not exceed 3 g/person/day from conventional food and dietary supplement sources.

The estimated daily intake of krill-based phosphatidylserine from its intended uses at the 90th percentile of 79 mg/person (1.84 mg/kg/day) is well below the safe levels of intake determined from human studies where no adverse effects from intakes of 300 mg of phosphatidylserine/person/ day (5 mg/kg body weight/day) were noted. Similarly the estimated intake of EPA+DHA of 33 mg/person/day from the intended uses is considerably lower than the established safe levels of 3000 mg/person/day of EPA+DHA. Additionally, any EPA/DHA intake from the intended uses of krill-based phophatidylserine will be substitutional. The estimated daily intake of krill-based phosphatidylserine, if ingested daily over a lifetime, is considered safe.

F. Availability of Information:

The data and information that forms the basis for this GRAS determination will be provided to Food and Drug Administration upon request and are located at the offices of:

Sigalit Zchut, Ph.D.
Regulatory Affairs Manager
Enzymotec Ltd.
P.O.Box 6, Migdal HaEmeq
Israel 23106

Telephone: 972-74-7177177
Facsimile: 972-74-7177001
Email: sigalit@enzymotec.com

II. Detailed Information About the Identity of the Notified Substance:

A. Chemical name

Phosphatidylserine derived from krill.
Phosphatidylserine. Per IUPAC-CBN nomenclature, it is a 1,2-diacyl-*sn*-glycero-3-phospho-L-serine.

B. Trade Name:

The subject of this notification will be marketed as PS-Omega 3

C. Chemical Abstract Registry Number:

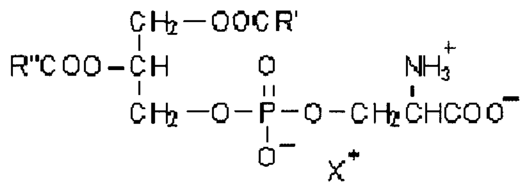
Phosphatidylserine: 84776-79-4

D. Chemical Formula:

The empirical formula of the most abundant krill-derived phosphatidylserine molecule in PS-Omega 3 is $C_{42}H_{71}O_{10}PNCa$

E. Structure:

Phosphatidylserine consists of a glycerophosphate skeleton conjugated with 2 fatty acids and L-serine *via* a phosphodiester linkage (Figure 1). The structural diagrams in Figure 1 show the general representation of the glycerophosphate backbone with R as fatty acids and the structure of most abundant molecule.



General structure of phosphatidylserine

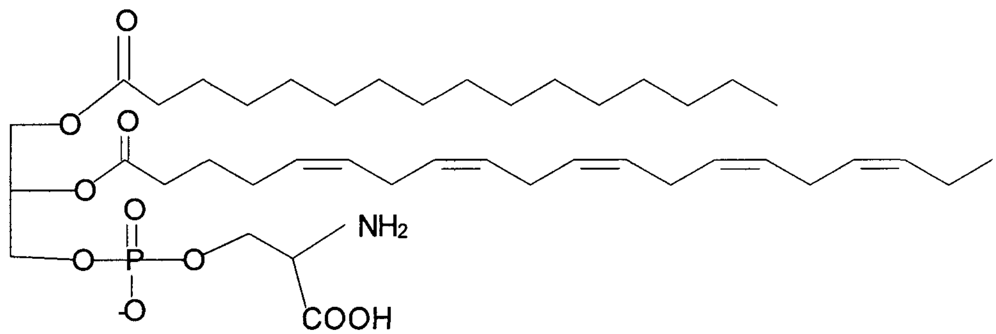


Figure 1. Structure of most abundant molecule present in PS-Omega3
(Phosphatidylserine with palmitic acid at sn-1 position and EPA at sn-2 position)

F. Fatty Acid Profile:

The typical fatty acid profile of krill-based phosphatidylserine is presented in Table 1.

000007

Table 1. Fatty acid profile of krill based phosphatidylserine

Fatty acid	Percentage*
C14 (Myristic)	2.0
C16 (Palmitic)	23.5
C16:1n7 (Palmitoleic)	1.8
C18 (Stearic)	1.0
C18:1n9 (Oleic)	6.5
C18:1 (Octadecenoic)	5.5
C18:2n6 (Linoleic)	2.0
C18:3n6 (gamma-Linolenic)	0.2
C18:3n3 (alpha-Linolenic)	1.2
C18:4n3 (Octadecatetraenoic)	1.9
C20:1n9 (Eicosenoic)	0.6
C20:4n6 (Arachidonic)	0.7
C20:4n3 (Eicosatetraenoic)	0.6
C20:5n3 (Eicosapentaenoic)	31.0
C22:1n11(Cetoleic)	0.3
C22:1n9 (Erucic)	1.0
C22:5n3 (Docosapentaenoic)	0.7
C22:6n3 (Docosahexaenoic)	14
C24:1n9 (Nervonic)	0.3
Others	5.0

*As percent of total fatty acids on phosphatidylserine

G. Molecular Weight

Based on the fatty acid profile given above, the average molecular weight of krill-based phosphatidylserine is 820 Daltons.

H. Physical Characteristics

Krill-based phosphatidylserine is produced as a brown to red colored powder.

I. Typical Composition and Specifications

Typical compositional analysis and specifications of krill-based phosphatidylserine are presented in Tables 2. The phospholipid profile of phosphatidylserine from krill is presented in Table 3.

Table 2. Typical compositional analysis and specifications of krill-based phosphatidylserine

Parameter	Typical values/ Specifications	Assay method
Physical properties		
Consistency	Powder	Visual
Color	Brown to red	Visual
Composition		
Phosphatidylserine (%w/w)	55	HPTLC/^{31}P-NMR
Total phospholipids (%w/w)	80	^{31}P-NMR
EPA+DHA (as % of FA on PS)	45	AM 030 based on AOCS official method Ce 1b-89
EPA+DHA (%w/w)	23	AOCS official method Ce 1b-89
Peroxide value (meq/Kg)	<0.5	AOCS official method Cd 8-53
Volatiles (g/100g)	1.5	Assay method LOD
Residual solvents (mg/Kg)	In compliance with GMP	GC/MS Headspace
Ethanol	<5000ppm	GC-MS headspace
Hexane	<30	GC-MS headspace
Ethyl acetate	<30	GC-MS headspace
Heavy metals		
Inorganic arsenic (mg/kg)	<0.2	EPA Method 1638 and HG-CT-AAS
Lead (mg/kg)	<0.2	ICP (inductively coupled plasma) emission spectroscopy
Cadmium (mg/kg)	<0.2	ICP (inductively coupled plasma) emission spectroscopy
Mercury (mg/kg)	<0.2	ICP (inductively coupled plasma) emission spectroscopy
Chemical contaminants		
Pesticides (ppb)	None detected	US EPA Method 505
Dioxins and Furans (pptWHO TEQs)	<5	US EPA method 1613B
Total PCBs (pptWHO TEQs)	<5	US EPA Method 1668A
Microbiology		
Total plate count (cfu/g)	<1000	Israel Standard SI 885 part 3 (1999)
Moulds (cfu/g)	<100	Israel Standard SI 885 part 8 (1999)
Yeasts (cfu/g)	<100	Israel Standard SI 885 part 8 (1999)
Coliforms (cfu/g)	Negative	Israel Standard SI 885 part 4 (1999)
Staphylococcus aureus (cfu/g)	Negative	Israel Standard SI 885 part 6 (1999)
Salmonella (cfu/20g)	Negative	Israel Standard SI 885 part 7 (1999)

cfu = colony forming units

Table 3. Typical phospholipids profile (as % of total phospholipids)

Phospholipids	Percent
Phosphatidylserine	70
Lysophosphatidylserine	1
Phosphatidylcholine	1
Phosphatidylinositol	2
Phosphatidylethanolamine	7
Phosphatidic acid	7
Other phospholipids	9

J. Manufacturing process

Lecithin from Krill (*Euphausia superba*), which is produced by solvent extraction from the Krill biomass, and which is the subject of a GRAS notification (GRN 000226, 2007), is used as the source material for the production of Krill-based phosphatidylserine.

The phosphatidylserine is produced through enzymatic transphosphatidylation of the Krill lecithin with L-serine using a phospholipase D enzyme, which catalyses the substitution of the choline head-group with serine to form phosphatidylserine. The enzymatic treatment does not alter either the fatty acids attached to the phospholipid molecules or their stereospecificity. Following the enzymatic reaction, the product is purified from the reaction medium and a final blending with food-grade silicon dioxide is carried out in order to produce a free-flowing powder. Small quantities of food-grade antioxidants (tocopherols, ascorbyl palmitate and rosemary extract[4]) are added to the product in accordance with good manufacturing practices. Processing aids, such as solvents (which are removed by vacuum evaporation) and buffer salts used in the manufacturing process are all of food-grade quality as specified in the 5th Edition of Food Chemicals Codex.

[4] The rosemary extract is equivalent to what is used as a flavor and is used at a level too low to provide flavor, hence this use is considered as GRAS.

K. Manufacturing process diagram

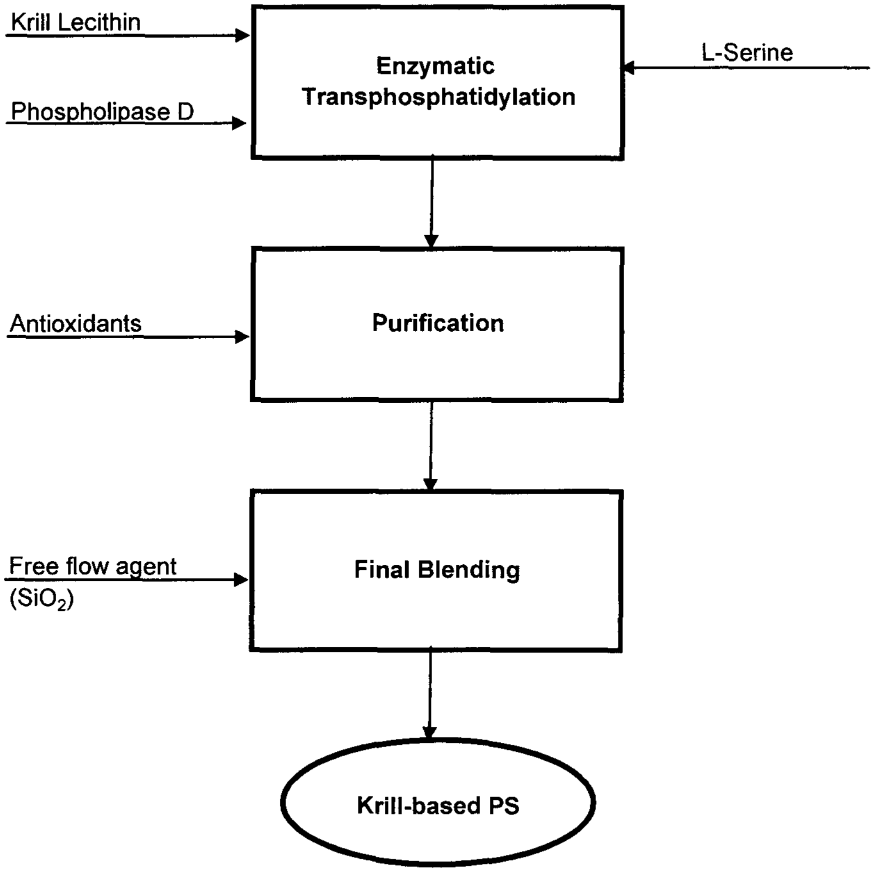


Figure 2. Manufacturing process of krill-based phosphatidylserine
(Abbreviations used: SiO_2 = silicon dioxide; PS = phosphatidylserine)

L. Intended Technical Effects

Krill-derived phosphatidylserine is intended for addition to a limited number of conventional foods as a nutritional ingredient. Its use is intended for the general population at the levels identified in this document for addition to the following food categories: Breakfast Cereals, Dairy Product Analogs, Grain Products and Pastas, Milk Products, Processed Fruits and Fruit Juices, and Medical Food. It is recognized that there are Standard of Identity requirements for some of these foods (i.e., certain bakery products), located in Title 21 of the Code of Federal Regulations. If used in such foods, the name will be changed so as not to be confused with the standardized food.

III. Summary of the Basis for the Notifier's Determination that Krill-Based Phosphatidylserine is GRAS

A comprehensive search of the scientific literature for safety and toxicity information on phosphatidylserine and omega-3 fatty acids was conducted through September 2009 and was also utilized for this review.

Based on a critical evaluation of the pertinent data and information summarized here and employing scientific procedures, it is determined that the addition of krill-based phosphatidylserine to the following foods containing no other ingredients that are good sources of EPA or DHA: Breakfast Cereals, Dairy Product Analogs, Grain Products and Pastas, Milk Products, Processed Fruits and Fruit Juices when not otherwise precluded by a Standard of Identity, and to Medical Foods meeting the specification cited above and manufactured according to current Good Manufacturing Practice, is Generally Recognized As Safe (GRAS) under the conditions of intended use in selected foods, as specified herein.

In coming to this decision that krill-derived phosphatidylserine is GRAS, we relied upon the conclusions that neither phosphatidylserine nor any of its degradation products pose any toxicological hazards or safety concerns at the intended use levels, as well as on published toxicology studies and other articles relating to the safety of the product. It is also our opinion that other qualified and competent scientists, reviewing the same publicly available toxicological and safety information, would reach the same conclusion.

IV. Basis for a Conclusion that Krill-Based Phosphatidylserine is GRAS for its Intended Use.

DETERMINATION OF THE GENERALLY RECOGNIZED AS SAFE (GRAS) STATUS OF KRILL-BASED PHOSPHATIDYLSERINE AS A NUTRIENT

1. INTRODUCTION

A comprehensive search of the scientific literature for safety and toxicity information on phosphatidylserine and omega-3 fatty acids was conducted through September 2009 and critically evaluated to determine the Generally Recognized As Safe (GRAS) status of krill-based phosphatidylserine as a nutrient [21 CFR 170.3(o)(20)][5] in Breakfast Cereals, Dairy Product Analogs, Grain Products and Pastas, Milk Products, and Processed Fruits and Fruit Juices at use levels of 30 mg phosphatidylserine *per* serving (reference amounts customarily consumed) and in Medical Foods at levels not to exceed 300 mg phosphatidylserine/person/day.

1.1. Occurrence

Phosphatidylserine, an aminophospholipid, is widely distributed among animals, plants and microorganisms. It is a component of mammalian cell membranes and play important roles in biological processes such as apoptosis and cell signaling. Of the total phospholipids found in the human body, phosphatidylserine comprises less than 10%. Approximately 30 g of phosphatidylserine is found in the human body and about half of this amount (~13 g) is present in brain tissue (Horrocks et al., 1982). In the brain, phosphatidylserine comprises 15% of the total phospholipid pool.

The fatty acid composition of endogenous phosphatidylserine depends on its localization and function in the cell. The brain and eye are also highly enriched with omega-3 fatty acids. It has been suggested that one of the fatty acids, docosahexaenoic acid (DHA) is maintained at certain levels in the retina despite reductions in dietary omega-3 fatty acids, and a decrease in dietary DHA has been observed to lead to a decrease in phosphatidylserine (Hodge et al., 2006). As the major structural and functional component of the central nervous system, DHA constitutes as much as 20-50% of the total fatty acid content of the adult human brain (Svennerholm, 1968; White et al., 1973; Neuringer et al., 1988). The omega-3 fatty acids, DHA and eicosapentaenoic acid (EPA) play an important role in brain function. Both DHA and EPA are orthomolecules whose functional sites are exclusively cell membranes, wherein they are structurally and functionally integrated via phospholipid molecules. In the krill-based phosphatidylserine that is the subject of this Notification, both EPA and DHA are chemically-bound components of the phosphatidylserine.

1.2. Chemistry and Biological Activity

Phosphatidylserine has three different chemical components: a glycerol backbone, a polar head group, and a hydrophobic moiety (Figure 1, page 5). The head group consists of the amino acid serine, which is attached to the 3-carbon backbone (glycerol) *via* a phosphate group; the two other hydroxy groups of the glycerol moiety are each

[5] "Nutrient supplements": Substances which are necessary for the body's nutritional and metabolic processes.

esterified with a fatty acid. The term phosphatidylserine refers to a group of molecules due to the fact that the fatty acid substituents on the two R positions can vary considerably, depending on their source. Phosphatidylserine derived from soy lecithin contains mainly polyunsaturated fatty acids (up to 18 carbons), while that from bovine brain contains mainly saturated and monounsaturated fatty acids, as well as some DHA (Hendler and Rorvik, 2001). Phosphatidylserine is amphophilic in nature because of the negatively charged head group (hydrophilic) and the fat-soluble fatty acid tails (lipophilic) (Hendler and Rorvik, 2001). Phosphatidylserine cannot be synthesized by the human body *de novo* and is produced only *via* exchange of the head group from phosphatidylcholine (PC) and/or phosphatidylethanolamine (PE), which are catalyzed by phosphatidylserine synthase-1 or -2. Thus, phosphatidylserine is formed from phosphatidylethanolamine by exchange of the ethanolamine head for L-serine.

Phosphatidylserine, primarily located in the internal layer of the cell membrane, performs a variety of structural and regulatory functions. In addition to regulating several metabolic processes such as activation of cell membrane bound enzymes, phosphatidylserine is involved in governing membrane fluidity and neuronal signaling. The sodium-potassium-stimulated enzyme ATPase is activated by phosphatidylserine. It also activates tyrosine hydroxylase, which in turn is involved in neurotransmitter synthesis, and thus influences dopaminergic and adrenergic signal transduction in the brain. Phosphatidylserine also facilitates calcium uptake into nerve cells. It is involved in processes of cell repair and removal. It can stimulate synthesis of the neurotransmitter acetylcholine to both improve receptor functioning and restore nerve signal transduction. Neurons are continuously forming axonal and dendritic extensions with accompanying cell membranes and as such, these growing membranes require fluidity for proper functioning. DHA has been identified as the most fluidizing element in cell membranes (Kidd, 2007; 2008).

1.3. Description, Manufacturing Process and Specifications

General descriptive parameters and properties of krill-based phosphatidylserine produced by Enzymotec are summarized in Table 4. The product is a brown to red-colored powder with slight fishy odor and flavor. Detailed information about the identity of phopshatidylserine along with specifications, composition and manufacturing are summarized earlier in Section II.

Analytical results of three lots from three different batches indicate that the product consistently meets the specifications (Appendix I). The results of pesticide, PCBs and dioxins and furans analyses from a combination of the three batches are presented in Appendix II. These results support previous analysis from krill-lecithin (GRN 000226, 2007). It should be noted that the source material, lecithin used in the manufacturing of krill-based phosphatidylserine is GRAS substance, the subject of our earlier GRAS notification (GRN 000226, 2007).

Table 4. General descriptive characteristics of krill-based phosphatidylserine

Parameter	Description
Chemical name	Phosphatidylserine; 1,2-diacyl-*sn*-glycero-3-phospho-L-serine
CAS Number	84776-79-4 (general, not specific to krill phosphatidylserine)
Average molecular weight	820
Chemical formula (most abundant)	$C_{42}H_{71}O_{10}PNCa$
Physical state	Powder
Solubility	Soluble in hexane, chloroform, toluene. Insoluble in water, acetone, ethanol
Stability	24 months
Color	Brown to red
Odor	Light fishy
Flavor	Light fishy
Storage	Below 25°C, in sealed containers, protected from light and moisture.

1.4. Technical effects

Krill-based phosphatidylserine is intended for use in food as a nutrient for individuals who wish to increase their daily intake of phosphatidylserine enriched with omega-3 fatty acids (EPA+DHA). It is intended for addition to a limited number of conventional foods such as Breakfast Cereals, Dairy Product Analogs, Grain Products and Pastas, Milk Products, Processed Fruits and Fruit Juices, and Medical Foods as a nutritional ingredient at the levels identified in this document. It is recognized that there are Standard of Identity requirements for some of these foods, and therefore, in these cases, it is not the intent to refer to them by their commonly recognized names such as milk, or yogurt.

1.5. Current Uses

Phosphatidylserine is extensively marketed as a dietary supplement. FDA has concluded that "the use of phosphatidylserine as a dietary supplement is safe and lawful under 21 CFR§101.14 provided that bovine-derived sources, if used, are not derived from bovine tissues from cattle born, raised, or slaughtered in any country where bovine spongiform encephalopathy (BSE) exists."[6] In reply to three separate GRAS notifications, FDA had no questions on the use of phosphatidylserine in different food categories under the conditions described in these notifications. In the first notification submitted by Lipogen (GRN 000186, 2006), use of phosphatidylserine in 28 different food categories/products resulting in estimated mean and 90th percentile intakes of approximately 140 and 240 mg/person/day, respectively, was determined to be GRAS. In the second GRAS notification from Degussa Food Ingredients GmbH (GRN 000197, 2006), use of phosphatidylserine at levels of 20 mg/serving in yogurt, powdered milk,

[6]Phosphatidylserine and Cognitive Dysfunction and Dementia (Qualified Health Claim: Final Decision Letter) http://www.cfsan.fda.gov/~dms/ds-ltr36.html

ready-to-drink soymilk, meal replacement, cereal bars, powdered beverage mixes, chewing gum, and breakfast cereals was considered as GRAS. In the third notice submitted by Enzymotec (GRN 000223, 2007), use of phosphatidylserine at levels of up to 100 mg/serving was considered as GRAS and in the fourth notice (GRN 000279, 2009) up to 30 mg/serving was considered as GRAS.

1.6. Intake from Natural Presence in Food

Phosphatidylserine is ubiquitous in all biological membranes of plants and animals. As such, it is a typical constituent of the human diet. It is found in small amounts in foods such as meats, eggs, soy products, certain legumes, and milk. Bruni et al. (1989) reported an estimated daily intake of phosphatidylserine of about 75 mg/day. On the basis of a scientific analysis of phosphatidylserine exposure, Hamm (2004) determined an average intake of phosphatidylserine as 130 mg/day, with light eaters of meat and fish consuming about 100 mg and vegans consuming less than 50 mg/day. In their GRAS notification, Degussa (GRN 000197, 2006) determined the estimated dietary intake of phosphatidylserine in the US population. Based on their analysis which was limited to the foods in which the presence of phosphatidylserine has been reported, the estimated average and 90th percentile intakes of phosphatidylserine for an adult from natural sources were determined as 98 and 184 mg/person/day, respectively. In another notification, Lipogen (GRN 000186, 2006) estimated that consumers of meat ingest approximately 80 mg of naturally-occurring phosphatidylserine *per* day. These reports demonstrate that the dietary intake of phosphatidylserine, from its natural occurrence in the diet, ranges from 75 to 184 mg/day.

1.7. Intended Use Levels and Food Categories

Enzymotec intends to use krill-derived phosphatidylserine as a nutrient [21 CFR §170.3(o)(20)] at levels of 30 mg/serving (reference amounts customarily consumed, 21 CFR 101.12) in the following food categories Breakfast Cereals, Dairy Product Analogs, Grain Products and Pastas, Milk Products, and Processed Fruits and Fruit Juices. In addition to these categories, phosphatidylserine is also intended for use in Medical Food at levels not to exceed 300 mg phosphatidylserine/person/day. The intended use levels and food categories for uses of krill-based phosphatidylserine are identical to those of Enzymotec's recent GRAS notice on Fish-based phosphatidylserine (GRN 000279, 2009). Further, this GRAS determination does not encompass foods containing other significant sources of DHA or EPA.

1.7.1. Estimated Daily Intake from the Intended Uses

As the intended use levels and food categories for uses of krill-based phosphatidylserine are identical to those of Enzymotec's recent GRAS notice (GRN 000279, 2009), previous analysis of fish-based as well as soy-based phosphatidylserine were used for the determinations of estimated daily intake. In the previous submissions, estimates of possible daily intake were determined using the CSFII 1994-96 database (USDA, 1998). Based on the exposure estimates prepared (Appendix III), approximately 60% of the total U.S. population was identified as potential consumers of krill-based phosphatidylserine (12,503 actual users identified). Although infants are included in the intake determinations, krill-based phosphatidylserine is not intended to be added to baby foods or infant formula. Consumption from the identified food categories intended for addition of krill-based phosphatidylserine by the total U.S. population resulted in estimated mean all-person and all-user intakes of phosphatidylserine of 22 mg/person/day (0.46 mg/kg body weight/day) and 39 mg/person/day (0.83 mg/kg body weight/day), respectively. The 90th percentile all-person and all-user intakes of phosphatidylserine from all intended food-uses by the total population were 61 mg/person/day (1.30 mg/kg body weight/day) and 79 mg/person/day (1.84 mg/kg body weight/day), respectively. Only male teenagers and adults slightly exceeded this mean. For details of the consumption analysis, please see Appendix III.

The resulting intake of EPA+DHA from the intended uses was determined using the amount of these two fatty acids (~23% w/w) and phosphatidylserine (~55%) present in the product (Table 2, page 7). Based on the consumption analysis discussed earlier, the estimated 90th percentile phosphatidylserine intake is 79 mg/day. As phosphatidylserine in the final product is 55%, the amount of product consumed will be 144 mg/day. The amount of EPA+DHA in the final product is ~23% (w/w), hence the resulting intake of EPA+DHA will be 33 mg/day. Thus the intended proposed use of krill-derived phosphatidylserine will result in an estimated 90th percentile intake of 33 mg EPA+DHA/person/day.

The FDA commonly uses the estimated daily intake for the 90th percentile consumer of a food additive as a measure of high chronic dietary intake. Hence, for the safety determination in this assessment the resulting 90th percentile intake of krill-based phosphatidylserine (79 mg/person/day) from its intended uses in the above described food categories are considered. A summary of the estimated daily intake of phosphatidylserine from the intended food categories is presented in Table 5.

Table 5. Intake of krill-based phosphatidylserine (mg/person/day) based on USDA data

Population Group	Age Group (Years)	% Users	Actual # of Total Users	All-Person Consumption (mg)		All-Users Consumption (mg)	
				Mean	90th Percentile	Mean	90th Percentile
Infant	0 to 2	53.1	1,902	13.59	36.37	22.71	45.50
Child	3 to 11	80.7	5,090	30.19	68.18	37.93	74.25
Female Teenager	12 to 19	54.8	385	22.54	61.20	40.24	75.00
Male Teenager	12 to 19	55.5	386	30.79	85.59	55.42	109.64
Female Adult	20 and Up	54.0	2,471	19.26	53.81	35.30	71.75
Male Adult	20 and Up	47.8	2,269	21.11	63.28	45.20	91.44
Total Population	**All Ages**	**60.7**	**12,503**	**21.97**	**61.09**	**39.42**	**79.14**

1.7.2. Cumulative intake

The intended uses of krill-derived phosphatidylserine in the specified food categories are identical to those described in our fish-derived phosphatidylserine GRAS (GRN 000279, 2009). Compared to our other GRAS Notification (GRN 000223, 2007) for soy-derived phosphatidylserine, the intended uses are substitutional and lower, except for milk powder. For the cumulative intake estimation, in the previous GRAS determination we compared the intended food uses of phosphatidylserine with the earlier GRAS Notifications [GRN 000186 (Lipogen) and GRN 000197 (Degussa)]. A comparison of food categories along with use levels from the FDA GRAS Notifications is compared with the present GRAS determination in Table 6. As presented in the table, all the food categories from the present GRAS determination are the same as those identified in previous notices. Thus the food category comparison shows that all the food categories from the present Notification are substitutional.

As noted previously in the GRAS notifications from Enzymotec (GRN 000223; GRN 000279), in order to gain some insight on the intake of phosphatidylserine, the estimated daily intake (EDI) of phosphatidylserine from all GRAS Notifications (GRN 000186, GRN 000197 and GRN 000223) were added to determine cumulative intake. The 90th percentile EDI for "all-users" from the intended uses of phosphatidylserine in all food categories for GRN 000186 was reported as 240 mg, for GRN 000197 it was reported as 52.4 mg, while for GRN 000223 it was 98.73 mg. The total (or cumulative) intake from all three GRAS Notifications was 391.13 mg. This determined cumulative EDI was the 'worst case' scenario, and the actual intakes are likely to be lower, because of simple addition of the values without considerations of overlapping intake (as the majority of the food categories in the three notices were substitutional). In a Health Claim Petition on phosphatidylserine, where FDA agreed to exercise enforcement discretion, the petitioner demonstrated that phosphatidylserine is safe at levels up to 500 mg/day. The intake value determined by simple addition of EDIs from the three separate referenced GRAS Notifications was below this value of 500 mg/day.

It should be noted that the intended use levels and food categories of the present GRAS determination are identical to those of Enzymotec's GRN 000279. In the present GRAS determination, the use levels of krill-based phosphatidylserine are significantly lower compared to the previous soy-based phosphatidylserine (GRN 000223). Additionally, compared to GRN 000223, all food categories are substitutional except for milk powder. However, it should be noted that milk powder was one of the food categories previously identified in GRAS notification GRN 000186 as well as in GRN 000279. For the present GRAS determination, all food categories for the proposed use of krill-based phosphatidylserine are substitutional, and the intended use levels are lower compared to these previous GRAS determinations (GRN 000186, GRN 000197 and GRN 000223). All these considerations suggest that the intended uses of krill-based phosphatidylserine in the specified categories at levels of 30 mg/serving will not add to the existing intake of phosphatidylserine and estimations of cumulative daily intake are not necessary.

Table 6. Comparison of GRAS food uses and use levels with previous GRAS Notifications

Lipogen (GRN 000186)		Degussa (GRN 000197)		Enzymotec (GRN 000223)		Enzymotec (GRN 000279) and current GRAS	
Food category	**Use levels %**	**Food category**	**Use levels %**	**Food category**	**Use levels %**	**Food category**	**Use levels %**
Breads and Rolls	0.05 to 0.08	NA	NA	NA	NA	NA	NA
Biscuits	0.08	NA	NA	NA	NA	NA	NA
Crackers	0.08	NA	NA	NA	NA	NA	NA
Waffles	0.05	NA	NA	NA	NA	NA	NA
Carbonated Beverages	0.05	NA	NA	NA	NA	NA	NA
NA	NA	**Powdered beverages**	**0.01-0.18**	NA	NA	**Powdered milk**	**0.01**
Meal Replacements (Liquid)	**0.05**	**Meal replacement**	NA	**Meal replacement (milk-based)**	**0.04**	**Meal replacement (milk-based)**	**0.01**
Ready-to-Eat Breakfast Cereals	**0.08**	**Cereals**	**0.03-0.08**	**Breakfast cereals**	**0.021**	**Breakfast cereals**	**0.01**
NA	NA	Chewing gum	0.7-1	NA	NA	NA	NA
Cheese	0.25	NA	NA	NA	NA	NA	NA
Cheese Spreads	0.25	NA	NA	NA	NA	NA	NA
Coffee	0.05	NA	NA	NA	NA	NA	NA
Tea	0.05	NA	NA	NA	NA	NA	NA
Ketchup	0.5	NA	NA	NA	NA	NA	NA
Soy-Based Milk	**0.05**	**Soy milk**	**0.01-0.67**	**Soy milk**	**0.04**	**Soy milk**	**0.01**
Margarine	0.25	NA	NA	NA	NA	NA	NA
Mayonnaise	0.05	NA	NA	NA	NA	NA	NA
Fat-based Spreads	0.25	NA	NA	NA	NA	NA	NA
Pasta	0.05	NA	NA	NA	NA	NA	NA
Grain-based Bars	**0.05**	**Breakfast bars**	**0.03-0.08**	**Grain based**	**0.25**	**Grain based bars**	**0.07**

				bars			
Commercial Jams	0.25	NA	NA	NA	NA	NA	NA
Milk	**0.05**	NA	NA	**Milk**	**0.02**	**Milk**	**0.01**
Milk Drinks	**0.05**	NA	NA	**Milk drink**	**0.04**	**Milk drink**	**0.01**
Yogurt	**0.05**	**Yogurt**	**0.01-0.01**	**Yogurt**	**0.04**	**Yogurt**	**0.01**
Peanut Spreads	0.25	NA	NA	NA	NA	NA	NA
Tofu	0.25	NA	NA	NA	NA	NA	NA
Soy-based Meat Substitutes	0.05	NA	NA	NA	NA	NA	NA
Fruit Juices	**0.05**	NA	NA	**Fruit juices**	**0.04**	**Fruit juices**	**0.01**
Salty Snacks	0.05	NA	NA	NA	NA	NA	NA
Chocolates	0.05	NA	NA	NA	NA	NA	NA

NA = Not applicable, as use of phosphatidylserine was not proposed in these categories by the notifier

1.8. Summary of Consumption

The estimated daily intake of krill-based phosphatidylserine for the various food categories identified above was determined using CSFII 1994-96 database (USDA, 1998). As such, the high users-only 90th percentile consumption value of krill-based phosphatidylserine as an ingredient for food uses in specified food categories [Breakfast Cereals, Dairy Product Analogs, Grain Products and Pastas, Milk Products, and Processed Fruits and Fruit Juices] was considered as the estimated daily intake resulting from the intended use of krill-based phosphatidylserine. Based on these considerations, the intended uses of krill-based phosphatidylserine will result in daily 90th percentile exposure of 79.14 mg/person/day (1.84 mg/kg body weight/day). Only male teenagers and adults slightly exceeded this mean. Phosphatidylserine is naturally present in food and its intake from the diet ranges from 75 to 184 mg/day and the 90th percentile intakes by male teenagers and adults are well within this range. In addition, the estimated 90th percentile intake of EPA+DHA from the intended uses of krill-derived phosphatidylserine is determined as 33 mg/person/day.

2. Toxicology

The safety of phosphatidylserine and omega-3 fatty acids is supported by multiple human clinical trials and a variety of animal as well as *in vitro* experimental studies that further corroborate the human observations. Because of the physiological role of phosphatidylserine and omega-3 fatty acids in human health, there have been considerable efforts to elucidate the biological role of these ingredients in humans. As a result, the literature is full of information on phosphatidylserine and omega-3 fatty acids. Relevant biological and toxicological studies on phosphatidylserine and omega-3 fatty acids are included in the following section in support of the safety conclusions determined in this assessment.

2.1. Source of Phosphatidylserine and Metabolism

Phosphatidylserine has been historically derived from bovine cortex. However, in recent years, because of potential contamination concerns from BSE prions, alternative sources of phosphatidylserine from soy and marine biomass have become common. The safety database of phosphatidylserine is primarily based on studies in which the ingredient was derived from plants and animals. Hence, it is important to compare the composition of different forms of phosphatidylserine for an understanding of the applicability of the various studies for safety assessment. Soy-derived phosphatidylserine contains mainly polyunsaturated fatty acids, while bovine cortex derived phosphatidylserine (BCPS) contains primarily saturated and monounsaturated fatty acids, as well as some docosahexaenoic acid (DHA) (Hendler and Rorvik, 2001).

Krill-based phosphatidylserine, the subject of this GRAS determination, contains typically 23% palmitic acid, 6.5% oleic acid, 31% EPA, and 14% DHA (Table 1). Phosphatidylserine derived from bovine brain cortex has been reported to contain 8-29% DHA, 38-44% palmitic acid, and 15-38% oleic acid (Claro et al., 1999; Salem, 1980; Yabuuchi et al., 1968), while phosphatidylserine derived from soy contains approximately 7% α-linolenic acid and 47% linoleic acid. Although, the fatty acid composition of krill-based, versus bovine cortex-derived and soy-derived phosphatidylserine differs, as discussed below, these differences are unlikely to affect the safety profile as long as the specific fatty acid intake stays within safe levels. Following ingestion, phosphatidylserine from any origin is modified in the gastrointestinal mucosa. In the human body, ingested phosphatidylserine is modified depending on the physiological need. Secondly, depending on the target tissue requirements, the fatty acid profile of phosphatidylserine is modified to meet the need of the tissue/organ. These facts demonstrate that the fatty acid composition of phosphatidylserine, and thus the source, is unlikely to play a role in its safety.

2.2. Absorption, Metabolism and Excretion

It is well established and recognized that phosphatidylserine from either plant or animal sources undergo a similar metabolic fate. Following dietary ingestion, pancreatic digestive enzymes cleave specific fatty acids leading to the formation of lysophospholipids that are absorbed by the mucosal cells of the intestine and can be reacylated into phosphatidylserine. The fatty acids released can be further used for triglyceride synthesis (Tso, 1994). Because of the high activity of decarboxylases in the mucosal cells, the majority of the phosphatidylserine is converted into other phospholipids, and notably, to phosphatidylethanolamine (Wise et al., 1965). The reacylated phosphatidylserine, phosphatidylethanolamine and other phospholipids enter the lymph and circulation, and are redistributed. Available evidence suggests that only part of the ingested phosphatidylserine reaches systemic circulation as part of the phospholipid pool.

The versatile biological actions of orally administered phosphatidylserine have been suggested to be due to shuffling of the fatty acid at tail 2 position on glycerol either during the course of absorption, while the molecule is in an intestinal cell, or after its delivery to an organ. The fatty acids of phosphatidylserine are also shuffled to meet the needs of the cell as they change over time, or as the phosphatidylserine "parent molecule" are transported from tissue to tissue, cell to cell, or perhaps even from spot to spot within a membrane. Enzymes such as hydrolases, acyltransferases that remove or replace the tail 2 position fatty acid are present in the digestive juices, in the intestinal lining cells, and in the membranes of all the other cells of the body. Depending on the functional needs of the cell, the acyltransferases remove fatty acids from phosphatidylserine (or other phospholipids), and replace them with other fatty acids (Kidd, 1998).

Orally administered phosphatidylserine has good bioavailability. Following oral administration of radio-labeled phosphatidylserine to rats at dose level of 20 mg/kg, radiolabel appeared in the blood at about 30 minutes. Immediately after its absorption, phosphatidylserine begins to accumulate in the liver and the brain (Toffano et al., 1987). Following intravenous administration of 20 mg/kg of phosphatidyl L-[U-^{14}C]-serine to mice, blood levels of radioactivity declined in a biphasic manner (Mazzari et al., 1982; Toffano et al., 1987). At 20 minutes after administration, the amount of radioactivity peaked in the brain and was 0.25% of the dose. The amount of radioactivity in liver was 36% of the dose and reached to peak after 20 minutes (Mazzari et al., 1982). Following a bolus intravenous injection of radio-labeled phosphatidylserine (2 mg/kg) to rats, plasma concentration showed a biphasic decline with half-lives of 0.85 and 40 min (Palatini et al., 1991). The initial decline was found to be due to the irreversible uptake of phosphatidylserine liposomes by the mononuclear phagocyte system, as demonstrated by the almost exclusive accumulation of phosphatidylserine in the liver and spleen. The slow decline phase reflects the elimination of that fraction of phosphatidylserine that has been incorporated into high density plasma lipoproteins. Comparative analysis of the biotransformation products found in tissues following either [^{3}H]-glycerol- or [^{14}C]-serine-phosphatidylserine injection show that parenterally administered phosphatidylserine follows two pathways: decarboxylation to phosphatidylethanolamine, and extensive hydrolytic degradation with release of the individual components of the molecule (Palatini et al., 1991). Unlike phosphatidylcholine, phosphatidylserine does not accumulate in the body, and approximately 60% of the orally ingested phosphatidylserine is excreted in feces, with 10% eliminated in urine. The major metabolite recovered was lysophosphatidylcholine and to a lesser extent lysophosphatidylserine (Toffano et al., 1987).

In summary, given the metabolic sequel described above, there is no scientific reason to conclude that the krill-derived phosphatidylserine and its associated lipids normally present in marine biomass would pose any health hazards different from plant oil, or fish oil, or bovine cortex-derived phosphatidylserine and lipids. Any differences in lipid content based on the marine origin versus plant or animal derived phosphatidylserine would also not pose a risk as these are natural constituents of the diet. The following section will also discuss the consequences of consumption of EPA and DHA resulting from krill-based phosphatidylserine, and the possible presence of source-based contaminants.

2.3. Human Studies

The clinical database of phosphatidylserine intake includes over 35 clinical trials of which 17 have been identified as double-blind protocols. The objective of the majority of these studies was to examine the effect of phosphatidylserine in reducing the symptoms of dementia and cognitive dysfunction in geriatric individuals. Although these investigations were designed to study the efficacy of phophatidylserine, clinical observations also included any adverse effects. In the clinical trials with phosphatidylserine, over 1500 subjects participated and the treatment lasted for periods of up to 6 months. The doses used in these trials ranged from 200 to 800 mg/day. Results from these studies show that oral administration of phosphatidylserine at doses of up to 600 mg/day for up to 3 months were without any significant adverse effects. In the largest double-blind, placebo-controlled trial (Cenacchi et al., 1993), of the 494 participants', only one subject dropped out because of a treatment-related adverse effect as compared to seven drop-outs from the placebo group that were considered as treatment-related. The reason for drop-out for these subjects was reported as dizziness. In the majority of the clinical trials (Table 7), phosphatidylserine derived from bovine cortex was used. In five trials, the source of phosphatidylserine was soy. As discussed earlier, the source of phosphatidylserine is unlikely to affect its safety. A summary of clinical trial design, doses and adverse effects noted in these investigations is presented in Table 7.

In a review article, Pepping (1999) reported that phosphatidylserine is well-tolerated at a dosage up to 300 mg/day. The only adverse effects reported were stomach upset in some individuals at doses higher than 300-400 mg/day and sleeplessness at doses ≥600 mg taken before bedtime. There are no reported drugs, nutritional supplement, food or herbal interactions with phosphatidylserine. Hendler and Rorvik (2001) reported occasional gastrointestinal side effects (nausea and indigestion) following phosphatidylserine ingestion. These effects were attributed to the oily nature of the ingested phosphatidylserine and vehicle used. These symptoms can be minimized by consuming phosphatidylserine with food.

In a randomized, double-blind, placebo controlled trial, Viseman et al. (2008) investigated the effects of EPA and DHA in phospholipid (phosphatidylserine) or triacylglycerol (fish oil) in children aged 8-13 years with impaired visual sustained attention performance. Children received placebo, 250 mg/day EPA + DHA esterified to phospholipid (300 mg/day phosphatidylserine), or fish oil for 3 months. Sixty of the 83 enrolled children completed the interventions (n = 18-21 per group). No treatment-related adverse effects were noted. A significant correlation between the alterations in fatty acids and increased TOVA (Test of Variables of Attention) scores mainly occurred in the phosphatidylserine group. The investigators concluded that consumption of EPA+DHA esterified to different carriers affected the incorporation of these fatty acids in blood fractions and improved the visual sustained attention performance in children.

Table 7. Reported adverse effects of phosphatidylserine in clinical trials

Reference; study design	PS Source	Number Subjects	Dose (mg/day); Duration	Adverse Effects Reported
Allegro et al., 1987	BC-PS	30	300; 60 days	No symptoms of adverse reactions were observed
Amaducci et al., 1988; DB-PC	BC-PS	142	200; 90 days	No change noted in pre- and post-dose clinical exams, clinical chemistries, and blood counts; no adverse events
Caffarra et al., 1987	BC-PS	30	300; 60 days	None reported
Cenacchi et al., 1987; DB-PC	BC-PS	130	300; 60 days	No treatment related clinically significant adverse effects.
Cenacchi et al., 1993; DB-PC	BC-PS	425	300; 180 days	Dizziness, vomiting and dyspepsia reported in a few patients, mainly in the placebo group. No pharmacological interactions
Crook et al., 1991; DB-PC	BC-PS	149	300; 12 wks.	Well tolerated; no adverse events
Crook et al., 1992; DB-PC	BC-PS	51	300; 12 wks.	Well tolerated; no adverse events
Crook, 1998	S-PS	50	300; 12 wks	No adverse effects noted
Delwaide et al., 1986; DB-PC	BC-PS	35	300; 6 wks.	No significant side effects noted
Engel et al., 1992; DB	BC-PS	33	300; 8 wks.	None reported
Fahey and Pearl, 1998; DB	S-PS	11	800; 2 wks.	None reported
Funfgeld et al., 1989; DB-PC	NA	62	300; NA	None reported
Granata & Michele, 1987	BC-PS	35	300; 60 days	None reported
Gindin et al., 1993; DB-PC	S-PS	57	300 mg; 3 mos.	No adverse effects noted
Heiss et al., 1994	BC-PS	18	400; 6 mos.	None reported
Hershkowitz et al., 1989; DB	BC-PS	24	300; 6 mos.	None reported
Jorissen et al., 2001; DB-PC	S-PS	81	300 or 600; 12 wks.	None reported
Maggioni et al., 1990; DB	BC-PS	10	300; 30 days	None reported
Monteleone et al., 1992	BC-PS	9	800; 10 days	None reported; BP unchanged
Palmieri et al., 1987; DB-PC	BC-PS	87	300; 60 days	No change noted in pre- and post-dose clinical and neurological exams, clinical chemistries, and EEG
Puca et al., 1987	BC-PS	27	300; 60 days	No change in pre- and post-dose blood biochemistry parameters
Rabboni et al., 1990	BC-PS	30	400; 60 days	No reported changes in liver and kidney function blood biochemistry or blood counts

Reference; study design	PS Source	Number Subjects	Dose (mg/day); Duration	Adverse Effects Reported
Ransmayr et al., 1987; DB-PC	BC-PS	39	300; 60 days	Few patients with epigastric pain associated with oil or gelatin caps
Schreiber et al., 2000	S-PS	18	300; 12 wks	No changes noted in serum electrolytes, glucose, thyroid function, and differential blood counts; no adverse effects noted
Sinforiani et al., 1987	BC-PS	34	300; 60 days	No remarkable side effects
Villardita et al., 1987; DB-PC	BC-PS	170	300; 90 days	None reported
Vaisman et al., 2008; DB-PC	F-PS	60	300; 90 days	No side effects. Well tolerated
PS = phosphatidylserine; DB-PC = double-blind placebo-controlled; BC-PS = Bovine cortex derived phosphatidylserine; S-PS = soy derived phosphatidylserine; wks = weeks; mos = months				

2.4. Subchronic Studies

Repeat-dose safety studies in rats and dogs show that oral administration of bovine cortex-derived phosphatidylserine (BCPS) at doses up to 1000 mg/kg/day for up to 6 months was without any significant adverse effects of toxicological concern (Heywood et al., 1987).

2.4.1. Rat Studies

Groups of Sprague-Dawley rats (20/sex/group) were administered daily *via* gavage with 0 (vehicle control), 10, 100, and 1000 mg/kg BCPS for 26 weeks (Heywood et al., 1987; Degussa, 2006). Compared to controls, BCPS administration did not significantly affect survival, weight gain and feed or water consumption. In some of the animals receiving 1000 mg/kg/day, post-dose salivation was noted. No significant hematological changes attributable to BCPS treatment were noted. In male and female rats receiving the highest dose of BCPS, a slight increase in alkaline phosphatase levels was observed. At week 13, in males receiving the highest dose of BCPS, serum albumin levels were slightly lower, potassium levels were elevated and serum sodium values were decreased. In males and females receiving the highest dose, lower urine pH values were recorded. Terminal necropsy did not reveal any adverse macroscopic or microscopic treatment-related findings. In the absence of dose-related effects, minimal changes, and lack of histological changes, the changes in serum and urine parameters in this study were considered minor and of no health or toxicological significance. The results of this study show that the no-observed-adverse-effect level (NOAEL) is lower than 1000 mg/kg/day but is close to this dose.

Blum et al. (2007a) investigated the subchronic toxicity of DHA-algal oil, produced through the use of the non-toxigenic and non-pathogenic marine protist, *Ulkenia* sp. Sprague-Dawley Crj:CD (SD) IGS rats (15/sex/group) were orally (*via* gavage) administered combinations of DHA-algal oil/DHA-fish oil at concentrations of 0/2000 (DHA-fish oil group), 500/1500 (500 mg/kg group), 1000/1000 (1000 mg/kg group), and 2000/ 0 mg/kg body weight/day (2000 mg/kg DHA-algal oil group). These dosing regimens provided daily doses of 540, 630, 720, and 900 mg DHA/kg. No treatment-related effects were observed in clinical observations, food and water consumption, mortality, gross pathology, and histopathology. Following treatment, a tendency toward increased body weights and increased absolute and relative liver weights was noted in males treated with DHA-fish oil compared to controls. Increased body weights and liver weights in both oil-treated groups were attributed to the large lipid load and were not regarded as toxicologically significant. No treatment-related differences in the measured parameters between the DHA-algal oil and fish oil groups were detected. The NOAEL for both fish-DHA oil and algal-DHA oil was considered as 2000 mg/kg/day, the highest dose tested. The 2000 mg/kg dose of fish-DHA oil corresponds to approximately 540 mg DHA/kg body weight/day.

2.4.2. Dog Studies

Groups of pure-bred beagle dogs (5/sex/group) were administered *via* gavage BCPS at dosage levels of 0 (vehicle control; corn oil), 10, 100, and 1000 mg/kg/day for 26 weeks (Heywood et al., 1987). No deaths were recorded in any of the groups during the treatment. During the first eight weeks of the study, animals in the high-dose group consumed less feed. Following week 9, animals were given moistened feed and the intake was matched with the control group. Compared to controls, animals receiving the highest dose showed a decrease in body weight gain. In dogs receiving 1000 mg/kg/day dose, blood glucose and cholesterol levels were significantly decreased. No treatment-related macroscopic findings or changes in organ weights were noted. Histological examinations of tissues did not reveal any treatment-related changes. The results of this study indicate that NOAEL is lower than 1000 mg/kg/day but is closer to this dose, the highest dose tested.

In another study, groups of pure-bred beagle dogs (4/sex/group) were administered BCPS at dosage levels of 0 (vehicle control), 5, 10, and 15 mg/kg/day *via* intramuscular injection for 13 weeks (Heywood et al., 1987). BCPS administration did not affect body weight gain. In animals receiving both the 10 and 15 mg/kg/day doses, pain reaction during the dosing procedure and subcutaneous hardening of injection site were noted. Hematological examinations revealed elevations in erythrocyte sedimentation rates and total white blood cell counts in animal receiving the highest dose of BCPS. Morphological examinations at termination did not reveal any changes, except for injection site changes in all groups. As the test material was administered by intramuscular injection rather than by oral dosing, these results have limited value for assessing safety of dietary exposure.

In a blinded cross-over study, Arujo et al. (2008) investigated the effects of a supplement containing phosphatidylserine, *Ginkgo biloba,* vitamin E, and pyridoxine on cognitive function in aged beagles. Four male and 5 female beagles, ranging in age from 7 to 12.7 years (mean = 8.2 years). Dogs were divided into 2 groups, based primarily on delayed-non-matching-to-position (DNMP) performance. For the 1st phase, one group received the supplement in a single meatball (n=5), the other group (n=4) served as control and received a single meatball without the supplement for over 60-day wash-in and for an additional 10 day on which they were tested on the DNMP, for a total of 70 day on their respective treatment conditions. For the 2nd phase, the groups were reversed. A single capsule containing contained 25 mg of phosphatidylserine, 50 mg of *Ginkgo biloba* extract, 20.5 mg of pyridoxine HCl, and 33.5 mg of vitamin E was administered per 5 kg of body weight once daily. Control subjects received only a single meatball. The results of DNMP performance was significantly improved in supplemented dogs. No adverse effects were reported.

2.5. Teratogenicity/Reproduction Studies

Heywood et al. (1987) also investigated the potential teratogenic effects of BCPS following daily gavage administration to pregnant Sprague-Dawley rats at daily doses of 0, 10, 100, and 200 mg/kg/day from Days 6 to 15 of gestation. Based on the results summarized for this study, the highest dose described in the methods section should be 1000 mg/kg/day, instead of 200 mg/kg/day. On Day 20, animals were euthanized, litter values were determined, and fetuses were examined for skeletal and visceral malformations. The only sign of toxicity noted in rats receiving 1000 mg BCPS/kg/day was a slight increase in salivation. BCPS treatment did not affect mean body weight gain during gestation. At terminal autopsy, no treatment-related macroscopic changes were noted. BCPS treatment did not affect litter values as assessed by litter size, post-implantation loss, litter and mean fetal weights, and the embryonic and fetal development. The NOAEL for this study was 1000 mg/kg body weight /day, the highest dose tested.

In another study, BCPS was administered *via* gavage to pregnant New Zealand white rabbits at daily doses of 0, 50, 150, and 450 mg/kg body weight/day from Days 6 to 18 of pregnancy (Heywood et al., 1987). At the highest dose level, BCPS administration resulted in a decrease in body weight gain during the first four days of dosing. On Day 29 of pregnancy, animals were euthanized, litters were examined macroscopically, and fetuses were examined for skeletal and visceral abnormalities. Mean fetal weights at the highest dose were slightly lower but did not reach statistical significance. BCPS administration did not affect embryonic and fetal development. The NOAEL for this study was 450 mg/kg body weight /day, the highest dose tested.

Blum et al. (2007b) assessed the reproductive toxicity of DHA-algal oil in a one-generation study. Sprague-Dawley rats were provided diets containing DHA-algal oil at concentrations of 1.5, 3.0, or 7.5% (~700, 1500 and 3750 mg/kg/day, respectively), and the control group received a diet containing 7.5% corn oil. Males and females were treated for 10 weeks prior to mating and during mating. Females continued to receive test diets during gestation and lactation. DHA-algal oil treatment did not affect clinical observations, mortality, fertility, and reproductive performance in parental animals. Changes noted in feed consumption, body weight, and liver weights in the treated groups were not considered to be due to an adverse effect of DHA-algal oil. Spleen weight increases in treated animals were associated with extramedullary hematopoiesis. Yellow discoloration of abdominal adipose tissue was observed in rats from the high-dose group, and histological examination revealed steatitis in all treated parental groups. Exposure to DHA-algal oil did not influence the physical development of F_1 animals. DHA-algal oil at dietary concentrations of up to 7.5% in rats did not affect reproductive capacity or pup development. The NOAEL for this study was 7.5% (~3750 mg/kg body weight /day), the highest dose tested.

In summary, the results of teratogenicity studies in rats at doses up to 200 mg/kg/day and in rabbits at doses up to 450 mg/kg/day showed that oral administration of phosphatidylserine did not affect embryonic and fetal development. Similarly, DHA-rich algal oil at dietary levels up to 7.5% (~3750 mg/kg/day) did not affect reproductive capacity or pup development.

2.6. Genotoxicity

The mutagenic potential of BCPS was investigated in a human lymphocyte chromosomal damage assay, mouse-lymphoma cell mutation test, cultured human epithelial cell DNA repair assay and in an *in vivo* mouse micronucleus assay (Heywood et al., 1987). Cultures of human lymphocytes were exposed to BCPS at concentrations of 0, 17, 83 and 166 μg/ml with and without metabolic activation. Compared to the control, no significant increase in chromosomal damage was noted in either the presence or absence of metabolic activation. In the *in vitro* mouse lymphoma L5178Y mammalian cell test system, which detects mutations from the heterozygous condition for the thymidine kinase locus (TK+/-) to the thymidine kinase deficient genotype (TK-/-), exposure to BCPS did not reveal any significant increases in the number of mutant colonies or mutation frequency either in the presence or absence of metabolic activation. In the DNA repair assay with autoradiographic techniques, cultured human epithelioid cells did not reveal any evidence of DNA repair synthesis in the HELA S3 cells either in the presence or absence of metabolic activation.

In the micronucleus test, BCPS was administered *via* gavage to mice at total dosages of 30, 150 and 300 mg/kg in two equal doses separated by 24-hours. The positive control group was treated with mytomycin C. Six hours after the second dose, mice were killed and bone marrow smears were examined for the presence of micronuclei in 1000 polychromatic erythrocytes per mouse and for the ratio of normochromatic and polychromatic erythrocytes. The results of the study did not reveal any evidence of mutagenic potential or bone marrow toxicity.

Under the conditions of the above *in vitro* and *in vivo* studies, BCPS is devoid of any genotoxic or clastogenic activity.

2.7. Safety of EPA and DHA

Typically, EPA and DHA are contained in oily fish, such as salmon, lake trout, tuna and herring. The composition of EPA and DHA in krill-based phosphatidylserine, which is the subject of this notification ranges from 15.4 to 15.9% w/w and 7.3 to 8.9 % w/w, respectively. The average total of EPA+DHA in krill-based phosphatidylserine is 23%. The safety of DHA and EPA, the principal fatty acids of phosphatidylserine, has been extensively evaluated by the FDA in the 1997 final rule on the GRAS affirmed use of menhaden oil as a direct food ingredient and also regarding the use of omega-3 fatty acids as a dietary supplement in 2005. In 1997, menhaden oil was affirmed as GRAS by FDA as a direct human food ingredient with specific limitations of use to ensure that the total daily intake of EPA and DHA would not exceed 3 g/person/day (62 FR 30751; June 5, 1997; 21 CFR 184.1472). Because of concerns over possible adverse effects of fish oil consumption on bleeding coagulation time, glycemic control, and LDL cholesterol, FDA established maximum use levels of menhaden oil in certain foods (62 FR 30751 at 30757; June 5, 1997; amended March 23, 2005). FDA reaffirmed that the intake of DHA and EPA must not exceed 3.0 g/day from all fish oil sources and in doing so, FDA placed specific limitations, including the category of foods, the functional use of the ingredient, and the level of use, to ensure that the consumption of EPA and DHA would not exceed 3.0 g/day.

In addition, FDA has not objected to certain GRAS notifications for additional sources of EPA and DHA as food ingredients (fish oils other than menhaden oil) (GRAS Notification Nos: GRN 000102, GRN 000105, GRN 000109, GRN 000138; GRN 000146; GRN 000193; GRN 000200; GRN 000217). These GRAS Notifications proposed maximum use levels consistent with those specified in the final rule affirming as GRAS, menhaden oil as a direct human food ingredient with specific limitations of use. FDA has also responded without objection to a GRAS notification from Martek Biosciences Corporation for high DHA algal oil DHA. Martek estimated that the use of algal oil in a number of food categories at the maximum proposed use levels would result in a mean exposure of no more than 1.5 g DHA/day (GRAS Notice No. GRN 000137).

To ensure that the intended use of krill-based phosphatidylserine will not add to the existing intake of DHA and EPA, we compared the FDA tables (identified in the Final Rule on menhaden oil) of concentrations of menhaden oil in various foods consistent with safe levels of DHA and EPA from menhaden oil with krill-based phosphatidylserine (Table 8). For this calculation, we used the FDA's statement that menhaden oil contains 8% DHA and 14% EPA. The total of DHA+EPA (22%) in menhaden oil is essentially similar (23%) to that in krill-based phosphatidylserine. Similarly, the individual levels of DHA (8% vs 8%) and EPA (14% vs 15.4%) are also essentially similar between menhaden and krill-derived phopshatidylserine. In different FDA GRAS Notifications, the total amount of DHA+EPA ranged from 20 to 41% and was reported as follows: GRN 000105 = 38%, GRN 000109 = 28%, GRN 000138 = 29%, GRN 000146 = 20%, GRN 000200 = 41%, and GRN 000279 = 22%. In all of these notices, the maximum levels of use in food categories were adjusted such that the resulting intake of DHA+EPA was similar to or lower than what is currently permitted for menhaden oil under 21 CFR 184.1472.

Table 8. Comparison of proposed maximum use levels of krill-based phosphatidylserine with current menhaden oil permitted under 21 CFR 184.1472

Category of food (and 21CFR §170.3(n) paragraph)	Maximum level of use of menhaden oil under 21CFR184.1472 and FR Vol 70 No 55 March 23 2005 Final Rule	Intended maximum level of use of krill-based phosphatidylserine
Cereals	4.0%	0.09%*
Dairy product analogs	5.0%	0.09%
Milk products	5.0%	0.44%
Processed fruit juices	1.0%	0.42%

*Also includes nutritional bars at phosphatidylserine use level of 0.25%

In addition to the food categories presented in Table 8, the menhaden oil rule cited above permits, and subsequent GRAS notices include, several other applications where krill-based phosphatidylserine would not be used. It is important to note that the levels of phosphatidylserine (consequently the levels of DHA+EPA) are substantially (2 - 40 fold) lower than those allowed for menhaden oil. Additionally, krill-based phosphatidylserine will not be added to foods containing other significant sources of DHA or EPA as ingredients. One of the food categories, breakfast bars, intended for application of krill-based phosphatidylserine is not the subject of the menhaden oil regulation. However, this food category would generally substitute for breakfast cereals, which are allowed under the menhaden oil rule. Based on reference amounts customarily consumed (21CFR 101.12) for breakfast cereal (15-55 g), the allowed concentration of menhaden oil in this category (4%) would result in an intake of 0.6 to 2.2 g menhaden oil. The intended use of krill-based phosphatidylserine (containing EPA+DHA) at maximum use levels of 0.25% in breakfast bars would simply substitute for breakfast cereals. The FDA permitted use levels of menhaden oil in breakfast cereals (4%) is approximately 16-fold higher than the intended use of krill-based phosphatidylserine in breakfast bars (0.25%). In addition, based on the consumption estimates, the estimated 90th percentile intake of EPA+DHA from the intended uses of krill-derived phosphatidylserine is determined as 33 mg/person/day, which is only about ~1% of the total FDA recommended maximum daily intake of EPA and DHA of 3000 mg/person/day for menhaden oil.

In summary, the intended uses of krill-based phosphatidylserine in specified foods identified in this document would result in far less DHA+EPA consumption than is currently authorized from the use of menhaden oil in these foods, as listed in 21 CFR 184.1472, or from other fish oils that are the subject of previous GRAS Notifications cited earlier. It is important to note that the intended uses of krill-based phosphatidylserine will not contribute to any increase in cumulative intake of DHA+EPA as krill-based phosphatidylserine would simply provide much less of these fatty acids than what has already been considered for these foods and concluded to be safe. While the intended use of krill-based phosphatidylserine may broaden the range of the selection of products with DHA+EPA, these would simply provide alternative products to those that are currently permitted and would not add to the current cumulative intake for users of such products.

2.8. Other Safety Considerations

As krill-based phosphatidylserine, the subject of this Notification, is derived from krill, it is important to characterize the nature and quantity of impurities/contaminants that might be stored in marine lipids that may pose a health hazard. As krill-based phosphatidylserine is derived from the lipid fraction of krill biomass, likely contaminants were analyzed from 2 representative batches. These results, presented in Appendix II, demonstrate the levels of contaminants are low and consistent with levels of other food ingredients. Similarly the levels of impurities generated during manufacturing/processing are minimal and consistent with levels reported for other food ingredients. In addition to contaminants, allergy to krill has been reported. Because krill are a species of crustacean shellfish, anyone with a crustacean shellfish allergy may be allergic to an ingredient derived from krill oil. Enzymotec will market krill-based phosphatidylserine in full compliance with the Food Allergen Labeling and Consumer Protection Act of 2004 (Title II of Public Law 108-282).

3. SUMMARY

Phosphatidylserine is a naturally-occurring structural component of virtually all biological membranes in plants and animals. It is essential to the functioning of all the cells of the body and is most concentrated in the brain. Phosphatidylserine plays an important role in several metabolic processes such as activation of cell-membrane bound enzymes and is involved in neuronal signaling. It is found naturally in small quantities in foods such as meats, eggs, soy products, certain legumes and milk. Enzymotec proposes to use a standardized krill lecithin-derived phosphatidylserine as a nutrient at levels up to 30 mg phosphatidylserine/serving in Breakfast Cereals, Dairy Product Analogs, Grain Products and Pastas, Milk Products, and Processed Fruits and Fruit Juices. The intended use of krill-derived phosphatidylserine will result in an estimated daily 90th percentile intake for "users only" of 79.14 mg/person (1.84 mg/kg/day). The resulting 90th percentile EPA+DHA intake is estimated as 33 mg/person/day. In addition to the above categories, phosphatidylserine is also intended for use in Medical Food at levels not to exceed 300 mg phosphatidylserine/person/day. Phosphatidylserine has been the subject of four GRAS Notices submitted to FDA for use as a nutrient. In each case, FDA responded that they had no questions on the proposed use and did not object to the respective GRAS determination.

Given the metabolic sequel of phosphatidylserine from different sources, there is no reason to believe that krill-based phosphatidylserine is metabolized differently or poses any different health hazards than phosphatidylserine derived from soy or bovine cortex, except, possibly, for differences in lipid content based on the marine origin. These fatty acids are typical components of the diet and are not anticipated to pose any risk at the levels consumed. Furthermore, the different fatty acid chains are unlikely to affect the overall oral toxicity, as the fatty acid portions of molecules are largely cleaved prior to absorption by mucosal cells. Among the fatty acids of krill-derived phosphatidylserine, there is a potential safety concern for DHA and EPA at high levels of intake. FDA has determined that daily intakes of EPA and DHA at doses up to 3000 mg/person/day from conventional food and dietary supplement sources are safe. Given the limited and substitutional (for substances with DHA and EPA) uses of krill-derived phosphatidylserine, the resulting intake of DHA and EPA (33 mg/person/day) is safe.

Multiple clinical studies (>35) with over 1500 subjects revealed that oral administration of phosphatidylserine at doses of 200 to 600 mg/day is without any adverse effect. In the majority of these studies, phosphatidylserine was derived from bovine cortex. Available scientific evidence indicates that phosphatidylserine derived from krill is toxicologically equivalent to phosphatidylserine naturally found in the diet or derived from bovine cortex, soy, or fish. Once inside the body, orally ingested phosphatidylserine is hydrolyzed in the intestine prior to its absorption. The absorbed phosphatidylserine is transported and rapidly converted into other endogenous constituents. In repeat-dose safety studies in rats and dogs, oral administration of phosphatidylserine at doses up to 1000 mg/kg/day for up to 6 months was without any significant toxicity. The adverse effects noted in animal safety studies at 1000 mg/kg/day were considered minor as these changes were small in magnitude, not supported by any histological damage, and lacked any dose-response correlation. In teratogenicity studies in rats and rabbits, phosphatidylserine did not affect embryonic or fetal development. Multiple genotoxicity studies show that phosphatidylserine did not reveal any genotoxic or clastogenic activity.

4. RISK ASSESSMENT

There is sufficient qualitative and quantitative scientific evidence, including human and animal data, to determine safety-in-use for krill-based phosphatidylserine. The safety data on phosphatidylserine includes over 35 human clinical trials, repeat-dose (6 month) studies in rats and dogs, teratogenicity/reproductive toxicity studies in rats and rabbits and *in vitro* and *in vivo* genotoxicity studies. Generally, ADIs are derived from a no-observed-adverse-effect level determined from animal studies with considerations of uncertainty factors to account for variability and uncertainties. The animal studies did not show significant toxicity at doses up to 1000 mg/kg/day. Several human clinical trials (>35) and historical use supports the safety of phosphatidylserine at even higher use levels than can be determined by available animal studies. The discussion presented in this dossier supports a safe intake level of 300 mg phosphatidylserine/day.

The clinical evidence of krill-based phosphatidylserine safety is supported by:

- Phosphatidylserine is commonly found in daily-consumed foods such as meat, fish, legume, etc.
- Phosphatidylserine is an endogenous substance found in human body.
- The bioavailability of the ingested phosphatidylserine is limited due to extensive hydrolysis in the intestine prior to absorption, and that the absorbed phosphatidylserine is transported and rapidly converted into other endogenous constituents.
- Given the metabolic sequel, the krill-derived phosphatidylserine and associated lipids would not pose any different health hazards than soy or bovine cortex-derived phosphatidylserine and lipids.
- The limited and substitutional intake of DHA and EPA from the intended uses of krill-derived phosphatidylserine is safe.
- In multiple (>35) human clinical studies, safety of phosphatidylserine was confirmed at doses of 300 mg/day.
- There is no evidence that consumption of phosphatidylserine either in foods or as a dietary supplement has a cumulative effect that would affect its safety.
- A variety of animal and *in vitro* studies corroborate the human clinical safety data.

Phosphatidylserine has been marketed as a dietary supplement for over 10 years without significant adverse effects (except gastrointestinal side effects such as nausea and indigestion) at high doses. The typical recommended doses of phosphatidylserine as a dietary supplement are 100 mg three times a day (300 mg/day). Additionally, recently FDA has agreed to exercise enforcement discretion with a Health Claim Petition[7] on phosphatidylserine. The petitioner in this submission demonstrated that soy-derived phosphatidylserine is safe at levels up to 500 mg/day[8]. In a notice submitted to the FDA, use of phosphatidylserine was determined as GRAS at an estimated 90th percentile intake of 240 mg/person/day. The intended use of krill-based phosphatidylserine will result in a daily estimated 90th percentile intake for "users only" of 79.14 mg phosphatidylserine/person (1.84 mg/kg body weight/day). The 90th percentile intake of phosphatidylserine is approximately 3-fold lower than the safe levels (300 mg/day) determined on the basis of available safety studies. The resulting intake of EPA+DHA from the intended uses is determined as 38 mg/person/day and is approximately 80-fold lower than the safe levels.

On the basis of scientific procedures[9] including knowledge from a history of exposure from natural sources, the consumption of phosphatidylserine as an added food ingredient is considered safe at levels up to 300 mg/day. The intended uses are compatible with current regulations, *i.e.,* phosphatidylserine is used in Breakfast Cereals, Dairy Product Analogs, Grain Products and Pastas, Milk Products, Processed Fruits and Fruit Juices, and Medical Foods and is produced according to current good manufacturing practices (cGMP).

[7] "Letter Regarding Dietary Supplement Health Claim for Phosphatidylserine and Cognitive Dysfunction and Dementia" (2003) Available at: http://www.cfsan.fda.gov/~dms/ds-ltr33.html

[8] Petition for Health Claim: Phosphatidylserine and Cognitive Dysfunction, Phosphatidylserine and Dementia (2002) Available at: http://www.fda.gov/ohrms/dockets/dailys/02/Sep02/091302/80027351.pdf

[9] 21 CFR §170.3 Definitions. (h) Scientific procedures include those human, animal, analytical, and other scientific studies, whether published or unpublished, appropriate to establish the safety of a substance.

5. CONCLUSION

In summary, based on the information provided above and the fact that the constituents of krill-based phosphatidylserine are commonly found in food, and because these lipids and phospholipids are essentially the same, and will be handled metabolically the same as those derived from other sources, it is concluded that scientific experts, generally, would recognize them to be as safe and as acceptable as bovine-, soy- or fish-derived phosphatidylserine and lipids. Further, we believe that there are no significant questions regarding the safety of krill-based phosphatidylserine that would appear to require additional safety studies, due to the prior consideration and acceptability by the Agency for plant- or fish-derived phosphatidylserine and other phospholipids, and fish oil-derived omega-3 fatty acids.

In light of the data and discussion presented above, Enzymotec Ltd. respectfully concludes that krill-based phosphatidylserine, meeting the specifications cited above, and when used as a nutrient [21 CFR 170.3(o)(20)] in Breakfast Cereals, Dairy Product Analogs, Grain Products and Pastas, Milk Products, Processed Fruits and Fruit Juices that do not contain other good sources of DHA or EPA, at maximum use levels of up to 30 mg phosphatidylserine/serving (reference amounts customarily consumed, 21 CFR 101.12) and when not otherwise precluded by Standards of Identity, and also in Medical Foods at levels not to exceed 300 mg phosphatidylserine/person/day is GRAS, as demonstrated through scientific procedures.

6. REFERENCES

Allegro, L., V. Favaretto, G. Ziliotto. 1987. Oral phosphatidylserine in elderly patients with cognitive deterioration. An open study. *Clinical Trials Journal* 24: 104–108.

Amaducci, L. and the SMID group. 1988. Phosphatidylserine in the treatment of Alzheimer's disease: Results of a multicenter study. *Psychopharmacol Bulletin* 24:130-134.

Araujo, J.A., Landsberg, G.M., Milgram, N.W., Miolo, A., 2008. Improvement of short-term memory performance in aged beagles by a nutraceutical supplement containing phosphatidylserine, *Ginkgo biloba*, vitamin E, and pyridoxine. Can. Vet. J. 49:379-385.

Blum, R., Kiy, T., Tanaka, S., Wong, A.W., Roberts, A., 2007a. Genotoxicity and subchronic toxicity studies of DHA-rich oil in rats. *Regulatory Toxicology Pharmacology* 49: 271-284.

Blum, R., Kiy, T., Waalkens-Berendsen, I., Wong, A.W., Roberts, A., 2007b. One-generation reproductive toxicity study of DHA-rich oil in rats. *Regulatory Toxicology Pharmacology* 49: 260-270.

Bruni, A., L. Mietto, F. Bellini, E. Boarato, G. Toffano, 1989. Pharmacological and autopharmacological action of phosphatidylserine. In Phospholipids in the Nervous System: Biochemical and Molecular Biology. Eds: Bazan, N.G., Horrocks, L.A. and Toffano, G. Fidia Research Series, Vol. 17 Livana Press, Padova, pp. 217-224.

Caffarra, P., V. Santamaria. 1987. The effects of phosphatidylserine in patients with mild cognitive decline. *Clinical Trials Journal* 24: 109 – 114.

Cenacchi, T., T. Bertoldin, C. Farina, M.G. Fiori, G. Crepaldi. 1993. Cognitive decline in the elderly: A double-blind, placebo-controlled multicenter study on the efficacy of phosphatidylserine administration. *Aging Clinical and Experimental Research* 5: 123-133.

Claro, F.T., Silva, R.H., Frussa-Filho, R. 1999. Bovine brain phosphatidylserine attenuates scopolamine-induced amnesia. *Physiology & Behavior* 67: 551-554.

Crook, T.H., Tinklenberg, J., Yesavage,J., Petrie,W., Nunzi, M.G., Massari, D.C., 1991. Effects of phosphatidylserine in age-associated memory impairment. *Neurology* 41: 644-649.

Crook, T.H., W. Petrie, C. Wells, D.C. Massari. 1992. Effects of phosphatidylserine in Alzheimer's Disease. *Psychopharmacol Bulletin* 28: 61-66.

Crook, T. H. 1998. Treatment of age-related cognitive decline: Effects of phosphatidylserine in anti-aging medical therapeutics, Vol II, edited by R.M. Klatz, Health Quest Publications, Chicago, pp. 20-29.

Degussa Food Ingredients GmbH. 2006. GRAS notification for phosphatidylserine. Information received from Food and Drug Administration under Freedom of Information Act (FOIA).

000036

Delwaide, P.J., A.M.Gyselynck-Mambourg, A. Hurlet, M. Ylieff. 1986. Double-blind randomized controlled study of phosphatidylserine in senile demented patients. *Acta Neurologica Scandinavica* 73: 136-140.

Engel, R.R., Satzger, W., Gunther, W., Kathmann, N., Bove, D., Gerke, S., Munch, U., Hippius, H., 1992. Double-blind cross-over study of phosphatidylserine vs. placebo in patients with early dementia of the Alzheimer type. *European Neuropsychopharmacol* 2: 149-155.

Fahey, T.D., M.S. Pearl. 1998. The hormonal and perceptive effects of phosphatidylserine administration during two weeks of weight training-induced over-training, *Biology of Sport* 15: 135-144.

Federal Register/62FR/ No.108/pp 30751-30757/ Thursday, June 5, 1997-21CFRPart 184 Substances Affirmed as Generally Recognized as Safe: Menhaden Oil. Final Rule.

Federal Register January 15, 2004 FR69 10 2313 - Substances Affirmed as Generally Recognized as Safe: Menhaden Oil http://www.cfsan.fda.gov/~lrd/fr040115.html

Federal Register - 70FR 14530 March 23, 2005: Substances Affirmed as Generally Recognized as Safe;menhaden oil http://www.cfsan.fda.gov/~lrd/fr050323.html

Funfgeld, E.W., M. Baggen, P. Nedwidek, B. Richstein, G. Mistlberger. 1989. Double-blind study with phosphatidylserine (PS) in Parkinsonian patients with senile dementia of Alzheimer's type (SDAT). *Alzheimer's Disease and Related Disorders*: 1235-1246.

Granata, Q., J. Di Michele. 1987. Phosphatidylserine in elderly patients. *Clinical Trials Journal* 24: 99-103.

GRN 000186. 2005. GRAS Notice Inventory. Agency response letter. http://www.accessdata.fda.gov/scripts/fcn/fcnNavigation.cfm?rpt=grasListing

GRN 000197. 2006. GRAS Notice Inventory. Agency response letter. http://www.accessdata.fda.gov/scripts/fcn/fcnNavigation.cfm?rpt=grasListing

GRN 000223. 2007. GRAS Notice Inventory. Agency response letter. http://www.accessdata.fda.gov/scripts/fcn/fcnNavigation.cfm?rpt=grasListing

GRN 000226. 2007. GRAS Notice Inventory. Agency response letter. http://www.accessdata.fda.gov/scripts/fcn/fcnNavigation.cfm?rpt=grasListing

GRN 000279. 2009. GRAS Notice Inventory. Agency response letter. http://www.accessdata.fda.gov/scripts/fcn/fcnNavigation.cfm?rpt=grasListing

Hamm, M., 2004. Nutritional- scientific statement on the change of nutritive provision with phosphatidylserine (PS). Article provided by Dr. Hamm. (Unpublished report cited in Degussa Food Ingredients GmbH. 2006).

Heiss, W.-D., J. Kessler, R. Mielke, B. Szelies, K. Herholz. 1994. Long-term effects of phosphatidylserine, pyritinol, and cognition training in Alzheimer's disease. *Dementia* 5: 88-98.

Hendler, S.S., D. Rorvik.2001. Phosphatidylserine. In: PDR for Nutritional Supplements. Medical Economics™ Thomson Healthcare, Montvale, NJ, pp. 354-356.

Hershkowitz, M., M. Fisher, D. Bobrov, M. Rabinowitz. 1989. Long-term treatment of dementia of Alzheimer type with phosphatidylserine: Effect on cognitive functioning and performance in daily life. In: Phospholipids in the Nervous System: Biochemical and Molecular Pathology. Eds. N.G. Bazan, L.A. Horrocks, G. Toffano (eds). Liviana Press, Padova; pp. 279-288.

Horrocks, L.A., M. Rollins, A.J. Yates. 1982. In: The Molecular Basis of Neuropathology. Eds. A.N. Davidson and R.H.S. Thompson. Edward Arnold Publishers, pp. 601-630.

Heywood, R., D.D. Cozens, M. Richold. 1987. Toxicology of a phosphatidylserine preparation from bovine brain (BC-PS). *Clinical Trials Journal* 24: 25-32.

Jorissen, B.L., F. Brouns, M.P.J. van Boxtel, R.W.H.M. Ponds. F.R.J. Verhey, J. Jolles, W.J. Riedel. 2001. The influence of soy-derived phosphatidylserine on cognition in age-associated memory impairment. *Nutritional Neuroscience* 4: 121-134.

Kidd, P.M., 2008. Alzheimer's disease, amnestic mild cognitive impairment, and age-associated memory impairment: current understanding and progress toward integrative prevention. *Alternative Medicine Review* 13: 85-115.

Kidd, P.M., 2007. Omega-3 DHA and EPA for cognition, behavior, and mood: clinical findings and structural-functional synergies with cell membrane phospholipids. *Alternative Medicine Review* 12: 207-227.

Kidd, P.M., 1998. Phosphatidylserine (PS): A Remarkable Cell Nutrient (3rd Edition). Monograph. Lucas Meyer, Inc., Decatur, Illinois, USA.

Maggioni, M., G.B. Picotti, G.P. Bondiolotti, A. Panerai, T. Cenacchi, P. Nobile, F. Brambilla. 1990. Effects of phosphatidylserine therapy in geriatric patients with depressive disorders. *Acta Psychiatrica Scandinavica* 81: 265-270.

Mazzari, S., A. Zanoti, P. Orlando. 1982. Pharmacokinetics and pharmacodynamics of phosphatidylserine liposomes in mice. In: Transport in biomembranes: Model systems and reconstructions. Eds. Antolini et al. New York, Raven Press, pp. 257-263.

Monteleone, P., M. Maj, L. Beinat, M. Natale, D. Kemali. 1992. Blunting by chronic phosphatidylserine administration of the stress-induced activation of the hypothalamo-pituitary-adrenal axis in healthy men. *European Journal of Clinical Pharmacology* 41:385–388.

Neuringer, M., Anderson, G.J., Connor, W.E., 1988. The essentiality of n-3 fatty acids for the development and function of the retina and brain. Annu. Rev. Nutr. 8, 517-541.

Palatini, P., G. Viola, E. Bigon, A.M. Menegus, A. Bruni. 1991. Pharmacokinetic characterization of phosphatidylserine liposomes in the rat. *British Journal of Pharmacology* 102:345-350.

Palmieri, G., R. Palmieri, M.R. Inzoll, A.M. Agrati, A Vargiu. 1987. Double-blind controlled trial of phosphatidylserine in patients with senile mental deterioration. *Clinical Trials Journal* 24: 73-83.

Pepping, J. 1999. Phosphatidylserine. *American Journal of Health-System Pharmacy* 56: 2043-2044.

Puca, F.M., M.A. Bavarese, M.G. Minervini. 1987. Exploratory trial of phosphatidylserine efficacy in mildly demented patients. *Clinical Trials Journal* 24: 94-98.

Rabboni, M., F.S. Maggioni, A. Giannelli, L. Beinat. 1990. Neuroendocrine and behavioral effects of phosphatidylserine in elderly patients with abiotrophic or vascular dementia or mild depression. *Clinical Trials Journal* 27: 230-240.

Ransmayr, G., S. Ploerer, F. Gerstenbrand, G. Bauer. 1987. Double-blind placebo-controlled trial of phosphatidylserine in elderly patients with arteriosclerotic encephalopaty. *Clinical Trials Journal* 24; 62-72.

Salem, N, Jr., Serpentino, P., Puskin, J.S., Abood, L.G. 1980. Preparation and spectroscopic characterization of molecular species of brain phosphatidylserines. *Chemistry and Physics of Lipids* 27: 289-304.

Schreiber, S., O. Kampf-Sherf, M. Gorfine, D. Kelly, Y. Oppenheim, B. Lerer. 2000. An open trial of plant-source derived Phosphatidylserine for the treatment of age-related cognitive decline. *Israel Journal of Psychiatry and Related Sciences* 37: 302-307.

Sinforiani, E., C. Agostinis, P. Merlo, S. Gualtieri, M. Mauri, A. Mancuso. 1987. Cognitive decline in ageing brain. Therapeutic approach with phosphatidylserine. *Clinical Trials Journal* 24: 115-124.

Toffano, G., A. Battistella, P. Orlando. 1987. Pharmacokinetics of radiolabeled brain phosphatidylserine. *Clinical Trials Journal* 24:18-24.

Tso, P. 1994. Intestinal lipid absorption. In L. R. Johnson (ed), *Physiology of the Gastrointestinal Tract.*, Vol. 56 (L. R. Johnson, ed), Raven Press, New York, pp. 1867–1907.

Villardita, J.C., S. Grioli, G. Salmeri, F. Nicoletti, G. Pennisi. 1987. Multicenter clinical trial of brain phosphatidylserine in elderly patients with intellectual deterioration, *Clinical Trials Journal* 24: 84-93.

Wise, E. M., Jr. and D. Elwyn. 1965. Rates of reactions involved in phosphatide synthesis in liver and small intestine of intact rats. *Journal of Biological Chemistry* 240: 1537-1548.

Yabuuchi, H. and O'Brien, J.S. 1968. Positional distribution of fatty acids in glycerophosphatides of bovine gray matter. *Journal of Lipid Research* 9: 65-67.

Vaisman, N., Kaysar, N., Zaruk-Adasha, Y., Pelled, D., Brichon, G., Zwingelstein, G., Bodennec, J., 2008. Correlation between changes in blood fatty acid composition and visual sustained attention performance in children with inattention: effect of dietary n-3 fatty acids containing phospholipids. *American Journal of Clinical Nutrition* 87: 1170-1180.

7. APPENDIX I

Analytical data from different manufacturing lots

Compositional analysis of krill-based phosphatidylserine from three different batches (Enzymotec, 2009)

Composition (%w/w)	**Batch PKM-1**	**Batch PKM-2**	**Batch PKM-3**
Phosphatidylserine	60.1	60.5	51.3
Other phospholipids	21.0	19.3	28.0
Volatiles	2.3	1.0	2.0
Free serine	0.13	0.36	0.27
Free Cl^-	0.02	0.03	0.02
Free Ca^{2+}	0.02	0.01	0.02
Free Na^+	0.02	0.04	0.01
DHA and EPA content (%w/w)			
DHA	7.3	8.2	8.9
EPA	15.4	15.9	15.9
DHA+EPA	22.7	24.1	24.8
Quality parameters/ Heavy metals			
Peroxide value (meq/Kg)	<0.5	<0.5	<0.5
Lead (ppm)	<0.20	<0.20	<0.20
Inorganic arsenic (ppm)		0.01	
Cadmium (ppm)	<0.03	<0.03	<0.01
Mercury (ppm)	<0.20	<0.20	<0.20
Residual solvents(ppm)			
Ethanol	92.0	85.7	100.0
Hexane	1.6	3.8	2.1
Ethyl acetate	<1.0	<1.0	<1.0
Microbiological			
Total Count (cfu/1 g)	<10	<10	<10
Molds Count (cfu/1 g)	<10	<10	<10
Yeasts Count (cfu/1 g)	<10	<10	<10
Coliforms Count (cfu/1 g)	negative	negative	negative
Salmonella Detection (cfu/20 g)	negative	negative	negative
Staphylococcus aureus (coa.+) (cfu/1 g)	negative	negative	negative

Phospholipids profile (as % of total phospholipids) of krill-based phosphatidylserine

Phospholipids	Typical value	PKM-1	PKM-2	PKM-3
Phosphatidylserine	70	74.1	75.8	64.7
Lysophosphatidylserine	1	1.4	0.9	0.9
Phosphatidylcholine	1	1.6	0.0	1.6
Phosphatidylinsitol	1.7	1.5	1.7	2.0
Phosphatidylethanolamine	7	7.4	9.0	5.3
Phosphatidic acid	7	5.0	4.8	12.6
Other phospholipids	9	8.9	7.8	12.9
Total		**100**	**100**	**100**

Fatty acid profile of Krill based phosphatidylserine

Fatty acid (As percent of total fatty acids on PS)	Typical value	PKM-1	PKM-2	PKM-3
C14 (Myristic)	2.0	2.2	2.1	2.2
C16 (Palmitic)	23.5	24.2	23.6	23.0
C16:1n7 (Palmitoleic)	1.8	1.9	1.7	1.9
C18 (Stearic)	1.0	1.0	0.9	1.2
C18:1n9 (Oleic)	6.5	6.2	6.9	5.8
C18:1 (Octadecenoic)	5.5	4.9	5.5	5.8
C18:2n6 (Linoleic)	2.0	2.4	1.7	2.3
C18:3n6 (gamma-Linolenic)	0.2	0.2	0.2	0.2
C18:3n3 (alpha-Linolenic)	1.2	1.4	1.1	1.1
C18:4n3 (Octadecatetraenoic)	1.9	2.4	1.9	1.6
C20:1n9 (Eicosenoic)	0.6	0.5	0.6	0.6
C20:4n6 (Arachidonic)	0.7	0.7	0.6	0.9
C20:4n3 (Eicosatetraenoic)	0.6	0.6	0.5	0.7
C20:5n3 (Eicosapentaenoic)	31.0	30.2	32.5	30.4
C22:1n11(Cetoleic)	0.3	0.4	0.2	0.3
C22:1n9 (Erucic)	1.0	0.2	1.4	1.4
C22:5n3 (Docosapentaenoic)	0.7	0.7	0.6	0.8
C22:6n3 (Docosahexaenoic)	14	13.1	14.4	14.7
C24:1n9 (Nervonic)	0.3	0.4	0.2	0.4
Others	5.0	6.4	3.5	4.6
Total		**100.0**	**100.0**	**100.0**

7.1. APPENDIX II

Chemical contaminants	Units	Mix of PKM-1, 2, 3 in ratio 1:1:1
Total PCBs	pptWHO TEQ	<1.39
Pesticides	ppb	None detected
Dioxins and furans WHO TEQ with DLs	pptWHO TEQ	0.407

7.2. APPENDIX III

ESTIMATED DAILY INTAKE OF PHOSPHATIDYLSERINE BY THE U.S. POPULATION FROM PROPOSED FOOD-USES

CANTOX
HEALTH SCIENCES INTERNATIONAL

ESTIMATED DAILY INTAKE OF PHOSPHATIDYLSERINE BY THE U.S. POPULATION FROM PROPOSED FOOD-USES

Prepared for: EAS Consulting
1940 Duke Street, Suite 200
Alexandria, VA 22314

Prepared by: Cantox Health Sciences International
2233 Argentia Road, Suite 308
Mississauga, Ontario, Canada
L5N 2X7

October 14, 2008

Mississauga, ON, CANADA 905-542-2900

Bridgewater, NJ, USA 908-429-9202

Fleet, Hampshire, UK +44 (0) 1252 400 505

Shinjuku, Tokyo JAPAN 81-3-5287-3522

CANTOX
HEALTH SCIENCES INTERNATIONAL

ESTIMATED DAILY INTAKE OF PHOSPHATIDYLSERINE BY THE U.S. POPULATION FROM PROPOSED FOOD-USES

Table of Contents

List of Appendices

List of Tables

ESTIMATED DAILY INTAKE OF PHOSPHATIDYLSERINE BY THE U.S. POPULATION FROM PROPOSED FOOD-USES

1.0 INTRODUCTION

Cantox Health Sciences International (Cantox) has completed an assessment of the consumption of phosphatidylserine by the U.S. population in breakfast cereals (instant and regular hot cereals and ready-to-eat cereals), dairy product analogs (imitation milk and soy milk), grain products and pastas [nutritional bars (breakfast, granola, protein)], milk products [flavored milk and milk drinks, fluid milk (regular), powdered milk, milk-based meal replacements, and yogurt], and processed fruits and fruit juices (fruit flavored drink).

Estimates for the intake of phosphatidylserine were based on the proposed food-uses and use-levels in conjunction with food consumption data included in the United States Department of Agriculture's (USDA) 1994-1996 Continuing Survey of Food Intakes by Individuals (CSFII 1994-1996) and the 1998 Supplemental Children's Survey (CSFII 1998) (USDA, 2000). Calculations for the mean and 90th percentile all-person and all-user intakes, and percent consuming were performed for each of the individual food-uses of phosphatidylserine. Similar calculations were used to determine the estimated total intake of phosphatidylserine from all proposed food-uses combined. In both cases, the per person and per kilogram body weight intakes were reported for the following population groups:

infants, ages 0 to 2;
children, ages 3 to 11;
female teenagers, ages 12 to 19;
male teenagers, ages 12 to 19;
female adults, ages 20 and up;
male adults, ages 20 and up; and
total population (all population and gender groups combined).

2.0 FOOD CONSUMPTION SURVEY DATA

2.1 Survey Description

Nationwide dietary intake data for the years 2001-2002 are now available for public use; however, only Day 1 interview data are included in the present release. It is well established that the length of a dietary survey affects the estimated consumption of individual users and that short-term surveys, such as the typical 1-day dietary survey, overestimate consumption over longer time periods (Anderson, 1988). Because two 24-hour dietary recalls administered on 2 non-consecutive days (Day 1 and Day 2) are available from the CSFII 1994-1996, 1998 surveys, these data were used to generate estimates for the current intake analysis.

USDA CSFII 1994-1996 provides food consumption data on persons of all ages, whereas, CSFII 1998 is limited to children from birth through 9 years of age. Combined, these surveys provide the most appropriate data for evaluating food-use and food consumption patterns in the United States, containing 4 years of data on individuals selected *via* stratified, multistage area probability sampling of American households within all 50 states.

CSFII 1994-1996, 1998 survey data were collected from individuals and households *via* 24-hour dietary recalls administered on 2 non-consecutive days (Day 1 and Day 2) throughout all 4 seasons of the year. Data were collected in-person, a minimum of 3 days apart, on different days of the week, to achieve the desired degree of statistical independence. CSFII 1994-1996 contains 2-day dietary food consumption data for more than 15,000 individuals of all ages, and 1-day data for 16,103 individuals. CSFII 1998 contributes data from an additional 5,559 children, birth through 9 years of age, to data reported for 4,253 children of the same ages within CSFII 1994-1996. The overall CSFII 1994-1996, 1998 response rate for individuals selected for participation in the survey was 81.5 and 77.5% for Day 1 and Day 2, respectively.

In addition to collecting information on the types and quantities of foods being consumed, CSFII 1994-1996, 1998 collected physiological and demographic information from individual participants in the survey, such as sex, age, self-reported height and weight, and other variables useful in characterizing consumption. The inclusion of this information allows for further assessment of food intake based on consumption by specific population groups of interest within the total population. USDA sample weights were developed and incorporated with CSFII 1994-1996, 1998 to compensate for the potential under-representation of intakes from specific population groups as a result of sample variability due to survey design, differential non-response rates, or other factors, such as deficiencies in the sampling frame (USDA, 2000).

2.2 Statistical Methods

Consumption data from individual dietary records, detailing food items ingested by each survey participant on each of the 2 survey days, were collated by computer and used to generate estimates for the intake of phosphatidylserine from all proposed food-uses by the U.S. population. Estimates for the daily intake of phosphatidylserine from all proposed food-uses represent projected 2-day averages for each individual from Day 1 and Day 2 of CSFII 1994-96, 1998 data. These average amounts comprised the distribution from which mean and percentile intake estimates were produced. Mean and percentile estimates were generated using ratio estimation and non-parametric techniques, respectively, incorporating USDA survey weights in order to provide representative intakes for the entire U.S. population. All-person intake refers to the estimated intake of phosphatidylserine averaged over all individuals surveyed, regardless of whether they consumed food products containing phosphatidylserine, and therefore includes "zero" consumers (those who reported no intake of phosphatidylserine during the 2 survey days). All-user intake refers to the estimated intake of phosphatidylserine by those individuals consuming food products containing phosphatidylserine, hence the 'all-user' designation. Individuals were considered users if they consumed 1 or more food products containing phosphatidylserine on either Day 1 or Day 2 of the survey.

2.3 Statistical Reliability

Mean or percentile intake estimates based on small sample sizes or with high variability relative to the mean [assessed using the coefficient of variation (CV)] may be less statistically reliable than estimates based on adequate sample sizes or low variability relative to the mean (LSRO, 1995). Data presented herein for the estimated daily intake of phosphatidylserine follow the guidelines proposed by the Human Nutrition Information Service/National Center for Health Statistics Analytic Working Group for evaluating the reliability of statistical estimates adopted in the "Third Report on Nutrition Monitoring in the United States", whereby an estimated mean may be unreliable if the CV is equal to or greater than 30% (LSRO, 1995). The CV is the ratio of the estimated standard error of the mean to the estimated mean, expressed as a percentage (LSRO, 1995). Therefore, for the estimated intakes of phosphatidylserine presented herein, values were considered statistically unreliable if the CV was equal to or greater than 30%. These values were not considered when assessing the relative contribution of specific food-uses to total phosphatidylserine consumption and are marked with an asterisk.

3.0 FOOD USAGE DATA

The individual proposed food-uses and use-levels for phosphatidylserine employed in the current intake analysis are summarized in Table 3-1. Food codes were chosen by the client from the CSFII 1994-1996, 1998 (USDA, 2000) and grouped in food-use categories according to Title 21, Section §170.3 of the *Code of Federal Regulations* (CFR, 2007a). All food codes included in the current intake assessment are listed in Appendix C.

Table 3.1 Summary of the Individual Proposed Food-Uses and Use-Levels for Phosphatidylserine in the United States

Food Category	Proposed Food-Use	Use-Level (mg/RACC)	RACC[2] (g or mL)	Use-Level (%)
Breakfast Cereals	Instant and Regular Hot Cereals	30	240	0.0125
	Ready-to-Eat Cereals[1]	30	15 30 55	0.2 0.1 0.0545
Dairy Product Analogs	Imitation and Soy Milk[1]	30	240	0.0125
Grain Products and Pastas	Nutritional Bars (Breakfast, Granola, Protein)	30	40	0.075
Milk Products	Flavored Milk and Milk Drinks[1]	30	240	0.0125
	Milk, Fluid (Regular)[1]	30	240	0.0125
	Milk, Powdered[1]	30	240 (prepared)	0.0125
	Milk-Based Meal Replacements	30	240	0.0125
	Yogurt[1]	30	225	0.0133
Processed Fruits and Fruit Juices	Fruit Flavored Drink[1]	30	240	0.0125

[1] A limited number of food codes within each food-use were selected (See Appendix C).
[2] RACC – Reference Amounts Customarily Consumed Per Eating Occasion (21 CFR §101.12) (CFR, 2007b). When a range of values is reported for a proposed food-use, particular foods within that food-use may differ with respect to their RACC.

4.0 FOOD SURVEY RESULTS

Estimates for the total daily intakes of phosphatidylserine from all proposed food-uses are provided in Tables 4.1-1 and 4.1-2. Estimates for the daily intake of phosphatidylserine from individual food-uses in the U.S. are summarized in Tables A-1 to A-7 and B-1 to B-7 of Appendices A and B, respectively. Tables A-1 to A-7 provide estimates for the daily intake of phosphatidylserine per person (mg/person/day), whereas Tables B-1 to B-7 provide estimates for the daily intake of phosphatidylserine on a per kilogram body weight basis (mg/kg body weight/day).

4.1 Estimated Daily Intake of Phosphatidylserine from All Proposed Food-Uses

The estimated total intake of phosphatidylserine from all proposed food-uses in the U.S. by population group is summarized in Table 4.1-1. Table 4.1-2 presents this data on a per kilogram body weight basis.

Approximately 60.7% of the total U.S. population was identified as consumers of phosphatidylserine (12,503 actual users identified). Consumption of these types of foods by the total U.S. population resulted in estimated mean all-person and all-user intakes of phosphatidylserine of 22.0 mg/person/day (0.5 mg/kg body weight/day) and 39.4 mg/person/day (0.8 mg/kg body weight/day), respectively (Tables 4.1-1 and 4.1-2). The 90th percentile all-person and all-user intakes of phosphatidylserine from all proposed food-uses by the total population were 61.1 mg/person/day (1.3 mg/kg body weight/day) and 79.1 mg/person/day (1.8 mg/kg body weight/day), respectively.

On an individual population basis, the greatest mean all-person and all-user intakes of phosphatidylserine on an absolute basis were determined in male teenagers, at 30.8 mg/person/day (0.5 mg/kg body weight/day), and 55.4 mg/person/day (0.9 mg/kg body weight/day), respectively. Children encompassed the greatest percentage of users of any population group at 80.7%. Infants had the lowest intakes of phosphatidylserine on an absolute basis, with all-person and all-user mean intakes of 13.6 and 22.7 mg/person/day, respectively. On a body weight basis, mean all-person intake of phosphatidylserine was highest in children, at 1.2 mg/kg body weight/day, and the mean all-user intake was highest in infants, at 1.8 mg/kg body weight/day. The lowest mean all-person on a per kilogram body weight basis were observed in male and female adults (0.3 mg/kg body weight/day), and lowest mean all-user intakes were observed in female adults (0.5 mg/kg body weight/day) (Table 4.2-2).

Table 4.1-1 Summary of the Estimated Daily Intake of Phosphatidylserine from All Proposed Food Categories in the U.S. by Population Group (1994-1996, 1998 USDA CSFII Data)

Population Group	Age Group (Years)	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
				Mean (mg)	90th Percentile (mg)	Mean (mg)	90th Percentile (mg)
Infant	0-2	53.1	1,902	13.6	36.4	22.7	45.5
Child	3-11	80.7	5,090	30.2	68.2	37.9	74.3
Female Teenager	12-19	54.8	385	22.5	61.2	40.2	75.0
Male Teenager	12-19	55.5	386	30.8	85.6	55.4	109.6
Female Adult	20 and Up	54.0	2,471	19.3	53.8	35.3	71.8
Male Adult	20 and Up	47.8	2,269	21.1	63.3	45.2	91.4
Total Population	All Ages	60.7	12,503	22.0	61.1	39.4	79.1

Table 4.1-2 Summary of the Estimated Daily Per Kilogram Body Weight Intake of Phosphatidylserine from All Proposed Food Categories in the U.S. by Population Group (1994-1996, 1998 USDA CSFII Data)

Population Group	Age Group (Years)	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
				Mean (mg/kg)	90th Percentile (mg/kg)	Mean (mg/kg)	90th Percentile (mg/kg)
Infant	0-2	53.1	1,902	1.1	2.9	1.8	3.5
Child	3-11	80.7	5,090	1.2	2.8	1.5	3.0
Female Teenager	12-19	54.8	385	0.4	1.1	0.7	1.4
Male Teenager	12-19	55.5	386	0.5	1.5	0.9	1.7
Female Adult	20 and Up	54.0	2,471	0.3	0.9	0.5	1.1
Male Adult	20 and Up	47.8	2,269	0.3	0.8	0.6	1.1
Total Population	All Ages	60.7	12,503	0.5	1.3	0.8	1.8

When heavy consumers (90^{th} percentile) were assessed, all-person and all-user intakes of phosphatidylserine from all proposed food-uses on an absolute basis also were determined to be greatest in male teenagers (85.6 and 109.6 mg/person/day, respectively). The lowest 90^{th} percentile all-person and all-users intakes of phosphatidylserine on an absolute basis were in infants (36.4 and 45.5 mg/person/day, respectively) (Table 4.1-1). On a body weight basis, infants were determined to have the greatest all-person 90^{th} percentile intakes (2.9 mg/kg body weight/day) and the greatest all-user 90^{th} percentile intakes of phosphatidylserine (3.5 mg/kg body weight/day) (Table 4.1-2). The lowest all-person and all-user 90^{th} percentile intakes of phosphatidylserine on a body weight basis were observed in male adults (0.8 mg/kg body weight/day), and male and female adults (1.1 mg/kg body weight/day) respectively.

4.1.1 All-Person Intakes

Estimates for the mean and 90^{th} percentile daily intakes of phosphatidylserine from each individual food category are summarized in Tables A-1 to A-7 and B-1 to B-7 on a mg/day and mg/kg body weight/day basis, respectively. Tables A-7 and B-7 summarize the estimates for the mean all-person intakes of phosphatidylserine by the total population (all ages) from each of the individual food-uses on a mg/person/day and mg/kg body weight/day basis, respectively. The total U.S. population was identified as being significant consumers of ready-to-eat cereals (43.5% users) and fluid milk products (13.7% users).

Consumption of ready-to-eat cereals made the highest mean and 90^{th} percentile all-person intakes of phosphatidylserine, at 11.9 mg/person/day (0.3 mg/kg body weight/day) and 39.9 mg/person/day (0.8 mg/kg body weight/day). On a body weight basis, the highest mean and 90^{th} percentile all-person intakes were 0.3 and 0.8 mg/kg body weight/day, respectively, for ready-to-eat cereals. Imitation and soy milk, powdered milk, milk-based meal replacements, and fruit flavored drinks had a negligible impact on the all-person intakes of phosphatidylserine.

Of the individual population groups, the consumption of ready-to-eat cereals made the most significant contribution to the mean all-person intakes of phosphatidylserine (Tables A-1 to A-6 and Tables B-1 to B-6). The consumption of ready-to-eat cereals also made a significant contribution to the 90th percentile intakes of phosphatidylserine in each population group. The highest mean all-person intakes of phosphatidylserine, on an absolute basis, were reported in male teenagers consuming ready-to-eat cereals, at 20.1 mg/person/day (0.3 mg/kg body weight/day), and the highest 90th percentile all-person intakes also were reported in male teenagers consuming ready-to-eat cereals, at 60.0 mg/person/day (1.0 mg/kg body weight/day). On a body weight basis, consumption of ready-to-eat cereals in children led to the highest mean and 90th percentile all-person intakes of phosphatidylserine, with values of 0.8 and 2.0 mg/kg body weight/day, respectively. The lowest mean all-person intakes of phosphatidylserine across the various individual population groups were identified consistently for powdered milk or soy and imitation milk.

4.1.2 All-User Intakes

Tables A-7 and B-7 also summarize the estimates for the mean all-user intakes of phosphatidylserine by the total population (all ages) from each of the individual food-uses on a mg/person/day and mg/kg body weight/day basis, respectively. The contribution of a food category is based on the estimated intake of phosphatidylserine resulting from the consumption of the food, as well as the percentage of users identified as consumers of the food. For example, for the total population, the consumption of milk-based meal replacements resulted in an estimated mean all-user phosphatidylserine intake of 53.0 mg/person/day; however, only 175 users (0.8% of the total population) of this food-use were identified and therefore, the contribution of milk-based meal replacements did not have a great impact on phosphatidylserine intake in comparison to ready-to-eat cereals. When the number of users of each individual food-use is taken into account, consumers of ready-to-eat cereals were identified as having the greatest contribution to the mean and 90th percentile all-user intakes of phosphatidylserine at 32.0 and 60.0 mg/person/day (0.7 and 1.5 mg/kg body weight/day), respectively. Of the other food categories with a significant number of users of phosphatidylserine in the total population, consumption of fluid milk, instant and regular hot cereals, yogurt, and nutritional bars also made significant contributions to the estimates for the mean (28.6, 19.1, 15.1, and 20.4 mg/person/ day, respectively) and 90th percentile (61.3, 33.1, 28.5, and 34.0 mg/person/day, respectively). The lowest contribution to the all-user mean and 90th percentile intakes of phosphatidylserine were observed for powdered milk, at 11.7 and 36.2 mg/person/day (0.2 and 0.7 mg/kg body weight/day), respectively.

On an individual population group basis, the consumption of ready-to-eat cereals made the most significant contribution to the all-user intakes of phosphatidylserine (Tables A-1 to A-6 and

Tables B-1 to B-6). On an absolute basis children consuming ready-to-eat were determined to have the highest reliable mean and 90th percentile all-user intakes of phosphatidylserine of 31.3 and 57.0 mg/person/day, respectively. On a per kilogram body weight basis, infants consuming milk-based meal replacements experienced the highest reliable mean and 90th percentile all-user intakes of phosphatidylserine of 4.99 and 17.9 mg/kg body weight/day, respectively.

The estimated intakes of phosphatidylserine were considered statistically unreliable if the CV was equal to or greater than 30%. Assessing the CV for all-user intake estimates found the intake for milk-based meal replacements and fruit flavored drinks to be statistically unreliable in the infant and female teenage population groups. Powdered milks were statistically unreliable in the children and male adult population groups. Soy and imitation milks were statistically unreliable in the male and female teenager and male adult population groups.

5.0 CONCLUSIONS

Consumption data and information pertaining to the individual food-uses of phosphatidylserine were used to estimate the all-person and all-user intakes of phosphatidylserine for specific demographic groups and for the total U.S. population. This type of intake methodology is generally considered to be 'worst case' as a result of several conservative assumptions made in the consumption estimates. For example, it is well established that the length of a dietary survey affects the estimated consumption of individual users. Short-term surveys, such as the typical 2- or 3-day dietary surveys, overestimate the consumption of food products that are consumed relatively infrequently.

In summary, on an all-user basis, the mean intake of phosphatidylserine by the total U.S. population from all proposed food-uses was estimated to be 39.4 mg/person/day or 0.8 mg/kg body weight/day. The heavy consumer (90th percentile) all-user intake of phosphatidylserine by the total U.S. population from all proposed food-uses was estimated to be 79.1 mg/person/day or 1.8 mg/kg body weight/day.

6.0 REFERENCES

Anderson, S.A. (Ed.). 1988. Estimation of Exposure to Substances in the Food Supply. Federation of American Societies for Experimental Biology (FASEB), Life Science Research Office (LSRO); Bethesda, MD. [Contract No. FDA 223-84-2059].

CFR. 2007a. Part 170—Food additives. Sections §170.3—Definitions. In: U.S. Code of Federal Regulations (CFR). Title 21: Food and Drugs (Food and Drug Administration). U.S. Government Printing Office (GPO); Washington, DC, pp. 5-9. Available from: http://a257.g.akamaitech.net/7/257/2422/26mar20071500/edocket.access.gpo.gov/cfr_2007/aprqtr/pdf/21cfr170.3.pdf.

CFR. 2007b. Part 101—Food labeling. Sections §101.12—Reference amounts customarily consumed per eating occasion. In: U.S. Code of Federal Regulations (CFR). Title 21: Food and Drugs (U.S. Food and Drug Administration). U.S. Food and Drug Administration (U.S. FDA). U.S. Government Printing Office (GPO); Washington, DC, pp. 46-56. Available from: http://a257.g.akamaitech.net/7/257/2422/26mar20071500/edocket.access.gpo.gov/cfr_2007/aprqtr/pdf/21cfr101.12.pdf.

LSRO. 1995. The Third Report on Nutrition Monitoring in the United States. Federation of American Societies for Experimental Biology (FASEB), Life Sciences Research Office (LSRO), Interagency Board for Nutrition Monitoring and Related Research; Bethesda, Maryland, Vol. 1, pp. 19-31 & III-1 to III-10 and Vol. 2, pp. VB-1 to VB-2.

USDA. 2000. 1994-1996, 1998 Continuing Survey of Food Intakes by Individuals (CSFII) and Diet and Health Knowledge Survey (DHKS) (On CD-ROM). U.S. Department of Agriculture (USDA); Riverdale, Maryland. PB2000-500027 Supercedes PB98-500457.

APPENDIX A

Estimated Daily Intake of Phosphatidylserine from Individual Proposed Food-Uses by Different Population Groups Within the United States

Table A-1 Estimated Daily Intake of Phosphatidylserine from Individual Proposed Food-Uses by Infants Aged 0 to 2 Years Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg)	90^{th} Percentile (mg)	Mean (mg)	90^{th} Percentile (mg)
Breakfast Cereals						
Instant and Regular Hot Cereals	12.0	429	2.0	7.6	14.0	27.7
Ready-to-Eat Cereal	39.8	1,424	7.7	24.3	17.3	35.0
Dairy Product Analogs						
Imitation and Soy Milk	0.3	9	0.1*	na	42.8	76.6
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	3.2	115	0.6	na	15.4	27.8
Milk and Milk Products						
Flavored Milk and Milk Drinks	3.1	110	0.3	na	10.6	31.3
Milk, Fluid Regular	5.3	190	1.6	na	26.3	67.0
Milk, Powdered	0	0	na	na	na	na
Milk-Based Meal Replacements	0.1	5	0.1*	na	56.2*	187.5*
Yogurt	8.8	317	1.2	na	11.6	18.8
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	< 0.1	1	< 0.1*	na	11.3*	11.3*

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000058

Table A-2 Estimated Daily Intake of Phosphatidylserine from Individual Proposed Food-Uses by Children Aged 3 to 11 Years Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg)	90th Percentile (mg)	Mean (mg)	90th Percentile (mg)
Breakfast Cereals						
Instant and Regular Hot Cereals	13.2	830	2.0	7.3	17.9	31.4
Ready-to-Eat Cereal	65.3	4,116	19.9	49.5	31.3	57.0
Dairy Product Analogs						
Imitation and Soy Milk	0.2	15	< 0.1*	na	29.7	45.9
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	5.8	363	1.1	na	17.4	31.5
Milk and Milk Products						
Flavored Milk and Milk Drinks	8.2	514	1.3	na	15.1	28.7
Milk, Fluid Regular	13.6	858	4.7	15.3	30.4	61.3
Milk, Powdered	< 0.1	2	< 0.1*	na	24.1*	24.2*
Milk-Based Meal Replacements	0.4	25	0.1	na	29.3	69.4
Yogurt	9.4	593	0.9	na	11.1	16.6
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	0.4	27	0.1	na	23.9	37.5

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000059

Table A-3 Estimated Daily Intake of Phosphatidylserine from Individual Proposed Food-Uses by Female Teenagers Aged 12 to 19 Years Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg)	90th Percentile (mg)	Mean (mg)	90th Percentile (mg)
Breakfast Cereals						
Instant and Regular Hot Cereals	5.8	41	1.1	na	18.8	30.3
Ready-to-Eat Cereal	40.7	286	13.2	40.0	31.4	56.7
Dairy Product Analogs						
Imitation and Soy Milk	0.1	1	< 0.1*	na	15.3*	15.3*
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	5.1	36	1.0	na	17.5	24.4
Milk and Milk Products						
Flavored Milk and Milk Drinks	5.4	38	1.0	na	19.6	35.1
Milk, Fluid Regular	13.5	95	4.5	15.3	30.9	76.6
Milk, Powdered	0	0	na	na	na	na
Milk-Based Meal Replacements	0.7	5	0.8*	na	104.6*	450.0*
Yogurt	3.6	25	0.5	na	13.8	22.6
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	0.3	2	0.4*	na	142.1*	180.0*

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000060

Table A-4	**Estimated Daily Intake of Phosphatidylserine from Individual Proposed Food-Uses by Male Teenagers Aged 12 to 19 Years Within the United States (1994-1996, 1998 USDA CSFII Data)**					
Food-Use Category	**% Users**	**Actual # of Total Users**	**All-Person Consumption**		**All-Users Consumption**	
			Mean (mg)	**90th Percentile (mg)**	**Mean (mg)**	**90th Percentile (mg)**
Breakfast Cereals						
Instant and Regular Hot Cereals	6.0	42	1.2	na	23.6	45.7
Ready-to-Eat Cereal	42.7	297	20.1	60.0	46.7	84.0
Dairy Product Analogs						
Imitation and Soy Milk	0.1	1	< 0.1*	na	22.9*	22.9*
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	4.9	34	1.0	na	20.2	32.3
Milk and Milk Products						
Flavored Milk and Milk Drinks	6.6	46	1.6	na	23.0	46.9
Milk, Fluid Regular	12.9	90	5.8	15.3	45.4	111.0
Milk, Powdered	0	0	na	na	na	na
Milk-Based Meal Replacements	0.6	4	0.3*	na	47.3	69.4
Yogurt	2.6	18	0.5	na	16.4	32.1
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	0.4	3	0.2*	na	55.6	71.3

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000061

Table A-5 Estimated Daily Intake of Phosphatidylserine from Individual Proposed Food-Uses by Female Adults Aged 20 and Over Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg)	90th Percentile (mg)	Mean (mg)	90th Percentile (mg)
Breakfast Cereals						
Instant and Regular Hot Cereals	12.8	586	2.0	7.6	17.2	30.4
Ready-to-Eat Cereal	31.9	1,458	8.8	30.5	27.4	49.5
Dairy Product Analogs						
Imitation and Soy Milk	0.2	10	< 0.1*	na	13.5	23.0
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	2.9	134	0.7	na	19.3	32.3
Milk and Milk Products						
Flavored Milk and Milk Drinks	1.9	85	0.3	na	16.3	32.8
Milk, Fluid Regular	19.2	876	4.9	19.1	24.7	53.4
Milk, Powdered	0.4	19	< 0.1*	na	10.9	38.5
Milk-Based Meal Replacements	1.7	76	0.8	na	42.4	69.8
Yogurt	7.7	354	1.3	na	16.0	30.2
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	1.1	52	0.4	na	34.3	75.0

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000062

Table A-6 **Estimated Daily Intake of Phosphatidylserine from Individual Proposed Food-Uses by Male Adults Aged 20 and Over Within the United States (1994-1996, 1998 USDA CSFII Data)**						
Food-Use Category	**% Users**	**Actual # of Total Users**	**All-Person Consumption**		**All-Users Consumption**	
			Mean (mg)	**90th Percentile (mg)**	**Mean (mg)**	**90th Percentile (mg)**
Breakfast Cereals						
Instant and Regular Hot Cereals	11.8	561	2.3	na	22.7	43.9
Ready-to-Eat Cereal	29.9	1,422	11.0	41.3	37.8	70.0
Dairy Product Analogs						
Imitation and Soy Milk	0.1	6	< 0.1*	na	18.6*	45.9*
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	2.6	123	0.8	na	25.8	48.8
Milk and Milk Products						
Flavored Milk and Milk Drinks	1.6	77	0.3	na	17.6	36.6
Milk, Fluid Regular	14.8	702	4.6	15.3	30.9	68.9
Milk, Powdered	0.2	9	< 0.1*	na	13.2*	36.2*
Milk-Based Meal Replacements	1.3	60	0.9	na	69.6	163.1
Yogurt	4.0	189	0.8	na	17.2	31.1
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	0.9	44	0.4	na	50.3	108.8

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000063

Table A-7 Estimated Daily Intake of Phosphatidylserine from Individual Proposed Food-Uses by the Total Population (All Ages) Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg)	90th Percentile (mg)	Mean (mg)	90th Percentile (mg)
Breakfast Cereals						
Instant and Regular Hot Cereals	12.1	2,489	2.0	4.8	19.1	33.1
Ready-to-Eat Cereal	43.7	9,003	11.9	39.9	32.0	60.0
Dairy Product Analogs						
Imitation and Soy Milk	0.2	42	< 0.1	na	19.4	45.9
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	3.9	805	0.8	na	20.4	34.0
Milk and Milk Products						
Flavored Milk and Milk Drinks	4.2	870	0.5	na	17.0	31.3
Milk, Fluid Regular	13.6	2,811	4.7	15.3	28.6	61.3
Milk, Powdered	0.1	30	< 0.1	na	11.7	36.2
Milk-Based Meal Replacements	0.8	175	0.7	na	53.0	108.8
Yogurt	7.3	1,496	1.0	na	15.1	28.5
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	0.6	129	0.3	na	42.1	75.0

na = not applicable.

000064

APPENDIX B

Estimated Daily Per Kilogram Body Weight Intake of Phosphatidylserine from Individual Proposed Food-Uses by Different Population Groups Within the United States

Table B-1 Estimated Daily Per Kilogram Body Weight Intake of Phosphatidylserine from Individual Proposed Food-Uses by Infants Aged 0 to 2 Years Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg/kg)	90th Percentile (mg/kg)	Mean (mg/kg)	90th Percentile (mg/kg)
Breakfast Cereals						
Instant and Regular Hot Cereals	12.0	429	0.2	0.7	1.2	2.4
Ready-to-Eat Cereal	39.8	1,424	0.6	1.9	1.3	2.7
Dairy Product Analogs						
Imitation and Soy Milk	0.3	9	< 0.1*	na	3.2	5.8
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	3.2	115	< 0.1	na	1.2	2.1
Milk and Milk Products						
Flavored Milk and Milk Drinks	3.1	110	< 0.1	na	0.8	2.2
Milk, Fluid Regular	5.3	190	0.1	na	2.1	4.5
Milk, Powdered	0	0	na	na	na	na
Milk-Based Meal Replacements	0.1	5	< 0.1*	na	5.0*	17.9*
Yogurt	8.8	317	0.1	0.1	1.0	1.8
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	< 0.1	1	< 0.1*	na	0.8*	0.8*

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

Table B-2 Estimated Daily Per Kilogram Body Weight Intake of Phosphatidylserine from Individual Proposed Food-Uses by Children Aged 3 to 11 Years Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg/kg)	90th Percentile (mg/kg)	Mean (mg/kg)	90th Percentile (mg/kg)
Breakfast Cereals						
Instant and Regular Hot Cereals	13.2	830	0.1	0.3	0.8	1.4
Ready-to-Eat Cereal	65.3	4,116	0.8	2.0	1.3	2.4
Dairy Product Analogs						
Imitation and Soy Milk	0.2	15	< 0.1*	na	1.2	1.7
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	5.8	363	< 0.1	na	0.7	1.3
Milk and Milk Products						
Flavored Milk and Milk Drinks	8.2	514	0.1	na	0.6	1.2
Milk, Fluid Regular	13.6	858	0.2	0.7	1.2	2.5
Milk, Powdered	< 0.1	2	< 0.1*	na	1.2*	1.8*
Milk-Based Meal Replacements	0.4	25	< 0.1	na	1.1	1.8
Yogurt	9.4	593	< 0.1	na	0.5	0.9
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	0.4	27	< 0.1	na	0.9	2.9

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000067

Table B-3 Estimated Daily Per Kilogram Body Weight Intake of Phosphatidylserine from Individual Proposed Food-Uses by Female Teenagers Aged 12 to 19 Years Within the United States (1994-1996, 1998 USDA CSFII Data)						
Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg/kg)	90th Percentile (mg/kg)	Mean (mg/kg)	90th Percentile (mg/kg)
Breakfast Cereals						
Instant and Regular Hot Cereals	5.8	41	< 0.1	na	0.3	0.6
Ready-to-Eat Cereal	40.7	286	0.2	0.7	0.6	1.0
Dairy Product Analogs						
Imitation and Soy Milk	0.1	1	< 0.1*	na	0.3*	0.3*
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	5.1	36	< 0.1	na	0.3	0.5
Milk and Milk Products						
Flavored Milk and Milk Drinks	5.4	38	< 0.1	na	0.4	0.6
Milk, Fluid Regular	13.5	95	0.1	0.3	0.5	1.3
Milk, Powdered	0	0	na	na	na	na
Milk-Based Meal Replacements	0.7	5	< 0.1*	na	1.4*	5.7*
Yogurt	3.6	25	< 0.1	na	0.2	0.4
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	0.3	2	< 0.1*	na	2.4*	3.1*

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000068

Table B-4 Estimated Daily Per Kilogram Body Weight Intake of Phosphatidylserine from Individual Proposed Food-Uses by Male Teenagers Aged 12 to 19 Years Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg/kg)	90th Percentile (mg/kg)	Mean (mg/kg)	90th Percentile (mg/kg)
Breakfast Cereals						
Instant and Regular Hot Cereals	6.0	42	< 0.1	na	0.4	0.8
Ready-to-Eat Cereal	42.7	297	0.3	1.0	0.8	1.5
Dairy Product Analogs						
Imitation and Soy Milk	0.1	1	< 0.1*	na	0.4*	0.4*
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	4.9	34	< 0.1	na	0.4	0.8
Milk and Milk Products						
Flavored Milk and Milk Drinks	6.6	46	< 0.1	na	0.4	0.6
Milk, Fluid Regular	12.9	90	0.1	0.2	0.8	1.6
Milk, Powdered	0	0	na	na	na	na
Milk-Based Meal Replacements	0.6	4	< 0.1*	na	0.7	0.8
Yogurt	2.6	18	< 0.1	na	0.3	0.4
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	0.4	3	< 0.1*	na	0.5	0.6

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000069

Table B-5 Estimated Daily Per Kilogram Body Weight Intake of Phosphatidylserine from Individual Proposed Food-Uses by Female Adults Aged 20 and Over Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg/kg)	90th Percentile (mg/kg)	Mean (mg/kg)	90th Percentile (mg/kg)
Breakfast Cereals						
Instant and Regular Hot Cereals	12.8	586	< 0.1	0.1	0.3	0.5
Ready-to-Eat Cereal	31.9	1,458	0.1	0.5	0.4	0.8
Dairy Product Analogs						
Imitation and Soy Milk	0.2	10	< 0.1*	na	0.2*	0.4*
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	2.9	134	< 0.1	na	0.3	0.6
Milk and Milk Products						
Flavored Milk and Milk Drinks	1.9	85	< 0.1	na	0.3	0.5
Milk, Fluid Regular	19.2	876	0.1	0.3	0.4	0.8
Milk, Powdered	0.4	19	< 0.1*	na	0.2*	0.7*
Milk-Based Meal Replacements	1.7	76	< 0.1	na	0.6	1.2
Yogurt	7.7	354	< 0.1	na	0.2	0.4
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	1.1	52	< 0.1	na	0.5	0.9

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000070

Table B-6 Estimated Daily Per Kilogram Body Weight Intake of Phosphatidylserine from Individual Proposed Food-Uses by Male Adults Aged 20 and Over Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg/kg)	90th Percentile (mg/kg)	Mean (mg/kg)	90th Percentile (mg/kg)
Breakfast Cereals						
Instant and Regular Hot Cereals	11.8	561	< 0.1	na	0.3	0.6
Ready-to-Eat Cereal	29.9	1,422	0.1	0.5	0.5	0.9
Dairy Product Analogs						
Imitation and Soy Milk	0.1	6	< 0.1*	na	0.3*	0.7*
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	2.6	123	< 0.1	na	0.3	0.6
Milk and Milk Products						
Flavored Milk and Milk Drinks	1.6	77	< 0.1	na	0.2	0.5
Milk, Fluid Regular	14.8	702	0.1	0.2	0.4	0.8
Milk, Powdered	0.2	9	< 0.1*	na	0.2*	0.6*
Milk-Based Meal Replacements	1.3	60	< 0.1	na	0.8	1.8
Yogurt	4.0	189	< 0.1	na	0.2	0.4
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	0.9	44	< 0.1	na	0.6	1.1

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000071

Table B-7 Estimated Daily Per Kilogram Body Weight Intake of Phosphatidylserine from Individual Proposed Food-Uses by the Total Population (All Ages) Within the United States (1994-1996, 1998 USDA CSFII Data)

Food-Use Category	% Users	Actual # of Total Users	All-Person Consumption		All-Users Consumption	
			Mean (mg/kg)	90th Percentile (mg/kg)	Mean (mg/kg)	90th Percentile (mg/kg)
Breakfast Cereals						
Instant and Regular Hot Cereals	12.1	2,489	< 0.1	0.1	0.4	0.9
Ready-to-Eat Cereal	43.7	9,003	0.3	0.8	0.7	1.5
Dairy Product Analogs						
Imitation and Soy Milk	0.2	42	< 0.1*	na	0.6	1.3
Grain and Pasta Products						
Nutritional Bars (Breakfast, Granola, Protein)	3.9	805	< 0.1	na	0.4	0.9
Milk and Milk Products						
Flavored Milk and Milk Drinks	4.2	870	< 0.1	na	0.4	0.9
Milk, Fluid Regular	13.6	2,811	0.1	0.3	0.5	1.2
Milk, Powdered	0.1	30	< 0.1	na	0.2	0.7
Milk-Based Meal Replacements	0.8	175	< 0.1	na	0.8	1.4
Yogurt	7.3	1,496	< 0.1	na	0.3	0.7
Processed Fruits and Fruit Juices						
Fruit Flavored Drink	0.6	129	< 0.1	na	0.6	0.9

na = not applicable.
*Indicates an intake estimate that may not be statistically reliable, as the CV of the mean is equal to or greater than 30% (see Section 2.3).

000072

APPENDIX C

Representative CSFII 1994-1996, 1998 Food Codes for All Proposed Food-Uses of Phosphatidylserine in the United States

Representative CSFII 1994-1996, 1998 Food Codes for All Proposed Food-Uses of Phosphatidylserine in the United States

Breakfast Cereals

Instant and Regular Hot Cereals
[Phosphatidylserine] = 0.0125%

56200300	Cereal, cooked, NFS
56200350	Cereal, cooked, instant, NS as to grain
56200990	Grits, cooked, corn or hominy, NS as to regular, quick or instant, NS as to fat added in cooking
56201000	Grits, cooked, corn or hominy, NS as to regular, quick, or instant, fat not added in cooking
56201010	Grits, cooked, corn or hominy, regular, fat not added in cooking
56201020	Grits, cooked, corn or hominy, regular, fat added in cooking
56201030	Grits, cooked, corn or hominy, regular, NS as to fat added in cooking
56201040	Grits, cooked, corn or hominy, NS as to regular, quick, or instant, fat added in cooking
56201060	Grits, cooked, corn or hominy, with cheese, NS as to regular, quick, or instant, NS as to fat added
56201061	Grits, cooked, corn or hominy, with cheese, NS as to regular, quick, or instant, fat not added in cooking
56201062	Grits, cooked, corn or hominy, with cheese, NS as to regular, quick, or instant, fat added in cooking
56201070	Grits, cooked, corn or hominy, with cheese, regular, NS as to fat added in cooking
56201071	Grits, cooked, corn or hominy, with cheese, regular, fat not added in cooking
56201072	Grits, cooked, corn or hominy, with cheese, regular, fat added in cooking
56201080	Grits, cooked, corn or hominy, with cheese, quick, NS as to fat added in cooking
56201081	Grits, cooked, corn or hominy, with cheese, quick, fat not added in cooking
56201082	Grits, cooked, corn or hominy, with cheese, quick, fat added in cooking
56201090	Grits, cooked, corn or hominy, with cheese, instant, NS as to fat added in cooking
56201091	Grits, cooked, corn or hominy, with cheese, instant, fat not added in cooking
56201092	Grits, cooked, corn or hominy, with cheese, instant, fat added in cooking
56201110	Grits, cooked, corn or hominy, quick, fat not added in cooking
56201120	Grits, cooked, corn or hominy, quick, fat added in cooking
56201130	Grits, cooked, corn or hominy, quick, NS as to fat added in cooking
56201210	Grits, cooked, corn or hominy, instant, fat not added in cooking
56201220	Grits, cooked, corn or hominy, instant, fat added in cooking
56201230	Grits, cooked, corn or hominy, instant, NS as to fat added in cooking
56201240	Grits, cooked, flavored, corn or hominy, instant, fat not added in cooking
56201250	Grits, cooked, flavored, corn or hominy, instant, fat added in cooking
56201260	Grits, cooked, flavored, corn or hominy, instant, NS as to fat added in cooking
56201300	Grits, cooked, corn or hominy, NS as to regular, quick, or instant, NS as to fat added in cooking
56201510	Cornmeal mush, made with water
56201520	Cornmeal mush, fried
56201530	Cornmeal mush, made with milk
56201540	Cornmeal, made with evaporated milk and sugar, Puerto Rican Style (Harina de maize con leche)
56201560	Cornmeal sticks, boiled
56201600	Cornmeal, lime-treated, cooked (Masa harina)
56201700	Cornstarch with milk, eaten as a cereal (2 tbsp cornstarch in 2-1/2 cups milk)
56201990	Millet, cooked, NS as to fat added in cooking
56202000	Millet, cooked, fat not added in cooking

56202100	Millet, cooked, fat added in cooking
56202500	Muesli, prepared, instant
56202960	Oatmeal, cooked, NS as to regular, quick or instant; NS as to fat added in cooking
56202970	Oatmeal, cooked, quick (1 or 3 minutes), NS as to fat added in cooking
56202980	Oatmeal, cooked, regular, NS as to fat added in cooking
56203000	Oatmeal, cooked, NS as to regular, quick or instant, fat not added in cooking
56203010	Oatmeal, cooked, regular, fat not added in cooking
56203020	Oatmeal, cooked, quick (1 or 3 minutes), fat not added in cooking
56203030	Oatmeal, cooked, instant, fat not added in cooking
56203040	Oatmeal, cooked, NS as to regular, quick, or instant, fat added in cooking
56203050	Oatmeal, cooked, regular, fat added in cooking
56203060	Oatmeal, cooked, quick (1 or 3 minutes), fat added in cooking
56203070	Oatmeal, cooked, instant, fat added in cooking
56203080	Oatmeal, cooked, instant, NS as to fat added in cooking
56203090	Oatmeal, fortified, cooked, instant, fat not added in cooking
56203100	Oatmeal, fortified, cooked, instant, fat added in cooking
56203110	Oatmeal with maple flavor, cooked
56203120	Oatmeal, with oat bran, fortified, cooked, instant, fat not added in cooking
56203140	Total Oatmeal, cooked, quick, fat not added in cooking
56203200	Oatmeal with fruit, cooked
56203210	Oatmeal, NS as to regular, quick, or instant, made with milk, fat not added in cooking
56203220	Oatmeal, NS as to regular, quick, or instant, made with milk, fat added in cooking
56203230	Oatmeal, NS as to regular, quick, or instant, made with milk, NS as to fat added in cooking
56203540	Oatmeal, made with evaporated milk and sugar, Puerto Rican style
56203600	Oatmeal, multigrain, cooked, NS as to fat added in cooking
56203610	Oatmeal, multigrain, cooked, fat not added in cooking
56203620	Oatmeal, multigrain, cooked, fat added in cooking
56206970	Wheat, cream of, cooked, quick, NS as to fat added in cooking
56206980	Wheat, cream of, cooked, regular, NS as to fat added in cooking
56206990	Wheat, cream of, cooked, NS as to regular, quick, or instant, NS as to fat added in cooking
56207000	Wheat, cream of, cooked, NS as to regular, quick, or instant, fat not added in cooking
56207010	Wheat, cream of, cooked, regular, fat not added in cooking
56207020	Wheat, cream of, cooked, quick, fat not added in cooking
56207030	Wheat, cream of, cooked, instant, fat not added in cooking
56207040	Wheat, cream of, cooked, made with milk
56207050	Wheat, cream of, cooked, made with milk and sugar, Puerto Rican style
56207060	Wheat, cream of, cooked, instant, fat added in cooking
56207070	Wheat, cream of, cooked, instant, NS as to fat added in cooking
56207080	Wheat, cream of, cooked, NS as to regular, quick, or instant, fat added in cooking
56207100	Wheat, rolled, cooked, fat not added in cooking
56207110	Bulgur, cooked or canned, fat not added in cooking
56207120	Bulgur, cooked or canned, fat added in cooking
56207130	Bulgur, cooked or canned, NS as to fat added in cooking
56207140	Wheat, rolled, cooked, NS as to fat added in cooking
56207150	Couscous, plain, cooked, fat not added in cooking
56207160	Couscous, plain, cooked, NS as to fat added in cooking
56207180	Couscous, plain, cooked, fat added in cooking
56207190	Whole wheat cereal, cooked, NS as to fat added in cooking
56207200	Whole wheat cereal, cooked, fat not added in cooking
56207210	Whole wheat cereal, cooked, fat added in cooking
56207220	Wheat, cream of, cooked, regular, fat added in cooking
56207230	Wheat, cream of, cooked, quick, fat added in cooking
56207290	Wheat hearts, cooked, NS as to fat added in cooking
56207300	Whole wheat cereal, wheat and barley, cooked, fat not added in cooking

56207310	Wheat hearts, cooked, fat not added in cooking
56207330	Whole wheat cereal, wheat and barley, cooked, fat added in cooking
56207340	Whole wheat cereal, wheat and barley, cooked, NS as to fat added in cooking
56207350	Wheat cereal, chocolate flavored, cooked, made with milk
56207360	Wheat cereal, chocolate flavored, cooked, fat not added in cooking
56207370	Wheat cereal, chocolate flavored, cooked, NS as to fat added in cooking
56208000	Multigrain cereal, cooked, fat not added in cooking
56208010	Multigrain cereal, cooked, fat added in cooking
56208020	Multigrain cereal, cooked, NS as to fat added in cooking
56208500	Oat bran cereal, cooked, fat not added in cooking
56208510	Oat bran cereal, cooked, fat added in cooking
56208520	Oat bran cereal, cooked, NS as to fat added in cooking
56208530	Oat bran cereal, cooked, made with milk, fat not added in cooking
56208540	Oat bran cereal, cooked, made with milk, fat added in cooking
56208550	Oat bran cereal, cooked, made with milk, NS as to fat added in cooking
56209000	Rye, cream of, cooked
56210000	Nestum cereal

Ready-to-Eat Cereals
[Phosphatidylserine] = 0.1%

57000000	Cereal, NFS
57000050	Kashi cereal, NS as to ready to eat or cooked
57000100	Oat cereal, NFS
57100100	Cereal, ready-to-eat, NFS
57101000	All-Bran
57101020	All-Bran with Extra Fiber
57101500	Almond Delight
57102000	Alpen
57103000	Alpha-Bits
57103020	Alpha-bits with marshmallows
57103050	Amaranth Flakes
57103100	Apple Cinnamon Cheerios
57103400	Apple Cinnamon Oh's Cereal
57103450	Apple Cinnamon Rice Krispies
57104000	Apple Jacks
57106000	Banana Frosted Flakes
57106200	Batman
57106250	Berry Berry Kix
57106300	Bigg Mixx
57106500	Bill and Ted's Excellent Adventure
57106530	Blueberry Morning, Post
57107000	Booberry
57109000	Body Buddies, natural fruit flavor
57110000	All-Bran Bran Buds, Kellogg's (formerly Bran Buds)
57111000	Bran Chex
57111300	Bran News
57111500	Bran Muffin Crisp
57112500	Breakfast with Barbie
57113000	Buc Wheats
57113300	Bunuelitos
57114000	C-3PO's
57116100	Cabbage Patch
57117000	Cap'n Crunch
57117500	Cap'n Crunch's Christmas Crunch
57118000	Cap'n Crunch's Choco Crunch

57119000	Cap'n Crunch's Crunch Berries
57119500	Cap'n Crunch's Deep Sea Crunch
57120000	Cap'n Crunch's Peanut Butter Crunch
57123000	Cheerios
57124000	Chex cereal, NFS
57124200	Chocolate flavored frosted puffed corn cereal
57124500	Cinnamon Grahams, General Mills
57125000	Cinnamon Toast Crunch
57125900	Honey Nut Clusters (formerly called Clusters)
57126000	Cocoa Krispies
57126500	Cocoa Blasts, Quaker
57127000	Cocoa Pebbles
57128000	Cocoa Puffs
57128880	Common Sense Oat Bran, plain
57128900	Common Sense Oat Bran, with raisins
57130000	Cookie-Crisp
57131000	Crunchy Corn Bran, Quaker
57132000	Corn Chex
57134000	Corn flakes, NFS
57134090	Corn flakes, low sodium
57135000	Corn flakes, Kellogg
57138000	Total Corn Flakes
57139000	Count Chocula
57141000	Cracker Jack
57144000	Crisp Crunch
57148000	Crispix
57148500	Crispy Brown Rice Cereal
57151000	Crispy Rice
57152000	Crispy Wheats'n Raisins
57152100	Croonchy Stars
57201700	Dino Pebbles
57202100	Donkey Kong
57203100	Donkey Kong Jr.
57204100	Donutz Cereal
57205100	Donutz Cereal, Chocolate
57205250	Double Chex
57205260	Double Dip Crunch, Kellogg's
57205300	E.T. Cereal
57206700	Fiber One
57206800	Fiber 7 Flakes, Health Valley
57212100	French Toast Crunch, General Mills
57213800	Frosted Bran, Kellogg's
57213850	Frosted Cheerios
57214100	Frosted Wheat Bites
57215000	Frosty O's
57216000	Frosted rice, NFS
57217000	Frosted Rice Krinkles
57218000	Frosted Rice Krispies
57219000	Fruit & Fibre (fiber), NFS
57220000	Fruit & Fibre (fiber) with apples and cinnamon
57221000	Fruit & Fibre (fiber) with dates, raisins, and walnuts
57221500	Fruit & Fibre (fiber) tropical fruit w/oat clusters
57221600	Fruit & Fibre (fiber) with peaches, raisins, almonds and oat clusters
57221700	Fruit Rings, NFS
57221800	Fruit Whirls
57222500	Fruit Wheats

57224000	Golden Grahams
57225000	Golden Harvest Proteinola
57231000	Grape-Nut Flakes
57231500	Gremlins
57232000	Halfsies
57232100	Healthy Choice Almond Crunch with raisins, Kellogg's
57232120	Healthy Choice Multi-Grain Flakes, Kellogg's
57235500	Heartwise, plain
57235600	Heartwise, with fruit nuggets
57237000	Honey Bran
57237100	Honey Bunches of Oats
57237300	Honey Bunches of Oats with Almonds, Post
57238000	Honeycomb, plain
57239000	Honeycomb, strawberry
57239100	Honey Crunch Corn Flakes, Kellogg's
57240000	Honey Graham Chex
57241000	Honey Nut Cheerios
57243000	Honey Smacks
57243600	Hot Wheels
57243900	Jetsons
57301100	Kaboom
57301700	Kenmei Rice Bran
57302100	King Vitaman
57303100	Kix
57304100	Life (plain and cinnamon)
57305100	Lucky Charms
57305150	Frosted oat cereal with marshmallows
57305170	Malt-O-Meal Coco-Roos
57305200	Malt-O-Meal Crispy Rice
57305500	Malt-O-Meal Honey and Nut Toasty O's
57306500	Malt-O-Meal Golden Puffs (formerly Sugar Puffs)
57306700	Malt-O-Meal Toasted Oat Cereal
57306800	Malt-O-meal Tootie Fruities
57307100	Fruity Marshmallow Krispies (formerly called Marshmallow Krispies)
57307550	Mini Buns Cereal (cinnamon)
57308000	Morning Funnies
57308300	Multi Bran Chex
57308400	Multi Grain Cheerios
57308410	Multi-Grain Cheerios Plus
57311600	Nintendo
57311790	Nut and Honey crunch biscuits
57311800	Nut and Honey Crunch (flakes)
57311900	Nutrific Oatmeal Flakes
57315000	Nutri-Grain Golden Wheat (formerly Nutri-Grain Wheat)
57316100	Nutri-Grain Almond Raisin
57316300	Oat Bran Flakes, Health Valley
57316350	Oat Bran Options
57316700	Oh's, Crunchy Nut
57316710	Oh's, Honey Graham
57316750	Oh's, Fruitangy, Quaker
57316800	O.J.'s
57317000	Oat flakes, fortified
57317200	Oat Flakes, Post
57322500	Oreo O's cereal, Post
57323000	Sweet Crunch, Quaker (formerly called Popeye)
57323050	Sweet Puffs, Quaker

57323200	Pop Tarts Crunch Cereal
57325000	Product 19
57327450	Quaker Oat Bran Cereal
57328000	Quisp
57328500	Rainbow Brite
57335530	Razzle Dazzle Rice Krispies
57335550	Reese's Peanut Butter Puffs cereal
57336000	Rice Chex
57337000	Rice Flakes, NFS
57339000	Rice Krispies
57339500	Rice Krispies Treats Cereal (Kellogg's)
57340200	Ripple Crisp Golden Corn
57340210	Ripple Crisp Honey Bran, General Mills
57340900	S.W. Graham
57342000	Slimer! & Ghostbusters
57342500	S'mores Crunch
57343000	Smurf Magic Berries
57344000	Special K
57344050	Spider-Man, Ralston
57344100	Sprinkle Spangles
57345000	Strawberry Krispies
57346500	Toasted Oatmeal, Honey Nut (Quaker)
57347000	Corn Pops
57347500	Strawberry Squares Mini-Wheats, Kellogg's (formerly Strawberry Squares)
57348000	Frosted corn flakes, NFS
57349000	Frosted Flakes, Kellogg
57349010	Cocoa Frosted Flakes, Kellogg's
57350000	Frosted Flakes, Ralston Purina
57352000	Sugar-Sparkled Flakes
57353000	Sugar-Sparkled Rice Krinkles
57354000	Sun Flakes
57355000	Golden Crisp (Formerly called Super Golden Crisp)
57401100	Toasted oat cereal
57402000	Team
57402250	Teddy Grahams Breakfast Bears
57402500	Teenage Mutant Ninja Turtles
57402600	Temptations, French Vanilla Almond, Kellogg's
57402610	Temptations, Honey Roasted Pecan, Kellogg's
57402750	Tiny Toon Adventures
57403100	Toasties, Post
57404100	Malt-O-Meal Toasty O's
57406100	Total
57406200	Triples
57408100	Uncle Sam's Hi Fiber Cereal
57409100	Waffle Crisp, Post
57410000	Weetabix Whole Wheat Cereal
57415000	Wheat'n Raisin Chex
57418000	Wheaties
57418200	Wheaties, Honey Frosted (formerly Wheaties Honey Gold)

Biscuit-Type Ready-to-Eat Cereals
[Phosphatidylserine] = 0.0545%

57103500	Apple Cinnamon Squares Mini-Wheats, Kellogg's (formerly Apple Cinnamon Squares)
57105000	Apple Raisin Crisp
57106100	Basic 4

57207000	Bran Flakes, NFS (formerly 40% Bran Flakes, NFS)
57208000	Complete Wheat Bran Flakes, Kellogg's (formerly 40% Bran Flakes)
57210100	40+ Bran Flakes
57214000	Frosted Mini-Wheats
57229000	Granola, lowfat, Kellogg's
57229500	Granola with Raisins, lowfat, Kellogg's
57232110	Healthy Choice Multi-Grain Squares, Kellogg's
57241200	Honey Nut Shredded Wheat, Post
57244000	Just Right
57245000	Just Right Fruit and Nut (formerly Just Right with raisins, dates, and nuts)
57308180	Mueslix Crispy Blend (formerly Mueslix Five Grain Muesli Cereal)
57308200	Mueslix golden crunch cereal
57308210	Muesli with apples and almonds, Ralston Purina
57308220	Strawberry muesli with pecans and raisins, Ralston
57312100	Nutri-Grain Biscuits, Whole Grain Shredded Wheat Cereal
57316410	Apple Cinnamon Oatmeal Crisp (formerly Oatmeal Crisp with Apples)
57316450	Oatmeal Crisp with Almonds
57316500	Oatmeal Raisin Crisp
57327500	Quaker Oatmeal Squares (formerly Quaker Oat Squares)
57329000	Raisin bran, NFS
57330000	Raisin Bran, Kellogg
57330500	Raisin Bran, Nutri System
57331000	Raisin Bran, Post
57332050	Raisin Bran, Total
57332100	Raisin Nut Bran
57332300	Super Raisin Bran, New Morning
57334000	Raisin Life
57335000	Raisins, Rice and Rye
57335500	Raisin Squares Mini-Wheats, Kellogg's (formerly Raisin Squares)
57341000	Shredded Wheat'N Bran
57417000	Shredded Wheat, 100%
57417500	Shredded Wheat with Oat Bran

Puffed Ready-to-Eat Cereals
[Phosphatidylserine] = 0.2%

57137000	Corn Puffs
57301500	Kashi, Puffed
57306100	Malt-O-Meal Puffed Rice
57306120	Malt-O-Meal Puffed Wheat
57307500	Millet, puffed
57340000	Rice, puffed
57416000	Wheat, puffed, plain
57416010	Wheat, puffed, presweetened with sugar

Dairy Product Analogs

Imitation Milk
[Phosphatidylserine] = 0.0125%

11310000 Milk, imitation, fluid, soy based

Soy Milk
[Phosphatidylserine] = 0.0125%

11320000 Milk, soy, ready-to-drink, not baby's

Grain Products and Pastas

Nutritional Bars (Breakfast, Granola, Protein)
[Phosphatidylserine] = 0.075%

41435010	High protein bar, soy base
41435110	High protein bar, candy-like, soy and milk base
41435200	High protein bar, cookie type, soy and milk base
53540000	Breakfast bar, nfs
53540100	Breakfast bar, cake-like
53540200	Breakfast bar, cereal crust with fruit filling, lowfat
53540250	Breakfast bar, cereal crust with fruit filling, fat free
53540500	Breakfast bar, date, with yogurt coating
53541100	Breakfast bar, diet meal type
53544450	PowerBar (fortified high energy bar)
53541200	Meal replacement bar (incl slim fast bar)
53542100	Granola bar w/ oats, sugar, raisins, coconut
53542200	Granola bar, oats, fruit, nuts, lowfat
53542210	Granola bar, nonfat
53543100	Granola bar w/ peanuts, oats, sugar, wheat germ
53544100	Granola bar, w/ nougat
53544200	Granola bar, chocolate-coated
53544210	Granola bar, w/ coconut, chocolate-coated
53544220	Granola bar w/ nuts, chocolate-coated
53544250	Granola bar, coated w/ nonchocolate coating
53544300	Granola bar, high fiber, yogurt coating, not choc
53544400	Granola bars, w/ rice cereal

Milk Products

Flavoured Milk and Milk Drinks
[Phosphatidylserine] = 0.0125%

11511000	Milk, chocolate, NFS
11513000	Cocoa & sugar mixture, milk added, ns type milk
11513400	Chocolate syrup milk added, ns as to type of milk
11514300	Cocoa with nonfat dry milk and low calorie sweetener, mixture, water added
11514500	Cocoa w/ whey, lo cal sweetnr, fortifd, water added
11515100	Cocoa and sugar mixture fortified with vitamins and minerals, milk added, NS as to type of milk, Puerto Rican style
11515400	Cocoa with nonfat dry milk and low calorie sweetener, high calcium, water added
11516000	Cocoa, whey, and low-calorie sweetener mixture, lowfat milk added
11518000	Milk beverage with nonfat dry milk and low calorie sweetener, water added, chocolate
11518050	Milk beverage with nonfat dry milk and low calorie sweetener, water added, flavors other than chocolate
11518100	Milk beverage with nonfat dry milk and low calorie sweetener, high calcium, water added, chocolate
11519000	Milk beverage, made with whole milk, flavors other than chocolate
11519050	Milk, flavors other than chocolate, whole milk-based
11519100	Milk beverage, beads, chocolate, whole milk added
11551050	Milk fruit drink
11551100	Milk fruit drink, Puerto Rican style (Champola de frutas)
11552200	Milk-based fruit drink
11553000	Fruit smoothie drink, w/ fruit and dairy products
11553100	Fruit smoothie drink, nfs

11560000	Chocolate-flavored drink, whey- and milk-based
11560020	Flavored milk drink, whey- and milk-based, flavors other than chocolate
11560100	Flavored milk drink, skim milk and cream-based, flavors other than chocolate
11560110	Chocolate-flavored milk drink, skim milk and cream-based

(Adjusted for not being reconstituted, 28 g of powder to 240 mL of water)
[Phosphatidylserine] = 0.1065%

11830100	Cocoa (or chocolate) with dry milk and sugar, dry mix, not reconstituted
11830110	Cocoa powder with nonfat dry milk and low calorie sweetener, dry mix, not reconstituted
11830120	Cocoa, whey, and low calorie sweetener, fortified, dry mix, not reconstituted
11830140	Chocolate, instant, dry mix, fortified with vitamins and minerals, not reconstituted, Puerto Rican style
11830150	Cocoa powder, not reconstituted (no dry milk)
11830160	Cocoa (or chocolate) flavored beverage powder with sugar, dry mix, not reconstituted
11830170	Cocoa, whey, and low-calorie sweetener mixture, not reconstituted
11830180	Cocoa (or chocolate) flavored beverage powder with low calorie sweetener, dry mix, not reconstituted
11830200	Milk, malted, dry mix, unfortified, not reconstituted, flavors other than chocolate
11830210	Milk, malted, dry mix, fortified, not reconstituted, flavors other than chocolate
11830250	Milk, malted, dry mix, unfortified, not reconstituted, chocolate
11830260	Milk, malted, dry mix, fortified, not reconstituted, chocolate
11830400	Milk beverage, powder, dry mix, not reconstituted, flavors other than chocolate
11830450	Milk beverage with sugar, dry milk, and egg white powder, dry mix, not reconstituted
11830500	Milk beverage, powder, with nonfat dry milk and low calorie sweetener, dry mix, not reconstituted, chocolate
11830550	Milk beverage, powder, with nonfat dry milk and low calorie sweetener, dry mix, not reconstituted, flavors other than chocolate

Milk, Fluid (Regular)
[Phosphatidylserine] = 0.0125%

11100000	Milk, nfs
11112000	Milk, cow's, fluid, not whole, ns as to % fat
11113000	Milk, cow's, fluid, skim or nonfat

Milk, Powdered
[Phosphatidylserine] = 0.0125%

(Adjusted for not being reconstituted, 28 g of powder to 240 mL of water)
[Phosphatidylserine] = 0.1065%

11810000	Milk, dry, not reconstituted, ns as to fat
11811000	Milk, dry, whole, not reconstituted
11812000	Milk, dry, lowfat, not reconstituted
11813000	Milk, dry, nonfat, not reconstituted

Milk-Based Meal Replacements
[Phosphatidylserine] = 0.0125%

11611000	Instant breakfast, fluid, canned
11612000	Instant breakfast, powder, milk added
11613000	Instant breakfast, powder, sweetener with low calorie sweetener, milk added
11621000	Diet beverage, liquid, canned
11622000	Diet beverage powder, milk added
11622010	Diet beverage, powder, reconstituted with skim (incl carnation)

11623000	Meal supplement / replacement, prepared, ready-to-drink
11631000	High calorie beverage, canned or powdered, reconstituted
11641000	Meal replacement, milk based, high protein, liquid
11651010	Meal replacement, cambridge, reconstituted, all flavors

(Adjusted for not being reconstituted, 16 g of powder to 240 mL of water)
[Phosphatidylserine] = 0.1875%

11830800	Instant breakfast powder, not reconstituted
11830810	Instant breakfast, powder, sweetened w/ low calorie sweetener, not reconstituted
11830850	High calorie milk beverage, powder, not reconstituted
11830900	Protein supplement, milk based, dry powder
11830940	Meal replacement, protein, milk based, fruit juice mix
11830950	Nutrient supp, milk-based, powdered, not reconstituted
11830960	Protein supp, milk base, sodium controlled, powder
11830970	Meal replacement, protein type, milk-base, powder
11830980	Protein supp, milk-base, powder (incl sustacal)
11830990	Nutrient supp, milk-base, powder (incl sustagen)
11831500	Nutrient supplement, milk-base, high protein, not reconstituted
11832000	Meal replacement, milk-&soy-base, powder, not reconstituted
11832500	Meal replacement, protein type, milk-base, w/sugar &art
11835000	Meal replacement, cambridge, powder, not reconstituted
11835100	Meal replacement, positrim drink mix, dry powder
11835150	Dynatrim, meal replacement, powder
11835200	Lose-it (nanci), meal replacement, powder
11840100	Milk, tiger's, dry, not reconstituted
11841100	Milk beverage beads, not choc, not reconstituted
11841110	Milk beverage beads, choc, not reconstituted
11940100	Nutramigen, dry, not reconstituted

Yogurt
[Phosphatidylserine] = 0.0133%

11410000	Yogurt, ns as to type of milk/flavor
11411100	Yogurt, plain, whole milk
11411200	Yogurt, plain, low fat milk
11411300	Yogurt, plain, nonfat milk
11420000	Yogurt, vanilla, lemon, coffee, ns as to milk type
11421000	Yogurt, vanilla, lemon, coffee, whole milk
11422000	Yogurt, vanilla, lemon, coffee, low fat milk
11423000	Yogurt, vanilla, lemon, coffee, nonfat milk
11424000	Yogurt, vanilla, lemon, coffee, nonfat milk, low cal sweet
11425000	Yogurt, chocolate, ns as to type of milk
11426000	Yogurt, chocolate, whole milk
11427000	Yogurt, chocolate, nonfat milk
11430000	Yogurt, fruit variety, ns as to milk type
11431000	Yogurt, fruit variety, whole milk
11432000	Yogurt, fruit variety, low fat milk
11433000	Yogurt, fruit variety, nonfat milk
11433500	Yogurt, fruited, nonfat milk, low cal sweetener
11444000	Yogurt, fruit & nuts, ns as to type of milk
11445000	Yogurt, fruit & nuts, low fat milk

Processed Fruits and Fruit Juices

Fruit Flavored Drink
[Phosphatidylserine] = 0.0125%

92741000 Fruit-flavored drink, non-carbonated, made from low calorie powdered mix

EAS CONSULTING GROUP

Specializing in FDA Regulatory Matters

RECEIVED
DEC 28 [illegible]
Division of Biotechnology and GRAS Notice Review

December 22, 2009

Robert Martin, Ph.D.
Office of Food Additive Safety (HFS-255)
Center for Food Safety and Applied Nutrition
Food and Drug Administration
5100 Paint Branch Parkway
College Park, MD 20740-3835

RE: Revision to Notification of GRAS Determination for Krill-based Phosphatidylserine in Food – Location of Information

Dear Dr. Martin:

In response to a request from your office, we are agreeable to the EAS Consulting Group office address being used as the location at which copies of the information that forms the basis for the GRAS determination are maintained. Accordingly, we have revised section IF. Availability of Information. We are providing you with a new page 4 of the GRAS Notice (in triplicate) to use as a replacement page.

Please let us know if you need any additional information.

Sincerely,

Edward A. Steele
President

Enclosure

EAS Consulting Group, LLC
1940 Duke Street, Suite 200, Alexandria, Virginia 22314
(877) 327-9808 Toll Free • (703) 684-4408 Local • (703) 684-4428 Fax
www.easconsultinggroup.com

Claim on dietary supplements containing EPA and DHA on October 31, 2000 as well as for conventional foods on September 8, 2004. FDA concluded that the use of EPA and DHA omega-3 fatty acids as dietary supplements is safe, provided that daily intakes of EPA and DHA do not exceed 3 g/person/day from conventional food and dietary supplement sources.

The estimated daily intake of krill-based phosphatidylserine from its intended uses at the 90th percentile of 79 mg/person (1.84 mg/kg/day) is well below the safe levels of intake determined from human studies where no adverse effects from intakes of 300 mg of phosphatidylserine/person/ day (5 mg/kg body weight/day) were noted. Similarly the estimated intake of EPA+DHA of 33 mg/person/day from the intended uses is considerably lower than the established safe levels of 3000 mg/person/day of EPA+DHA. Additionally, any EPA/DHA intake from the intended uses of krill-based phophatidylserine will be substitutional. The estimated daily intake of krill-based phosphatidylserine, if ingested daily over a lifetime, is considered safe.

F. Availability of Information:

The data and information that forms the basis for this GRAS determination will be provided to Food and Drug Administration upon request and are located at the offices of:

EAS Consulting Group, LLC
1940 Duke Street, Suite 200
Alexandria, VA 22314

Telephone: 703-684-4408
Facsimile: 703-684-4428
Email: esteele@easconsultinggroup.com

II. Detailed Information About the Identity of the Notified Substance:

A. Chemical name

Phosphatidylserine derived from krill.
Phosphatidylserine. Per IUPAC-CBN nomenclature, it is a 1,2-diacyl-*sn*-glycero-3-phospho-L-serine.

B. Trade Name:

The subject of this notification will be marketed as PS-Omega 3

C. Chemical Abstract Registry Number:

Phosphatidylserine: 84776-79-4

D. Chemical Formula:

The empirical formula of the most abundant krill-derived phosphatidylserine molecule in PS-Omega 3 is $C_{42}H_{71}O_{10}PNCa$

SUBMISSION END