Industria y Comercio

SUPERINTENDENCIA

Delegatura de propiedad Industrial

División de Nuevas Creaciones

PCT

SOLICITUD

PATENTE DE INVENCIÓN

21. **EXPEDIENTE No.** 048857

54. **TÍTULO** Antibioticos tipo florfenicol novedosos

51. **CLASIFICACIÓN INTERNACIONAL** A61K 31/165

71. **SOLICITANTE** schering-plough, Ltd

DOMICILIO Weystrasse 20, CH-6000, Lucerne 6, Switzerland.

74. **APODERADO** Carlos D Oberte

22. **BOGOTÁ, D. C.,** Septiembre 8, 2004

(FORMA P-10)

24/114 01

OLARTERAISBECK

Industria y Comercio
SUPERINTENDENCIA

SUPERINDUSTRIA Y COMERCIO
Radicación : 04000857 00000000 Folios: 232
Fecha (AMD): 2004-09-08 17:42:24
Tramite : 011 PCT-PATENT 379 FASE NAC1 411 PRESENTAC
Dependencia: 2020 DIVISION DE NUEVAS CREACIONES

FORMULARIO ÚNICO DE SOLICITUD DE PATENTE
PCT
ENTRADA EN FASE NACIONAL

(1) **SOLICITUD DE:**

[X] Patente de Invención. [ ] Patente de Modelo de Utilidad.

[ ] Capítulo I. [X] Capítulo II.

(2) **SOLICITANTE (71)**

Nombre: SCHERING-PLOUGH, LTD.
Dirección: Weystrasse 20, CH-6000, Lucerne 6, Switzerland
Nacionalidad o Domicilio: SUIZA
Lugar de Constitución: SUIZA
Teléfono: _____ Fax: _____
E-mail: _____

IDENTIFICACIÓN
C.C. [ ] NIT [ ]
C.E. [ ] Otro [ ]
Cual _____
Número _____

(3) **REPRESENTANTE O APODERADO**

Nombre: CARLOS R. OLARTE
Dirección: World Trade Center Torre A, Calle 100 Nº. 8A-37 Piso 10
Teléfono: 601-7700 Fax: 601-7799
E-mail: office@olarteraisbeck.com

IDENTIFICACIÓN
C.C. [X] NIT [ ]
C.E. [ ] Otro [ ]
Cual _____
Número 79.782.747 Btá
TP 74.295

(4) **INVENTOR (ES) (72)**

Nombre: VER PAGINA ADJUNTA
Dirección: VER PAGINA ADJUNTA
Nacionalidad o Domicilio: VER PAGINA ADJUNTA

(5) **PCT (86)**

Solicitud Internacional No. PCT/IB03/03140 Fecha 7 de Marzo de 2003
Publicación Internacional No. WO 03/077828 Fecha 25 de Septiembre de 2003

(6) **Título (54)** ANTIBIOTICOS TIPO FLORFENICOL NOVEDOSOS.

(7) **Clasificación Internacional (51)** A61K 31/165

(8) **Prioridad** SI [X] NO [ ]

(33) País de Origen	(32) Fecha	(31) Número de Solicitud
US	Marzo 8, 2002	10/094.688

(9) **Comprobante de pago No.** 66,565 **Fecha** 24 de Agosto de 2004

02

(10) ANEXOS

- [x] Comprobante de pago de la tasa de presentación de la solicitud.
- [ ] Documento que acredita la existencia y representación legal cuando el solicitante sea persona jurídica.
- [x] Poderes, si fuere el caso.
- [x] Documento de cesión del inventor al solicitante o a su causante.
- [ ] Declaraciones.
- [x] Copia de la solicitud internacional. (Resumen, descripción, reivindicaciones, dibujos).
- [x] Traducción de la solicitud internacional al castellano. (Resumen, descripción, reivindicaciones, dibujos).
- [x] 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 del 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.
- [x] De ser el caso, modificaciones efectuadas a la solicitud internacional.
- [x] Otros. Búsqueda Internacional

(11) FIGURA CARACTERÍSTICA:

(12) Solicito la concesión de la patente,

NOMBRE: CARLOS R. OLARTE FIRMA:

C.C. 79.782.747 Btá T.P. 74.295

ATTACHMENT A

BOOJAMRA, Constantine, G., citizen of the United States of America, residing at 1068 Ashbury Street, San Francisco, California, 94117, USA

CHONG, Lee, S., citizen of the United States of America, residing at 37469 Marsten Drive, Newark, California, 94560, USA

HECKER, Scott, J., citizen of the United States of America, residing at 16387 Englewood Avenue, Los Gatos, California, 95032, USA

GLINKA, Tomasz, W., citizen of the United States of America, residing at 21122 Patriot Way, Cupertino, California, 95014, USA

SHUSTER, Dale, E., citizen of the United States of America, residing at 267 Beechspring Road, South Orange, New Jersey, 07079, USA

04

SUPERINDUSTRIA Y COMERCIO
Radicacion : 04088857 00000000 Folios: 232
Fecha (AMD): 2004-09-08 17:42:24
Tramite : 811 PCT-PATENT 379 FASE NACI 411 PRESENTAC
Dependencia: 2020 DIVISION DE NUEVAS CREACIONES

NIT : 900176099-2

RECIBO OFICIAL DE CAJA : 04 - 66,365
FECHA : AGOSTO 24 DE 2004

C O N S I G N A C I O N

DEPOSITANTE	TIPO PAGO	BANCO	CUENTA	No. PAGO	FECHA PGO	Vr. PAGO
FREDERICK TORRES	CONSIGNACION	BANCO POPULAR	050-00110-6	31332018	23/08/2004	809,000.00

C O N C E P T O

CANT.	RENTISTICO	CONCEPTO	TOTAL CONCEPTO
1	50005-01-01 SOLICITUDES	1 TRAMITES DE SOL. DE PATENTE DE IN	422,000.
		TOTAL : $	422,000.00

SON: CUATROCIENTOS VEINTIDOS MIL PESOS

RESPONSABLE : _____

RECIBO DE CAJA APLICADO AL EXPEDIENTE No._____

Traducción oficial del Inglés por Marlén Neira Castro, resolución 2117 del Ministerio de Justicia de Colombia

APOSTILLA EN PODER DE REPRESENTACIÓN DE

SCHERING- PLOUGH LTD.

APOSTILLA

(Convención de la Haya del 5 de octubre de 1961)

1. País: Estados Unidos de América
   - Este documento Público
2. ha sido firmado por Marguerite La Corte
3. Quien ejerce funciones de Notario público de Nueva Jersey
4. Lleva el sello de Marguertie La Corte Notario

CERTIFICADO

5. En Trenton, Nueva Jersey
6. El 29 de agosto 2003
7. Por John E. McCormac Contador Publico- Tesorero de Nueva Jersey
8. No. A 180348
9. Gran Sello del Estado de Nueva Jersey
10. Firma legible de John MacCormac

PODER DE REPRESENTACIÓN

Bilingüe inglés- español otorgado en Kenilworth, Nueva Jersey, Estados Unidos, el 7 de agosto de 2003 y firmado por Edward H. Mazer - Representante Autorizado

CERTIFICADO NOTARIAL

Bilingüe inglés- español, fechado el 7 de agosto, 2003 certificando las funciones de Edward H. Mazer -Representante autorizado de Schering - Plough Ltd.

Las certificaciones anteriores se basan en la Resolución de Junta Directiva de Schering- Plough Ltd. fechada el 7 noviembre de 2002.

Sello: Marguerite La Corte- Notario público,

Estado de Nueva Jersey
Su comisión vence el 7 de octubre de 2003.

Sello en alto relieve del Notario Público

Es traducción fiel del inglés, con copia archivo para confrontación. Bogotá 6 septiembre, 2003.

10 OCT 2003

BOGOTÁ, D.C. - COLOMBIA

MARLEN NEIRA CASTRO
Traductor e Intérprete Oficial
Inglés - Español - Inglés
Resolución No. 2117 del 7 Oct 1987

APOSTILLE

(CONVENTION DE LA HAYE DU 5 OCTOBRE 1961)

1. COUNTRY: UNITED STATES OF AMERICA

2. THIS PUBLIC DOCUMENT HAS BEEN SIGNED BY:
MARGUERITE LA CORTE

3. ACTING IN THE CAPACITY OF:
NOTARY PUBLIC OF NEW JERSEY

4. BEARS THE SEAL/STAMP OF:
MARGUERITE LA CORTE, NOTARY

CERTIFIED

5. AT TRENTON, NEW JERSEY

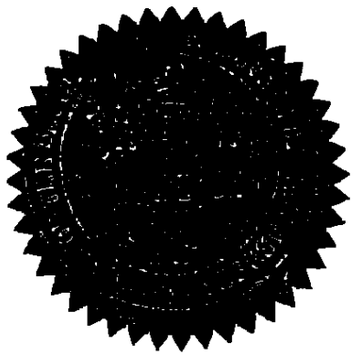
6. THE 29TH DAY OF AUGUST 2003

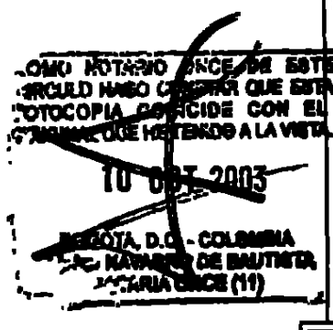
7. BY: John E McCormac, CPA, NEW JERSEY TREASURER

8. NO. A180348

9. SEAL/STAMP:

10. SIGNATURE





07

OLARTERAISBECK

OLARTE RAISBECK & FRIERI, LTDA.
LEGAL & PROFESSIONAL SERVICES

WORLD TRADE CENTER – TORRE C
CALLE 100 No. 8A-55 Piso 10
BOGOTA, COLOMBIA
TELEPHONE: +57-1 638-6200
Fax: +57-1 638-6201

www.olarteraisbeck.com

Power of Attorney

The undersigned,
Schering-Plough Ltd.
domiciled in
Toepferstrasse 5, CH 6004 Lucerne, Switzerland
by means of these presents hereby grants to Carlos R. Olarte and/or Ana María Frieri full and sufficient Power of Attorney to file, prosecute, obtain, defend and enforce any and all of the company's Patents, Utility Models and Industrial Designs in the Republic of Colombia, to which end it empowers them to take all steps necessary before any administrative or judicial authority of any class for the objects stated, to file petitions, appeals, replies and oppositions, pay taxes and annuities, request testimonies, receive documents and securities, abandon applications and collect refunds. Sufficient power is hereby granted to receive notifications on behalf of the Company, to confer powers on those who are to represent the company judicially and extrajudicially, to answer in court in the matter of all claims and demands that may be presented in connection with the objects stated, giving them likewise power to substitute these presents and revoke such substitutions.

Poder

El suscrito,
domiciliado en
por medio del presente otorga a Carlos R. Olarte y/o Ana María Frieri poder especial, amplio y suficiente para presentar, tramitar, obtener, defender y hacer respetar todas y cualquiera de Patentes, Modelos de Utilidad y Diseños Industriales de la compañía en la República de Colombia, a cuyo efecto los faculta para dar todos los pasos necesarios ante cualquier autoridad administrativa o judicial de cualquier jerarquía para cumplir con lo encomendado, elevar solicitudes, apelaciones, respuestas y oposiciones, pagar impuestos y anualidades, solicitar testimonios, recibir documentos y valores, desistir y percibir. Se concede poder bastante para recibir notificaciones a nombre de la compañía, otorgar poderes a quienes vayan a representar a la compañía judicial y extrajudicialmente, responder en juicio a todas las reclamaciones o demandas que por motivo de lo encomendado se presentaren, dándoles asimismo facultad para sustituir el presente y revocar dichas sustituciones.

Signature/Firma: _____
Edward H. Mazer, Duly Authorized Representative

Date/Fecha: August 7, 2003

City,Country/Ciudad,Pais: Kenilworth, New Jersey, U.S.A.

COMO NOTARIO ONCE DE ESTE CIRCULO HAGO CONSTAR QUE ESTA FOTOCOPIA COINCIDE CON EL ORIGINAL QUE HE TENIDO A LA VISTA.
10 OCT 2003
BOGOTA, D.C. - COLOMBIA
NOTARIA ONCE (11)

08

OLARTERAISBECK

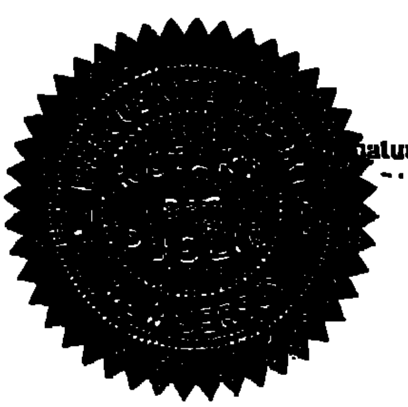
OLARTE RAISBECK & FRIERI, LTDA.
LEGAL & PROFESSIONAL SERVICES

WORLD TRADE CENTER – TORRE C
CALLE 100 NO. 8A-55 PISO 10
BOGOTA, COLOMBIA
TELEPHONE: +57-1 638-6200
FAX +57-1 638-6201

www.olarteraisbeck.com

Notarial Certification	Certificación Notarial
On this date, August 7, 2003	En esta fecha,
The undersigned Notary Public certifies:	el Notario Público suscrito certifica:
1. That Edward H. Mazer	1. Que
of legal age and domiciled in 119 Greenwood Drive, Millburn, New Jersey 07041, U.S.A. signed the foregoing document.	mayor de edad y domiciliado suscribió el documento que antecede.
2. That the grantor has executed the foregoing document as Duly Authorized Representative of Schering-Plough Ltd.	2. Que el otorgante ha suscrito el referido documento en su carácter de de
3. That Schering-Plough Ltd. having its principal place of business located in Toepferstrasse 5, CH 6004 Lucerne, Switzerland is duly incorporated, is currently in existence, and that the purposes for which the Power of Attorney is granted are within the scope of its corporate purposes.	4. Que con sede principal ubicada en está legalmente constituida, que tiene existencia legal vigente y que los propósitos para los cuales se otorga el poder se encuentra dentro de los que comprenden su objeto social.
4. That the grantor has the authority to execute the Power of Attorney and that his/her representation is legal.	4. Que el otorgante tiene la representación para suscribir el poder, y que ésta es legítima.
5. The foundation for my certifications contained in points 2, 3 and 4 was the exhibition of the following authentic document(s):	5. El fundamento de mis certificaciones contenidas en los puntos 2, 3 y 4 fue la exhibición de los (del) siguiente(s) documento(s) auténtico(s):

Resolution of the Board of Directors
of Schering-Plough Ltd. of November 7, 2002.



(Signature):

COINCIDE CON EL ORIGINAL QUE HE TENIDO A LA VISTA.
OCT 2003
BOGOTA, D.C. - COLOMBIA
NOTARIA ONCE (11)

Marguerite La Corte
Notary Public of New Jersey
My Commission Expires October 7, 2003

ASSIGNMENT

WHEREAS, **SCHERING-PLOUGH ANIMAL HEALTH C RPORATION**, a corporation organized and existing under the laws of the State of Delaware, United States of America, having its principal office at 1095 Morris Avenue, Union, New Jersey 07083-7143, United States of America ("**ASSIGNOR**"), is the owner of record of patent applications entitled:

NOVEL FLORFENICOL-TYPE ANTIBIOTICS

and officially identified by the United States Patent and Trademar< Office as Application Number 10/094,688, filed on March 8, 2002, Application Number 10/410,330, filed on April 8, 2003, and/or by the World Intellectual Property Organization as International Patent Application Number PCT/IB03/03140, filed on March 7, 2003, by virtue of an assignment recorded on Reel_____, Frame _____, in the United States Patent and Trademark Office, Washington, D.C., including all inventions described and/or claimed in said patent(s) and/or patent application(s);

WHEREAS, **SCHERING-PLOUGH LTD.**, a corporation organized and existing under the laws of Switzerland, having its principal office at Weystrasse 20, P.O. Box CH-6000, Lucerne 6, Switzerland ("**ASSIGNEE**"), is desirous of acquiring **ASSIGNOR'S** interest in said inventions, patent applications and all resulting patents based thereon excluding the patents granted in the United States;

NOW, THEREFORE, for good and valuable consideration, the receipt of which is hereby acknowledged, **ASSIGNOR** hereby sells, assigns, and transfers to **ASSIGNEE**, all of **ASSIGNOR'S** right, title, and interest in said inventions and patent applications, including all counterpart applications, patents, extensions, supplementary protection certificates, and the like, based upon said inventions and patent applications with the exception of those held in the United States, together with **ASSIGNOR'S** right to sue and collect damages for any infringement, for its own use and benefit and for its successors and assigns, as fully and entirely as the same would have been held by **ASSIGNOR** had this Assignment and transfer not been made.

Executed on this 26th day of August, 2004.

Assignor: SCHERING-PLOUGH ANIMAL HEALTH CORPORATION

By: _____

Name: Joel Wiener

Title: Vice President and Counsel

ACKNOWLEDGEMENT

GLOBAL LAW DEPT. REVIEW

State of New Jersey)

) s.s.:

County of Union)

On 26th day of August, 2004, personally appeared before me Joel Wiener to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he executed the same, of his own free will for the purpose set forth.

(Seal)

_____

Notary Public

Assignor: SCHERING-PLOU H ANIMAL HEALTH CORPORATION

By: _____

Name: Joel Wiener

Title: Vice President and Counsel

GLOBAL LAW DEPT. REVIEW

ACKNOWLEDGEMENT

State of New Jersey)

) s.s.:

County of Union)

On 26th day of August, 2004, personally appeared before me Joel Wiener to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he executed the same, of his own free will for the purpose set forth.

(Seal)

_____

Notary Public

SHERYL A. HOHMANN
NOTARY PUBLIC OF NEW JERSEY
Commission Expires 10/30/2007

Assignee: SCHERING-PLOUGH LTD.

By: Richard A. Elder

Name: Richard A. Elder

Title: Director, Patents

ACKNOWLEDGEMENT

State of New Jersey)
) s.s.:
County of Union)

On 26th day of August, 2004, personally appeared before me Richard A. Elder to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he executed the same, of his own free will for the purpose set forth.

Sheryl A. Hohmann

Notary Public

SHERYL A. HOHMANN
NOTARY PUBLIC OF NEW JERSEY
Commission Expires 10/26/2007

35574

Case AH06114

ACKNOWLEDGEMENT

State of New Jersey }
} s.s
County of Morris }

I hereby certify that the annexed is a true copy of the original Assignment to **SCHERING-PLOUGH ANIMAL HEALTH CORPORATION**, of a patent application entitled **"NOVEL FLORFENICOL-TYPE ANTIBIOTICS"**, said Assignment having been executed on December 12, 2003, by Constantine G. Boojamra.

August 5, 2004

August 5, 2004

Shawn Armstrong
Notary Public of New Jersey
My Commission Expires May of 2005

SHAWN M ARMSTRONG
A Notary Public of New Jersey
My Commission Expires May 15, 2005

(Seal)

138150-1

(Joint)

PATENT CASE: AH06114 01

ASSIGNMENT

For good and valuable consideration paid to us,

(1) Constantine G. Boojamra / (4) Tomasz W. Glinka /

(2) Lee S. Chong / (5) Dale E. Shuster /

(3) Scott J. Hecker /

respectively residing at

(1) San Francisco, California (4) Cupertino, California

(2) Newark, California (5) South Orange, New Jersey

(3) Lost Gatos, California

by **SCHERING-PLOUGH ANIMAL HEALTH CORPORATION**, a corporation organized under the laws of the State of Delaware, United States of America, having its principal office at 1095 Morris Avenue, Union, New Jersey 07083 , United States of America, (hereinafter called "**ASSIGNEE**"), we each do hereby sell, assign and transfer to said **ASSIGNEE** each of our entire right, title and interest in all countries of the world in and to any and all of our inventions and discoveries described in the patent application entitled

NOVEL FLORFENICOL-TYPE ANTIBIOTICS

and officially identified* by the United States Patent and Trademark Office as Application Number 10/410,330 filed on April 8, 2003, and/or by the World Intellectual Property Organization as International Patent Application Number IB03/03140 filed on March 7, 2003 , in and to the right to file patent applications in the name of **ASSIGNEE**, its designee, or in any or all of our names, at its election, on the aforesaid inventions and discoveries in all countries of the world, together with all rights of priority in the aforesaid countries deriving from the above-identified patent application under the International Convention for the Protection of Industrial Property, under the Inter-American Convention relating to Inventions, Patents, Designs and Industrial Models and under any other international arrangement to which the United States now is or hereafter becomes a signatory, in and to any and all Letters Patent that issue on any of the aforesaid patent applications, in and to any applications claiming priority benefits to a provisional patent application or other applications filed on said inventions, and in and to any continuations, divisions, reissues, renewals and extensions thereof of any of said Letters Patent, the same to be held and enjoyed by said **ASSIGNEE**, its successors, assigns and other legal representatives, to the full ends of the terms for which a Letters Patent therefor may be granted, as fully and entirely as the same would have been held and enjoyed by us if this assignment and sale had not been made.

And we hereby covenant and agree that we will at any time, upon the request and at the expense of **ASSIGNEE**, execute and deliver any and all documents that may be necessary or desirable to perfect the title to the foregoing inventions and discoveries, patent applications, and Letters Patent and continuations, divisions, reissues, renewals and extensions thereof in **ASSIGNEE**, its successors, assigns or other legal representatives, including the execution and procurement of any and all further documents evidencing this assignment and sale as may be necessary or desirable for recording the same in the Patent Office of any country concerned, and that we will, at any time, upon the request and at the expense of **ASSIGNEE**,

execute any additional or divisional applications for patents for said inventions and discoveries, or any part or parts thereof, and applications for patents of confirmation, registration and importation based on said Letters Patent and on Letters Patent issuing from said additional or divisional applications and reissues, renewals and extensions therefor, and will make all rightful oaths and declarations and do all lawful acts requisite for procuring the same or for aiding therein, without further compensation, but at the expense of **ASSIGNEE**, its successors, assigns or other legal representatives.

"We hereby authorize **ASSIGNEE** to insert in this instrument the Application Number and the filing date of said application for Letters Patent when officially notified thereof.

Executed this 12th day of Dec, 2006.

_____ L.S.

CONSTANTINE G. BOOJAMRA

ACKNOWLEDGEMENT

State of California)
) s.s.:
County of San Mateo)

On this 12th day of Dec, 2006, personally appeared before me Constantine G. Boojamra to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (~~she~~) executed the same, of his (~~her~~) own free will for the purpose set forth.

(Seal)

JOSE J. SAN GABRIEL
Commission # 1428433
Notary Public - California
San Mateo County
My Comm. Expires Jul 26, 2007

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

LEE S. CHONG

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Lee S. Chong** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

SC TT J. HECKER

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Scott J. Hecker** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

T MASZ W. GLINKA

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me Tomasz W. Glinka to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

DALE E. SHUSTER

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me Dale E. Shuster to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Case AH06114

ACKNOWLEDGEMENT

State of New Jersey }
} s.s
County of Morris }

I hereby certify that the annexed is a true copy of the original Assignment to **SCHERING-PLOUGH ANIMAL HEALTH CORPORATION**, of a patent application entitled **"NOVEL FLORFENICOL-TYPE ANTIBIOTICS"**, said Assignment having been executed on November 21, 2003, by Lee S. Chong.

August 5, 2004
August 5, 2004

Shawn Armstrong
Notary Public of New Jersey
My Commission Expires May of 2005

SHAWN M ARMSTRONG
A Notary Public of New Jersey
My Commission Expires May 15, 2005

(Seal)
136150-1

(Joint)

PATENT CASE: AH06114 01

ASSIGNMENT

For good and valuable consideration paid to us,

(1) Constantine G. Boojamra (4) Tomasz W. Glinka

(2) Lee S. Chong (5) Dale E. Shuster

(3) Scott J. Hecker

respectively residing at

(1) San Francisco, California (4) Cupertino, California

(2) Newark, California (5) South Orange, New Jersey

(3) Lost Gatos, California

by **SCHERING-PLOUGH ANIMAL HEALTH CORPORATION**, a corporation organized under the laws of the State of Delaware, United States of America, having its principal office at 1095 Morris Avenue, Union, New Jersey 07083 , United States of America, (hereinafter called "**ASSIGNEE**"), we each do hereby sell, assign and transfer to said **ASSIGNEE** each of our entire right, title and interest in all countries of the world in and to any and all of our inventions and discoveries described in the patent application entitled

NOVEL FLORFENICOL-TYPE ANTIBIOTICS

and officially identified* by the United States Patent and Trademark Office as Application Number 10/410,330 filed on April 8, 2003, and/or by the World Intellectual Property Organization as International Patent Application Number IB03/03140 filed on March 7, 2003 , in and to the right to file patent applications in the name of **ASSIGNEE**, its designee, or in any or all of our names, at its election, on the aforesaid inventions and discoveries in all countries of the world, together with all rights of priority in the aforesaid countries deriving from the above-identified patent application under the International Convention for the Protection of Industrial Property, under the Inter-American Convention relating to Inventions, Patents, Designs and Industrial Models and under any other international arrangement to which the United States now is or hereafter becomes a signatory, in and to any and all Letters Patent that issue on any of the aforesaid patent applications, in and to any applications claiming priority benefits to a provisional patent application or other applications filed on said inventions, and in and to any continuations, divisions, reissues, renewals and extensions thereof of any of said Letters Patent, the same to be held and enjoyed by said **ASSIGNEE**, its successors, assigns and other legal representatives, to the full ends of the terms for which a Letters Patent therefor may be granted, as fully and entirely as the same would have been held and enjoyed by us if this assignment and sale had not been made.

And we hereby covenant and agree that we will at any time, upon the request and at the expense of **ASSIGNEE**, execute and deliver any and all documents that may be necessary or desirable to perfect the title to the foregoing inventions and discoveries, patent applications, and Letters Patent and continuations, divisions, reissues, renewals and extensions thereof in **ASSIGNEE**, its successors, assigns or other legal representatives, including the execution and procurement of any and all further documents evidencing this assignment and sale as may be necessary or desirable for recording the same in the Patent Office of any country concerned, and that we will, at any time, upon the request and at the expense of **ASSIGNEE**,

execute any additional or divisional applications for patents for said inventions and discoveries, or any part or parts thereof, and applications for patents of confirmation, registration and importation based on said Letters Patent and on Letters Patent issuing from said additional or divisional applications and reissues, renewals and extensions therefor, and will make all rightful oaths and declarations and do all lawful acts requisite for procuring the same or for aiding therein, without further compensation, but at the expense of **ASSIGNEE**, its successors, assigns or other legal representatives.

*We hereby authorize **ASSIGNEE** to insert in this instrument the Application Number and the filing date of said application for Letters Patent when officially notified thereof.

Executed this _____ day of _____ , 20_____.

_____ L.S.

C NSTANTINE G. BOOJAMRA

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Constantine G. Boojamra** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

 PATENT CASE: AH06114 01

Executed this 21 day of Nov, 20 03.

_____ L.S.

LEE S. CHONG

ACKNOWLEDGEMENT

State of California)
) s.s.:
County of San Mateo)

On this 21st day of November, 2003 personally appeared before me Lee S. Chong to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

JOSE J. SAN GABRIEL
Commission # 1428433
Notary Public - California
San Mateo County
My Comm. Expires Jul 28, 2007

_____
Notary Public

Executed this _____ day of _____, 20_____.

_____ L.S.

SCOTT J. HECKER

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____, 20____, personally appeared before me Scott J. Hecker to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

TOMASZ W. GLINKA

ACKN WLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Tomasz W. Glinka** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

DALE E. SHUSTER

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Dale E. Shuster** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Case AH06114

ACKNOWLEDGEMENT

State of New Jersey }
} s.s
County of Morris }

I hereby certify that the annexed is a true copy of the original Assignment to **SCHERING-PLOUGH ANIMAL HEALTH CORPORATION**, of a patent application entitled **"NOVEL FLORFENICOL-TYPE ANTIBIOTICS"**, said Assignment having been executed on December 5, 2003, by Scott J. Hecker.

August 5, 2004

Shawn Armstrong
Notary Public of New Jersey
My Commission Expires May of 2005

SHAWN M ARMSTRONG
A Notary Public of New Jersey
My Commission Expires May 15, 2005

(Seal)

136150-1

(Joint)

PATENT CASE: AH06114 01

ASSIGNMENT

For good and valuable consideration paid to us,

(1) Constantine G. Boojamra
(2) Lee S. Chong
(3) Scott J. Hecker
(4) Tomasz W. Glinka
(5) Dale E. Shuster

respectively residing at

(1) San Francisco, California
(2) Newark, California
(3) Lost Gatos, California
(4) Cuptertino, California
(5) South Orange, New Jersey

by SCHERING-PLOUGH ANIMAL HEALTH CORPORATION, a corporation organized under the laws of the State of Delaware, United States of America, having its principal office at 1095 Morris Avenue, Union, New Jersey 07083 , United States of America, (hereinafter called "ASSIGNEE"), we each do hereby sell, assign and transfer to said ASSIGNEE each of our entire right, title and interest in all countries of the world in and to any and all of our inventions and discoveries described in the patent application entitled

NOVEL FLORFENICOL-TYPE ANTIBIOTICS

and officially identified* by the United States Patent and Trademark Office as Application Number 10/410,330 filed on April 8, 2003, and/or by the World Intellectual Property Organization as International Patent Application Number IB03/03140 filed on March 7, 2003 , in and to the right to file patent applications in the name of ASSIGNEE, its designee, or in any or all of our names, at its election, on the aforesaid inventions and discoveries in all countries of the world, together with all rights of priority in the aforesaid countries deriving from the above-identified patent application under the International Convention for the Protection of Industrial Property, under the Inter-American Convention relating to Inventions, Patents, Designs and Industrial Models and under any other international arrangement to which the United States now is or hereafter becomes a signatory, in and to any and all Letters Patent that issue on any of the aforesaid patent applications, in and to any applications claiming priority benefits to a provisional patent application or other applications filed on said inventions, and in and to any continuations, divisions, reissues, renewals and extensions thereof of any of said Letters Patent, the same to be held and enjoyed by said ASSIGNEE, its successors, assigns and other legal representatives, to the full ends of the terms for which a Letters Patent therefor may be granted, as fully and entirely as the same would have been held and enjoyed by us if this assignment and sale had not been made.

And we hereby covenant and agree that we will at any time, upon the request and at the expense of ASSIGNEE, execute and deliver any and all documents that may be necessary or desirable to perfect the title to the foregoing inventions and discoveries, patent applications, and Letters Patent and continuations, divisions, reissues, renewals and extensions thereof in ASSIGNEE, its successors, assigns or other legal representatives, including the execution and procurement of any and all further documents evidencing this assignment and sale as may be necessary or desirable for recording the same in the Patent Office of any country concerned, and that we will, at any time, upon the request and at the expense of ASSIGNEE,

execute any additional or divisional applications for patents for said inventions and discoveries, or any part or parts thereof, and applications for patents of confirmation, registration and importation based on said Letters Patent and on Letters Patent issuing from said additional or divisional applications and reissues, renewals and extensions therefor, and will make all rightful oaths and declarations and do all lawful acts requisite for procuring the same or for aiding therein, without further compensation, but at the expense of **ASSIGNEE**, its successors, assigns or other legal representatives.

*We hereby authorize **ASSIGNEE** to insert in this instrument the Application Number and the filing date of said application for Letters Patent when officially notified thereof.

Executed this _____ day of _____ , 20_____.

_____ L.S.

CONSTANTINE G. BOOJAMRA

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Constantine G. Boojamra** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

LEE S. CHONG

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Lee S. Chong** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this 5 day of December , 2003 .

Scott J Hecker L.S.

SCOTT J. HECKER

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Scott J. Hecker** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

SEE ATTACHED CALIFORNIA ALL-PURPOSE ACKNOWLEDGMENT

CALIF RNIA ALL-PURPOSE ACKN WLEDGMENT

State of California
County of San Diego } ss.

On December 5, 2003 (Date) before me. Elisa Lynne Weston, Notary Public (Name and Title of Officer e.g. "Jane Doe, Notary Public")

personally appeared Scott J. Hecker (Name(s) of Signer(s))

X personally known to me
~~proved to me on the basis of satisfactory evidence~~

to be the person(s) whose name(s) is/are subscribed to the within instrument and acknowledged to me that he/she/they executed the same in his/her/their authorized capacity(ies), and that by his/her/their signature(s) on the instrument the person(s), or the entity upon behalf of which the person(s) acted, executed the instrument.

WITNESS my hand and official seal.

ELISA LYNNE WESTON
Commission # 1244152
Notary Public - California
San Diego County
My Comm. Expires Dec 25, 2003

OPTIONAL

Though the information below is not required by law, it may prove valuable to persons relying on the document and could prevent fraudulent removal and reattachment of this form to another document.

Description of Attached Document

Title or Type of Document: Assignment

Document Date: Signed 12/5/03 Number of Pages 4

Signer(s) Other Than Named Above: Constantine G. Boojamra, Lee S. Chong, Thomasz W. Glinka, and Dale E. Shuster

Capacity(ies) Claimed by Signer

Signer's Name: Scott J. Hecker

RIGHT THUMBPRINT OF SIGNER

XX Individual
Corporate Officer — Title(s):
Partner — Limited General
Attorney-in-Fact
Trustee
Guardian or Conservator
Other:

Signer Is Representing: Self

Executed this _____ day of _____ , 20_____.

_____ L.S.

TOMASZ W. GLINKA

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me Tomasz W. Glinka to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

DALE E. SHUSTER

ACKN WLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me Dale E. Shuster to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Case AH06114

ACKNOWLEDGEMENT

State of New Jersey }
} s.s
County of Morris }

I hereby certify that the annexed is a true copy of the original Assignment to **SCHERING-PLOUGH ANIMAL HEALTH CORPORATION**, of a patent application entitled **"NOVEL FLORFENICOL-TYPE ANTIBIOTICS"**, said Assignment having been executed on November 13, 2003, by Tomasz W. Glinka.

August 5, 2004
August 5, 2004

Shawn Armstrong
Notary Public of New Jersey
My Commission Expires May of 2005

SHAWN M ARMSTRONG
A Notary Public of New Jersey
My Commission Expires May 15, 2005

(Seal)
136150-1

(Joint)

PATENT CASE: AH06114 01

ASSIGNMENT

For good and valuable consideration paid to us,

(1) Constantine G. Boojamra (4) Tomasz W. Glinka

(2) Lee S. Chong (5) Dale E. Shuster

(3) Scott J. Hecker

respectively residing at

(1) San Francisco, California (4) Cuptertino, California

(2) Newark, California (5) South Orange, New Jersey

(3) Lost Gatos, California

by **SCHERING-PLOUGH ANIMAL HEALTH CORPORATION**, a corporation organized under the laws of the State of Delaware, United States of America, having its principal office at 1095 Morris Avenue, Union, New Jersey 07083 , United States of America, (hereinafter called "**ASSIGNEE**"), we each do hereby sell, assign and transfer to said **ASSIGNEE** each of our entire right, title and interest in all countries of the world in and to any and all of our inventions and discoveries described in the patent application entitled

NOVEL FLORFENICOL-TYPE ANTIBIOTICS

and officially identified* by the United States Patent and Trademark Office as Application Number 10/410,330 filed on April 8, 2003, and/or by the World Intellectual Property Organization as International Patent Application Number IB03/03140 filed on March 7, 2003 , in and to the right to file patent applications in the name of **ASSIGNEE**, its designee, or in any or all of our names, at its election, on the aforesaid inventions and discoveries in all countries of the world, together with all rights of priority in the aforesaid countries deriving from the above-identified patent application under the International Convention for the Protection of Industrial Property, under the Inter-American Convention relating to Inventions, Patents, Designs and Industrial Models and under any other international arrangement to which the United States now is or hereafter becomes a signatory, in and to any and all Letters Patent that issue on any of the aforesaid patent applications, in and to any applications claiming priority benefits to a provisional patent application or other applications filed on said inventions, and in and to any continuations, divisions, reissues, renewals and extensions thereof of any of said Letters Patent, the same to be held and enjoyed by said **ASSIGNEE**, its successors, assigns and other legal representatives, to the full ends of the terms for which a Letters Patent therefor may be granted, as fully and entirely as the same would have been held and enjoyed by us if this assignment and sale had not been made.

And we hereby covenant and agree that we will at any time, upon the request and at the expense of **ASSIGNEE**, execute and deliver any and all documents that may be necessary or desirable to perfect the title to the foregoing inventions and discoveries, patent applications, and Letters Patent and continuations, divisions, reissues, renewals and extensions thereof in **ASSIGNEE**, its successors, assigns or other legal representatives, including the execution and procurement of any and all further documents evidencing this assignment and sale as may be necessary or desirable for recording the same in the Patent Office of any country concerned, and that we will, at any time, upon the request and at the expense of **ASSIGNEE**,

execute any additional or divisional applications for patents for said inventions and discoveries, or any part or parts thereof, and applications for patents of confirmation, registration and importation based on said Letters Patent and on Letters Patent issuing from said additional or divisional applications and reissues, renewals and extensions therefor, and will make all rightful oaths and declarations and do all lawful acts requisite for procuring the same or for aiding therein, without further compensation, but at the expense of **ASSIGNEE**, its successors, assigns or other legal representatives.

*We hereby authorize **ASSIGNEE** to insert in this instrument the Application Number and the filing date of said application for Letters Patent when officially notified thereof.

Executed this _____ day of _____ , 20_____.

_____ L.S.

CONSTANTINE G. BOOJAMRA

ACKN WLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Constantine G. Boojamra** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

LEE S. CHONG

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me Lee S. Chong to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

SCOTT J. HECKER

ACKN WLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me Scott J. Hecker to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

 PATENT CASE: AH06114 01

Executed this 13 day of November, 2003.

_____ L.S.

TOMASZ W. GLINKA

ACKNOWLEDGEMENT

State of California)
) s.s.:
County of San Diego)

On this 13th day of Nov., 2003, personally appeared before me Tomasz W. Glinka ~~to me known, and known by me~~ to be the same person described in and who executed the foregoing instrument, and acknowledged that ~~he (she) executed~~ the same, of his ~~(her)~~ own free will for the purpose set forth.

(Seal)

RAJENDRA K. DHAR, #1241591, NOTARY PUBLIC-CALIFORNIA, San Diego County, My Comm. Expires Jan 27, 2007

_____
Notary Public

Executed this _____ day of _____, 20_____.

_____ L.S.

DALE E. SHUSTER

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____, 20____, personally appeared before me Dale E. Shuster ~~to me known, and known by me~~ to be the same person described in and who executed the foregoing instrument, and acknowledged that ~~he (she)~~ executed the same, of his ~~(her)~~ own free will for the purpose set forth.

(Seal)

_____
Notary Public

Case AH06114

ACKNOWLEDGEMENT

State of New Jersey }
} s.s
County of Morris }

I hereby certify that the annexed is a true copy of the original Assignment to **SCHERING-PLOUGH ANIMAL HEALTH CORPORATION**, of a patent application entitled **"NOVEL FLORFENICOL-TYPE ANTIBIOTICS"**, said Assignment having been executed on November 17, 2003, by Dale E. Shuster.

August 5, 2004

August 5, 2004

Shawn Armstrong
Notary Public of New Jersey
My Commission Expires May of 2005

SHAWN M ARMSTRONG
A Notary Public of New Jersey
My Commission Expires May 15, 2005

(Seal)

136150-1

(Joint)

PATENT CASE: AH06114 01

ASSIGNMENT

For good and valuable consideration paid to us,

(1) Constantine G. Boojamra (4) Tomasz W. Glinka

(2) Lee S. Chong (5) Dale E. Shuster

(3) Scott J. Hecker

respectively residing at

(1) San Francisco, California (4) Cupertino, California

(2) Newark, California (5) South Orange, New Jersey

(3) Lost Gatos, California

by **SCHERING-PLOUGH ANIMAL HEALTH CORPORATION**, a corporation organized under the laws of the State of Delaware, United States of America, having its principal office at 1095 Morris Avenue, Union, New Jersey 07083 , United States of America, (hereinafter called "**ASSIGNEE**"), we each do hereby sell, assign and transfer to said **ASSIGNEE** each of our entire right, title and interest in all countries of the world in and to any and all of our inventions and discoveries described in the patent application entitled

NOVEL FLORFENICOL-TYPE ANTIBIOTICS

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And we hereby covenant and agree that we will at any time, upon the request and at the expense of **ASSIGNEE**, execute and deliver any and all documents that may be necessary or desirable to perfect the title to the foregoing inventions and discoveries, patent applications, and Letters Patent and continuations, divisions, reissues, renewals and extensions thereof in **ASSIGNEE**, its successors, assigns or other legal representatives, including the execution and procurement of any and all further documents evidencing this assignment and sale as may be necessary or desirable for recording the same in the Patent Office of any country concerned, and that we will, at any time, upon the request and at the expense of **ASSIGNEE**,

execute any additional or divisional applications for patents for said inventions and discoveries, or any part or parts thereof, and applications for patents of confirmation, registration and importation based on said Letters Patent and on Letters Patent issuing from said additional or divisional applications and reissues, renewals and extensions therefor, and will make all rightful oaths and declarations and do all lawful acts requisite for procuring the same or for aiding therein, without further compensation, but at the expense of **ASSIGNEE**, its successors, assigns or other legal representatives.

*We hereby authorize **ASSIGNEE** to insert in this instrument the Application Number and the filing date of said application for Letters Patent when officially notified thereof.

Executed this _____ day of _____, 20_____.

_____ L.S.

CONSTANTINE G. BOOJAMRA

ACKN WLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____, 20____, personally appeared before me Constantine G. Boojamra to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

LEE S. CHONG

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Lee S. Chong** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____ , 20_____.

_____ L.S.

SCOTT J. HECKER

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____ , 20____, personally appeared before me **Scott J. Hecker** to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this _____ day of _____, 20_____.

_____ L.S.

TOMASZ W. GLINKA

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this _____ day of _____, 20____, personally appeared before me Tomasz W. Glinka to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

Executed this 17th day of November, 2003.

_____ L.S.

DALE E. SHUSTER

ACKNOWLEDGEMENT

State of _____)
) s.s.:
County of _____)

On this 17th day of November, 2003, personally appeared before me Dale E. Shuster to me known, and known by me to be the same person described in and who executed the foregoing instrument, and acknowledged that he (she) executed the same, of his (her) own free will for the purpose set forth.

(Seal)

_____
Notary Public

DELIA NAAB
Notary Public, State of New Jersey
Qualified in Union/Middlesex Counties
My Commission Expires Aug. 20. 2007

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

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25 September 2003 (25.09.2003)

PCT

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10/094,688 8 March 2002 (08.03.2002) US

(71) Applicant: SCHERING-PLOUGH, LTD. [CH/CH]; Toepferstrasse 5, CH-6000 Lucerne 6 (CH).

(72) Inventors: BOOJAMRA, Constantine, G.; 1068 Ashbury Street, San Francisco, CA 94117 (US). CHONG, Lee, S.; 37469 Marsten Drive, Newark, Ca 94560 (US). HECKER, Scott, J.; 16387 Englewood Avenue, Los Gatos, CA 95032 (US). GLINKA, Tomasz, W.; 21122 Patriot Way, Cupertino, CA 95014 (US). SCHUSTER, Dale, E.; 267 Beechspring Road, South Orange, NJ 07079 (US).

(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, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VC, VN, YU, ZA, ZM, ZW.

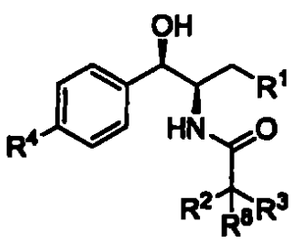
(84) Designated States *regional*: ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

Published:
without international search report and to be republished upon receipt of that 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.

WO 03/077828 A2

(54) Title: NOVEL FLORFENICOL-TYPE ANTIBIOTICS



(I)

(57) Abstract: The present invention relates to novel florfenicol compounds having the chemical structure (I) wherein the compounds are useful for the treatment and/or prevention of bacterial infections in a broad range of patients such as, without limitation, birds, fish, shellfish and mammals.

NOVEL FLORFENICOL-TYPE ANTIBIOTICS

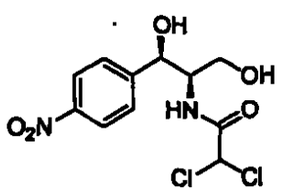
FIELD OF THE INVENTION

The present invention relates to the fields of organic chemistry, pharmaceutical chemistry, biochemistry and medicine. In particular, it relates to novel florfenicol-type antibiotics.

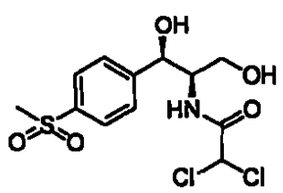
BACKGROUND OF THE INVENTION

Florfenicol is a broad spectrum antibiotic with activity against many gram-negative and gram-positive bacteria. Florfenicol is useful for the prevention and treatment of bacterial infections due to susceptible pathogens in birds, reptiles, fish, shellfish and mammals. One of its primary uses is in the treatment of pneumonia and associated respiratory infections in cattle (often referred to generically as Bovine Respiratory Disease or BRD) caused by *Mannhemia haemolytica*, *Pasturella multocida* and(or) *Haemophilus somnus*. It is also indicated in the treatment of pododermatitis in cattle caused by *Fusobacterium necrophorum* and *Bacterioides melaninogenicus*, swine respiratory disease caused by *Pasteurella multocida*, *Actinobacillus pleuropneumoniae*, *Streptococcus suis*, *Salmonella cholerasuis* and(or) *Mycoplasma* spp., colibacillosis in chickens caused by *Escherichia coli*, enteric septicemia in catfish caused by *Edwardsiella ictaluri*, and furunculosis in salmon caused by *Aeromonas salmonicida*. Other genera of bacteria that have exhibited susceptibility to florfenicol include *Enterobacter*, *Klebsiella*, *Staphylococcus*, *Enterococcus*, *Bordetella*, *Proteus*,and *Shigella*. In particular, chloramphenicol resistant strains of organisms such as *K. pneumoniae*, *E. cloacae*, *S. typhus* and *E. coli* are susceptible to florfenicol.

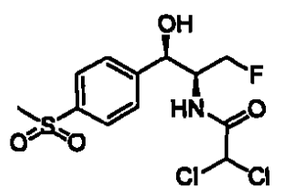
Florfenicol is a structural analog of thiamphenicol, which in turn is a derivative of chloramphenicol in which the aromatic nitro group, which nitro group has been implicated in



chloramphenicol



thiamphenicol



florfenicol

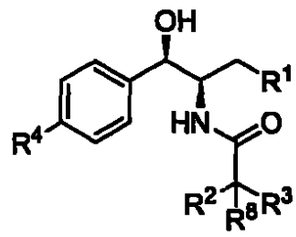
chloramphenicol-induced, non-dose related irreversible aplastic anemia in humans, is replaced with a methylsulfonyl group. Florfenicol has a fluorine atom in place of the primary hydroxyl group of chloramphenicol and thiamphenicol. This renders florfenicol less susceptible to deactivation by bacteria containing the plasmid-encoded enzyme, chloramphenicol acetyl transferase (CAT), which acetylates the primary hydroxyl group of chloramphenicol and thiamphenicol, thereby preventing them from binding to ribosomal subunits of susceptible bacteria. Ribosomal binding is the primary mechanism of action of the chloramphenicol antibiotics and results in inhibition of peptidyl transferase, which is responsible for the transfer of amino acids to growing peptide chains and subsequent protein formation in bacteria. Nonetheless, compounds having the primary hydroxyl group do have utility in the treatment of bacterial infections, as evidenced by the continuing use of chloramphenicol and thiamphenicol throughout the world.

In recent years, a number of bacterial genera and species have begun to exhibit some resistance to florfenicol. For example, resistance has been observed in *Salmonella* species (Bolton, L. F., et al., *Clin. Microbiol.* **1999**, *37*, 1348), *E. coli* (Keyes, K., et al., *Antimicrob. Agents Chemother.*, **2000**, *44*, 421.), *Klebsiella pneumoniae* (Cloeckaert, A., et al., *Antimicrob. Agents Chemother.*, **2001**, *45*, 2381), and in the aquacultural pathogen, *Photobacterium damselae* subsp. *piscicida* (formerly *Pasteurella piscicida)* (Kim, E., et al., Microbiol. Immunol., **1996**, *40*, 665). This resistance has been traced to a highly conserved gene (*flo*) that produces an antibiotic efflux pump (Flo).

The emergence, and threatened spread, of resistance to florfenicol has fostered the need for new antibiotics that retain or exceed the activity of florfenicol, maintain its imperviousness to the CAT enzyme and, in addition, are not substrates for the Flo efflux pump. The compounds of the present invention are such antibiotics.

SUMMARY

Thus, an embodiment of this invention is a compound having the chemical formula:



wherein:

R^1 is selected from the group consisting of -OH and -F;

R^2 and R^3 are independently selected from the group consisting of hydrogen, (1C – 4C)alkyl, halo, $-CF_3$, $-NH_2$, -CN and N_3;

R^4 is selected from the group consisting of:

$-C(=R^5)R^6$,

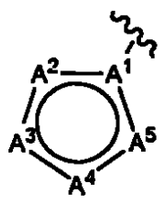
wherein:

R^5 is selected from the group consisting of oxygen, N-C≡N, and NOR^7,

wherein:

R^7 is selected from the group consisting of hydrogen, alkyl, aryl, heteroaryl, alicyclic and heteroalicyclic;

R^6 is selected from the group consisting of hydrogen, (1C – 4C)alkyl, (3C – 6C)cycloalkyl, (1C – 4C)alkoxy, aryl, heteroaryl, alicyclic and heteroalicyclic;



wherein:

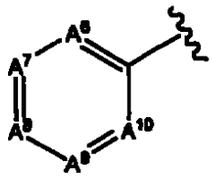
A^1 is carbon or nitrogen;

A^2, A^3, A^4 and A^5 are independently selected from the group consisting of carbon, nitrogen, oxygen and sulfur, provided that at least one of $A^1 – A^5$ is not carbon, that the total number of nitrogen, oxygen and sulfur atom in the ring does not exceed 4 and that the ring is aromatic;

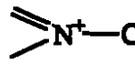
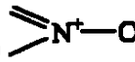
carbon atoms in the ring are independently substituted with an entity selected from the group consisting of hydrogen, (1C – 4C)alkyl, (3C – 6C)cycloalkyl,

(1C – 4C)alkylO-, -CF_3, -OH, -CN, halo, (1C – 4C)alkylS(O)-, (1C – 4C)alkylS$(O)_2$-, NH_2SO_2-, (1C – 4C)alkylNHSO_2-, ((1C – 4C)alkyl)$_2$NSO_2-, -NH_2, (1C – 4C)alkylNH-, ((1C – 4C)alkyl)$_2$N-, (1C – 4C)alkylSO_2NH-, (1C – 4C)alkylC(O)-, (3C – 6C)cycloalkylC(O)-, (1C – 4C)alkylOC(O)-, (1C – 4C)alkylC(O)NH-, -C(O)NH_2, (1C – 4C)alkylNHC(O)- and ((1C – 4C)alkyl)$_2$NC(O)-, wherein any of the alkyl groups in any of the substituents may optionally be substituted with a group selected from halo and -OH;

if A^1 is carbon and the ring does not contain oxygen or sulfur, one of the nitrogen atoms may optionally be substituted with an entity selected from the group consisting of (1C – 4C)alkyl, (1C – 4C)alkylS$(O)_2$- and –NH_2;



wherein:

A^6, A^7, A^8, A^9 and A^{10} are independently selected from the group consisting of carbon, nitrogen and  , provided that only one of A^6 – A^{10} at a time can be  ;

carbon atoms in the ring are independently substituted with an entity selected from the group consisting of hydrogen, (1C – 4C)alkyl, (3C – 6C)cycloalkyl, (1C – 4C)alkylO-, -CF_3, -OH, -CN, halo, (1C – 4C)alkylS(O)-, (1C – 4C)alkylS$(O)_2$-, NH_2SO_2-, (1C – 4C)alkylNHSO_2-, ((1C – 4C)alkyl)$_2$NSO_2-, -NH_2, (1C – 4C)alkylNH-, ((1C – 4C)alkyl)$_2$N-, (1C – 4C)alkylSO_2NH-, (1C – 4C)alkylC(O)-, (3C – 6C)cycloalkylC(O)-, (1C – 4C)alkylOC(O)-, (1C – 4C)alkylC(O)NH-, -C(O)NH_2, (1C – 4C)alkylNHC(O)-, ((1C – 4C)alkyl)$_2$NC(O)- and –OCH_2O-, the oxygen atoms in the –OCH_2O- substituent being bonded to adjacent ring carbon atoms, wherein any of the alkyl groups in any of the substituents may optionally be substituted with a group selected from halo and -OH;

R^8 is hydrogen in all compounds, except when R^2 and R^3 are both F, in which case R^8 is hydrogen or F; and,

the compound is either a racemate having the relative stereochemistry shown or is substantially enantiomerically pure and has the absolute stereochemistry shown.

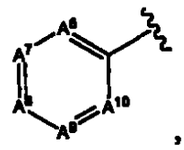
In an embodiment of this invention, R^1 is -F.

In an embodiment of this invention, R^2 and R^3 are independently selected from the group consisting of Cl and F.

In an embodiment of this invention, R^8 is hydrogen.

In an embodiment of this invention, R^4 is $-C(=R^5)R^6$ wherein R^5 and R^6 are as defined above.

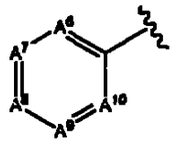
In an embodiment of this invention, R^4 is $CH_3C(O)$-.

In an embodiment of this invention, R^4 is  ,

wherein:

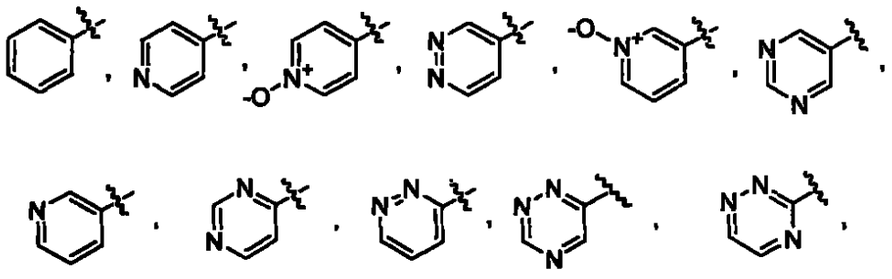
A^6, A^7, A^8, A^9 and A^{10} are as defined above, and,

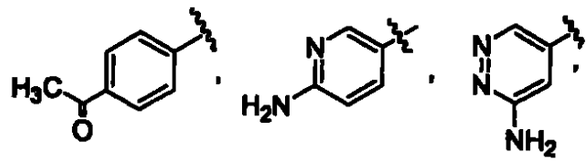
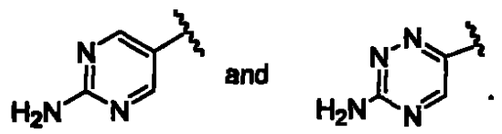
any of $A^6 - A^{10}$ that is carbon is substituted with an entity selected from the group consisting of hydrogen, $-NH_2$, halo-, -CN, (1C – 4C)alkyl-, (1C – 4C)alkylC(O)-, (1C – 4C)alkylS(O)-, (1C – 4C)alkylS$(O)_2$-, NH_2SO_2-, (1C – 4C)alkylSO_2NH-, (1C – 4C)alkyl$NHSO_2$-, ((1C – 4C)alkyl$)_2NSO_2$-, wherein any of the alkyl groups in any of the substituents may optionally be substituted with halo or –OH.

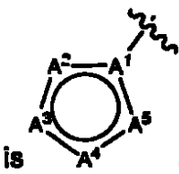
In an embodiment of this invention, R^4 is  and one, two or three of $A^6 - A^{10}$ is/are nitrogen; and,

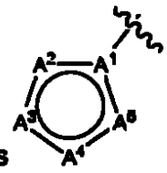
one or two of the remaining carbon atoms in the ring is/are optionally substituted with $-NH_2$, all other carbon atoms in the ring being unsubstituted.

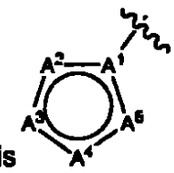
In a presently preferred embodiment of this invention, R^4 is selected from the group consisting of:



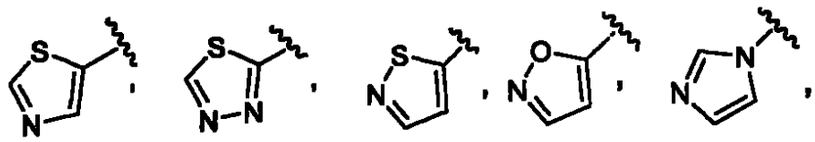



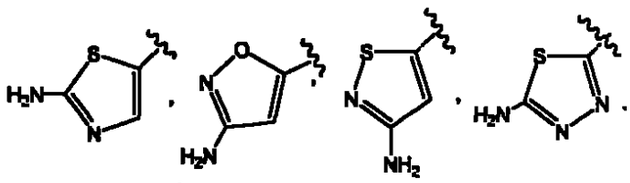
In an embodiment of this invention, R^4 is  as defined above.

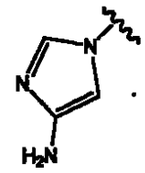
In an embodiment of this invention, R^4 is  and all carbon atoms and nitrogen atoms are unsubstituted.

In an embodiment of this invention, R^4 is  and one of $A^2 - A^5$ that is carbon is substituted with an $-NH_2$ group, all other carbon and, if applicable, nitrogen atoms in the ring being unsubstituted.

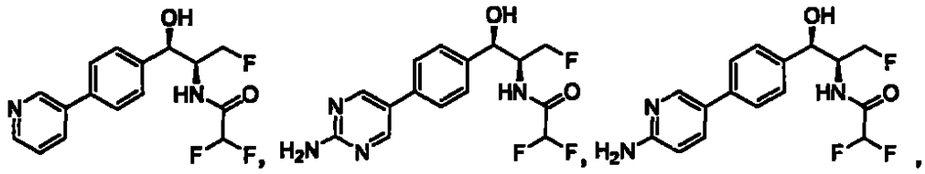
In a presently preferred embodiment of this invention, R^4 is selected from the group consisting of:

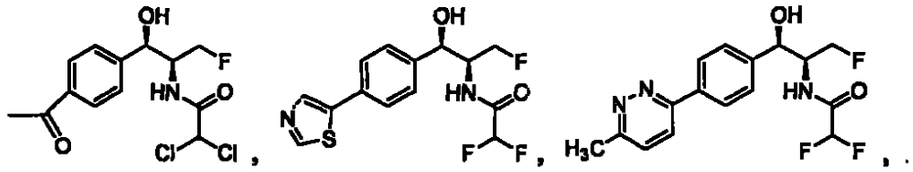


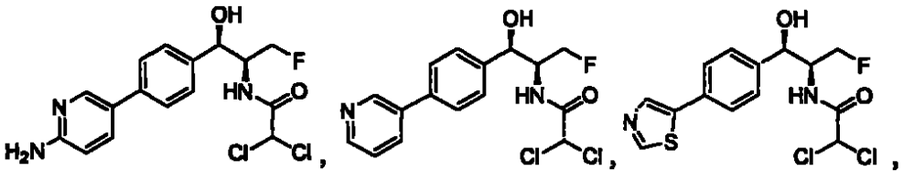


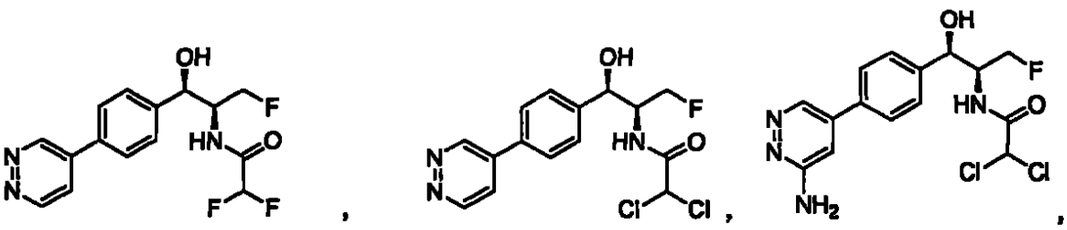
and 

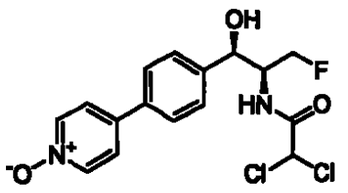
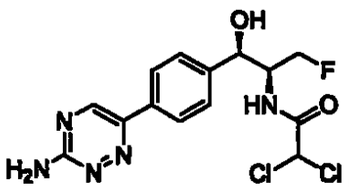
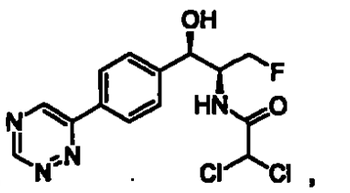
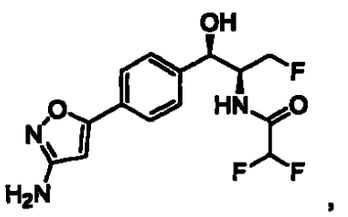
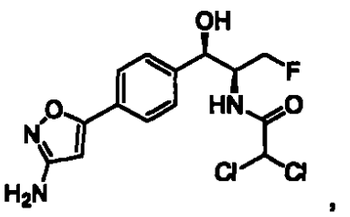
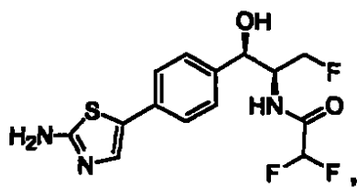
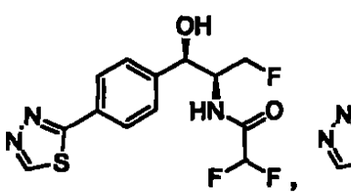
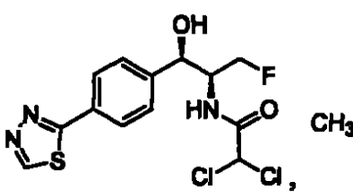
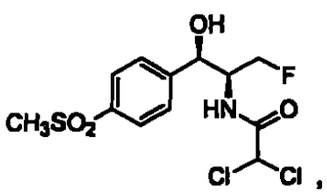
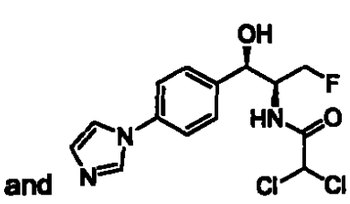
A presently preferred embodiment of this invention is a compound selected from the group consisting of:





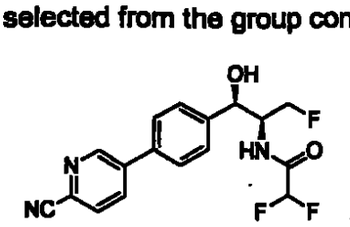
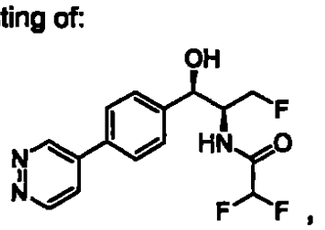
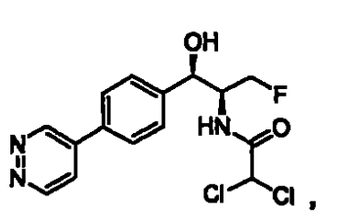


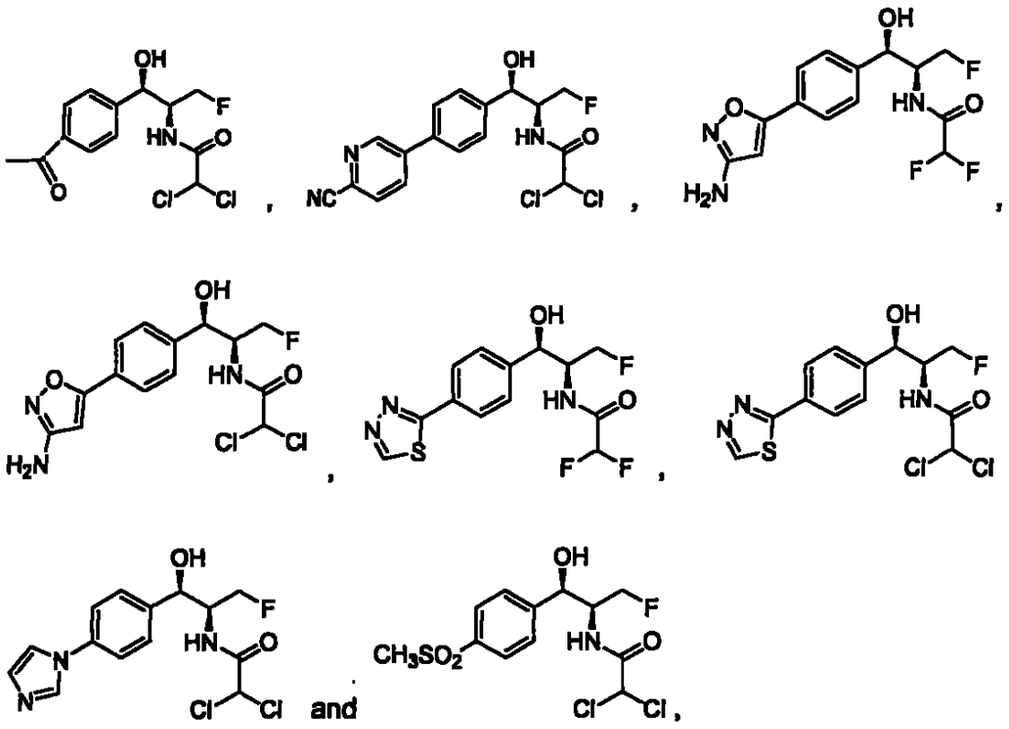












and

wherein the compound is either a racemate having the relative stereochemistry shown or is substantially enantiomerically pure and has the absolute stereochemistry shown.

A presently particularly preferred embodiment of this invention is a compound selected from the group consisting of:



wherein the compound is either a racemate having the relative stereochemistry shown or is substantially enantiomerically pure and has the absolute stereochemistry shown.

In another particularly preferred embodiment of this invention, the compound herein is substantially enantiomerically pure and has a 1-(R)-2-(S) absolute configuration.

An embodiment of this invention is a method of treating or preventing a bacterial infection, comprising administering to a patient in need thereof a pharmaceutically effective amount of a compound hereof.

In an embodiment of this invention, the bacterial infection is caused by a bacteria of the genus *Pasteurella*, *Haemophilus*, *Fusobacterium*, *Bacterioides*, *Aeromonas*, *Enterobacter*, *Escherichia*, *Klebsiella*, *Salmonella*, *Shigella*, *Actinobacillus*, *Streptococcus*, *Mycoplasma*, *Edwardsiella*, *Staphylococcus*, *Enterococcus*, *Bordetella*, *Proteus*, or *Mannheimia*.

In an embodiment of this invention the bacterial infection is caused by *Mannhemia haemolytica*, *Pasteurella multocida*, *Haemophilus somnus*, *Fusobacterium necrophorum*, *Bacterioides melaninogenicus*, *Actinobacillus*

pleuropneumoniae, Streptococcus suis, Salmonella choleresuis, Mycoplasma bovis, Mycoplasma hyopneumoniae, Mycoplasma hyorhinis, Mycoplasma gallisepticum, Edwardsiella ictaluri, Escherichia coli, Enterobacter cloacae, Staphylococcus aureus, Staphylococcus intermedius, Enterococcus faecalis, Enterococcus faecium, Klebsiella pneumoniae, Klebsiella oxytoca, Enterobacter cloacae, Proteus mirabilis, or *Aeromonas salmonicida.*

DETAILED DESCRIPTION OF THE INVENTION

Brief description of the Tables

Table 1 shows structures of representative compounds of this invention. The table and the compounds therein are not intended, nor should they be construed, to limit this invention in any manner whatsoever.

Table 2 is a list of the microorganisms against which the compounds of this invention were tested. The list is not intended, nor should it be construed, to limit the scope of this invention in any manner whatsoever.

Definitions

As used herein, "halo" refers to fluorine, chlorine, bromine or iodine.

As used herein, "alkyl" refers to a saturated (containing no multiple bonds) aliphatic (no delocalized π-electron system), hydrocarbon (containing, if unsubstituted, only carbon and hydrogen). The designation (n_1C – n_2C)alkyl, wherein n_1 and n_2 are integers from 1 – 6, refers to a straight chain or branched chain alkyl comprising from n_1 to n_2 carbon atoms. For example, (1C – 4C)alkyl refers to CH_3-, CH_3CH_2-, $CH_3CH_2CH_2$-, $CH_3CH(CH_3)$-, $CH_3CH_2CH_2CH_2$-, $CH_3CH_2CH(CH_3)$- or $(CH_3)_3C$-. The alkyl group may be unsubstituted or substituted with one or more moieties selected from the group consisting of halo, -OH, OCH_3, and -C≡N.

As used herein, "cycloalkyl" refers to an all-carbon cyclic or fused multicyclic ring, which, although it may contain one or more double bonds, maintains an essentially aliphatic character; that is, the double bonds do not interact to form a delocalized π-electron system around the ring. For the purposes of this invention, the ring may contain up to 7 carbon atoms. The designation (3C – 6C)cycloalkyl refers to 3, 4, 5, and 6-member all-carbon-atom rings. As used herein, "fused"

means that two cycloalkyl groups share at least one ring atom between them. Thus, such compounds as spiro[4.4]nonane are considered "fused" for the purposes of this invention. More commonly, fused rings share two adjacent ring carbon atoms. An example of such a fused system is decalin. A cycloalkyl ring may be unsubstituted or substituted with a moiety selected from the group consisting of –OH, -OCH_3, halo and -C≡N.

As used herein, "aryl" refers to an all-carbon 6-member ring or two fused six-member rings, the ring or fused rings having a delocalized π-electron system. By "fused" is meant that each ring of the system shares two adjacent ring carbon atoms with at least one other ring. An aryl ring may be unsubstituted or substituted with one or more moieties selected from the group consisting of –OH, -OCH_3, halo and -C≡N.

As used herein, "heteroaryl" refers to a five-member or six-member ring or to two rings, i.e., two 5-member, two six-member or a five- and a six-member ring fused together wherein the ring or fused ring has a delocalized π-electron system. If a ring is six-membered, it must consist of carbon and nitrogen only and may contain from one to four nitrogen atoms. If a ring is five-membered, it must contain one nitrogen, oxygen or sulfur atom and may contain one, two or three additional nitrogen atoms. A five-member ring with a circle in the center indicates that the ring is heteroaromatic. The circle is used to emphasize the fact that the location of the double bonds that participate in making the ring heteroaromatic is not static, rather it is dependent on the nature of the atoms forming the ring, i.e., whether they are carbon, nitrogen, oxygen or sulfur and what groups, if any, are bonded to them. The actual structure of any five-member heteroaromatic will be immediately apparent to those skilled in the art once the ring atoms are designated. With regard to heteroaromatic groups, the term fused has the same meaning as in the case of aryl groups. A heteroaryl group may be unsubstituted or substituted with any of the moieties described above with regard to aryl groups.

As used herein, "heteroalicyclic" refers to a cyclic or fused cyclic ring system containing atoms selected from the group consisting of carbon, nitrogen, oxygen and sulfur but no delocalized π-electron system. "Fused" has the same meaning set forth above with regard to cycloalkyl rings. Likewise, a heteroallcyclic ring may be

unsubstituted or substituted with the same moieties described above for cycloalkyl rings.

Whenever a ring carbon atom is stated to be "unsubstituted," it is understood that any unfilled valences are in fact occupied by hydrogen atoms. Likewise, if a ring nitrogen atom is capable of being further substituted and it is stated to be unsubstituted, it means that the nitrogen is bonded to a hydrogen atom.

As used herein, "relative stereochemistry" refers to the positioning in space of substituents relative to one another.

As used herein, "absolute stereochemistry" refers to the exact positioning of substitutents in three-dimensional space as determined by the Cahn-Ingold-Prelog rules, the application of which are well-known to those skilled in the art.

As used herein, an "enantiomer" refers to one of the two absolute stereochemical configurations of a molecule that rotates plane polarized light in one direction or the other (i.e., counterclockwise from its original axis, conventionally called "left," or clockwise, conventionally referred to as "right"). By "substantially enantiomerically pure" is meant that the compound consists of greater than 90% of the one enantiomer, preferably greater than 95%, and most preferably greater than 99%.

As used herein, a "racemate" refers to a 1:1 mixture of the two enantiomers of a compound. Racemic mixtures are designated by a (+/-) indicator. Substantially enantiomerically pure compounds are shown without the indicator.

As used herein, "patient" refers to birds, reptiles, fish, shellfish, and mammals. In particular it refers to birds such as, without limitation, chickens and turkeys, fish such as, without limitation, salmon, trout, catfish and yellowtail, mammals such as, without limitation, cats, dogs, rabbits, sheep, cattle, pigs, horses and goats and to human beings.

Discussion

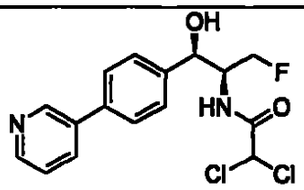
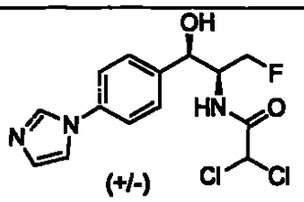
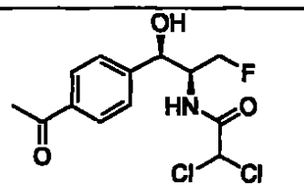
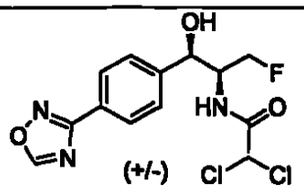
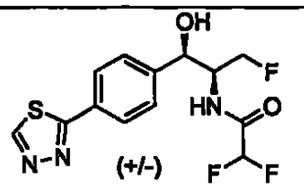
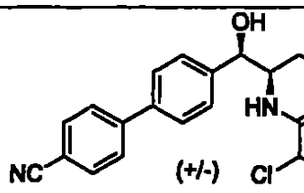
The compounds of this invention are expected to be useful for the treatment of bacterial infections in patients.

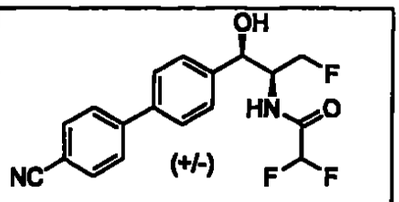
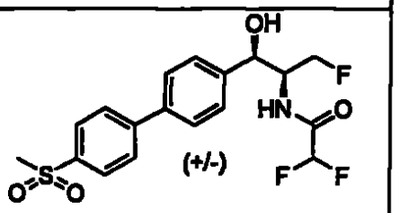
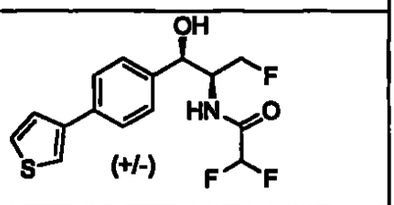
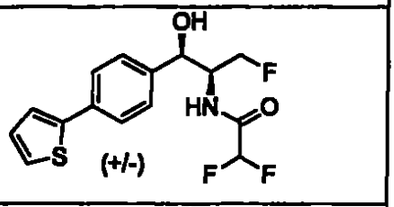
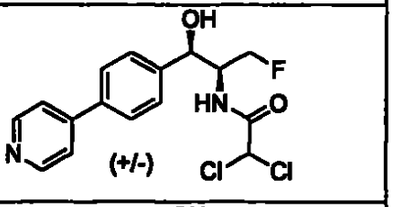
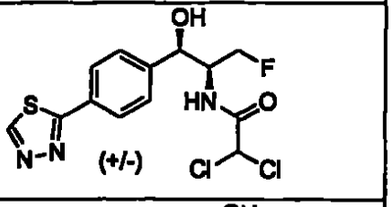
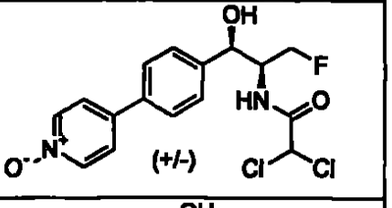
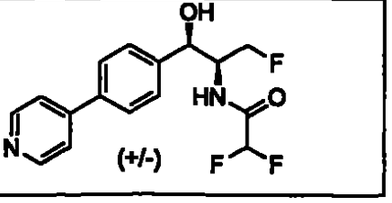
Compounds

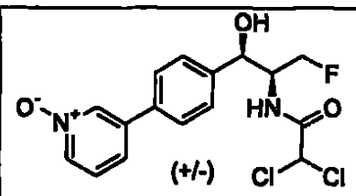
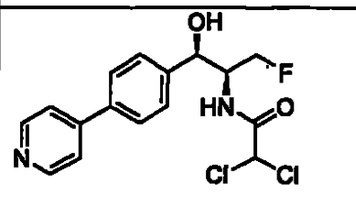
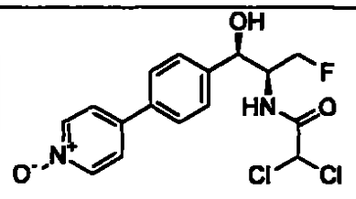
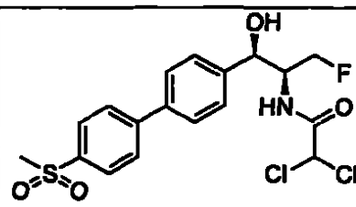
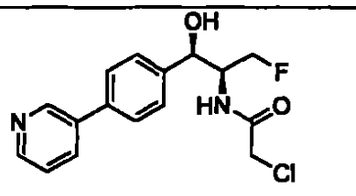
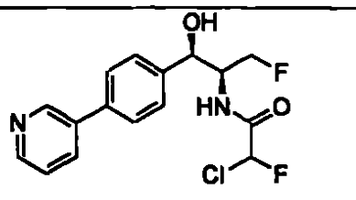
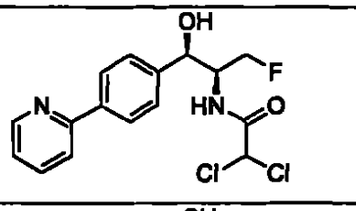
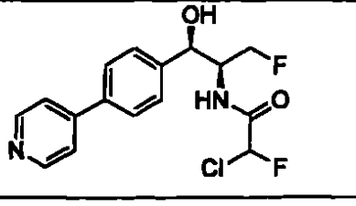
The compounds of the present invention are set forth generally in the Summary, above. Exemplary compounds of this invention are shown in Table 1.

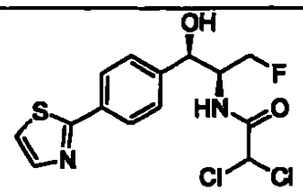
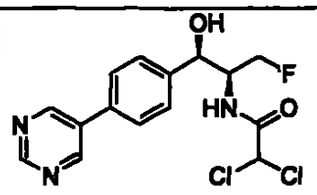
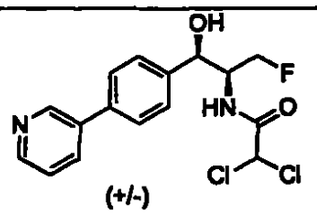
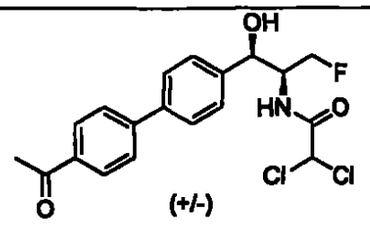
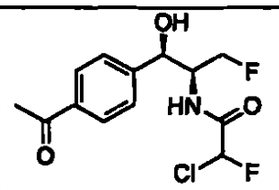
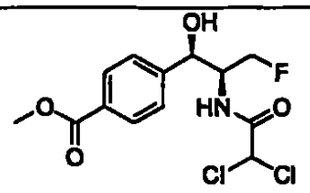
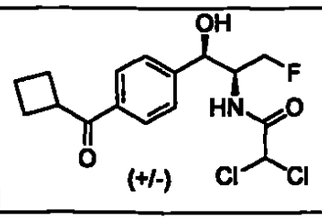
Neither the table nor the compounds shown therein are intended, or are to be construed, as limiting the scope of this invention in any manner whatsoever.

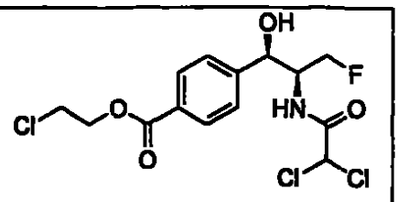
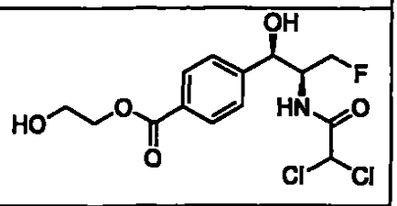
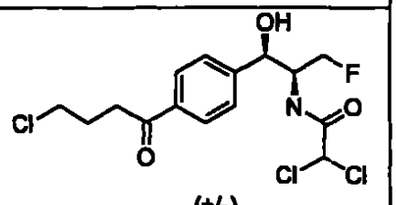
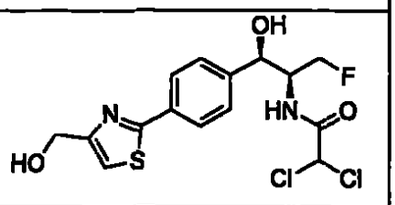
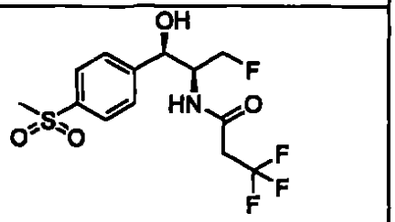
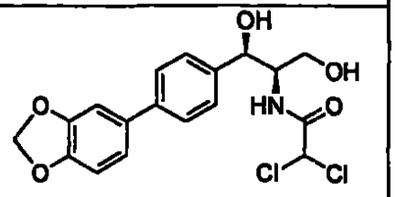
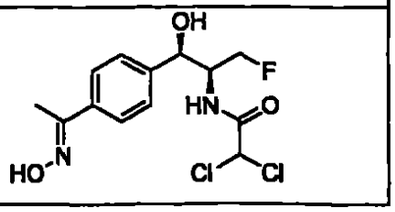
TABLE 1

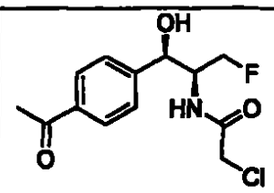
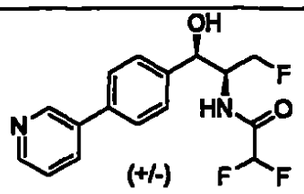
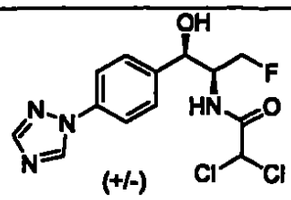
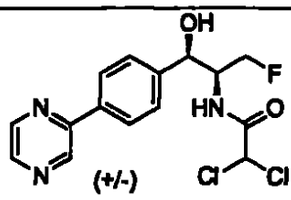
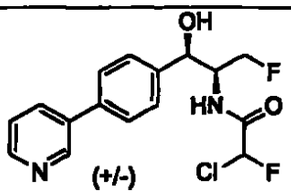
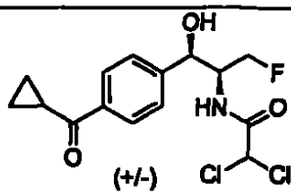
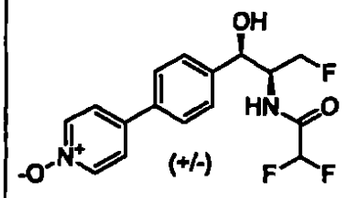
Compound #	Structure
28	
29	
41	
47	
48	
49	

50	
51	
52	
53	
54	
55	
56	
57	

58	
59	
60	
61	
62	
63	
64	
65	

66	
67	
68	 (+/-)
69	 (+/-)
71	
72	
73	 (+/-)

74	
75	
76	 (+/-)
77	
78	
79	
80	

81	
82	
84	
85	
86	
91	
92	
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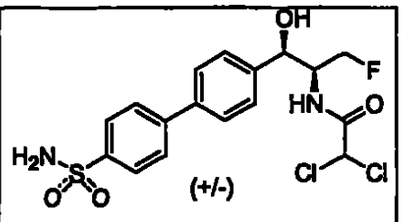
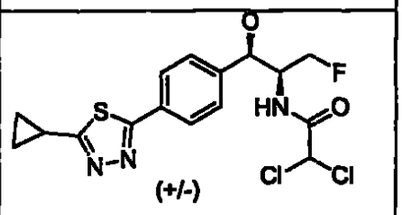
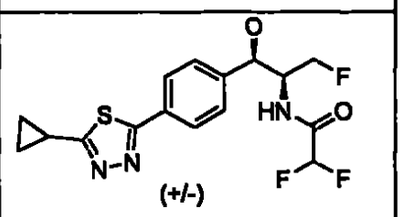
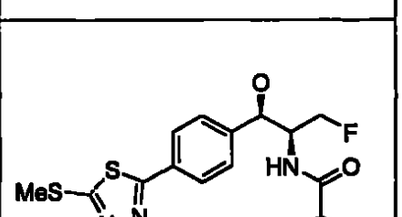
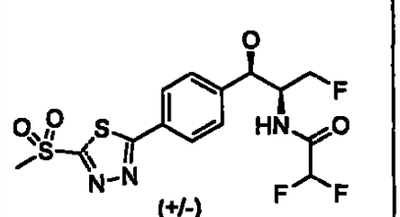
	
94	
95	
97	
98	
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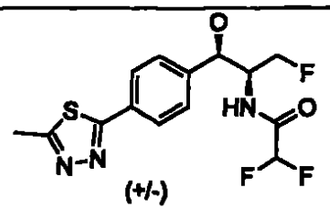
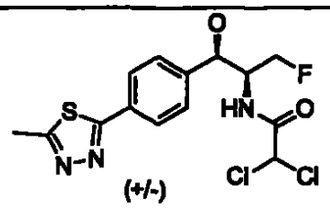
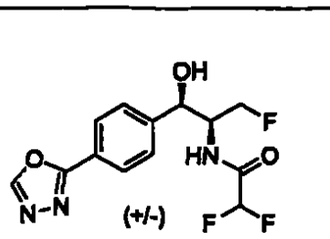
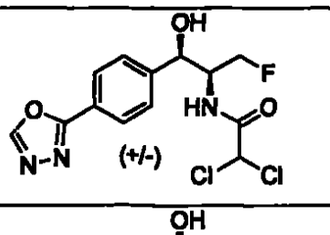
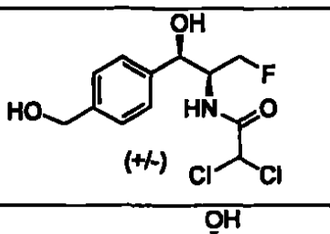
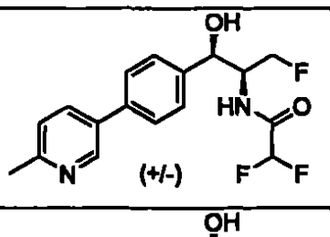
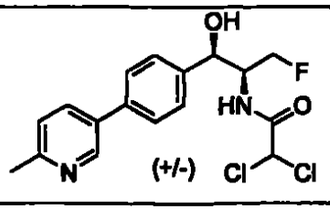
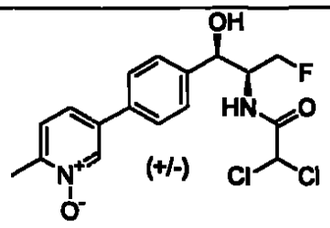
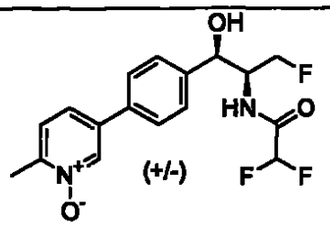
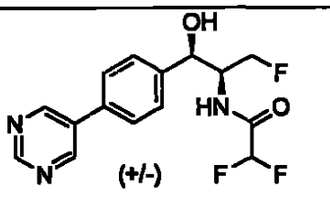
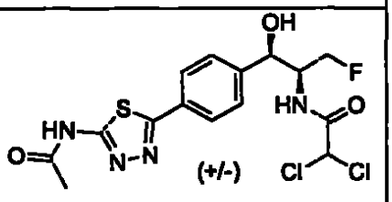
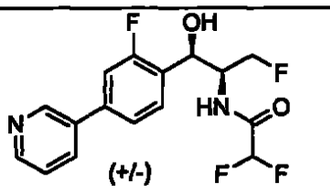
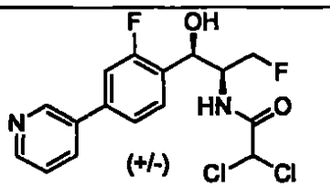
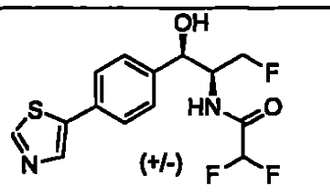
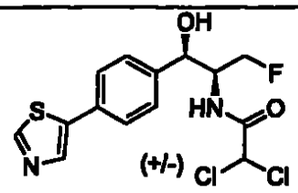
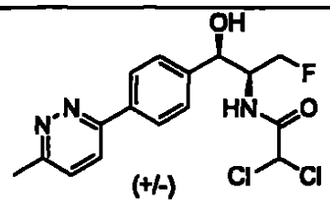
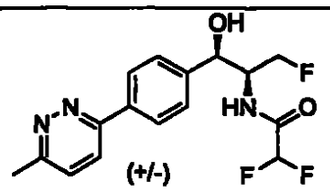
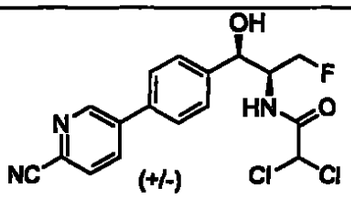
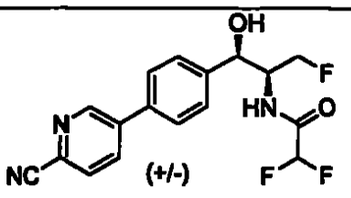
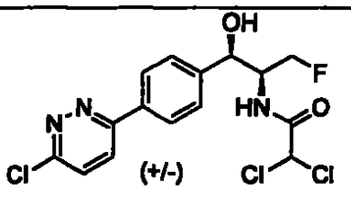
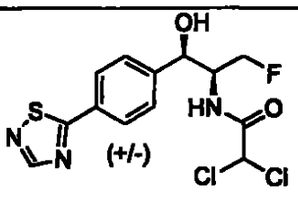
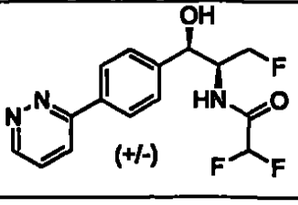
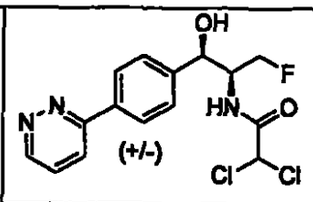
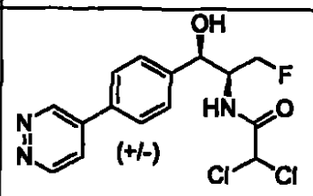
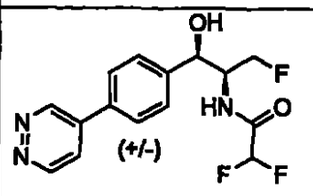
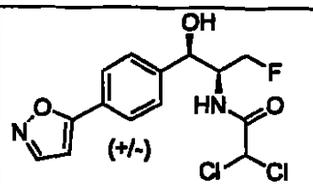
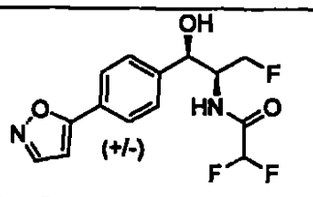
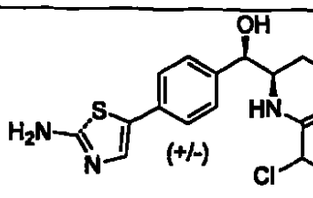
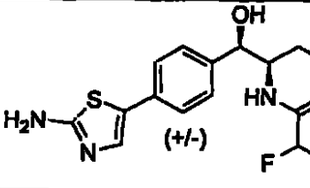
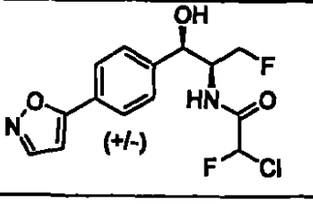
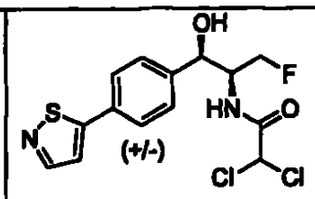
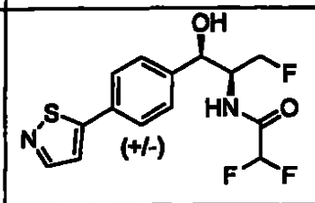
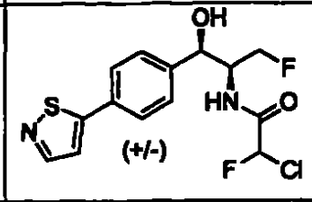
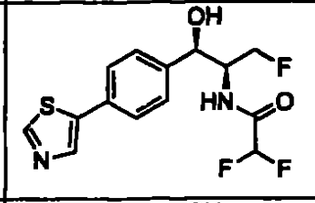
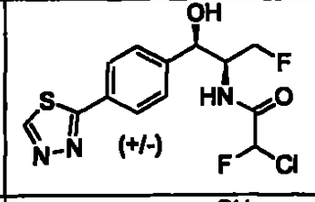
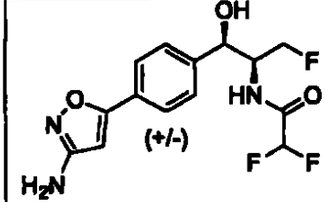
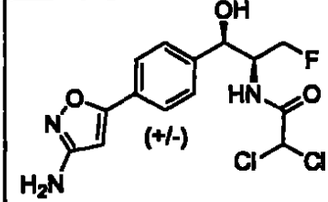
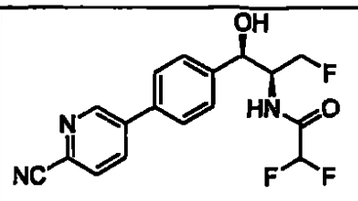
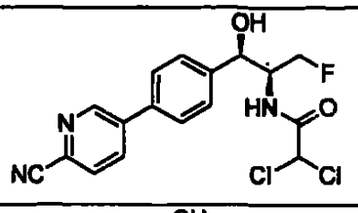
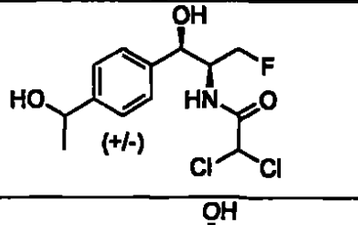
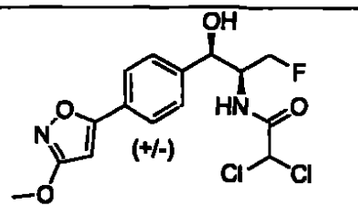
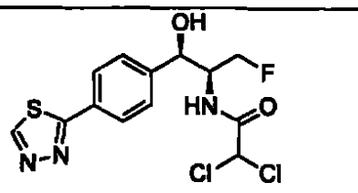
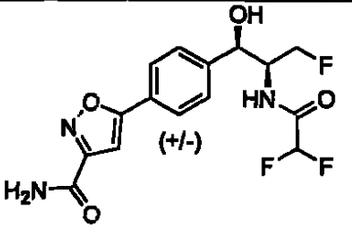
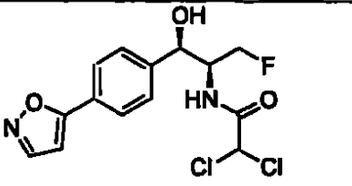
	
101	
106	
107	
109	
110	
111	

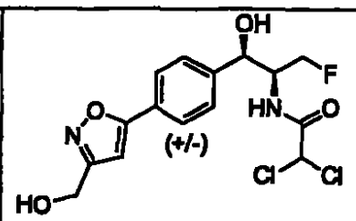
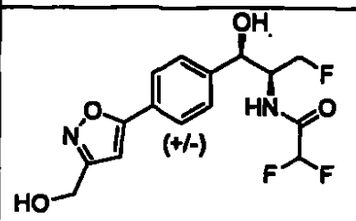
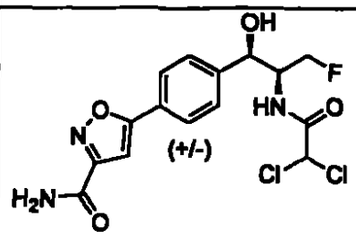
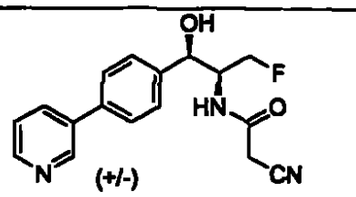
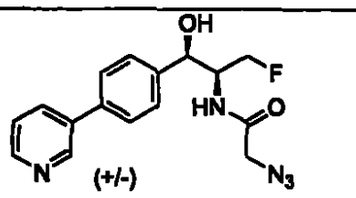
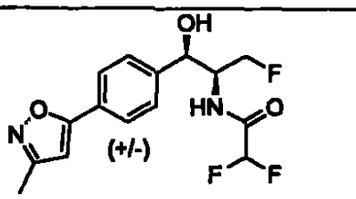
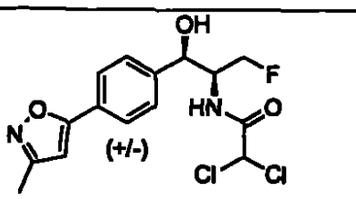
112	
113	
114	
115	
116	
117	
118	

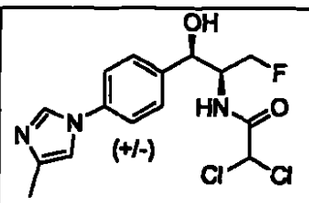
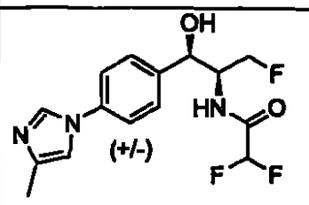
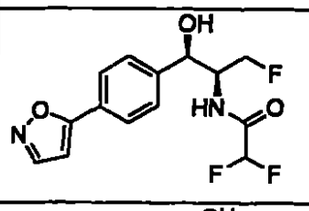
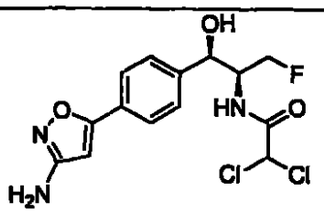
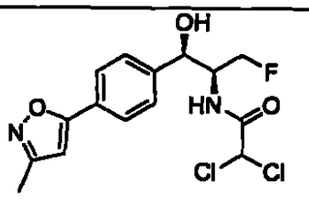
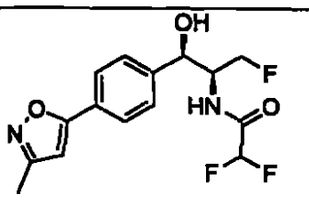
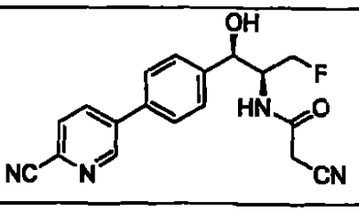
120	
121	
122	
123	
124	
125	
126	
127	

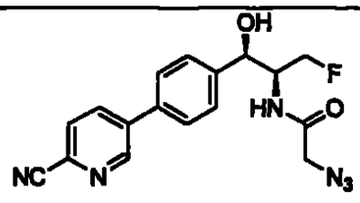
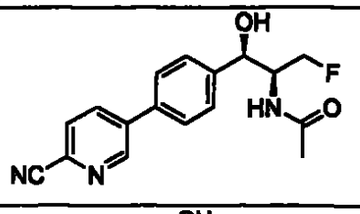
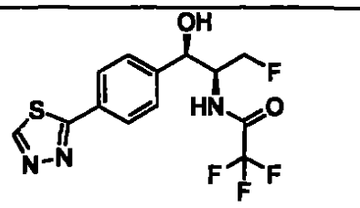
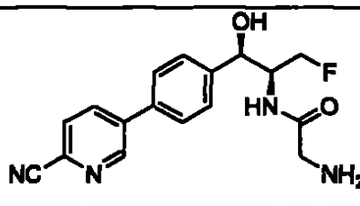
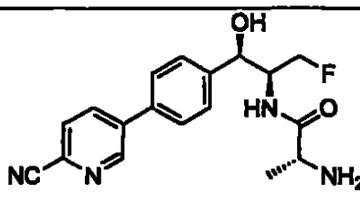
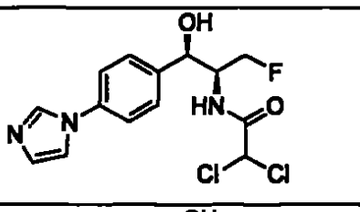
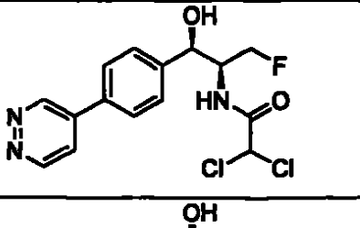
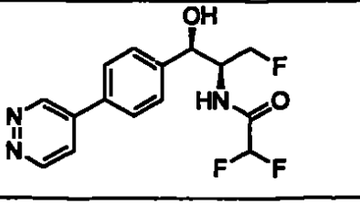
128	
129	
130	
131	
132	
133	
134	
135	

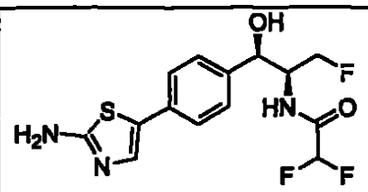
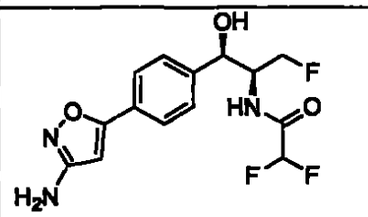
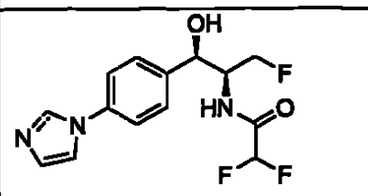
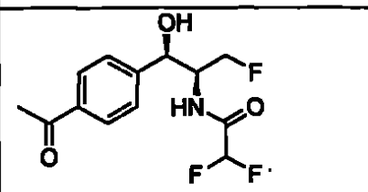
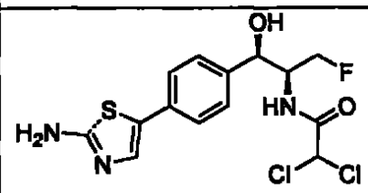
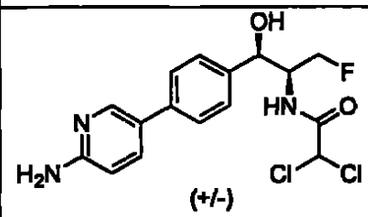
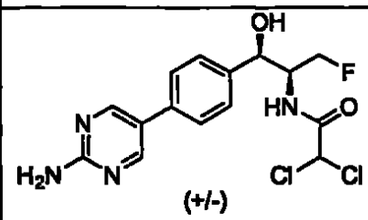
136	
137	
138	
139	
140	
141	
142	

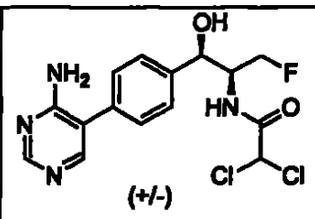
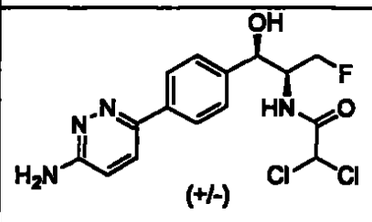
143	
144	
145	
146	
147	
148	
149	

150	
151	
152	
153	
154	
155	
156	

157	
158	
159	
160	
161	
162	
163	

164	
165	
166	
167	
168	
169	
170	
171	

172	
173	
174	
175	
176	
177	 (+/-)
178	 (+/-)

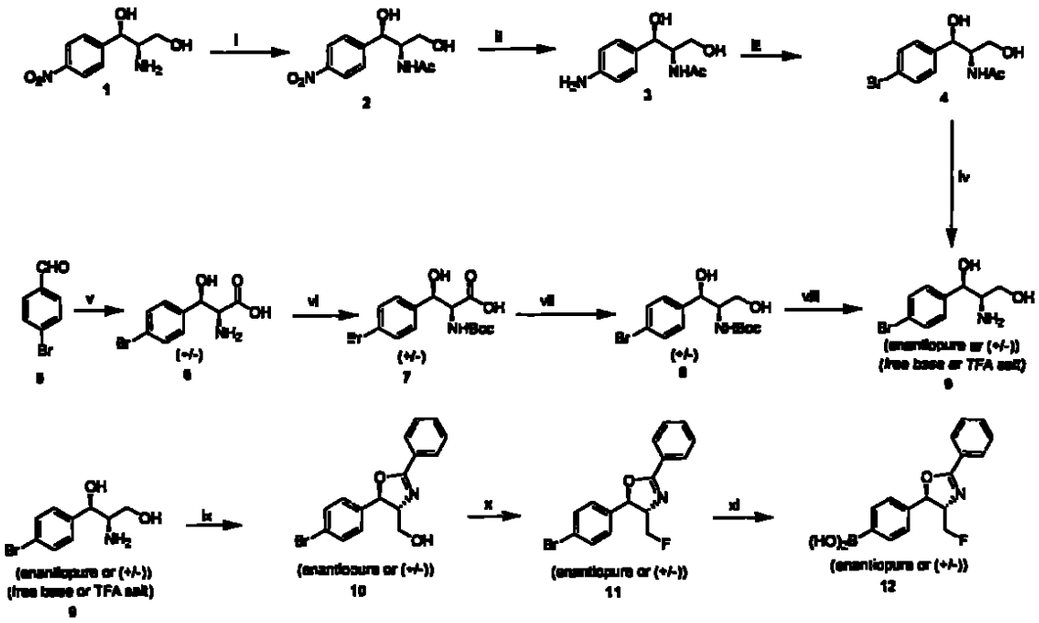
179	 (+/-)
180	 (+/-)

Syntheses

Intermediates were prepared in enantiomerically pure form, starting with commercially available chloramphenicol base (**1**), or as racemic mixtures by condensation of *p*-bromobenzaldehyde with glycine under basic conditions. For the enantiomerically pure synthesis of intermediates such as **9**, chloramphenicol base was converted to compound **3** (Rebstock, M. C., et al., *J. Am, Chem. Soc.*, **1949**, *71*, 2458; Evans, D. D., et al., *J. Chem. Soc.*, **1954**, 1687; Morris, D. S. and Smith, S. D., *J. Chem. Soc.*, **1954**, 1680). Compound **3** was subjected to the Sandmeyer reaction after which the acetate protecting group was removed under acidic conditions to provide enantiomerically pure **9**.

Alternatively, *p*-bromobenzaldehyde (**5**) can be converted to (*d/l*)-*threo*-*p*-bromophenylglycine (**6**, Scheme 1), (Bolhoffer, W. A. *J. Am. Chem. Soc.* **1954**, *76*, 1322; Herbert, R. B.; Wilkinson, B.; Ellames, G. J. Can. J. Chem. **1994**, *72*, 114), which can be protected as N-Boc derivative **7** and then reduced in two-steps: activation with DCC and *N*-hydroxysuccinimide followed by treatment with $NaBH_4$ to provide **8**. Compound **9** can be isolated as its TFA salt or as the free base. Regioselective introduction of fluoride is accomplished by protecting the amine and benzylic hydroxyl group as phenyl oxazoline **10** followed by fluorination with (diethylamino)sulfur trifluoride (DAST) or [bis(2-methoxyethyl)amino]sulfur trifluoride (Lal, G. S., *J. Org. Chem.*, **1999**, *64*, 7048) to give **11**. Compound **11** is used in the Suzuki cross-coupling reaction for the synthesis of biaryl derivatives. The other Suzuki partner, an aryl boronic acid such as **12**, can be prepared as shown and reacted with aryl halides.

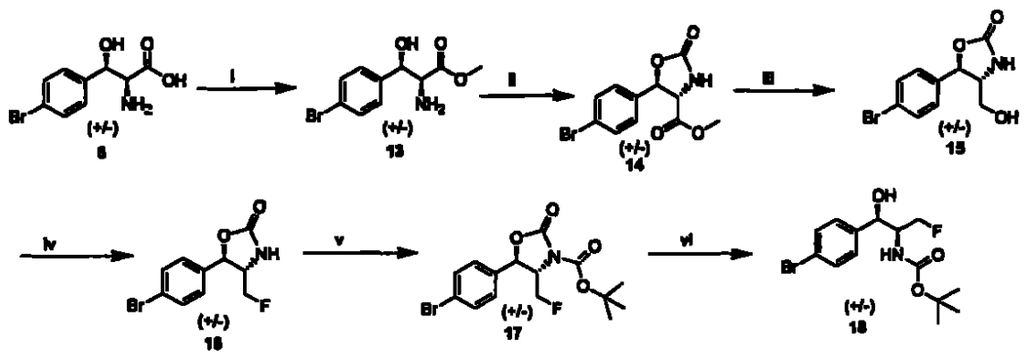
Scheme 1



i) Ac_2O neat; ii) H_2(g), 10% Pd/C; iii) $NaNO_2$/aq HBr, 0 °C, then CuBr/aq. HBr; iv) 10% H_2SO_4, Δ, then NaOH workup; v) KOH, glycine, then aq. HCl, then NH_4OH; vi) Boc_2O, dioxane, water; vii) DCC, HOSu then $NaBH_4$ THF then H_2O; viii) TFA then NaOH workup; ix) ethylbenzimidate hydrochloride, Et_3N; x) DASTor [bis(2-methoxyethyl)amino]sulfur trifluoride, -78 °C to room temperature; xi) first *n*-BuLi at -78 °C, then $B(OMe)_3$ -78 °C to room temperature.

The conditions for removal of the phenyloxazoline protecting group proved to be incompatible with many functional groups at the *p*-position of the aromatic ring. Thus, a new protecting group motif was developed based on literature methods (Scheme 2; Jommi, G., et al., *Gazz. Chim. Ital.*, **1986**, 116:485). Phenylserine **6** was converted to methyl ester **13**, which was protected as oxazolidinone **14**, which, in turn, was reduced with $NaBH_4$ to **15**. Fluorination with either DAST or [bis(2-methoxyethyl)amino]sulfur trifluoride gave intermediate **16**, which did not give consistently good Suzuki reaction yields. Thus, **16** was converted to **17**. Cleavage of the oxazolidinone protecting group was facilitated by the introduction of a Boc group (Grehn, L., et al., *Acta Chem. Scand. B*, **1986**, *40*, 745) followed by base-catalyzed cleavage to give **18** (Ishizuka, T. and Kunieda, T., *Tetrahedron Lett.*, **1987**, *28*, 4185; Jommi, G., et al., *Gazz. Chim. Ital.*, **1988**, 118:75).

Scheme 2

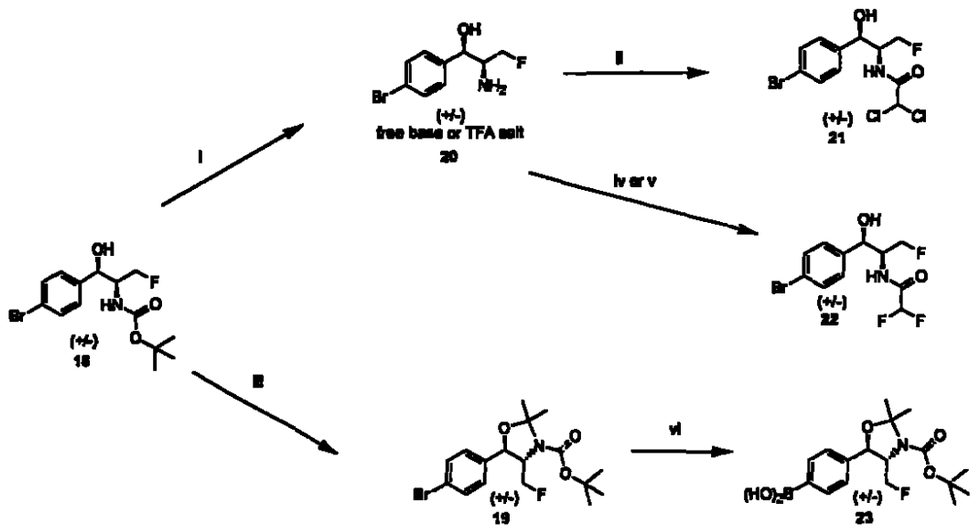


I) HCl, MeOH 0 °C to room temperature 16 h, then reflux 16 h; II) CDI, Et_3N, THF/1,2-dichloroethane; III) $NaBH_4$, MeOH 0 °C; IV) DAST or [bis(2-methoxyethyl)aminosulfur trifluoride -78 °C 10 min then room temperature until complete; v) Boc_2O, DMAP, MeCN; vi) Cs_2CO_3, MeOH.

Compound **18** was protected as isopropylidene derivative **19**, which was converted to boronic acid **23** (Scheme 3). Compounds **18**, **19**, **22** and **23** were used in Suzuki cross-coupling reactions. Compound **23** provided the most versatility since it could be cross-coupled with any aryl bromide or iodide and the protecting groups could be easily removed under mild conditions. Compound **22** was attractive as a Suzuki cross-coupling partner in that the coupling reactions yielded the desired florfenicol analogs directly without the need for deprotection and dihaloacetylation. However, cross-coupling yields with **22** were lower than those with **23**. Cross-couplings using **21** gave the desired products contaminated with acetate and monochloroacetate analogs.

Racemic compound **20** was identical spectroscopically to a sample synthesized from semi-synthetic **11** by hydrolytic cleavage and basic work-up. This provided unequivocal proof that the condensation of phenylserine **6** had proceeded with the appropriate relative *threo* or *syn* stereoselectivity.

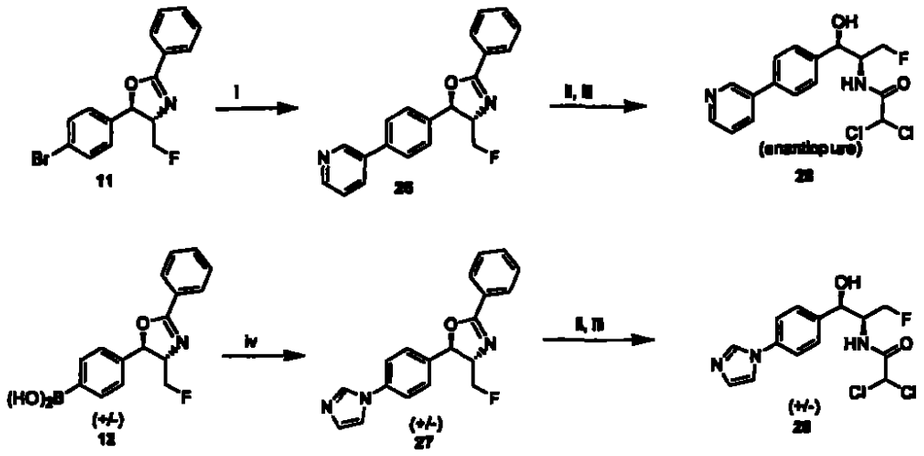
Scheme 3



I) 8:1 TFA, H_2O, then NaOH work-up if free-base is desired; II) methyl dichloroacetate, Et_3N, MeoH, Δ; III) 2-methoxypropene, *p*-TsOH·H_2O; IV) methyl difluoroacetate, Et_3N, MeoH, Δ; V) difluoroacetyl chloride, Et_3N, followed by 1:8:1 Et_3N, MeOH, H_2O; VI) first *n*-BuLi at -78 °C, then $B(OMe)_3$, -78 °C to room temperature.

Compounds **11** and **12** were used to prepare compounds **28** and **29** (Scheme 4). Compound **11** was cross-coupled with 3-pyridine boronic acid under standard biphasic Suzuki conditions to yield protected intermediate **26**. The *N-C* cross-coupling product **27** was prepared according to the recently reported methods of Lam and coworkers (Lam, P. Y. S., et al., *Synlett*, **2000**, 674; Lam, P. Y. S., *Tetrahedron, Lett.*, **1998**, *39*, 2941; Lam, P. Y. S., *Tetrahedron Lett.*, **2001**, *42*, 3415). Both **26** and **27** were deprotected and dichloroacetylated to give **28** and **29**.

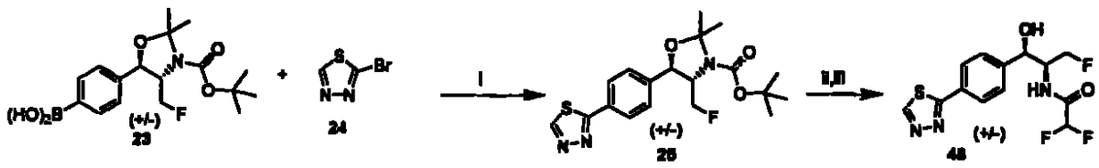
SCHEME 4



i) *m*-pyridine boronic acid, Na_2CO_3, $Pd(PPh_3)_4$, THF, H_2O, Δ; ii) 6 N aqueous HCl, sea led tube 100 °C then basic work-up; iii) methyl dichloroacetate, Et_3N, MeOH, Δ; iv) $Cu(OAc)_2$, pyr., CH_2Cl_2, room temperature, open to air, 40 h.

Compound **23** and 2-bromo-1,3,4-thiadiazole (**24**) were reacted to give **48** (Scheme 5). Suzuki cross-coupling produced the heteroblaryl **25**. Compound **25** was deprotected with 9:1 TFA/H_2O, which removed the Boc and isopropylidene groups simultaneously, and then was difluoroacetyated to give **48**.

Scheme 5

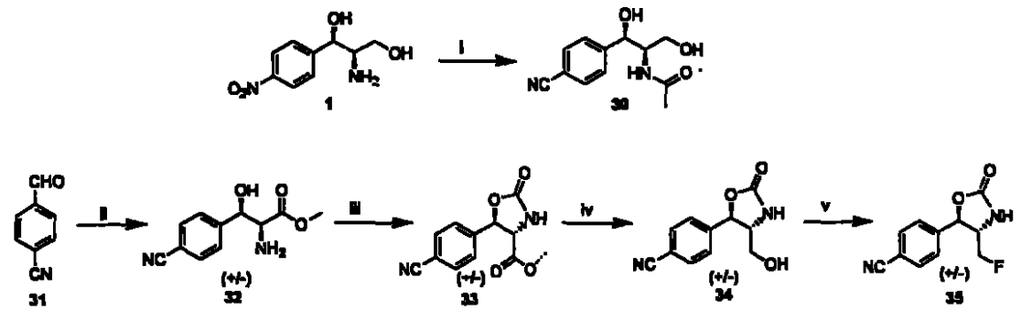


i) Cs_2CO_3, $PdCl_2(dppf)$, THF, DMF, H_2O; ii) 9:1 TFA/H_2O; iii) Et_3N, methyl difluoroacetate, Δ

While *p*-bromo functional groups of derivatives such as **11** and the corresponding boronic acids such as **12** have substantial utility as synthetic intermediates to florfenicol analogs, *p*-cyano compounds have also served as important intermediates. Using literature methods (Morris, D. S.; Smith, S. D., above), intermediates such as **30** have been prepared in enantiomerically pure form (Scheme 6). Larger amounts of intermediate were prepared using racemic, totally synthetic intermediates such as **35**. The aldol condensation used to produce **32**

from p-cyanobenzaldehyde and glycine methyl ester was adapted from the literature (Pines, S. H. and Kazlowski, M. A., *J. Org. Chem.*, **1972**, *37*, 292).

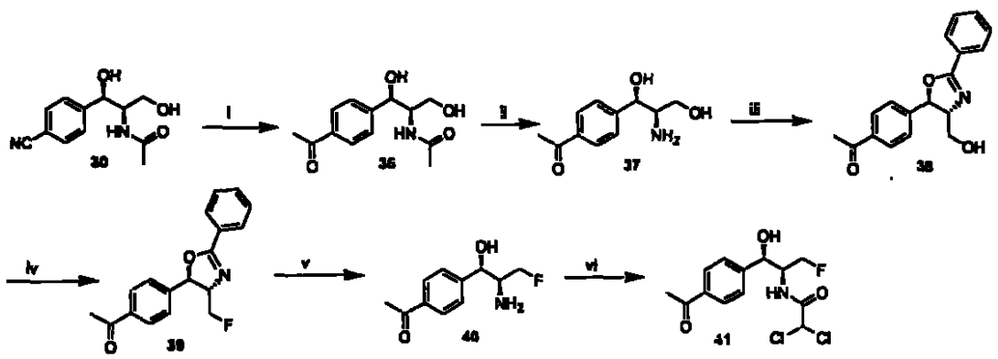
Scheme 6



i) literature procedure–see text; ii) literature procedure–see text; iii) CDI, TEA, THF or 1,2-dichloroethane; iv) $NaBH_4$, MeOH, 0 °C; v) DAST or [bis(2-methoxyethyl)amino]sulfur trifluoride -78 °C 10 min then room temperature until complete.

Compound **41** (Scheme 7) was prepared from intermediate **36**, the preparation of which from **30** has been described (von Strandtmann, M., et al., *J. Med. Chem.*, **1967**, *10*, 888). Compound **36** was deprotected with aqueous H_2SO_4 and then regioselectively protected as phenyloxazoline **38**. Compound **39** was obtained on treatment with DAST and then was deprotected and dichloroacetylated to give **41**.

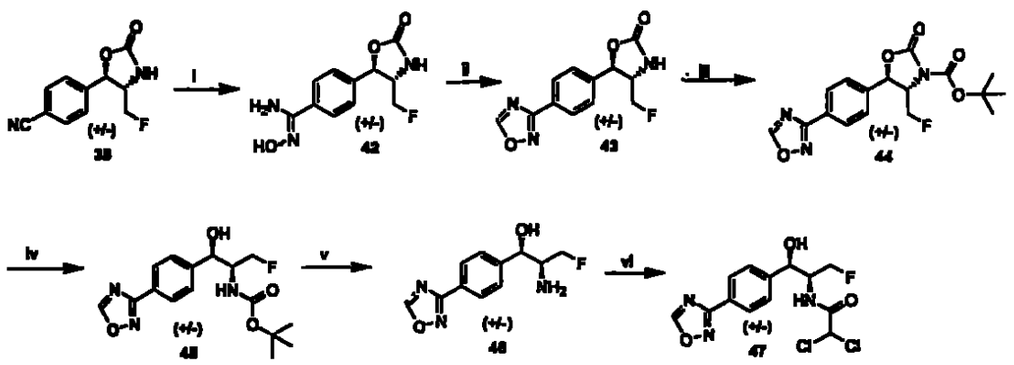
Scheme 7



i) MeLi, THF, then H_2O; ii) 16% aqueous H_2SO_4, Δ; iii) Et_3N, ethyl benzimidate hydrochloride, Δ; iv) DAST -78 °C to room temperature; v) 6N HCl, Δ; vi) methyldichloroacetate, Et_3N, MeOH, Δ.

Compound **47** was prepared from intermediate **35** by treatment with hydroxylamine hydrochloride followed by triethyl orthoformate to give **43**, which was deprotected to give **46**, which was then dichloroacetylated.

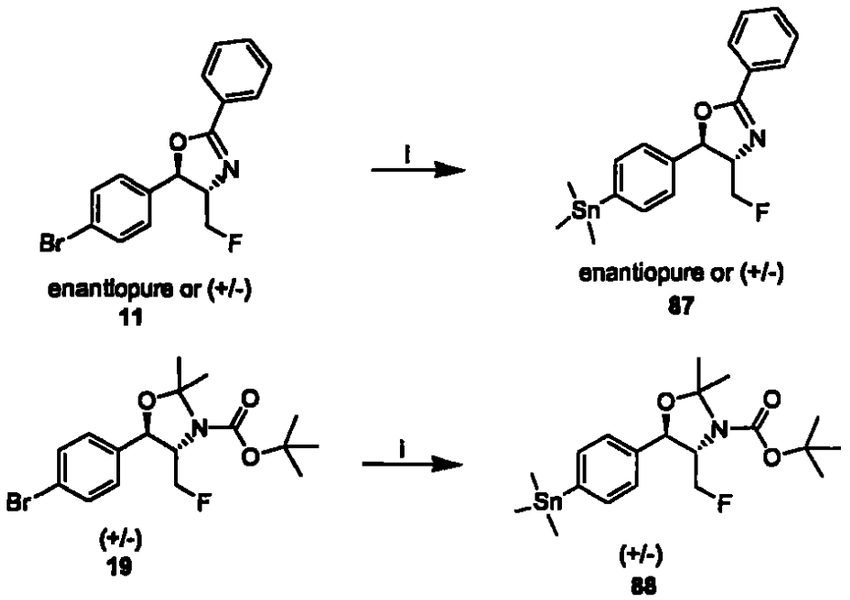
Scheme 8



i) NH_2OH-HCl, EtOH, Δ; ii) triethyl orthoformate, Δ; iii) Boc_2O, DMAP, MeCN; iv) Cs_2CO_3, MeOH; v) TFA/H_2O 8:1; vi) methyldichloroacetate, Et_3N, Δ.

p-Acyl derivatives were obtained by Stille coupling reactions of the protected intermediates (Scheme 9, compounds **87** and **88**) with acid chlorides. Trimethylstannyl groups were introduced by Pd-mediated reactions using hexamethylditin.

Scheme 9

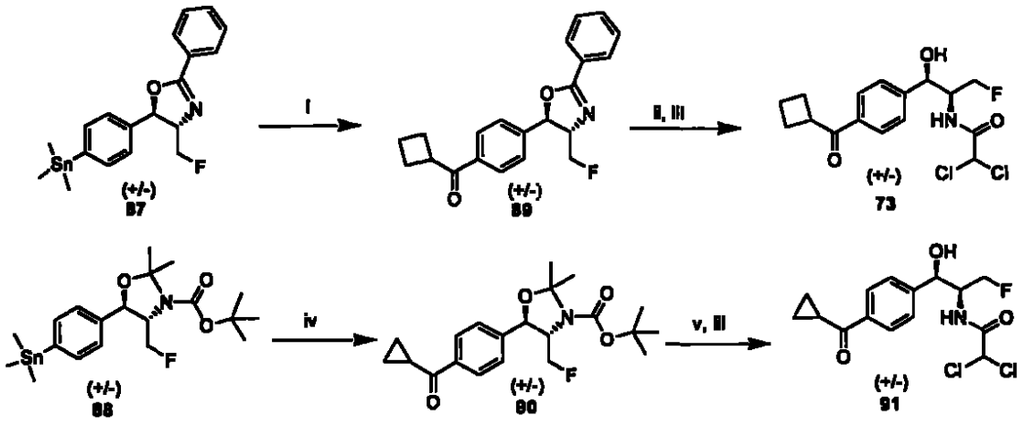


i) $Me_3SnSnMe_3$, $Pd(PPh_3)_4$, Δ, C_6H_6 or PhMe

The cyclobutyl derivative **73** was prepared from **87** via a Stille coupling reaction that produced intermediate **89** (Scheme 10) which was deprotected and dichloro- acetylated.

Attempted deprotection of the cyclopropyl derivative corresponding to **89** led to HCl-mediated ring opening. Thus, to prepare **91**, the boc/isopropylidene approach was employed since deprotection occurs under conditions that do not affect the cyclopropyl group.

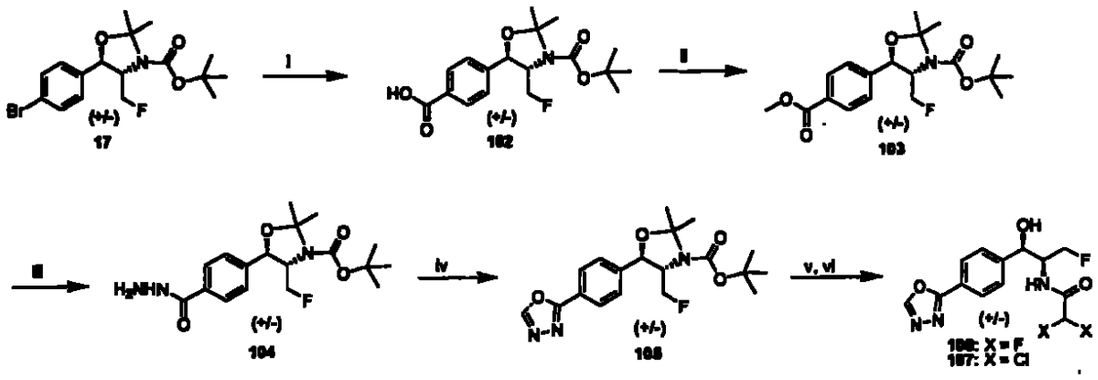
Scheme 10



i) Pd_2dba_3, K_2CO_3, Et_3N, cyclobutanecarbonyl chloride, room temperature, 3h; ii) 6N HCl sealed tube, Δ; iii) methyldichloroacetate, Et_3N, MeOH, Δ; iv) Pd_2dba_3, K_2CO_3, Et_3N, cyclopropanecarbonyl chloride, room temperature, 3h; v) TFA/H_2O

When required, *p*-carboxyphenyl derivatives of protected phenicol intermediates could be prepared in two ways. For example, hydrolysis of the *p*-nitrile analog of **17** gives the corresponding carboxylic acid. However, nitriles generally could not be obtained as readily as the bromo derivatives. Thus, the preferred approach to carboxylation was replacement of the bromo group. For example, carboxylic acid derivative **102** (Scheme 11) was prepared by lithiation of **17** followed by treatment with CO_2 and acid work-up. Compound **102** was converted to methyl ester **103**, which was reacted with hydrazine to give **104**. Cyclization of **104** with triethyl orthoformate gave oxadiazole **105**, which was deprotected and dihaloacetylated to give **106** and **107**.

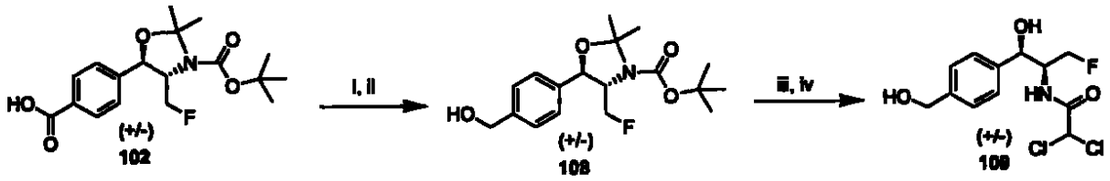
Scheme 11



i) *n*-BuLi, -78 °C, THF, then bubble dry CO_2 then warm to room temperature, then H_3O^+; CH_2N_2; ii) hydrazine, EtOH, Δ; iv) triethylorthoformate, Δ; v) TFA/H_2O; vi) for 106 Et_3N, ethyldifluoroacetate, room temperature; for 107 same as 106, but replace ethyl difluoroacetate with methyl dichloroacetate.

The carboxylic acid derivative of **102** was also used to prepare compounds such as **109** (Scheme 12), by reduction to alcohol **108** followed by deprotection and dichloroacetylation

Scheme 12



i) DCC, pentafluorophenol, EtOAc; ii) $NaBH_4$, MeOH; iii) TFA/H_2O; iv) methyl dichloroacetate, Et_3N, Δ.

Biological evaluation

All of the compounds of this invention are expected to demonstrate antimicrobial activity against the same bacteria as the other members of the chloramphenicol family. In addition, they may be expected to be active against species of bacteria that are resistant to current chloramphenicol antibiotics, in particular florfenicol. It is also expected that the present compounds may exhibit activity against genera and species of bacteria against which current chloramphenicol-type antibiotics are not active.

It is also understood that, with regard to bioactivity, one enantiomer of a compound may be more active than the other. In such case, whether expressly

stated or not, the more active isomer is considered the preferred embodiment of this invention. Particularly preferred is the most active enantiomer of the 1-(R)-2(S) absolute configuration of any compound herein.

To determine the range and level of activity of the compounds of this invention, the following protocols may be used. Other such protocols will become apparent to those skilled in the art based on the disclosures herein and are within the scope of this invention. Some compounds herein are expected to not only exhibit substantial antibacterial activity but to also be less susceptible to current chloramphenicol resistance mechanisms. The screening protocols herein may be used to determine such characteristics also.

Susceptibility testing

Compounds were evaluated against a panel of bacterial strains using a broth microdilution assay performed as recommended by the NCCLS (National Committee for Clinical Laboratory Standards (NCCLS) 2000, Methods for Dilution of Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically - Fifth Edition, Approved Standard, NCCLS Document M7-A5, Vol 20, No. 2). The minimum inhibitory concentration (MIC) is defined as the lowest concentration of a compound that prevents the growth of the bacteria.

The following 10 organisms constituted the primary panel of evaluation:

TABLE 2

Bacteria	Strain	Efflux Pump	Phenotype
Escherichia coli	ECM1194	AcrAB[a]	Wild type
Escherichia coli	ECM1694	None	Δ*acrAB*::Tn903kmr
Escherichia coli	ECM 1642	AcrAB	*MarR*
Escherichia coli	ECM 1888	MdfA	*tolC*::Tn mdfR
Escherichia coli	ECM 1750	CmlA	Δ*acrAB*::Tn903kmr/pLQ821
Escherichia coli	ECM 1958	Flo	Δ*acrAB*::Tn903kmr/p1956
Escherichia coli	ECM 1970	AcrAB+Flo	*marR*/p1956
Escherichia coli	ECM 1197	AcrAB[a]	EMR::Cm (cat)
	or ECM 2024	AcrAB[a]	ECM1197 acrAB::Kan
Pasteurella multocida	ATCC43134		Wild type
Mannhemia haemolytica	ATCC33396		Wild type

[a] AcrAB efflux pump expressed at a low level

Assays were performed in Cation-Adjusted Mueller-Hinton Broth (CAMHB) at a bacterial inoculum of 5 x 10^5 CFU/ml and a final volume of 100 µl. Florfenicol and chloramphenicol controls and test compounds were prepared at four times the desired final concentration. Dilution to the desired concentration was accomplished directly on the plates by serial 2-fold dilution using a multi-channel pipette. After dilution, 25 µl of CAMHB was added to each well.

The bacterial inocula were prepared as follows. For each strain, one isolated colony was used to inoculate a volume of 5 ml of CAMHB. The cultures were incubated overnight (20 hours) at 35 °C in a shaking incubator. They were then diluted in sterile saline to a density equivalent to a 0.5 McFarland suspension (approx. 10^8 CFU/ml). The suspensions were further diluted in CAMHB to approximately 5 x 10^5 CFU/ml. A volume of 50 µl of the inoculum was added to each well. Positive and negative growth controls were included on each plate. The original inocula were determined by applying 10 µl of several 10-fold dilutions on TSA plates. Agar plates were incubated overnight at 35 °C and colony-forming units (CFU) counted. Microtiter plates were incubated for 20 hours at 35 °C and were read using a microtiterplate reader (Molecular Devices) at 650 nm and by visual observation using a microtiterplate reading mirror to determine the MIC.

Pharmaceutical compositions

A compound of the present invention, a prodrug thereof or a physiologically acceptable salt of either the compound or its prodrug, can be administered as such to a patient or can be administered in pharmaceutical compositions in which the foregoing materials are mixed with suitable excipient(s). Techniques for formulation and administration of drugs may be found in *Remington's Pharmacological Sciences*, Mack Publishing Co., Easton, PA, latest edition. The formulations and techniques discussed in Remington relate primarily to use with human patients; however, they may readily modified for use with non-human patients by techniques well-known to those skilled in the veterinary art.

Routes of Administration

As used herein, "administer" or "administration" refers to the delivery of a compound, salt or prodrug of the present invention or of a pharmaceutical composition containing a compound, salt or prodrug of this invention to an organism for the purpose of treating or preventing a microbial infection.

Suitable routes of administration may include, without limitation, oral, rectal, transmucosal, intramuscular, subcutaneous, intramedullary, intrathecal, direct intraventricular, intravenous, intravitreal, intraperitoneal, intranasal, aural or intraocular. The preferred routes of administration are oral and parenteral.

Alternatively, one may administer the compound in a local rather than systemic manner, for example, by preparation as a salve that is applied directly to the infected area or by injection of the compound directly into infected tissue. In either case, a sustained release formulation may be used.

Composition/Formulation

Pharmaceutical compositions of the present invention may be manufactured by processes well known in the art, e.g., using a variety of well-known mixing, dissolving, granulating, dragee-making, levigating, emulsifying, encapsulating, entrapping or lyophilizing processes. The compositions may be formulated in conjunction with one or more physiologically acceptable carriers comprising excipients and auxiliaries which facilitate processing of the active compounds into preparations which can be used pharmaceutically. Proper formulation is dependent upon the route of administration chosen.

For injection, including, without limitation, intravenous, intramusclular and subcutaneous injection, the compounds of the invention may be formulated in aqueous solutions, preferably in physiologically compatible buffers such as Hanks' solution, Ringer's solution, physiological saline buffer or polar solvents including, without limitation, N-methyl-2-pyrrolidone, 2-pyrrolidone, other pyrrolidones, N,N-dimethylacetamide, N,N-dimethylformamide, dimethylsulfoxide, acetone and glycerol formal. For transmucosal administration, penetrants appropriate to the barrier to be permeated are used in the formulation. Such penetrants are generally known in the art.

For oral administration, the compounds can be formulated by combining the active compounds with pharmaceutically acceptable carriers well-known in the art. Such carriers enable the compounds of the invention to be formulated as tablets, pills, lozenges, dragees, capsules, liquids, gels, syrups, pastes, slurries, solutions, suspensions, concentrated solutions and suspensions for diluting in the drinking water of a patient, premixes for dilution in the feed of a patient, and the like, for oral ingestion by a patient. Pharmaceutical preparations for oral use can be made using

a solid excipient, optionally grinding the resulting mixture, and processing the mixture of granules, after adding other suitable auxiliaries if desired, to obtain tablets or dragee cores. Useful excipients are, in particular, fillers such as sugars, including lactose, sucrose, mannitol, or sorbitol, cellulose preparations such as, for example, maize starch, wheat starch, rice starch and potato starch and other materials such as gelatin, gum tragacanth, methyl cellulose, hydroxypropyl- methylcellulose, sodium carboxy- methylcellulose, and/or polyvinylpyrrolidone (PVP). If desired, disintegrating agents may be added, such as cross-linked polyvinyl pyrrolidone, agar, or alginic acid. A salt such as sodium alginate may also be used.

Dragee cores are provided with suitable coatings. For this purpose, concentrated sugar solutions may be used which may optionally contain gum arabic, talc, polyvinyl pyrrolidone, carbopol gel, polyethylene glycol, and/or titanium dioxide, lacquer solutions, and suitable organic solvents or solvent mixtures. Dyestuffs or pigments may be added to the tablets or dragee coatings for identification or to characterize different combinations of active compound doses.

Pharmaceutical compositions that can be used orally include push-fit capsules made of gelatin, as well as soft, sealed capsules made of gelatin and a plasticizer, such as glycerol or sorbitol. The push-fit capsules can contain the active ingredients in admixture with a filler such as lactose, a binder such as starch, and/or a lubricant such as talc or magnesium stearate and, optionally, stabilizers. In soft capsules, the active compounds may be dissolved or suspended in suitable liquids, such as fatty oils, liquid paraffin, or liquid polyethylene glycols. Stabilizers may be added in these formulations, also.

For administration by inhalation, the compounds of the present invention can conveniently be delivered in the form of an aerosol spray using a pressurized pack or a nebulizer and a suitable propellant, e.g., without limitation, dichlorodifluoro-methane, trichlorofluoromethane, dichlorotetrafluoroethane or carbon dioxide. In the case of a pressurized aerosol, the dosage unit may be controlled by providing a valve to deliver a metered amount. Capsules and cartridges of, for example, gelatin for use in an inhaler or insufflator may be formulated containing a powder mix of the compound and a suitable powder base such as lactose or starch.

The compounds may also be formulated for parenteral administration, e.g., by bolus injection or continuous infusion. Formulations for injection may be presented in

unit dosage form, e.g., in ampoules or in multi-dose containers. Useful compositions include, without limitation, suspensions, solutions or emulsions in oily or aqueous vehicles, and may contain adjuncts such as suspending, stabilizing and/or dispersing agents. Pharmaceutical compositions for parenteral administration include aqueous solutions of a water soluble form, such as, without limitation, a salt, of the active compound. Additionally, suspensions of the active compounds may be prepared in a lipophilic vehicle. Suitable lipophilic vehicles include fatty oils such as sesame oil, synthetic fatty acid esters such as ethyl oleate and triglycerides, or materials such as liposomes. Aqueous injection suspensions may contain substances that increase the viscosity of the suspension, such as sodium carboxymethyl cellulose, sorbitol, or dextran. Optionally, the suspension may also contain suitable stabilizers and/or agents that increase the solubility of the compounds to allow for the preparation of highly concentrated solutions. Alternatively, the active ingredient may be in powder form for constitution with a suitable vehicle, e.g., sterile, pyrogen-free water, before use.

The compounds may also be formulated in rectal compositions such as suppositories or retention enemas, using, e.g., conventional suppository bases such as cocoa butter or other glycerides.

In addition to the formulations described previously, the compounds may also be formulated as depot preparations. Such long acting formulations may be administered by implantation (for example, subcutaneously or intramuscularly) or by intramuscular injection. A compound of this invention may be formulated for this route of administration with suitable polymeric or hydrophobic materials (for instance, in an emulsion with a pharmacologically acceptable oil), with ion exchange resins, or as a sparingly soluble derivative such as, without limitation, a sparingly soluble salt.

Other delivery systems for relatively hydrophobic pharmaceutical compounds may be employed. Liposomes and emulsions are well-known examples of delivery vehicles or carriers for hydrophobic drugs. In addition, organic solvents such as dimethylsulfoxide may be used, although often at the risk of greater toxicity.

Additionally, the compounds may be delivered using a sustained-release system, such as semi-permeable matrices of solid hydrophobic polymers containing the therapeutic agent. Various sustained-release materials have been established and are well known by those skilled in the art. Sustained-release capsules may,

depending on their chemical nature, release the compounds for a few weeks up to over 100 days. Depending on the chemical nature and the biological stability of the particular compound, additional stabilization strategies may be employed.

Pharmaceutical compositions useful herein also may comprise solid or gel phase carriers or excipients. Examples of such carriers or excipients include, but are not limited to, calcium carbonate, calcium phosphate, various sugars, starches, cellulose derivatives, gelatin, and polymers such as polyethylene glycols.

Dosage

A therapeutically effective amount refers to an amount of compound effective to prevent, alleviate or ameliorate symptoms of a microbial infection. Determination of a therapeutically effective amount is well within the capability of those skilled in the art, especially in light of the disclosure herein.

For any compound used in the methods of the invention, the therapeutically effective amount can be estimated initially from cell culture assays. Then, the d sage can be formulated for use in animal models so as to achieve a circulating concentration range that includes the MIC as determined in cell culture. Such information can then be used to more accurately determine dosages useful in patients.

Toxicity and therapeutic efficacy of the compounds described herein can be determined by standard pharmaceutical procedures in cell cultures or experimental animals. For example, the MIC and the LD_{50} for a particular compound can be determined by methods well-known in the art. The data obtained can be used to formulat a range of dosages useful in patients. The dosage, of course, may vary depending upon the dosage form and route of administration. The exact formulation, route of administration and dosage can be selected by the individual physician in view of the patient's condition. (See e.g., Fingl, et al., 1975, in "The Pharmacological Basis of Therapeutics", Ch. 1 p.1). In general, however, the presently preferred dosage range for systemic delivery of a compound of this invention will be from about 1 to about 100 mg/Kg. The presently preferred dosage range for topical use will generally be from about 0.1 mg to about 1 gm.

Dosage amount and interval may be adjusted individually to provide plasma levels of the compound that are sufficient to maintain a concentration equal to the MIC or any other desired level. Such plasma levels are often referred to as minimum

effective concentrations (MECs). The MEC will vary for each compound but can be estimated from in vitro data, e.g., the concentration necessary to achieve 80+% inhibition of a microbe, may be ascertained using the assays described herein. Dosages necessary to achieve the MEC will depend on individual characteristics and route of administration. HPLC assays or bioassays can be used to determine plasma concentrations.

Dosage intervals can also be determined using MEC value. Compounds should be administered using a regimen that maintains plasma levels above the MEC for 10-90% of the time, preferably between 30-90% and most preferably between 50-90%.

In cases of local administration or selective uptake, the effective local concentration of the drug may not be related to plasma concentration and other procedures known in the art may be employed to determine the correct dosage amount and interval.

The amount of a composition administered will, of course, be dependent on the patient being treated, the severity of the infection, the manner of administration, the judgment of the prescribing physician, etc.

Packaging

The compositions may, if desired, be presented in a pack or dispenser device, such as an FDA approved kit, which may contain one or more unit dosage forms containing the active ingredient. The pack may for example comprise metal or plastic foil, such as a blister pack. The pack or dispenser device may be accompanied by instructions for administration. The pack or dispenser may also be accompanied by a notice associated with the container in a form prescribed by a governmental agency regulating the manufacture, use or sale of pharmaceuticals, which notice is reflective of approval by the agency of the form of the compositions or of human or veterinary administration. Such notice, for example, may be of the labeling approved by the U.S. Food and Drug Administration for prescription drugs or of an approved product insert. Compositions comprising a compound of the invention formulated in a compatible pharmaceutical carrier may also be prepared, placed in an appropriate container, and labeled for treatment of an indicated condition.

Examples

The following examples are provided to illustrate certain embodiments of this invention and are not intended, nor are they to be construed, to limit its scope in any manner whatsoever.

Starting materials were obtained from commercial suppliers and used without further purification unless otherwise noted. Chemical suppliers included Aldrich, Fluka, and Lancaster. $Pd(PPh_3)_4$ was obtained from Lancaster or Strem and immediately transferred into N_2 flushed vials (between 10 and 100 mg each) in an N_2 glove bag. The vials were wrapped in aluminum foil and stored in N_2-flushed, zip-lock baggies at –20 °C. $PdCl_2(dppf)$ was obtained from Aldrich and used from the bottle. Standard reagent grade solvents, which were not necessarily anhydrous, were used. Anhydrous solvents were purchased from chemical suppliers and used as is.

^1H NMR spectra were recorded on a 300 MHz Varian FT-NMR spectrometer and are reported in the format "chemical shift (multiplicity, integration, coupling constant)." Coupling constants are reported in Hz. Mass spectra were obtained on a Micromass Platform II single quadrupole mass spectrometer equipped with electrospray ionization (ESI).

The following three Suzuki cross-coupling methods were used:

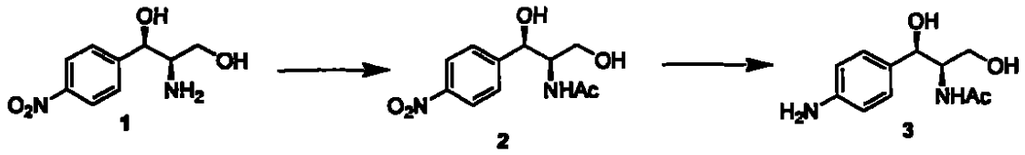
Method A: the aryl boronic acid (0.167 mmol) and aryl bromide (0.334 mmol) were combined in a mixture of aqueous Na_2CO_3 (3 mL of a 10% (w/w) solution) and THF (5 mL). The mixture was purged briefly with N_2. $Pd(PPh_3)_4$ (10 mol%, 0.0167 mmol) was added, the mixture purged with N_2, and then refluxed for 16 hours. The reaction mixture was diluted with ethyl acetate (EtOAc), washed with a saturated brine solution, dried over anhydrous Na_2SO_4 and concentrated. The residue was then purified by chromatography.

Method B: the aryl boronic acid (0.301 mmol), the aryl bromide (0.602 mmol), and Cs_2CO_3 (0.903 mmol) were combined in THF (2.0 mL), DMF (2,0 mL), and H_2O (0.5 mL) at room temperature. The mixture was purged with N_2 for 5 min and [1,1'-bis(diphenylphosphino)ferrocene]dichloropalladium(II) dichloromethane complex (0.0301 mmol) was added. The mixture was purged with N_2 for 5 minutes and then stirred at 55 °C for 16 hours. The mixture was concentrated under vacuum, diluted

with EtOAc and washed with brine. The EtOAc was dried over anhydrous Na_2SO_4 and concentrated. The residue was purified by chromatography.

Method C: the aryl boronic acid (0.934 mmol), the aryl bromide (2.34 mmol), and Cs_2CO_3 (2.80 mmol) were suspended in a mixture of toluene (4 mL), *n*-butanol (4 mL), and H_2O (2 mL). The mixture was purged at room temperature with N_2 after which $Pd(PPh_3)_4$ (0.280 mmol) was added. The mixture was purged with N_2 for an additional 5 minutes and then heated to 75 °C. After 12 hours, the mixture was cooled to room temperature and concentrated under vacuum. The residue was partitioned between H_2O (75 mL) and EtOAc (75 mL). The EtOAc layer was washed with brine (2 x 50 mL), dried over anhydrous Na_2SO_4, and concentrated under vacuum. The residue was purified by chromatography.

Example 1: Compound 3



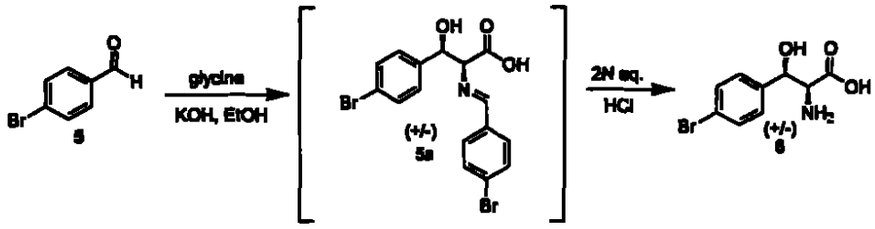
In 80 mL of methanol was dissolved 8.13 g (32.0 mmol) of chloramphenicol base as the *N*-acetate, prepared by literature methods (Rebstock, M. C., *et al.*, *supra*, Crooks H. M. *et al.*, *supra*; Evans, D. D.; Morris, D. S. *et al.*, *supra*). After purging with N_2, Pd/C was added and the mixture was stirred under H_2(1 atm) for 16 hours. The Pd/C was then removed by filtration through Celite. The solution was concentrated under vacuum and the residue purified by silica gel chromatography, eluting with 15% MeOH in CH_2Cl_2 to give 3 (7.13 g, 31.7 mmol). ^1H NMR (300 MHz, CD_3OD): δ 1.92 (s, 3H), 3.40 (dd, 1H, *J* = 11.1, 6.0), 3.61 (dd, 1H, *J* = 11.1, 5.7), 4.01 (m, 1H), 4.73 (d, 1H, *J* = 5.4), 6.69 (d, 2H, *J* = 8.4), 7.11 (d, 2H, *J* = 8.4).

Example 2: Compound 4

An aqueous solution of $NaNO_2$ (1.27 g, 18.5 mmol, 50 mL H_2O) was added dropwise to a solution of 3 (3.76 g, 16.8 mmol) in 35 mL of 48% aqueous HBr at 0

°C. After addition was completed, the mixture was stirred for 30 min at 0 °C. The mixture was then added dropwise to a solution of CuBr (2.65 g, 18.45 mmol) in 15 mL 48% aqueous HBr. The mixture was warmed to room temperature and stirred for an additional 16 hours. The reaction mixture was neutralized with 3 M aqueous NaOH, filtered through a pad of Celite, and extracted with EtOAc (3 x 100 mL). The combined organic fractions were dried over anhydrous Na_2SO_4 and concentrated under vacuum to give 1.2 g (4.17 mmol) of crude product that was used without further purification. ^{1}H NMR (300 MHz, CD_3OD): δ 1.87 (s, 3H), 3.48 (dd, 1H, J = 11.0, 5.9), 3.69 (dd, 1H, J = 11.0, 6.5), 4.01 (m, 1H), 4.91 (d, 1H, J = 3.6), 7.30 (d, 2H, J = 8.3), 7.45 (d, 2H, J = 8.3).

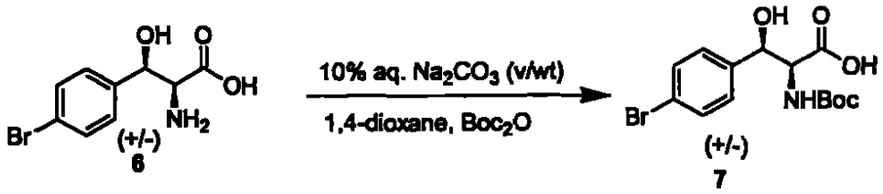
Example 3: Compound 6



4-Bromobenzaldehyde (100 g, 0.540 mol) was dissolved in ethanol (EtOH) in a 2 L round-bottom flask. With rapid stirring, glycine (0.5 molar equivalents, 20.3 g, 0.270 mol) and then, in one portion, KOH (30.3 g, 0.540 mol) were added. It is essential to add the KOH all at once. Thus, an ice bath should be used when performing the reaction on a larger scale to control the exotherm. After addition of the KOH, the turbid suspension became yellow and homogeneous. After about 15 minutes, a thick white precipitate began to form. The mixture was stirred for 12 hours at room temperature under N_2. Enough 2 N aqueous HCl was added (~400 mL) to make the solution red to pH paper. The mixture was then stirred at approximately 60 °C until it again become a homogeneous yellow solution. The EtOH was removed under vacuum to give an aqueous suspension of white precipitate. The precipitate was filtered and the remaining aqueous solution washed three times with EtOAc. The aqueous solution was then basified to about pH 9 with concentrated aqueous NH_3 and excess ammonia was removed under vacuum. As the NH_3 was removed, the product, 6, began to precipitate. Evaporation continued to one-quarter the volume of solution where precipitation was first observed. The

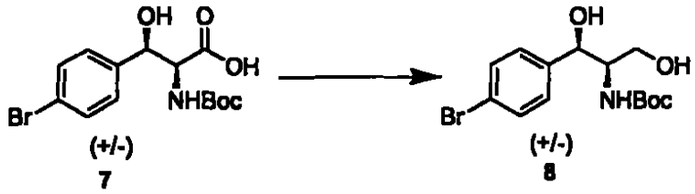
product was then collected on a vacuum filter and dried under vacuum to a constant weight (51.3 g). If NMR indicates the presence of the undesired trans stereoisomer, it can be removed by recrystallization from H_2O/ EtOH). ^1H NMR (300 MHz, CD_3OD): δ 3.64 (d, 1H, *J* = 3.6), 5.25 (d, 1H, *J* = 3.6), 7.41 (d, 2H, *J* = 8.4), 7.53 (d, 2H, *J* = 8.4).

Example 4: Compound 7



Compound **6** (32.60 g, 12.53 mmol) was dissolved in 10% aqueous K_2CO_3 (10% w/v; 500 mL). Di-*tert*-butyl dicarbonate (34.2 g, 157 mmol) was dissolved in 1,4-dioxane (500 mL) and added to the aqueous solution, after which the mixture was stirred at room temperature for 72 hours. The mixture was concentrated under vacuum and taken up in 100 mL 1 N aqueous NaOH and washed with Et_2O (2 x 100 mL). The aqueous layer was acidified with 1 N aqueous HCl and the product extracted into EtOAc (3 x 200 mL). The combined EtOAc extracts were dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum. The product (26.7 g, 74.0 mmol) was isolated and used without further purification. ^1H NMR (300 MHz, CD_3OD): δ 1.32 (s, 9H), 4.37 (d, 1H, *J* = 2.7), 5.23 (d, 1H, *J* = 2.7), 7.32 (d, 2H, 8.4), 7.46 (d, 2H, *J* = 8.4).

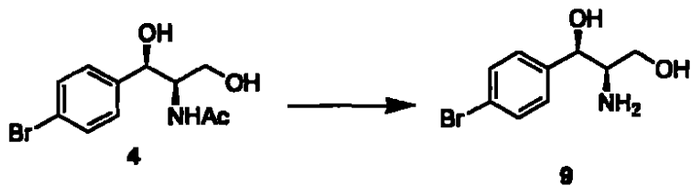
Example 5: Compound 8



Compound **7** (5.29 g, 14.7 mmol) and *N*-hydroxysuccinimide (1.69 g, 14.7 mol) were dissolved in EtOAc (200 mL) and the mixture cooled to 0 °C. *N,N'*-dicyclohexyl- carbodiimide (3.04 g, 14.7 mmol) was added and the mixture stirred for 30 minutes, warmed to room temperature and stirred for 30 additional minutes. It

was then cooled to 0 °C and filtered to remove the precipitated *N,N'*-dicyclohexylurea byproduct. The filtrate was concentrated under vacuum and the residue dissolved in THF (100 mL). The solution was cooled to 0 °C, $NaBH_4$ (5.6 g, 150 mmol) added, and the mixture stirred for two minutes. Water was then added to the mixture dropwise until bubbling ceased, then a volume of water equal to the volume of THF was added over 30 minutes, after which the mixture was warmed to room temperature and stirred for 5 hours. The dioxane was removed under vacuum and EtOAc (200 mL) was added, followed by gradual acidification of the aqueous layer with 1 N aqueous HCl. The EtOAc layer was washed with brine and dried over anhydrous Na_2SO_4. Filtration followed by concentration under vacuum gave 4.31 g (12.4 mmol) of **8**, which was used without further purification. ^1H NMR (300 MHz, CD_3OD): δ 1.32 (s, 9H), 3.46-3.51 (m, 1H), 3.63-3.73 (m, 2H), 4.88 (br s, 1H), 7.29 (d, 2H, J = 8.3), 7.45 (d, 2H, J = 8.3).

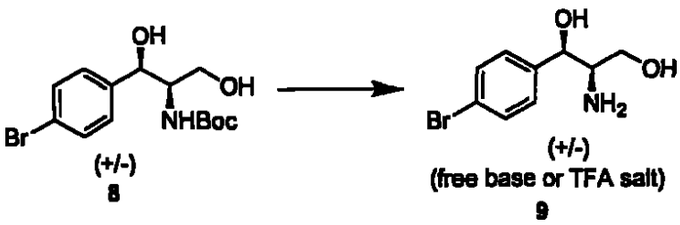
Example 6: Compound 9 (from 4)



4 **9**

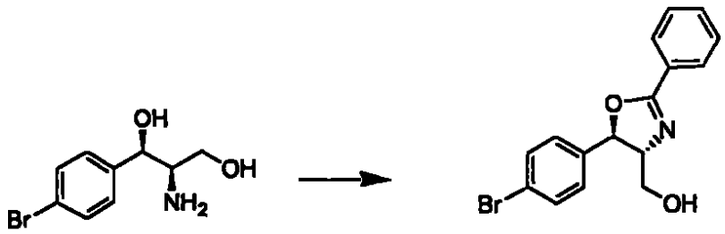
Compound **4** (1.0 g, 3.5 mmol) was dissolved in 10% aqueous H_2SO_4 (10% v/v, 15 mL total) and the solution refluxed for 10 h. The reaction mixture was basified with 3 M aqueous NaOH and extracted with EtOAc (3 x 50 mL). The combined EtOAc fractions were dried over anhydrous Na_2SO_4 and concentrated under vacuum to give 710 mg (2.9 mmol) of **4**, which was used without further purification. ^1H NMR (300 MHz, CD_3OD): δ 2.80-2.90 (br m, 1H), 3.30-3.39 (br m, 1H), 3.40-3.50 (br m, 1H), 4.52-4.58 (m, 1H), 7.30 (d, 2H, J = 8.1), 7.49 (d, 2H, J = 8.1).

Example 7: Compound 9 (from 8)



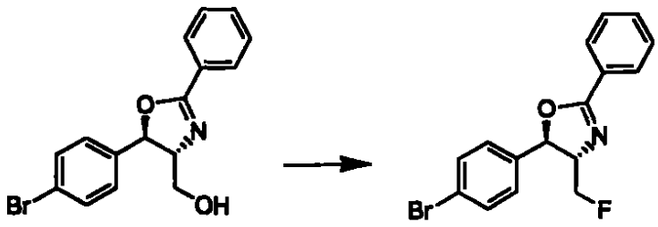
Compound **8** (4.31 g, 12.4 mmol) was dissolved in trifluoroacetic acid (TFA, 40 mL). The mixture was stirred for 3 hours at room temperature and concentrated under vacuum. The TFA salt was partitioned between 50 mL of 2 N aqueous NaOH and an equal volume of EtOAc. The layers were separated and the aqueous layer was washed with EtOAc (50 mL). The combined EtOAc fractions were washed with brine, dried over anhydrous Na_2SO_4 and concentrated under vacuum to give a waxy solid (2.72 g, 11.1 mmol), which was used without further purification. ^{1}H NMR (300 MHz, CD_3OD): δ 2.83-2.90 (m, 1H), 3.30-3.35 (m, 1H), 3.46 (dd, 1H, *J* = 10.8, 4.8), 4.56 (d, 1H, *J* = 6.6), 7.29 (d, 2H, *J* = 8.6), 7.49 (d, 2H, *J* = 8.6).

Example 8: Compound 10



Compound **9** (710 mg, 2.89 mmol), ethyl benzimidate hydrochloride (533 mg, 2.89 mmol) and triethylamine (Et_3N, 0.40 mL, 2.89 mmol) were combined in 25 mL 1,2-dichloroethane. The mixture was refluxed with stirring under N_2 for 16 hours at which time TLC indicated one major product. After cooling to room temperature, the mixture was diluted with EtOAc, washed twice with saturated aqueous NH_4Cl, twice with saturated aqueous $NaHCO_3$ and then dried over anhydrous Na_2SO_4. The solution was filtered and the EtOAc removed under vacuum to give 885 mg (2.66 mmol) of **10**, which was used without further purification. ^{1}H NMR (300 MHz, CD_3OD): δ 3.74-3.89 (m, 2H), 4.13-4.18 (m, 1H), 5.59 (d, 1H, *J* = 6.6), 7.30 (d, 2H, *J* = 8.4), 7.46-7.57 (m, 5H), 7.99-8.02 (m, 2H).

Example 9: Compound 11

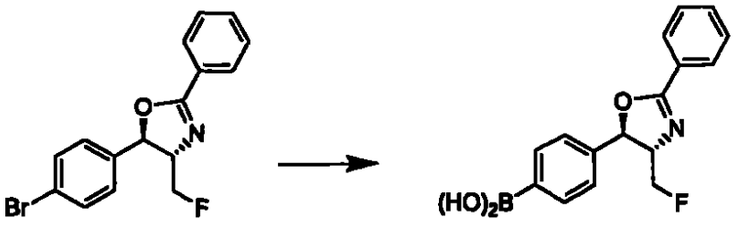


(enantiopure or (+/-))
10

(enantiopure or (+/-))
11

Compound 10 (885 mg, 2.66 mmol) was suspended in CH_2Cl_2 (15 mL) and cooled to –78 °C. Diethylaminosulfur trifluoride (DAST, 0.53 mL, 4.00 mmol) was added by syringe, the mixture warmed over 1 hour to room temperature and then stirred for 16 hours under N_2. Excess DAST was quenched by slow addition of H_2O after which the mixture was diluted with additional CH_2Cl_2. The organic layer was washed with H_2O and saturated aqueous $NaHCO_3$, dried over anhydrous Na_2SO_4 and concentrated under vacuum. The residue (950 mg) was chromatographed on silica gel, eluting with 15:85 EtOAc/hexanes to give 11 as an oil (420 mg, 1.25 mmol). ^1H NMR (300 MHz, $CDCl_3$): δ 4.23-4.35 (m, 1H), 4.45-4.75 (m, 2H), 5.43 (d, 1H, J = 6.9), 7.17(d, 2H, 8.3), 7.36-7.47 (m, 5H), 7.96 (d, 2H, J = 8.3).

Example 10: Compound 12



(enantiopure or (+/-))
11

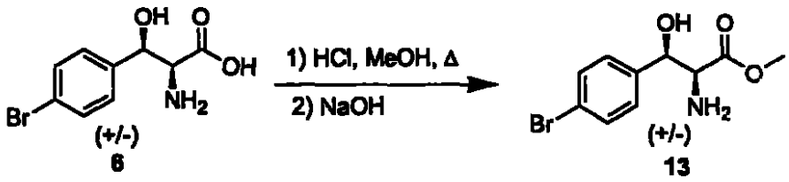
(enantiopure or (+/-))
12

Compound 11 (807 mg, 2.41 mmol, enantiopure from chloramphenicol or racemic from (+/-)-*threo*-*p*-bromophenylserine), was dissolved in anhydrous THF (10

65
66

mL) in a flame-dried round bottom flask and cooled to –78 °C. *n*-BuLi (1.6 M in hexanes, 3.02 mL, 4.83 mmol) was added with vigorous stirring. After 10 minutes, trimethyl borate (0.55 mL, 4.83 mmol) was added, the mixture warmed to room temperature over 1 hour and then stirred for 6 hours. The mixture was quenched with 1 N aqueous HCl and extracted three times with EtOAc. The combined EtOAc fractions were washed three times with brine and dried over anhydrous Na_2SO_4. The EtOAc was removed under vacuum to give 805 mg of material, which was purified by flash column chromatography, eluting initially with 1:1 hexanes/EtOAc to remove impurities followed by elution with 1:9 MeOH/CH_2Cl_2 to obtain the product as an oil (285 mg, 0.953 mmol). ^{1}H NMR (300 MHz, CD_3OD): δ 4.29-4.39 (m, 1H), 4.69 (dd, 2H, *J* = 47.1, 4.2), 5.64 (d, 1H, *J* = 6.6), 7.37 (d, 2H, *J* = 8.1), 7.47-7.52 (m, 2H), 7.56-7.62 (m, 1H), 7.67 (d, 1H, *J* = 7.5) 7.74-7.82 (m, 1H), 8.01 (d, 2H, *J* = 7.2).). LRMS (ESI$^-$) m/z: 298.0 (M-H$^+$ $C_{16}H_{14}BFNO_3$ requires 298.1).

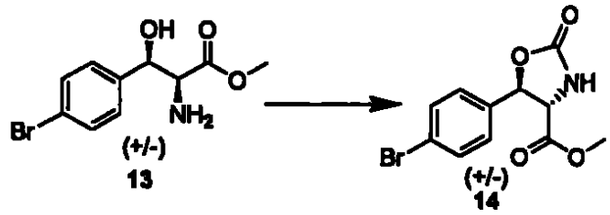
Example 11: Compound 13



Compound **6** (20.6 g, 0.079 mol) was suspended in 400 mL of anhydrous MeOH. The mixture was stirred rapidly and cooled in an ice bath to 0 °C. Anhydrous HCl gas was slowly bubbled in. After 10 minutes, all solids had dissolved. Acidification was continued for 10 minutes after the mixture became homogeneous, at which point the MeOH turned pH paper very red. The apparatus was fitted with a drying tube and the reaction mixture was refluxed for 8 hours followed by stirring at room temperature for 12 hours. The mixture was concentrated under vacuum and then suspended in a mixture of 300 mL of EtOAC and 100 mL of water. With rapid stirring, 3 N NaOH was added very slowly until the suspended material dissolved. Addition of base was continued until a pH of 10 was attained. The layers were separated and the EtOAc fraction was washed with brine (2x) and dried over anhydrous Na_2SO_4. Concentration under vacuum gave **13** as a white powder (15.3 g), which was used without further purification. ^{1}H NMR (300 MHz,

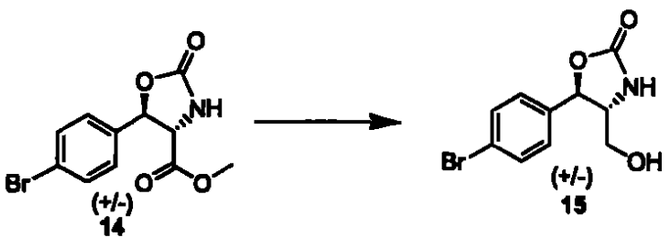
CD_3OD): δ 3.59 (d, 1H, *J* = 4.5), 3.66 (s, 3H), 4.92 (d, 1H, *J* = 4.5), 7.29 (d, 2H, *J* = 8.3), 7.49 (d, 2H, *J* = 8.3).

Example 12: Compound 14



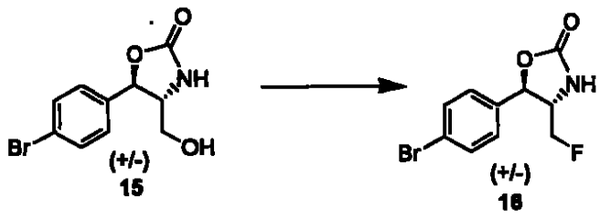
1,1'-Carbonyldiimidazole (23.6 g, 0.146 mol) was dissolved in 250 mL of anhydrous 1,2-dichloroethane at room temperature followed by addition of triethylamine (Et_3N, 12.2 g, 16.8 mL). In a separate flask, 13 (33.26 g, 0.121 mol) was dissolved in 125 mL of anhydrous tetrahydrofuran (THF) and an equivalent of triethylamine. The THF solution was diluted with 125 mL of 1,2-dichloroethane and then added, under N_2, over 2 hours to the 1,1'-carbonyldiimidazole solution. After addition was complete, the mixture was stirred for 2 hours under N_2 at room temperature. The reaction mixture was concentrated under vacuum and the residue diluted with 300 mL of EtOAc, which was washed with 5 x 200 mL of 2 N HCl, 3 x 300 mL of saturated $NaHCO_3$, and 2 x 200 mL of brine. The EtOAc layer was dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum to give 34.27 g of 14, 80-90% pure by ^1H NMR. Compound 14 was used without further purification. ^1H NMR(300 MHz, CD_3OD): δ 3.83 (s, 3H), 4.34 (d, 1H, *J* = 5.0), 5.65 (d, 1H, *J* = 5.0), 7.36 (d, 2H, *J* = 8.7), 7.60 (d, 2H, *J* = 8.7).

Example 13: Compound 15



Compound 14 (55.3 g, 0.184 mmol) was dissolved in 375 mL MeOH. The solution was cooled to 0 °C and $NaBH_4$ (2 equivalents, 13.9 g, 0.369 mol) was added in portions, care being taken to not let the temperature exceed 20 °C. After the last portion was added, cooling was ceased and the mixture stirred for 2 hours. Glacial acetic acid was added slowly until a pH of 7 was achieved. The mixture was filtered through Celite and concentrated under vacuum. The residue was partitioned between EtOAc (375 mL) and 2 N HCl (500 mL). The organic layer was washed with 2 N HCl (2 x 300 mL), saturated aqueous $NaHCO_3$ (2 x 250 mL) and brine (250 mL), dried over anhydrous Na_2SO_4, filtered and concentrated under vacuum. The product was crystallized from hexanes/EtOAc to give 26.3 g of 15. ^1H NMR(300 MHz, CD3OD): δ 3.64-3.72 (m, 3H), 5.38 (d, 1H, J = 5.1), 7.32 (d, 2H, J = 8.6), 7.57 (d, 2H, J = 8.6).

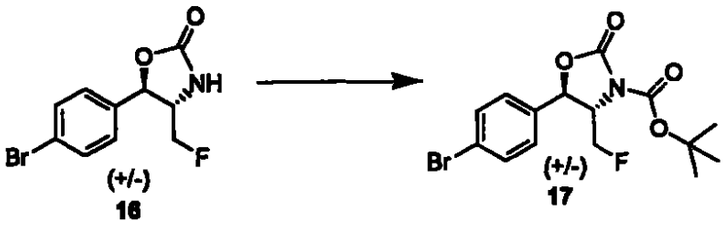
Example 14: Compound 16



Compound 15 (7.57 g, 0.0278 mol) was suspended in 275 mL of CH_2Cl_2. Under N_2 and with rapid stirring, the biphasic mixture was cooled to –78 °C in a dry ice/acetone bath. DAST (5.51 mL, 6.72 g, 0.0417 mol) was added dropwise by syringe over 2-3 minutes. After 30 minutes the mixture was transferred to an ice water bath and stirred for 30 minutes, during which time the solids dissolved. Another 1 mL of DAST was added to ensure completion of the reaction. The mixture was stirred for 20 minutes at 0 °C and then warmed to room temperature. The mixture was stirred at room temperature for 20 minutes and then cooled to 0 °C. Excess DAST was quenched by dropwise addition of saturated aqueous $NaHCO_3$ over 30 minutes with very rapid stirring. After bubbling ceased, the aqueous layer was slightly basic (7 < pH < 9). The CH_2Cl_2 was diluted with enough EtOAc (~600 mL) to bring the organic layer to the top during aqueous extraction. The organic layer was washed with saturated aqueous $NaHCO_3$ (1 x 300 mL), 1 N HCl (2 x 200 mL), saturated aqueous $NaHCO_3$ (3 x 200 mL) and brine (1 x 150 mL) and dried

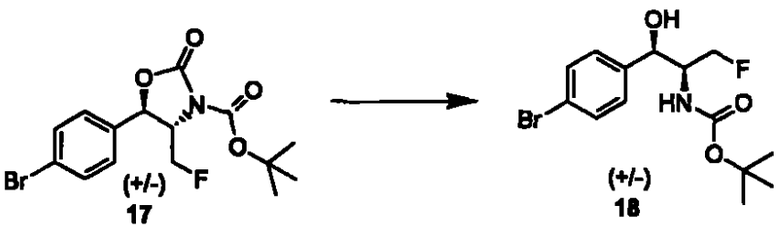
over anhydrous Na_2SO_4. After filtration, the mixture was concentrated under vacuum to give 8.13 g of brown oil. ^1H NMR(300 MHz, CD_3OD): δ 3.87-3.98 (dm, 1H, *J* = 20 (F-CH_N)), 4.54 (dd, 2H, *J* = 46.7 (CH_2-F), 4.2), 5.42 (d, 1H, *J* = 5.1), 7.34 (d, 2H, *J* = 8.4), 7.59 (d, 2H, *J* = 8.4).

Example 15: Compound 17



Compound **16** (8.13 g or 0.0297) was dissolved in 200 mL of CH_3CN. Boc_2O (9.71 g, 1.5 equivalents, 0.0445 mol) and DMAP (362 mg, 0.00297 mol, 0.1 equivalent) were then added. The mixture was stirred for 2 hours at room temperature under N_2. Concentration of the mixture under vacuum was followed by partitioning between 200 mL of EtOAc and 200 mL of 1 N HCl. The EtOAc layer was washed with 2 N HCl (3 x 100 mL), saturated aqueous $NaHCO_3$ (2 x 100 mL) and brine. The EtOAc layer was filtered, dried over anhydrous Na_2SO_4 and concentrated under vacuum. The residue was recrystallized twice from hexanes/EtOAc. ^1H NMR (300 MHz, CD_3OD): δ 1.53 (s, 9H), 4.29-4.40 (dm, 1H, *J* = 24.6), 4.63-4.98 (m, 2H), 5.53 (d, 1H, *J* = 4.2), 7.34 (d, 2H, *J* = 8.4), 7.61, (d. 2H, *J* = 8.4).

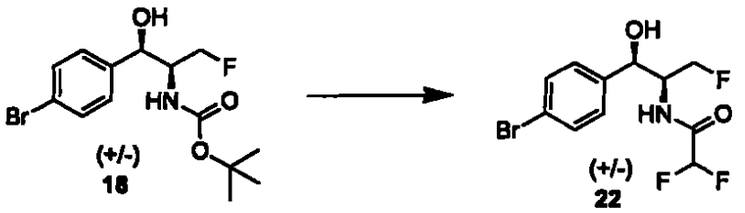
Example 16: Compound 18



Compound **17** (0.050 g, 0.134 mmol) was suspended in 5 mL of CH_3OH. Cs_2CO_3 (8.7 mg, 20 mol%, 0.0267 mmol) was added with rapid stirring at room temperature. The solids dissolved in about 10 minutes after which stirring was continued for 10 minutes, at which time TLC (3:1 hexanes/EtOAc) indicated that the

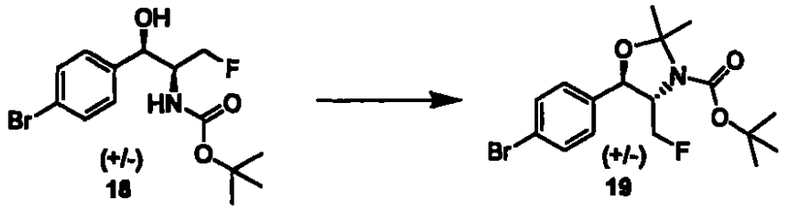
reaction was complete. The reaction mixture was concentrated under vacuum and the residue partitioned between 20 mL of EtOAc and 10 mL of H_2O. The H_2O layer was acidified slightly with 1N aqueous HCl, the mixture vigorously shaken and the layers separated. The EtOAc layer was washed with saturated aqueous $NaHCO_3$ (1 x 20 mL) and brine (1 x 20 mL). The EtOAc was dried over anhydrous Na_2SO_4 and concentrated under vacuum to give 43.4 mg (0.124 mmol) of **18** as a white foam. ^1H NMR (300 MHz, CD_3OD): δ 1.33 (s, 9H), 3.90-4.57 (m, 4H), 7.29 (d, 2H, J = 8.1), 7.46 (d. 2H, J = 8.1)

Example 17: Compound 22



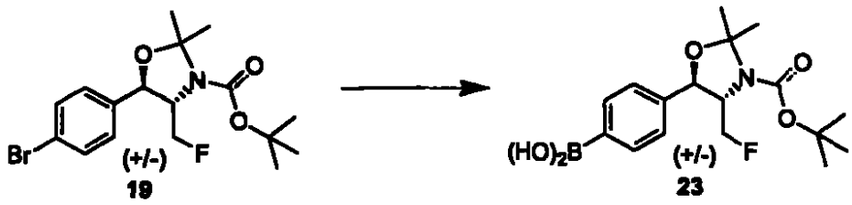
Compound **18** (0.740 g, 2.13 mmol) was stirred in 10 mL TFA/H_2O (9/1, v/v) at room temperature for one hour. The mixture was concentrated under vacuum and partitioned between 30 mL of EtOAc and 30 mL of 1N aqueous NaOH. The mixture was shaken vigorously and the layers allowed to separate. The organic layer was washed with brine (2 x), dried over anhydrous Na_2SO_4 and concentrated under vacuum to give 0.484 g (1.95 mmol, 92%) of product, which was dissolved in 20 mL of CH_2Cl_2. To this was added 1 mL of Et_3N and 1 mL of difluoroacetyl chloride, prepared according to literature procedure (Yu, K.-L., et al., *J. Med. Chem.*, **1996**, 39, 2411-2421.). After 1 hour, the mixture was concentrated under vacuum. The residue was placed in 10 mL of a 1:8:1 (v/v/v) solution of Et_3N/MeOH/H_2O and stirred for one hour at room temperature to cleave any difluoroacetyl groups on the benzylic oxygen. The mixture was then concentrated under vacuum, and partitioned between EtOAc (50 mL) and 1 N HCl (50 ml). The EtOAc layer was washed twice with 20 mL of 1 N HCl, twice with 20 mL saturated aqueous $NaHCO_3$ and twice with 20 mL brine. The EtOAc was then dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum. Compound **22** (584 mg, 1.79 mmol) was used without further purification. ^1H NMR (300 MHz, CD_3OD): δ 4.25-4.67 (m, 3H), 4.90 (d, 1H, J = 3.9), 5.98 (t, 1H, J = 53.9), 7.30 (d. 2H, J = 8.6), 7.48 (d, 2H, J = 8.6).

Example 18: Comp und 19



Compound **18** (1.03 g, 2.94 mmol) was dissolved in CH_2Cl_2 (30 mL) in a 50 mL round-bottom flask. 2-Methoxypropene (339 µL, 3.54 mmol) was added by syringe, followed by a single crystal of *p*-toluenesulfonic acid (*p*-TsOH) hydrate. After about 1 minute, the mixture turned yellow. TLC (4:1 hexanes/EtOAc) showed some **18** to still be present. An additional 5 drops of 2-methoxypropene were added followed two minutes later by another 5 drops, a which time TLC indicated the reaction was complete. The CH_2Cl_2 was removed under vacuum and the residue partitioned between EtOAc (75 mL) and saturated aqueous $NaHCO_3$ (50 mL). The EtOAc layer was washed twice with 50 mL $NaHCO_3$ and twice with brine. The EtOAc layer was dried over anhydrous Na_2SO_4, filtered and evaporated to give a yellow oil (1.08 g, 2.78 mmol) that solidified to a waxy solid, which was used without further purification. ^1H NMR (300 MHz, CD_3OD): δ 1.49 (s, 9H), 1.55 (s, 3H), 1.67 (s, 3H), 3.70-3.85 (m, 1H), 4.34-4.53 (m, 2H), 5.08, (d. 1H, J = 7.5), 7.37 (d, 2H, J = 8.6), 7.54 (d, 2H, J = 8.6).

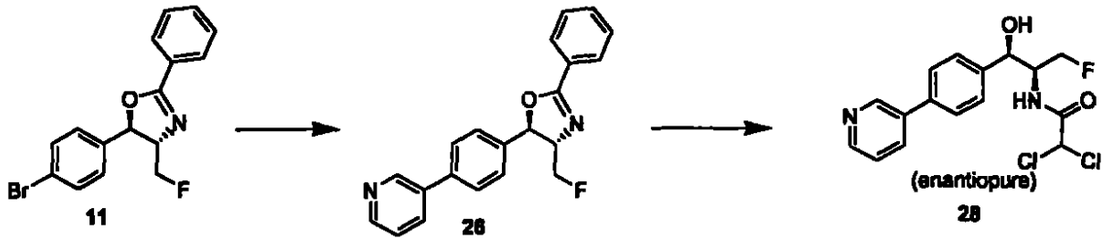
Example 19: Compound 23



Compound **19** (309 mg, 0.796 mmol) was placed in an oven-dried 50 mL round-bottom flask equipped with a magnetic stir bar. The flask was immediately capped with a rubber septum and flushed with dry N_2. Anhydrous THF (15 mL) was added by syringe and a long cannula was used to flush the solvent with N_2 for 10 minutes. The mixture was then cooled to –78 °C. With vigorous stirring, *n*-BuLi (850 µL, 0.995 mmol) was added dropwise by syringe over 3 minutes to give a clear

orange solution. After 20 minutes, $B(OMe)_3$ (158 µL, 1.39 mmol), which had been stored over 4Å activated molecular sieves for at least 48 hours, was added dropwise over one minute using an oven-dried, gas-tight syringe. The mixture was warmed to room temperature and stirred under N_2 for 16 hours. Approximately 20 mL of saturated aqueous NH_4Cl was added with vigorous stirring, which resulted in a white precipitate dispersed in the two liquid phases. The mixture was stirred for an additional 90 minutes and then diluted with EtOAc and H_2O until all the precipitate dissolved. The organic layer was separated, washed with brine and dried over anhydrous Na_2SO_4. After filtration, the EtOAc was removed under vacuum to give 328 mg of yellow oil. The product was purified by silica gel chromatography to give **23** (163 mg). ^1H NMR (300 MHz, CD_3OD): δ 1.49 (s, 9H), 1.56 (s, 3H), 1.67 (s, 3H), 3.74-3.88 (m, 1H), 4.33-1.52 (m, 2H), 5.10 (d, 1H, J = 7.5), 7.43 (d, 2H, J = 8.1), 7.64 (d, 2H, J = 8.1). LRMS (ESI^-) m/z: 352.2 ($M\text{-}H^+$ $C_{17}H_{24}BFNO_5$ requires 352.7).

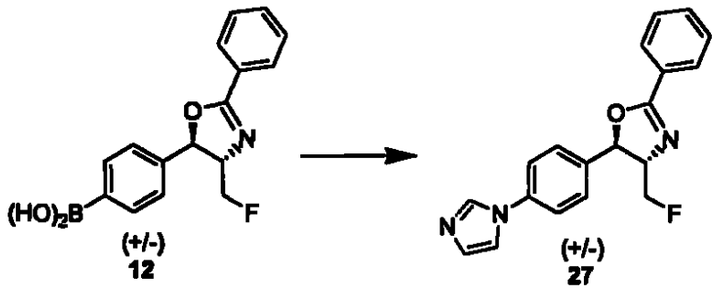
Example 20: Compound 28



This is an example of Suzuki coupling method A. Compound **11** (47 mg, 0.14 mmol) and pyridine-3-boronic acid (21 mg, 0.17 mmol) were dissolved in 5.0 mL of THF. $Pd(PPh_3)_4$ (11 mg, 0.0098 mmol) and 3.0 mL of 10% (w/v) aqueous Na_2CO_3 were added. The reaction mixture was refluxed for 16 hours, cooled, diluted with EtOAc and washed twice with 10% (w/v) aqueous Na_2CO_3 and twice with brine. The EtOAc layer was then dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum. Silica gel chromatography (eluting with 1:1 hexanes/EtOAc) gave 47 mg (0.14 mmol) of **26**. Intermediate **26** was heated to 105 °C in a sealed tube with 6M aqueous HCl and held at that temperature for 16 hours. The mixture was cooled to room temperature and basified to pH 10 with 3 N NaOH. The product was extracted into EtOAc, which was dried over Na_2SO_4, filtered, and concentrated under vacuum to give 42 mg of the deprotected free amine, contaminated with the

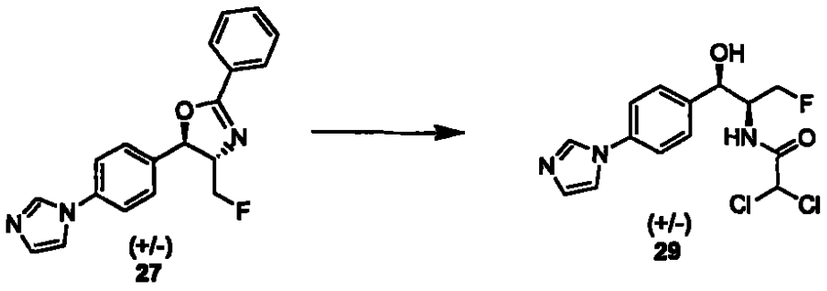
expected benzoic acid-related by-product arising from cleavage of the protecting group. The crude residue was dichloroacetylated as in the synthesis of **29**. Silica gel chromatography, eluting with 7.5% MeOH in CH_2Cl_2, gave 10 mg of product. ^1H NMR (300 MHz, CD_3OD): δ 4.31-4.74 (m, 3H), 5.02 (d, 1H, *J* = 3.6), 6.27 (s, 1H), 7.49-7.56 (m, 3H), 7.64 (d, 2H, *J* = 8.4), 8.09 (ddd, 1H, *J* = 8.0, 2.3, 1.5), 8.50 (br d, 1H, *J* = 3.9), 8.78 (br s, 1H). LRMS (ESI^+) m/z: 356.9 (calc. for $M+H^+$: $C_{16}H_{16}Cl_2FN_2O_2$ 357.0).

Example 21: Compound 27



Boronic acid **12** (43 mg, 0.144 mmol), imidazole (15 mg, 0.215 mmol) and 4Å powdered molecular sieves (110 mg) were combined in CH_2Cl_2 (4.0 mL) and pyridine (23 μL). With rapid stirring, $Cu(OAc)_2$ (26 mg, 0.144 mmol) was added and the mixture stirred, exposed to air, for 40 hours at room temperature. The mixture was quenched with 3 mL of 2 M NH_3 in MeOH. The mixture was filtered through Celite and concentrated under vacuum. The product was purified by flash column chromatography (19:1 CH_2Cl_2/MeOH). Although ^1H NMR indicated that a significant side-product had eluted with the desired product, the mixture was used without further purification. LRMS (ESI^+) m/z: 321.9 (calc. for $M+H^+$ $C_{19}H_{17}FN_3O$ 322.1).

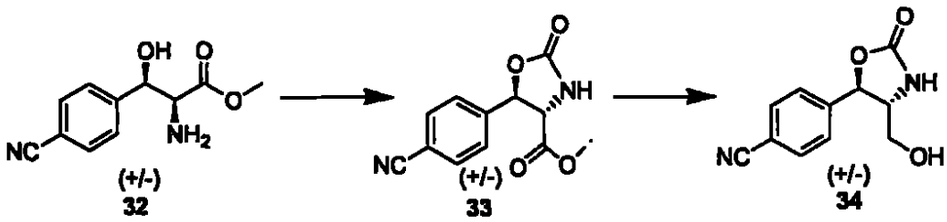
Example 22: Compound 29



Compound **27** (27 mg, 0.084 mmol) was heated with 6 N HCl (3.0 mL) to 100 °C in a sealed tube and held for 16 hours. The mixture was cooled to room temperature, basified with 3 N aqueous NaOH and extracted three times with EtOAc. The combined EtOAc fractions were dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum to give 9 mg of residue. LRMS (ESI^+) m/z: 236.1 (Calc. for $M+H^+$ $C_{12}H_{15}FN_3O$ 236.1).

The residue was refluxed in MeOH (5.0 mL) containing Et_3N (26 µL 0.189 mmol) and methyl dichloroacetate (13 µL, 0.126 mmol) for 3 hours. The mixture was concentrated under vacuum and purified by silica gel chromatography (7.5% MeOH in CH_2Cl_2) to give **29** (6.0 mg, 0.017 mmol). ^1H NMR (300 MHz, CD_3OD): δ 4.33-4.75 (m, 3H), 5.03 (d, 1H, *J* = 3.3), 6.26 (s, 1H), 7.13 (s, 1H), 7.51-7.58 (m, 5H), 8.10 (s, 1H). LRMS (ESI^+) m/z: 345.8 (calc. for $M+H^+$ $C_{14}H_{15}Cl_2FN_3O_2$ 346.0).

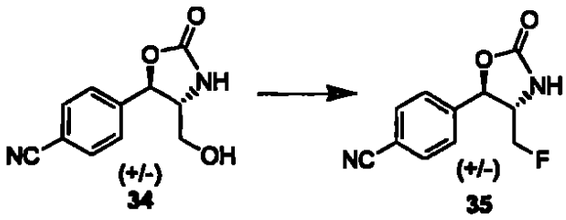
Example 23: Compound 34



Compound **32** was prepared by the procedure of Pines, et al., *supra*. In a 250-mL round-bottom flask was placed 1,1'-carbonyldiimidazole (CDI, 5.06 g, 31.2 mmol) and Et_3N (2.17 mL, 15.6 mmol) in 1,2-dichloroethane (50 mL). Compound **32** (4.00 g, 15.6 mmol) was added to a beaker containing 1,2-dichloroethane (50 mL). Et_3N (4.34 mL, 31.2 mmol) was added to the beaker upon which a thick suspension formed. Additional 1,2-dichloroethane (20 mL) and THF (50 mL) were added to thin the suspension. The mixture was added portion-wise to the CDI suspension over 30 min. The mixture, which became mostly homogenous and turned yellow was stirred for 12 hours. The solvent was removed under vacuum and the residue was partitioned between 2 N aqueous HCl and EtOAc. The EtOAc layer was washed with 2 N HCl (2 x 45 mL), saturated aqueous $NaHCO_3$ (2 x 30 mL) and brine. It was dried over anhydrous Na_2SO_4, filtered, and evaporated to give 3.27 g of **33** as a fluffy yellow solid, which was used without further purification.

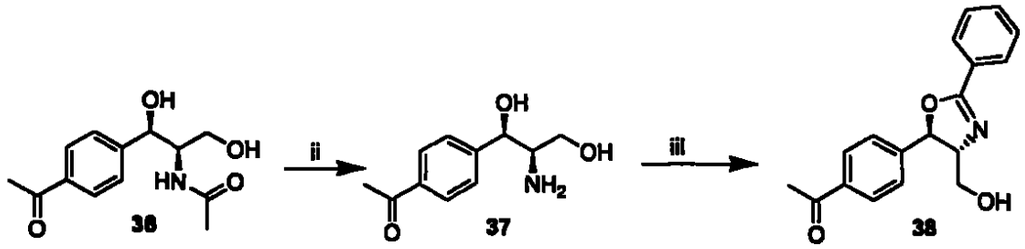
Compound **33** (1.00 g, 4.06 mmol) was dissolved in MeOH (50 mL) and cooled to 0 °C in an ice bath. Over 5 minutes, $NaBH_4$ (308 mg, 8.13 mmol) was added portion-wise. After bubbling ceased, TLC (5% MeOH in CH_2Cl_2) indicated that starting material remained so 100 mg additional $NaBH_4$ was added. After bubbling ceased, the mixture was quenched with glacial HOAc until a pH of approximately 7 was achieved. The mixture was concentrated under vacuum and partitioned between EtOAc and 1 N HCl. The EtOAc layer was washed with saturated aqueous $NaHCO_3$ (2 x 40 mL) and brine (40 mL) and dried over anhydrous Na_2SO_4. The product was isolated by silica gel chromatography (2% to 3% to 4% to10% MeOH in CH_2Cl_2) to give 128 mg of **34** as an oil. Additional product was obtained by evaporation of the aqueous layer followed by trituration with hot EtOAc (153 mg, 281 mg total). ^1H NMR (300 MHz, CD_3OD): δ 3.69 (br s, 3H), 5.50 (br s, 1H), 7.58 (d, 2H, J = 7.8), 7.79 (d, 2H, J = 7.8).

Example 24: Compound 35



Fluorination was performed using DAST as in the synthesis of compound **11**. ^1H NMR (300 MHz, CD_3OD): δ 3.89-3.97 (m, 1H), 4.49-4.66 (m, 2 H), 5.55 (d, 1H, J = 4.8), 7.59 (d, 2H, J = 8.1), 7.80 (d, 2H, J = 8.4).

Example 25: Compound 38

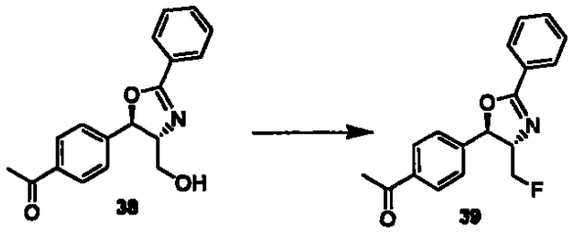


Compound **36** (2.12 g), was prepared by the procedure of von Strandtmann, *supra*. It was heated with 10% aqueous H_2SO_4 to 100 °C in a sealed tube and held for 4 hours. After cooling to room temperature, the mixture was basified with 1 M

NaOH and extracted three times with *n*-butanol. The *n*-butanol fractions were combined and dried over anhydrous Na_2SO_4. The solvent was evaporated to give 1.61 g of **37**.

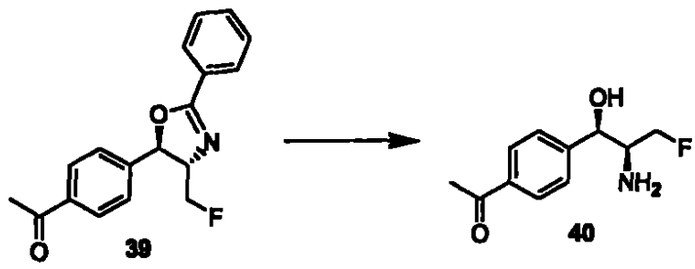
Compound **37** (1.61 g, 7.70 mmol) was dissolved in 1,2-dichloroethane (100 mL), along with ethyl benzimidate hydrochloride (1.42 g, 7.70 mmol), and Et_3N (1.06 mL, 7.70 mmol). The mixture was refluxed for 12 hours, cooled to room temperature and diluted with EtOAc. The solution was washed with saturated aqueous NH_4Cl (3 x) and saturated $NaHCO_3$ (1 x) and dried over anhydrous Na_2SO_4. The product precipitated from the EtOAc on cooling and addition of several drops of hexanes to give 0.62 g of **38** (2.1 mmol). ^1H NMR (300 MHz, CD_3OD): δ 2.60 (s, 3H), 3.79 (dd, 1H, *J* = 11.3, 5.9), 3.89 (dd, 1H, *J* = 11.3, 4.1), 4.14-4.20 (m, 1H), 5.70 (d, 1H, 6.3), 7.47-7.61 (m, 5H), 8.01-8.05 (m, 4H).

Example 26: Compound 39



Compound **38** (0.62 g, 2.1 mmol) was suspended in CH_2Cl_2 (15 mL) and cooled to –78 °C. DAST (0.42 mL, 3.1 mmol) was added by syringe and the solution stirred overnight, during which time it was allowed to come to room temperature as the cold bath warmed. The solvent was removed under vacuum and the product isolated by silica gel chromatography (1:4 EtOAc/hexanes) to give 0.17 g of **39**. ^1H NMR (300 MHz, CD_3OD): δ 2.60 (s, 3H), 4.30-4.40 (m, 1H), 4.63-4.67 (m, 1H), 4.79-4.81 (m, 1H), 5.74 (d, 1H, 6.9), 7.48-7.63 (m, 5H), 8.01-8.07 (m, 4H).

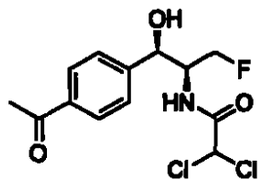
Example 27: Compound 40



39 40

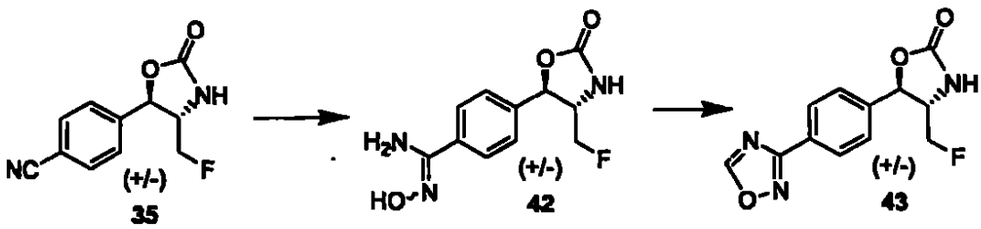
Compound **39** (0.17 g, 0.58 mmol) was suspended in 6 N aqueous HCl (5 mL) in a sealed tube. The mixture was heated to 100 °C and held for 12 hours. After cooling to room temperature, the mixture was basified with 3 N NaOH and the product extracted into CH_2Cl_2 (3 x). The combined CH_2Cl_2 fractions were dried over anhydrous Na_2SO_4, filtered and concentrated to give **40** (0.113 g, 0.541 mmol). ^1H NMR (300 MHz, CD_3OD): δ 2.60 (s, 3H), 3.04-3.15 (m, 1H), 4.09-4.48 (m, 3H), 4.71 (d, 1H, *J* = 6.0), 7.52 (d, 2H, *J* = 8.1), 7.90 (d, 2H, *J* = 8.1).

Example 28: Compound 41



Compound **40** was dichloroacetylated to give **41** in the same manner that **27** was converted to **29**. ^1H NMR (300 MHz, CD_3OD): δ 2.57 (s, 3H), 4.31-4.78 (m, 3H,), 5.03 (d, 1H, *J* = 3.3), 6.23 (s, 1H), 7.53 (d, 2H, *J* = 8.6), 7.95 (d, 2H, *J* = 8.6).

Example 29: Compound 43

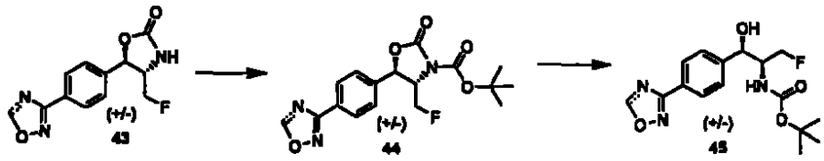


35 42 43

Compound **35** (100 mg, 0.454 mmol) was dissolved in 3 mL of EtOH and transferred to a 25 mL round-bottom flask. Hydroxylamine hydrochloride (38 mg, 0.545 mmol) was added followed by Et_3N (127 µL, 0.909 mmol). The mixture was stirred at reflux for 3 hours, at which point TLC indicated complete consumption of

starting material. The mixture was concentrated under vacuum to 232 mg of yellow oil, which was used without further purification. A portion of the oil (115 mg) was dissolved in 10 mL of triethylorthoformate. The mixture was stirred at 120 °C under N_2 for 2 hours and then at room temperature for 48 hours. TLC (10% MeOH in CH_2Cl_2) a major new spot. The triethyl orthoformate was removed under vacuum and the residue partitioned between EtOAc and 1 N aqueous NaOH. The organic layer was washed with 1 N aqueous NaOH (2 x) and brine (2 x). The EtOAc layer was dried over anhydrous Na_2SO_4, filtered, and evaporated, to give 84 mg of oil. The product was purified by chromatotron (1 mm silica plate, 60:40 to 50:50 to 40:60 hexanes/EtOAc). Another purification by chromatotron (2% MeOH in CH_2Cl_2) gave **43** (20 mg) as a white solid. ^1H NMR (300 MHz, CD_3OD): δ 3.93-4.04 (m, 1H), 4.49-4.67 (m, 2 H), 5.53 (d, 1H, *J* = 5.1), 7.59 (d, 2H, *J* = 8.3), 8.17 (d, 2H, *J* = 8.3), 9.28 (s, 1H).

Example 30: Compound 45

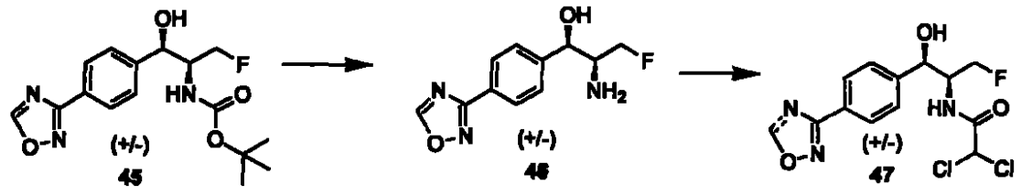


Compound **43** (20 mg, 0.076 mmol) was dissolved in CH_3CN (2 mL). Boc_2O (25 mg, 0.114 mmol) was added followed by a single crystal of DMAP (approximately 1 mg, 0.008 mmol). After 2 hours, TLC (1:1 hexanes/EtOAc) indicated that the reaction was complete. The CH_3CN was removed under vacuum and the resulting white solid dissolved in EtOAc, which was washed with 1 N aqueous HCl (2 x), followed by $NaHCO_3$ (2 x) then brine. The organic layer was dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum to give **44** as a white solid (28 mg).

Compound **44** (0.026 mg, 0.072 mmol) was dissolved in MeOH (2 mL) and Cs_2CO_3 (5 mg, 0.014 mmol) was added in a single portion. After 1 hour, TLC (1:1 hexanes/EtOAc) showed a single product. The solvent was removed under vacuum, EtOAc added and the mixture stirred rapidly at room temperature. Then, H_2O was added, followed by 0.5 M HCl dropwise until all solids dissolved. The organic phase was washed with 0.5 N HCl, followed by saturated aqueous $NaHCO_3$ (2 x) and brine.

The organic layer was then dried over anhydrous Na_2SO_4, filtered and concentrated to give a yellow oil, which was purified by chromatotron (1 mm silica plate, 4:1 hexanes/EtOAc to 1:1 hexanes/EtOAc) to give **45** (13 mg, 0.039 mmol). ^{1}H NMR (300 MHz, CD_3OD): δ 1.31 (s, 9H), 4.01-4.63 (m, 3 H), 4.94 (d, 1H, *J* = 2.7), 7.55 (d, 2H, *J* = 8.4), 8.05 (d, 2H, *J* = 8.4), 9.24 (s, 1H).

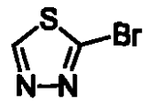
Example 31: Compound 47



Compound **45** was dissolved in 2.5 mL of 9:1 TFA/H_2O in a 10 mL flask. The mixture was stirred at room temperature for 30 minutes, at which time TLC indicated completion of the reaction. The mixture was concentrated under vacuum and partitioned with rapid stirring between 1 N aqueous NaOH and EtOAc. The EtOAc layer was washed with brine (1 x), dried over anhydrous Na_2SO_4, filtered and evaporated to give 11 mg of **46** as an oil .

To 6 mg (0.025 mmol) of **46** in a 10 mL round bottom flask was added MeOH (2 mL), followed by Et_3N (3.5 µL, 0.025 mmol) and methyl dichloroacetate (13.1 µL, 0.025 mmol). The mixture was refluxed for 4 hours at which time TLC indicated no reaction had occurred. An additional 26.2 µL of methyl dichloroacetate was added followed by 7 µL of Et_3N. The mixture was refluxed for 20 hours after which TLC indicated that the reaction was complete. The product was purified via chromatotron (1mm silica gel plate, 99:1 CH_2Cl_2/MeOH) to give 5 mg **47**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.72 (m, 3H), 5.03 (d, 1H, *J* = 2.7), 6.25 (s, 1H), 7.56 (d, 2H, *J* = 8.3), 8.05 (d, 2H, *J* = 8.3), 9.24 (s, 1H). LRMS (ESI$^-$) m/z: 346.0 (M-H$^+$ $C_{13}H_{11}Cl_2FN_3O_3$ requires 346.0).

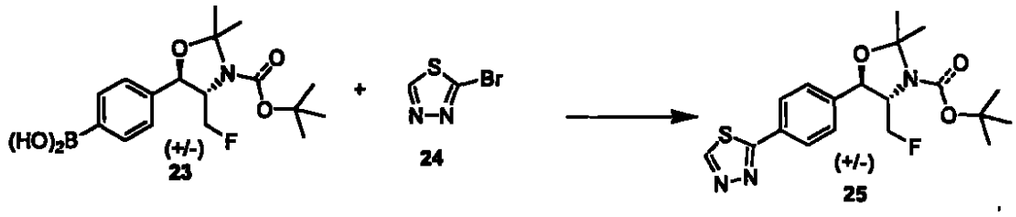
Example 32: Compound 24



24

In a 200 mL round-bottom flask, 2-amino-1,3,4-thiadiazole (1.00 g, 9.89 mmol) was added to 10 mL of 48% aqueous HBr. Water (10 mL) was added to give a yellow solution in which most solids dissolved. The mixture was cooled to 0 °C and CuBr (142 mg, 0.989 mmol) was added to give an opaque brown solution with some precipitate. $NaNO_2$ (682 mg, 9.89 mmol) was dissolved in 25 mL H_2O and added dropwise over 45 minutes to the thiadiazole mixture. The mixture became dark green and opaque with the first drops of $NaNO_2$ solution. Slowly, the solution became brownish yellow and brown gas evolved. The mixture was stirred for an additional 10 minutes at 0 °C from the time gas evolution began. The mixture was warmed to room temperature over 30 minutes. Saturated aqueous $NaHCO_3$ was added dropwise until bubbling ceased and the pH was 8.5. To this was added 50 mL of EtOAc and the biphasic mixture stirred rapidly. The mixture was filtered through a pad of Celite to remove solids and then the layers were separated. The organics were washed with brine (2 x). The aqueous layer was extracted with EtOAc and washed with brine (1 x). The combined organic layers were dried over Na_2SO_4. The remaining aqueous material was extracted once more by vigorous stirring with EtOAc (50 mL) for 12 hours. This EtOAc layer was washed with brine and combined with the EtOAc that was already drying over Na_2SO_4. The solution was filtered and evaporated under vacuum to give **24** as a tan solid (1.22 g, 7.36 mmol), which was used without further purification.

Example 33: Compound 25

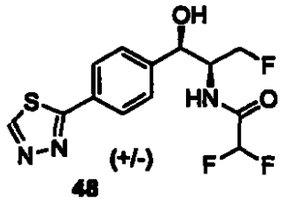


This is an example of Suzuki coupling method B. Compound **23** (19 mg, 0.0538 mmol) was dissolved in THF (1 mL), DMF (1 mL), and H_2O (0.5 mL). The mixture was stirred at room temperature until all material dissolved and then 2-bromo-1,3,4-thiadiazole (**24**, 5.0 mg, 0.027 mmol) was added. The solution was purged with N_2 for 5 minutes by means of a long needle. $PdCl_2(dppf)$ was added and then the solution was again purged with N_2 for 5 minutes. The mixture was then

stirred at 55 °C for 18 hours under N_2 and then at room temperature for an additional 30 hours. The resulting clear brown solution was concentrated under vacuum. Compound **25** (5.4 mg, 0.0137 mmol) was obtained as a yellow oil by chromatotron purification (silica gel, 1 mm plate, 1:9 EtOAc/hexanes to 1:4 EtOAc/hexanes).

It was found that the yield could be substantially improved by switching to coupling method C. Compounds **23** (0.330 g, 0.934 mmol) and **24** (0.385 g, 2.34 mmol) were dissolved in a mixture of toluene, *n*-butanol, and H_2O (8 mL:8 mL:2 mL) and Cs_2CO_3 (0.912 g, 2.80 mmol) was added. The mixture was purged with N_2 and $Pd(PPh_3)_4$ was added. The mixture was again purged with N_2 for 5 minutes and then stirred rapidly at 70 °C under N_2 for 12 hours. The mixture was concentrated under vacuum and the residue partitioned between H_2O (75 mL) and EtOAc (75 mL. The layers were separated and the EtOAc was washed with brine (2 x 50 mL). The EtOAc was dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum to give 0.720 g of yellow oil. Compound **25** (0.220 g) was obtained by silica gel chromatography, eluting with 5:1 to 2:1 to 1:1 hexanes/EtOAc. ^{1}H NMR (300 MHz, CD_3OD): δ 1.50 (s, 9H), 1.58 (s, 3H), 1.70 (s, 3H), 3.80-3.94 (m, 1H), 4.42-4.61 (m, 2H), 5.20, (d. 1H, *J* = 7.5), 7.64 (d, 2H, *J* = 8.3), 8.05 (d, 2H, *J* = 8.3), 9.45 (s, 1H).

Example 34: Compound 48

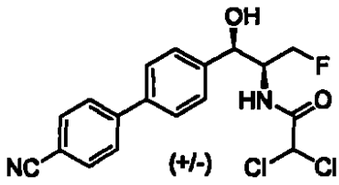


48

Compound **25** (5.4 mg, 0.014 mmol) was transferred to a 25 mL round-bottom flask and 2.5 mL of 9:1 TFA/H_2O (v/v) were added. The resulting bright yellow mixture was stirred at room temperature for 18 hours. The solvent was removed under vacuum and the residue dissolved three times in a mixture of MeOH and toluene, which was evaporated to near dryness each time. The product was recovered as an oil (4.9 mg, 0.013 mmol), which was used without further purification. ^{1}H NMR (300 MHz, CD_3OD): δ 3.59-3.70 (m, 1H), 4.30-4.71 (m, 2H), 4.92 (d, 1H, *J* = 8.1), 7.64 (d, 2H, *J* = 8.3), 8.08 (d, 2H, *J* = 8.3), 9.47 (s, 1H).

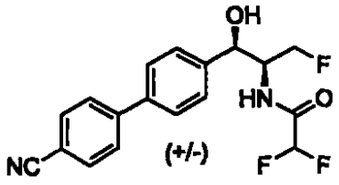
In a 10 mL round-bottom flask, the above product was dissolved in 2 mL of MeOH. To this was added Et_3N (9.7 μL, 0.070 mmol) followed by methyl difluoroacetate (3.0 μL, 0.035 mmol). The mixture was refluxed for 16 hours, at which point TLC indicated that some starting material still remained. An additional 3 drops of Et_3N was added followed by 2 drops of methyl difluoroacetate. After 3 hours, all starting material had been consumed. The mixture was cooled to room temperature and evaporated to dryness under vacuum. Compound **48** (4.1mg, 012 mmol) was obtained by chromatotron purification (silica gel, 1 mm plate). ^1H NMR (300 MHz, CD_3OD): δ 4.31-4.72 (m, 3H), 5.03 (d, 1H, J = 3.9), 5.98 (t, 1H, J = 53.9), 7.58 (d, 2H, J = 8.3), 7.99 (d, 2H, J = 8.3), 9.43 (s, 1H). LRMS (ESI$^-$) m/z: 330.1 (M-H^+ $C_{13}H_{11}F_3N_3O_2S$ requires 330.1).

Example 35: Compound 49



The biaryl intermediate was prepared from **18** and the appropriate boronic acid using Suzuki coupling method A. Removal of the Boc group was accomplished by brief treatment with 90/10 (v/v) TFA/H_2O. Dichloroacetylation was performed as in the synthesis of **29**. ^1H NMR (300 MHz, CD_3OD): δ 4.31-4.80 (m, 3H), 5.01 (d, 1H, J = 3.6), 6.26 (s, 1H), 7.52 (d, 2H, 8.3), 7.65 (d, 2H, J = 8.3), 7.79 (s. 4H). LRMS (ESI$^-$) m/z: 379.0 (M-H^+ $C_{18}H_{14}Cl_2FN_2O_2$ 379.0).

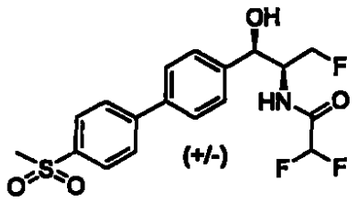
Example 36: Compound 50



The biaryl intermediate was prepared from **22** and the appropriate boronic acid using Suzuki coupling method A. ^1H NMR (300 MHz, CD_3OD): δ 4.27-4.68 (m, 3H), 4.99 (d, 1H, J = 4.5), 6.00 (t, 1H, J = 53.9), 7.52 (d, 2H, J = 8.1), 7.67 (d, 2H, J =

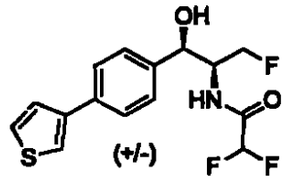
8.1), 7.76-7.82 (m, 2H). LRMS (ESI⁻) m/z: 347.1 ($M-H^+$ $C_{18}H_{14}F_3N_2O_2$ requires 347.1).

Example 37: Compound 51



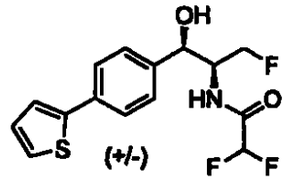
The biaryl intermediate was prepared from **22** and the appropriate boronic acid using Suzuki coupling method A. ^1H NMR (300 MHz, CD_3OD): δ 3.14 (s, 3H), 4.30-4.68 (m, 3H), 5.00 (d, 1H, J = 4.5), 6.00 (t, 1H, J = 54), 7.53 (d, 2H, J = 8.3), 7.70 (d, 2H, J = 8.3), 7.88 (d, 2H, J = 8.6), 8.00 (d, 2H, J = 8.6). LRMS (ESI⁻) m/z: 400.1 ($M-H^+$ $C_{18}H_{17}F_3NO_4S$ requires 400.1).

Example 38: Compound 52



The biaryl intermediate was prepared from **22** and the appropriate boronic acid using Suzuki coupling method B. ^1H NMR (300 MHz, CD_3OD): δ 4.26-4.64 (m, 3H), 4.93 (d, 1H, J = 4.8), 6.01 (t, 1H, J = 53.9), 7.40-7.45 (m, 4H), 7.60-7.66 (m, 3H). LRMS (ESI⁻) m/z: 328.1 ($M-H^+$ $C_{15}H_{13}F_3NO_2S$ requires 328.1).

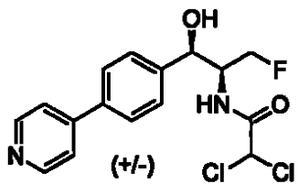
Example 39: Compound 53



The biaryl intermediate was prepared from **22** and the appropriate boronic acid using Suzuki coupling method B. ^1H NMR (300 MHz, CD_3OD): δ 4.25-4.65 (m, 3H), 4.92 (d, 1H, J = 4.5), 6.01 (t, 1H, J = 53.9), 7.06-7.08 (m, 1H), 7.36-7.42 (m,

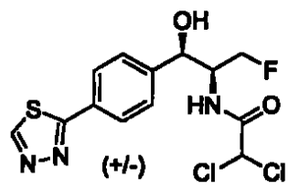
4H), 7.61 (d, 2H, J = 8.4). LRMS (ESI^+) m/z: 352.0 (calc. for $M+Na^+$ $C_{15}H_{14}F_3NNaO_2S$ 352.1).

Example 40: Compound 54



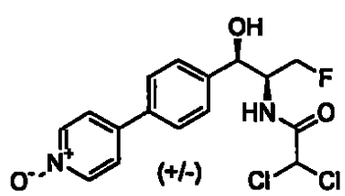
The biaryl intermediate was prepared from **18** and the boronic acid using Suzuki coupling method B. Removal of the Boc group was accomplished by brief treatment with 9/1 (v/v) TFA/H_2O. Dichloroacetylation was performed as in the synthesis of **29**. ^1H NMR (300 MHz, CD_3OD): δ 4.33-4.79 (m, 3H), 5.03 (d, 1H, J = 3.3), 6.26 (s, 1H), 7.55 (d, 2H, J = 8.4), 7.69-7.75 (m, 4H), 8.56 (d, 2H, J = 5.7). LRMS (ESI^+) m/z: 357.1 (calc. for $M+H^+$ $C_{16}H_{16}Cl_2FN_2O_2$ 357.1).

Example 41: Compound 55



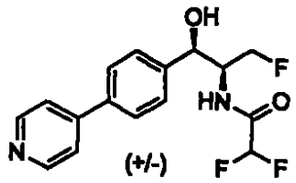
The biaryl intermediate was prepared from boronic acid **23** and compound **24** using Suzuki coupling method B. Removal of the protecting groups was accomplished by brief treatment with 9/1 (v/v) TFA/H_2O. Dichloroacetylation was performed as in the synthesis of **29**. ^1H NMR (300 MHz, CD_3OD): δ 4.34-4.76 (m, 3H), 5.04 (d, 1H, J = 3.0), 6.24 (s, 1H), 7.58 (d, 2H, J = 8.3), 7.97 (d, 2H, J = 8.3), 9.42 (s, 1H). LRMS (ESI^-) m/z: 362.0 (calc. for $M-H^+$ $C_{13}H_{11}Cl_2FN_3O_2S$ 362.0).

Example 42: Compound 56



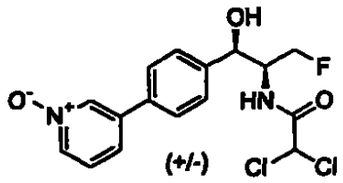
Compound **54** (29.4 mg, 0.0824 mmol) was dissolved in 5 mL of CH_2Cl_2. The mixture was cooled to 0 °C and m-chloroperbenzoic acid (*m*-CPBA, 38 mg, 0.16 mmol) was added with stirring. After 5 minutes, the mixture was warmed to room temperature and stirred for 12 hours. The mixture was concentrated under vacuum and the product purified by silica gel chromatography eluting successively with 2%, 3%, 4%, 6%, and10% MeOH in CH_2Cl_2. Compound **56** was obtained as a white solid (22.5 mg, 0.060 mmol). ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.75 (m, 3H), 5.03 (d, 1H, *J* = 3.3), 6.25 (s, 1H), 7.56 (d, 2H, *J* = 8.4), 7.74 (d, 2H, *J* = 8.4), 7.84 (d, 2H, *J* = 7.2), 8.34 (d, 2H, *J* = 7.2). LRMS (ESI$^-$) m/z: 371.0 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_3$ 371.0).

Example 43: Compound 57



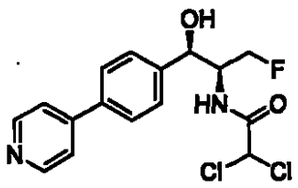
The biaryl intermediate was prepared from **18** and the appropriate boronic acid using Suzuki coupling method B. Removal of the protecting group was accomplished by brief treatment with 9/1 (v/v) TFA/H_2O. Difluoroacetylation was performed as in the synthesis of **48**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.28-4.68 (m, 3H), 5.00 (d, 1H, *J* = 4.2), 5.99 (t, 1H, *J* = 53.9), 7.55 (d, 2H, *J* = 8.1), 7.70-7.77 (m, 4H), 7.84 (d, 2H, *J* = 7.2), 8.56 (d, 2H, *J* = 6.3). LRMS (ESI$^+$) m/z: 325.2 (calc. for M+H$^+$ $C_{16}H_{16}F_3N_2O_2$ 325.1).

Example 44: Compound 58



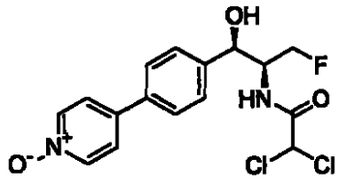
Compound **58** was prepared from **68** as **56** was from **54**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.30-4.75 (m, 3H), 5.02 (d, 1H, *J* = 3.3), 6.25 (s, 1H), 7.55-7.67 (m, 5H), 7.87 (d, 1H, *J* = 8.1), 8.30 (d, 1H, *J* = 7.2), 8.58 (br s, 1H). LRMS (ESI$^-$) m/z: 371.0 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_3$ 371.0).

Example 45: Compound 59



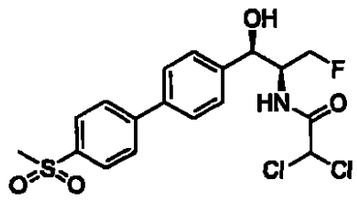
The biaryl intermediate was prepared from **11** and the appropriate boronic acid using Suzuki coupling method A. The phenyloxazoline protecting group was removed and the dichloroacetate group introduced as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.30-4.75 (m, 3H), 5.02 (d, 1H, *J* = 3.6), 6.26 (s, 1H), 7.54-8.70 (m, 8H). LRMS (ESI$^-$) m/z: 354.8 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_2$ 355.0).

Example 46: Compound 60



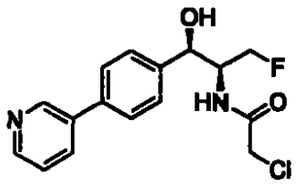
Compound **60** was prepared from compound **59** in the same manner that other pyridine N-oxides in these examples were formed from the corresponding pyridine. ^{1}H NMR (300 MHz, CD_3OD): δ 4.33-4.78 (m, 3H), 5.03 (d, 1H, *J* = 3.3), 6.25 (s, 1H), 7.57 (d, 2H, *J* = 8.4), 7.75 (d, 2H, *J* = 8.4), 7.87 (d, 2H, *J* = 7.5), 8.38 (d, 2H, *J* = 7.5). LRMS (ESI$^-$) m/z: 370.8 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_3$ 371.0).

Example 47: Compound 61



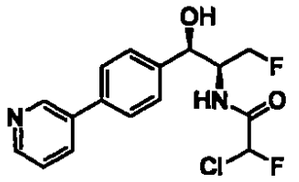
The biaryl intermediate was prepared from **11** and the appropriate boronic acid using Suzuki coupling method A. The phenyl oxazoline protecting group was cleaved and dichloroacetate introduced as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 3.14 (s, 3H), 4.36-4.75 (m, 3H), 5.02 (d, 1H, *J* = 3.6), 6.27 (s, 1H), 7.54 (d, 2H, *J* = 8.3), 7.68 (d, 2H, *J* = 8.3), 7.87 (d, 2H, *J* = 8.7), 8.01 (d, 2H, *J* = 8.7). LRMS (ESI$^-$) m/z: 431.8 (calc. for M-H$^+$ $C_{18}H_{17}Cl_2FNO_4S$ 432.0).

Example 48: Compound 62



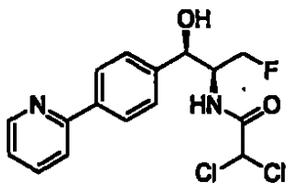
The free amine intermediate was prepared as in the synthesis of **28**. The nitrogen was monochloroacetylated using chloroacetyl chloride. ^{1}H NMR (300 MHz, CD_3OD): δ 4.00-4.03 (m, 2H), 4.27-4.70 (m, 3H), 5.00 (d, 1H, *J* = 3.9), 7.49-7.55 (m, 3H), 7.66 (d, 2H, *J* = 8.4), 8.09 (d app t, 1H, *J* = 7.8, 2.4), 8.50 (dd, 1H, *J* = 4.8, 1.5), 8.78 (br d, 1H, *J* = 2.4). LRMS (ESI$^+$) m/z: 321.0 (calc. for M+H$^+$ $C_{16}H_{15}ClFN_2O_2$ 357.1).

Example 49: Compound 63



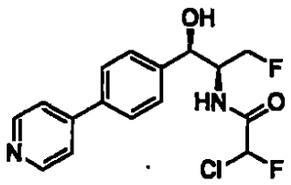
Prepared in the same manner as **28** using ethyl chlorofluoroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 4.31-4.70 (m, 3H), 5.01 (d, 1H, *J* = 3.9), major diastereomer 6.49 (d, 1H, *J* = 49.8), minor diastereomer 6.51 (d, 1H, *J* = 49.8), 7.49-7.56 (m, 3H), 7.63-7.67 (m, 2H), 8.07-8.11 (m, 1H), 8.49-8.51 (m, 1H), 8.78 (br d, 1H, *J* = 1.5). LRMS (ESI) m/z: 338.9 (calc. for M-H$^+$ $C_{16}H_{14}ClF_2N_2O_2$ 339.1).

Example 50: Compound 64



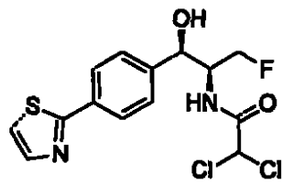
The biaryl intermediate was prepared from **12** and 2-bromopyridine using Suzuki coupling method A. ^{1}H NMR (300 MHz, CD_3OD): δ 4.28-4.74 (m, 3H), 5.02 (d, 1H, *J* = 3.6), 6.27 (s, 1H), 7.33-7.37 (m, 1H), 7.53 (d, 2H, *J* = 8.1), 7.18-7.93 (m, 4H), 8.58-8.60 (m, 1H). LRMS (ESI$^-$) m/z: 354.8 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_2$ 355.0)

Example 51: Compound 65



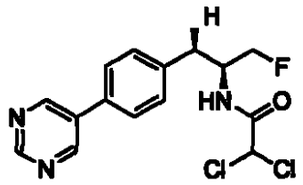
Compound **65** was prepared in the same manner as **59** using ethyl chlorofluoroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.70 (m, 3H), 5.02 (d, 1H, *J* = 3.6), major diastereomer 6.48 (d, 1H, *J* = 49.8), minor diastereomer 6.50 (d, 1H, *J* = 50.1), 7.55-7.58 (m, 2H), 7.73-7.78 (m, 4H) 8.57 (br d, 2H, *J* = 5.7). LRMS (ESI$^-$) m/z: 338.8 (calc. for M-H$^+$ $C_{16}H_{14}ClF_2N_2O_2$ 339.1).

Example 52: Compound 66



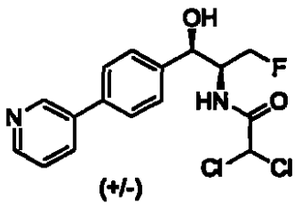
The biaryl intermediate was prepared from **12** and the appropriate bromide using Suzuki coupling method A. Removal of the protecting group and dichloroacetylation was accomplished as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.34-4.72 (m, 3H), 5.01 (d, 1H, *J* = 2.4), 6.25 (s, 1H), 7.52 (d, 2H, *J* = 6.3), 7.57 (d, 1H, *J* = 2.6) 7.84 (d, 1H, *J* = 2.6), 7.90 (d, 2H, *J* = 6.3). LRMS (ESI$^-$) m/z: 360.7 (calc. for M-H$^+$ $C_{14}H_{12}Cl_2FN_2O_2S$ 361.0).

Example 53: Compound 67



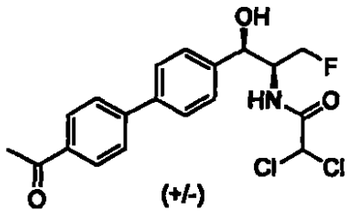
The biaryl intermediate was prepared from **12** and the appropriate bromide using Suzuki coupling method A. Removal of the protecting group and dichloroacetylation was accomplished as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.75 (m, 3H), 5.04 (d, 1H, *J* = 3.6), 6.26 (s, 1H), 7.59 (d, 2H, *J* = 8.3), 7.70 (d, 2H, *J* = 8.3) 9.06 (s, 2H,), 9.13 (s, 1H). LRMS (ESI$^+$) m/z: 357.8 (calc. for M+H$^+$ $C_{15}H_{15}Cl_2FN_3O_2$ 358.0).

Example 54: Compound 68



The biaryl intermediate was prepared from bromide **11** and the appropriate boronic acid using Suzuki coupling method A. Removal of the protecting group and dichloroacetylation was accomplished as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.31-4.74 (m, 3H), 5.02 (d, 1H, *J* = 3.6), 6.27 (s, 1H), 7.48-7.51 (m, 1H), 7.54 (d, 2H, *J* = 8.3), 7.64 (d, 2H, *J* = 8.3) 8.08 (ddd,1H, *J* = 8.0, 2.4, 1.8), 8.50 (dd, 1H, 4.8, 1.5), 8.77 (dd, 1H, *J* = 2.4, 0.9). LRMS (ESI$^-$) m/z: 354.8 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_2$ 355.0).

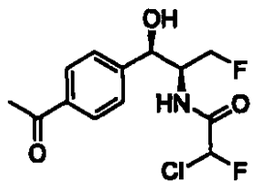
Example 55: Compound 69



The biaryl intermediate was prepared from bromide **11** and the appropriate boronic acid using Suzuki coupling method A. Removal of the protecting group and

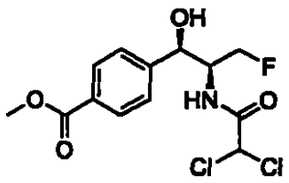
dichloroacetylation was performed as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 2.63 (s, 3H), 4.31-4.74 (m, 3H), 5.01 (d, 1H, *J* = 3.9), 6.27 (s, 1H), 7.51 (d, 2H, *J* = 8.3), 7.67 (d, 2H, *J* = 8.3), 7.75 (d, 2H, *J* = 8.6) 8.06 (d, 2H, *J* = 8.6).

Example 56: Compound 71



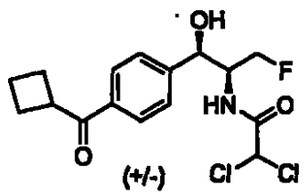
This compound was prepared in analogous fashion to **41** using ethyl chlorofluoroacetate in place of methyl dichloroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 2.58 (s, 3H), 4.31-4.71 (m, 3H), 5.03 (br d, 1H, *J* = 3.9), major diastereomer 6.46 (d, 1H, *J* = 49.8), minor diastereomer (d, 1H, *J* = 50.1), major diastereomer 7.53 (d, 2H, *J* = 8.1), minor diastereomer 7.51 (d, 2H, *J* = 8.1), 7.94-7.96 (m, 2H). LRMS (ESI$^-$) m/z: 304.1 (calc. for M-H$^+$ $C_{13}H_{13}ClF_2NO_3$ 304.1).

Example 57: Compound 72



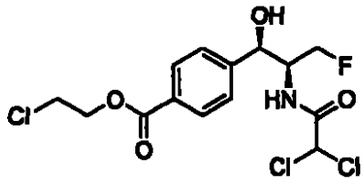
Initially, the *p*-cyano analog of compound **11** had been prepared by methods analogous to those used to prepare **11** itself. Attempts to remove the phenyloxazoline protecting group (as described in the synthesis of **29**) led to some of the desired deprotected nitrile intermediate. However, much of the mass recovered was the deprotected *p*-carboxylic acid corresponding to acidic hydrolysis of the nitrile functional group. This material was dichloroacetylated using the same procedure as that employed in the synthesis of **29** and the product was converted to its methyl ester with CH_2N_2. ^{1}H NMR (300 MHz, CD_3OD): δ 3.83 (s, 3H), 4.25-4.66 (m, 3H), 4.98 (d, 1H, *J* = 3), 6.17 (s, 1H), 7.46 (d, 2H, *J* = 8.3), 7.91 (d, 2H, *J* = 8.3).

Example 58: Compound 73



Compound **87** (36.5 mg, 0.0873 mmol) was dissolved in THF (5 mL anhydrous) under N_2. After addition of anhydrous powdered K_2CO_3 (24 mg, 0.175 mmol), the mixture was purged with N_2. Et_3N (31 mL, 0.218 mmol) and cyclobutanecarbonyl chloride (13 mL, 0.113 mmol) were added and the mixture purged gently with N_2 for another 5 min. Pd_2dba_3 was added and the mixture purged again with N_2. The mixture was then stirred under N_2 for 3 hours after which it was diluted with EtOAc and H_2O and filtered through a cotton plug. The EtOAc layer was washed with 1N aqueous HCl (x 2), brine (x 2), and dried over anhydrous Na_2SO_4. The mixture was filtered and the solvent removed to give 16.9 mg, 0.0501 mmol of material that was purified by chromatotron (1 mm plate, eluting with 4:1 hexanes/EtOAc). The protected intermediate obtained was subjected to phenyloxazoline cleavage and dichloroacetylation as in the synthesis of **29** to give **73**. ^1H NMR (300 MHz, CD_3OD): δ 1.73-1.85 (m, 1H), 1.99-2.11 (m, 1H), 2.20-2.27 (m, 4H), 3.97-4.09 (m, 1H), 4.23-4.67 (m, 3H), 4.95 (d, 1H, J = 3.3), 6.16 (s, 1H), 7.45 (d, 2H, J = 8.4), 7.81 (d, 2H, J = 8.4). LRMS (ESI$^-$) m/z: 360.0 (calc. for M-H$^+$ $C_{16}H_{17}Cl_2FNO_3$ 360.0).

Example 59: Compound 74

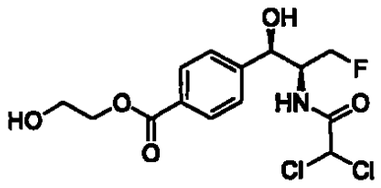


The carboxylic acid intermediate from the synthesis of **72** was dichloroacetylated as in the synthesis of **29**. The resulting intermediate (95.8 mg, 0.297 mmol) was dissolved in 2 mL of MeOH, several drops of H_2O were added and then a 20% (w/v) aqueous Cs_2CO_3 solution was added dropwise until the solution reached pH 7. The mixture was concentrated under vacuum. One drop of Et_3N and 10 mL of 2-chloroethanol were added and the mixture stirred at 135 °C for 2 hours. Residual chloroethanol was removed under vacuum and the residue

78
79

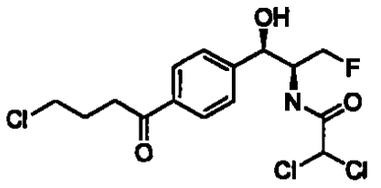
chromatographed. Compound **74** (13.3 mg) was the major product and **75** (9.2 mg) the minor product. ^{1}H NMR (300 MHz, CD_3OD): δ 3.78-3.82 (m, 2H), 4.23-4.68 (m, 5H), 4.97 (d, 1H, *J* = 3.0), 6.17 (s, 1H), 7.46 (d, 2H, *J* = 8.3), 7.93 (d, 2H, *J* = 8.3). LRMS (ESI$^+$) m/z: 407.9 (calc. for M+Na$^+$ $C_{14}H_{15}Cl_3FNNaO_4$ 408.0).

Example 60: Compound 75



^{1}H NMR (300 MHz, CD_3OD): δ 3.83-3.39 (m, 2H), 4.31-4.74 (m, 5H), 5.02 (d, 1H, *J* = 3.3), 6.23 (s, 1H), 7.52 (d, 2H, *J* = 8.4), 8.02 (d, 2H, *J* = 8.4). LRMS (ESI$^-$) m/z: 366.0 (calc. for M-H$^+$ $C_{14}H_{15}Cl_2FNO_5$ 366.0).

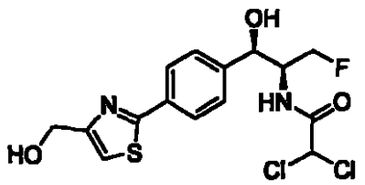
Example 61: Compound 76



(+/-)

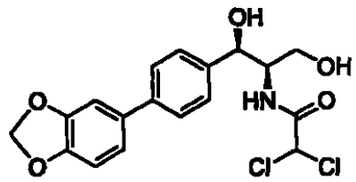
The cyclopropyl analog of intermediate **89** was prepared by a Stille coupling analogous to that used to synthesize **89**. Acidic cleavage of the phenyloxazoline group resulted in the HCl-mediated opening of the cyclopropyl ring. This material was deprotected and dichloroacetylated to give **76**. ^{1}H NMR (300 MHz, CD_3OD): δ 2.10-2.19 (m, 2H), 3.19 (t, 2H, *J* = 7.1), 3.66 (t, 2H, *J* = 6.6), 4.31-4.74 (m, 3H), 5.03 (d, 1H, *J* = 3.3), 6.23 (s, 1H), 7.53 (d, 2H, *J* = 8.3), 7.96 (d, *J* = 8.3). LRMS (ESI$^-$) m/z: 381.9 (calc. for M-H$^+$ $C_{15}H_{18}Cl_3FNO_3$ 382.0).

Example 62: Compound 77



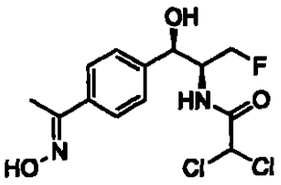
The thiazole ring of this analog was formed by conversion of the protected nitrile intermediate to the corresponding thioamide by reaction with H_2S in pyridine. The thioamide was then reacted with the appropriate protected α-halocarbonyl compound to form the substituted thiazole shown. Deprotection and dichloroacetylation was performed as for other compounds of this invention. ^1H NMR (300 MHz, CD_3OD): δ 4.32-4.75 (m, 5H), 5.01 (d, 1H, J = 3.3), 6.27 (s, 1H), 7.38 (s, 1H), 7.51 (d, 2H, J = 8.3), 7.91 (d, 2H, J = 8.3). LRMS (ESI$^-$) m/z: 390.9 (calc. for M-H$^+$ $C_{15}H_{14}Cl_2FN_2O_3S$ 391.0).

Example 63: Compound 79



This compound was formed by Suzuki coupling method A from an intermediate in which the primary hydroxyl group was not converted to a fluoride. Deprotection and dichloroacetylation were performed as with other compounds of this invention. ^1H NMR (300 MHz, CD_3OD): δ 3.55 (dd, 1H, J = 11.0, 5.9), 3.77 (dd, 1H, J = 11.0, 6.5), 4.07-4.11 (m, 1H), 5.02 (d, 1H, J = 3.6), 5.96 (s, 2H), 6.29 (s, 1H), 6.86 (d, 1H, J = 8.4), 7.05-7.07 (m, 2H), 7.40-7.50 (m, 4H). LRMS (ESI$^-$) m/z: 395.9 (calc. for M-H$^+$ $C_{18}H_{16}Cl_2NO_5$ 396.0).

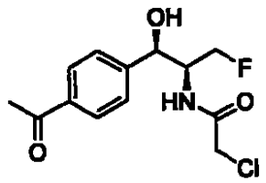
Example 64: Compound 80



Compound 41 (11.5 mg, 0.0346 mmol) and hydroxylamine hydrochloride (2.8 mg, 0.0415 mmol) were stirred in EtOH (3 mL) for 12 hours and then refluxed for an

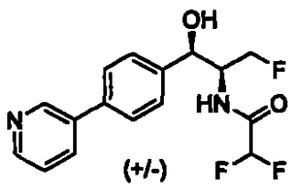
additional 12 hours. The solvent was removed under vacuum and the product isolated as a single isomer (it was not determined whether the isomer was the *cis* or *trans* oxime). ^{1}H NMR (300 MHz, CD_3OD): δ 2.21 (s, 3H), 4.26-4.68 (m, 3H,), 4.95 (d, 1H, *J* = 3.9), 6.27 (s, 1H), 7.39 (d, 2H, *J* = 8.3), 7.61 (d, 2H, *J* = 8.3).

Example 65: Compound 81



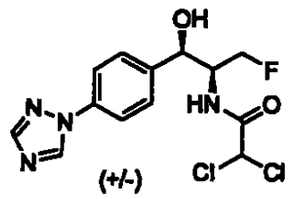
Free amine **37** (4.7 mg, 0.0224 mmol) was dissolved in MeOH (1 mL) and the mixture cooled to 0 °C. Chloroacetic anhydride (5 drops) and triethylamine (5 drops) were added and the mixture warmed to room temperature and stirred for 2 hours. The solvent was removed under vacuum and the product purified by silica gel chromatography (2:3 EtOAc/hexanes) to give 3.3 mg (0.011 mmol) of product. ^{1}H NMR (300 MHz, CD_3OD): δ 2.59 (s, 3H), 3.92-4.03 (m, 2H,), 4.29-4.68 (m, 3H), 5.02 (d, 1H, *J* = 3.3), 7.53 (d, 2H, *J* = 8.4), 7.96 (d, 2H, *J* = 8.4).

Example 66: Compound 82



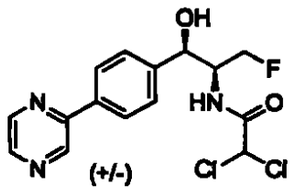
The biaryl intermediate was formed by Suzuki coupling method A using **22** and the appropriate boronic acid. ^{1}H NMR (300 MHz, CD_3OD): δ 4.28-4.68 (m, 3H), 5.00 (d, 1H, *J* = 4.2), 6.01 (t, 1H, *J* = 54), 7.49-7.60 (m, 3H), 7.66 (d, 2H, *J* = 8.4), 8.07-8.11 (m, 1H), 8.50 (dd, 1H, *J* = 4.8, 1.5), 8.79-8.79 (m, 1H). LRMS (ESI$^-$) m/z: 322.9 (M-H$^+$ $C_{16}H_{14}F_3N_2O_2$ requires 323.1).

Example 67: Compound 84



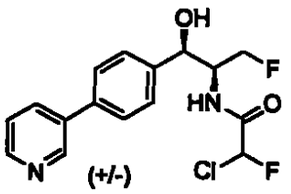
This compound was prepared by procedures analogous to those described in Schemes 2 and 3. The 4-(1,2,4-triazol-1-yl)phenylserine analog of **6** was synthesized by the same methods used to make **6**. The required 4-(1,2,4-triazol-1-yl)benzaldehyde was prepared as described in the literature (Tanaka, A., et al., *J. Med. Chem.* **1998**, *41*, 2390-2410). ^{1}H NMR (300 MHz, CD_3OD): δ 4.33-4.76 (m, 3H), 5.04 (d, 1H, *J* = 3.3), 6.26 (s, 1H), 7.59 (d, 2H, *J* = 8.6), 7.78 (d, 2H, *J* = 8.6), 8.14 (s, 1H), 9.05 (s, 1H). LRMS (ESI$^-$) m/z: 345.0 (calc. for M-H$^+$ $C_{13}H_{12}Cl_2FN_4O_2$ 345.0).

Example 68: Compound 85



The biaryl intermediate was prepared from boronic acid **12** and 2-iodopyrazine. Deprotection and dichloroacetylation were accomplished as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.75 (m, 3H), 5.04 (d, 1H, *J* = 3.3), 6.27 (s, 1H), 7.57 (d, 2H, *J* = 8.4), 8.05 (d, 2H, *J* = 8.4), 8.51 (d, 1H, *J* = 2.7), 8.65-8.66 (m, 1H), 9.08 (d, 1H, *J* = 1.8). LRMS (ESI$^-$) m/z: 355.8 (calc. for M-H$^+$ $C_{15}H_{13}Cl_2FN_3O_2$ 356.0).

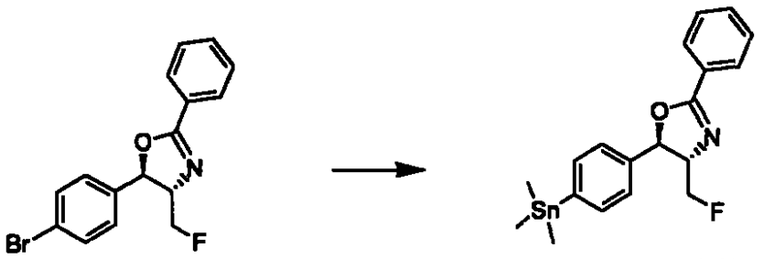
Example 69: Compound 86



Same procedure as that for the synthesis of **68**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.29-4.73 (m, 3H), 5.01 (br d, 1H, *J* = 3.9), major diastereomer 6.49 (d, 1H, *J* =

49.8), minor diastereomer 6.51 (d, 1H, J = 49.8), 7.48-7.56 (m, 3H), 7.64-7.67 (m, 2H), 8.06-8.10 (m, 1H), 8.49-8.51 (m, 1H), 8.78-8.79 (m, 1H). LRMS (ESI$^-$) m/z: 338.9 (calc. for M-H$^+$ $C_{16}H_{14}ClF_2N_2O_2$ 339.1).

Example 70: Compound 87

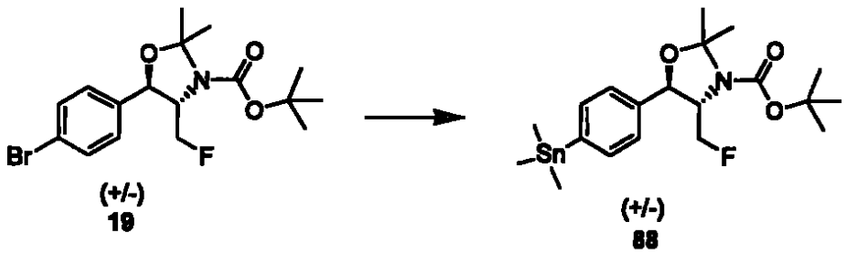


enantiopure or (+/-)
11

enantiopure or (+/-)
87

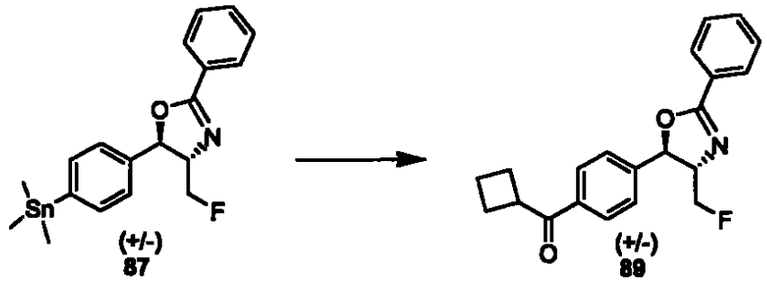
Compound **11** (0.496 g, 1.48 mmol) was dissolved in anhydrous benzene (15 mL) under N_2. The flask was fitted with a reflux condenser and a long needle was dropped through the condenser and the solution purged with a gentle stream of dry N_2 for 5 minutes. Hexamethyldiltin (0.58 g, 1.78 mmol) was added and the mixture purged for another 5 minutes. $Pd(PPh_3)_4$ (0.171 g, 0.148 mmol) was added and purging with N_2 continued. After 5 minutes, the needle was removed and a N_2 inlet was placed at the top of the reflux condenser. The mixture was refluxed for approximately 2 hours under N_2. Over the course of the reaction, the mixture turned from orange to yellow to black. The solvent was removed under vacuum and the product purified by chromatotron (silica gel, 1 mm plate) to give 440 mg of product as a clear oil. ^{1}H NMR (300 MHz, CD_3OD): δ 0.27 (s, 9H - this resonance contains significant satellite peaks, which are due to NMR-active isotopes of tin), 4.27-4.41 (m, 1H), 4.59-4.76 (m, 2H), 5.60 (d, 1H, J = 6.9), 7.33 (d, 2H, J = 8.1), 7.45-7.31 (m, 5H), 7.99-8.02 (m, 2H).

Example 71: Compound 88



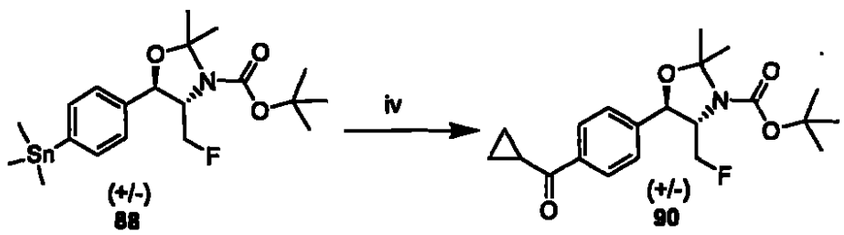
This compound was prepared in the same manner as **87**. ^{1}H NMR (300 MHz, CD_3OD): δ 0.027 (s, 9H, with significant satellite peaks caused by NMR-active tin isotopes), 1.49 (s, 9H), 1.55 (s, 3H), 1.67 (s, 3H), 3.73-3.83 (m, 1H), 4.30-4.49 (m, 2H), 5.06 (d, 2H, J = 7.5), 7.36-7.51 (m, 4H).

Example 72: Compound 89



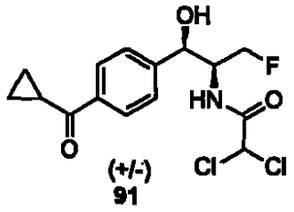
Compound **87** (36.5 mg, 0.0873 mmol) was dissolved in anhydrous THF under N_2. With stirring, K_2CO_3 (24mg, 0.16 mmol) was added. The mixture was purged with a gentle stream of N_2 for 5 minutes. Et_3N (31 µL, 0.22 mmol) and cyclobutanecarbonyl chloride (13 µL, 0.11 mmol) were added and the mixture purged with N_2 for 5 minutes. Pd_2dba_3 was added, the mixture purged for 5 min and then stirred under N_2 for 3 hours. The mixture was diluted with EtOAc (25 mL) and H_2O (25 mL) and filtered through a cotton plug to remove solids. The EtOAc was washed with 1 N HCl (2 x) and brine (2 x), dried over Na_2SO_4 and concentrated under vacuum. The product was purified by chromatotron (silica plate, 1 mm, 1:4 EtOAc/hexanes) to give 16:9 mg (0.050 mmol) of product. LRMS (ESI$^+$) m/z: 338.2 ($M+H^+$ $C_{21}H_{21}FNO_2$ requires 338.2).

Example 73: Compound 90



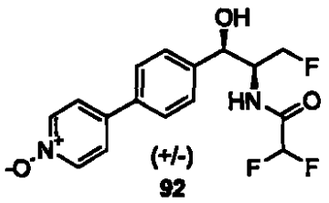
Same procedure as in the synthesis of **89**. ^{1}H NMR (300 MHz, CD_3OD): δ 0.027 (s, 9H, with significant satellite peaks caused by NMR-active tin isotopes), 1.49 (s, 9H), 1.55 (s, 3H), 1.67 (s, 3H), 3.73-3.83 (m, 1H), 4.30-4.49 (m, 2H), 5.06 (d, 2H, *J* = 7.5), 7.36-7.51 (m, 4H).

Example 74: Compound 91



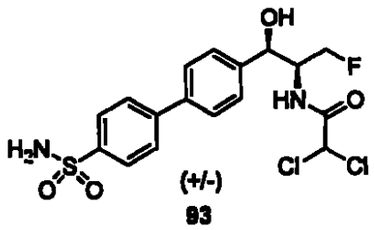
Compound **91** was deprotected by brief treatment of **90** with 9/1 TFA/H_2O. The dichloroacetyl group was introduced as in the synthesis of **29**. LRMS (ESI$^-$) m/z: 345.8 (calc. for M-H$^+$ $C_{15}H_{18}Cl_2FNO_3$ 346.0).

Example 75: Compound 92



The same procedure as that used in the synthesis of **56** was employed. ^{1}H NMR (300 MHz, CD_3OD): δ 4.28-4.74 (m, 3H), 5.01 (d, 1H, *J* = 3.9), 5.98 (t, 1H, *J* = 53.8), 7.55 (d, 2H, *J* = 8.4). 7.76 (d, 2H, *J* = 8.4), 7.86 (d, 2H, *J* = 7.1), 8.35 (d, 2H, *J* = 7.1).

Example 76: Compound 93

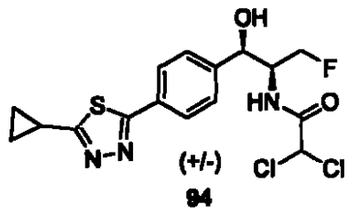


(+/-)

93

The biaryl intermediate was prepared by reaction of **23** and the appropriate bromide using Suzuki coupling A. Deprotection and dichloroacetylation were carried out as described above in the synthesis of **29**. LRMS (ESI$^-$) m/z: 433.0 ($M-H^+$ $C_{17}H_{16}Cl_2FN_2O_4S$ requires 433.0).

Example 77: Compound 94

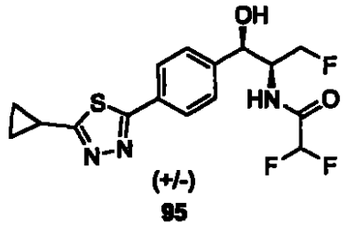


(+/-)

94

The biaryl intermediate was prepared by reaction of **23** with 2-amino-4-cyclopropyl-1,3,4-thiadiazole (prepared as in the synthesis of **24**) using Suzuki coupling B. Deprotection was accomplished by brief treatment of the protected biaryl intermediate with 90/10 TFA/H_2O and dichloroacetylation was performed as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 1.12-1.31 (m, 4H), 2.46-2.52 (m, 1H), 4.30-4.75 (m, 3H), 5.02 (d, 1H, J = 3.3), 6.24 (s, 1H), 7.54 (d, 2H, J = 8.4), 7.87 (d, 2H, J = 8.4).

Example 78: Compound 95



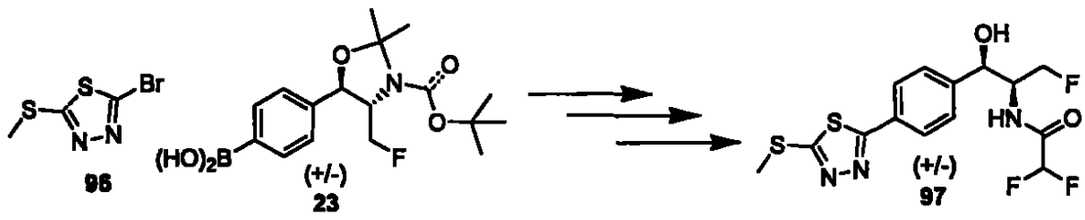
(+/-)

95

Same procedure as in the synthesis of **94** using methyl difluoroacetate in place of methyl dichloroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 1.13-1.15 (m, 2H), 1.28-1.32 (m, 2H), 2.47-2.52 (m, 1H), 4.29-4.70 (m, 3H), 5.01 (d, 1H, J = 4.2); 5.98

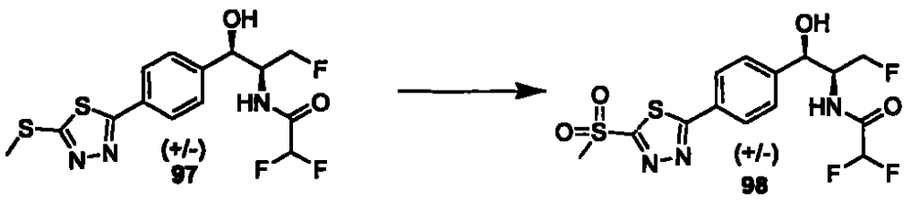
(t, 1H, J = 53.7), 7.54 (d, 2H, J = 8.7), 7.88 (d, 2H, J = 8.7). LRMS (ESI$^-$) m/z: 370.0 (M-H$^+$ $C_{16}H_{15}F_3N_3O_2S$ requires 370.1).

Example 79: Compound 97



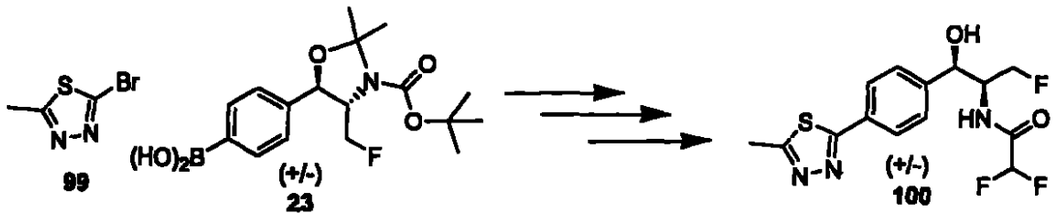
Compound **96** was prepared in analogous fashion to **24**. Compound **97** was prepared in analogous fashion to **48**. Suzuki coupling method B was used, followed by deprotection and difluoroacetylation. LRMS (ESI$^-$) m/z: 376.0 (M-H$^+$ $C_{14}H_{13}F_3N_3O_2S_2$ requires 376.0)

Example 80: Compound 98



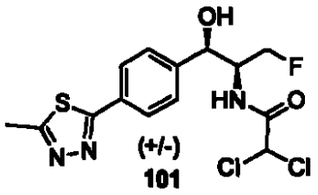
Compound **97** (11.1 mg or 0.0294 mmol) was dissolved in 5 mL of CH_2Cl_2, with stirring at room temperature. To this was added *m*-CPBA (36 mg, 0.147 mmol). The mixture was stirred for 23 hours. Attempted Chromatotron (1 mm plate) purification, eluting with 80:20 then 70:30 then 50:50 hexanes/EtOAc, failed to remove *m*-CPBA-related materials. Thus, material from the Chromotron was subjected to C-8 reversed-phase HPLC, which gave 8.8 mg (.021 mmol) of **98**. ^{1}H NMR (300 MHz, CD_3OD): δ 3.52 (s, 3H), 4.34-4.72 (m, 3H), 5.05 (d, 1H, J = 3.6), 5.97 (t, 1H, J = 53.9), 7.61 (d, 2H, J = 7.61), 8.05 (d, 2H, J = 8.1).

Example 81: Compound 100



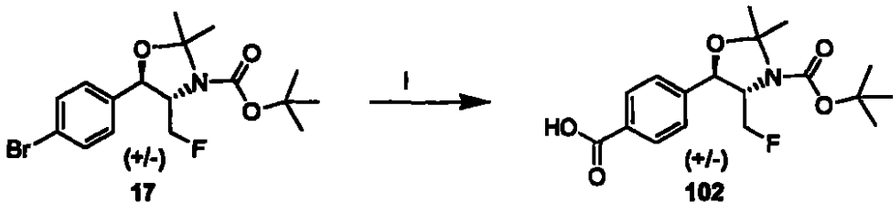
Compound **99** was prepared in analogous fashion to **24** and **100** was prepared in analagous fashion to **48**. Suzuki coupling method B was used followed by deprotection and difluoroacetylation. ^{1}H NMR (300 MHz, CD_3OD): δ 2.80 (s, 3H), 4.30-4.70 (m, 3H), 5.01 (d, 1H, *J* = 3.6), 5.98 (t, 1H, J = 53.9), 7.55 (d, 2H, *J* = 8.4), 7.91 (d, 2H, *J* = 8.3).

Example 82: Compound 101



Compound 101 was prepared using the same procedure as in the synthesis of **100** but with methyldichloroacetate in place of methyldifluoroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 2.79 (s, 3H), 4.31-4.75 (m, 3H), 5.03 (d, 1H, *J* = 3.0), 6.24 (s, 1H), 7.55 (d, 2H, *J* = 8.4), 7.89 (d, 2H, *J* = 8.4).

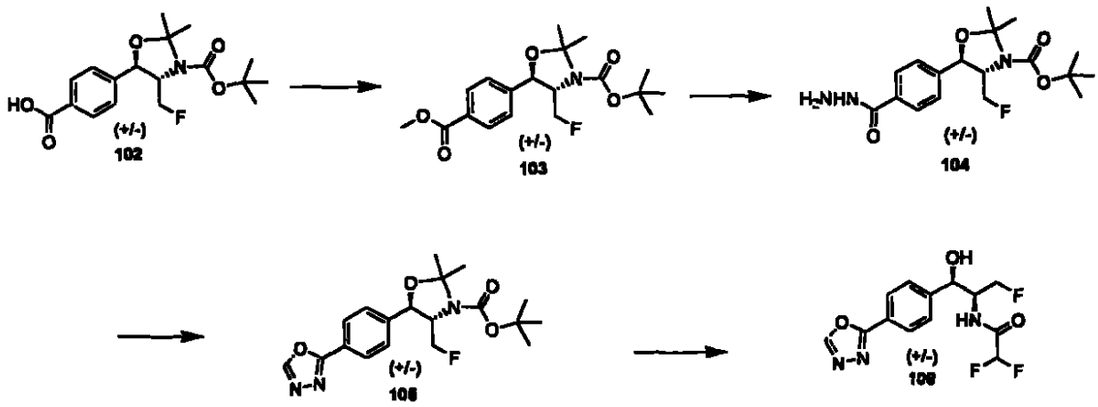
Example 83: Compound 102



Compound **17** (100 mg, 0.258 mmol) was placed in a dry round-bottom flask equipped with a stir bar. The flask was charged with 5 mL of dry THF and the contents cooled to –78 °C under N_2. With rapid stirring, *n*-BuLi (1.30 M in hexanes, 0.322 mmol, 0.248 mL) was added, which produced a clear brown/yellow solution. The mixture was stirred for an additional 10 minutes. Excess CO_2, generated by sublimation of dry ice, was passed though a drying tube charged with Drierite and

bubbled directly into the –78 °C mixture, now equipped with a venting needle in the septum to prevent buildup of CO_2 pressure. The mixture was warmed to room temperature and stirred an additional 30 minutes by which time it was clear yellow and TLC indicated a product had formed. The mixture was quenched by addition of 10% (v/w) aqueous NH_4Cl (5 drops), producing a cloudy yellow suspension. The mixture was concentrated under vacuum and re-suspended in EtOAc (50 mL) and enough 10% aqueous citric acid to produce a biphasic mixture having a pH of 2.5. The layers were separated and the EtOAc layer was washed with brine (1 x 10 mL). The mixture was dried over Na_2SO_4, filtered and evaporated to give 105 mg of yellow oil. The product was isolated by chromatotron (1 mm plate eluting with 4:1 hexanes/EtOAc to 65:35 hexanes/EtOAc to 1:1 hexanes/EtOAc to 2% MeOH in CH_2Cl_2 to 5% MeOH in CH_2Cl_2 and, finally, 10% MeOH in CH_2Cl_2 to which several drops of acetic acid had been added). The appropriate fractions were combined, diluted with toluene and evaporated to give 67.5 mg (0.191 mmol) of **102**. ^1H NMR (300 MHz, CD_3OD): δ 1.49 (s, 9H), 1.57 (S, 3H), 1.69 (s, 3H), 3.77-3.90 (m, 1H), 4.39-4.57 (m, 1H), 4.90-5.10 (br s, 1H), 5.18 (d, 1H, J = 7.2), 7.56 (d, 2H, J = 8.1), 8.04 (d, 2H, J = 8.1). LRMS (ESI^-) m/z: 352 ($M\text{-}H^+$ $C_{18}H_{23}FNO_5$ requires 352).

Example 84: Compound 106



Compound **102** (106 mg, 0.299 mmol) was dissolved in EtOAc and diazomethane, generated using an Aldrich Chemical Company diazomethane kit, was added dropwise until a slight yellow color persisted. Excess of diazomethane was allowed to evaporate overnight in a well-ventilated fume hood, and the remaining EtOAc solution was concentrated under vacuum. The residue was

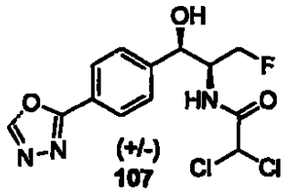
dissolved in Et_2O and washed with aqueous $NaHCO_3$ and then brine. The layers were separated and the organic layer was dried over anhydrous Na_2SO_4, filtered and concentrated to give 0.087 g (0.24 mmol, 79%) of methyl ester **103**, which was used without further purification or characterization.

Compound **103** (0.037g, 0.136 mmol) was dissolved in 1 mL of EtOH. To this was added hydrazine hydrate (.007 g, 0.177 mmol). The mixture was refluxed for 12 hours. The solvent was removed under vacuum and the product purified by chromatotron (1 mm plate, 10% MeOH in CH_2Cl_2). Compound **104** was isolated (40.0 mg, 0.109 mmol) and was used without further characterization. LRMS (ESI$^-$) m/z: 366 (M-H$^+$ $C_{18}H_{25}FN_3O_4$ requires 366).

Compound **104** (0.050 g, 0.133 mmol) was dissolved in 5 mL of triethyl orthoformate and stirred at 120 °C for 24 hours. The mixture was evaporated to give 26 mg of crude **105**, which was used without further characterization or purification.

Compound **105** (0.0051 g, 0.014 mmol) was stirred at room temperature with 10 mL of 9:1 TFA/H_2O for 10 minutes. The mixture was concentrated under vacuum, dissolved three times in a MeOH/toluene mixture, the solvents being evaporated each time, and dried to a constant weight to give 0.5 mg of deprotected material. The residue was dissolved in 1 mL of MeOH in an open container and 5 drops of methyl difluoroacetate and 15 drops of Et_3N were added. The mixture was stirred at room temperature for 12 hours. After 12 hours, TLC indicated a single product. The mixture was concentrated under vacuum and the product isolated by silica gel chromatography eluting with 20:1 CH_2Cl_2/CH_3OH to give 2.3 mg of **106**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.31-4.74 (m, 3H), 5.04 (d, 1H, J = 3.9), 5.96 (t, 1H, J = 53.9), 7.61 (d, 2H, J = 8.4), 8.05 (d, 2H, J = 8.4), 8.99 (d, 1H). LRMS (ESI$^-$) m/z: 314 (M-H$^+$ $C_{13}H_{11}F_3N_3O_3$ requires 314).

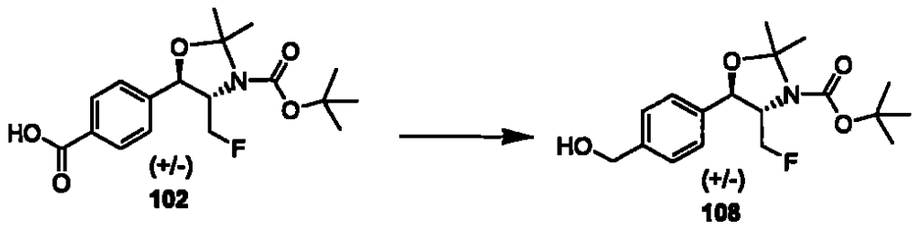
Example 85: Compound 107



Compound **107** was prepared in analogous manner to **106** using methyl dichloroacetate in place methyl difluoroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-

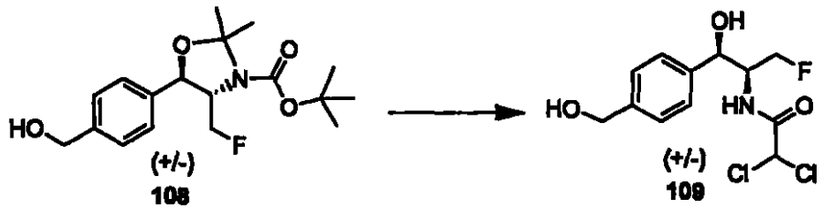
4.80 (m, 3H), 5.06 (d, 1H, J = 3.0), 6.23 (S, 1H), 7.62 (d, 2H, J = 8.4), 8.03 (d, 2H, J = 8,4), 8.98 (s. 1H). LRMS (ESI$^-$) m/z: 346 (M-H$^+$ $C_{13}H_{11}Cl_2FN_3O_3$ requires 346).

Example 86:Compound 108



Compound **102** (32.7 mg, 0.0925 mmol) was dissolved in EtOAc (8 mL) and cooled with stirring to 0 °C. Pentafluorophenol (17 mg, 0.093 mmol) was added. Once all solids had dissolved, DCC (19 mg, 0.093 mmol) was added and the mixture stirred at 0 °C for 1.25 hours. The mixture was evaporated to one-quarter the original volume at which time a DCU precipitate formed. The DCU was removed by filtration and the pentafluorophenyl ester was isolated by evaporation. The residue was dissolved in 4 mL of MeOH and cooled to 0 °C with stirring. $NaBH_4$ (18 mg, 0.46 mmol) was added portionwise. When bubbling ceased, the mixture was warmed to room temperature. After 2 hour, excess $NaBH_4$ was quenched by addition of 4 drops of glacial HOAc. The mixture was evaporated to dryness and the residue partitioned between 1N aq. HCl and EtOAc. The EtOAc was separated and washed with 1 N aqueous HCl (2 x 15 mL), saturated aqueous $NaHCO_3$ (2 x 10 mL) and finally brine (1 x 25 mL). The EtOAc layer was dried over anhydrous Na_2SO_4, filtered, and evaporated to give 50 mg of yellow oil. Chromatotron purification (1 mm plate, eluting with 5% EtOAc in hexanes to 10% EtOAc in hexanes) gave **108** (10.3 mg, 0.030 mmol). ^{1}H NMR (300 MHz, CD_3OD): δ 1.49 (s, 9H), 1.57 (s, 3H), 1.68 (s. 3H), 3.75-3.89 (m, 1H), 3.90 (s, 2H), 4.39-4.58 (m, 1H), 4.90-5.05 (br s, 1H), 5.19 (d, 1H, J = 7.2), 7.57 (d, 2H, J = 8.3), 8.04 (d, 2H, J = 8.3).

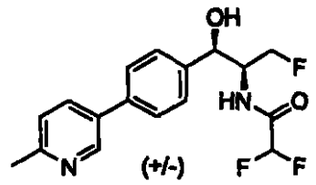
Example 87: Compound 109



Compound **108** (10 mg, 0.30 mmol) was dissolved in 5 mL of 9:1 TFA/H_2O and stirred at room temperature for 1 hour. The mixture was then concentrated under vacuum to give approximately 9 mg of deprotected material as the TFA salt. ^{1}H NMR (300 MHz, CD_3OD): δ 3.54-3.68 (m, 1H), 3.91 (s, 2H), 4.24-4.67 (m, 2H), 4.90 (d, 1H, solvent obscured), 7.57 (d, 2H, *J* = 8.4), 8.07 (d, 2H, *J* = 8.4).

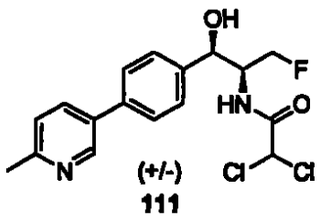
The 9 mg of material (0.03 mmol) were dissolved in 2 mL of MeOH in an open container. To this was added 15 drops of Et_3N and 15 drops of methyl dichloroacetate. The mixture was stirred for 56 hour at room temperature. By the end of the reaction the solvent had completely evaporated. The residue was loaded onto a chromatotron (1 mm plate, eluting with 2% MeOH in CH_2Cl_2 to 4% MeOH in CH_2Cl_2, providing 7.5 mg of **109**. ^{1}H NMR (300 MHz, CD_3OD): δ 3.88 (s, 2H), 4.28-4.73 (m, 3H), 5.02 (d, 1H, *J* = 3.3), 6.22 (s, 1H), 7.51 (d, 2H, *J* = 8.4), 7.97 (d, 2H, *J* = 8.4).

Example 88: Compound 110



Prepared in analogous fashion to other pyridine-containing biaryl compounds herein. ^{1}H NMR (300 MHz, CD_3OD): δ 2.54 (s, 3H), 4.30-4.68 (m, 3H), 4.99 (d, *J* = 3.3), 5.98 (t, 1H, *J* = 40.5), 7.35 (d, 1H, *J* = 6.0), 7.49 (d, 2H, *J* = 6.2), 7.61 (d, 2H, *J* = 6.2), 7.95 (dd 1H, *J* = 6.0, 1.7), 8.61 (d, 1H, *J* = 1.7). LRMS (ESI$^-$) m/z: 336.9 (M-H$^+$ $C_{17}H_{16}F_3N_2O_2$ requires 337.1).

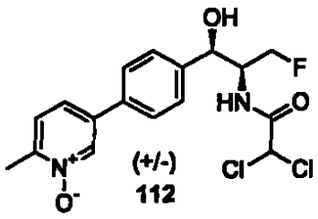
Example 89: Compound 111



111

Prepared in analogous fashion to other pyridine-containing biaryl compounds herein. ^{1}H NMR (300 MHz, CD_3OD): δ 2.56 (s, 3H), 4.30-4.74 (m, 3H), 5.01 (d, 1H, *J* = 3.9), 6.27 (s, 1H), 7.36 (d, 1H, *J* = 8.1), 7.56 (d, 2H, *J* = 8.4), 7.61 (d, 2H, *J* = 8.4), 7.96 (1H, dd, *J* = 8.1, 2.3), 8.62 (d, 1H, *J* = 2.31).

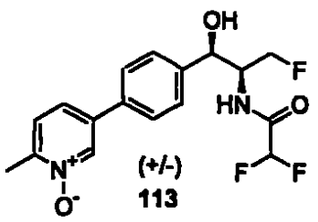
Example 90: Compound 112



112

Compound **111** (0.0185g, 0.0499 mmol) was dissolved in 10 mL CH_2Cl_2 at 0 °C. To this was added *m*-CPBA (0.246 g, 0.0997 mmol). The mixture was stirred until the ice bath melted and the mixture came to room temperature, approximately 12 hours. The mixture was concentrated under vacuum and **112** was isolated by preparative plate silica gel chromatography (15% MeOH in CH_2Cl_2) followed by a silica gel plug (3% MeOH in CH_2Cl_2). LRMS (ESI$^-$) m/z: 385 (M-H$^+$ $C_{17}H_{16}Cl_2FN_2O_3$ requires 385.1).

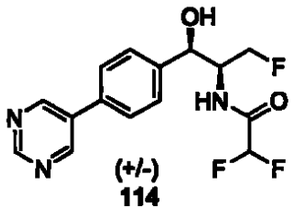
Example 91: Compound 113



113

Compound **113** was synthesized from compound **110** in the same manner **112** was synthesized from **111**. LRMS (ESI$^-$) m/z: 353 (M-H$^+$ $C_{17}H_{16}Cl_2FN_2O_3$ requires 353.1).

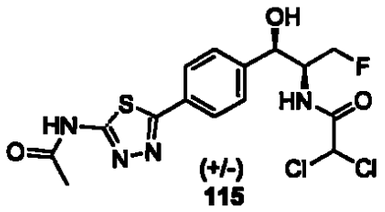
Example 92: Compound 114



114

This compound was prepared in the same manner as other biaryls herein. LRMS (ESI$^-$) m/z: 324 (M-H$^+$ $C_{15}H_{13}F_3N_3O_2$ requires 324.1).

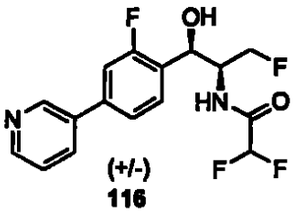
Example 93: Compound 115



115

Prepared in same manner as compound **48**. The required aryl bromide was prepared in two steps from 2-amino-1,3,4-thiadiazole by bromination with Br_2 followed by acetylation. LRMS (ESI$^-$) m/z: 419 (M-H$^+$ $C_{15}H_{14}Cl_2FN_4O_3S$ requires 419.0).

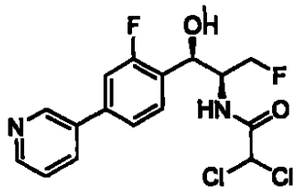
Example 94: Compound 116



116

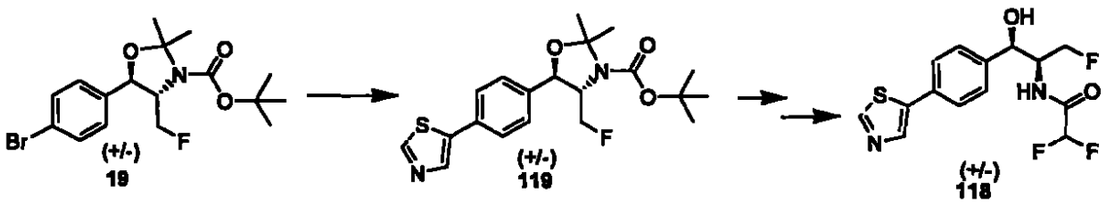
Compound **116** was synthesized as set forth in Schemes 2 and 3. The phenylserine analog was obtained by condensation of 4-bromo-2-fluorobenzaldehyde with glycine. Suzuki coupling method B was used to couple the bromide intermediate with *m*-pyridineboronic acid. Deprotection and difluoroacetylation were carried out as described previously herein. LRMS (ESI^+) m/z: 343 ($M+H^+$ $C_{16}H_{15}F_4N_2O_2$ requires 343.1).

Example 95: Compound 117



Same procedure as that used for the synthesis of **116**. LRMS (ESI^+) m/z: 375 ($M+H^+$ $C_{16}H_{15}F_4N_2O_2$ requires 375.1).

Example 96: Compound 118

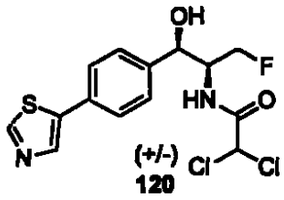


Compound **19** (100 mg, 0.258 mmol), potassium acetate (38 mg, 0.386 mmol), and 2 mL of *N,N*-dimethylacetamide (DMAC) were combined in a glass pressure tube. Thiazole (112 mg, 1.31 mmol) was added by syringe, and the syringe rinsed with an additional 1 mL of DMAC, which was added to the pressure tube. The mixture was stirred at room temperature while purging with N_2 for 10 minutes. $Pd(PPh_3)_4$ (15 mg, 0.013 mmol) was added, the mixture purged with N_2 for 5 minutes, the tube sealed and heated to 150 °C behind a blast shield and held at that temperature for 12 hours. The mixture became dark brown. After cooling to room temperature, the contents of the tube were filtered through a pad of Celite and the

filtrate evaporated to dryness. The residue was dissolved in 40 mL of 3:1 EtOAc/hexanes and washed with H_2O (2 x 15 mL) and brine (2 x 15 mL) and then dried over anhydrous Na_2SO_4. The dried organic layer was filtered to remove the drying agent and concentrated under vacuum to give 109 mg of brown oil. Compound **119** was purified by silica gel chromatography eluting with 80:20 to 50:50 hexanes/EtOAc (54 mg, 0.14 mmol). LRMS (ESI^+) m/z: 393.1 ($M+H^+$ $C_{20}H_{26}FN_2O_3S$ requires 393.2).

All of compound **119** was dissolved in 9:1 (v/v) TFA/H_2O (7.5 mL) and stirred at room temperature for 30 minutes. The mixture was then concentrated under vacuum and the residue dissolved twice in a mixture of toluene and methanol, the solvents being evaporated each time. A portion of the product (9.7 mg, 0.027 mmol) was dissolved in 2 mL of MeOH and 10 drops of Et_3N and 12 drops of methyl difluoroacetate were added. The mixture was left open to the air and stirred rapidly for 16 hours. The mixture was evaporated to dryness and the residue purified by chromatotron (1 mm plate) eluting with 3% MeOH in CH_2Cl_2 to give **118** (6.0mg, 0.018 mmol. ^1H NMR (300 MHz, CD_3OD): δ 4.27-4.69 (m, 3H), 4.97(d, 1H, J = 4.2), 5.99 (t, 1H, J = 53.9), 7.47 (d, 2H, J = 8.3), 7.65 (d, 2H, J = 8.3), 8.16 (s, 1H), 8.94 (s, 1H). LRMS (ESI^+) m/z: 331.1 ($M+H^+$ $C_{14}H_{14}F_3N_2O_2S$ requires 331.1).

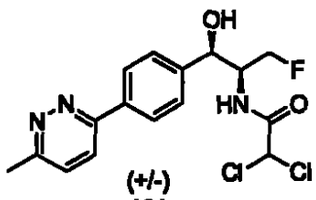
Example 97: Compound 120



120

Compound **120** was prepared in a manner analogous to that used to prepare **118** using dichloroacetylation instead of difluoroacetylation. LRMS (ESI^+) m/z: 363.0 ($M+H^+$ $C_{14}H_{14}Cl_2FN_2O_2S$ requires 363.0).

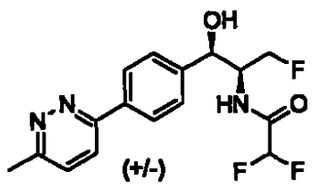
Example 98: Compound 121



(+/-)
121

The protected biaryl intermediate was synthesized by Suzuki coupling method B from **23** and 3-chloro-6-methylpyridazine. Deprotection and dichloroacetylation were performed as for other compounds of this invention. LRMS (ESI^+) m/z: 372.0 ($M+H^+$ $C_{16}H_{17}Cl_2FN_3O_2$ requires 372.1).

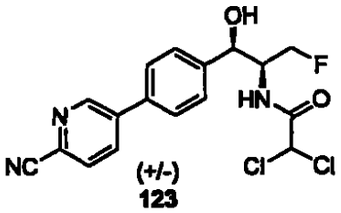
Example 99: Compound 122



(+/-)
122

Prepared in analogous fashion to **121**. LRMS (ESI^+) m/z: 340.1 ($M+H^+$ $C_{16}H_{17}F_3N_3O_2$ requires 340.1).

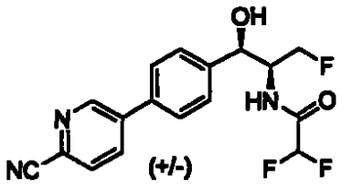
Example 100: Compound 123



(+/-)
123

The protected biaryl intermediate was prepared by Suzuki coupling method B from boronic acid **23** and the appropriate aryl bromide. This intermediate was then deprotected by brief treatment with 9:1 (v/v) TFA/H_2O and dichloroacetylated as described for **29**. LRMS (ESI^+) m/z: 380.0 ($M-H^+$ $C_{17}H_{13}Cl_2FN_3O_2$ requires 380.0).

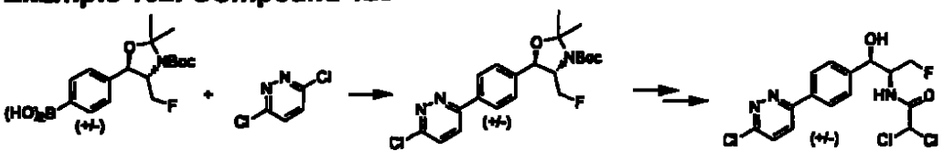
Example 101: Compound 124



124

Prepared in analogous fashion to **123**. Difluoroacetylation was performed as with other compounds of this invention. LRMS (ESI^+) m/z: 348.0 ($M+H^+$ $C_{17}H_{13}F_3N_3O_2$ requires 348.1).

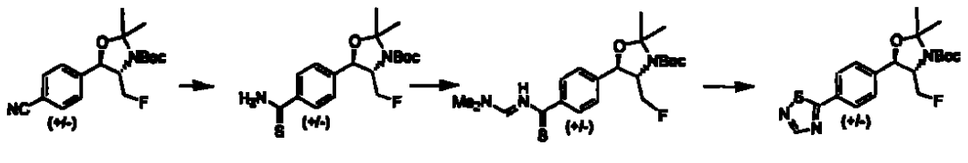
Example 102: Compound 125



23 125

The biaryl intermediate was prepared from boronic acid **23** using Suzuki coupling. LRMS (ESI^+) m/z: 392.0 ($M-H^+$ $C_{15}H_{13}Cl_3FN_3O_2$ requires 392.0).

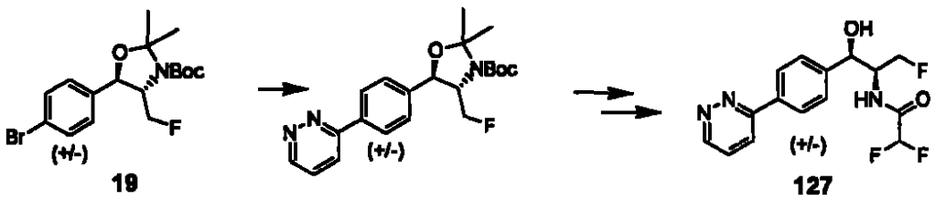
Example 103: Compound 126



126

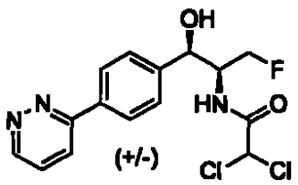
The starting nitrile was prepared from **35** by analogy to the preparation of bromo intermediate **19**. Treatment with $Ph_2P(S)SH$ gave the thiobenzamide which was converted to the thiobenzamidine intermediate by reaction with dimethylformamide dimethylacetal. Cyclization with hydroxylamine-O-sulfonic acid in methanol/pyridine gave the biaryl intermediate. LRMS (ESI^+) m/z: 364.0 ($M-H^+$ $C_{13}H_{12}Cl_2FN_3O_2S$ requires 364.0)

Example 104: Compound 127



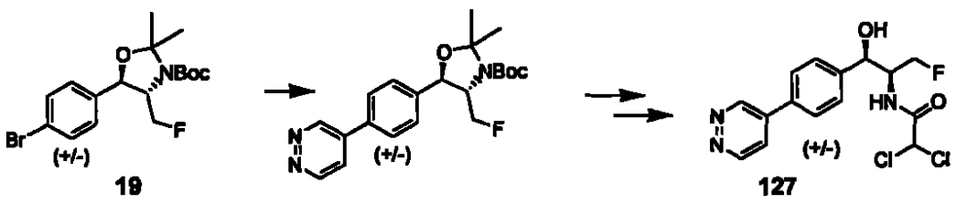
The phenyllithium intermediate was prepared by treatment of intermediate **19** with n-butyllithium in THF at -78 °C. The intermediate was reacted with pyridazine and the mixture of the resulting 2- and 3-position adducts was oxidized with DDQ. The desired 3-pyridazyl regioisomer was isolated by chromatography. LRMS (ESI^+) m/z: 326.0 ($M-H^+$ $C_{15}H_{14}F_3N_3O_2$ requires 326.0)

Example 105: Compound 128



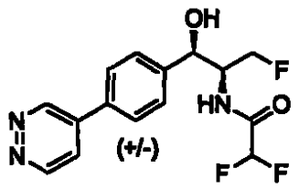
Compound **128** was synthesized using the procedure in Example 104. LRMS (ESI^+) m/z: 358.0 ($M-H^+$ $C_{15}H_{14}Cl_2FN_3O_2$ requires 358.0)

Example 106: Compound 129



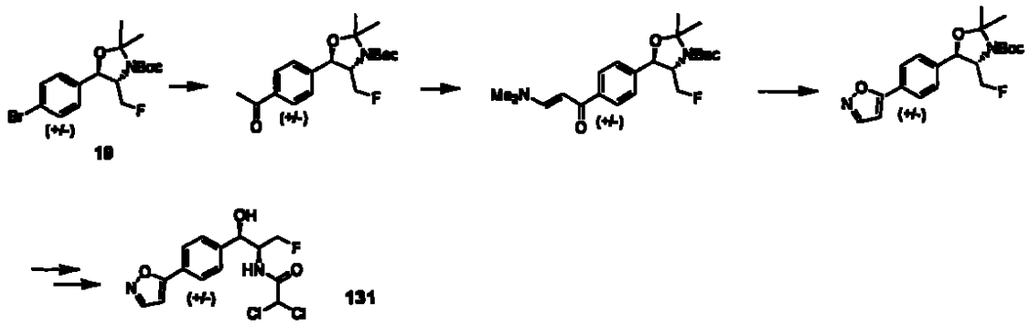
The procedure discussed in Example 104 was used. LRMS (ESI^+) m/z: 358.0 ($M-H^+$ $C_{15}H_{14}Cl_2FN_3O_2$ requires 358.0)

Example 107: Compound 130



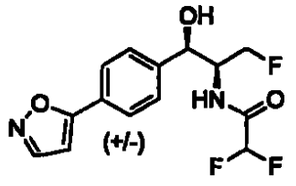
The procedure of Example 104 was used. LRMS (ESI^+) m/z: 326.0 ($M-H^+$ $C_{15}H_{14}F_3N_3O_2$ requires 326.0)

Example 108: Compound 131



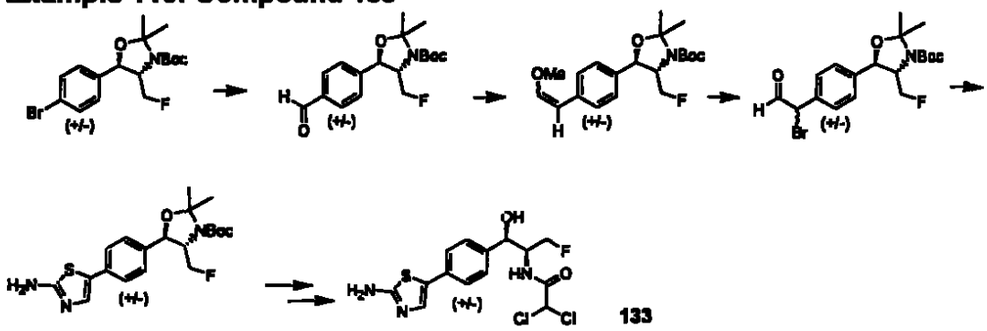
The phenyllithium intermediate was reacted with N-methoxy-N-methylacetamide and the resulting acetophenone was reacted with dimethylformamide dimethylacetal and hydroxylamine-O-sulfonic acid in methanol in presence of pyridine to give the isoxazole. LRMS (ESI^+) m/z: 347.0 ($M-H^+$ $C_{14}H_{13}Cl_2N_2O_3$ requires 347.0)

Example 109: Compound 132



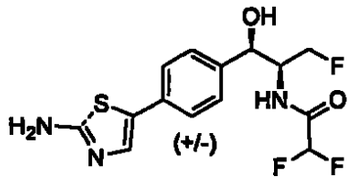
Compound **132** was synthesized using the procedure in Example 108. LRMS (ESI$^+$) m/z: 315.0 (M-H$^+$ $C_{14}H_{13}F_3N_2O_3$ requires 315.0.

Example 110: Compound 133



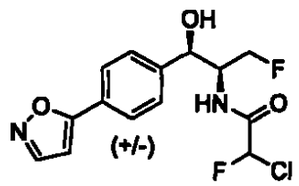
The phenyllithium intermediate was reacted with dimethylformamide to give the formyl intermediate which was then reacted with the ylide generated from methoxymethyltriphenyl phosphonium bromide. The resulting enol ether was brominated with bromine to give the bromoaldehyde. Cyclization with thiourea gave the aminothiazole. LRMS (ESI$^+$) m/z: 378.0 (M-H$^+$ $C_{14}H_{14}Cl_2FN_3O_2S$ requires 378.0)

Example 111: Compound 134



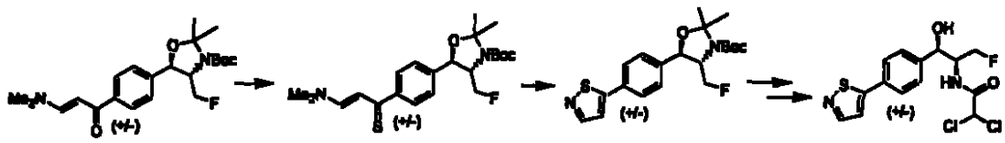
Compound **134** was synthesized using the procedure in Example 110. LRMS (ESI$^+$) m/z: 346.0 (M-H$^+$ $C_{14}H_{14}F_3N_3O_2S$ requires 346.0)

Example 112: Compound 135 (mixture of diastereoisomers)



Compound **135** was synthesized using the procedure in Example 108. LRMS (ESI$^+$) m/z: 331.0 (M-H$^+$ $C_{14}H_{13}F_2N_2O_3$ requires 331.0)

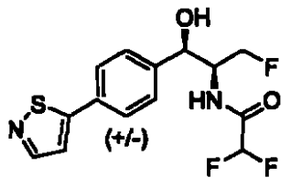
Example 113: Compound 136



136

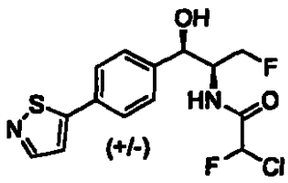
The starting material in Example 108 was reacted with Lawesson's reagent and the product was cyclized to the biaryl intermediate with hydroxylamine-O-sulfonic acid in methanol in presence of pyridine. LRMS (ESI^+) m/z: 363.0 ($M-H^+$ $C_{14}H_{13}Cl_2FN_2O_2S$ requires 363.0)

Example 114: Compound 137



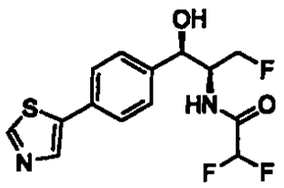
Compound **137** was synthesized using the procedure in Example 113. LRMS (ESI^+) m/z: 331.0 ($M-H^+$ $C_{14}H_{13}F_3N_2O_2S$ requires 331.0)

Example 115: Compound 138 (mixture of diastereoisomers)



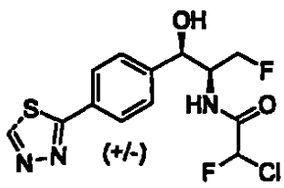
Compound **138** was synthesized using the procedure in Example 113. LRMS (ESI^+) m/z: 347.0 ($M-H^+$ $C_{14}H_{13}ClF_2N_2O_2S$ requires 347.0)

Example 116: Compound 139



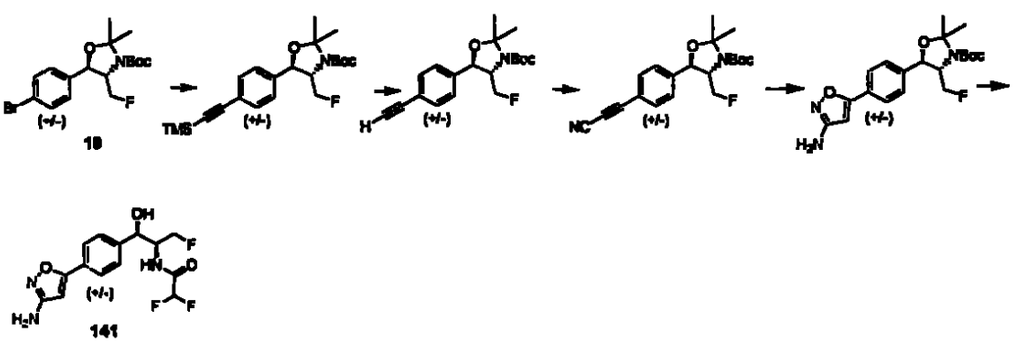
Compound **139** was synthesized using the procedure in Example 96 starting from the desired enantiomer of intermediate **19**. LRMS (ESI^+) m/z: 331.0 ($M-H^+$ $C_{14}H_{13}F_3N_2O_2S$ requires 331.0)

Example 117: Compound 140 (mixture of diastereoisomers)



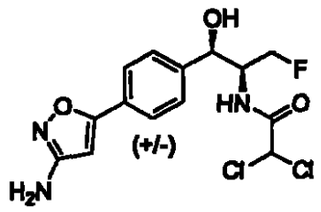
Compound **140** was synthesized using the procedure in Example 41. LRMS (ESI^+) m/z: 348.0 ($M-H^+$ $C_{13}H_{12}ClF_2N_3O_2S$ requires 348.0)

Example 118: Compound 141



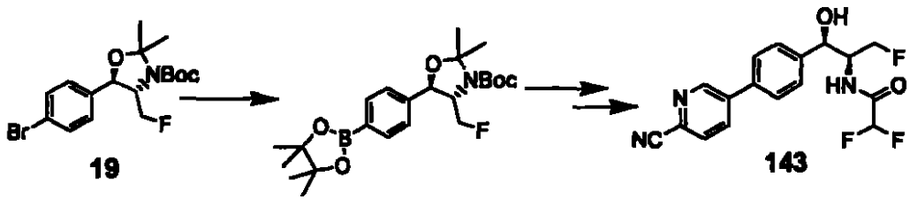
Intermediate **19** was reacted with trimethylsilylacetylene in presence of copper iodide and $PdCl_2(PPh_3)_2$. The product was desilylated by treatment with potassium carbonate in methanol. The cyanoacetylene intermediate was reacting acetylene with n-butyllithium, which was then reacted with tosylcyanide. Cyclization to the 3-aminoisoxazole was performed by treatment of the cyanoacetylene with hydroxylamine in ethanol. LRMS (ESI^+) m/z: 330.0 ($M-H^+$ $C_{14}H_{14}F_3N_3O_3$ requires 330.0)

Example 119: Compound 142



Compound **142** was synthesized using the procedure in Example 118. LRMS (ESI$^+$) m/z: 362.0 (M-H$^+$ $C_{14}H_{14}Cl_2FN_3O_3$ requires 362.0)

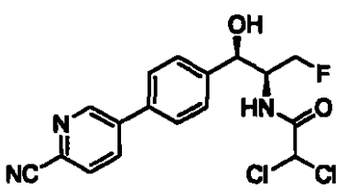
Example 120: Compound 143



Compound **19**

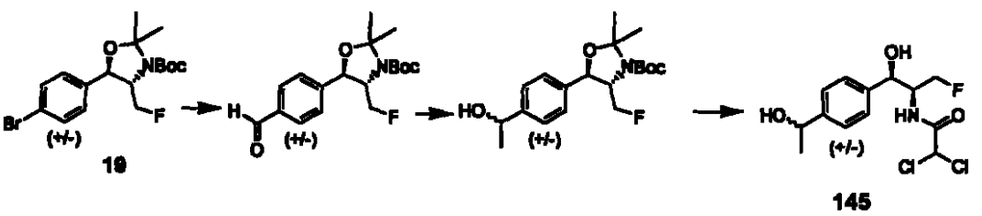
The desired enantiomer of intermediate **19** was reacted with bis(pinacolato)diboron in presence of 1,1'-bis(diphenylphosphino)ferrocenepalladium (II) dichloride dichloromethane. The pinacolboronate ester was coupled with 5-bromo-2-cyanopyridine using the Suzuki reaction. LRMS (ESI$^+$) m/z: 350.0 (M-H$^+$ $C_{17}H_{14}F_3N_3O_2$ requires 350.0)

Example 121: Compound 144



Compound **144** was synthesized using the procedure in Example 120. LRMS (ESI$^+$) m/z: 382.0 (M-H$^+$ $C_{17}H_{14}Cl_2FN_3O_2$ requires 382.0)

Example 122: Compound 145 (mixture of diastereoisomers)

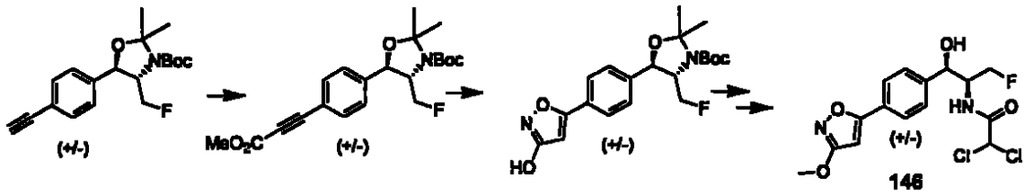


Compound **19**

Compound **145**

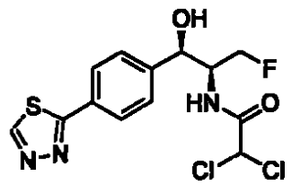
Intermediate **19** was reacted with n-butyllithium and then with dimethylformamide. The formyl intermediate was reacted with methylmagnesium bromide to give the benzylic alcohol as a mixture of diastereisomers. LRMS (ESI^+) m/z: 324.0 ($M-H^+$ $C_{13}H_{16}Cl_2FNO_3$ requires 324.0)

Example 123: Compound 146



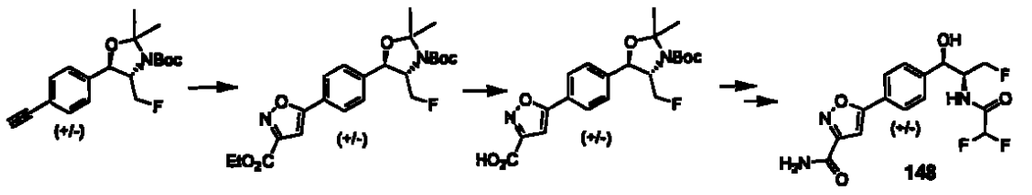
The starting acetylene, prepared as described in Example 118, was reacted with n-butyllithium followed by addition of carbon dioxide and the resulting acid was esterified with diazomethane in ethyl acetate/diethyl ether. The 3-hydroxyisoxazole was obtained by reacting the ester with hydroxylamine and was then methylated with diazomethane in ethyl acetate/diethyl ether. LRMS (ESI^+) m/z: 377.0 ($M-H^+$ $C_{15}H_{15}Cl_2FN_2O_4$ requires 377.0)

Example 124: Compound 147



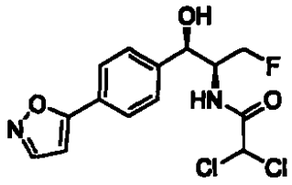
Compound **147** was synthesized by the method of Example 120 using 2-bromo-1,3,4-thiadiazole in a Suzuki coupling. LRMS (ESI^+) m/z: 364.0 ($M-H^+$ $C_{13}H_{12}Cl_2FN_3O_2S$ requires 364.0)

Example 125: Compound 148



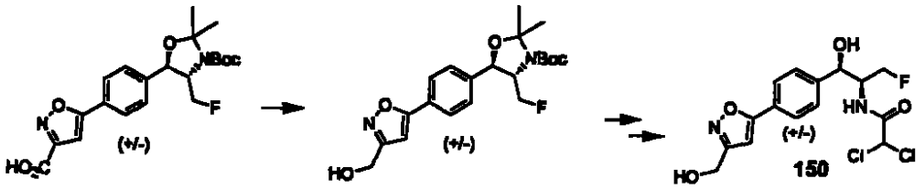
The starting acetylene was obtained as described in Example 118. The 3-carboethoxy isoxazole intermediate was obtained by cycloaddition of the nitrile oxide generated *in situ* from ethyl nitroacetate, di-t-butyldicarbonate and 4-dimethylaminopyridine. The amide was obtained from the 3-carboethoxy isoxazole intermediate by hydrolysis to the acid, conversion to the acyl chloride and reaction with ammonia. LRMS (ESI$^+$) m/z: 358.0 (M-H$^+$ $C_{15}H_{14}F_3N_3O_4$ requires 358.0)

Example 126: Compound 149



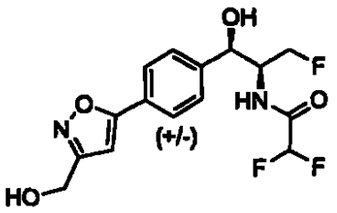
Compound **149** was synthesized using the procedure in Example 108 starting from the desired enantiomer of intermediate **19**. LRMS (ESI$^+$) m/z: 347.0 (M-H$^+$ $C_{14}H_{13}Cl_2FN_2O_3$ requires 347.0)

Example 127: Compound 150



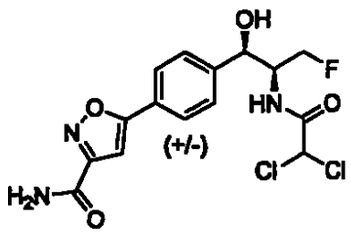
The 3-carboxyisoxazole intermediate obtained as described in Example 125 was converted into the acid chloride using Vilsmeyer reagent in dimethylformamide. The crude product was reduced to the 3-hydroxymethyl isoxazole intermediate with tetrabutylammonium borohydride in THF. LRMS (ESI$^+$) m/z: 377.0 (M-H$^+$ $C_{15}H_{15}Cl_2FN_2O_4$ requires 377.0)

Example 128: Compound 151



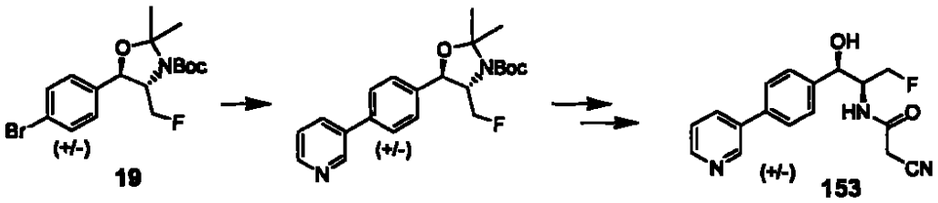
Compound **151** was synthesized using the procedure in Example 127.
LRMS (ESI$^+$) m/z: 345.0 (M-H$^+$ $C_{16}H_{15}F_3N_2O_4$ requires 345.0)

Example 129: Compound 152



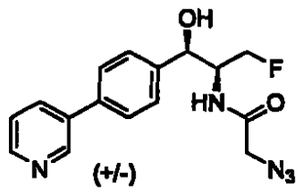
Compound **152** was synthesized using the procedure in Example 125.
LRMS (ESI$^+$) m/z: 390.0 (M-H$^+$ $C_{15}H_{14}Cl_2FN_3O_4$ requires 390.0)

Example 130: Compound 153



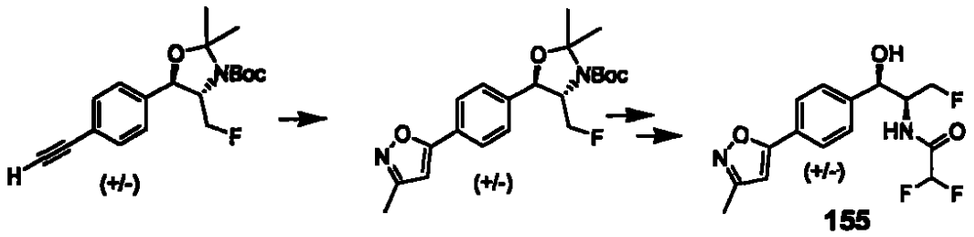
Compound **153** was synthesized using the procedure in Example 40.
EDC/HOBT was used to acylate the amino intermediate with cyanoacetic acid.
LRMS (ESI$^+$) m/z: 314.0 (M-H$^+$ $C_{17}H_{16}FN_3O_2$ requires 314.0)

Example 131: Compound 154



Compound **154** was synthesized using the procedure in Example 130.
LRMS (ESI$^+$) m/z: 330.0 (M-H$^+$ $C_{16}H_{16}FN_5O_2$ requires 330.0)

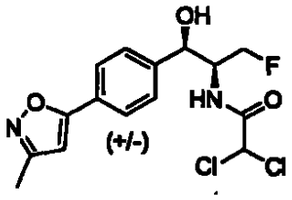
Example 132: Compound 155



155

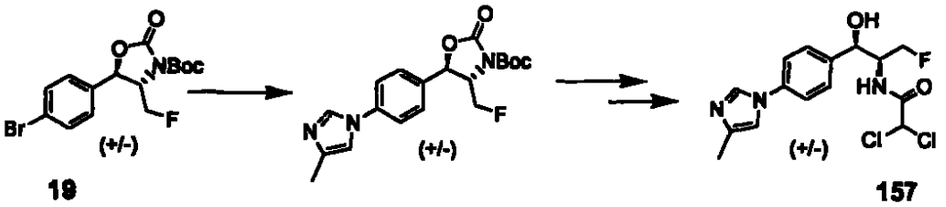
The starting acetylene was obtained as described in Example 118. The 3-methyl isoxazole intermediate was obtained by cycloaddition of the nitrile oxide generated *in situ* from nitroethane, di-t-butyldicarbonate and 4-dimethylaminopyridine. LRMS (ESI^+) m/z: 329.0 ($M-H^+$ $C_{15}H_{15}F_3N_2O_3$ requires 329.0)

Example 133: Compound 156



Compound **156** was synthesized using the procedure in Example 132. LRMS (ESI^+) m/z: 361.0 ($M-H^+$ $C_{15}H_{15}Cl_2FN_2O_3$ requires 361.0)

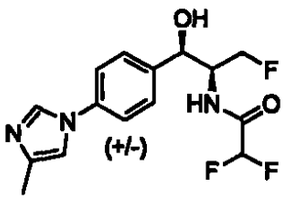
Example 134 : Compound 157



19 157

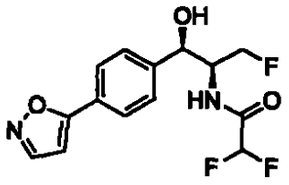
Intermediate **19** *was reacted with 4-methylimidazole in refluxing* dimethylformamide in the presence of copper powder. LRMS (ESI^+) m/z: 3460.0 ($M-H^+$ $C_{14}H_{14}Cl_2FN_3O_2$ requires 346.0)

Example 135: Compound 158



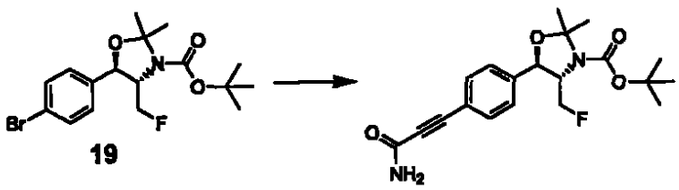
Compound **158** was synthesized using the procedure in Example 134.
LRMS (ESI$^+$) m/z: 328.0 (M-H$^+$ $C_{15}H_{16}F_3N_3O_2$ requires 328.0)

Example 136: Compound 159



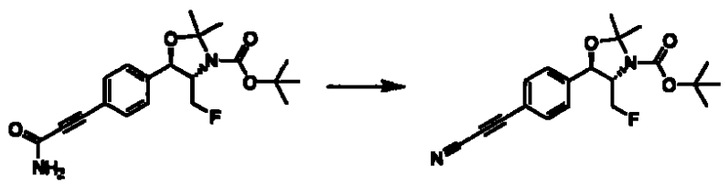
Compound **159** was synthesized using the procedure in Example 126.
LRMS (ESI$^+$) m/z: 315.0 (M-H$^+$ $C_{14}H_{13}F_3N_2O_3$ requires 315.0)

Example 137: Compound 160

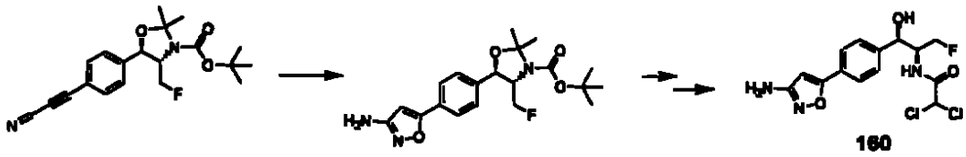


19

A mixture of the desired enantiomer of intermediate **19** (1 g, 2.5 mmol), potassium carbonate (712 mg, 5 mmol), and propiolamide (1.07 g, 15 mmol) in N,N-dimethylacetamide (4 mL) was stirred at room temperature while purging with N_2 for 10 minutes. $Pd(PPh_3)_4$ catalyst (15 mg, 0.013 mmol) was added and the mixture was purged with N_2 for another 5 minutes. The resulting mixture was stirred at 100 °C for 9 hrs. After cooling to room temperature, the crude was flash silica gel chromatographed to give the coupling product (601 mg). ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.49 (s, 9H), 1.59 (s, 3H), 1.70 (s, 3H), 3.75-3.90 (m, 1H), 4.35-4.60 (m, 2H), 5.15 (d, 1H, J = 7.5), 5.65 (s, 1H), 5.85 (s, 1H), 7.45 (d, 2H, J = 8.4), and 7.55 (d, 2H, J = 8.4).



To dimethylformamamide (2 mL) was slowly added thionyl chloride (2 mL) at 0 °C and the reaction mixture was stirred at that temperature for 30 min. The solution was then cannulated to a solution of the propiolamide intermediate (600 mg) in DMF (4 mL) at 0 °C. After stirring at room temperature for 30 min, the mixture was poured into ice/water (50 mL). The pH was adjusted to 7 with saturated sodium bicarbonate. The solution was extracted with ethyl acetate three times. The combined organic layers were washed with water, dried over anhydrous sodium sulfate, and purified by column chromatography to give propiolonitrile intermediate (326 mg). ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.49 (s, 9H), 1.59 (s, 3H), 1.70 (s, 3H), 3.75-3.90 (m, 1H), 4.35-4.60 (m, 2H), 5.15 (d, 1H, J = 7.5), 7.49 (d, 2H, J = 8.4), and 7.63 (d, 2H, J = 8.4).

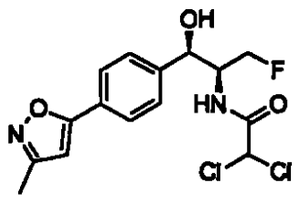


160

To a solution of sodium hydroxide (206 mg, 7 eq.) in water (2 mL), was added hydroxylamine hydrochloride (256 mg, 5 eq.). The solution was added to the propiolonitrile intermediate (263 mg) dissolved in ethanol (7 mL). The resulting mixture was stirred at room temperature for 6 hrs. Ethyl acetate was added, the organic layer was separated, washed with water, and dried with anhydrous sodium sulfate. The crude material was purified by column chromatography to give the protected 3-aminoisoxazole which was then dissolved in THF (6 mL) and 4 N HCl (6 mL), and stirred at 80 °C for 2.5 hrs. The reaction mixture was evaporated to dryness under reduced pressure. The residue was co-evaporated with methanol twice and dried overnight under vacuum. To the crude material dissolved in methanol (2 mL) was added triethylamine (2 mL), followed by methyl dichloroacetate (1 mL). The mixture was stirred at room temperature for 24 hrs. The solvent was evaporated and the mixture was purified by flash column chromatography to give compound **160** (167 mg). ^{1}H NMR (300 MHz, $CDCl_3$ +

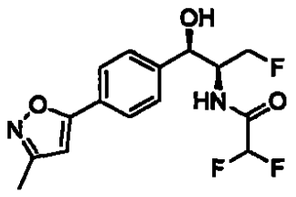
CD$_3$OD): δ 4.35-4.70 (m, 3H), 5.05 (m, 1H), 5.85 (s, 1H), 6.16 (s, 1H), 7.44 (d, 2H, J = 8.1), and 7.65 (d, 2H, J = 8.1). LRMS (ESI$^+$) m/z: 362.0 (M-H$^+$ $C_{14}H_{14}Cl_2FN_3O_3$ requires 362.0)

Example 138: Compound 161



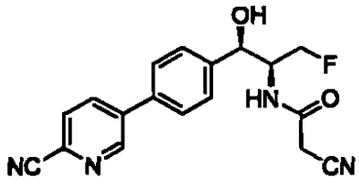
Compound **161** was synthesized using the procedure in Example 132 starting from the desired enantiomer of intermediate **19**. LRMS (ESI$^+$) m/z: 361.0 (M-H$^+$ $C_{15}H_{15}Cl_2FN_2O_3$ requires 361.0)

Example 139: Compound 162



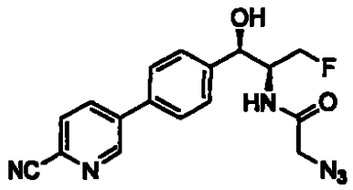
Compound **162** was synthesized using the procedure in Example 138. LRMS (ESI$^+$) m/z: 329.0 (M-H$^+$ $C_{15}H_{15}F_3N_2O_3$ requires 329.0)

Example 140: Compound 163



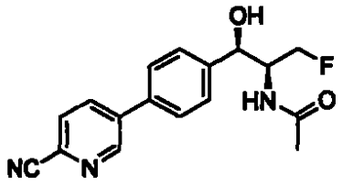
Compound **163** was synthesized using the procedure in Example 120. Dicyclohexylcarbodiimide was used to acylate the amino intermediate with cyanoacetic acid. LRMS (ESI$^+$) m/z: 339.0 (M-H$^+$ $C_{18}H_{15}FN_4O_2$ requires 339.0)

Example 141: Compound 164



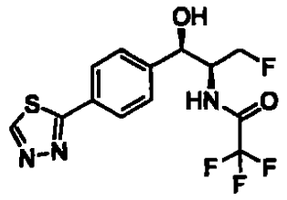
Compound **164** was synthesized using the procedure in Example 120. Dicyclohexylcarbodiimide was used to acylate the amino intermediate with azidoacetic acid. LRMS (ESI^+) m/z: 355.0 ($M-H^+$ $C_{17}H_{15}FN_6O_2$ requires 355.0)

Example 142: Compound 165



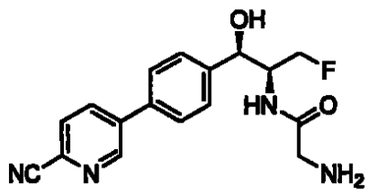
Compound **165** was synthesized using the procedure in Example 120. The amino intermediate was acetylated with acetic anhydride. LRMS (ESI^+) m/z: 314.0 ($M-H^+$ $C_{17}H_{16}FN_3O_2$ requires 314.0)

Example 143: Compound 166



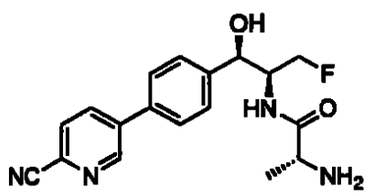
Compound **166** was synthesized using the procedure in Example 120. LRMS (ESI^+) m/z: 350.0 ($M-H^+$ $C_{13}H_{11}F_4N_3O_2S$ requires 350.0)

Example 144: Compound 167



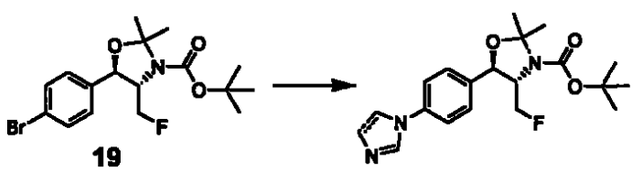
Compound **167** was synthesized using the procedure in Example 120. Dicyclohexylcarbodiimide was used to acylate the amino intermediate with N-Boc-glycine and the resulting glycinamide was deprotected with hydrochloric acid in methanol. LRMS (ESI^+) m/z: 329.0 ($M-H^+$ $C_{17}H_{17}FN_4O_2$ requires 329.0)

Example 145: Compound 168



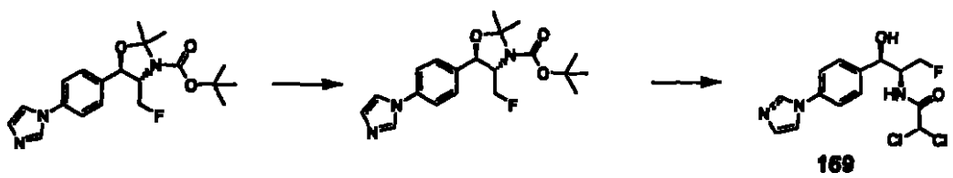
Compound **168** was synthesized using the procedure in Example 144. LRMS (ESI^+) m/z: 343.0 ($M-H^+$ $C_{18}H_{19}FN_4O_2$ requires 343.0)

Example 146 : Compound 169



19

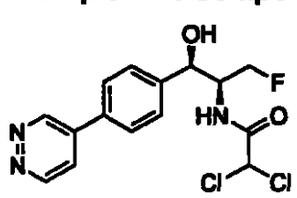
The desired enantiomer of intermediate **19** (1.16 g, 3 mmol), potassium carbonate (1.659 g, 12 mmol), imidazole (1.225 g, 18 mmol), and copper powder (191 mg, 3 mmol) in 20 mL of DMF were stirred vigorously at reflux for 4 hrs. The reaction mixture was cooled to room temperature and poured into water (100 mL). After extraction with ethyl acetate and washing of the organic extract with water (3X), the organic layer was passed through a silica gel and anhydrous sodium sulfate plug. Evaporation of solvent gave the imidazole (1.06 g). ^1H NMR (300 MHz, $CDCl_3$): δ 1.45 (s, 9H), 1.60 (s, 3H), 1.75 (s, 3H), 3.75-3.85 (m, 1H), 4.35-4.55 (m, 2H), 5.20 (d, 1H, J = 7.5), 7.22 (s, 1H), 7.28 (s, 1H), 7.41 (d, 2H, J = 8.4), 7.58 (d, 2H, J = 8.4), and 7.89 (s, 1H).



169

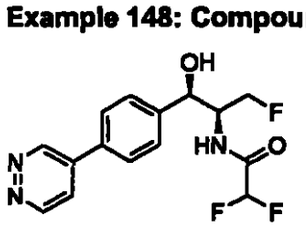
The imidazole intermediate (2.77 g) was dissolved in THF (20 mL) and 4 N HCl (20 mL), and stirred at 80 °C for 2.5 hrs. The reaction mixture was evaporated to dryness under reduced pressure. The residue was co-evaporated with methanol twice and dried overnight under vacuum. The crude material was dissolved in methanol (20 mL) and triethylamine (5 mL) was added, followed by methyl dichloroacetate (5 mL). The mixture was stirred at room temperature for 20 hrs. The solvent was evaporated and the mixture was purified by flash column chromatography to give compound **169** (1.1 g) as white solid. ^{1}H NMR (300 MHz, $CDCl_3$): δ 4.30-4.75 (m, 3H), 5.21 (d, 1H, J = 7.5), 5.86 (s, 1H), 7.05 (d, 1H, J=7.5), 7.10 (s, 1H), 7.28 (s, 1H), 7.39 (d, 2H, J =8.4), 7.52 (d, 2H, J=8.4 Hz), and 7.81 (s, 1H). LRMS (ESI$^+$) m/z: 360.0 (M-H$^+$ $C_{15}H_{16}Cl_2FN_3O_2$ requires 360.0)

Example 147: Compound 170



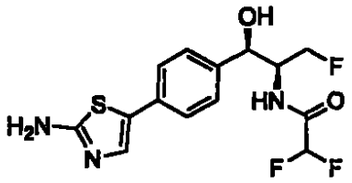
Compound **170** was synthesized by the method of Example 106 using the desired enantiomer of Intermediate **19**. LRMS (ESI$^+$) m/z: 358.0 (M-H$^+$ $C_{15}H_{14}Cl_2FN_3O_2$ requires 358.0)

Example 148: Compound 171



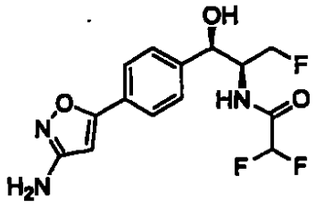
Compound **171** was synthesized using the procedure in Example 147. LRMS (ESI$^+$) m/z: 326.0 (M-H$^+$ $C_{15}H_{14}F_3N_3O_2$ requires 326.0)

Example 149: Compound 172



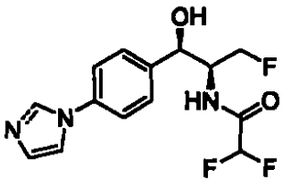
Compound **172** was synthesized by the method of Example 110 using the desired enantiomer of Intermediate **19**. LRMS (ESI^+) m/z: 346.0 ($M-H^+$ $C_{14}H_{14}F_3N_3O_2S$ requires 346.0)

Example 150: Compound 173



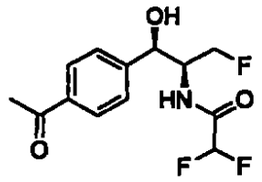
Compound **173** was synthesized using the procedure in Example 137. LRMS (ESI^+) m/z: 330.0 ($M-H^+$ $C_{14}H_{14}F_3N_3O_3$ requires 330.0)

Example 151: Compound 174



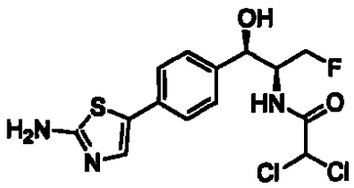
Compound **174** was synthesized using the procedure in Example 146. LRMS (ESI^+) m/z: 314.0 ($M-H^+$ $C_{14}H_{14}F_3N_3O_2$ requires 314.0)

Example 152: Compound 175



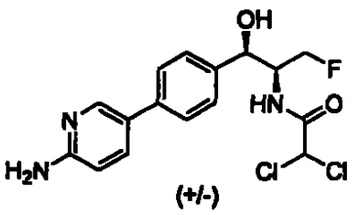
Compound **175** was synthesized using the procedure in Example 28. LRMS (ESI^+) m/z: 290.0 ($M-H^+$ $C_{13}H_{14}F_3NO_3$ requires 290.0)

Example 153: Compound 176



Compound **176** was synthesized using the procedure in Example 149. LRMS (ESI^+) m/z: 378.0 ($M-H^+$ $C_{14}H_{14}Cl_2FN_3O_2S$ requires 378.0)

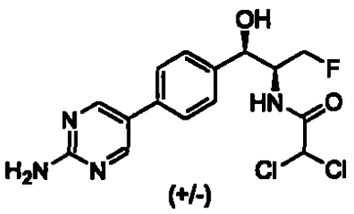
Example 154: Compound 177



(+/-)

Compound **177** was synthesized by the method of Example 33 using the bromopyridine derivative and boronic acid **23**. LRMS (ESI^+) m/z: 372.0 ($M-H^+$ $C_{16}H_{16}Cl_2FN_3O_2$ requires 372.0)

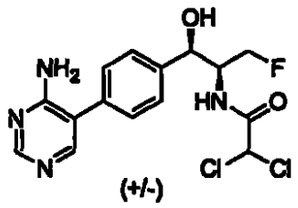
Example 155: Compound 178



(+/-)

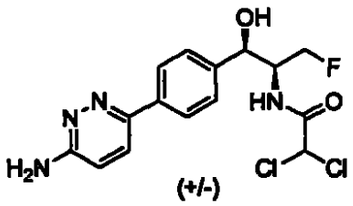
Compound **178** was synthesized by the method of Example 33 using a bromopyrimidine derivative and boronic acid **23**. LRMS (ESI^+) m/z: 373.0 ($M-H^+$ $C_{15}H_{15}Cl_2FN_4O_2$ requires 373.0)

Example 156: Compound 179



Compound **179** was synthesized by the method of Example 33 using the bromopyrimidine derivative and boronic acid **23**. Standard conditions were used for the removal of protecting groups and the introduction of dichloroacetamide functionality. LRMS (ESI$^+$) m/z: 373.0 (M-H$^+$ $C_{15}H_{15}Cl_2FN_4O_2$ requires 373.0)

Example 157: Compound 180



Compound **180** was synthesized by the method of Example 33 using the requisite chloropyridazine derivative and boronic acid **23**. Standard conditions were used for the removal of protecting groups and the introduction of dichloroacetamide functionality. LRMS (ESI$^+$) m/z: 373.0 (M-H$^+$ $C_{15}H_{15}Cl_2FN_4O_2$ requires 373.0)

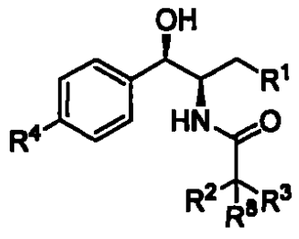
CONCLUSION

Thus, it will be appreciated that the present invention provides novel florfenicol-like compounds and methods for their use in the treatment or prevention of bacterial infection in animals or humans.

Although certain embodiments and examples have been used to describe the present invention, it will be apparent to those skilled in the art that changes in the embodiments and examples shown may be made without departing from the scope of this invention.

WHAT IS CLAIMED:

1. A compound having the chemical formula:



wherein:

R^1 is selected from the group consisting of -OH and -F;

R^2 and R^3 are independently selected from the group consisting of hydrogen, (1C – 4C)alkyl, halo, $-CF_3$, $-NH_2$, -CN and N_3;

R^4 is selected from the group consisting of:

$-C(=R^5)R^6$,

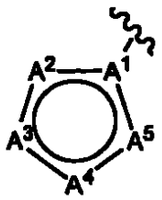
wherein:

R^5 is selected from the group consisting of oxygen, N-C≡N, and NOR^7,

wherein:

R^7 is selected from the group consisting of hydrogen, alkyl, aryl, heteroaryl, alicyclic and heteroalicyclic;

R^6 is selected from the group consisting of hydrogen, (1C – 4C)alkyl, (3C – 6C)cycloalkyl, (1C – 4C)alkoxy, heteroaryl, alicyclic and heteroalicyclic;



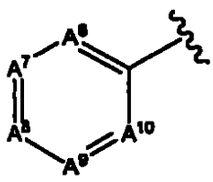
wherein:

A^1 is carbon or nitrogen;

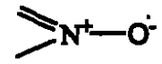
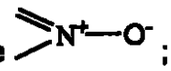
A^2, A^3, A^4 and A^5 are independently selected from the group consisting of carbon, nitrogen, oxygen and sulfur, provided that at least one of $A^1 - A^5$ is not carbon, that the total number of nitrogen, oxygen and sulfur atoms in the ring does not exceed 4 and that the ring is aromatic;

carbon atoms in the ring are independently substituted with an entity selected from the group consisting of hydrogen, (1C – 4C)alkyl, (3C – 6C)cycloalkyl, (1C – 4C)alkylO-, $-CF_3$, -OH, -CN, halo, (1C – 4C)alkylS(O)-, (1C – 4C)alkylS$(O)_2$-, NH_2SO_2-, (1C – 4C)alkylNHSO_2-, ((1C – 4C)alkyl)$_2$NSO_2-, $-NH_2$, (1C – 4C)alkylNH-, ((1C – 4C)alkyl)$_2$N-, (1C – 4C)alkylSO_2NH-, (1C – 4C)alkylC(O)-, (3C – 6C)cycloalkylC(O)-, (1C – 4C)alkylOC(O)-, (1C – 4C)alkylC(O)NH-, $-C(O)NH_2$, (1C – 4C)alkylNHC(O)- and ((1C – 4C)alkyl)$_2$NC(O)-, wherein any of the alkyl groups in any of the substituents may be unsubstituted or substituted with a group selected from halo and -OH;

if A^1 is carbon and the ring does not contain oxygen or sulfur, one of the nitrogen atoms may optionally be substituted with an entity selected from the group consisting of (1C – 4C)alkyl, (1C – 4C)alkylS$(O)_2$- and $-NH_2$;



wherein:

A^6, A^7, A^8, A^9 and A^{10} are independently selected from the group consisting of carbon, nitrogen and  , provided that only one of A^6 – A^{10} at a time can be  ;

carbon atoms in the ring are independently substituted with an entity selected from the group consisting of hydrogen, (1C – 4C)alkyl, (3C – 6C)cycloalkyl, (1C – 4C)alkylO-, $-CF_3$, -OH, -CN, halo, (1C – 4C)alkylS(O)-, (1C – 4C)alkylS$(O)_2$-, NH_2SO_2-, (1C – 4C)alkylNHSO_2-, ((1C – 4C)alkyl)$_2$NSO_2-, $-NH_2$, (1C – 4C)alkylNH-, ((1C – 4C)alkyl)$_2$N-, (1C – 4C)alkylSO_2NH-, (1C – 4C)alkylC(O)-, (3C – 6C)cycloalkylC(O)-, (1C – 4C)alkylOC(O)-, (1C – 4C)alkylC(O)NH-, $-C(O)NH_2$, (1C – 4C)alkylNHC(O)-, ((1C – 4C)alkyl)$_2$NC(O)- and $-OCH_2O$-, the oxygen atoms of the $-OCH_2O$- group being bonded to adjacent ring carbon atoms, and wherein any of the alkyl groups in any substituent may be unsubstituted or substituted with a group selected from halo and -OH;

R^8 is hydrogen in all compounds, except when R^2 and R^3 are both F, in which case R^8 is hydrogen or F; and,

the compound is either a racemate having the relative stereochemistry shown or is substantially enantiomerically pure and has the absolute stereochemistry shown.

2. The compound of claim 1, wherein R^1 is -F.

3. The compound of claim 2, wherein:

R^2 and R^3 are independently selected from the group consisting of Cl and F; and,

R^8 is hydrogen.

100
99

4. The compound of claim 3, wherein:

R^4 is $-C(=R^5)R^6$,

wherein:

R^5 is selected from the group consisting of oxygen, N-C≡N, and NOR^7,

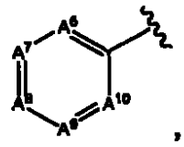
wherein:

R^7 is selected from the group consisting of hydrogen, alkyl, aryl, heteroaryl, alicyclic and heteroalicyclic; and,

R^6 is selected from the group consisting of hydrogen, (1C – 4C)alkyl, (3C – 6C)cycloalkyl, (1C – 4C)alkoxy, aryl, heteroaryl, alicyclic and heteroalicyclic.

5. The compound of claim 4, wherein R^4 is $CH_3C(O)$-.

6. The compound of claim 2, wherein:

R^4 is 

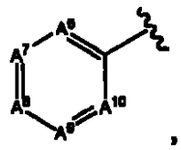
,

wherein:

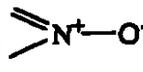
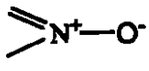
A^6, A^7, A^8, A^9 and A^{10} are as defined in claim 1; and,

any of $A^6 – A^{10}$ that is carbon is substituted with an entity selected from the group consisting of hydrogen, $-NH_2$, halo-, -CN, (1C – 4C)alkyl-, (1C – 4C)alkylC(O)-, (1C – 4C)alkylS(O)-, (1C – 4C)alkylS$(O)_2$-, NH_2SO_2-, (1C – 4C)alkylSO_2NH-, (1C – 4C)alkyl$NHSO_2$-, ((1C – 4C)alkyl)$_2NSO_2$-, wherein any alkyl group in any substituent may be unsubstituted or substituted with halo or –OH.

7. The compound of claim 3, wherein:

R^4 is  ,

wherein:

A^6, A^7, A^8, A^9 and A^{10} are independently selected from the group consisting of carbon, nitrogen and  , provided that only one of A^6 – A^{10} at a time can be  ; and,

any of A^6 – A^{10} that is carbon is substituted with an entity selected from the group consisting of hydrogen, $-NH_2$, halo-, -CN, (1C – 4C)alkyl-, (1C – 4C)alkylC(O)-, (1C – 4C)alkylS(O)-, (1C – 4C)alkylS(O)$_2$-, NH_2SO_2-, (1C – 4C)alkylSO_2NH-, (1C – 4C)alkyl$NHSO_2$-, ((1C – 4C)alkyl)$_2NSO_2$-, wherein any of the alkyl groups within any of the substituents may be unsubstituted or substituted with halo or –OH.

8. The compound of claim 2, wherein:

one, two or three of A^6 – A^{10} is/are nitrogen; and,

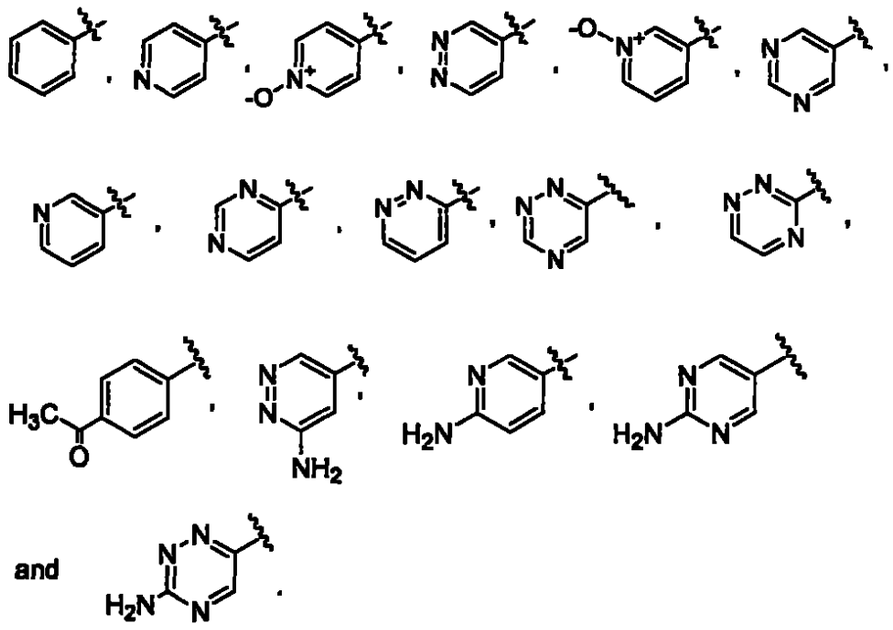
one or two of the remaining carbon atoms in the ring is/are optionally substituted with $-NH_2$, all other carbon atoms in the ring being unsubstituted.

9. The compound of claim 3, wherein:

one, two or three of A^6 – A^{10} is/are nitrogen; and,

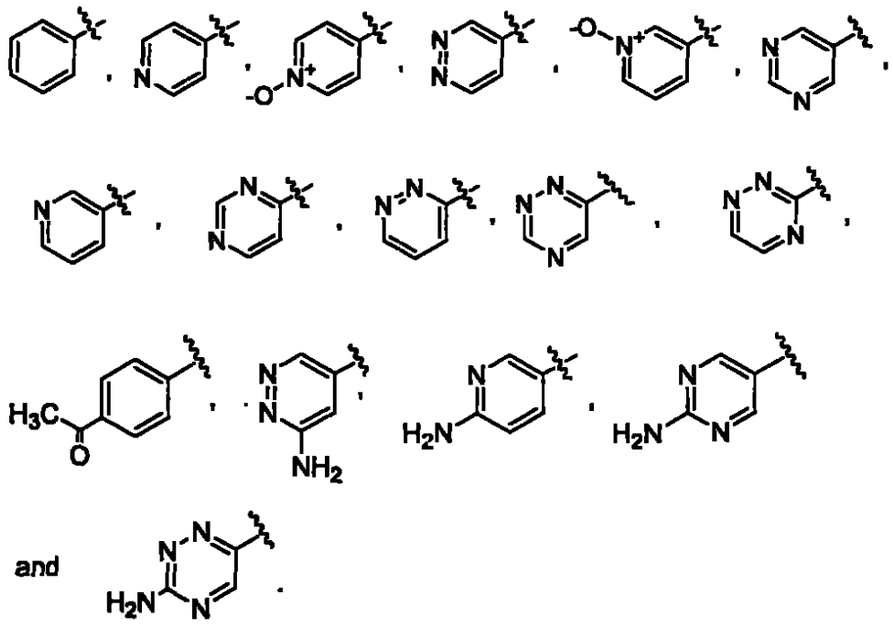
one or two of the remaining carbon atoms in the ring is/are optionally substituted with $-NH_2$, all other carbon atoms in the ring being unsubstituted.

10. The compound of claim 2, wherein R^4 is selected from the group consisting of:



and .

11. The compound of claim 3, wherein R^4 is selected from the group consisting of :



and .

12. The compound of claim 2, wherein:

R^4 is 

,

wherein A^1, A^2, A^3, A^4 and A^5 are as defined in claim 1.

13. The compound of claim 3, wherein:

R^4 is 

,

wherein A^1, A^2, A^3, A^4 and A^5 are as defined in claim 1.

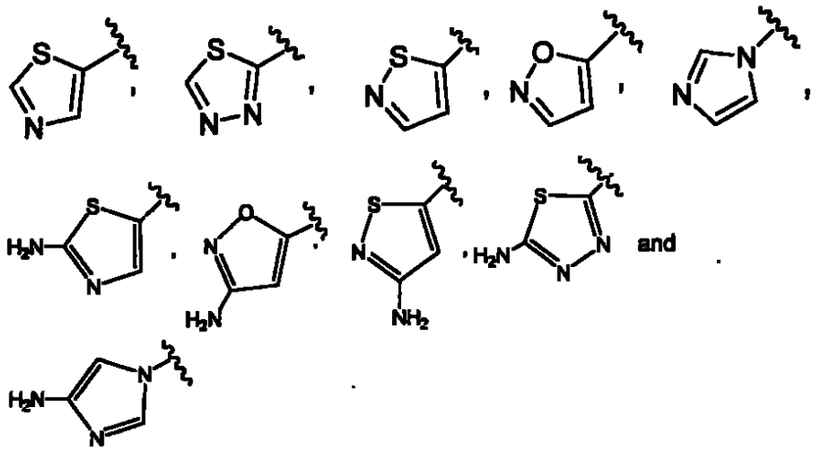
14. The compound of claim 12, wherein all carbon atoms and nitrogen atoms are unsubstituted.

15. The compound of claim 13, wherein all carbon atoms and nitrogen atoms are unsubstituted.

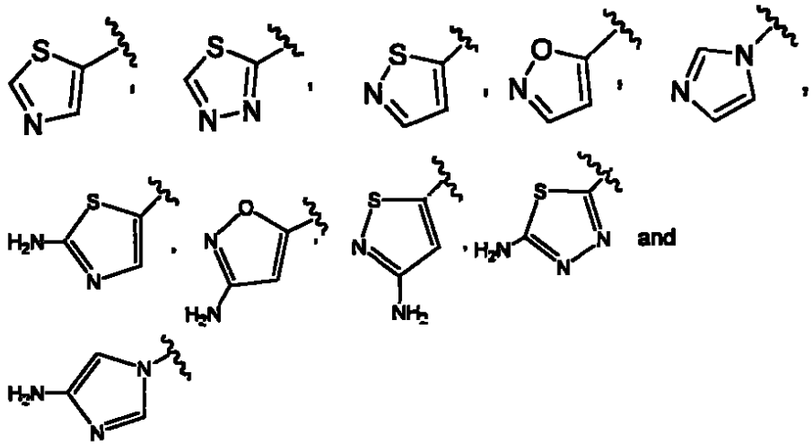
16. The compound of claim 12, wherein one of $A^2 - A^5$ that is carbon is substituted with an $-NH_2$ group, all other carbon and, if applicable, nitrogen atoms in the ring being unsubstituted.

17. The compound of claim 13, wherein one of $A^2 - A^5$ that is carbon is substituted with an $-NH_2$ group, all other carbon and, if applicable, nitrogen atoms in the ring being unsubstituted.

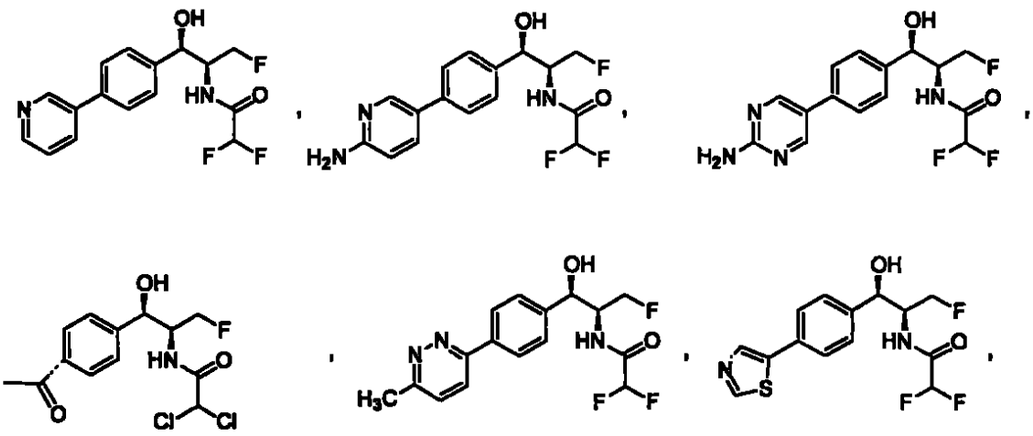
18. The compound of claim 2, wherein R^4 is selected from the group consisting of:

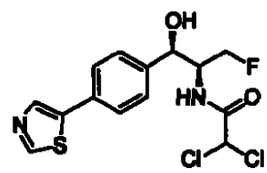
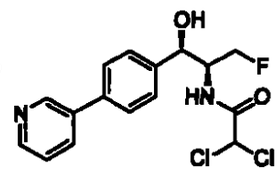
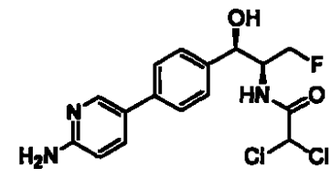
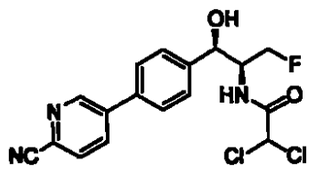
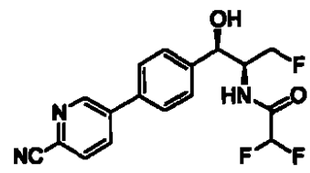
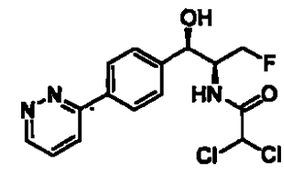
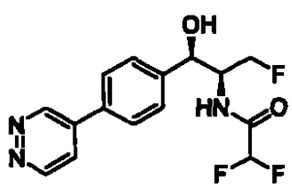
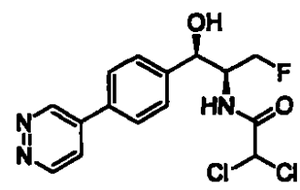
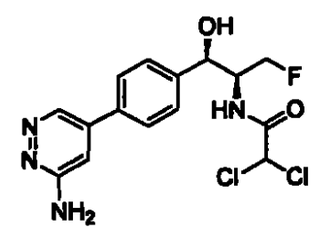
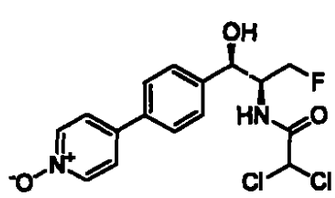
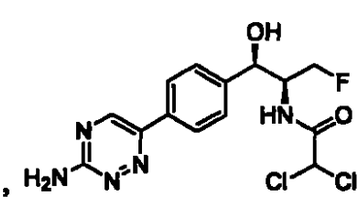
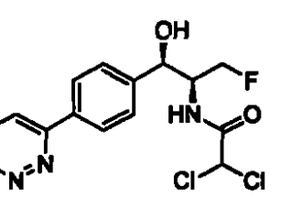
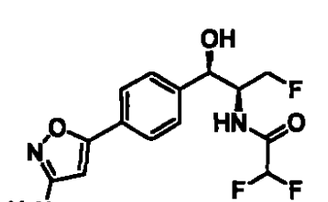
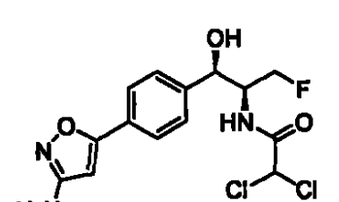
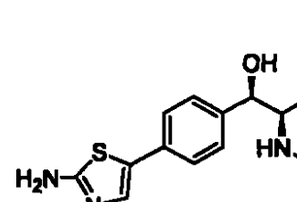


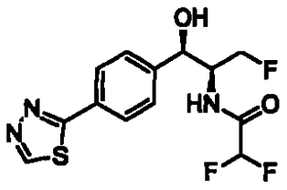
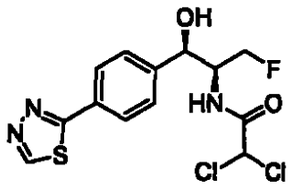
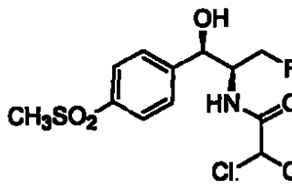
19. The compound of claim 3, wherein R^4 is selected from the group consisting of:



20. A compound of claim 1 selected from the group consisting of:

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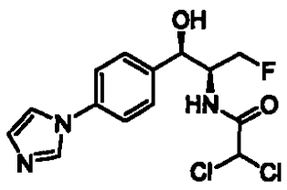
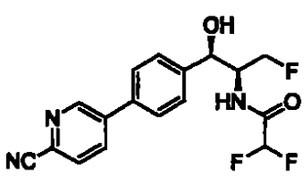
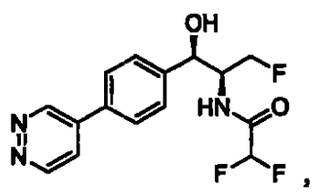
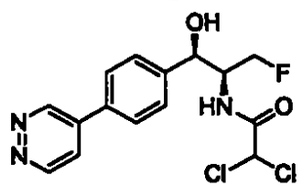
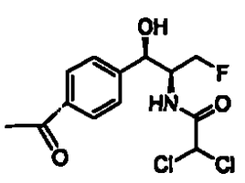
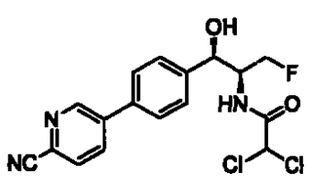
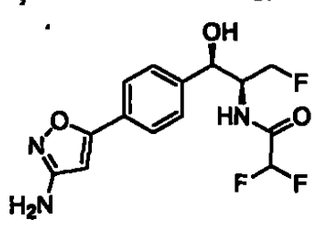
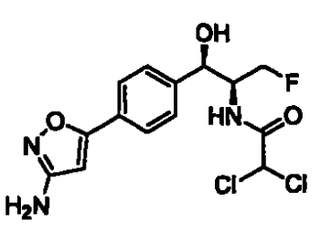
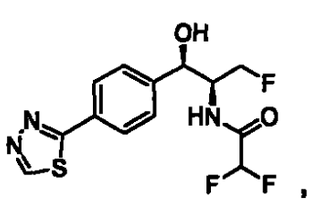
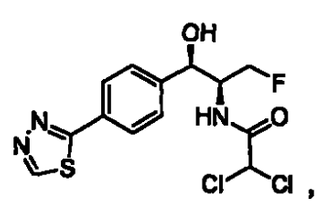
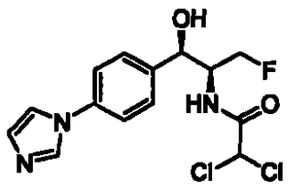


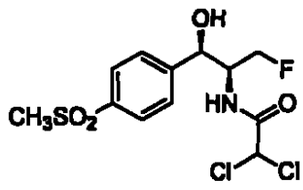
wherein the compound is either a racemate having the relative stereochemistry shown or is substantially enantiomerically pure and has the absolute stereochemistry shown.

21. A compound of claim 1 selected from the group consisting of:



, and



,

wherein the compound is either a racemate having the relative stereochemistry shown or is substantially enantiomerically pure and has the absolute stereochemistry shown.

22. The compound of claim 1, wherein the compound is substantially enantiomerically pure and has a 1-(R)-2-(S) absolute configuration.

23. A method of treating or preventing a bacterial infection, comprising administering to a patient in need thereof a pharmaceutically effective amount of a compound of claim 1.

24. The method of claim 30, wherein the bacterial infection is caused by a bacteria of the genus *Pasteurella*, *Haemophilus*, *Fusobacterium*, *Bacterioides*, *Aeromonas*, *Enterobacter*, *Escherichia*, *Klebsiella*, *Salmonella*, *Shigella*, *Actinobacillus*, *Streptococcus*, *Mycoplasma*, *Edwardsiella*, *Staphylococcus*, *Enterococcus*, *Bordetella*, *Proteus*, or *Mannheimia*.

25. The method of claim 31, wherein the bacterial infection is caused by Mannhemia haemolytica, Pasteurella multocida, Haemophilus somnus, Fusobacterium necrophorum, Bacterioides melaninogenicus, Actinobacillus pleuropneumoniae, Streptococcus suis, Salmonella cholerasuis, Mycoplasma bovis, Mycoplasma hyopneumoniae, Mycoplasma hyorhinis, Mycoplasma gallisepticum, Edwardsiella ictaluri, Escherichia coli, Enterobacter cloacae, Staphylococcus aureus, Staphylococcus intermedius, Enterococcus faecalis, Enterococcus faecium, Klebsiella pneumoniae, Klebsiella oxytoca, Enterobacter cloacae, Proteus mirabilis, or Aeromonas salmonicida.

NUEVOS ANTIBIÓTICOS TIPO FLORFENICOL

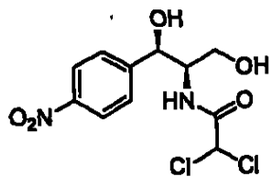
CAMPO EL INVENTO

El presente invento se relaciona con los campos de la química orgánica, química farmacéutica, bioquímica y medicina. En particular, se relaciona con nuevos antibióticos tipo florfenicol.

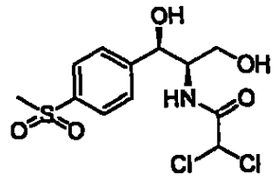
FUNDAMENTO DEL INVENTO

El florfenicol es un antibiótico de amplio espectro con una actividad contra muchas bacterias gram-negativas y gram-positivas. El florfenicol es útil para la prevención y tratamiento de infecciones bacterianas por patógenos susceptibles en aves, reptiles, peces, moluscos y mamíferos. Uno de sus usos principales está en el tratamiento de neumonía e infecciones respiratorias asociadas en ganado (a menudo genéricamente conocida como Enfermedad Respiratoria Bovina o BRD) *Mannhemia hemolitica, Pasturella multocida (o) Haemophilus somnus.* También se indica en el tratamiento de pododermatitis en un ganado producida por *Fusobacterium necrophorum y Bacterioides melaninogénicus,* enfermedad respiratoria porcina producida por *Pasteurella multocida, Actinobacillus pleuropneumoniae, Streptococcus suis, Salmonella cholerasuis* y(o) *Mycoplasma* spp., colibacilosis en pollos producida por Escherichia coli, septicemia entérica en bagre producida por *Edwardsiella ictaluri*, y furunculosis en salmón producida por *Aeromonas salmonicida.* Otros géneros de bacterias que han exhibido susceptibilidad al florfenicol incluyen *Enterobactor, Klebsiella, Staphylococcus, Enterococcus, Bordetella, Proteus y Shigella.* En particular, las cepas resistentes a clorafenicol de organismos tales como *K.pneumoniae, E. cloacae, S. typhus* y *E. coli* son susceptibles a florfenicol.

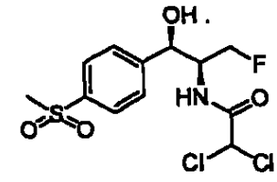
El florfenicol es un análogo estructural de tianfenicol, el cual a su vez es un derivado de cloranfenicol en el cual el grupo nitro aromático, cuyo grupo nitro ha sido implicado en



chloramphenicol



thiamphenicol



florfenicol

anemia aplástica irreversible no relacionada con dosis inducida por clorafenicol en humanos, es reemplazado con un grupo metilsulfonilo. El florfenicol tiene un átomo de flúor en cambio del grupo hidróxilo primario de cloranfenicol y tianfenicol. Este hace al florfenicol menos susceptible a desactivación mediante bacterias que contienen la enzima codificada mediante plásmida, cloranfenicol acetil transferasa (CAT) la cual acetila el grupo hidróxilo primario de cloranfenicol y tianfenicol, evitando así la unión de estos a subunidades ribosomales de bacterias susceptibles. La unión ribosomal es el principal mecanismo de acción de los antibióticos de cloranfenicol y produce la inhibición de la peptidilo transferasa, la cual es responsable de la transferencia de aminoácidos hacia cadenas peptídicas crecientes y subsecuente formación proteica en bacterias. Sin embargo, los compuestos y que tienen el grupo hidróxilo primario tienen utilidad en el tratamiento de

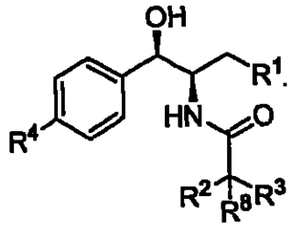
infecciones bacterianas, tal como se evidenció por el uso continuo de cloranfenicol y tianfenicol en todo el mundo.

En años recientes, un número de géneros y especies bacterianas han comenzado a exhibir alguna resistencia al florfenicol. Por ejemplo, se ha observado resistencia en especies de salmonella (Bolton, L.F., *Clin. Microbiol.* 1999, 37, 1348), *E. coli* (Keyes, K., *Antimicrob.* Agents Chomother., 2000, 44, 421.), *Klebsiella pneumoniae* (Cloeckaert, A., *Antimicrob. Agents Chemother.*, 2001, 45, 2381), y el patogeno acuicultural, *Photobacterium damselae subsp. piscicida* (anteriormente *Pasteurella piscicida*) (Kim, E., Microbiol. Immunol., 1996, *40, 665).* Esta resistencia ha sido rastreada hasta un gen altamente conservado (*flo*) que produce una bomba de efusión antibiótica (Flo).

La aparición y la amenaza de diseminación, de resistencia al florfenicol ha fomentado la necesidad de nuevos antibióticos que retengan o excedan la actividad del florfenicol, mantengan su impermeabilidad a la enzima CAT y, además, no sean substratos para la bomba de efusión Flo. Los compuestos del presente invento son tales antibióticos.

RESUMEN

Por lo tanto, una realización de este invento es un compuesto con la formula química:



en donde:
R1 es seleccionado del grupo que consta de -OH y -F;
R2 y R3 son independientemente seleccionados del grupo que consta de hidrógeno, (1C-4C)alquilo, halo, -CF3, -NH2, -CN y N3;
R4 es seleccionado del grupo que consta de:
-C(=R6)R6,

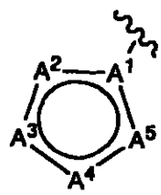
en donde:

R5 es seleccionado del grupo que consta de oxígeno, N-C=N, y NOR7,

en donde:

R7 es seleccionado del grupo que consta de hidrógeno, alquilo, arilo, heteroarilo, alicíclico y heteroalicíclico;

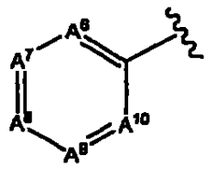
R6 es seleccionado del grupo que consta de hidrógeno, (1C-4C)alquilo, (3C-6C)cicloalquilo, (1C-4C)alcoxilo, arilo, heteroarilo, alicíclico y heteroalicíclico;



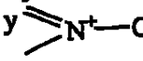
en donde:

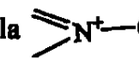
A1 es carbono o nitrógeno;

A2, A3, A4 y A5 son independientemente seleccionados del grupo que consta de carbono, nitrógeno, oxígeno y azufre siempre y cuando al menos uno entre A1 - A5 no sea carbono, que el número total de átomos de nitrógeno, oxígeno y azufre en el anillo no exceda 4 y que el anillo sea aromático; los átomos de carbono en el anillo son independientemente sustituidos con una entidad seleccionada del grupo que consta de hidrógeno, (1C-4C)alquilo, (3C-6C)cicloalquilo, (1C-4C)alquilO, -CF3, OH, -CN, halo, (1C-4C)alquilS(O)-, (1C-4C)alquilS(O)2, NH2SO2-, (1C-4C)alquilNHSO2, ((1C-4C)alquilo)2NSO2-, NH2, (1C-4C)alquilNH, ((1C-4C)alquilo)2N, (1C-4C)alquilSO2NH, (1C-4C)alquilC(O), (3C-6C)cicloalquilC(O), (1C-4C)alquilOC(O)-, (1C-4C)alquilC(O)NH-, -C(O)NH2, (1C-4C)alquilNHC(O)-, y (alquilo(1C-4C))2NC(O)-, en donde cualquiera de los grupos alquilo en cualquiera de los substituyentes puede opcionalmente ser substituido con un grupo seleccionado de halo y -OH; si A1 es carbono y el anillo no contiene oxígeno o azufre, uno de los átomos de nitrógeno puede opcionalmente ser substituido con una entidad seleccionada del grupo que consta de (1C-4C)alquilo, (1C-4C)AlquilS(O)2, y -NH2;



en donde

A6, A7, A8, A9 y A10 son independientemente seleccionados del grupo que consta de carbono, nitrógeno y  , siempre y cuando solamente uno entre A8 - A10 a

la vez pueda ser formula  ;

los átomos de carbono en el anillo son independientemente sustituidos con una entidad seleccionada del grupo que consta de hidrógeno, (1C-4C)alquilo, (3C-6C)cicloalquilo, (1C-4C)alquilO, -CF3, OH, -CN, halo, (1C-4C)alquilS(O)-, (1C-4C)alquilS(O)2, NH2SO2-, (1C-4C)alquilNHSO2, ((1C-4C)alquilo)2NSO2-, NH2, (1C-4C)alquilNH, ((1C-4C)alquilo)2N, (1C-4C)alquilSO2NH, (1C-4C)alquilC(O), (3C-6C)cicloalquilC(O), (1C-4C)alquilOC(O)-, (1C-4C)alquilC(O)NH-, -C(O)NH2, (1C-4C)alquilNHC(O)-, (alquilo(1C-4C))2NC(O)-, y OCH2O-, los átomos de oxígeno en el substituyente -OCH2O- son unidos a los átomos de carbono del anillo adyacente, en donde cualquiera de los grupos alquilo en cualquiera de los sustituyentes puede opcionalmente ser sustituido con un grupo seleccionado de halo y -OH;

R8 es hidrógeno en todos los compuestos, excepto cuando R2 y R3 son ambos F, en cuyo caso R8 es hidrógeno o F; y, el compuesto es un racemato que tiene la estereoquímica relativa mostrada o es sustancialmente enantioméricamente puro y tiene la estereoquímica absoluta mostrada.

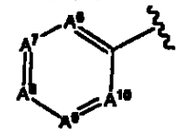
En una realización de este invento, R1 es -F.

En una realización de este invento, R2 y R3 son independientemente seleccionados del grupo que consta de Cl y F.

En una realización de este invento, R8 es hidrógeno.

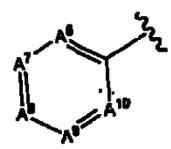
En una realización de este invento, R4 es -C(=R5)R6 en donde R5 y R6 son tal como se definieron antes.

En una realización de este invento, R4 es CH3C(O)-;



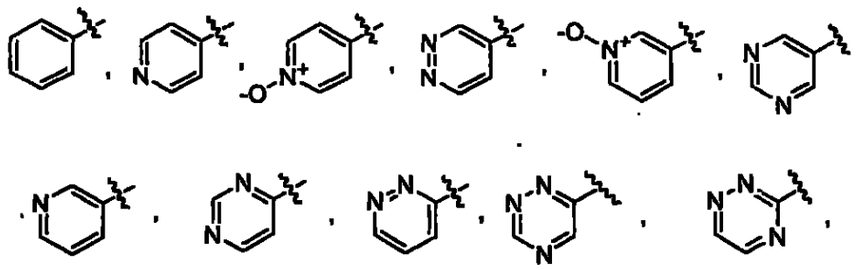
En una realización de este invento, R4 es , en donde:

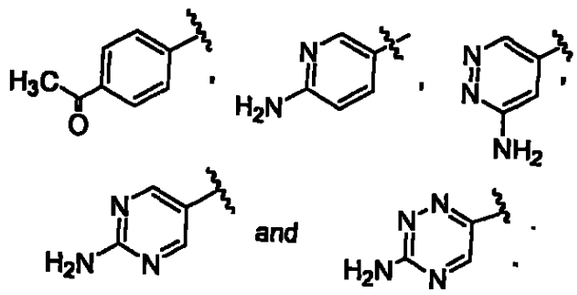
A6, A7, A8, A9 y A10 son tal como se definieron anteriormente, y, cualquiera entre A6 - A10 que es carbono es substituido con una entidad que es seleccionado del grupo que consta de hidrógeno, -NH2, halo, -CN, (1C-4C)alquilo-, (1C-4C)alquiloC(O)-, (1C-4C)alquilS(O)-, (1C-4C)alquilS(O)2-, NH2SO2-, (1C-4C)alquilSO2NH-, (1C-4C)alquilNHSO2-, ((1C-4C)alquilo)2NSO2-, en donde cualquiera de los grupos alquilo en cualquiera de los sustituyentes puede opcionalmente ser sustituido con halo o -OH.

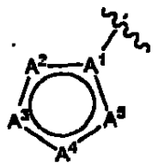
En una realización de este invento, R4 es  y uno, dos o tres de A6 – A10 es/son nitrógeno; y,

uno o dos de los restantes átomos de carbono en el anillo es/son opcionalmente sustituidos con -NH2, todos los demás átomos en el anillo no son sustituidos.

En una realización actualmente preferida de este invento, R4 es seleccionado del grupo que consta de:



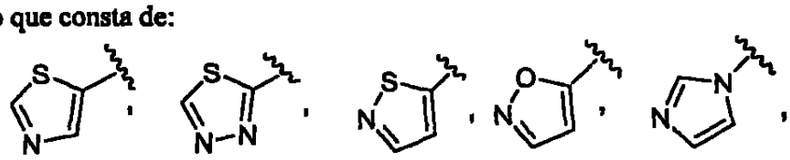


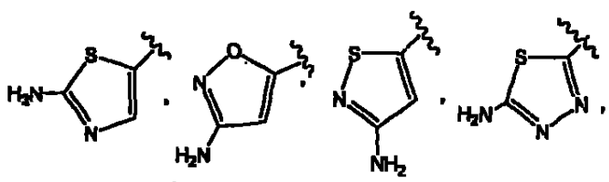
En una realización de este invento, R4 es  tal como se definió anteriormente.

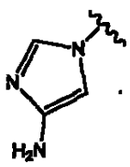
En una realización de este invento, R4 es  y todos los átomos de carbono y átomos de nitrógeno no son sustituidos

En una realización de este invento, R4 es  y uno entre A2 - A5 que es carbono es substituido con un grupo -NH2, todos los demás átomos de carbono y, si es aplicable, de nitrógeno en el anillo no son substituidos.

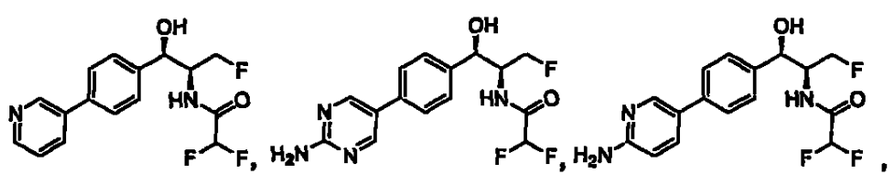
En una realización actualmente preferida de este invento, R4 es seleccionado del grupo que consta de:

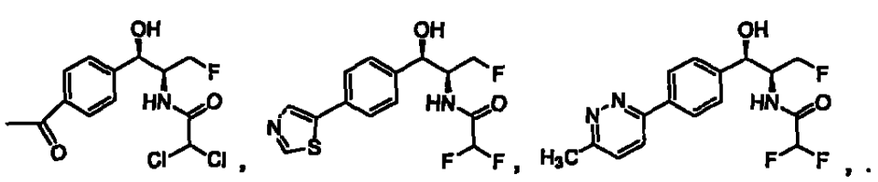


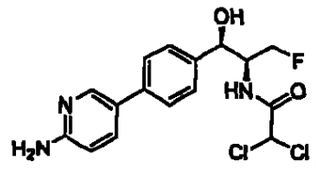
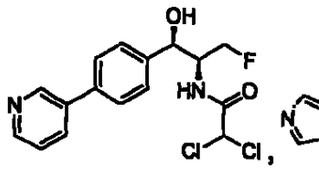
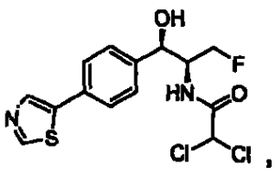


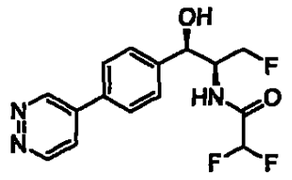
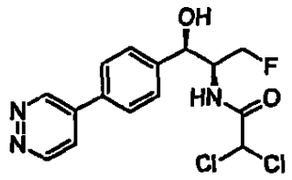
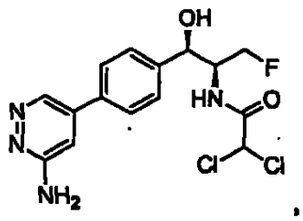
and 

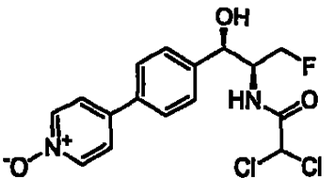
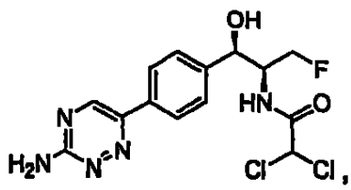
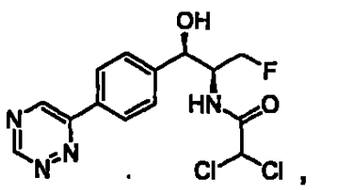
Una realización actualmente preferida de este invento es un compuesto seleccionado del grupo que consta de:

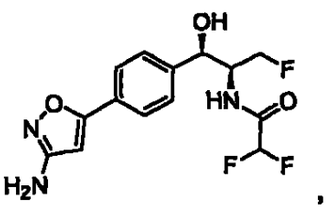
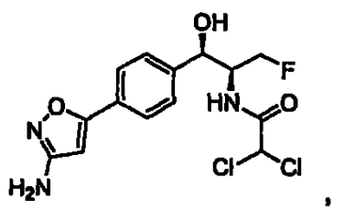
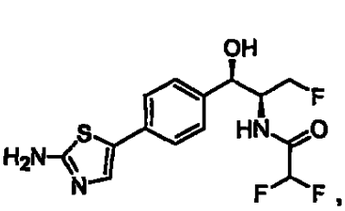


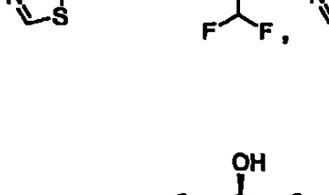
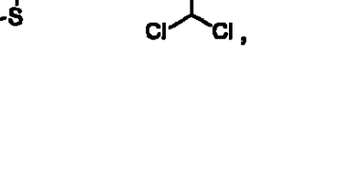
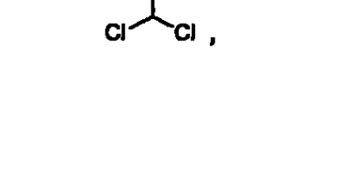


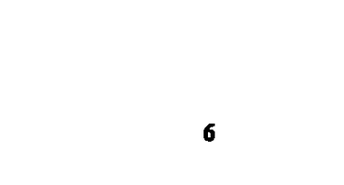




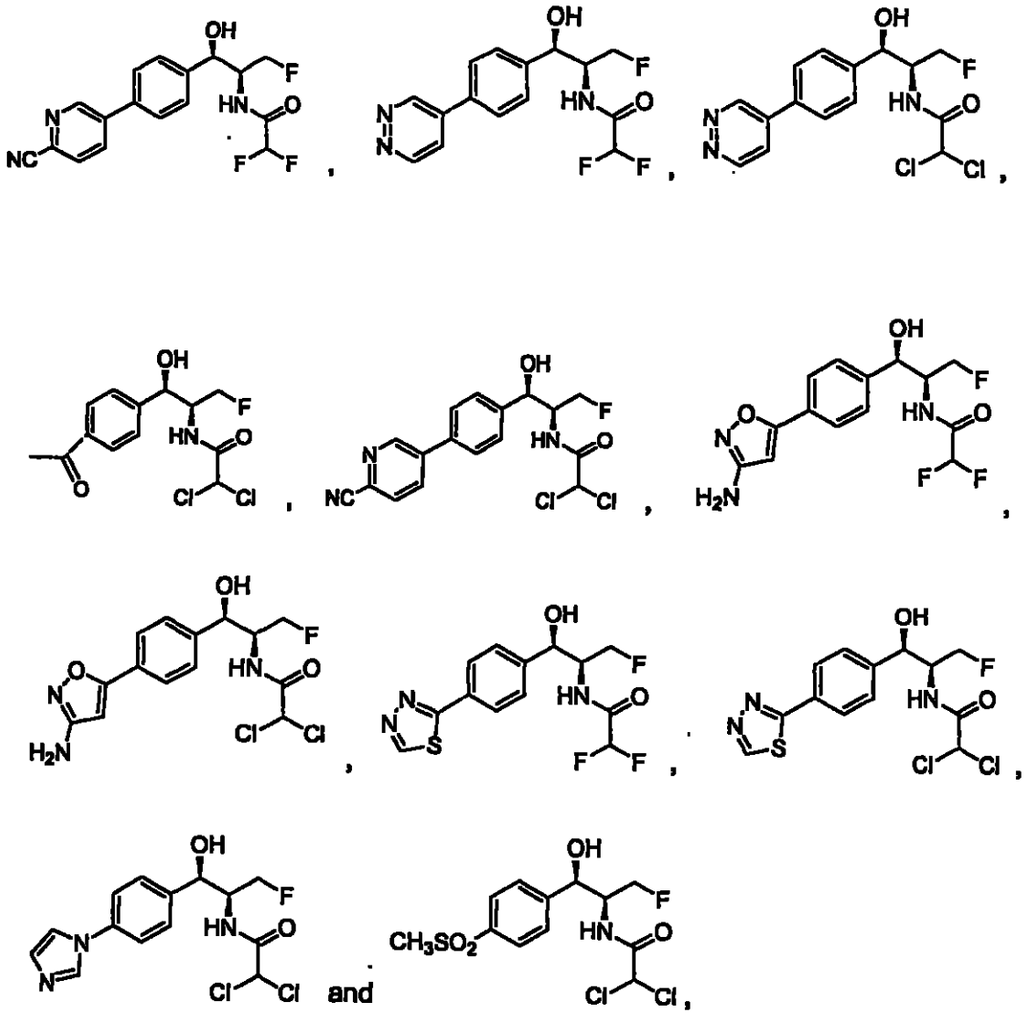






en donde el compuesto es un racemato que tiene la estereoquímica relativa mostrada o es sustancialmente enantioméricamente puro y tiene la estereoquímica absoluta mostrada.

Una realización actualmente particularmente preferida de este invento es un compuesto seleccionado del grupo que consta de:



en donde el compuesto es un racemato que tiene la estereoquímica relativa mostrada o es sustancialmente enantioméricamente puro y tiene la estereoquímica absoluta mostrada.

En otra realización particularmente preferida de éste invento, el compuesto en la presente es sustancialmente enantiomericamente puro y tiene una configuración absoluta 1-(R)-2-(S).

Una realización de este invento es un método para tratar o prevenir una infección bacteriana, que comprende la administración a un paciente en necesidad del mismo de una cantidad farmacéuticamente efectiva de un compuesto.

En una realización de este invento, la infección bacteriana es producida por una bacteria del género *Pasteurella, Haemophilus, Fusobacterium, Bacterioides, Aeromonas, Enterobacter, Escherichia, Klebsiella, Salmonella, Shigella, Actinobacillus, Streptococcus, Mycoplasma, Edwardsiella, Estaphylococcus, Enterococcus, Bordetella, Ptroteus, o Mannheimia.*

En una realización de este invento la infección bacteriana es producida por *Mannhemia haemolítica, Pasteurella multocida, Haemophilus somnus, Fusobacterium*

necrophorum, Bacterioides melaninogenicus, Actinobacillus pleuropneumoniae, Streptococcus suis, salmonella cholerasuis, Mycoplasma bovis, Mycoplasma hyopneumoniae, Mycoplasma hyorhinis, Mycoplasma gallisepticum, Edwardsiella ictaluri, Escherichia coli, Enteroacter cloacae, Staphylococcus aureus, Staphylococcus intermedius, Enterococcus faecalis, Enterococcus faecium, Klebsiella pneumoniae, Klebsiella oxytoca, Enterobacter cloacae, Proteus mirabilis, o Aeromonas salmonicida.

DESCRIPCION DETALLADA DEL INVENTO

Breve descripción de las tablas

La Tabla 1 muestra estructuras de compuestos representativos de este invento. La tabla y los compuestos en la misma no pretenden ni se deben interpretar como limitación de este invento en forma alguna.

La Tabla 2 es una lista de los microorganismos contra los cuales los compuestos de este invento fueron probados. La lista no pretende ni se debe interpretar como una limitación del alcance del invento en forma alguna.

Definiciones

Como se utiliza la presente, "halo" se refiere a flúor, cloro, bromo o yodo.

Como se utiliza en la presente, "alquilo" se refiere a un hidrocarburo saturado (no contiene enlaces multiples) alifático (sistema pi-electrónico no deslocalizado) (conteniendo si no es substituido solamente carbono e hidrógeno). La designación (n1C-n2C)alquilo, en donde n1 y n2 son teneros entre 1 - 6, se refiere a un alquilo cadena lineal o cadena ramificada que comprende entre n1 y n2 átomos de carbono. Por ejemplo, (1C - 4C)alquilo se refiere a CH3-, CH3CH2-, CH3CH2CH2-, CH3CH(CH3)-, CH3CH2CH2CH2-, CH3CH2CH(CH3)-, o (CH3)3C-. El grupo alquilo puede ser substituido o no substituido con una o más fracciones seleccionadas del grupo que consta de halo, OH, OCH3, y -C≡N.

Como se utiliza en la presente, "cicloalquilo" se refiere a un anillo multicíclico fusionado o cíclico constituido enteramente de carbonos, el cual aunque puede contener uno o más enlaces dobles, mantiene un carácter esencialmente alifático; es decir, los enlaces dobles no interactúan para formar un sistema pi-electrónico deslocalizado alrededor del anillo. Para propósitos de este invento, el anillo puede contener hasta 7 átomos de carbono. La designación (3C-6C)cicloalquilo se refiere a anillos de 3, 4, 5, y 6 miembros enteramente de átomos de carbono. Como se utiliza en la presente, "fusionado" significa que dos grupos cicloalquilo comparten al menos un átomo cíclico entre estos. Por lo tanto, compuestos tales como spiro[4,4]nonano son considerados "fusionados" para los propósitos de este invento. Más comúnmente, los anillos fusionados comparten dos átomos de carbono cíclicos adyacentes. Un ejemplo de tal sistema fusionado es decalina. Un anillo de cicloalquilo puede ser substituido o no substituido con una fracción seleccionada del grupo que consta de -OH, -OCH3, halo y -C≡N.

Como se utiliza en la presente, "arilo" se refiere a un anillo de seis miembros enteramente de carbono o dos anillos de seis miembros fusionados, el anillo o anillos fusionados con un sistema pi-electrónico deslocalizado. Por "fusionado" se entiende que cada anillo del sistema comparte dos átomos de carbono cíclicos adyacentes con al menos otro anillo. Un anillo de arilo puede ser substituido o no substituido con uno o más fracciones seleccionadas del grupo que consta de -OH, -OCH3, halo y -C≡N.

Como se utiliza la presente, "heteroarilo" se refiere a un anillo de cinco miembros o seis miembros o a dos anillos, es decir dos anillos de cinco miembros, dos anillos de seis miembros o un anillo de cinco miembros y un anillo de seis miembros fusionados juntos en donde el anillo o el anillo fusionado tiene un sistema pi-eletrónico deslocalizado. Si un anillo es de seis miembros, este debe constar solamente de carbono y nitrógeno y puede contener entre 1 y 4 átomos de nitrógeno. Presión anillo es de cinco miembros, este debe contener un átomo de nitrógeno, oxígeno o azufre y puede contener uno, dos o tres átomos adicionales de nitrógeno. Un anillo de cinco miembros con un círculo en el centro indica que el anillo es heteroaromático. El círculo es utilizado para enfatizar el hecho que la ubicación de enlaces dobles que participan en la composición del anillo heteroaromático no es estática, en cambio dependerá de la naturaleza de los átomos que forman el anillo, es decir, si son carbono, nitrógeno, oxígeno o azufre y que grupos, si los hay, están unidos a los mismos. La estructura actual de cualquier heteroaromático de cinco miembros será inmediatamente manifiesta a aquellos con experiencia en el estado de la técnica una vez que los átomos cíclicos son designados. Con respecto a grupos heteroaromáticos, el termino fusionado tiene el mismo significado que en el caso de grupos arilo. Un grupo heteroarilo puede ser substituido o no substituido con cualquiera las fracciones descritas anteriormente con respecto a grupos arilo.

Como se utiliza en la presente, "heteroalicíclico" se refiere a un sistema cíclico o cíclico fusionado que contiene átomos seleccionados del grupo que consta de carbono, nitrógeno, oxígeno y azufre pero sin sistema pi-electrónico deslocalizado. "Fusionado" tiene el mismo significado divulgado con respecto a anillos de cicloalquilo. Igualmente, un anillo heteroalicíclico puede ser substituido o no substituido con las mismas fracciones descritas anteriormente para anillos de cicloalquilo.

Cuando quiera que un átomo de carbono cíclico es declarado como "no substituido," se entiende que cualquier valencia sin llenar es en realidad ocupadas por átomos de hidrógeno. Igualmente, si un átomo de nitrógeno cíclico es capaz de ser adicionalmente substituido y es declarado como no substituido, significa que el nitrógeno está unido a un átomo de hidrógeno.

Como se utiliza en la presente, "estereoquímica absoluta" se refiere al posicionamiento exacto de substituyentes en el espacio tridimensional tal como se determinó mediante las reglas de Cahn-Ingold-Prelog, la aplicación de estas es bien conocida por aquellos con experiencia en el estado la técnica.

Como se utiliza en la presente, "enantiomero" se refiere a una de las dos configuraciones estereoquímicas absolutas de una molécula que rota la luz polarizada plana en una dirección o la otra (es decir, en dirección contraria a las manecillas del reloj desde su eje original, convencionalmente llamada "izquierda," o en dirección de las manecillas del reloj, convencionalmente conocida como "derecha"). Por "substancialmente enantiomericamente puro" se entiende que el compuesto consta de más de 90% del enantiomero, preferiblemente más de 95%, y lo más preferiblemente más del 99%.

Como se utiliza en la presente, un "racemato" se refiere a una mezcla 1:1 de los dos enantiomeros de un compuesto. Las mezclas racémicas son designadas por un indicador (+/-). Compuestos substancialmente enantiomericamente puros son mostrados sin el indicador.

Como se utiliza en la presente, "paciente" se refiere a aves, reptiles, peces, moluscos, y mamíferos. En particular se refiere a aves tales como, sin limitación, pollos y pavos, peces tales como, sin limitación, salmón, trucha, bagre y pez de cola amarilla,

mamíferos tales como, sin limitación, gatos, perros, conejos, ovejas, vacas, cerdos, caballos y cabras y seres humanos.

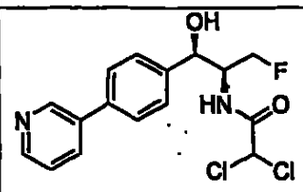
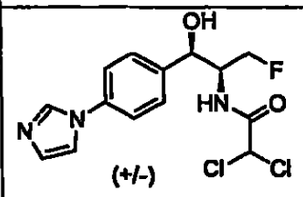
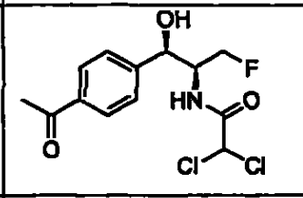
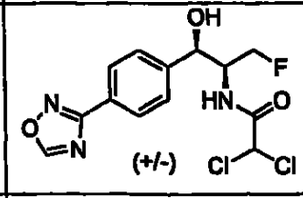
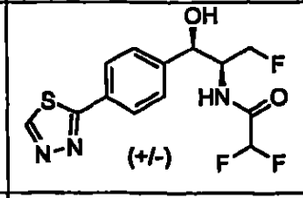
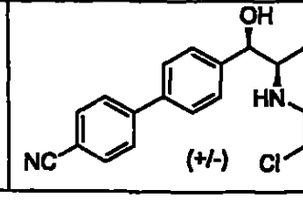
Discusión

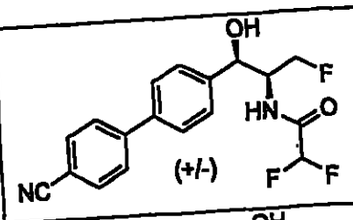
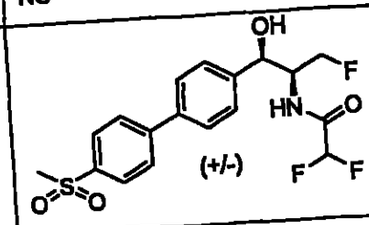
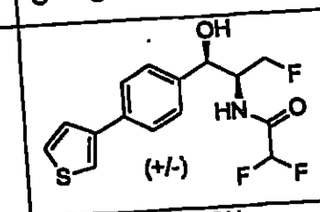
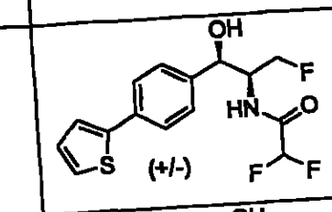
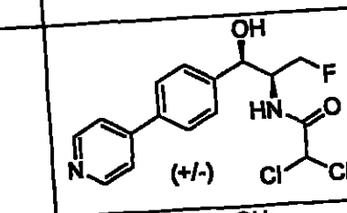
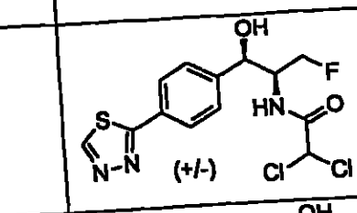
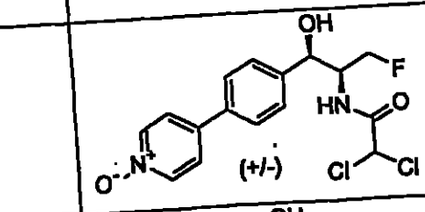
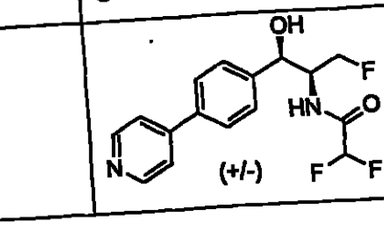
Se espera que los compuestos de este invento sean útiles para el tratamiento de infecciones bacterianas en pacientes.

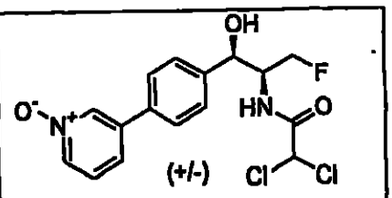
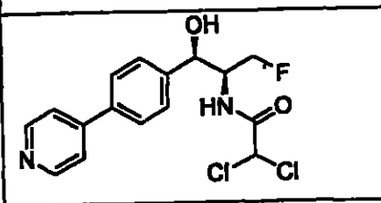
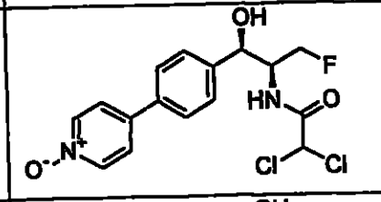
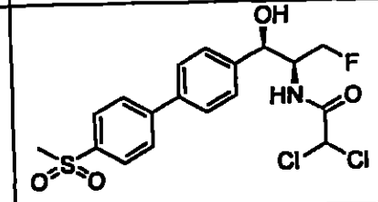
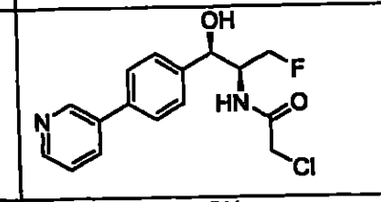
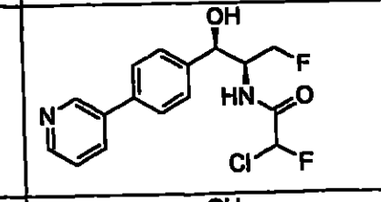
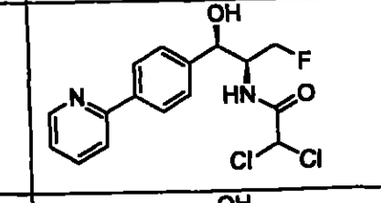
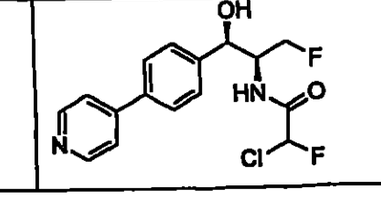
Compuestos

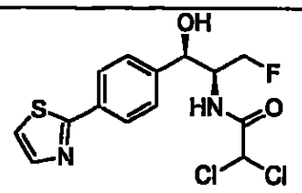
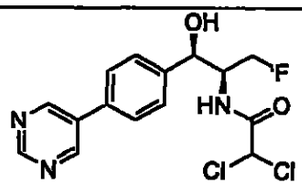
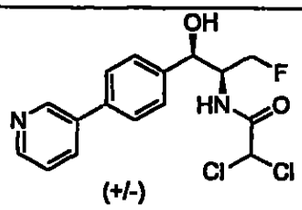
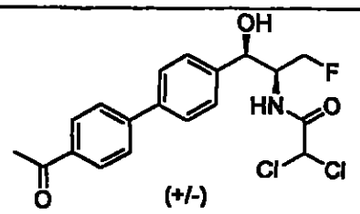
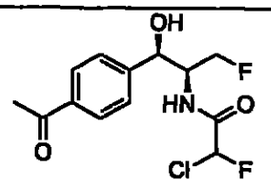
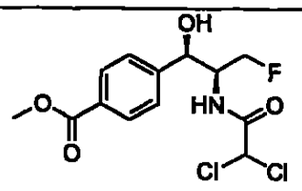
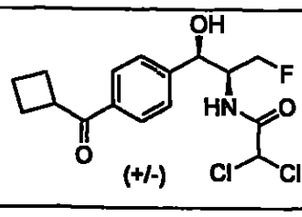
Los compuestos del presente invento son divulgados generalmente en el resumen anterior. Compuestos de ejemplo de este invento son mostrados en la tabla 1. Ni la tabla ni los compuestos expuestos pretenden limitar el alcance de este invento de ninguna forma.

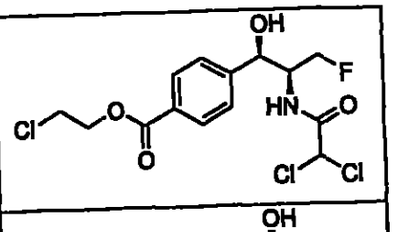
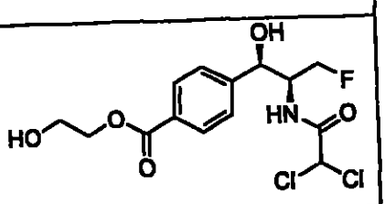
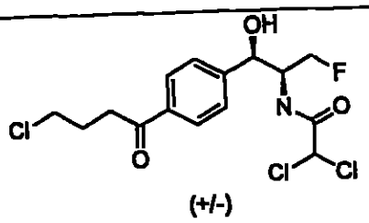
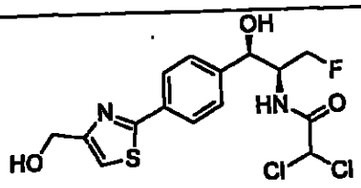
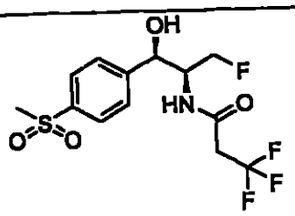
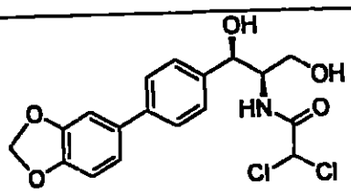
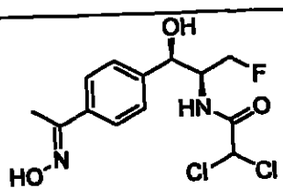
TABLE 1

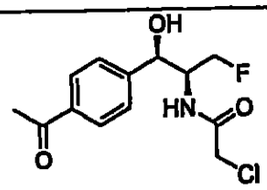
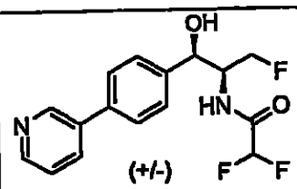
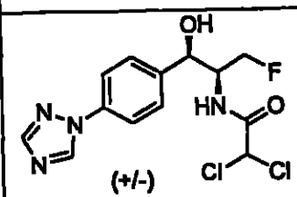
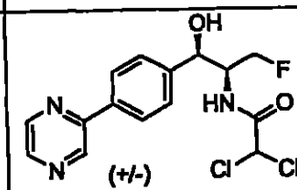
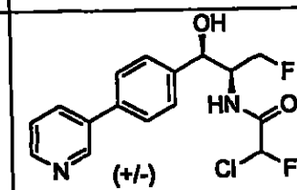
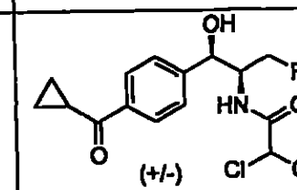
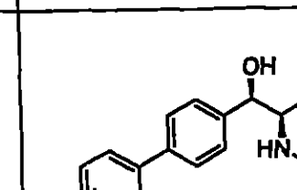
Compound #	Structure
28	
29	
41	
47	
48	
49	

50	
51	
52	
53	
54	
55	
56	
57	

58	
59	
60	
61	
62	
63	
64	
65	

66	
67	
68	 (+/-)
69	 (+/-)
71	
72	
73	 (+/-)

74	
75	
76	 (+/-)
77	
78	
79	
80	

81	
82	
84	
85	
86	
91	
92	
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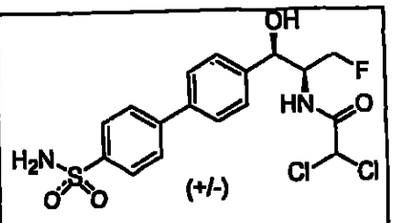
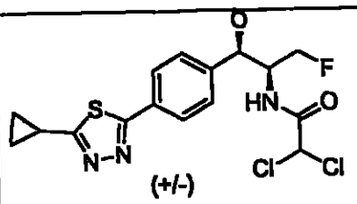
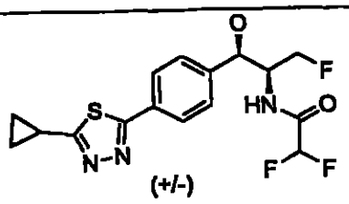
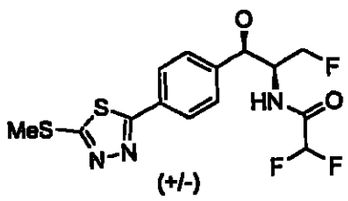
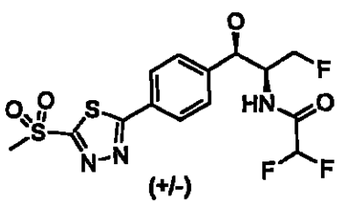
	 (+/-)
94	 (+/-)
95	 (+/-)
97	 (+/-)
98	 (+/-)
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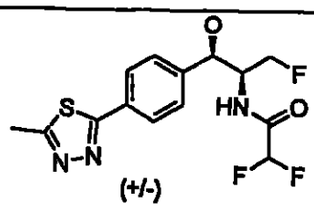
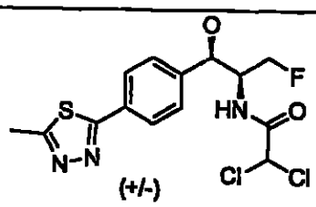
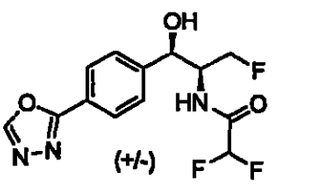
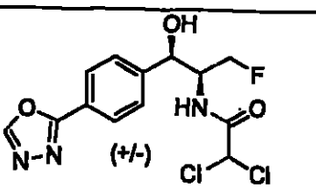
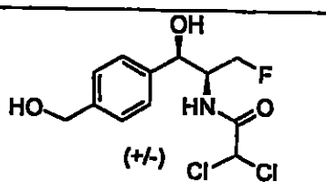
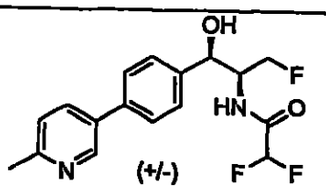
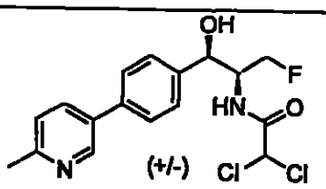
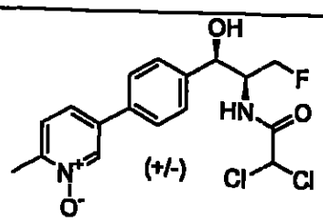
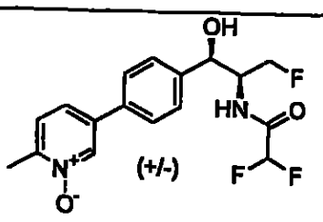
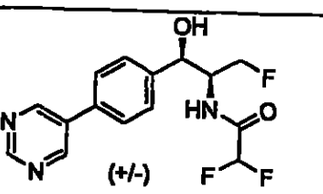
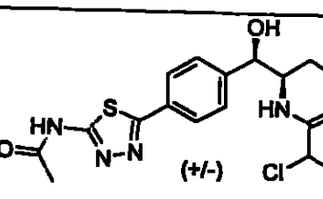
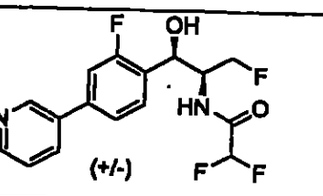
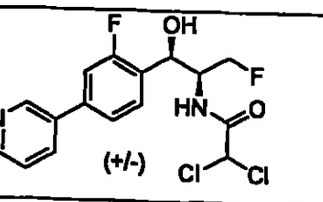
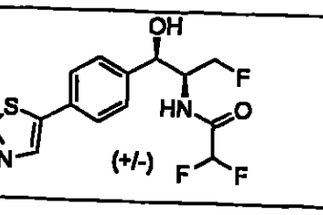
	
101	
106	
107	
109	
110	
111	

112	
113	
114	
115	
116	
117	
118	

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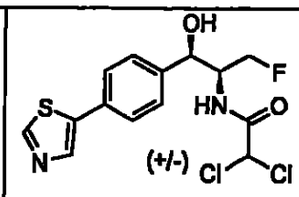
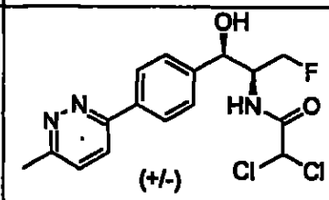
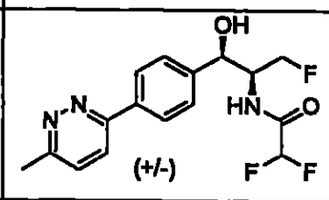
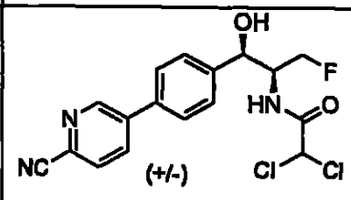
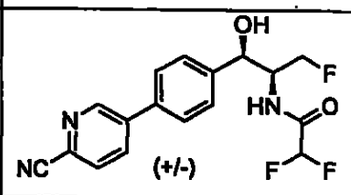
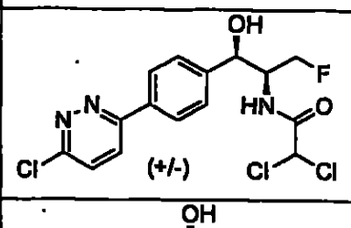
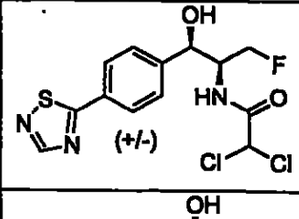
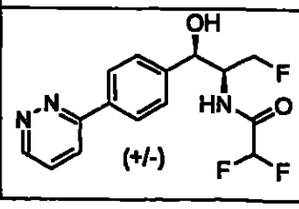
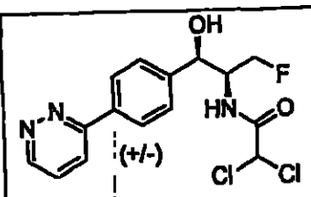
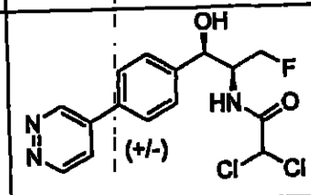
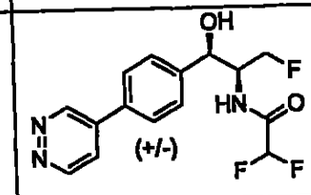
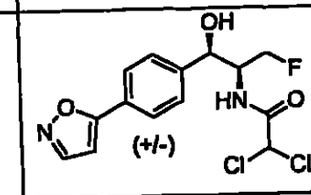
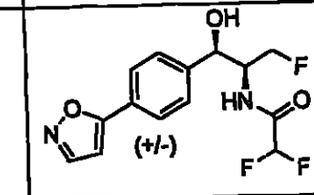
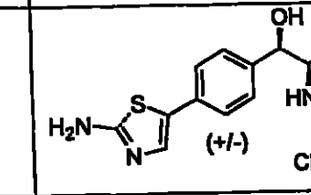
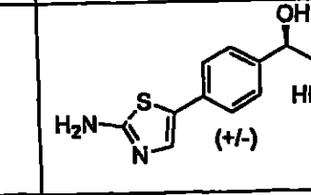
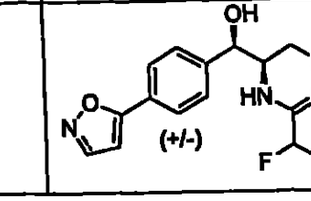
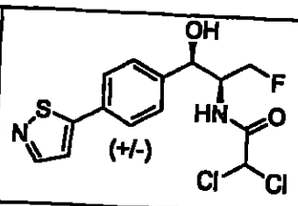
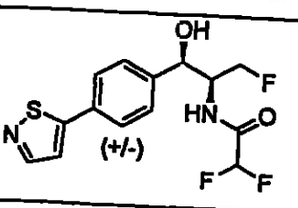
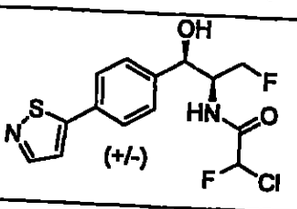
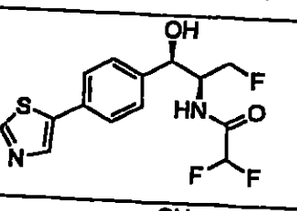
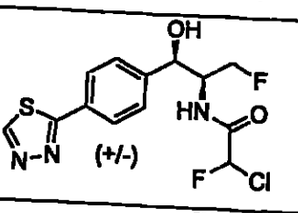
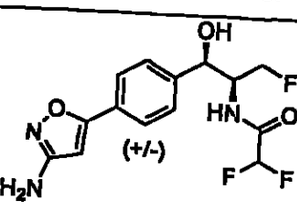
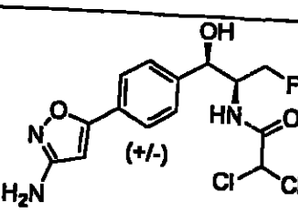
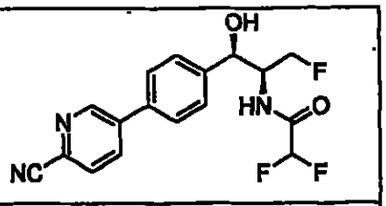
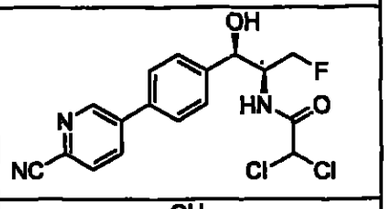
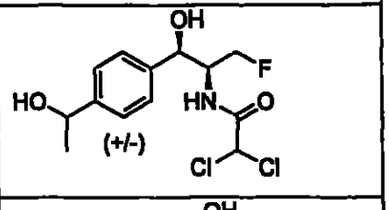
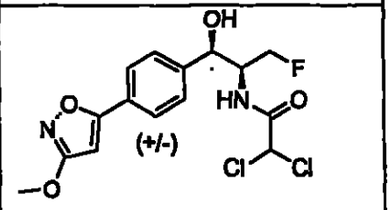
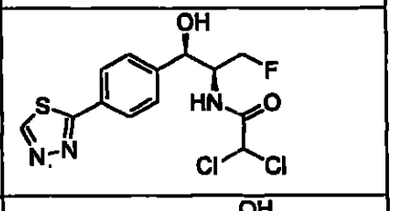
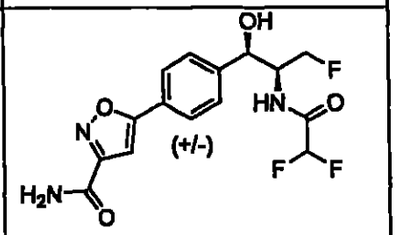
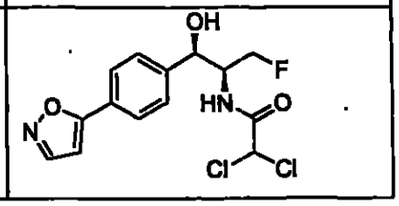
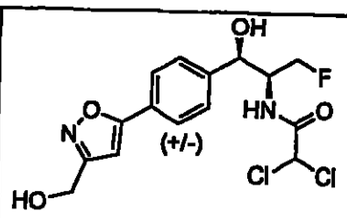
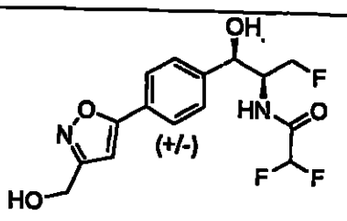
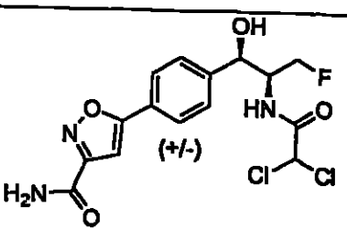
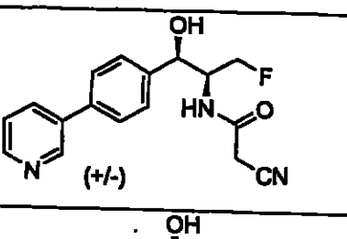
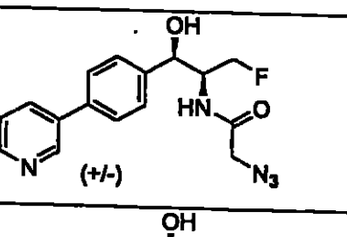
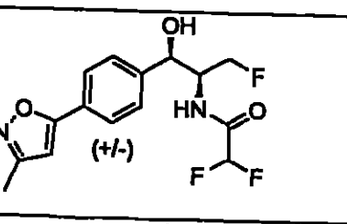
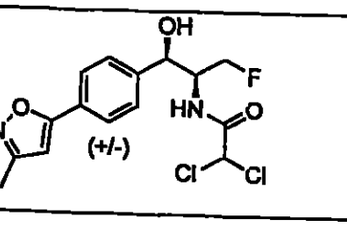
120	
121	
122	
123	
124	
125	
126	
127	

128	
129	
130	
131	
132	
133	
134	
135	

136	
137	
138	
139	
140	
141	
142	

143	
144	
145	
146	
147	
148	
149	

150	
151	
152	
153	
154	
155	
156	

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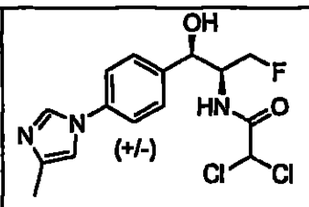
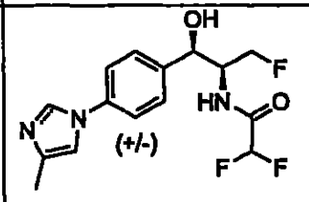
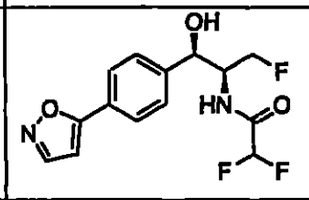
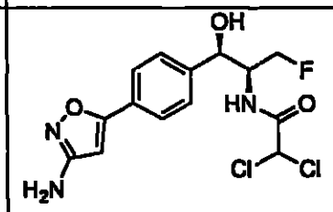
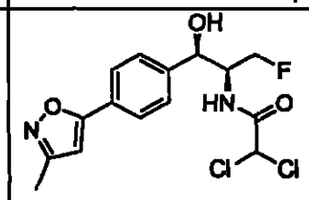
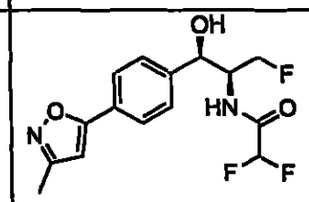
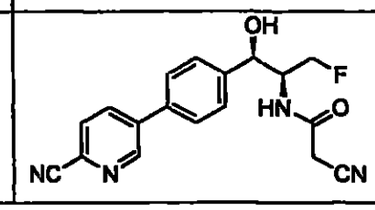
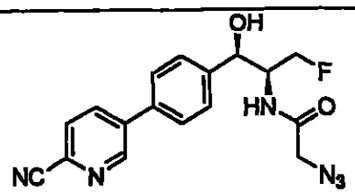
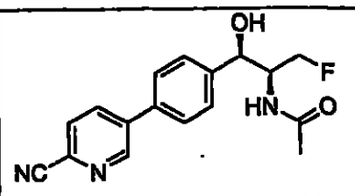
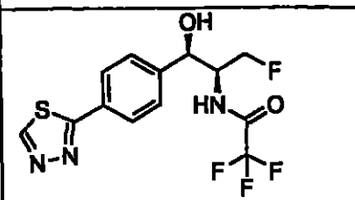
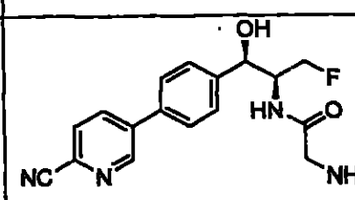
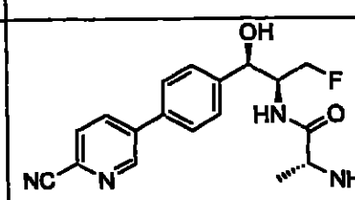
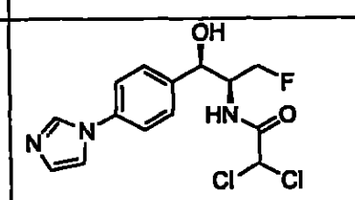
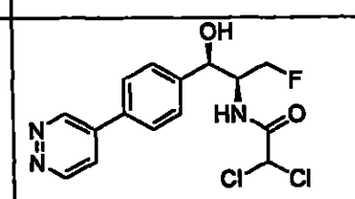
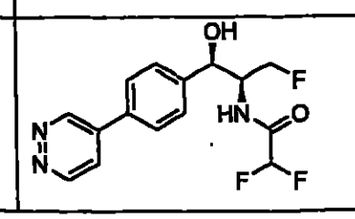
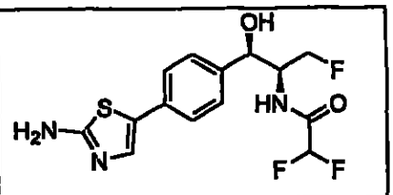
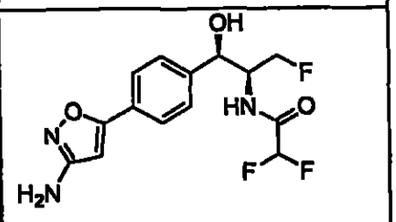
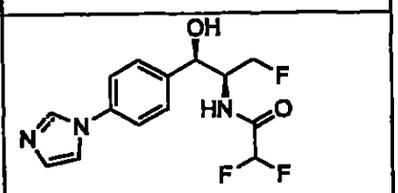
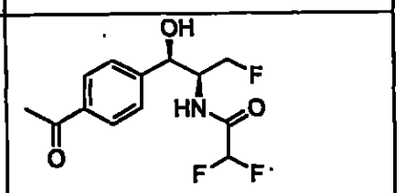
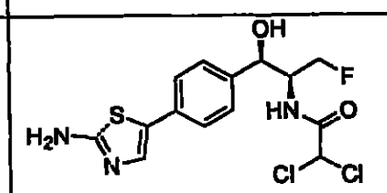
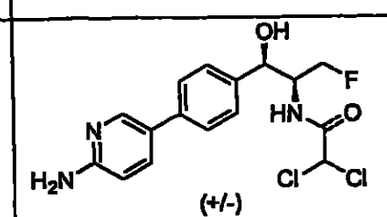
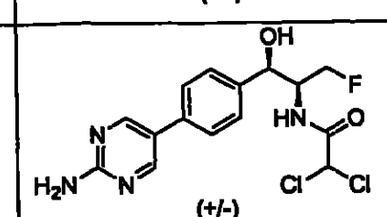
157	
158	
159	
160	
161	
162	
163	

164	
165	
166	
167	
168	
169	
170	
171	

172	
173	
174	
175 .	
176	
177	 (+/-)
178	 (+/-)

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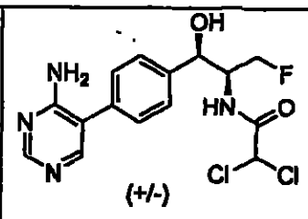
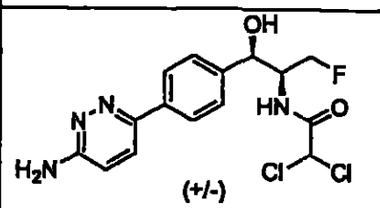
179	 (+/-)
180	 (+/-)

Síntesis

Se prepararon compuestos intermedios en forma enantioméricamente pura, comenzando con base de cloranfenicol comercialmente disponible (1), o como mezclas racémicas mediante la condensación de p-bromobenzaldehido con glicina bajo condiciones básicas. Por la síntesis enantiomericamente pura de los compuestos intermedios tales como 9, la base de cloranfenicol fue convertida en el compuesto 3 (Rebstock, M.C., *J. Am, Chem. Soc.*, 1949, 71, 2458; Evans, D.D., *J. Chem. Soc.*, 1954, 1687; Morris, D. S. y Smith, S. D., *J. Chem. Soc.*, 1954, 1680*)*. El compuesto 3 fue sometido a la reacción de Sandmeyer después de la cual el grupo protector de acetato fue removido bajo condiciones acídicas para proporcionar el compuesto 9 enantiomericamente puro.

Alternativamente, el p-bromobenzaldehido (5) puede ser convertido en (d/l)-treo-p-bromofenilglicina (6, Esquema 1), (Bolhoffer, W. A. *J. Am. Chem. Soc. 1954, 76 1322; Herbert*, R. B.; Wilkinson, B. Ellames, G. J. Can. J. Chem. 1994, *72, 114)*, el cual puede ser protegido N-Boc derivado 7 y luego reducido en dos pasos: la activación con DCC y N-hidroxisuccinimida seguido por el tratamiento con NaBH4 para proporcionar el compuesto 8. El compuesto 9 puede ser aislado como su sal TFA o como la base libre. La introducción regioselectiva de fluor es realizada mediante la protección de la amina y el grupo hidroxilo bencílico como oxazolina de fenilo 10 seguido por fluorinación con trifluoruro de (dietilamino)azufre (DAST) o trifluoruro de [bis(2-metoxietil)amino]azufre (Lal, G. S., *J. Org. Chem.*, 1999, *64* 7048) para proporcionar el compuesto 11. El compuesto 11 es utilizado en la reacción de acople cruzado de Suzuki para la sintesis de derivados de biarilo. El otro socio Suzuki, un ácido borónico de arilo tal como 12, puede ser preparado como se muestra y se puede hacer reaccionar con haluros de arilo.

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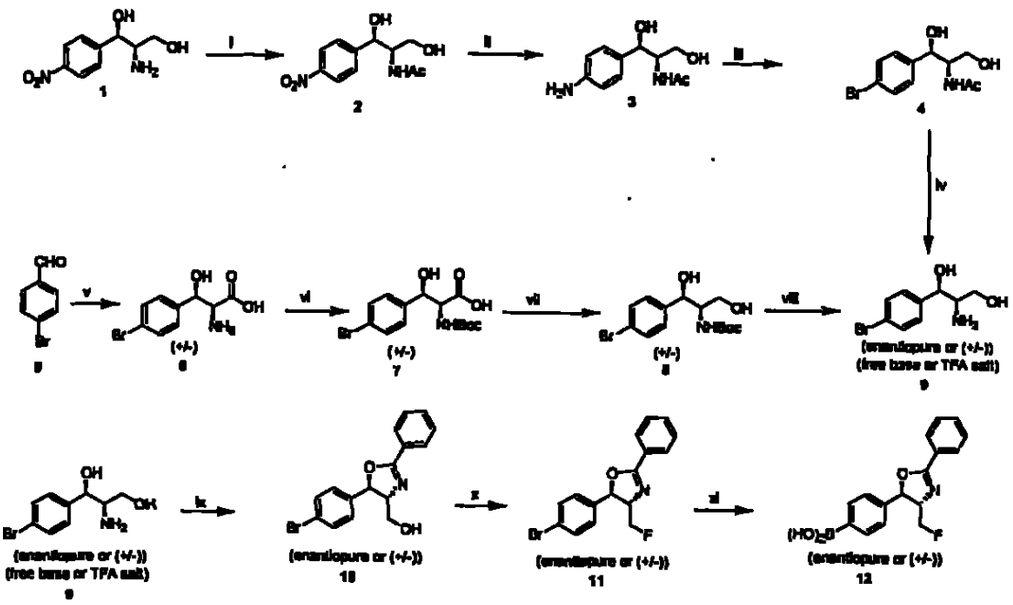
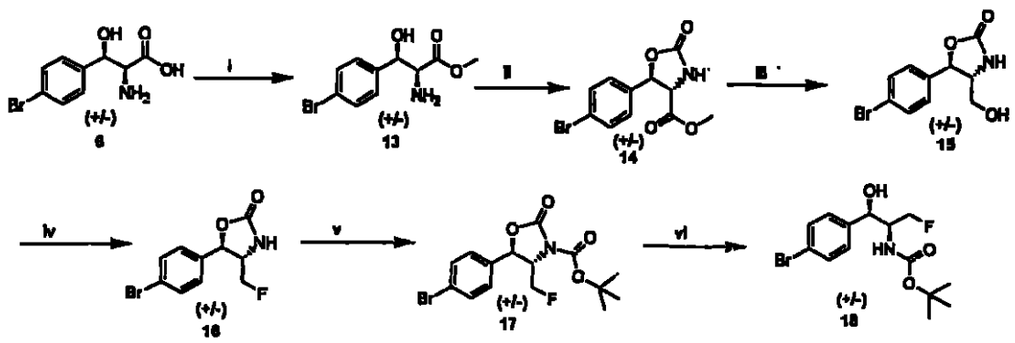
Esquema 1



I) Ac_2O neat; II) H_2(g), 10% Pd/C; III) $NaNO_2$/aq. HBr, 0 °C, then CuBr/aq. HBr; IV) 10% H_2SO_4, Δ, then NaOH workup; v) KOH, glycine, then aq. HCl, then NH_4OH; vi) Boc_2O, dioxane, water; vii) DCC, HOBu then $NaBH_4$ THF then H_2O; viii) TFA then NaOH workup; ix) ethylbenzimidate hydrochloride, Et_3N; x) DAST or [bis(2-methoxyethyl)amino]sulfur trifluoride, -78 °C to room temperature; xi) first *n*-BuLi at -78 °C, then $B(OMe)_3$ -78 °C to room temperature.

Las condiciones para la remoción del grupo protector de feniloxazolina probaron ser incompatibles con muchos grupos funcionales en la posición-p de anillo aromático. Por lo tanto, un nuevo grupo protector fue desarrollado con base en métodos de literatura (Esquema 2; Jommi, G., Gazz. Chim. Ital., 1986, 116:485). La fenilserina 6 fue convertida en éster de metilo 13, el cual fue protegido como oxazolidinona 14, la cual a su vez fue reducida con NaBH4 a 15. La fluorinación con DAST o trifluoruro de [bis(2-metoxietil)amino]azufre proporcionó el compuesto intermedio 16, el cual no dió rendimientos de la reacción de Suzuki consistentemente buenos. Por lo tanto, el compuesto 16 fue convertido en el compuesto 17. La escisión del grupo protector de oxazolidinona fue facilitada mediante la introducción de un grupo Boc (Grehn, L., *Acta Chem. Scand. B.* 1986, 40, 745) seguido por escisión catalizada con base para proporcionar el compuesto 18 (Ishizuka, T. y Kunieda, T. *Tetrahedron Lett.*, 1987, 28, 4185; Jommi, G., Gazza. Chim, Ital, 1988, 118:75).

Esquema 2

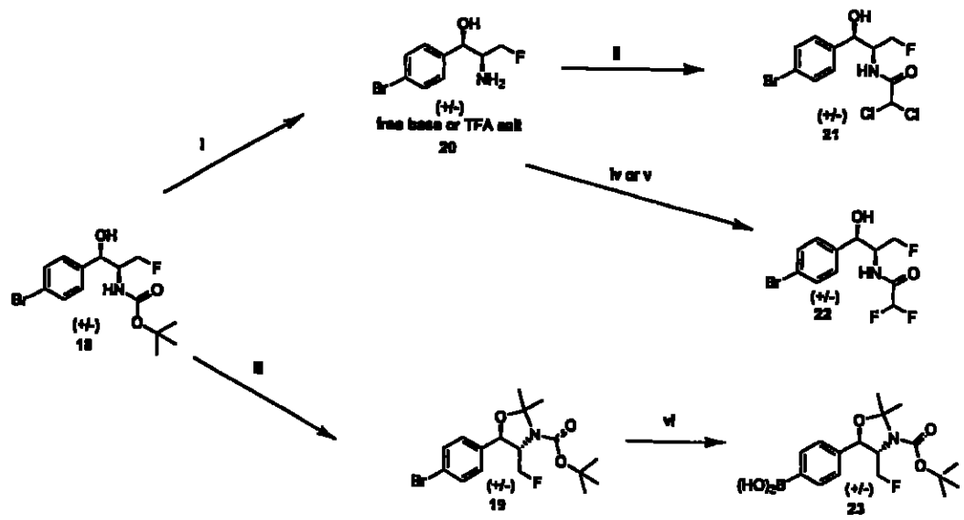


I) HCl, MeOH 0 °C to room temperature 18 h, then reflux 18 h; II) CDI, Et_3N, THF/1,2-dichloroethane; III) $NaBH_4$, MeOH 0 °C; IV) DAST or [bis(2-methoxyethyl)amino]sulfur trifluoride -78 °C 10 min then room temperature until complete; v) Boc_2O, DMAP, MeCN; vi) Cs_2CO_3, MeOH.

El compuesto 18 fue protegido como derivado de isopropilideno 19, el cual fue convertido en ácido borónico 23 (Esquema 3). Los compuestos 18, 19, 22 y 23 fueron utilizados en reacciones de acoplamiento cruzado de Suzuki. El compuesto 23 proporcionó la mayor versatilidad puesto que este pudo ser acoplado de manera cruzada con cualquier bromuro de arilo o yodo y los grupos protectores podían ser fácilmente removidos bajo condiciones suaves. El compuesto 22 fue atractivo como un socio de acoplamiento cruzado Suzuki puesto que las reacciones de acoplamiento produjeron los analogos de florfenicol deseados directamente sin necesidad de desprotección y dihaloacetilación. Sin embargo, los rendimientos de acoplamiento cruzado con 22 no fueron menores que aquellos con 23. Los acoplamientos cruzados utilizando 21 proporcionaron los productos deseados contaminados con análogos de acetato y monocloroacetato.

El compuesto racémico 20 fue espectroscopicamente idéntico a una muestra sintetizada a partir del compuesto 11 semi-sintético mediante depuración hidrolítica y crecimiento básico. Esto proporcionó la prueba inequívoca que la condensación de fenilserina 6 procedió con la *treo* o *syn* estereoselectividad relativa apropiada.

Esquema 3

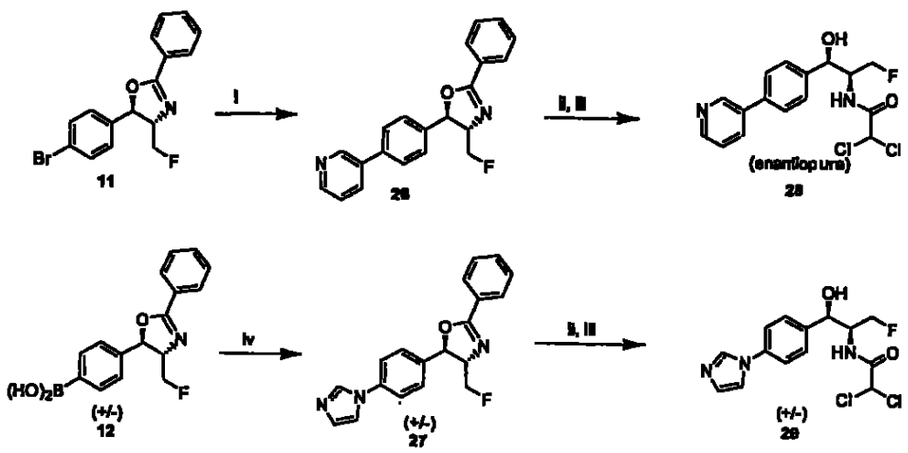


I) 9:1 TFA, H_2O, then NaOH work-up if free-base is desired; II) methyl dichloroacetate, Et_3N, MeoH, Δ; III) 2-methoxypropene, *p*-TsOH·H_2O; IV) methyl difluoroacetate, Et_3N, MeoH, Δ; V) difluoroacetyl chloride, Et_3N, followed by 1:1:1 Et_3N, MeOH, H_2O; VI) first *n*-BuLi at -78 °C, then $B(OMe)_3$, -78 °C to room temperature.

Los compuestos 11 y 12 fueron utilizados para preparar los compuestos 28 y 29 (Esquema 4). El compuesto 11 fue acoplado con ácido borónico de 3-piridina bajo condiciones de Suzuki bifásicas estándares para proporcionar el compuesto intermedio 26 protegido. El producto de acoplamiento cruzado N-C 27 fue preparado según los métodos recientemente reportados de Lam y colegas (Lam, P. Y. S., Synlett, 2000, 674; Lam, P. Y. S., *Tetrahedron, Lett., 1998, 39, 2941; Lam, P. Y. S., Tetrahedron Lett.*, 2001, *42, 3415).* Tanto 26 como 27 fueron desprotegidos y dicloroacetilados para

proporcionar 28 y 29.

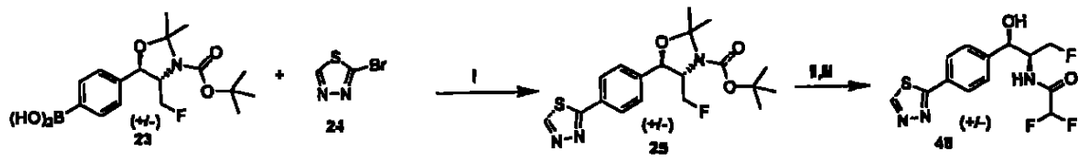
Esquema 4



I) *m*-pyridine boronic acid, Na_2CO_3, $Pd(PPh_3)_4$, THF, H_2O, Δ; II) 6 N aqueous HCl, sea led tube 100 °C then basic work-up; III) methyl dichloroacetate, Et_3N, MeOH, Δ; IV) $Cu(OAc)_2$, pyr., CH_2Cl_2, room temperature, open to air, 40 h.

El compuesto 23 y 2-bromo-1,3,4-tiadiazol (24) fueron hechos reaccionar para proporcionar 48 (Esquema 5). El acoplamiento cruzado de Suzuki proporcionó el heterobiarilo 25. El compuesto 25 fue desprotegido con 9:1 TFA/H2O, el cual removió los grupos Boc e isopropilideno simultáneamente, y luego fue difluoroacetilado para proporcionar 48.

Esquema 5

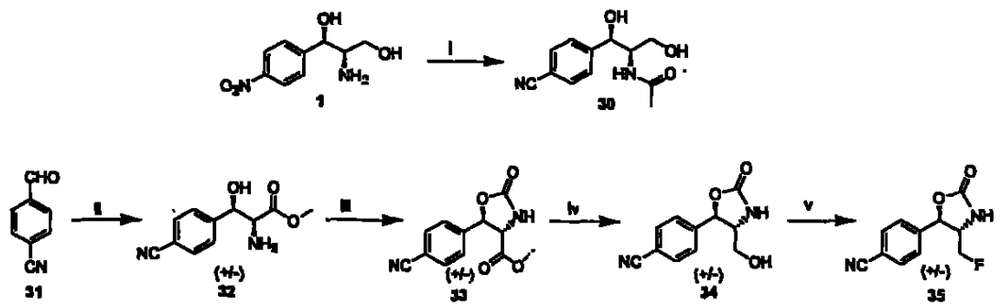


I) Cs_2CO_3, $PdCl_2(dppf)$, THF, DMF, H_2O; II) 9:1 TFA/H_2O; III) Et_3N, methyl difluoroacetate, Δ

Mientras los grupos funcionales de p-bromo de derivados tales como 11 y los correspondientes ácidos borónicos tales como 12 la tienen utilidad sustancial como intermedios sintéticos a análogos de florfenicol, los compuestos p-ciano también sirvieron como intermedios importantes. Utilizando los métodos de la literatura (Morris, D. S.; Smith, S. D., anterior), los intermedios tales como 30 han sido

preparados en forma enantiomericamente pura (Esquema 6). Grandes cantidades del intermedio fueron preparadas utilizando intermedios racémicos totalmente sintéticos tales como 35. La condensación de aldol utilizada para producir 32 a partir de p-cianobenzaldehido y éster metílico de glicina fue adaptada a partir de la literatura (Pines, S. H. y Kazlowski, M. A., J. Org. Chem., 1972, 37, 292).

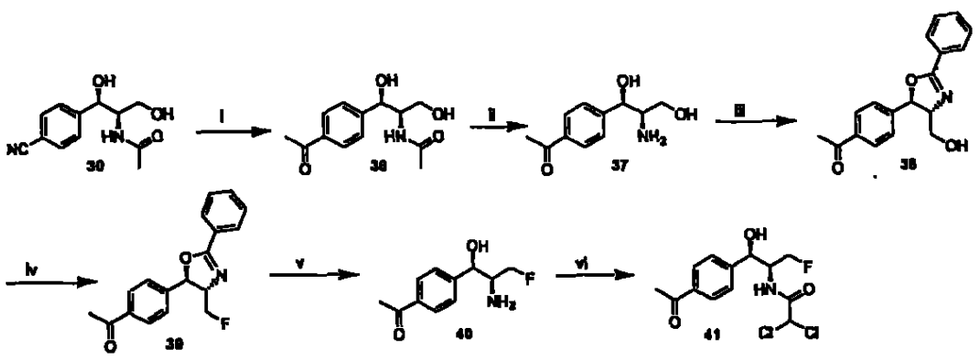
Esquema 6



i) literature procedure—see text; ii) literature procedure—see text; iii) CDI, TEA, THF or 1,2-dichloroethane; iv) $NaBH_4$, MeOH, 0 °C; v) DAST or [bis(2-methoxyethyl)amino]sulfur trifluoride -78 °C 10 min then room temperature until complete.

El compuesto 41 (Esquema 7) fue preparado a partir del intermedio 36, la preparación de este a partir de 30 ha sido descrita (von Strandtmann. M. *J. Med. Chem.*, 1967, 10, 888). El compuesto 36 fue desprotegido con H2SO4 acuoso y luego regioselectivamente protegido como feniloxazolina 38. El compuesto 39 fue tenido sobre tratamiento con DAST y luego fue desprotegido y dicloroacetilado para proporcionar 41.

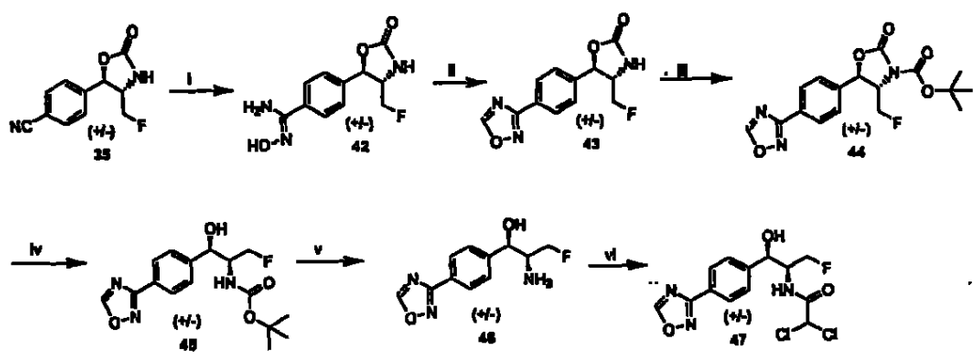
Esquema 7



i) MeLi, THF, then H_2O; ii) 10% aqueous H_2SO_4, Δ; iii) Et_3N, ethyl benzimidate hydrochloride, Δ; iv) DAST -78 °C to room temperature; v) 6N HCl, Δ; vi) methyldichloroacetate, Et_3N, MeOH, Δ.

El compuesto 47 fue preparado a partir del intermedio 35 mediante tratamiento con hidrocloruro de hidroxilamina seguido por ortoformato de trietilo para proporcionar 43, el cual fue desprotegido para proporcionar 46, el cual fue luego dicloroacetilado.

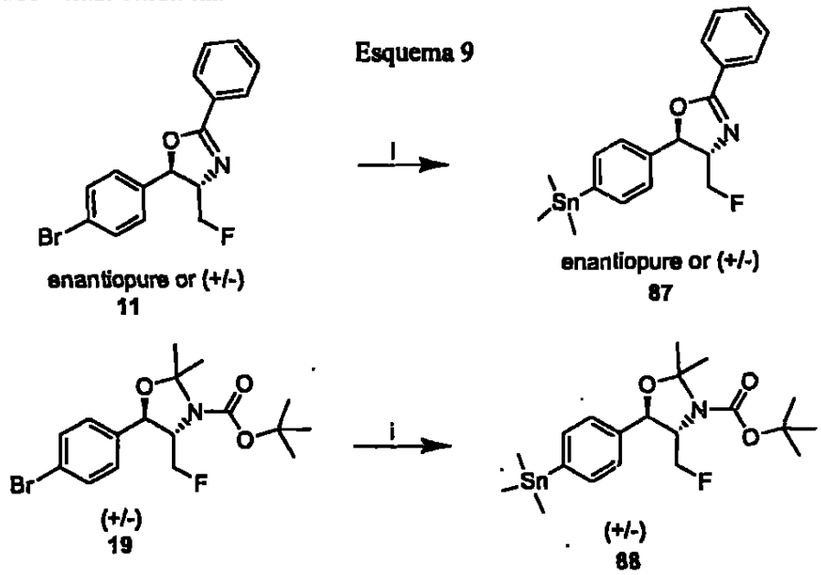
Esquema 8



i) NH_2OH·HCl, EtOH, Δ; ii) triethyl orthoformate, Δ; iii) Boc_2O, DMAP, MeCN; iv) Cs_2CO_3, MeOH; v) TFA/H_2O 9:1; vi) methyldichloroacetate, Et_3N, Δ.

Los derivados de p-acilo fueron obtenidos mediante reacciones de acoplamiento de Stille de los intermedios protegidos (Esquema 9, compuestos 87 y 88) con cloruros ácidos. Los grupos timetilstanilo fueron introducidos mediante reacciones Pd-mediadas utilizando hexametilditina.

Esquema 9



enantiopure or (+/-)
11

enantiopure or (+/-)
87

(+/-)
19

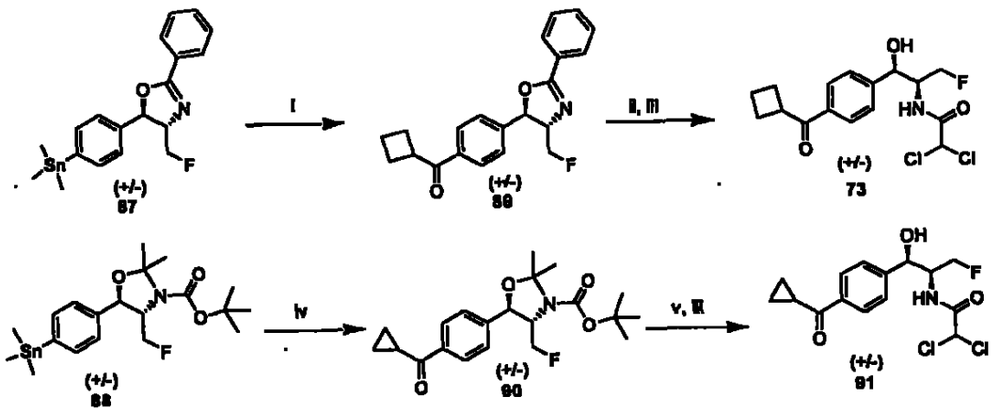
(+/-)
88

i) $Me_3SnSnMe_3$, $Pd(PPh_3)_4$, Δ, C_6H_6 or PhMe

El derivado de ciclobutilo 73 fue preparado a partir de 87 a través de la reacción de acoplamiento de Stille que proporcionó el intermedio 89 (Esquema 10) el cual fue desprotegido y dicloroacetilado.

La desprotección intentada del derivado de ciclopropilo correspondiente a 89 llevó a abertura de anillo HCl-mediada. Por lo tanto, para preparar 91, se utilizó el enfoque de boc/isopropilideno puesto que la desprotección ocurre bajo condiciones que no afectan el grupo ciclopropilo.

Esquema 10

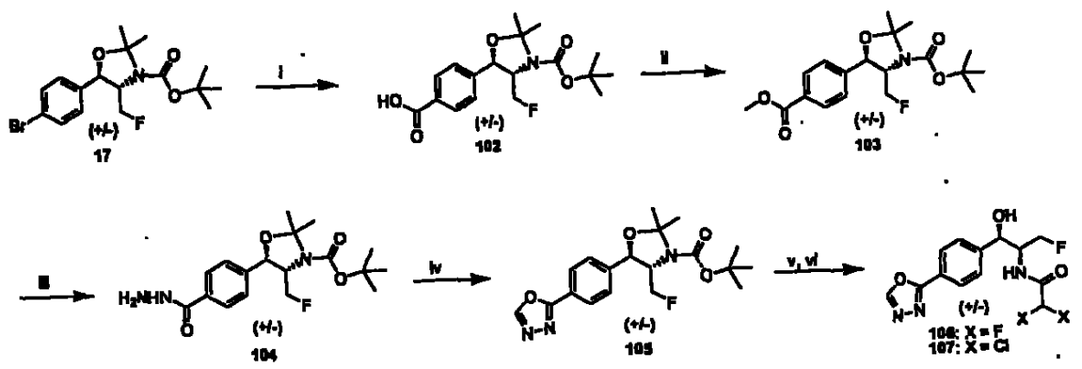


I) Pd_2dba_3, K_2CO_3, Et_3N, cyclobutanecarbonyl chloride, room temperature, 3h; II) 6N HCl sealed tube, Δ; III) methyldichloroacetate, Et_3N, MeOH, Δ; IV) Pd_2dba_3, K_2CO_3, Et_3N, cyclopropanecarbonyl chloride, room temperature, 3h; V) TFA/H_2O

Cuando se requirió, los derivados de p-carboxifenilo de intermedios de fenicol protegido se pudieron preparar en dos formas. Por ejemplo, la hidrólisis del análogo de p-nitrilo de 17 proporciona el correspondiente ácido carboxílico. Sin embargo, los nitrilos generalmente no pueden ser obtenidos con la facilidad de los derivados de bromo. Por lo tanto, el enfoque preferido para carboxilación fue el reemplazo del grupo bromo. Por ejemplo, el derivado del ácido carboxílico 102 (Esquema 11) fue preparado mediante litiación de 17 seguido por el tratamiento con CO2 y crecimiento con ácido. El compuesto 102 fue convertido en éster de metilo 103, el cual fue hecho reaccionar con hidrazina para proporcionar 104. La ciclización de 104 con ortoformato de trietilo proporcionó oxadiazol 105, el cual fue desprotegido y dihaloacetilado para proporcionar 106 y 107.

~~137~~
138

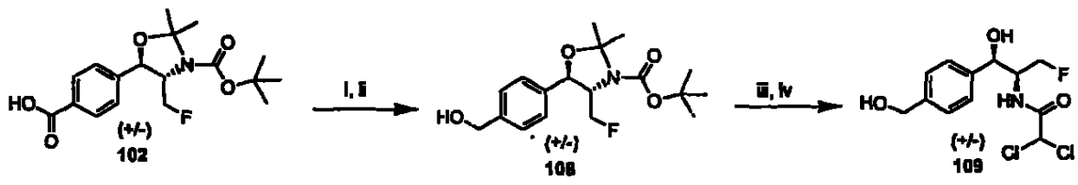
Esquema 11



i) *n*-BuLi, -78 °C, THF, then bubble dry CO_2 then warm to room temperature, then H_3O^+; ii) CH_2N_2; iii) hydrazine, EtOH, Δ; iv) triethylorthoformate, Δ; v) TFA/H_2O; vi) for 106 Et_3N, ethyldifluoroacetate, room temperature; for 107 same as 106, but replace ethyl difluoroacetate with methyl dichloroacetate.

El derivado del ácido carboxílico 102 también fue utilizado para preparar compuestos tales como 109 (esquema 12), mediante reducción a alcohol 108 seguido por desprotección y dicloroacetilación

Esquema 12



I) DCC, pentafluorophenol, EtOAc; II) $NaBH_4$, MeOH; III) TFA/H_2O; iv) methyl dichloroacetate, Et_3N, Δ.

Evaluación Biológica

Se espera que todos los compuestos de este invento demuestren actividad antimicrobiana contra las mismas bacterias que los demás miembros de la familia del

cloranfenicol. Además, se espera que sean activos contra especies de bacterias que son resistentes a los actuales antibióticos de cloranfenicol, en particular florfenicol. También se espera que los presentes compuestos puedan exhibir actividad contra géneros y especies de bacterias contra los cuales los antibióticos tipo florfenicol actuales no son activos.

También se entiende, con respecto a la bioactividad, que un enantiomero de un compuesto pueda ser más activo que otro. En tal caso, si se declara expresamente o no, el isomero más activo es considerado la realización más preferida del invento. Particularmente preferido es el enantiomero más activo de la configuración absoluta 1-(R)-2(S) de cualquier compuesto en la presente.

Para determinar el rango y nivel de actividad de los compuestos de este invento, se pueden utilizar los siguientes protocolos. Otros protocolos serán manifiestos para aquellos con experiencia en el estado de la técnica con base en la divulgaciones en la presente y están dentro del alcance de este invento. Se espera que algunos compuestos en la presente no solamente exhiban actividad antibacteriana sustancial sino también que sean menos susceptibles a los mecanismos de resistencia del cloranfenicol. Los protocolos de clasificación en la presente se pueden utilizar para determinar tales caracteristicas también.

Prueba de Susceptibilidad

Los compuestos fueron evaluados contra un panel de cepas bacterianas utilizando un ensayo de microdilución de caldo tal como se recomendó mediante el NCCLS (Comité Nacional para Estándares de Laboratorio Clínico (NCCLS) 2000, Métodos para Dilución de Pruebas de Susceptibilidad Antimicrobiana para Bacterias que Crecen Aerobicamente - Quinta Edición, Estandar Aprobado, Documento NCCLS M7-A5, vol 20, No. 2). La concentración inhibitoria mínima (MIC) es definida como la concentración menor de un compuesto que evita el crecimiento de la bacteria.

Los siguientes 10 organismos constituyeron el panel principal de evaluación:

TABLA 2

Bacteria	Cepa	Bomba de salida de flujo	fenotipo
Escherichia coli	ECM 1194	AcrAB	tipo silvestre
Escherichia coli	ECM 1694	Ninguno	ΔacrAB::Tn903kmr
Escherichia coli	ECM 1642	AcrAB	MarR
Escherichia coli	ECM 1888	MdfA	tolC::Tn mdfR
Escherichia coli	ECM 1750	CmlA	ΔacrAB::Tn903kmr/pLQ821
Escherichia coli	ECM 1958	Flo	ΔacrAB::Tn903kmr/p1956
Escherichia coli	ECM 1970	AcrAB+Flo	marR/p1956
Escherichia coli	ECM 1197	AcrAB	EMR::Cm (cat)
	ó ECM 2024	AcrAB	ECM1197 acrAB::Kan
Pasteurella multocida	ATCC43134		tipo silvestre
Mannhemia haemolytica	ATCC33396		tipo silvestre

Los ensayos fueron realizados en Caldo Cation-Ajustado Mueller-Hinton a un inoculo bacteriano de 5 x 10^5 CFU/ml y un volumen final de 100 ul. Se prepararon controles de florfenicol y cloranfenicol y compuestos de prueba a cuatro veces la

concentración final deseada. La dilución a la concentración deseada fue realizada directamente sobre las placas mediente dilución serial doble utilizando una pipeta multicanal. Después de la dilución, se agregó 25 ul de CAMHB a cada pozo.

Los inoculos bacterianos fueron preparados como sigue. Para cada cepa, una colonia aislada fue utilizada para inocular un volumen de 5 ml de CAMHB. Los cultivos fueron incubados durante la noche (20 horas) a 35°C en un incubador con agitación. Luego fueron diluidos en solución salina estéril hasta una densidad equivalente a una suspensión 0.5 McFarland (10^8 CFU/ml aprox). Las suspensiones fueron adicionalmente diluidas en CAMHB hasta aproximadamente 5 x 10^5 CFU/ml. Se agregó un volumen de 50 □l del inoculo a cada pozo. Se incluyeron controles de crecimiento positivos y negativos en cada placa. Los inoculos originales fueron determinados mediante la aplicación de 10 □l de varias diluciones de 10 veces sobre placas TSA. Las placas de agar fueron incubadas durante la noche a 35°C y se contaron las unidades formadoras de colonias (CFU). Las placas de microtitulos fueron incubadas durante 20 horas a 35°C y fueron leídas utilizando un lector de microtitulos (dispositivos moleculares) a 650 nm y mediante observación visual utilizando un espejo de lectura de plato de microtitulación para determinar el MIC.

Composiciones farmacéuticas

Un compuesto del presente invento, un profármaco del mismo o una sal fisiológicamente aceptable de cualquiera el compuesto o su profármaco, pueden ser administrados como tal a un paciente o se pueden administrar en composiciones farmacéuticas en las cuales los anteriores materiales son mezclados con excipientes apropiados. Las técnicas para formulación y administración de fármacos se pueden encontrar en *Pharmacological Sciences de Remington*, Mack Publishing Co. Easton, PA, última edición. Las formulaciones y técnicas discutidas en Remington se relacionan principalmente con el uso en pacientes humanos; sin embargo, se pueden modificar fácilmente para utilizar en pacientes no humanos mediante técnicas bien conocidas por aquellos con experiencia en el estado de la técnica veterinaria.

Rutas de Administración

Como se utiliza en la presente, "administrar" o "administración" se refiere a la liberación de un compuesto, sal o profármaco del presente invento o de una composición farmacéutica que contiene un compuesto, sal o profármaco de este invento a un organismo para el propósito de tratar o prevenir una infección microbiana.

Rutas de administración apropiadas pueden incluir, sin limitación, oral, rectal, transmucosal, intramuscular, subcutánca, intramedular, intratecal, intraventrícular directa, intravenosa, intravítrea, intraperitoneal, intranasal, aural, o intraocular. Las rutas preferidas de administración son la oral y la parenteral.

Alternativamente, el compuesto se puede administrar en forma local más que sistémica, por ejemplo, mediante la preparación como un ungüento que es aplicado directamente al área infectada o mediante inyección del compuesto directamente en tejido infectado. En cualquier caso, se puede utilizar una formulación de liberación sostenida.

Composición/Formulación

Las composiciones farmacéuticas del presente invento se pueden fabricar mediante procesos bien conocidos en el estado de la técnica, ej., utilizando una variedad de procesos bien conocidos de mezclado, disolución, granulación, fabricación de grageas, levigación, emulsificación, encapsulación o liofilización. Las composiciones se

puede formular en conjunción con uno o más vehículos fisiológicamente aceptables que comprenden excipientes y auxiliares que facilitan el procesamiento de los compuestos activos en preparaciones que pueden ser utilizadas farmacéuticamente. La formulación adecuada es dependiente de la ruta de administración elegida.

Para inyección, incluyendo, sin limitación, inyección intravenosa, intramuscular y subcutánea, los compuestos del invento puede ser formulados en soluciones acuosos, preferiblemente en amortiguadores fisiológicamente compatibles tales como solución de Hanks, la solución de Ringer, amortiguador salino fisiológico o solventes polares incluyendo, sin limitación, N-metil-2-pirrolidona, 2-pirrolidona, otras pirrolidonas, N,N-dimetilacetamida, N,N-dimetilformamida, dimetilsulfoxido, acetona y glicerol formal. Para administración transmucosal, se utilizan en la formulación, penetrantes apropiados para la barrera a ser permeada. Tales penetrantes generalmente son conocidos en el estado de la técnica. Para administración oral, los compuestos se pueden formular mediante combinación de los compuestos activos con vehículos farmacéuticamente aceptables bien conocidos en el estado de la técnica. Tales vehículos permiten que los compuestos del invento sean formulados como tabletas, píldoras, pastillas, grageas, cápsulas, líquidos, geles, jarabes, pastas, pastas aguadas, soluciones, suspensiones, soluciones concentradas y suspensiones para diluir en el agua potable de un paciente, premezclas para dilución en el alimento de un paciente, y similares, para ingestión oral por parte del paciente. Las preparaciones farmacéuticas para uso oral se pueden hacer utilizando un excipiente sólido, opcionalmente triturando la mezcla resultante, y procesando la mezcla de gránulos, después de agregar auxiliares apropiados si se desea, para obtener tabletas o núcleos de grageas. Excipientes útiles son, en particular, rellenos tales como azúcares, incluyendo lactosa, sacarosa, manitol, o sorbitol, preparaciones de celulosa tales como, por ejemplo, almidón de maíz, almidón de cebada, almidón de arroz y almidón de papa y otros materiales tales como gelatina, goma tragacanto, celulosa de metilo, hidroxipropilmetilcelulosa, carboximetilcelulosa sódica, y/o polivinilpirrolidona (PVP). Si se desea, se pueden agregar agentes desintegrantes, tales como pirrolidona de polivinilo entrecruzada, o ácido algínico. También se puede utilizar una sal tal como alginato de sodio.

Los núcleos de grageas son proporcionados con revestimientos apropiados. Para este propósito, se pueden utilizar soluciones de azucar concentradas las cuales opcionalmente pueden contener goma arabiga, talco, polivinilpirrolidona, gel de carbopol, glicol de polietileno, y/o dióxido de titanio, soluciones de laca, y solventes orgánicos apropiados o mezclas de solventes. Se pueden agregar tintes o pigmentos a las tabletas o revestimientos de grageas para identificación o para caracterizar las diferentes combinaciones de dosis de compuestos activos.

Las composiciones farmacéuticas que se pueden utilizar oralmente incluyen cápsulas *push-fit* hechas de gelatina, así como también cápsulas suaves, selladas hechas de gelatina y un plastificante tal como glicerol o sorbitol. Las cápsulas push-fit pueden contener los ingredientes activos en mezcla con un relleno tal como lactosa, un aglomerante tal como almidón, y/o un lubricante tal como talco o estearato de magnesio y, opcionalmente, estabilizantes. En cápsulas suaves, los compuestos activos se pueden disolver o suspender en líquidos apropiados, tales como aceites grasos, parafina liquida, o glicoles de polietileno líquidos. Los estabilizantes también se pueden agregar en estas formulaciones.

Para administración por inhalación, los compuestos del presente invento se pueden liberar convenientemente en la forma de una atomización en aerosol utilizando en empaque presurizado o un nebulizador y un propulsor apropiado, ej., sin limitación, diclorodifluorometano, triclorofluorometano, diclorotetrafluoroetano o dióxido de

carbono. En el caso de un aerosol presurizado, la unidad de dosificación puede ser controlada proporcionando una válvula para liberar una cantidad medida. Las cápsulas y cartuchos de, por ejemplo, gelatina para utilizar en un inhalador o insuflator se pueden formular conteniendo una mezcla en polvo del compuesto y una base en polvo apropiada tal como lactosa o almidón.

Los compuestos también se pueden formular para administración parenteral, ej., mediante inyección de bolo o infusión contínua. Formulaciones para inyección se pueden presentar en forma de dosificación unitaria, ej., en ampollas o en contenedores de múltiple dosis. Composiciones útiles incluyen, sin limitación, suspensiones, soluciones o emulsiones en vehículos aceitosos o acuosos, y pueden contener aditamentos tales como agentes de suspensión, estabilización y/o dispersión. Las composiciones farmacéuticas para administración parenteral incluyen soluciones acuosas de una forma soluble en agua, tal como, sin limitación, una sal, del compuesto activo. Adicionalmente, las suspensiones de los compuestos activos se pueden preparar en un vehículo lipofílico. Vehículos lipofílicos apropiados incluyen aceites grados tales como aceite de cacahuate, ésteres de ácidos grados sintéticos tales como oleato de etilo y triglicéridos, o materiales tales como liposomas. Las suspensiones de inyección acuosa pueden contener substancias que aumentan la viscosidad de la suspensión, tal como carboximetilcelulosa sódica, sorbital o dextran. Opcionalmente, la suspensión también puede contener estabilizantes apropiados y/o agentes que aumentan la solubilidad de los compuestos para permitir la preparación de las soluciones altamente concentradas. Alternativamente, el ingrediente activo también puede estar en forma de polvo para constitución con un vehículo apropiado, ej., agua esteril libre de pirogenos, antes de utilizar.

Los compuestos también se pueden formular en las composiciones rectales tales como supositorios o enemas de retención, utilizando, ej., bases supositorias convencionales tales como manteca de cacao u otros glicéridos.

Además de las formulaciones descritas anteriormente, los compuestos también pueden ser formulados como preparaciones depot. Tales formulaciones de acción prolongada se pueden administrar mediante implante (por ejemplo, subcutáneamente o intramuscularmente) o mediante inyección intramuscular. Un compuesto de este invento se puede formular para esta ruta de administración con materiales poliméricos o hidrofóbicos apropiados (por ejemplo, en una emulsión con un aceite farmacológicamente aceptable), con resinas de intercambio iónico, o con un derivado apenas soluble tal como, sin limitación, una sal apenas soluble.

Se pueden utilizar otros sistemas de liberación para compuestos farmacéuticos relativamente hidrofóbicos. Los liposomas y emulsiones son ejemplos bien conocidos de portadores o vehículos de liberación para fármacos hidrofóbicos. Además, se pueden utilizar solventes orgánicos tales como dimetilsulfóxido, aunque a menudo con riesgo de mayor toxicidad.

Adicionalmente, los compuestos se pueden liberar utilizando un sistema de liberación sostenida, tales como matrices semi-permeables de polímeros hidrofóbicos que contienen el agente terapéutico. Se han establecido varios materiales de liberación sostenida y son bien conocidos por aquellos con habilidad en el estado de la técnica. Las cápsulas de liberación sostenida pueden dependiendo de su naturaleza química, liberar los compuestos durante pocas semanas hasta 100 días. Dependiendo de la naturaleza química y la estabilidad biológica del compuesto particular, se pueden emplear estrategias similares de estabilización.

Las composiciones farmacéuticas útiles en la presente también pueden comprender vehículos o excipientes de fase sólida o en gel. Ejemplos de tales vehículos

o excipientes incluyen, pero no están limitados a, carbonato de calcio, fosfato de calcio, varios azucares, almidones, derivados de celulosa, gelatina y polímeros tales como glicoles de polietileno.

Dosificación

Una cantidad terapéuticamente efectiva se refiere a una cantidad efectiva de un compuesto para prevenir, aliviar, o mejorar los síntomas de una infección microbiana. La determinación de una cantidad terapéuticamente efectiva es una habilidad general para aquellos capacitados en el arte, especialmente a la luz de la divulgación en la presente.

Para cualquier compuesto usado en los métodos del invento, la cantidad terapéuticamente efectiva puede ser estimada de manera inicial a partir ensayos de cultivos celulares. Luego, la dosificación puede ser formulada para usarse en modelos animales para así lograr un rango de concentración en circulación que incluya el MIC tal como se determina en cultivos celulares. Tal información puede luego ser usada para determinar de manera más precisa las dosificaciones útiles en los pacientes.

La toxicidad y la eficacia terapéutica de los compuestos descritos en la presente pueden ser determinados mediante procedimientos farmacéuticos estándares en cultivos celulares o en animales experimentales. Por ejemplo, el MIC y el LD_{50} para un compuesto en particular pueden ser determinado mediante métodos bastante conocidos en el arte. Los datos obtenidos pueden ser usados para formular un rango de dosificaciones útiles en los pacientes. Por supuesto que la dosificación puede variar dependiendo de la forma de dosificación y la vía de administración. La formulación exacta, la vía de administración y la dosificación puede ser escogida por el médico particular conociendo la condición particular del paciente. (ver, Fingl, et al., 1975, "The Pharmacological Basis of Therapuetics", capítulo 1, página 1). En general, sin embargo, el rango de dosificación preferido en la presente para administración sistémica de un compuesto de este invento oscilará entre 1 y 100 mg/kg aproximadamente. El rango de dosificación preferido en la presente para uso tópico generalmente oscilará entre 0.1 mg y 1 g aproximadamente.

La cantidad e intervalo de dosificación puede ser ajustado de manera individual para proporcionar niveles de plasma del compuesto que sean suficientes para mantener una concentración igual al MIC o cualquier otro nivel deseado. Tales niveles de plasma usualmente se conocen como concentraciones efectivas mínimas (MECs). El MEC variará para cada compuesto puede estimarse a partir de datos in vitro, como por ejemplo, la concentración necesaria para lograr un 80%+ de inhibición de un microbio, podría averiguarse usando los ensayos descritos en la presente. Las dosificaciones necesarias para lograr la MEC dependerá de características particulares y la vía de administración. Los ensayos de HPLC o bioensayos pueden usarse para determinar las concentraciones de plasma.

Los intervalos de dosificación también pueden determinarse usando el valor MEC. Los compuestos deben ser administrados usando un régimen que mantenga los niveles de plasma por encima del MEC entre 10 y 90% del tiempo, preferiblemente entre 30 y 90% y de manera más preferida entre 50 y 90%.

Para los casos de una administración local o reabsorción selectiva, la concentración local efectiva del fármaco no podrá relacionarse con la concentración de plasma y otros procedimientos conocidos en el arte pueden ser empleados para determinar el monto de dosis correcta y el intervalo.

La cantidad de una composición administrada por supuesto que será dependiente del paciente a tratar, la severidad de la infección, la manera de administración, el juicio del médico que receta, etc.

Empaques

Las composiciones pueden, si se desea, ser presentadas en un empaque (*pack*) o dispositivo dispensador, tal como un kit aprobado por la FDA, el cual puede contener uno o más formas de dosificación unitarias que contengan el ingrediente activo. El empaque puede, por ejemplo, comprender lámina metálica o plástica, como lo es un blister pack. El empaque o dispositivo dispensador puede estar acompañado por instrucciones para su administración. El empaque o dispensador también puede estar acompañado por una nota asociada con el recipiente en una forma dada por un ente gubernamental regulando la manufactura, uso o venta de farmacéuticos; la nota refleja la aprobación de la forma de las composiciones por parte de la agencia o para su administración human o veterinaria. Tal notificación, por ejemplo, puede ser del tipo de rotulación aprobada por la FDA de los EEUU para fármacos de venta bajo receta médica o de un inserto de producto aprobado. Las composiciones que comprenden un compuesto del invento formulado en un vehículo farmacéutico compatible también pueden prepararse, colocarse en un recipiente apropiado, y rotularse para un tratamiento de una condición indicada.

Ejemplos

Los siguientes ejemplos son proporcionados para ilustrar ciertas realizaciones de este invento y no pretenden, ni deben considerarse como límite del alcance de ninguna manera.

Los materiales iniciales fueron obtenidos de proveedores comerciales y usados sin ninguna purificación adicional a menos que se indique lo contrario. Los proveedores químicos incluyen Aldrich, Fluka y Lancaster. El $Pd(PPh_3)_4$ fue obtenido de Lancaster o Strem e inmediatamente transferido a frascos lavados con N_2 (entre 10 y 100 mg cada uno) en una bolsa tipo guante de N_2. Los frascos fueron envueltos en lámina de aluminio y almacenados en bolsas de cierre hermético lavados con N_2 a -20°C. El $PdCl_2(dppf)$ fue obtenido de Aldrich y utilizado desde la botella. Los solventes de grado reactivo estándares, los cuales no necesariamente eran anhidros, fueron usados. Los solventes anhidros fueron comprados de proveedores químicos y utilizados tal cual.

Los espectros ^1H NMR fueron registrados en un espectrómetro Varian FT-NMR de 300 MHz y se reportan en el formato "desplazamiento químico (multiplicidad, integración, constante de acople)." Las constantes de acople están reportadas en Hz. Los espectros de masa fueron obtenidos en un espectrómetro de masa de cuadrupolo sencillo Micromass Platform II equipado con ionización por electroespray. (ESI)

Los siguientes tres métodos de acoplamiento cruzado Suzuki fueron usados:

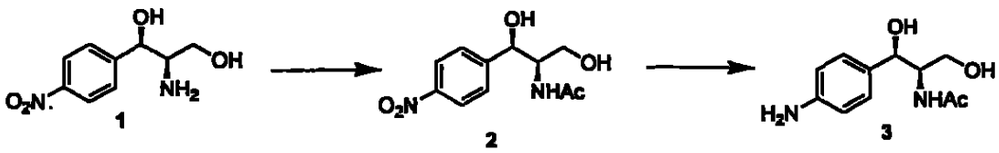
Método A: el ácido arilborónico (0.167 mmol) y el arilbromuro (0.334 mmol) fueron combinados en una mezcla de Na_2CO_3 acuoso (3 mL de una solución (peso/peso) al 10%) y THF (5 mL). La mezcla fue purgada brevemente con N_2. Se añadió $Pd(PPh_3)_4$ (10 mol%, 0.0167 mmol),la mezcla fue purgada con N_2, y luego sometida a reflujo por 16 horas. La mezcla de reacción fue diluida con etilacetato (EtOAc), lavada con solución de salmuera saturada, secada sobre Na_2SO_4 anhidro y concentrada. El residuo luego fue purificado mediante cromatografía.

Método B: el ácido arilborónico (0.301 mmol), el arilbromuro (0.602 mmol) y Cs_2CO_3 (0.903 mmol) fueron combinados en THF (2.0 mL), DMF (2.0 mL), y H_2O (0.5 mL) a

temperatura ambiente. La mezcla fue purgada con N_2 por 5 minutos y se añadió complejo de [1,1'-bis(difenilfosfino)ferroceno]dicloropaladio(II) y diclorometano (0.0301 mmol). La mezcla fue purgada con N_2 por cinco minutos y luego agitada a 55°C por 16 horas. La mezcla fue concentrada bajo vacío, diluida con EtOAc y lavada con salmuera. El EtOAc fue secado sobre $NaSO_4$ anhidro y concentrado. El residuo fue purificado mediante cromatografía.

Método C: el ácido arilborónico (0.934 mmol), el arilbromuro (2.34 mmol) y Cs_2CO_3 (2.80 mmol) fueron suspendidos en una mezcla de tolueno (4 mL), n-butanol (4 mL), y H_2O (2 mL). La mezcla fue purgada a temperatura ambiente con N_2 y luego se añadió $Pd(PPh_3)_4$ (0.280 mmol). La mezcla fue purgada con N_2 por otros cinco minutos adicionales y luego calentada a 75°C. Después de 12 horas, la mezcla fue enfriada hasta temperatura ambiente y concentrada bajo vacío. El residuo fue dividido entre agua (75 mL) y EtOAc (75 mL). La capa de EtOAc fue lavada con salmuera (2 x 50mL), secada sobre $NaSO_4$ anhidro y concentrada al vacío. El residuo fue purificado mediante cromatografía.

Example 1: Compound 3



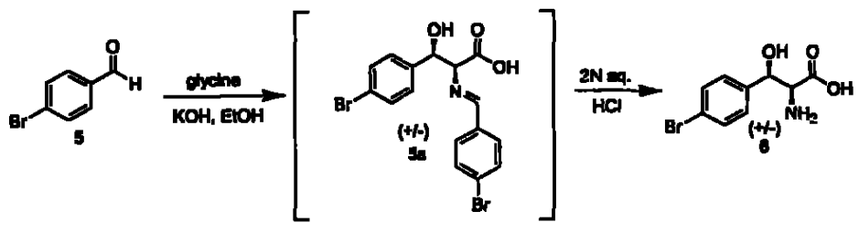
In 80 mL of methanol was dissolved 8.13 g (32.0 mmol) of chloramphenicol base as the *N*-acetate, prepared by literature methods (Rebstock, M. C., *et al.*, *supra*, Crooks H. M. *et al.*, *supra*; Evans, D. D.; Morris, D. S. *et al.*, *supra*). After purging with N_2, Pd/C was added and the mixture was stirred under H_2(1 atm) for 16 hours. The Pd/C was then removed by filtration through Celite. The solution was concentrated under vacuum and the residue purified by silica gel chromatography, eluting with 15% MeOH in CH_2Cl_2 to give 3 (7.13 g, 31.7 mmol). ^{1}H NMR (300 MHz, CD_3OD): δ 1.92 (s, 3H), 3.40 (dd, 1H, *J* = 11.1, 6.0), 3.61 (dd, 1H, *J* = 11.1, 5.7), 4.01 (m, 1H), 4.73 (d, 1H, *J* = 5.4), 6.69 (d, 2H, *J* = 8.4), 7.11 (d, 2H, *J* = 8.4).

Example 2: Compound 4

An aqueous solution of $NaNO_2$ (1.27 g, 18.5 mmol, 50 mL H_2O) was added dropwise to a solution of 3 (3.76 g, 16.8 mmol) in 35 mL of 48% aqueous HBr at 0

°C. After addition was completed, the mixture was stirred for 30 min at 0 °C. The mixture was then added dropwise to a solution of CuBr (2.65 g, 18.45 mmol) in 15 mL 48% aqueous HBr. The mixture was warmed to room temperature and stirred for an additional 16 hours. The reaction mixture was neutralized with 3 M aqueous NaOH, filtered through a pad of Celite, and extracted with EtOAc (3 x 100 mL). The combined organic fractions were dried over anhydrous Na_2SO_4 and concentrated under vacuum to give 1.2 g (4.17 mmol) of crude product that was used without further purification. ^{1}H NMR (300 MHz, CD_3OD): δ 1.87 (s, 3H), 3.48 (dd, 1H, J = 11.0, 5.9), 3.69 (dd, 1H, J = 11.0, 6.5), 4.01 (m, 1H), 4.91 (d, 1H, J = 3.6), 7.30 (d, 2H, J = 8.3), 7.45 (d, 2H, J = 8.3).

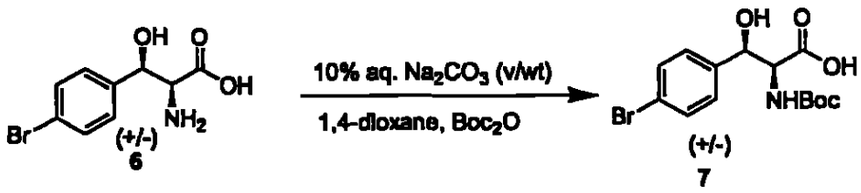
Example 3: Compound 6



4-Bromobenzaldehyde (100 g, 0.540 mol) was dissolved in ethanol (EtOH) in a 2 L round-bottom flask. With rapid stirring, glycine (0.5 molar equivalents, 20.3 g, 0.270 mol) and then, in one portion, KOH (30.3 g, 0.540 mol) were added. It is essential to add the KOH all at once. Thus, an ice bath should be used when performing the reaction on a larger scale to control the exotherm. After addition of the KOH, the turbid suspension became yellow and homogeneous. After about 15 minutes, a thick white precipitate began to form. The mixture was stirred for 12 hours at room temperature under N_2. Enough 2 N aqueous HCl was added (~400 mL) to make the solution red to pH paper. The mixture was then stirred at approximately 60 °C until it again become a homogeneous yellow solution. The EtOH was removed under vacuum to give an aqueous suspension of white precipitate. The precipitate was filtered and the remaining aqueous solution washed three times with EtOAc. The aqueous solution was then basified to about pH 9 with concentrated aqueous NH_3 and excess ammonia was removed under vacuum. As the NH_3 was removed, the product, 6, began to precipitate. Evaporation continued to one-quarter the volume of solution where precipitation was first observed. The

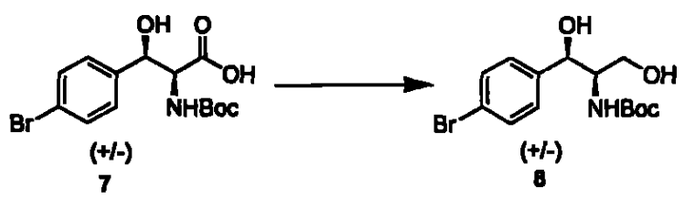
product was then collected on a vacuum filter and dried under vacuum to a constant weight (51.3 g). If NMR indicates the presence of the undesired trans stereoisomer, it can be removed by recrystallization from H_2O/ EtOH). ^1H NMR (300 MHz, CD_3OD): δ 3.64 (d, 1H, J = 3.6), 5.25 (d, 1H, J = 3.6), 7.41 (d, 2H, J = 8.4), 7.53 (d, 2H, J = 8.4).

Example 4: Compound 7



Compound 6 (32.60 g, 12.53 mmol) was dissolved in 10% aqueous K_2CO_3 (10% w/v; 500 mL). Di-*tert*-butyl dicarbonate (34.2 g, 157 mmol) was dissolved in 1,4-dioxane (500 mL) and added to the aqueous solution, after which the mixture was stirred at room temperature for 72 hours. The mixture was concentrated under vacuum and taken up in 100 mL 1 N aqueous NaOH and washed with Et_2O (2 x 100 mL). The aqueous layer was acidified with 1 N aqueous HCl and the product extracted into EtOAc (3 x 200 mL). The combined EtOAc extracts were dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum. The product (26.7 g, 74.0 mmol) was isolated and used without further purification. ^1H NMR (300 MHz, CD_3OD): δ 1.32 (s, 9H), 4.37 (d, 1H, J = 2.7), 5.23 (d, 1H, J = 2.7), 7.32 (d, 2H, 8.4), 7.46 (d, 2H, J = 8.4).

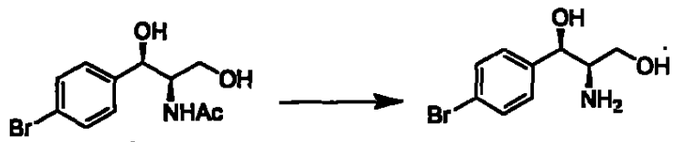
Example 5: Compound 8



Compound 7 (5.29 g, 14.7 mmol) and *N*-hydroxysuccinimide (1.69 g, 14.7 mol) were dissolved in EtOAc (200 mL) and the mixture cooled to 0 °C. *N,N'*-dicyclohexyl- carbodiimide (3.04 g, 14.7 mmol) was added and the mixture stirred for 30 minutes, warmed to room temperature and stirred for 30 additional minutes. It

was then cooled to 0 °C and filtered to remove the precipitated *N,N'*-dicyclohexylurea byproduct. The filtrate was concentrated under vacuum and the residue dissolved in THF (100 mL). The solution was cooled to 0 °C, $NaBH_4$ (5.6 g, 150 mmol) added, and the mixture stirred for two minutes. Water was then added to the mixture dropwise until bubbling ceased, then a volume of water equal to the volume of THF was added over 30 minutes, after which the mixture was warmed to room temperature and stirred for 5 hours. The dioxane was removed under vacuum and EtOAc (200 mL) was added, followed by gradual acidification of the aqueous layer with 1 N aqueous HCl. The EtOAc layer was washed with brine and dried over anhydrous Na_2SO_4. Filtration followed by concentration under vacuum gave 4.31 g (12.4 mmol) of **8**, which was used without further purification. ^{1}H NMR (300 MHz, CD_3OD): δ 1.32 (s, 9H), 3.46-3.51 (*m*, 1H), 3.63-3.73 (*m*, 2H), 4.88 (br s, 1H), 7.29 (d, 2H, *J* = 8.3), 7.45 (d, 2H, *J* = 8.3).

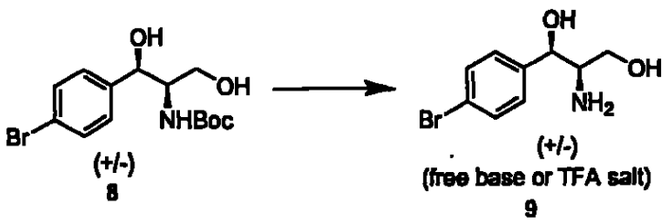
Example 6: Compound 9 (from 4)



4 **9**

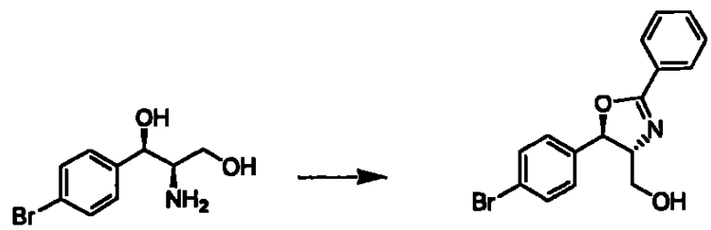
Compound **4** (1.0 g, 3.5 mmol) was dissolved in 10% aqueous H_2SO_4 (10% v/v; 15 mL total) and the solution refluxed for 10 h. The reaction mixture was basified with 3 M aqueous NaOH and extracted with EtOAc (3 x 50 mL). The combined EtOAc fractions were dried over anhydrous Na_2SO_4 and concentrated under vacuum to give 710 mg (2.9 mmol) of **4**, which was used without further purification. ^{1}H NMR (300 MHz, CD_3OD): δ 2.80-2.90 (br m, 1H), 3.30-3.39 (br m, 1H), 3.40-3.50 (br m, 1H), 4.52-4.58 (m, 1H), 7.30 (d, 2H, *J* = 8.1), 7.49 (d, 2H, *J* = 8.1).

Example 7: Compound 9 (from 8)



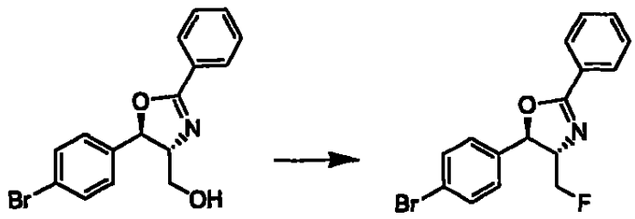
Compound **8** (4.31 g, 12.4 mmol) was dissolved in trifluoroacetic acid (TFA, 40 mL). The mixture was stirred for 3 hours at room temperature and concentrated under vacuum. The TFA salt was partitioned between 50 mL of 2 N aqueous NaOH and an equal volume of EtOAc. The layers were separated and the aqueous layer was washed with EtOAc (50 mL). The combined EtOAc fractions were washed with brine, dried over anhydrous Na_2SO_4 and concentrated under vacuum to give a waxy solid (2.72 g, 11.1 mmol), which was used without further purification. ^{1}H NMR (300 MHz, CD_3OD): δ 2.83-2.90 (m, 1H), 3.30-3.35 (m, 1H), 3.46 (dd, 1H, J = 10.8, 4.8), 4.56 (d, 1H, J = 6.6), 7.29 (d, 2H, J = 8.6), 7.49 (d, 2H, J = 8.6).

Example 8: Compound 10



Compound **9** (710 mg, 2.89 mmol), ethyl benzimidate hydrochloride (533 mg, 2.89 mmol) and triethylamine (Et_3N, 0.40 mL, 2.89 mmol) were combined in 25 mL 1,2-dichloroethane. The mixture was refluxed with stirring under N_2 for 16 hours at which time TLC indicated one major product. After cooling to room temperature, the mixture was diluted with EtOAc, washed twice with saturated aqueous NH_4Cl, twice with saturated aqueous $NaHCO_3$ and then dried over anhydrous Na_2SO_4. The solution was filtered and the EtOAc removed under vacuum to give 885 mg (2.66 mmol) of **10**, which was used without further purification. ^{1}H NMR (300 MHz, CD_3OD): δ 3.74-3.89 (m, 2H), 4.13-4.18 (m, 1H), 5.59 (d, 1H, J = 6.6), 7.30 (d, 2H, J = 8.4), 7.46-7.57 (m, 5H), 7.99-8.02 (m, 2H).

Example 9: Compound 11

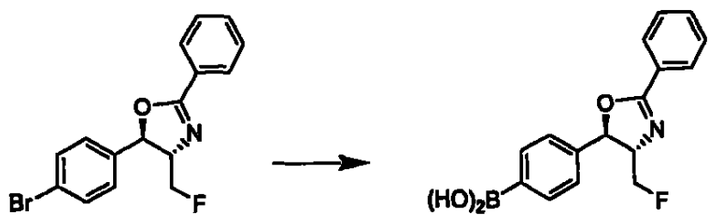


(enantiopure or (+/-))
10

(enantiopure or (+/-))
11

Compound **10** (885 mg, 2.66 mmol) was suspended in CH_2Cl_2 (15 mL) and cooled to –78 °C. Diethylaminosulfur trifluoride (DAST, 0.53 mL, 4.00 mmol) was added by syringe, the mixture warmed over 1 hour to room temperature and then stirred for 16 hours under N_2. Excess DAST was quenched by slow addition of H_2O after which the mixture was diluted with additional CH_2Cl_2. The organic layer was washed with H_2O and saturated aqueous $NaHCO_3$, dried over anhydrous Na_2SO_4 and concentrated under vacuum. The residue (950 mg) was chromatographed on silica gel, eluting with 15:85 EtOAc/hexanes to give **11** as an oil (420 mg, 1.25 mmol). ^1H NMR (300 MHz, $CDCl_3$): δ 4.23-4.35 (m, 1H), 4.45-4.75 (m, 2H), 5.43 (d, 1H, J = 6.9), 7.17(d, 2H, 8.3), 7.36-7.47 (m, 5H), 7.96 (d, 2H, J = 8.3).

Example 10: Compound 12



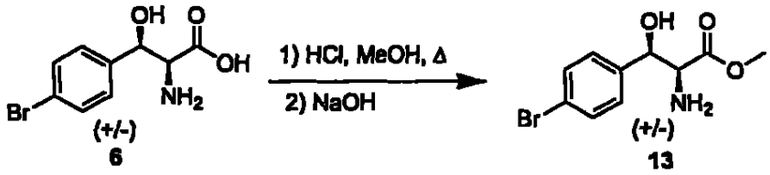
(enantiopure or (+/-))
11

(enantiopure or (+/-))
12

Compound **11** (807 mg, 2.41 mmol, enantiopure from chloramphenicol or racemic from (+/-)-*threo*-*p*-bromophenylserine), was dissolved in anhydrous THF (10

mL) in a flame-dried round bottom flask and cooled to –78 °C. *n*-BuLi (1.6 M in hexanes, 3.02 mL, 4.83 mmol) was added with vigorous stirring. After 10 minutes, trimethyl borate (0.55 mL, 4.83 mmol) was added, the mixture warmed to room temperature over 1 hour and then stirred for 6 hours. The mixture was quenched with 1 N aqueous HCl and extracted three times with EtOAc. The combined EtOAc fractions were washed three times with brine and dried over anhydrous Na_2SO_4. The EtOAc was removed under vacuum to give 805 mg of material, which was purified by flash column chromatography, eluting initially with 1:1 hexanes/EtOAc to remove impurities followed by elution with 1:9 MeOH/CH_2Cl_2 to obtain the product as an oil (285 mg, 0.953 mmol). ^{1}H NMR (300 MHz, CD_3OD): δ 4.29-4.39 (m, 1H), 4.69 (dd, 2H, *J* = 47.1, 4.2), 5.64 (d, 1H, *J* = 6.6), 7.37 (d, 2H, *J* = 8.1), 7.47-7.52 (m, 2H), 7.56-7.62 (m, 1H), 7.67 (d, 1H, *J* = 7.5) 7.74-7.82 (m, 1H), 8.01 (d, 2H, *J* = 7.2).). LRMS (ESI$^-$) m/z: 298.0 (M-H$^+$ $C_{16}H_{14}BFNO_3$ requires 298.1).

Example 11: Compound 13



Compound **6** (20.6 g, 0.079 mol) was suspended in 400 mL of anhydrous MeOH. The mixture was stirred rapidly and cooled in an ice bath to 0 °C. Anhydrous HCl gas was slowly bubbled in. After 10 minutes, all solids had dissolved. Acidification was continued for 10 minutes after the mixture became homogeneous, at which point the MeOH turned pH paper very red. The apparatus was fitted with a drying tube and the reaction mixture was refluxed for 8 hours followed by stirring at room temperature for 12 hours. The mixture was concentrated under vacuum and then suspended in a mixture of 300 mL of EtOAC and 100 mL of water. With rapid stirring, 3 N NaOH was added very slowly until the suspended material dissolved. Addition of base was continued until a pH of 10 was attained. The layers were separated and the EtOAc fraction was washed with brine (2x) and dried over anhydrous Na_2SO_4. Concentration under vacuum gave **13** as a white powder (15.3 g), which was used without further purification. ^{1}H NMR (300 MHz,

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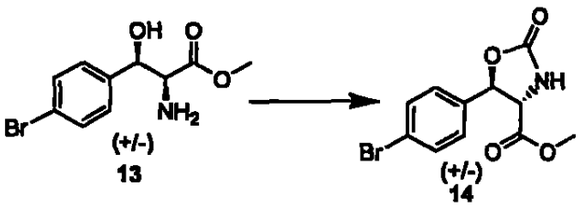
152

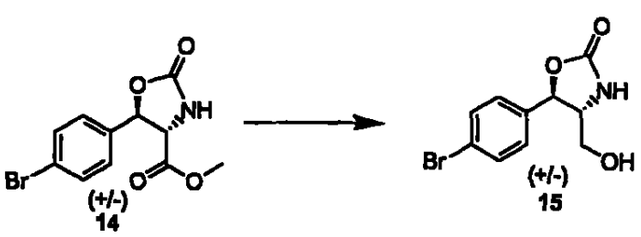
CD_3OD): δ 3.59 (d, 1H, J = 4.5), 3.66 (s, 3H), 4.92 (d, 1H, J = 4.5), 7.29 (d, 2H, J = 8.3), 7.49 (d, 2H, J = 8.3).

Example 12: Compound 14



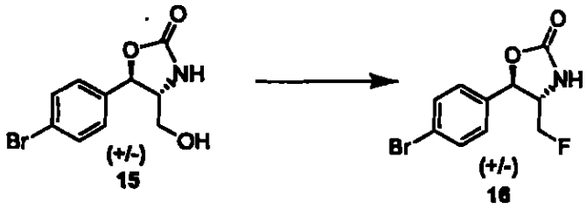
1,1'-Carbonyldiimidazole (23.6 g, 0.146 mol) was dissolved in 250 mL of anhydrous 1,2-dichloroethane at room temperature followed by addition of triethylamine (Et_3N, 12.2 g, 16.8 mL). In a separate flask, 13 (33.26 g, 0.121 mol) was dissolved in 125 mL of anhydrous tetrahydrofuran (THF) and an equivalent of triethylamine. The THF solution was diluted with 125 mL of 1,2-dichloroethane and then added, under N_2, over 2 hours to the 1,1'-carbonyldiimidazole solution. After addition was complete, the mixture was stirred for 2 hours under N_2 at room temperature. The reaction mixture was concentrated under vacuum and the residue diluted with 300 mL of EtOAc, which was washed with 5 x 200 mL of 2 N HCl, 3 x 300 mL of saturated $NaHCO_3$, and 2 x 200 mL of brine. The EtOAc layer was dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum to give 34.27 g of 14, 80-90% pure by ^1H NMR. Compound 14 was used without further purification. ^1H NMR(300 MHz, CD_3OD): δ 3.83 (s, 3H), 4.34 (d, 1H, J = 5.0), 5.65 (d, 1H, J = 5.0), 7.36 (d, 2H, J = 8.7), 7.60 (d, 2H, J = 8.7).

Example 13: Compound 15



Compound 14 (55.3 g, 0.184 mmol) was dissolved in 375 mL MeOH. The solution was cooled to 0 °C and $NaBH_4$ (2 equivalents, 13.9 g, 0.369 mol) was added in portions, care being taken to not let the temperature exceed 20 °C. After the last portion was added, cooling was ceased and the mixture stirred for 2 hours. Glacial acetic acid was added slowly until a pH of 7 was achieved. The mixture was filtered through Celite and concentrated under vacuum. The residue was partitioned between EtOAc (375 mL) and 2 N HCl (500 mL). The organic layer was washed with 2 N HCl (2 x 300 mL), saturated aqueous $NaHCO_3$ (2 x 250 mL) and brine (250 mL), dried over anhydrous Na_2SO_4, filtered and concentrated under vacuum. The product was crystallized from hexanes/EtOAc to give 26.3 g of 15. ^1H NMR(300 MHz, CD3OD): δ 3.64-3.72 (m, 3H), 5.38 (d, 1H, J = 5.1), 7.32 (d, 2H, J = 8.6), 7.57 (d, 2H, J = 8.6).

Example 14: Compound 16



Compound 15 (7.57 g, 0.0278 mol) was suspended in 275 mL of CH_2Cl_2. Under N_2 and with rapid stirring, the biphasic mixture was cooled to –78 °C in a dry ice/acetone bath. DAST (5.51 mL, 6.72 g, 0.0417 mol) was added dropwise by syringe over 2-3 minutes. After 30 minutes the mixture was transferred to an ice water bath and stirred for 30 minutes, during which time the solids dissolved. Another 1 mL of DAST was added to ensure completion of the reaction. The mixture was stirred for 20 minutes at 0 °C and then warmed to room temperature. The mixture was stirred at room temperature for 20 minutes and then cooled to 0 °C. Excess DAST was quenched by dropwise addition of saturated aqueous $NaHCO_3$ over 30 minutes with very rapid stirring. After bubbling ceased, the aqueous layer was slightly basic (7 < pH < 9). The CH_2Cl_2 was diluted with enough EtOAc (~600 mL) to bring the organic layer to the top during aqueous extraction. The organic layer was washed with saturated aqueous $NaHCO_3$ (1 x 300 mL), 1 N HCl (2 x 200 mL), saturated aqueous $NaHCO_3$ (3 x 200 mL) and brine (1 x 150 mL) and dried

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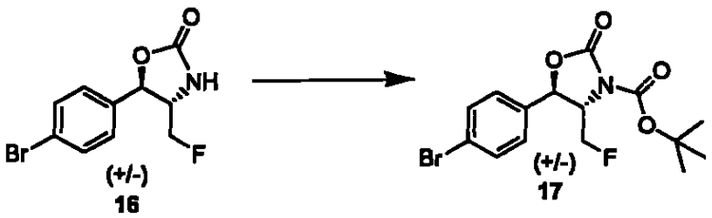
154

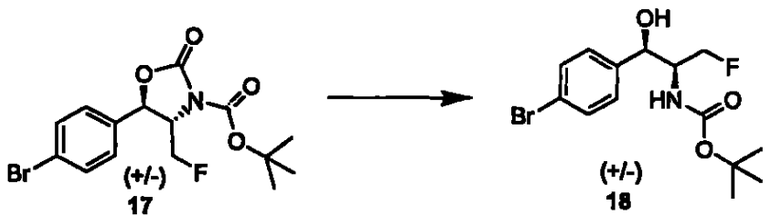
over anhydrous Na_2SO_4. After filtration, the mixture was concentrated under vacuum to give 8.13 g of brown oil. ^1H NMR(300 MHz, CD_3OD): δ 3.87-3.98 (dm, 1H, J = 20 ($F\text{-}CH_N$)), 4.54 (dd, 2H, J = 46.7 (CH_2-F), 4.2), 5.42 (d, 1H, J = 5.1), 7.34 (d, 2H, J = 8.4), 7.59 (d, 2H, J = 8.4).

Example 15: Compound 17



Compound 16 (8.13 g or 0.0297) was dissolved in 200 mL of CH_3CN. Boc_2O (9.71 g, 1.5 equivalents, 0.0445 mol) and DMAP (362 mg, 0.00297 mol, 0.1 equivalent) were then added. The mixture was stirred for 2 hours at room temperature under N_2. Concentration of the mixture under vacuum was followed by partitioning between 200 mL of EtOAc and 200 mL of 1 N HCl. The EtOAc layer was washed with 2 N HCl (3 x 100 mL), saturated aqueous $NaHCO_3$ (2 x 100 mL) and brine. The EtOAc layer was filtered, dried over anhydrous Na_2SO_4 and concentrated under vacuum. The residue was recrystallized twice from hexanes/EtOAc. ^1H NMR (300 MHz, CD_3OD): δ 1.53 (s, 9H), 4.29-4.40 (dm, 1H, J = 24.6), 4.63-4.98 (m, 2H), 5.53 (d, 1H, J = 4.2), 7.34 (d, 2H, J = 8.4), 7.61, (d. 2H, J = 8.4).

Example 16: Compound 18



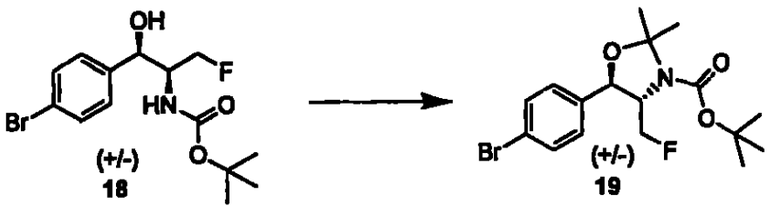
Compound 17 (0.050 g, 0.134 mmol) was suspended in 5 mL of CH_3OH. Cs_2CO_3 (8.7 mg, 20 mol%, 0.0267 mmol) was added with rapid stirring at room temperature. The solids dissolved in about 10 minutes after which stirring was continued for 10 minutes, at which time TLC (3:1 hexanes/EtOAc) indicated that the

reaction was complete. The reaction mixture was concentrated under vacuum and the residue partitioned between 20 mL of EtOAc and 10 mL of H_2O. The H_2O layer was acidified slightly with 1N aqueous HCl, the mixture vigorously shaken and the layers separated. The EtOAc layer was washed with saturated aqueous $NaHCO_3$ (1 x 20 mL) and brine (1 x 20 mL). The EtOAc was dried over anhydrous Na_2SO_4 and concentrated under vacuum to give 43.4 mg (0.124 mmol) of **18** as a white foam. ^1H NMR (300 MHz, CD_3OD): δ 1.33 (s, 9H), 3.90-4.57 (m, 4H), 7.29 (d, 2H, J = 8.1), 7.46 (d. 2H, J = 8.1)

Example 17: Compound 22

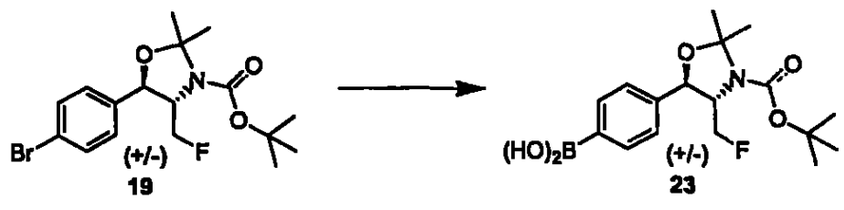
Compound **18** (0.740 g, 2.13 mmol) was stirred in 10 mL TFA/H_2O (9/1, v/v) at room temperature for one hour. The mixture was concentrated under vacuum and partitioned between 30 mL of EtOAc and 30 mL of 1N aqueous NaOH. The mixture was shaken vigorously and the layers allowed to separate. The organic layer was washed with brine (2 x), dried over anhydrous Na_2SO_4 and concentrated under vacuum to give 0.484 g (1.95 mmol, 92%) of product, which was dissolved in 20 mL of CH_2Cl_2. To this was added 1 mL of Et_3N and 1 mL of difluoroacetyl chloride, prepared according to literature procedure (Yu, K.-L., et al., *J. Med. Chem.*, **1996**, *39*, 2411-2421.). After 1 hour, the mixture was concentrated under vacuum. The residue was placed in 10 mL of a 1:8:1 (v/v/v) solution of Et_3N/MeOH/H_2O and stirred for one hour at room temperature to cleave any difluoroacetyl groups on the benzylic oxygen. The mixture was then concentrated under vacuum, and partitioned between EtOAc (50 mL) and 1 N HCl (50 ml). The EtOAc layer was washed twice with 20 mL of 1 N HCl, twice with 20 mL saturated aqueous $NaHCO_3$ and twice with 20 mL brine. The EtOAc was then dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum. Compound **22** (584 mg, 1.79 mmol) was used without further purification. ^1H NMR (300 MHz, CD_3OD): δ 4.25-4.67 (m, 3H), 4.90 (d, 1H, J = 3.9), 5.98 (t, 1H, J = 53.9), 7.30 (d. 2H, J = 8.6), 7.48 (d, 2H, J = 8.6).

Example 18: Compound 19



Compound **18** (1.03 g, 2.94 mmol) was dissolved in CH_2Cl_2 (30 mL) in a 50 mL round-bottom flask. 2-Methoxypropene (339 µL, 3.54 mmol) was added by syringe, followed by a single crystal of *p*-toluenesulfonic acid (*p*-TsOH) hydrate. After about 1 minute, the mixture turned yellow. TLC (4:1 hexanes/EtOAc) showed some **18** to still be present. An additional 5 drops of 2-methoxypropene were added followed two minutes later by another 5 drops, a which time TLC indicated the reaction was complete. The CH_2Cl_2 was removed under vacuum and the residue partitioned between EtOAc (75 mL) and saturated aqueous $NaHCO_3$ (50 mL). The EtOAc layer was washed twice with 50 mL $NaHCO_3$ and twice with brine. The EtOAc layer was dried over anhydrous Na_2SO_4, filtered and evaporated to give a yellow oil (1.08 g, 2.78 mmol) that solidified to a waxy solid, which was used without further purification. ^1H NMR (300 MHz, CD_3OD): δ 1.49 (s, 9H), 1.55 (s, 3H), 1.67 (s, 3H), 3.70-3.85 (m, 1H), 4.34-4.53 (m, 2H), 5.08, (d. 1H, J = 7.5), 7.37 (d, 2H, J = 8.6), 7.54 (d, 2H, J = 8.6).

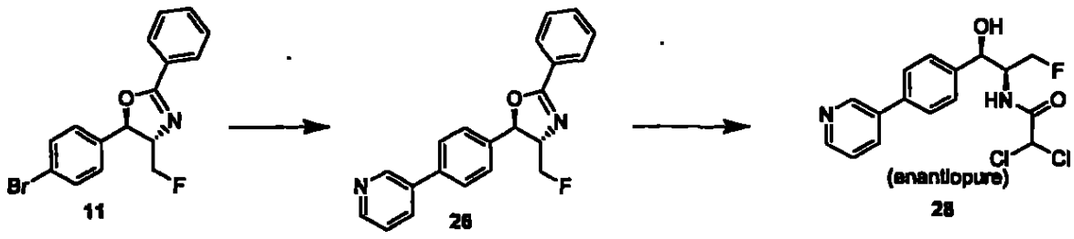
Example 19: Compound 23



Compound **19** (309 mg, 0.796 mmol) was placed in an oven-dried 50 mL round-bottom flask equipped with a magnetic stir bar. The flask was immediately capped with a rubber septum and flushed with dry N_2. Anhydrous THF (15 mL) was added by syringe and a long cannula was used to flush the solvent with N_2 for 10 minutes. The mixture was then cooled to –78 °C. With vigorous stirring, *n*-BuLi (850 µL, 0.995 mmol) was added dropwise by syringe over 3 minutes to give a clear

orange solution. After 20 minutes, $B(OMe)_3$ (158 µL, 1.39 mmol), which had been stored over 4Å activated molecular sieves for at least 48 hours, was added dropwise over one minute using an oven-dried, gas-tight syringe. The mixture was warmed to room temperature and stirred under N_2 for 16 hours. Approximately 20 mL of saturated aqueous NH_4Cl was added with vigorous stirring, which resulted in a white precipitate dispersed in the two liquid phases. The mixture was stirred for an additional 90 minutes and then diluted with EtOAc and H_2O until all the precipitate dissolved. The organic layer was separated, washed with brine and dried over anhydrous Na_2SO_4. After filtration, the EtOAc was removed under vacuum to give 328 mg of yellow oil. The product was purified by silica gel chromatography to give **23** (163 mg). ^1H NMR (300 MHz, CD_3OD): δ 1.49 (s, 9H), 1.56 (s, 3H), 1.67 (s, 3H), 3.74-3.88 (m, 1H), 4.33-1.52 (m, 2H), 5.10 (d, 1H, J = 7.5), 7.43 (d, 2H, J = 8.1), 7.64 (d, 2H, J = 8.1). LRMS (ESI$^-$) m/z: 352.2 ($M\text{-}H^+$ $C_{17}H_{24}BFNO_5$ requires 352.7).

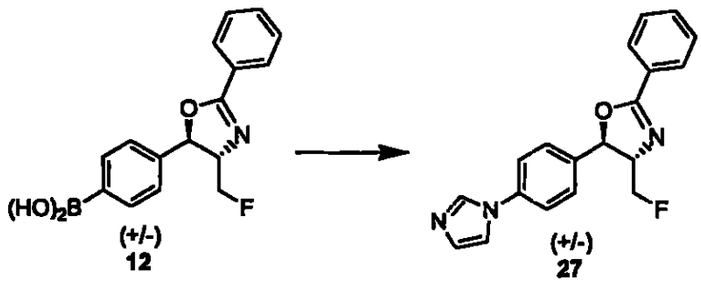
Example 20: Compound 28



This is an example of Suzuki coupling method A. Compound **11** (47 mg, 0.14 mmol) and pyridine-3-boronic acid (21 mg, 0.17 mmol) were dissolved in 5.0 mL of THF. $Pd(PPh_3)_4$ (11 mg, 0.0098 mmol) and 3.0 mL of 10% (w/v) aqueous Na_2CO_3 were added. The reaction mixture was refluxed for 16 hours, cooled, diluted with EtOAc and washed twice with 10% (w/v) aqueous Na_2CO_3 and twice with brine. The EtOAc layer was then dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum. Silica gel chromatography (eluting with 1:1 hexanes/EtOAc) gave 47 mg (0.14 mmol) of **26**. Intermediate **26** was heated to 105 °C in a sealed tube with 6M aqueous HCl and held at that temperature for 16 hours. The mixture was cooled to room temperature and basified to pH 10 with 3 N NaOH. The product was extracted into EtOAc, which was dried over Na_2SO_4, filtered, and concentrated under vacuum to give 42 mg of the deprotected free amine, contaminated with the

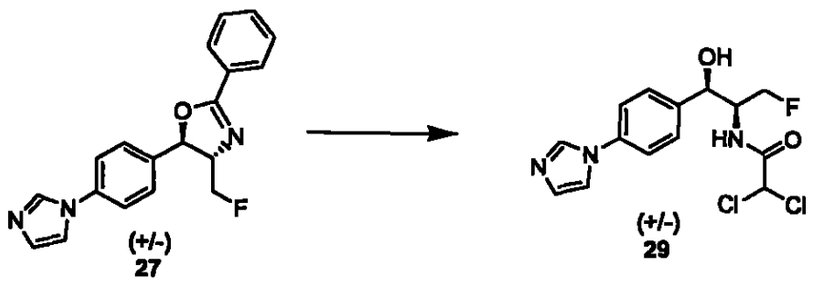
expected benzoic acid-related by-product arising from cleavage of the protecting group. The crude residue was dichloroacetylated as in the synthesis of **29**. Silica gel chromatography, eluting with 7.5% MeOH in CH_2Cl_2, gave 10 mg of product. ^1H NMR (300 MHz, CD_3OD): δ 4.31-4.74 (m, 3H), 5.02 (d, 1H, *J* = 3.6), 6.27 (s, 1H), 7.49-7.56 (m, 3H), 7.64 (d, 2H, *J* = 8.4), 8.09 (ddd, 1H, *J* = 8.0, 2.3, 1.5), 8.50 (br d, 1H, *J* = 3.9), 8.78 (br s, 1H). LRMS (ESI^+) m/z: 356.9 (calc. for $M+H^+$: $C_{16}H_{16}Cl_2FN_2O_2$ 357.0).

Example 21: Compound 27



Boronic acid **12** (43 mg, 0.144 mmol), imidazole (15 mg, 0.215 mmol) and 4Å powdered molecular sieves (110 mg) were combined in CH_2Cl_2 (4.0 mL) and pyridine (23 μL). With rapid stirring, $Cu(OAc)_2$ (26 mg, 0.144 mmol) was added and the mixture stirred, exposed to air, for 40 hours at room temperature. The mixture was quenched with 3 mL of 2 M NH_3 in MeOH. The mixture was filtered through Celite and concentrated under vacuum. The product was purified by flash column chromatography (19:1 CH_2Cl_2/MeOH). Although ^1H NMR indicated that a significant side-product had eluted with the desired product, the mixture was used without further purification. LRMS (ESI^+) m/z: 321.9 (calc. for $M+H^+$ $C_{19}H_{17}FN_3O$ 322.1).

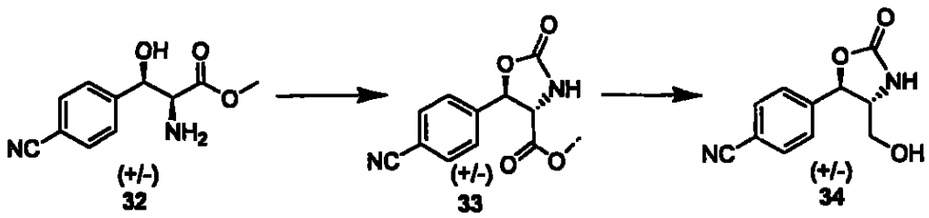
Example 22: Compound 29



Compound 27 (27 mg, 0.084 mmol) was heated with 6 N HCl (3.0 mL) to 100 °C in a sealed tube and held for 16 hours. The mixture was cooled to room temperature, basified with 3 N aqueous NaOH and extracted three times with EtOAc. The combined EtOAc fractions were dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum to give 9 mg of residue. LRMS (ESI^+) m/z: 236.1 (Calc. for $M+H^+$ $C_{12}H_{15}FN_3O$ 236.1).

The residue was refluxed in MeOH (5.0 mL) containing Et_3N (26 µL 0.189 mmol) and methyl dichloroacetate (13 µL, 0.126 mmol) for 3 hours. The mixture was concentrated under vacuum and purified by silica gel chromatography (7.5% MeOH in CH_2Cl_2) to give 29 (6.0 mg, 0.017 mmol). ^1H NMR (300 MHz, CD_3OD): δ 4.33-4.75 (m, 3H), 5.03 (d, 1H, J = 3.3), 6.26 (s, 1H), 7.13 (s, 1H), 7.51-7.58 (m, 5H), 8.10 (s, 1H). LRMS (ESI^+) m/z: 345.8 (calc. for $M+H^+$ $C_{14}H_{15}Cl_2FN_3O_2$ 346.0).

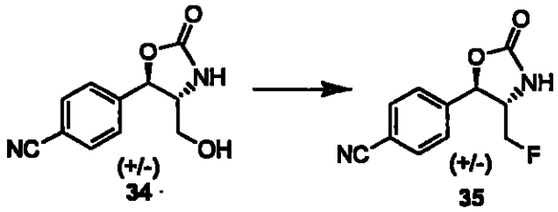
Example 23: Compound 34



Compound 32 was prepared by the procedure of Pines, et al., *supra*. In a 250-mL round-bottom flask was placed 1,1'-carbonyldiimidazole (CDI, 5.06 g, 31.2 mmol) and Et_3N (2.17 mL, 15.6 mmol) in 1,2-dichloroethane (50 mL). Compound 32 (4.00 g, 15.6 mmol) was added to a beaker containing 1,2-dichloroethane (50 mL). Et_3N (4.34 mL, 31.2 mmol) was added to the beaker upon which a thick suspension formed. Additional 1,2-dichloroethane (20 mL) and THF (50 mL) were added to thin the suspension. The mixture was added portion-wise to the CDI suspension over 30 min. The mixture, which became mostly homogenous and turned yellow was stirred for 12 hours. The solvent was removed under vacuum and the residue was partitioned between 2 N aqueous HCl and EtOAc. The EtOAc layer was washed with 2 N HCl (2 x 45 mL), saturated aqueous $NaHCO_3$ (2 x 30 mL) and brine. It was dried over anhydrous Na_2SO_4, filtered, and evaporated to give 3.27 g of 33 as a fluffy yellow solid, which was used without further purification.

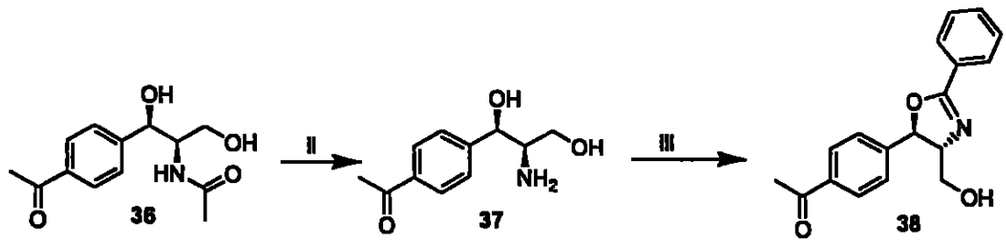
Compound 33 (1.00 g, 4.06 mmol) was dissolved in MeOH (50 mL) and cooled to 0 °C in an ice bath. Over 5 minutes, $NaBH_4$ (308 mg, 8.13 mmol) was added portion-wise. After bubbling ceased, TLC (5% MeOH in CH_2Cl_2) indicated that starting material remained so 100 mg additional $NaBH_4$ was added. After bubbling ceased, the mixture was quenched with glacial HOAc until a pH of approximately 7 was achieved. The mixture was concentrated under vacuum and partitioned between EtOAc and 1 N HCl. The EtOAc layer was washed with saturated aqueous $NaHCO_3$ (2 x 40 mL) and brine (40 mL) and dried over anhydrous Na_2SO_4. The product was isolated by silica gel chromatography (2% to 3% to 4% to10% MeOH in CH_2Cl_2) to give 128 mg of 34 as an oil. Additional product was obtained by evaporation of the aqueous layer followed by trituration with hot EtOAc (153 mg, 281 mg total). ^1H NMR (300 MHz, CD_3OD): δ 3.69 (br s, 3H), 5.50 (br s, 1H), 7.58 (d, 2H, J = 7.8), 7.79 (d, 2H, J = 7.8).

Example 24: Compound 35



Fluorination was performed using DAST as in the synthesis of compound 11. ^1H NMR (300 MHz, CD_3OD): δ 3.89-3.97 (m, 1H), 4.49-4.66 (m, 2 H), 5.55 (d, 1H, J = 4.8), 7.59 (d, 2H, J = 8.1), 7.80 (d, 2H, J = 8.4).

Example 25: Compound 38

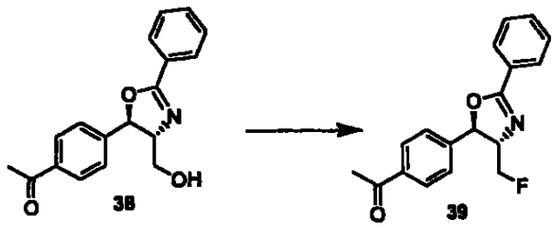


Compound 36 (2.12 g), was prepared by the procedure of von Strandtmann, *supra*. It was heated with 10% aqueous H_2SO_4 to 100 °C in a sealed tube and held for 4 hours. After cooling to room temperature, the mixture was basified with 1 M

NaOH and extracted three times with *n*-butanol. The *n*-butanol fractions were combined and dried over anhydrous Na_2SO_4. The solvent was evaporated to give 1.61 g of **37**.

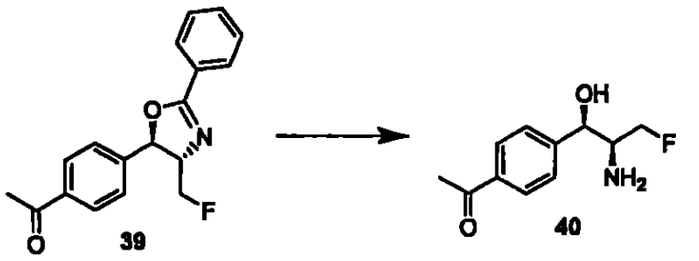
Compound **37** (1.61 g, 7.70 mmol) was dissolved in 1,2-dichloroethane (100 mL), along with ethyl benzimidate hydrochloride (1.42 g, 7.70 mmol), and Et_3N (1.06 mL, 7.70 mmol). The mixture was refluxed for 12 hours, cooled to room temperature and diluted with EtOAc. The solution was washed with saturated aqueous NH_4Cl (3 x) and saturated $NaHCO_3$ (1 x) and dried over anhydrous Na_2SO_4. The product precipitated from the EtOAc on cooling and addition of several drops of hexanes to give 0.62 g of **38** (2.1 mmol). ^1H NMR (300 MHz, CD_3OD): δ 2.60 (s, 3H), 3.79 (dd, 1H, J = 11.3, 5.9), 3.89 (dd, 1H, J = 11.3, 4.1), 4.14-4.20 (m, 1H), 5.70 (d, 1H, 6.3), 7.47-7.61 (m, 5H), 8.01-8.05 (m, 4H).

Example 26: Compound 39



Compound **38** (0.62 g, 2.1 mmol) was suspended in CH_2Cl_2 (15 mL) and cooled to –78 °C. DAST (0.42 mL, 3.1 mmol) was added by syringe and the solution stirred overnight, during which time it was allowed to come to room temperature as the cold bath warmed. The solvent was removed under vacuum and the product isolated by silica gel chromatography (1:4 EtOAc/hexanes) to give 0.17 g of **39**. ^1H NMR (300 MHz, CD_3OD): δ 2.60 (s, 3H), 4.30-4.40 (m, 1H), 4.63-4.67 (m, 1H), 4.79-4.81 (m, 1H), 5.74 (d, 1H, 6.9), 7.48-7.63 (m, 5H), 8.01-8.07 (m, 4H).

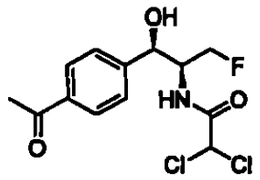
Example 27: Compound 40



39 40

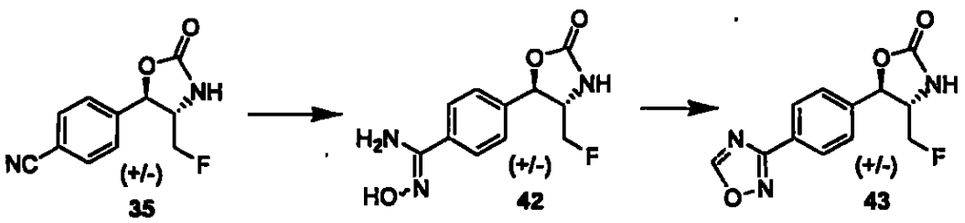
Compound **39** (0.17 g, 0.58 mmol) was suspended in 6 N aqueous HCl (5 mL) in a sealed tube. The mixture was heated to 100 °C and held for 12 hours. After cooling to room temperature, the mixture was basified with 3 N NaOH and the product extracted into CH_2Cl_2 (3 x). The combined CH_2Cl_2 fractions were dried over anhydrous Na_2SO_4, filtered and concentrated to give **40** (0.113 g, 0.541 mmol). ^1H NMR (300 MHz, CD_3OD): δ 2.60 (s, 3H), 3.04-3.15 (m, 1H), 4.09-4.48 (m, 3H), 4.71 (d, 1H, J = 6.0), 7.52 (d, 2H, J = 8.1), 7.90 (d, 2H, J = 8.1).

Example 28: Compound 41



Compound **40** was dichloroacetylated to give **41** in the same manner that **27** was converted to **29**. ^1H NMR (300 MHz, CD_3OD): δ 2.57 (s, 3H), 4.31-4.78 (m, 3H,), 5.03 (d, 1H, J = 3.3), 6.23 (s, 1H), 7.53 (d, 2H, J = 8.6), 7.95 (d, 2H, J = 8.6).

Example 29: Compound 43

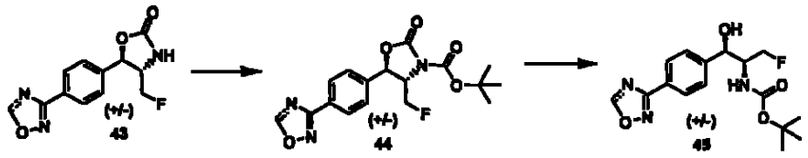


35 42 43

Compound **35** (100 mg, 0.454 mmol) was dissolved in 3 mL of EtOH and transferred to a 25 mL round-bottom flask. Hydroxylamine hydrochloride (38 mg, 0.545 mmol) was added followed by Et_3N (127 µL, 0.909 mmol). The mixture was stirred at reflux for 3 hours, at which point TLC indicated complete consumption of

starting material. The mixture was concentrated under vacuum to 232 mg of yellow oil, which was used without further purification. A portion of the oil (115 mg) was dissolved in 10 mL of triethylorthoformate. The mixture was stirred at 120 °C under N_2 for 2 hours and then at room temperature for 48 hours. TLC (10% MeOH in CH_2Cl_2) a major new spot. The triethyl orthoformate was removed under vacuum and the residue partitioned between EtOAc and 1 N aqueous NaOH. The organic layer was washed with 1 N aqueous NaOH (2 x) and brine (2 x). The EtOAc layer was dried over anhydrous Na_2SO_4, filtered, and evaporated, to give 84 mg of oil. The product was purified by chromatotron (1 mm silica plate, 60:40 to 50:50 to 40:60 hexanes/EtOAc). Another purification by chromatotron (2% MeOH in CH_2Cl_2) gave **43** (20 mg) as a white solid. ^{1}H NMR (300 MHz, CD_3OD): δ 3.93-4.04 (m, 1H), 4.49-4.67 (m, 2 H), 5.53 (d, 1H, *J* = 5.1), 7.59 (d, 2H, *J* = 8.3), 8.17 (d, 2H, *J* = 8.3), 9.28 (s, 1H).

Example 30: Compound 45

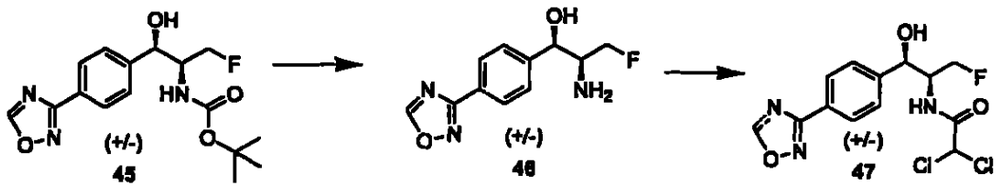


Compound **43** (20 mg, 0.076 mmol) was dissolved in CH_3CN (2 mL). Boc_2O (25 mg, 0.114 mmol) was added followed by a single crystal of DMAP (approximately 1 mg, 0.008 mmol). After 2 hours, TLC (1:1 hexanes/EtOAc) indicated that the reaction was complete. The CH_3CN was removed under vacuum and the resulting white solid dissolved in EtOAc, which was washed with 1 N aqueous HCl (2 x), followed by $NaHCO_3$ (2 x) then brine. The organic layer was dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum to give **44** as a white solid (28 mg).

Compound **44** (0.026 mg, 0.072 mmol) was dissolved in MeOH (2 mL) and Cs_2CO_3 (5 mg, 0.014 mmol) was added in a single portion. After 1 hour, TLC (1:1 hexanes/EtOAc) showed a single product. The solvent was removed under vacuum, EtOAc added and the mixture stirred rapidly at room temperature. Then, H_2O was added, followed by 0.5 M HCl dropwise until all solids dissolved. The organic phase was washed with 0.5 N HCl, followed by saturated aqueous $NaHCO_3$ (2 x) and brine.

The organic layer was then dried over anhydrous Na_2SO_4, filtered and concentrated to give a yellow oil, which was purified by chromatotron (1 mm silica plate, 4:1 hexanes/EtOAc to 1:1 hexanes/EtOAc) to give **45** (13 mg, 0.039 mmol). ^{1}H NMR (300 MHz, CD_3OD): δ 1.31 (s, 9H), 4.01-4.63 (m, 3 H), 4.94 (d, 1H, *J* = 2.7), 7.55 (d, 2H, *J* = 8.4), 8.05 (d, 2H, *J* = 8.4), 9.24 (s, 1H).

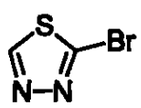
Example 31: Compound 47



Compound **45** was dissolved in 2.5 mL of 9:1 TFA/H_2O in a 10 mL flask. The mixture was stirred at room temperature for 30 minutes, at which time TLC indicated completion of the reaction. The mixture was concentrated under vacuum and partitioned with rapid stirring between 1 N aqueous NaOH and EtOAc. The EtOAc layer was washed with brine (1 x), dried over anhydrous Na_2SO_4, filtered and evaporated to give 11 mg of **46** as an oil .

To 6 mg (0.025 mmol) of **46** in a 10 mL round bottom flask was added MeOH (2 mL), followed by Et_3N (3.5 μL, 0.025 mmol) and methyl dichloroacetate (13.1 μL, 0.025 mmol). The mixture was refluxed for 4 hours at which time TLC indicated no reaction had occurred. An additional 26.2 μL of methyl dichloroacetate was added followed by 7 μL of Et_3N. The mixture was refluxed for 20 hours after which TLC indicated that the reaction was complete. The product was purified via chromatotron (1mm silica gel plate, 99:1 CH_2Cl_2/MeOH) to give 5 mg **47**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.72 (m, 3H), 5.03 (d, 1H, *J* = 2.7), 6.25 (s, 1H), 7.56 (d, 2H, *J* = 8.3), 8.05 (d, 2H, *J* = 8.3), 9.24 (s, 1H). LRMS (ESI$^-$) m/z: 346.0 ($M-H^+$ $C_{13}H_{11}Cl_2FN_3O_3$ requires 346.0).

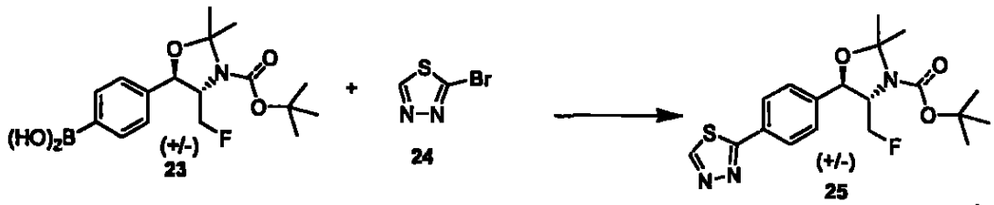
Example 32: Compound 24



24

In a 200 mL round-bottom flask, 2-amino-1,3,4-thiadiazole (1.00 g, 9.89 mmol) was added to 10 mL of 48% aqueous HBr. Water (10 mL) was added to give a yellow solution in which most solids dissolved. The mixture was cooled to 0 °C and CuBr (142 mg, 0.989 mmol) was added to give an opaque brown solution with some precipitate. $NaNO_2$ (682 mg, 9.89 mmol) was dissolved in 25 mL H_2O and added dropwise over 45 minutes to the thiadiazole mixture. The mixture became dark green and opaque with the first drops of $NaNO_2$ solution. Slowly, the solution became brownish yellow and brown gas evolved. The mixture was stirred for an additional 10 minutes at 0 °C from the time gas evolution began. The mixture was warmed to room temperature over 30 minutes. Saturated aqueous $NaHCO_3$ was added dropwise until bubbling ceased and the pH was 8.5. To this was added 50 mL of EtOAc and the biphasic mixture stirred rapidly. The mixture was filtered through a pad of Celite to remove solids and then the layers were separated. The organics were washed with brine (2 x). The aqueous layer was extracted with EtOAc and washed with brine (1 x). The combined organic layers were dried over Na_2SO_4. The remaining aqueous material was extracted once more by vigorous stirring with EtOAc (50 mL) for 12 hours. This EtOAc layer was washed with brine and combined with the EtOAc that was already drying over Na_2SO_4. The solution was filtered and evaporated under vacuum to give 24 as a tan solid (1.22 g, 7.36 mmol), which was used without further purification.

Example 33: Compound 25

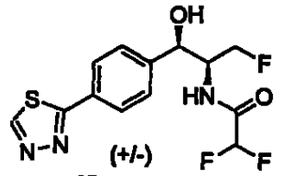


This is an example of Suzuki coupling method B. Compound 23 (19 mg, 0.0538 mmol) was dissolved in THF (1 mL), DMF (1 mL), and H_2O (0.5 mL). The mixture was stirred at room temperature until all material dissolved and then 2-bromo-1,3,4-thiadiazole (24, 5.0 mg, 0.027 mmol) was added. The solution was purged with N_2 for 5 minutes by means of a long needle. $PdCl_2(dppf)$ was added and then the solution was again purged with N_2 for 5 minutes. The mixture was then

stirred at 55 °C for 18 hours under N_2 and then at room temperature for an additional 30 hours. The resulting clear brown solution was concentrated under vacuum. Compound **25** (5.4 mg, 0.0137 mmol) was obtained as a yellow oil by chromatotron purification (silica gel, 1 mm plate, 1:9 EtOAc/hexanes to 1:4 EtOAc/hexanes).

It was found that the yield could be substantially improved by switching to coupling method C. Compounds **23** (0.330 g, 0.934 mmol) and **24** (0.385 g, 2.34 mmol) were dissolved in a mixture of toluene, *n*-butanol, and H_2O (8 mL:8 mL:2 mL) and Cs_2CO_3 (0.912 g, 2.80 mmol) was added. The mixture was purged with N_2 and $Pd(PPh_3)_4$ was added. The mixture was again purged with N_2 for 5 minutes and then stirred rapidly at 70 °C under N_2 for 12 hours. The mixture was concentrated under vacuum and the residue partitioned between H_2O (75 mL) and EtOAc (75 mL. The layers were separated and the EtOAc was washed with brine (2 x 50 mL). The EtOAc was dried over anhydrous Na_2SO_4, filtered, and concentrated under vacuum to give 0.720 g of yellow oil. Compound **25** (0.220 g) was obtained by silica gel chromatography, eluting with 5:1 to 2:1 to 1:1 hexanes/EtOAc. ^{1}H NMR (300 MHz, CD_3OD): δ 1.50 (s, 9H), 1.58 (s, 3H), 1.70 (s, 3H), 3.80-3.94 (m, 1H), 4.42-4.61 (m, 2H), 5.20, (d. 1H, *J* = 7.5), 7.64 (d, 2H, *J* = 8.3), 8.05 (d, 2H, *J* = 8.3), 9.45 (s, 1H).

Example 34: Compound 48

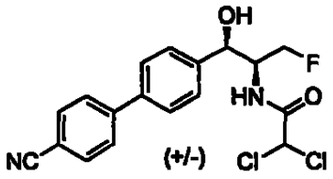


48

Compound **25** (5.4 mg, 0.014 mmol) was transferred to a 25 mL round-bottom flask and 2.5 mL of 9:1 TFA/H_2O (v/v) were added. The resulting bright yellow mixture was stirred at room temperature for 18 hours. The solvent was removed under vacuum and the residue dissolved three times in a mixture of MeOH and toluene, which was evaporated to near dryness each time. The product was recovered as an oil (4.9 mg, 0.013 mmol), which was used without further purification. ^{1}H NMR (300 MHz, CD_3OD): δ 3.59-3.70 (m, 1H), 4.30-4.71 (m, 2H), 4.92 (d, 1H, *J* = 8.1), 7.64 (d, 2H, *J* = 8.3), 8.08 (d, 2H, *J* = 8.3), 9.47 (s, 1H).

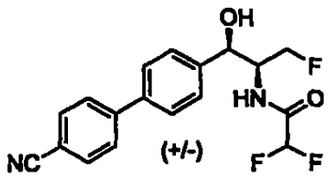
In a 10 mL round-bottom flask, the above product was dissolved in 2 mL of MeOH. To this was added Et_3N (9.7 µL, 0.070 mmol) followed by methyl difluoroacetate (3.0 µL, 0.035 mmol). The mixture was refluxed for 16 hours, at which point TLC indicated that some starting material still remained. An additional 3 drops of Et_3N was added followed by 2 drops of methyl difluoroacetate. After 3 hours, all starting material had been consumed. The mixture was cooled to room temperature and evaporated to dryness under vacuum. Compound **48** (4.1mg, 012 mmol) was obtained by chromatotron purification (silica gel, 1 mm plate). ^1H NMR (300 MHz, CD_3OD): δ 4.31-4.72 (m, 3H), 5.03 (d, 1H, J = 3.9), 5.98 (t, 1H, J = 53.9), 7.58 (d, 2H, J = 8.3), 7.99 (d, 2H, J = 8.3), 9.43 (s, 1H). LRMS (ESI^-) m/z: 330.1 ($M-H^+$ $C_{13}H_{11}F_3N_3O_2S$ requires 330.1).

Example 35: Compound 49



The biaryl intermediate was prepared from **18** and the appropriate boronic acid using Suzuki coupling method A. Removal of the Boc group was accomplished by brief treatment with 90/10 (v/v) TFA/H_2O. Dichloroacetylation was performed as in the synthesis of **29**. ^1H NMR (300 MHz, CD_3OD): δ 4.31-4.80 (m, 3H), 5.01 (d, 1H, J = 3.6), 6.26 (s, 1H), 7.52 (d, 2H, 8.3), 7.65 (d, 2H, J = 8.3), 7.79 (s. 4H). LRMS (ESI^-) m/z: 379.0 ($M-H^+$ $C_{18}H_{14}Cl_2FN_2O_2$ 379.0).

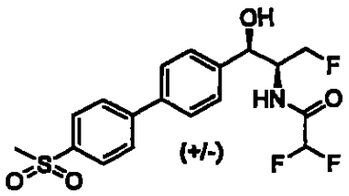
Example 36: Compound 50



The biaryl intermediate was prepared from **22** and the appropriate boronic acid using Suzuki coupling method A. ^1H NMR (300 MHz, CD_3OD): δ 4.27-4.68 (m, 3H), 4.99 (d, 1H, J = 4.5), 6.00 (t, 1H, J = 53.9), 7.52 (d, 2H, J = 8.1), 7.67 (d, 2H, J =

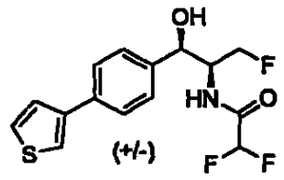
8.1), 7.76-7.82 (m, 2H). LRMS (ESI⁻) m/z: 347.1 (M-H⁺ $C_{18}H_{14}F_3N_2O_2$ requires 347.1).

Example 37: Compound 51



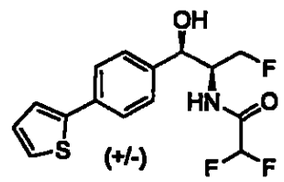
The biaryl intermediate was prepared from **22** and the appropriate boronic acid using Suzuki coupling method A. ^{1}H NMR (300 MHz, CD_3OD): δ 3.14 (s, 3H), 4.30-4.68 (m, 3H), 5.00 (d, 1H, *J* = 4.5), 6.00 (t, 1H, *J* = 54), 7.53 (d, 2H, *J* = 8.3), 7.70 (d, 2H, *J* = 8.3), 7.88 (d, 2H, *J* = 8.6), 8.00 (d, 2H, *J* = 8.6). LRMS (ESI⁻) m/z: 400.1 (M-H⁺ $C_{18}H_{17}F_3NO_4S$ requires 400.1).

Example 38: Compound 52



The biaryl intermediate was prepared from **22** and the appropriate boronic acid using Suzuki coupling method B. ^{1}H NMR (300 MHz, CD_3OD): δ 4.26-4.64 (m, 3H), 4.93 (d, 1H, *J* = 4.8), 6.01 (t, 1H, *J* = 53.9), 7.40-7.45 (m, 4H), 7.60-7.66 (m, 3H). LRMS (ESI⁻) m/z: 328.1 (M-H⁺ $C_{16}H_{13}F_3NO_2S$ requires 328.1).

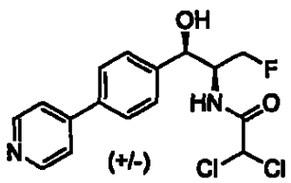
Example 39: Compound 53



The biaryl intermediate was prepared from **22** and the appropriate boronic acid using Suzuki coupling method B. ^{1}H NMR (300 MHz, CD_3OD): δ 4.25-4.65 (m, 3H), 4.92 (d, 1H, *J* = 4.5), 6.01 (t, 1H, *J* = 53.9), 7.06-7.08 (m, 1H), 7.36-7.42 (m,

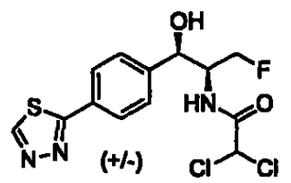
4H), 7.61 (d, 2H, J = 8.4). LRMS (ESI^+) m/z: 352.0 (calc. for $M+Na^+$ $C_{15}H_{14}F_3NNaO_2S$ 352.1).

Example 40: Compound 54



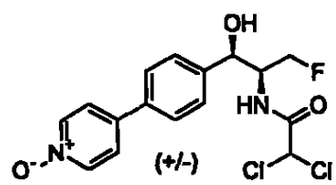
The biaryl intermediate was prepared from **18** and the boronic acid using Suzuki coupling method B. Removal of the Boc group was accomplished by brief treatment with 9/1 (v/v) TFA/H_2O. Dichloroacetylation was performed as in the synthesis of **29**. ^1H NMR (300 MHz, CD_3OD): δ 4.33-4.79 (m, 3H), 5.03 (d, 1H, J = 3.3), 6.26 (s, 1H), 7.55 (d, 2H, J = 8.4), 7.69-7.75 (m, 4H), 8.56 (d, 2H, J = 5.7). LRMS (ESI^+) m/z: 357.1 (calc. for $M+H^+$ $C_{16}H_{16}Cl_2FN_2O_2$ 357.1).

Example 41: Compound 55



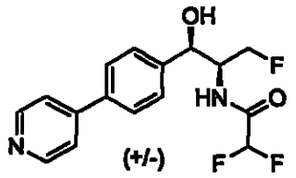
The biaryl intermediate was prepared from boronic acid **23** and compound **24** using Suzuki coupling method B. Removal of the protecting groups was accomplished by brief treatment with 9/1 (v/v) TFA/H_2O. Dichloroacetylation was performed as in the synthesis of **29**. ^1H NMR (300 MHz, CD_3OD): δ 4.34-4.76 (m, 3H), 5.04 (d, 1H, J = 3.0), 6.24 (s, 1H), 7.58 (d, 2H, J = 8.3), 7.97 (d, 2H, J = 8.3), 9.42 (s, 1H). LRMS (ESI^-) m/z: 362.0 (calc. for $M-H^+$ $C_{13}H_{11}Cl_2FN_3O_2S$ 362.0).

Example 42: Compound 56



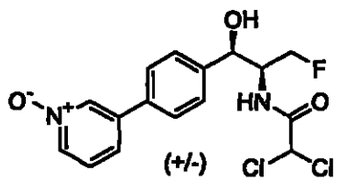
Compound **54** (29.4 mg, 0.0824 mmol) was dissolved in 5 mL of CH_2Cl_2. The mixture was cooled to 0 °C and m-chloroperbenzoic acid (*m*-CPBA, 38 mg, 0.16 mmol) was added with stirring. After 5 minutes, the mixture was warmed to room temperature and stirred for 12 hours. The mixture was concentrated under vacuum and the product purified by silica gel chromatography eluting successively with 2%, 3%, 4%, 6%, and10% MeOH in CH_2Cl_2. Compound **56** was obtained as a white solid (22.5 mg, 0.060 mmol). ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.75 (m, 3H), 5.03 (d, 1H, *J* = 3.3), 6.25 (s, 1H), 7.56 (d, 2H, *J* = 8.4), 7.74 (d, 2H, *J* = 8.4), 7.84 (d, 2H, *J* = 7.2), 8.34 (d, 2H, *J* = 7.2). LRMS (ESI$^-$) m/z: 371.0 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_3$ 371.0).

Example 43: Compound 57



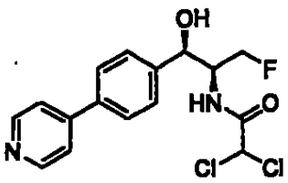
The biaryl intermediate was prepared from **18** and the appropriate boronic acid using Suzuki coupling method B. Removal of the protecting group was accomplished by brief treatment with 9/1 (v/v) TFA/H_2O. Difluoroacetylation was performed as in the synthesis of **48**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.28-4.68 (m, 3H), 5.00 (d, 1H, *J* = 4.2), 5.99 (t, 1H, *J* = 53.9), 7.55 (d, 2H, *J* = 8.1), 7.70-7.77 (m, 4H), 7.84 (d, 2H, *J* = 7.2), 8.56 (d, 2H, *J* = 6.3). LRMS (ESI$^+$) m/z: 325.2 (calc. for M+H$^+$ $C_{16}H_{16}F_3N_2O_2$ 325.1).

Example 44: Compound 58



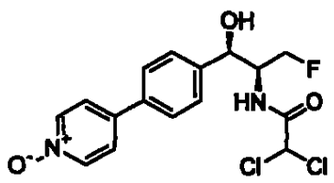
Compound **58** was prepared from **68** as **56** was from **54**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.30-4.75 (m, 3H), 5.02 (d, 1H, *J* = 3.3), 6.25 (s, 1H), 7.55-7.67 (m, 5H), 7.87 (d, 1H, *J* = 8.1), 8.30 (d, 1H, *J* = 7.2), 8.58 (br s, 1H). LRMS (ESI$^-$) m/z: 371.0 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_3$ 371.0).

Example 45: Compound 59



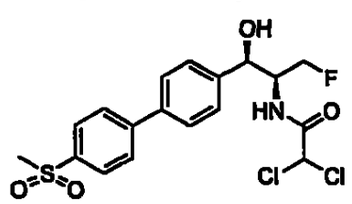
The biaryl intermediate was prepared from **11** and the appropriate boronic acid using Suzuki coupling method A. The phenyloxazoline protecting group was removed and the dichloroacetate group introduced as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.30-4.75 (m, 3H), 5.02 (d, 1H, J = 3.6), 6.26 (s, 1H), 7.54-8.70 (m, 8H). LRMS (ESI$^-$) m/z: 354.8 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_2$ 355.0).

Example 46: Compound 60



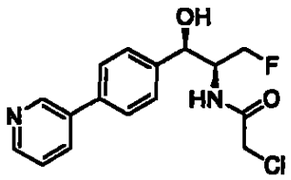
Compound **60** was prepared from compound **59** in the same manner that other pyridine N-oxides in these examples were formed from the corresponding pyridine. ^{1}H NMR (300 MHz, CD_3OD): δ 4.33-4.78 (m, 3H), 5.03 (d, 1H, J = 3.3), 6.25 (s, 1H), 7.57 (d, 2H, J = 8.4), 7.75 (d, 2H, J = 8.4), 7.87 (d, 2H, J = 7.5), 8.38 (d, 2H, J = 7.5). LRMS (ESI$^-$) m/z: 370.8 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_3$ 371.0).

Example 47: Compound 61



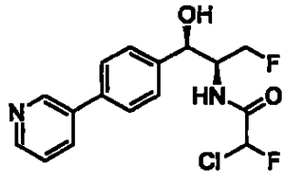
The biaryl intermediate was prepared from **11** and the appropriate boronic acid using Suzuki coupling method A. The phenyl oxazoline protecting group was cleaved and dichloroacetate introduced as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 3.14 (s, 3H), 4.36-4.75 (m, 3H), 5.02 (d, 1H, *J* = 3.6), 6.27 (s, 1H), 7.54 (d, 2H, *J* = 8.3), 7.68 (d, 2H, *J* = 8.3), 7.87 (d, 2H, *J* = 8.7), 8.01 (d, 2H, *J* = 8.7). LRMS (ESI$^-$) m/z: 431.8 (calc. for M-H$^+$ $C_{18}H_{17}Cl_2FNO_4S$ 432.0).

Example 48: Compound 62



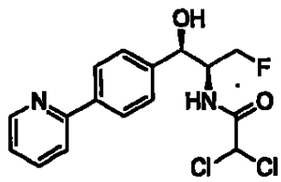
The free amine intermediate was prepared as in the synthesis of **28**. The nitrogen was monochloroacetylated using chloroacetyl chloride. ^{1}H NMR (300 MHz, CD_3OD): δ 4.00-4.03 (m, 2H), 4.27-4.70 (m, 3H), 5.00 (d, 1H, *J* = 3.9), 7.49-7.55 (m, 3H), 7.66 (d, 2H, *J* = 8.4), 8.09 (d app t, 1H, *J* = 7.8, 2.4), 8.50 (dd, 1H, *J* = 4.8, 1.5), 8.78 (br d, 1H, *J* = 2.4). LRMS (ESI$^+$) m/z: 321.0 (calc. for M+H$^+$ $C_{16}H_{15}ClFN_2O_2$ 357.1).

Example 49: Compound 63



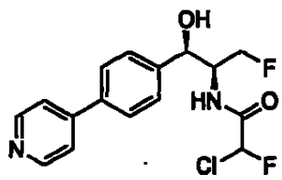
Prepared in the same manner as **28** using ethyl chlorofluoroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 4.31-4.70 (m, 3H), 5.01 (d, 1H, *J* = 3.9), major diastereomer 6.49 (d, 1H, *J* = 49.8), minor diastereomer 6.51 (d, 1H, *J* = 49.8), 7.49-7.56 (m, 3H), 7.63-7.67 (m, 2H), 8.07-8.11 (m, 1H), 8.49-8.51 (m, 1H), 8.78 (br d, 1H, *J* = 1.5). LRMS (ESI) m/z: 338.9 (calc. for M-H$^+$ $C_{16}H_{14}ClF_2N_2O_2$ 339.1).

Example 50: Compound 64



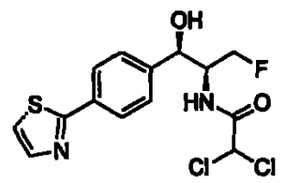
The biaryl intermediate was prepared from **12** and 2-bromopyridine using Suzuki coupling method A. ^{1}H NMR (300 MHz, CD_3OD): δ 4.28-4.74 (m, 3H), 5.02 (d, 1H, *J* = 3.6), 6.27 (s, 1H), 7.33-7.37 (m, 1H), 7.53 (d, 2H, *J* = 8.1), 7.18-7.93 (m, 4H), 8.58-8.60 (m, 1H). LRMS (ESI$^-$) m/z: 354.8 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_2$ 355.0)

Example 51: Compound 65



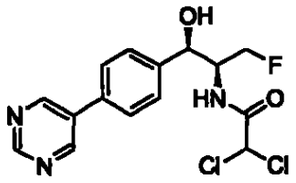
Compound **65** was prepared in the same manner as **59** using ethyl chlorofluoroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.70 (m, 3H), 5.02 (d, 1H, *J* = 3.6), major diastereomer 6.48 (d, 1H, *J* = 49.8), minor diastereomer 6.50 (d, 1H, *J* = 50.1), 7.55-7.58 (m, 2H), 7.73-7.78 (m, 4H) 8.57 (br d, 2H, *J* = 5.7). LRMS (ESI$^-$) m/z: 338.8 (calc. for M-H$^+$ $C_{16}H_{14}ClF_2N_2O_2$ 339.1).

Example 52: Compound 66



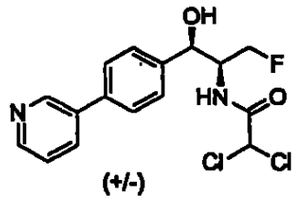
The biaryl intermediate was prepared from **12** and the appropriate bromide using Suzuki coupling method A. Removal of the protecting group and dichloroacetylation was accomplished as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.34-4.72 (m, 3H), 5.01 (d, 1H, *J* = 2.4), 6.25 (s, 1H), 7.52 (d, 2H, *J* = 6.3), 7.57 (d, 1H, *J* = 2.6) 7.84 (d, 1H, *J* = 2.6), 7.90 (d, 2H, *J* = 6.3). LRMS (ESI$^-$) m/z: 360.7 (calc. for M-H$^+$ $C_{14}H_{12}Cl_2FN_2O_2S$ 361.0).

Example 53: Compound 67



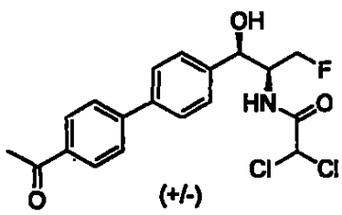
The biaryl intermediate was prepared from **12** and the appropriate bromide using Suzuki coupling method A. Removal of the protecting group and dichloroacetylation was accomplished as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.75 (m, 3H), 5.04 (d, 1H, *J* = 3.6), 6.26 (s, 1H), 7.59 (d, 2H, *J* = 8.3), 7.70 (d, 2H, *J* = 8.3) 9.06 (s, 2H,), 9.13 (s, 1H). LRMS (ESI$^+$) m/z: 357.8 (calc. for M+H$^+$ $C_{15}H_{15}Cl_2FN_3O_2$ 358.0).

Example 54: Compound 68



The biaryl intermediate was prepared from bromide **11** and the appropriate boronic acid using Suzuki coupling method A. Removal of the protecting group and dichloroacetylation was accomplished as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.31-4.74 (m, 3H), 5.02 (d, 1H, *J* = 3.6), 6.27 (s, 1H), 7.48-7.51 (m, 1H), 7.54 (d, 2H, *J* = 8.3), 7.64 (d, 2H, *J* = 8.3) 8.08 (ddd,1H, *J* = 8.0, 2.4, 1.8), 8.50 (dd, 1H, 4.8, 1.5), 8.77 (dd, 1H, *J* = 2.4, 0.9). LRMS (ESI$^-$) m/z: 354.8 (calc. for M-H$^+$ $C_{16}H_{14}Cl_2FN_2O_2$ 355.0).

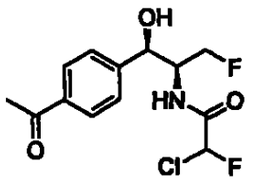
Example 55: Compound 69



The biaryl intermediate was prepared from bromide **11** and the appropriate boronic acid using Suzuki coupling method A. Removal of the protecting group and

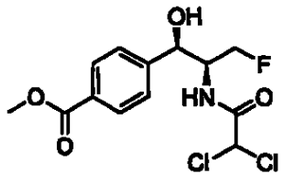
dichloroacetylation was performed as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 2.63 (s, 3H), 4.31-4.74 (m, 3H), 5.01 (d, 1H, *J* = 3.9), 6.27 (s, 1H), 7.51 (d, 2H, *J* = 8.3), 7.67 (d, 2H, *J* = 8.3), 7.75 (d, 2H, *J* = 8.6) 8.06 (d, 2H, *J* = 8.6).

Example 56: Compound 71



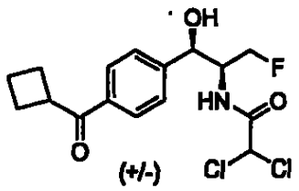
This compound was prepared in analogous fashion to **41** using ethyl chlorofluoroacetate in place of methyl dichloroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 2.58 (s, 3H), 4.31-4.71 (m, 3H), 5.03 (br d, 1H, *J* = 3.9), major diastereomer 6.46 (d, 1H, *J* = 49.8), minor diastereomer (d, 1H, *J* = 50.1), major diastereomer 7.53 (d, 2H, *J* = 8.1), minor diastereomer 7.51 (d, 2H, *J* = 8.1), 7.94-7.96 (m, 2H). LRMS (ESI$^-$) m/z: 304.1 (calc. for M-H$^+$ $C_{13}H_{13}ClF_2NO_3$ 304.1).

Example 57: Compound 72



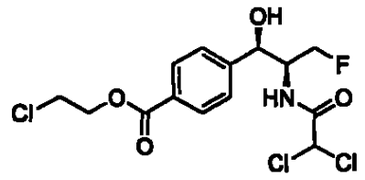
Initially, the *p*-cyano analog of compound **11** had been prepared by methods analogous to those used to prepare **11** itself. Attempts to remove the phenyloxazoline protecting group (as described in the synthesis of **29**) led to some of the desired deprotected nitrile intermediate. However, much of the mass recovered was the deprotected *p*-carboxylic acid corresponding to acidic hydrolysis of the nitrile functional group. This material was dichloroacetylated using the same procedure as that employed in the synthesis of **29** and the product was converted to its methyl ester with CH_2N_2. ^{1}H NMR (300 MHz, CD_3OD): δ 3.83 (s, 3H), 4.25-4.66 (m, 3H), 4.98 (d, 1H, *J* = 3), 6.17 (s, 1H), 7.46 (d, 2H, *J* = 8.3), 7.91 (d, 2H, *J* = 8.3).

Example 58: Compound 73



Compound **87** (36.5 mg, 0.0873 mmol) was dissolved in THF (5 mL anhydrous) under N_2. After addition of anhydrous powdered K_2CO_3 (24 mg, 0.175 mmol), the mixture was purged with N_2. Et_3N (31 mL, 0.218 mmol) and cyclobutanecarbonyl chloride (13 mL, 0.113 mmol) were added and the mixture purged gently with N_2 for another 5 min. Pd_2dba_3 was added and the mixture purged again with N_2. The mixture was then stirred under N_2 for 3 hours after which it was diluted with EtOAc and H_2O and filtered through a cotton plug. The EtOAc layer was washed with 1N aqueous HCl (x 2), brine (x 2), and dried over anhydrous Na_2SO_4. The mixture was filtered and the solvent removed to give 16.9 mg, 0.0501 mmol of material that was purified by chromatotron (1 mm plate, eluting with 4:1 hexanes/EtOAc). The protected intermediate obtained was subjected to phenyloxazoline cleavage and dichloroacetylation as in the synthesis of **29** to give **73**. ^{1}H NMR (300 MHz, CD_3OD): δ 1.73-1.85 (m, 1H), 1.99-2.11 (m, 1H), 2.20-2.27 (m, 4H), 3.97-4.09 (m, 1H), 4.23-4.67 (m, 3H), 4.95 (d, 1H, J = 3.3), 6.16 (s, 1H), 7.45 (d, 2H, J = 8.4), 7.81 (d, 2H, J = 8.4). LRMS (ESI$^-$) m/z: 360.0 (calc. for M-H$^+$ $C_{18}H_{17}Cl_2FNO_3$ 360.0).

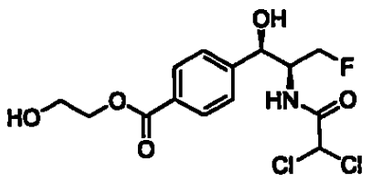
Example 59: Compound 74



The carboxylic acid intermediate from the synthesis of **72** was dichloroacetylated as in the synthesis of **29**. The resulting intermediate (95.8 mg, 0.297 mmol) was dissolved in 2 mL of MeOH, several drops of H_2O were added and then a 20% (w/v) aqueous Cs_2CO_3 solution was added dropwise until the solution reached pH 7. The mixture was concentrated under vacuum. One drop of Et_3N and 10 mL of 2-chloroethanol were added and the mixture stirred at 135 °C for 2 hours. Residual chloroethanol was removed under vacuum and the residue

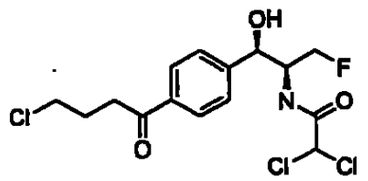
chromatographed. Compound 74 (13.3 mg) was the major product and 75 (9.2 mg) the minor product. ^{1}H NMR (300 MHz, CD_3OD): δ 3.78-3.82 (m, 2H), 4.23-4.68 (m, 5H), 4.97 (d, 1H, J = 3.0), 6.17 (s, 1H), 7.46 (d, 2H, J = 8.3), 7.93 (d, 2H, J = 8.3). LRMS (ESI$^+$) m/z: 407.9 (calc. for M+Na$^+$ $C_{14}H_{15}Cl_3FNNaO_4$ 408.0).

Example 60: Compound 75



^{1}H NMR (300 MHz, CD_3OD): δ 3.83-3.39 (m, 2H), 4.31-4.74 (m, 5H), 5.02 (d, 1H, J = 3.3), 6.23 (s, 1H), 7.52 (d, 2H, J = 8.4), 8.02 (d, 2H, J = 8.4). LRMS (ESI$^-$) m/z: 366.0 (calc. for M-H$^+$ $C_{14}H_{15}Cl_2FNO_5$ 366.0).

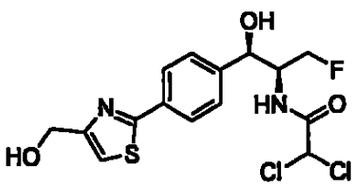
Example 61: Compound 76



(+/-)

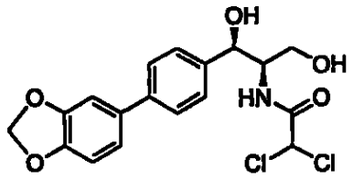
The cyclopropyl analog of intermediate 89 was prepared by a Stille coupling analogous to that used to synthesize 89. Acidic cleavage of the phenyloxazoline group resulted in the HCl-mediated opening of the cyclopropyl ring. This material was deprotected and dichloroacetylated to give 76. ^{1}H NMR (300 MHz, CD_3OD): δ 2.10-2.19 (m, 2H), 3.19 (t, 2H, J = 7.1), 3.66 (t, 2H, J = 6.6), 4.31-4.74 (m, 3H), 5.03 (d, 1H, J = 3.3), 6.23 (s, 1H), 7.53 (d, 2H, J = 8.3), 7.96 (d, J = 8.3). LRMS (ESI$^-$) m/z: 381.9 (calc. for M-H$^+$ $C_{15}H_{16}Cl_3FNO_3$ 382.0).

Example 62: Compound 77



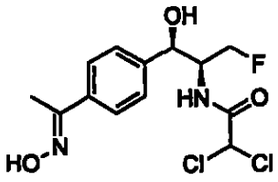
The thiazole ring of this analog was formed by conversion of the protected nitrile intermediate to the corresponding thioamide by reaction with H_2S in pyridine. The thioamide was then reacted with the appropriate protected α-halocarbonyl compound to form the substituted thiazole shown. Deprotection and dichloroacetylation was performed as for other compounds of this invention. ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.75 (m, 5H), 5.01 (d, 1H, *J* = 3.3), 6.27 (s, 1H), 7.38 (s, 1H), 7.51 (d, 2H, *J* = 8.3), 7.91 (d, 2H, *J* = 8.3). LRMS (ESI$^-$) m/z: 390.9 (calc. for M-H$^+$ $C_{15}H_{14}Cl_2FN_2O_3S$ 391.0).

Example 63: Compound 79



This compound was formed by Suzuki coupling method A from an intermediate in which the primary hydroxyl group was not converted to a fluoride. Deprotection and dichloroacetylation were performed as with other compounds of this invention. ^{1}H NMR (300 MHz, CD_3OD): δ 3.55 (dd, 1H, *J* = 11.0, 5.9), 3.77 (dd, 1H, *J* = 11.0, 6.5), 4.07-4.11 (m, 1H), 5.02 (d, 1H, *J* = 3.6), 5.96 (s, 2H), 6.29 (s, 1H), 6.86 (d, 1H, *J* = 8.4), 7.05-7.07 (m, 2H), 7.40-7.50 (m, 4H). LRMS (ESI$^-$) m/z: 395.9 (calc. for M-H$^+$ $C_{18}H_{16}Cl_2NO_5$ 396.0).

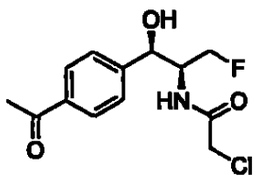
Example 64: Compound 80



Compound 41 (11.5 mg, 0.0346 mmol) and hydroxylamine hydrochloride (2.8 mg, 0.0415 mmol) were stirred in EtOH (3 mL) for 12 hours and then refluxed for an

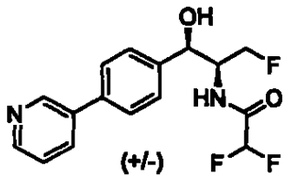
additional 12 hours. The solvent was removed under vacuum and the product isolated as a single isomer (it was not determined whether the isomer was the *cis* or *trans* oxime). ^{1}H NMR (300 MHz, CD_3OD): δ 2.21 (s, 3H), 4.26-4.68 (m, 3H,), 4.95 (d, 1H, *J* = 3.9), 6.27 (s, 1H), 7.39 (d, 2H, *J* = 8.3), 7.61 (d, 2H, *J* = 8.3).

Example 65: Compound 81



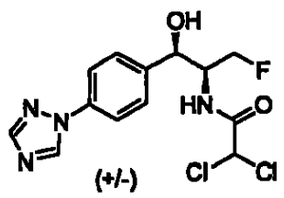
Free amine 37 (4.7 mg, 0.0224 mmol) was dissolved in MeOH (1 mL) and the mixture cooled to 0 °C. Chloroacetic anhydride (5 drops) and triethylamine (5 drops) were added and the mixture warmed to room temperature and stirred for 2 hours. The solvent was removed under vacuum and the product purified by silica gel chromatography (2:3 EtOAc/hexanes) to give 3.3 mg (0.011 mmol) of product. ^{1}H NMR (300 MHz, CD_3OD): δ 2.59 (s, 3H), 3.92-4.03 (m, 2H,), 4.29-4.68 (m, 3H), 5.02 (d, 1H, *J* = 3.3), 7.53 (d, 2H, *J* = 8.4), 7.96 (d, 2H, *J* = 8.4).

Example 66: Compound 82



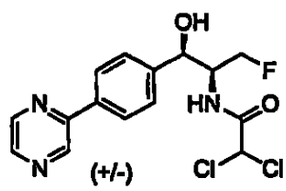
The biaryl intermediate was formed by Suzuki coupling method A using 22 and the appropriate boronic acid. ^{1}H NMR (300 MHz, CD_3OD): δ 4.28-4.68 (m, 3H), 5.00 (d, 1H, *J* = 4.2), 6.01 (t, 1H, *J* = 54), 7.49-7.60 (m, 3H), 7.66 (d, 2H, *J* = 8.4), 8.07-8.11 (m, 1H), 8.50 (dd, 1H, *J* = 4.8, 1.5), 8.79-8.79 (m, 1H). LRMS (ESI$^-$) m/z: 322.9 ($M\text{-}H^+$ $C_{18}H_{14}F_3N_2O_2$ requires 323.1).

Example 67: Compound 84



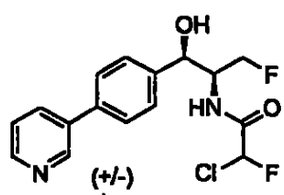
This compound was prepared by procedures analogous to those described in Schemes 2 and 3. The 4-(1,2,4-triazol-1-yl)phenylserine analog of **6** was synthesized by the same methods used to make **6**. The required 4-(1,2,4-triazol-1-yl)benzaldehyde was prepared as described in the literature (Tanaka, A., et al., *J. Med. Chem.* **1998**, *41*, 2390-2410). ^{1}H NMR (300 MHz, CD_3OD): δ 4.33-4.76 (m, 3H), 5.04 (d, 1H, *J* = 3.3), 6.26 (s, 1H), 7.59 (d, 2H, *J* = 8.6), 7.78 (d, 2H, *J* = 8.6), 8.14 (s, 1H), 9.05 (s, 1H). LRMS (ESI$^-$) m/z: 345.0 (calc. for M-H$^+$ $C_{13}H_{12}Cl_2FN_4O_2$ 345.0).

Example 68: Compound 85



The biaryl intermediate was prepared from boronic acid **12** and 2-iodopyrazine. Deprotection and dichloroacetylation were accomplished as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.32-4.75 (m, 3H), 5.04 (d, 1H, *J* = 3.3), 6.27 (s, 1H), 7.57 (d, 2H, *J* = 8.4), 8.05 (d, 2H, *J* = 8.4), 8.51 (d, 1H, *J* = 2.7), 8.65-8.66 (m, 1H), 9.08 (d, 1H, *J* = 1.8). LRMS (ESI$^-$) m/z: 355.8 (calc. for M-H$^+$ $C_{15}H_{13}Cl_2FN_3O_2$ 356.0).

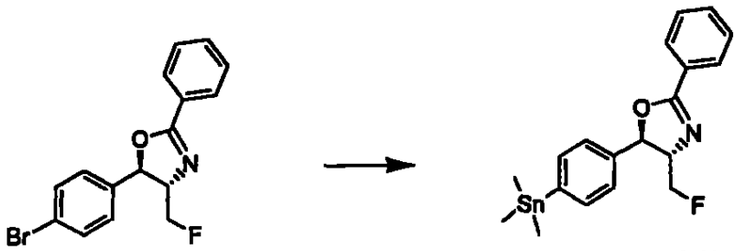
Example 69: Compound 86



Same procedure as that for the synthesis of **68**. ^{1}H NMR (300 MHz, CD_3OD): δ 4.29-4.73 (m, 3H), 5.01 (br d, 1H, *J* = 3.9), major diastereomer 6.49 (d, 1H, *J* =

49.8), minor diastereomer 6.51 (d, 1H, J = 49.8), 7.48-7.56 (m, 3H), 7.64-7.67 (m, 2H), 8.06-8.10 (m, 1H), 8.49-8.51 (m, 1H), 8.78-8.79 (m, 1H). LRMS (ESI$^-$) m/z: 338.9 (calc. for M-H$^+$ $C_{16}H_{14}ClF_2N_2O_2$ 339.1).

Example 70: Compound 87

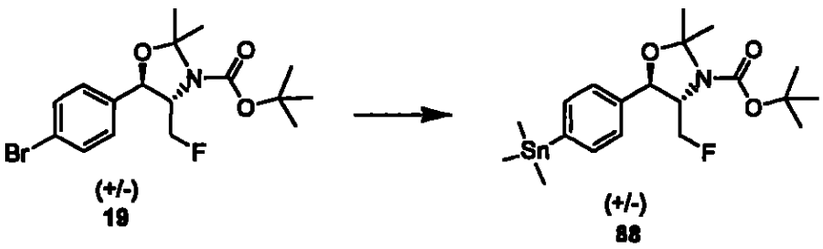


enantiopure or (+/-)
11

enantiopure or (+/-)
87

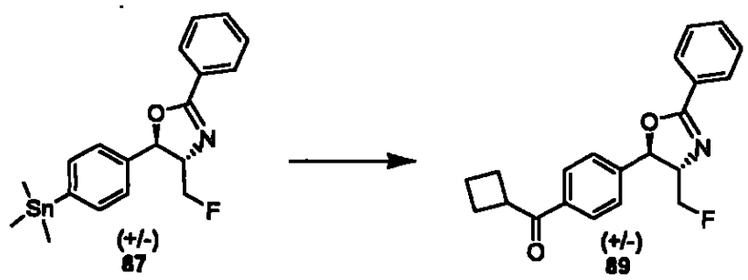
Compound 11 (0.496 g, 1.48 mmol) was dissolved in anhydrous benzene (15 mL) under N_2. The flask was fitted with a reflux condenser and a long needle was dropped through the condenser and the solution purged with a gentle stream of dry N_2 for 5 minutes. Hexamethylditin (0.58 g, 1.78 mmol) was added and the mixture purged for another 5 minutes. $Pd(PPh_3)_4$ (0.171 g, 0.148 mmol) was added and purging with N_2 continued. After 5 minutes, the needle was removed and a N_2 inlet was placed at the top of the reflux condenser. The mixture was refluxed for approximately 2 hours under N_2. Over the course of the reaction, the mixture turned from orange to yellow to black. The solvent was removed under vacuum and the product purified by chromatotron (silica gel, 1 mm plate) to give 440 mg of product as a clear oil. ^{1}H NMR (300 MHz, CD_3OD): δ 0.27 (s, 9H - this resonance contains significant satellite peaks, which are due to NMR-active isotopes of tin), 4.27-4.41 (m, 1H), 4.59-4.76 (m, 2H), 5.60 (d, 1H, J = 6.9), 7.33 (d, 2H, J = 8.1), 7.45-7.31 (m, 5H), 7.99-8.02 (m, 2H).

Example 71: Compound 88



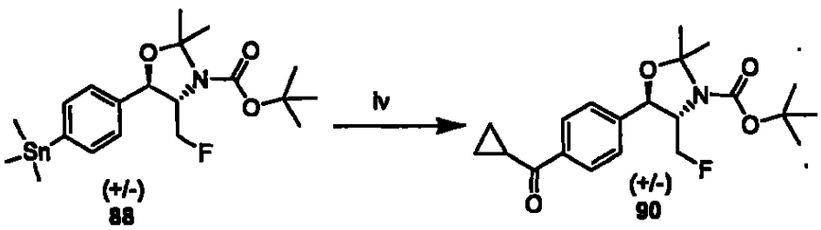
This compound was prepared in the same manner as 87. ^{1}H NMR (300 MHz, CD_3OD): δ 0.027 (s, 9H, with significant satellite peaks caused by NMR-active tin isotopes), 1.49 (s, 9H), 1.55 (s, 3H), 1.67 (s, 3H), 3.73-3.83 (m, 1H), 4.30-4.49 (m, 2H), 5.06 (d, 2H, J = 7.5), 7.36-7.51 (m, 4H).

Example 72: Compound 89



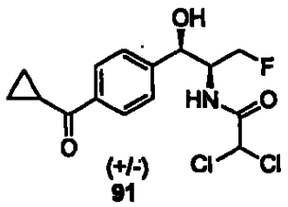
Compound 87 (36.5 mg, 0.0873 mmol) was dissolved in anhydrous THF under N_2. With stirring, K_2CO_3 (24mg, 0.16 mmol) was added. The mixture was purged with a gentle stream of N_2 for 5 minutes. Et_3N (31 μL, 0.22 mmol) and cyclobutanecarbonyl chloride (13 μL, 0.11 mmol) were added and the mixture purged with N_2 for 5 minutes. Pd_2dba_3 was added, the mixture purged for 5 min and then stirred under N_2 for 3 hours. The mixture was diluted with EtOAc (25 mL) and H_2O (25 mL) and filtered through a cotton plug to remove solids. The EtOAc was washed with 1 N HCl (2 x) and brine (2 x), dried over Na_2SO_4 and concentrated under vacuum. The product was purified by chromatotron (silica plate, 1 mm, 1:4 EtOAc/hexanes) to give 16.9 mg (0.050 mmol) of product. LRMS (ESI^+) m/z: 338.2 ($M+H^+$ $C_{21}H_{21}FNO_2$ requires 338.2).

Example 73: Compound 90



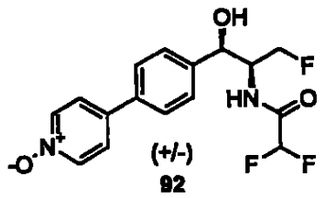
Same procedure as in the synthesis of **89**. ^{1}H NMR (300 MHz, CD_3OD): δ 0.027 (s, 9H, with significant satellite peaks caused by NMR-active tin isotopes), 1.49 (s, 9H), 1.55 (s, 3H), 1.67 (s, 3H), 3.73-3.83 (m, 1H), 4.30-4.49 (m, 2H), 5.06 (d, 2H, *J* = 7.5), 7.36-7.51 (m, 4H).

Example 74: Compound 91



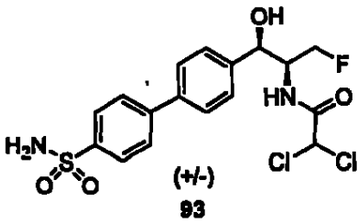
Compound **91** was deprotected by brief treatment of **90** with 9/1 TFA/H_2O. The dichloroacetyl group was introduced as in the synthesis of **29**. LRMS (ESI$^-$) m/z: 345.8 (calc. for M-H$^+$ $C_{15}H_{16}Cl_2FNO_3$ 346.0).

Example 75: Compound 92



The same procedure as that used in the synthesis of **56** was employed. ^{1}H NMR (300 MHz, CD_3OD): δ 4.28-4.74 (m, 3H), 5.01 (d, 1H, *J* = 3.9), 5.98 (t, 1H, *J* = 53.8), 7.55 (d, 2H, *J* = 8.4). 7.76 (d, 2H, *J* = 8.4), 7.86 (d, 2H, *J* = 7.1), 8.35 (d, 2H, *J* = 7.1).

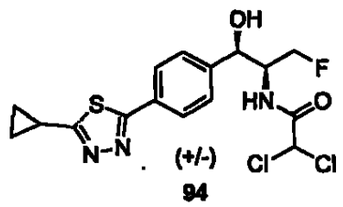
Example 76: Compound 93



93

The biaryl intermediate was prepared by reaction of **23** and the appropriate bromide using Suzuki coupling A. Deprotection and dichloroacetylation were carried out as described above in the synthesis of **29**. LRMS (ESI$^-$) m/z: 433.0 (M-H$^+$ $C_{17}H_{16}Cl_2FN_2O_4S$ requires 433.0).

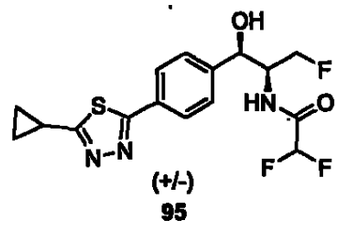
Example 77: Compound 94



94

The biaryl intermediate was prepared by reaction of **23** with 2-amino-4-cyclopropyl-1,3,4-thiadiazole (prepared as in the synthesis of **24**) using Suzuki coupling B. Deprotection was accomplished by brief treatment of the protected biaryl intermediate with 90/10 TFA/H_2O and dichloroacetylation was performed as in the synthesis of **29**. ^{1}H NMR (300 MHz, CD_3OD): δ 1.12-1.31 (m, 4H), 2.46-2.52 (m, 1H), 4.30-4.75 (m, 3H), 5.02 (d, 1H, *J* = 3.3), 6.24 (s, 1H), 7.54 (d, 2H, *J* = 8.4), 7.87 (d, 2H, *J* = 8.4).

Example 78: Compound 95

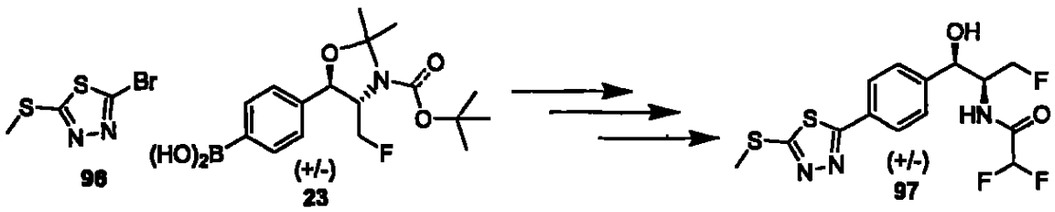


95

Same procedure as in the synthesis of **94** using methyl difluoroacetate in place of methyl dichloroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 1.13-1.15 (m, 2H), 1.28-1.32 (m, 2H), 2.47-2.52 (m, 1H), 4.29-4.70 (m, 3H), 5.01 (d, 1H, *J* = 4.2); 5.98

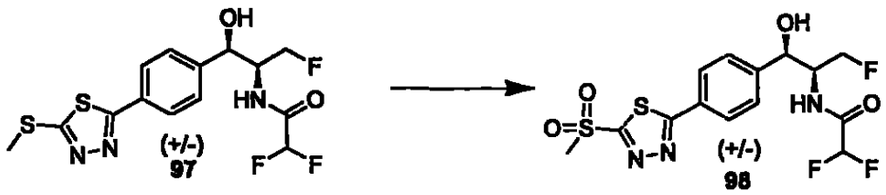
(t, 1H, J = 53.7), 7.54 (d, 2H, J = 8.7), 7.88 (d, 2H, J = 8.7). LRMS (ESI$^-$) m/z: 370.0 (M-H$^+$ $C_{16}H_{15}F_3N_3O_2S$ requires 370.1).

Example 79: Compound 97



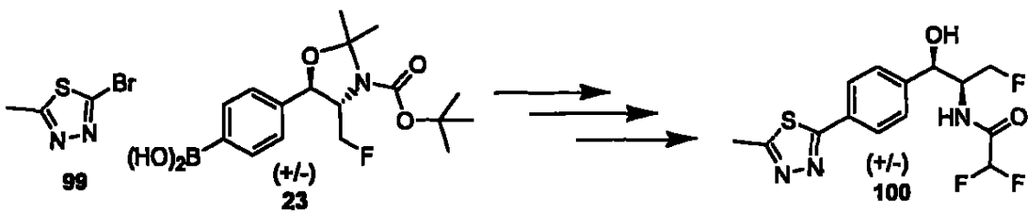
Compound **96** was prepared in analogous fashion to **24**. Compound **97** was prepared in analogous fashion to **48**. Suzuki coupling method B was used, followed by deprotection and difluoroacetylation. LRMS (ESI$^-$) m/z: 376.0 (M-H$^+$ $C_{14}H_{13}F_3N_3O_2S_2$ requires 376.0)

Example 80: Compound 98



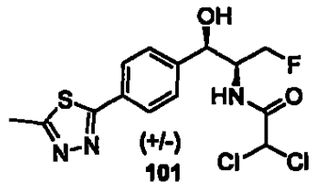
Compound **97** (11.1 mg or 0.0294 mmol) was dissolved in 5 mL of CH_2Cl_2, with stirring at room temperature. To this was added *m*-CPBA (36 mg, 0.147 mmol). The mixture was stirred for 23 hours. Attempted Chromatotron (1 mm plate) purification, eluting with 80:20 then 70:30 then 50:50 hexanes/EtOAc, failed to remove *m*-CPBA-related materials. Thus, material from the Chromotron was subjected to C-8 reversed-phase HPLC, which gave 8.8 mg (.021 mmol) of **98**. ^{1}H NMR (300 MHz, CD_3OD): δ 3.52 (s, 3H), 4.34-4.72 (m, 3H), 5.05 (d, 1H, J = 3.6), 5.97 (t, 1H, J = 53.9), 7.61 (d, 2H, J = 7.61), 8.05 (d, 2H, J = 8.1).

Example 81: Compound 100



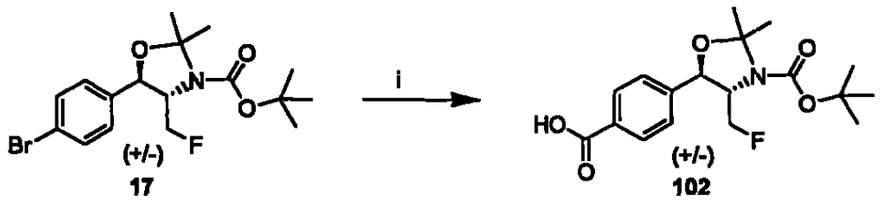
Compound **99** was prepared in analogous fashion to **24** and **100** was prepared in analagous fashion to **48**. Suzuki coupling method B was used followed by deprotection and difluoroacetylation. ^{1}H NMR (300 MHz, CD_3OD): δ 2.80 (s, 3H), 4.30-4.70 (m, 3H), 5.01 (d, 1H, *J* = 3.6), 5.98 (t, 1H, J = 53.9), 7.55 (d, 2H, *J* = 8.4), 7.91 (d, 2H, *J* = 8.3).

Example 82: Compound 101



Compound 101 was prepared using the same procedure as in the synthesis of **100** but with methyldichloroacetate in place of methyldifluoroacetate. ^{1}H NMR (300 MHz, CD_3OD): δ 2.79 (s, 3H), 4.31-4.75 (m, 3H), 5.03 (d, 1H, *J* = 3.0), 6.24 (s, 1H), 7.55 (d, 2H, *J* = 8.4), 7.89 (d, 2H, *J* = 8.4).

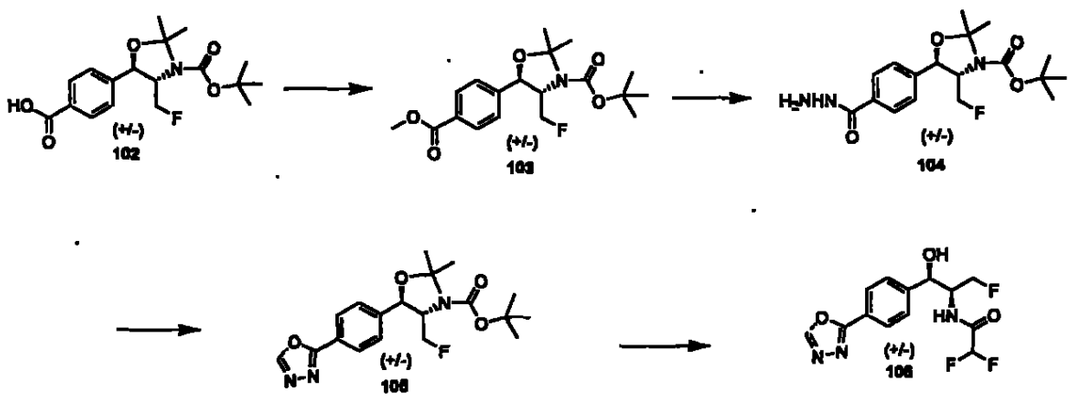
Example 83: Compound 102



Compound **17** (100 mg, 0.258 mmol) was placed in a dry round-bottom flask equipped with a stir bar. The flask was charged with 5 mL of dry THF and the contents cooled to –78 °C under N_2. With rapid stirring, *n*-BuLi (1.30 M in hexanes, 0.322 mmol, 0.248 mL) was added, which produced a clear brown/yellow solution. The mixture was stirred for an additional 10 minutes. Excess CO_2, generated by sublimation of dry ice, was passed though a drying tube charged with Drierite and

bubbled directly into the –78 °C mixture, now equipped with a venting needle in the septum to prevent buildup of CO_2 pressure. The mixture was warmed to room temperature and stirred an additional 30 minutes by which time it was clear yellow and TLC indicated a product had formed. The mixture was quenched by addition of 10% (v/w) aqueous NH_4Cl (5 drops), producing a cloudy yellow suspension. The mixture was concentrated under vacuum and re-suspended in EtOAc (50 mL) and enough 10% aqueous citric acid to produce a biphasic mixture having a pH of 2.5. The layers were separated and the EtOAc layer was washed with brine (1 x 10 mL). The mixture was dried over Na_2SO_4, filtered and evaporated to give 105 mg of yellow oil. The product was isolated by chromatotron (1 mm plate eluting with 4:1 hexanes/EtOAc to 65:35 hexanes/EtOAc to 1:1 hexanes/EtOAc to 2% MeOH in CH_2Cl_2 to 5% MeOH in CH_2Cl_2 and, finally, 10% MeOH in CH_2Cl_2 to which several drops of acetic acid had been added). The appropriate fractions were combined, diluted with toluene and evaporated to give 67.5 mg (0.191 mmol) of **102**. ^1H NMR (300 MHz, CD_3OD): δ 1.49 (s, 9H), 1.57 (S, 3H), 1.69 (s, 3H), 3.77-3.90 (m, 1H), 4.39-4.57 (m, 1H), 4.90-5.10 (br s, 1H), 5.18 (d, 1H, J = 7.2), 7.56 (d, 2H, J = 8.1), 8.04 (d, 2H, J = 8.1). LRMS (ESI$^-$) m/z: 352 (M-H$^+$ $C_{18}H_{23}FNO_5$ requires 352).

Example 84: Compound 106



Compound **102** (106 mg, 0.299 mmol) was dissolved in EtOAc and diazomethane, generated using an Aldrich Chemical Company diazomethane kit, was added dropwise until a slight yellow color persisted. Excess of diazomethane was allowed to evaporate overnight in a well-ventilated fume hood, and the remaining EtOAc solution was concentrated under vacuum. The residue was

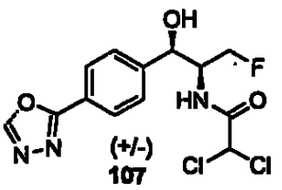
dissolved in Et_2O and washed with aqueous $NaHCO_3$ and then brine. The layers were separated and the organic layer was dried over anhydrous Na_2SO_4, filtered and concentrated to give 0.087 g (0.24 mmol, 79%) of methyl ester **103**, which was used without further purification or characterization.

Compound **103** (0.037g, 0.136 mmol) was dissolved in 1 mL of EtOH. To this was added hydrazine hydrate (.007 g, 0.177 mmol). The mixture was refluxed for 12 hours. The solvent was removed under vacuum and the product purified by chromatotron (1 mm plate, 10% MeOH in CH_2Cl_2). Compound **104** was isolated (40.0 mg, 0.109 mmol) and was used without further characterization. LRMS (ESI^-) m/z: 366 ($M\text{-}H^+$ $C_{18}H_{25}FN_3O_4$ requires 366).

Compound **104** (0.050 g, 0.133 mmol) was dissolved in 5 mL of triethyl orthoformate and stirred at 120 °C for 24 hours. The mixture was evaporated to give 26 mg of crude **105**, which was used without further characterization or purification.

Compound **105** (0.0051 g, 0.014 mmol) was stirred at room temperature with 10 mL of 9:1 TFA/H_2O for 10 minutes. The mixture was concentrated under vacuum, dissolved three times in a MeOH/toluene mixture, the solvents being evaporated each time, and dried to a constant weight to give 0.5 mg of deprotected material. The residue was dissolved in 1 mL of MeOH in an open container and 5 drops of methyl difluoroacetate and 15 drops of Et_3N were added. The mixture was stirred at room temperature for 12 hours. After 12 hours, TLC indicated a single product. The mixture was concentrated under vacuum and the product isolated by silica gel chromatography eluting with 20:1 CH_2Cl_2/CH_3OH to give 2.3 mg of **106**. ^1H NMR (300 MHz, CD_3OD): δ 4.31-4.74 (m, 3H), 5.04 (d, 1H, J = 3.9), 5.96 (t, 1H, J = 53.9), 7.61 (d, 2H, J = 8.4), 8.05 (d, 2H, J = 8.4), 8.99 (d, 1H). LRMS (ESI^-) m/z: 314 ($M\text{-}H^+$ $C_{13}H_{11}F_3N_3O_3$ requires 314).

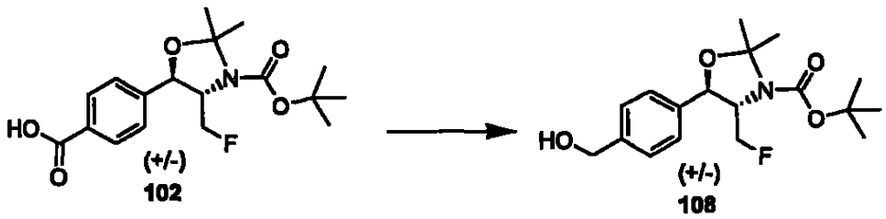
Example 85: Compound 107



Compound **107** was prepared in analogous manner to **106** using methyl dichloroacetate in place methyl difluoroacetate. ^1H NMR (300 MHz, CD_3OD): δ 4.32-

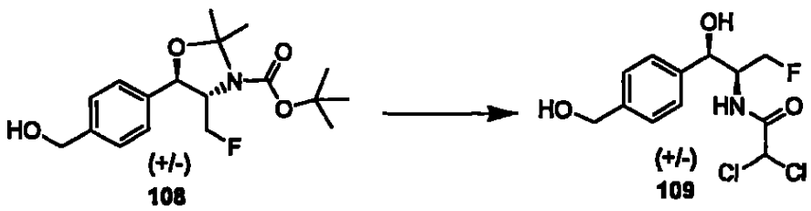
4.80 (m, 3H), 5.06 (d, 1H, J = 3.0), 6.23 (S, 1H), 7.62 (d, 2H, J = 8.4), 8.03 (d, 2H, J = 8,4), 8.98 (s. 1H). LRMS (ESI$^-$) m/z: 346 (M-H$^+$ $C_{13}H_{11}Cl_2FN_3O_3$ requires 346).

Example 86:Compound 108



Compound **102** (32.7 mg, 0.0925 mmol) was dissolved in EtOAc (8 mL) and cooled with stirring to 0 °C. Pentafluorophenol (17 mg, 0.093 mmol) was added. Once all solids had dissolved, DCC (19 mg, 0.093 mmol) was added and the mixture stirred at 0 °C for 1.25 hours. The mixture was evaporated to one-quarter the original volume at which time a DCU precipitate formed. The DCU was removed by filtration and the pentafluorophenyl ester was isolated by evaporation. The residue was dissolved in 4 mL of MeOH and cooled to 0 °C with stirring. $NaBH_4$ (18 mg, 0.46 mmol) was added portionwise. When bubbling ceased, the mixture was warmed to room temperature. After 2 hour, excess $NaBH_4$ was quenched by addition of 4 drops of glacial HOAc. The mixture was evaporated to dryness and the residue partitioned between 1N aq. HCl and EtOAc. The EtOAc was separated and washed with 1 N aqueous HCl (2 x 15 mL), saturated aqueous $NaHCO_3$ (2 x 10 mL) and finally brine (1 x 25 mL). The EtOAc layer was dried over anhydrous Na_2SO_4, filtered, and evaporated to give 50 mg of yellow oil. Chromatotron purification (1 mm plate, eluting with 5% EtOAc in hexanes to 10% EtOAc in hexanes) gave **108** (10.3 mg, 0.030 mmol). ^{1}H NMR (300 MHz, CD_3OD): δ 1.49 (s, 9H), 1.57 (s, 3H), 1.68 (s. 3H), 3.75-3.89 (m, 1H), 3.90 (s, 2H), 4.39-4.58 (m, 1H), 4.90-5.05 (br s, 1H), 5.19 (d, 1H, J = 7.2), 7.57 (d, 2H, J = 8.3), 8.04 (d, 2H, J = 8.3).

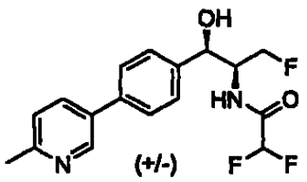
Example 87: Compound 109



Compound **108** (10 mg, 0.30 mmol) was dissolved in 5 mL of 9:1 TFA/H_2O and stirred at room temperature for 1 hour. The mixture was then concentrated under vacuum to give approximately 9 mg of deprotected material as the TFA salt. ^{1}H NMR (300 MHz, CD_3OD): δ 3.54-3.68 (m, 1H), 3.91 (s, 2H), 4.24-4.67 (m, 2H), 4.90 (d, 1H, solvent obscured), 7.57 (d, 2H, *J* = 8.4), 8.07 (d, 2H, *J* = 8.4).

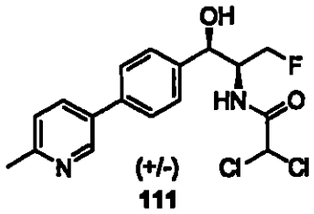
The 9 mg of material (0.03 mmol) were dissolved in 2 mL of MeOH in an open container. To this was added 15 drops of Et_3N and 15 drops of methyl dichloroacetate. The mixture was stirred for 56 hour at room temperature. By the end of the reaction the solvent had completely evaporated. The residue was loaded onto a chromatotron (1 mm plate, eluting with 2% MeOH in CH_2Cl_2 to 4% MeOH in CH_2Cl_2, providing 7.5 mg of **109**. ^{1}H NMR (300 MHz, CD_3OD): δ 3.88 (s, 2H), 4.28-4.73 (m, 3H), 5.02 (d, 1H, *J* = 3.3), 6.22 (s, 1H), 7.51 (d, 2H, *J* = 8.4), 7.97 (d, 2H, *J* = 8.4).

Example 88: Compound 110



Prepared in analogous fashion to other pyridine-containing biaryl compounds herein. ^{1}H NMR (300 MHz, CD_3OD): δ 2.54 (s, 3H), 4.30-4.68 (m, 3H), 4.99 (d, *J* = 3.3), 5.98 (t, 1H, *J* = 40.5), 7.35 (d, 1H, *J* = 6.0), 7.49 (d, 2H, *J* = 6.2), 7.61 (d, 2H, *J* = 6.2), 7.95 (dd 1H, *J* = 6.0, 1.7), 8.61 (d, 1H, *J* = 1.7). LRMS (ESI$^-$) m/z: 336.9 (M-H$^+$ $C_{17}H_{16}F_3N_2O_2$ requires 337.1).

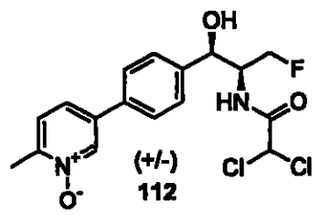
Example 89: Compound 111



111

Prepared in analogous fashion to other pyridine-containing biaryl compounds herein. ^{1}H NMR (300 MHz, CD_3OD): δ 2.56 (s, 3H), 4.30-4.74 (m, 3H), 5.01 (d, 1H, *J* = 3.9), 6.27 (s, 1H), 7.36 (d, 1H, *J* = 8.1), 7.56 (d, 2H, *J* = 8.4), 7.61 (d, 2H, *J* = 8.4), 7.96 (1H, dd, *J* = 8.1, 2.3), 8.62 (d, 1H, *J* = 2.31).

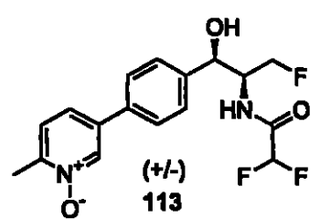
Example 90: Compound 112



112

Compound **111** (0.0185g, 0.0499 mmol) was dissolved in 10 mL CH_2Cl_2 at 0 °C. To this was added *m*-CPBA (0.246 g, 0.0997 mmol). The mixture was stirred until the ice bath melted and the mixture came to room temperature, approximately 12 hours. The mixture was concentrated under vacuum and **112** was isolated by preparative plate silica gel chromatography (15% MeOH in CH_2Cl_2) followed by a silica gel plug (3% MeOH in CH_2Cl_2). LRMS (ESI$^-$) m/z: 385 ($M-H^+$ $C_{17}H_{16}Cl_2FN_2O_3$ requires 385.1).

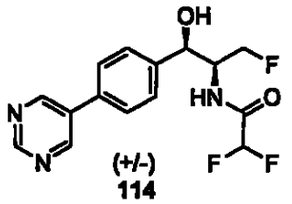
Example 91: Compound 113



113

Compound **113** was synthesized from compound **110** in the same manner **112** was synthesized from **111**. LRMS (ESI$^-$) m/z: 353 (M-H$^+$ $C_{17}H_{16}Cl_2FN_2O_3$ requires 353.1).

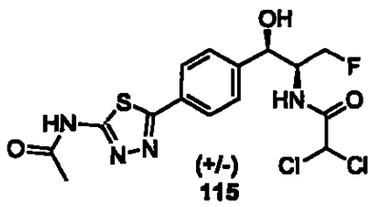
Example 92: Compound 114



114

This compound was prepared in the same manner as other biaryls herein. LRMS (ESI$^-$) m/z: 324 (M-H$^+$ $C_{15}H_{13}F_3N_3O_2$ requires 324.1).

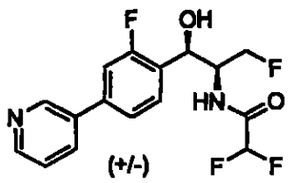
Example 93: Compound 115



115

Prepared in same manner as compound **48**. The required aryl bromide was prepared in two steps from 2-amino-1,3,4-thiadiazole by bromination with Br_2 followed by acetylation. LRMS (ESI$^-$) m/z: 419 (M-H$^+$ $C_{15}H_{14}Cl_2FN_4O_3S$ requires 419.0).

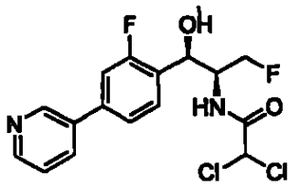
Example 94: Compound 116



116

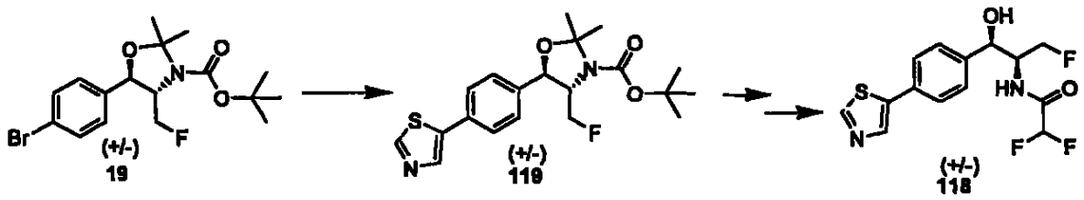
Compound **116** was synthesized as set forth in Schemes 2 and 3. The phenylserine analog was obtained by condensation of 4-bromo-2-fluorobenzaldehyde with glycine. Suzuki coupling method B was used to couple the bromide intermediate with *m*-pyridineboronic acid. Deprotection and difluoroacetylation were carried out as described previously herein. LRMS (ESI^+) m/z: 343 ($M+H^+$ $C_{16}H_{15}F_4N_2O_2$ requires 343.1).

Example 95: Compound 117



Same procedure as that used for the synthesis of **116**. LRMS (ESI^+) m/z: 375 ($M+H^+$ $C_{16}H_{15}F_4N_2O_2$ requires 375.1).

Example 96: Compound 118

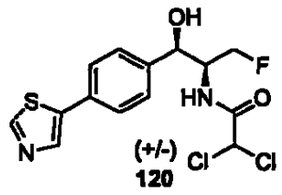


Compound **19** (100 mg, 0.258 mmol), potassium acetate (38 mg, 0.386 mmol), and 2 mL of *N,N*-dimethylacetamide (DMAC) were combined in a glass pressure tube. Thiazole (112 mg, 1.31 mmol) was added by syringe, and the syringe rinsed with an additional 1 mL of DMAC, which was added to the pressure tube. The mixture was stirred at room temperature while purging with N_2 for 10 minutes. $Pd(PPh_3)_4$ (15 mg, 0.013 mmol) was added, the mixture purged with N_2 for 5 minutes, the tube sealed and heated to 150 °C behind a blast shield and held at that temperature for 12 hours. The mixture became dark brown. After cooling to room temperature, the contents of the tube were filtered through a pad of Celite and the

filtrate evaporated to dryness. The residue was dissolved in 40 mL of 3:1 EtOAc/hexanes and washed with H_2O (2 x 15 mL) and brine (2 x 15 mL) and then dried over anhydrous Na_2SO_4. The dried organic layer was filtered to remove the drying agent and concentrated under vacuum to give 109 mg of brown oil. Compound **119** was purified by silica gel chromatography eluting with 80:20 to 50:50 hexanes/EtOAc (54 mg, 0.14 mmol). LRMS (ESI^+) m/z: 393.1 ($M+H^+$ $C_{20}H_{26}FN_2O_3S$ requires 393.2).

All of compound **119** was dissolved in 9:1 (v/v) TFA/H_2O (7.5 mL) and stirred at room temperature for 30 minutes. The mixture was then concentrated under vacuum and the residue dissolved twice in a mixture of toluene and methanol, the solvents being evaporated each time. A portion of the product (9.7 mg, 0.027 mmol) was dissolved in 2 mL of MeOH and 10 drops of Et_3N and 12 drops of methyl difluoroacetate were added. The mixture was left open to the air and stirred rapidly for 16 hours. The mixture was evaporated to dryness and the residue purified by chromatotron (1 mm plate) eluting with 3% MeOH in CH_2Cl_2 to give **118** (6.0mg, 0.018 mmol. ^1H NMR (300 MHz, CD_3OD): δ 4.27-4.69 (m, 3H), 4.97(d, 1H, J = 4.2), 5.99 (t, 1H, J = 53.9), 7.47 (d, 2H, J = 8.3), 7.65 (d, 2H, J = 8.3), 8.16 (s, 1H), 8.94 (s, 1H). LRMS (ESI^+) m/z: 331.1 ($M+H^+$ $C_{14}H_{14}F_3N_2O_2S$ requires 331.1).

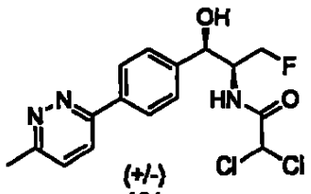
Example 97: Compound 120



120

Compound **120** was prepared in a manner analogous to that used to prepare **118** using dichloroacetylation instead of difluoroacetylation. LRMS (ESI^+) m/z: 363.0 ($M+H^+$ $C_{14}H_{14}Cl_2FN_2O_2S$ requires 363.0).

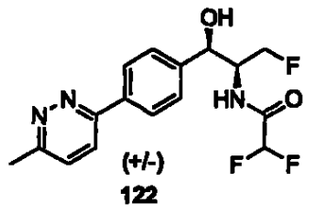
Example 98: Compound 121



(+/-)
121

The protected biaryl intermediate was synthesized by Suzuki coupling method B from 23 and 3-chloro-6-methylpyridazine. Deprotection and dichloroacetylation were performed as for other compounds of this invention. LRMS (ESI$^+$) m/z: 372.0 (M+H$^+$ $C_{16}H_{17}Cl_2FN_3O_2$ requires 372.1).

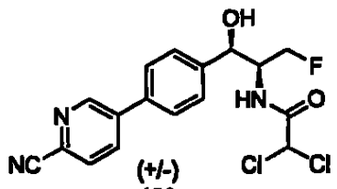
Example 99: Compound 122



(+/-)
122

Prepared in analogous fashion to 121. LRMS (ESI$^+$) m/z: 340.1 (M+H$^+$ $C_{16}H_{17}F_3N_3O_2$ requires 340.1).

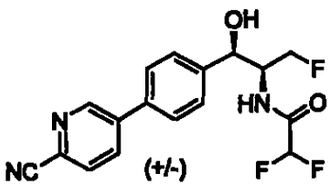
Example 100: Compound 123



(+/-)
123

The protected biaryl intermediate was prepared by Suzuki coupling method B from boronic acid 23 and the appropriate aryl bromide. This intermediate was then deprotected by brief treatment with 9:1 (v/v) TFA/H_2O and dichloroacetylated as described for 29. LRMS (ESI$^+$) m/z: 380.0 (M-H$^+$ $C_{17}H_{13}Cl_2FN_3O_2$ requires 380.0).

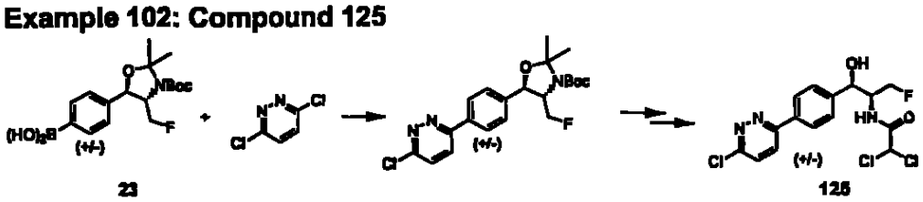
Example 101: Compound 124



124

Prepared in analogous fashion to **123**. Difluoroacetylation was performed as with other compounds of this invention. LRMS (ESI^+) m/z: 348.0 ($M+H^+$ $C_{17}H_{13}F_3N_3O_2$ requires 348.1).

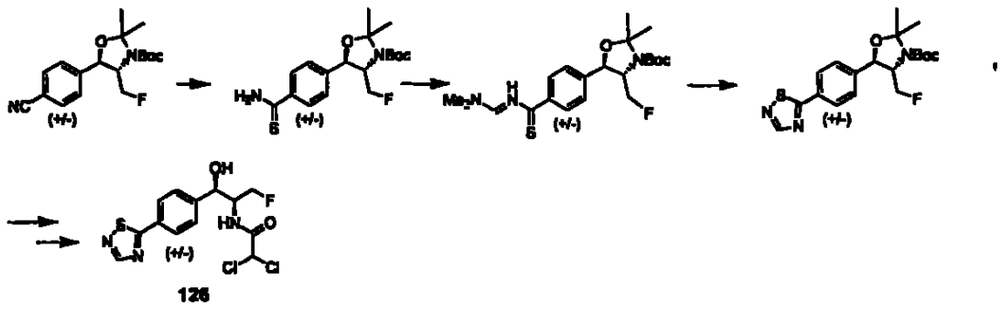
Example 102: Compound 125



23 125

The biaryl intermediate was prepared from boronic acid **23** using Suzuki coupling. LRMS (ESI^+) m/z: 392.0 ($M-H^+$ $C_{15}H_{13}Cl_3FN_3O_2$ requires 392.0).

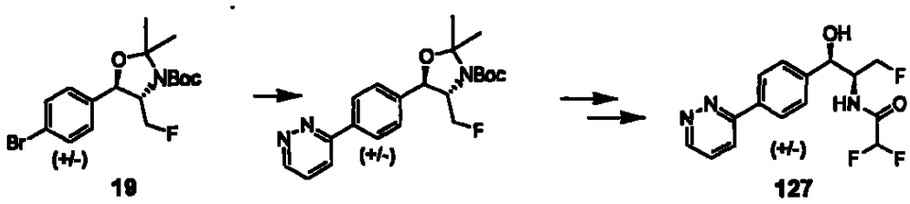
Example 103: Compound 126



126

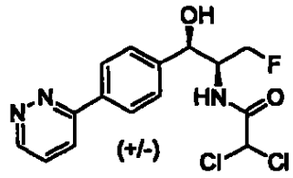
The starting nitrile was prepared from **35** by analogy to the preparation of bromo intermediate **19**. Treatment with $Ph_2P(S)SH$ gave the thiobenzamide which was converted to the thiobenzamidine intermediate by reaction with dimethylformamide dimethylacetal. Cyclization with hydroxylamine-O-sulfonic acid in methanol/pyridine gave the biaryl intermediate. LRMS (ESI^+) m/z: 364.0 ($M-H^+$ $C_{13}H_{12}Cl_2FN_3O_2S$ requires 364.0)

Example 104: Compound 127



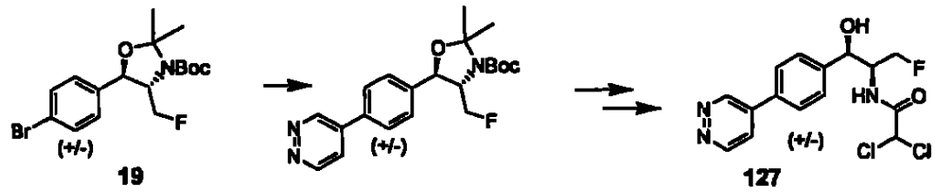
The phenyllithium intermediate was prepared by treatment of intermediate **19** with n-butyllithium in THF at -78 °C. The intermediate was reacted with pyridazine and the mixture of the resulting 2- and 3-position adducts was oxidized with DDQ. The desired 3-pyridazyl regioisomer was isolated by chromatography. LRMS (ESI^+) m/z: 326.0 ($M-H^+$ $C_{15}H_{14}F_3N_3O_2$ requires 326.0)

Example 105: Compound 128



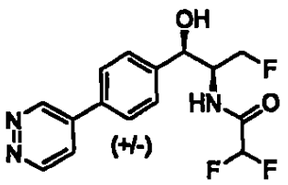
Compound **128** was synthesized using the procedure in Example 104. LRMS (ESI^+) m/z: 358.0 ($M-H^+$ $C_{15}H_{14}Cl_2FN_3O_2$ requires 358.0)

Example 106: Compound 129



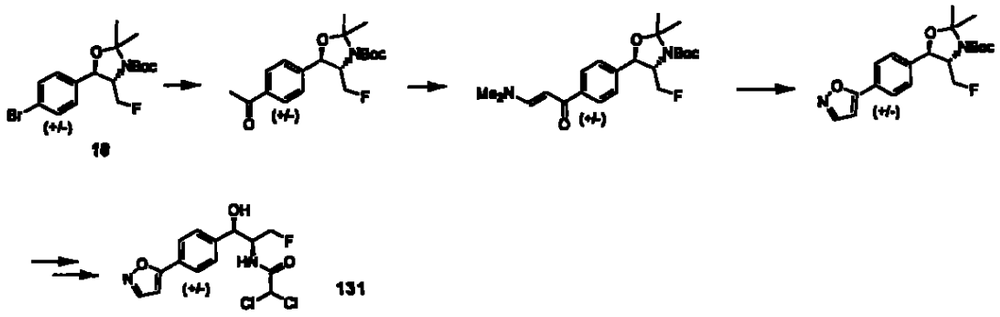
The procedure discussed in Example 104 was used. LRMS (ESI^+) m/z: 358.0 ($M-H^+$ $C_{15}H_{14}Cl_2FN_3O_2$ requires 358.0)

Example 107: Compound 130



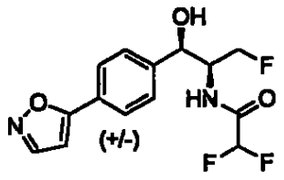
The procedure of Example 104 was used. LRMS (ESI^+) m/z: 326.0 ($M-H^+$ $C_{15}H_{14}F_3N_3O_2$ requires 326.0)

Example 108: Compound 131



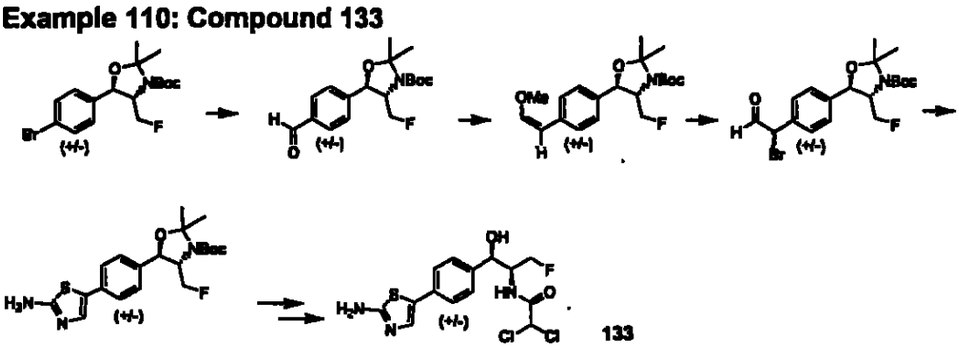
The phenyllithium intermediate was reacted with N-methoxy-N-methylacetamide and the resulting acetophenone was reacted with dimethylformamide dimethylacetal and hydroxylamine-O-sulfonic acid in methanol in presence of pyridine to give the isoxazole. LRMS (ESI^+) m/z: 347.0 ($M-H^+$ $C_{14}H_{13}Cl_2N_2O_3$ requires 347.0)

Example 109: Compound 132



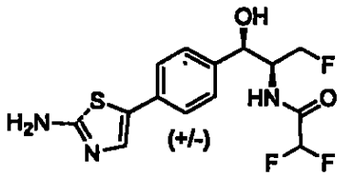
Compound **132** was synthesized using the procedure in Example 108. LRMS (ESI$^+$) m/z: 315.0 (M-H$^+$ $C_{14}H_{13}F_3N_2O_3$ requires 315.0.

Example 110: Compound 133



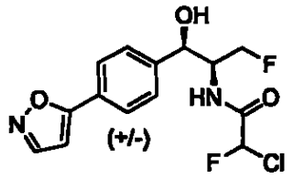
The phenyllithium intermediate was reacted with dimethylformamide to give the formyl intermediate which was then reacted with the ylide generated from methoxymethyltriphenyl phosphonium bromide. The resulting enol ether was brominated with bromine to give the bromoaldehyde. Cyclization with thiourea gave the aminothiazole. LRMS (ESI$^+$) m/z: 378.0 (M-H$^+$ $C_{14}H_{14}Cl_2FN_3O_2S$ requires 378.0)

Example 111: Compound 134



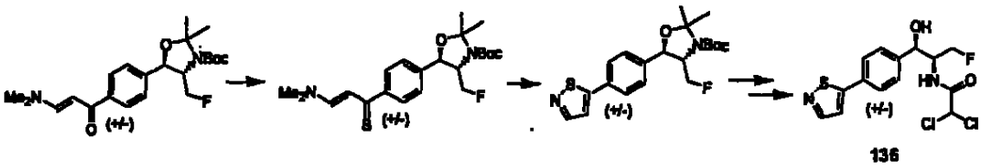
Compound **134** was synthesized using the procedure in Example 110. LRMS (ESI$^+$) m/z: 346.0 (M-H$^+$ $C_{14}H_{14}F_3N_3O_2S$ requires 346.0)

Example 112: Compound 135 (mixture of diastereoisomers)



Compound **135** was synthesized using the procedure in Example 108. LRMS (ESI$^+$) m/z: 331.0 (M-H$^+$ $C_{14}H_{13}F_2N_2O_3$ requires 331.0)

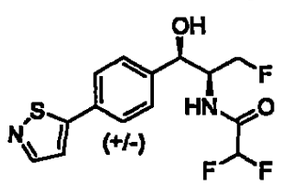
Example 113: Compound 136



136

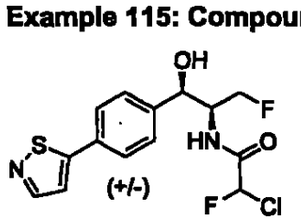
The starting material in Example 108 was reacted with Lawesson's reagent and the product was cyclized to the biaryl intermediate with hydroxylamine-O-sulfonic acid in methanol in presence of pyridine. LRMS (ESI^+) m/z: 363.0 ($M-H^+$ $C_{14}H_{13}Cl_2FN_2O_2S$ requires 363.0)

Example 114: Compound 137



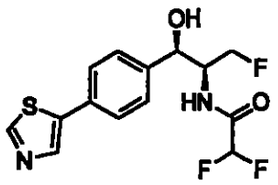
Compound **137** was synthesized using the procedure in Example 113. LRMS (ESI^+) m/z: 331.0 ($M-H^+$ $C_{14}H_{13}F_3N_2O_2S$ requires 331.0)

Example 115: Compound 138 (mixture of diastereoisomers)



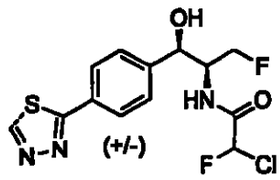
Compound **138** was synthesized using the procedure in Example 113. LRMS (ESI^+) m/z: 347.0 ($M-H^+$ $C_{14}H_{13}ClF_2N_2O_2S$ requires 347.0)

Example 116: Compound 139



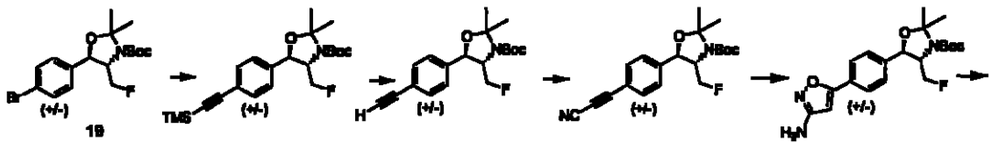
Compound **139** was synthesized using the procedure in Example 96 starting from the desired enantiomer of intermediate **19**. LRMS (ESI^+) m/z: 331.0 ($M-H^+$ $C_{14}H_{13}F_3N_2O_2S$ requires 331.0)

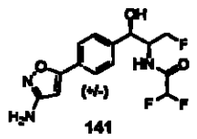
Example 117: Compound 140 (mixture of diastereoisomers)



Compound **140** was synthesized using the procedure in Example 41. LRMS (ESI^+) m/z: 348.0 ($M-H^+$ $C_{13}H_{12}ClF_2N_3O_2S$ requires 348.0)

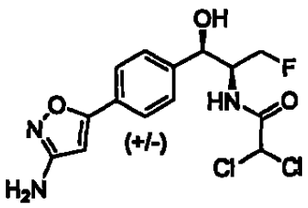
Example 118: Compound 141





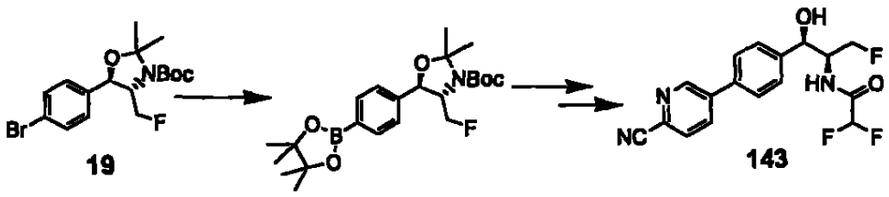
Intermediate **19** was reacted with trimethylsilylacetylene in presence of copper iodide and $PdCl_2(PPh_3)_2$. The product was desilylated by treatment with potassium carbonate in methanol. The cyanoacetylene intermediate was reacting acetylene with n-butyllithium, which was then reacted with tosylcyanide. Cyclization to the 3-aminoisoxazole was performed by treatment of the cyanoacetylene with hydroxylamine in ethanol. LRMS (ESI^+) m/z: 330.0 ($M-H^+$ $C_{14}H_{14}F_3N_3O_3$ requires 330.0)

Example 119: Compound 142



Compound **142** was synthesized using the procedure in Example 118. LRMS (ESI^+) m/z: 362.0 ($M-H^+$ $C_{14}H_{14}Cl_2FN_3O_3$ requires 362.0)

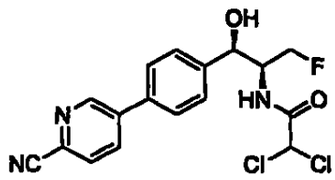
Example 120: Compound 143



19 **143**

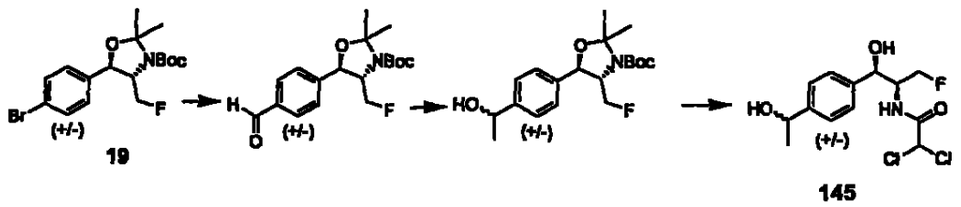
The desired enantiomer of intermediate **19** was reacted with bis(pinacolato)diboron in presence of 1,1'-bis(diphenylphosphino)ferrocenepalladium (II) dichloride dichloromethane. The pinacolboronate ester was coupled with 5-bromo-2-cyanopyridine using the Suzuki reaction. LRMS (ESI^+) m/z: 350.0 ($M-H^+$ $C_{17}H_{14}F_3N_3O_2$ requires 350.0)

Example 121: Compound 144



Compound **144** was synthesized using the procedure in Example 120. LRMS (ESI^+) m/z: 382.0 ($M-H^+$ $C_{17}H_{14}Cl_2FN_3O_2$ requires 382.0)

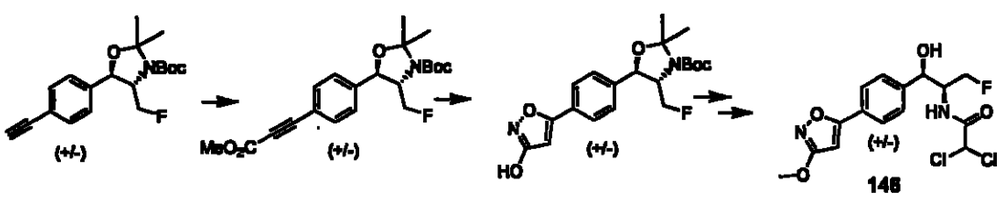
Example 122: Compound 145 (mixture of diastereoisomers)



19 **145**

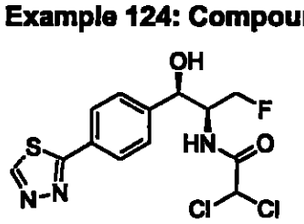
Intermediate 19 was reacted with n-butyllithium and then with dimethylformamide. The formyl intermediate was reacted with methylmagnesium bromide to give the benzylic alcohol as a mixture of diastereisomers. LRMS (ESI^+) m/z: 324.0 ($M-H^+$ $C_{13}H_{16}Cl_2FNO_3$ requires 324.0)

Example 123: Compound 146



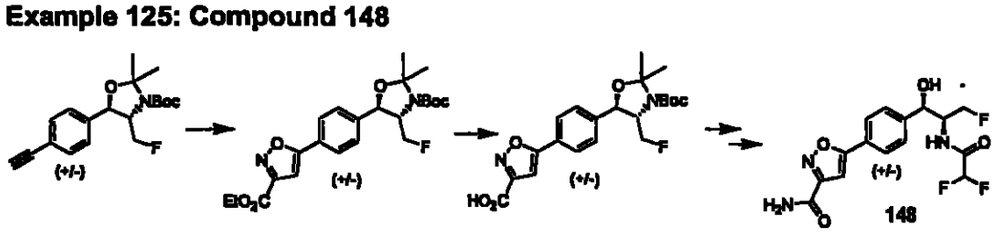
The starting acetylene, prepared as described in Example 118, was reacted with n-butyllithium followed by addition of carbon dioxide and the resulting acid was esterified with diazomethane in ethyl acetate/diethyl ether. The 3-hydroxyisoxazole was obtained by reacting the ester with hydroxylamine and was then methylated with diazomethane in ethyl acetate/diethyl ether. LRMS (ESI^+) m/z: 377.0 ($M-H^+$ $C_{15}H_{15}Cl_2FN_2O_4$ requires 377.0)

Example 124: Compound 147



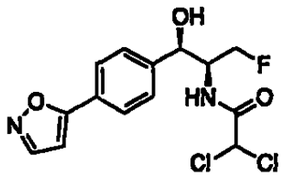
Compound 147 was synthesized by the method of Example 120 using 2-bromo-1,3,4-thiadiazole in a Suzuki coupling. LRMS (ESI^+) m/z: 364.0 ($M-H^+$ $C_{13}H_{12}Cl_2FN_3O_2S$ requires 364.0)

Example 125: Compound 148



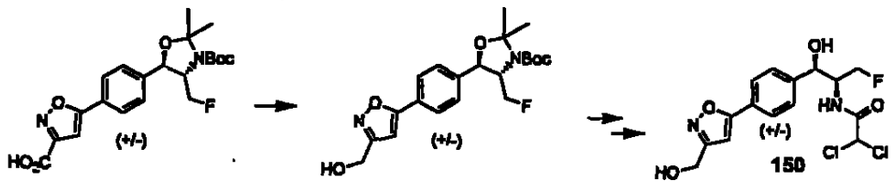
The starting acetylene was obtained as described in Example 118. The 3-carboethoxy isoxazole intermediate was obtained by cycloaddition of the nitrile oxide generated *in situ* from ethyl nitroacetate, di-t-butyldicarbonate and 4-dimethylaminopyridine. The amide was obtained from the 3-carboethoxy isoxazole intermediate by hydrolysis to the acid, conversion to the acyl chloride and reaction with ammonia. LRMS (ESI^+) m/z: 358.0 ($M-H^+$ $C_{15}H_{14}F_3N_3O_4$ requires 358.0)

Example 126: Compound 149



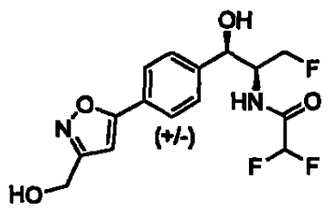
Compound **149** was synthesized using the procedure in Example 108 starting from the desired enantiomer of intermediate **19**. LRMS (ESI^+) m/z: 347.0 ($M-H^+$ $C_{14}H_{13}Cl_2FN_2O_3$ requires 347.0)

Example 127: Compound 150



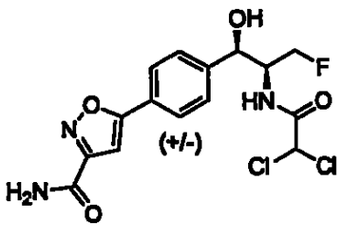
The 3-carboxyisoxazole intermediate obtained as described in Example 125 was converted into the acid chloride using Vilsmeyer reagent in dimethylformamide. The crude product was reduced to the 3-hydroxymethyl isoxazole intermediate with tetrabutylammonium borohydride in THF. LRMS (ESI^+) m/z: 377.0 ($M-H^+$ $C_{15}H_{15}Cl_2FN_2O_4$ requires 377.0)

Example 128: Compound 151



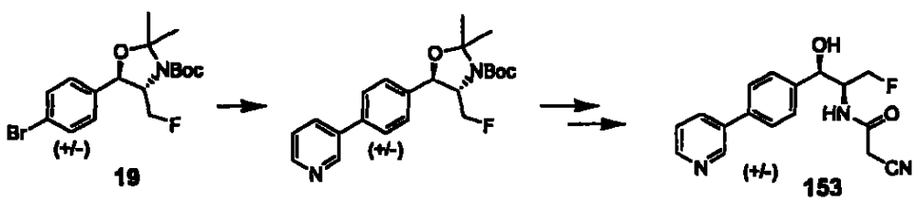
Compound **151** was synthesized using the procedure in Example 127.
LRMS (ESI$^+$) m/z: 345.0 (M-H$^+$ $C_{15}H_{15}F_3N_2O_4$ requires 345.0)

Example 129: Compound 152



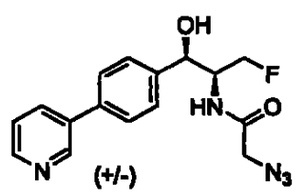
Compound **152** was synthesized using the procedure in Example 125.
LRMS (ESI$^+$) m/z: 390.0 (M-H$^+$ $C_{15}H_{14}Cl_2FN_3O_4$ requires 390.0)

Example 130: Compound 153



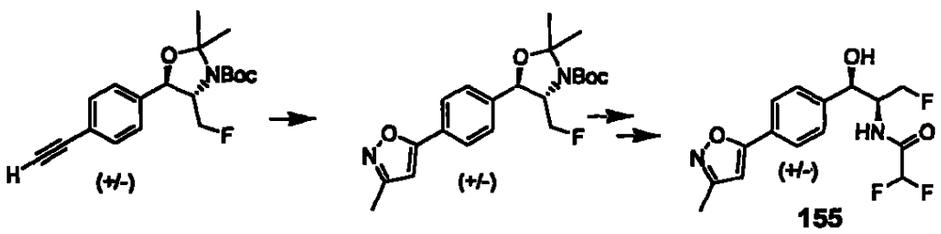
Compound **153** was synthesized using the procedure in Example 40.
EDC/HOBT was used to acylate the amino intermediate with cyanoacetic acid.
LRMS (ESI$^+$) m/z: 314.0 (M-H$^+$ $C_{17}H_{16}FN_3O_2$ requires 314.0)

Example 131: Compound 154



Compound **154** was synthesized using the procedure in Example 130.
LRMS (ESI$^+$) m/z: 330.0 (M-H$^+$ $C_{16}H_{16}FN_5O_2$ requires 330.0)

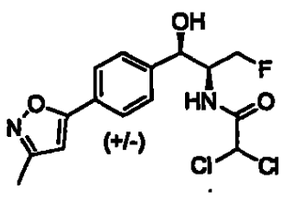
Example 132: Compound 155



155

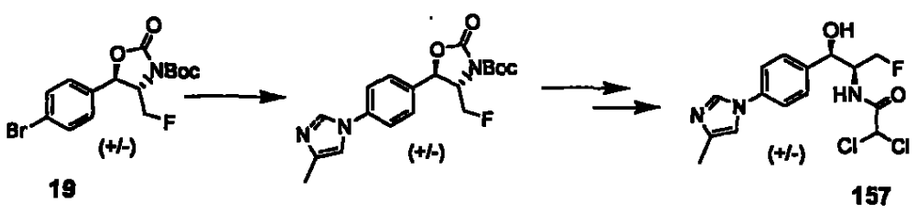
The starting acetylene was obtained as described in Example 118. The 3-methyl isoxazole intermediate was obtained by cycloaddition of the nitrile oxide generated *in situ* from nitroethane, di-t-butyldicarbonate and 4-dimethylaminopyridine. LRMS (ESI^+) m/z: 329.0 ($M-H^+$ $C_{15}H_{15}F_3N_2O_3$ requires 329.0)

Example 133: Compound 156



Compound **156** was synthesized using the procedure in Example 132. LRMS (ESI^+) m/z: 361.0 ($M-H^+$ $C_{15}H_{15}Cl_2FN_2O_3$ requires 361.0)

Example 134 : Compound 157

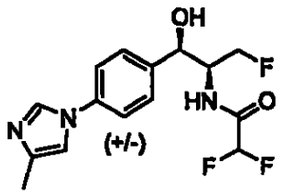


19

157

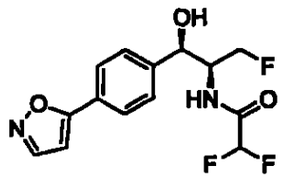
Intermediate **19** was reacted with 4-methylimidazole in refluxing dimethylformamide in the presence of copper powder. LRMS (ESI^+) m/z: 3460.0 ($M-H^+$ $C_{14}H_{14}Cl_2FN_3O_2$ requires 346.0)

Example 135: Compound 158



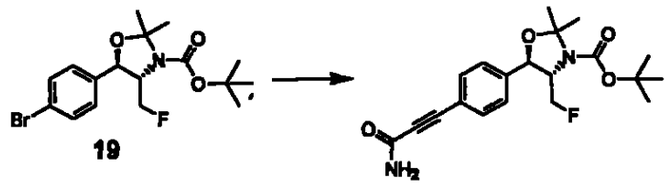
Compound **158** was synthesized using the procedure in Example 134. LRMS (ESI^+) m/z: 328.0 ($M-H^+$ $C_{15}H_{16}F_3N_3O_2$ requires 328.0)

Example 136: Compound 159



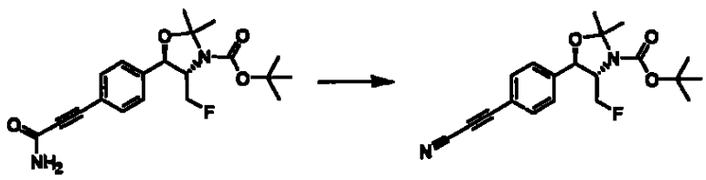
Compound **159** was synthesized using the procedure in Example 126. LRMS (ESI^+) m/z: 315.0 ($M-H^+$ $C_{14}H_{13}F_3N_2O_3$ requires 315.0)

Example 137: Compound 160

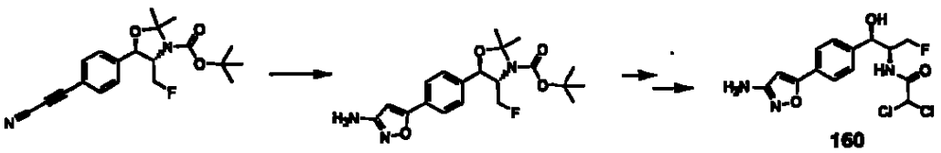


19

A mixture of the desired enantiomer of Intermediate **19** (1 g, 2.5 mmol), potassium carbonate (712 mg, 5 mmol), and propiolamide (1.07 g, 15 mmol) in N,N-dimethylacetamide (4 mL) was stirred at room temperature while purging with N_2 for 10 minutes. $Pd(PPh_3)_4$ catalyst (15 mg, 0.013 mmol) was added and the mixture was purged with N_2 for another 5 minutes. The resulting mixture was stirred at 100 °C for 9 hrs. After cooling to room temperature, the crude was flash silica gel chromatographed to give the coupling product (601 mg). ^1H NMR (300 MHz, $CDCl_3$): δ 1.49 (s, 9H), 1.59 (s, 3H), 1.70 (s, 3H), 3.75-3.90 (m, 1H), 4.35-4.60 (m, 2H), 5.15 (d, 1H, J = 7.5), 5.65 (s, 1H), 5.85 (s, 1H), 7.45 (d, 2H, J = 8.4), and 7.55 (d, 2H, J = 8.4).



To dimethylformamamide (2 mL) was slowly added thionyl chloride (2 mL) at 0 °C and the reaction mixture was stirred at that temperature for 30 min. The solution was then cannulated to a solution of the propiolamide intermediate (600 mg) in DMF (4 mL) at 0 °C. After stirring at room temperature for 30 min, the mixture was poured into ice/water (50 mL). The pH was adjusted to 7 with saturated sodium bicarbonate. The solution was extracted with ethyl acetate three times. The combined organic layers were washed with water, dried over anhydrous sodium sulfate, and purified by column chromatography to give propiolonitrile intermediate (326 mg). ^{1}H NMR (300 MHz, $CDCl_3$): δ 1.49 (s, 9H), 1.59 (s, 3H), 1.70 (s, 3H), 3.75-3.90 (m, 1H), 4.35-4.60 (m, 2H), 5.15 (d, 1H, J = 7.5), 7.49 (d, 2H, J = 8.4), and 7.63 (d, 2H, J = 8.4).

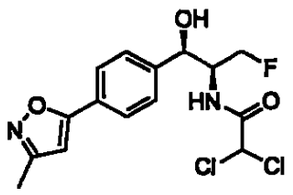


160

To a solution of sodium hydroxide (206 mg, 7 eq.) in water (2 mL), was added hydroxylamine hydrochloride (256 mg, 5 eq.). The solution was added to the propiolonitrile intermediate (263 mg) dissolved in ethanol (7 mL). The resulting mixture was stirred at room temperature for 6 hrs. Ethyl acetate was added, the organic layer was separated, washed with water, and dried with anhydrous sodium sulfate. The crude material was purified by column chromatography to give the protected 3-aminoisoxazole which was then dissolved in THF (6 mL) and 4 N HCl (6 mL), and stirred at 80 °C for 2.5 hrs. The reaction mixture was evaporated to dryness under reduced pressure. The residue was co-evaporated with methanol twice and dried overnight under vacuum. To the crude material dissolved in methanol (2 mL) was added triethylamine (2 mL), followed by methyl dichloroacetate (1 mL). The mixture was stirred at room temperature for 24 hrs. The solvent was evaporated and the mixture was purified by flash column chromatography to give compound **160** (167 mg). ^{1}H NMR (300 MHz, $CDCl_3$ +

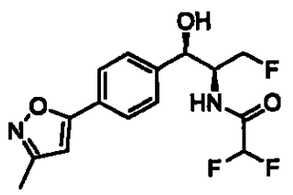
CD_3OD): δ 4.35-4.70 (m, 3H), 5.05 (m, 1H), 5.85 (s, 1H), 6.16 (s, 1H), 7.44 (d, 2H, J = 8.1), and 7.65 (d, 2H, J = 8.1). LRMS (ESI^+) m/z: 362.0 ($M-H^+$ $C_{14}H_{14}Cl_2FN_3O_3$ requires 362.0)

Example 138: Compound 161



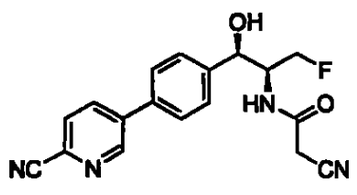
Compound **161** was synthesized using the procedure in Example 132 starting from the desired enantiomer of intermediate **19**. LRMS (ESI^+) m/z: 361.0 ($M-H^+$ $C_{15}H_{15}Cl_2FN_2O_3$ requires 361.0)

Example 139: Compound 162



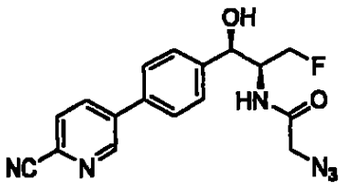
Compound **162** was synthesized using the procedure in Example 138. LRMS (ESI^+) m/z: 329.0 ($M-H^+$ $C_{15}H_{15}F_3N_2O_3$ requires 329.0)

Example 140: Compound 163



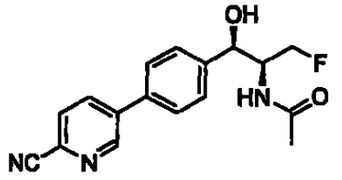
Compound **163** was synthesized using the procedure in Example 120. Dicyclohexylcarbodiimide was used to acylate the amino intermediate with cyanoacetic acid. LRMS (ESI^+) m/z: 339.0 ($M-H^+$ $C_{18}H_{15}FN_4O_2$ requires 339.0)

Example 141: Compound 164



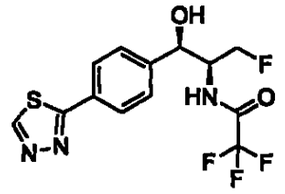
Compound **164** was synthesized using the procedure in Example 120. Dicyclohexylcarbodiimide was used to acylate the amino intermediate with azidoacetic acid. LRMS (ESI^+) m/z: 355.0 ($M-H^+$ $C_{17}H_{15}FN_6O_2$ requires 355.0)

Example 142: Compound 165



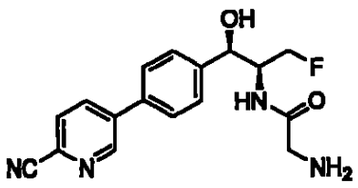
Compound **165** was synthesized using the procedure in Example 120. The amino intermediate was acetylated with acetic anhydride. LRMS (ESI^+) m/z: 314.0 ($M-H^+$ $C_{17}H_{16}FN_3O_2$ requires 314.0)

Example 143: Compound 166



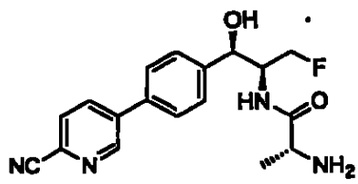
Compound **166** was synthesized using the procedure in Example 120. LRMS (ESI^+) m/z: 350.0 ($M-H^+$ $C_{13}H_{11}F_4N_3O_2S$ requires 350.0)

Example 144: Compound 167



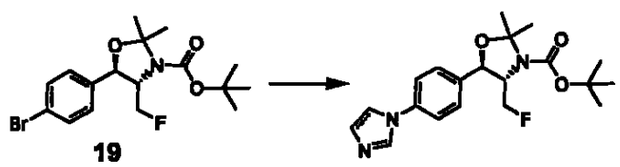
Compound **167** was synthesized using the procedure in Example 120. Dicyclohexylcarbodiimide was used to acylate the amino intermediate with N-Boc-glycine and the resulting glycinamide was deprotected with hydrochloric acid in methanol. LRMS (ESI^+) m/z: 329.0 ($M-H^+$ $C_{17}H_{17}FN_4O_2$ requires 329.0)

Example 145: Compound 168



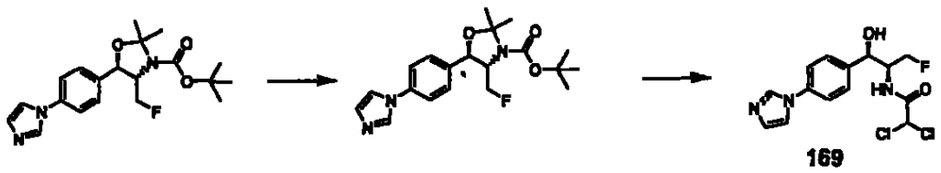
Compound **168** was synthesized using the procedure in Example 144. LRMS (ESI^+) m/z: 343.0 ($M-H^+$ $C_{18}H_{19}FN_4O_2$ requires 343.0)

Example 146 : Compound 169



19

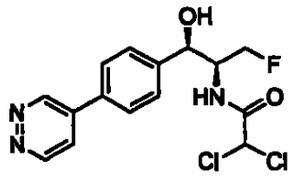
The desired enantiomer of intermediate **19** (1.16 g, 3 mmol), potassium carbonate (1.659 g, 12 mmol), imidazole (1.225 g, 18 mmol), and copper powder (191 mg, 3 mmol) in 20 mL of DMF were stirred vigorously at reflux for 4 hrs. The reaction mixture was cooled to room temperature and poured into water (100 mL). After extraction with ethyl acetate and washing of the organic extract with water (3X), the organic layer was passed through a silica gel and anhydrous sodium sulfate plug. Evaporation of solvent gave the imidazole (1.06 g). ^1H NMR (300 MHz, $CDCl_3$): δ 1.45 (s, 9H), 1.60 (s, 3H), 1.75 (s, 3H), 3.75-3.85 (m, 1H), 4.35-4.55 (m, 2H), 5.20 (d, 1H, J = 7.5), 7.22 (s, 1H), 7.28 (s, 1H), 7.41 (d, 2H, J = 8.4), 7.58 (d, 2H, J = 8.4), and 7.89 (s, 1H).



169

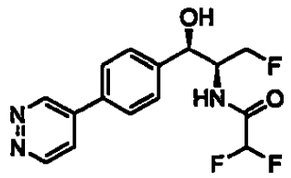
The imidazole intermediate (2.77 g) was dissolved in THF (20 mL) and 4 N HCl (20 mL), and stirred at 80 °C for 2.5 hrs. The reaction mixture was evaporated to dryness under reduced pressure. The residue was co-evaporated with methanol twice and dried overnight under vacuum. The crude material was dissolved in methanol (20 mL) and triethylamine (5 mL) was added, followed by methyl dichloroacetate (5 mL). The mixture was stirred at room temperature for 20 hrs. The solvent was evaporated and the mixture was purified by flash column chromatography to give compound **169** (1.1 g) as white solid. ^{1}H NMR (300 MHz, $CDCl_3$): δ 4.30-4.75 (m, 3H), 5.21 (d, 1H, J = 7.5), 5.86 (s, 1H), 7.05 (d, 1H, J=7.5), 7.10 (s, 1H), 7.28 (s, 1H), 7.39 (d, 2H, J =8.4), 7.52 (d, 2H, J=8.4 Hz), and 7.81 (s, 1H). LRMS (ESI$^+$) m/z: 360.0 (M-H$^+$ $C_{15}H_{16}Cl_2FN_3O_2$ requires 360.0)

Example 147: Compound 170



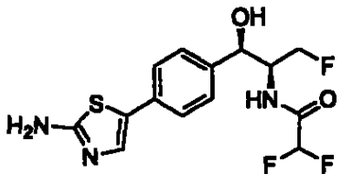
Compound **170** was synthesized by the method of Example 106 using the desired enantiomer of intermediate **19**. LRMS (ESI$^+$) m/z: 358.0 (M-H$^+$ $C_{15}H_{14}Cl_2FN_3O_2$ requires 358.0)

Example 148: Compound 171



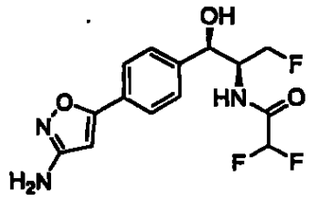
Compound **171** was synthesized using the procedure in Example 147. LRMS (ESI$^+$) m/z: 326.0 (M-H$^+$ $C_{15}H_{14}F_3N_3O_2$ requires 326.0)

Example 149: Compound 172



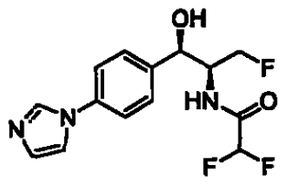
Compound **172** was synthesized by the method of Example 110 using the desired enantiomer of Intermediate **19**. LRMS (ESI^+) m/z: 346.0 ($M-H^+$ $C_{14}H_{14}F_3N_3O_2S$ requires 346.0)

Example 150: Compound 173



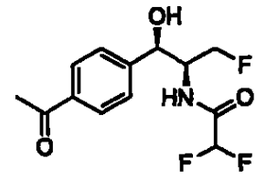
Compound **173** was synthesized using the procedure in Example 137. LRMS (ESI^+) m/z: 330.0 ($M-H^+$ $C_{14}H_{14}F_3N_3O_3$ requires 330.0)

Example 151: Compound 174



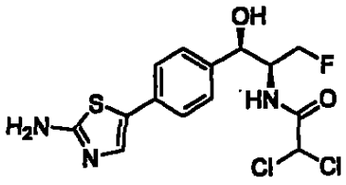
Compound **174** was synthesized using the procedure in Example 146. LRMS (ESI^+) m/z: 314.0 ($M-H^+$ $C_{14}H_{14}F_3N_3O_2$ requires 314.0)

Example 152: Compound 175



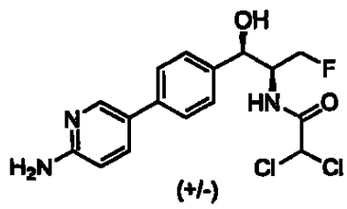
Compound **175** was synthesized using the procedure in Example 28. LRMS (ESI^+) m/z: 290.0 ($M-H^+$ $C_{13}H_{14}F_3NO_3$ requires 290.0)

Example 153: Compound 176



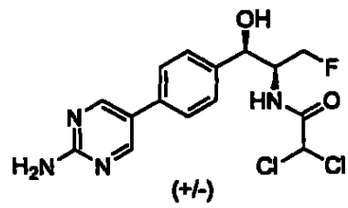
Compound **176** was synthesized using the procedure in Example 149. LRMS (ESI^+) m/z: 378.0 ($M-H^+$ $C_{14}H_{14}Cl_2FN_3O_2S$ requires 378.0)

Example 154: Compound 177



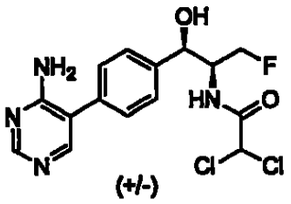
Compound **177** was synthesized by the method of Example 33 using the bromopyridine derivative and boronic acid **23**. LRMS (ESI^+) m/z: 372.0 ($M-H^+$ $C_{16}H_{16}Cl_2FN_3O_2$ requires 372.0)

Example 155: Compound 178



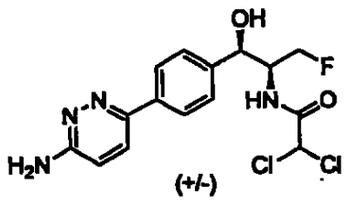
Compound **178** was synthesized by the method of Example 33 using a bromopyrimidine derivative and boronic acid **23**. LRMS (ESI^+) m/z: 373.0 ($M-H^+$ $C_{15}H_{15}Cl_2FN_4O_2$ requires 373.0)

Example 156: Compound 179



Compound **179** was synthesized by the method of Example 33 using the bromopyrimidine derivative and boronic acid **23**. Standard conditions were used for the removal of protecting groups and the introduction of dichloroacetamide functionality. LRMS (ESI^+) m/z: 373.0 ($M-H^+$ $C_{15}H_{15}Cl_2FN_4O_2$ requires 373.0)

Example 157: Compound 180



Compound **180** was synthesized by the method of Example 33 using the requisite chloropyridazine derivative and boronic acid **23**. Standard conditions were used for the removal of protecting groups and the introduction of dichloroacetamide functionality. LRMS (ESI^+) m/z: 373.0 ($M-H^+$ $C_{15}H_{15}Cl_2FN_4O_2$ requires 373.0)

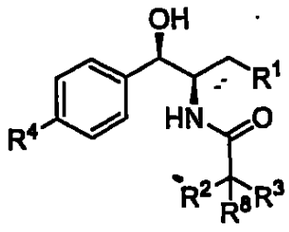
CONCLUSION

Thus, it will be appreciated that the present invention provides novel florfenicol-like compounds and methods for their use in the treatment or prevention of bacterial infection in animals or humans.

Although certain embodiments and examples have been used to describe the present invention, it will be apparent to those skilled in the art that changes in the embodiments and examples shown may be made without departing from the scope of this invention.

REIVINDICACIONES:

1. Un compuesto de la fórmula química:



en donde:

R^1 es seleccionado entre del grupo que consiste de –OH y –F;

R^2 y R^3 son cada uno seleccionados de manera independiente del grupo que consiste de hidrógeno, alquilo (1C-4C), halo, $-CF_3$, $-NH_2$, -CN y N_3;

R^4 es seleccionado del grupo que consiste de:

$-C(=R^5)R^6$,

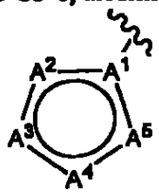
en donde;

R^5 es seleccionado del grupo que consiste de oxígeno, N-C≡N, y NOR^7,

en donde:

R^7 es seleccionado del grupo que consiste de hidrógeno, alquilo, arilo, heteroarilo, alicíclico y heteroalicíclico;

R^6 es seleccionado del grupo que consiste de hidrógeno, alquilo C1-4, cicloalquilo C3-6, alcoxilo C1-4, heteroarilo, alicíclico y heteroalicíclico;



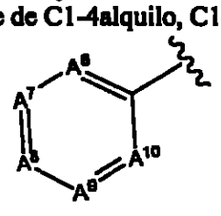
en donde:

A^1 es carbono o nitrógeno;

A^2, A^3, A^4 y A^5 son cada uno, de manera independiente, seleccionados del grupo que consiste de carbono, nitrógeno, oxígeno, y azufre, siempre y cuando por lo menos uno entre A^1 y A^5 no sea carbono, que el número total de átomos de nitrógeno, oxígeno y azufre en el anillo no exceda 4 y que el anillo sea aromático;

los átomos de carbono en el anillo son sustituidos de manera independiente con una entidad seleccionada del grupo que consiste de hidrógeno, alquilo C1-4, cicloalquilo C3-6, C1-4 alquilo O-, $-CF_3$, -OH, -CN, halo, C1-4alquilo S(O)-, C1-4alquilo $S(O)_2$, NH_2SO_2-, C1-4alquilo $NHSO_2$-, $((1C-4C)alquilo)_2NH$-, C1-4alquilo C(O)-, C3-6cicloalquilo C(O)-, C1-4alquilo OC(O)-, C1-4alquilo C(O)NH-, $-C(O)NH_2$, C1-4alquilo NHC(O)- y $((1C-4C)alquilo)_2NC(O)$-, en donde cualquiera de los grupos alquilo en cualquiera de los sustituyentes pueden ser no sustituidos o sustituidos con un grupo seleccionado entre halo y –OH;

si A^1 es carbono y el anillo no contiene oxígeno o azufre, uno de los átomos de nitrógeno puede estar opcionalmente sustituido con una entidad seleccionada a partir del grupo que consiste de C1-4alquilo, C1-4alquilo $S(O)_2$- y $-NH_2$;



en donde:

A^6, A^7, A^8, A^9 y A^{10} son seleccionados, de manera independiente, del grupo que consiste de carbono, nitrógeno y $>N^+—O^-$, siempre y cuando que solo uno a la vez entre A^6 y A^{10} sea $>N^+—O^-$;

los átomos de carbono en el anillo son sustituidos de manera independiente con una entidad seleccionada del grupo que consiste de hidrógeno, alquilo C1-4, cicloalquilo C3-6, C1-4 alquilo O-, $-CF_3$, -OH, -CN, halo, C1-4alquilo S(O)-, C1-4alquilo $S(O)_2$, NH_2SO_2-, C1-4alquilo $NHSO_2$-, $((1C-4C)alquilo)_2NH$-, C1-4alquilo C(O)-, C3-6cicloalquilo C(O)-, C1-4alquilo OC(O)-, C1-4alquilo C(O)NH-, $-C(O)NH_2$, C1-4alquilo NHC(O)-, $((1C-4C)alquilo)_2NC(O)$-, y $-OCH_2O$-, los átomos de oxígeno del grupo $-OCH_2O$- están enlazados a átomos de carbono adyacentes en el anillo, y en donde cualquiera de los grupos alquilo en cualquier sustituyente pueden ser no sustituidos o sustituidos con un grupo seleccionado entre halo y –OH;

R^8 es hidrógeno en todos los compuestos, excepto que cuando R^2 y R^3 son ambos F, entonces R^8 es hidrógeno o F; y

el compuesto es tanto un racemato, mostrando la estereoquímica relativa, o es sustancialmente enantioméricamente pura y tiene la estereoquímica absoluta mostrada.

2. El compuesto de la reivindicación 1, en donde R^1 es –F.

3. El compuesto de la reivindicación 2, en donde:

R^2 y R^3 son cada uno seleccionados de manera independiente, del grupo que consiste de Cl y F; y

R^8 es hidrógeno.

4. El compuesto de la reivindicación 3, en donde:

R^4 es $-C(=R^5)R^6$,

en donde:

R^5 es seleccionado del grupo que consiste de oxígeno, N-C≡N, y NOR^7,

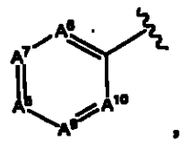
en donde:

R^7 es seleccionado del grupo que consiste de hidrógeno, alquilo, arilo, heteroarilo, alicíclico y heteroalicíclico; y,

R^6 es seleccionado del grupo que consiste de hidrógeno, alquilo C1-4, cicloalquilo C3-6, alcoxilo C1-4, heteroarilo, alicíclico y heteroalicíclico.

5. El compuesto de la reivindicación 4, en donde R^4 es $CH_3C(O)$-.

6. El compuesto de la reivindicación 2, en donde:



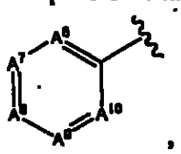
R^4 es

en donde:

A^6, A^7, A^8, A^9 y A^{10} son tal como se definen en la reivindicación 1; y,

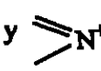
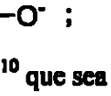
cualquiera entre A^6 y A^{10} que sea carbono es sustituido por una entidad seleccionada del grupo que consiste de hidrógeno, $-NH_2$, halo, -CN, alquilo C1-4, C1-4alquilo C(O)-, C1-4alquilo S(O)-, C1-4alquilo $S(O)_2$, NH_2SO_2-, C1-4alquilo SO_2NH-, C1-4alquilo $NHSO_2$-, $((1C\text{-}4C)alquilo)_2NSO_2$-, en donde cualquier grupo alquilo en cualquier sustituyente puede ser no sustituido o sustituido con un grupo seleccionado entre halo y –OH.

7. El compuesto de la reivindicación 3, en donde:



R^4 es

en donde:

A^6, A^7, A^8, A^9 y A^{10} son seleccionados, de manera independiente, del grupo que consiste de carbono, nitrógeno y  , siempre y cuando que solo uno a la vez entre A^1 y A^5 sea  ;

cualquiera entre A^6 y A^{10} que sea carbono es sustituido por una entidad seleccionada del grupo que consiste de hidrógeno, $-NH_2$, halo, -CN, alquilo C1-4, C1-4alquilo C(O)-, C1-4alquilo S(O)-, C1-4alquilo $S(O)_2$, NH_2SO_2-, C1-4alquilo SO_2NH-, C1-4alquilo $NHSO_2$-, $((1C\text{-}4C)alquilo)_2NSO_2$-, en donde cualquier grupo alquilo en

cualquier sustituyente puede ser no sustituido o sustituido con un grupo seleccionado entre halo y –OH.

8. El compuesto de la reivindicación 2, en donde:

uno, dos o tres entre A^6 y A^{10} es/son nitrógeno; y,

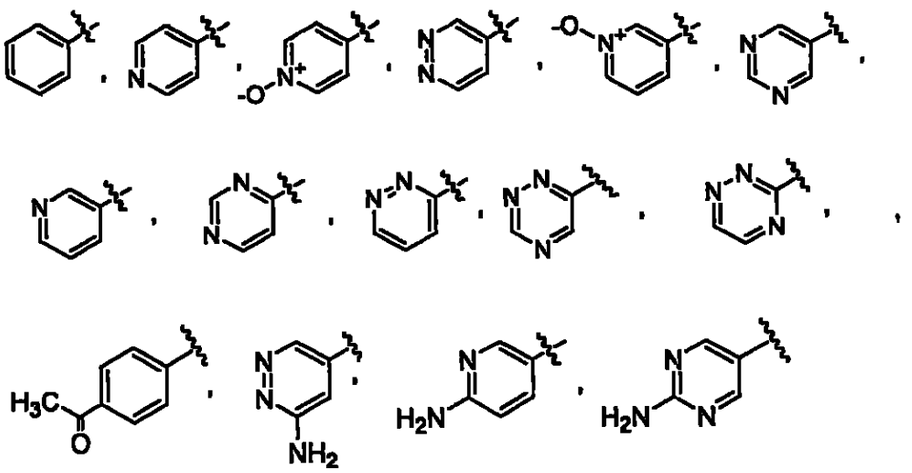
uno o dos de los átomos de carbono restantes en el anillo es/son opcionalmente sustituidos por $–NH_2$; todos los otros átomos de carbono en el anillo son no sustituidos.

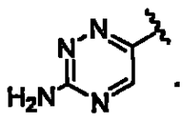
9. El compuesto de la reivindicación 3, en donde:

uno, dos o tres entre A^6 y A^{10} es/son nitrógeno; y,

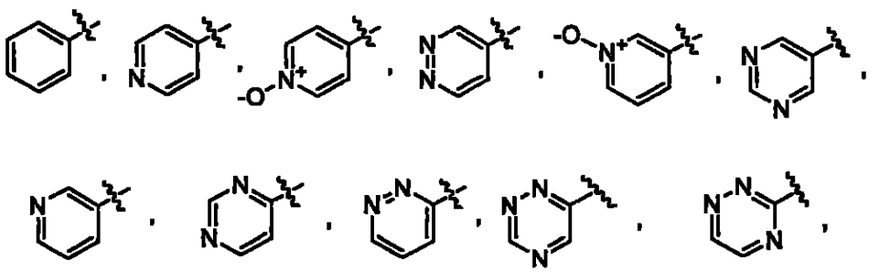
uno o dos de los átomos de carbono restantes en el anillo es/son opcionalmente sustituidos por $–NH_2$; todos los otros átomos de carbono en el anillo son no sustituidos.

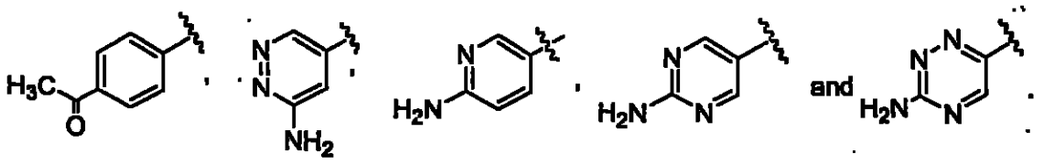
10. El compuesto de la reivindicación 2, en donde R^4 es seleccionado entre el grupo que consiste de:



and 

11. El compuesto de la reivindicación 3, en donde R^4 es seleccionado entre el grupo que consiste de:





12. El compuesto de la reivindicación 2, en donde:

R^4 es  ,

en donde A^1, A^2, A^3, A^4 y A^5 son tal como se definen en la reivindicación 1.

13. El compuesto de la reivindicación 3, en donde:

R^4 es  ,

en donde A^1, A^2, A^3, A^4 y A^5 son tal como se definen en la reivindicación 1.

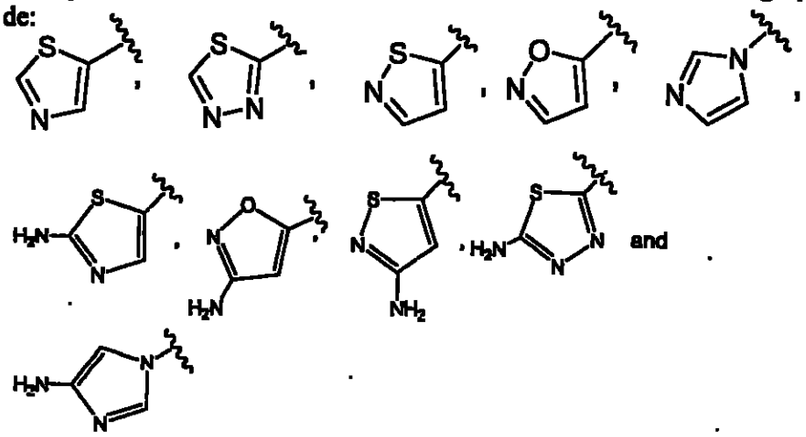
14. El compuesto de la reivindicación 12, en donde todos los átomos de carbono y nitrógeno son no sustituidos.

15. El compuesto de la reivindicación 13, en donde todos los átomos de carbono y nitrógeno son no sustituidos.

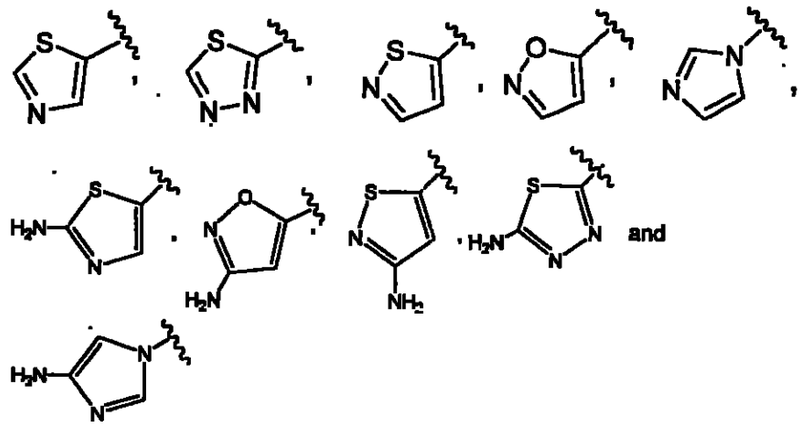
16. El compuesto de la reivindicación 12, en donde uno entre A^2 y A^5 que sea carbono es sustituido por un grupo $-NH_2$, todos los otros carbonos, y, si aplica, los átomos de nitrógeno en el anillo, son no sustituidos.

17. El compuesto de la reivindicación 13, en donde uno entre A^2 y A^5 que sea carbono es sustituido por un grupo $-NH_2$, todos los otros carbonos, y, si aplica, los átomos de nitrógeno en el anillo, son no sustituidos.

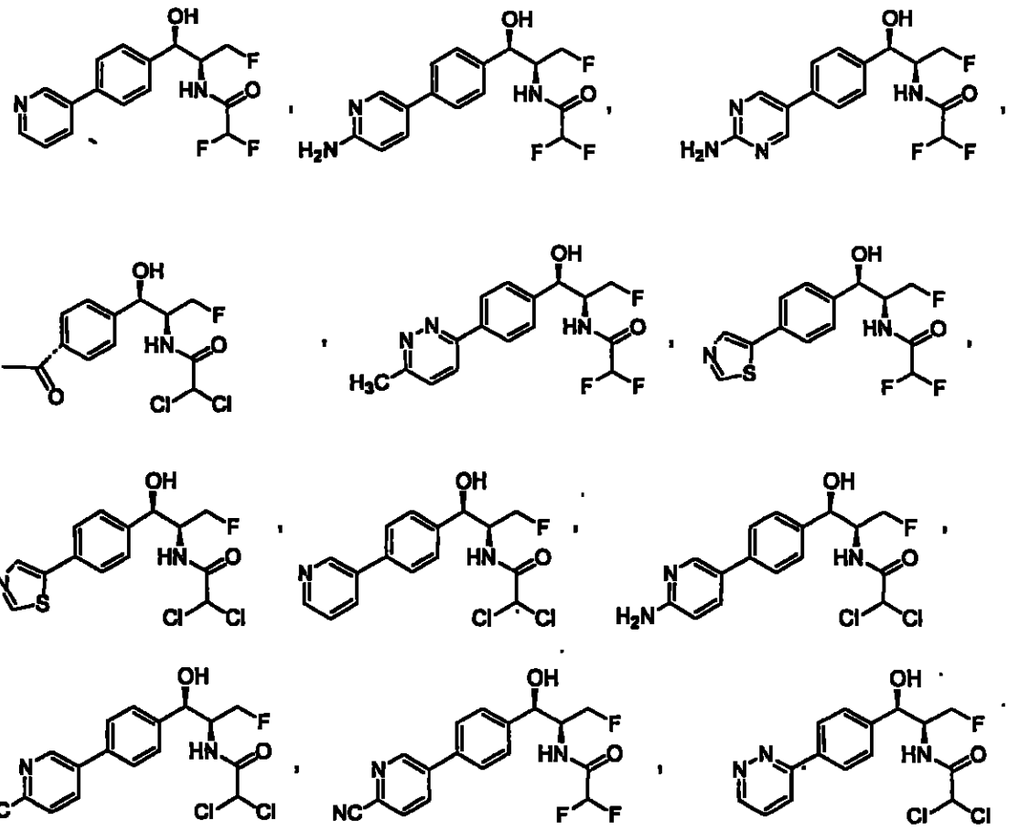
18. El compuesto de la reivindicación 2, en donde R^4 es seleccionado del grupo que consiste de:

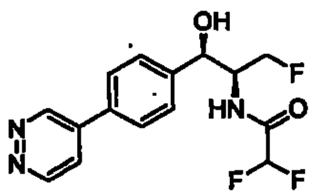
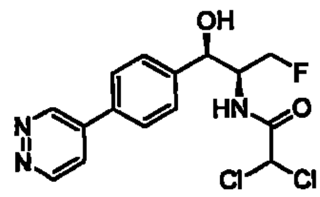
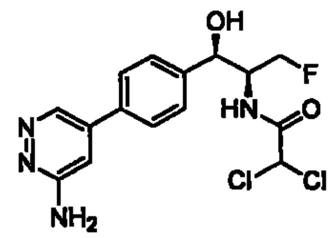


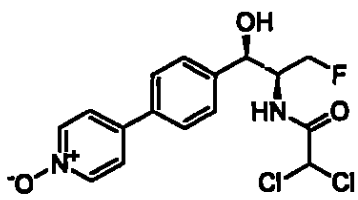
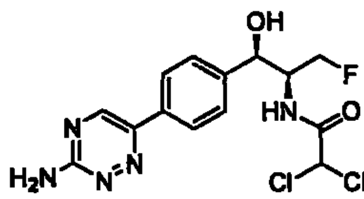
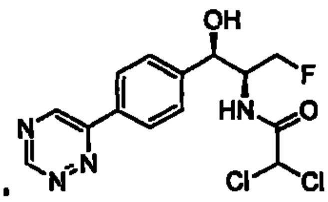
19. El compuesto de la reivindicación 3, en donde R^4 es seleccionado del grupo que consiste de:

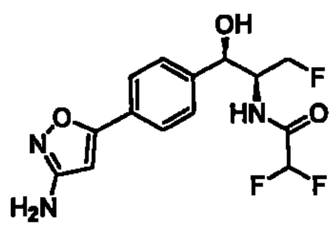
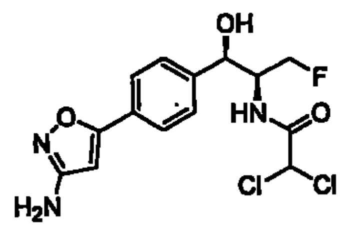
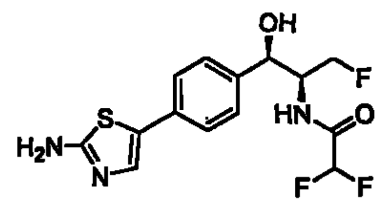


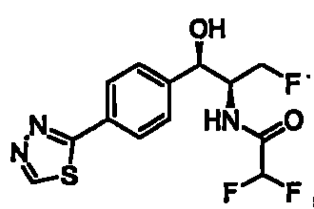
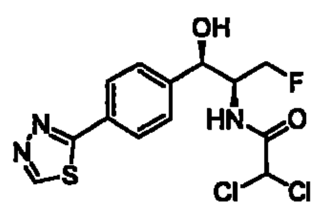
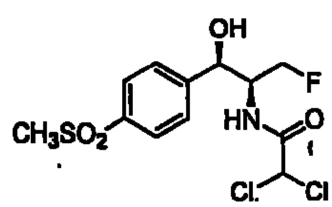
20. Un compuesto de la reivindicación 1, seleccionado entre:

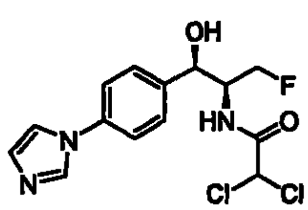


 ,



 ,
 ,


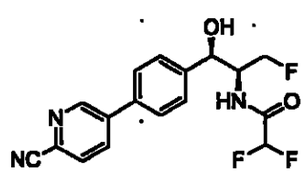
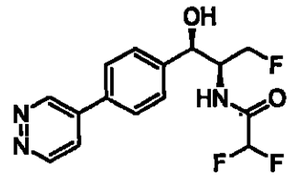
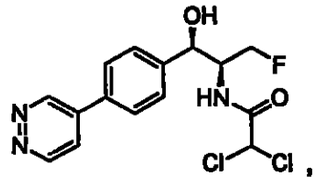
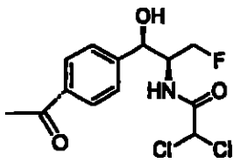
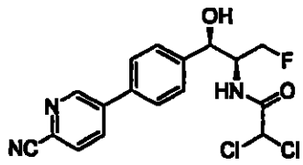
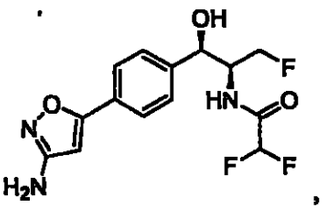
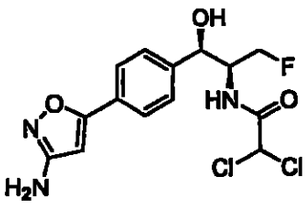
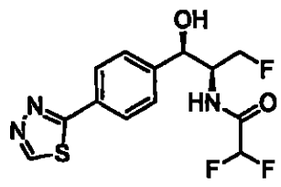
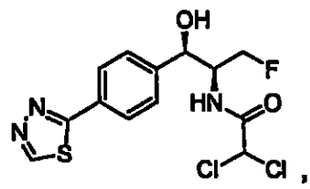
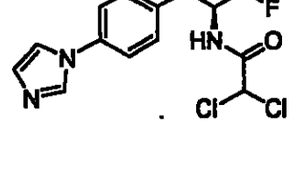
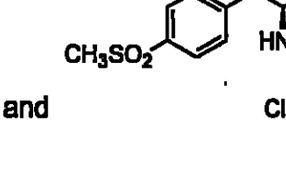
 ,
 ,
 ,

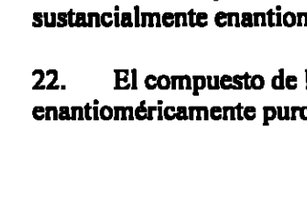
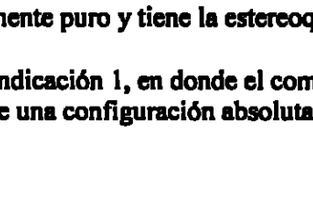
 ,
 ,
 , and

 ,

en donde el compuesto es un racemato, mostrando la estereoquímica relativa, o es sustancialmente enantioméricamente puro y tiene la estereoquímica absoluta mostrada.

21. Un compuesto de la reivindicación 1 seleccionado del grupo que consiste de:

en donde el compuesto es un racemato, mostrando la estereoquímica relativa, o es sustancialmente enantioméricamente puro y tiene la estereoquímica absoluta mostrada.

22. El compuesto de la reivindicación 1, en donde el compuesto es sustancialmente enantioméricamente puro y tiene una configuración absoluta de 1-(R)-2-(S).

23. Un método para tratar o prevenir una infección bacteriana, el cual comprende administrar una cantidad farmacéuticamente efectiva de un compuesto de la reivindicación 1, a un paciente en necesidad del mismo.

24. El método de la reivindicación 23, en donde la infección bacteriana es causada por una bacteria del género *Pasteurella, Haemophilus, Fusobacterium, Bacterioides, Aeromonas, Enterobacter, Escherichia, Klebsiella, Salmonella, Shigella, Actinobacillus, Streptococcus, Mycoplasma, Edwardsiella, Staphylococcus, Enterococcus, Bordetella, Proteus,* o *Mannheimia.*

25. El método de la reivindicación 31, en donde la infección bacteriana es causada por *Mannhemia haemolytica, Pasteurella multocida, Haemophilus somnus, Fusobacterium necrophorum, Bacterioides melaninogenicus, Actinobacillus pleuropneumoniae, Streptococcus suis, Salmonella cholerasuis, Mycoplasma bovis, Mycoplasma hyopneumoniae, Mycoplasma hyorhinis, Mycoplasma gallisepticum, Edwardsiella ictaluri, Escherichia coli, Enterobacter cloacae, Staphylococcus aureus, Staphylococcus intermedius, Enterococcus faecalis, Enterococcus faecium, Klebsiella pneumoniae, Klebsiella oxytoca, Enterobacter cloacae, Proteus mirabilis, o Aeromonas salmonicida.*

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PATENT COOPERATION TREATY

From the
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Schering-Plough Corporation
Patent Department K-6-1 1990
2000 Galloping Hill Road
Kenilworth, New Jersey 07033-0530
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NOTIFICATION OF TRANSMITTAL OF INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Rule 71.1)

Date of mailing *(day;month;year)*	02/02/2004
Applicant's or agent's file reference: AH06114	IMPORTANT NOTIFICATION

International application No.	International filing date *(day;month;year)*	Priority date *(day;month;year)*
PCT/IB03/03140	07/03/2003	08/03/2002

Applicant

SCHERING-PLOUGH, LTD.

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.

The applicant's attention is drawn to Article 33(5), which provides that the criteria of novelty, inventive step and industrial applicability described in Article 33(2) to (4) merely serve the purposes of international preliminary examination and that "any Contracting State may apply additional or different criteria for the purposes of deciding whether, in that State, the claimed invention is patentable or not" (see also Article 27(5)). Such additional criteria may relate, for example, to exemptions from patentability, requirements for enabling disclosure, clarity and support for the claims.

Name and mailing address of the IPEA:	Authorized officer
European Patent Office D-80298 Munich Tel. (+49-89) 2399-0, Tx: 523656 epmu d Fax: (+49-89) 2399-4465	AIMALE MURILLO J Tel. (+49-89) 2399 2828

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PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

(Rationalised Report according to the Notice of the President of the EPO published in the OJ11/2001)

Applicant's or agent's file reference	FOR FURTHER ACTION	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
AH06114		
International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/IB03/03140	07/03/2003	08/03/2002
International Patent Classification (IPC) or national classification and IPC		
C07C233/18		
Applicant		
SCHERING-PLOUGH, LTD.		

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 2 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 consists of a total of _____ 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	Date of completion of this report
06/10/2003	28/01/2004
Name and mailing address of the IPEA/	Authorized officer
European Patent Office D-80298 Munich Tel. (+49-89) 2399-0, Tx: 523656 epmu d Fax: (+49-89) 2399-4465	JONES M H Tel. (+49-89) 2399 2828

Form PCT/IPEA/409 (cover sheet) P20476 (October 2002)

INTERNATIONAL PRELIMINARY EXAMINATI N REPORT

International application No. PCT/IB03/03140

I. Basis of the report

The basis of this international preliminary examination 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 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

To the extent that the international preliminary examination has been carried out (see item III above), the following is pointed out:

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).

PATENT COOPERATION TREATY

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

ACTION RESPONDED TO
DATE
ATTY INITIALS

PCT Computer Input
Date:
PA:
Atty:
FILE ☐ CHECK FILE

WRITTEN OPINION (PCT Rule 66)

To:
Richard A. Elder
Schering-Plough Corporation
Patent Department K-6-1 1990
2000 Galloping Hill Road
Kenilworth, New Jersey 07033-0530
ETATS-UNIS D'AMERIQUE

RESPONSE DUE:
OFFICE ACTION DUE:

Date of mailing *(day/month/year)*	27/11/2003
Applicant's or agent's file reference: AH06114	REPLY DUE within 1 / 00 months/days from the above date of mailing

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/IB 03/03140	07/03/2003	08/03/2002

International Patent Classification (IPC) or both national classification and IPC
C07C233/18

Applicant
SCHERING-PLOUGH, LTD.

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 [X] Basis of the opinion
- II [ ] Priority
- III [X] Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
- IV [ ] Lack of unity of invention
- V [X] 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 66.4*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: 08/07/2004.

Name and mailing address of the IPEA/	Authorized officer
European Patent Office D-80298 Munich Tel. (+49-89) 2399-0, Tx: 523656 epmu d Fax: (+49-89) 2399-4465	Examiner Formalities officer (incl. extension of time limits) Tel. (+49-89) 2399 2828

Form PCT/IPEA/408 (cover sheet) (march 2002)

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.

B U R E A U
D.A. CASALONGA - JOSSE

CONSEILS EN PROPRIÉTÉ INDUSTRIELLE
EUROPEAN PATENT AND TRADEMARK ATTORNEYS
PAUL-HEYSE-STRASSE 33 D - 80336 MUNICH
TEL: 89 22 30 05
FAX: 89 22 47 53
E-mail : munich@casalonga.com
http://www.casalonga.com

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EUROPEAN PATENT OFFICE

80298 MUNICH

O/ Ref: E 04079 SCHER

Munich, 1 September 2004

International Patent Application PCT/IB2003/003140
In the name of SCHERING-PLOUGH LTD.

Dear Sir/Madam,

We hereby inform you that there is a change of address for the applicant SCHERING-PLOUGH LTD. in connection with the above-referenced application.

The new address is: SCHERING PLOUGH LTD.
Weystrasse 20
CH- 6000
Lucerne 6
SWITZERLAND

Thank you for your attention in this matter.

Yours very truly,

Axel CASALONGA
European Patent Attorney

Conseils en Propriété Industrielle (CPI)
European Patent Attorneys (EPA)
European Trademark Attorneys (ETMA)

Axel CASALONGA (CPI-EPA-ETMA)
Gérard DOSSMANN (CPI-EPA-ETMA)

[illegible] (CPI-EPA-ETMA)
[illegible] (CPI-ETMA)
[illegible] (CPI-EPA-ETMA)
[illegible] (CPI-ETMA)
Philippe Conte (CPI-ETMA)
[illegible] (CPI-EPA)
Caroline Casalonga (CPI-ETMA)
[illegible] (CPI-EPA)
[illegible] (CPI-ETMA)
[illegible] (EPA)
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* [illegible] Alicante
** admitted New York Bar

BUREAU DE PARIS
8, AVENUE PERCIER
F - 75008 PARIS
TEL: 33 (0)1 45 61 94 64
FAX: 33 (0)1 45 63 94 21

BUREAU DE MUNICH
PAUL-HEYSE-STRASSE 33
D - 80336 MUNICH
TEL: 49 (0) 89 22 30 05
FAX: 49 (0) 89 22 47 53

BUREAU D'ALICANTE
AVENIDA MAISONNAVE, 41
E - 03003 ALICANTE
TEL: 34 96 513 17 95
FAX: 34 96 513 16 89

BUREAU DE GRENOBLE
7, CHEMIN DES PRÉS ZIRST 4403
F - 38944 MEYLAN CEDEX
TEL: 33 (0)4 76 90 22 25
FAX: 33 (0)4 76 90 19 96

(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
25 September 2003 (25.09.2003)

PCT

(10) International Publication Number
WO 2003/077828 A3

(51) International Patent Classification[7]: C07C 233/18, A61K 31/165, A61P 31/04

(21) International Application Number: PCT/IB2003/003140

(22) International Filing Date: 7 March 2003 (07.03.2003)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:
10/094,688 8 March 2002 (08.03.2002) US

(71) Applicant: SCHERING-PLOUGH, LTD. [CH/CH]; Toepferstrasse 5, CH-6000 Lucerne 6 (CH).

(72) Inventors: BOOJAMRA, Constantine, G.; 1068 Ashbury Street, San Francisco, CA 94117 (US). CHONG, Lee, S.; 37469 Marsten Drive, Newark, Ca 94560 (US). HECKER, Scott, J.; 16387 Englewood Avenue, Los Gatos, CA 95032 (US). GLINKA, Tomasz, W.; 21122 Patriot Way, Cupertino, CA 95014 (US). SCHUSTER, Dale, E.; 267 Beechspring Road, South Orange, NJ 07079 (US).

(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, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, TJ, TM, TN, TR, TT, TZ, UA, UG, UZ, VC, VN, YU, ZA, ZM, ZW.

(84) Designated States *(regional)*: ARIPO patent (GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG).

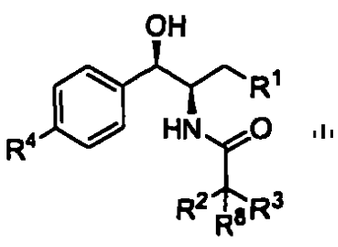
Published:
— *with international search report*
— *before the expiration of the time limit for amending the claims and to be republished in the event of receipt of amendments*

(88) Date of publication of the International search report:
8 January 2004

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.

WO 2003/077828 A3

(54) Title: NOVEL FLORFENICOL-TYPE ANTIBIOTICS



(I)

(57) Abstract: The present invention relates to novel florfenicol compounds having the chemical structure (I) wherein the compounds are useful for the treatment and/or prevention of bacterial infections in a broad range of patients such as, without limitation, birds, fish, shellfish and mammals.

INTERNATIONAL SEARCH REPORT

Internatic pplication No
PCT/IB 03/03140

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C07C233/18 A61K31/165 A61P31/04

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 C07C A61K

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

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No
X	FR 4 604 M (WARNER-LAMBERT PHARMACEUTICAL COMPANY) 21 November 1966 (1966-11-21) example 1	1-25
X	BE 669 982 A (WARNER-LAMBERT PHARMACEUTICAL COMPANY) 22 March 1966 (1966-03-22) example 13	1-25
X	BE 638 755 A (WARNER-LAMBERT PHARMACEUTICAL COMPANY) 16 April 1964 (1964-04-16) page 4, line 13	1-25

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[X] Further documents are listed in the continuation of box C

[X] 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	Date of mailing of the international search report
3 November 2003	11/11/2003

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Tx. 31 651 epo nl, Fax: (+31-70) 340-3016	Janus, S

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

Internat Application No
PCT/IB 03/03140

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No
X	NIELSEN P.E. ET AL.: "Light-Sensitive Chloramphenicol Analogs" ACTA CHEMICA SCANDINAVICA B, vol. B29, no. 6, 1975, pages 662-666, XP009012343 example V	1-25
X	HANSCH, C. ET AL.: "Structure-Activity Relation of Chloramphenicols" JOURNAL OF MEDICINAL CHEMISTRY, vol. 16, no. 8, 1973, pages 917-922, XP001152917 examples 21,32,34; table I	1-25
X	MITSCHER, L.A. ET AL.: "Circular Dichroism of Aryl Diastereoisomers. 3. Cupra A Spectra of Chloramphenicol Derivatives" JOURNAL OF MEDICINAL CHEMISTRY, vol. 16, no. 2, 1973, pages 98-101, XP001152918 examples XIII,XVI,XVII,XXV	1-25
X	VON STRANDTMANN M. ET AL.: "Synthesis of p-Acyl Analogs of Chloramphenicol and their Antimicrobial Properties" JOURNAL OF MEDICINAL CHEMISTRY, vol. 10, no. 5, 1967, pages 888-890, XP001152919 examples I,IA,IB,II,IA,III,IIIA,IIIB,IIIC,IV,IVA; table I	1-25

INTERNATIONAL SEARCH REPORT

International application No.
PCT/IB 03/03140

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. [X] Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

Although claims 23-25 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound.

2. [ ] Claims Nos.:
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:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(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/IB 03/03140

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
FR 4604 M		US 3294818 A	27-12-1966
		GB 1033270 A	22-06-1966
BE 669982 A	22-03-1966	CH 465638 A	30-11-1968
		CH 465644 A	30-11-1968
		FR 1457622 A	24-01-1966
		GB 1117617 A	19-06-1968
		GB 1117616 A	19-06-1968
		SE 322207 B	06-04-1970
BE 638755 A		US 3183265 A	11-05-1965
		CH 455838 A	15-05-1968
		CH 460812 A	15-08-1968
		CH 437354 A	15-06-1967
		DE 1518439 A1	09-03-1972
		DK 115265 B	22-09-1969
		GB 1065125 A	12-04-1967
		GB 1065124 A	12-04-1967
		GB 1065123 A	12-04-1967
		SE 315876 B	13-10-1969
		SE 322237 B	06-04-1970
		SE 301470 B	10-06-1968

30 233

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SUPERINDUSTRIA Y COMERCIO
Radicacion : 04006857 00000000 Folios: 232
Fecha (AMD): 2004-09-08 17:42:24
Tramite : 811 PCT-PATENT 379 FASE NACI 411 PRESENTA
Dependencia: 0020 DIVISION DE NUEVAS CREACIONES

REQUISITOS NECESARIOS PARA PRESENTACION Y AGILIZACION DEL TRAMITE

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

Fecha:

234
235

Industria y Comercio
SUPERINTENDENCIA

2020
Bogotá D C

Doctor(a)
CARLOS R. OLARTE
Apoderado(a)
SCHERING – PLOUGH, LTD

Oficio N°068859

ASUNTO:

Radicación N°	04-88857
Solicitud internacional	PCT/IB03/03140
Fecha inicio fase nacional	08/10/2004
Trámite	011
Evento	379
Actuación	430
Folios	001

Como resultado del examen de forma, realizado con fundamento en el artículo 38 de la Decisión 486 de la Comisión de la Comunidad Andina, artículo 27 del Tratado de Cooperación en Materia de Patentes (PCT), regla 51 bis del Reglamento y Circular Única de la Superintendencia de Industria y Comercio, se le comunica que:

- El solicitante debe allegar la traducción completa de la solicitud Internacional, dado que los folios 47 a 117 fueron presentados en idioma extranjero.

En cumplimiento de lo dispuesto por el artículo 39 de la Decisión 486, se le requiere para que dentro del término de dos meses siguientes a la fecha de notificación del presente oficio, complete los requisitos anteriormente enunciados. En caso de no dar cumplimiento al presente requerimiento, la solicitud se considerará abandonada y perderá su prelación.

Notifíquese el presente oficio conforme a lo dispuesto en el numeral 5.2, literal e), del capítulo quinto del título primero de la Circular Única N° 10 de 2001.

ALIX CARMENZA CÉSPEDES DE VERGEL
Jefe de la División de Nuevas Creaciones.

El anterior requerimiento fue notificado por fijación en lista de fecha,

24 SET. 2004

202044

Sede Centro: Carrera 13 No. 27-00 Pisos 2, 5, 7 y 10
Conmutador: 3 820840 Fax: 3 505220
Sede CAN: Tr. 40A No. 38-50 3153265 al 69
Fax: 3 505220. Línea 9800-910 165
Web: www.sic.gov.co e-mail: info@sic.gov.co
Bogotá D.C. - Colombia