ORIGINAL

IN THE UNITED STATES DISTRICT COURT
FOR THE NORTHERN DISTRICT OF ILLINOIS
EASTERN DIVISION

GENPHARM INC., A Canadian corporation,	)	
	)	No. 04 C 6732
Plaintiff,	)	
	)	Judge Coar
v.	)	
	)	Magistrate Judge Mason
ABBOTT LABORATORIES, An Illinois corporation,	)	
	)	
Defendant.	)	

NOTICE OF FILING

To: Edward L. Foote
R. Mark McCareins
Winston & Strawn LLP
35 West Wacker Drive
Chicago, Illinois 60601

PLEASE TAKE NOTICE that on March 23, 2005, the undersigned caused a copy of Plaintiff's "Amended Complaint for Declaratory Judgment" to be filed with the Clerk of the Court, leave to make such filing having previously been granted. A true and correct copy of such pleading has been previously served upon you.

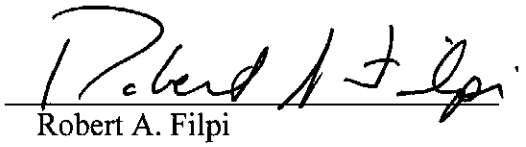
One of the Attorneys for Plaintiff

Paul F. Stack
Robert A. Filpi
Stack & Filpi Chartered
140 S. Dearborn St. Suite 411
Chicago, Illinois 60603
(312) 782-0690

CERTIFICATE OF SERVICE

The undersigned hereby certifies that he caused the foregoing notice to be served upon the attorneys for the Defendant by depositing a copy thereof in the United States Mail with first class postage prepaid on March 23, 2005, and that a copy of the First Amended Complaint for Declaratory Judgment was hand delivered on the same date to the following persons:

Edward L. Foote
R. Mark McCareins
Winston & Strawn LLP
35 West Wacker Drive
Chicago, Illinois 60601



Robert A. Filpi

In the United States District Court
for the Northern District of Illinois
Eastern Division

GENPHARM INC.,
a Canadian corporation,

Plaintiff,

vs.

ABBOTT LABORATORIES,
an Illinois corporation,

Defendant.

No. 04 C 6732

Judge Coar

Magistrate Judge Mason

Jury Trial Demanded

AMENDED COMPLAINT FOR DECLARATORY JUDGMENT

Plaintiff Genpharm Inc. ("Genpharm"), for its Complaint against Abbott Laboratories ("Abbott"), alleges:

THE PARTIES

1. Genpharm is a Canadian corporation with its principal place of business at 37 Advance Road, Etibicoke, Ontario, Canada M8Z 2S6. Among other things, Genpharm develops and manufactures generic pharmaceutical products that are distributed and sold in the United States.

2. On information and belief, Abbott is an Illinois corporation with its principal office at 100 Abbott Park Road, Abbott Park, Illinois 60064-3500.

3. On information and belief, Abbott owns U.S. Patent No. 5,844,105 ("the '105 patent"), entitled "Preparation of a Crystal Form II of Clarithromycin" (copy attached as Exhibit A).

4. On information and belief, Abbott owns U.S. Patent No. 5,858,986 ("the '986 patent"), entitled "Crystal Form I of Clarithromycin" (copy attached as Exhibit B).

5. On information and belief, Abbott owns U.S. Patent No. 5,945,405 ("the '405 patent"), entitled "Crystal Form 0 of Clarithromycin" (copy attached as Exhibit C).

6. On information and belief, Abbott holds New Drug Application ("NDA") No. 50-662 for 250 mg and 500 mg clarithromycin oral tablets, which are marketed as BIAXIN® in the United States. BIAXIN® is an oral antibiotic.

JURISDICTION AND VENUE

7. This Court has original jurisdiction over the subject matter of this action pursuant to 28 U.S.C. §§ 1331 and 1338(a), as it involves substantial claims arising under the United States Patent Act, 35 U.S.C. § 1 *et seq.*

8. This Court may declare the rights and other legal relations of the parties under 28 U.S.C. §§ 2201 and 2202, because this is a case of actual controversy within the Court's jurisdiction seeking a declaratory judgment that the '105, '986, and '405 patents would not be infringed by Genpharm's proposed clarithromycin product.

9. Personal jurisdiction exists in this Court over Abbott by virtue of Abbott being incorporated in Illinois and having its principal place of business within this District, and because Abbott does business within this District.

10. Venue is proper in this District under 28 U.S.C. § 1391(b).

11. Venue is also proper as to Abbott in that Abbott is subject to personal jurisdiction in this District, is doing and transacting business and has substantial contacts in this District, and Abbott's registered agent resides in this District.

GENERAL ALLEGATIONS

12. On information and belief, Abbott has been marketing, directly or indirectly, BIAXIN® in the United States since first obtaining Food and Drug Administration ("FDA") approval in 1991.

13. Abbott is the assignee listed on the face of the '105 patent, which will expire on July 29, 2016.

14. Abbott is the assignee listed on the face of the '986 patent, which will expire on July 29, 2016.

15. Abbott is the assignee listed on the face of the '405 patent, which will expire on January 17, 2017.

EXISTENCE OF ACTUAL CASE AND CONTROVERSY

16. Genpharm filed Abbreviated New Drug Application ("ANDA") No. 65-195, directed to a generic version of BIAXIN®, with the FDA on November 3, 2003. The FDA accepted this ANDA for review on January 22, 2004. By preparing and filing this ANDA, Genpharm has made substantial preparations to import, offer to sell, and sell generic versions of BIAXIN® in the United States.

17. Genpharm expects FDA approval of its ANDA for a generic version of BIAXIN® before Abbott's '105 and '986 patents expire on July 29, 2016, and before Abbott's '405 patent expires on January 17, 2017. Genpharm does not expect to make any material changes to the products described in its ANDA before FDA approval, so that the products that Genpharm will market in the United States will be the same products that are described in that ANDA.

18. Abbott's 2001 annual report announces that "Abbott believes that no single patent, license, trademark (or related group of patents, licenses, or trademarks), except for those related to clarithromycin (which is sold under the trademarks Biaxin®, Klacid® and Klaricid®) and those related to divalproex sodium (which is sold under the trademark Depakote®), are material in relation to Abbott's business as a whole." Abbott's '105, '986, and '405 patents each contain claims directed to clarithromycin, a component of BIAXIN® tablets, and thus, on information and belief, Abbott considers the '105, '986, and '405 patents to be material company assets.

19. On information and belief, Abbott would assert the '105, '986, and '405 patents against Genpharm for alleged infringement of those patents if Genpharm commercially marketed generic versions of BIAXIN®.

20. Abbott has enforced its divalproex sodium (i.e., Abbott's other avowed material asset) patents against Alra Laboratories, Inc. (in *Abbott Labs. v. Alra Labs., Inc.*, No. 92 C 5806 (N.D. Ill.)) and TorPharm, Inc., Apotex Corporation, and Apotex, Inc. (in *Abbott Labs. v. TorPharm, Inc.*, No. 97 C 7515 (N.D. Ill.)) for their respective efforts to market generic divalproex sodium in the United States.

21. Abbott has demonstrated its intention to enforce its patents relating to BIAXIN® and clarithromycin in the United States. Specifically, in *Teva Pharms. USA, Inc. v. Abbott Labs.*, Civ. Act. No. 03 C 5455 (N.D. Ill.), Abbott claims that Teva Pharmaceuticals USA, Inc. ("Teva") would infringe (by commercial manufacture, use, offer for sale, sale, or importation of generic versions of BIAXIN®), infringes (by filing an ANDA for

clarithromycin 250 mg and 500 mg tablets), and willfully infringes the '105, '986, and '405 patents.

22. Abbott has demonstrated its intention to enforce patents relating to BIAXIN® and clarithromycin against Genpharm. Specifically, Abbott instituted a patent infringement lawsuit against Genpharm in Canada (Federal Court, Trial Division, Court File No. T-2274-03) requesting an Order prohibiting Genpharm from marketing a generic version of BIAXIN® until Abbott's Canadian Patent No. 2,261,732, the Canadian equivalent to the '105 patent, expires on July 28, 2017.

23. Abbott has demonstrated its intention to enforce patents relating to BIAXIN® and clarithromycin in Canada. Specifically, Abbott sued the following companies in Canada for alleged infringement of Abbott's Canadian Patent No. 2,261,732: (a) Ratiopharm (Federal Court, Trial Division, Court File No. T-1847-02 and No. T-1656-03); (b) Novopharm Ltd. (Federal Court, Trial Division, Court File No. T-1236-02); (c) Apotex Inc. (Federal Court, Trial Division, Court File No. T-1133-02 and No. T-1847-03); and (d) Pharmascience Inc. (Federal Court, Trial Division, Court File No. T-1035-02 and No. T-2295-03).

24. Based on (a) Abbott's suits against Genpharm and other generic pharmaceutical manufacturers to enforce the Canadian equivalent of the '105 patent, (b) Abbott's claims in the United States against Teva for infringement and willful infringement of the '105, '986, and '405 patents, and (c) Abbott's prior lawsuits to protect divalproex sodium from generic competition, Genpharm is under a reasonable apprehension that Abbott will sue Genpharm, alleging infringement of the '105, '986, and '405 patents.

25. To avoid legal uncertainty and to protect its substantial investment and anticipated future investments in its generic BIAXIN® products, Genpharm has instituted this declaratory judgment action.

COUNT I
DECLARATORY JUDGMENT OF NONINFRINGEMENT

26. Genpharm repeats and realleges the allegations of Paragraphs 1 though 25 above as if fully set forth herein.

27. Genpharm's activities related to seeking FDA approval to market a generic version of BIAXIN® do not, and would not, infringe any properly construed, valid claim of the '105 patent.

28. The manufacture, importation, use, offer for sale, or sale of Genpharm's proposed generic version of BIAXIN® does not, and would not, infringe any properly construed, valid claim of the '105 patent.

COUNT II
DECLARATORY JUDGMENT OF NONINFRINGEMENT

29. Genpharm repeats and realleges the allegations of Paragraphs 1 though 25 above as if fully set forth herein.

30. Genpharm's activities related to seeking FDA approval to market a generic version of BIAXIN® do not, and would not, infringe any properly construed, valid claim of the '986 patent.

31. The manufacture, importation, use, offer for sale, or sale of Genpharm's proposed generic version of BIAXIN® does not, and would not, infringe any properly construed, valid claim of the '986 patent.

COUNT III
DECLARATORY JUDGMENT OF NONINFRINGEMENT

32. Genpharm repeats and realleges the allegations of Paragraphs 1 though 25 above as if fully set forth herein.

33. Genpharm's activities related to seeking FDA approval to market a generic version of BIAXIN® do not, and would not, infringe any properly construed, valid claim of the '405 patent.

34. The manufacture, importation, use, offer for sale, or sale of Genpharm's proposed generic version of BIAXIN® does not, and would not, infringe any properly construed, valid claim of the '405 patent.

COUNT IV
DECLARATORY JUDGMENT OF PATENT INVALIDITY

35. Genpharm repeats and realleges the allegations of Paragraphs 1 though 25 above as if fully set forth herein.

36. The claims of the '105 patent are invalid under 35 U.S.C. § 101 *et seq.*

COUNT V
DECLARATORY JUDGMENT OF PATENT INVALIDITY

37. Genpharm repeats and realleges the allegations of Paragraphs 1 though 25 above as if fully set forth herein.

38. The claims of the '986 patent are invalid under 35 U.S.C. § 101 *et seq.*

COUNT IV
DECLARATORY JUDGMENT OF PATENT INVALIDITY

39. Genpharm repeats and realleges the allegations of Paragraphs 1 though 25 above as if fully set forth herein.

40. The claims of the '405 patent are invalid under 35 U.S.C. § 101 *et seq.*

PRAYER FOR RELIEF

WHEREFORE, Genpharm respectfully requests the Court to enter judgment against Abbott to include:

A. A declaration that United States Patent No. 5,844,105 will not be infringed by Genpharm's activities related to its seeking FDA approval to make a generic version of BIAXIN®;

B. A declaration that United States Patent No. 5,844,105 will not be infringed by the manufacture, importation, use, offer for sale, or sale of Genpharm's generic version of BIAXIN®;

C. A declaration that United States Patent No. 5,858,986 will not be infringed by Genpharm's activities related to its seeking FDA approval to make a generic version of BIAXIN®;

D. A declaration that United States Patent No. 5,858,986 will not be infringed by the manufacture, importation, use, offer for sale, or sale of Genpharm's generic version of BIAXIN®;

E. A declaration that United States Patent No. 5, 945,405 will not be infringed by Genpharm's activities related to its seeking FDA approval to make a generic version of BIAXIN®;

F. A declaration that United States Patent No. 5,945,405 will not be infringed by the manufacture, importation, use, offer for sale, or sale of Genpharm's generic version of BIAXIN®;

G. A declaration that United States Patent No. 5,844,105 is invalid.

H. A declaration that United States Patent No. 5,858,986 is invalid.

I. A declaration that United States Patent No. 5,945,405 is invalid.

J. An award of Genpharm's reasonable costs and attorneys' fees incurred in connection with this action; and

K. All such other and further relief as this Court may deem just and proper.

DEMAND FOR JURY TRIAL

Genpharm demands a jury trial of all issues in this action so triable pursuant to Rule 38 of the Federal Rules of Civil Procedure.

Respectfully submitted,

STACK & FILPI CHARTERED

Dated: February 14, 2005

By: _____

Paul F. Stack
Roger H. Dusberger
Cori A. Szczucki
Suite 411
140 South Dearborn Street
Chicago, Illinois 60603-5298
(312) 782-0690 (tel)
(312) 782-0936 (fax)

Attorneys for Plaintiff
GENPHARM INC.

Of Counsel:

Edgar H. Haug
Jeffrey A. Hovden
Richard E. Parke
Frommer Lawrence & Haug LLP
745 Fifth Avenue
New York, New York 10151
(212) 588-0800 (tel)
(212) 588-0500 (fax)

Exhibit A

US005844105A

United States Patent [19]

Liu et al.

[11] Patent Number: 5,844,105

[45] Date of Patent: Dec. 1, 1998

[54] PREPARATION OF CRYSTAL FORM II OF CLARITHROMYCIN

[75] Inventors: Jih-Hua Liu, Green Oaks, Ill.; David A. Riley, Kenosha, Wis.

[73] Assignee: Abbott Laboratories, Abbott Park, Ill.

[21] Appl. No.: 681,695

[22] Filed: Jul. 29, 1996

[51] Int. Cl.[6] C07G 3/00; C07H 15/00; C07H 17/08, C07H 1/00

[52] U.S. Cl. 536/18.5; 536/7.2; 536/7.5; 536/124

[58] Field of Search 536/7.2, 124, 7.5, 536/18.5

[56] References Cited

U.S. PATENT DOCUMENTS

4,331,660 5/1982 Watanabe et al. 536/7.2
4,990,602 2/1991 Morimoto et al. 536/7.4

FOREIGN PATENT DOCUMENTS

WO 9[illegible] 19096 5 1993[illegible] WIPO .

OTHER PUBLICATIONS

Quantitative Structure–Activity Relationships In Drug Design, vol. 291 (1989), pp. 325–328, Kim et al., "Conformational Study of Erythromycin Analogues".

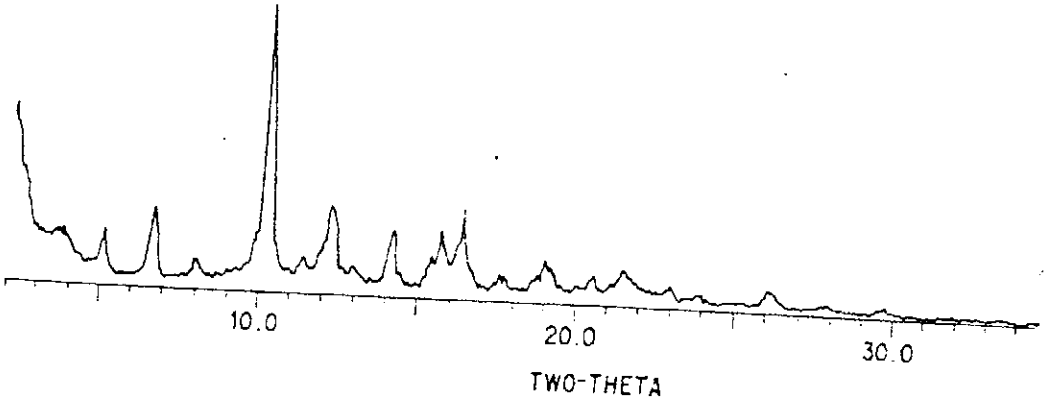
Acta Crystallographica, vol. c49, No. 5 (May 1993), pp. 1227–1230, Iwasaki et al., "Structure of 6-O-Methylerythromycin A (Clarithromycin)".

Primary Examiner—John Kight
Assistant Examiner—Everett White
Attorney, Agent, or Firm—Mona Anand

[57] ABSTRACT

The present invention provides a process for the preparation of 6-O-methylerythromycin A Form II comprising converting erythromycin A to 6-O-methylerythromycin A and treating the 6-O-methylerythromycin A with a number of common organic solvents or mixtures of common organic solvents.

16 Claims, 6 Drawing Sheets



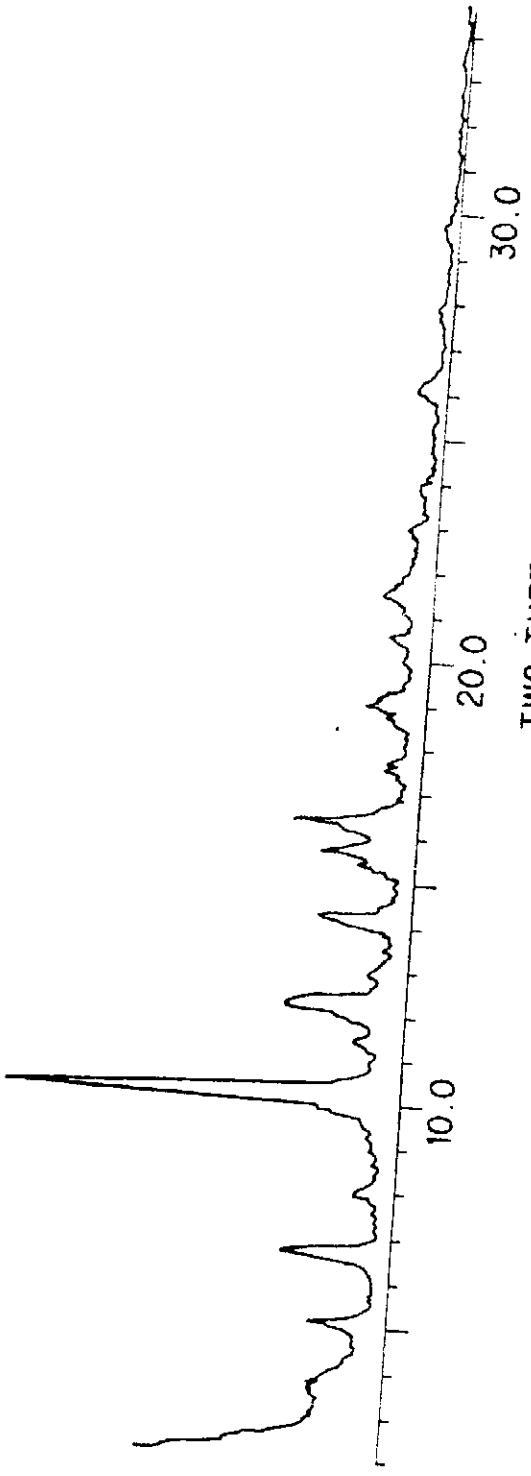


FIG.1a

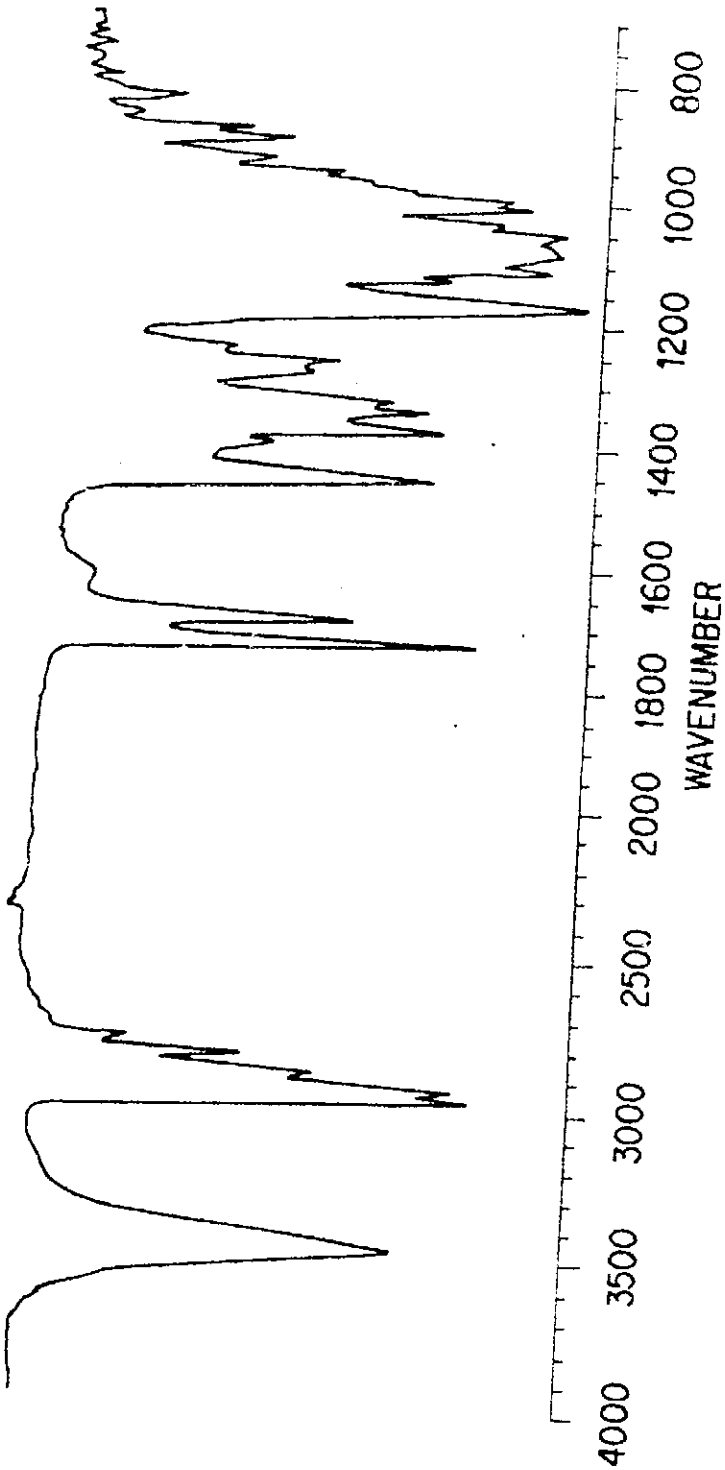

800
1000
1200
1400
1600
1800
2000
2500
3000
3500
4000
WAVENUMBER

FIG.1b

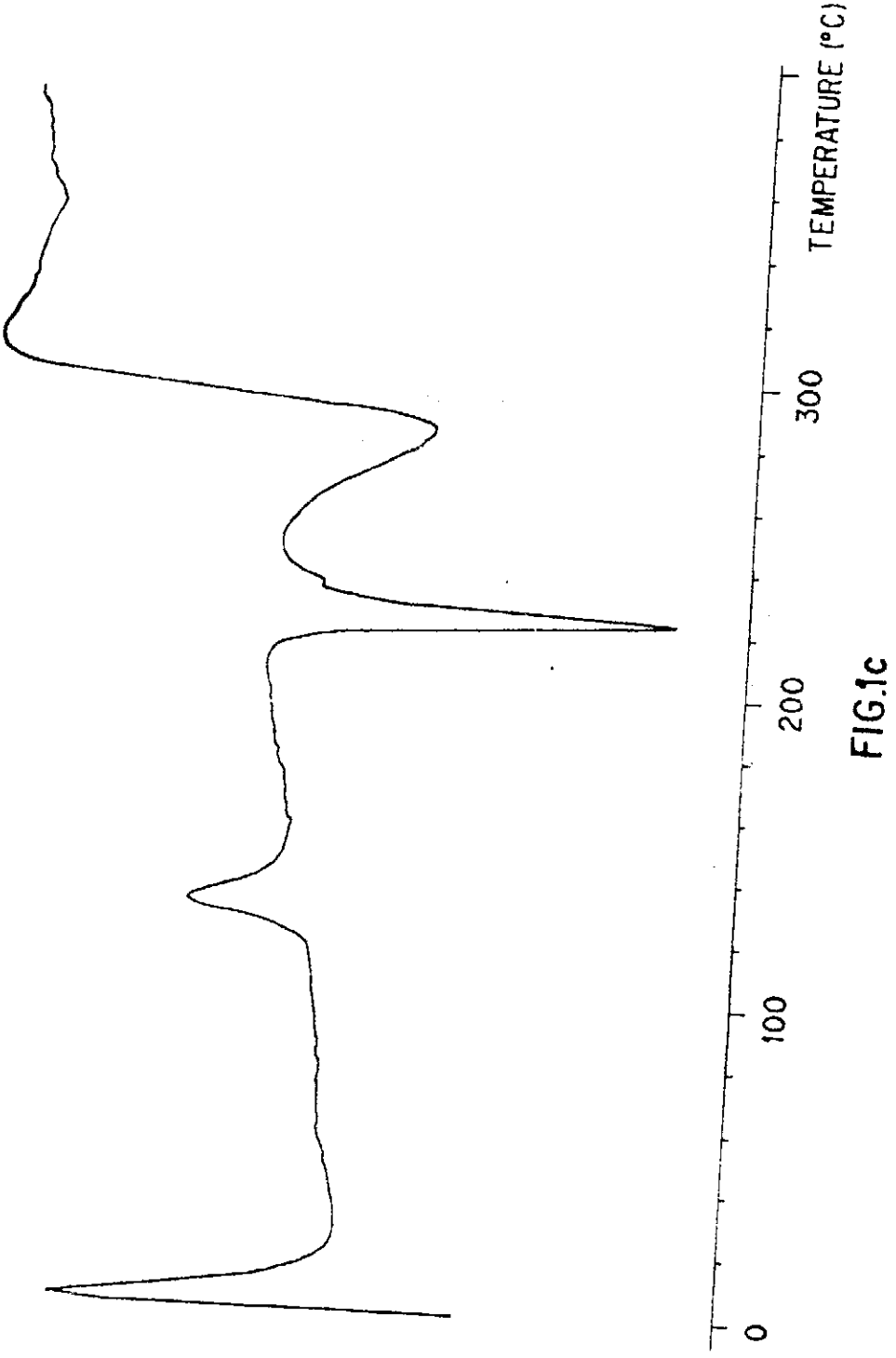


FIG.1c

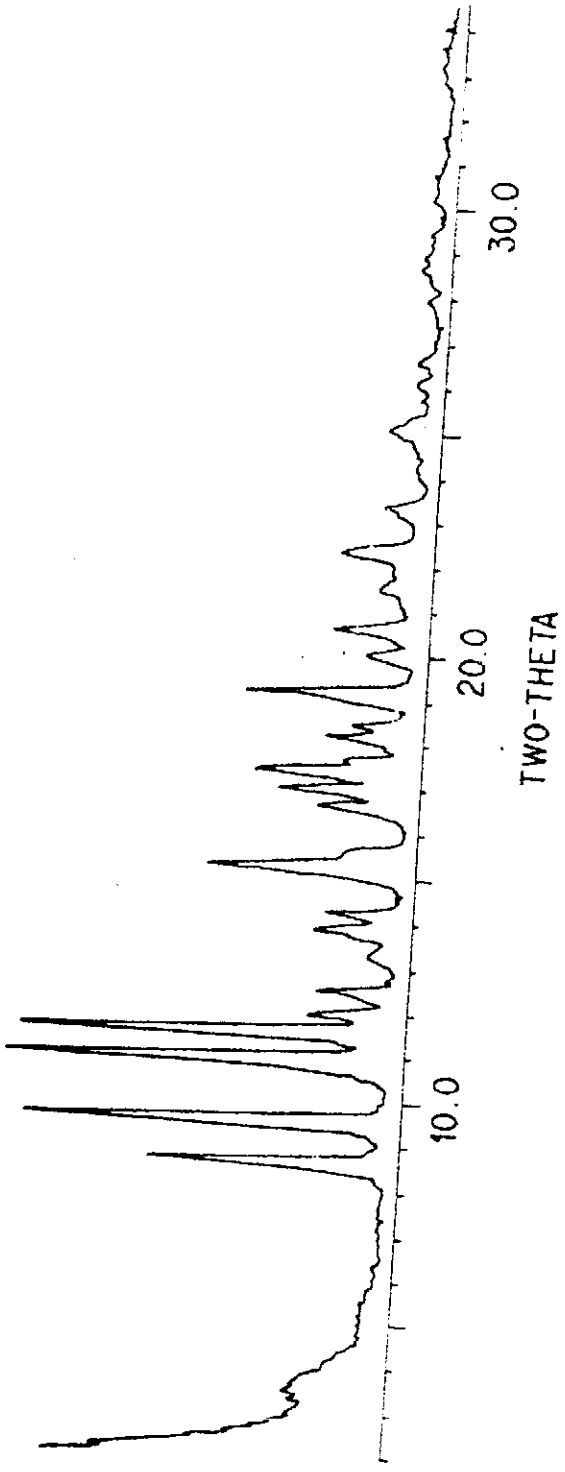


FIG.2a

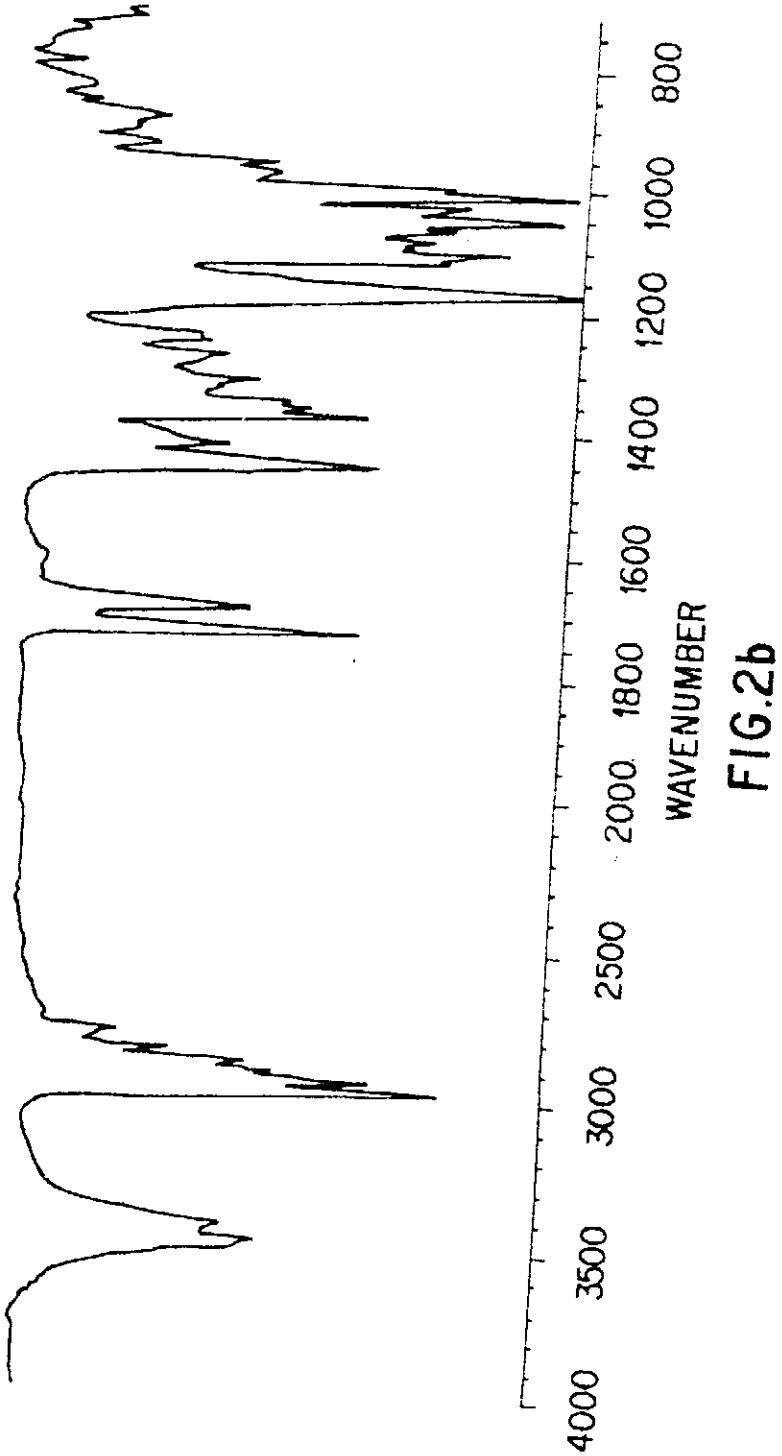

800
1000
1200
1400
1600
1800
2000
2500
3000
3500
4000
WAVENUMBER

FIG.2b

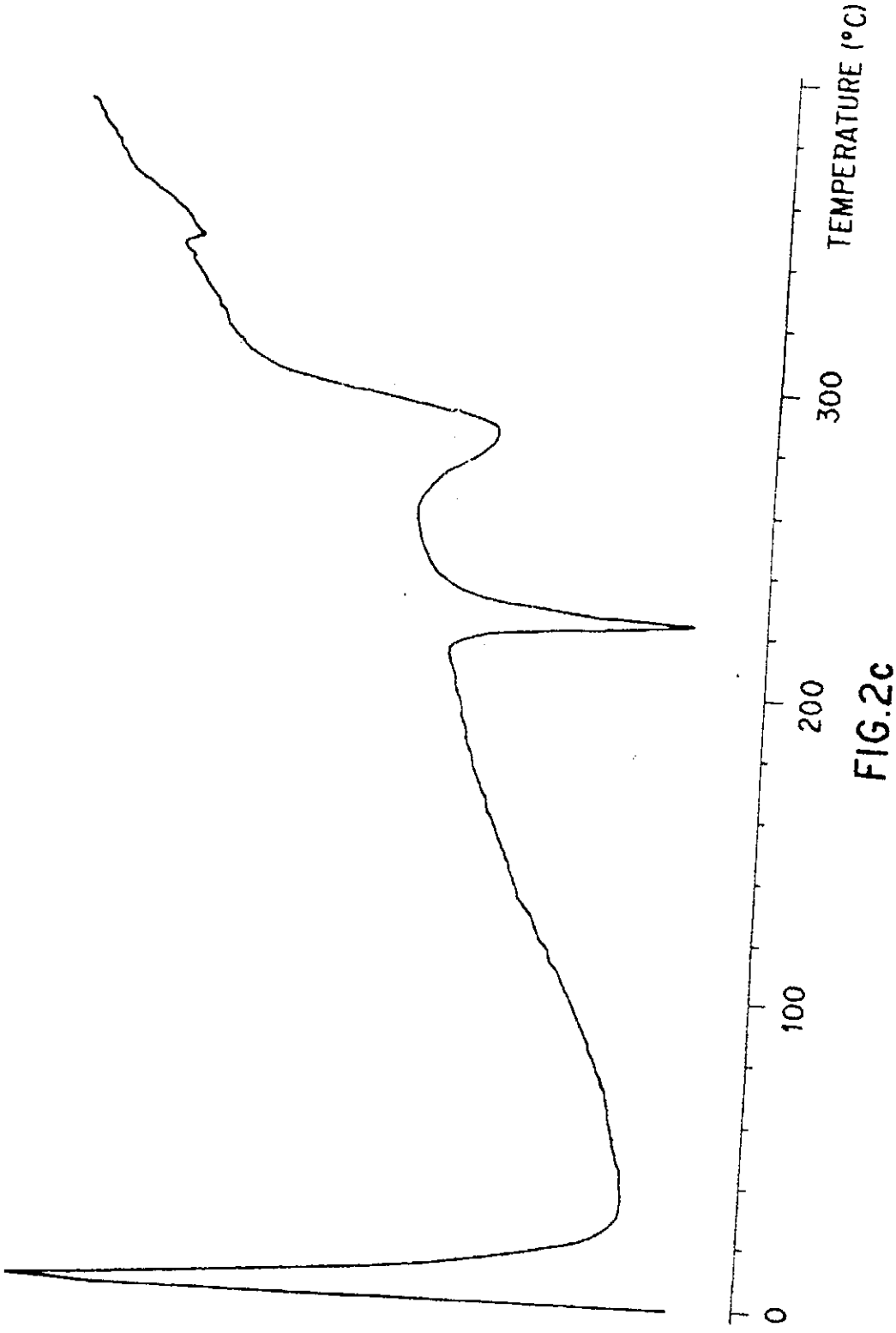


FIG.2c

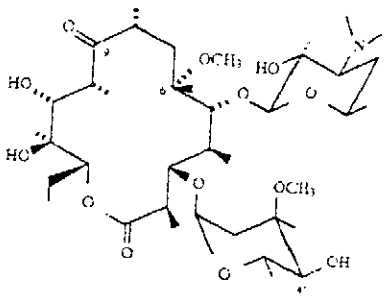
PREPARATION OF CRYSTAL FORM II OF CLARITHROMYCIN

TECHNICAL FIELD

This invention relates to a compound having therapeutic utility and to a method for its preparation. More particularly, the present invention concerns a process for the direct isolation of 6-O-methylerythromycin A crystal form II.

BACKGROUND OF THE INVENTION

6-O-methylerythromycin A (Clarithromycin, Biaxin) is a semisynthetic macrolide antibiotic of formula



6-O-methyl erythromycin A

which exhibits excellent antibacterial activity against gram-positive bacteria, some gram-negative bacteria, anaerobic bacteria, Mycoplasma, and Chlamidia. It is stable under acidic conditions and is efficacious when administered orally. Clarithromycin is a useful therapy for infections of the upper respiratory tract in children and adults.

BRIEF DESCRIPTION OF THE DRAWING

FIGS. *1a*, *1b* and *1c* show, respectively, the powder X-ray diffraction spectrum, the infrared spectrum, and the differential scanning calorimetric (DSC) thermogram of 6-O-methylerythromycin A form I.

FIGS. *2a*, *2b* and *2c* show, respectively, the powder X-ray diffraction spectrum, the infrared spectrum, and the differential scanning calorimetric (DSC) thermogram of 6-O-methylerythromycin A form II.

SUMMARY OF THE INVENTION

We have discovered that 6-O-methylerythromycin A can exist in at least two distinct crystalline forms, which for the sake of identification are designated "Form I" and "Form II". The crystal forms are identified by their infrared spectrum and powder x-ray diffraction pattern. Investigations in our laboratory have revealed that 6-O-methylerythromycin A prepared by the various methods described in the patent literature summarized below, in which the compound is purified by recrystallization from ethanol, result in exclusive initial formation of crystal form I.

Drugs currently on the market are formulated from the thermodynamically more stable form II. Therefore, preparation of the current commercial entity requires converting the form I crystals to form II. Typically this is done by heating the form I crystals under vacuum at a temperature of greater than 80° C. Therefore, a process for the preparation of 6-O-methylerythromycin A form II which does not require the high temperature treatment would produce substantial processing cost savings.

Although recrystallization from ethanol, tetrahydrofuran, isopropanol or isopropyl acetate results in exclusive formation of form I crystals, 6-O-methylerythromycin A Form II can be isolated directly by using a number of other common organic solvents, or mixtures of common organic solvents, thereby eliminating the additional conversion step.

Accordingly, the present invention provides a process for preparing 6-O-methylerythromycin A Form II comprising

(a) converting erythromycin A to 6-O-methylerythromycin A;

(b) treating the 6-O-methylerythromycin A prepared in step (a) with a solvent selected from the group consisting of (i) an alkanol of from 1 to 5 carbon atoms, provided said alkanol is not ethanol or isopropanol, (ii) a hydrocarbon of from 5 to 12 carbon atoms, (iii) a ketone of from 3 to 12 carbon atoms, (iv) a carboxylic ester of from 3 to 12 carbon atoms, provided said carboxylic ester is not isopropyl acetate, (v) an ether of from 4 to 10 carbon atoms, (vi) benzene, (vii) benzene substituted with one or more substituents selected from the group consisting of alkyl of from one to four carbon atoms, alkoxy of from one to four carbon atoms, nitro, and halogen, (viii) a polar aprotic solvent, (ix) a compound having the formula HNR^1R^2 wherein R^1 and R^2 are independently selected from hydrogen and alkyl of one to four carbon atoms, provided that R^1 and R^2 are not both hydrogen, (x) water and a second solvent selected from the group consisting of a water miscible organic solvent and a water miscible alkanol, (xi) methanol and a second solvent selected from the group consisting of a hydrocarbon of from 5 to 12 carbon atoms, an alkanol of from 2 to 5 carbon atoms, a ketone of from 3 to 12 carbon atoms, a carboxylic ester of from 3 to 12 carbon atoms, an ether of from 4 to 10 carbon atoms, benzene, and benzene substituted with one or more substituents selected from the group consisting of alkyl of from one to four carbon atoms, alkoxy of from one to four carbon atoms, nitro, and halogen, and (xii) a hydrocarbon of from 5 to 12 carbon atoms and a second solvent selected from the group consisting of a ketone of from 3 to 12 carbon atoms, a carboxylic ester of from 3 to 12 carbon atoms, an ether of from 4 to 10 carbon atoms, benzene, benzene substituted with one or more substituents selected from the group consisting of alkyl of from one to four carbon atoms, alkoxy of from one to four carbon atoms, nitro, and halogen, and a polar aprotic; and

(c) isolating the 6-O-methylerythromycin A form II crystals.

DETAILED DESCRIPTION

6-O-methylerythromycin A is prepared by methylation of the 6-hydroxy group of erythromycin A. However, in addition to the 6 position, erythromycin A contains hydroxy groups at the 11, 12, 2' and 4" positions, and a nitrogen at 3' position, all of which are potentially reactive with alkylating agents. Therefore, it is necessary to protect the various reactive functionalities prior to alkylation of the 6-hydroxy group. Representative 6-O-methylerythromycin A preparations are described in U.S. Pat. Nos. 4,331,803, 4,670,549, 4,672,109 and 4,990,602 and European Patent Specification 260 938 B 1 which are incorporated herein by reference. Following final removal of the protecting groups, the 6-O-methylerythromycin A may exist as a solid, a semisolid, or a syrup containing residual solvents from the deprotection reactions, inorganic salts, and other impurities. 6-O-methylerythromycin A form II may be crystallized directly from the syrup or semisolid using the solvent systems

described above. Alternatively, if the crude reaction product solidifies, the solid may be recrystallized from any of the solvent systems described above. Pure 6-O-methylerytaromycin A form II may also be obtained by recrystallizing form I or mixtures of form I and form II from any of the solvent systems described above. The term "6-O-methylerythromycin A" as used herein is meant to include 6-O-methylerythromycin A Form I or II in any state of purity, or mixtures thereof.

The term "treating" refers to crystallizing or recrystallizing 6-O-methylerythromycin A as defined above from any of the solvent systems described above.

The term "hydrocarbon" as used herein refers to straight chain or branched alkanes having the formula C_nH_{2n+2}. Hydrocarbons suitable for use in isolating 6-O-methylerythromycin A form II crystals include hexane, heptane, octane and the like.

The term "alkyl" refers to a monovalent group derived from a straight or branched chain saturated hydrocarbon by the removal of a single hydrogen atom. Alkyl groups are exemplified by methyl, ethyl, n- and iso-propyl, n-, sec-, iso- and tert-butyl, and the like.

The term "ketone" refers to a solvent of formula $RC(O)R'$ where R and R' are straight or branched alkyl. Ketones suitable for use in isolating 6-O-methylerythromycin A form II crystals include acetone, methyl ethyl ketone, 2-, and 3-pentanone, and the like.

The term "carboxylic ester" means a solvent of formula RCO_2R' where R and R' are straight or branched alkyl. Carboxylic esters suitable for use in isolating 6-O-methylerythromycin A form II crystals include methyl acetate, ethyl acetate, isobutyl acetate, and the like.

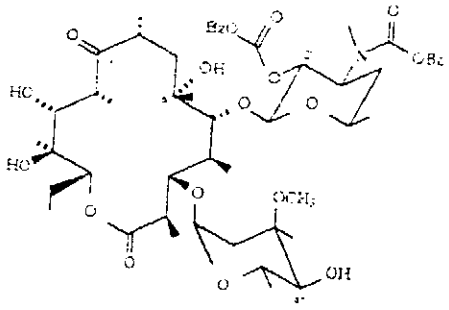
The term "ether" means a solvent of formula ROR' where R and R' are straight or branched alkyl. Ethers suitable for use in isolating 6-O-methylerythromycin A form II crystals include ethyl ether, diisopropyl ether, methyl tert-butyl ether, and the like.

The term "polar aprotic" refers to solvents which do not contain hydroxy groups but have a relatively high dipole moment. Polar aprotic solvents suitable for use in isolating 6-O-methylerythromycin A form II crystals include acetonitrile, N,N-dimethylformamide (DMF), dimethyl sulfoxide (DMSO), 1,1-dimethoxyethane (DME), hexamethylphosphoric triamide (HMPA), and the like.

The term "water miscible organic solvents" means organic solvents which are substantially miscible with water. Examples of water miscible organic solvents suitable for use in isolating 6-O-methylerythromycin A form II crystals from water miscible organic solvent-water mixtures include acetone, acetonitrile, N,N-dimethylformamide (DMF), dimethyl sulfoxide (DMSO), tetrahydrofuran, dioxane, ethylene glycol diethyl ether, ethylene glycol dimethyl ether (glyme), and diethylene glycol dimethyl ether (diglyme).

The term "alkanol" refers to a hydrocarbon as defined above substituted with one or more hydroxy groups. Representative alkanols include methanol, propanol, isopropanol, butanol, isobutanol, ethylene glycol, and the like.

The term "water miscible alkanols" means an alkanol as defined above which is substantially miscible with water. Examples of water miscible alkanols suitable for use in isolating 6-O-methylerythromycin A form II crystals from water miscible alkanol-water mixtures include methanol, ethanol, propanol, isopropanol, butanol, isobutanol and tert-butanol. 6-O-methylerythromycin A is prepared from erythromycin A by a variety of synthetic routes. In one method, erythromycin A is converted to 2'-O-3'-N-bis (benzyloxycarbonyl)-N-

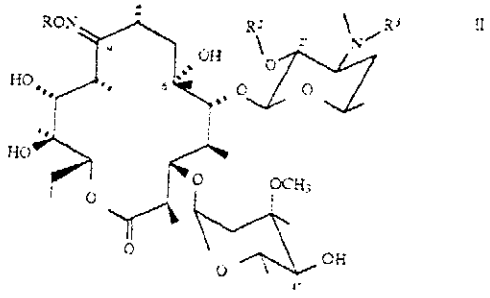


I

demethylerythromycin A (I). The 6-hydroxy group is then methylated by reaction with an alkylating agent such as bromomethane or iodomethane and a base. Removal of the benzoyl groups by catalytic hydrogenation and reductive methylation of the 3' N gives 6-O-methylerythromycin A. See U.S. Pat. No. 4,331,803

An alternative synthetic route involves methylation of 6-O-methylerythromycin A-9-oxime. 6-O-methylerythromycin A-9-oxime is prepared by methods well known in the art such as reaction of erythromycin A with hydroxylamine hydrochloride in the presence of base, or by reaction with hydroxylamine in the presence of acid as described in U.S. Pat. No. 5,274,085. Reaction of the oxime with RX wherein R is allyl or benzyl and X is halogen results in formation of 2'-O,3'-N-diallyl or dibenzylerythromycin A-9-O-allyl or benzyloxime halide. Methylation of this quartemary salt as described above, followed by elimination of the R groups and deoximination gives 6-O-methylerythromycin A. See U.S. Pat. No. 4,670,549.

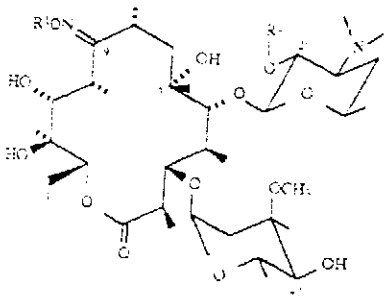
Methylation of 6-O-methylerythromycin A oxime derivatives of formula II,



II

wherein R is alkyl, alkenyl, substituted or unsubstituted benzyl, oxyalkyl, or substituted phenylthioalkyl, R^2 is benzoyl, and R^3 is methyl or benzoyl, followed by deprotection, deoximation, and reductive methylation when R^3 is benzoyl gives 6-O-methylerytaromycin A. See U.S. Pat. Nos. 4,672,109.

A particularly useful preparation of 6-O-methylerythromycin A involves methylation of the

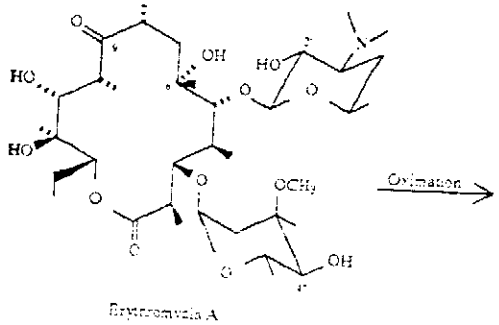


III

oxime derivative III, wherein R^1 is alkenyl, substituted or unsubstituted benzyl, or alkoxyalkyl, R^2 is substituted silyl, and R^3 is R^2 or H. Removal of the protecting groups and deoximation is then accomplished in a single step by treatment with acid to give 6-O-methylerythromycin A. See European Patent Specification 260 938 B 1 and U.S. Pat. No. 4,990,602.

A preferred route of 6-O-methylerythromycin A is outlined in Scheme 1. Erythromycin A, prepared by fermentation of *Streptomyces erythreus* is oximated to give oxime 4 wherein R^1 is alkoxyalkyl. The group R^1 may be introduced by reaction of erythromycin A with the substituted hydroxylamine R^1ONH_2, or by reaction of erythromycin A with hydroxylamine hydrochloride in the presence of base, or hydroxylamine in the presence of acid, followed by reaction with R^1X. The two hydroxy groups are then protected simultaneously, in which R^2 or R^3 are the same, or sequentially in which R^2 and R^3 are different. Particularly useful protecting groups are substituted silyl groups such as trimethylsilyl, tert-butyldimethylsilyl, tert-butyldiphenylsilyl and the like. The protecting groups are then removed and the compound is deoximated to produce 6-O-methylerythromycin A. The order of deprotection/deoximation is not critical. When the protecting groups are substituted silyl, deprotection and deoximation can be accomplished in a single step by treatment with acid, for example using formic acid or sodium hydrogen sulfite. See U.S. Pat. No. 4,990,602.

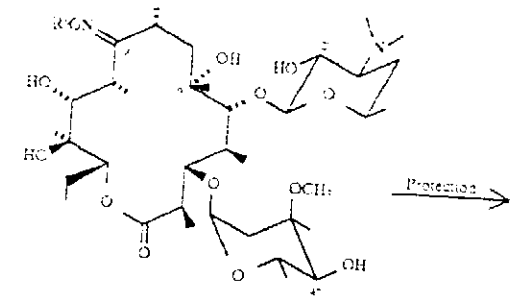
Scheme 1



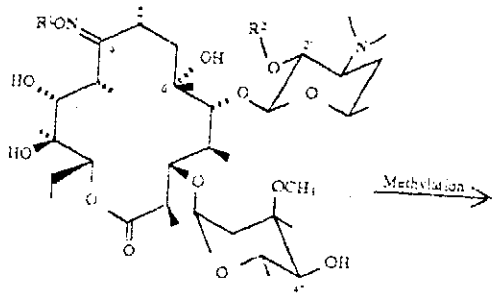
Erythromycin A

-Continued

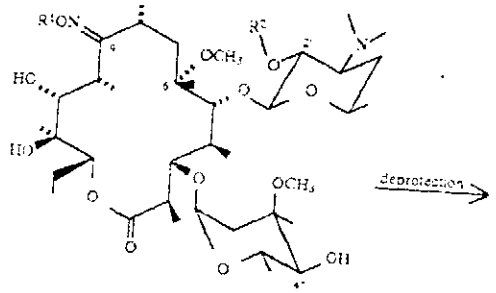
Scheme 1



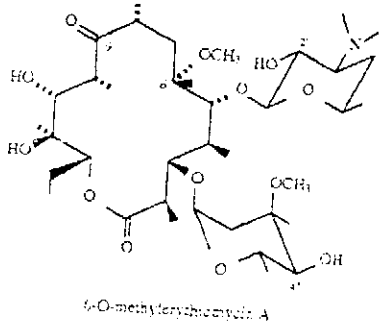
IV



V



VI



6-O-methylerythromycin A

The 6-O-methylerythromycin A prepared as described above is suspended in the desired solvent and heated to about the reflux temperature of the solvent. Heating is then continued and the suspension is stirred for an amount of time sufficient to dissolve most of the solid, generally about 10 minutes to 2 hours. The suspension is then filtered hot. If necessary, the filtrate may be heated to at or about the reflux temperature of the solvent to form a clear solution. The filtrate is then slowly cooled to ambient temperature with

optional further cooling in an ice-water bath. For purposes of this specification, ambient temperature is from about 20° to about 25° C. 6-O-methylerythromycin A crystal form II is isolated by filtration and dried in a vacuum oven at a temperature of between ambient temperature and about 50° C., and a pressure of between about 2 inches of mercury and atmospheric pressure to remove any remaining solvent.

When 6-O-methylerythromycin A is treated with a water miscible organic solvent and water or a water miscible alkanol and water, a suspension of 6-O-methylerythromycin A in the organic solvent or alkanol is heated to reflux and hot filtered. If necessary, the filtrate is heated at about the reflux temperature of the solvent until a clear solution is obtained. The clear solution is then mixed with water and cooled to ambient temperature with optional further cooling in an ice bath. The upper limit on the amount of water occurTs when the mixture separates into two liquid phases. A preferred ratio is about 1:1 parts by volume of water. After cooling, 6-O-methylerythromycin A crystal form II is isolated by filtration and dried as described above. A preferred water miscible organic solvent is tetrahydrofuran. Preferred water miscible alkanols include methanol, ethanol, propanol and isopropanol.

In another aspect of the present invention, 6-O-methylerythromycin A may is treated with mixtures of methanol and a second solvent. In this case, the second solvent my include solvents such as ethanol, isopropanol, tetrahydrofuran or isopropyl acetate which normally result in formation of form I crystals. Because the drug may have comparable solubilities in methanol and the second solvent, the amount of methanol must be carefully controlled to ensure maximum recovery. Preferred amounts of methanol are from about 1:3 to about 1:1 parts by volume. An especially preferred ratio is about 1:1 parts by volume of methanol. In accordance with this aspect of the invention, a suspension of 6-O-methylerythromycin A in the second solvent is heated to reflux and hot filtered. If necessary, the filtrate is heated at about the reflux temperature of the second solvent until a clear solution is obtained. The hot solution is then mixed with methanol and cooled to ambient temperature with optional further cooling in an ice bath. Alternatively, when 6-O-methylerythromycin A has comparable solubility in both the second solvent and methanol, the second solvent and methanol are premixed in a ratio of about 1:1 parts by volume and the drug is suspended in a the solvent mixture, followed by heating, filtration, and cooling as described above. After cooling, 6-O-methylerythromycin A crystal form II is isolated by filtration and dried as described above.

In accordance with the aspects of this invention wherein 6-O-methylerythromycin A is treated with hydrocarbon-second solvent mixtures, 6-O-methylerythromycin A is suspended in the desired second solvent and heated to about the reflux temperature of the second solvent The suspension is then heated and stirred for an amount of time sufficient to dissolve most of the solid, generally about 10 minutes to 2 hours. The suspension is then filtered hot. The filtrate may be heated to reflux to form a clear solution if necessary. A hydrocarbon solvent is then added to the hot filtrate and the mixture is cooled slowly to ambient temperature with optional further cooling in an ice bath. After cooling, 6-O-methylerythromycin A crystal form II is isolated by filtration and dried as described above. The amount of hydrocarbon solvent added is dependent on the solubility of the drug in the second solvent and the hydrocarbon solvent, and can be readily determined by one of ordinary skill in the art. Typical ratios fall in the range of about 1:10 to about 1:1 parts by volume of hydrocarbon solvent.

In a preferred embodiment, 6-O-methylerythromycin A crystal form II is isolated by treating 6-O-methylerythromycin A with a solvent selected from the group consisting of acetone, heptane, toluene, methyl tert-butyl ether, N,N-dimethylformamide, ethyl acetate, xylene, ethyl ether, amyl acetate, diisopropyl ether, and isopropyl butyrate.

In a more preferred embodiment, 6-O-methylerythromycin A crystal form II is isolated by treating 6-O-methylerythromycin A with water and a solvent selected from the group consisting of a water miscible organic solvent and a water miscible alkanol. An especially preferred water miscible organic solvent is tetrahydrofuran. Especially preferred water miscible alkanols are methanol, ethanol, propanol, and isopropanol.

When water is replaced with methanol in solvent mixtures, drying times are shortened or drying can be accomplished at a lower temperature. Therefore, in a still more preferred embodiment, 6-O-methylerythromycin A crystal form II is isolated by treating 6-O-methylerythromycin A with a solvent comprising methanol and a second solvent selected from the group consisting of a hydrocarbon of from 5 to 12 carbon atoms, an alkanol of from 2 to 5 carbon atoms, a ketone of from 3 to 12 carbon atoms, a carboxylic ester of from 3 to 12 carbon atoms, an ether of from 4 to 10 carbon atoms, benzene, and benzene substituted with one or more substituents selected from the group consisting of alkyl of from one to four carbon atoms, alkoxy of from one to four carbon atoms, nitro, and halogen. Preferred solvents according to this embodiment are methanol and alkanols of from 2 to 5 carbon atoms, and methanol and carboxylic esters of from 3 to 12 carbon atoms. Especially preferred solvents are methanol-ethanol and methanol-isopropyl acetate.

In the most preferred embodiment of the present invention, 6-O-methylerythromycin A crystal form II is isolated by treating 6-O-methylerythromycin A with a solvent having the formula HNR^1R^2 wherein R^1 and R^2 are independently selected from hydrogen and alkyl of one to four carbon atoms, provided that R^1 and R^2 are not both hydrogen. Alkyl and dialkylamines are preferred because 6-O-methylerythromycin A is substantially soluble in these solvents and the solvents are readily evaporated, resulting in lower solvent and energy costs. The most preferred solvent is isopropylamine.

The foregoing may be better understood by reference to the following examples which are provided for illustration and not intended to limit the scope of the inventive concept.

REFERENCE EXAMPLE

6-O-methylerythromycin A was prepared from erythromycin A by oximation of the C-9 carbonyl, protection of the C-2' and C-4" hydroxy groups, methylation of the C-6 hydroxy group, deoximation and removal of the protecting groups, and recrystallization from ethanol according to the method of U.S. Pat. No. 4,990,502 to give 6-O-methylerythromycin A form I. The form I crystals (0.40 g) were placed in a vial and heated in the vacuum oven (4–9 in Hg, 100°–110° C.) for 18 hours to give 6-O-methylerythromycin A form II crystals

6-O-methylerythromycin A form II melts at 223.4° C. in the differential scanning calorimetric thermogram of 6-O-methylerythromycin A form II there can be seen an endothermic peak at 283.3° C. which may be due to decomposition. After the DSC scan the color of the sample was black. The 2-theta angle positions in the powder x-ray diffraction pattern of 6-O-methylerythromycin A form I are 8.52°±0.2,

9.48°±0.2, 10.84°±0.2, 11.48°±0.2, 11.48°±0.2, 12.36°±0.2, 13.72°±0.2, 14.12°±0.2, 15.16°±0.2, 16.48°±0.2, 16.92°±0.2, 17.32°±0.2, 18.08°±0.2, 18.40°±0.2, 19.04°±0.2, 19.88°±0.2, and 20.48°±0.2.

EXAMPLE 1

Recrystallization from Acetone

A suspension of 6-O-methylerythromycin A (30 g) in acetone (200 mL) was heated at reflux for 15 minutes. The hot solution was filtered and 5.53 g of solid was removed. The filter flask was rinsed with acetone (5 mL). The combined filtrate and rinse was warmed to reflux and acetone (45 mL) was added to dissolve all remaining solid. The solution was cooled to ambient temperature and then in an ice-water bath. The resulting solid was filtered and dried overnight in a vacuum oven (4–9 in Hg, 40°–45° C.) to give 6-O-methylerythromycin A form II (17.8 g).

EXAMPLE 2

Recrystallization from Heptane

A suspension of 6-O-methylerythromycin A (10 g) in heptane (1000 mL) was heated at reflux (98° C.) for 1.5 hours. The hot solution was filtered and 1.91 g of solid was removed. The filtrate was warmed to reflux and heated for 35 minutes. The clear solution was cooled to ambient temperature and then in an ice-water bath. The resulting solid was filtered and dried overnight in a vacuum oven (4–9 in Hg, 40°–45° C.) to give 6-O-methylerythromycin A form II (5.7 g).

EXAMPLE 3

Recrystallization from Toluene

A suspension of 6-O-methylerythromycin A (30 g) in toluene (100 mL) was heated at reflux (110°–112° C.) for 1.5 hours. The hot solution was filtered and the filter flask was rinsed with toluene (10 mL). The combined filtrate and rinse was warmed to reflux (110° C.) and heated for 35 minutes. The solution was cooled to ambient temperature and then in an ice-water bath. The resulting solid was filtered and dried overnight in a vacuum oven (4–9 in Hg, 40°–45° C.) to give 6-O-methylerythromycin A form II (5.7 g).

EXAMPLE 4

Recrystallization from Methyl tert-Butyl Ether

A suspension of 6-O-methylerythromycin A (10 g) in methyl tert-butyl ether (200 mL) was heated at reflux (55° C.) for 15 minutes. The hot solution was filtered and 2.6 g of solid was removed. The filtrate was warmed to reflux and methyl tert-butyl ether (70 mL) was added to dissolve the remaining solid. The solution was cooled to ambient temperature and then in an icewater bath. The resulting solid was filtered and dried overnight in a vacuum oven (4–9 in Hg, 40°–45° C.) to give 6-O-methylerythromycin A form II (4.5 g).

EXAMPLE 5

Recrystallization from N,N-Dimethylformamide

A suspension of 6-O-methylerythromycin A (20 g) in N,N-dimethylformamide (200 mL) was heated at reflux (153° C.) for 15 minutes. The hot solution was filtered and

the filtrate was warmed to reflux. The clear solution was cooled slowly to ambient temperature and stirred for four days. The resulting solid was filtered and dried overnight in a vacuum oven (4–9 in Hg, 40°–45° C.) to give 6-O-methylerythromycin A form II (7.4 g).

EXAMPLE 6

Recrystallization from Ethyl Acetate

A suspension of 6-O-methylerythromycin A (15 g) in ethyl acetate (100 mL) was heated at reflux (77° C.) for 30 minutes. The hot solution was filtered and the filtrate was warmed to reflux. To the cloudy solution was added ethyl acetate (15 mL). The resulting clear solution was cooled to ambient temperature overnight. The resulting solid was filtered and dried in a vacuum oven (4–9 in Hg, 40°–45° C.) for 91 hours to give 6-O-methylerythromycin A form II (8.7 g).

EXAMPLE 7

Recrystallization from Xylene

A suspension of 6-O-methylerythromycin A (35 g) in xylene (105 mL) was heated to 140° C. at which point a clear solution was obtained. Additional 6-O-methylerythromycin A (5.0 g) was added and the hot solution was filtered to remove a trace amount of insoluble material. The filter flask was rinsed with xylene (5 mL) and the combined filtrate and rinse were heated at reflux for 15 minutes. The solution was cooled to ambient temperature and then in an ice water bath. The resulting solid was filtered and dried overnight in a vacuum oven (4–9 in Hg, 40°–45° C.) to give 6-O-methylerythromycin A form II (29 g).

EXAMPLE 8

Recrystallization from Isopropanol-Water

A suspension of 6-O-methylerythromycin A (20 g) and isopropanol (100 mL) was heated to reflux (82° C.). The hot solution was filtered and 1.16 g of solid was removed. The filtrate was diluted with isopropanol (20 mL) and was again warmed to reflux. The hot suspension was filtered and 3.5 g of 6-O-methylerythromycin A was collected. To the filtrated was added isopropanol (50 mL) and the mixture was heated at reflux until a clear solution was obtained. To the clear solution was added water (100 mL) and the solution was cooled in an ice bath. The resulting solid was filtered and dried overnight in a vacuum oven (4–9 in Hg, 40°–45° C.) to give 6-O-methylerythromycin A form II (9.5 g).

EXAMPLE 9

Recrystallization from Tetrahydrofuran-Water

A suspension of 6-O-methylerythromycin A (30 g) in THF (100 mL) was heated at reflux (66.5° C.) for 20 minutes. The hot solution was filtered to remove a trace amount of insoluble material. The filtrate was warmed to (66.5° C.) and water (100 mL) was added at which point a solid formed. The suspension was cooled to ambient temperature and filtered. The solid was dried in a vacuum oven (4–9 in Hg, 40°–45° C.) for four days to give 6-O-methylerythromycin A form II (24 g).

EXAMPLE 10

Recrystallization from Ethanol-Water

A suspension of 6-O-methylerythromycin A (20 g) in ethanol (200 mL) was heated to 78° C. The hot solution was

filtered and 12.6 g of solid was removed. The filtrate was warmed to reflux and water (200 mL) was added. The mixture was cooled to ambient temperature and filtered. The solid was dried in a vacuum oven (4–9 in Hg, 40°–45° C.) to give 6-O-methylerythromycin A form II (8.8 g).

EXAMPLE 11

Recrystallization from Ethyl Ether

A suspension of 6-O-methylerythromycin A (5.0 g) in ethyl ether (150 mL) was warmed to reflux. The insoluble solids were removed by filtration and the filtrate was cooled to ambient temperature. A precipitate slowly appeared and was isolated by filtration to give 6-O-methylerythromycin A form II (0.8 g). The filtrate was stirred overnight at ambient temperature to give an additional 0.65 g of 6-O-methylerythromycin A form II.

EXAMPLE 12

Recrystallization from Amyl Acetate

A suspension of 6-O-methylerythromycin A in amyl acetate (100 mL) was warmed 93° C. at which point the solution was almost clear. A trace amount of insoluble solids were removed by filtration of the hot solution and the filtrate was cooled to ambient temperature. A precipitate slowly appeared and was isolated by filtration to give 6-O-methylerythromycin A form II (6.9 g) after drying overnight at ambient temperature (4–9 in Hg).

EXAMPLE 13

Recrystallization from Isopropyl Acetate-Methanol

A suspension of 6-O-methylerythromycin A (12 g) in isopropyl acetate (100 mL) was warmed to reflux. The hot solution was filtered and the filtrate was transferred to another vessel. The filter flask was rinsed with isopropyl acetate (10 mL) and the combined filtrate and rinse were warmed to reflux. Methanol (100 mL) was added and the clear solution was cooled slowly to ambient temperature during which time a precipitate formed. After three hours at ambient temperature the precipitate was collected by filtration. The solid was dried in a vacuum oven (4–9 in Hg, 40°–45° C.) to give 6-O-methylerythromycin A form II (6.8 g).

EXAMPLE 14

Recrystallization from Diisopropyl Ether

A suspension of 6-O-methylerythromycin A (3.0 g) and diisopropyl ether (150 mL) was warmed to reflux. The hot solution was filtered rapidly and the filtrate was cooled to ambient temperature over two hours. The resulting solid was collected by filtration and dried in the vacuum oven (7–9 in Hg, 45°–50° C.) to give 6-O-methylerythromycin A form II (0.27 g).

EXAMPLE 15

Recrystallization from Isopropyl Butyrate

A suspension of 6-O-methylerythromycin A (5.0 g) in ispropyl butyrate (100 mL) was warmed to 90° C. The resulting clear solution was cooled to ambient temperature over three hours and then was cooled for 30 minutes in an ice-water bath. The resulting solid was collected by filtration and dried in the vacuum oven (2–4 in Hg, 45°–50° C.) to give 6-O-methylerythromycin A form II (2.8 g).

EXAMPLE 16

Recrystallization from Isopropylamine

A clear solution resulting from addition of 6-O-methylerythromycin A (8.0 g) to isopropylamine (50 mL) was stirred overnight at ambient temperature. When no precipitate formed, and additional 10.4 g of 6-O-methylerythromycin A was added. The clear solution was stirred overnight at ambient temperature during which time a precipitate formed. The solid was collected by filtration and dried in the vacuum oven (2–4 in Hg, 45°–50° C.) to give 6-O-methylerythromycin A form II (16.2 g).

EXAMPLE 17

Recrystallization from Methanol-Ethanol

A mixture of 6-O-methylerythromycin A (15 g),ethanol (100 mL) and methanol (100 mL) was warmed to 69° C. and stirred for 30 minutes. The hot solution was filtered and the filtrate was transferred to another vessel. The clear solution was cooled to ambient temperature over two hours and then was stirred for 30 minutes in an ice-water bath. The resulting solid was collected by filtration and dried in the vacuum oven (2–4 in Hg, 45°–50° C.) to give 6-O-methylerythromycin A form II (7.1 g).

The foregoing examples are presented for purposes of illustration and are not intended to limit the scope of the invention. Variations and changes which are obvious to one skilled in the art are intended to be within the scope and nature of the invention as defined in the appended claims.

We claim:

1. A method of preparing 6-O-methylerythromycin A crystal form II comprising

(a) converting erythromycin A to 6-O-methylerythromycin A;

(b) treating the 6-O-methylerythromycin A prepared in step a with a solvent selected from the group consisting of

(i) an alkanol of from 1 to 5 carbon atoms, provided said alkanol is not ethanol or isopropanol,

(ii) a hydrocarbon of from 5 to 12 carbon atoms,

(iii) a ketone of from 3 to 12 carbon atoms,

(iv) a carboxylic ester of from 3 to 12 carbon atoms, provided said carboxylic ester is not isopropyl acetate,

(v) an ether of from 4 to 10 carbon atoms,

(vi) benzene,

(vii) benzene substituted with one or more substituents selected from the group consisting of

alkyl of from one to four carbon atoms,

alkoxy of from one to four carbon atoms,

nitro, and

halogen,

(viii) a polar aprotic solvent,

(ix) a compound having the formula HNR^1R^2 wherein R^1 and R^2 are independently selected from hydrogen and alkyl of one to four carbon atoms, provided that R^1 and R^2 are not both hydrogen,

(x) water and a water miscible solvent selected from the group consisting of

a water miscible organic solvent and

a water miscible alkanol,

(xi) methanol and a second solvent selected from the group consisting of

a hydrocarbon of from 5 to 12 carbon atoms,
an alkanol of from 2 to 5 carbon atoms,
a ketone of from 3 to 12 carbon atoms,
a carboxylic ester of from 3 to 12 carbon atoms,
an ether of from 4 to 10 carbon atoms,
benzene, and
benzene substituted with one or more substituents selected from the group consisting of
alkyl of from one to four carbon atoms,
alkoxy of from one to four carbon atoms,
nitro, and
halogen, and

(xii) a hydrocarbon of from 5 to 12 carbon atoms and a second solvent selected from the group consisting of
a ketone of from 3 to 12 carbon atoms,
a carboxylic ester of from 3 to 12 carbon atoms,
an ether of from 4 to 10 carbon atoms,
benzene,
benzene substituted with one or more substituents selected from the group consisting of
alkyl of from one to four carbon atoms,
alkoxy of from one to four carbon atoms,
nitro, and
halogen, and
a polar aprotic; and

(c) isolating the 6-O-methylerythromycin A form II crystals.

2. The process of claim 1 wherein step (a) comprises

(i) converting erythromycin A into an erythromycin A 9-oxime derivative;

(ii) protecting the 2' and 4" hydroxy groups of the erythromycin A 9-oxime derivative prepared in step (i);

(iii) reacting the product of step (ii) with a methylating agent; and

(iv) deprotecting and deoximating the product of step (iii) to form 6-O-methylerythromycin A.

3. A method according to claim 2 wherein the solvent comprises water and a water miscible organic solvent or a water miscible alkanol.

4. A method according to claim 3 wherein the solvent comprises water and a water miscible organic solvent or water miscible alkanol in a ratio of about 1:1 parts by volume.

5. A method according to claim 4 wherein the solvent comprises water and a water miscible organic solvent.

6. A method of preparing 6-O-methylerythromycin A crystal foim II according to claim 5 wherein water miscible organic solvent is tetrahydrofuran.

7. A method according to claim 4 wherein the solvent comprises water and a water miscible alkanol.

8. A method according to claim 7 wherein the water miscible alkanol is selected from the group consisting of methanol, ethanol, and isopropanol.

9. A method of according to claim 2 wherein the solvent comprises methanol and a second solvent selected from the group consisting of

a hydrocarbon of from 5 to 12 carbon atoms,
an alkanol of from 2 to 5 carbon atoms,
a ketone of from 3 to 12 carbon atoms,
a carboxylic ester of from 3 to 12 carbon atoms,
an ether of from 4 to 10 carbon atoms,
benzene, or
benzene substituted with one or more substituents selected from the group consisting of
alkyl of from one to four carbon atoms,
alkoxy of from one to four carbon atoms,
nitro, and
halogen.

10. A method according to claim 9 wherein the solvent comprises methanol and

an alkanol of from 2 to 5 carbon atoms, or
a carboxylic ester of from 3 to 12 carbon atoms.

11. A method according to claim 10 wherein the solvent comprises methanol and an alkanol of from 2 to 5 carbon atoms, or a carboxylic ester of from 3 to 12 carbon atoms in a ratio of about 1:1 parts by volume

12. A method according to claim 11 wherein the solvent comprises methanol and a second solvent selected from ethanol and isopropyl acetate.

13. A method according to claim 2 wherein the solvent comprises a compound of formula HNR^1R^2 wherein R^1 and R^2 are independently selected from hydrogen and alkyl of one to four carbon atoms, provided that R^1 and R^2 are not both hydrogen.

14. A method according to claim 13 wherein the solvent is isopropylamine.

15. A method according to claim 2 wherein the solvent is selected from the group consisting of
acetone,
heptane,
toluene,
methyl tert-butyl ether,
N,N-dimethylformamide,
ethyl acetate,
xylene,
isopropanol-water,
tetrahydrofurali-water,
ethanol-water,
ethyl ether,
amyl acetate,
isopropyl acetate-methanol
diisopropyl ether,
isopropyl butyrate,
isopropylamine, and
methanol-ethanol.

16. 6-O-methylerythromycin A form II prepared according to the process of claim 2.

* * * * *

Exhibit B

US005858986A

United States Patent [19]

Liu et al.

[11] Patent Number: 5,858,986

[45] Date of Patent: Jan. 12, 1999

[54] CRYSTAL FORM I OF CLARITHROMYCIN

[75] Inventors: Jih-Hua Liu, Green Oaks, Ill.; David A. Riley, Kenosha, Wis.; Steven G. Spanton, Green Oaks, Ill.

[73] Assignee: Abbott Laboratories, Abbott Park, Ill.

[21] Appl. No.: 681,723

[22] Filed: Jul. 29, 1996

[51] Int. Cl.[6] A61K 31/70; C07H 1.00; C07H 17/08

[52] U.S. Cl. 514/29; 536/7.2; 536/18.5; 536/127

[58] Field of Search 536/7.2, 7.5, 18.5, 536/127; 574/29

[56] References Cited

U.S. PATENT DOCUMENTS

4,670,549 6/1987 Morimoto et al. 536/7.4
4,672,109 6/1987 Wantanabe et al. 536/7.2
4,990,602 2/1991 Morimoto et al. .

FOREIGN PATENT DOCUMENTS

97/19096 A 5/1997 WIPO .

OTHER PUBLICATIONS

Quantitative Structure–Activity Relationships In Drug Design, vol. 291 (1989), pp. 325–328, Kim et al., "Conformational Study of Erythromycin Analogues".

Acta Crystallographica, vol. c49, No. 5 (May 1993), pp. 1227–1230, Iwasaki et al., "Structure of 6–O–Methylerythromycin A (Clarithromycin)".

Primary Examiner—Elli Peselev
Attorney, Agent, or Firm—Mona Anand

[57] ABSTRACT

The present invention concerns the novel antiobiotic 6-O-methylerythromycin A crystal form I, a process for its preparation, pharmaceutical compositions comprising this compound and a method of use as a therapeutic agent.

14 Claims, 6 Drawing Sheets

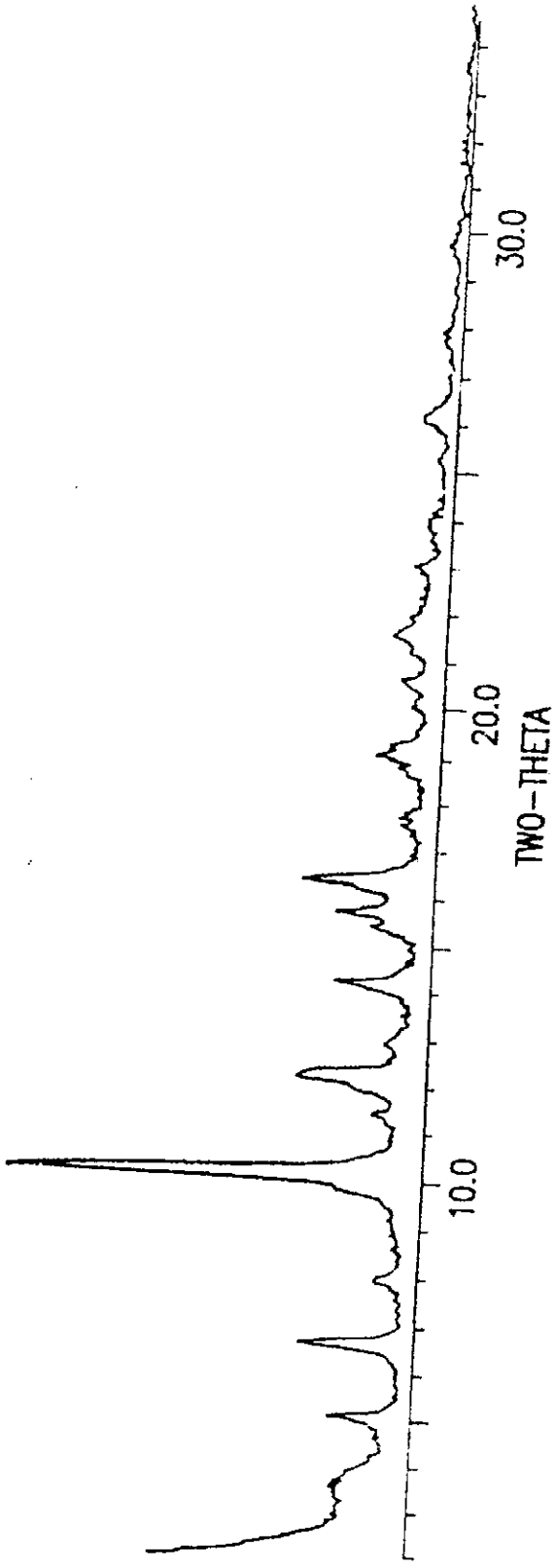


FIG. 1a

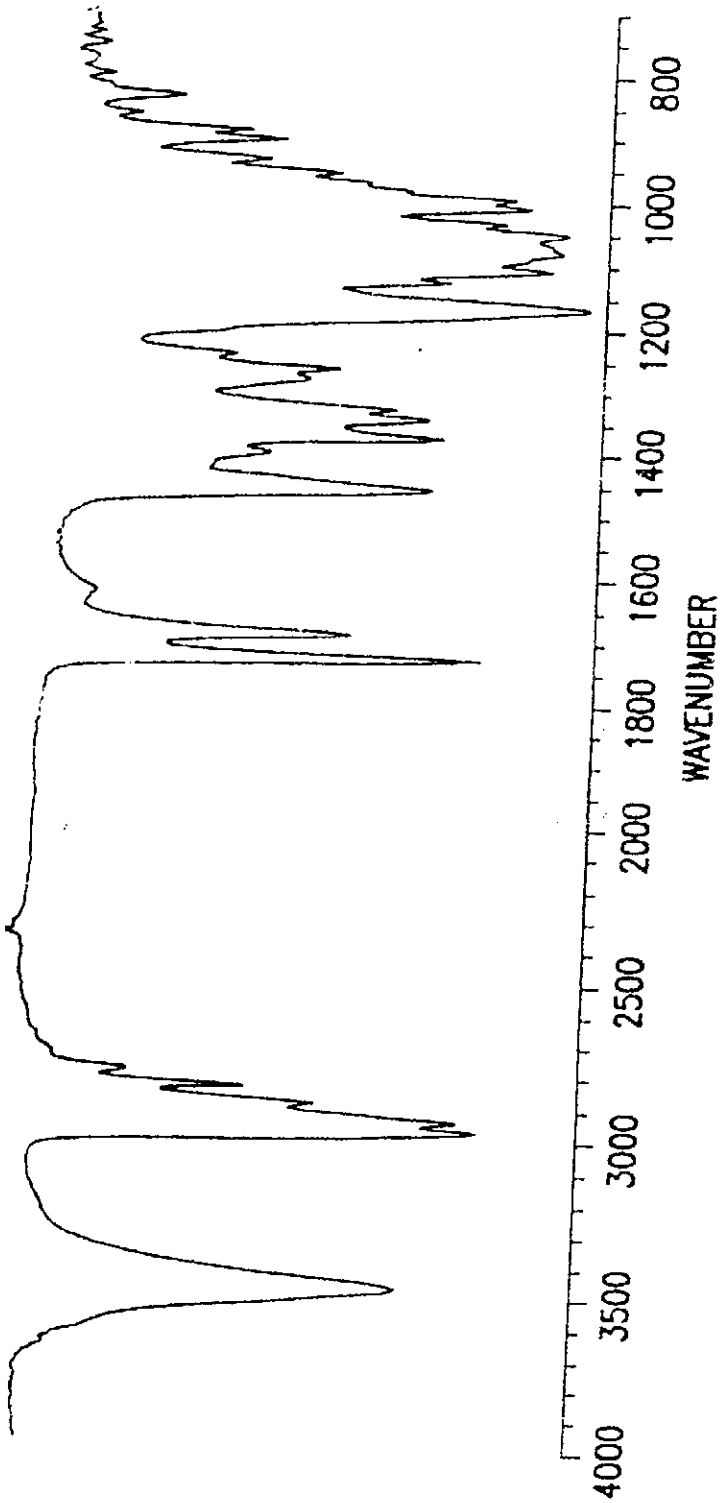

4000
3500
3000
2500
2000
1800
1600
1400
1200
1000
800
WAVENUMBER

FIG. 1b

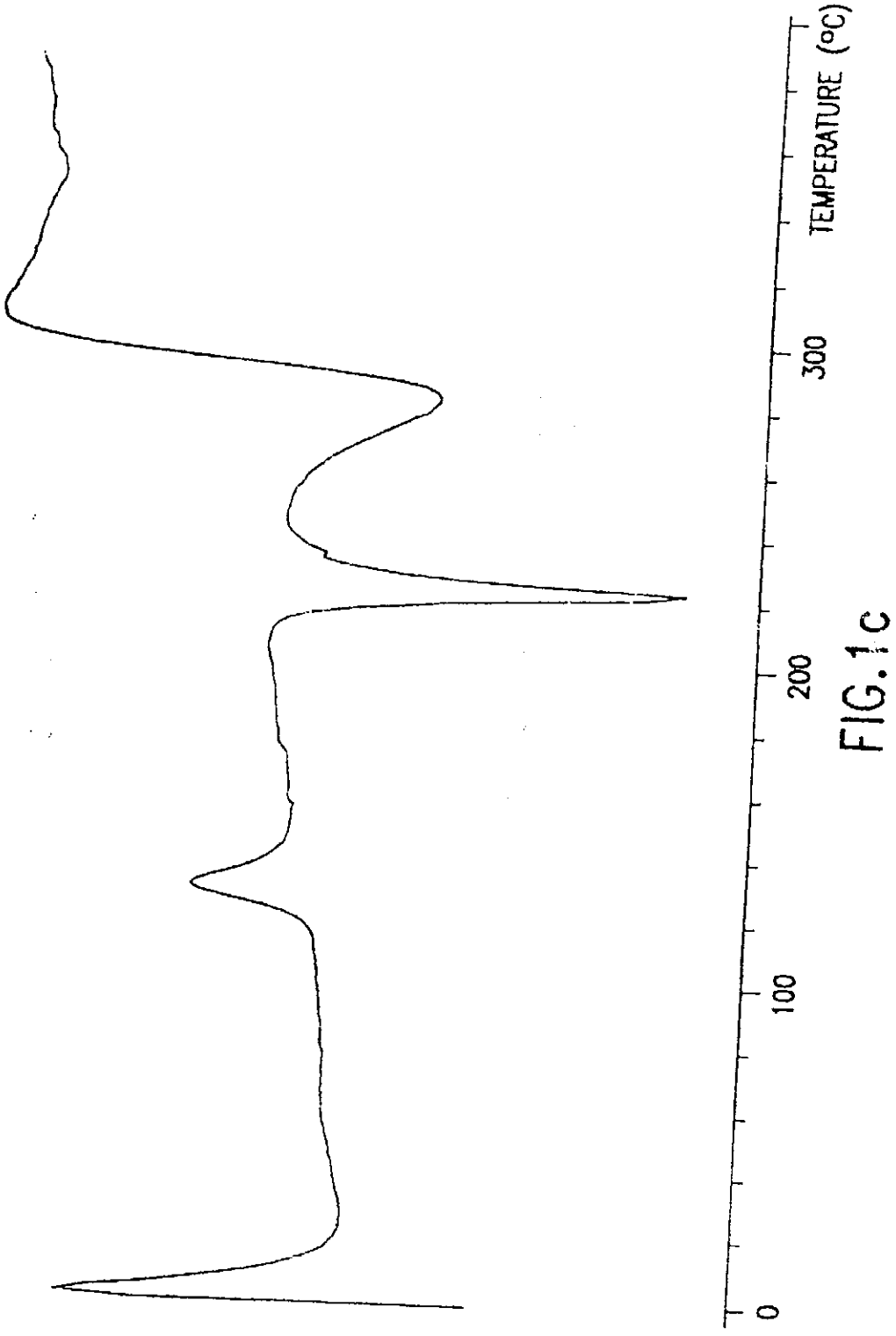


FIG. 1C

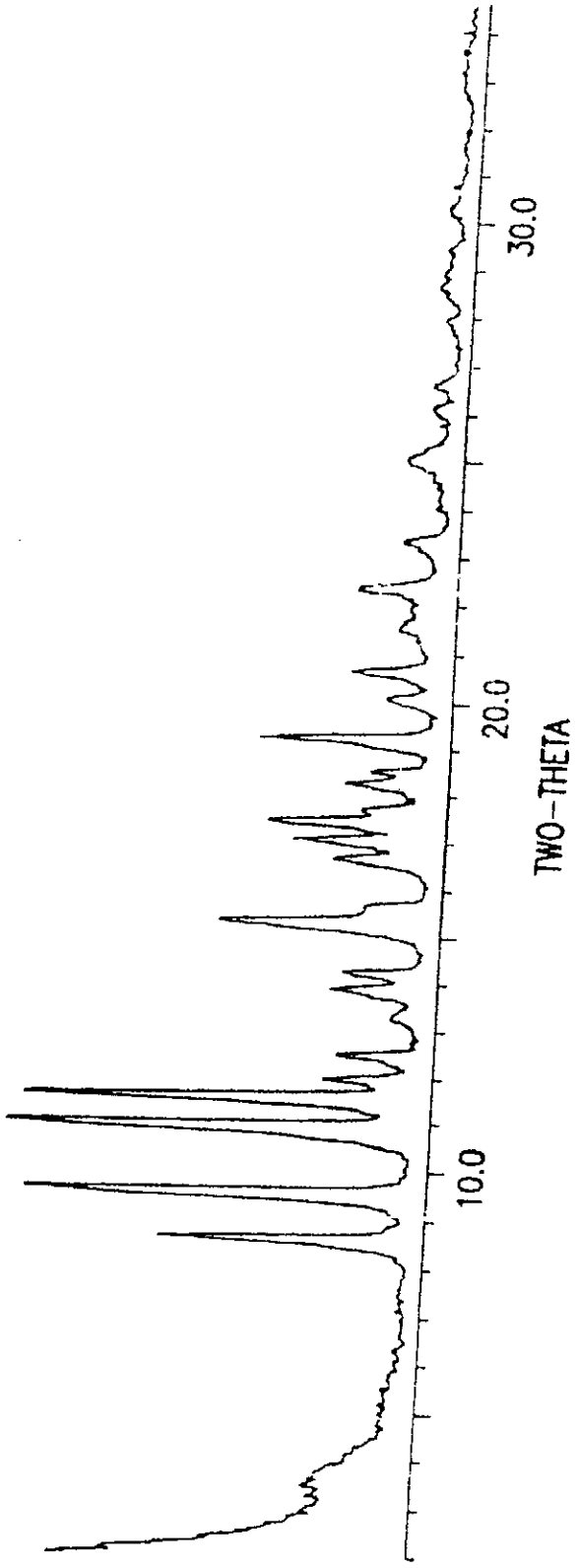


FIG.2a

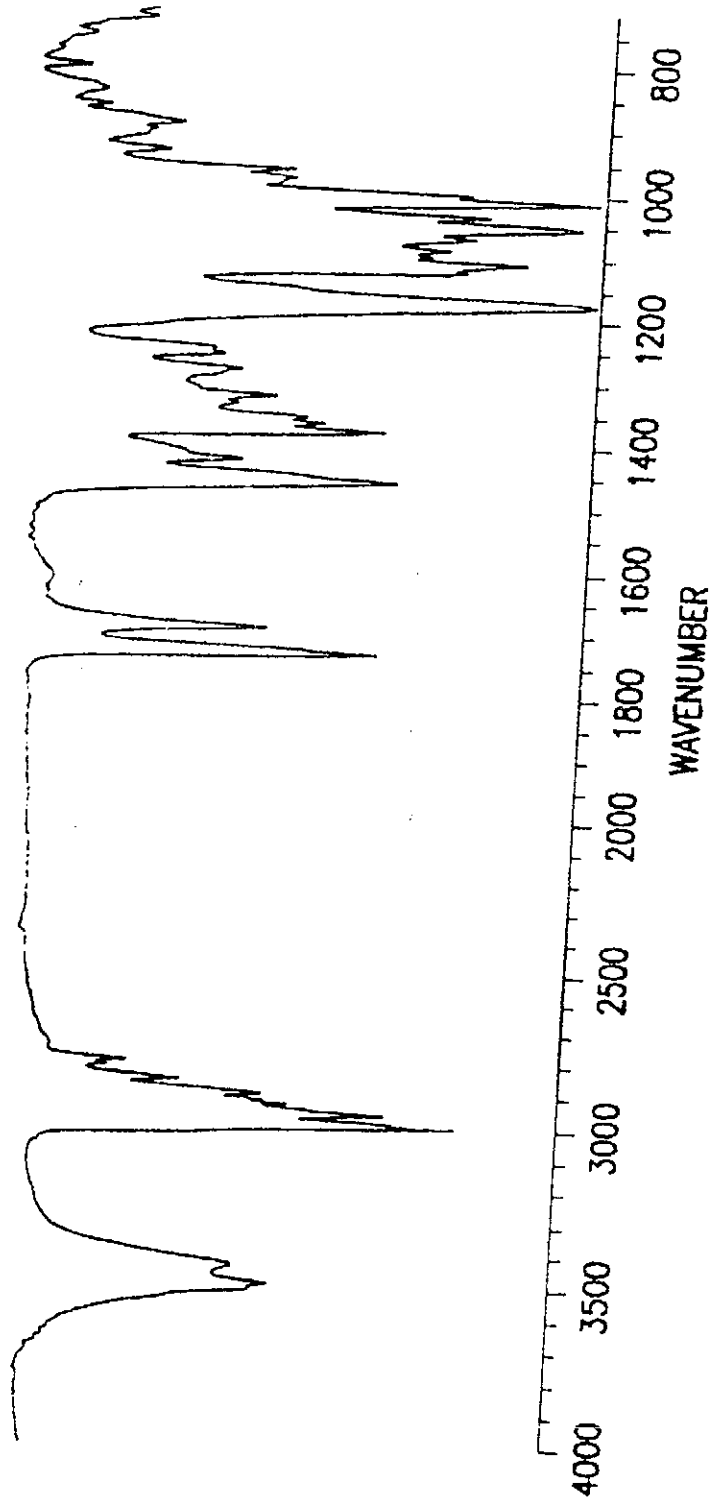

800
1000
1200
1400
1600
1800
2000
2500
3000
3500
4000
WAVENUMBER

FIG. 2b

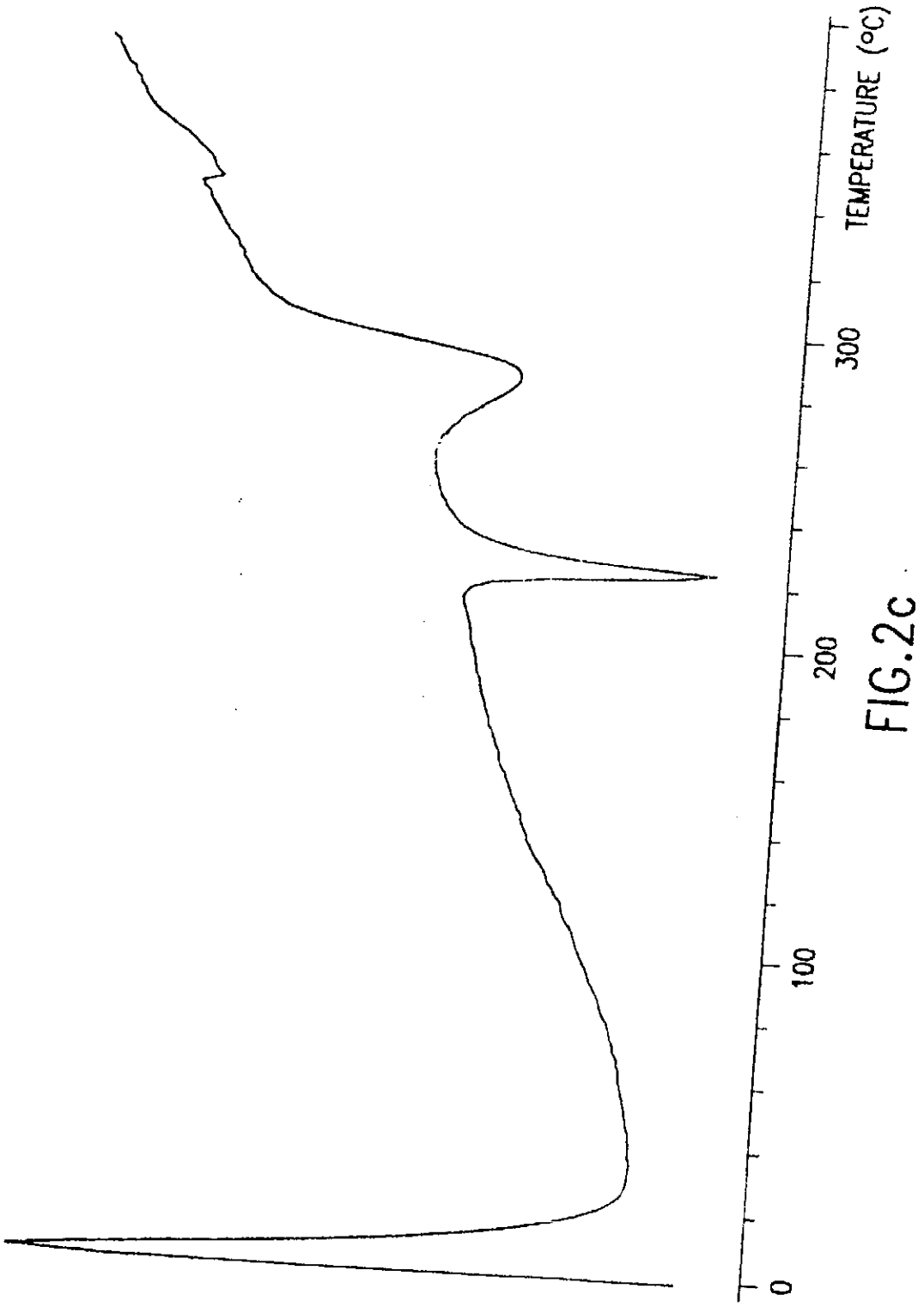


FIG. 2c

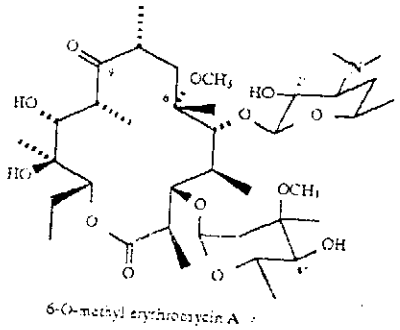
CRYSTAL FORM I OF CLARITHROMYCIN

TECHNICAL FIELD

This invention relates to a compound having therapeutic utility and to a method for its preparation. More particularly, the present invention concerns the novel compound 6-O-methylerythromycin A crystal form I, a process for its preparation, pharmaceutical compositions comprising this compound and a method of use as a therapeutic agent.

BACKGROUND OF THE INVENTION

6-O-methylerythromycin A (Clarithromycin) is a semi-synthetic macrolide antibiotic of formula



6-O-methyl erythromycin A

which exhibits excellent antibacterial activity against gram-positive bacteria, some gram-negative bacteria, anaerobic bacteria, Mycoplasma, and Chlamidia. It is stable under acidic conditions and is efficacious when administered orally. Clarithromycin is a useful therapy for infections of the upper respiratory tract in children and adults.

BRIEF DESCRIPTION OF THE DRAWING

FIGS. 1*a*, 1*b* and 1*c* show, respectively, the powder X-ray diffraction spectrum, the infrared spectrum, and the differential scanning calorimetric (DSC) thermogram of 6-O-methylerythromycin A form I.

FIGS. 2*a*, 2*b* and 2*c* show, respectively, the powder X-ray diffraction spectrum, the infrared spectrum, and the differential scanning calorimetric (DSC) thermogram of 6-O-methylerythromycin A form II.

SUMMARY OF THE INVENTION

We have discovered that 6-O-methylerythromycin A can exist in at least two distinct crystalline forms, which for the sake of identification are designated "Form I" and "Form II". The crystal forms are identified by their infrared spectrum and powder x-ray diffraction pattern. Form I and form II crystals have an identical spectrum of antibacterial activity, but form I crystals unexpectedly have an intrinsic rate of dissolution about three times that of form II crystals. Investigations in our laboratory have revealed that 6-O-methylerythromycin A prepared by the various methods described in the patent literature summarized below, in which the compound is purified by recrystallization from ethanol, result in exclusive initial formation of crystal form I. Further investigation revealed that recrystallization from tetrahydrofuran, isopropyl acetate, and isopropanol, or mixtures of ethanol, tetrahydrofuran, isopropyl acetate, or isopropanol with other common organic solvents result in exclusive formation of form I crystals.

Drugs currently on the market are formulated from the thermodynamically more stable form II crystals. Therefore, preparation of the current commercial entity requires converting the form I crystals to form II. Typically this is done by heating the form I crystals under vacuum at a temperature of greater than 80° C. Therefore, the discovery of a novel form of 6-O-methylerythromycin A which can be prepared without the high temperature treatment results in substantial processing cost savings. In addition, the favorable dissolution characteristics of form I relative to form II increases bioavailability of the antibiotic and provides significant formulation advantages.

Accordingly, the present invention in its principle embodiment provides a novel crystalline antibiotic designated 6-O-methylerythromycin A form I, or a pharmaceutically acceptable salt thereof.

The present invention also provides pharmaceutical compositions which comprise a therapeutically effective amount of 6-O-methylerythromycin A form I in combination with a pharmaceutically acceptable carrier.

The invention further relates to a method of treating bacterial infections in a host mammal in need of such treatment comprising administering to the mammal a therapeutically effective amount of 6-O-methylerythromycin A form I.

In another embodiment, the present invention provides a process for preparing 6-O-methylerythromycin A form I comprising

(a) converting erythromycin A to 6-O- methylerythromycin A;

(b) treating the 6-O-methylerythromycin A with a solvent selected from the group consisting of (i) ethanol, (ii) isopropyl acetate, (iii) isopropanol, (iv) tetrahydrofuran, and (v) a mixture of a first solvent selected from the group consisting of ethanol, isopropyl acetate, isopropanol, and tetrahydrofuran and a second solvent selected from the group consisting of a hydrocarbon of from 5 to 12 carbon atoms, a ketone of from 3 to 12 carbon atoms, a carboxylic ester of from 3 to 12 carbon atoms, an ether of from 4 to 10 carbon atoms, benzene, benzene substituted with one or more substituents selected from the group consisting of alkyl of from one to four carbon atoms, alkoxy of from one to four carbon atoms, nitro, and halogen, and a polar aprotic solvent;

(c) isolating the crystalline 6-O-methylerythromycin A formed in step (b); and

(d) drying 6-O-methylerythromycin A isolate in step (c) at a temperature of between ambient temperature and about 70° C. to form 6-O-methylerythromycin A form I.

DETAILED DESCRIPTION

6-O-methylerythromycin A is prepared by methylation of the 6-hydroxy group of erythromycin A. However, in addition to the 6 position, erythromycin A contains hydroxy groups at the 11, 12, 2' and 4" positions, and a nitrogen at 3' position, all of which are potentially reactive with alkylating agents. Therefore, it is necessary to protect the various reactive functionalities prior to alkylation of the 6-hydroxy group. Representative 6-O-methylerythromycin A preparations are described in U.S. Pat. Nos. 4,331,803, 4,670,549, 4,672,109 and 4,990,602 and European Patent Specification 260 938 B1 which are incorporated herein by reference. Following final removal of the protecting groups, the 6-O-methylerythromycin A may exist as a solid, a semisolid, or a syrup containing residual solvents from the deprotection

reactions, inorganic salts, and other impurities. 6-O-methylerythromycin A form I may be crystallized directly from the syrup or semisolid using the solvents described above. Alternatively, if the crude reaction product solidifies, the solid may be recrystallized from any of the solvents described above. Pure 6-O-methylerythromycin A form I may also be obtained by recrystallizing form II or mixtures of form I and form II from any of the solvent systems described above. The term "6-O-methylerythromycin A" as used herein is meant to include 6-O-methylerythromycin A Form I or II in any state of purity, or mixtures thereof.

The term "treating" refers to crystallizing or recrystallizing 6-O-methylerythromycin A as defined above from any of the solvents described above.

The term "hydrocarbon" as used herein refers to straight chain or branched alkanes having the formula C_nH_{2n+2}. Hydrocarbons useful in the solvent mixtures of the present invention include hexane, heptane, octane and the like.

The term "alkyl" refers to a monovalent group derived from a straight or branched chain saturated hydrocarbon by the removal of a single hydrogen atom. Alkyl groups are exemplified by methyl, ethyl, n- and iso-propyl, n-, sec-, iso- and tert-butyl, and the like.

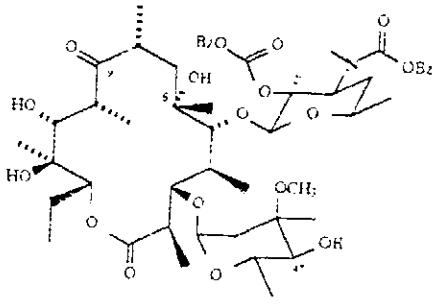
The term "ketone" refers to a solvent of formula RC(O)R' where R and R' are straight or branched alkyl. Ketones useful in the solvent mixtures of the present invention include acetone, methyl ethyl ketone, 2-, and 3-pentanone, and the like.

The term "carboxylic ester" means a solvent of formula RCO_2R' where R and R' are straight or branched alkyl. Carboxylic esters useful in the solvent mixtures of the present invention include methyl acetate, ethyl acetate, isobutyl acetate, and the like.

The term "ether" means a solvent of formula ROR' where R and R' are straight or branched alkyl. Ethers useful in the solvent mixtures of the present invention include ethyl ether, diisopropyl ether, methyl tert-butyl ether, and the like.

The term "polar aprotic" refers to solvents which do not contain hydroxy groups but have a relatively high dipole moment. Polar aprotic solvents useful in the solvent mixtures of the present invention include acetonitrile, N,N-dimethylformamide (DMF), dimethyl sulfoxide (DMSO), 1,1-dimethoxyethane (DME), hexamethylphosphoric triamide (HMPA), and the like.

By "pharmaceutically acceptable salt" it is meant those salts which are, within the scope of sound medical judgement, suitable for use in contact with the tissues of humans and lower animals without undue toxicity, irritation, allergic response and the like, and are commensurate with a reasonable benefit/risk ratio. Pharmaceutically acceptable salts are well known in the art. For example, S. M. Berge, et al. describe pharmaceutically acceptable salts in detail in *J. Pharmaceutical Sciences*, 1977, 66: 1–19. The salts can be prepared in situ during the final isolation and purification of the compounds of the invention, or separately by reacting the free base function with a suitable organic acid. Representative acid addition salts include acetate, adipate, alginate, ascorbate, aspartate, benzenesulfonate, benzoate, bisulfate, borate, butyrate, camphorate, camphersulfonate, citrate, cyclopentanepropionate, digluconate, dodecylsulfate, ethanesulfonate, fumarate, glucoheptonate, glycerophosphate, hemisulfate, heptonate, hexanoate, hydrobromide, hydrochloride, hydroiodide, 2-hydroxyethanesulfonate, lactobionate, lactate, laurate, lauryl sulfate, malate, maleate, malonate, methanesulfonate, 2-naphthalenesulfonate, nicotinate, nitrate, oleate, oxalate, palmitate, pamoate, pectinate, persulfate, 3-phenylpropionate, phosphate, picrate, pivalate, propionate, stearate, succinate, sulfate, tartrate, thiocyanate, toluenesulfonate, undecanoate, valerate salts, and the like. Representative alkali or alkaline earth metal salts include sodium, lithium, potassium, calcium, magnesium, and the like, as well as nontoxic ammonium, quaternary ammonium, and amine cations, including, but not limited to ammonium, tetramethylammonium, tetraethylammonium, methylamine, dimethylamine, trimethylamine, triethylamine, ethylamine, and the like. 6-O-methylerythromycin A is prepared from erythromycin A by a variety of synthetic routes. In one method, erythromycin A is converted to 2'-O-3'-N-bis (benzyloxycarbonyl)-N-demethylerythromycin A (I).

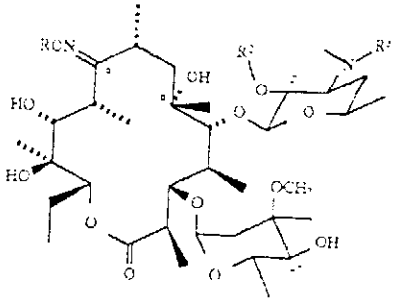


I

The 6-hydroxy group is then methylated by reaction with an alkylating agent such as bromomethane or iodomethane and a base. Removal of the benzoyl groups by catalytic hydrogenation and reductive methylation of the 3' N gives 6-O-methylerythromycin A. See U.S. Pat. No. 4,331,803.

An alternative synthetic route involves methylation of 6-O-methylerythromycin A-9-oxime. 6-O-methylerythromycin A-9-oxime is prepared by methods well known in the art such as reaction of erythromycin A with hydroxylamine hydrochloride in the presence of base, or by reaction with hydroxylamine in the presence of acid as described in U.S. Pat. No. 5,274,085. Reaction of the oxime with RX wherein R is allyl or benzyl and X is halogen results in formation of 2'-O,3+-N-diallyl or dibenzylerythromycin A-9-O-allyl or benzyloxime halide. Methylation of this quarternary salt as described above, followed by elimination of the R groups and deoximation gives 6-O-methylerythromycin A. See U.S. Pat. No. 4,670,549.

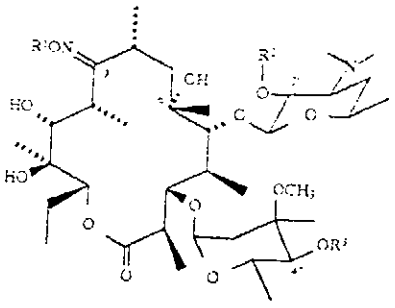
Methylation of 6-O-methylerythromycin A oxime derivatives of formula II,



II

wherein R is alkyl, alkenyl, substituted or unsubstituted benzyl, oxyalkyl, or substituted phenylthioalkyl. R^2 is benzoyl, and R^3 is methyl or benzoyl, followed by deprotection, deoximation, and reductive methylation when R^3 is benzoyl gives 6-O-methylerythromycin A. See U.S. Pat. No. 4,672,109.

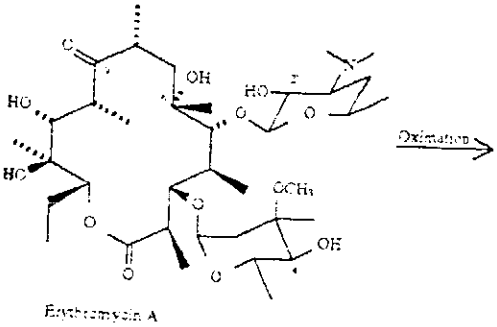
A particularly useful preparation of 6-O-methylerythromycin A involves methylation of the oxime derivative III,



wherein R^1 is alkenyl, substituted or unsubstituted benzyl, or alkoxyalkyl, R^2 is substituted silyl, and R^3 is R^2 or H. Removal of the protecting groups and deoximation is then accomplished in a single step by treatment with acid to give 6-O-methylerythromycin A. See European Patent Specification 260 938 B1 and U.S. Pat. No. 4,990,602.

A preferred route of 6-O-methylerythromycin A is outlined in Scheme 1. Erythromycin A, prepared by fermentation of *Streptomyces erythreus* is oximated to give oxime 4 wherein R^1 is alkoxyalkyl. The group R^1 may be introduced by reaction of erythromycin A with the substituted hydroxylamine R^1ONH_2, or by reaction of erythromycin A with hydroxylamine hydrochloride in the presence of base, or hydroxylamine in the presence of acid, followed by reaction with R^1X. The two hydroxy groups are then protected simultaneously, in which R^2 or R^3 are the same, or sequentially in which R^2 and R^3 are different. Particularly useful protecting groups are substituted silyl groups such as trimethylsilyl, tert-butyldimethylsilyl, tert-butyldiphenylsilyl and the like. The protecting groups are then removed and the compound is deoximated to produce 6-O-methylerythromycin A. The order of deprotection/deoximation is not critical. When the protecting groups are substituted silyl, deprotection and deoximation can be accomplished in a single step by treatment with acid, for example using formic acid or sodium hydrogen sulfite. See U.S. Pat. No. 4,990,602.

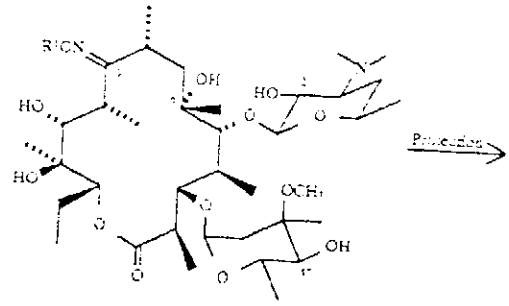
Scheme 1



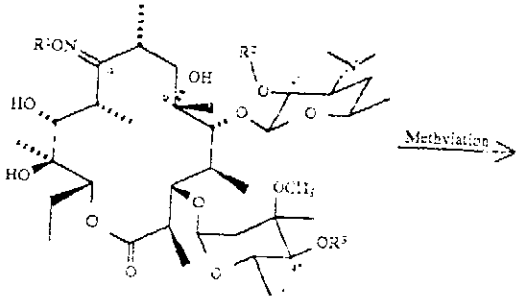
Erythromycin A

-continued

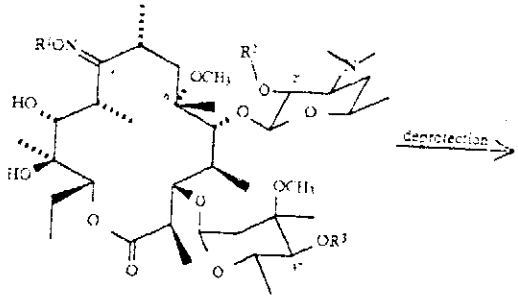
Scheme 1



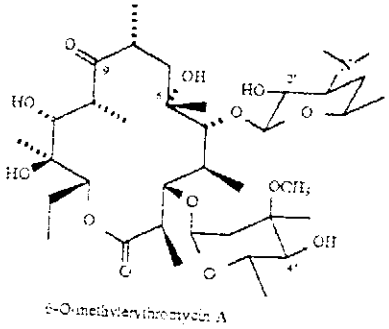
IV



V



VI



6-O-methylerythromycin A

In accordance with the process aspect of the present invention, 6-O-methylerythromycin A prepared by any of the methods described above is suspended in the desired solvent and heated to about the reflux temperature of the solvent. Heating is then continued and the suspension is stirred for an amount of time sufficient to dissolve most of the solid, generally about 10 minutes to 2 hours. The suspension is then filtered hot. If necessary, the filtrate may be heated to at or about the reflux temperature of the solvent

to form a clear solution. The filtrate is then slowly cooled to ambient temperature with optional further cooling in an ice-water bath. For purposes of this specification, ambient temperature is from about 20° C. to about 25° C. Crystalline 6-O-methylerythromycin A is then isolated, preferably by filtration, and the wet solid is converted to 6-O-methylerythromycin A form I by drying in a vacuum oven at a temperature of between ambient temperature and about 70° C., preferably from about 40 to about 50° C. and a pressure of between about 2 inches of mercury and atmospheric pressure to remove any remaining solvent.

In accordance with the aspects of this invention wherein 6-O-methylerythromycin A is recrystallized from solvent mixtures, 6-O-methylerythromycin A is suspended in the first solvent and heated to about the reflux temperature of the solvent. Heating is then continued and the suspension is stirred for an amount of time sufficient to dissolve most of the solid, generally about 10 minutes to 2 hours. The suspension is then filtered hot. The filtrate may be heated to reflux to form a clear solution if necessary. A second solvent is then added to the hot filtrate and the mixture is cooled slowly to ambient temperature with optional further cooling in an ice bath. Representative second solvents include, but are not limited to, hexane, heptane, octane, acetone, methyl ethyl ketone, 2-, and 3-pentanone, methyl acetate, ethyl acetate, isobutyl acetate, ethyl ether, diisopropyl ether, methyl tert-butyl ether, acetonitrile, N,N-dimethylformamide, dimethyl sulfoxide, 1,1-dimethoxyethane, hexamethylphosphoric triamide, benzene, toluene, and chlorobenzene. Hydrocarbons of from 5 to 12 carbon atoms are preferred second solvents. The most preferred second solvent is heptane. After cooling, 6-O-methylerythromycin A crystal form I is isolated by filtration and drying as described above. The amount of second solvent added is dependent on the solubility of the drug in the first solvent and the second solvent, and can be readily determined by one of ordinary skill in the art. Typical ratios fall in the range of about 1:10 to about 2:1 parts by volume of second solvent. A preferred ratio of first solvent to second solvent is 1:1 parts by volume.

Preferred solvents for the isolation of 6-O-methylerythromycin A form I are ethanol, isopropyl acetate, tetrahydrofuran, and isopropanol.

The most preferred solvent for the isolation of 6-O-methylerythromycin A form I is ethanol.

Pharmaceutical Compositions

The present invention also provides pharmaceutical compositions which comprise 6-O-methylerythromycin A form I formulated together with one or more non-toxic pharmaceutically acceptable carriers. The pharmaceutical compositions may be specially formulated for oral administration in solid or liquid form, for parenteral injection, or for rectal administration.

The pharmaceutical compositions of this invention can be administered to humans and other animals orally, rectally, parenterally, intracisternally, intravaginally, intraperitoneally, topically (as by powders, ointments, or drops), bucally, or as an oral or nasal spray. The term "parenteral" administration as used herein refers to modes of administration which include intravenous, intramuscular, intraperitoneal, intrasternal, subcutaneous and intraarticular injection and infusion.

Pharmaceutical compositions of this invention for parenteral injection comprise pharmaceutically acceptable sterile aqueous or nonaqueous solutions, dispersions, suspensions or emulsions as well as sterile powders for reconstitution into sterile injectable solutions or dispersions just prior to use. Examples of suitable aqueous and nonaqueous carriers, diluents, solvents or vehicles include water, ethanol, polyols (such as glycerol, propylene glycol, polyethylene glycol, and the like), and suitable mixtures thereof, vegetable oils (such as olive oil), and injectable organic esters such as ethyl oleate. Proper fluidity can be maintained, for example, by the use of coating materials such as lecithin, by the maintenance of the required particle size in the case of dispersions, and by the use of surfactants.

These compositions may also contain adjuvants such as preservative, wetting agents, emulsifying agents, and dispersing agents. Prevention of the action of microorganisms may be ensured by the inclusion of various antibacterial and antifungal agents, for example, paraben, chlorobutanol, phenol sorbic acid, and the like. It may also be desirable to include isotonic agents such as sugars, sodium chloride, and the like. Prolonged absorption of the injectable pharmaceutical form may be brought about by the inclusion of agents which delay absorption such as aluminum monostearate and gelatin.

In some cases, in order to prolong the effect of the drug, it is desirable to slow the absorption of the drug from subcutaneous or intramuscular injection. This may be accomplished by the use of a liquid suspension of crystalline or amorphous material with poor water solubility. The rate of absorption of the drug then depends upon its rate of dissolution which, in turn, may depend upon crystal size and crystalline form. Alternatively, delayed absorption of a parenterally administered drug form is accomplished by dissolving or suspending the drug in an oil vehicle.

Injectable depot forms are made by forming microencapsule matrices of the drug in biodegradable polymers such as polylactide-polyglycolide. Depending upon the ratio of drug to polymer and the nature of the particular polymer employed, the rate of drug release can be controlled. Examples of other biodegradable polymers include poly (orthoesters) and poly(anhydrides) Depot injectable formulations are also prepared by entrapping the drug in liposomes or microemulsions which are compatible with body tissues.

The injectable formulations can be sterilized, for example, by filtration through a bacterial-retaining filter, or by incorporating sterilizing agents in the form of sterile solid compositions which can be dissolved or dispersed in sterile water or other sterile injectable medium just prior to use.

Solid dosage forms for oral administration include capsules, tablets, pills, powders, and granules. In such solid dosage forms, the active compound is mixed with at least one inert, pharmaceutically acceptable excipient or carrier such as sodium citrate or dicalcium phosphate and/or a) fillers or extenders such as starches, lactose, sucrose, glucose, mannitol, and silicic acid, b) binders such as, for example, carboxymethylcellulose, alginates, gelatin, polyvinylpyrrolidone, sucrose, and acacia, c) humectants such as glycerol, d) disintegrating agents such as agar-agar, calcium carbonate, potato or tapioca starch, alginic acid, certain silicates, and sodium carbonate, e) solution retarding agents such as paraffin, f) absorption accelerators such as quaternary ammonium compounds, g) wetting agents such as, for example, cetyl alcohol and glycerol monostearate, h) absorbents such as kaolin and bentonite clay, and i) lubricants such as talc, calcium stearate, magnesium stearate, solid polyethylene glycols, sodium lauryl sulfate, and mixtures thereof. In the case of capsules, tablets and pills, the dosage form may also comprise buffering agents.

Solid compositions of a similar type may also be employed as fillers in soft and hard-filled gelatin capsules

using such excipients as lactose or milk sugar as well as high molecular weight polyethylene glycols and the like.

The solid dosage forms of tablets, dragees, capsules, pills, and granules can be prepared with coatings and shells such as enteric coatings and other coatings well known in the pharmaceutical formulating art. They may optionally contain opacifying agents and can also be of a composition that they release the active ingredient(s) only, or preferentially, in a certain part of the intestinal tract, optionally, in a delayed manner. Examples of embedding compositions which can be used include polymeric substances and waxes.

The active compounds can also be in micro-encapsulated form, if appropriate, with one or more of the above-mentioned excipients.

Liquid dosage form for oral administration include pharmaceutically acceptable emulsions, solutions, suspensions, syrups and elixirs. In addition to the active compounds, the liquid dosage forms may contain inert diluents commonly used in the art such as, for example, water or other solvents, solubilizing agents and emulsifiers such as ethyl alcohol, isopropyl alcohol, ethyl carbonate, ethyl acetate, benzyl alcohol, benzyl benzoate, propylene glycol, 1,3-butylene glycol, dimethyl formamide, oils (in particular, cottonseed, groundnut, corn, germ, olive, castor, and sesame oils), glycerol, tetrahydrofurfuryl alcohol, polyethylene glycols and fatty acid esters of sorbitan, and mixtures thereof.

Besides inert diluents, the oral compositions can also include adjuvants such as wetting agents, emulsifying and suspending agents, sweetening, flavoring, and perfuming agents.

Suspensions, in addition to the active compounds, may contain suspending agents as, for example, ethoxylated isostearyl alcohols, polyoxyethylene sorbitol and sorbitan esters, microcrystalline cellulose, aluminum metahydroxide, bentonite, agar-agar, and tragacanth, and mixtures thereof.

Compositions for rectal or vaginal administration are preferably suppositories which can be prepared by mixing the compounds of this invention with suitable non-irritating excipients or carriers such as cocoa butter, polyethylene glycol or a suppository wax which are solid at room temperature but liquid at body temperature and therefore melt in the rectum or vaginal cavity and release the active compound.

Compounds of the present invention can also be administered in the form of liposomes. As is known in the art, liposomes are generally derived from phospholipids or other lipid substances. Liposomes are formed by mono- or multilamellar hydrated liquid crystals that are dispersed in an aqueous medium. Any non-toxic, physiologically acceptable and metabolizable lipid capable of forming liposomes can be used. The present compositions in liposome form can contain, in addition to a compound of the present invention, stabilizers, preservatives, excipients, and the like. The preferred lipids are the phospholipids and the phosphatidyl cholines (lecithins), both natural and synthetic.

Methods to form liposomes are known in the art. See, for example, Prescott, Ed., *Methods in Cell Biology*, Volume XIV, Academic Press, New York, N.Y. (1976), p. 33 et seq.

Dosage forms for topical administration of a compound of this invention include powders, sprays, ointments and inhalants. The active compound is mixed under sterile conditions with a pharmaceutically acceptable carrier and any needed preservatives, buffers, or propellants which may be required. Opthalmic formulations, eye ointments, powders and solutions are also contemplated as being within the scope of this invention.

Actual dosage levels of active ingredients in the pharmaceutical compositions of this invention may be varied so as to obtain an amount of the active compound(s) that is effective to achieve the desired therapeutic response for a particular patient, compositions, and mode of administration. The selected dosage level will depend upon the activity of the particular compound, the route of administration, the severity of the condition being treated, and the condition and prior medical history of the patient being treated. However, it is within the skill of the art to start doses of the compound at levels lower than required for to achieve the desired therapeutic effect and to gradually increase the dosage until the desired effect is achieved.

Generally dosage levels of about 1 to about 1000, more preferably of about 5 to about 200 mg of 6-O-methylerythromycin A form I per kilogram of body weight per day are administered to a mammalian patient. If desired, the effective daily dose may be divided into multiple doses for purposes of administration, e.g. two to four separate doses per day.

The following Examples are provided to enable one skilled in the art to practice the invention and are merely illustrative of the invention. They should not be read as limiting the scope of the invention as defined in the claims.

EXAMPLE 1

Preparation of 6-O-methylerythromycin Form I

6-O-methylerythromycin A was prepared from erythromycin A by oximation of the C-9 carbonyl, protection of the C-2' and C-4" hydroxy groups, methylation of the C-6 hydroxy group, deoximation and removal of the protecting groups, and recrystallization from ethanol according to the method of U.S. Pat. No. 4,990,602. The material obtained from the recrystallization was dried in a vacuum oven (40°–45° C., 4–8 in. Hg) to give 6-O-methylerythromycin A form I.

In the differential scanning calorimetric thermogram of 6-O-methylerythromycin A form I there can be seen an exothermic transition at 132.2° C. which is believed to be due to a phase transition and an endothermic peak at 223.4° C. which may be due to melting. Another endothermic peak at 283.3° C. followed by an exothermic peak at 306.9° C. may be due to decomposition. After the DSC scan the color of the sample was black. The 2-theta angle positions in the powder x-ray diffraction pattern of 6-O-methylerythromycin A form I are 5.16°±0.2, 6.68°±0.2, 10.20°±0.2, 12.28°±0.2, 14.20°±0.2, 15.40°±0.2, 15.72°±0.2, and 16.36°±0.2.

EXAMPLE 2

Conversion of 6-O-methylerythromycin Form I Crystals to Form II Crystals

6-O-methylerythromycin A form I crystals (0.40 g), prepared as in Example 1, were placed in a vial and heated in the vacuum oven (4–9 in Hg, 100°–110° C.) 00 C) for 18 hours to give 6-O-methylerythromycin A form II crystals. 6-O-methylerythromycin A form II melts at 223.4° C. In the differential scanning calorimetric thermogram of 6-O-methylerythromycin A form II there can be seen an endothermic peak at 283.3° C. which may be due to decomposition. After the DSC scan the color of the sample was black. The 2-theta angle positions in the powder x-ray diffraction pattern of 6-O-methylerythromycin A form I are 8.52°±0.2, 9.48°±0.2, 10.84°±0.2, 11.48°±0.2, 11.88°±0.2, 12.36°±0.2, 13.72°±0.2, 14.12°±0.2, 15.16°±0.2, 16.48°±0.2,

15.92°±0.2, 17.32°±0.2, 18.08°±0.2, 18.40°±0.2, 19.04°±0.2, 19.88°±0.2, 20.48°±0.2.

EXAMPLE 3

Isolation of 6-O-methylerythromycin Form I by Recrystallization

Recrystallization from Tetrahydrofuran

A mixture of 6-O-methylerythromycin A (20 g), prepared as described in Example 1, in tetrahydrofuran (100 mL) was warmed to reflux and stirred for 15 minutes. The hot solution was filtered to remove traces of insoluble material and cooled to ambient temperature. No crystallization occurred so 10 g of 6-O-methylerythromycin A was added to the solution and the suspension was again heated to reflux, hot filtered, and cooled in an ice bath. The resulting solid was collected by filtration and dried in the vacuum oven (40°–45° C., 4–8 in. Hg) to give 6-O-methylerythromycin A form I (16.74 g).

Recrystallization from isopropyl alcohol

A mixture of 6-O-methylerythromycin A (15 g), prepared as described in Example 1, and isopropyl alcohol (100 mL) was warmed to reflux and heated for 20 minutes. The hot solution was filtered to remove traces of insoluble material. The filtrate was transferred to another flask along with a 50 mL isopropanol rinse, and the solution was again heated to reflux. The clear solution was then cooled slowly to ambient temperature and left standing for seven hours. The resulting solid was collected by filtration and dried in the vacuum oven (40°–45° C., 4–8 in. Hg) to give 6-O-methylerythromycin A form I (13.3 g).

Recrystallization from isopropyl acetate

A mixture of 6-O-methylerythromycin A (10 g), prepared as described in Example 1, and isopropyl acetate (100 mL) was warmed to 73° C. The hot solution was filtered to remove traces of insoluble material. The clear solution was then cooled slowly to ambient temperature. The resulting solid was collected by filtration and dried in the vacuum oven (40°–45° C., 4–8 in. Hg) to give 6-O-methylerythromycin A form I (3.6 g).

Recrystallization from Isopropyl Acetate-Heptane

A mixture of 6-O-methylerythromycin A (10 g), prepared as described in Example 1, and isopropyl acetate (100 mL) was warmed to reflux. A small amount of insoluble material was removed by filtration and the filtrate was transferred to another vessel. The filter flask was rinsed with isopropyl acetate (5 mL) and the filtrate and rinse were combined and heated to reflux. To the resulting clear solution was added heptane (100 mL) and the clear solution was cooled to ambient temperature over 1.5 hours during which time a precipitate formed. The solid was collected by filtration and dried overnight in the vacuum oven (45°–50° C., 4–8 in. Hg) to give 6-O-methylerythromycin A form I (7.0 g).

Recrystallization from Isopropyl Acetate-N,N-dimethylformamide

A mixture of 6-O-methylerythromycin A (12 g), prepared as described in Example 1, and isopropyl acetate (100 mL) was warmed to reflux. A small amount of insoluble material was removed by filtration and the filtrate was transferred to another vessel. The filtrate was heated to reflux and N,N-dimethylformamide (30 mL) was added. The clear solution was cooled to ambient temperature over 1.5 hours during which time a precipitate formed. The solid was collected by filtration and dried overnight in the vacuum oven (40°–50° C., 4–8 in. Hg) to give 6-O-methylerythromycin A form I (6.4 g).

Recrystallization from Tetrahydrofuran-Heptane

To a clear solution of 6-O-methylerythromycin A (10 g), prepared as described in Example 1, in tetrahydrofuran (75 mL) was added heptane (150 mL). The resulting cloudy solution was heated to 71.5° C. at which point it turned clear. The mixture was cooled to ambient temperature over 2 hours, and then was cooled in an ice-water bath for 0.5 hours. The resulting solid was filtered and dried in the vacuum oven (45°–50° C., 3–4 in. Hg) for four days to give 6-O-methylerythromycin A form I (0.50 g).

Recrystallization from Ethanol-Heptane

A mixture of 6-O-methylerythromycin A (10 g), prepared as described in Example 1, and ethanol (100 mL) was warmed to reflux. A small amount of insoluble material was removed by filtration and the filtrate was transferred to another vessel. The filter flask was rinsed with ethanol (20 mL) and the filtrate and rinse were combined and heated at 78° C. until a clear solution was obtained. To the clear solution was added heptane (100 mL) and the clear solution was cooled slowly to ambient temperature and stirred for four days. The resulting solid was collected by filtration and dried in the vacuum oven (45°–50° C., 4–8 in. Hg) to give 6-O-methylerythromycin A form I (4.5 g).

Recrystallization from Isopropanol-Heptane

A mixture of 6-O-methylerythromycin A (4.0 g), prepared as described in Example 1, and isopropanol (50 mL) was warmed to reflux. Heptane (50 mL) was added and the solution was cooled slowly to ambient temperature and then was cooled in an ice-water bath. The resulting solids were collected by filtration and dried in the vacuum oven (4–8 in. Hg) to give 6-O-methylerythromycin A form I (3.6 g).

EXAMPLE 4

Dissolution Rates of 6-O-methylerythromycin Forms I and II

Dissolution studies were carried out at 60 rpm in 300 mL of 0.05M phosphate buffer at 37° C. using a constant surface area (3/8" diameter) drug compact. Alliquots were removed periodically and assayed directly by HPLC (5cm×4.6 mm 3μODS-2 "Little Champ" (Regis) column; 50:50 acetonitrile-0.05M pH 4.0 phosphate buffer mobile phase; 1.0 mL/min flow rate). As shown in Table 1, 6-O-methylerythromycin A form I has an intrinsic rate of dissolution about three times greater than form II.

TABLE 1

Intrinsic Dissolution Rates of 6-O-methylerythromycin A forms I and II

Crystal Form	Dissolution Rate ± S.D. (μg min/cm²)
I	636 ± 2.5
II	203 ± 24

The foregoing examples are presented for purposes of illustration and are not intended to limit the scope of the

invention. Variations and changes which are obvious to one skilled in the art are intended to be within the scope and nature of the invention as defined in the appended claims.

We claim:

1. A compound having the name 6-O-methylerythromycin A form I exhibiting an exothermic transition at 132.2° C. and characterized by peaks in the powder x-ray diffraction at values of two theta of 5.16°±0.2, 6.68°±0.2, 10.20°±0.2, 12.28°±0.2, 14.20°±0.2, 15.40°±0.2, 15.72°±0.2, and 16.36°±0.2, or a pharmaceutically acceptable salt thereof.

2. A composition comprising a therapeutically effective amount of 6-O-methylerythromycin A form I exhibiting an exothermic transition at 132.2° C. in combination with a pharmaceutically acceptable carrier.

3. A method of treating bacterial infections in a host mammal in need of such treatment comprising administering to the mammal a therapeutically effective amount of 6-O-methylerythromycin A form I exhibiting an exothermic transition at 132.2° C.

4. A process for preparing 6-O-methylerythromycin A form I exhibiting an exothermic transition at 132.2° C. comprising

(a) converting erythromycin A to 6-O-methylerythromycin A;

(b) treating the 6-O-methylerythromycin A with a solvent selected from the group consisting of

(i) ethanol,

(ii) isopropyl acetate,

(iii) isopropanol,

(iv) tetrahydrofuran, and

(v) a mixture of a first solvent selected from the group consisting of ethanol, isopropyl acetate, isopropanol, and tetrahydrofuran and a second solvent selected from the group consisting of

a hydrocarbon of from 5 to 12 carbon atoms,

a ketone of from 3 to 12 carbon atoms,

a carboxylic ester of from 3 to 12 carbon atoms,

an ether of from 4 to 10 carbon atoms, benzene,

benzene substituted with one or more substituents selected from the group consisting of

alkyl of from one to four carbon atoms,

alkoxy of from one to four carbon atoms,

nitro, and

halogen, and

a polar aprotic solvent;

(c) isolating the crystalline 6-O-methylerythromycin A formed in step (b); and

(d) drying 6-O-methylerythromycin A isolate in step (c) at a temperature of between ambient temperature and about 70° C. to form 6-O-methylerythromycin A form I.

5. The process of claim 4 wherein the 6-O-methylerythromycin A is dried at a temperature of from about 40° C. to about 50° C.

6. The process of claim 4 wherein step (a) comprises

(i) converting erythromycin A into an erythromycin A 9-oxime derivative;

(ii) protecting the 2' and 4" hydroxy groups of the erythromycin A 9-oxime derivative prepared in step a;

(iii) reacting the product of step b with a methylating agent;

(iv) deprotecting and deoximating the product of step c to form 6-O-methylerythromycin A.

7. The process of claim 6 wherein the 6-O-methylerythromycin A is dried at a temperature of from about 40° C. to about 50° C.

8. A process for preparing 6-O-methylerythromycin A form I according to claim 7 wherein the solvent is selected from the group consisting of

(a) ethanol,

(b) isopropyl acetate,

(c) isopropanol, and

(d) tetrahydrofuran.

9. A process for preparing 6-O-methylerythromycin A form I according to claim 7 wherein the solvent is ethanol.

10. A process for preparing 6-O-methylerythromycin A form I according to claim 7 wherein the solvent comprises a mixture of a first solvent selected from the group consisting of ethanol, isopropyl acetate, isopropanol, and tetrahydrofuran and a second solvent selected from the group consisting of

a hydrocarbon of from 5 to 12 carbon atoms,

a ketone of from 3 to 12 carbon atoms,

a carboxylic ester of from 3 to 12 carbon atoms,

an ether of from 4 to 10 carbon atoms, benzene,

benzene substituted with one or more substituents selected from the group consisting of

alkyl of from one to four carbon atoms,

alkoxy of from one to four carbon atoms,

nitro, and

halogen, and

a polar aprotic solvent.

11. A process for preparing 6-O-methylerythromycin A form I according to claim 10 wherein the second solvent is a hydrocarbon from 5 to 12 carbon atoms.

12. A process for preparing 6-O-methylerythromycin A form I according to claim 11 wherein the second solvent is heptane.

13. A process for preparing 6-O-methylerythromycin A form I according to claim 7 wherein the solvent is selected from the group consisting of

(a) ethanol,

(b) isopropyl acetate,

(c) isopropanol,

(d) tetrahydrofuran,

(e) isopropyl acetate-heptane,

(f) isopropyl acetate-N,N-dimethylformamide,

(g) tetrahydrofuran-heptane,

(h) ethanol-heptane, and

(i) isopropanol-heptane

14. 6-O-methylerythromycin Form I prepared according to the method of claim 7.

* * * * *

Exhibit C

US005945405A

United States Patent [19]

Spanton et al.

[11] Patent Number: 5,945,405

[45] Date of Patent: Aug. 31, 1999

[54] CRYSTAL FORM O OF CLARITHROMYCIN

[75] Inventors: Stephen G. Spanton, Green Oaks; Rodger F. Henry, Waukegan, both of Ill.; David A. Riley, Kenosha, Wis.; Jih-Hua Liu, Green Oaks, Ill.

[73] Assignee: Abbott Laboratories, Abbott Park, Ill.

[21] Appl. No.: 08/785,623

[22] Filed: Jan. 17, 1997

[51] Int. Cl.[6] A61K 31/70; C07H 1.00; C07H 17/08

[52] U.S. Cl. 514/29; 536/7.2; 536/18.5

[58] Field of Search 536/7.2, 18.5; 514/29

[56] References Cited

U.S. PATENT DOCUMENTS

4,331,803	5/1982	Watanabe et al.	536/7.2
4,670,549	6/1987	Morimoto et al.	536/7.4
4,672,109	6/1987	Watanabe et al.	536/72
4,808,411	2/1989	Lu et al.	424/441
4,990,602	2/1991	Morimoto et al.	536/7.3
5,256,839	9/1996	Greene et al.	574/29

FOREIGN PATENT DOCUMENTS

B 260 938	9/1992	European Pat. Off. .
9804573	2/1998	WIPO .
9804574	2/1998	WIPO .

OTHER PUBLICATIONS

Acta Crystalographica Section C. Crystal Structure Communications, vol. C49, No. 5 (May 1993), pp. 1227–1230, Iwaska et al., "Structure of 6–O–Methylerythromycin A (Clarithromycin)".

Journal of Antibiotics, vol. XLII, No. 6 (Jun. 1989), pp. 1012–1014, Takashi Adachi et al., "Crystal and Molecular Structure of (14R)–14–Hydroxy–6–O–Methylerythromycin A".

Cremer et al., *Proceed. Intern. Symp. Control. Rel. Bioact. Mater.*, 1995, 22, 732–33

Primary Examiner—Elli Peselev

Attorney, Agent, or Firm—Portia Chen; Mona Anand

[57] ABSTRACT

The present invention concerns the novel antibiotic 6-O-methylerythromycin A form O solvate, a process for its preparation, pharmaceutical compositions comprising this compound and a method of use as a therapeutic agent.

25 Claims, No Drawings

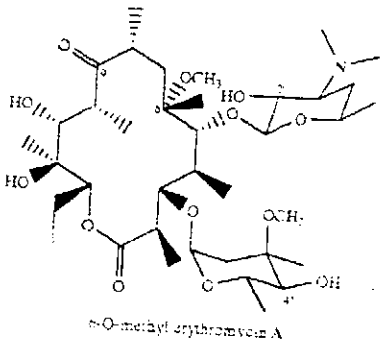
CRYSTAL FORM O OF CLARITHROMYCIN

TECHNICAL FIELD

This invention relates to a compound having therapeutic utility and to a method for its preparation. More particularly, the present invention concerns the novel compound 6-O-methylerythromycin A crystal form 0 solvate, a process for its preparation, pharmaceutical compositions comprising this compound and a method of use as a therapeutic agent.

BACKGROUND OF THE INVENTION

6-O-methylerythromycin A (Clarithromycin) is a semi-synthetic macrolide antibiotic of formula



6-O-methyl erythromycin A

Two distinct crystal forms of 6-O-methylerythromycin A, designated "form I" and "form II" have been identified. The crystal forms are identified by their single crystal or powder diffraction patterns.

6-O-methylerythromycin A exhibits excellent antibacterial activity against gram-positive bacteria, some gram-negative bacteria, anaerobic bacteria, Mycoplasma, and Chlamidia. It is stable under acidic conditions and is efficacious when administered orally. It is a useful therapy for infections of the upper respiratory tract in children and adults. 6-O-methylerythromycin A is available as tablets and as an oral suspension. Drugs currently on the market are formulated using the thermodynamically more stable 6-O-methylerythromycin A form II.

The oral suspension is particularly useful for patients such as children and the elderly who have difficulty swallowing the tablets. However, because 6-O-methylerythromycin A has such pronounced bitterness conventional approaches to taste masking failed to produce palatable suspensions. Ultimately, it was discovered that 6-O-methylerythromycin A-carbomer (acrylic acid copolymer) complexes provided particles sufficiently palatable for use in the oral suspension. See U.S. Pat. No. 4,808,411.

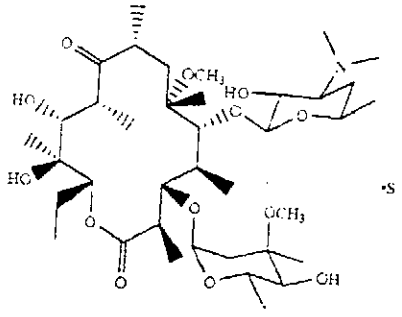
The 6-O-methylerythromycin A-carbomer complexes used in the oral suspension are prepared by dispersing 6-O-methylerythromycin A in an organic solvent, preferably ethanol, separately dispersing the carbomer in ethanol, mixing the two solutions to allow formation of the desired reaction product, evaporating most of the solvent and diluting the mixture with water to precipitate the carbomer-6-O-methylerythromycin A form 0 solvate complex.

SUMMARY OF THE INVENTION

6-O-methylerythromycin A can exist in a third crystal form, designated "form 0". Form 0, I, and II crystals have an identical spectrum of antibacterial activity. 6-O-methylerythromycin A prepared by the various methods described in the patent literature summarized below, in which the compound is purified by recrystallization from ethanol, result in initial formation of the crystalline form 0-ethanolate. Form 0 solvates are also formed with tetrahydrofuran, isopropanol, and isopropyl acetate. The form 0 solvate is converted to the non solvated form I by removing the solvent from the crystal lattice by drying at a temperature of from about 0° C. to about 50° C. Form 0 is converted to the non-solvated crystal form II by heating under vacuum at a temperature of between about 70° C. and 110° C.

The 6-O-methylerythromycin A-carbomer complexes described above are prepared using 6-O-methylerythromycin A form II. Substantial savings in energy and material handling could be realized by forming the carbomer complexes from 6-O-methylerythromycin A form 0 solvate, thereby eliminating the vacuum drying step required to prepare form II crystals. 6-O-methylerythromycin A form 0 solvate is also a useful intermediate in the preparation of the non-solvated 6-O-methylerythromycin A forms I and II.

Accordingly, the present invention in its principle embodiment provides a novel crystalline antibiotic designated 6-O-methylerythromycin A form 0 solvate having the structure



wherein S is a solvating molecule selected from the group consisting of ethanol, isopropyl acetate, isopropanol and tetrahydrofuran.

6-O-methylerythromycin A form 0 solvate is characterized by 2-theta angle positions in the powder x-ray diffraction pattern of 4.581°±0.2, 6.498°±0.2, 7.615°±0.2, 9.169°±0 2, 10.154°±0.2, 11.009°±0.2, 11.618°±0.2, 12.495°±0.2, 13.772°±0.2, 14.820°±0.2, 16.984°±0.2, 18.221°±0.2, 18.914°±0.2 and 19.495°±0.2.

In another embodiment, the present invention provides a composition comprising a therapeutically effective amount of 6-O-methylerythromycin A form 0 solvate in combination with a pharmaceutically acceptable carrier.

In yet another embodiment, the present invention provides a method of treating bacterial infections in a host mammal in need of such treatment comprising administering to the mammal a therapeutically effective amount of 6-O-methylerythromycin A form 0 solvate.

In yet another embodiment, the present invention provides a process for preparing 6-O-methylerythromycin A form 0 solvate comprising

(a) converting erythromycin A to 6-O-methylerythromycin A;

(b) treating the 6-O-methylerythromycin A with a solvent selected from the group consisting of (i) ethanol, (ii) isopropyl acetate, (iii) isopropanol, and (iv) tetrahydrofuran; and

(c) isolating the 6-O-methylerythromycin A form 0 solvate.

In yet another embodiment, the present invention provides 6-O-methylerythromycin A form 0-ethanolate prepared according to the foregoing process.

In yet another embodiment, the present invention provides 6-O-methylerythromycin A form 0-isopropyl acetate prepared according to the foregoing process.

In yet another embodiment, the present invention provides 6-O-methylerythromycin A form 0-tetrahydrofuran prepared according to the foregoing process.

In yet another embodiment, the present invention provides 6-O-methylerythromycin A form 0-isopropanolate prepared according to the foregoing process.

In yet another embodiment, the present invention provides a composition comprising from about 25% to about 95% of 6-O-methylerythromycin A form 0 solvate and from about 5% to about 75% of a carbomer.

In yet another embodiment, the present invention provides a method of treating bacterial infections in a host mammal in need of such treatment comprising administering to the mammal a therapeutically effective amount of 6-O-methylerythromycin A form 0 solvate-carbomer complex.

In yet another embodiment, the present invention provides a suspension for oral administration comprising 6-O-methylerythromycin form 0 solvate-carbomer complex suspended in a pharmaceutically acceptable liquid medium.

In yet another embodiment, the present invention provides a process for the preparation of 6-O-methylerythromycin A form 0 solvate-carbomer complex of from about 25% to about 95% of 6-O-methylerythromycin A form 0 solvate and from about 5% to about 75% of a carbomer comprising

(a) dispersing a carbomer in an organic solvent, and

(b) mixing the dispersion of step (a) with 6-O-methylerythromycin form 0 solvate to allow formation of the reaction product.

In yet another embodiment, the present invention provides 6-O-methylerythromycin A form 0 solvate-carbomer complex prepared according to the foregoing process.

In yet another embodiment, the present invention provides a process for the preparation of 6-O-methylerythromycin A form I comprising drying 6-O-methylerythromycin A form 0 solvate at a temperature of from about 0° C. to about 50° C.

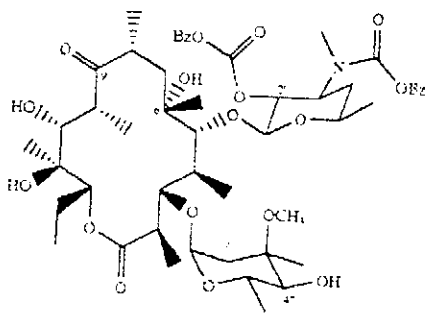
In yet another embodiment, the present invention provides a process for the preparation of 6-O-methylerythromycin A form II comprising heating 6-O-methylerythromycin A form 0 solvate under vacuum at a temperature of between about 70° C. and 110° C.

DETAILED DESCRIPTION

6-O-methylerythromycin A is prepared by methylation of the 6-hydroxy group of erythromycin A. However, in addition to the 6 position, erythromycin A contains hydroxy groups at the 11, 12, 2' and 4" positions, and a nitrogen at 3' position, all of which are potentially reactive with alkylating agents. Therefore, it is necessary to protect the various reactive functionalities prior to alkylation of the 6 hydroxy group. Representative 6-O-methylerythromycin A preparations are described in U.S. Pat. Nos. 4,331,803, 4,670,549, 4,672,109 and 4,990,602 and European Patent Specification 260 938 B1 which are incorporated herein by reference. Following final removal of the protecting groups, the 6-O-methylerythromycin A may exist as a solid, a semisolid, or a syrup containing residual solvents from the deprotection reactions, inorganic salts, and other impurities. 6-O-methylerythromycin A form 0 solvate may be crystallized directly from the syrup or semisolid using the solvents listed above. Alternatively, if the crude reaction product solidifies, the solid may be recrystallized from any of the solvents described above. Pure 6-O-methylerythromycin A form 0 solvate may also be obtained by recrystallizing form II or mixtures of form I and form II from any of the solvents described above. The term "6-O-methylerythromycin A" as used herein is meant to include 6-O-methylerythromycin A in any crystalline form or mixtures thereof, as well as amorphous solids, syrups, or semisolids comprising 6-O-methylerythromycin A in any state of purity.

The term "treating" refers to crystallizing or recrystallizing 6-O-methylerythromycin A as defined above from any of the solvents described above.

6-O-methylerythromycin A is prepared from erythromycin A by a variety of synthetic routes. In one method, erythromycin A is converted to 2'-O-3'-N-bis (benzyloxycarbonyl)-N-demethylerythromycin A (I).

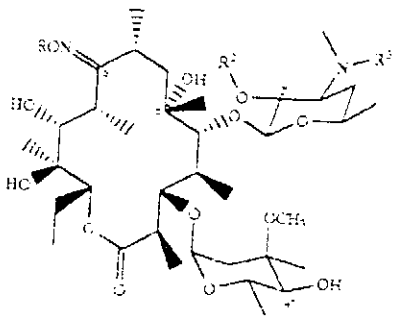


I

The 6-hydroxy group is then methylated by reaction with an alkylating agent such as bromomethane or iodomethane and a base. Removal of the benzoyl groups by catalytic hydrogenation and reductive methylation of the 3' N gives 6-O-methylerythromycin A. See U.S. Pat. No. 4,331,803.

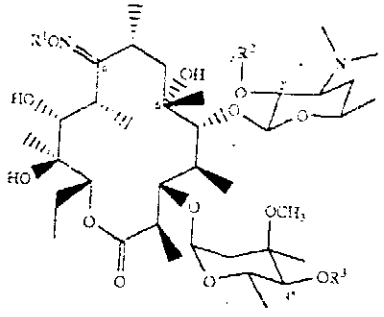
An alternative synthetic route involves methylation of 6-O-methylerythromycin A-9-oxime. 6-O-methylerythromycin A-9-oxime is prepared by methods well known in the art such as reaction of erythromycin A with hydroxylamine hydrochloride in the presence of base, or by reaction with hydroxylamine in the presence of acid as described in U.S. Pat. No. 5,274,085. Reaction of the oxime with RX wherein R is allyl or benzyl and X is halogen results in formation of 2'-O,3'-N-diallyl or dibenzylerythromycin A-9-O-allyl or benzyloxime halide. Methylation of this quaternary salt as described above, followed by elimination of the R groups and deoximination gives 6-O-methylerythromycin A. See U.S. Pat. No. 4,670,549.

Methylation of 6-O-methylerythromycin A oxime derivatives of formula II,



wherein R is alkyl, alkenyl, substituted or unsubstituted benzyl, oxyalkyl, or substituted phenylthioalkyl, R^2 is benzoyl, and R^3 is methyl or benzoyl, followed by deprotection, deoximation, and reductive methylation when R^3 is benzoyl gives 6-O-methylerythromycin A. See U.S. Pat. No. 4,672,109

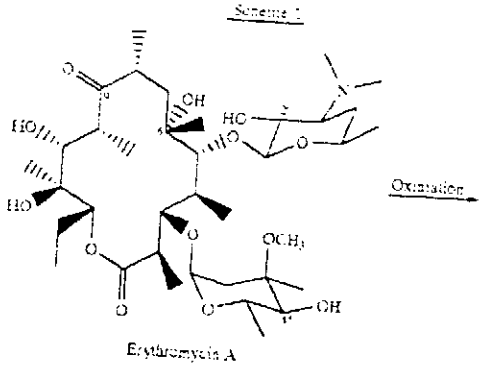
A particularly useful preparation of 6-O-methylerythromycin A involves methylation of the oxime derivative III.



wherein R^1 is alkenyl, substituted or unsubstituted benzyl, or alkoxyalkyl, R^2 is substituted silyl, and R^3 is R^2 or H. Removal of the protecting groups and deoximation is then accomplished in a single step by treatment with acid to give 6-O-methylerythromycin A. See European Patent Specification 260 938 B1 and U.S. Pat. No. 4,990,602.

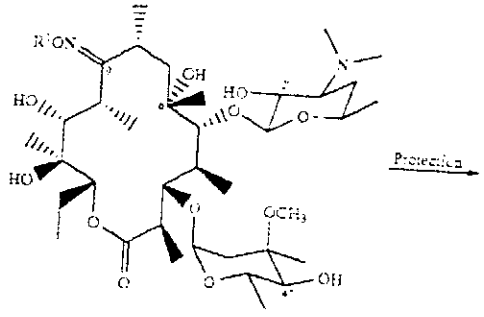
A preferred route of 6-O-methylerythromycin A is outlined in Scheme 1. Erythromycin A, prepared by fermentation of *Streptomyces erythreus* is oximated to give oxime 4 wherein R^1 is alkoxyalkyl. The group R^1 may be introduced by reaction of erythromycin A with the substituted hydroxylamine R^1ONH_2, or by reaction of erythromycin A with hydroxylamine hydrochloride in the presence of base, or hydroxylamine in the presence of acid, followed by reaction with R^1X. The two hydroxy groups are then protected simultaneously, in which R^2 or R^3 are the same, or sequentially in which R^2 and R^3 are different. Particularly useful protecting groups are substituted silyl groups such as trimethylsilyl, tert-butyldimethylsilyl, tert-butyldiphenylsilyl and the like. The protecting groups are then removed and the compound is deoximated to produce 6-O-methylerythromycin A. The order of deprotection deoxmation is not critical. When the protecting groups are substituted silyl, deprotection and deoximation can be accomplished in a single step by treatment with acid, for example using formic acid or sodium hydrogen sulfite. See U.S. Pat. No. 4,990,602.

Scheme 1



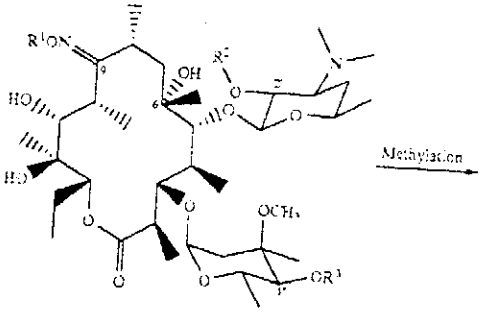
Erythromycin A

Oximation



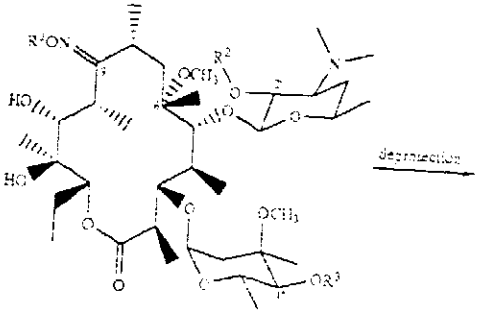
IV

Protection



V

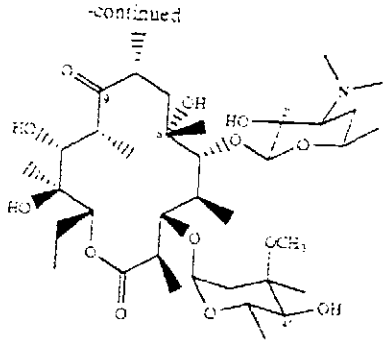
Methylation



VI

deprotection

-continued



6-O-methylerythromycin A

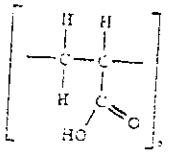
In accordance with the process aspects of the present invention, 6-O-methylerythromycin A prepared by any of the methods described above is suspended in the desired solvent and heated to about the reflux temperature of the solvent. Heating is then continued and the suspension is stirred for an amount of time sufficient to dissolve most of the solid, generally about 10 minutes to 2 hours. The suspension is then filtered hot. If necessary, the filtrate may be heated to at or about the reflux temperature of the solvent to form a clear solution. The filtrate is then slowly cooled to ambient temperature with optional further cooling in an ice-water bath. For purposes of this specification, ambient temperature is from about 20 to about 25° C. 6-O-methylerythromycin A form 0 solvate is then isolated, preferably by filtration, and the wet crystals are transferred to a sealed container.

The most preferred solvent for the isolation of 6-O-methylerythromycin A form 0 solvate is ethanol.

6-O-methylerythromycin A form 0 solvate-carbomer complexes are prepared by dispersing from about 5% to about 75% by weight of a carbomer in an organic solvent and mixing the dispersion with from about 95% to about 5% of 6-O-methylerythromycin A form 0 solvate. A preferred organic solvent is acetone. The mixture is then stirred for a period of time sufficient to allow formation of the antibiotic-carbomer complex, generally from about 0.5 to about 12 hours. The solid complex is then isolated, preferably by filtration. If necessary, water may be added to the mixture to promote precipitation of the complex. The collected precipitate is then dried and milled to the desired particle size by conventional methods.

Alternatively, the carbomer complex may be prepared by mixing 6-O-methylerythromycin A form 0 solvate and the dry carbomer in a limited amount of organic solvent. The solvent is then removed by evaporation, thereby eliminating the filtration step.

The carbomers employed in the foregoing process are branched acrylic acid polymers with a high degree of cross linking and thickening capacity. They have the general formula



where n is from about 10,000 to about 60,000. The average equivalent weight is 76, while the molecular weight is approximately 3 million. A preferred material is in the U.S. Pharmacopoeia as Carbomer 934P. This carbomer is classified as a water soluble resin and has been used in other pharmaceutical compositions for its thickening and suspending properties. In its presolvated state, the carbomer is a tightly coiled molecule and its thickening properties are limited. However, due to its relatively high molecular weight and extensive resin cross linking, the carbomer can generate a high viscosity gel. This gelation is initially achieved to occur as a result of hydration and partial uncoiling. Neutralization of the acidic groups of the carbomer with a suitable base organic or inorganic base is required to further uncoil the molecule and generate high viscosity solutions.

The term "6-O-methylerythromycin A form 0 solvate-carbomer complex or granule" refers to the product obtained in the above process. While not intending to be limited by theory, the granule is believed to be held together by both ionic attraction between the amino group of 6-O-methylerythromycin A form 0 solvate and the carboxyl group of the carbomer, and the gel properties of the carbomer.

The antibiotic-carbomer complexes of this invention can be employed in dry form, preferably in the form of particles. Such particles can be mixed with foods or beverages, can be used to prepare liquid suspensions for oral administration, or can be formed into conventional whole or chewable tablets for oral administration.

In such solid dosage forms, the antibiotic-carbomer complex is mixed with at least one inert, pharmaceutically acceptable excipient or carrier such as sodium citrate or dicalcium phosphate and/or a) fillers or extenders such as starches, lactose, sucrose, glucose, mannitol, and silicic acid, b) binders such as, for example, carboxymethylcellulose, alginates, gelatin, polyvinylpyrrolidone, sucrose, and acacia, c) humectants such as glycerol, d) disintegrating agents such as agar-agar, calcium carbonate, potato or tapioca starch, alginic acid, certain silicates, and sodium carbonate, e) solution retarding agents such as paraffin, f) absorption accelerators such as quaternary ammonium compounds, g) wetting agents such as, for example, cetyl alcohol and glycerol monostearate, h) absorbents such as kaolin and bentonite clay, and i) lubricants such as talc, calcium stearate, magnesium stearate, solid polyethylene glycols, sodium lauryl sulfate, and mixtures thereof. The dosage form may also comprise buffering agents.

Solid compositions of a similar type may also be employed as fillers in soft and hard-filled gelatin capsules using such excipients as lactose or milk sugar as well as high molecular weight polyethylene glycols and the like.

The solid dosage forms can be prepared with coatings and shells such as enteric coatings and other coatings well known in the pharmaceutical formulating art. They may optionally contain opacifying agents and can also be of a composition that they release the active ingredient(s) only, or preferentially, in a certain part of the intestinal tract, optionally, in a delayed manner. Examples of embedding compositions which can be used include polymeric substances and waxes.

The active compounds can also be in micro-encapsulated form, if appropriate, with one or more of the above-mentioned excipients.

Suspensions for oral administration may contain, in addition to the antibiotic-carbomer complex, inert diluents commonly used in the art such as, for example, water or other solvents, solubilizing agents and emulsifiers such as ethyl alcohol, isopropyl alcohol, ethyl carbonate, ethyl acetate, benzyl alcohol, benzyl benzoate, propylene glycol, 1,3-

butylene glycol, dimethyl formamide, oils (in particular, cottonseed, groundnut, corn, germ, olive, castor, and sesame oils), glycerol, tetrahydrofurfuryl alcohol, polyethylene glycols and fatty acid esters of sorbitan, and mixtures thereof.

Besides inert diluents, the oral compositions can also include adjuvants such as wetting agents, emulsifying and suspending agents, sweetening, flavoring, and perfuming agents.

Suspensions, in addition to the active compounds, may contain suspending agents as, for example, ethoxylated isostearyl alcohols, polyoxyethylene sorbitol and sorbitan esters, microcrystalline cellulose, aluminum metahydroxide, bentonite, agar-agar, and tragacanth, and mixtures thereof.

Preferably, fine particles having average diameters smaller than 40 mesh (420 microns) will be employed. For use in a pediatric suspension a mean particle diameter of less than 50 mesh (297 microns) will be desirable. In some products, the particles will be larger, having a mean diameter of less than 10 mesh (2000 microns), or more preferably less than 1000 microns (about 16 mesh).

To further reduce dissolution of the active drug in the mouth, the complexes provided in accordance with the present invention can be polymer coated. A variety of polymeric materials can be employed. Non-limiting examples of such materials include ethyl cellulose, hydroxypropyl cellulose, hydroxypropylmethyl cellulose, polyvinyl acetate phthalate, cellulose acetate phthalate, hydroxypropylmethyl cellulose phthalate and shellac, as well as numerous other polymers familiar to those of ordinary skill in the pharmaceutical arts. Such other polymers commonly known by tradenames include Eudragit® E-100, S-100 and L-100 polymers, available from the Rohm and Haas Company. Most preferable is hydroxypropylmethyl cellulose phthalate.

The use of pH sensitive coatings offer advantages in addition to taste coverage. A coating insoluble at neutral pH, but soluble in acid (e.g. Eudragit® E-100) can give complete taste coverage in the neutral pH of the mouth, while still allowing rapid dissolution in the strongly acidic stomach contents after swallowing. Conversely, an enteric coating can be insoluble in acid or water while dissolving rapidly in a neutral buffer above pH 5 or 6. This offers the opportunity to prepare a suspension of antibiotic-carbomer complex that remains intact in the formulation but rapidly releases the antibiotic in the intestine. The drug thereby remains protected from the hostile environment of the stomach, but are rapidly dissolved in the higher pH of the intestinal tract.

Actual dosage levels of active ingredients in the pharmaceutical compositions of this invention may be varied so as to obtain an amount of the active compound(s) that is effective to achieve the desired therapeutic response for a particular patient, compositions, and mode of administration. The selected dosage level will depend upon the activity of the particular compound, the route of administration, the severity of the condition being treated, and the condition and prior medical history of the patient being treated. However, it is within the skill of the art to start doses of the compound at levels lower than required for to achieve the desired therapeutic effect and to gradually increase the dosage until the desired effect is achieved.

Generally dosage levels of about 1 to about 1000, more preferably of about 5 to about 200 mg of 6-O-methylerythromysin A form 0 solvate per kilogram of body weight per day are administered to a mammalian patient If desired, the effective daily dose may be divided into multiple doses for purposes of administration, e.g. two to four separate doses per day.

The following Examples are provided to enable one skilled in the art to practice the invention and are merely illustrative of the invention. They should not be read as limiting the scope of the invention as defined in the claims.

EXAMPLE 1

Preparation of 6-O-methylerythromycin Form 0.Ethanolate

6-O-methylerythromycin A was prepared from erythromycin A by oximation of the C-9 carbonyl, protection of the C-2' and C-4" hydroxy groups, methylation of the C-6 hydroxy group, deoximation and removal of the protecting groups, and recrystallization from ethanol according to the method of U.S. Pat. No. 4,990,602.

A mixture of 6-O-methylerythromycin A (20 g), prepared as described above, and ethanol (200 mL) was warmed to reflux and the insoluble material (11.2 g) was removed by filtration. The filtrate was transferred to a clean flask and heated to reflux. The clear solution was allowed to cool to ambient temperature and then was further cooled in an ice bath. The liquid was decanted to leave 6-O-methylerythromycin A form 0 ethanolate which was sealed in a container without further drying. The 2-theta angle positions in the single crystal x-ray diffraction pattern of 6-O-methylerythromycin A form 0-ethanolate are 4.72°±0.2, 6.60°±0.2, 7.72°±0.2, 9.30°±0.2, 10.40°±0.2, 11.10°±0.2, 11.86°±0.2, 12.72°±0.2, 13.90°±0.2, 15.02°±0.2, 17.18°±0.2, 18.50°±0.2, 19.08°±0.2, 19.68°±0.2, 23.14°±0.2 and 23.98°±0.2.

EXAMPLE 2

Preparation of 6-O-methylerythromycin Form 0.Isopropyl Acetate

A mixture of 6-O-methylerythromycin A (10 g), prepared as described in Example 1, and isopropyl acetate (100 mL) was warmed to 73° C. The hot solution was filtered to remove traces of insoluble material. The clear solution was then cooled slowly to ambient temperature. The liquid was decanted and the wet solid sealed in a container without further drying.

The 2-theta angle positions in the single crystal x-ray diffraction pattern of 6-O-methylerythromycin A form 0.Isopropyl Acetate are 4.76°±0.2, 6.70°±0.2, 7.80°±0.2, 9.128°±0.2, 10.56°±0.2, 11.96°±0.2, 12.24°±0.2, 12.36°±0.2, 12.60°±0.2, 12.84°±0.2, 13.96°±0.2, 15.16°±0.2, 16.68°±0.2, 17.28°±0.2, 18.52°±0.2, 19.18°±0.2, 19.80°±0.2, 20.56°±0.2, 21.52°±0.2 and 23.96°±0.2.

EXAMPLE 3

Preparation of 6-O-methylerythromycin Form 0.Tetrahydrofuran

A mixture of 6-O-methylerythromycin A (10 g), prepared as described in Example 1, and tetrahydrofuran (20 mL) was warmed to 50° C. The hot solution was filtered and the filtrate was cooled slowly to ambient temperture. The liquid was decanted and the wet solid sealed in a container without further drying.

EXAMPLE 4

Preparation of 6-O-methylerythromycin Form 0.isopropanolate

A mixture of 6-O-methylerythromycin A (5 g), prepared as described in Example 1, and isopropanol (20 mL) was

warmed to 60° C. The hot solution was gravity filtered to give 10 mL of clear filtrate which was cooled slowly to ambient temperature. The liquid was decanted and the wet solid sealed in a container without further drying.

EXAMPLE 5

Conversion of 6-O-methylerythromycin A form 0 solvate to 6-O-methylerythromycin A form I.

6-O-methylerythromycin A form 0 solvate prepared as in Examples 1–4 is dried in a vacuum oven (40–45° C., 4–8 in. Hg) to give 6-O-methylerythromycin A form I. The 2-theta angle positions in the powder x-ray diffraction pattern of 6-O-methylerythromycin A form I are 5.10°±0.2, 6.68°±0.2, 10.20°±0.2, 12.28°±0.2, 14.20°±0.2, 15.40°±0.2, 15.72°±0.2, and 16.36°±0.2.

EXAMPLE 6

Conversion of 6-O-methylerythromycin A form 0 solvate to 6-O-methylerythromycin A form II.

6-O-methylerythromycin A form 0 solvate, prepared as in Examples 1–4, is placed in a vial and heated in the vacuum oven (4–9 in Hg, 100–110° C.) for 18 hours to give 6-O-methylerythromycin A form II crystals. 6-O-methylerythromycin A form II melts at 223.4° C. The 2-theta angle positions in the powder x-ray diffraction pattern of 6-O-methylerythromycin A form II are 8.52°±0.2, 9.48°±0.2, 10.84°±0.2, 11.48°±0.2, 11.88°±0.2, 12.36°±0.2, 13.72°±0.2, 14.12°±0.2, 15.16°±0.2, 16.48°±0.2, 16.92°±0.2, 17.32°±0.2, 18.08°±0.2, 18.40°±0.2, 19.04°±0.2, 19.88°±0.2, and 20.48°±0.2.

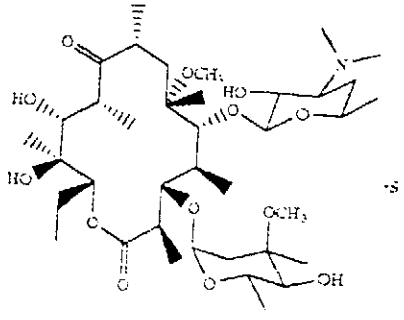
EXAMPLE 7

Preparation of 6-O-methylerythromycin A form 0-carbomer complex

The desired complex is prepared by stirring a mixture in acetone of about 1.5 parts by weight of 6-O-methylerythromycin form 0.ethanolate and 1 part by weight of Carbomer 934P until the mixture is uniform. Water is then added with stirring and the resulting precipitate is stirred for about 30 minutes. The solids are separated by vacuum filtration and washed with water. The damp filter cake is then passed through a 30 mesh screen and dried in a vacuum oven at about 40° C. The potency of the complex is determined by colorimetric analysis.

We claim:

1. An isolated crystalline antibiotic designated 6-O-methylerythromycin A form 0 solvate having the structure



·S

wherein S is a solvating molecule selected from the group consisting of ethanol, isopropyl acetate, isopropanol and tetrahydrofuran.

2. 6-O-methylerythromycin A form 0 solvate according to claim 1 characterized by 2-theta angle positions in the powder x-ray diffraction pattern of 4.581°±0.2, 6.498°±0.2, 7.315°±0.2, 9.169°±0.2, 10.154°±0.2, 11.009°±0.2, 11.618°±0.2, 12.495°±0.2, 13.772°±0.2, 14.820°±0.2, 16.984°±0.2, 18.221°±0.2, 18.914°±0.2 and 19.495°±0.2.

3. A crystalline antibiotic according to claim 1 having the name 6-O-methylerythromycin A form 0.ethanolate characterized by 2-theta angle positions in the powder x-ray diffraction pattern of 4.72°±0.2, 6.50°±0.2, 7.72°±0.2, 9.30°±0.2, 10.40°±0.2, 11.10°±0.2, 11.86°±0.2, 12.72°±0.2, 13.90°±0.2, 15.02°±0.2, 17.18°±0.2, 18.50°±0.2, 19.08°±0.2, 19.68°±0.2, 23.14°±0.2 and 23.98°±0.2.

4. A crystalline antibiotic according to claim 1 having the name 6-O-methylerythromycin A form 0.isopropyl acetate characterized by 2-theta angle positions in the powder x-ray diffraction pattern of 4.76°±0.2, 6.70°±0.2, 7.80°±0.2, 9.128°±0.2, 10.56°±0.2, 11.96°±0.2, 12.24°±0.2, 12.36°±0.2, 12.60°±0.2, 12.84°±0.2, 13.96°±0.2, 15.16°±0.2, 16.68°±0.2, 17.28°±0.2, 18.52°±0.2, 19.18°±0.2, 19.80°±0.2, 20.56°±0.2, 21.52°±0.2 and 23.96°±0.2.

5. A crystalline antibiotic according to claim 1 having the name 6-O-methylerythromycin A form 0.tetrahydrofuran.

6. A crystalline antibiotic according to claim 1 having the name 6-O-methylerythromycin A form 0.isopropanolate.

7. A composition comprising a therapeutically effective amount of an isolated 6-O-methylerythromycin A form 0 solvate in combination with a pharmaceutically acceptable carrier.

8. A method of treating bacterial infections in a host mammal in need of such treatment comprising administering to the mammal a therapeutically effective amount of an isolated 6-O-methylerythromycin A form 0 solvate.

9. A process for preparing 6-O-methylerythromycin A form 0 solvate comprising

(a) converting erythromycin A to 6-O-methylerythromycin A;

(b) treating the 6-O-methylerythromycin prepared in step a with a solvent selected from the group consisting of ethanol, isopropyl acetate, isopropanol, and tetrahydrofuran; and

(c) isolating the 6-O-methylerythromycin form 0 solvate crystals.

10. The process of claim **9** wherein step (a) comprises

(i) converting erythromycin A into an erythromycin A 9-oxime derivative;

(ii) protecting the 2' and 4" hydroxy groups of the erythromycin A 9-oxime derivative prepared in step i;

(iii) reacting the product of step ii with a methylating agent;

(iv) deprotecting and deoximating the product of step iii to form 6-O-methylerythromycin A.

11. The process of claim **10** wherein the solvent is ethanol.

12. 6-O-methylerythromycin form 0-ethanolate prepared according to the process of claim **11**.

13. The process of claim **10** wherein the solvent is isopropyl acetate.

14. 6-O-methylerythromycin form 0.isopropyl acetate prepared according to the process of claim **13**.

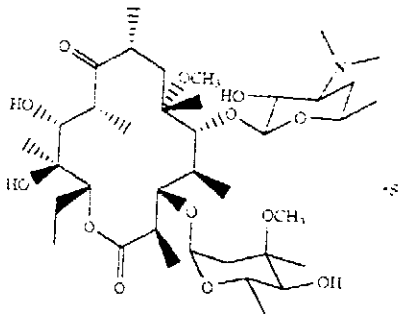
15. The process of claim **10** wherein the solvent is isopropanol.

16. 6-O-methylerythromycin form 0.isopropanolate prepared according to the process of claim **15**.

17. The process of claim **10** wherein the solvent is tetrahydrofuran.

18. 6-O-methylerythromycin form 0.tetrahydrofuran prepared according to the process of claim 17.

19. A complex comprising from about 25% to about 95% of an isolated crystalline antibiotic designated 6-O-methylerythromycin A form 0 solvate having the structure

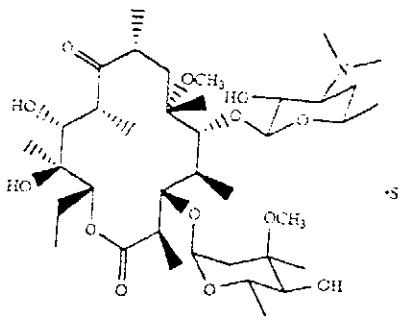


wherein S is a solvating molecule selected from the group consisting of ethanol, isopropyl acetate, isopropanol and tetrahydrofuran and from about 5% to about 75% of a carbomer.

20. A method of treating bacterial infections in a host mammal in need of such treatment comprising administering to the mammal a therapeutically effective amount of the complex of claim 19.

21. A suspension for oral administration comprising the complex of claim 19 suspended in a pharmaceutically acceptable inert diluent.

22. A process for the preparation of 6-O-methylerythromycin A form 0 solvate-carbomer complex of from about 25% to about 95% of an isolated crystalline antibiotic designated 6-O-methylerythromycin A form 0 solvate having the structure



wherein S is a solvating molecule selected from the group consisting of ethanol, isopropyl acetate, isopropanol and tetrahydrofuran and from about 5% to about 75% of a carbomer comprising

(a) dispersing a carbomer in an organic solvent; and

(b) mixing the dispersion of step (a) with 6-O-methylerythromycin form 0 solvate to allow formation of the reaction product.

23. 6-O-methylerythromycin A form 0 solvate-carbomer complex prepared according to the process of claim 22.

24. A process for the preparation of 6-O-methylerythromycin A form I comprising drying an isolated 6-O-methylerythromycin A form 0 solvate at a temperature of from about 0° C. to about 50° C.

25. A process for the preparation of 6-O-methylerythromycin A form II comprising heating an isolated 6-O-methylerythromycin A form 0 solvate under vacuum at a temperature of between about 70° C. and 110° C.

* * * * *