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(54) Title: SPRAY FORMULATION COMPRISING CHOLECALCIFEROL (VITAMIN D3) WITH IMPROVED STABILITY

(57) Abstract: This invention relates to a spray compositions comprising vitamin D in natural fatty oil, preferably olive oil, sunflower oil and coconut oil having no alcohol, sugar, artificial sweeteners, artificial fragrance, preservatives and antioxidants with improved stability properties. This product is used thereof for human beings, particularly, breast-feeding infants. This invention also provides an ease option of use by applying one puff into the mouth and the one puff comprises specific amount of vitamin D3.

SPRAY FORMULATION COMPRISING CHOLECALCIFEROL (VITAMIN D3) WITH IMPROVED STABILITY

DESCRIPTION

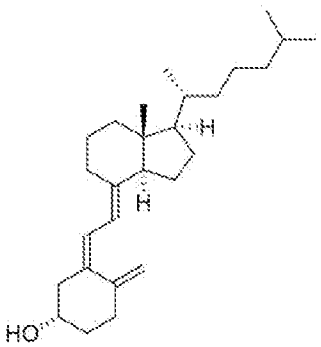
Technical field:

This invention relates to a stable and ease of application spray formulation comprising cholecalsiferol (vitamin D3) in natural fatty oil, preferably olive oil, sunflower oil and coconut oil; having no alcohol, sugar, artificial sweeteners, artificial fragrance and preservatives. It is used thereof for human beings, particularly, breast-feeding infants.

Prior Art:

Vitamins are complex organic substances found variously in most foods and are essential in small amounts, for the normal functioning of most living organisms. For example, vitamins are a critically necessary component of the diet of higher animals, including humans. When the normal diet fails to furnish minimum quantities of essential vitamins, it is a common practice to supplement the diet by artificially inducing the required amounts into the body. Common methods of introduction include oral ingestion of tablets or liquid solutions containing the vitamins (which may require concurrent ingestion of a suitable carrier liquid such as water or other drinkable liquid to aid in swallowing the vitamin composition and washing it into the gastro-intestinal system of the human). Similarly, but less frequently, vitamins are introduced by parenteral injection.

The invention relates to specifically Vitamin D. It exists in two forms as vitamin D3 (cholecalciferol) and vitamin D2 (ergocalciferol). In the present invention, "vitamin D" means vitamin D3 (cholecalciferol). Its chemical name is **(5Z-7E)-(3S)-9,10-seco-5,7,10(19)-**cholestatrien-3-ol. The structural formula Cholecalciferol (vitamin D3) is represented below (Formula I):



Formula (I)

Vitamin D2 is produced by plants whereas vitamin D3 is formed in human as the result of exposure to the ultraviolet radiation of sunlight in skin. Both vitamin D2 and vitamin D3 are effective in the body as transformed to 25-hydroxyvitamin D. This form goes to the kidneys through the circulation and there turns into calcitriol, the active form of vitamin D. Vitamin D level is low in nutrients except nutrients enriched with Vitamin D and nutrients cannot meet the daily needs. When daily requirement of vitamin D for bone and muscle health cannot meet by food and sun, vitamin D supplementation is necessarily required. When vitamin D is taken orally, rapidly absorbed from the intestines and converted into active Vitamin D by the body and vitamin D needs are resolved. According to the data obtained from the scientific study, vitamin D3 is absorbed faster and more from the digestive system and exerts more powerful than vitamin D2.

For biological activity, vitamin D must go through two metabolic steps. Vitamin D is metabolized by the liver to 25-hydroxyvitamin D [25(OH)D], which is measured in serum to reflect vitamin D nutritional status. 25-hydroxyvitamin D intrinsically has little biological activity. The kidney metabolizes 25-hydroxyvitamin D into the active hormone, 1,25-dihydroxyvitamin D, which affects calcium transport across cell membranes. The body, according to its mineral requirements, carefully regulates production and breakdown of 1,25-dihydroxyvitamin D to regulate plasma calcium concentrations. Very few foods naturally contain vitamin D. Mellanby, J. Physiol (London) volume 52 (1919), instituted the idea that an artificial supplement, cod-liver oil, contained an agent that prevented rickets; the agent became known as vitamin D. Most of the vitamin D in our food is supplementary, synthetic material, which is either in the form of vitamin D3, the form naturally produced in animals, or it can be vitamin D2, which is derived from a plant steroid.

Vitamin D has been recognized as essential to health. It appears to increase the efficiency of the intestines to absorb calcium and also mobilizes calcium from bone tissue when required. Both deficiency and sufficiency of vitamin D have negative results, such as excessive vitamin D intake has been known as toxic. On the other hand, vitamin D deficiency results in impaired bone mineralization and leads to bone softening diseases such as rickets and osteoporosis, and may also be a contributing factor to high blood pressure, tuberculosis, cancer, heart disease, stroke, periodontal disease, MS, seasonal affective disorder and memory loss. Vitamin D deficiency is commonly observed in children, the aged and those of lower socio-economic status. Protection of vitamin D levels is important not only for calcium and phosphorus metabolism of bone, but also for general health and well-being. It is also a vitamin which is absolutely necessary in children for normal bone growth and development. Vitamin D deficiency is now recognized as a global epidemic.

The skin is the major site of cholesterol production and humans acquire vitamin D through the natural action of ultraviolet light on the skin. 7-Dehydrocholesterol, which is unstable to ultraviolet light, is normally a precursor to cholesterol. However, ultraviolet light breaks open the B-ring of the 7-dehydrocholesterol molecule to generate previtamin D3, which spontaneously isomerizes over hours and days into vitamin D3, which is also known as cholecalciferol. An unknown proportion of the vitamin D from the skin is absorbed into the circulation. Vitamin D3 is not soluble in water, and in the circulation, there is a protein that specifically binds to and carries vitamin D and its metabolites. The advantage of ultraviolet exposure is that it is natural, and has no vitamin D toxicity associated with it. The disadvantage is that the availability of ultraviolet light is unreliable, and too much of it causes sunburn or skin cancer. At northern latitudes there is often not enough ultraviolet light intensity outdoors to generate previtamin D.

In pharmacology, the international unit (IU) is a unit of measurement for the amount of a substance; the mass or volume that constitutes one international unit varies based on which substance is being measured, and the variance is based on the biological activity or effect, for the purpose of easier comparison across substances. International units are used to quantify vitamins, hormones, some medications, vaccines, blood products, and similar biologically active substances. In brief, IU is defined as a quantity of a biologic (such as a vitamin) that produces a particular biological effect agreed upon as an international standard.

Pediatric associations in Canada and the United States now strongly encourage starting vitamin D supplementation from birth onwards (Health Canada, Vitamin D supplementation for breastfed infants). Breast-feeding results in fewer infections and allergies during the first year of life compared to babies fed formula. Breast milk provides nearly optimal nutrition for newborns; however, it provides little vitamin D. Vitamin D has well-recognized effects on bone, but beyond that, better vitamin D nutrition during infancy is associated with less risk of other diseases that develop later in life. These disease reductions include less risk of juvenile diabetes, and schizophrenia (Hypponen, E., Laara, E., Reunanen, A., Jarvelin, M. R., and Virtanen, S. M. (2001) Lancet 358, 1500-1503) (McGrath, J., Saari, K., Hakko, H., Jokelainen, J., Jones, P., Jarvelin, M. R., Chant, D., and Isohanni, M. (2004) Schizophr. Res. 67, 237-245). However, because it is normally obtained through sun exposure instead of orally, vitamin D is the one nutrient that is most often deficient in breast milk (Centers for Disease Control, Vitamin D Expert Panel Meeting, October 11 - 12, 2001 Atlanta, Georgia Final Report).

In 2005, the American Academy of Pediatrics issued a new policy statement concerning breast-feeding (American Academy of Pediatrics Policy Statement 2005; Pediatrics 115; 496-506). One major change was that all breast-fed infants should receive 200 IU (5 meg) of oral vitamin D drops daily, beginning during the first 2 months of life. The older recommendation had been that vitamin

D supplementation should start at around 2 months of life. The latest American Academy of Pediatrics recommendation follows what Health Canada has been advising, that vitamin D supplementation should be from birth, with 400 IU (10 meg) of vitamin D per day (Pediatrics 2005; 115; 496-506). Moreover, Health Canada recommends that infants in the north be given 800 IU (20 meg) of vitamin D per day.

The need to provide vitamin D at an earlier age makes the problem of providing vitamin D nutrition more complicated. Smaller infants are more difficult to handle. What makes the problem worse is that the recommendations from pediatric societies and government bodies provide no detail or any method for exactly how to give vitamin D to the breast-feeding infant. They simply advise that parents should be giving vitamin D.

Parents of infants express great frustration about existing products and methods for providing infants with vitamin D, especially about the taste and the fact that infants often spit out at least some of the liquid. Direct administration of liquid vitamin D preparations is commonly done with an eyedropper, and with the infant lying on its back. One risk associated with direct administration of vitamin D solutions into the mouth is that infants can gag on, or inhale some of the liquid.

There are vitamin D drops available in oil, at 400 IU per drop in the United States (naturalhealthsupply.com). The vitamin D for this is dissolved in olive oil and/or corn oil and/or coconut or flax-seed oil. The problem with these is that the oils are comprised of long-chain unsaturated fatty-acid triglycerides that will become rancid with repeated opening of the container, they carry a flavor, and they have a greasy feel on pacifier.

EP2068885 relates to compositions comprising vitamin D in a medium-chain triglyceride medium and use thereof for human beings, particularly, breast-feeding infants, wherein said composition is applied as a single drop to a breast feeding mother's nipple, to a pacifier or a nipple of a baby bottle, and having human being suck the composition from said object surface.

US4525341 discloses a method of administering a sprayable vitamin composition which may include other ingredients such as breath fresheners, flavoring agents and the like. In the development of vitamin D analogs in spray areas, spray containing vitamins are formulated with ethanol, ethanol - water mixture or water as a solvent carrier, especially many products on the market mainly with ethanol as a solvent carrier.

The low viscosity of ethanol in the formulation is considered to be an advantage for the dissolution of the drug; however, since ethanol is volatile, the liquid may be concentrated in the spray pump causing variance in dosage form and also blockage and not spout spray smoothly.

This is the most important disadvantage of using ethanol in vitamin D spray formulation especially for breast-feeding infants.

Apart from the formulation, another important point is stability of vitamins in dietary supplements. A number of common physical and chemical factors affect the stability of vitamins in products (Gadient, 1986; Frye, 1994; Reddy and Love, 1999). Exposure to multiple stresses generally multiplies the effect on vitamin stability. For example, exposure to moisture through high relative humidity during storage significantly increases the rate at which vitamins are degraded by chemical reactions, such as oxidation. A similar statement could be made about exposure of vitamins to elevated temperature or strong light during storage. According to Shurson et al. (1996) vitamin D3 is sensitive to temperature, humidity, light and acidic pH; also it is highly sensitive to oxygen. Therefore there are several studies on stability of vitamin D product and it is still a necessity to develop a stable vitamin D product especially for breast-feeding infants.

Notwithstanding the above teachings of the prior art, there remains a need for a safe, convenient and efficacious method of administering nutritional or therapeutic amounts of vitamin D to a human being, particularly a breast-feeding infants. There is also a utility for adults to be able to deliver a dose of vitamin D efficiently placing the supplement directly into the mouth with a convenient way.

Accordingly, it would be highly advantageous to provide more convenient compositions and methods for introducing supplemental vitamins into the human body, particularly breast-feeding infants. Because applying vitamin D drop into the mouth or any object surface concludes unreliable dose delivering to the infant. Difficulty with administration result in lower compliance rates, consequently poor compliance results in under-dosing and ineffective treatment. These factors also diminish nutrient supplementation with vitamin D. Consequently, an object of the invention is to provide a spray containing a vitamin D analog with natural oil having no alcohol, sugar, artificial sweeteners, artificial fragrance and preservatives with improved stability properties. This product also has dose uniformity with spraying the design dose even in the first dose and after the storage time.

Description of the Invention:

The present invention relates to liquid compositions comprising vitamin D, methods for the preparation thereof, and also a spray containing a vitamin D analogue not having any conservative and providing simple application procedure with high stability.

In this invention, the compound "vitamin D" means (5Z-7E)-(3S)-9,10-seco-5,7, 10(19)-cholestatrien-3-ol also having the trivial names cholecalciferol or calciol (D3).

The invention relates to a method of administering a sprayable vitamin to the human being; the vitamin D compositions can be simply sprayed into the mouth, it finds the way into the proper body processes by absorption through the mucous membranes and/or simple swallowing according to normal salivary mechanisms.

The human being may be an adult or an infant.

Spray drug products contain therapeutically active ingredients (drug substances) dissolved or suspended in solutions or mixtures of excipients (e.g., preservatives, viscosity modifiers, emulsifiers, buffering agents) in non-pressurized dispensers that deliver a spray containing a metered dose of the active ingredient. The dose can be metered by the spray pump or could have been premetered during manufacture.

In this invention, the liquid carrier is pharmaceutically acceptable natural fatty oil, like olive oil, sunflower oil and coconut oil. Olive oil is a fat obtained from the olive. The composition of olive oil varies with the cultivar, altitude, time of harvest and extraction process. It consists mainly of oleic acid (up to 83%), with smaller amounts of other fatty acids including linoleic acid (up to 21%) and palmitic acid (up to 20%). Sunflower oil is the non-volatile oil compressed from the seeds of sunflower. It is a monounsaturated (MUFA)/polyunsaturated (PUFA) mixture of mostly oleic acid (omega-9)-linoleic acid (omega-6) group of oils. The oil content of the seed ranges from 22% to 36% (average, 28%): the kernel contains 45-55% oil. Coconut oil is an edible oil extracted from the kernel or meat of mature coconuts harvested from the coconut palm. It has a distinctive nutty aroma and taste. It is composed of the following fatty acids: lauric acid (48% of total), myristic acid (16%), palmitic acid (9.5%), decanoic acid (8%) and others in small amounts.

Vitamin D3 contains doses of vitamin D3 recommended by scientific medical guidelines for children and adults. When used as a food supplement with recommended doses, it does not accumulate in the body and does not cause toxic effects. In this invention, it does not lead to over-consumption because of spray form. Each puff applied to mouth provides a safe amount of vitamin D support.

The use of preservatives, like alcohols, benzoates, sorbates, and parabens is common in liquid formulations. Preservatives are effective in controlling mold, inhibiting yeast growth and protecting against bacterial proliferation, thus, finally, to allow compliance with the European Pharmacopoeia microbiological specifications (Ph. Eur. 6.7, S5.1.4) for "aqueous preparations for oral use" or "aqueous preparations for oromucosal use". Consequently most of the vitamin products contain preservatives; especially butylated hydroxyanisole (BHA) is used in vitamin products containing oil.

Butylated hydroxyanisole (BHA) is an antioxidant consisting of a mixture of two isomeric organic compounds, 2-teri-butyl-4-hydroxyanisole and 3-teri-butyl-4-hydroxyanisole. It is prepared from 4-methoxyphenol and isobutylene. It is a waxy solid used as a food additive with the E number of E320. The primary use for BHA is as an antioxidant and preservative in food, food packaging, animal feed, cosmetics, rubber, and petroleum products. BHA is commonly used in medicines. Since 1947, BHA has been added to edible fats and fat-containing foods and medicines for its antioxidant properties as it prevents rancidification of food which creates objectionable odors. Like butylated hydroxytoluene (BHT), the conjugated aromatic ring of BHA is able to stabilize free radicals, sequestering them. By acting as free radical scavengers, further free radical reactions are prevented. However, The U.S. National Institutes of Health report that BHA is reasonably anticipated to be a human carcinogen based on evidence of carcinogenicity in experimental animals. In particular, when administered in high doses as part of their diet, BHA causes papillomas and squamous cell carcinomas of the forestomach in rats and Syrian golden hamsters. The State of California, has, however, listed it as a carcinogen.

Since babies and kids have much smaller bodies, any harmful foods or additives can do much more proportionate damage to them. Furthermore, breast-feeding mothers may not want to give their infants foreign liquids or compounds that are not natural for them to be taking.

In this invention, the product contains no alcohol, sugar, artificial sweeteners, artificial fragrance and preservatives; especially the product is BHA free.

The spray apparatus of the present invention (including spray bottle and spray pump) material may be selected from high density polyethylene, polypropylene, metal (e.g., aluminum), glass, etc. in one or more thereof, so that the drug can get better stability. Also, the amount of vitamin D3 per puff is fixed as 400 IU, 600 IU and 1000 IU. Especially, 400 IU/puff is used for breast-feed infants (0-1 age); more than 600 IU/puff and 1000 IU/puff doses are used for adults.

In the present invention, vitamin D3 is available in spray cans. According to 400 IU/puff dose recommended for breast-feed infants, it provides an ease option of use by applying one puff into the mouth and the one puff has exactly 400 IU of Vitamin D3. In spray form, transportation and storage of the product is very practical. According to Republic of Turkey Ministry of Food, Agriculture and Livestock: Regulations on Turkish Food Codex Food Supplement, maximum of 500 IU vitamin D is recommended for ages 4-10; and also maximum of 1000 IU vitamin D is recommended for ages higher than 11.

Compositions of this invention are detailed below showing examples. However, pharmaceutical compounds of this invention are not restricted to the following examples.

Example 1.

Vitamin D Oral Spray (400 IU/puff)	**% (a/a)**
Olive oil	99.98553
Vitamin D3 (Cholecalciferol)	0.01447
Total	**100**

Crystalline vitamin D3 (cholecalciferol, United States Pharmacopea grade) was dissolved into required amount of olive oil (United States Pharmacopea grade), to make a solution containing 125 meg vitamin D per one mL of final product. It was dispensed into a spray bottle and spray pumps tightened.

Example 2.

Vitamin D Oral Spray (600 IU/puff)	**% (a/a)**
Olive oil	99.9783
Vitamin D3 (Cholecalciferol)	0.02170
Total	**100**

Crystalline vitamin D3 (cholecalciferol, United States Pharmacopea grade) was dissolved into required amount of olive oil (United States Pharmacopea grade), to make a solution containing 187.5 meg vitamin D per one mL of final product. It was dispensed into a spray bottle and spray pumps tightened.

Example 3.

Vitamin D Oral Spray (1000 IU/puff)	**% (a/a)**
Olive oil	99.96383
Vitamin D3 (Cholecalciferol)	0.03617
Total	**100**

Crystalline vitamin D3 (cholecalciferol, United States Pharmacopea grade) was dissolved into required amount of olive oil (United States Pharmacopea grade), to make a solution containing 312.5 meg vitamin D per one mL of final product. It was dispensed into a spray bottle and spray pumps tightened.

Example 4.

Vitamin D Oral Spray (600 IU/puff)	**% (a/a)**
Sunflower oil	99.9783
Vitamin D3 (Cholecalciferol)	0.02170
Total	**100**

Crystalline vitamin D3 (cholecalciferol, United States Pharmacopea grade) was dissolved into required amount of sunflower oil (United States Pharmacopea grade), to make a solution containing 187.5 meg vitamin D per one mL of final product. It was dispensed into a spray bottle and spray pumps tightened.

Example 5.

Vitamin D Oral Spray (600 IU/puff)	**% (a/a)**
Coconut oil	99.9783
Vitamin D3 (Cholecalciferol)	0.02170
Total	**100**

Crystalline vitamin D3 (cholecalciferol, United States Pharmacopea grade) was dissolved into required amount of coconut oil (United States Pharmacopea grade), to make a solution containing 187.5 meg vitamin D per one mL of final product. It was dispensed into a spray bottle and spray pumps tightened.

Stability Studies

The stability of a drug substance is an important factor in the manufacture of safe and effective pharmaceutical products. Stability studies are required to be submitted by any applicant seeking approval for a new pharmaceutical product. Stability study requirements are covered, for example in the United States Pharmacopeia, in the Good Manufacturing Practices (GMP) as well as in FDA and ICH Guidelines. It is known that many drugs exhibit poor or modest shelf stability. The diminution of the concentration of a drag as a result of its degradation is inherently undesirable, as it makes therapy with the drug less certain. Stability issues can be caused by environmental factors such as humidity, temperature and the like.

Furthermore, it is hypothesized that the stability of vitamin D3 decreases rapidly after the preparation has been opened because of the influence of oxygen, temperature, humidity and light exposure, leading to a significant decrease in vitamin D content.

Vitamin D3 content should be regulated due to the risk of toxicity from excessive intake. Hypervitaminosis D can occur in patients who take vitamin D supplements and are associated with hypercalcemia and other effects such as hypercalciuria, anorexia, weight loss, weakness, fatigue, disorientation, vomiting, constipation and even irreversible renal and cardiovascular damage. Due to this reason, no excess dose is used in this invention.

Most of the liquid dosage forms of vitamin D3 on market are packaged in dark glass containers that protected the product from light and oxygen. However, once the preparations are opened, they are exposed to both of these factors, making them further susceptible to oxidation and degradation of the vitamin. It has been discovered in this invention; using spray bottles having little contact with air highly prevent the oxidation and degradation of vitamin D3.

A literature search on Medline and Embase indicated that there was limited published data on the stability of oral vitamin D preparations. In one study, Huyghebaert et al. reported that cholecalciferol prepared as a liquid dose form with 0.15% Tween 80 and various stabilizers, the product was stable just for 6 months at 17 °C and for 2 months at room temperature. EP2068885 B1 discloses experimental observation of potential liquids for suitability for vitamin D as stability of vitamin D is poor in water; evaporation of solvent changes concentration of vitamin D in ethanol; oxidation and rancidization occur in formulation of vitamin D with canola oil, olive oil and coconut oil.

In the development of vitamin D spray dosage form, stability was assessed under three different isothermal conditions (25°C ± 2°C, 30°C ± 2°C and 40°C ± 2°C) in temperature-programmable control cabinets. A temperature of 25°C ± 2°C represents ambient temperature, 30°C ± 2°C represents intermediate temperature and 40°C ± 2°C is a temperature that can be reached under extreme conditions in homes without air conditioning in the summer. The extreme condition, temperature of 40°C ± 2°C and 75 % ± 5 % RH (Relative Humidity), accelerated stability studies is carried out on vitamin D spray dosage form up to 6 months. Stability data for all examples given over 40°C ± 2°C and 75% ± 5 % relative humidity are shown in Table 1.

Even if no excess dose is used in the products explained in example 1 to 5, the results of assay determination for cholecalciferol (vitamin D3) were within the limit for 6 months period and 40°C ± 2°C, 75% ± 5 % RH humidity extreme conditions. Also, the cholecalciferol spray form products, not containing any preservatives such as BHA, were found in acceptable limits and no degradation products were observed or levels of degradation product were below the limits of quantification according to the analytical method adopted. In addition to that, all formulations were tested for microbial contamination of total viable aerobic count, total combined yeast and mould count and Escherichia coli at each month. Example 1-5 passed microbial testing for 6 months period and 40°C ± 2°C, 75% ± 5 % RH humidity extreme conditions.

Table 1. Stability data of examples in olive oil formulations over 6 months' period and 40°C ± 2°C, 75% ± 5 % RH humidity

	Mean assay determination of cholecalciferol (IU/puff)				
Time (months)	**Example 1 (400 IU/puff in olive oil)**	**Example 2 (600 IU/puff in olive oil)**	**Example 3 (1000 IU/puff in olive oil)**	**Example 4 (600 IU/puff in sun-flower oil)**	**Example 5 (600 IU/puff in coconut oil)**
Initial	401,3	613,5	1034,2	620,4	600,5
1	410,8	621,6	1020,3	612,6	615,3
2	402,8	625,5	1036,3	603,4	594,4
3	398,1	618,6	1016,7	615,7	605,4
4	385,6	620,4	995,7	598,5	610,6
5	391,4	627,6	1015,1	605,3	610,5
6	384,2	615,8	991,5	611,4	602,4

It is concluded that the product developed in the present invention is biologically natural in the content and it is almost safe for infant consumption. Also, the product intrinsically is not subject to rancidity, oxidation or degradation over the shelf life of the composition according to results

obtained 6 months' period of 40°C ± 2°C, 75% ± 5 % **RH** humidity. There is little change in assay of cholecalciferol (IU/puff) within 6 months with different oil content and dosage forms. Despite the product contains no preservative, the stability of product is provided with spray form having little contact with air. Therefore this formulation achieves the improvement of stability for the vitamin D3 spray form.

CLAIMS

1. A stable liquid oral pharmaceutical composition in a form of a spray comprising vitamin D, or a salt thereof, in natural fatty oil having no alcohol, sugar, artificial sweeteners, artificial fragrance, preservatives and antioxidants, wherein the vitamin D is cholecalciferol (vitamin D3).
2. The composition according to claim 1, wherein used natural fatty oil is olive oil.
3. The composition according to claim 1, wherein used natural fatty oil is sunflower oil.
4. The composition according to claim 1, wherein used natural fatty oil is coconut oil.
5. The composition according to any of the claims 1 to 4, wherein the product is BHA free.
6. The composition according to claim 1, wherein amount of vitamin D per puff is fixed in order to administer nutritional or therapeutic amounts of vitamin D.
7. The composition according to claim 6, wherein the amount of vitamin D is 400 IU, 600 IU or 1000 IU per puff.
8. The composition according to claim 7, wherein the amount of vitamin D is 400 IU/puff for breast-feeding infants.
9. The composition according to claim 7, wherein the amount of vitamin D is 600 IU and/or 1000 IU per puff for adults.

INTERNATIONAL SEARCH REPORT

International application No
PCT/TR2017/05Q101

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61K31/593 A23L33/155 A61K9/00 A61K9/08
ADD. A61P19/08

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)
A23L A61K

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 practicable, search terms used)
EPO-Internal, BIOSIS, FSTA, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	AU 2014 201 329 Al (BIOMEDICA NEUTRACEUTICALS PTY LTD) 25 September 2014 (2014-09-25)	1,5-9
Y	abstract paragraphs [0001], [0011], [0013], [0015] - [0018], [0023] - [0024], [0027] - [0029], [0032] examples 1,2	1-9
X	EP 2 942 050 Al (S I I T S R L UNIPERS0NALE [IT]) 11 November 2015 (2015-11-11)	1,4-9
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[X] Further documents are listed in the continuation of Box C.

[X] See patent family annex.

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5 May 2017	15/05/2017

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Couzy, Frangois

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International application No
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