INJECTION COMPRISING DOCETAXEL AND ITS PREPARATION

BACKGROUND OF THE INVENTION

FIELD OF THE INVENTION

The present invention discloses a injection containing Docetaxel compounds, particularly a injection containing Docetaxel compounds with a prolonged stability and convenience for medical personnel, said injection is a three-part injection containing Docetaxel compounds.

DESCRIPTION OF RELATED ART

Paclitaxel, which belongs to the class of taxane, was approved to be a new anti-cancer drug by U.S. Food and Drug Administration (FDA) in 1992, and have been recognized as one of the most important anti-cancer drugs since then. However, it was not very successful when the drug first entered the market, for that, in addition to the limited resource of its raw material such as Taxus brevifolia and Taxus sumatrana (Miq.) de Laub, it is also found that the patient administered with the Paclitaxel suffered from serious allergy in Phase I clinical trials. The allergen is not Paclitaxel itself, but the solvent to dissolve Paclitaxel, Cremophor EL. To prevent the allergy from Cremophor EL, the current research direction has focused on finding a new solvent to replace Cremophor EL.

Due to the high hydrophobicity of taxane, whose solubility in water is less than 10μg/mL, the process to find a new solvent to replace Cremophor EL for dissolving the Paclitaxel is not very successful. The only Paclitaxel-containing drug approved currently is

Taxol® IV injection developed by U.S. Bristol-Myers Squibb company. However, the solvent of the IV injection used for dissolving Paclitaxel is still a mixture of ethanol and Cremophor EL at 1:1, and it is still possible to cause allergy in patients even with low Cremophor EL content.

Some researchers have proposed to use organic solvent in place of Cremophor EL to dissolve Paclitaxel (see, Andersson, B, "Parenteral Paclitaxel in a Stable Non-toxic Formulation", USP5,877,205, March 2, 1999; Hausheer, F.H., Murali, D., and, Seetharamulu, P., USP6,040,330, "Pharmaceutical Formulations of Taxanes", March 21, 2000). Even though the preparation process disclosed by these prior art is simple and has high potential in the market, it is not widely accepted due to high toxicity of the organic solvent.

To find a substitute solvent, many researchers forward the study in two different aspects:

1. Improvement of the formula of the injection

It mainly uses liposomal agent and emulsifying agent to dissolve Paclitaxel. However, the liposomal formulation forms precipitation during storage, which lowers the stability of the drug. In order to solve the problem, the injection is stored as lyophilized crystal powder. However, it is required to freeze dry the liposomal formulation into lyophilized crystal powder by using a costly freeze drying preparation technique, which is not economical and results in a product unworthy to be commercialized.

The research of emulsion have been disclosed by Panayiotis, P. et al, WO02/26208, "Emulsion Vehicle for Poorly Soluble Drugs", April 4, 2002; Kaufman, R.J., and Richard,

T.J., USP5,616,330, "Stable Oil-In-Water Emulsions Incorporating a Taxine (Taxol) and Method of Making Same", April 1, 1997; Chu, I.M., and Wang, T.R., USP6,348,491, "Oil-In-Water Emulsion for Encapsulation Paclitaxel", Feb.19, 2002, and the Paclitaxel emulsion produced with an emulsifying agent shows excellent effects. But the preparation process thereof is complicated and costly, and it also forms crystal precipitations like the liposomal formulation during storage, therefore this pharmaceutical formulation has an insufficient stability.

2. Synthesis of Docetaxel having a higher solubility than Paclitaxel

It is very difficult to find a good solvent for taxane due to its low solubility in water, so Docetaxel with a higher solubility than Paclitaxel is synthesized chemically. Taxotere® IV injection made by Docetaxel has been approved by FDA to enter the market, wherein the injection is dissolved with a surfactant. It is a great breakthrough in this field.

However, there are still some disadvantages for dissolving Docetaxel with surfactant instead of Cremophor EL. While Tween is selected as the surfactant with a lower toxicity than typical organic solvents, a formulation in which Docetaxel is dissolved with Tween needs to be mixed with perfusate such as 5% glucose solution prior to injection. As a result, the mixture may be so viscous to form a gel, thus the dissolution requires a vigorous stirring. To improve the defects above, Rhone-Poulenc proposed a two-part injection composition with the addition of an intermediate solution, thereby prevent the mixture from gelling (see, JEAN-MARC BOBEE, PATRICK DE LANTY, GILLES GUERIN, MICHEL VEILLARD, TW00271395, " Two-part injection composition comprising a taxane derivative in surfactant and an additive to prevent gelling

on dilution"), wherein the intermediate solution includes an additive such as organic compounds (MW<200) or sodium chloride and water. The intermediate solution is an aqueous solution having the additive at a concentration higher 6% by weight than that of surfactant. According to the claims of said patent, the intermediate solution is a solution of 13% ethanol diluted in water. However, the addition of the intermediate solution may result in the following problems:

(a) Problems in usage: According to USP27 General Chapters<1151> pharmaceutical dosage forms-injections, it is required to fill the syringe with excess volume when injecting a general viscous fluid. For instance, the excess volume to fill is 0.12 ml (24% by weight) for a 0.5 ml injection amount. The viscosity of the injection containing Docetaxel and surfactant is so high that a high excess volume is required as well, thus increasing the cost and it takes long time for medical personnel to withdraw the viscous injection. Moreover, this would lead to a poor control on the actual amount withdrawn, thus affecting the dosage accuracy and therapeutic effect. Even though the addition of intermediate solution facilitates to reduce the viscosity of the injection, this reduction is limited and the problem still exists.

(b) Preparation risk: A commercially available ethanol solution has a ethanol concentration of 95%. In order to prepare 13% by weight ethanol intermediate solution, a 95% ethanol solution must be weighted and mixed accordingly with water. However, there are may be some variance among the concentration of ethanol solution prepared by each human at each time. Besides, an additional sterilization step is required for fulfilling the safety issue, which greatly increases the complexity and risk of the manufacturing process.

Therefore, the use of the commercially available 95% ethanol solution would enable a relatively easier production and a lower risk in the sterilization step, with an obvious progress.

(c) Stability problem: The formulation prepared can not be stable in a prolonged period. Moreover, it is emphasized in the instruction of the product provided by Rhone-Poulenc that the product must be used within 4 hours once mixed with the perfusion. Moreover, a crystal precipitation forms after 12 hours in the mixture of said product, it has been proved by an experimentation. The crystal precipitation may clog blood vessels, and cause fatal danger to the patient.

SUMMARY OF THE INVENTION

In view of the low stability of the IV injection of Docetaxel (Taxotere®) which is dissolved with the aforementioned solvent, a three-part injection containing Docetaxel is developed.

An object of the present invention is to provide a three-part injection containing Docetaxel with a prolonged stability and convenience for medical personnel.

For the above purpose, the present invention provides a three-part injection containing Docetaxel, which includes:

a mother liquid, which is a solution comprising Docetaxel and a pharmaceutically acceptable surfactant;

ethanol, which is an anti-gelling agent for preventing gel formation after the injection is mixed; and

water, which serves as a flow agent to keep the mother liquid in a homogenized state, in synergy with the ethanol.

Wherein, the weight ratio of the anti-gelling agent to the surfactant is not less than 1%; the weight ratio of the flow agent to the surfactant is not less than 10%.

The present invention can prevent, by adding the anti-gelling agent, gel formation when the mother liquid is mixed with perfusion solution, such as an aqueous 5% glucose solution, and the flow agent can adjust the viscosity of the mother liquid, together with the anti-gelling agent. Therefore, the three-part injection prepared by the method according to the present invention has a reduced viscosity and is homogeneous without gel formation. This provides convenience for medical personnel to withdraw the injection with a syringe and confirm the amount withdrawn. Furthermore, the period for Docetaxel to form a crystal precipitate from the injection solution is prolonged, thereby increasing the stability of the injection, reducing the fatal danger to patients administered with medicament and enhancing the therapeutic quality.

In the injection containing Docetaxel according to the present invention, the pharmaceutically acceptable surfactant is preferably Tween. The concentration of ethanol used therein is preferably not less than 90%, more preferably not less than 95%. The concentration of Docetaxel in the mother liquid is preferably 20 mg/ml to 60 mg/ml. The weight of the anti-gelling agent of ethanol is preferably at least 1% of that of Tween; the weight of the water is preferably at least 10% of that of Tween.

The present invention also provides a method for preparing a injection containing Docetaxel, which comprises steps of: (a) mixing a mother liquid and ethanol evenly into a

mixture liquid; and (b) adding water into the aforementioned mixture liquid and mixing it evenly.

BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 shows a schematic top view of a cylindrical injection vial in the homogeneity test of the present invention.

DETAILED DESCRIPTION OF THE INVENTION

The following detailed description to the present invention is made referring to the accompanied figures and does not intend to limit the scope of implementing the present invention.

The injection containing Docetaxel according to this invention including:

a mother liquid, comprising a solution of Docetaxel and one or more pharmaceutically acceptable surfactants, wherein the surfactant is Tween (i.e. polysorbate), and the concentration of Docetaxel dissolved in Tween is 20 mg/ml to 60 mg/ml;

an anti-gelling agent, which prevents gel formation when the mother liquid is mixed with a perfusate, such as 5% glucose solution, wherein the anti-gelling agent is preferably 95% ethanol, and the weight of the ethanol is not less than 1% of that of Tween (ethanol $\geqslant$ 1% by weight of Tween); and

a flow agent, which is water to keep the mother liquid in a homogenized state, in synergy with the ethanol, and serves to adjust the viscosity of the mother liquid, wherein the flow agent is pure water or water for injection, and the weight the flow agent is not less than 10% of that of Tween (water $\geqslant$ 10% by weight of Tween).

The three-part injection, which comprises the anti-gelling agent, the flow agent and the mother liquid, can be prepared in a more flexible way, and the adjustment to viscosity of the mother liquid can be more flexibly, comparing with the conventional composition comprising the intermediate solution and mother liquid.

The method of preparing a three-part injection containing Docetaxel according to the present invention includes mixing the mother liquid and the anti-gelling agent evenly, then adding and evenly mixing the flow agent.

The preparation method described above enables each component mixed to reach a homogeneous state, at the time to use, by mixing them directly through hand shake or a machine (such as desk Vortex) at an ambient temperature.

The advantages of the injection containing Docetaxel compounds of the present invention are further depicted below with the illustration of examples, which however should not be construed as limitations to the scope of the present invention.

Example 1: Observation on gel formation resulted from different methods of mixing

The present invention adopts different methods of mixing to ensure ease of use, and the gel formation was observed.

Control group:

The test in this group was performed by mixing firstly the anti-gelling agent with the flow agent, which is similar to the intermediate solution in prior art, then carrying out different mixing steps. The mother liquid, anti-gelling agent and flow agent used are listed in Table 1, whereas Table 2 and 3 list different methods of mixing and corresponding

observation results.

Table 1

Mother liquid	Intermediate solution comprising mixture of anti-gelling agent and flow agent
Docetaxel and surfactant Tween (0.5 ml in total)	1.5 ml 13% by weight ethanol formed by mixing 0.24 ml ethanol and 1.26 ml water

Table 2

Mixing method 1		Observation Result
Step 1	inject the intermediate solution of Table 1 into the mother liquid to form a mixture solution	No gel formation
Step 2	inject the mixture solution into 500 ml perfusate	No gel formation

Table 3

Mixing method 2		Observation Result
Step 1	inject the intermediate solution of Table 1 into 500 ml perfusate	No gel formation
Step 2	then inject the mother liquid to perfusate (containing anti-gelling agent and flow agent)	Gel formation

It can be found, by comparing the two mixing methods above, that the final mixture liquids had the same composition, but different local compositions surrounding Tween molecules. In mixing method 1, the anti-gelling agent and flow agent would attach to the Tween molecules firstly, so that there were more anti-gelling agent molecules surrounding

Tween molecules (i.e. the local concentration of anti-gelling agent around surfactant molecule is higher) to prevent gel formation. In mixing method 2, the anti-gelling agent and flow agent had already been diluted by the perfusate before the mother liquid containing Tween was injected into the perfusate, which lowered the concentration of the anti-gelling agent molecule around Tween molecules, so the gel formed in variable extents throughout the whole solution.

It is thus known that the main cause to prevent gel formation is not the amount of the anti-gelling agent used, but is whether the anti-gelling agent can be mixed evenly with surfactant molecule in the mother liquid during the process of mixing so as to prevent aggregation with other surfactant molecules.

Experimental Group:

The mother liquid, anti-gelling agent and flow agent used in this group are listed in Table 4, but the mixing steps are different from those described in the control group in that: the anti-gelling agent and the flowing agent were mixed with the mother liquid separately. Table 5 lists the mixing steps and corresponding observation results which can be compared with those in the control group.

Table 4

Mother liquid	Anti-gelling agent	Flow agent
Docetaxel and surfactant Tween (2ml in total)	0.1 ml ethanol	0.53 ml water

Table 5

Mixing method 3	Observation Result

Step 1	inject the anti-gelling agent into the mother liquid	No gel formation
Step 2	inject the flow agent into the mother liquid (containing the anti-gelling agent)	No gel formation
Step 3	inject the mother liquid (containing the anti-gelling agent and the flow agent) into the perfusate	No gel formation

In this example, the anti-gelling agent and the flow agent were added to the mother liquid sequentially (i. e., the flow agent and the anti-gelling agent were mixed with the mother liquid), which is different from mixing method 2. It was observed that the concentration of anti-gelling agent molecules around surfactant molecules was higher. According to this mixing method, the ratio of anti-gelling agent molecule to surfactant molecule can be 5 % by weight (< 6 % by weight) without gelling.

Example 2: Fluidity test

The injections with or without the flow agent are listed in Table 6, and mixing of each of the injections was observed and compared.

Table 6

Mother liquid	Anti-gelling agent	Flow agent	Observation Result
0.5 ml	0.24 ml ethanol		No gel formation
0.5 ml	0.24 ml ethanol	1.26 ml water	No gel formation and the fluidity was better than sample item 1
1.0 ml	0.24 ml ethanol		No gel formation

1.0 ml	0.24 ml ethanol	1.26 ml water	No gel formation and the fluidity was better than sample item 1

From the results of this example, it was known that the addition of flow agent in the injection reduces the viscosity of the solution, and therefore reduces the excess volume filled so as to reduce the production cost. Furthermore, it is more convenient to withdraw the injection, which allows a precise control on the dosage of injection withdrawn.

Example 3: Homogeneity test

After each components are mixed, the injections were produced by different stirring processes, and then tested for their homogeneity.

As shown in FIG. 1, homogeneity test was performed by using the top view of a cylindrical injection vial as a reference, taking the central point as well as points at 0°, 90°, 180° and 270° of the circumference of the cross section of the vial as sampling points for homogeneity, and then analyzing by High Performance Liquid Chromatography.

Control group:

Similar to the intermediate solution in prior art, an intermediate solution was formed in this group by mixing the anti-gelling agent with the flow agent firstly. And then the intermediate solution thus formed was withdrawn, injected slowly into the mother liquid, stirred by different stirring processes, and analyzed. Table 7 shows the used mother liquid, anti-gelling agent and flow agent, whereas Tables 8 and 9 show the observation results after stirring.

Table 7 composition of two-part injection composition

Sample item	Mother liquid	Intermediate solution comprised the mixture of anti-gelling agent and flow agent
1	0.5 ml	1.5 ml 13 % by weight ethanol (mixture of 0.24 ml ethanol and 1.26 ml water)
2	2.0 ml	6.0 ml 13 % by weight ethanol (mixture of 0.96 ml ethanol and 5.04 ml water)

The intermediate solutions of sample 1 and 2 were withdrawn and injected slowly into the mother liquid, and then the resulting mixture liquid was mildly spinned and stirred for at least 40 seconds so as to prevent the formation of gas bubbles during the mixing of mother liquid and intermediate solution. The analysis results obtained are shown below:

Table 8 Homogeneity test of two-part injection composition after mild stirring

Test item	Central points	0°	90°	180°	270°	Relative standard deviation
1	5.5	4.4	1.7	8.2	4.3	49%
2	7.5	8.9	7.3	6.6	9.0	13%

(the unit of concentration is mg/ml)

The intermediate solutions of sample item 1 and 2 were withdrawn, injected slowly into the mother liquid, and then stirred vigorously by a desk Vortex for at least 40 seconds to create gas bubbles in the mixture liquid of intermediate solution and mother liquid. The analysis results obtained are shown below:

Table 9 Homogeneity test of two-part injection composition after vigorous stirring

Sample item	Central points	0°	90°	180°	270°	Relative standard deviation
1	9.4	8.8	10	9.8	9.1	5%
2	9.1	9.4	9.5	10.3	8.8	5%

(the unit of concentration is mg/ml)

From the above results, it is known that the injection stirred vigorously is mixed more homogeneous than the injection stirred mildly.

Experimental group:

In the experimental group, the anti-gelling agent and the flow agent were injected into the mother liquid separately, stirred by different ways, and then analyzed. The used mother liquid, anti-gelling agent and flow agent are shown in Table 10, whereas the analysis results after stirring are shown in Tables 11 and 12 and were compared with those of control group.

Table 10 composition of three-part injection composition

Sample item	Mother liquid	Anti-gelling agent	Flow agent
3	0.5 ml	0.24 ml ethanol	1.26 ml water
4	2.0 ml	0.96 ml ethanol	5.04 ml water
5	1.5 ml	0.24 ml ethanol	1.26 ml water
6	2.0 ml	0.48 ml ethanol	2.52 ml water

Each anti-gelling agent in the three-part injection composition from sample items 3, 4, 5, 6 was withdrawn and added into the mother liquid, and then was stirred vigorously by a desk Vortex for at least 20 seconds. After that, each flow agent was respectively added and mildly spinned and stirred for at least 20 seconds so as to prevent the formation of gas

bubbles in the final solution during stirring. The analysis results obtained are shown below:

Table 11 Homogeneity test of three-part injection composition after mild stirring

Sample item	Central points	0°	90°	180°	270°	Relative standard deviation
3	9.8	10.2	9.8	10.1	9.7	2%
4	10.2	9.7	9.6	9.9	20	2%
5	19.8	20.1	20.2	19.6	19.5	1%
6	15.7	16.3	16.1	15.6	15.4	2%

(the unit of concentration is mg/ml)

Each anti-gelling agent of the three-part injection composition from sample items 3, 4, 5, 6 was withdrawn and added into the mother liquid, and then was vigorously stirred by a desk Vortex for at least 20 seconds. After that, each flow agent was respectively added and vigorously stirred by the desk Vortex for at least 20 seconds to creates gas bubbles in the mixture liquid of anti-gelling agent and mother liquid. The analysis results obtained are shown below:

Table 12 Homogeneity test of three-part injection composition after vigorous stirring

Sample item	Central points	0°	90°	180°	270°	Relative standard deviation
3	10	9.9	10.1	9.9	9.9	1%
4	9.9	10	10	9.8	10.1	1%
5	20.1	19.9	20	20.2	19.8	1%
6	16.2	16	15.9	15.8	16	1%

(the unit of concentration is mg/ml)

It is known from example 1 that the local concentration of anti-gelling agent

around the surfactant molecule is higher, therefore, in addition to reduce the ratio of gel in the solution, the resistance against homogeneity from the gel itself can be reduced. Compared to the experimental group of present invention, the surfactant in the two-part injection composition of the control group was pre-diluted, which leads to a low local concentration of the anti-gelling agent around surfactant molecule, so that the ratio of gel in the solution and the resistance against homogeneity from the gel itself cannot be reduced. Even after a vigorous stirring, the homogeneity of control group is still lower than that of the three-part injection composition of the present invention.

Example 4: Stability test

The injection composition was mixed, and placed at room temperature to observe the formation of crystal at different time.

Control group:

The intermediate solutions of sample item 1 and 2 in Table 7 were withdrawn, injected slowly into the mother liquid respectively, then stirred vigorously by a desk Vortex for at least 40 seconds, and tested for their stabilities. The results are shown in Table 13:

Table 13 Crystal test for two-part injection composition after vigorous stirring

Sample item	0 hour	4 hours	8 hours	12 hours	16 hours	24 hours
1	No crystal	No crystal	No crystal	Few crystals	Obvious crystals	Massive crystals
2	No crystal	No crystal	No crystal	Few crystals	Obvious crystals	Massive crystals

Experimental group:

Each anti-gelling agent of the three-part injection composition of sample item 3, 4, 5, 6 in Table 10 was withdrawn and added to the mother liquid and then was stirred vigorously by a desk Vortex for at least 20 seconds. After that, each flow agent was respectively added and vigorously stirred by the desk Vortex for at least 20 seconds, then was tested for their stabilities. The results are shown in Table 14, and compared with control group:

Table 14 Crystal test for three-part injection composition after vigorous stirring

Sample item	0 hour	4 hours	8 hours	12 hours	24 hours	48 hours
3	No crystal	No crystal	No crystal	No crystal	No crystal	No crystal
4	No crystal	No crystal	No crystal	No crystal	No crystal	No crystal
5	No crystal	No crystal	No crystal	No crystal	No crystal	No crystal
6	No crystal	No crystal	No crystal	No crystal	No crystal	No crystal

It is known from examples 1 and 3 that the local concentration of anti-gelling agent around the surfactant molecule is higher, therefore, in addition to reduce the ratio of gel in the solution and the resistance against homogeneity of the gel itself, it is also possible to increase the energy barrier between surfactant molecules to prevent crystal precipitation due to the intermolecular aggregation of Docetaxel molecules. Accordingly, the local concentration of anti-gelling agent around surfactant molecules in the two-part injection

composition of control group is lower, and therefore the energy barrier between surfactant molecules is reduced. Thus, the two-part injection composition forms crystal precipitate after standing for 12 hours, whereas all the three-part injections of the experimental group of the present invention have no crystal precipitate within 48 hours.

The addition of anti-gelling agent prevents gel formation in the mother liquid when it is mixed with the perfusate at the time of use, and the flow agent can adjust the viscosity of mother liquid, in synergy with the anti-gelling agent. Therefore, the three-part injection prepared by the method of present invention has a reduced viscosity and is homogeneous without gel formation. The three-part injection of the present invention allows medical personnel to withdraw it more conveniently with a syringe, and confirm the amount withdrawn. Besides, the period for Docetaxel to form a crystal precipitate from the injection solution is prolonged, thereby increasing the stability of the injection, reducing the fatal risk to patients and improving the medical quality.

What is claimed is

1. A three-part injection containing Docetaxel, comprising:

a mother liquid, which is a solution comprising Docetaxel and a pharmaceutically acceptable surfactant;

ethanol, which is an anti-gelling agent for preventing gel formation after the injection is mixed; and

water, which serves as a flow agent to keep the mother liquid in a homogenized state, in synergy with the ethanol,

wherein the mother liquid, ethanol and water are mixed in sequence to reach a homogenous state at the time of use, and then diluted into the perfusate.

2. The injection containing Docetaxel according to claim 1, wherein the pharmaceutically acceptable surfactant is polysorbate.

3. The injection containing Docetaxel according to claim 1, wherein the ethanol has a concentration of no less than 90%.

4. The injection containing Docetaxel according to claim 1, wherein the ethanol has a concentration of no less than 95%.

5. The injection containing Docetaxel according to claim 2, wherein the Docetaxel in the mother liquid has a concentration of 20 mg/ml to 60 mg/ml.

6. The injection containing Docetaxel according to claim 2, wherein the weight of the ethanol is at least 1% of the weight of the polysorbate.

7. The injection containing Docetaxel according to claim 2, wherein the weight of the water is at least 10% of the weight of the polysorbate.

8. A method for preparing the three-part injection containing Docetaxel according to claim 1, comprising steps of:

(a) mixing the mother liquid and the ethanol evenly into a mixture liquid; and

(b) adding the water into the mixture liquid and mixing it evenly.