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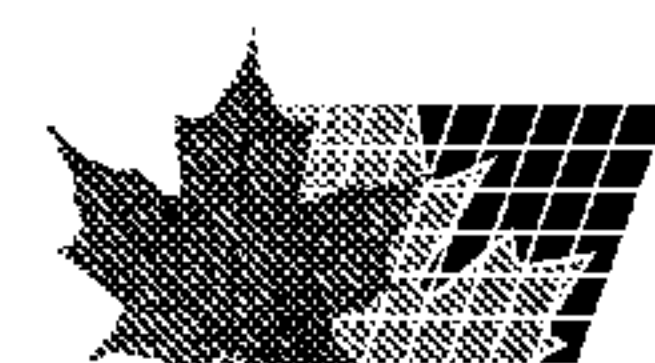
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(54) Titre : FORMULATIONS LIQUIDES ET LYOPHILISEES
(54) Title: LIQUID AND FREEZE DRIED FORMULATIONS

(57) **Abrégé/Abstract:**

The present invention relates to stable parenteral formulations comprising a new class of compounds, which is a non proteinic compound, a method for preparing such formulations as well as the use of certain compounds for stabilizing these formulations.



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(54) Title: LIQUID AND FREEZE DRIED FORMULATIONS

(57) Abstract: The present invention relates to stable parenteral formulations comprising a new class of compounds, which is a non proteinic compound, a method for preparing such formulations as well as the use of certain compounds for stabilizing these formulations.



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Liquid and freeze dried formulations

The present invention relates to liquid and freeze dried formulations of a new class of compounds, which is a non proteinic compound, a method for preparing such formulations as well as the use of certain compounds for stabilizing these formulations.

The active substance used in the present invention is a chimeric molecule consisting of two dopamine agonist, small molecule moieties covalently linked to the N-terminus of a cyclic peptide somatostatin analogue and acts as a chimeric compound.

Specifically, the invention concerns stable formulations at temperatures that can range from + 5° C to + 40° C and which can be reconstituted in liquid form by adding a solvent for its administration through parenteral administration.

As further object, the present invention encompasses a stable liquid pharmaceutical formulation at temperatures that can range from + 5° C to + 40° C and 60 to 85 % RH (relative humidity) preferably 75 % RH. This liquid formulation may be kept in a vial or as a ready to use formulation in a syringe type device.

It is known that freeze-drying process may have a considerable effect on degradation of the pharmaceutically active substance in the formulation, as well as a strong influence on their stability in freeze dried form or after reconstitution of the suspension or solution.

Difficulties are also encountered for liquid formulations in order to maintain activity, efficiency and ratio of the active substances as well as clarity of the liquid over time.

Various approaches to stabilizes freeze-dried and liquid formulations have been discussed in the literature.

For instance, pharmaceutical compositions in the form of freeze-dried preparation for parenteral injection use are described in the scientific publication "*Stabilisation of Octastation, a somatostatin analogue. Preparation of freeze-dried products for parenteral injection*", H. Pourrat & Al, Biological and Pharmaceutical Bulletin, Vol. 18, No. 5, pp 766-771, The Pharmaceutical society of Japan. The publication describes a formulation with glutamic acid-sodium glutamate buffer as a stabilizing agent and the freeze-drying procedure found is appropriate for subsequent industrial production.

The use of sugars as stabilizing agent is known with proteins in pharmaceutical formulation. In the international patent application WO 0056365, compositions comprising an antigen which consists of a polysaccharide bound to a carrier protein and trehalose are reported.

The interest for novel dopamine-somatostatin chimeric molecules with differing, enhanced activity at specific receptor has been demonstrated in the scientific publication "*Efficacy of chimeric molecules directed towards multiple somatostatin and dopamine receptors on inhibition of GH and prolactin secretion from GH-secreting pituitary adenomas classified as partially responsive to somatostatin analog therapy*", P Jaquet & Al, European Journal of Endocrinology, Vol 153, Issue 1, pp 135-141. These class of compounds, consistently produced significantly greater suppression of GH (growth hormone) and PRL (prolactin) than either octreotide or single-receptor-interacting ligands in tumors from patients classified as only partially responsive to octreotide therapy.

The problem solved by the present invention is to provide stable formulations over time containing this chimeric compound in freeze-dried form or liquid form by means of the specific freeze-dried process and/or adequate choice of excipients. The present invention provides a composition which is stable for at least 24 months in liquid or freeze dried form and additionally a process for freeze drying such compositions.

The following definitions are set forth to illustrate and define the meaning and scope of the various terms used to describe the invention herein.

The terms "lyophilised" "freeze-dried" "lyophilization" as used herein refers to a method of isolating a solid substance from solution by freezing the solution and evaporating the ice under vacuum.

The term "lyophilisate" as used herein refers to a drug substance after freeze-drying process on its own or with different injectable bulking agents.

The term "bulking agents" as used herein consists of a non reducing sugar or a polyhydric alcohol (polyol).

The term "stability" as used in the present invention refers to chemical and physical stability.

The term "powder for solution for infusion" as used herein consists of a powder, such as lyophilisate that can be quickly reconstituted (extemporaneously) in solution with water to be administered by parenteral route.

The term "immediate release formulation" as used herein consists of a release of the active substance for a duration of one day to one week.

The term "amino acid" as used in the present invention denotes an amino acid comprising but not limited to arginin, glycine, lysine, histidine, glutamic acid, aspartic acid, isoleucine, leucine, alanine, phenylalanine, tryptophane, serine, methionine, proline the amino acid may used singly or in combination.

The term "pharmaceutically acceptable" means in this context physiologically well-tolerated by a mammal or a human.

The term "high concentrated" in the present invention generally means a content in active substance of 0.1 mg/ml to 20 mg/ml preferably from 1 to 10 mg per 1 vial.

The term "buffered isotonic" and "buffered saline" solution refers to a solution containing a salt in addition to a buffer.

The term "water for injection" as used herein means sterile water obtained after filtration or by known techniques from the skilled person.

Solutol[®] used in liquid formulation is macrogol 12 Hydroxystearate, a non ionic solubilizer for injection solutions.

Description of figure 1 :

Figure 1 : shows the content in active substance (% initial) of BIM23A760 in function of storage for 3, 6, 9, and 18 months at various temperatures.

According to the invention, a further object is the use of sugars such as saccharides or disaccharides or polyols or a mixture thereof for the stability of the formulation as well as for storage stability of the liquid or freeze-dried preparation.

Accordingly, saccharides disaccharides and polyols include xylitol, maltose, lactose, galactose, mannitol, sorbitol, dextran and threulose or combination thereof although not limited to these.

Preferred are mannitol, xylitol and threulose even more preferred is threulose.

In a preferred embodiment, the present formulation, freeze dried and/or liquid formulation, comprises between 1 to 50 % by weight of the active substance BIM23A760 (BIM23A760 is Dop2-DLys(Dop2)-c(Cys-Tyr-DTrp-Lys-Abu-Cys)-Thr-NH₂) in the formulation or other dopamine derivatives or salt thereof preferably 15 to 30 % by weight of BIM23A760 in the formulation or other dopamine derivatives or salt thereof.

The quantity of polyols, sugars, saccharides or disaccharides according to the freeze-dried formulations of the present invention, comprises about 50 to 95 % by weight of the formulation, preferably the amount of polyol or sugars such as saccharides or disaccharides is between 70 % to 85 % by total weight of the formulation.

The quantity of polyols, sugars, saccharides or disaccharides according to the liquid formulations of the present invention, comprises about 1 to 15 % by weight of the formulation, preferably the amount of polyol or sugars such as saccharides or disaccharides is between 5 % to 8 % by total weight of the formulation.

The preferred pH range of the formulation is from 3.0 to 7.0, wherein 4.0 to 6.0 is particularly preferred.

Preferably the compositions are in the form of freeze-dried product, to be reconstituted by addition of a solvent. The solvent may be an isotonic solution, a buffered isotonic solution, a buffered saline solution or water for injection.

In addition, the compositions of the invention may further comprise a conventional buffering agent, such as amino acid or organic salts which include boric acid, phosphoric acid, acetic acid, and citric acid and/or a salt thereof.

The formulations disclose herein may also contain an isotonic agent. A suitable isotonic agent for the formulation according to the invention include propylene glycol, glycerol, sodium chloride, potassium chloride in the amount of 0.1 to 5 % (w/w), preferably in the amount of 0.9 to 3 % (w/w).

Pharmaceutical salts of active substance usable for compositions according to the invention may be produced by acid addition salts with organic and inorganic acids. Examples of acid addition salts of compounds are salts with mineral acids, for example hydrohalic acids such as hydrochloric acid, hydrogen bromide and hydrogen iodide, sulphuric acid, nitric acid, phosphoric acid and the like, salts with organic sulphonc acids, for example with alkyl- and arylsulphonic acids such as methanesulphonic acid, p-toluenesulphonic acid, benzenesulphonic acid and the like, as well as salts with

organic carboxylic acids, for example with acetic acid, tartaric acid, maleic acid, citric acid, benzoic acid, fumaric acid, oxalic acid, stearic acid, salicylic acid, ascorbic acid or insoluble salts such as pamoic acid and the like.

Whether the active substance contains a carboxyl group also form pharmaceutically acceptable salts with bases. Examples of such salts are alkali metal salts, for example sodium and potassium salts, ammonium salts, salts with organic bases, for example with amines such as diisopropylamine, benzylamine, dibenzylamine, triethanolamine, triethylamine, N,N-dibenzylethylenediamine, N-methylmorpholine, pyridine, piperazine, N-ethylpiperidine, N-methyl-D-glucamine and procaine, or with amino acids such as arginine and lysine.

Preferred peptide salt is a salt formed with an organic acid.

The object of the present invention is to provide freeze-dried and liquid formulations for dopamine derivatives such as BIM 23A760, showing an increased stability during manufacture of the formulation and/or during subsequent storage therefore as further object the invention also encompasses a process.

Methods of preparing the compound of the present invention are illustrated in illustrative purposes and should not be constructed as limiting the invention disclosed.

The process according to the invention for the preparation of the liquid or freeze dried formulation may be carried out as follows :

- a) introduction and weighing of the active substance and excipients ;
- b) dissolution of compounds by means of water under stirring ;
- c) use of Nitrogen for solution bubbling ;
- d) filtration ;
- e) filling into a vial and pre-stoppering or stoppering under nitrogen for the solution form ;
- f) freeze-drying step under vacuum ;
- g) releasing vacuum and stoppering under nitrogen.

In accordance with the invention the pharmaceutical formulations are suitably prepared :

- By introducing an appropriate amount of the active drug and excipients.
- Dissolving under stirring, optionally water is used for dissolving the active substance and excipients and may be water for injection, buffered solution or isotonic solution. The solution containing a predetermined concentration of the active substance and excipients may be bubbled under nitrogen. Thereafter the solution may be sterilized by sterile filtration through PVDF (polyvinylidene fluoride) membrane filters and thereafter optionally aseptically filled into sterile vials.
- Optionally stoppering vials under bubbling with nitrogen at room temperature for the liquid form in order to maintain a protected atmosphere and prevent oxidation.
- A further step consisting of pre stoppering vials at room temperature and then pre frozen step may be carried out. The pre frozen temperature could be from about -20° C to -50° C preferably from about -20° C to -30° C at even more preferred at about -25° C to -30° C under inert conditions.
- Subsequently the solution may be frozen, dried optionally under nitrogen atmosphere or other controlled atmosphere known in the art, by primary drying under reduced pressure of about 100 to 400 microbars preferably about 150 to 300 microbars for about 8 to 12 hours at a temperature of about -50° C to + 5° C preferably at a temperature of about -25° C to + 5° C.
- Thereafter a secondary drying step may be carried out under nitrogen with a reduced pressure the reduced pressure may be about 40 to 150 preferably about 60 to 100 microbars for 3 to 5 hours and at a temperature of about -5° C to + 25° C preferably at a temperature of about + 5° C to + 15° C.
- Removing vacuum by a suitable method known by the skilled practitioner using sterile filtered nitrogen and thereafter the vials may be stoppered under nitrogen and brought out the freeze-drier for crimping.

The freeze-dried preparation according to this process can be re-dissolved with great ease.

The present invention further provides an immediate release formulation (IRF) and/or a continuous release of the active substance over a period of one day to one week when the pharmaceutical formulation is placed in an aqueous physiological environment.

In a particular embodiment of the invention, the immediate release formulation is injectable. This pharmaceutical composition will be used by parenteral way such as subcutaneous, intramuscular, intravenous or infusion route.

As further preferred embodiment the pharmaceutical formulation of the present invention is an immediate release formulation (IRF) administered by parenteral route.

Further, according to the present invention the parenteral administration is preferably a subcutaneous administration with daily administration repeated for 7 days and according to patient response repeated treatment several days or weeks, preferably in continuous treatment but not limited to this method of administration.

As further object of the invention the pharmaceutical formulations show a high control of the release, expressed by a zero order curve or pseudo zero order curve (zero order kinetics curve means a constant rate of release of the active substance), and a low C_{max} (C_{max} is a maximum peak observed in plasma concentration of the active substance) therefore provide a control of the initial burst.

The object has been achieved by means of in-vivo and in-vitro tests and stability data provided in the invention.

In addition, various techniques such as radiosterilisation or autoclave sterilisation could be used for the formulations as disclosed in the invention to obtain an aseptic preparation.

The active substances described before, can be used for the treatment and/or prevention of chronic disorders or diseases such as of acromegaly, pituitary adenoma and symptoms associated with neuroendocrine (particularly carcinoid) tumours.

The compound is also indicated for long term treatment of symptomatic, non functional pituitary adenoma.

The following examples serve as illustration of the invention without limiting it.

Example 1

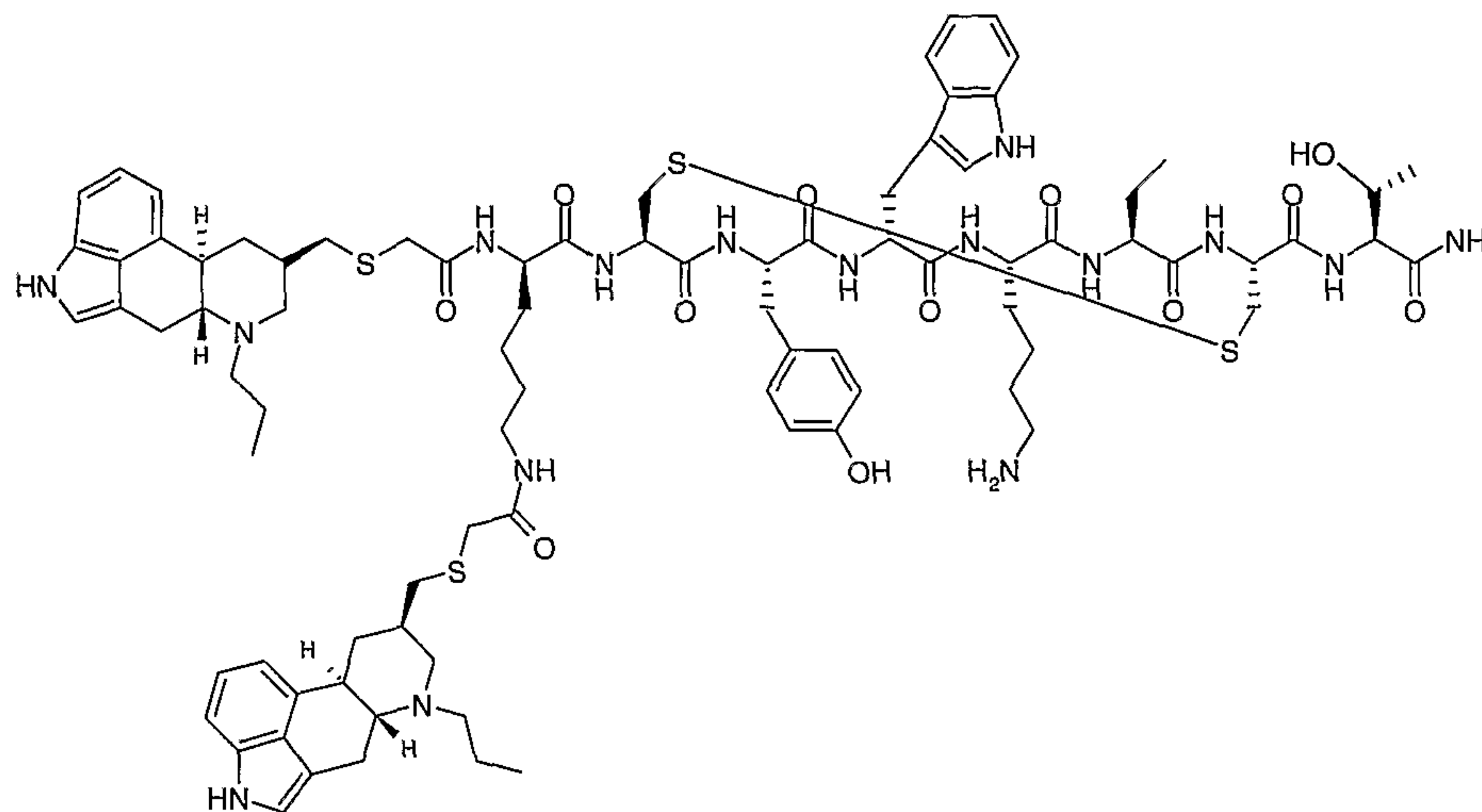
Drawing and Formula of BIM23A760 which corresponds to Dop2-DLys(Dop2)-c(Cys-Tyr-DTrp-Lys-Abu-Cys)-Thr-NH₂

BIM23A760 is a powder product, white to off white color.

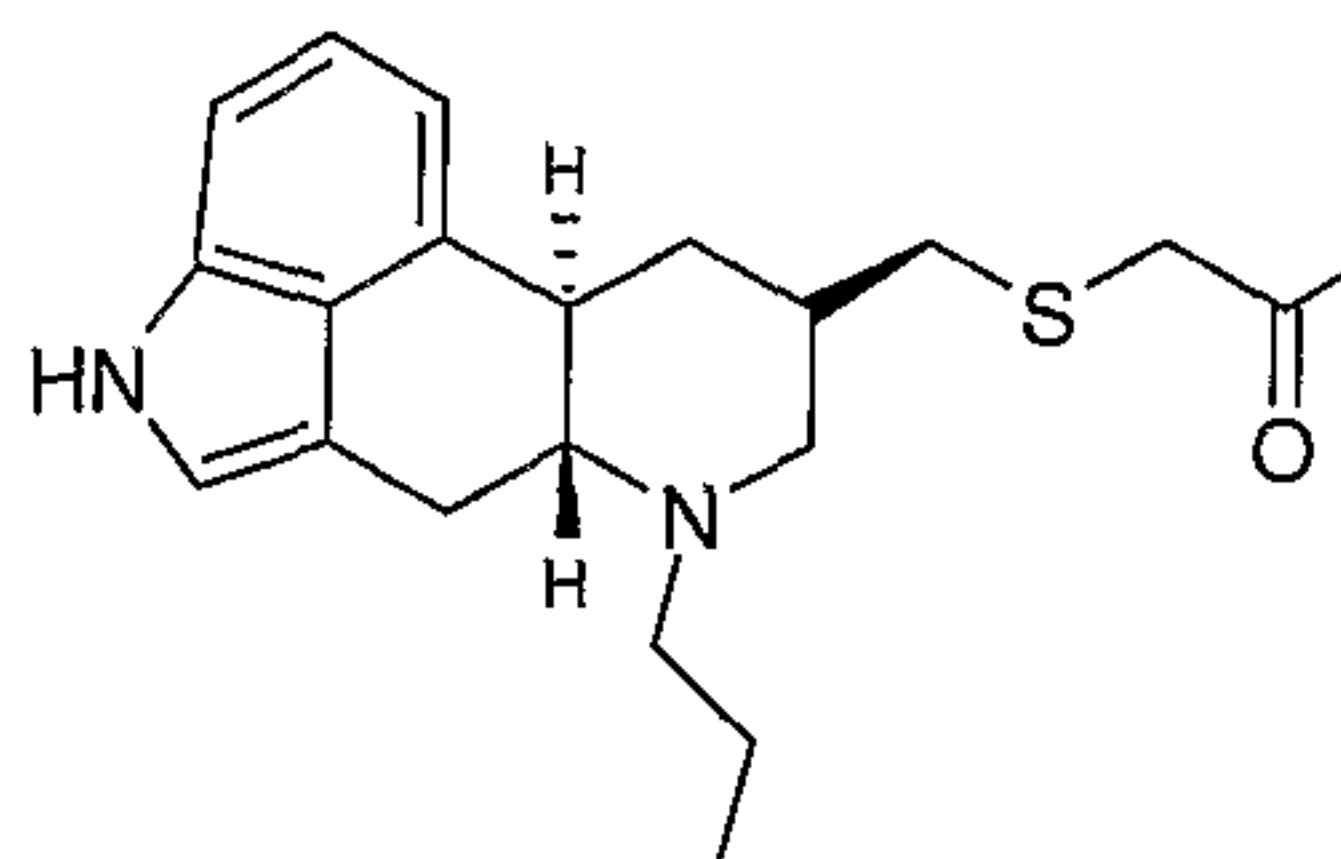
Molecular Weight : ~1690 da

Formula : Dop2-DLys(Dop2)-c(Cys-Tyr-DTrp-Lys-Abu-Cys)-Thr-NH₂

Molecular structure : C₈₆H₁₁₆N₁₆O₁₂S₄



In the formula Dop2-DLys(Dop2)-c(Cys-Tyr-DTrp-Lys-Abu-Cys)-Thr-NH₂, by Dop2 it is meant the following formula :



Example 2

Formulation stabilities- method of measurements and results

Stabilities measurements were performed by means of High Performance Liquid Chromatographic (HPLC) method. The HPLC system a HPLC Waters Alliance 2695 completed by detection system 2487 Dual λ Absorbance Detector, the column used was a C₁₈, 250 x 4.6 mm. All details concerning instrumental conditions are gathered in table 6. Elution conditions are gathered in table 7.

Two pharmaceutical forms were studied in parallel for stability assays over six months at + 5° C and + 25° C / 60 % RH (relative humidity) and + 40° C / 75 % RH. Measurements were carried out in parallel with (in either wfi, or 5 % dextrose as solvent) and a lyophilisate (drug substance on its own) or as second variant of formulations with an additional bulking agents such as mannitol, glycine and trehalose) in WFI.

Composition and timing for stabilities durations are reported in the Tables 1 and

2.

Table 1

Solution form		<i>per ml</i>
Formulation référence	BIM23A760 pure material wfi	0.3 % w/v qs
Formulation 1	BIM23A760 pure material Galactose wfi	0.5 % w/v 5% w/v qs
Formulation 2	BIM23A760 pure material Mannitol wfi	0.5 % w/v 5% w/v qs
Formulation 3	BIM23A760 pure material Fructose wfi	0.5 % w/v 5% w/v qs
Formulation 4	BIM23A760 pure material Maltose wfi	0.5 % w/v 5% w/v qs
Formulation 5	BIM23A760 pure material Trehalose Maltose wfi	0.5 % w/v 4% w/v 4% w/v qs
Formulation 6	BIM23A760 pure material Fructose Galactose wfi	0.5 % w/v 2.5% w/v 2.5% w/v qs
Formulation 7	BIM23A760 pure material Trehalose wfi	0.5 % w/v 8 % w/v qs

Table 2

<i>Lyophilisate form</i>	<i>per vial</i>
Formulation 1	BIM23A760 pure material Galactose 9.1 % w/w 90.9% w/w
Formulation 2	BIM23A760 pure material Mannitol 9.1 % w/w 90.9% w/w
Formulation 3	BIM23A760 pure material Fructose 9.1 % w/w 90.9% w/w
Formulation 4	BIM23A760 pure material Maltose 9.1 % w/w 90.9% w/w
Formulation 5	BIM23A760 pure material Trehalose Maltose 5.9 % w/w 47.1% w/w 47.1% w/w
Formulation 6	BIM23A760 pure material Fructose Galactose 9.1 % w/w 45.5% w/w 45.5% w/w
Formulation 7	BIM23A760 pure material Trehalose 22.72 % w/w 77.28% w/w
Formulation 8	BIM23A760 pure material Trehalose 5.88 % w/w 94.11% w/w

Table 3

Temperature storage	Time points
+ 5° C	T3 month, T6 month
+ 25° C / 60 % RH	Tzero, T3 month, T6 month
+ 40° C / 75 % RH	T1 month, T3 month, T6 month

Stability results are gathered in table 4 and 5.

Table 4

<i>Lyophilisate form</i>	+ 5 °C			+ 25°C / 60 % RH			+ 40°C / 75 % RH		
	C (% initial)	P (% a/a)	pH	C (% initial)	P (% a/a)	pH	C (% initial)	P (% a/a)	pH
BIM23A760									
T0	/	/	/	100 %	95.90 %	6.8	/	/	/
T1 Month	/	/	/	/	/	/	52.0%	95.9 %	6.87
T3 Months	103.6 %	96.10 %	6.64	98.8 %	93.70 %	6.12	29.0%	95.5 %	5.60
T6 Months	73.0 %	96.80 %	5.81	NA	NA	6.63	NA	NA	5.62
BIM23A760 / Mannitol									
T0	/	/	/	100 %	95.80 %	6.40	/	/	/
T1 Month	/	/	/	/	/	/	65.0%	95.9 %	6.87
T3 Months	100.2%	96.60 %	6.28	97.7 %	92.60 %	6.12	42.0%	95.9 %	5.85
T6 Months	98.8 %	96.60 %	6.45	NA	NA	6.39	NA	NA	6.51
BIM23A760 / Glycine									
T0	/	/	/	100 %	96.50 %	6.8	/	/	/
T1 Month	/	/	/	/	/	/	65.0%	96.50 %	5.71
T3 Months	104.8 %	97.90 %	6.48	65.0 %	97.00 %	6.07	33.0%	92.70 %	5.67
T6 Months	74.4 %	96.80 %	5.70	NA	NA	6.14	NA	NA	5.71
BIM23A760 / Trehalose									
T0	/	/	/	100 %	96.00 %	5.90	/	/	/
T1 Month	/	/	/	/	/	/	103.0%	95.70 %	6.78
T3 Months	106.5 %	97.60 %	5.44	106.8%	96.60 %	6.09	98.9%	94.40 %	6.79
T6 Months	106.8 %	96.50 %	5.41	98.9 %	96.00 %	6.06	23.74%	87.30 %	6.80

NA : Not Analysed

Table 5

Solution form	+ 5 °C			+ 25 °C / 60% HR			+ 40 °C / 75% HR		
	C (% initial)	P (% a/a)	pH	C (% initial)	P (% a/a)	pH	C (% initial)	P (% a/a)	pH
BIM23A760 / wfi T0	/	/	/	/	/	/	100%	98.17%	/
T 1 Month	/	/	/	/	/	/	94.23%	95.85%	/
T 3 Months	/	/	/	/	/	/	69.40%	79.84%	/
BIM23A760 / Galactose T0	/	/	/	100%	98.47%	4.92	/	/	/
T 3 Months	/	/	/	98.85%	98.11%	/	92.40%	93.01%	/
T 6 Months	99.98 %	98.57%	/	96.27%	96.98%	/	/	/	/
BIM23A760 / Mannitol T0	/	/	/	100%	98.37%	4.93	/	/	/
T 3 Months	/	/	/	99.67%	98.43%	/	98.47%	96.88%	/
T 6 Months	100.14 %	98.61%	/	98.49%	98.03%	/	/	/	/
BIM23A760 / Fructose T0	/	/	/	100%	98.52%	4.92	/	/	/
T 3 Months	/	/	/	100.37%	98.45%	/	98.80%	96.87%	/
T 6 Months	99.81 %	98.62%	/	98.28%	98.06%	/	/	/	/
BIM23A760 / Maltose T0	/	/	/	100%	98.52%	4.91	/	/	/
T 3 Months	/	/	/	99.47%	98.15%	/	89.23%	93.58%	/
T 6 Months	99.19 %	98.52%	/	94.96%	96.85%	/	/	/	/
BIM23A760 / Trehalose / Maltose T0	/	/	/	100%	98.56%	4.87	/	/	/
T 3 Months	/	/	/	97.71%	98.19%	/	88.63%	94.62%	/
T 6 Months	98.37 %	98.61%	/	94.94%	97.30%	/	/	/	/
BIM23A760 / Fructose / Galactose T0	/	/	/	100%	98.50%	4.92	/	/	/
T 3 Months	/	/	/	99.55%	98.31%	/	91.31%	95.17%	/
T 6 Months	98.24 %	98.60%	/	96.08%	97.62%	/	/	/	/
BIM23A760 / 8%Trehalose T0	/	/	/	100%	98.20%	4.80	/	/	4.80
T 1 Month	/	/	/	/	/	/	98.50 %	97.50%	4.80
T 3 Months	96.02 %	98.08%	4.80	96.00 %	97.87%	4.80	85.77 %	91.29%	4.80
T 6 Months	98.11 %	98.18%	4.90	96.02 %	96.11%	4.80	84.06 %	89.30%	4.80
T 9 months	97.73 %	98.09%	4.80	91.09 %	94.53%	4.80	/	/	/

The dilutions were made by means of a micropipette.

The stability results showed that the trehalose lyophilisate formulation in comparison to the tested liquid formulation was the most stable form at + 5° C and + 25° C over the first 3 months. After 6 months at + 5° C and + 25° C, the trehalose-based lyophilisate remained the most stable in terms of cake appearance, reconstitution, pH, chromatographic profile and assay. The lyophilisate and the liquid formulations are stable for at least 24 months at ± 5° C.

The trehalose-based lyophilisate ensures solubilisation, and stability of the active drug over time. Mannitol and glycine were also experimented on freeze dried and stored at + 25° C or + 40° C over 3 or 6 months.

Saccharides or di-sacharrides have been tested (xylitol, maltose, lactose, galactose, dextran) to check the compound stability in such formulation.

Nevertheless based on the results trehalose ensures and improves BIM23A760 physico-chemical stability. Trehalose allows to protect and stabilize the active drug in the freeze dried formulations.

The molecule BIM23A760 for subcutaneous administration was supplied as a "powder for solution for infusion" in clear vials. The finished drug product was reconstituted either with water for injections or dextrose 5 % for infusion.

The reference solution (BIM23A760 in water for injection) was analyzed at T zero, T 1 month and T 3 months at 40° C / 75 % RH.

Stability results are gathered in Tables 4 and 5 and represented in Figure 1.

C (% initial) represents the content expressed in percentage at the moment T with regard to the content at the moment Tzero .

$$C(\% \text{ initial}) = \frac{C(\text{final})}{C(\text{initial})} \times 100$$

P (% a/a) represents the purity of the solution expressed by areas (a) ratio in percentage :

$$P(\% \text{ a/a}) = \frac{\text{area (main peak)}}{\sum \text{peaks area}} \times 100$$

HPLC Method used for the exploratory stability testing

Table 6 discloses the method and apparatus for measurement of stabilities of the active substance in the formulation disclosed in the present invention.

Table 6

Instrumental conditions	
HPLC System	HPLC Waters Alliance 2695
Detection System	2487 Dual λ Absorbance Detector
Column (Type, Dimension and packing)	C ₁₈ , 250 x 4.6 mm ID, YMC ODS-AM, 5 μ m, 12 nm
Mobile Phase	A : 0.05M NaH ₂ PO ₄ in water, pH adjusted to 3.0 with H ₃ PO ₄ .
	B : 100 % Acetonitrile
Flow Rate	1.0 mL/min
Run Time	50 min
Detection (UV,nm)	UV, 220 nm
Injection Volume (μ l)	10 μ l
Target concentration	0,5 mg / mL
Sample preparation diluent	0,1N Acetic Acid
Column temperature	30° C

Table 7

In table 7 are gathered the elution program, the ratio of each eluant (mobile phase) used for mesurment of formulation stabilities.

Elution Gradient Conditions		
Time (min)	Eluant A (%)	Eluant B (%)
0	75	25
30	60	40
35	40	60
40	40	60
41	75	25
50	75	25

Standard solution with a concentration of 0.5 mg/ml of BIM 23A760 was prepared by dissolving about 10 mg of active substance BIM23A760 in a flask glass. Thereafter 20 ml of acetic acid 0,1 N were added under shaking by means of a vortex and if required additionally dissolved in ultrasonic bath in order to obtain a total dissolution. Weight of brut BIM23A 760 was calculated by means of the following relation :

$$\text{Weight.of.brut BIM23A760} = \frac{10\text{mg of pure BIM23A760}}{\text{Potency of BIM23A760}} * 100$$

The dilutions were made by means of a micropipette.

The results showed that the trehalose lyophilisate formulation in comparison to the tested liquid formulation is the most stable form at + 5° C and + 25° C over the first 3 months. After 6 months at + 5° C and + 25° C, the trehalose-based lyophilisate remains the most stable in terms of cake appearance, reconstitution, pH, chromatographic profile and assay. The lyophilisate and the liquid formulations are stable for at least 24 months at ± 5° C.

The trehalose-based lyophilisate ensures solubilisation, and stability of the active drug over time. Mannitol and Glycine were also experimented on freeze dried and stored at + 25° C or + 40° C over 3 or 6 months.

Based on the results, trehalose ensures and improves BIM23A760 physico-chemical stability. Trehalose allows to protect and stabilize the active drug in the freeze dried formulations.

The molecule BIM23A760 for subcutaneous administration is supplied as a "powder for solution for infusion" in clear vials. The finished drug product is reconstituted either with water for injections or dextrose 5 % for infusion.

Example 3**Example of a freeze-dried formulation**

Material	Doses
BIM23A760 pur	5.0 mg
Trehalose	17.0 mg
Nitrogen	vial closed under nitrogen

The lyophilisate is available in 11 ml type I glass vial and stoppered with a grey chlorobutyl stopper under nitrogen.

The lyophilisate was reconstituted either with an adequate volume of water for injection or dextrose 5 % (= 1 ml) according to the required clinical dosage. The volume of reconstitution is from 1 ml to 10 ml.

Example 4**Process manufacturing of the freeze-dried formulation**

BIM23A760 lyophilisate 5.0 mg was manufactured by dissolving the active drug and the ingredients in water for injections under stirring. This solution was then bubbled under nitrogen sterilised by sterile filtration through 0.22 micrometer PVDF membrane filters and aseptically filled into sterile vials. The theoretical filling was $1.0 \text{ g} \pm 3 \%$ of solution.

Filled vials were pre-stoppered and transferred into the freeze-drier. The time for freezing was about 3 hours 30 min at -25°C .

The primary desiccation was carried out under nitrogen and vacuum at 150 to 300 microbars for 10 hours from -25°C to $+5^{\circ} \text{C}$.

The secondary desiccation was carried out under nitrogen and the vacuum was adjusted from 60 to 100 microbars for 4 hours 30 min at the temperature from $+5^{\circ} \text{C}$ to $+15^{\circ} \text{C}$.

Stoppered vials were automatically crimped. Then vials were externally washed with single use wipes. Bulk product is 100 % visually inspected then was stored at $+5^{\circ} \text{C} \pm 3^{\circ} \text{C}$ in sealed containers.

Process flow chart

Table 8 summarizes the process of lyophilisation and the preparation of formulations.

Table 8

Step	Operations
A	Automatic Vials washing OVEN Depyrogenisation Stoppers Caps, Filters, ... autoclave Active and ingredients weighing wfi weighing dissolution under stirring / Bubbling
	Sterilizing Filtration
	Automatic aseptic filling into 11 ml glass vial. Vial pre stoppering
	Freeze dryer sterilisation
D	Freeze drying Vial stoppering under sterile nitrogen
	Vial crimping
E	External cleaning and inspection
	Labelling and bulk packing
F	Storage at + 5°C ± 3 °C

Example 5**Pharmacokinetics data and methods used for providing the data**

Reconstituted Lyophilisate (+ 1 ml of wfi) has been administered on animals by subcutaneous route, in order to evaluate the pharmacokinetic, on Sprague Dawleys rats versus liquid formulation used for preclinical development (2 % Solutol Formulation in wfi).

Two series of diluted solution were made, one serie with solutol 2 % and the second with threalose. Different dilutions were prepared with 0.1 mg/ml, 1 mg/ml and 10 mg/ml of BIM23A760.

Preliminary pharmacokinetic study after single subcutaneous injection of BIM23A760 at different concentrations to Sprague-Dawley Rats are disclosed in Table 9.

Table 9

Formulation	Group ID	Actual Dose mg/kg	Dose Concentration mg/mL	t_{max} h	C_{max} ng/mL	C_{max}/D -	AUC_t (ng·h/mL)	AUC_{all} (ng·h/mL)	AUC_t/D -	MRT_t h
2 % Solutol in wfi	1	0.183	0.1	8	6.896	37.68	127.5	133.9	696.7	14.06
	2	0.181	1	0.25	13.66	75.47	52.62	61.54	290.7	15.54
	3	0.179	10	0.25	66.16	369.6	124.2	132.4	693.7	12.64
	4	0.896	10	0.25	127.8	142.6	582.2	592.4	649.8	18.12
	5	1.028	25	0.25	215.1	209.2	1072	1085	1043	16.42
Trehalose Formulation	6	0.191	0.1	0.25	125.8	658.6	133.1	139.2	697.0	8.013
	7	0.184	1	0.25	65.55	356.3	95.81	108.7	520.7	12.30
	8	0.187	10	1	46.90	250.8	241.4	248.9	1290.8	13.55
	9	0.933	10	1	70.52	75.58	705.2	719.6	755.8	24.24
	10	1.072	25	1	97.83	91.3	723.9	731.2	675.3	25.62

Claims

1. A stable solid pharmaceutical formulation at temperatures that can range from + 5° C to + 40° C obtainable by lyophilisation comprising a chimeric molecule and at least one sugar or a polyols or a mixture thereof.
2. The formulation according to claim 1 characterized in that the chimeric molecule is BIM23A760 or a dopamine derivatives.
3. The formulation according to claim 1 or 2 characterized in that the sugar of the stable solid pharmaceutical formulation is selected from saccharides or disaccharides or polyols which include xylitol, maltose, lactose, galactose, mannitol, sorbitol, dextran and threalose or a mixture thereof preferably xylitol, mannitol or threalose.
4. A process for the preparation of the pharmaceutical formulation of claim 1 comprising :
 - (a) Introduction and weighing of the active substance and excipients.
 - (b) Dissolution of compounds by means of water under stirring / bubbling under nitrogen.
 - (c) Filtration.
 - (d) Filling into a vial and pre-stoppering.
 - (e) Pre frozen step and freeze-drying step under vacuum.
 - (f) Releasing vacuum and stoppering under controlled atmosphere
5. The process according to claim 4 is characterized in that the pre frozen temperature of step (e) is comprised from about -20° C to -50° C preferably from about -20° C to -30° C more preferably at about -25° C to -30° C under inert conditions.
6. The process according to claim 4 or 5 characterized in that release of vacuum is made under controlled atmosphere consisting in bubbling with nitrogen.
7. A stable liquid pharmaceutical formulation at temperatures that can range from +5° C to +40° C obtainable according to steps (a) to (d) of the process of claim 4

comprising a chimeric molecule and at least one sugar or a polyols or a mixture thereof and optionally containing an isotonic agent and/or a buffer.

8. The stable liquid pharmaceutical formulation according to claim 7 characterized in that the chimeric molecule is BIM23A760 or a dopamine derivatives.
9. The stable liquid pharmaceutical formulation according to claim 7 or 8 is characterized in that the isotonic agent is selected from propylene glycol, glycerol, sodium chloride or potassium chloride.
10. The stable liquid pharmaceutical formulation according to claims 7 to 9 is characterized in that the sugar is selected from saccharides or disaccharides or polyols which include xylitol, maltose, lactose, galactose, mannitol, sorbitol, dextran and threalose or a mixture thereof preferably threalose, xylitol or mannitol.
11. Use of BIM BIM23A760 in freeze dried formulation prepared according to the process of claims 4 to 6 for the treatment and/or prevention of chronic disorders or diseases such as acromegaly, pituitary adenoma and symptoms associated with neuroendocrine (particularly carcinoid) tumours.
12. Use of BIM23A760 in liquid formulation prepared according to the process of claim 4 steps (a) to (d), for the treatment and/or prevention of chronic disorders or diseases such as acromegaly, pituitary adenoma and symptoms associated with neuroendocrine (particularly carcinoid) tumours.
13. The invention as herein before defined.

