(19) Europäisches Patentamt
European Patent Office
Office européen des brevets

(11) EP 1 235 566 B1

(12) **EUROPEAN PATENT SPECIFICATION**

(45) Date of publication and mention of the grant of the patent:
23.03.2005 Bulletin 2005/12

(21) Application number: **00990671.0**

(22) Date of filing: **07.12.2000**

(51) Int Cl.[7]: **A61K 31/22**, A61K 47/12, A61K 47/10, A61P 31/00

(86) International application number:
PCT/EP2000/012346

(87) International publication number:
WO 2001/041758 (14.06.2001 Gazette 2001/24)

(54) **INJECTABLE VALNEMULIN FORMULATION**

INIERZIERBARE VALNEMULIN FORMULIERUNG

FORMULATION INJECTABLE DE VALNEMULIN

(84) Designated Contracting States:
AT BE CH CY DE DK ES FI FR GB GR IE IT LI LU MC NL PT SE TR

(30) Priority: **09.12.1999 EP 99811128**

(43) Date of publication of application:
04.09.2002 Bulletin 2002/36

(73) Proprietors:
- **Novartis AG**
  4056 Basel (CH)
  Designated Contracting States:
  BE CH CY DE DK ES FI FR GB GR IE IT LU MC NL PT SE TR LI
- **Novartis Pharma GmbH**
  1230 Wien (AT)
  Designated Contracting States:
  AT

(72) Inventors:
- **KOLLER, Kurt**
  A-6020 Innsbruck (AT)
- **SCHWARZ, Franz**
  A-6300 Wörgl (AT)

(74) Representative: **Gros, Florent et al**
Novartis AG
4002 Basel (CH)

(56) References cited:
WO-A-98/01127

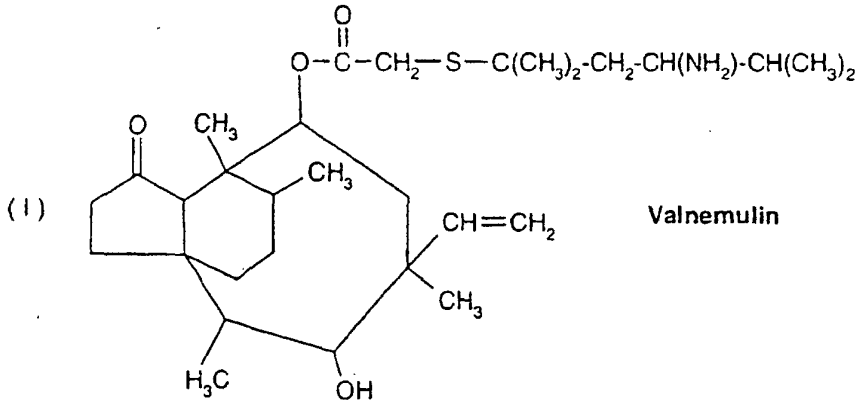
EP 1 235 566 B1

Printed by Jouve, 75001 PARIS (FR)

Description

[0001] The present invention relates to the preparation of an improved galenic delivery form of valnemulin, which is notable for its good tolerance and stability in storage. The new delivery form in question is a non-aqueous or oily injection formulation, which is obtainable by means of *in-situ* preparation and subsequent stabilisation of the free, relatively unstable base of the active ingredient. A further aspect of the present invention concerns the oily injection formulation as such and its usage in the manufacture of a medicament for treating infectious diseases in productive livestock or domestic animals.

[0002] In connection with the present invention, valnemulin is understood to be the compound shown in the following formula I



(I) **Valnemulin**

[0003] Valnemulin is known from EP-0.153.277 and is described specifically therein in example 12. Valnemulin is also known by the commercial name Econor®.

[0004] As is generally known, this compound has antibacterial properties, e.g. following oral or parenteral administration, and is used for the prevention or cure of a series of bacterial infections in the field of animal health. The broad spectrum of activity includes *Streptococcus aronson, Staphylococcus aureus*, *Mycoplasma arthritidis, Mycoplasma bovigenitalium, Mycoplasma bovimastitidis, Mycoplasma bovirhinis*, *Mycoplasma sp.*, *Mycoplasma canis, Mycoplasma felis, Mycoplasma fermentans, Mycoplasma gallinarum, Mycoplasma gallisepticum, A. granularum, Mycoplasma hominis, Mycoplasma hyorhinis*, *Actinobacillus laidlawii, Mycoplasma meleagridis, Mycoplasma neurolyticum, Mycoplasma pneumonia* und *Mycoplasma hyopneumoniae*.

[0005] WO 98/01127 describes its excellent activity against an illness complex that can arise whenever animals are kept in a very restricted space (increased stocking density) e.g. for transport purposes, and are thus exposed to great stress. The most frequent pathogens that play a decisive role in this instance are *Mycoplasma hyopneumoniae, Serpulina* (formerly *Treponema*) *hyodysenteriae, Serpulina pilosicoli, Lawsonia intracellularis, Mycoplasma gallisepticum, Pasteurella multocida, Actinobacillus* (*Haemophilus*) *pleuropneumoniae* and *Haemophilus parasuis*, whereby diseases of the respiratory tract and other infections often occur together and lead to a complex clinical picture. All herd animals are affected, e.g. cattle, sheep and pigs, but also poultry.

[0006] In its free form (valnemulin base), valnemulin is relatively unstable and is therefore primarily used in the form of its salts, particularly as the hydrochloride. A current method of administering antibiotics in the field of animal health is the injection, since it is suitable for administering a controlled single dose and thus a quantity tailored to individual needs. This is often crucial to successful control of many infectious diseases in the field of animal medicines. In contrast, oral administration cannot be controlled nearly so well, and is more customary in human medicine.

[0007] However, it has been shown that aqueous injection solutions and even oily injection suspensions of the salts of valnemulin are poorly tolerated by most domestic animals and in particular by pigs. Damage ranging from mild skin irritation to poorly healing necroses, has been observed. This is also one of the reasons that valnemulin has mainly been used orally until now. In addition, aqueous solutions usually do not show the desired depot action. A further problem is that valnemulin cannot be produced in technical quantities in the free form, as the so-called valnemulin base, but occurs as the salt, and has therefore been used for therapy as the salt.

[0008] However, for commercial usage, it would be extremely desirable to have stable, storable, oily and, in addition, tolerable injection preparations.

[0009] It has now surprisingly been found that chemically stable, non-aqueous injection preparations of valnemulin can be produced *in situ* and can be stabilised in non-aqueous or oily solvents.

[0010] Suitable non-aqueous or oily solvents (i) for the *in situ* production of the valnemulin free base in connection

with the present invention are isopropyl myristate, semi-synthetic and synthetic esters of glycerol, or ethylene or propylene glycol with short-chained to medium-chained mono- or dicarboxylic acids, for example mono-, di- and triglycerides (e.g. neutral oils or miglyol).

[0011] In order to stabilise the free base, other solvents (ii) are added to these solvents (i), e.g. esters of medium-chained to long-chained carboxylic acids (e.g. lactic, lauric, myristic, palmitic, stearic and oleic acid etc.) with monovalent alcohols (e.g. ethanol, n-propanol, 2-propanol, etc.), 1,2-O-isopropylidene glycerol, glycerol, ethanol, N,N-dimethylacetamide, benzyl benzoate or tetraethylene glycol, so that mixtures (iii) of solvents (i) with solvents (ii) are present in the end product. The addition of solvents (ii) serves to improve the galenic properties. Especially preferred as solvent (i) is isopropyl myristate with the addition of solvent (ii) benzyl benzoate or ethanol.

[0012] A preferred embodiment of the present invention is notable for the fact that solvents (i) and (ii) are present in the end product in a ratio of ca. 70 : 15.

[0013] As already mentioned, the free base of valnemulin has until now not been obtainable on a large scale. Therefore, in the following, a new and surprising way is shown of preparing this free base *in situ* and formulating it in a stable form.

[0014] To this end, 0.5 to 30% by weight, preferably 5 to 10% by weight of a salt of valnemulin is suspended in a non-aqueous solvent (i) in a concentration range of 50 to 99% by weight. Appropriate alkaline excipients are added to the suspension whilst stirring and heating gently at a temperature ranging from ca. 50 to 80°C, in order to release the valnemulin base *in situ,* whereby the free base immediately dissolves in the solvent (i).

[0015] Suitable alkaline excipients for the *in situ* release of the free valnemulin base are, for example, alkali and alkaline earth carbonates, hydrogen carbonates and hydroxides, or organic amines, such as triethylamine.

[0016] After completion of this process, the two phases which are immiscible together, the aqueous and the non-aqueous phase, are separated from one another at a temperature of ca. 50 to ca. 80°C This phase separation may be assisted by centrifuging. The separated non-aqueous phase, which now contains the free base of (I), is washed many times with water at room temperature and undergoes fresh phase separation at a temperature of ca. 50 to 80°C. The non-aqueous phase is subsequently dried e.g. using a vacuum, in order to remove residual water and volatile substances such as organic amines, and is mixed with a solvent or solvent mixture from group (ii) to stabilise it. This addition of (ii) improves the galenic properties of the formulations.

[0017] To stabilise against oxidising influences, physiologically acceptable antioxidants may be added to the solution obtained, e.g. esters of ascorbic acid, butyl hydroxy toluene, butyl hydroxy anisole, propyl gallate, tocopherols or tocopherol derivatives, etc., and to stabilise against microbial infestation, physiologically acceptable preservatives may be added, e.g. benzyl alcohol, chlorocresol, chlorobutanol, esters of parahydroxybenzoic acid, phenoxyethanol, phenol and phenol derivatives, sorbic acid, etc.

[0018] The finished solutions are sterile-filtered or sterilised in the final container, e.g. in ampoules.

[0019] The present invention thus comprises essentially the following preferred aspects:

> A method of producing a non-aqueous injection formulation which contains as active ingredient the free valnemulin base, optionally a stabiliser to protect against oxidising influences, and likewise optionally a stabiliser to protect against microbial infestation, and which is characterised in that the free valnemulin base is produced *in situ* from a salt form in a physiologically acceptable non-aqueous solvent or solvent mixture from the above-mentioned category (i) and is stabilised by adding a further solvent from the above-mentioned category (ii).

[0020] A preferred embodiment is characterised in that 0.5 to 30% by weight of a salt of valnemulin in a physiologically acceptable non-aqueous solvent or solvent mixture of the above-mentioned category (i) is released *in situ* whilst heating gently and adding an appropriate alkaline excipient.

[0021] Preferably one or more solvents are used as the non-aqueous solvent or solvent mixture from the above-mentioned category (i), these being selected from the series isopropyl myristate, semi-synthetic and synthetic esters of glycerol, or ethylene or propylene glycol with short-chained to medium-chained mono- or dicarboxylic acids, medium-chained to long-chained carboxylic acids with monovalent alcohols.

[0022] A further notable embodiment is characterised in that a salt of valnemulin is suspended in a solvent in a concentration range of 50 to 99% by weight, and the free base of valnemulin is produced *in situ* using an appropriate alkaline excipient, whilst heating gently and stirring.

[0023] In a preferred variant of the process, to physically stabilise the valnemulin base, an ester of medium-chained to long-chained carboxylic acids with monovalent alcohols, 1,2-O-isopropylidene glycerol, glycerol, ethanol, N,N-dimethylacetamide, benzyl benzoate or tetraethylene glycol is added as a stabilising solvent of the above-mentioned category (ii).

[0024] In the preferred embodiments, solvents (i) and (ii) are present in the end product in a ratio of ca. 70:15.

[0025] An especially preferred variant of the process is characterised in that a salt of valnemulin is suspended in a solvent or solvent mixture (i), an appropriate alkaline excipient is added to the suspension whilst stirring and heating

gently in the range of ca. 50 to ca. 80°C in order to effect *in situ* release of the valnemulin base, the resulting free base being absorbed by this solvent (i); after completion of this process, the two phases which are immiscible together, the aqueous and the non-aqueous phase, are separated from one another at a temperature of ca. 50 to ca. 80°C; the separated non-aqueous phase, which now contains the free base of (I), is washed many times with water at room temperature and undergoes fresh phase separation at a temperature of ca. 50 to 80°C; the non-aqueous phase is subsequently dried and volatile components are removed, and it is mixed with a solvent or solvent mixture from group (ii) to stabilise it

[0026] A further object of the present invention is formed by an oily, stabilised injection formulation, containing as active ingredient the free valnemulin base, which is obtainable by one of the above-characterised releasing and stabilising processes. The present invention also includes the usage of the described oily injection formulation in the manufacture of a medicament for treating infectious diseases of productive livestock or domestic animals.

Formulation examples

Example 1: Injection formulations

[0027] Each 100 ml of the ready injection formulation contains 5 g of valnemulin base. The amounts of grams indicated before the parenthesis (formulation 1: 90.0 g, formulation 2: 85.0 g and formulation 3: 86.0 g) are a result of the different densities of the mixtures of isopropyl myristate, ethanol and benzyl benzoate. The reason for giving two figures lies in the preparation, since the ingredients are weighed in grams, but volumetric (ml) amounts are measured in.

Formulation 1	g
valnemulin	5.0
benzyl benzoate	15.1
isopropyl myristate	69.9
	90.0 (100.0 ml)

Formulation 2	g
valnemulin	5.0
ethanol	13.5
isopropyl myristate	66.5
	85.0 (100.0 ml)

Formulation 3	g
valnemulin	5.0
isopropyl myristate	81.0
	86.0 (100.0 ml)

Chemical stability data

Stability of an oily injection solution of valnemulin in %:

[0028]

valnemulin content after	25°C/60% relative humidity			40°C/75% relative humidity		
	form. 1	form. 2	form. 3	form. 1	form. 2	form. 3
0 months	99.2	99.8	98.5	99.2	99.8	98.5
1 month	99.9	99.2	98.9	99.7	100.4	98.4

(continued)

valnemulin content after	25°C/60% relative humidity			40°C/75% relative humidity		
	form. 1	form. 2	form. 3	form. 1	form. 2	form. 3
2 months	100.2	100.5	100.3	99.4	100.4	99.4
3 months	101.2	102.0	99.6	100.2	101.2	100.8
6 months	101.4	100.4	n.a.	98.9	99.2	n.a.
n.a. indicates not tested. Values greater than 100% result from deviations in the analysis method.						

[0029] All three formulations proved to be chemically stable.

Stability of an oily suspension of valnemulin hydrochloride in %:

[0030]

valnemulin content after	25°C/60% relative humidity	40°C/75% relative humidity
0 weeks	102.0	102.0
8 weeks	104.8	102.8
Values greater than 100% result from deviations in the analysis method.		

Tolerance data

[0031] An investigation of tolerance was made by means of an intramuscular injection of the injection formulation to pigs and evaluating its effect on the adjacent tissue.

	diameter of necrosis in cm			
	formulation 1	placebo	formulation 2	placebo
neck area	1	0	4	n.a.
thigh	1	n.a.	3	5

[0032] The formulations are compared with the placebo. It is shown that the two formulations do not exhibit significantly poorer tolerance than the placebos.

Claims

1. Method of producing a non-aqueous injection formulation containing

   (a) as active ingredient the free valnemulin base;
   (b) optionally a stabiliser to protect against oxidising influences and
   (c) likewise optionally a stabiliser to protect against microbial infestation;

   whereby the free valnemulin base is produced *in situ* from a salt form in a physiologically acceptable non-aqueous solvent or solvent mixture (i) and is stabilised by adding a further solvent (ii).

2. Method according to claim 1, in which 0.5 to 30% by weight of a salt of valnemulin in a physiologically acceptable

non-aqueous solvent or solvent mixture (i) is released *in situ* whilst heating gently and adding an appropriate alkaline excipient.

3. Method according to claim 1 or 2, in which one or more solvents are used as the non-aqueous solvent or solvent mixture (i), these being selected from the series isopropyl myristate, semi-synthetic and synthetic esters of glycerol, or ethylene or propylene glycol with short-chained to medium-chained mono- or dicarboxylic acids, medium-chained to long-chained carboxylic acids with monovalent alcohols.

4. Method according to one of claims 1 to 3, in which a salt of valnemulin is suspended in a solvent in a concentration range of 50 to 99% by weight, and the free base of valnemulin is produced *in situ* using an appropriate alkaline excipient, whilst heating gently and stirring.

5. Method according to one of claims 1 to 5, whereby in order to physically stabilise the valnemulin base, an ester of medium-chained to long-chained carboxylic acids with monovalent alcohols, 1,2-O-isopropylidene glycerol, glycerol, ethanol, N,N-dimethylacetamide, benzyl benzoate or tetraethylene glycol is added as a stabilising solvent (ii).

6. Method according to one of claims 1 to 5, whereby the solvents (i) and (ii) are present in the end product in a ratio of ca. 70:15.

7. Method according to one of claims 1 to 5, whereby a salt of valnemulin is suspended in a solvent or solvent mixture (i), an appropriate alkaline excipient is added to the suspension whilst stirring and heating gently in a temperature range of ca. 50 to ca. 80°C in order to effect *in situ* release of the valnemulin base, the resulting free base is absorbed by this solvent (i); after completion of this process, the two phases which are immiscible together, the aqueous and the non-aqueous phase, are separated from one another at a temperature of ca. 50 to ca. 80°C; the separated non-aqueous phase, which now contains the free base of (I), is washed many times with water at room temperature and undergoes fresh phase separation at a temperature of ca. 50 to 80°C; the non-aqueous phase is subsequently dried and volatile components are removed, and it is mixed with a solvent or solvent mixture from group (ii) to stabilise it

8. Oily, stabilised injection formulation containing as active ingredient the free valnemulin base; obtainable by a method according to one of claims 1 to 7.

9. The use of the in situ-formed free valnemulin base according to any of claims 1-8 for the manufacture of a non-aqueous injection formulation for the treatment of infectious diseases of productive livestock or domestic animals.

Patentansprüche

1. Verfahren zur Herstellung einer nicht wässrigen Injektionsformulierung, die enthält

   a) als Wirkstoff die freie Base von Valnemulin,
   b) optional einen Stabilisator zum Schutz gegen oxidierende Einflüsse und
   c) ebenfalls optional einen Stabilisator zum Schutz gegen einen Überfall durch Mikroben,

   wobei die freie Base von Valnemulin in situ gebildet wird aus einer Salzform in einem physiologisch annehmbaren nicht wässrigen Lösemittel oder Lösemittelgemisch (i) und durch Zusatz eines weiteren Lösemittels (ii) stabilisiert wird.

2. Verfahren nach Anspruch 1, wobei 0,5 bis 30 Gew.-% eines Salzes von Valnemulin in einem physiologisch annehmbaren nicht wässrigen Lösemittel oder Lösemittelgemisch (i) in situ freigesetzt werden während eines schwachen Erhitzens und eines Zusatzes eines geeigneten alkalischen Hilfsstoffs.

3. Verfahren nach Anspruch 1 oder 2, wobei als nicht wässriges Lösemittel oder Lösemittelgemisch (i) ein oder mehr Lösemittel verwendet werden, die ausgewählt werden aus Isopropylmyristat, halbsynthetischen und synthetischen Estern von Glycerin oder Ethylenglykol oder Propylenglykol mit kurzkettigen bis mittelkettigen Mono- oder Dicarbonsäuren, mittelkettigen bis langkettigen Carbonsäuren mit einwertigen Alkoholen.

4. Verfahren nach einem der Ansprüche 1 bis 3, wobei ein Salz von Valnemulin in einem Lösemittel in einem Kon-

zentrationsbereich von 50 bis 99 Gew.-% suspendiert wird und die freie Base von Valnemulin in situ gebildet wird unter Verwendung eines geeigneten alkalischen Hilfsstoffs unter schwachem Erhitzen und Rühren.

5. Verfahren nach einem der Ansprüche 1 bis 5, wobei zur physikalischen Stabilisierung der Base von Valnemulin ein Ester von mittelkettigen bis langkettigen Carbonsäuren mit einwertigen Alkoholen, 1,2-O-Isopropylidenglycerin, Glycerin, Ethanol, N,N-Dimethylacetamid, Benzylbenzoat oder Tetraethylenglykol als stabilisierendes Lösemittel (ii) zugesetzt wird.

6. Verfahren nach einem der Ansprüche 1 bis 4, worin die Lösemittel (i) und (ii) im Endprodukt in einem Verhältnis von etwa 70:15 vorhanden sind.

7. Verfahren nach einem der Ansprüche 1 bis 5, wobei ein Salz von Valnemulin in einem Lösemittel oder Lösemittelgemisch (i) suspendiert wird, die Suspension mit einem geeigneten alkalischen Hilfsstoff unter Rühren und schwachem Erhitzen auf eine Temperatur im Bereich von etwa 50 bis etwa 80 °C versetzt wird, um in situ eine Freisetzung von Valnemulin zu bewirken, die erhaltene freie Base durch dieses Lösemittel (i) absorbiert wird, nach Beendigung dieses Prozesses die beiden Phasen, die zusammen nicht vermischbar sind, nämlich die wässrige und die nicht wässrige Phase, bei einer Temperatur von etwa 50 bis etwa 80 °C voneinander getrennt werden, die abgetrennte nicht wässrige Phase, die nun die freie Base von (i) enthält, mehrmals mit Wasser bei Raumtemperatur gewaschen und einer erneuten Auftrennung der Phasen bei einer Temperatur von etwa 50 bis 80 °C unterzogen wird, die nicht wässrige Phase dann getrocknet wird und die flüchtigen Komponenten entfernt werden und dann zur Stabilisierung eine Vermischung mit einem Lösemittel oder einem Lösemittelgemisch aus der Gruppe (ii) vorgenommen wird.

8. Ölige stabilisierte Injektionsformulierung, die als Wirkstoff die freie Base von Valnemulin enthält, erhältlich durch ein Verfahren nach einem der Ansprüche 1 bis 7.

9. Verwendung einer in situ gebildeten freien Base von Valnemulin nach einem der Ansprüche 1 bis 8 zur Herstellung einer nicht wässrigen Injektionsformulierung für die Behandlung von Infektionskrankheiten bei Nutzvieh oder Haustieren.

Revendications

1. Procédé de préparation d'une formulation injectable non aqueuse contenant :

   (a) comme ingrédient actif, la valnémuline base libre ;
   (b) éventuellement un stabilisant pour protéger des influences oxydantes, et
   (c) éventuellement également, un stabilisant pour protéger d'une infestation microbienne ;

   la valnémuline base libre étant produite *in situ* à partir d'une forme sel dans un solvant non aqueux ou un mélange de solvants physiologiquement acceptable (i), et stabilisée par addition d'un autre solvant (ii).

2. Procédé selon la revendication 1, dans lequel 0,5 à 30 % en poids d'un sel de valnémuline dans un solvant non aqueux ou un mélange de solvants physiologiquement acceptable (i) sont libérés *in situ*, tout en chauffant doucement et en ajoutant un excipient alcalin approprié.

3. Procédé selon la revendication 1 ou 2, dans lequel on utilise un ou plusieurs solvants comme solvant non aqueux ou mélange de solvants (i), ceux-ci étant choisis dans la série du myristate d'isopropyle, des esters de glycérol semi-synthétiques et synthétiques, ou de l'éthylène- ou du propylèneglycol avec des acides mono- ou dicarboxyliques à chaîne courte à moyenne, des acides carboxyliques à chaîne moyenne à longue avec des alcools monovalents.

4. Procédé selon l'une des revendications 1 à 3, dans lequel un sel de valnémuline est mis en suspension dans un solvant dans un intervalle de concentrations de 50 à 99 % en poids, et la base libre de valnémuline est produite *in situ* en utilisant un excipient alcalin approprié, tout en chauffant doucement et en agitant.

5. Procédé selon l'une des revendications 1 à 5 dans lequel, afin de stabiliser physiquement la valnémuline base, on ajoute un ester d'acides carboxyliques à chaîne moyenne à longue avec des alcools monovalents, du 1,2-O-

isopropylidèneglycérol, du glycérol, de l'éthanol, du N,N-diméthylacétamide, du benzoate de benzyle ou du tétraéthylèneglycol comme solvant stabilisant (ii).

6. Procédé selon l'une des revendications 1 à 5, dans lequel les solvants (i) et (ii) sont présents dans le produit final selon un rapport d'environ 70:15.

7. Procédé selon l'une des revendications 1 à 5, dans lequel un sel de valnémuline est mis en suspension dans un solvant ou un mélange de solvants (i), un excipient alcalin approprié est ajouté à la suspension, tout en agitant et en chauffant doucement, dans un intervalle de températures d'environ 50 à 80 °C, afin d'effectuer la libération *in situ* de la valnémuline base, la base libre résultante est absorbée par ce solvant (i) ; après l'achèvement de ce processus, les deux phases qui ne sont pas miscibles l'une à l'autre, la phase aqueuse et la phase non aqueuse, sont séparées l'une de l'autre à une température d'environ 50 à environ 80 °C ; la phase non aqueuse séparée, qui contient maintenant la base libre de (i) est lavée plusieurs fois avec de l'eau à température ambiante et subit une nouvelle séparation de phase à une température d'environ 50 à 80 °C ; la phase non aqueuse est ultérieurement déshydratée et les composants volatils sont éliminés, et celle-ci est mélangée avec un solvant ou un mélange de solvants du groupe (ii) pour la stabiliser.

8. Formulation injectable huileuse, stabilisée, contenant comme ingrédient actif la valnémuline base libre pouvant être obtenue par un procédé selon l'une des revendications 1 à 7.

9. Utilisation de la valnémuline base libre formée *in situ* selon l'une quelconque des revendications 1 à 8 dans la fabrication d'une formulation injectable non aqueuse pour le traitement de maladies infectieuses du bétail productif ou des animaux domestiques.