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(71) Applicant (for all designated States except US): **MON-SANTO TECHNOLOGY LLC** [US/US]; 800 North Lindbergh Boulevard, St. Louis, Missouri 63167 (US).

(72) Inventor; and

(75) Inventor/Applicant (for US only): **WILKES, Richard S.** [US/US]; c/o Monsanto Technology LLC, 800 North Lindbergh Boulevard, St. Louis, Missouri 63167 (US).

(74) Agents: **GOFF, Christopher M.** et al.; Armstrong Teasdale LLP, 7700 Forsyth Blvd., Suite 1800, St. Louis, Missouri 63105-1847 (US).

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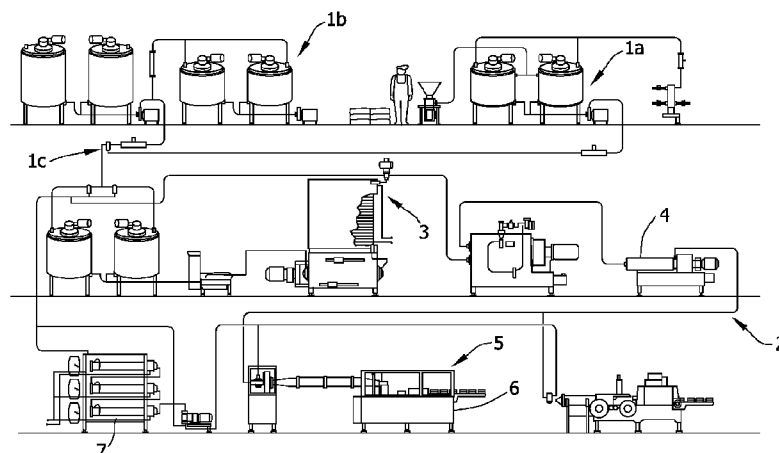
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(54) Title: SPREAD FORMULATIONS INCLUDING STEARIDONIC ACID

FIG. 1



(57) Abstract: The present disclosure provides for improved spread formulations and methods of producing such formulations by incorporating healthy lipids containing stearidonic acid into the formulations. In one embodiment of the disclosure, a spread formulation including a SDA-enriched oil is disclosed. In another embodiment of the disclosure, a margarine spread formulation including a SDA-enriched soybean oil is disclosed.

SPREAD FORMULATIONS INCLUDING STEARIDONIC ACID

FIELD OF THE DISCLOSURE

[0001] The disclosure relates to the enhancement of desirable characteristics in spread formulations such as margarine spreads through the incorporation of beneficial fatty acids. More specifically, it relates to spread formulations comprising polyunsaturated fatty acids including stearidonic acid and to methods of producing the formulations thereof. These modified spread formulations show an improvement in nutritional quality while maintaining shelf-life compared to conventional margarine spread formulations.

BACKGROUND OF THE DISCLOSURE

[0002] The present disclosure is directed to spread formulations such as margarine spreads including stearidonic acid ("SDA") or SDA-enriched oil. Specifically, the present disclosure provides margarine spread formulations that have improved nutritional quality and methods of producing the spread formulations.

[0003] Many studies have made a physiological link between dietary fats and pathologies such as obesity and atherosclerosis. In some instances, consumption of fats has been discouraged by the medical establishment. More recently, the qualitative differences between dietary fats and their health benefits have been recognized.

[0004] Recent studies have determined that despite their relatively simple biological structures, there are some types of fats that appear to improve body function in some ways. Some fats may, in fact, be essential to certain physiological processes. The wider class of fat molecules includes fatty acids, isoprenols, steroids, other lipids and oil-soluble vitamins. Among these are the fatty acids. The fatty acids are carboxylic acids, which have from 2 to 26 carbon atoms in their "backbone," with none or few desaturated sites in their carbohydrate structure. They generally have dissociation constants (pKa) of about 4.5 indicating that in normal body conditions (physiological pH of 7.4) the vast majority will be in a dissociated form.

[0005] With continued experimentation, workers in the field have begun to understand the nutritional need for fats and in particular fatty acids in the diet. For this reason, many in the food industry have begun to focus on fatty acids and lipid technology as a new focus for food production, with its consequent benefits for the consumers consuming the modified products. This focus has been particularly intense for the production and incorporation of omega-3 fatty acids into the diet. Omega-3 fatty acids are long-chain polyunsaturated fatty acids (18-22 carbon atoms in chain length) (LC-PUFAs) with the first of the double bonds (“unsaturations”) beginning with the third carbon atom from the methyl end of the molecule. They are called “polyunsaturated” because their molecules have two or more double bonds “unsaturations” in their carbohydrate chain. They are termed “long-chain” fatty acids since their carbon backbone has at least 18 carbon atoms. In addition to stearidonic acid “SDA” the omega-3 family of fatty acids includes alpha-linolenic acid (“ALA”), eicosatetraenoic acid (ETA), eicosapentaenoic acid (“EPA”), docosapentaenoic acid (DPA), and docosahexaenoic acid (“DHA”). ALA can be considered a “base” omega-3 fatty acid, from which EPA and DHA are made in the body through a series of enzymatic reactions, including the production of SDA. Most nutritionists point to DHA and EPA as the most physiologically important of the omega-3 fatty acids with the most beneficial effects. However, SDA has also been shown to have significant health benefits. See for example, US patent 7,163,960 herein incorporated by reference. Furthermore, it has now been shown that SDA readily enriches the EPA level in red blood cells.

[0006] The synthesis process from ALA is called “elongation” (i.e., the molecule becomes longer by incorporating new carbon atoms) and “desaturation” (i.e., new double bonds are created), respectively. In nature, ALA is primarily found in certain plant leaves and seeds (e.g., flax) while EPA and DHA mostly occur in the tissues of cold-water predatory fish (e.g., tuna, trout, sardines and salmon), and in some marine algae or microbes that they feed upon.

[0007] In addition to difficulties with simply securing an appropriate supply of LC-PUFAs for societal consumption, often the cost to process LC-PUFAs into food products is restrictive. These omega-3 fatty acids, and some of the other

LC-PUFAs can be quickly oxidized leading to undesirable odors and flavors. To reduce the rate of oxidation food processors must therefore either distribute the oil in a frozen condition or encapsulate the desirable fatty acids, each greatly increasing the cost of processing and consequent cost to the consumer. Despite this increased expense, food companies are interested in supplying omega-3 fatty acids and generally healthier food oils because they believe that health conscious consumers may be willing to pay a small premium for an improved diet if a reliable supply can be developed.

[0008] Along with the movement of food companies to develop and deliver essential fats and oils as an important component in a healthy human diet, governments have begun developing regulations pushing for the adoption of PUFAs in the diet. There has been difficulty in supplying these needs, however, as there has been an inability to develop a large enough supply of omega-3 containing oil to meet growing marketplace demand.

[0009] Furthermore, as already mentioned, the omega-3 fatty acids commercially deemed to be of highest value, EPA and DHA, which are provided in marine sources, also chemically oxidize very quickly over time limiting commercial availability. Importantly, during the rapid process of EPA and DHA degradation these long chain fatty acids develop rancid and profoundly unsatisfactory sensory properties (e.g., fishy odor and taste) that make their inclusion in many foodstuffs or products difficult or impossible from a commercial acceptance perspective. As such, previous attempts to incorporate omega-3 fatty acids into spread formulations have not met with much success as they have included the addition of highly unstable EPA or DHA.

[0010] Furthermore, attempts at incorporating traditional omega-3 fatty acids such as alpha-linolenic acid (ALA) are not practical as these fatty acids are not converted to the beneficial forms efficiently enough. Nutritional studies have shown that, compared to ALA, SDA is 3 to 4 times more efficiently converted in vivo to EPA in humans (Ursin, 2003).

[0011] These limitations on supply, stability and sourcing greatly increase cost and correspondingly limit the availability of dietary omega-3 fatty acids. Accordingly, a need exists to enhance the nutritional quality and shelf-life of foodstuffs, and in particular, of spread formulations. The SDA-containing compositions of the current disclosure not only provide needed dietary fat for specific consumers, but also provide other dietary improvements for the commercial production of spread formulation.

[0012] In addition, a need exists to provide a consumer-acceptable means of delivering EPA and DHA or critical precursors in spread formulations in a commercially acceptable way. The current disclosure provides an alternative to fish or microbe-supplied omega-3 fatty acids in the form of spread formulations comprising beneficial omega-3 fatty acids and does so utilizing a comparatively chemically stable omega-3 fatty acid, SDA, as a source that offers improved cost-effective production and abundant supply as derived from transgenic plants.

SUMMARY OF THE DISCLOSURE

[0013] The present disclosure includes the incorporation of oil from transgenic plants engineered to contain significant quantities of stearidonic acid (18:4 ω 3) (SDA) for use in spread formulations to improve the fatty acid profile in the resulting formulations and/or the health of an end consumer. According to embodiments of the current disclosure, SDA-containing oils provide enhanced nutritional quality relative to traditional omega-3 alternatives such as flaxseed and lack negative taste and low stability characteristics associated with fish oil. Therefore, a preferred embodiment of this disclosure includes a spread formulation with an increased level of beneficial polyunsaturated fatty acids such as SDA.

[0014] In another embodiment of the disclosure, an oil-in-water emulsion spread formulation is provided. The spread formulation includes an oil phase and an aqueous phase. The oil phase includes a SDA-enriched oil.

[0015] In another embodiment of the disclosure, a margarine spread formulation including SDA-enriched soybean oil is provided.

[0016] Furthermore, methods of making spread formulations as described above are disclosed. These methods may include providing an oil phase including a stearidonic acid-enriched oil; providing an aqueous phase; and contacting the oil phase and the aqueous phase to make an oil-in-water emulsion spread formulation.

[0017] Exemplary stearidonic acid sources for obtaining the stearidonic acid-enriched oil may include transgenic soybeans, transgenic soybean oil, transgenic canola, transgenic canola oil, echium, and echium oil. Additional stearidonic acid sources may include seeds such as soybeans, safflower, canola, echium and corn.

[0018] In at least one embodiment, the SDA-enriched oil includes from about 10% (by weight) to about 60% (by weight) of SDA. In another embodiment, the SDA-enriched oil includes from about 10% (by weight) to about 30% (by weight) of SDA. In an even more particularly preferred embodiment, the SDA-enriched oil includes about 20% (by weight) SDA.

[0019] In at least one embodiment, the spread formulation including the SDA-enriched oil includes about 375 mg SDA-enriched oil in a 14-gram serving of the spread formulation. This amount ensures providing the end consumer with the minimum amount of SDA per day needed to enrich EPA in tissues based on James, et al. (2003). In another embodiment, the spread formulation includes about 1.875 g SDA-enriched oil in a 14-gram serving. The amount of SDA in the enriched oil may vary due to Germplasm, environmental effects, and the like. Typically, however, the SDA-enriched oil provides from about 10% (by weight) to about 60% (by weight) SDA, more preferably from about 10% (by weight) to about 30% (by weight), and even more preferably, about 20% (by weight) SDA.

[0020] Other features and advantages of this disclosure will become apparent in the following detailed description of preferred embodiments of this disclosure, taken with reference to the accompanying figures.

BRIEF DESCRIPTION OF THE FIGURE

[0021] Fig. 1 depicts one embodiment of the present disclosure for producing the spread formulation including an SDA-enriched oil.

DEFINITIONS

[0022] The following definitions are provided to aid those skilled in the art to more readily understand and appreciate the full scope of the present disclosure. Nevertheless, as indicated in the definitions provided below, the definitions provided are not intended to be exclusive, unless so indicated. Rather, they are preferred definitions, provided to focus the skilled artisan on various illustrative embodiments of the disclosure.

[0023] As used herein the term “spread formulation” refers to an oil-in-water emulsion including about 80% (by weight) fat or less. Typically, the spread formulations of the present disclosure include from about 20% (by weight) to about 80% (by weight) fat, and more suitably, from about 25% (by weight) to about 75% (by weight) fat. In some particularly preferred embodiments, the spread formulations include about 60% (by weight) fat.

[0024] As used herein the term “margarine spread formulation” refers to an oil-in-water emulsion including about 80% (by weight) fat, and would include margarines as defined in USCFR21:166.40, margarine and spread formulations blended with butter and butter.

[0025] As used herein the term “SDA-enriched oil” refers to an oil including at least about 10% (by weight) SDA.

[0026] As used herein the term “interesterified oils” refers to an oil produced by mixing small amounts of fully hydrogenated oils with liquid polyunsaturated oils.

DETAILED DESCRIPTION OF THE DISCLOSURE

Production of SDA:

[0027] The present disclosure relates to a system for an improved method for the plant based production of stearidonic acid and its incorporation into the diets of humans in an effort to improve human health. This production is made possible through the utilization of transgenic plants engineered to produce SDA in sufficiently high yield so as to allow commercial incorporation into food products. For the purposes of the current disclosure the acid and salt forms of fatty acids, for instance, butyric acid and butyrate, arachidonic acid and arachidonate, will be considered interchangeable chemical forms.

[0028] All higher plants have the ability to synthesize the main 18 carbon PUFAs, LA and ALA, and in some cases SDA (C18:4n3, SDA), but few are able to further elongate and desaturate these to produce arachidonic acid (AA), EPA or DHA. Synthesis of EPA and/or DHA in higher plants therefore requires the introduction of several genes encoding all of the biosynthetic enzymes required to convert LA into AA, or ALA into EPA and DHA. Taking into account the importance of PUFAs in human health, the successful production of PUFAs (especially the n-3 class) in transgenic oilseeds, according to the current disclosure can then provide a sustainable source of these essential fatty acids for dietary use. The “conventional” aerobic pathway which operates in most PUFA-synthesizing eukaryotic organisms, starts with $\Delta 6$ desaturation of both LA and ALA to yield γ -linolenic (GLA, 18:3n6) and SDA.

[0029] Turning to Table 1, it is important to provide a basis of what constitutes “normal” ranges of oil composition vis-à-vis the oil compositions of the current disclosure. A significant source of data used to establish basic composition criteria for edible oils and fats of major importance has been the Ministry of Agriculture, Fisheries and Food (MAFF) and the Federation of Oils, Seeds and Fats Associations (FOSFA) at the Leatherhead Food International facility in the United Kingdom.

[0030] To establish meaningful standards data, it is preferred that sufficient samples be collected from representative geographical origins and that these oils are pure. In the MAFF/FOSFA work, over 600 authentic commercial samples of vegetable oilseeds of known origin and history, generally of ten different geographical origins, were studied for each of 11 vegetable oils. The extracted oils were analyzed to determine their overall fatty acid composition ("FAC"). The FAC at the 2-position of the triglyceride, sterol and tocopherol composition, triglyceride carbon number and iodine value, protein values in the oil, melting point and solid fat content as appropriate are determined.

[0031] Prior to 1981, FAC data were not included in published standards because data of sufficient quality was not available. In 1981, standards were adopted that included FAC ranges as mandatory compositional criteria. The MAFF/FOSFA work provided the basis for later revisions to these ranges.

[0032] In general, as more data became available, it was possible to propose fatty acid ranges much narrower and consequently more specific than those adopted in 1981. Table 1 gives examples of FAC of oils that were adopted by the Codex Alimentarius Commission (CAC) in 1981 and ranges for the same oils proposed at the Codex Committee on Fats and Oils (CCFO) meeting held in 1993.

TABLE 1 - Standards For Fatty Acid Composition Of Oils (% Of Oil)

Fatty acid	Soybean oil		Groundnut oil		Cottonseed oil		Sunflower-seed oil	
	981	993	981	993	981	993	981	993
14:0	0.5	0.2	0.6	0.1	.4-2	.6-1	0.5	0.2
16:0	-14	-13.3	-16	.3-14	7-31	1.4-26.4	-10	.6-7.6
16:1	0.5	0.2	1	0.2	.5-2	-1.2	1	0.3
18:0	.4-5.5	.4-5.4	.3-6.5	.9-4.4	-4	.1-3.3	-10	.7-6.5
18:1	9-30	7.7-26.1	5-72	6.4-67.1	3-44	4.7-21.7	4-65	4-39.4
18:2	4-62	9.8-57.1	3-45	4-43	3-59	6.7-58.2	0-75	8.3-74

18:3	-11	.5-9.5	1	0.1	.1-2.1	-0.4	-0.7	-0.2
20:0	1	.1-0.6	-3	.1-1.7	-0.7	.2-0.5	-1.5	.2-0.4
20:1	1	0.3	.5-2.1	.7-1.7	-0.5	-0.1	-0.5	-0.2
22:0	0.5	.3-0.7	-5	.1-4.4	-0.5	-0.6	-1	.5-1.3
22:1		0.3	2	0.3	-0.5	-0.3	-0.5	-0.2
22:2								-0.3
24:0		0.4	.5-3	.1-2.2	-0.5	-0.1	-0.5	.2-0.3
24:1				0.3			0.5	

Sources: Codex Alimentarius Commission, 1983 and 1993.

[0033] More recently, oils from transgenic plants have been created. Some embodiments of the present disclosure may incorporate products of transgenic plants such as transgenic soybean oil. Transgenic plants and methods for creating such transgenic plants can be found in the literature. See for example, WO2005/021761A1. As shown in Table 2, the composition of the transgenic soy oil is substantially different than that of the accepted standards for soy oil.

Table 2. A comparison of transgenic soy oil and traditional soy oil fatty acid compositions (% of Oil)

	Low SDA Soy Oil	Medium SDA Soy Oil	High SDA Soy Oil
C14:0 (Myristic)	0.10	0.11	0.10
C16:0 (Palmitic)	12.23	12.33	12.52
C16:1 (Palmitoleic)	0.10	0.10	0.15
C18:0 (Stearic)	3.95	3.99	4.10
C18:1 (Oleic)	16.21	15.50	15.17
C18:2 (Linoleic)	34.04	29.40	18.46
C18:3 n6 (Gamma Linolenic)	4.30	5.50	4.71
C18:3 n3 (Alpha-Linolenic)	11.64	11.14	12.78
C18:4 n3 (Stearidonic)	14.51	18.86	28.92
C20:0 (Arachidic)	0.34	0.35	0.38
C20:1 (Eicosenoic)	0.21	0.21	0.22
C22:0 (Behenic)	0.32	0.32	0.34
C24:0 (Lignoceric)	0.10	0.09	0.09
Other fatty acids	0.56	0.60	0.69

[0034] According to embodiments of the current disclosure, the preferred plant species that could be modified to reasonably supply demand are: soybeans, canola, and echium but many other plants could also be included as needed and as scientifically practicable. For the present disclosure, the preferred source of SDA is transgenic soybeans which have been engineered to produce high levels of SDA. The soybeans may be processed at an oil processing facility and oil may be extracted consistent with the methods described in US Patent Applications 2006/0111578A1, 2006/0110521A1, and 2006/0111254A1.

[0035] It should be recognized that once produced, the SDA of the disclosure can be used to improve the health characteristics of a great variety of spread formulations. This production offers a sustainable crop-based source of omega-3 fatty acids that enriches EPA in red blood cells and other tissues, and has

improved flavor and stability as compared to many alternative omega-3 fatty acid sources available today.

Oil Phase:

[0036] As noted above, the spread formulations of the present disclosure include an oil phase and an aqueous phase. In one embodiment, in addition to the SDA-enriched oil, the oil phase further include oils such as hydrogenated oils, partially hydrogenated oils, and interesterified oils. Exemplary of these additional oils include partially hydrogenated oils having a solids fat index of from about 19% to 25.5% at 50°F (10°C), from about 10.5% to about 15.5% at 70°F (21°C), and from about 0.5% to about 4.0% at 92°F (33°C). Hydrogenated oils having this solids fat index will provide the spread formulation with the desired plastic texture. Furthermore, these oils will allow the spread formulations to adequately melt on food products and in the mouth, such as is desired of the spread formulations. Commercial hydrogenated and partially hydrogenated oils typically used in spread formulations are available from ADM (Decatur, Illinois). Specifically, one particularly preferred partially hydrogenated oil is Product No. 86-334-0, available from ADM (Decatur, Illinois). Other commercially available hydrogenated and partially hydrogenated oils can be obtained, for example, from Cargill (Minneapolis, Minnesota), Bunge (St. Louis, Missouri), CHS (Inver Grove Heights, Minnesota), AGP (Omaha, Nebraska), and Perdue (Salisbury, Maryland). Interesterified oils are commercially available from ADM (Decatur, Illinois),

[0037] Typically, when used, the oil phase includes these additional oils in amounts of from about 20% (by weight) to about 80% (by weight). In one particularly preferred embodiment, the oil phase includes these additional oils in an amount of about 58% (by weight).

[0038] In addition to the oils mentioned above, in some embodiments, the oil phase further includes a liquid oil such as soybean oil, canola oil, rapeseed oil, palm, oil, and the like, and combinations thereof. Typically, these oils are refined, bleached and deodorized. These liquid oils provide improved flavor

to the spread formulation. Liquid oils, such as soybean oil, further provide for improved texture and spreadability to the spread formulations. Furthermore, some liquid oils, such as palm oil provide a non-trans fat option to the spread formulations, thereby providing health benefits to the consumer along with improved flavor.

[0039] Typically, when used, the oil phase includes these additional liquid oils in amounts of from about 20% (by weight) to about 80% (by weight). In one particular embodiment, the oil phase includes the liquid oils in an amount of about 20% (by weight).

[0040] Other particularly preferred liquid oils that may be used in the oil phase of the spread formulation to stabilize the spread formulation include high stability oils. These oils can replace the conventional liquid oils described above to further slow down oxidation and off flavor development due to the polyunsaturated fat content in the omega-3 oils. Exemplary suitable high stability oils include low linolenic soybean oils, high oleic soybean oils, high oleic/low saturate soybean oils, high oleic canola oils, sunflower oils, and the like, and combinations thereof.

[0041] Typically, when used, the oil phase includes these high stability liquid oils in amounts of from about 0.1% (by weight) to about 35% (by weight). In one particular embodiment, the oil phase includes at least one high stability oil in an amount of about 19% (by weight).

[0042] Apart from the above fat blend of oils, the oil phase of the spread formulation may include minor fat-soluble ingredients such as emulsifiers, lecithin, flavoring agents, coloring agents, and combinations thereof.

[0043] Exemplary emulsifiers that can be included in the oil phase include monoglycerides and diglycerides, which can disperse the water particles in the oil-in-water emulsion spread formulation and prevent water spattering. Additionally, the monoglycerides and diglycerides can stabilize the emulsion spread formulation. Exemplary monoglycerides and diglycerides include those commercially available from Eastman Chemical Company (Kingsport, Tennessee) and Danisco (Copenhagen,

Denmark). Particularly suitable monoglycerides are Dimodan Distilled monoglycerides, commercially available from Danisco (Copenhagen, Denmark).

[0044] Typically, when used, the oil phase includes monoglycerides and diglycerides in amounts of from about 0.1% (by weight) to about 0.6% (by weight). In one particular embodiment, the oil phase includes monoglycerides and diglycerides in an amount of about 0.2% (by weight).

[0045] Lecithin may also be included in the oil phase to provide improved stability of the emulsion spread formulation. Additionally, it has been found that the inclusion of lecithin may aid in release of the product in frying applications.

[0046] Typically, when used, the oil phase includes lecithin in amounts of from about 0.1% (by weight) to about 0.2% (by weight). In one particular embodiment, the oil phase includes lecithin in an amount of about 0.2% (by weight).

[0047] Coloring agents may include any coloring agents known in the food processing agent. The coloring agents provide aesthetic value to the spread formulation. For example, when the spread formulation is a margarine spread formulation, beta carotene can be added to the oil phase of the spread formulation to provide adequate orange-yellow coloring.

[0048] Additionally, beta carotene, as with many of the other coloring agents, serves multiple functions in the spread formulations. More particularly, beta carotene can provide activity as Vitamin A in addition to behaving as a coloring agent. Fortification of all margarine spread formulations is mandatory under FDA guidelines. This mandatory Vitamin A level is typically attained by the addition of beta-carotene into the margarine spread formulation, which can be added as a vitamin blend, such as with Vitamin D.

[0049] Typically, when used, the oil phase includes one or more coloring agents in amounts of from about 0.001% (by weight) to about 0.3% (by weight). In one embodiment, the oil phase includes a coloring agent in an amount of

about 0.002% (by weight). When beta-carotene is the coloring agent, and is further added to meet Vitamin A requirements, the level of beta-carotene is determined by the other components in the formulation and the required amounts of Vitamin A in the final spread formulation.

[0050] In many spread formulations provided in the present disclosure, it is desirable to enhance the flavoring. Particularly, when the spread formulation is a margarine spread formulation, it is desirable to include an artificial butter flavoring agent in the oil phase. It should be recognized by one skilled in the art, however, that any suitable flavoring agent known in the art may be used.

[0051] Typically, when used, the oil phase includes one or more flavoring agents in amounts of from about 0.1% (by weight) to about 0.6% (by weight). In one particular embodiment, the oil phase includes a flavoring agent in an amount of about 0.2% (by weight).

[0052] In at least one embodiment, a lower fat content is desirable. Specifically, in recent years, the consumption of reduced and low fat products has increased and intensive research has been made in processing and ingredients in order to achieve better low fat products. In such an embodiment, the oil phase of the spread formulation may further include thickening agents such as a starch and/or a hydrocolloid to be used as fat replacements. One particularly preferred fat replacement is gelatin. Other suitable thickening agents include pectin, carrageenans, agar, Xanthan gum, starch alginates, methocellulose derivatives and combinations thereof.

Aqueous Phase:

[0053] In addition to the oil phase, the oil-in-water spread formulations of the present disclosure include an aqueous phase. Typically, at least about 95% (by weight) of the aqueous phase is water. In addition to the water, the aqueous phase may include one or more of salt or brine, dairy protein, antioxidants, and preservatives.

[0054] When used, salts, such as sodium chloride and potassium chloride, are typically included in the aqueous phase in amounts of from about 1.5% (by weight) to about 3.0% (by weight). In one particular embodiment, the aqueous phase includes salt in an amount of about 1.5% (by weight) to behave as both a flavoring agent and a preservative.

[0055] Other preservatives that may be included in the aqueous phase include antimicrobial preservatives, antioxidants, and metal scavengers. Common antimicrobial preservatives include benzoic acid, sorbic acid, sodium benzoate and potassium sorbate.

[0056] When included, antimicrobial preservatives are typically present in the aqueous phase in amounts of from about 0.1% (by weight) to about 0.2% (by weight)

[0057] Exemplary antioxidants that will further improve stability of the fatty acids within the formulations, include ethylenediaminetetraacetic acid (EDTA), tocopherols (Vitamin E), ascorbic acid (Vitamin C), Vitamin C salts (e.g., L-sodium, L-calcium ascorbate), Vitamin C esters (e.g., ascorbyl-5,6-diacetate, ascorbyl-6-palmitate), ethoxquin, citric acid, calcium citrate, butylated hydroxyl anisole (BHA), butylated hydroxytoluene (BHT), tertiary butyl hydroquinone (TBHQ), natural antioxidants (e.g., rosemary extract), and the like, and combinations thereof. EDTA further acts as an antioxidant synergist, which performs two functions: (1) it increases the antioxidant effectiveness; and (2) it ties up or chelates the trace metals, which are oxidative catalysts. EDTA is also effective as an agent to retard oxidative bleaching of the carotenoid coloring agents used in the oil phase as described above.

[0058] Amounts of antioxidants to be added to the formulations will typically depend on the antioxidant to be added, and further, on the other components in the spread formulation. Exemplary amounts of antioxidants to be added include from about 1 ppm to about 200 ppm. More preferably, antioxidants can be added in amounts of from about 10 ppm to about 150 ppm, and even more preferably, from

about 10 ppm to about 50 ppm. In one particularly preferred embodiment, the antioxidant is EDTA and the formulation includes about 100 ppm.

[0059] Dairy proteins may be included in the aqueous phase to provide improved nutritional value to the spread formulations. Exemplary dairy proteins for use in the aqueous phase may include whole milk, non fat dry milk, sodium caseinates, whey, and combinations thereof.

[0060] When used, dairy proteins are included in the aqueous phase in amounts of from about 1% (by weight) to about 10% (by weight). In one particular embodiment, the aqueous phase includes at least one dairy protein in an amount of about 1.2% (by weight).

Methods of Producing Spread Formulations:

[0061] Additionally, the present disclosure is directed to methods of making the spread formulations including SDA. Generally, the spread formulations of the present disclosure are produced by: providing an oil phase comprising a stearidonic acid (SDA)-enriched oil; providing an aqueous phase; and contacting the oil phase and the aqueous phase to make an oil-in-water emulsion spread formulation.

[0062] To prepare the oil phase for the spread formulation, the various oils and fats or fat blends may be transferred to an emulsion tank for blending together. Specifically, as shown in Figure 1 at 1a, the oils and fats are transferred to the tank where the oils and fats are melted and blended. Typically, it is desirable to add the highest melting fats first, followed by the lower melting fats and liquid oils.

[0063] Once the oils and fats have been added, emulsifiers and other oil-soluble minor ingredients as described above (e.g., monoglycerides, diglycerides, coloring agents, flavoring agents, etc.) are added to the blend.

[0064] Typically, the oils and fats (and any other additional ingredients) are blended at a temperature approximately 5°C to 8°C higher than the melting point of the oil phase. More particularly, the oils and fats are blended at a temperature of from about 105°F (41°C) to about 110°F (43°C).

[0065] Once blended together, the oil phase is kept at stable storage temperature above the melting point of the fat and under agitation in order to avoid fractionation of the fat and oils and to allow easy handling.

[0066] The aqueous phase is often prepared batch-wise by mixing all ingredients in the aqueous phase in an aqueous phase tank (as shown in Figure 1 at 1b). The water in the aqueous phase should be of good drinking quality. If drinking quality water cannot be guaranteed, the water can be subjected to pre-treatment by means of e.g., a UV or filter system. In one embodiment, the aqueous phase is mixed and further pasteurized at a temperature of greater than about 150°F (66°C) for a time period of about 30 minutes.

[0067] The aqueous phase is then added to the oil phase and the oil-in-water emulsion is created under intensive but controlled mixing (see Figure 1 at 1c). In one embodiment, the emulsion spread formulation is produced and then held at a temperature of from about 105°F (41°C) to about 110°F (43°C) for a time period of from about 1 hour to about 2 hours. By holding the emulsion spread formulation at these temperatures, the spread formulation is pasteurized. Alternatively, the formulation can be pasteurized using a plate heat exchanger (PHE), as described below.

[0068] In another embodiment, as shown in Figure 1 at 3, the emulsion spread formulation is continuously pumped through either a PHE or a low pressure scraped surface heat exchanger (SSHE), or a high pressure SSHE to be pasteurized. Specifically, the emulsion is heated to a temperature of from about 167°F (75°C) to about 176°F (80°C) for 15 to 20 seconds and then cooled to a temperature of from about 113°F (45°C) to about 122°F (50°C) or 5°C to 8°C higher than the melting point of the oil phase in the emulsion.

[0069] For full fat products, a PHE is typically used for pasteurization. For lower fat versions where the emulsion is expected to exhibit a relatively high viscosity and for heat-sensible emulsions (e.g., emulsions with high protein content) a low pressure SSHE or high pressure SSHE is recommended.

[0070] The pasteurization process has several advantages. It ensures inhibition of bacterial growth and growth of other micro-organisms, thus improving the microbiological stability of the emulsion. Pasteurization of the emulsion will minimize the residence time from pasteurized product to filling or packing of the final product.

[0071] Furthermore pasteurization of the complete emulsion spread formulation ensures that the emulsion is fed to a crystallization line, described below, at a constant temperature achieving constant processing parameters, product temperatures and product texture. In addition, occurrence of pre-crystallized emulsion fed to the crystallization equipment is prevented when the emulsion is properly pasteurized and fed to the high pressure pump at a temperature approximately 5°C to 10°C higher than the melting point of the oil phase.

[0072] A typical pasteurization process will, after preparation of the emulsion at a temperature of from about 105°F (41°C) to about 110°F (43°C), include a heating and holding sequence of the emulsion at from about 167°F (75°C) to about 185°F (85°C) for 16 seconds and subsequently a cooling process to a temperature of from about 113°F (45°C) to about 131°F (55°C). The end temperature will depend on the melting point of the oil phase: the higher the melting point, the higher the temperature.

[0073] Referring to Figure 1, the emulsion spread formulation is further pumped to a crystallization line 2, typically by means of a high pressure piston pump (HPP) 101. The crystallization line 2 for the production of spread formulations such as margarine spread formulations typically consists of a high pressure SSHE 4 which is cooled by ammonia or Freon type cooling media (not shown). Pin mixers and/or intermediate crystallizers (not shown) are often included in the line in order to

add extra kneading intensity and time for the production of plastic products. A resting tube 7 is the final step of the crystallization line 2 and is only included if the end product is packaged.

[0074] The high pressure SSHE 4 super-cools and crystallizes the warm emulsion spread formulation on the inner surface of the chilling tube. The emulsion is efficiently scraped off by the rotating knives, thus the emulsion is chilled and kneaded simultaneously. When the fats in the emulsion crystallize, the fat crystals form a three-dimensional network entrapping droplets of the aqueous phase and the liquid oil of the oil phase, resulting in products with properties of a plastic semi-solid nature.

[0075] Depending on the type of spread formulation to be manufactured and the type of fats used for the particular formulation, the configuration of the crystallization line 2 (i.e., the order of the chilling tubes and the pin mixer) can be adjusted to provide the optimum configuration for the particular formulation.

[0076] After the formulation is chilled in the SSHE 4, it enters the pin mixer and/or intermediate crystallizers in which it is kneaded for a certain period of time and with a certain intensity in order to assist the promotion of the three-dimensional network, which on the macroscopic level is the plastic structure. If the formulation is meant to be distributed as a wrapped formulation, it will enter the SSHE 4 again before it settles in the resting tube 7 prior to wrapping. If the formulation is filled into cups, no resting tube is included in the crystallization line.

[0077] In one embodiment, the warm emulsion spread formulation is pumped into a SSHE to cool the spread formulation to a temperature of from about 34°F (1°C) to about 41°F (5°C) and then pumped into a pin mixer for kneading.

[0078] Various packaging and filling machines are available on the market and will not be described herein. However, the consistency of the formulation is very different if it is produced to be packaged or filled into cups or wrapped. If the

formulation is packaged, it must exhibit a firmer texture than a filled formulation, and if this texture is not optimal, the formulation will be diverted to the remelting system (*see* Figure 1 at 5), melted and added to the buffer tank 6 for re-processing. Different remelting systems are available, but the most common systems used are PHE or low pressure SSHE.

[0079] Surprisingly, the inventors have found that including SDA compositions from transgenic plant sources in spread formulations as described above is highly effective in increasing the omega-3 fatty acid levels of SDA (18:4) and EPA (eicosapentaenoic acid). Furthermore, plant sources, such as soybean oil, have been found to provide more stable fatty acids to the formulations. Specifically, SDA soybean oil was shown to take 5 to 10 times longer to oxidize as measured by peroxide values and anisidine values as compared to fish oils in stability tests.

[0080] Furthermore, there has been found to be little difference in the palatability, flavor, texture, or overall consumer acceptability, of the spread formulation including SDA as compared to conventional spread formulations without omega-3 fatty acids. Specifically, as shown in the Example below, SDA-containing spread formulations at 2 weeks had similar flavor, aroma, appearance, mouthfeel, and spreadability as compared to conventional spread formulations without omega-3 fatty acids. At 2 months, the differences perceived between the SDA-containing formulation and the control formulation were in appearance, but, again, no differences in flavor, aroma, mouthfeel, or spreadability. Differences in flavor and aftertaste were not perceived between the SDA-containing formulation and control formulation until 4 months, however aroma, mouthfeel, and spreadability remained similar.

[0081] Furthermore, as compared to spread formulations including alternative omega-3 fatty acids, such as spread formulations using fish oils or algal oils, the shelf life of the spread formulation including SDA after nine months was similar to a conventional spread formulation without omega-3 fatty acids. Similar results were obtained for off flavor aftertaste where the spread formulation with SDA

was less different from conventional spread formulations as compared to spread formulations using alternate sources of omega-3 fatty acids.

ILLUSTRATIVE EMBODIMENTS OF THE DISCLOSURE

[0082] The following example is included to demonstrate general embodiments of the disclosure. It should be appreciated by those of skill in the art that the techniques disclosed in the example which follows represent techniques discovered by the inventors to function well in the practice of the disclosure, and thus can be considered to constitute preferred modes for its practice. However, those of skill in the art should, in light of the present disclosure, appreciate that many changes can be made in the specific embodiments which are disclosed and still obtain a like or similar result without departing from the disclosure.

[0083] All of the compositions and methods disclosed and claimed herein can be made and executed without undue experimentation in light of the present disclosure. While the compositions and methods of this disclosure have been described in terms of preferred embodiments, it will be apparent to those of skill in the art that variations may be applied without departing from the concept and scope of the disclosure.

[0084] In the examples below, transgenic soybean oil containing SDA was used. Similar results would be obtained when using oil derived from other transgenic plants such as corn or canola.

Example 1: Margarine Spread Formulations – A 9-month Study

[0085] A 9-month study was conducted to determine whether a margarine spread formulation containing SDA had an equivalent sensory shelf life as compared to a control margarine spread formulation (i.e., conventional margarine spread formulations without SDA) and to other spread formulations using alternative omega-3 fatty acids.

[0086] The spread formulations analyzed are shown in Table 3.

Table 3. Spread Formulations

Ingredient	Control Formulation (wt. %)	Spread Formulation with 20% (by weight) SDA (wt. %)	WT. % in Phase of Spread Formulation with SDA	Spread Formulation with 4.64% (by weight) Menhaden Oil (wt. %)	Spread Formulation with 3.07% (by weight) Meg-3 Oil (wt. %)	Spread Formulation with 2.65% (by weight) Algal Oil (wt. %)
Oil Phase						
Liquid Soybean Oil (ADM)	25.0000	11.6100	19.18165	20.3600	21.9300	22.3500
Margarine Soybean Oil (ADM)	35.0000	35.0000	57.82582	35.0000	35.0000	35.0000
SDA-enriched soybean oil (Monsanto)	0	13.3900	22.1225	0	0	0
Menhaden-enriched soybean oil (Monsanto)	0	0	0	4.6400	0	0
Meg-3-enriched soybean oil (Monsanto)	0	0	0	0	3.0700	0
Algal-enriched soybean Oil (Monsanto)	0	0	0	0	0	2.6500
Lecithin	0.2000	0.2000	0.330433	0.2000	0.2000	0.2000

(Solae Co.)						
Mono & Di-Glycerides (Danisco)	0.2000	0.2000	0.330433	0.2000	0.2000	0.2000
Coloring Agent	0.0016	0.0016	0.002643	0.0016	0.0016	0.0016
Flavoring Agent (Givaudan)	0.1250	0.1250	0.206521	0.1250	0.1250	0.1250
Aqueous Phase						
Water	37.4634	37.4634	94.90796	37.4634	37.4634	37.4634
Salt (Morton)	1.5000	1.5000	3.800027	1.5000	1.5000	1.5000
Non Fat Dry Milk (Land O'Lakes)	0.5000	0.5000	1.266676	0.5000	0.5000	0.5000
EDTA (Brenntag North America)	0.0100	0.0100	0.025334	0.0100	0.0100	0.0100
Total	100.0000	100.0000		100.0000	100.0000	100.0000

[087] The spread formulation samples were stored at a temperature of from about 38°F (3.33°C) to about 42°F (5.56°C) throughout the duration of the study. Two-ounce samples were then submitted for sensory analysis.

[088] A panel of trained assessors (9) participated in discussion and training sessions to identify and define key descriptive attributes that discriminated well between the formulations. In subsequent rating sessions the panel used Quantitative Descriptive Analysis (Tragon Corp., Redwood Shores, California), with verbal anchors to rate the perceived intensity of each attribute. Each panelist assessed

one replicate of each sample at five time points (e.g., 2 weeks, 2 mos., 4 mos., 6 mos., and 9 mos.) over a period of nine months. Plain crackers and mineral water were used as palate cleansers between samples. Samples were tasted and chewed, and then spat out rather than being swallowed. The aftertaste of samples was determined five seconds after the samples had been removed from the mouth.

[089] The sensory attributes and definitions produced for the formulations were:

Appearance

Color The intensity of color from pale yellow to darker yellow (light-dark).
(evaluated without stirring)

Press sample down with spoon a couple of times and evaluate:

Firm The degree to which sample feels firm and dense (soft-dense).

Glossy The degree to which the sample appears glossy, shiny or has a sheen after being mashed with a spoon (slight-very).

Smooth The degree to which the samples appears smooth, not grainy (grainy-smooth).

Whipped The degree to which sample appears whipped with air, fluffy, not dense, not heavy (slight-very).

Aroma (lift lid and evaluate immediately)

Overall Intensity The intensity of overall aroma, regardless of type (weak-strong).

Artificial Butter The intensity of artificial butter aroma as in theater popcorn, microwave popcorn imitation butter (weak-strong).

Oily	The amount of oily aroma like vegetable oil, greasy or lardy (weak-strong).
Milk Dairy Sweet	The intensity of milky, dairy, sweet dairy aroma like the sweet aroma found in butter or dairy cream (weak-strong).
Chemical	The intensity of chemical aroma (weak-strong).
Dairy Sour	The intensity of dairy sour aroma like sour cream, yogurt, buttermilk (weak-strong).
Fruity (Artificial)	The intensity of fruity aroma like an artificial pineapple (weak-strong).
Sweet (Artificial)	The intensity of sweet aroma like artificial vanilla (weak-strong).

Mouthfeel

Melt Rate	The rate at which formulation melts in your mouth or dissolves (slow-fast).
Inconsistent Texture	The degree to which formulation feels lumpy like it contains teeny tiny bumps or lumps that go away right away (butterlumps) (slight-very).
Greasy/Oily	The degree to which formulation feels greasy or oily (watery-greasy).

Flavor

Fruity (Artificial)	The intensity of fruity flavor like an artificial pineapple flavor (weak-strong).
Salty	The intensity of salty flavor (weak-strong).

Oily	The amount of oil flavor like vegetable oil (weak-strong).
Artificial Butter	The intensity of artificial butter flavor as in theater popcorn, microwave popcorn imitation butter (weak-strong).
Dairy Sour	The intensity of dairy sour flavor, like buttermilk, sour cream, or yogurt (weak-strong).
Real Butter	The intensity of butter flavor like that of real butter (weak-strong).
Milky Dairy Sweet	The intensity of milky, dairy, sweet dairy flavor like the sweet flavor found in butter or dairy cream (weak-strong).
Chemical	The intensity of chemical flavor, not natural (weak-strong).
Bitter	The intensity of bitter flavor (weak-strong).
Old	The intensity of old flavor, not fresh, has been in refrigerator too long (weak-strong).
Off Flavor	The flavor typically not found in spread formulations that provides a negative sensory perception.
<u>Aftertaste</u> (5 seconds after removing formulation from mouth)	
Salty	The amount of salty flavor remaining (weak-strong).
Dairy Sour	The amount of dairy sour flavor remaining, like buttermilk, yogurt, or sour cream (weak-strong).
Milky Dairy Sweet	The amount of milky, dairy, or sweet dairy flavor remaining, like the sweet flavor found in butter or dairy cream (weak-strong).

Real Butter	The amount of butter flavor remaining, like that of real butter (weak-strong).
Oily Aftertaste	The amount of oil flavor remaining, like vegetable oil, greasy or lardy taste (weak-strong).
Old	The amount of old flavor remaining (weak-strong).
Bitter	The amount of bitter flavor remaining (weak-strong).

Afterfeel

Astringent Mouthfeel	The degree to which the formulation leaves your tongue feeling astringent, drying, dries the tongue as from too much salt on your tongue (slight-very).
Oily Mouthfeel	The degree to which the formulation leaves an oily, greasy coating remaining in the mouth and lips (slight-very).

Spreading onto Bread (spread with serrated side of a knife 7 times in all different directions)

Spreadable	The degree to which formulation appears spreadable and spreads easily from doesn't spread, to hard to spread, to spreads easily (slight-very).
Rips Bread	The degree to which the formulation rips or tears the bread while spreading, puts holds in the bread (slight-very).

Appearance on Bread

Shiny	The degree to which the formulation appears shiny from a dull matte appearance with no shine to high gloss (slight-very).
Color on Bread	The degree of color from pale to dark yellow (light-dark).

[090] The results of the sensory analysis are summarized in Tables 4-8. Differences that were perceived in the SDA-containing formulation as compared to the control formulation at 2 weeks were associated with the salty aftertaste and appearance on bread of the formulation, but not its flavor, aroma, appearance, mouthfeel, or spreadability. At 2 months, the differences perceived between the SDA-containing formulation and the control formulation were in appearance, but, again, no differences in flavor, aroma, mouthfeel, or spreadability. Differences in flavor and aftertaste were not perceived between the SDA-containing formulation and control formulation until 4 months, however aroma, mouthfeel, and spreadability remained similar. At both 6 and 9 months, the SDA-containing formulation was perceived to have a difference in flavor and appearance on bread.

Table 4. Effect of omega-3 enriched oil (spread formulation with omega-3 fatty acids) and conventional soybean oil (control spread formulation) on the sensory attributes of margarine spread formulations at 2 weeks.

	Spread Formulation				
Attributes	Control Spread Formulation	Spread Formulation with SDA	Spread Formulation with Menhadin Oil	Spread Formulation with Meg-3 Oil	Spread Formulation with Algal Oil
Appearance Attributes					
Color	12.042abc	13.25ab	10.88d	12.74abc	11.55cd
Firm	26.02a	25.92a	21.57c	23.71abc	24.83abc
Glossy	33.38abc	30.33cd	32.18bc	28.48de	33.73abc
Smooth	45.41ab	43.55b	44.57ab	44.72ab	45.65ab
Whipped	25.76	25.75	29.19	27.57	28.62
Aroma Attributes					
Overall Intensity	15.12	14.63	15.88	15.30	16.44

Artificial Butter	8.52	8.30	8.22	8.63	8.45
Oily	9.68	9.84	8.72	8.90	10.01
Milk Dairy Sweet	6.88	7.32	6.53	6.68	7.93
Chemical	3.78	4.04	3.33	3.85	4.12
Dairy Sour	4.79b	5.40b	5.54ab	5.10b	6.35a
Fruity (Artificial)	6.31	6.09	6.92	6.16	7.11
Sweet (Artificial)	4.78abc	4.48bc	4.58bc	3.96c	4.68abc
Mouthfeel Attributes					
Melt Rate	34.26b	35.82ab	36.13ab	36.48ab	34.34b
Inconsistent Texture	2.36	2.20	2.22	2.04	2.06
Greasy/Oily	19.32	18.85	18.52	18.49	19.26
Flavor Attributes					
Fruity (Artificial)	11.51	12.95	11.96	12.55	12.54
Salty	22.23	24.78	24.28	25.05	23.71
Oily	15.29bc	15.60abc	17.38a	14.93c	17.32ab
Artificial Butter	13.82	14.70	14.52	15.24	14.34
Dairy Sour	8.06	7.98	8.98	8.46	8.38
Real Butter	7.18	6.68	5.98	7.34	6.44
Milky Dairy Sweet	11.65	10.52	9.91	11.62	10.86

Chemical	5.32b	5.08b	8.18a	5.22b	7.13a
Bitter	3.16c	3.81bc	4.69a	3.63bc	4.13ab
Old	1.78	1.72	2.08	1.71	1.95
Aftertaste Attributes					
Salty	17.98b	21.07a	19.66ab	19.60ab	19.57ab
Dairy Sour	7.50	7.95	8.45	7.96	8.07
Milky Dairy Sweet	10.42a	9.74ab	8.65b	10.44a	9.48ab
Real Butter	7.36a	6.85abc	6.08c	7.28ab	6.25bc
Oily Aftertaste	12.78abcd	13.68abc	14.10ab	11.20d	14.53a
Old	1.84	2.02	2.23	2.02	2.01
Bitter	3.51	3.57	4.05	3.49	3.88
Afterfeel Attributes					
Astringent	13.95	14.71	15.53	15.62	15.12
Oily Mouthfeel	13.47	13.12	13.07	12.35	13.19
Spreading onto Bread Attributes					
Spreadable	51.98	51.62	51.60	51.05	51.75
Appearance on Bread Attributes					
Shiny	40.88a	38.22bc	42.08a	40.52ab	40.36ab
Color on Bread	13.98bcd	15.16ab	12.47e	13.70cde	13.42cde

Table 5. Effect of omega-3 enriched oil (spread formulation with omega-3 fatty acids) and conventional soybean oil (control spread formulation) on the sensory attributes of margarine spread formulations at 2 months.

	Spread Formulation				
Attributes	Control Spread Formulation	Spread Formulation with SDA	Spread Formulation with Menhadin Oil	Spread Formulation with Meg-3 Oil	Spread Formulation with Algal Oil
Appearance Attributes					
Color	11.02bc	12.28a	10.53bc	11.49ab	10.34c
Firm	22.61ab	24.81a	19.39b	22.99ab	24.77a
Glossy	28.56a	24.17c	28.01ab	24.86bc	27.23abc
Smooth	44.81	44.59	44.40	43.25	43.07
Whipped	24.19	23.35	24.01	24.28	22.15
Aroma Attributes					
Overall Intensity	15.54abc	14.08bc	15.65abc	16.07abc	15.06bc
Artificial Butter	10.30	10.01	10.61	11.27	10.63
Oily	7.78	6.77	7.01	7.40	7.21
Milk Dairy Sweet	7.57	6.51	7.21	6.60	6.54
Chemical	3.96	3.91	4.06	4.59	3.96
Dairy Sour	4.69	4.75	5.40	4.29	4.97
Fruity (Artificial)	8.18	7.87	8.41	8.26	7.98
Sweet (Artificial)	5.24	4.99	5.57	5.29	4.92

Mouthfeel Attributes					
Melt Rate	35.95	36.66	37.35	36.91	36.93
Inconsistent Texture	2.81	1.96	1.81	1.76	2.15
Greasy/Oily	15.66	16.39	16.62	16.77	17.04
Flavor Attributes					
Fruity (Artificial)	13.10	11.30	12.50	12.50	10.89
Salty	20.69	21.17	21.02	21.11	20.22
Oily	13.14	14.02	13.71	13.99	14.61
Artificial Butter	7.14	6.72	15.19	15.34	16.02
Dairy Sour	7.49	7.66	8.44	8.04	7.46
Real Butter	7.14ab	6.72abc	6.86abc	7.51a	6.01bc
Milky Dairy Sweet	10.54a	9.52abc	10.09ab	10.22ab	8.69bcd
Chemical	4.16d	5.12abcd	5.33abcd	4.69bcd	4.91bcd
Bitter	2.89b	3.29ab	3.69ab	3.66ab	3.71ab
Old	2.17	2.76	2.97	2.56	2.59
Off Flavor	6.10d	6.69cd	8.13bcd	7.65bcd	10.14abc
Aftertaste Attributes					
Salty	17.24	17.16	18.94	17.92	16.93
Dairy Sour	7.03	7.05	7.20	7.40	6.85
Milky Dairy Sweet	8.86ab	8.72abc	8.16abc	9.14a	7.81bc

Real Butter	7.19a	6.85ab	5.85abc	7.04ab	5.87abc
Oily Aftertaste	10.44ab	11.51ab	11.70a	12.06a	11.94a
Old	2.34	2.72	2.45	3.00	2.83
Off Flavor	5.91cd	5.41cd	6.31cd	6.74bcd	8.89abc
Bitter	2.84bc	2.89bc	3.56ab	3.46abc	3.56ab
Afterfeel Attributes					
Astringent	11.87	12.15	12.91	12.96	13.76
Oily Mouthfeel	10.36	10.76	10.25	10.54	9.83
Spreading onto Bread Attributes					
Spreadable	50.31	50.11	49.64	50.31	50.03
Appearance on Bread Attributes					
Shiny	41.77ab	38.43cd	43.24a	40.17bc	42.01ab
Color on Bread	12.97bc	14.19ab	12.81c	14.44a	12.57c

Table 6. Effect of omega-3 enriched oil (spread formulation with omega-3 fatty acids) and conventional soybean oil (control spread formulation) on the sensory attributes of margarine spread formulations at 4 months.

	Spread Formulation				
Attributes	Control Spread Formulation	Spread Formulation with SDA	Spread Formulation with Menhadin Oil	Spread Formulation with Meg-3 Oil	Spread Formulation with Algal Oil
Appearance Attributes					
Color	12.24ab	12.10abc	10.12ef	11.51bcd	10.91cde
Firm	28.67	28.19	22.97	26.96	26.17
Glossy	26.76ab	21.77bcd	26.33ab	25.92abc	26.59ab
Smooth	46.18a	44.83ab	44.96ab	44.46ab	44.87ab
Whipped	28.16	26.81	28.77	27.46	28.11
Aroma Attributes					
Overall Intensity	17.83	16.68	17.65	16.09	17.81
Artificial Butter	10.32	10.25	9.52	8.73	9.77
Oily	10.22	9.70	9.82	9.19	9.69
Milk Dairy Sweet	6.52	7.08	6.79	6.89	6.78
Chemical	3.58ab	3.81ab	4.11ab	3.17b	4.02ab
Dairy Sour	4.57	5.45	4.82	4.31	4.68
Fruity (Artificial)	7.77	9.24	8.97	7.71	8.25
Sweet (Artificial)	5.82ab	7.43a	5.47b	6.21ab	5.84ab

Mouthfeel Attributes					
Melt Rate	36.99	35.99	36.42	39.72	38.22
Inconsistent Texture	4.81	5.15	4.12	4.51	4.66
Greasy/Oily	21.74	22.16	22.37	23.52	22.48
Flavor Attributes					
Fruity (Artificial)	16.36	15.77	16.07	14.84	15.08
Salty	25.48abcd	25.90abcde	26.00abc	23.33d	25.84abc
Oily	17.65de	19.22abcde	18.02cde	19.77abc	18.52bcde
Artificial Butter	16.53a	15.88a	14.68ab	13.22b	15.03ab
Dairy Sour	9.19	8.83	8.80	8.12	8.08
Real Butter	7.32a	6.00bcd	6.37abc	5.45cd	5.51cd
Milky Dairy Sweet	10.65a	9.28abc	9.33abc	8.15c	8.49c
Chemical	4.53c	5.70bc	6.10bc	6.59ab	6.11bc
Bitter	2.51b	3.85a	3.74a	3.92a	3.83a
Old	1.93b	2.00b	2.73ab	2.58ab	2.33b
Off Flavor	4.74c	8.11abc	8.00abc	9.65ab	10.92a
Aftertaste Attributes					
Salty	20.43	21.76	21.09	21.12	21.73
Dairy Sour	8.24ab	7.59ab	7.79ab	8.48ab	7.11b
Milky Dairy Sweet	9.62a	8.98abc	8.78abcd	8.28abcd	7.84bcd

Real Butter	7.38a	5.62cd	6.29bc	5.49cd	5.80bcd
Oily Aftertaste	14.27b	16.58a	15.90ab	16.32a	16.03ab
Old	2.25	2.12	2.54	2.62	2.32
Off Flavor	4.49b	6.57ab	6.98ab	8.45ab	9.76a
Bitter	2.95	3.49	2.99	3.37	3.48
Afterfeel Attributes					
Astringent	13.77bc	15.98ab	14.49abc	13.81bc	12.28c
Oily Mouthfeel	11.88	13.38	14.28	13.52	13.05
Spreading onto Bread Attributes					
Spreadable	50.78	51.36	51.39	51.41	51.19
Appearance on Bread Attributes					
Shiny	40.67ab	37.42bc	40.76ab	39.38abc	40.32abc
Color on Bread	12.39abc	12.83ab	11.22cd	12.06bcd	11.12cd

Table 7. Effect of omega-3 enriched oil (spread formulation with omega-3 fatty acids) and conventional soybean oil (control spread formulation) on the sensory attributes of margarine spread formulations at 6 months.

	Spread Formulation				
Attributes	Control Spread Formulation	Spread Formulation with SDA	Spread Formulation with Menhadin Oil	Spread Formulation with Meg-3 Oil	Spread Formulation with Algal Oil
Appearance Attributes					
Color	12.84abc	13.59a	12.12bcd	12.95abc	12.62abcd
Firm	24.26	25.47	23.26	23.04	24.45
Glossy	23.80cd	22.53cde	25.58bc	21.55de	25.57bc
Smooth	43.78bcd	44.97abc	44.12abcd	45.60ab	43.92bcd
Whipped	24.85	23.65	26.74	26.23	25.42
Aroma Attributes					
Overall Intensity	14.22b	15.28ab	16.28ab	16.06ab	16.90a
Artificial Butter	8.45	8.83	9.52	10.01	9.41
Oily	9.07ab	9.93ab	9.93ab	9.84ab	9.92ab
Milk Dairy Sweet	8.93	8.50	8.16	6.99	7.72
Chemical	3.62c	4.08bc	4.65abc	4.63abc	4.94ab
Dairy Sour	5.14	5.36	5.90	5.88	5.65
Fruity (Artificial)	5.99	6.75	6.98	7.03	6.79
Sweet (Artificial)	4.90	5.60	5.47	5.34	5.27

Mouthfeel Attributes					
Melt Rate	41.43	42.00	42.17	42.38	41.74
Inconsistent Texture	2.33	2.94	2.14	2.28	2.27
Greasy/Oily	17.85	17.64	18.22	18.32	18.97
Flavor Attributes					
Fruity (Artificial)	1.42ab	12.53ab	13.31a	10.96cd	10.19d
Salty	25.63	25.59	25.32	24.89	24.77
Oily	14.59c	16.63b	16.89b	17.49ab	18.69a
Artificial Butter	13.47	13.67	13.95	14.09	13.00
Dairy Sour	10.52	10.11	11.29	10.83	10.64
Real Butter	8.19a	6.23b	6.02b	5.01b	4.97b
Milky Dairy Sweet	11.67ab	10.38bcd	10.59bc	8.82de	8.12e
Chemical	5.61d	6.90c	6.77c	7.64abc	8.22ab
Bitter	4.43de	5.27cd	5.07cde	6.37ab	6.67a
Old	2.15	2.81	2.45	2.85	2.98
Off Flavor	7.69d	13.76c	13.27c	17.88a	18.94a
Aftertaste Attributes					
Salty	20.88	21.37	20.59	21.40	21.32
Dairy Sour	9.13	10.30	10.34	10.35	10.68
Milky Dairy Sweet	11.11ab	10.29bc	10.41abc	9.86cd	8.68d

Real Butter	8.07a	6.89bc	6.89bc	5.86cd	5.42d
Oily Aftertaste	11.02c	11.43bc	12.65ab	13.12a	13.57a
Old	6.65	10.23	2.38	2.68	2.71
Off Flavor	6.65f	10.23de	8.91e	12.62bc	15.27a
Bitter	4.00cd	4.88abc	4.51bc	5.34ab	5.89a
Afterfeel Attributes					
Astringent	13.07	12.67	13.60	13.21	12.76
Oily Mouthfeel	12.38	11.64	12.28	12.47	12.20
Spreading onto Bread Attributes					
Spreadable	52.69	52.22	52.91	52.74	53.00
Appearance on Bread Attributes					
Shiny	44.73abc	44.05bc	46.70a	44.44bc	45.98ab
Color on Bread	15.15bcd	16.29a	14.09d	15.32abc	14.60cd

Table 8. Effect of omega-3 enriched oil (spread formulation with omega-3 fatty acids) and conventional soybean oil (control spread formulation) on the sensory attributes of margarine spread formulations at 9 months.

	Spread Formulation				
Attributes	Control Spread Formulation	Spread Formulation with SDA	Spread Formulation with Menhadin Oil	Spread Formulation with Meg-3 Oil	Spread Formulation with Algal Oil
Appearance Attributes					
Color	16.76ab	17.50ab	16.01bc	16.43bc	14.93c
Firm	24.23	24.94	23.17	25.54	25.02
Glossy	19.09bc	16.57bc	18.52bc	20.27ab	23.49a
Smooth	40.06abc	38.64bc	39.89bc	40.39abc	42.71a
Whipped	19.88	19.83	19.39	17.00	19.63
Aroma Attributes					
Overall Intensity	14.37	13.81	15.28	13.77	16.66
Artificial Butter	9.64	8.74	10.66	9.89	11.10
Oily	8.72	8.14	8.84	8.50	9.66
Milk Dairy Sweet	4.48	5.04	4.69	4.53	5.47
Chemical	3.62	4.24	5.42	4.51	5.11
Dairy Sour	4.51	4.72	4.22	4.34	5.00
Fruity (Artificial)	6.13	6.49	6.21	6.60	6.77
Sweet (Artificial)	4.21	4.91	4.57	4.73	4.54

Mouthfeel Attributes					
Melt Rate	36.13	37.72	36.82	36.07	36.16
Inconsistent Texture	2.16	2.73	1.87	2.37	1.56
Greasy/Oily	20.37	19.87	19.86	20.83	22.32
Flavor Attributes					
Fruity (Artificial)	10.94abc	11.54abc	9.90c	10.22bc	10.31bc
Salty	22.48	23.19	22.57	20.46	20.59
Oily	16.33	17.50	16.89	17.89	17.08
Artificial Butter	13.18	12.71	13.81	12.89	13.53
Dairy Sour	7.47ab	6.30ab	6.66ab	5.76b	6.16b
Real Butter	4.49ab	4.08abc	3.41bcde	2.72de	2.37e
Milky Dairy Sweet	5.27ab	4.53abc	4.32abcd	3.30d	3.57cd
Chemical	5.31d	7.76bcd	11.50abc	11.73ab	11.71ab
Bitter	3.87c	4.77bc	6.78ab	7.10a	6.90ab
Old	2.03	2.96	3.66	3.43	2.72
Off Flavor	7.14c	12.23bc	16.21ab	18.26a	19.50a
Aftertaste Attributes					
Salty	20.74	20.94	19.43	19.51	19.41
Dairy Sour	6.36	6.38	6.77	5.59	5.93
Milky Dairy Sweet	5.37a	4.22bc	3.70c	3.56c	3.58c

Real Butter	4.23ab	3.21bc	3.06c	2.39c	2.40c
Oily Aftertaste	13.21	13.19	13.13	12.99	14.10
Old	1.81	2.00	3.10	2.99	3.08
Off Flavor	5.34e	10.10cd	12.97bc	15.88ab	17.66a
Bitter	3.58e	4.94abcd	6.01a	6.06a	5.68ab
Afterfeel Attributes					
Astringent	12.61	12.43	12.08	12.03	11.73
Oily Mouthfeel	13.92	13.10	13.40	13.04	13.51
Spreading onto Bread Attributes					
Spreadable	50.33	50.10	50.88	50.62	51.32
Appearance on Bread Attributes					
Shiny	36.87ab	33.37bcd	35.74abc	35.17abc	35.82abc
Color on Bread	18371bcd	19.80abc	19.49abcd	20.22ab	18.08cd

^{a,b} Within a row, values without a common superscript differ significantly ($P < 0.05$)

[091] Conclusion: Throughout the shelf life, the flavor attributes of the spread formulation with SDA closely resembled the control spread formulation. In comparison to spread formulations made with a competitive set of omega-3 oils including two sources of fish oil and algal oil, off flavor after nine months of shelf life of the spread formulation with SDA was not significantly different from the control spread formulation, wherein the alternate forms of omega-3 oils were all significantly different from the control. Similar results were obtained for off flavor aftertaste where the spread formulation with SDA was less different from the control spread formulation than the alternate sources of omega-3.

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The references cited in this application, both above and below, are specifically incorporated herein by reference.

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WHAT IS CLAIMED IS:

1. A spread formulation comprising an oil phase and an aqueous phase, wherein the oil phase comprises stearidonic acid (SDA)-enriched oil.
2. The spread formulation of claim 1, wherein the SDA-enriched oil is SDA-enriched soybean oil.
3. The spread formulation of claim 1, wherein the oil phase further comprises an oil selected from the group consisting of hydrogenated oils, partially hydrogenated oils, interesterified oils, and combinations thereof.
4. The spread formulation of claim 3, wherein the oil phase further comprises a liquid oil selected from the group consisting of soybean oil, canola oil, rapeseed oil, palm oil, and combinations thereof.
5. The spread formulation of claim 4, wherein the oil phase further comprises at least one of monoglycerides, diglycerides, and lecithin.
6. The spread formulation of claim 1, wherein the oil phase further comprises at least one high stability oil selected from the group consisting of low linolenic soybean oil, high oleic soybean oil, high oleic/low saturate soybean oil, high oleic canola oil, sunflower oil, and combinations thereof.
7. The spread formulation of claim 1, wherein the aqueous phase further comprises at least one of a salt, a dairy protein, ethylenediaminetetraacetic acid (EDTA), and a preservative.
8. A margarine spread formulation comprising an oil phase and an aqueous phase, the oil phase comprising stearidonic acid (SDA)-enriched soybean oil.
9. The margarine spread formulation of claim 8, wherein the oil phase further comprises an oil selected from the group consisting of hydrogenated oils, partially hydrogenated oils, interesterified oils, and combinations thereof.

10. The margarine spread formulation of claim 9, wherein the oil phase further comprises at least one of a liquid oil selected from the group consisting of soybean oil, canola oil, rapeseed oil, palm oil, and combinations thereof; monoglycerides; diglycerides; lecithin; and a high stability oil selected from the group
5 consisting of low linolenic soybean oil, high oleic soybean oil, high oleic/low saturate soybean oil, high oleic canola oil, sunflower oil, and combinations thereof.

11. The margarine spread formulation of claim 8, wherein the aqueous phase further comprises at least one of a salt, a dairy protein, ethylenediaminetetraacetic acid (EDTA), and a preservative.

12. A method of producing a spread formulation for human consumption comprising:

- a. providing an oil phase comprising a stearidonic acid (SDA)-enriched oil;
- 5 b. providing an aqueous phase; and
- c. contacting the oil phase and the aqueous phase to make an oil-in-water emulsion spread formulation.

13. The method of claim 12, wherein the oil phase is provided by blending the SDA-enriched oil with at least one of a liquid oil, a hydrogenated oil, a partially hydrogenated oil, and an interesterified oil.

14. The method of claim 13, further comprising blending at least one of monoglycerides, diglycerides, and lecithin into the oil phase.

15. The method of claim 14, further comprising blending at least one high stability oil selected from the group consisting of low linolenic soybean oil, high oleic soybean oil, high oleic/low saturate soybean oil, high oleic canola oil, and sunflower oil into the oil phase.

16. The method of claim 14, wherein the blending is conducted at a temperature of from about 105°F (41°C) to about 110°F (43°C).

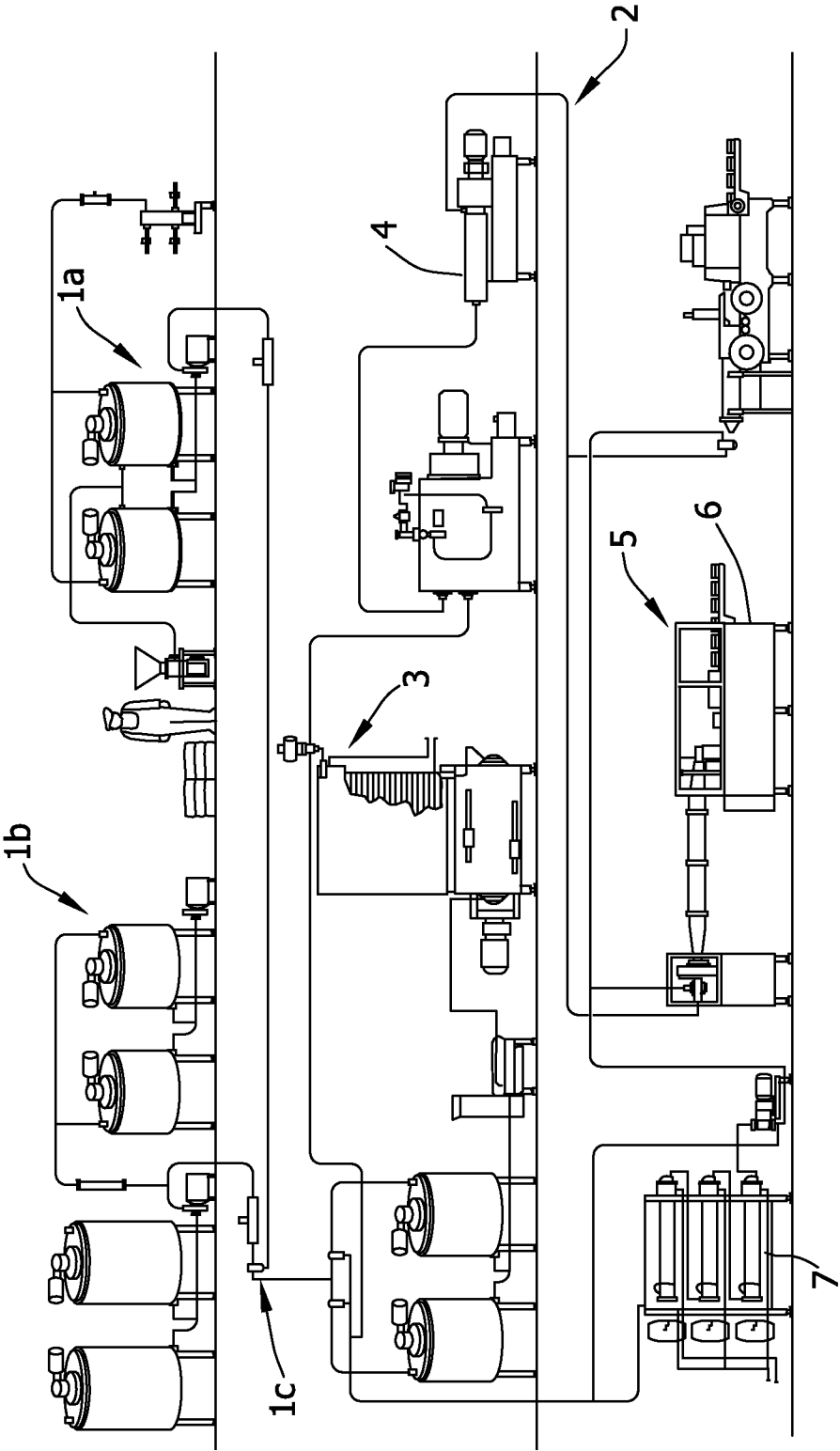
17. The method of claim 12, wherein the aqueous phase is provided by blending water with at least one of a salt, a dairy protein, and ethylenediaminetetraacetic acid (EDTA).

18. The method of claim 12 further comprising pasteurizing the aqueous phase at a temperature of greater than about 150°F (66°C) for a time period of about 30 minutes.

19. The method of claim 12, wherein the oil phase and aqueous phase are blended together to form the oil-in-water emulsion spread formulation, and wherein the spread formulation is held at a temperature of from about 105°F (41°C) to about 110°F (43°C) for a time period of from about 1 hour to about 2 hours.

20. The method of claim 19 further comprising pumping the spread formulation into a scraped surface heat exchanger to cool the spread formulation to a temperature of from about 34°F (1°C) to about 41°F (5°C) and then pumping the cooled spread formulation into a pin mixer.

FIG. 1



INTERNATIONAL SEARCH REPORT

International application No

PCT/US2010/031336

A. CLASSIFICATION OF SUBJECT MATTER

INV. A23D7/00

ADD.

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

A23D

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, BIOSIS, COMPENDEX, FSTA, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

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X	WO 2008/085840 A2 (MONSANTO TECHNOLOGY LLC [US]; WILKES RICHARD S [US]) 17 July 2008 (2008-07-17) paragraph [0011] - paragraph [0016]; claims 1,11,23,31,56,62,71; examples 3,6 paragraph [0032] ----- -/--	1-20



Further documents are listed in the continuation of Box C.



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Date of the actual completion of the international search

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Date of mailing of the international search report

30/08/2010

Name and mailing address of the ISA/

European Patent Office, P.B. 5818 Patentlaan 2
NL - 2280 HV Rijswijk
Tel. (+31-70) 340-2040,
Fax: (+31-70) 340-3016

Authorized officer

Saettel, Damien

INTERNATIONAL SEARCH REPORT

International application No
PCT/US2010/031336

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WHELAN J ET AL: "INNOVATIVE DIETARY SOURCES OF N-3 FATTY ACIDS" ANNUAL REVIEW OF NUTRITION, ANNUAL REVIEWS INC., PALO ALTO, CA, US LNKD-DOI:10.1146/ANNUREV.NUTR.25.050304.092605, vol. 26, 1 January 2006 (2006-01-01), pages 75-103, XP002485599 ISSN: 0199-9885 [retrieved on 2006-05-09] page 79 - page 80 page 95, paragraph 4 - page 96, paragraph 1 -----	1-20
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Information on patent family members

International application No

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