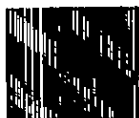


Industria y Comercio
SUPERINTENDENCIA



03 047585 00

SUPERINDUSTRIA Y COMERCIO

Radicación : 03047585 00000000

Folios: 114

Fecha (ARD): 2003-06-05 16:09:23

Trámite : 011 PCT-PATENT 379 FASE NACI 411 PRESENTAC

Dependencia: 2020 DIVISION DE NUEVAS CREACIONES

**FORMULARIO ÚNICO DE SOLICITUD DE PATENTE
PCT
ENTRADA EN FASE NACIONAL**

SOLICITUD DE:



Patente de Invención



Patente de Modelo de Utilidad



Capítulo I



Capítulo II

2

**SOLICITANTE
(71)**

Nombre: **NOVARTIS AG**

Dirección: Lichtstrasse 35

Nacionalidad o Domicilio: CH -4056 Basilea,
Suiza.

Lugar de Constitución : Suiza

Teléfono: 41613241111 Fax: 41613227532

E-mail

IDENTIFICACIÓN

C.C. ☐

NIT ☐

C.E. ☐

Otro ☐

Número

3

**REPRESENTANTE
O APODERADO
(74)**

Nombre: JIMENA ESCOBAR URIBE

Dirección: Cra. 11A No. 94-76 (Of. 305)

Teléfono: 6 162051/61 Fax: 6 161889

E-mail: tm-patent@escobaruribe.com

IDENTIFICACIÓN

C.C. ☒

NIT ☐

C.E. ☐

Otro ☐

Número 39.779.452T.P 68228

4

**INVENTOR (ES)
(72)**

Nombre: **VER ANEXOS**

Dirección:

Nacionalidad o domicilio:

5

PCT (86)

Solicitud Internacional No. PCT/EP01/14771

Fecha Diciembre 8, 2001

Publicación Internacional No. WO 02/40186 A1

Fecha Junio 13, 2002

6 TÍTULO (84) PROCESO PARA EL AISLAMIENTO Y PURIFICACIÓN DE EPOTILONAS.

7

Clasificación Internacional (51)

C 07 D 493 / 04

A61K 31 / 335



8

Prioridad

SI



NO



(33) País de Origen

REINO UNIDO

(32) Fecha

Diciembre 7, 2000

(31) Número de Solicitud

0029895.0

9

Comprobante de pago No. 03 - 32 228

Fecha Mayo 9, 2003

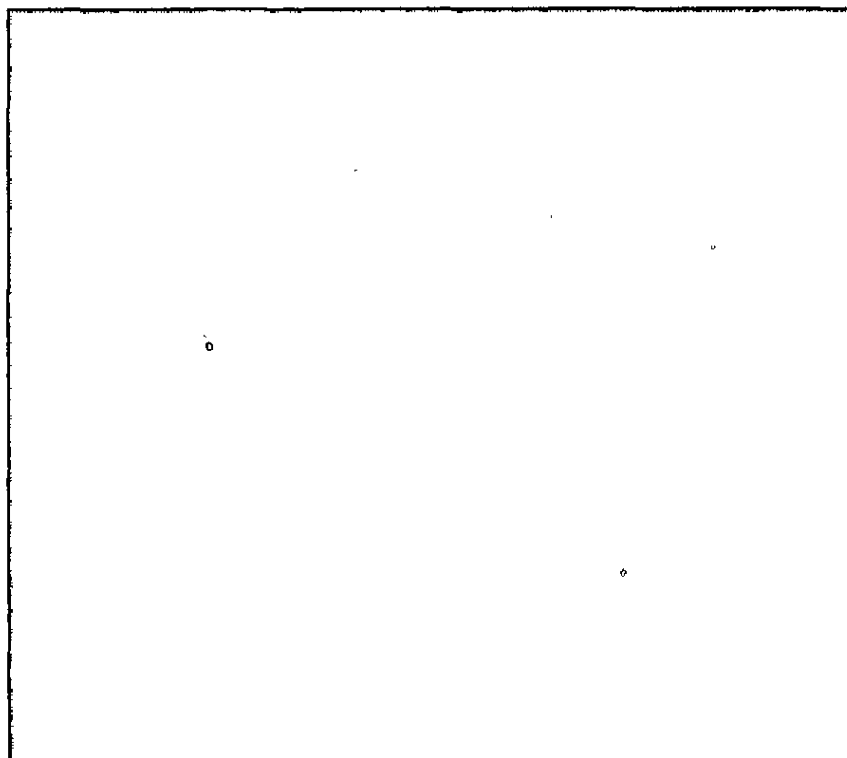
10

ANEXOS

- ☐ Comprobante de pago de la tasa de presentación de la solicitud
- ☐ Documento que acredita la existencia y representación legal cuando el solicitante sea persona
- ☐ Prot. 17.398
- ☐ Poderes si fuere el caso **Protocolo No. 17.398**
- ☐ Documento de cesión del inventor al solicitante o a su causante
- ☐ Declaraciones
- ☐ Copia de la solicitud internacional. (Resumen, descripción, reivindicaciones, dibujos)
- ☐ Traducción de la solicitud internacional al castellano. (Resumen, descripción, reivindicaciones, dibujos)
- ☐ Copia del examen preliminar internacional efectuado por la Autoridad Internacional de Examen Preliminar (IPEA).
- ☐ Traducción del examen preliminar internacional efectuado por la IPEA
- ☐ De ser el caso copia de contrato de acceso.
- ☐ De ser el caso, documento que acredite la licencia o autorización de uso de conocimientos tradicionales de las comunidades indígenas
- ☐ De ser el caso, certificado de depósito del material biológico
- ☐ Arte final 12 X 12 .
- ☐ De ser el caso, información sobre otras solicitudes de patente o títulos obtenidos en el extranjero por el mismo titular o su causante, relacionadas parcial o totalmente con la invención de esta solicitud.
- ☐ De ser el caso, modificaciones efectuadas a la solicitud internacional

Otros

11 FIGURA CARACTERÍSTICA



12 Solicito la concesión de la patente .

NOMBRE: JIMENA ESCOBAR URIBEFIRMA: 

G.C. 30.779.452 DE Usaquén T.P. 68.228

As. 2562/PCT

2562

SUPERINDUSTRIA Y COMERCIO
 Radicacion : 03047585 00000000 Folios: 114
 Fecha (AMD): 2003-06-05 16:09:23
 Trámite : 011 PCT-PATENT 379 FASE MACI 411 PRESENTAC
 Dependencia: 2020 DIVISION DE NUEVAS CREACIONES

MIT : B00176089-2

RECIBO OFICIAL DE CAJA : 03 - 32,228
 FECHA : MAYO 9 DE 2003

***** CONSIGNACION *****

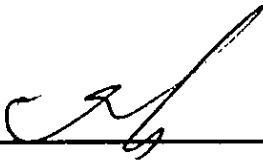
DEPOSITANTE	TIPO PAGO	BANCO	CUENTA	No. PAGO	FECHA PAGO	Vr. PAGO
ESCOBAR URIBE ASOCIADOS	CONSIGNACION	BANCO POPULAR	030-00110-6	9935764	08/05/2003	4,550,000.00

***** CONCEPTO *****

CANT. RENTISTICO	CONCEPTO	TOTAL CONCEPTO
1 50003-01-01 SOLICITUDES	1 TRAMITES DE SOL. DE PATENTE DE IN	400,000.00
	TOTAL :	\$ 400,000.00

SOM: CUATROCIENTOS MIL PESOS

RESPONSABLE :



RECIBO DE CAJA APLICADO AL EXPEDIENTE No. _____

ANEXOS**INVENTORES****1.)****ERNST KÜSTERS
(79427 Eschbach, Alemania);****2.)****HEINZ UNTERNÄHRER
(4303 Kaiseraugst, Suiza).**

4

Solicito expresamente a nombre de la sociedad
NOVARTIS AG., sociedad constituida de
conformidad con las leyes de Suiza, domiciliada en
Lichtstrasse 35, CH -4056 Basilea, Suiza el
otorgamiento de Patente para la invención titulada
**"PROCESO PARA EL AISLAMIENTO Y
PURIFICACIÓN DE EPOTILONAS".**

Clase: C 07


JIMENA ESCOBAR URIBE
C.C. 39.779.452 de Usaquen
T.P. 68.228

PATENT COOPERATION TREATY

16

5

From the RECEIVING OFFICE

P + TM Dept.

PCT

PR

To:

Becker, Konrad
NOVARTIS AG
Corporate Intellectual Property
Patent & Trademark Department
CH-4002 Basel
SUISSE

- 7. Feb. 2002

NOTIFICATION OF THE INTERNATIONAL
APPLICATION NUMBER AND OF THE
INTERNATIONAL FILING DATE

(PCT Rule 20.5(c))

Date of mailing
(day/month/year)

05. 02. 2002

Applicant's or agent's file reference

4-31672A

IMPORTANT NOTIFICATION

International application No.

PCT/EP 01/14771

International filing date (day/month/year)

05/12/2001

Priority date (day/month/year)

07/12/2000 ✓

Applicant

NOVARTIS AG

Title of the invention

1. The applicant is hereby notified that the international application has been accorded the international application number and the international filing date indicated above.
2. The applicant is further notified that the record copy of the international application was transmitted to the International Bureau on the above date of mailing.

3.

☐

Other: _____

* The International Bureau monitors the transmittal of the record copy by the receiving Office and will notify the applicant (with Form PCT/IB/301) of its receipt. Should the record copy not have been received by the expiration of 14 months from the priority date, the International Bureau will notify the applicant (Rule 22.1(c)).

Name and mailing address of the Receiving Office



European Patent Office, P.B. 5818 Patentlaan 2
NL-2280 HV Rijswijk
Tel. (+ 31-70) 340-2040
Fax: (+ 31-70) 340-3016

Authorized officer

N. MAILLIARD

Tel.: (070) 340.28.55
The Hague

N. Mailliard

Sheet No. ...2...

Continuation of Box No. III FURTHER APPLICANT(S) AND/OR (FURTHER) INVENTOR(S)	
<i>If none of the following sub-boxes is used, this sheet should not be included in the request.</i>	
Name and address: (Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country. The country of the address indicated in this box is the applicant's State (that is, country) of residence if no State of residence is indicated below.) KÜSTERS, Ernst Gallenweilerweg 1 79427 Eschbach DE	This person is: ☐ applicant only ☒ applicant and inventor ☐ inventor only (If this check-box is marked, do not fill in below.) Applicant's registration No. with the Office
State (that is, country) of nationality: DE	State (that is, country) of residence: DE
This person is applicant for the purposes of: ☐ all designated States ☐ all designated States except the United States of America ☒ the United States of America only ☐ the States indicated in the Supplemental Box	
Name and address: (Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country. The country of the address indicated in this box is the applicant's State (that is, country) of residence if no State of residence is indicated below.) UNTERNÄHRER, Heinz Widhagweg 46 4303 Kaiseraugst CH	This person is: ☐ applicant only ☒ applicant and inventor ☐ inventor only (If this check-box is marked, do not fill in below.) Applicant's registration No. with the Office
State (that is, country) of nationality: CH	State (that is, country) of residence: CH
This person is applicant for the purposes of: ☐ all designated States ☐ all designated States except the United States of America ☒ the United States of America only ☐ the States indicated in the Supplemental Box	
Name and address: (Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country. The country of the address indicated in this box is the applicant's State (that is, country) of residence if no State of residence is indicated below.)	This person is: ☐ applicant only ☐ applicant and inventor ☐ inventor only (If this check-box is marked, do not fill in below.) Applicant's registration No. with the Office
State (that is, country) of nationality:	State (that is, country) of residence:
This person is applicant for the purposes of: ☐ all designated States ☐ all designated States except the United States of America ☐ the United States of America only ☐ the States indicated in the Supplemental Box	
Name and address: (Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country. The country of the address indicated in this box is the applicant's State (that is, country) of residence if no State of residence is indicated below.)	This person is: ☐ applicant only ☐ applicant and inventor ☐ inventor only (If this check-box is marked, do not fill in below.) Applicant's registration No. with the Office
State (that is, country) of nationality:	State (that is, country) of residence:
This person is applicant for the purposes of: ☐ all designated States ☐ all designated States except the United States of America ☐ the United States of America only ☐ the States indicated in the Supplemental Box	
☐ Further applicants and/or (further) inventors are indicated on another continuation sheet.	

Sheet No. ...3...

Box No.V DESIGNATION OF STATES

Mark the applicable check-boxes below; at least one must be marked.

The following designations are hereby made under Rule 4.9(a):

Regional Patent

- ☐ AP ARIPO Patent: GH Ghana, GM Gambia, KE Kenya, LS Lesotho, MW Malawi, MZ Mozambique, SD Sudan, SL Sierra Leone, SZ Swaziland, TZ United Republic of Tanzania, UG Uganda, ZW Zimbabwe, and any other State which is a Contracting State of the Harare Protocol and of the PCT
- ☒ EA Eurasian Patent: AM Armenia, AZ Azerbaijan, BY Belarus, KG Kyrgyzstan, KZ Kazakhstan, MD Republic of Moldova, RU Russian Federation, TJ Tajikistan, TM Turkmenistan, and any other State which is a Contracting State of the Eurasian Patent Convention and of the PCT
- ☒ EP European Patent: AT Austria, BE Belgium, CH & LI Switzerland and Liechtenstein, CY Cyprus, DE Germany, DK Denmark, ES Spain, FI Finland, FR France, GB United Kingdom, GR Greece, IE Ireland, IT Italy, LU Luxembourg, MC Monaco, NL Netherlands, PT Portugal, SE Sweden, TR Turkey, and any other State which is a Contracting State of the European Patent Convention and of the PCT
- ☐ OA OAPI Patent: BF Burkina Faso, BJ Benin, CF Central African Republic, CG Congo, CI Côte d'Ivoire, CM Cameroon, GA Gabon, GN Guinea, GW Guinea-Bissau, ML Mali, MR Mauritania, NE Niger, SN Senegal, TD Chad, TG Togo, and any other State which is a member State of OAPI and a Contracting State of the PCT (if other kind of protection or treatment desired, specify on dotted line)

National Patent (if other kind of protection or treatment desired, specify on dotted line):

- | | | |
|---|--|---|
| ☒ AE United Arab Emirates | ☒ GE Georgia | ☐ MWMalawi |
| ☒ AG Antigua and Barbuda | ☒ GH Ghana | ☒ MX Mexico |
| ☒ AL Albania | ☐ GM Gambia | ☐ MZ Mozambique |
| ☒ AM Armenia | ☒ HR Croatia | ☒ NO Norway |
| ☒ AT Austria | ☒ HU Hungary | ☒ NZ New Zealand |
| ☒ AU Australia | ☒ ID Indonesia | ☒ PL Poland |
| ☒ AZ Azerbaijan | ☒ IL Israel | ☒ PT Portugal |
| ☒ BA Bosnia and Herzegovina | ☒ IN India | ☒ RO Romania |
| | ☒ IS Iceland | ☒ RU Russian Federation |
| ☒ BB Barbados | ☒ JP Japan | |
| ☒ BG Bulgaria | ☒ KE Kenya | ☐ SD Sudan |
| ☒ BR Brazil | ☒ KG Kyrgyzstan | ☒ SE Sweden |
| ☒ BY Belarus | ☒ KP Democratic People's Republic of Korea | ☒ SG Singapore |
| ☒ BZ Belize | ☒ KR Republic of Korea | ☒ SI Slovenia |
| ☒ CA Canada | ☒ KZ Kazakhstan | ☒ SK Slovakia |
| ☒ CH & LI Switzerland and Liechtenstein | ☒ LC Saint Lucia | ☐ SL Sierra Leone |
| ☒ CN China | ☒ LK Sri Lanka | ☒ TJ Tajikistan |
| ☒ CO Colombia | ☐ LR Liberia | ☒ TM Turkmenistan |
| ☒ CR Costa Rica | ☐ LS Lesotho | ☒ TR Turkey |
| ☒ CU Cuba | ☒ LT Lithuania | ☒ TT Trinidad and Tobago |
| ☒ CZ Czech Republic | ☒ LU Luxembourg | |
| ☒ DE Germany | ☒ LV Latvia | ☐ TZ United Republic of Tanzania |
| ☒ DK Denmark | ☒ MA Morocco | ☒ UA Ukraine |
| ☒ DM Dominica | ☒ MD Republic of Moldova | ☐ UG Uganda |
| ☒ DZ Algeria | | ☒ US United States of America |
| ☒ EE Estonia | ☐ MG Madagascar | ☒ UZ Uzbekistan |
| ☒ ES Spain | ☒ MK The former Yugoslav Republic of Macedonia | ☒ VN Viet Nam |
| ☒ FI Finland | ☒ MN Mongolia | ☒ YU Yugoslavia |
| ☒ GB United Kingdom | | ☒ ZA South Africa |
| ☒ GD Grenada | | ☒ ZW Zimbabwe |

Check-boxes below reserved for designating States which have become party to the PCT after issuance of this sheet:

- | | | |
|--|--|--------------------------------|
| ☒ EC Ecuador | ☒ .. OM Oman | ☐ |
| ☒ PH Philippines | ☒ .. TR Tunisia (to be designated) | ☐ |

Precautionary Designation Statement: In addition to the designations made above, the applicant also makes under Rule 4.9(b) all other designations which would be permitted under the PCT except any designation(s) indicated in the Supplemental Box as being excluded from the scope of this statement. The applicant declares that those additional designations are subject to confirmation and that any designation which is not confirmed before the expiration of 15 months from the priority date is to be regarded as withdrawn by the applicant at the expiration of that time limit. (Confirmation (including fees) must reach the receiving Office within the 15-month time limit.)

Supplemental Box

If the Supplemental Box is not used, this sheet should not be included in the request.

1. If, in any of the Boxes, except Boxes Nos. VIII (i) to (v) for which a special continuation box is provided, the space is insufficient to furnish all the information: in such case, write "Continuation of Box No." (Indicate the number of the Box) and furnish the information in the same manner as required according to the captions of the Box in which the space was insufficient, in particular:

Continuation of Box No. II
Novartis AG is applicant for all designated States with the exception of: AT (Austria) and US (USA)

 - (i) If more than two persons are to be indicated as applicants and/or inventors and no "continuation sheet" is available: in such case, write "Continuation of Box No. III" and indicate for each additional person the same type of information as required in Box No. III. The country of the address indicated in this Box is the applicant's State (that is, country) of residence if no State of residence is indicated below;

Continuation of Box No. III
Novartis-Erfindungen Verwaltungsgesellschaft m.b.H. is applicant for AT (Austria) only.
 - (ii) If, in Box No. II or in any of the sub-boxes of Box No. III, the indication "the States indicated in the Supplemental Box" is checked: in such case, write "Continuation of Box No. II" or "Continuation of Box No. III" or "Continuation of Boxes No. II and No. III" (as the case may be), indicate the name of the applicant(s) involved and, next to (each) such name, the State(s) (and/or, where applicable, ARIPO, Eurasian, European or OAPI patent) for the purposes of which the named person is applicant;
 - (iii) If, in Box No. II or in any of the sub-boxes of Box No. III, the inventor or the inventor/applicant is not inventor for the purposes of all designated States or for the purposes of the United States of America: in such case, write "Continuation of Box No. II" or "Continuation of Box No. III" or "Continuation of Boxes No. II and No. III" (as the case may be), indicate the name of the inventor(s) and, next to (each) such name, the State(s) (and/or, where applicable, ARIPO, Eurasian, European or OAPI patent) for the purposes of which the named person is inventor;
 - (iv) If, in addition to the agent(s) indicated in Box No. IV, there are further agents: in such case, write "Continuation of Box No. IV" and indicate for each further agent the same type of information as required in Box No. IV;
 - (v) If, in Box No. V, the name of any State (or OAPI) is accompanied by the indication "patent of addition," or "certificate of addition," or if, in Box No. V, the name of the United States of America is accompanied by an indication "continuation" or "continuation-in-part": in such case, write "Continuation of Box No. V" and the name of each State involved (or OAPI), and after the name of each such State (or OAPI), the number of the parent title or parent application and the date of grant of the parent title or filing of the parent application;
 - (vi) If, in Box No. VI, there are more than five earlier applications whose priority is claimed: in such case, write "Continuation of Box No. VI" and indicate for each additional earlier application the same type of information as required in Box No. VI.
2. If, with regard to the precautionary designation statement contained in Box No. V, the applicant wishes to exclude any State(s) from the scope of that statement: in such case, write "Designation(s) excluded from precautionary designation statement" and indicate the name or two-letter code of each State so excluded.

Box No. VI PRIORITY CLAIM

The priority of the following earlier application(s) is hereby claimed:

Filing date of earlier application (day/month/year)	Number of earlier application	Where earlier application is:		
		national application: country	regional application: regional Office	international application: receiving Office
item (1) 07 December 2000 (07.12.00)	0029895.0	GB		
item (2)				
item (3)				
item (4)				
item (5)				

☐ Further priority claims are indicated in the Supplemental Box.

The receiving Office is requested to prepare and transmit to the International Bureau a certified copy of the earlier application(s) (only if the earlier application was filed with the Office which for the purposes of this international application is the receiving Office) identified above as:

☐ all items ☐ item (1) ☐ item (2) ☐ item (3) ☐ item (4) ☐ item (5) ☐ other, see Supplemental Box

* Where the earlier application is an ARIPO application, indicate at least one country party to the Paris Convention for the Protection of Industrial Property or one Member of the World Trade Organization for which that earlier application was filed (Rule 4.10(b)(ii)):

Box No. VII INTERNATIONAL SEARCHING AUTHORITY

Choice of International Searching Authority (ISA) (if two or more International Searching Authorities are competent to carry out the international search, indicate the Authority chosen; the two-letter code may be used):

ISA /

Request to use results of earlier search; reference to that search (if an earlier search has been carried out by or requested from the International Searching Authority):

Date (day/month/year) Number Country (or regional Office)

Box No. VIII DECLARATIONS

The following declarations are contained in Boxes Nos. VIII (i) to (v) (mark the applicable check-boxes below and indicate in the right column the number of each type of declaration):

Number of
declarations

- | | | |
|---|--|---|
| ☐ Box No. VIII (i) | Declaration as to the identity of the inventor | : |
| ☐ Box No. VIII (ii) | Declaration as to the applicant's entitlement, as at the international filing date, to apply for and be granted a patent | : |
| ☐ Box No. VIII (iii) | Declaration as to the applicant's entitlement, as at the international filing date, to claim the priority of the earlier application | : |
| ☐ Box No. VIII (iv) | Declaration of inventorship (only for the purposes of the designation of the United States of America) | : |
| ☐ Box No. VIII (v) | Declaration as to non-prejudicial disclosures or exceptions to lack of novelty | : |

Sheet No. ...6...

Box No. IX CHECK LIST; LANGUAGE OF FILING																												
This international application contains: (a) the following number of sheets in paper form: request (including declaration sheets) : 6 description (excluding sequence listing part) : 19 claims : 3 abstract : 1 drawings : 0 Sub-total number of sheets : 29 sequence listing part of description (actual number of sheets if filed in paper form, whether or not also filed in computer readable form; see (b) below) : 0 Total number of sheets : 29 (b) sequence listing part of description filed in computer readable form (i) ☐ only (under Section 801(a)(i)) (ii) ☐ in addition to being filed in paper form (under Section 801(a)(ii)) Type and number of carriers (diskette, CD-ROM, CD-R or other) on which the sequence listing part is contained (additional copies to be indicated under item 9(ii), in right column):	This international application is accompanied by the following item(s) (mark the applicable check-boxes below and indicate in right column the number of each item): <table style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 80%;">1. ☒ fee calculation sheet</td> <td style="width: 20%; text-align: right;">1</td> </tr> <tr> <td>2. ☒ original separate power of attorney</td> <td style="text-align: right;">1</td> </tr> <tr> <td>3. ☐ original general power of attorney</td> <td style="text-align: right;"></td> </tr> <tr> <td>4. ☒ copy of general power of attorney; reference number, if any: AV.36671 + 37586</td> <td style="text-align: right;">2</td> </tr> <tr> <td>5. ☐ statement explaining lack of signature</td> <td style="text-align: right;"></td> </tr> <tr> <td>6. ☒ priority document(s) identified in Box No. VI as item(s):</td> <td style="text-align: right;">1</td> </tr> <tr> <td>7. ☐ translation of international application into (language):</td> <td style="text-align: right;"></td> </tr> <tr> <td>8. ☐ separate indications concerning deposited microorganism or other biological material</td> <td style="text-align: right;"></td> </tr> <tr> <td>9. ☐ sequence listing in computer readable form (indicate also type and number of carriers (diskette, CD-ROM, CD-R or other))</td> <td style="text-align: right;"></td> </tr> <tr> <td colspan="2">(i) ☐ copy submitted for the purposes of international search under Rule 13ter only (and not as part of the international application)</td> </tr> <tr> <td colspan="2">(ii) ☐ (only where check-box (b)(i) or (b)(ii) is marked in left column) additional copies including, where applicable, the copy for the purposes of international search under Rule 13ter</td> </tr> <tr> <td colspan="2">(iii) ☐ together with relevant statement as to the identity of the copy or copies with the sequence listing part mentioned in left column</td> </tr> <tr> <td>10. ☐ other (specify):</td> <td style="text-align: right;"></td> </tr> </table>	1. ☒ fee calculation sheet	1	2. ☒ original separate power of attorney	1	3. ☐ original general power of attorney		4. ☒ copy of general power of attorney; reference number, if any: AV.36671 + 37586	2	5. ☐ statement explaining lack of signature		6. ☒ priority document(s) identified in Box No. VI as item(s):	1	7. ☐ translation of international application into (language):		8. ☐ separate indications concerning deposited microorganism or other biological material		9. ☐ sequence listing in computer readable form (indicate also type and number of carriers (diskette, CD-ROM, CD-R or other))		(i) ☐ copy submitted for the purposes of international search under Rule 13ter only (and not as part of the international application)		(ii) ☐ (only where check-box (b)(i) or (b)(ii) is marked in left column) additional copies including, where applicable, the copy for the purposes of international search under Rule 13ter		(iii) ☐ together with relevant statement as to the identity of the copy or copies with the sequence listing part mentioned in left column		10. ☐ other (specify):		Number of items
1. ☒ fee calculation sheet	1																											
2. ☒ original separate power of attorney	1																											
3. ☐ original general power of attorney																												
4. ☒ copy of general power of attorney; reference number, if any: AV.36671 + 37586	2																											
5. ☐ statement explaining lack of signature																												
6. ☒ priority document(s) identified in Box No. VI as item(s):	1																											
7. ☐ translation of international application into (language):																												
8. ☐ separate indications concerning deposited microorganism or other biological material																												
9. ☐ sequence listing in computer readable form (indicate also type and number of carriers (diskette, CD-ROM, CD-R or other))																												
(i) ☐ copy submitted for the purposes of international search under Rule 13ter only (and not as part of the international application)																												
(ii) ☐ (only where check-box (b)(i) or (b)(ii) is marked in left column) additional copies including, where applicable, the copy for the purposes of international search under Rule 13ter																												
(iii) ☐ together with relevant statement as to the identity of the copy or copies with the sequence listing part mentioned in left column																												
10. ☐ other (specify):																												
Figure of the drawings which should accompany the abstract: -	Language of filing of the international application: English																											
Box No. X SIGNATURE OF APPLICANT, AGENT OR COMMON REPRESENTATIVE <i>Next to each signature, indicate the name of the person signing and the capacity in which the person signs (if such capacity is not obvious from reading the request).</i>																												
In the name of the applicants The representative <i>K. Becker</i> BECKER, Konrad, AV 36671 + 37586 14.11.2001																												
For receiving Office use only <table style="width: 100%; border-collapse: collapse;"> <tr> <td style="width: 40%; border: 1px solid black; padding: 2px;"> 1. Date of actual receipt of the purported international application: (05.12.01) </td> <td style="width: 20%; border: 1px solid black; padding: 2px;"> 05 DEC 2001 </td> <td style="width: 40%; border: 1px solid black; padding: 2px;"> 2. Drawings: ☐ received: ☐ not received: </td> </tr> <tr> <td colspan="2" style="border: 1px solid black; padding: 2px;"> 3. Corrected date of actual receipt due to later but timely received papers or drawings completing the purported international application: </td> <td rowspan="2" style="border: 1px solid black; padding: 2px;"></td> </tr> <tr> <td colspan="2" style="border: 1px solid black; padding: 2px;"> 4. Date of timely receipt of the required corrections under PCT Article 11(2): </td> </tr> <tr> <td style="border: 1px solid black; padding: 2px;"> 5. International Searching Authority (if two or more are competent): ISA / </td> <td colspan="2" style="border: 1px solid black; padding: 2px;"> 6. ☐ Transmittal of search copy delayed until search fee is paid </td> </tr> </table>			1. Date of actual receipt of the purported international application: (05.12.01)	05 DEC 2001	2. Drawings: ☐ received: ☐ not received:	3. Corrected date of actual receipt due to later but timely received papers or drawings completing the purported international application:			4. Date of timely receipt of the required corrections under PCT Article 11(2):		5. International Searching Authority (if two or more are competent): ISA /	6. ☐ Transmittal of search copy delayed until search fee is paid																
1. Date of actual receipt of the purported international application: (05.12.01)	05 DEC 2001	2. Drawings: ☐ received: ☐ not received:																										
3. Corrected date of actual receipt due to later but timely received papers or drawings completing the purported international application:																												
4. Date of timely receipt of the required corrections under PCT Article 11(2):																												
5. International Searching Authority (if two or more are competent): ISA /	6. ☐ Transmittal of search copy delayed until search fee is paid																											
For International Bureau use only Date of receipt of the record copy by the International Bureau:																												

(19) World Intellectual Property Organization
International Bureau



(43) International Publication Date
13 June 2002 (13.06.2002)

PCT

(10) International Publication Number
WO 02/46196 A1

- (51) International Patent Classification⁷: C07D 493/04
- (21) International Application Number: PCT/EP01/14771 ✓
- (22) International Filing Date: 5 December 2001 (05.12.2001)
- (25) Filing Language: English
- (26) Publication Language: English
- (30) Priority Data:
0629895.0 7 December 2000 (07.12.2000) GB
- (71) Applicant (for all designated States except AT, US): NOVARTIS AG [CH/CH]; Lichtstrasse 35, CH-4056 Basle (CH).
- (71) Applicant (for AT only): NOVARTIS-ERFINDUNGEN VERWALTUNGSGESELLSCHAFT M.B.H. [AT/AT]; Brunner Strasse 59, A-1230 Vienna (AT).
- (72) Inventors; and ✓
- (75) Inventors/Applicants (for US only): KÜSTERS, Ernst [DE/DE]; Gallenweilerweg 1, 79427 Eschbach (DE).
- INTERNÄHRER, Heinz [CH/CH]; Widhagweg 46, CH-4303 Kaiseraugst (CH).
- (74) Agent: BECKER, Konrad; Novartis AG, Corporate Intellectual Property, Patent & Trademark Dept., CH-4002 Basel (CH).
- (81) Designated States (national): AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LT, LU, LV, MA, MD, MK, MN, MX, NO, NZ, OM, PH, PL, PT, RO, RU, SE, SG, SI, SK, TJ, TM, TR, TT, UA, US, UZ, VN, YU, ZA, ZM, ZW.
- (84) Designated States (regional): Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR).
- Published:
— with international search report
- For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.

(54) Title: PROCESS FOR THE ISOLATION AND PURIFICATION OF EPOTHILONES

(57) Abstract: The invention relates to a desorption process for epothilones, especially epothilone A and/or epothilone B, from resins and new production, work-up or purification processes or manufacturing methods for epothilones comprising said desorption process, as well as the use of certain solvents for the desorption of epothilones from resins.

WO 02/46196 A1

22.05.2002
4-31672A
Peter Roth

VT
PCT
EP01/14771

12

84

From the INTERNATIONAL SEARCHING AUTHORITY

To:
NOVARTIS AG
Corporate Intellectual Property
Attn. Becker, Konrad
Patent & Trademark Department
CH-4002 Basel
SWITZERLAND

PCT

NOTIFICATION OF TRANSMITTAL OF
THE INTERNATIONAL SEARCH REPORT
OR THE DECLARATION

B + TM Dept.

22 MAR 2002

(P) (A) (P) (M) (S) (I) F/L (P) (S) (T) (B)

Kopien:

Date of mailing
(day/month/year) 22/03/2002

Applicant's or agent's file reference
4-31672A

FOR FURTHER ACTION See paragraphs 1 and 4 below

International application No.
PCT/EP 01/14771

International filing date
(day/month/year) 05/12/2001

Applicant

NOVARTIS AG

1. ☒ The applicant is hereby notified that the International Search Report has been established and is transmitted herewith.

Filing of amendments and statement under Article 19:

The applicant is entitled, if he so wishes, to amend the claims of the International Application (see Rule 46):

When? The time limit for filing such amendments is normally 2 months from the date of transmittal of the International Search Report; however, for more details, see the notes on the accompanying sheet.

Where? Directly to the International Bureau of WIPO
34, chemin des Colombettes
1211 Geneva 20, Switzerland
Facsimile No.: (41-22) 740.14.35

For more detailed instructions, see the notes on the accompanying sheet.

2. ☐ The applicant is hereby notified that no International Search Report will be established and that the declaration under Article 17(2)(a) to that effect is transmitted herewith.

3. ☐ With regard to the protest against payment of (an) additional fee(s) under Rule 40.2, the applicant is notified that:

☐ the protest together with the decision thereon has been transmitted to the International Bureau together with the applicant's request to forward the texts of both the protest and the decision thereon to the designated Offices.

☐ no decision has been made yet on the protest; the applicant will be notified as soon as a decision is made.

4. Further action(s): The applicant is reminded of the following:

Shortly after 18 months from the priority date, the International application will be published by the International Bureau. If the applicant wishes to avoid or postpone publication, a notice of withdrawal of the International application, or of the priority claim, must reach the International Bureau as provided in Rules 90bis.1 and 90bis.3, respectively, before the completion of the technical preparations for International publication.

Within 19 months from the priority date, a demand for international preliminary examination must be filed if the applicant wishes to postpone the entry into the national phase until 30 months from the priority date (in some Offices even later).

Within 20 months from the priority date, the applicant must perform the prescribed acts for entry into the national phase before all designated Offices which have not been elected in the demand or in a later election within 19 months from the priority date or could not be elected because they are not bound by Chapter II.

Name and mailing address of the International Searching Authority



European Patent Office, P.B. 5818 Patentaan 2
NL-2280 HV Rijswijk
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl,
Fax: (+31-70) 340-3016

Authorized officer

Sandrine Parriche

NOTES TO FORM PCT/ISA/220

These Notes are intended to give the basic instructions concerning the filing of amendments under Article 19. The Notes are based on the requirements of the Patent Cooperation Treaty, the Regulations and the Administrative Instructions under that Treaty. In case of discrepancy between these Notes and those requirements, the latter are applicable. For more detailed information, see also the PCT Applicant's Guide, a publication of WIPO.

In these Notes, "Article", "Rule", and "Section" refer to the provisions of the PCT, the PCT Regulations and the PCT Administrative Instructions respectively.

INSTRUCTIONS CONCERNING AMENDMENTS UNDER ARTICLE 19

The applicant has, after having received the international search report, one opportunity to amend the claims of the international application. It should however be emphasized that, since all parts of the international application (claims, description and drawings) may be amended during the international preliminary examination procedure, there is usually no need to file amendments of the claims under Article 19 except where, e.g. the applicant wants the latter to be published for the purposes of provisional protection or has another reason for amending the claims before international publication. Furthermore, it should be emphasized that provisional protection is available in some States only.

What parts of the international application may be amended?

Under Article 19, only the claims may be amended.

During the international phase, the claims may also be amended (or further amended) under Article 34 before the International Preliminary Examining Authority. The description and drawings may only be amended under Article 34 before the International Examining Authority.

Upon entry into the national phase, all parts of the international application may be amended under Article 28 or, where applicable, Article 41.

When?

Within 2 months from the date of transmittal of the international search report or 18 months from the priority date, whichever time limit expires later. It should be noted, however, that the amendments will be considered as having been received on time if they are received by the International Bureau after the expiration of the applicable time limit but before the completion of the technical preparations for international publication (Rule 46.1).

Where not to file the amendments?

The amendments may only be filed with the International Bureau and not with the receiving Office or the International Searching Authority (Rule 46.2).

Where a demand for international preliminary examination has been filed, see below.

How?

Either by cancelling one or more entire claims, by adding one or more new claims or by amending the text of one or more of the claims as filed.

A replacement sheet must be submitted for each sheet of the claims which, on account of an amendment or amendments, differs from the sheet originally filed.

All the claims appearing on a replacement sheet must be numbered in Arabic numerals. Where a claim is cancelled, no renumbering of the other claims is required. In all cases where claims are renumbered, they must be renumbered consecutively (Administrative Instructions, Section 205(b)).

The amendments must be made in the language in which the international application is to be published.

What documents must/may accompany the amendments?

Letter (Section 205(b)):

The amendments must be submitted with a letter.

The letter will not be published with the international application and the amended claims. It should not be confused with the "Statement under Article 19(1)" (see below, under "Statement under Article 19(1)").

The letter must be in English or French, at the choice of the applicant. However, if the language of the international application is English, the letter must be in English; if the language of the international application is French, the letter must be in French.

The letter must indicate the differences between the claims as filed and the claims as amended. It must, in particular, indicate, in connection with each claim appearing in the international application (it being understood that identical indications concerning several claims may be grouped), whether

- (i) the claim is unchanged;
- (ii) the claim is cancelled;
- (iii) the claim is new;
- (iv) the claim replaces one or more claims as filed;
- (v) the claim is the result of the division of a claim as filed.

The following examples illustrate the manner in which amendments must be explained in the accompanying letter:

1. [Where originally there were 48 claims and after amendment of some claims there are 51]:
"Claims 1 to 29, 31, 32, 34, 35, 37 to 48 replaced by amended claims bearing the same numbers; claims 30, 33 and 36 unchanged; new claims 49 to 51 added."
2. [Where originally there were 15 claims and after amendment of all claims there are 11]:
"Claims 1 to 15 replaced by amended claims 1 to 11."
3. [Where originally there were 14 claims and the amendments consist in cancelling some claims and in adding new claims]:
"Claims 1 to 6 and 14 unchanged; claims 7 to 13 cancelled; new claims 15, 16 and 17 added." or
"Claims 7 to 13 cancelled; new claims 15, 16 and 17 added; all other claims unchanged."
4. [Where various kinds of amendments are made]:
"Claims 1-10 unchanged; claims 11 to 13, 18 and 19 cancelled; claims 14, 15 and 16 replaced by amended claim 14; claim 17 subdivided into amended claims 15, 16 and 17; new claims 20 and 21 added."

"Statement under article 19(1)" (Rule 46.4)

The amendments may be accompanied by a statement explaining the amendments and indicating any impact that such amendments might have on the description and the drawings (which cannot be amended under Article 19(1)).

The statement will be published with the international application and the amended claims.

It must be in the language in which the international application is to be published.

It must be brief, not exceeding 500 words if in English or if translated into English.

It should not be confused with and does not replace the letter indicating the differences between the claims as filed and as amended. It must be filed on a separate sheet and must be identified as such by a heading, preferably by using the words "Statement under Article 19(1)."

It may not contain any disparaging comments on the international search report or the relevance of citations contained in that report. Reference to citations, relevant to a given claim, contained in the international search report may be made only in connection with an amendment of that claim.

Consequence if a demand for international preliminary examination has already been filed

If, at the time of filing any amendments under Article 19, a demand for international preliminary examination has already been submitted, the applicant must preferably, at the same time of filing the amendments with the International Bureau, also file a copy of such amendments with the International Preliminary Examining Authority (see Rule 62.2(a), first sentence).

Consequence with regard to translation of the international application for entry into the national phase

The applicant's attention is drawn to the fact that, where upon entry into the national phase, a translation of the claims as amended under Article 19 may have to be furnished to the designated/elected Office, instead of, or in addition to, the translation of the claims as filed.

For further details on the requirements of each designated/elected Office, see Volume II of the PCT Applicant's Guide.

PCT

INTERNATIONAL SEARCH REPORT

(PCT Article 18 and Rules 43 and 44)

Applicant's or agent's file reference 4-31672A	FOR FURTHER ACTION see Notification of Transmittal of International Search Report (Form PCT/ISA/220) as well as, where applicable, item 5 below.	
International application No. PCT/EP 01/ 14771	International filing date (day/month/year) 05/12/2001	(Earliest) Priority Date (day/month/year) 07/12/2000
Applicant NOVARTIS AG		

This International Search Report has been prepared by this International Searching Authority and is transmitted to the applicant according to Article 18. A copy is being transmitted to the International Bureau.

This International Search Report consists of a total of 5 sheets.

☒ It is also accompanied by a copy of each prior art document cited in this report.

1. Basis of the report

- a. With regard to the language, the international search was carried out on the basis of the international application in the language in which it was filed, unless otherwise indicated under this item.

☐ the international search was carried out on the basis of a translation of the international application furnished to this Authority (Rule 23.1(b)).

- b. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of the sequence listing:

☐ contained in the international application in written form.

☐ filed together with the international application in computer readable form.

☐ furnished subsequently to this Authority in written form.

☐ furnished subsequently to this Authority in computer readable form.

☐ the statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

☐ the statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished

2. ☒ Certain claims were found unsearchable (See Box I).

3. ☐ Unity of invention is lacking (see Box II).

4. With regard to the title,

☒ the text is approved as submitted by the applicant.

☐ the text has been established by this Authority to read as follows:

5. With regard to the abstract,

☒ the text is approved as submitted by the applicant.

☐ the text has been established, according to Rule 38.2(b), by this Authority as it appears in Box III. The applicant may, within one month from the date of mailing of this international search report, submit comments to this Authority.

6. The figure of the drawings to be published with the abstract is Figure No.

☐ as suggested by the applicant.

☐ because the applicant failed to suggest a figure.

☐ because this figure better characterizes the invention.

☒ None of the figures.

INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 01/14771

16

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C07D493/04

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 C07D

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the International search (name of data base and, where practical, search terms used)

EPO-Internal, WPI Data, PAJ, CHEM ABS Data, BEILSTEIN Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 99 42602 A (NOVARTIS ERFIND VERWALT GMBH; NOVARTIS AG (CH)) 26 August 1999 (1999-08-26) Claims 12-14; pages 37-38, example 3; page 9, paragraph 2.	1,7,9,12
A	WO 93 10121 A (CIBA GEIGY AG; BIOTECHNOLOG FORSCHUNG GMBH (DE)) 27 May 1993 (1993-05-27) Page 8, last paragraph.	1-13
A	WO 98 22461 A (BIOTECHNOLOG FORSCHUNG GMBH; GERTH KLAUS (DE); HOEFLE GERHARD (DE)) 28 May 1998 (1998-05-28) Claim 1, step c.	1-13

	-/--	



Further documents are listed in the continuation of box C.



Patent family members are listed in annex.

*** Special categories of cited documents:**

- *A* document defining the general state of the art which is not considered to be of particular relevance
- *E* earlier document but published on or after the international filing date
- *L* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- *O* document referring to an oral disclosure, use, exhibition or other means
- *P* document published prior to the international filing date but later than the priority date claimed

- *T* later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- *X* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
- *Y* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
- *&* document member of the same patent family

Date of the actual completion of the international search

6 March 2002

Date of mailing of the international search report

22/03/2002

Name and mailing address of the ISA

European Patent Office, P.B. 5818 Patentlaan 2
NL - 2260 HV Rijswijk
Tel. (+31-70) 340-2040, Tx. 31 651 epo nl.
Fax: (+31-70) 340-3016

Authorized officer

Weisbrod, T

INTERNATIONAL SEARCH REPORT

International Application No

PCT/EP 01/14771

17

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	NICOLAOU K C ET AL: "Probing the ring size of epothilones: total synthesis of '14!-, '15!-, '17!-, and '18!Epothilones A" ANGEWANDTE CHEMIE. INTERNATIONAL EDITION, VERLAG CHEMIE. WEINHEIM, DE, vol. 37, no. 1/2, 1998, pages 81-84, XP002131226 ISSN: 0570-0833 the whole document	1-13
A	NICOLAOU K C ET AL: "Chemical Biology of Epothilones" ANGEWANDTE CHEMIE. INTERNATIONAL EDITION, VERLAG CHEMIE. WEINHEIM, DE, vol. 37, no. 15, August 1998 (1998-08), pages 2014-2045, XP002131418 ISSN: 0570-0833 Tables 2-6.	1-13
A	REICHHARDT, C.: "Solvents and Solvent Effects in Organic Chemistry, 2nd. ed." 1988, VCH, WEINHEIM XP002192137 Page 60, lines 5-7: "the term polarity itself has, until now, not been precisely defined".	1-13

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

Continuation of Box I.2

Claims Nos.: 1 and 3-13 (all part)

The present claims 1 and 3-13 relate to processes comprising desorbing epothilones from a resin respectively the use of a weakly polar or apolar solvent for desorbing epothilones from a resin. The term epothilone, however, is not precisely defined in the technical field. Along with the parent compounds, i.e. the 16-membered thiazole-comprising macrolides epothilones A and B, the term epothilone and epothilone derivative includes inter alia 14- to 18-membered macrolides with or without annelated oxirane ring, compounds with modified configuration of stereocentres or modified geometry of double bonds, as well as compounds wherein the side-chain thiazole has been replaced (cf. Angew. Chem. Int. Ed. 1998, 37, pp. 81-83, and pp. 2015-2045). The said claims relate thus to methods and uses for the desorption of an extremely large number of diverse possible compounds. Support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT is to be found, however, for only a very small proportion of the claimed methods and uses i.e. for the desorption of natural epothilone B (cf. examples). In the present case, the claims so lack support, and the application so lacks disclosure, that a meaningful search over the whole of the claimed scope is impossible. Consequently, the search has been carried out for those parts of the claims which appear to be supported and disclosed, namely those parts directed to methods and uses relating to the natural 16-membered epothilones A-F.

The applicant's attention is drawn to the fact that claims, or parts of claims, relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/EP 01/14771

Box I Observations where certain claims were found unsearchable (Continuation of Item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:
2. ☒ Claims Nos.: 1 and 3-13 (all part)
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:
see FURTHER INFORMATION sheet PCT/ISA/210
3. ☐ Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 8.4(a).

Box II Observations where unity of invention is lacking (Continuation of Item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
☐ No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Information on patent family members

International Application No

PCT/EP 01/14771

20

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9942602 A	26-08-1999	AU 3028799 A	06-09-1999
		BR 9908119 A	24-10-2000
		CN 1291239 T	11-04-2001
		WO 9942602 A2	26-08-1999
		EP 1054994 A2	29-11-2000
		HU 0100855 A2	28-06-2001
		NO 20004114 A	17-10-2000
		PL 342423 A1	04-06-2001
		SK 12402000 A3	10-05-2001
		TR 200002431 T2	22-01-2001
		US 6194181 B1	27-02-2001
WO 9310121 A	27-05-1993	DE 4138042 A1	27-05-1993
		AU 2943792 A	15-06-1993
		WO 9310121 A1	27-05-1993
WO 9822461 A	28-05-1998	AU 5483798 A	10-06-1998
		BR 9713363 A	25-01-2000
		CN 1237970 A	08-12-1999
		CZ 9901750 A3	15-09-1999
		WO 9822461 A1	28-05-1998
		EP 0941227 A1	15-09-1999
		HU 0000497 A2	28-06-2000
		JP 2001504474 T	03-04-2001
		NO 992338 A	14-05-1999
		PL 333435 A1	06-12-1999
		TW 408119 B	11-10-2000
		ZA 9710384 A	18-05-1999

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

PCT

To:

GROSS, Florent
NOVARTIS AG
Corporate Intellectual Property
Patent & Trademark Department
CH-4002 Basel
SUISSE

WRITTEN OPINION

P + TM Dept.

12. Juni 2002

(PCT Rule 66)

APPL M/D F/L PS/TS
Kopien:

Date of mailing
(day/month/year)

10/06/2002

Applicant's or agent's file reference

4-31672A

REPLY DUE

within 2 / 00 months/days
from the above date of mailing

International application No.

PCT/EP 01/ 14771

International filing date (day/month/year)

05/12/2001

Priority date (day/month/year)

07/12/2000

International Patent Classification (IPC) or both national classification and IPC

C07D493/04

Applicant

NOVARTIS AG ET AL.

1. This written opinion is the first drawn up by this International Preliminary Examining Authority.

2. This opinion contains indications relating to the following items:

I ☒ Basis of the opinion

II ☐ Priority

III ☒ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

IV ☐ Lack of unity of invention

V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

VI ☐ Certain documents cited

VII ☐ Certain defects in the international application

VIII ☐ Certain observations on the international application

3. The applicant is hereby invited to reply to this opinion.

When? See the time limit indicated above. The applicant may, before the expiration of that time limit, request this Authority to grant an extension, see Rule 66.2(d).

How? By submitting a written reply, accompanied, where appropriate, by amendments, according to Rule 66.3. For the form and the language of the amendments, see Rules 66.8 and 66.9.

Also For an additional opportunity to submit amendments, see Rule 66.4. For the examiner's obligation to consider amendments and/or arguments, see Rule 66.4bis. For an informal communication with the examiner, see Rule 66.6.

If no reply is filed, the international preliminary examination report will be established on the basis of this opinion.

4. The final date by which the international preliminary examination report must be established according to Rule 69.2 is:

07/04/2003

Name and mailing address of the IPEA/

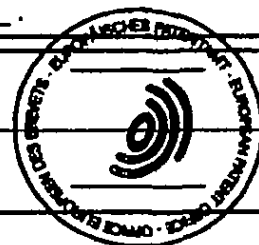


European Patent Office
D-80298 Munich
Tel. (+49-89) 2399-0, Tx: 523656 epmu d
Fax: (+49-89) 2399-4465

Authorized officer

Examiner

Formalities officer
(incl. extension of time limits)
Tel. (+49-89) 2399 2828



WRITTEN OPINION

International application No PCT/EP 01/14771

I. Basis of the opinion

The basis of this written opinion is the application as originally filed.

III. Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

The question of whether the claimed invention appears to be novel, to involve an inventive step, or to be industrially applicable has not been and will not be the subject of the international preliminary examination in respect of the claims which have not been searched (Article 17(2)(a) or (3) and Rule 66.1(e) PCT; see also international search report).

V. Reasoned statement under Rule 66.2(a)(II) with regard to novelty, inventive step or industrial applicability

1. To the extent that the international preliminary examination has been carried out (see item III above), the following is pointed out:
2. In light of the documents cited in the international search report, it is considered that the invention as defined in at least some of the claims, which have been the subject of an international search report, does not appear to meet the criteria mentioned in Article 33(1) PCT, i.e. does not appear to be novel and/or to involve an inventive step (see international search report, in particular the documents cited X and/or Y and corresponding claim references).
3. If amendments are filed, the applicant should comply with the requirements of Rule 66.8 PCT and indicate the basis of the amendments in the documents of the application as originally filed (Article 34 (2) (b) PCT) otherwise these amendments may not be taken into consideration for the establishment of the international preliminary examination report. The attention of the applicant is drawn to the fact that if the application contains an unnecessary plurality of independent claims, no examination of any of the claims will be carried out.

NB: Should the applicant decide to request detailed substantive examination, then an international preliminary examination report will normally be established directly. Exceptionally the examiner may draw up a second written opinion, should this be explicitly requested.

PATENT COOPERATION TREATY

23

16.12.2002
4-31672A
Peter Roth

VT
PCT
EP01/14771

Pat TM Dept.

19. Sep. 2002

PCT

WRITTEN OPINION

(PCT Rule 66)

GROSS, Florent
NOVARTIS AG
Corporate Intellectual Property
Patent & Trademark Department
CH-4002 Basel
SUISSE

APPL M/D F/L PS/TS

Kopian:

Date of mailing
(day/month/year)

16.09.2002

Applicant's or agent's file reference

4-31672A

REPLY DUE

within 3 month(s)
from the above date of mailing

International application No.

PCT/EP01/14771

International filing date (day/month/year)

05/12/2001

Priority date (day/month/year)

07/12/2000

International Patent Classification (IPC) or both national classification and IPC

C07D483/04

Applicant

NOVARTIS AG ET AL.

1. This written opinion is the first drawn up by this International Preliminary Examining Authority.

2. This opinion contains indications relating to the following items:

- I ☒ Basis of the opinion
- II ☐ Priority
- III ☒ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- IV ☐ Lack of unity of invention
- V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
- VI ☐ Certain document cited
- VII ☐ Certain defects in the international application
- VIII ☐ Certain observations on the international application

3. The applicant is hereby invited to reply to this opinion.

When? See the time limit indicated above. The applicant may, before the expiration of that time limit, request this Authority to grant an extension, see Rule 66.2(d).

How? By submitting a written reply, accompanied, where appropriate, by amendments, according to Rule 66.3. For the form and the language of the amendments, see Rules 66.8 and 66.9.

Also: For an additional opportunity to submit amendments, see Rule 66.4.
For the examiner's obligation to consider amendments and/or arguments, see Rule 66.4 bis.
For an informal communication with the examiner, see Rule 66.6.

If no reply is filed, the international preliminary examination report will be established on the basis of this opinion.

4. The final date by which the international preliminary examination report must be established according to Rule 69.2 is: 07/04/2003.

Name and mailing address of the international preliminary examining authority:



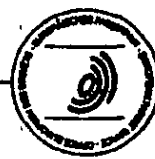
European Patent Office
D-80298 Munich
Tel. +49 89 2399 - 0 Tlx 523656 epmu d
Fax: +49 89 2399 - 4465

Authorized officer / Examiner

Weisbrod, T

Formalities officer (incl. extension of time limits)

Ferreiro Vasconcelos, M
Telephone No. +49 89 2399 7906



WRITTEN OPINION

International application No. PCT/EP01/14771

I. Basis of the opinion

1. With regard to the elements of the international application (Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this opinion as "originally filed").

Description, pages:

1-19 as originally filed

Claims, No.:

1-9,10 (part) as originally filed

10 (part),11-14 with telefax of 05/08/2002

2. With regard to the language, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

- ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
- ☐ the language of publication of the international application (under Rule 48.3(b)).
- ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- ☐ contained in the international application in written form.
- ☐ filed together with the international application in computer readable form.
- ☐ furnished subsequently to this Authority in written form.
- ☐ furnished subsequently to this Authority in computer readable form.
- ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
- ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

- ☐ the description, pages:
- ☐ the claims, Nos.:
- ☐ the drawings, sheets:

WRITTEN OPINION

International application No. PCT/EP01/14771

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)):

(Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

III. Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non-obvious), or to be industrially applicable have not been and will not be examined in respect of:

☐ the entire international application,

☒ claims Nos. 1,3-14,

because:

☐ the said international application, or the said claims Nos. relate to the following subject matter which does not require an international preliminary examination (*specify*):

☐ the description, claims or drawings (*indicate particular elements below*) or said claims Nos. are so unclear that no meaningful opinion could be formed (*specify*):

☐ the claims, or said claims Nos. are so inadequately supported by the description that no meaningful opinion could be formed.

☒ no international search report has been established for the said claims Nos. 1,3-14.

2. A written opinion cannot be drawn due to the failure of the nucleotide and/or amino acid sequence listing to comply with the standard provided for in Annex C of the Administrative Instructions:

☐ the written form has not been furnished or does not comply with the standard.

☐ the computer readable form has not been furnished or does not comply with the standard.

V. Reasoned statement under Rule 68.2(a)(II) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N) Claims 1,7,9,12,13

Inventive step (IS) Claims 1-4,7-13

Industrial applicability (IA) Claims

WRITTEN OPINION

International application No. **PCT/EP01/14771**

2. Citations and explanations
see separate sheet

**WRITTEN OPINION
SEPARATE SHEET**

International application No. PCT/EP01/14771

Re Item I**Basis of the opinion**

- 1 The amendments filed by FAX of 05.08.2002 comply with the requirements of Article 19 PCT, since they do not introduce subject-matter which extends beyond the application as originally filed. oe
- 2 The application is directed to
 - (i) a process for desorbing epothilones from a resin, said process making use of a weakly polar or apolar solvent (claims 1-6 and 8);
 - (ii) a process for the isolation of epothilones comprising
 - (a) the desorption of the epothilones from a synthetic resin with a weakly polar or apolar solvent, and
 - (b) separation of the epothilones by reversed-phase chromatography with a nitrile/water mixture (claims 7, 8, 11, 12);
 - (iii) a process for working up epothilones comprising the steps of
 - (a) desorbing the epothilones with a weakly polar or apolar solvent,
 - (b) removing the solvent used for desorption by evaporation,
 - (c) separating the epothilones by reversed-phase chromatography by eluting with a nitrile/water mixture, and removing the nitrile by evaporation, and
 - (d) recrystallizing a purified epothilone (claims 9-12); as well as
 - (iv) the use of a weakly polar or apolar solvent for the desorption of epothilones from a resin (claims 13-14).

Re Item III**Non-establishment of opinion with regard to novelty, inventive step and industrial applicability**

The ISA has issued only a partial search report for the original claims 1 and 3-13, insofar as these claims concern methods or uses which relate to the natural epothilones A-F. For such partially searched claims it is not possible to determine whether, and if so to what extent, the matter to which said claims relate is novel and might involve an inventive step. No full International Preliminary Examination will thus be carried out with regard to novelty and inventive step insofar as the amended claims

**WRITTEN OPINION
SEPARATE SHEET**

International application No. PCT/EP01/14771

1 and 3-14 do not relate to the natural epothilones A-F (cf. Rule 66.1(e) PCT).

Re Item V

**Reasoned statement under Rule 66.2(a)(II) with regard to novelty, inventive step
r industrial applicability; citations and explanations supporting such statement**

1 Reference is made to the following documents:

D1: WO-A-99 42602, 26.08.1999.

D2: WO-A-93 10121, 17.05.1993.

D3: WO-A-98 22461, 28.05.1998.

D4: Nicolaou, K. C. *et al. Angew. Chem. Int. Ed.* 1998, 37, 81-83.

D5: Nicolaou, K. C. *et al. Angew. Chem. Int. Ed.* 1998, 37, 2015-2045.

D6: Reichardt, C. *Solvents and Solvent Effects in Organic Chemistry*, 2nd. ed., VCH, Weinheim 1988, p. 60.

D7: Snyder, L. R. *J. Chromatogr. Sci.* 1978, 16, 223-234.

The document D7 has not been cited in the International search report, but is mentioned in the application. A copy is annexed to this opinion.

2 The application does not comply with the requirements of the Articles 5 and 6 PCT for the following reasons.

2.1 Present claims 1 and 3-14 relate to processes comprising desorbing epothilones from a resin respectively the use of a weakly polar or apolar solvent for desorbing epothilones from a resin. The term epothilone, however, is not precisely defined in the technical field. Along with the parent compounds, i.e. the 16-membered thiazole-comprising macrolides epothilone A and B, the term epothilone includes inter alia 14- to 18-membered macrolides with or without annelated oxirane ring, compounds with modified configuration of stereocentres or modified geometry of double bonds, as well as compounds wherein the side-chain thiazole has been replaced (see e.g. D4 and D5). The said claims relate thus to methods and uses for the desorption of an extremely large number of diverse possible compounds. Support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT is to be found, however, for only a very small proportion of the claimed methods and uses i.e. for the desorption of natural epothilone B (cf. examples). In the present case, the claims so lack support, and the application so

lacks disclosure.

- 3.2 The vague term "weakly polar solvent" used in the claims renders the claims 1-4, 7-11, and 13 unclear (see e.g. D6, page 60, lines 5-7). Furthermore, the term "lower" in lower alkyl etc. has no precise meaning with regard to the number of carbon atoms which define such lower groups, thereby resulting in a lack of clarity of the claims 5, 6, 9, 10, and 14. *amend X*

- 3.3 In addition claim 9 has to be objected under Article 6 PCT for listing a plethora of optional and thus unnecessary, non-limiting technical features (see e.g. "in particular ...", "optionally ...", "especially ...", "if desired ...", etc.), thereby resulting in a lack of conciseness of the claim. *amend*

3 Novelty

D1 describes a biotechnological process for the production of epothilones wherein the culture medium is worked-up by

- (a) mixing the aqueous centrifugate with a synthetic resin,
- (b) desorbing the epothilones from the resin with a polar solvent (especially an alcohol, cf. page 14, lines 4-5)
- (c) concentrating the resulting solution if necessary,
- (d) separating the epothilones by reversed-phase chromatography,
- (e) followed by recrystallization if desired (cf. claims 12-14; and page 37, example 3 wherein the polar solvent is isopropanol).

The term "weakly polar solvent" used in present claims 1, 7, 9, and 13 does not unequivocally distinguish the present used solvents from the polar solvents used in D1. Consequently, present claims 1, 7, 9, and 13 lack novelty in view of D1. Furthermore, amended claim 12 overlaps with the disclosure of D1 insofar as the alcohol methoxyethanol is concerned (cf. D7, page 226, methoxyethanol with $x_o = 0.38$, $x_d = 0.24$, $x_n = 0.38$). Claim 12 lacks thus novelty in view of D1.

D2 describes a biotechnological process for the production of epothilones A and B, and D3 a biotechnological manufacture process for the epothilones C-F. These processes comprise the step of desorbing the epothilones from an absorber resin with methanol (D2, page 8, last paragraph; D3, claim 1, step c). In view of D2 and D3 the present claimed matter may be regarded novel since it refers to a "weak

polar solvent" instead of methanol for the desorption of epothilones from a resin.

The documents D4 to D7 do not appear relevant to the question of novelty of the present claimed matter, since the said documents do not disclose the desorption of epothilones from a resin.

4 Inventive Step

Insofar as the application relates to novel processes or uses involving lower alkyl halogenides and aromatic solvents for desorbing epothilones from a resin, the following observations apply to the requirements of inventive step.

- 4.1 The application exemplifies the desorption of epothilone B from Amberlite XAD-16 with toluene instead of isopropanol as exemplified in D1 (present application: page 13, example 1; D1, page 37, example 3). The application furthermore provides comparative data for the desorption of epothilone B with toluene and dichloromethane in comparison with isopropanol (pages 16-18, table 1). The data show that the use of toluene as desorbant results in a decreased desorption time and increased yield of desorbed epothilone B compared with isopropanol (cf. page 17, runs 10-12 in comparison with run 9); and the comparison of the dichloromethane and isopropanol runs indicate a positive trend in favour of dichloromethane (cf. page 16, run 4 in comparison with run 3).
- 4.2 In view of D1 the problem underlying the present application may be seen in the provision of an alternative, improved, process for the desorption of epothilones from absorber resins. Since the favourable effects of toluene and dichloromethane on the desorption of epothilone B does not appear obvious in view of the cited prior art, an inventive step may be acknowledged for processes involving lower alkyl halogenide or aromatic solvents as desorbants as well as for the use of these solvents for the desorption of epothilones i.e. the claims 5, 6 and 14 (with regard to "lower alkyl halogenide" see however item 2.2 above).

Amendments

The applicant's attention is drawn to the fact that the application may not be amended in such a way that it contains subject-matter which extends beyond the content of the application as filed (Article 19(2) and 34(2)(b) PCT). When filing new claims, the complete set of claims should be filed rather than single amended sheets. Additionally,

**WRITTEN OPINION
SEPARATE SHEET**

International application No. PCT/EP01/14771

one set indicating the amendments in hand-writing should be provided. The applicant, furthermore, should clearly indicate the location of those places in the original application which would support each of the specific amendments.

Classification of the Solvent Properties of Common Liquids

XP-001062572

L.R. Snyder, Technicon Instruments Corp., Tarrytown, New York 10591

Abstract

A previous scheme for the classification of various solvents according to their polarity P' and selectivity (x_D, x_G, x_H) has been modified, leading to greater precision in these calculated solvent parameters. This scheme has been further extended for the case of liquids used as gas chromatography (GC) stationary phases, allowing classification of additional solvents studied by McReynolds. The greater precision of these resulting data allows a number of further checks on the utility of this approach to solvent classification, and allows most common solvents to be characterized according to different selectivity groups.

The present study also confirms that three polar test-solutes suffice to establish the primary selectivity characteristics of various solvents. Dispersion interactions in solutions of polar solvents appear to play an unimportant role in contributing to solvent selectivity.

Introduction

An article with the same title was published in 1974 (1). In that paper a scheme was described for the classification of common solvents according to their "polarity" or chromatographic strength (P' values), and according to their selectivity or relative ability to engage in hydrogen bonding or dipole interactions (x_i values; x_D , proton acceptor; x_G , proton donor; x_H , strong dipole). On the basis of this scheme, data from an extensive experimental study by Rohrschneider (2) were reduced to give values of P' and x_i for 75 common solvents that could be used in liquid chromatography (LC) or other separation processes (see discussion of 3). It was further found that these various solvents could be grouped into 8 selectivity classes (I, II, ... VIII) for easy selection of solvents of significantly differing selectivity. The major validation of this classification scheme was provided by the observation that solvents of similar functionality were grouped into the same selectivity class, as would be expected on theoretical grounds.

More recently, Poppe (4) has noted a small error in the calculation of the various P' and x_i values in (1). As we will see, corrected values of P' and x_i do not differ substantially from the values reported in (1). However, these corrected values of x_i do show better agreement among solvents of similar functionality than was found originally, and this has led to a review of these x_i values in terms of further substantiating the utility of this solvent classification scheme.

Finally, it has been found possible to calculate P' and x_i values for a large group of additional solvents: the less-volatile liquids used as stationary phases in gas chromatography (GC) and previously characterized by so-called Rohrschneider or McReynolds constants (e.g., 5,6). Thus, the original base of 75 volatile solvents classified by P' and x_i values in (1) is here expanded to include various GC phases. These latter data provide additional opportunities to test the consistency and utility of the present solvent classification scheme.

Calculations

Correction of Data for LC Solvents

The solvents originally discussed will be referred to in (1) as "LC solvents," because they are generally volatile and applicable as mobile phases for use in LC, and to distinguish them from the "GC solvents" or stationary phases described in (5,6). The treatment of (1) began with experimental solute distribution coefficients K_D for several test solutes: *n*-octane (*o*), ethanol (*e*), dioxane (*d*) and nitromethane (*n*). These K_D values were first corrected for solvent molecular weight by use of the solvent molar volume V_s :

$$K_D' = K_D V_s \quad (1)$$

These adjusted distribution coefficients K_D' were in turn used to calculate polar distribution coefficients K_D'' , which are corrected for the molar volume of the test-solutes ethanol, dioxane, or nitromethane:

$$\log K_D'' = \log K' - \log K_v \quad (2)$$

Here, K_v is the K_D' value of the hypothetical *n*-alkane with the same molar volume V_x as the solute *X*. K_v is in turn calculated from the K_D' value of *n*-octane (K_o) as:

$$\log K_v = (V_x/163) (\log K_o) \quad (3)$$

Finally, P' is calculated as the sum of ($\log K_D''$) values for the three solutes ethanol, dioxane, and nitromethane, and x_D, x_G and x_H are calculated as the ratio ($\log K_D''/P'$) for each solute (*e*, ethanol; *d*, dioxane; *n*, nitromethane).

Poppe (4) has noted that the values of P' and x_i reported in (1) are in error, due to the inadvertent use of K_D rather than K_D' values for the solutes ethanol, dioxane, and nitromethane in equation 2. This mistake was hidden in (1) by the observation that values of $\log K'$ thus calculated were quite close to zero for nonpolar solvents such as the aliphatic hydrocarbons. It was incorrectly argued in (1) that zero values of $\log K'$ were in fact expected for nonpolar solvents. Actually, this is not the case, for a variety of reasons (inductive effects, entropy effects, etc.). As seen in Table I, values of $\log K_D'$ for the C_6 - C_8 saturated hydrocarbon solvents are significantly different from zero. Therefore, as in (1), final values of $\log K_D''$ were corrected to give values of zero for the saturated hydrocarbon solvents, by subtracting the values of Table I from the appropriate $\log K_D''$ value from equation 2. For example, $\log K_D''$ values (equation 2) for ethanol as solute were corrected by subtraction of 1.773 (see Table I), prior to calculating P' and x_i values.

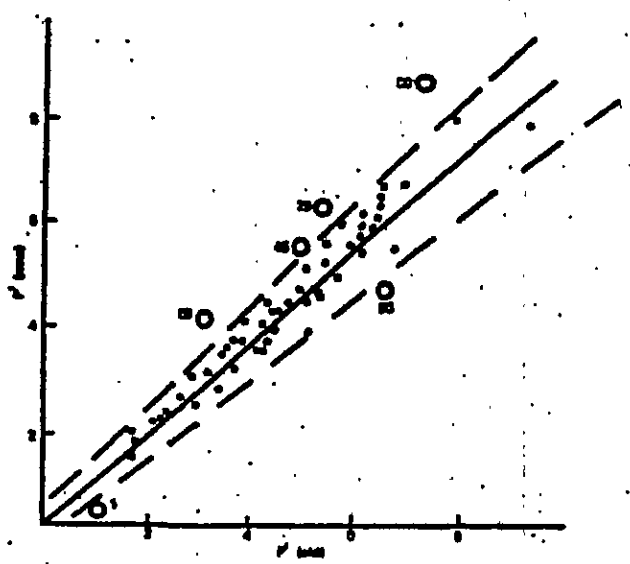
Reproduction (photocopying) of editorial content of this journal is prohibited without publisher's permission.

Table II. Classification of Solvents According to Present Scheme

No.	Solvent	ρ^a	Group	x_e	x_d	x_H	x_I	x_M
1	carbon disulfide	0.3	— ^b					
2	cyclohexane	0.2	— ^b					
3	triethyl amine	1.9	I ^c	0.56	0.12	0.32	0.05	0.15
4	ethyl ether	2.8	I	0.53	0.13	0.34	0.09	0.15
5	ethyl bromide ^d							
6	n-hexane	0.1	— ^b					
7	iso-octane	0.1	— ^b					
8	tetrahydrofuran	4.0	III	0.38	0.20	0.42	0.14	0.21
9	i-propyl ether	2.4	I	0.48	0.14	0.38	0.07	0.20
10	toluene	2.4	VII	0.25	0.28	0.47	— ^e	0.28
11	benzene	2.7	VII	0.23	0.32	0.45	0.16	0.29
12	p-xylene	2.5	VII	0.27	0.28	0.45	0.13	0.29
13	chloroform	4.1	VIII ^c	0.25	0.41	0.33	0.15	0.39
14	carbon tetrachloride	1.6	— ^b					
15	butyl ether	2.1	I	0.44	0.18	0.38	0.11	0.21
16	methylene chloride	3.1	V	0.29	0.18	0.53	0.20	— ^c
17	n-decane	0.4	— ^b					
18	chlorobenzene	2.7	VII	0.23	0.33	0.44	0.17	0.34
19	bromobenzene	2.7	VII	0.24	0.33	0.43	0.18	0.33
20	fluorobenzene	3.2	VII	0.24	0.32	0.45	0.18	0.33
21	2,6-lutidine	4.5	III	0.45	0.20	0.36	0.11	0.22
22	squalane	1.2	— ^b					
23	hexafluorobenzene ^d							
24	ethoxybenzene	3.3	VII	0.28	0.28	0.44	0.16	0.30
25	2-picoline	4.9	III	0.44	0.21	0.36	0.12	0.23
26	ethylene chloride ^d	3.5	V	0.30	0.21	0.49	0.20	0.42
27	ethyl acetate	4.4	VIa	0.34	0.23	0.43	0.13	— ^c
28	iodobenzene	2.8	VII	0.24	0.35	0.41	0.19	0.33
29	methylethyl ketone	4.7	VIa	0.35	0.22	0.43	0.13	— ^c
30	di-(2-ethoxy ethyl) ether	4.6	VIa	0.37	0.21	0.43	0.13	0.22
31	anisole	3.8	VII	0.27	0.29	0.43	0.14	0.30
32	n-octanol	3.4	II	0.56	0.18	0.25	0.07	0.22
33	cyclohexanone	4.7	VIa	0.36	0.22	0.42	0.14	0.24
34	i-butanol	4.1	II	0.56	0.20	0.24	0.02	— ^c
35	tetramethyl guanidine	6.1	I	0.47	0.17	0.35	0.11	0.19
36	i-pentanol	3.7	II	0.56	0.19	0.26	0.04	0.23
37	pyridine	5.3	III	0.41	0.22	0.36	0.13	0.24
38	dioxane ^d	4.8	VIa	0.36	0.24	0.40	0.14	0.24
39	n-butanol	3.9	II	0.59	0.19	0.25	0.07	0.24
40	i-propanol	3.9	II	0.55	0.19	0.27	0.03	0.26
41	n-propanol	4.0	II	0.54	0.19	0.27	0.06	0.24
42	phenyl ether	3.4	VII	0.27	0.32	0.41	0.19	0.38
43	acetone	5.1	VIa	0.35	0.23	0.42	0.12	0.28
44	benzonitrile	4.8	VIb	0.31	0.27	0.42	0.15	0.29
45	tetramethyl urea	6.0	III	0.42	0.19	0.39	0.14	0.22
46	benzyl ether	4.1	VII	0.30	0.28	0.42	0.17	0.28
47	acetophenone	4.8	VIa	0.33	0.26	0.41	0.15	0.27

Table II. Comparison of Solvent Parameters for GC Phases Reported by Rohrschneider (5) and McReynolds (6)

Solvent ^a	P'	x_e	x_d	x_n
Diethyl-hexyl sebacate	2.6 (2.8)	0.35 (0.33)	0.22 (0.22)	0.43 (0.45)
Di-isodecyl phthalate	2.8 (2.9)	0.31 (0.31)	0.23 (0.23)	0.46 (0.46)
Tricresyl phosphate	3.9 (4.1)	0.32 (0.33)	0.25 (0.25)	0.42 (0.42)
Carbowax 20M	4.9 (5.3)	0.33 (0.32)	0.41 (0.42)	0.26 (0.26)
Diethyleneglycol succinate	5.9 (6.0)	0.30 (0.31)	0.29 (0.29)	0.41 (0.40)
Tri-cyanoethoxypropane	6.4 (6.7)	0.29 (0.30)	0.29 (0.30)	0.42 (0.40)



^a values for McReynolds solvents given first; values for Rohrschneider solvents (equation 4d) in parentheses.

Figure 1. Comparison of new P' values (Table II) with values of (1).

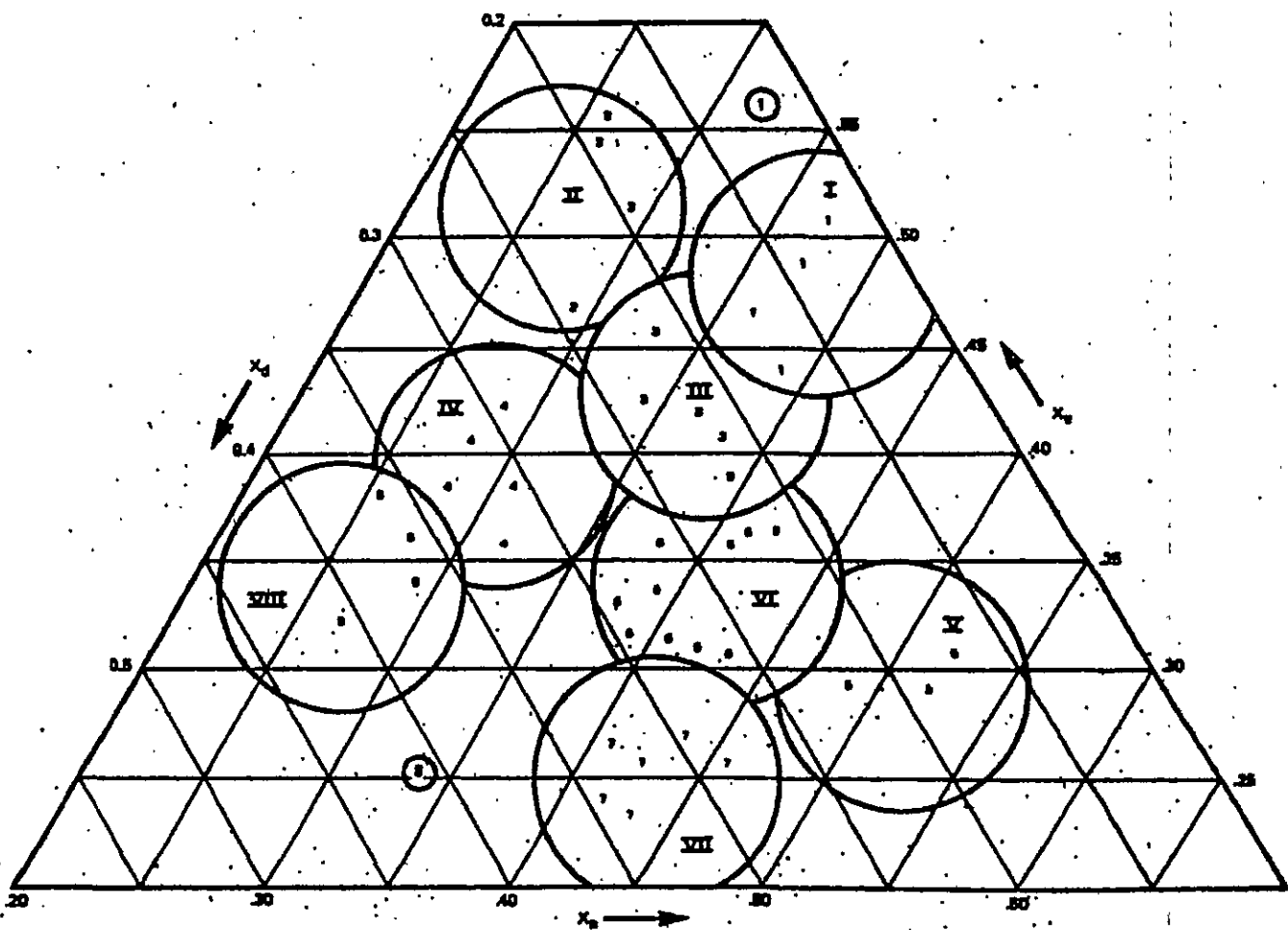


Figure 2. Selectivity grouping of solvents of Table II.

group	average x_m
I	0.18±0.03
III	0.23±0.01
II	0.25±0.02
VIa	0.26±0.01
IV	0.29±0.04
VIb	0.29±0.01
VII	0.31±0.03
VIII	0.36±0.02
V	0.43

* Group VI is subdivided into two adjacent groups (Table IV)

The overall deviation of x_m values from these averages for each group is ±0.02 (1 SD), which is not significantly different from the value of ±0.016 obtained for the solvents of Table V. Therefore it is concluded that the present scheme based on three polar test-solutes adequately characterizes the various solvents in terms of their potential selectivity toward other polar solutes such as methylethyl ketone.

In the case of the relatively nonpolar solute toluene, dispersion interactions should be relatively important—if such interactions are ever important in polar-solvent solutions. The reason is that polar interactions involving the nonpolar toluene as solute are less important, while its higher refractive index and greater electron polarizability provide a greater potential role for dispersion interactions (e.g., 7). Obviously, such dispersion interactions should not correlate with the various polarity indices (x_p , x_d , x_n) of the solvent, while any residual polar interactions would so correlate. Therefore the constancy of x_i values for solvents within a given (polar) selectivity group provides a direct assessment of the importance of dispersion interactions for toluene as solute. The data of Table II provide the following summary groupings:

group	avg. x_i	group	avg. x_i
II	0.05±0.02	IV	0.13±0.03
VIII	0.09±0.03	VI	0.15±0.02
I	0.09±0.03	VII	0.17±0.02
III	0.13±0.02	V	0.20±0.00

As in the case of methylethyl ketone as solute, x_i values for a particular solvent selectivity group are found to be relatively constant. The overall deviation of these x_i values from the above averages is ±0.03 (1 SD), which is only slightly larger than for the polar solute methylethyl ketone (this increase in SD is expected, because of smaller $\log K_p$ and P' values for toluene). This strongly suggests, even in cases which favor a role for dispersion forces in solvent selectivity, that these interactions are relatively unimportant. The predominant solvent-selectivity effects for nonpolar solutes such as toluene dissolved in polar solvents appear to arise as a result of polar interactions.

It is interesting to further examine x_i values within a group of solvents of the same polar selectivity, to see if dispersion interactions play even a secondary role in affecting solvent selectivity. If this were the case, x_i values for such a group of solvents should correlate with solvent refractive index (7). x_i values have been arbitrarily chosen from selectivity group III as a test of this hypothesis, since a large number of solvents

fall within group III, and both solvent refractive index and x_i values vary significantly within this group of solvents. The resulting plot of group III x_i values vs. solvent refractive index is shown in Figure 3. No correlation whatever is apparent, and the author concludes that in the usual chromatographic systems involving polar solvents there is no predictable effect of dispersion interactions or solvent refractive index on solvent selectivity. Why this is so is somewhat puzzling, but it nevertheless seems to be true.

It is possible (but unlikely) that solvent mixtures which include both polar and nonpolar solvents will exhibit some selectivity from the nonpolar solvent as a result of differing dispersion interactions between the nonpolar solvent and the solute. This possibility, which requires experimental study, would then result in a variation of solvent selectivity as the refractive index of the nonpolar solvent component varies.

A further test of the dependence of solvent x_m and x_i values on the three standard x_j values is shown in Figure 4. Here, values of x_m (underlined) and x_i are superimposed on the selectivity triangle of Figure 2. Values of x_m or x_i would be expected to be similar for adjacent selectivity groups, and to change continuously in moving toward one of the three apexes of the triangle (i.e., $x_j = 1.00$). This is observed to be the case, in Figure 4: x_i values generally increase for larger x_p , while x_m values increase for smaller x_p . Thus, the plot in Figure 4 further substantiates the concept that solvent selectivity for all solutes can be adequately represented by the solvent classification based on the three standard x_j values (or the selectivity groups of Table II).

Comparison of Parameters for LC vs. GC Solvents

It appears that P' and x_i values for the various solvents included in both the LC and GC groups are essentially comparable, and therefore calculated values of P' and x_i for a solvent included in either group are identical. This is illustrated in Table VI. The first two entries are for two pure compounds, a nitrile/ether and an aryl phosphate, where values of both P' and x_i can be compared. The remaining entries in Table VI are for compound-type groups, which allow comparison of x_i values only. P' values from the data for LC solvents (first two compounds) are slightly higher than result from the GC solvents, which can be attributed to the use of 25°-data for the LC solvents and 120°-data for the GC solvents. The various x_i values are of greater interest, and agreement among these

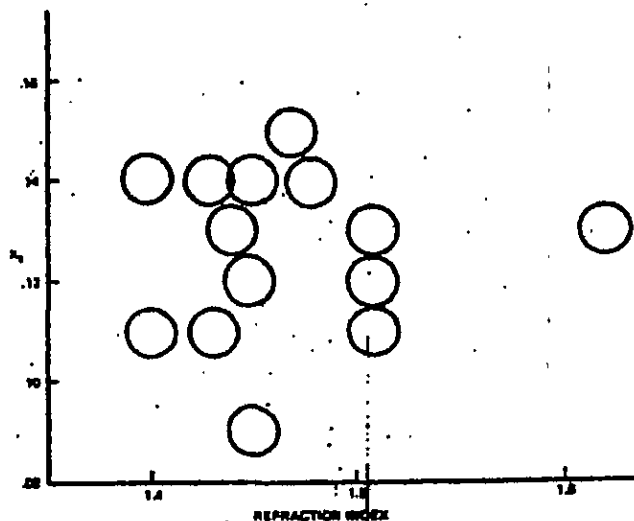


Figure 3. Noncorrelation of toluene selectivity values with solvent refractive index (Group III solvents).

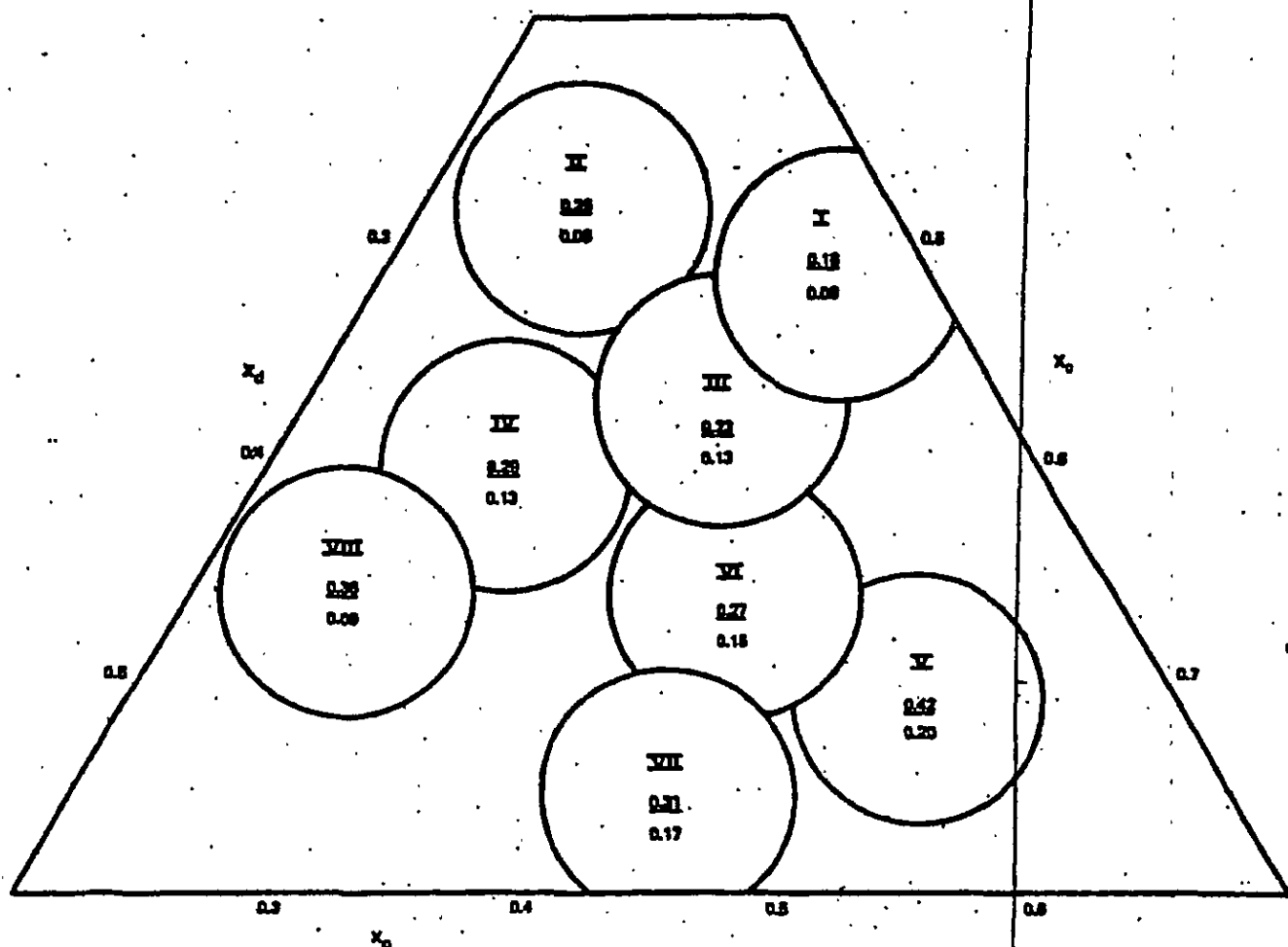


Figure 4. Solvent selectivity toward methyl ethyl ketone (x_H , values underlined) and toluene (x_T); see text.

Table VI. Similarity of Solvent Parameters for Calculation by Either by LC^a or GC^b Procedures

Solvent or solvent type	Solvent parameters								
	GC data ^a				LC data ^b				
	<i>P'</i>	x_e	x_d	x_n	<i>P'</i>	x_e	x_d	x_n	
tri-cyanoethoxy propane	6.4	0.29	0.29	0.42	6.6	0.32	0.27	0.41	
tricresyl phosphate	4.1	0.33	0.25	0.42	4.6	0.36	0.23	0.41	
amides		0.41 ±0.04	0.22 ±0.05	0.37 ±0.03		0.41	0.21	0.38	
fatty acids		0.39	0.30	0.31		0.39	0.30	0.31	
(CH ₂ CH ₂ O) _n		0.34 ±0.02	0.26 ±0.01	0.40 ±0.01		0.37	0.22	0.41	
esters of mono-alcohols		0.33 ±0.02	0.24 ±0.02	0.43 ±0.02		0.34	0.25	0.41	
nitriles		0.30 ±0.01	0.30 ±0.01	0.40 ±0.01		0.31	0.28	0.41	
poly-phenylethers		0.27 ±0.01	0.31 ±0.01	0.42 ±0.02		0.27	0.32	0.41	

^a data of (6); calculated by "GC solvent" procedure

^b data of Tables II or V; calculated by "LC solvent" procedure

Appendix I

Calculation of P' and x_i Values

LC Solvents. A model calculation is provided by 1-octanol as solvent, and is summarized in Table I-I. The K_g values of the various test solutes are shown in line 1, and the molar volumes of these compounds in line 2. The molar volume of the solvent V_s is 158.0 ml. $\log K_g'$ values are listed on line 3, as calculated from equation 1. $\log K_g$ values for each solute are given in line 4, based on equation 3. $\log K_g''$ values from equation 2 appear on line 5. Corrected $\log K_g''$ values (line 6) are obtained by subtracting the $\log K_g'$ values for the C_4 - C_6 hydrocarbons (Table I) from the values of line 5. Now a preliminary value of P' is calculated, equal to the sum of corrected $\log K_g''$ values for ethanol (1.757), dioxane (0.572) and nitromethane (0.785): 3.11. Values of x_i are next calculated:

$$\begin{aligned} x_i(\text{toluene}) &= (\log K_g')_{\text{tol}}/P' \\ &= 0.22/3.11 = 0.07 \\ x_e(\text{ethanol}) &= 1.757/3.11 = 0.56 \\ x_m(\text{methyl ethyl ketone}) &= 0.678/3.11 = 0.22 \\ x_d(\text{dioxane}) &= 0.572/3.11 = 0.18 \\ x_n(\text{nitromethane}) &= 0.785/3.11 = 0.25. \end{aligned}$$

A final value of $P' = 1.1 \times 3.11 = 3.4$ is determined.

Similarly, K for octane (solute) and water (solvent) could be estimated from the solubility of octane in water (1 ppm by weight) and the vapor pressure of pure octane at 25° (15 mm). Resulting values of K for these various solute/solvent combinations were:

solute/solvent	K
dioxane/dioxane	5510
nitromethane/nitromethane	10,300
ethanol/ethanol	5900
octane/water	0.0109

In a few other cases, K values were estimated simply by extrapolation from K values for related compounds (e.g., ethylene chloride vs. methylene chloride).

GC Solvents. An example of the calculation of P' and x_i values using the data of Rohrschneider will be given, for diethyleneglycol succinate as solvent. The calculation is also given for similar data reported by McReynolds (values in parentheses). Values of ΔI for the solutes of interest are summarized in Table I-II, along with values of b . One first sums ΔI values for the three test-solutes (e, d, n): $758 + 712 + 950 = 2420$. In the case of the McReynolds data, the ΔI value for 1-nitropropane is first multiplied by 1.18: $1.18 \times 840 = 991$.

Table I-I. Calculation of P' and x_i Values for LC Solvents of Rohrschneider (1): 1-octanol as example.

	Solute					
	octane (o)	toluene (t)	ethanol (e)	methyl ethyl ketone (m)	dioxane (d)	nitromethane (n)
K_g	2,339	1,895	2,168	639	1,518	411
V_x	163	106.6	58.6	89.9	85.5	53.8
$\log K_g'$	5.568	5.476	5.535	5.004	5.380	4.812
$\log K_g$		3.641	2.004	3.073	2.923	1.837
$\log K_g''$		1.835	3.530	1.930	2.457	2.975
corrected $\log K_g''$		0.22	1.757	0.678	0.572	0.785

In some cases, values of K_g for certain solute/solvent combinations were not reported by Rohrschneider (2). These cases are indicated by "d" or "e" notations in Table II. For some of these solvents with missing data, estimates of the unreported K_g values were possible. Thus, in the case of a solute which is the same as the solvent (e.g., ethanol as solute, ethanol as solvent), values of K_g can be estimated from the vapor pressure of the pure liquid by application of the ideal gas law:

$$\begin{aligned} K &= (g/ml)_g / (g/ml)_l \\ &= d MW P / (24,600 \times 760) \end{aligned} \quad (3-1)$$

Here, $(g/ml)_l$ and $(g/ml)_g$ refer to the concentrations of solute in liquid (l) and gas (g) phases, respectively; d is the density of the pure liquid at 25°, MW is its molecular weight, P is its vapor pressure (mm of Hg) at 25°, and the quantity (24,600) is the volume of a perfect gas (ml) at 25°.

The (e, d, n) sum for the McReynolds data is then: $751 + 725 + 991 = 2467$.

Values of x_i are next calculated, equal to ΔI for the test-solute of interest divided by the latter (e, d, n) sum:

$$\begin{aligned} x_t &= 493/2420 = 0.20 \text{ (Rohrschneider)} \\ x_l &= 499/2467 = 0.20 \text{ (McReynolds)} \end{aligned}$$

Similar calculations for the other x_i values of Table I-II are shown there. Finally, P' values are calculated from equations 4a-c, as the sum of the three (e, d, n) $\log K_g''$ values. This is equivalent to multiplying the above (e, d, n) sums of the ΔI values by $b/100$: $P' = 2420 \times 0.198/100 = 4.79$ (Rohrschneider) and 4.69 (McReynolds). Finally, since the ΔI values are relative to squalane as reference solvent, and P' for squalane is 1.2 (722, Table II), 1.2 is added to the latter values of P' to give final values on the same basis as for the LC solvents: $P' = 6.0$ (Rohrschneider) and 5.9 (McReynolds).

European Patent Office
D-80298 Munich

TELEFAX

From: +41 61 322 75 32
To: 0049 89 2399 4465
Total pages: 3

August 05, 2002

Your Ref:

Our Ref:
PR

PCT / EP01 / 14771 - Priority Date 07/12/2000 - Our Case 4-31672A
Written Opinion of June 10, 2002

Dear Sirs

Thank you for the rationalised Written Opinion of June 10, 2002.

Attached please find new page 22 and page 22 as originally filed with indicated amendments. New claim 12 was introduced and the following claims renumbered. New claim 12 is based on the disclosure in the passage on page 5 running from "A weakly ..." to "... 0,38-0,60; ..."

Based on the Notice of the President of the EPO dated Nov. 2, 2001 concerning rationalisation of the international preliminary examination procedure at the EPO, we herewith request under Rule 66.3 PCT a detailed preliminary examination of the above-identified application acc. to Section II, point 7, of the Notice of the President.

The Search Report completed March 06, 2002 comprises only one citation having the category X, which is WO 99 42602 A published August 26, 2002. In such prior art, only the desorption with a polar solvent is disclosed, whereas the present invention claimed in independent claims 1, 7, 9 and 12 to desorption processes employing a weakly polar or an apolar solvent.

In the name of
Novartis AG



Dr. Peter R. Roth

Professional Representative
GA No. 36671

Encl.:

- new page 22
- page 22 as originally filed with indicated amendments

 *** SENDEBERICHT ***

39

SENDUNG OK

SE/EM NR	0428
RUFNR. GEGENSTELLE	0004988923994485
NEBENADRESSE	
NAME GEGENSTELLE	
ANF. ZEIT	08/08 08:59
ÜB. ZEIT	00'30
SEITEN GEGENDET	3
ERGEBNIS	OK

Dr. Peter R. Roth
 European Patent Attorney

Novartis AG
 CH-4002 Basel
 Switzerland
 Tel: +41 61 324 33 82
 Fax: +41 61 322 75 32

 **NOVARTIS**

European Patent Office
 D-80298 Munich

TELEFAX
 From: +41 61 322 75 32
 To: 0049 89 2399 4465
 Total pages: 3

August 05, 2002

Your Ref:

Our Ref:
 PR

PCT / EP01 / 14771 - Priority Date 07/12/2000 - Our Case 4-31672A
 Written Opinion of June 10, 2002

Dear Sirs

Thank you for the rationalised Written Opinion of June 10, 2002.

Attached please find new page 22 and page 22 as originally filed with indicated amendments. New claim 12 was introduced and the following claims renumbered. New claim 12 is based on the disclosure in the passage on page 5 running from "A weakly ..." to "... 0,38-0,60; ..."

Based on the Notice of the President of the EPO dated Nov. 2, 2001 concerning rationalisation of the international preliminary examination procedure at the EPO, we herewith request under Rule 66.3 PCT a detailed preliminary examination of the above-identified application acc. to Section II, point 7, of the Notice of the President.

The Search Report completed March 06, 2002 comprises only one citation having the category X, which is WO 99 42602 A published August 26, 2002. In such prior art, only the description with a polar solvent is disclosed, whereas the present invention claimed in independent claims 1, 7, 8 and 12 is characterised by a non-polar solvent.

European Patent Office
D-80298 Munich

TELEFAX
From: +41 61 322 75 32
To: 0049 89 2399 4465
Total pages: 9

December 11, 2002

Your Ref:

Our Ref:
PR

PCT/EP01/14771 - Priority Date 07/12/2000 - Our Case 4-31672A

Dear Sirs

Thank you for the Written Opinion under Rule 66 PCT dated September 16, 2002.

Enclosed please find amended retyped pages 20 to 23 that shall replace the pages 20 to 62 on file. In addition, please find enclosed draft pages 20 to 22 with handwritten amendments.

All amendments are to be regarded as suggestions only and do not mean that any subject matter that has been removed from the claims is regarded as being unpatentable.

The following amendments were made:

- (a) At the end of amended claims 5, 6, 9, 10 and 15, the wording "; wherein the prefix "lower" means that the radical contains up to 7 carbon atoms" was added.
- (b) Several non-limiting features were deleted in amended claim 9.
- (c) Amended claim 12 is also depending from claim 1.
- (d) New claims 13 and 16 were introduced.

Comments:

I. Basis for the Amendments / Additions

The amendment under (a) is based on the disclosure on page 6, 3rd paragraph.

New claim 13 is based on the disclosure on page 5, 4th paragraph.

New claim 16 is based on the Examples (epothilones A and B) and the disclosure on page 5, first paragraph.

II. Items 2.2 and 2.3 of the Written Opinion

The amendment cited under (a) above should render moot the objection under 2.2, 2nd sentence. The amendment cited under (b) above should render moot the objection under 2.3.

III. Novelty

The Examiner alleges on Sheet 3 of the Written Opinion that amended claim 12 overlaps with the disclosure of D1 insofar as the alcohol methoxyethanol is concerned (4th full paragraph). The alcohol methoxyethanol is neither mentioned in the present application nor in D1. The Examiner is kindly requested to clarify in more detail why he believes that D1 can be regarded as novelty destroying for amended claim 12.

In the name of
Novartis AG



Dr. Peter R. Roth

Professional Representative
GA No. 36671

Encl.:

- retyped pages 20 to 23 (replacement pages; in single copy, in three copies with the confirmation copy)
- draft pages 20 to 22 with handwritten amendments

*** SENDEBERICHT ***

42

SENDUNG OK

SE/EM NR 1730
RUFNR. GEGENSTELLE 000498923994465
NEBENADRESSE
NAME GEGENSTELLE
ANF. ZEIT 11/12 15:32
UB. ZEIT 01'22
SEITEN GESENDET 9
ERGEBNIS OK

Dr. Peter R. Roth
European Patent Attorney

Novartis AG
CH-4002 Basel
Switzerland
Tel: +41 61 324 33 82
Fax: +41 61 322 75 32

 NOVARTIS

European Patent Office
D-80298 Munich

TELEFAX

From: +41 61 322 75 32
To: 0049 89 2399 4465
Total pages: 9

December 11, 2002

Your Ref:

Our Ref:
PR

PCT/EP01/14771 - Priority Date 07/12/2000 - Our Case 4-31672A

Dear Sirs

Thank you for the Written Opinion under Rule 66 PCT dated September 16, 2002.

Enclosed please find amended retyped pages 20 to 23 that shall replace the pages 20 to 62 on file. In addition, please find enclosed draft pages 20 to 22 with handwritten amendments.

All amendments are to be regarded as suggestions only and do not mean that any subject matter that has been removed from the claims is regarded as being unpatentable.

The following amendments were made:

- (a) At the end of amended claims 5, 6, 9, 10 and 15, the wording "; wherein the prefix "lower" means that the radical contains up to 7 carbon atoms" was added.
- (b) Several non-limiting features were deleted in amended claim 9.

The demand must be filed directly with the competent International Preliminary Examining Authority or, if two or more Authorities are competent, with the one chosen by the applicant. The full name or two-letter code of that Authority may be indicated by the applicant on the line below:

IPEA/ _____

PCT**CHAPTER II****DEMAND**

under Article 31 of the Patent Cooperation Treaty:

The undersigned requests that the international application specified below be the subject of international preliminary examination according to the Patent Cooperation Treaty and hereby elects all eligible States (except where otherwise indicated).

For International Preliminary Examining Authority use only	
Identification of IPEA	Date of receipt of DEMAND
Box No. I IDENTIFICATION OF THE INTERNATIONAL APPLICATION	
Applicant's or agent's file reference 4-31672A	
International application No. PCT/EP 01/14771	International filing date (day/month/year) 05 December 2001 (05.12.01)
(Earliest) Priority date (day/month/year) 07 December 2000 (07.12.00)	
Title of invention PROCESS FOR THE ISOLATION AND PURIFICATION OF EPOTHILONES	
Box No. II APPLICANT(S)	
Name and address: (Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)	
Novartis AG Lichtstrasse 35 4056 Basel CH	
Telephone No. +4161 324 11 11	
Facsimile No. +4161 322 75 32	
Teleprinter No.	
Applicant's registration No. with the Office	
State (that is, country) of nationality: CH	State (that is, country) of residence: CH
Name and address: (Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)	
Novartis-Erfindungen Verwaltungsgesellschaft m.b.H. Brunner Strasse 59 1230 Vienna AT	
State (that is, country) of nationality: AT	State (that is, country) of residence: AT
Name and address: (Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)	
KÜSTERS, Ernst Gallenweilerweg 1 79427 Eschbach DE	
State (that is, country) of nationality: DE	State (that is, country) of residence: DE
☒ Further applicants are indicated on a continuation sheet.	

Continuation of Box No. II APPLICANT(S)

*If none of the following sub-boxes is used, this sheet should not be included in the demand.*Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*UNTERNÄHRER, Heinz
Widhagweg 46
4303 Kaiseraugst
CHState *(that is, country)* of nationality:
CHState *(that is, country)* of residence:
CHName and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*State *(that is, country)* of nationality:State *(that is, country)* of residence:Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*State *(that is, country)* of nationality:State *(that is, country)* of residence:Name and address: *(Family name followed by given name; for a legal entity, full official designation. The address must include postal code and name of country.)*State *(that is, country)* of nationality:State *(that is, country)* of residence:☐ Further applicants are indicated on another continuation sheet.

Box No. III AGENT OR COMMON REPRESENTATIVE; OR ADDRESS FOR CORRESPONDENCE

The following person is ☒ agent ☐ common representativeand ☐ has been appointed earlier and represents the applicant(s) also for international preliminary examination.☐ is hereby appointed and any earlier appointment of (an) agent(s)/common representative is hereby revoked.☒ is hereby appointed, specifically for the procedure before the International Preliminary Examining Authority, in addition to the agent(s)/common representative appointed earlier.Name and address: (Family name followed by given names; for a legal entity, full official designation.
The address must include postal code and name of country.)GROS, Florent
Novartis AG
Corporate Intellectual Property
Patent & Trademark Department
4002 Basel
CH

Telephone No.

+4161 324 11 11

Facsimile No.

+4161 322 75 32

Teleprinter No.

Agent's registration No. with the Office

☐ Address for correspondence: Mark this check-box where no agent or common representative is/has been appointed and the space above is used instead to indicate a special address to which correspondence should be sent.

Box No. IV BASIS FOR INTERNATIONAL PRELIMINARY EXAMINATION

Statement concerning amendments:*

1. The applicant wishes the international preliminary examination to start on the basis of:

☒ the international application as originally filedthe description ☐ as originally filed☐ as amended under Article 34the claims ☐ as originally filed☐ as amended under Article 19 (together with any accompanying statement)☐ as amended under Article 34the drawings ☐ as originally filed☐ as amended under Article 342. ☐ The applicant wishes any amendment to the claims under Article 19 to be considered as reversed.3. ☐ The applicant wishes the start of the international preliminary examination to be postponed until the expiration of 20 months from the priority date unless the International Preliminary Examining Authority receives a copy of any amendments made under Article 19 or a notice from the applicant that he does not wish to make such amendments (Rule 69.1(d)). (This check-box may be marked only where the time limit under Article 19 has not yet expired.)

* Where no check-box is marked, international preliminary examination will start on the basis of the international application as originally filed or, where a copy of amendments to the claims under Article 19 and/or amendments of the international application under Article 34 are received by the International Preliminary Examining Authority before it has begun to draw up a written opinion or the international preliminary examination report, as so amended.

Language for the purposes of international preliminary examination: English☒ which is the language in which the international application was filed.☐ which is the language of a translation furnished for the purposes of international search.☒ which is the language of publication of the international application.☐ which is the language of the translation (to be) furnished for the purposes of international preliminary examination.

Box No. V ELECTION OF STATES

The applicant hereby elects all eligible States (that is, all States which have been designated and which are bound by Chapter II of the PCT)

excluding the following States which the applicant wishes not to elect:

Box No. VI CHECK LIST

The demand is accompanied by the following elements, in the language referred to in Box No. IV, for the purposes of international preliminary examination:

- | | | |
|--|---|--------|
| 1. translation of international application | : | sheets |
| 2. amendments under Article 34 | : | sheets |
| 3. copy (or, where required, translation) of amendments under Article 19 | : | sheets |
| 4. copy (or, where required, translation) of statement under Article 19 | : | sheets |
| 5. letter | : | sheets |
| 6. other (specify) | : | sheets |

For International Preliminary
Examining Authority use only

- | | |
|--------------------------|--------------------------|
| received | not received |
| ☐ | ☐ |
| ☐ | ☐ |
| ☐ | ☐ |
| ☐ | ☐ |
| ☐ | ☐ |
| ☐ | ☐ |

The demand is also accompanied by the item(s) marked below:

- | | |
|--|---|
| 1. ☒ fee calculation sheet | 5. ☐ statement explaining lack of signature |
| 2. ☐ original separate power of attorney | 6. ☐ sequence listing in computer readable form |
| 3. ☐ original general power of attorney | 7. ☒ other (specify): copy of general sub-authorisation |
| 4. ☐ copy of general power of attorney; reference number, if any: | |

Box No. VII SIGNATURE OF APPLICANT, AGENT OR COMMON REPRESENTATIVE

Next to each signature, indicate the name of the person signing and the capacity in which the person signs (if such capacity is not obvious from reading the demand).

In the name of the applicants
The representative



14.05.2002

GROS, Florent AV 36671 + 37586

For International Preliminary Examining Authority use only

1. Date of actual receipt of DEMAND:

2. Adjusted date of receipt of demand due to CORRECTIONS under Rule 60.1(b):

3. ☐ The date of receipt of the demand is AFTER the expiration of 19 months from the priority date and item 4 or 5, below, does not apply.☐ The applicant has been informed accordingly.4. ☐ The date of receipt of the demand is WITHIN the period of 19 months from the priority date as extended by virtue of Rule 80.5.5. ☐ Although the date of receipt of the demand is after the expiration of 19 months from the priority date, the delay in arrival is EXCUSED pursuant to Rule 82.

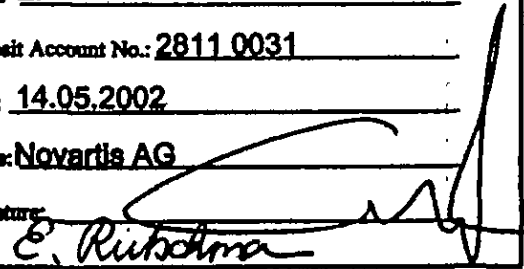
For International Bureau use only

Demand received from IPEA on:

PCT

FEE CALCULATION SHEET

Annex to the Demand

International application No. PCT/EP 01/14771	For International Preliminary Examining Authority use only	
Applicant's or agent's file reference 4-31672A	Date stamp of the IPEA	
Applicant Novartis AG et al.		
CALCULATION OF PRESCRIBED FEES		
1. Preliminary examination fee	EUR 1530.--	P
2. Handling fee (<i>Applicants from certain States are entitled to a reduction of 75% of the handling fee. Where the applicant is (or all applicants are) so entitled, the amount to be entered at H is 25% of the handling fee.</i>)	EUR 159.--	H
3. Total of prescribed fees Add the amounts entered at P and H and enter total in the TOTAL box	EUR 1689.--	
TOTAL		
MODE OF PAYMENT		
☒ authorization to charge deposit account with the IPEA (see below)	☐ cash	
☐ cheque	☐ revenue stamps	
☐ postal money order	☐ coupons	
☐ bank draft	☐ other (specify):	
AUTHORIZATION TO CHARGE (OR CREDIT) DEPOSIT ACCOUNT (This mode of payment may not be available at all IPEAs)		
☒ Authorization to charge the total fees indicated above.	IPEA/ EP	
☒ (This check-box may be marked only if the conditions for deposit accounts of the IPEA so permit) Authorization to charge any deficiency or credit any overpayment in the total fees indicated above.	Deposit Account No.: 2811 0031	
	Date: 14.05.2002	
	Name: Novartis AG	
	Signature: 	

48

TRATADO DE COOPERACION EN MATERIA DE PATENTES

PCT

INFORME EXAMEN PRELIMINAR INTERNACIONAL (PCT artículo 36 y r gla 70)

Referencia del solicitante o agente 4-31672A	PARA PROSEGUIR Ver notificación de la transmisión del examen preliminar internacional (formato PCT/IPEA/416)	
No. Solicitud Internacional PCT/EP01/14771	Fecha de presentación internacional (día/mes/año) 05/12/2001	Fecha de prioridad (día/mes/año) 07/12/2000
Clasificación Internacional de Patentes (IPC) o clasificación nacional e IPC CO7D483/04		
Solicitante NOVARTIS AG et al.		

1. Este examen preliminar internacional ha sido preparado por esta Autoridad encargada del Examen Preliminar Internacional y es transmitida al solicitante de acuerdo con el artículo 36. 2. Este INFORME consiste de un total de 8 páginas, incluyendo esta hoja. <u>X</u> Este informe también está acompañado por ANEXOS, esto es hojas de la descripción, reivindicaciones y/o dibujos, los cuales han sido modificados y son la base para este informe y/o hojas que contienen rectificaciones presentadas ante esta Autoridad (ver Regla 70.16 y Sección 607 de las Instrucciones Administrativas bajo el PCT). Estos anexos consisten en un total de <u>4</u> hojas.	
3. Este informe contiene indicaciones que se relacionan con los siguientes puntos: I. <u>X</u> Base del Informe II. <u> </u> Prioridad III. <u>X</u> El no-establecimiento de opinión en relación con la novedad, nivel inventivo y aplicación industrial IV. <u> </u> Falta de unidad de invención V. <u>X</u> Declaración motivada bajo el Art. 35(2) con relación a la novedad, nivel inventivo o aplicación industrial; citaciones y explicaciones que soportan dicha declaración VI. <u> </u> Ciertos documentos citados VII. <u> </u> Ciertos defectos en la solicitud internacional VIII. <u> </u> Ciertas observaciones en la solicitud internacional	

Fecha de presentación de la demanda 17/05/2002	Fecha en que se completó este informe 14.01.2003
Nombre y dirección de la autoridad encargada del examen preliminar internacional: Oficina de Patentes Europea D-10958 Berlín Tel. +49 89 2399-0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Oficial autorizado WEISBROD, T Teléfono: +49 30 25901 344

49

INFORME DEL EXAMEN PRELIMINAR INTERNACIONAL

Solicitud internacional No. PCT/EP01/14771

I. BASE DEL INFORME

1. En relación con los **elementos** de la solicitud internacional (las hojas de reemplazo que han sido suministradas a la oficina receptora como respuesta a la invitación bajo el artículo 14 serán referidas como "presentadas originalmente" y no están anexas a este informe debido a que no contienen modificaciones (Regla 70.16 y 70.17)):

CAPITULO DESCRIPTIVO, PAGINAS:

1-19 Como fueron presentadas originalmente

REIVINDICACIONES, No.:

1-16 Con telefax del 11/12/2002

2. En relación con el **idioma**, todos los elementos marcados anteriormente fueron suministrados a esta Autoridad en el idioma en que la solicitud internacional fue presentada, a menos que se indique lo contrario bajo este punto.

Estos elementos fueron suministrados o estaban disponibles para esta autoridad en el siguiente idioma, el cual es:

- ☒ El idioma de la traducción aportada a efectos de la búsqueda internacional (bajo la Regla 23.1(b))
- ☒ El idioma de publicación de la solicitud internacional (bajo la Regla 48.3(b))
- ☒ El idioma de la traducción suministrada para efecto del examen preliminar internacional (bajo la Regla 55.2 y/o 55.3)

3. Respecto a cualquier secuencia **nucleótido y/o aminoácido** revelado en la solicitud internacional, el examen preliminar internacional fue llevado a cabo con base en la lista de secuencias:

- ☐ Contenidas en la solicitud internacional de forma escrita
- ☐ Presentadas junto con la solicitud internacional en forma legible por computador
- ☐ Aportada posteriormente a esta Autoridad en forma escrita
- ☐ Suministrada posteriormente a esta Autoridad en forma legible por computador
- ☐ La declaración de que la lista de secuencias aportada posteriormente en forma escrita no va más allá que lo revelado en la solicitud internacional como fue presentada.
- ☐ La declaración de que la información inscrita en forma legible por computador es idéntica a la lista de secuencias en forma escrita aportada.

50

- | | |
|-----------------------------|----------|
| ___ El capítulo descriptivo | páginas: |
| ___ Las reivindicaciones | Nos.: |
| ___ Los dibujos | hojas: |

- (A cualquier hoja de reemplazo que contenga dichas modificaciones debe hacerse referencia bajo el punto 1 y ser anexada a este informe)

- ### III. El no-establecimiento de opinión en relación con la novedad, nivel inventivo y aplicación industrial

- X re/indicaciones Nos. 1, 3-15.

☒ las reivindicaciones Nos. 1.3-15 no han sido objeto de búsqueda internacional.

- ____ la forma legible por computador no ha sido suministrada o no cumple con los estándares.

V. Declaración motivada, bajo el Art. 35(2) con relación a la novedad, nivel inventivo o aplicación industrial; citaciones y explicaciones que sustentan dicha declaración

Declaración

Novedad	SI: Reivindicaciones 5,6,13,15
	NO: Reivindicaciones 1,7,9,12,14,16
Nivel inventivo	SI: Reivindicaciones 5,6,13,15
	NO: Reivindicaciones 1,7,9,12,14,16
Aplicación Industrial	SI: Reivindicaciones 1-16
	NO: Reivindicaciones

2. CITACIONES Y EXPLICACIONES

Referencia Punto I

Base de la opinión

1. Las modificaciones presentadas por FAX el 11.12.2002 cumplen con los requisitos del Art. 19 PCT, debido a que no incluye materia que vaya más allá de la solicitud como fue presentada originalmente.
2. La solicitud está dirigida a
 - (i) un proceso para resorber epotilonas a partir de una resina haciendo este proceso uso de un solvente débilmente polar o apolar (reivindicaciones 1-6, 8, 12-13 y 16);
 - (ii) Un proceso para el aislamiento de epotilonas que comprende
 - a) la desorción de las epotilonas a partir de la resina sintética con un solvente débilmente polar o apolar, y
 - b) la separación de las epotilonas mediante cromatografía de fase inversa con una mezcla de nitrilo y agua (reivindicaciones 7,8, 11-13, y 16).
 - (iii) Un proceso para procesar epotilonas que comprende los pasos de
 - a) desorber las epotilonas con un solvente débilmente polar o apolar,
 - b) remover el solvente utilizado para la desorción mediante la evaporación,
 - c) separar las epotilonas mediante cromatografía de fase inversa y fluir con una mezcla de nitrilo y agua, y remover el nitrilo por evaporación, y
 - d) recristalizar una epotilona purificada (reivindicaciones 9-13 y 16); al igual que el uso
 - (iv) de un solvente débilmente polar o apolar para la desorción de epotilonas a partir de una resina (reivindicaciones 14-16).

Referencia Punto III

El no-establecimiento de opinión en relación con la novedad, nivel inventivo y aplicación industrial

La Autoridad de Búsqueda Internacional (ISA) ha emitido únicamente un informe de búsqueda internacional parcial para las reivindicaciones originales 1 y 3-13, debido a que estas reivindicaciones hacen referencia a métodos o usos que se relacionan con las epotilonas A-F naturales. Para tales reivindicaciones parcialmente buscadas no es posible determinar si, y que alcance tendría, la materia a la cual se refieren dichas reivindicaciones es novedosa y podría contener nivel inventivo. Por lo tanto ningún Examen Internacional Preliminar completo se llevará a cabo con relación a la novedad y nivel inventivo con relación a las reivindicaciones modificadas 1 y 3-15 que no se relacionan con las epotilonas A-F naturales (c.f. Regla 66.1 (e) PCT).

Referencia Punto V

Declaración motivada bajo el Artículo 35(2) con relación a la novedad, nivel inventivo o aplicación industrial; citaciones y explicaciones que sustentan dicha declaración

1. Se hace referencia a los siguientes documentos:

D1: WO-A-99 42602, 26.08.1999
D2: WO-A-93 10121, 17.05.1993.
D3: WO-A-98 22461, 28.05.1998
D4 Nicolaou, K.C. et al. Angew. Chem.Int. Ed. 1998, 37, 81-83.
D5: Nicolaou, K.C. et al. Angew. Chem. Int. Ed. 1998, 37, 2015-2045
D6: Riechardt, C. Solvents and Solvent Effects in Organic Chemistry, 2nd. Ed., VCH, Weinheim 1988, p.60.
D7: Snyder, L.R. J. Chromatogr. Sci. 1978, 16, 223-234

El documento D7 no fue citado en el Informe de Búsqueda Internacional, pero fue mencionado en la memoria descriptiva.

2. La solicitud no cumple con los requerimientos de los Art. 5 y 6 PCT por las siguientes razones:

2.1 Las reivindicaciones 1 y 3-15 se relacionan con procesos que comprenden la desorción de las epotilonas a partir de la resina que se efectúa con un solvente débilmente polar o apolar. Sin embargo, el término epotilona no está definido de forma precisa en el campo técnico. Además de los compuestos principales, esto es, el tiazol de 16 miembros que comprende las macrolidas epotilonas A y B, el término epotilona incluye Inter. Alia macrolidas de 14- a 18- miembros con o sin anillo oxirano templado, los compuestos con configuración modificada de estereo centros o con geometría modificada de enlaces dobles, al igual que compuestos en donde la cadena lateral tiazol ha sido remplazada (ver ejemplo D4 y D5). Dichas reivindicaciones se relacionan por lo tanto a métodos y usos para la desorción de epotilona natural B. En el presente caso, las reivindicaciones no contienen soporte, y la solicitud tampoco.

2.2 El término "solvente débilmente polar" utilizado en las reivindicaciones 1-4, 7-11, 14 y 16 no es claro (ver ejemplos D6, página 60, líneas 5-7).

2.3 El término "inferior" en alquilo inferior etc. no tiene ningún significado preciso con relación al número de átomos de carbono que define tal grupo inferior, por lo tanto resulta que falta claridad en la reivindicación 15.

2.4 La nueva reivindicación 16 está dirigida a la materia de dos categorías, esto es, un proceso y un uso, por lo tanto esta reivindicación no es concisa.

3. Novedad

D1 describe un proceso biotecnológico para la producción de epotilonas en donde el medio de cultivo es llevado a cabo por

- mezclando el centrifugo acuoso con la resina sintética
- desorbiendo las epotilonas de la resina con un solvente polar (especialmente en alcohol, ver página 14, líneas 4-5)
- concentrando la solución resultante si fuera necesario
- separando las epotilonas a través de la cromatografía de fase inversa
- seguida por la recristalización si se desea (ver reivindicaciones 12-14, página 37, ejemplo 3 donde el solvente polar es isopropanol).

El término "solvente débilmente polar" utilizado en las reivindicaciones 1,7,9,14 y 16 de la presente solicitud no distingue inequívocamente los solventes utilizados aquí con los solventes utilizados en D1. Consecuentemente, las reivindicaciones 1,7,9,14 y 16 de la presente solicitud no contienen novedad frente a D1.

Más aún, la reivindicación 12 se relaciona con un proceso en donde el solvente débilmente polar está caracterizado por el parámetro $x_o = 0,20-0,40$; $x_d = 0,17-0,34$; y $x_n = 0,38-0,054$.

Dado que D1 revela un proceso para desorber epotilonas de una resina con un solvente polar, especialmente alcohol, por ejemplo metoxietanol está caracterizado por los parámetros $X_o=0.38$, $X_d=0.24$ y $X_n=0.38$ el proceso de acuerdo con la presente reivindicación 12 sobrelapa con lo revelado en D1 teniendo en lo relacionado con el alcohol metoxietanol.

D2 describe un proceso biotecnológico para la producción de epotilonas A y B, y D3 describe un proceso de manufactura biotecnológica para las epotilonas C-F. Estos procesos comprenden el paso de desorber las epotilonas de una resina absorbente con metanol (D2, página 8, último párrafo; D3, reivindicación 1, paso c). En vista de D2 y D3, la presente materia reivindicada puede ser considerada como novedosa debido a que se refiere a un "solvente débilmente polar" en vez de a metanol para la desorción de epotilonas de la resina.

Los documentos D4 a D7 no aparentan ser relevantes a la pregunta de novedad de la materia reivindicada en la presente solicitud de patente debido a que dichos documentos no revelan la desorción de las epotilonas de una resina.

4. Altura Inventiva

Debido a que la presente solicitud se refiere al proceso o uso novedoso que involucra halogenidas de alquilo inferior, solventes aromáticos al igual que solventes caracterizados por los parámetros de la reivindicación 13 para la desorción de epotilonas de una resina, la siguiente observaciones se aplican a los requerimientos de nivel inventivo.

- 4.1 La solicitud ejemplifica la desorción de epotilona B de Amberlite XAD-16 con tolueno en vez de isopropanol como se ejemplifica en D1 (la presente solicitud: página 13, ejemplo 1; D1, página 37, ejemplo 3). Más aún, la solicitud provee información comparativa para la desorción de epotilona B con tolueno y diclorometano en comparación con isopropanol (páginas 16-18, tabla 1). Los datos muestran que el uso de tolueno como desorbente resulta en un tiempo de desorción menor y en un aumento de epotilona B desorbida comparada con isopropanol (página 17, 10-12 en comparación con 9); y la comparación del diclorometano e isopropanol indica un uso positivo a favor del diclorometano (página 16, 4 en comparación con 3).
- 4.2 En vista de D1, el problema de la presente solicitud puede ser visto en la provisión de un proceso mejorado, alternativo para la desorción de epotilonas de resinas absorbentes. Teniendo en cuenta que los efectos favorables del tolueno y diclorometano sobre la desorción de epotilona B no aparenta ser obvia en vista de los documentos citados como arte previo, se puede reconocer que existe nivel inventivo para el proceso que involucra un halogenida de alquilo inferior, solventes aromáticos o solventes caracterizados por los parámetros de la reivindicación 13 como desorbientes al igual que el uso de estos solventes para la desorción de epotilonas, esto es, las reivindicaciones 5, 6, 13 y 15.

REIVINDICACIONES MODIFICADAS

1. Un proceso para desorber epotilonas a partir de una resina haciendo este proceso uso de un solvente débilmente polar o apolar.
2. El proceso de acuerdo con la reivindicación 1, en donde las epotilonas que se van a desorber son epotilona A ó epotilona B.
3. El proceso de acuerdo con cualquiera de las reivindicaciones 1 ó 2, en donde la resina es una resina sintética.
4. El proceso de acuerdo con la reivindicación 3, en donde la resina es una resina sintética basada en copolímeros de estireno/divinilbenceno.
5. El proceso de acuerdo con cualquiera de las reivindicaciones 1 a 4, en donde el solvente débilmente polar o apolar se selecciona a partir del grupo que consiste en haluros de alquilo inferior y solventes aromáticos, o una mezcla de dos o más de estos solventes; en donde el prefijo "inferior" significa que el radical contiene hasta 7 átomos de carbono.
6. El proceso de acuerdo con la reivindicación 5, en donde se utiliza un solvente aromático seleccionado a partir del grupo que consiste en naftaleno, benceno, o naftaleno o benceno sustituidos por una o más fracciones seleccionadas a partir del grupo que consiste en alquilo inferior, alcóxido inferior, halógeno, nitro, y alcóxido inferior-alquilo inferior; en donde el prefijo "inferior" significa que el radical contiene hasta 7 átomos de

carbón.

7. Un proceso para el aislamiento de epotilonas adsorbidas a una resina sintética, cuyo proceso comprende:

(iii) la desorción de las epotilonas a partir de la resina sintética con un solvente débilmente polar o apolar; y

(iv) la separación de las epotilonas mediante cromatografía de fase inversa después de recuperarse en un solvente adecuado, y la elución con una mezcla de nitrilo y agua.

8. Un proceso de acuerdo con cualquiera de las reivindicaciones 1 a 7, el cual incluye además cualesquiera pasos de purificación adicionales deseables o requeridos para llegar a las epotilonas respectivas en una forma pura.

9. Un proceso para procesar epotilonas a partir de una mezcla de reacción, comprendiendo este proceso:

- desorber las epotilonas con un solvente débilmente polar o apolar, en donde el paso de desorción se puede repetir con el objeto de lograr una desorción más completa;

- remover el solvente utilizado para la desorción a partir de las soluciones resultantes mediante evaporación;

- opcionalmente cristalizar las epotilonas después de la desorción; en especial, para la cristalización de la epotilona B, agregar una mezcla de un alcohol y un hidrocarburo, y evaporar la fase de alcohol a sequedad, y cristalizar la epotilona B a partir de una mezcla de solventes apropiada;

- (paso obligatorio) separar las epotilonas mediante cromatografía de fase inversa, recuperando el residuo a partir del paso anterior en un solvente adecuado, y eluir con una mezcla de nitrilo y agua, y remover el nitrilo de las fracciones que comprenden la epotilona recolectada por evaporación; si se desea, el agua restante con la epotilona se extrae entonces con un éster, con evaporación subsecuente de la fase de éster que contiene epotilona a sequedad;

- opcionalmente, purificar además mediante cromatografía de adsorción;

- y finalmente recrystalizar una epotilona purificada a partir de solventes o mezclas de solventes apropiados;

en cuyo proceso, si es necesario y/o se desea, entre cada caso que se emplee, las soluciones o suspensiones resultantes se concentran, y/o los componentes líquidos y sólidos se separan unos de otros; en donde el prefijo "inferior" significa que el radical contiene hasta 7 átomos de carbono.

10. Un proceso de acuerdo con la reivindicación 9, en donde la desorción de las epotilonas a partir de la resina se efectúa con un solvente débilmente polar o apolar, en especial como se mencionó como preferido en la reivindicación 5 ó 6; luego se remueve el solvente tanto como sea necesario, de preferencia hasta que se obtenga un residuo seco, donde sea apropiado, el residuo se recupera en una mezcla de alcohol/hidrocarburo en un volumen relativamente bajo; la fase de alcohol se evapora, de

preferencia a sequedad, y luego se cristaliza el extracto de alcohol a partir de una mezcla de un alcohol y un hidrocarburo, y luego el material cristalizado sólido resultante se disuelve en una mezcla de nitrilo/agua como se describe en la reivindicación 8, especialmente una mezcla de 2:3 (volumen/volumen) de acetonitrilo/agua, y la solución de alimentación resultante se vierte, si se requiere, después de dividir en más de una pasada, sobre la columna de fase inversa de preparación; seguido por elución con nitrilo/agua; luego sigue la remoción de nitrilo de las fracciones resultantes que contienen epotilona por evaporación, y se extrae la fase de agua resultante con un éster; entonces se evapora el extracto de éster, y subsecuentemente se recrystaliza el material resultante; en donde el prefijo "inferior" significa que el radical contiene hasta 7 átomos de carbono.

11. Un proceso de acuerdo con cualquiera de las reivindicaciones 7, 9, ó 10, en donde la separación de las epotilonas A y B se logra mediante cromatografía de lecho en movimiento simulado (SMB).

12. Un proceso de acuerdo con cualquiera de las reivindicaciones 1, 7, 9 ó 10, en donde el solvente débilmente polar o apolar, o una mezcla de estos solventes, muestra las siguientes características en la fila eluotrópica, determinadas de acuerdo con Snyder y colaboradores:

$x_e = 0,20-0,40$; $x_d = 0,15-0,36$; y $x_n = 0,38-0,60$;
en donde x_e es el parámetro del aceptor de protones, x_d es el parámetro del donador de protones, y x_n es el parámetro

de dipolo, con la condición de que $x_e + x_d + x_n = 1$.

13. Un proceso de acuerdo con la reivindicación 12, en donde el solvente débilmente polar o apolar, o una mezcla de estos solventes, muestra las siguientes características en la fila eluotrópica, determinadas de acuerdo con Snyder y colaboradores:

$$x_e = 0,22-0,32; x_d = 0.17-0.34; \text{ y } x_n = 0,39-0.54$$

en donde x_e es el parámetro del aceptor de protones, x_d es el parámetro del donador de protones, y x_n es el parámetro de dipolo, con la condición de que $x_e + x_d + x_n = 1$.

14. El uso de un solvente débilmente polar o apolar para la desorción de epotilonas a partir de una resina.

15. El uso de acuerdo con la reivindicación 14, en donde el solvente débilmente polar o apolar se selecciona a partir del grupo que consiste en haluros de alquilo inferior y solventes aromáticos, en especial naftaleno, benceno, o naftaleno o benceno sustituidos por una o más fracciones seleccionadas a partir del grupo que consiste en alquilo inferior, alcoxilo inferior, halógeno, nitro y alcoxilo inferior-alquilo inferior; o una mezcla de dos o más de estos solventes; y la resina es una resina sintética.

16. Un proceso de acuerdo con cualquiera de las reivindicaciones 1 ó 3 a 13, o un uso de acuerdo con la reivindicación 14 ó 15, en donde las epotilonas se seleccionan a partir de las epotilonas A, B, D, E, y F.

due 7/4/03

PATENT COOPERATION TREATY

2562

557

Corporate Intellectual Property				
16. Jan. 2003 59				
APPL.	M/O	F/L	PS	Copy:

PCT

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

To:
GROSS, Florent NOVARTIS AG Corporate Intellectual Property Patent & Trademark Department CH-4002 Basel SUISSE

NOTIFICATION OF TRANSMITTAL OF
THE INTERNATIONAL PRELIMINARY
EXAMINATION REPORT
(PCT Rule 71.1)

Date of mailing (day/month/year)	14.01.2003
-------------------------------------	------------

Applicant's or agent's file reference 4-31672A

IMPORTANT NOTIFICATION

International application No. PCT/EP01/14771	International filing date (day/month/year) 05/12/2001	Priority date (day/month/year) 07/12/2000
---	--	--

Applicant NOVARTIS AG ET AL.

1. The applicant is hereby notified that this International Preliminary Examining Authority transmits herewith the international preliminary examination report and its annexes, if any, established on the international application.
2. A copy of the report and its annexes, if any, is being transmitted to the International Bureau for communication to all the elected Offices.
3. Where required by any of the elected Offices, the International Bureau will prepare an English translation of the report (but not of any annexes) and will transmit such translation to those Offices.

4. REMINDER

The applicant must enter the national phase before each elected Office by performing certain acts (filing translations and paying national fees) within 30 months from the priority date (or later in some Offices) (Article 39(1)) (see also the reminder sent by the International Bureau with Form PCT/IB/301).

Where a translation of the international application must be furnished to an elected Office, that translation must contain a translation of any annexes to the international preliminary examination report. It is the applicant's responsibility to prepare and furnish such translation directly to each elected Office concerned.

For further details on the applicable time limits and requirements of the elected Offices, see Volume II of the PCT Applicant's Guide.

For the purpose of deciding whether the claimed invention is patentable or not, the elected Offices may apply criteria additional to or different from the criteria on which the international preliminary examination report is based (see Articles 27(5), 33(5)). Additional criteria may include e.g. exemptions from patentability and the requirements of enabling disclosure and of clarity and support of claims.

Name and mailing address of the IPEA/  European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tlx 523658 epmu d Fax: +49 89 2399 - 4465	Authorized officer Ulrich, J Tel. +49 89 2399-9048 
--	---

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 4-31672A	FOR FURTHER ACTION See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)	
International application No. PCT/EP01/14771	International filing date (day/month/year) 05/12/2001	Priority date (day/month/year) 07/12/2000
International Patent Classification (IPC) or national classification and IPC C07D493/04		
Applicant NOVARTIS AG ET AL.		
1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36. 2. This REPORT consists of a total of 8 sheets, including this cover sheet. ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT). These annexes consist of a total of 4 sheets.		
3. This report contains indications relating to the following items: - I ☒ Basis of the report - II ☐ Priority - III ☒ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability - IV ☐ Lack of unity of invention - V ☒ Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement - VI ☐ Certain documents cited - VII ☐ Certain defects in the international application - VIII ☐ Certain observations on the international application		
Date of submission of the demand 17/05/2002	Date of completion of this report 14.01.2003	
Name and mailing address of the international preliminary examining authority:  European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized officer Weisbrod, T Telephone No. +49 89 2399 8931 	

62
61

**INTERNATIONAL PRELIMINARY
EXAMINATION REPORT**

International application No. PCT/EP01/14771

I. Basis of the report

1. With regard to the elements of the international application (*Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17)*):

Description, pages:

1-19 as originally filed

Claims, No.:

1-16 with telefax of 11/12/2002

2. With regard to the language, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

These elements were available or furnished to this Authority in the following language: , which is:

- ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).
☐ the language of publication of the international application (under Rule 48.3(b)).
☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

- ☐ contained in the international application in written form.
☐ filed together with the international application in computer readable form.
☐ furnished subsequently to this Authority in written form.
☐ furnished subsequently to this Authority in computer readable form.
☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.
☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

- ☐ the description, pages:
☐ the claims, Nos.:
☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)):

**INTERNATIONAL PRELIMINARY
EXAMINATION REPORT**

International application No. PCT/EP01/14771

(Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

III. Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non-obvious), or to be industrially applicable have not been examined in respect of:

☐ the entire international application.

☒ claims Nos. 1,3-15 (all part).

because:

☐ the said international application, or the said claims Nos. relate to the following subject matter which does not require an international preliminary examination (*specify*):

☐ the description, claims or drawings (*indicate particular elements below*) or said claims Nos. are so unclear that no meaningful opinion could be formed (*specify*):

☐ the claims, or said claims Nos. are so inadequately supported by the description that no meaningful opinion could be formed.

☒ no international search report has been established for the said claims Nos. 1,3-15 (all part).

2. A meaningful international preliminary examination cannot be carried out due to the failure of the nucleotide and/or amino acid sequence listing to comply with the standard provided for in Annex C of the Administrative Instructions:

☐ the written form has not been furnished or does not comply with the standard.

☐ the computer readable form has not been furnished or does not comply with the standard.

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	5,6,13,15
	No:	Claims	1,7,9,12,14,16
Inventive step (IS)	Yes:	Claims	5,6,13,15
	No:	Claims	1,7,9,12,14,16
Industrial applicability (IA)	Yes:	Claims	1-16

**INTERNATIONAL PRELIMINARY
EXAMINATION REPORT**

International application No. PCT/EP01/14771

No: Claims

2. Citations and explanations
see separate sheet

65
64

Re Item I

Basis of the opinion

- 1 The amendments filed by FAX of 11.12.2002 comply with the requirements of Article 19 PCT, since they do not introduce subject-matter which extends beyond the application as originally filed.
- 2 The application is directed to
 - (i) a process for desorbing epothilones from a resin, said process making use of a weakly polar or apolar solvent (claims 1-6, 8, 12-13 and 16);
 - (ii) a process for the isolation of epothilones comprising
 - (a) the desorption of the epothilones from a synthetic resin with a weakly polar or apolar solvent, and
 - (b) separation of the epothilones by reversed-phase chromatography with a nitrile/water mixture (claims 7, 8, 11-13, and 16);
 - (iii) a process for working up epothilones comprising the steps of
 - (a) desorbing the epothilones with a weakly polar or apolar solvent,
 - (b) removing the solvent used for desorption by evaporation,
 - (c) separating the epothilones by reversed-phase chromatography by eluting with a nitrile/water mixture, and removing the nitrile by evaporation, and
 - (d) recrystallizing a purified epothilone (claims 9-13 and 16); as well as
 - (iv) the use of a weakly polar or apolar solvent for the desorption of epothilones from a resin (claims 14-16).

Re Item III

Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

The ISA has issued only a partial search report for the original claims 1 and 3-13, insofar as these claims concern methods or uses which relate to the natural epothilones A-F. For such partially searched claims it is not possible to determine whether, and if so to what extent, the matter to which said claims relate is novel and might involve an inventive step. No full International Preliminary Examination will thus be carried out with regard to novelty and inventive step insofar as the amended claims

66
65

1 and 3-15 do not relate to the natural epothilones A-F (cf. Rule 66.1(e) PCT).

Re Item V

Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1 Reference is made to the following documents:

D1: WO-A-99 42602, 26.08.1999.

D2: WO-A-93 10121, 17.05.1993.

D3: WO-A-98 22461, 28.05.1998.

D4: Nicolaou, K. C. *et al. Angew. Chem. Int. Ed.* 1998, 37, 81-83.

D5: Nicolaou, K. C. *et al. Angew. Chem. Int. Ed.* 1998, 37, 2015-2045.

D6: Reichardt, C. *Solvents and Solvent Effects in Organic Chemistry*, 2nd. ed., VCH, Weinheim 1988, p. 60.

D7: Snyder, L. R. *J. Chromatogr. Sci.* 1978, 16, 223-234.

The document D7 has not been cited in the International search report, but is mentioned in the application.

2 The application does not comply with the requirements of the Articles 5 and 6 PCT for the following reasons.

2.1 Present claims 1 and 3-15 relate to processes comprising desorbing epothilones from a resin respectively the use of a weakly polar or apolar solvent for desorbing epothilones from a resin. The term epothilone, however, is not precisely defined in the technical field. Along with the parent compounds, i.e. the 16-membered thiazole-comprising macrolides epothilone A and B, the term epothilone includes inter alia 14- to 18-membered macrolides with or without annelated oxirane ring, compounds with modified configuration of stereocentres or modified geometry of double bonds, as well as compounds wherein the side-chain thiazole has been replaced (see e.g. D4 and D5). The said claims relate thus to methods and uses for the desorption of an extremely large number of diverse possible compounds. Support within the meaning of Article 6 PCT and/or disclosure within the meaning of Article 5 PCT is to be found, however, for only a very small proportion of the claimed methods and uses i.e. for the desorption of natural epothilone B (cf. examples). In the present case, the claims so lack support, and the application so

lacks disclosure.

- 2.2 The vague term "weakly polar solvent" used in the claims renders the claims 1-4, 7-11, 14 and 16 unclear (see e.g. D6, page 60, lines 5-7).
- 2.3 The term "lower" in lower alkyl etc. has no precise meaning with regard to the number of carbon atoms which define such lower groups, thereby resulting in a lack of clarity of claim 15.
- 2.4 New claim 16 is directed to subject matter of two categories i.e. a process and a use, thereby resulting in a lack of conciseness of the claim.

3 Novelty

D1 describes a biotechnological process for the production of epothilones wherein the culture medium is worked-up by

- (a) mixing the aqueous centrifugate with a synthetic resin,
- (b) desorbing the epothilones from the resin with a polar solvent (especially an alcohol, cf. page 14, lines 4-5)
- (c) concentrating the resulting solution if necessary,
- (d) separating the epothilones by reversed-phase chromatography,
- (e) followed by recrystallization if desired (cf. claims 12-14; and page 37, example 3 wherein the polar solvent is isopropanol).

The term "weakly polar solvent" used in present claims 1, 7, 9, 14 and 16 does not unequivocally distinguish the present used solvents from the polar solvents used in D1. Consequently, present claims 1, 7, 9, 14 and 16 lack novelty in view of D1.

Furthermore, claim 12 relates to a process wherein the weakly polar solvent is characterized by the parameters $x_o = 0.20-0.40$, $x_d = 0.17-0.34$ and $x_n = 0.38-0.54$. As D1 discloses a process for desorbing epothilones from a resin with a polar solvent, especially an alcohol (cf. page 14, lines 4-5) and, according to D7, one such alcohol i.e. methoxyethanol is characterized by the parameters $x_o = 0.38$, $x_d = 0.24$ and $x_n = 0.38$ the process according to present claim 12 overlaps with the disclosure of D1 insofar as the alcohol methoxyethanol is concerned. Claim 12 lacks thus novelty in view of D1.

D2 describes a biotechnological process for the production of epothilones A and

68
67

B, and D3 a biotechnological manufacture process for the epothilones C-F. These processes comprise the step of desorbing the epothilones from an absorber resin with methanol (D2, page 8, last paragraph; D3, claim 1, step c). In view of D2 and D3 the present claimed matter may be regarded novel since it refers to a "weak polar solvent" instead of methanol for the desorption of epothilones from a resin.

The documents D4 to D7 do not appear relevant to the question of novelty of the present claimed matter, since the said documents do not disclose the desorption of epothilones from a resin.

4 Inventive Step

Insofar as the application relates to novel processes or uses involving lower alkyl halogenides, aromatic solvents as well as solvents characterized by the parameters of claim 13 for desorbing epothilones from a resin, the following observations apply to the requirements of inventive step.

- 4.1 The application exemplifies the desorption of epothilone B from Amberlite XAD-16 with toluene instead of isopropanol as exemplified in D1 (present application: page 13, example 1; D1, page 37, example 3). The application furthermore provides comparative data for the desorption of epothilone B with toluene and dichloromethane in comparison with isopropanol (pages 16-18, table 1). The data show that the use of toluene as desorbant results in a decreased desorption time and increased yield of desorbed epothilone B compared with isopropanol (cf. page 17, runs 10-12 in comparison with run 9); and the comparison of the dichloromethane and isopropanol runs indicate a positive trend in favour of dichloromethane (cf. page 16, run 4 in comparison with run 3).
- 4.2 In view of D1 the problem underlying the present application may be seen in the provision of an alternative, improved, process for the desorption of epothilones from absorber resins. Since the favourable effects of toluene and dichloromethane on the desorption of epothilone B does not appear obvious in view of the cited prior art, an inventive step may be acknowledged for processes involving lower alkyl halogenide, aromatic solvents or solvents characterized by the parameters of claim 13 as desorbants as well as for the use of these solvents for the desorption of epothilones i.e. the claims 5, 6, 13 and 15.

What is claimed is:

1. A process for desorbing epothilones from a resin, said process making use of a weakly polar or apolar solvent.
2. The process according to claim 1 wherein the epothilones to be desorbed are epothilone A or epothilone B.
3. The process according to any one of claims 1 or 2 wherein the resin is a synthetic resin.
4. The process according to claim 3 wherein the resin is a synthetic resin based on styrene/divinylbenzene copolymers.
5. The process according to any one of claims 1 to 4 wherein the weakly polar or apolar solvent is selected from the group consisting of lower alkyl halogenides and aromatic solvents or a mixture of two or more such solvents; wherein the prefix "lower" means that the radical contains up to 7 carbon atoms.
6. The process according to claim 5, wherein an aromatic solvent is used selected from the group consisting of naphthalene, benzene, or naphthalene or benzene substituted by one or more moieties selected from the group consisting of lower alkyl, lower alkoxy, halogen, nitro and lower alkoxy-lower alkyl; wherein the prefix "lower" means that the radical contains up to 7 carbon atoms.
7. A process for the isolation of epothilones adsorbed to a synthetic resin which process comprises
 - (iii) the desorption of the epothilones from said synthetic resin with a weakly polar or apolar solvent; and
 - (iv) separation of the epothilones by reversed-phase chromatography after being taken up in a suitable solvent and elution with a mixture of nitrile and water.
8. A process according to any one of claims 1 to 7, further including any further purification steps desirable or required to come to the respective epothilones in pure form.
9. A process of working up epothilones from a reaction mixture, said process comprising
 - desorbing the epothilones with a weakly polar or apolar solvent, where the step of desorption may be repeated in order to achieve more complete desorption;

AMENDED SHEET

70
69

- removing the solvent used for desorption from the resulting solutions by evaporation;
- optionally crystallizing the epothilone(s) after desorption; especially, for the crystallization of epothilone B, adding a mixture of an alcohol and a hydrocarbon and evaporating the alcohol phase to dryness, and crystallizing the epothilone B from an appropriate solvent mixture;
- (obligatory step) separating the epothilones by reversed-phase chromatography by taking up the residue from the preceding step in a suitable solvent and eluting with a mixture of nitrile and water and removing the nitrile from the collected epothilone comprising fractions by evaporation; if desired, the remaining water with the epothilone is then extracted with an ester with subsequent evaporation of the epothilone containing ester phase to dryness;
- optionally further purifying by adsorption chromatography;
- and finally recrystallizing a purified epothilone from appropriate solvents or solvent mixtures;

in which process, if necessary and/or desired, between each step being employed, the resulting solutions or suspensions are concentrated, and/or liquid and solid components are separated from one another; wherein the prefix "lower" means that the radical contains up to 7 carbon atoms.

10. A process according to claim 9, where desorption of the epothilones from the resin is effected with a weakly polar or apolar solvent, especially as mentioned as being preferred in claim 5 or 6; the solvent is then removed as far as necessary, preferably until a dry residue is obtained, where appropriate, the residue is taken up in an alcohol/hydrocarbon mixture in a relatively low volume; the alcohol phase is evaporated, preferably to dryness, and the alcohol extract is then crystallized from a mixture of an alcohol and a hydrocarbon the resulting solid crystallized material is then dissolved in a nitrile/water mixture as described in claim 8, especially a 2:3 (v/v) acetonitrile/water mixture, and the resulting

AMENDED SHEET

Enforcement of the Patent

71
70

feed solution is poured, if required, after splitting in more than one run, onto the preparative reversed phase column; followed by elution with nitrile/water; then removal of the nitrile of the resulting fractions containing epothilone by evaporation follows and the resulting water phase is extracted with an ester; said ester extract is then evaporated and subsequently the resulting material is recrystallized; wherein the prefix "lower" means that the radical contains up to 7 carbon atoms.

11. A process according to any one of claims 7, 9 or 10 wherein the separation of epothilone A and B is achieved by simulated moving bed (SMB) chromatography.
12. A process according to any one of claims 1, 7, 9 or 10 wherein the weakly polar or apolar solvent or a mixture of such solvents shows the following characteristics in the elutropic row determined according to Snyder et al.:

$$x_a = 0,20-0,40; x_d = 0,15-0,36; \text{ and } x_n = 0,38-0,60$$

where x_a is the proton acceptor parameter, x_d is the proton donor parameter and x_n is the dipole parameter, with the proviso that $x_a + x_d + x_n = 1$.

13. A process according to claim 12 wherein the weakly polar or apolar solvent or a mixture of such solvents shows the following characteristics in the elutropic row determined according to Snyder et al.:

$$x_a = 0,22-0,32; x_d = 0,17-0,34; \text{ and } x_n = 0,39-0,54$$

where x_a is the proton acceptor parameter, x_d is the proton donor parameter and x_n is the dipole parameter, with the proviso that $x_a + x_d + x_n = 1$

14. The use of a weakly polar or apolar solvent for the desorption of epothilones from a resin.
15. The use according to claim 14, wherein the weakly polar or apolar solvent is selected from the group consisting of lower alkyl halogenides and aromatic solvents, especially naphthalene, benzene or naphthalene or benzene substituted by one or more moieties selected from the group consisting of lower alkyl, lower alkoxy, halogen, nitro and lower

AMENDED SHEET
End of page 12

-12-200211672A

- 23 -

alkoxy-lower alkyl; or a mixture of two or more such solvents; the resin is a synthetic resin.

16. A process according to any one of claims 1 or 3 to 13 or a use according to claim 14 or 15, wherein the epothilones are selected from the epothilones A, B, D, E and F.

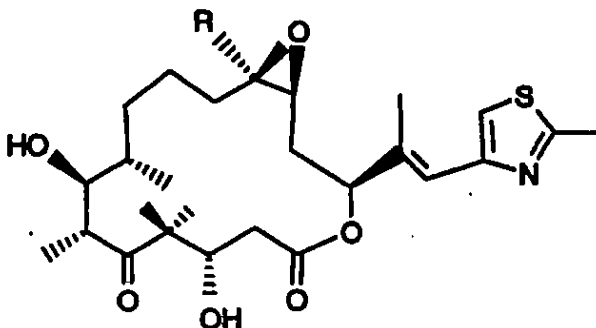
Empfangs AMENDED SHEET

PROCESO PARA EL AISLAMIENTO Y PURIFICACION DE EPOTILONAS

La invención se refiere a un nuevo proceso de desorción para epotilonas, especialmente epotilona A y/o epotilona B, a partir de resinas, y a nuevos procesos de producción, procesamiento, o purificación, o métodos de fabricación para epotilonas que comprenden dicho proceso de desorción, así como al uso de ciertos solventes para la desorción de epotilonas a partir de resinas.

Antecedentes de la Invención:

Las epotilonas A y B representan una nueva clase de ingredientes activos citotóxicos estabilizantes de microtúbulos (ver Gerth, K. y colaboradores, J. Antibiot. 49, 560-3 (1966)) de la fórmula:



en donde R significa hidrógeno (epotilona A) o metilo (epotilona B).

Desde de la descripción de estas epotilonas (ver la
Publicación Internacional Número WO 93/10121), se han dado a
conocer varios métodos para la síntesis y fabricación de
ambas epotilonas, y principalmente de numerosos derivados de
5 epotilona (colectivamente denominadas "epotilonas"
posteriormente en la presente), por ejemplo las dadas a
conocer en las Patentes Números WO 99/03848, WO 00/49020, WO
00/49021, WO 00/47584, WO 00/00485, WO 00/23452, WO 99/03848,
WO 00/49019, WO 99/07692, WO 98/22461, WO 99/65913, WO
10 98/38192, WO 00/50423, WO 00/22139, WO 99/58534, WO 97/19086,
WO 98/25929, WO 99/67252, WO 99/67253, WO 00/31247, WO
99/42602, WO 99/28324, WO 00/50423, WO 00/39276, WO 99/27890,
WO 99/54319, WO 99/54318, WO 99/02514, WO 99/59985, WO
00/37473, WO 98/08849, US 6,043,372, US 5,969,145, WO
15 99/40047, WO 99/01124 y WO 99/43653. En adición a la
epotilona A y la epotilona B, especialmente las epotilonas D
y E, descritas en las Publicaciones Internacionales Números
WO 97/19086 y WO 98/22461, y las epotilonas E y F, descritas
en la Publicación Internacional Número WO 98/22461, así como
20 las epotilonas descritas en la Publicación Internacional
Número WO 99/02514 muestran propiedades interesantes.

Como un ejemplo para el uso terapéutico, la
Solicitud Internacional Número WO 99/43320 describe un número
de modos de administración de epotilonas como agentes contra
25 enfermedades proliferativas, en especial enfermedades

tumorales, que, debido a su mecanismo similar, actúan de una manera comparable a la del Taxol®, un agente contra el cáncer bien conocido y comercializado. La Publicación Internacional Número WO 99/39694 da a conocer algunas formulaciones específicas de las epotilonas, en especial A y B.

Las epotilonas, especialmente la epotilona A, y más preferiblemente la epotilona B, ofrecen un número de ventajas en comparación con los tratamientos establecidos, en especial también en los casos en donde los tumores han llegado a ser refractarios al tratamiento con Taxol. Por consiguiente, se requieren urgentemente métodos para su síntesis en mayores cantidades con el objeto de satisfacer las demandas anticipadas.

Los procesos de producción más eficientes hasta ahora comprenden cuando menos algunos pasos biosintéticos y el aislamiento de las epotilonas a partir de un medio de cultivo o similar.

Originalmente, ya se describió en la literatura la extracción de sustancias naturales por medio mixobacterias, especialmente las epotilonas a partir de la cepa celular *Sorangium Cellulosum* Soce90 (depositada bajo el número 6773 en la German Collection of Microorganisms, ver la Publicación Internacional Número WO 93/10121). Con el objeto de obtener una concentración satisfactoria de las sustancias naturales, en especial las epotilonas, anteriormente siempre se agregaba

una resina de adsorbato basada en poliestireno al medio de cultivo para la adsorción al medio, por ejemplo Amberlite XAD-1180 (Rohm & Haas, Frankfurt, Alemania).

Sin embargo, el inconveniente de este proceso es que, a una gran escala, conduce a una abundancia de problemas. Las válvulas se perjudican por los glóbulos de la resina, los conductos pueden bloquearse, y el aparato se puede someter a un mayor desgaste debido a la fricción mecánica. Los glóbulos de resina son porosos, y por consiguiente, tienen una gran área superficial interna (aproximadamente 825 metros cuadrados/gramo de resina). La esterilización llega a ser un problema, debido a que no se pasa por autoclave el aire encerrado en la resina. Por consiguiente, el proceso no se puede realizar prácticamente a una gran escala utilizando adición de resina durante el cultivo de los microorganismos que producen las epotilonas.

Por consiguiente, se descubrió un proceso avanzado para la producción de epotilonas, especialmente de epotilonas A y B, y se describe en la Publicación Internacional Número 99/42602. Este método comprende formar complejo de las epotilonas a partir de un medio de cultivo de microorganismos productores de epotilonas, comprendiendo este medio ciclodextrinas u otros agentes formadores de complejos, mezclar el medio de cultivo exento de células (por ejemplo el filtrado o centrifugado del medio de cultivo) con una resina

sintética, por ejemplo una resina basada en copolímeros de estireno/divinilbenceno como matriz, tal como Amberlite XAD-16 (Rohm & Haas Germany GmbH, Frankfurt, Alemania) o Diaion HP-20 (Resindion S.R.L., Mitsubishi Chemical Co., Milán, Italia) con el objeto de absorber las epotilonas, y la desorción, en especial con un alcohol, en su mayor parte de preferencia isopropanol. Esto es seguido por la adición de agua a la fase de alcohol, la remoción de la fase de solvente (de preferencia por evaporación), la separación de fases del residuo resultante en la presencia de un éster, especialmente acetato de etilo o acetato de isopropilo, usualmente filtración molecular (cromatografía en gel) de la fase de éster secada, la separación de la mezcla de epotilonas resultante mediante HPLC de fase inversa (de preferencia mediante elución con una mezcla de nitrilo/agua, por ejemplo acetonitrilo/agua), y opcionalmente la purificación adicional mediante separación de fases en la presencia de una mezcla de agua/éter, de preferencia la subsecuente cromatografía de adsorción sobre gel de sílice, con el objeto de lograr la remoción adicional de impurezas, y la cristalización/recristalización.

Aunque es un progreso útil y apropiado para la producción a escala industrial, es método todavía sufre de ciertas desventajas.

Por ejemplo, con el objeto de obtener suficiente

pureza, es recomendable hacer uso del paso de filtración molecular, o bien del paso de cromatografía por adsorción de gel de sílice, o ambos. Vienen más dificultades de la separación de fases en la presencia de un éster, tal como acetato de etilo, lo cual (especialmente debido al largo tiempo para la separación de fases de la fase de agua/éster a gran escala industrial) es muy tardado, así como la subsecuente evaporación, que en adición es difícil de manejar en vista de la espumación y el chisporroteo.

Por consiguiente, éste es un problema que debe ser resuelto por la presente invención para evitar muchas de las dificultades anteriores tanto como sea posible, y encontrar nuevas vías convenientes para el aislamiento de las epotilonas, en especial de las epotilonas A y B, después de su adsorción a una resina.

Descripción General de la Invención

De una manera muy sorprendente, ahora se ha encontrado que mediante el simple reemplazo de los alcoholes utilizados como solventes de desorción con otros solventes determinados (denominados solventes débilmente polares o apolares, como se especifica más adelante), se proporciona un descubrimiento real que se puede lograr con el objeto de obtener una solución a los problemas mencionados anteriormente, conduciendo a ventajas adicionales, tales como

una mejor desorción y un rendimiento final más alto. Entre las ventajas adicionales están: (i) más alta selectividad de la desorción; (ii) más alta cantidad de epotilonas desorbidas, indicando una desorción más completa; (iii) no hay reextracción con éster (por ejemplo acetato de etilo), con la altamente problemática separación de fases de agua/éster, y no hay la subsecuente evaporación de éter requerida; (iv) no hay filtración molecular, y usualmente no se requiere cromatografía de adsorción; (v) se reduce considerablemente el tiempo requerido para la desorción; (vi) menos pasos de proceso, (vii) riesgo reducido de contaminación (importante con las epotilonas altamente tóxicas) y/o (viii) manejo mejor y más seguro (dependiendo del solvente utilizado para la desorción = extracción); y (ix) también, dependiendo del solvente, inesperadamente se necesitan remover inclusive cantidades más bajas de subproductos o impurezas con una polaridad similar a la de las epotilonas, especialmente a la de la epotilona B, después de la cromatografía de fase inversa. Estas y otras ventajas se pueden deducir a partir de los detalles dados en la siguiente Descripción Detallada de la Invención:

Descripción Detallada de la Invención

Un aspecto de la invención se refiere a un proceso para desorber epotilonas, en especial epotilonas A y/o B, en

particular epotilona B, a partir de una resina, especialmente una resina sintética, con un solvente débilmente polar o apolar.

Un aspecto adicional se refiere a un proceso para
5 procesar (o purificar) epotilonas, especialmente después de su producción en un medio convencional para síntesis química, o de preferencia un medio de cultivo que comprenda microorganismos, en especial mixobacterias, en particular del género *Sorangium*, que son adecuadas para producir epotilonas,
10 especialmente epotilona A y/o B, y un componente formador de complejo, comprendiendo este proceso el uso de un solvente débilmente polar o apolar para la desorción de las epotilonas a partir de una resina.

Un tercer aspecto de la invención se refiere al uso
15 de un solvente débilmente polar o apolar para la desorción de epotilonas, especialmente epotilona A y/o B, en particular epotilona B, a partir de una resina, en especial una resina sintética.

Los términos generales utilizados anteriormente en
20 la presente y más adelante en la presente de preferencia tienen los significados dados enseguida:

El término "epotilonas" se refiere de preferencia a cualquier epotilona o derivado de epotilona mencionado en las Solicitudes de Patente Números WO 99/03848, WO 00/49020, WO
25 00/49021, WO 00/47584, WO 00/00485, WO 00/23452, WO 99/03848,

WO 00/49019, WO 99/07692, WO 98/22461, WO 99/65913, WO 98/38192, WO 00/50423, WO 00/22139, WO 99/58534, WO 97/19086, WO 98/25929, WO 99/67252, WO 99/67253, WO 00/31247, WO 99/42602, WO 99/28324, WO 00/50423, WO 00/39276, WO 99/27890, 5 WO 99/54319, WO 99/54318, WO 99/02514, WO 99/59985, WO 00/37473, WO 98/08849, US 6,043,372, US 5,969,145, WO 99/40047, WO 99/01124 y/o WO 99/43653, más preferiblemente a la epotilona A, y en especial a la epotilona B, pero en un aspecto más amplio de la invención, también a las epotilonas 10 D y E, descritas en las Publicaciones Internacionales Números WO 97/19086 y WO 98/22461, las epotilonas E y F, descritas en la Publicación Internacional Número WO 98/22461, o las epotilonas descritas en la Publicación Internacional Número WO 99/02514. Todos estos documentos, especialmente con 15 respecto a los derivados de epotilona, y los derivados de epotilona preferidos mencionados en los mismos, se adjuntan a la presente como referencia.

Un solvente débilmente polar o apolar de preferencia tiene las siguientes características:

20 Se prefiere un solvente que, en la fila eluotrópica determinada de acuerdo con Snyder y colaboradores, J. Chromatogr. Sci. 16, 223 (1978), muestra las siguientes características, en donde X_s es el parámetro del aceptor de protones (indicador de la tendencia a formar enlaces de 25 hidrógeno como aceptores de hidrógeno), X_d es un parámetro

del donador de protones (indicador de la tendencia a formar enlaces de hidrógeno como donador de hidrógeno), y x_n es el parámetro de dipolo (que indica el carácter de dipolo), con la condición de que $x_e + x_d + x_n = 1$, ó una mezcla de estos solventes:

$x_e = 0,20-0,40$; $x_d = 0,15-0,36$; y $x_n = 0,38-0,60$; de preferencia

$x_e = 0,22-0,32$; $x_d = 0,17-0,34$; y $x_n = 0,39-0,54$.

Entre los solventes débilmente polares o apolares muy preferidos están las alquilo inferior-alquilo inferior-cetonas o las cetonas cíclicas, tales como acetona, metiletilcetona, 2-pentanona, metilisobutilcetona, o ciclohexanona, más preferiblemente éteres, en especial éteres cíclicos, muy especialmente tetrahidrofurano o dioxano; todavía de una manera más preferible compuestos de halógeno alifáticos, en especial haluros de alquilo inferior, especialmente dicloruro de metileno (= cloruro de metileno), o dicloruro de etileno; o muy especialmente solventes aromáticos, en especial naftaleno o (de preferencia) benceno o naftalina, o (de preferencia) benceno sustituido por una o más, de preferencia una a tres fracciones seleccionadas a partir del grupo que consiste en alquilo inferior, especialmente metilo, etilo o isopropilo, alcoxilo inferior, especialmente metoxilo o etoxilo, halógeno, especialmente

flúor, cloro, bromo o yodo, nitro y alcoxilo inferior-alquilo inferior, especialmente etoximetilo; en especial tolueno, etilbenceno, xileno, especialmente o-, m-, ó p-xileno, mesitileno, pseudo-cumeno, hemelitenos, cumenos, isopropil-tolueno, haluros de fenilo, especialmente fluorobenceno, clorobenceno, bromobenceno o yodobenceno, alcoxilo inferior-bencenos, especialmente etoxibenceno o metoxibenceno, o alcoxilo inferior-alquilo inferior-bencenos, especialmente etoximetilbenceno (benciléter); o cualquier mezcla de dos o más, por ejemplo de 2 a 4 de estos solventes; se prefieren más alquilo inferior-bencenos, especialmente etilbenceno, xileno, especialmente o-, m-, ó p-xileno, mesitileno, pseudo-cumeno, hemelitenos, cumenos, isopropil-tolueno, y más especialmente tolueno.

El término "solvente débilmente polar o apolar" también incluye mezclas de dos o más de los solventes descritos anteriormente en la presente y posteriormente en la presente, por ejemplo de 2 a 4 de estos solventes.

El prefijo "inferior" siempre indica que el radical correspondientemente mencionado contiene de preferencia hasta un máximo de 7 átomos de carbono, en particular hasta 4 átomos de carbono, y está ramificado o no ramificado. Alquilo inferior puede estar, por ejemplo, no ramificado o ramificado una o más veces, y es, por ejemplo, metilo, etilo, propilo tal como isopropilo o propilo normal, butilo tal como

isobutilo, butilo secundario, butilo terciario o butilo normal, o también pentilo tal como amilo o pentilo normal.

Halógeno es de preferencia yodo, bromo, cloro, o flúor.

5 Un hidrocarburo de preferencia es un compuesto orgánico que tiene de 4 a 32, más preferiblemente de 4 a 20, todavía más preferiblemente de 6 a 16 átomos de carbono, y puede ser alifático, por ejemplo un alcano saturado lineal, ramificado, o cíclico, por ejemplo ciclohexano, un compuesto
10 orgánico lineal, ramificado, o cíclico (no aromático) con uno o más dobles y/o triples enlaces, o un hidrocarburo aromático, siendo este último insustituido o sustituido por uno o más, por ejemplo hasta tres, sustituyentes seleccionados a partir del grupo que consiste en alquilo
15 inferior, especialmente metilo, etilo o isopropilo, alcóxido inferior, especialmente metóxido o etóxido, halógeno, especialmente flúor, cloro, bromo o yodo, nitro, y alcóxido inferior-alquilo inferior, especialmente etoximetilo; de preferencia por 1 a 3 fracciones de alquilo inferior.

20 Un alcohol es de preferencia un hidroxialcano inferior, especialmente metanol, etanol, o n- o iso-propanol.

 Una resina es especialmente una resina sintética, de preferencia una resina basada en copolímeros de estireno y divinilbenceno, más preferiblemente Amberlite XAD-4, o de
25 preferencia Amberlite XAD-16 [Rohm & Haas Germany GmbH,

Frankfurt] ó Diaion HP-20 [Resindion S.R.L., Mitsubishi Chemical Co., Milán]. Queda sin decir que una resina a partir de la cual se vaya a desorber una epotilona o epotilonas de acuerdo con la invención, tiene epotilonas no covalentemente en contacto con ella (por ejemplo, reversiblemente enlazadas con ella o adsorbidas a ella). En otros términos, cuando se utiliza el término "resina", significa "resina que tiene una o más epotilonas en contacto con ella, en especial reversiblemente enlazadas o absorbidas a ella".

10

Aspectos Preferidos de la Invención:

En un aspecto preferido de la invención, la invención se refiere a un proceso que comprende una desorción a partir de una resina con un solvente débilmente polar o apolar como se describe anteriormente o más adelante, el cual comprende cualesquiera pasos de purificación adicionales deseables o requeridos para llegar a las epotilonas respectivas, especialmente epotilona A y más especialmente epotilona B, en forma pura, de preferencia las descritas como preferibles en la presente invención.

15
20

El procesamiento de las epotilonas es precedido por la reacción o el proceso que conduce a un producto de reacción que comprende epotilonas para procesarse, las cuales se aíslan (i) ya sea a partir de las mezclas de reacción químicas después de la solución en un medio polar acuoso

25

apropiado, o (ii) más preferiblemente a partir del sobrenadante (por ejemplo, una ciclodextrina que contenga una descrita en la Publicación Internacional Número WO 99/42602) de un medio de cultivo con microorganismos productores de las
5 epotilonas, mediante la separación de un cultivo en la fase líquida (por ejemplo centrifugado o filtrado) y la fase sólida (células), especialmente por medio de filtración o centrifugación (centrífugo tubular o separador).

Este tratamiento previo de preferencia se continúa entonces mediante mezcla directa de la solución ((i)) o de la
10 fase líquida ((ii)) con una resina, especialmente una resina sintética, en especial una resina basada en copolímeros de estireno y divinilbenceno como matriz (referida posteriormente en la presente también simplemente como resina
15 de poliestireno), tal como Amberlite XAD-16 ó Diaion HP-20 (de preferencia en una proporción del volumen del centrifugado:resina de aproximadamente 10:1 a 100:1, de preferencia de aproximadamente 50:1). Después de un período de contacto de preferencia de 0.25 a 50 horas, especialmente
20 de 0.8 a 22 horas, la resina se separa, por ejemplo mediante filtración, sedimentación, o centrifugación. Si se requiere, después de la adsorción, la resina se lava con un solvente fuertemente polar, de preferencia con agua.

Luego se inicia el proceso de procesamiento
25 preferido de acuerdo con la presente invención: La desorción

de las epotilonas (formando como tal, un aspecto muy preferido de la invención) se efectúa con un solvente débilmente polar o apolar, en particular con haluros de alquilo inferior, especialmente dicloruro de metileno o dicloruro de etileno, o más preferiblemente solventes aromáticos, en especial naftaleno o (de preferencia) benceno, o naftalina o (de preferencia) benceno sustituido por una o más, de preferencia una a tres fracciones seleccionadas a partir del grupo que consiste en alquilo inferior, especialmente metilo, etilo o isopropilo, alcoxilo inferior, especialmente metoxilo o etoxilo, halógeno, especialmente fluoro, cloro, bromo o yodo, nitro y alcoxilo inferior-alquilo inferior, especialmente etoximetilo; de una manera muy preferible tolueno. El paso de extracción se puede repetir una o más veces, de preferencia de 0 a 3 veces, especialmente una vez, con el objeto de obtener una desorción más completa cuando sea necesario o se desee.

El procesamiento (en el caso de una extracción repetida combinada) de las soluciones de epotilonas desorbidas que se pueden obtener de esta manera, de preferencia continúa con la remoción del solvente utilizado para la desorción a partir de las soluciones resultantes mediante evaporación (destilación), de preferencia por medio de concentración en un reactor, y subsecuentemente en un evaporador giratorio al vacío.

Subsecuentemente, tiene lugar un procesamiento adicional utilizando los siguientes pasos, en donde es obligatorio el paso de purificación por medio de cromatografía de fase inversa con elución con un nitrilo, mientras que los demás pasos son opcionales:

- cristalización de las epotilonas después de la desorción; especialmente, para la cristalización de la epotilona B, se agrega una mezcla de un alcohol y un hidrocarburo, en especial de un alcohol inferior y un hidrocarburo alifático cíclico con 3 a 10 átomos del anillo, más especialmente de metanol y ciclohexano (de preferencia en una proporción por volumen/volumen de 1:10 a 10:1, en especial de 1:3 a 3:1). La adición de agua conduce a una rápida separación de fases, y la fase de alcohol se evapora a sequedad, por ejemplo por medio de un evaporador giratorio al vacío. El extracto resultante, el cual comprende la epotilona B, se cristaliza después a partir de una mezcla de solventes apropiada, en especial a partir de una mezcla de alcohol/hidrocarburo alifático cíclico, más preferiblemente isopropanol/ciclohexano, de preferencia en una proporción por volumen/volumen de 1:10 a 10:1, más preferiblemente de 1:6 a 6:1, y de una manera muy preferible de 1:6 a 1:4;

- separación (obligatoria) de las epotilonas mediante cromatografía de fase inversa después de recuperarse en un solvente adecuado, en especial una mezcla de un nitrilo

y agua, de preferencia acetonitrilo/agua, en una proporción por volumen/volumen preferida de 1:10 a 10:1, en especial de 1:3 a 1:1, y elución con una mezcla de nitrilo y agua, de preferencia caracterizada porque la cromatografía se realiza sobre una columna de un material de fase inversa, que se carga con cadenas de hidrocarburo, tales como cadenas de hidrocarburo que contengan 18 átomos de carbono, en especial un material RP-18, y se utiliza un eluyente que comprenda un nitrilo, en especial un alquilo inferior-nitrilo, en particular acetonitrilo, en particular se utiliza una mezcla de nitrilo/agua, en especial una mezcla de acetonitrilo/agua, de preferencia en una proporción del nitrilo al agua de aproximadamente 1:99 a 99:1, primordialmente entre 1:9 y 9:1, por ejemplo entre 2:8 y 7:3, por ejemplo 3:7 ó 4:6; y remoción del nitrilo a partir de la epotilona recolectada (en especial epotilona A, o más especialmente epotilona B) que comprende fracciones por evaporación (destilación); si se desea, entonces el agua restante con la epotilona se extrae con un éster, en especial un alcanato inferior de alquilo inferior, de preferencia acetato de isopropilo, con evaporación subsecuente (de preferencia primero en un reactor, luego en un evaporador giratorio al vacío) de la epotilona que contiene la fase de éster, a sequedad; (si se requiere, la solución de epotilona de partida se puede dividir y separar en más de una pasada separada de fase

90
89

inversa);

- solamente si se requiere (por ejemplo, como una alternativa a la cristalización después de la desorción), cromatografía de adsorción, en particular mediante la adición
5 a una columna de gel de sílice y elución con un solvente o mezcla de solventes apropiada, en especial una mezcla de éster/hidrocarburo, por ejemplo alcanato de alquilo inferior/alcano de 4 a 10 átomos de carbono, en especial acetato de etilo o isopropilo/hexano normal, en donde la
10 proporción entre el éster y el hidrocarburo de preferencia está en el rango de 99:1 a 1:99, de preferencia de 10:1 a 1:10, por ejemplo 4:1;

- y finalmente, recristalización, por ejemplo a partir de solventes o mezclas de solventes apropiados, por
15 ejemplo consistentes en ésteres, mezclas de éster/hidrocarburo, o alcoholes, en especial acetato de etilo o de isopropilo:tolueno, 1:10 a 10:1, de preferencia 2:3 (epotilona A), o metanol o acetato de etilo (epotilona B);

en cuyo proceso, si es necesario y/o se desea,
20 entre cada paso que se emplee, se concentran las soluciones o suspensiones resultantes, y/o se separan los componentes líquidos y sólidos unos de otros, en particular mediante sedimentación, filtración o centrifugación de las soluciones/suspensiones. Las definiciones más precisas mencionadas
25 anteriormente y más adelante se pueden utilizar de

96
90

preferencia en los pasos individuales anteriores.

Un aspecto preferido de la invención se refiere también a un proceso para el aislamiento de las epotilonas adsorbidas a una resina sintética, en especial epotilona A, o
5 más especialmente epotilona B, cuyo proceso comprende:

(i) la desorción de las epotilonas a partir de la resina sintética con un solvente débilmente polar o apolar, en particular con haluros de alquilo inferior, en especial dicloruro de metileno o dicloruro de etileno, o más
10 preferiblemente solventes aromáticos, en especial naftaleno o (de preferencia) benceno, o naftalina o (de preferencia) benceno sustituido por una o más, de preferencia una a tres fracciones seleccionadas a partir del grupo que consiste en alquilo inferior, en especial metilo, etilo o isopropilo,
15 alcoxilo inferior, en especial metoxilo o etoxilo, halógeno, en especial flúor, cloro, bromo o yodo, nitro y alcoxilo inferior-alquilo inferior, en especial etoximetilo; más preferiblemente tolueno; o en un aspecto más amplio de la invención, una mezcla de dos o más de estos solventes; y

20 (ii) la separación de las epotilonas mediante cromatografía de fase inversa después de recuperarse en un solvente adecuado, en especial una mezcla de un nitrilo y agua, de preferencia acetonitrilo/agua, en una proporción por volumen/volumen preferida de 1:10 a 10:1, en especial de 1:3
25 a 1:1, y elución con una mezcla de nitrilo y agua, de

preferencia caracterizada porque la cromatografía se realiza sobre una columna de un material de fase inversa, en especial un material RP-18, que se carga con cadenas de hidrocarburo, tales como cadenas de hidrocarburo que contengan 18 átomos de carbono, y se utiliza un eluyente que comprenda un nitrilo, en especial un alquilo inferior-nitrilo, en particular acetonitrilo, y se utiliza en particular una mezcla de nitrilo/agua, en especial una mezcla de acetonitrilo/agua, de preferencia en una proporción del nitrilo al agua de aproximadamente 1:99 a 99:1, primordialmente entre 1:9 y 9:1, por ejemplo entre 2:8 y 7:3, por ejemplo 3:7 ó 4:6.

En un aspecto preferido adicional de la invención, el proceso del último párrafo, empezando con el paso (i) y comprendiendo el paso (ii), implica además cualesquiera pasos de purificación adicionales deseables o requeridos para llegar a las epotilonas respectivas, en especial epotilona A, y más especialmente epotilona B, en forma pura, de preferencia las descritas como preferibles en la presente invención.

La preparación para el procesamiento de preferencia se realiza como sigue: la adsorción de las epotilonas, en especial a partir de mezclas de reacción química, o más preferiblemente a partir del sobrenadante de cultivos de microorganismos, se puede lograr como se describe en la Publicación Internacional Número WO 99/42602, o en analogía a

la misma; brevemente, las epotilonas se encuentran en el centrifugado, que luego se mezcla directamente con una resina sintética, en especial una resina de copolímero de estireno/divinilbenceno, tal como Amberlite XAD-16 ó Diaion HP-20 (de preferencia en una proporción por volumen del centrifugado:resina de aproximadamente 10:1 a 100:1, de preferencia de aproximadamente 50:1), y se agita en un agitador. En este paso, las epotilonas se transfieren a la resina. Después de un período suficiente para la adsorción, por ejemplo un período de contacto de aproximadamente 0.2 a 10 horas, la resina se separa mediante centrifugación o filtración. La adsorción de las epotilonas sobre la resina también se puede efectuar en una columna de cromatografía, colocando la resina en la columna, y pasando el centrifugado sobre la resina. Después de la adsorción, la resina se lava con agua.

Entonces empiezan los procesos preferidos de acuerdo con la invención y proceden como sigue: La desorción de las epotilonas a partir de la resina de preferencia se efectúa con un solvente débilmente polar o apolar de acuerdo con la invención, en especial uno descrito como preferido anteriormente o más adelante, en especial cloruro de metileno, o más especialmente tolueno. Luego se remueve el solvente tanto como sea necesario, de preferencia hasta que se obtenga un residuo seco. Cuando sea apropiado, el residuo

se recupera en una mezcla de alcohol/hidrocarburo, en especial en metanol/ciclohexano, de preferencia en las proporciones descritas anteriormente, en un volumen relativamente bajo. La fase de alcohol se evapora, de preferencia a sequedad, y luego se cristaliza el extracto de alcohol a partir de una mezcla de un alcohol, en especial isopropanol, y un hidrocarburo, en especial ciclohexano, de preferencia en las proporciones descritas anteriormente. Luego se disuelve el material cristalizado sólido resultante en una mezcla de nitrilo/agua, de preferencia como se describió anteriormente, en especial una mezcla de 2:3 (volumen/volumen) de acetonitrilo/agua, y la solución de alimentación resultante se vierte, si se requiere, después de dividirse en más de una pasada, sobre una columna de fase inversa de preparación. Sigue la elución con nitrilo/agua, en especial como se mencionó. El (aceto)nitrilo de las fracciones resultantes que contienen epotilona, en especial epotilona A, y más especialmente epotilona B, se remueve por evaporación (destilación), y la fase de agua resultante se extrae con un éster, en especial acetato de isopropilo. Luego se evapora el extracto de éster, de preferencia a sequedad, y subsecuentemente se recrystaliza el material resultante, por ejemplo se cristaliza una fracción de epotilona A directamente a partir de una mezcla de éster/hidrocarburo, por ejemplo acetato de etilo:tolueno =

98
94

2:3, y la fracción de epotilona B a partir de un éster, en especial acetato de etilo, o de preferencia a partir de un alcohol, en especial metanol.

Especialmente con el tolueno, se ha descubierto un
5 eluyente altamente selectivo (desorbente) que permite lograr un rendimiento de aproximadamente el 100 por ciento en la mitad del tiempo de desorción necesario con el isopropanol utilizado en la Publicación Internacional Número WO 99/42602. De una manera sorprendente, se incrementa la cantidad de
10 epotilonas desorbidas, por ejemplo hasta el 130 por ciento después de la desorción con tolueno, al compararse con la desorción con isopropanol. (Aunque esto parece *prima facie* imposible desde un punto de vista teórico, ilustra bien una importante ventaja de la presente invención: El resultado
15 está relacionado con el ensayo de la resina cargada. Debido a que el ensayo hizo uso de isopropanol para la desorción, se tuvo que tomar como base un procedimiento de desorción incompleto, conduciendo originalmente a valores de ensayo más bajos, que ahora resultan ser decepcionantes). La mezcla de
20 epotilona (sin o con la cristalización precedente) se puede aplicar directamente a la columna de fase inversa. El proceso llega a ser altamente robusto con respecto a las cantidades de solvente, las velocidades de agitación, y las temperaturas. Aunque para los alcoholes (por ejemplo etanol o
25 isopropanol) bajo agitación, se puede demostrar una desorción

de dos fases, en donde se desorbe una primera cantidad de material que comprende epotilona en un primer período de tiempo (que se puede explicar posiblemente por la distribución de tamaños de poros que se encuentran en el poliestireno XAD-16 que tiene dos máximas de distribución), y una segunda cantidad después de un período de tiempo adicional, no se encuentra este comportamiento indeseable con el tolueno, o tampoco con el diclorometano, en donde se desorbe todo el material ya en el primer período de tiempo.

5 Bajo condiciones comparables, el residuo de la evaporación, en el caso del isopropanol para las epotilonas A y B, en un ejemplo, se ha encontrado que es de 40 gramos, con el cloruro de metileno de 3.3 gramos, en el caso de tolueno solamente de 0.9 gramos, con por ejemplo 17-18 g-% de epotilona B después

10 de la desorción a partir de resinas de copolímero de estireno/divinilbenceno obtenidas a partir de sobrenadantes de cultivo que comprenden ciclodextrina, como se describe en la Publicación Internacional Número WO 99/42602, indicando una pureza mucho más alta. Aunque el tolueno tiene las

15 ventajas más significativas, el diclorometano tiene como una ventaja su facilidad de remoción, debido al bajo punto de ebullición.

La separación de las epotilonas A y B también se puede lograr mediante la realización de la cromatografía dada

20 a conocer en la presente, en su planteamiento de lecho en

movimiento simulado (SMB). Las cromatografías de lecho en movimiento simulado se utilizan ampliamente para la separación de mezclas binarias, por ejemplo la separación de racematos sobre fases estacionarias quirales, por ejemplo los procesos SORBEX en las industrias petroquímicas, como Parex ó Molex, o el proceso SAREX en la industria del azúcar. Comparándose con la cromatografía por lotes, la cromatografía de lecho en movimiento simulado proporciona la ventaja de una operación unitaria a contra-corriente continua, que conduce a una mayor productividad y a un consumo reducido de la fase móvil. Los expertos en la técnica conocen varios procedimientos sistemáticos para el desarrollo del método de las cromatografías de lecho en movimiento simulado. Estos procedimientos son descritos, por ejemplo, por R.-M. Nicoud, M. Bailly, J. Kinkel, R.M. Devant, T.R.E. Hampe y E. Küsters en *Proceedings of the 1st European Meeting on Simulated Moving Bed Chromatography*, (1993), ISBN 2-905-267-21-6 páginas 65-88; E. Küsters, G. Gerber, y F.D. Antia, *Chromatographia*, 40 (1995) 387; T. Pröll y E. Küsters, *J. Chromatogr. A*, 800 (1998) 135; ó C. Heuer, E. Küsters, T. Plattner y A. Seidel-Morgenstern, *J. Chromatogr. A*, 827 (1998) 175.

Los parámetros básicos para la separación de las epotilonas A y B con cromatografía de lecho en movimiento simulado, se pueden tomar directamente a partir de la

98
97

separación de LC convencional. De preferencia, el gel de sílice de fase inversa (RP 18) se toma como la fase estacionaria, y las mezclas de agua/acetonitrilo como las fases móviles. El conjunto final de velocidades de flujo
5 (para las zonas individuales del lecho en movimiento simulado y el tiempo de conmutación, respectivamente) se puede tomar a partir de un simple esquema de flujo, como fue desarrollado por E. Küsters y colaboradores en *Chromatographia*, 40 (1995) 387, o después de una estimación cuidadosa de las isothermas
10 de adsorción estipuladas en *J. Chromatogr. A*, 800 (1998) 135 y en *J. Chromatogr. A*, 827 (1998) 175. El procesamiento de las corrientes de extracto y refinado se puede realizar de nuevo como se describe para la separación de LC convencional.

La invención se refiere de una manera más
15 preferible a los procesos y métodos descritos en los siguientes ejemplos:

Ejemplos:

Los siguientes ejemplos sirven para ilustrar la
20 invención, sin limitar su alcance.

Precaución: Cuando se manejan epotilonas, se deben tomar medidas de protección apropiadas, donde sea necesario, en vista de su alta toxicidad.

Ejemplo 1: Procedimiento de procesamiento para la epotilona B

La desorción de 591.7 kilogramos de resina cargada (resina de copolímero de estireno/divinilbenceno XAD-16 cargada con epotilonas A y B a partir de un medio de cultivo) se efectúa mediante la agitación de la resina en dos porciones, cada una con 720 litros de tolueno en 4 porciones durante aproximadamente 8 horas. La separación de la fase de tolueno de la resina tiene lugar utilizando un filtro de succión. Las fases de tolueno combinadas se lavan en dos porciones, cada una con 250 litros de agua. Después de la separación de fases, el extracto de tolueno se concentra en un reactor de 1000 litros hasta aproximadamente 20 a 40 litros, y después se concentra a sequedad en un evaporador giratorio al vacío. El resultado es un extracto de tolueno de 4.095 kilogramos que contiene 209 gramos de la epotilona B. El extracto de tolueno se disuelve en 16.5 litros de metanol y 24.5 litros de ciclohexano. Después de la adición de 0.8 litros de agua, se presenta inmediatamente la separación de fases. La fracción de metanol se evapora a sequedad en un evaporador giratorio al vacío, produciendo 1.025 kilogramos de residuo de evaporación que contiene 194 gramos de la epotilona B. El extracto de metanol se cristaliza después en una mezcla de solventes consistente en 2.05 litros de isopropanol y 10.25 litros de ciclohexano, produciendo 0.4 kilogramos de material cristalizado que

contiene 184 gramos de la epotilona B. Los cristales se disuelven en 3.2 litros de acetonitrilo/agua = 2/3 (volumen/volumen), y la solución de alimentación resultante se transfiere en 3 pasadas separadas, a una columna de fase inversa de preparación (25 kilogramos de gel de sílice esférica RP-18, YMC-Gel ODS-A 120; 5-15 micras; Waters Corp., Milford, Massachusetts, EUA). La elución se efectúa con acetonitrilo/agua = 2/3 (volumen/volumen) como la fase móvil, con una velocidad de flujo de 2.3 litros/minuto; tiempo de retención de epotilona A = 77-96 minutos, tiempo de retención de epotilona B = 96-119 minutos. El fraccionamiento se monitorea con un detector de ultravioleta a 250 nanómetros. El acetonitrilo de las fracciones combinadas de epotilona B (de las tres pasadas) se destila, y la fase de agua restante se extrae con 504 litros de acetato de isopropilo. El extracto de acetato de isopropilo se concentra en un reactor de 630 litros hasta aproximadamente 20 a 40 litros, y después se concentra a sequedad en un evaporador giratorio al vacío. El peso del residuo de la evaporación de las fracciones de epotilona B es de 170 gramos, y tiene un contenido del 98.4 por ciento de acuerdo con la HPLC (estándar externo). El material resultante finalmente se cristaliza en 2.89 litros de metanol a 0-5°C, produciendo 150 gramos de cristalizado puro de epotilona B.

Punto de fusión: 124-125°C;

Datos de ^1H RMN para epotilona B (500 MHz-RMN, solvente: DMSO- d_6 . Desplazamiento químico δ en ppm en relación con TMS. S = singulete, d = doblete, m = multiplete):

	δ (multiplicidad)	Integral (número de H)
5	7.34 (s)	1
	6.50 (s)	1
	5.28 (d)	1
	5.08 (d)	1
	4.46 (d)	1
10	4.08 (m)	1
	3.47 (m)	1
	3.11 (m)	1
	2.83 (dd)	1
	2.64 (s)	3
15	2.36 (m)	2
	2.09 (s)	3
	2.04 (m)	1
	1.83 (m)	1
	1.61 (m)	1
20	1.47-1.24 (m)	4
	1.18 (s)	6
	1.13 (m)	2
	1.06 (d)	3
	0.89 (d + s, traslapados)	6
25	$\Sigma = 41$	

Ejemplo 2: Comparación de diferentes procesos de desorción para la preparación de epotilona B:

360 mililitros de cada una de una suspensión acuosa de la resina de copolímero de estireno/divinilbenceno XAD-16 cargada con epotilonas A y B a partir de un cultivo de mixobacterias mediante el método descrito en la Publicación Internacional Número WO 99/42602 (correspondientes a 194 gramos de Amberlite® XAD-16 húmeda), se extraen con los solventes y bajo las condiciones mencionadas en la siguiente tabla, en un reactor de vidrio agitado (agitador de ancla de laboratorio común) con un fritta sinterizada en el fondo (reactor por lotes en fase sólida hecho en casa, diámetro interno de 10 centímetros por 20 centímetros de longitud, "Agitador/Fritta" posteriormente en la presente).

15

Tabla 1: Comparación de los métodos de desorción para la fabricación de Epotilona B.

Resina de partida (resina con epotilona adsorbida):

20

- # 1001 carga teórica de 70 miligramos por 360 mililitros de suspensión de resina (corresponden a 70 miligramos por 194 gramos de XAD-16 húmeda)

25

- # 1003 carga teórica de 114 miligramos por 360 mililitros de suspensión de resina (corresponden a 114 miligramos por 194 gramos de XAD-16 húmeda).

Variante	1	2	3	4	5	6
Reactor	Agitador/frit a (TA)	Agitador/frita (TA)	Agitador/Frita (TA)	Agitador/Frita (TA)	Reflujo (40°C)	Reflujo (42-45°C)
Resina de partida	#1001	#1001	#1003	#1003	#1003	#1002
Cantidad de suspensión de resina (ml)	360	360	360	360	360	180
Solvente de desorción	Isopropanol	Cloruro de metileno	Isopropanol	Cloruro de metileno	Cloruro de metileno	Isopropanol
Agitación (rpm)	250	250	250	250	250	250
Volumen (l)	8 x 0.2 = 1.6	3 x 0.36 = 1.1 ^a	5 x 0.72 ¹ = 3.6	3 x 0.72 ^b = 2.2	1 x 2.2	1 x 1.8
Tiempo de desorción (h)	8	3	5	3	2	2
Evaporación (h)	2 ^e	0.5	4 ^e	Ninguno	Ninguno	2
Adición de agua (l)	0.7	Ninguno ^b	1.15	Ninguno	Ninguno	0.54
Extracción con acetato de etilo (l)	1.1	Ninguno	2.2	Ninguno	Ninguno	1.1
Tiempo de separación de fases (h)	24 ^c	Ninguno	15	Ninguno	Ninguno	15
Evaporación (h)	2	Ninguno	2	1	1	1
Residuo de evaporación (g)	5.84 ^f	3.37 ^d	3.31	2.68 ^d	3.24 ^f	1.70
Contenido (%)	1.26	1.75	2.9	3.7	3.1	3.3
Rendimiento: (mg) %	74 (105) ^g	59 (84)	96 (84)	99 (87)	101 (89)	56 (98)

5

10

15

5

10

15

Variante	7	8	9	10	11	12
Reactor	Erl./Agitador Magnético*	Agitador/frita (TA)	Reflujo (42-45°C)	Agitador/Frita (TA)	Agitador/Frita (TA)	Agitador/Frita (43°C)
Resina de partida	#1003	#1003	#1003	#1003	#1003	#1003
Cantidad de suspensión de resina (ml)	1000	360	180	360	360	360
Solvente de desorción	Cloruro de metileno	Tolueno	Isopropanol	Tolueno	Tolueno	Tolueno
Agitación (rpm)	?	640	250	250	640	250
Volumen (l)	1 x 0.5 = 0.5	3 x 0.72 = 2.2	1 x 1.8 = 1.8	4 x 0.36 = 1.44	4 x 0.36 = 1.44	4 x 0.36 = 1.44
Tiempo de desorción (h)	5	3	6	4	4	4
Evaporación (h)	Ninguno		6	Ninguno	Ninguno	Ninguno
Adición de agua (l)	Ninguno	0.5 ^j	0.54	0.5 ^k	0.5 ^k	0.5 ^k
Extracción con acetato de etilo (l)	Ninguno	Ninguno	1.1	Ninguno	Ninguno	Ninguno
Tiempo de separación de fases (h)	Ninguno	0.1	15	0.1	0.1	0.1
Evaporación (h)	1	1	1	1	1	1
Residuo de evaporación (g)	4.38	0.88	2.32	0.75	0.88	0.95
Contenido (%)	0.5	17.4	2.69	17.6	17.1	15.8
Rendimiento: (mg) %	22 (69)	153 (134)	62.5(110)	132 (116)	150 (132)	150 (132)

104

- a No optimizado; en el caso de 2 extracciones, se pierde aproximadamente el 5 por ciento del rendimiento.
- b La retroextracción triple con agua no remueve ningún componente polar.
- 5 c Primera de 3 separaciones de fases durante la noche.
- d El material, al compararse con el residuo de la evaporación de acetato de etilo, se puede pasar por cromatografía mucho mejor sobre gel de sílice.
- 10 e Corresponde a un residuo de evaporación de aproximadamente 25 a 30 gramos.
- f Horno de secado (alto vacío) durante el fin de semana.
- 15 g Posiblemente un resultado, con respecto al tiempo utilizado, de la generosa primera separación de fases.
- h Recuperación: primera extracción = 700 mililitros; segunda extracción = 660 mililitros; tercera extracción = 680 mililitros.

104

- i Recuperación: primera extracción = 670 mililitros; segunda extracción = 710 mililitros; tercera extracción = 720 mililitros.
- j Todavía no se ha confirmado la necesidad de adición de agua.
- 5 k Simplifica la remoción del agua originalmente presente (la XAD-16 se recupera mediante agua tal como se utilizó).
- 10 l En vista de los resultados de las variantes 8, 11 y 12, y las curvas de elución para todos los experimentos con tolueno, no se puede excluir el uso de una cantidad más pequeña de resina.

«Erl./agitador magnético» significa la agitación magnética en un matraz Erlenmeyer

«Cloruro de metileno» («Clor.de metileno») es diclorometano.

- 15 «Agitador/Frita» significa agitador/frita.

«TA» significa temperatura ambiente.

A partir de estos experimentos y de los datos adicionales, se puede deducir que, en comparación con el isopropanol, la extracción con cloruro de metileno ofrece una mejor selectividad, un tiempo más corto de extracción (aproximadamente por un factor de 2), una destilación de solvente más rápida (el punto de ebullición del cloruro de metileno es de aproximadamente 40°C, y el del isopropanol es de 81-83°C), ya no se requiere la separación de fases de acetato de etilo/agua muy intensa en tiempo y muy problemática, ya no se requiere más la segunda destilación del solvente, de tal manera que se puede utilizar un número más bajo de pasos de proceso, dando como resultado un riesgo de contaminación más bajo, y un manejo mejor y más seguro; es posible el procesamiento en la mitad del volumen (por ejemplo, en reactores de 1000 litros en lugar de reactores de 2000 litros); el producto de epotilona B tiene un mejor perfil de pureza (menos subproductos con una polaridad comparable a la de la epotilona B), y el residuo de la evaporación no chisporrotea ni espuma, como en el caso con la extracción con acetato de etal. Para la desorción con tolueno, se obtiene un rendimiento más alto en comparación con la extracción con isopropanol (aproximadamente el 100 en lugar de aproximadamente el 80 por ciento), se puede observar una mejor selectividad (con el isopropanol, se desorbe aproximadamente 10 veces la cantidad de subproductos), se

reduce el tiempo de extracción considerablemente (por un factor de 3), se simplifica la difícil filtración después de la desorción con isopropanol (la extracción con isopropanol era difícil de implementar a una mayor escala), ya no se
5 requiere la segunda destilación del solvente, es posible el procesamiento en reactores más pequeños (nuevamente, por ejemplo, es posible utilizar un reactor de 1000 litros en lugar de un reactor de 2000 litros), es posible abandonar la cromatografía de gel de sílice (el residuo de la evaporación
10 después de la desorción contiene ya aproximadamente el 40 por ciento de una mezcla de epotilonas A/B), y el residuo de la evaporación después de la desorción no muestra la espumación y el chisporroteo observados para el residuo a partir de la extracción con acetato de etilo.

15

20

25

REIVINDICACIONES

1. Un proceso para desorber epotilonas a partir de una resina haciendo este proceso uso de un solvente débilmente polar o apolar.

2. El proceso de acuerdo con la reivindicación 1, en donde las epotilonas que se van a desorber son epotilona A ó epotilona B.

3. El proceso de acuerdo con cualquiera de las reivindicaciones 1 ó 2, en donde la resina es una resina sintética.

4. El proceso de acuerdo con la reivindicación 3, en donde la resina es una resina sintética basada en copolímeros de estireno/divinilbenceno.

5. El proceso de acuerdo con cualquiera de las reivindicaciones 1 a 4, en donde el solvente débilmente polar o apolar se selecciona a partir del grupo que consiste en haluros de alquilo inferior y solventes aromáticos, o una mezcla de dos o más de estos solventes.

6. El proceso de acuerdo con la reivindicación 5, en donde se utiliza un solvente aromático seleccionado a partir del grupo que consiste en naftaleno, benceno, o naftaleno o benceno sustituidos por una o más fracciones seleccionadas a partir del grupo que consiste en alquilo inferior, alcoxilo inferior, halógeno, nitro, y alcoxilo

inferior-alquilo inferior.

7. Un proceso para el aislamiento de epotilonas adsorbidas a una resina sintética, cuyo proceso comprende:

(iii) la desorción de las epotilonas a partir de la resina sintética con un solvente débilmente polar o apolar; y

(iv) la separación de las epotilonas mediante cromatografía de fase inversa después de recuperarse en un solvente adecuado, y la elución con una mezcla de nitrilo y agua.

8. Un proceso de acuerdo con cualquiera de las reivindicaciones 1 a 7, el cual incluye además cualesquiera pasos de purificación adicionales deseables o requeridos para llegar a las epotilonas respectivas en una forma pura.

9. Un proceso para procesar epotilonas a partir de una mezcla de reacción, comprendiendo este proceso:

- desorber las epotilonas con un solvente débilmente polar o apolar, en donde el paso de desorción se puede repetir con el objeto de lograr una desorción más completa;

- remover el solvente utilizado para la desorción a partir de las soluciones resultantes mediante evaporación;

- opcionalmente cristalizar las epotilonas después de la desorción; en especial, para la cristalización de la epotilona B, agregar una mezcla de un alcohol y un

hidrocarburo, especialmente de un alcohol inferior y un hidrocarburo cíclico alifático con 3 a 10 anillos de átomos, y evaporar la fase de alcohol a sequedad, y cristalizar la epotilona B a partir de una mezcla de solventes apropiada, especialmente a partir de una mezcla de hidrocarburo alifático cíclico/alcohol;

- (paso obligatorio) separar las epotilonas mediante cromatografía de fase inversa, recuperando el residuo a partir del paso anterior en un solvente adecuado, especialmente una mezcla de un nitrilo y agua, y eluir con una mezcla de nitrilo y agua, preferiblemente caracterizado por que la cromatografía se lleva a cabo en una columna en el reverso de la fase de un material el cual se carga con cadenas de hidrocarburo, especialmente un material RP-18 que comprende cadenas de hidrocarburo que comprenden 18 átomos de carbono, y una elución que comprende nitrilo, especialmente se utiliza un alquilonitrilo inferior, en particular se utiliza una mezcla de nitrilo/agua, especialmente una mezcla de acetonanitrilo/agua, y remover el nitrilo de las fracciones que comprenden la epotilona recolectada por evaporación; si se desea, el agua restante con la epotilona se extrae entonces con un éster, con evaporación subsecuente de la fase de éster que contiene epotilona a sequedad;

- opcionalmente, purificar además mediante cromatografía de adsorción, particularmente adicionando a la

columna de gel sílice y eluyendo con un solvente apropiado o mezcla solvente;

- y finalmente recrystalizar una epotilona purificada a partir de solventes o mezclas de solventes apropiados;

en cuyo proceso, si es necesario y/o se desea, entre cada caso que se emplee, las soluciones o suspensiones resultantes se concentran, y/o los componentes líquidos y sólidos se separan unos de otros, particularmente al sedimentar, filtrar o centrifugar las soluciones o suspensiones.

10. Un proceso de acuerdo con la reivindicación 9, en donde la desorción de las epotilonas a partir de la resina se efectúa con un solvente débilmente polar o apolar, en especial como se mencionó como preferido en la reivindicación 5 ó 6; luego se remueve el solvente tanto como sea necesario, de preferencia hasta que se obtenga un residuo seco, donde sea apropiado, el residuo se recupera en una mezcla de alcohol/hidrocarburo en un volumen relativamente bajo; la fase de alcohol se evapora, de preferencia a sequedad, y luego se cristaliza el extracto de alcohol a partir de una mezcla de un alcohol y un hidrocarburo, y luego el material cristalizado sólido resultante se disuelve en una mezcla de nitrilo/agua como se describe en la reivindicación 8, especialmente una mezcla de 2:3 (volumen/volumen) de

acetonitrilo/agua, y la solución de alimentación resultante se vierte, si se requiere, después de dividir en más de una pasada, sobre la columna de fase inversa de preparación; seguido por elución con nitrilo/agua; luego sigue la remoción de nitrilo de las fracciones resultantes que contienen epotilona por evaporación, y se extrae la fase de agua resultante con un éster; entonces se evapora el extracto de éster, y subsecuentemente se recristaliza el material resultante.

10 11. Un proceso de acuerdo con cualquiera de las reivindicaciones 7, 9, ó 10, en donde la separación de las epotilonas A y B se logra mediante cromatografía de lecho en movimiento simulado (SMB).

15 12. El uso de un solvente débilmente polar o apolar para la desorción de epotilonas a partir de una resina.

20 15. El uso de acuerdo con la reivindicación 12, en donde el solvente débilmente polar o apolar se selecciona a partir del grupo que consiste en haluros de alquilo inferior y solventes aromáticos, en especial naftaleno, benceno, o naftaleno o benceno sustituidos por una o más fracciones seleccionadas a partir del grupo que consiste en alquilo inferior, alcoxilo inferior, halógeno, nitro y alcoxilo inferior-alquilo inferior; o una mezcla de dos o más de estos
25 solventes; y la resina es una resina sintética.

RESUMEN

La invención se refiere a un proceso de desorción
5 para epotilonas, en especial epotilona A y/o epotilona B, a
partir de resinas, y a nuevos procesos de producción,
procesamiento o purificación, o a métodos de fabricación para
epotilonas que comprenden dicho proceso de desorción, así
como al uso de ciertos solventes para la desorción de
10 epotilonas a partir de resinas.

* * * * *

15

20

REQUISITOS NECESARIOS PARA PRESENTACION Y AGILIZACION DEL TRAMITE

	USUARIO	RADICADOR
PATENTE DE INVENCION	()	(X)
MODELO DE UTILIDAD	()	(X)
- Indicación de que se solicita la concesión de una patente.	()	(X)
- Datos de identificación del solicitante o de la persona que se presenta la solicitud.	()	(X)
- Descripción de la invención.	()	()
- Dibujos de ser estos pertinentes.	()	(X)
- Comprobante de Pago de las tasas establecidas.	()	(X)

COMPLETA () INCOMPLETA ()

DISEÑO INDUSTRIAL ()		
- Indicación de que se solicita el registro de Diseño Industrial	()	()
- Datos de identificación del solicitante o de la persona que presenta la solicitud.	()	()
- Representación gráfica y fotográfica del Diseño Industrial o muestra del Material que incorpora el diseño.	()	()
- Comprobante de pago de las tasas establecidas	()	()

COMPLETA () INCOMPLETA ()

ESQUEMA DE TRAZADO ()		
- Indicación de que solicita el registro de un Esquema de trazado.	()	()
- Datos de identificación del solicitante o de la persona que presenta La solicitud.	()	()
- Representación gráfica del esquema de trazado	()	()
- Comprobante de pago de las tasas establecidas.	()	()

COMPLETA () INCOMPLETA ()

Firma Responsable:

Edwin Navarro
05-06-2003

Fecha:

