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INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

| (51) International Patent Classification 5 : | | (11) International Publication Number: |
|---|---|---|
| A61K 9/14, 9/107 | A1 | WO 92/17162 |
| | | (43) International Publication Date: 15 October 1992 (15.10.92) |

(21) International Application Number: PCT/US91/02353

(22) International Filing Date: 5 April 1991 (05.04.91)

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(81) Designated States: AT (European patent), AU, BE (European patent), CA, CH (European patent), DE (European patent), DK (European patent), ES (European patent), FR (European patent), GB (European patent), GR (European patent), IT (European patent), JP, LU (European patent), NL (European patent), SE (European patent).

**Published**
*With international search report.*

(54) Title: LYOPHILIZED EMULSION

(57) Abstract

The present invention provides a novel method of lyophilizing an oil-in-water emulsion.

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# LYOPHILIZED EMULSION

## FIELD OF INVENTION

The present invention relates to a process for lyophilizing an oil-in-water emulsion and the emulsion resulting from said process.

## BACKGROUND OF INVENTION AND PRIOR ART STATEMENT

European Application 86109369.8 (Publication Number 211,257) describes a lyophilized dry composition which can be reconstituted with water to form an oil-in-water emulsion suitable for parenteral administration. The freeze-dried composition contains 5% to 60% of a pharmaceutically acceptable lipid, 0.1% to 10% of an emulsifier, and 40% to 90% of a solid carbohydrate. The freeze-drying step is accomplished by spraying the emulsion as fine droplets into a bath of boiling fluid having a boiling point below -20°C, e.g., a fluorocarbon, collecting the dried particles, then sterilizing and packaging them. Although the freeze-dried composition is similar to the composition of the present invention, the lyophilization process is markedly different. The composition also differs from the present invention, e.g., in the quantity of carbohydrate employed. Also, the description of the "manufacture" of the composition indicates that the carbohydrate is added to the emulsion just prior to lyophilization.

U.S. Patent 4,616,047 describes a lyophilized oil-in-water emulsion suitable for oral administration. Although many of the components of

this composition are the same as or similar to those employed in the present invention, other of the components are not suitable for parenteral administration. For example, the polysorbates and sorbitan esters useful as nonionic surfactants are not suitable for parenteral use. The emulsion is placed in alveolar packets and freeze-dried. The amount of oil or lipid used in this formulation (60%-100%) is much higher than the amount employed in the present invention (5%-30%).

Japan Application Number 50-96910 (Disclosure Number 60-239417) describes a freeze-dried oil-in-water emulsion which can be reconstituted and used for parenteral administration. No details of the freeze-drying step are described. The disclosure states that the emulsion is portioned into containers and freeze-dried by an ordinary freeze-drying program. The composition differs from the composition of the present invention in that the prior art composition contains a water soluble polymer in the aqueous phase.

European Application 87111680.2 (Publication Number 257,454) describes an oil-in-water emulsion containing 1-[2,4-dichlorophenyl)-3-methyl-1-pentenyl]-1H-imidazole as the active ingredient. The emulsion can be freeze-dried but no details of the method are given. The disclosure specifically excludes the use of egg phosphatides as emulsifiers, stating that it has a low phosphatidylcholine content and does not show sufficient emulsifying effect.

F. Geyl-Hansen et al., J. Food Proc. Preserv. 2, 205-228 (1978) describes freeze-dried emulsions. The article indicates that Span 80 and Tween 80 were used as emulsifiers, neither of which is suitable for parenteral administration.

## SUMMARY OF INVENTION

The present invention provides a novel method for preparing lyophilized oil-in-water emulsions which comprises freezing and lyophilizing the emulsion in a single cycle. In the process of the present invention the emulsion is maintained at the following temperatures for the times indicated:

(a) 0-24 hours: -40°C;

(b) 24-36 hours: slowly raise temperature to +15°C;

(c) 36-48 hours: +15°C;

(d) 48-72 hours: +30°C;

(e) 72 hours: remove lyophilized product from lyophilizer.

Once the emulsion is placed in the freeze-drying apparatus the condenser is cooled to approximately -60°C. The goal is to begin with a temperature differential of about 20°C between the shelf temperature and the condenser temperature. After the initial 0 to 24 hours at -40°C, a vacuum is applied to obtain a pressure of less than about 60 millitorr. Also, following step (d) the chamber is pressurized with nitrogen.

The present invention also provides an oil-in-water emulsion composition which comprises a lipid or an oil suitable for injection; a surfactant; an agent to improve the isotonicity; a carbohydrate; and water. Preferably, the oil-in-water emulsion contains from 5% to 30% of a lipid or an oil suitable for injection; from 0.5% to 5.5% of a surfactant; from 5% to 15% of a carbohydrate; and from 2% to 4% of an agent which provides isotonicity to the emulsion; the active ingredient; and water for the balance of the

composition. The composition is lyophilized and suitable for injection upon reconstituting the lyophilized compositions with sterile water. The composition of the present invention contains a pharmaceutically active compound in the lipid phase of the emulsion.

## DETAILED DESCRIPTION OF INVENTION

Active ingredients which may be incorporated into the lipid phase of the emulsion of the present invention are any lipophilic compound and includes, for example, anticancer compounds such as adriamycin, trimetrexate, carmustine, semustine, lomustine, streptozotocin, methotrexate, cyclophosphamide, bleomycin; barbiturates such as hexobarbital, thiopental, pentobarbital, secobarbital, cyclobarbital; antiinflammatories such as phenyl butazone; cognition activators such as physostigmine salicylate; steroids such as prednisone, progestin, tamoxifen, androgens, dexamethasone palmitate; tranquilizers such as diazepam; antiepileptics such as phenytoin; antivirals such as acyclovir, vidarabine, idoxuridine; anti-AIDS drugs such as zidovudine (AZT); and cytotoxic and antifungal agents such as penclomedine and rhizoxin.

Oils suitable for use in the oil-in-water emulsion include medium chain triglycerides, linoleic acid, and vegetable oils such as soybean, safflower, sesame, sunflower, olive, rapeseed, and bran oils. Mixtures of the oils may be used also.

The emulsifier or surfactant used in the present invention is, for example, egg yolk phospholipids, i.e., egg lecithin, and the quantity employed varies from 1% to 2%. Additionally, non-ionic surfactants

can be employed and the amount of such surfactant used varies from 0.5% to 2%. Illustrative examples of suitable non-ionic surfactants include Pluronic F68, non-ionic esters of glycerol stearate, glycerol distearate, propylene glycol monostearate, glyceryl monostearate, tetraglyceryl monooleate, sorbitan mono-, di-, and tri-acylates, sucrose mono-, di-, and tri-acylates, polysorbate 40, polyoxyethylene, and sorbitan monopalmitate. One can also use acetylated monoglycerides, in which case the amount employed can be up to 5%. Combinations of egg lecithin and non-ionic surfactant or egg lecithin and acetylated monoglycerides are useful in forming suitable compositions of the present invention.

Agents used to improve the isotonicity of the present composition include dextrose, glucose, glycerin, sorbitol, and xylitol or lower alcohols such as ethanol.

Other agents such as preservatives or anti-oxidants may find use in the invention.

A carbohydrate is employed to provide some bulk to the composition. The preferred carbohydrate is lactose although others such as dextrose, xylose, mannitol, dextran, maltose, and sucrose may be used.

It has been found that certain oils and carbohydrates are not particularly compatible. For example, dextran and lactose are more compatible with soybean and safflower oils than are some of the other oils.

As indicated above, the oil-in-water composition comprises about 5% to 30% of a lipid or oil; about 5% to 15% of a carbohydrate; from about 0.5% to 5% of emulsifier; from about 2% to 4% of glycerin or equivalent agent thereof; and from 60% to 70% of

water. All percentages as used herein are on a weight per volume basis.

The concentration of active ingredient employed in the present invention would be an amount equivalent to provide the quantity known to be suitable for an injectable unit dose. The emulsion is placed in a container, preferably a sterile vial, prior to lyophilization. The vial can be any of the known types available in the market including the type into which sterile water is injected to achieve reconstitution of the lyophilized emulsion cake as well as the two-chamber type vials wherein the lyophilized emulsion cake is contained in one chamber and the sterile water for reconstitution is contained in the other chamber.

The lyophilization process comprises subjecting the oil-in-water emulsion to a series of temperature gradients whereby the emulsion is frozen then gradually warmed to about 30°C. The entire process requires about 72 hours and provides the product in final packaged form ready for use. The major advantage of this process is the ease of manufacture of the finished product formulation and a stable formulation for compounds known to be unstable in the presence of water and insoluble in water but soluble in oil. Another major advantage is the fact that the entire process is carried out in a sterile environment so opportunity for contamination of the composition is virtually nonexistent.

The oil-in-water emulsion is prepared at about 50° to 55°C by combining the water and lactose and dispersing the egg lecithin in the aqueous phase. The drug or active ingredient is combined with the oil and the oil phase is added to the aqueous phase and emulsified by homogenizing the mixture. Following

homogenization, glycerin is added with stirring and additional water is added as needed. The emulsion is placed in vials and lyophilized by subjecting the emulsion to the temperature gradient set forth above.

The following examples are illustrative of the basic formula for the composition of the present invention. An active ingredient could be incorporated in the lipid phase.

EXAMPLE 1

Composition:

| | |
|---|---|
| Safflower Oil | 5% |
| Soybean Oil | 5% |
| Egg Phosphatides | 1.2% |
| Glycerin | 2.5% |
| Lactose USP, Spray Dried | 10% |
| Water for Injection, q.s. ad | 100% |

Lyophilization steps:

The composition is placed in vials containing 10 mL each and lyophilized as follows:

| | | |
|---|---|---|
| (a) | 0-24 hours: | -40°C |
| (b) | 24-36 hours: | Raise the temp. slowly |
| (c) | 36-48 hours: | +15°C |
| (d) | 48-72 hours: | 30°C |
| (e) | 72 hours: | Out |

Vacuum is applied between steps (a) and (b) to approximately less than 60 millitorr and the chamber is repressurized with nitrogen after step (d). Also at the beginning of the freeze-drying process, the chamber is cooled to about -60°C. The pH of the

emulsion is approximately 8.0 and the particle size is about 280 nm. After the lyophilization cycle is completed, the product is obtained in a dry, lyophilized form. The reconstitution of the product was achieved instantaneously with a mean particle size of approximately 340 nm.

In a similar manner the following compositions were lyophilized.

EXAMPLE 2

| | Quantity (%) |
|---|---|
| Hexobarbital (Sedative/Hypnotic) | 3.75 |
| Soybean Oil | 10.0 |
| Ethanol | 25.0 |
| Egg Phosphatides | 1.00 |
| Myrj 52 (Tradename ICI Americas) | 0.50 |
| Lactose USP, Spray Dried | 10.0 |
| Water for Injection qs ad | 100.0 |

EXAMPLE 3

| | Quantity (%) |
|---|---|
| Phenylbutazone (Antiinflammatory) | 2.00 |
| Soybean Oil | 10.0 |
| Acetylated monoglycerides | 5.00 |
| Glycerol | 2.50 |
| Pluronic F68 (Tradename BASF) | 0.50 |
| Lactose USP, Spray Dried | 10.0 |
| Water for Injection qs ad | 100.0 |

EXAMPLE 4

| | Quantity (%) |
|---|---|
| Diazepam (Tranquilizer) | 0.50 |
| Soybean Oil | 15.0 |
| Acetylated monoglycerides | 5.00 |
| Egg Phosphatides | 1.2 |
| Glycerol | 2.50 |
| Lactose USP, Spray Dried | 10.0 |
| Water for Injection qs ad | 100.0 |

EXAMPLE 5

| | Quantity (%) |
|---|---|
| Penclomedine (Cytotoxic agent) | 1.00 |
| Safflower Oil | 10.0 |
| Egg Phosphatides | 1.20 |
| Glycerol | 2.50 |
| Lactose USP, Spray Dried | 10.0 |
| Water for Injection qs ad | 100.0 |

EXAMPLE 6

| | Quantity (%) |
|---|---|
| Rhizoxin (cytotoxic and antifungal) | 0.04 |
| Soybean Oil | 10.0 |
| Egg Phosphatides | 1.20 |
| Glycerol | 2.50 |
| Lactose USP, Spray Dried | 10.0 |
| Water for Injection qs ad | 100.0 |

EXAMPLE 7

| | Quantity (%) |
|---|---|
| Physostigmine Salicylate (Cognition) | 0.10 |
| Oleic Acid | 6.00 |
| Soybean Oil | 1.20 |
| Egg Phosphatides | 1.20 |
| Pluronic F68 | 2.00 |
| Glycine | 4.20 |
| Glycerol | 2.25 |
| Methylparaben | 0.20 |
| Butylparaben | 0.075 |
| Ascorbic Acid | 0.1 |
| α-Tocopherol | 0.02 |
| Lactose USP, Spray Dried | 10.0 |
| Water for Injection qs ad | 100.0 |

CLAIMS

1. A process for lyophilizing an oil-in-water emulsion which comprises
(a) cooling the emulsion at -40°C for 0 to 24 hours;
(b) slowly raising the temperature to +15°C over a 12-hour period;
(c) holding the temperature at +15°C for 12 hours; and raising the temperature to +30°C for 24 hours.

2. The process of Claim 1 wherein the oil-in-water emulsion is a composition comprising
(a) from 5% to 30% of an oil suitable for injection;
(b) from 0.5% to 5.5% of a surfactant;
(c) from 5% to 15% of a carbohydrate;
(d) from 2% to 4% of an agent to provide isotonicity; and
(e) from 60% to 70% water.

3. The process of Claim 2 wherein the oil contains a lipophilic pharmaceutically active compound.

4. The process of Claim 3 wherein the emulsion is placed in a sterile vial prior to lyophilization.

5. The process of Claim 3 wherein the surfactant is egg lecithin present in an amount of from 1% to 2%.

6. The process of Claim 3 wherein the surfactant is egg lecithin in combination with a non-ionic surfactant.

7. The process of Claim 5 wherein the carbohydrate is lactose.

8. The process of Claim 3 wherein the agent employed to provide isotonicity is glycerin.

9. The process of Claim 4 wherein the pharmaceutically active compound is selected from an anticancer agent, a barbiturate, an antiinflammatory agent, a steroid, an antiepileptic, an antiviral, an anti-AIDS agent, a cytotoxic agent, an antifungal agent, a cognition activator, or a tranquilizer.

10. The process of Claim 9 wherein the compound is selected from AZT, hexobarbital, phenylbutazone, diazepam, penclomedine, rhizoxin, or physostigmine salicylate.

11. An oil-in-water emulsion comprising
(a) from 5% to 30% of an oil suitable for injection;
(b) from 0.5% to 5.5% of a surfactant;
(c) from 5% to 15% of a carbohydrate;
(d) from 2% to 4% of an agent to provide isotonicity; and
(e) from 60% to 70% water.

12. An emulsion of Claim 11 wherein the oil contains a pharmaceutically active compound.

13. An emulsion of Claim 12 wherein the surfactant in egg lecithin present in an amount of from 1% to 2%.

14. An emulsion of Claim 12 wherein the surfactant is egg lecithin in combination with a non-ionic surfactant.

15. An emulsion of Claim 12 wherein the carbohydrate is lactose.

16. An emulsion of Claim 12 wherein the agent employed to provide isotonicity is glycerin.

17. An emulsion of Claim 12 wherein the pharmaceutically active compound is selected from an anticancer agent, a barbiturate, an antiinflammatory agent, a steroid, an analeptic, an antiviral, an anti-AIDS agent, a cytotoxic agent, an antifungal agent, a cognition activator, or a tranquilizer.

18. An emulsion of Claim 17 wherein the compound is selected from AZT, hexobarbital, phenylbutazone, diazepam, penclomedine, rhizoxin, or physostigmine salicylate.

# INTERNATIONAL SEARCH REPORT

International Appl on No PCT/US 91/02353

## I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all)[6]

According to International Patent Classification (IPC) or to both National Classification and IPC

Int.Cl.5 A 61 K 9/14 A 61 K 9/107

## II. FIELDS SEARCHED

Minimum Documentation Searched[7]

| Classification System | Classification Symbols |
|---|---|
| Int.Cl.5 | A 61 K |

Documentation Searched other than Minimum Documentation to the Extent that such Documents are Included in the Fields Searched[8]

## III. DOCUMENTS CONSIDERED TO BE RELEVANT[9]

| Category ° | Citation of Document,[11] with indication, where appropriate, of the relevant passages[12] | Relevant to Claim No.[13] |
|---|---|---|
| X | EP,A,0211257 (ABBOTT LABORATORIES) 25 February 1987, see claims 1-6,10; page 2, line 33 - page 3, line 17; page 5, lines 26-30; page 9, lines 8-10, 19-25; page 10, line 30 - page 11, line 7; page 14, line 10 - page 15, line 14; examples (cited in the application) --- | 1-5,7-10,12-18 |
| X | EP,A,0159237 (LABORATOIRE L. LAFON) 23 October 1985, see claim 1; page 19, line 5 - page 20, line 9 ----- | 1 |

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## IV. CERTIFICATION

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| EUROPEAN PATENT OFFICE | Mme Dagmar FRANK |

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# ANNEX TO THE INTERNATIONAL SEARCH REPORT ON INTERNATIONAL PATENT APPLICATION NO. US 9102353 SA 46959

This annex lists the patent family members relating to the patent documents cited in the above-mentioned international search report. The members are as contained in the European Patent Office EDP file on 06/11/91
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