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JUNE 2, 2023

THE CONTROLLER OF PATENT
THE PATENT OFFICE
NEW DELHI

Re: Opposition u/s 25(2) of the Patent act – By LOW COST STANDARD THERAPEUTICS against Indian Patent No. 397784 (Formerly Indian Patent Application No. 201817001590) dated 29/06/2016
Patentee**:** GILEAD SCIENCES INC.

Respected Sir,

We submit herewith Post-Grant Opposition under Section 25(2) of the Patent Act, 2005 along with evidence and Form 7.

We crave leave of the Controller to submit additional documents or evidence, if necessary to support any averments in the representation as may be necessitated in the proceeding.

The Controller is requested to take the documents on record and proceed further in the matter and keep the Petitioner advised of each and every step taken in the matter.

Lastly, we request the Controller to grant us an opportunity of being heard before the above Opposition is finally decided.

Thanking you,

Yours faithfully,

PRAGYA SINGH THAKUR (IN /PA – 3329)
OF RAJESHWARI AND ASSOCIATES
AGENT FOR THE OPPONENT

Encl.: As stated

C.C.: K&S PARTNERS (Email: ipo@knspartners.com);

BEFORE THE CONTROLLER OF PATENTS, NEW DELHI

IN THE MATTER OF:

The Patents Act, 1970 as amended by the Patents (Amendment) Act 2005, and The Patents Rules, 2003, as amended by The Patents (Amendment) Rules, 2006

AND

IN THE MATTER of Post-Grant Opposition under Section 25(2)

AND

IN THE MATTER of Indian Patent No. 397784

(Formerly Indian Patent Application No. 201817001590)

IN THE MATTER OF:

LOW COST STANDARD THERAPEUTICS .....OPPONENT

VS.

GILEAD SCIENCES INC. .....PATENTEE

POST-GRANT OPPOSITION BY LOW COST STANDARD THERAPEUTICS

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Dated this 02nd day of June, 2023

PRAGYA SINGH THAKUR (IN /PA – 3329)
OF RAJESHWARI AND ASSOCIATES
AGENT FOR THE OPPONENT

TO

THE CONTROLLER OF PATENTS

THE PATENT OFFICE, NEW DELHI

FORM 7
THE PATENTS ACT, 1970
(39 OF 1970)
AND
THE PATENTS RULES, 2003
NOTICE OF OPPOSITION
[See Section 25(2) and rule 55A]

We, **LOW COST STANDARD THERAPEUTICS,** an Indian Company of I Floor, Premananda Sahitya Bhavan, Opposite Lakadipul, Dandia Bazar, Vadodara, 390 001, Gujarat, India, hereby give Notice of opposition to the grant of patent in respect of Indian Patent No. 397784 (Formerly Indian Patent Application No. 201817001590) dated 29/06/2016 made by **GILEAD SCIENCES INC.** on the grounds:

(a) Section 25(2)(b): Lack of novelty
(b) Section 25(2)(e): Lack of inventive step
(c) Section 25(2)(f): Invention is not patentable under section 3(d) and 3(e)
(d) Section 25(2)(d): Invention is publicly known before the priority date.
(e) Section 25(2)(g): The complete specification does not sufficiently and clearly describe the invention or the method by which it is to be performed.
(f) Section 25(2)(h): Failure to disclose the information required by section 8 of the Patents Act.

(Detailed grounds are set out in the Opposition)

Our address for service in India is:

RAJESHWARI & ASSOCIATES
S – 357, FIRST FLOOR
NEAR HDFC BANK
PANCHSHEEL PARK
NEW DELHI – 110017, INDIA
Tel: + 91-11-41038911
Mobile No. 8368982401
Email: pragya@ralegal.co.in;

Dated this 02nd day of June, 2023

PRAGYA SINGH THAKUR (IN /PA – 3329)
OF RAJESHWARI AND ASSOCIATES
AGENT FOR THE OPPONENT

TO,
THE CONTROLLER OF PATENTS
THE PATENT OFFICE, NEW DELHI

BEFORE THE CONTROLLER OF PATENTS, THE PATENT OFFICE, DELHI

In the matter of Section 25(2) of The Patents Act,1970 as amended by The Patents (Amendment) Act 2005;

And

In the matter of Rule 55 of The Patents Rules 2003 as amended by the Patent (Amendment) Rules, 2006

And

In the matter of Indian Patent number 397784 (formerly Indian Patent Application No.: 201817001590), dated 29/06/2016, in the name of **GILEAD SCIENCES INC.**

NOTICE OF OPPOSITION BY:

LOW COST STANDARD THERAPEUTICS .....OPPONENT

VS.

GILEAD SCIENCES INC. .....APPLICANT

NOTICE OF POST-GRANT OPPOSITION UNDER SECTION 25(2) OF THE PATENTS ACT, 1970

STATEMENT OF CASE OF OPPONENT

The Opponent herein respectfully submits as under:

1. A Post-grant opposition under Section 25(2) of the Patents Act, 1970, is being submitted by the Opponent against Indian Patent No. 397784 (hereinafter referred to as the "impugned patent") which has been granted in favour of GILEAD SCIENCES INC. (Hereinafter referred to as the "Applicant/Patentee").

2. The Opponent, Low Cost Standard Therapeutics (hereinafter LOCOST) is a not-for profit organisation having its office at I Floor, Premananda Sahitya Bhavan, Opposite Lakadipul, Dandia Bazar, Vadodara, 390 001, Gujarat, India. LOCOST was established

to make essential medicines of quality at reduced costs by eliminating the margins shared with prescribers, distributers and related market costs. LOCOST makes essential medicines for those working with urban and rural poor in India. The goal of LOCOST is to provide good quality medicines at affordable prices for those working in remote areas.

3. Registered as Low-Cost Standard Therapeutics in 1983, it is a public charitable trust that is concerned with public health aspects of medicines: their genesis, costs, clinical trials, licensing, patenting, production, distribution and consumption till it reaches the end users, namely patients. LOCOST is also concerned about the long-term impact of medicines – their use, misuse and overuse, adverse reactions and side effects - and the public regulation of all related issues.

4. LOCOST has its own well-equipped production facility in Vadodara where it produces more than 100 essential and rational medicines. The establishment makes more than 60 essential medicines in 80 formulations (liquids, capsules, tablets). LOW LOCOST is also active in pharmaceutical policy advocacy at regional and national levels. LOCOST products are preferred by many not-for-profit health institutions across India The sales are directed at institutions and individuals that work on a not-for profit basis or cater to the poorest of people.

5. Manufacturing at LOCOST conforms to the highest standards of production set by the Government of India. LOCOST's production facilities have Schedule M certification of the Government of India and anytime now it is expected to have WHO-GMP certification too. LOCOST's management practises ethical business norms that further assure our long-standing clients that no corners are cut on quality standards.

6. LOCOST realizes that mere production of essential medicines is not enough. LOCOST also engages in educating prescribers as well as users. Its education cell focuses on issues related to education and training for rational use of medicines. LOCOST publishes a monthly in Gujarati, namely *Apnu Swasthya*, and other publications for the general public including but not limited to the Gujarati version of classics such as *Where there is no Doctor* and *A Lay Person's Guide to Medicine* (a guide on the use and political economy of medicines).

7. The Opponent is also engaged in the manufacture and sale of various antiviral compounds. Additionally, the Opponent has been conducting research in respect of antiviral compounds. The existence of the impugned patent is against public interest and research in respect of anti-viral compounds. Thus, the Opponent is a person interested having direct and tangible interest in filing the present opposition.

8. The present opposition is against Indian patent no. 397784 which was originally filed as Indian Patent Application No. 201817001590 on 29/06/2016.The said application was granted on 26/05/2022 with patent number 397784 (hereinafter referred as the Impugned Patent). The Impugned patent is titled "PHARMACEUTICAL FORMULATIONS COMPRISING TENOFOVIR AND EMTRICITABINE". The priority date of the impugned patent is 30/06/2015.

9. The opponent by way of this presents post-grant opposition submits that the claims currently on record are not patentable under the provisions of the Patents Act. The claims currently on record are annexed herewith as Annexure-1 and are reproduced herein below for ready reference:

1. A tablet consisting of:
(a) 28 mg tenofovir alafenamide hemifumarate, and
(b) 200 mg emtricitabine as the only active ingredients, and
(c) one or more excipients;
wherein the tablet comprises 7% to 9% by weight tenofovir alafenamide hemifumarate, and
wherein the tablet is uncoated or coated by a polymeric film coating.

2. The tablet as claimed in claim 1 comprising 8% by weight tenofovir alafenamide hemifumarate and at least 55% by weight emtricitabine.

3. The tablet as claimed in any one of claims 1-2, wherein the one or more excipients comprise croscarmellose sodium, microcrystalline cellulose, and magnesium stearate.

4. The tablet as claimed in any one of claims 1-3, wherein the one or more excipients comprise 20-35 mg croscarmellose sodium, 70-120 mg microcrystalline cellulose and 1-7 mg magnesium stearate.

5. The tablet as claimed in claim 1, wherein the tablet consists of 200 mg emtricitabine, 28 mg tenofovir alafenamide hemifumarate, 28 mg croscarmellose sodium, 88.70 mg microcrystalline cellulose, 5.25 mg magnesium stearate, and a polymeric film coating.

6. The tablet as claimed in claim 1, wherein the tablet consists of 28 mg tenofovir alafenamide hemifumarate, 200 mg emtricitabine, croscarmellose sodium, microcrystalline cellulose, magnesium stearate, and a polymeric film coating.

7. The tablet as claimed in claim 1, wherein the tablet has a total weight of 350 mg ± 25 mg.

8. The tablet as claimed in claim 1 consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±3
Microcrystalline cellulose	89 ±9
Magnesium stearate	5.2 ±1.1.

9. The tablet as claimed in claim 1, consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±1.4
Microcrystalline cellulose	89 ±4
Magnesium stearate	5.2 ±0.5.

10. The tablet as claimed in claim 1, consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28
Microcrystalline cellulose	89
Magnesium stearate	5.3.

11. The tablet as claimed in claim 1, consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±3
Microcrystalline cellulose	89 ±9
Magnesium stearate	5.2 ±1.1

and a polymeric film coating.

12. The tablet as claimed in claim 1, consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±1.4
Microcrystalline cellulose	89 ±4
Magnesium stearate	5.2 ±0.5

and a polymeric film coating.

13. The tablet as claimed in claim 1, consisting of:

Ingredient	Mass (mg)
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28
Microcrystalline cellulose	89
Magnesium stearate	5.3

and a polymeric film coating.

14. A tablet consisting of:

(a) 11 mg tenofovir alafenamide hemifumarate, and

(b) 200 mg emtricitabine as the only active ingredients; and

(c) one or more excipients;

wherein the tablet comprises 3% to 4% by weight tenofovir alafenamide hemifumarate, and

wherein the tablet is uncoated or coated by a polymeric film coating.

15. The tablet as claimed in claim 14, wherein the tablet comprises between 100 mg and 150 mg of excipients.

16. The tablet as claimed in claim 14 or 15, wherein the tablet comprises 3% by weight tenofovir alafenamide hemifumarate.

17. The tablet as claimed in claim 14, wherein the total weight of the tablet is 360.5 mg.

SUMMARY OF GROUNDS FOR OPPOSITION:

10. The Opponent brings this opposition under the following grounds, amongst others, each of which is without prejudice to one another:

(a) Section 25(2)(b): Lack of novelty

(b) Section 25(2)(e): Lack of inventive step

(c) Section 25(2)(f): Invention is not patentable under section 3(d) and 3(e)

(d) Section 25(2)(d): Invention is publicly known before the priority date.

(e) Section 25(2)(g): The complete specification does not sufficiently and clearly describe the invention or the method by which it is to be performed.

(f) Section 25(2)(h): Failure to disclose the information required by section 8 of the Patents Act.

11. As per the Patentee, impugned invention relates to pharmaceutical formulations suitable for treating viral infections such as HIV, in particular solid oral dosage forms including emtricitabine and tenofovir alafenamide.

12. **PRIOR ARTS:** The opponent wishes to rely on the following prior arts as evidence in support of the grounds of opposition.

i. US 2015/0105350 published on 16 April 2015 **(Annexed herewith as Annexure 2)**
ii. WO2014184553 (WO'553) published on 20 November 2014 **(Annexed herewith as Annexure 3)**
iii. Tenofovir Alafenamide Vs. Tenofovir Disoproxil Fumarate in Single Tablet Regimens for Initial HIV-1 Therapy: A Randomized Phase 2 Study, Paul E. Sax, J Acquir Immune Defic Syndr 2014;67:52–58 **(Annexed herewith as Annexure 4)**
iv. Gilead press release entitled "*European Medicines Agency Validates Gilead's Marketing Application for Fixed-Dose Combination of Emtricitabine and Tenofovir Alafenamide for HIV Treatment*"; published on May 28, 2015 **(Annexed herewith as Annexure 5).**

GROUND 1: Section 25(2)(b) Lack of Novelty

9. It is submitted that the invention as claimed in the impugned patent lacks novelty in view of the disclosure published prior to the earliest priority date of the impugned patent i.e. prior to June 30, 2015.

10. It is submitted that claims 1-17 of the impugned patent lack novelty in view of the disclosure of US 2015/0105350.

11. The Opponent submits that US 2015/0105350 (US'350) i.e. Annexure 2, which was published on 05 August 2004, discloses the alleged invention claimed in the impugned patent in its entirety.

12. US'350 discloses the use of the hemifumarate form of 9-(R)-2-(S)-(S)- 1-(isopropoxycarbonyl)ethylaminophenoxyphosphinyl methoxypropylladenine (tenofovir alafenamide hemifumarate, TAF, GS-7340) in combination with other drugs including emtricitabine.

13. It is disclosed in the document US350 that tenofovir alafenamide hemifumarate can be formulated as a unit dosage form by combining tenofovir alafenamide hemifumarate with emtricitabine in which emtricitabine is in dose of 200mg and tenofovir alafenamide fumarate is in the dose of 25 mg or 10 mg which is equivalent to tenofovir alafenamide hemifumarate 28mg or 11mg, respectively.

14. It is also disclosed in US350 that the unit dosage form can be a tablet comprising microcrystalline cellulose, croscarmellose sodium and magnesium stearate with a polymeric coating.

15. The document also discloses that though the drugs are used in their standard known doses, as aforementioned, it is preferred to have the amount of tenofovir alafenamide in 3±1mg weight % or 8±2mg weight % in the composition.

16. In view of the above, independent claim 1 (and claims 2-13 that depend from it) as well as independent claim 11 (and claims 15-17 that depend from it) lack novelty in view of US'350 and ought to be rejected on this ground alone.

GROUND 2: Section 25(2)(e) Lack of Inventive step

17. It is submitted that the invention as claimed is obvious and does not involve any inventive step in view of the disclosures published prior to the earliest priority date of the impugned patent i.e. prior to 30/06/2015.

18. It is submitted that claims 1-17 of the impugned patent lack inventive step and are obvious in view of common general knowledge in art and combined teachings of the cited prior arts.

19. US 2015/0105350 (hereinafter referred to as US'350), published on Apr. 16, 2015 discloses The use of the hemifumarate form of 9-(R)-2-(S)-(S)- 1-(isopropoxycarbonyl)ethylaminophenoxyphosphinyl methoxypropylladenine (also known

as tenofovir alafenamide hemifumarate or TAF or GS-7340) in combination with other drugs including emtricitabine. The document teaches that can be used in the dose of 8mg, 25mg, 40 mg as shown in Figure 1 and 2 of the document US350. Further, as seen in Paragraph [0157] to [0162]) and in Figure 4A and 4b, TAF can be combined with Emtricitabine in the dose of TAF 25mg + Emtricitabine 200mg.

20. In para 0290 it is disclosed that GS-7340, or a pharmaceutically acceptable salt thereof can be used in the dose of 10 mg.

21. In para 0015, 0219 to 220, US'350 discloses a composition comprising a unit-dosage form of GS-7340 or a pharmaceutically acceptable salt thereof, said composition includes GS-7340 or a pharmaceutically acceptable salt thereof in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg of the composition.

22. Para 0088 of the document discloses that TAF and emtricitabine can be formulated into a unit dosage which may also include other drugs.

23. The percentage of tenofovir alafenamide hemifumarate in the compositions and preparations may, of course, be varied and may conveniently be between about 2% to about 60% or more of the weight of a given unit dosage form [para 248]. Para 0232 a tenofovir alafenamide hemifumarate composition comprises less than about 5%, preferably less than 1%, and more preferably less than 0.5% by weight of tenofovir alafenamide monofumarate.

24. Para 0250 discloses that Tenofovir alafenamide hemifumarate is preferably administered as part of a pharmaceutical composition, together with one or more pharmaceutically acceptable carriers/excipients, and optionally other therapeutic ingredients. Such excipients includes substances that can serve as a vehicle or medium for tenofovir alafenamide hemifumarate (e.g., a diluent carrier). Para 0251 discloses that the composition could be a polymer coated tablet composition and the excipients could be filler (diluents): micro crystalline cellulose, a disintegrating agent: croscarmellose sodium, a lubricant: magnesium stearate.

25. In para 0293 it is disclosed that one of skill in the art will know that, in the case of administering a pharmaceutically acceptable salt or complex of an agent, the amount administered will be adjusted relative to the weight of the component added to produce the salt or complex. Therefore, the dose of 10 mg and 25mg tenofovir alafenamide comes to be 11mg and 28mg of tenofovir alafenamide hemifumarate by following caluculation.

26. A simple calculation shows that an amount of Tenofovir alafenamide hemifumarate incorporating 28 mg of the molecule is equivalent to an amount of Tenofovir alafenamide incorporating 25 mg of said molecule. The calculation is as follows:
    - ❖ Molecular weight of Tenofovir alafenamide hemifumarate = 1069
    - ❖ Number of units of Tenofovir alafenamide per molecule of Tenofovir alafenamide hemifumarate = 02
    - ❖ Molecular weight of Tenofovir alafenamide = 476.5
    - ❖ Molecular weight of fumaric acid = 116.07
    - ❖ Tenofovir alafenamide hemifumarate molecular weight is therefore 476.5 + 476.5 + 116.07 = 1069.07
    - ❖ Number of equivalents of tenofovir alafenamide hemifumarate = 28/1069 = 0.026192
    - ❖ We already know that tenofovir alafenamide hemifumarate contains two molecules of tenofovir alafenamide
    - ❖ Thus, no. of equivalents for tenofovir alafenamide = 0.026192 × 2 = 0.052384
    - ❖ Therefore, the weight of tenofovir alafenamide = 0.052384 × 476.5 = 24.960976≈ 25mg

27. A calculation shows that an amount of Tenofovir alafenamide hemifumarate incorporating 11 mg of the molecule is equivalent to an amount of Tenofovir alafenamide incorporating 10 mg of said molecule. The calculation is as follows:
    - ❖ Molecular weight of Tenofovir alafenamide hemifumarate = 1069
    - ❖ Number of units of Tenofovir alafenamide per molecule of Tenofovir alafenamide hemifumarate = 02
    - ❖ Molecular weight of Tenofovir alafenamide = 476.5
    - ❖ Molecular weight of fumaric acid = 116.07
    - ❖ Tenofovir alafenamide hemifumarate molecular weight is therefore 476.5 + 476.5 + 116.07 = 1069.07

- ❖ Number of equivalents of tenofovir alafenamide hemifumarate = 11/1069 = 0.010289
- ❖ We already know that tenofovir alafenamide hemifumarate contains two molecules of tenofovir alafenamide
- ❖ Thus, no. of equivalents for tenofovir alafenamide = 0.010289 × 2 = 0.020579
- ❖ Therefore, the weight of tenofovir alafenamide = 0.020579 × 476.5 = 9.80589 ≈ 10mg

28. In para 0219 and 0221 the document discloses that the preferred weight of tenofovir alafenamide hemifumarate in a unit composition is 3 mg, 3±2 mg, or 3±1 mg and 8±2 mg of GS-7340, or a pharmaceutically accept able salt thereof. Therefore, it is taught that it is best to restrict the weight of tenofovir alafenamide hemifumarate in a tablet to 3 mg, 3±2 mg, or 3±1 mg and 8±2 mg of the tablet.

29. Thus, a person skilled in the art preparing a tablet composition of tenofovir alafenamide whether as a singular drug or in combination with other drug(s) is motivated to prepare a tablet containing 3±1 and 8±2% of tenofovir alafenamide hemifumarate in a tablet since even though US'350 as well as Sax et al teach that the dose of tenofovir to be used in combination is 25mg and 10mg i.e. 28mg and 11mg, respectively of tenofovir alafenamide hemifumarate, US'350 discloses preferred amount of tenofovir alafenamide hemifumarate to be present in a unit dosage form i.e. tablet to be 3±1mg and 8±2 mg i.e. 3±1 and 8±2 by weight of tenofovir alafenamide hemifumarate in a tablet.

30. WO2014184553 discloses pharmaceutical antiretroviral combination compositions for HIV treatment. Two of the specifically exemplified formulations of the document namely composition of Example 1 and 2 comprise of a combination of tenofovir in the form of tenofovir disoproxil fumarate and Emtricitabine. The compositions of tenofovir and emtricitabine are tablet compositions comprising microcrystalline cellulose, croscarmellose sodium and have a polymer coating.

31. A publication by Sax et al discloses that in a 10-day monotherapy study in HIV-1–positive patients, those who received 25 mg of TAF had an approximately 0.5 log10 greater decline in plasma HIV-1 RNA than did patients who received the standard 300-mg dose of TDF (tenofovir disoproxil fumarate). In vitro, higher intracellular TFV-DP

levels enable TAF to retain activity against viruses that have reduced susceptibility to TDF suggesting the potential use of TAF in a broader range of patients.

32. Sax et al also state that to confirm the antiviral activity and safety profile of TAF compared with that of TDF, a randomized, double-blind Phase 2 clinical trial of 2 STRs— elvitegravir 150 mg, cobicistat 150 mg, emtricitabine 200 mg, and TAF 10 mg (E/C/F/TAF) compared with elvitegravir 150 mg, cobicistat 150 mg, emtricitabine 200 mg, and TDF 300 mg (E/C/F/TDF), licensed as Stribild (Gilead Sciences, Foster City, CA) were conducted where the combination containing TAF fared better than the combination containing TDF.

33. A person skilled in the art gets the teaching from Sax et al. and WO553 that it would be better to replace tenofovir disoproxil fumarate with tenofovir alafenamide fumarate in a combination of tenofovir + emtricitabine and that a composition comprising of said combination would be effective as anti-HIV. Sax also teaches that 300 mg tenofovir disoproxil fumarate can be replaced with 10 or 25 mg tenofovir alafenamide fumarate. In addition, US350 teaches that such a combination is stable when the weight of TAF comprising the composition is 3±1mg or 8±2 mg of the composition.

34. It is also well within the purview of a person skilled in the art that 28 mg of tenofovir alafenamide hemifumarate is simply equivalent to 25 mg of tenofovir alafenamide. Thus, the dosage of tenofovir alafenamide hemifumarate as claimed in claim 1 (and dependent claims 6, 8, 9, 10, 11, 12 and 13), which is 28 mg, is necessarily equivalent to 25 mg of tenofovir alafenamide.

35. It is evident from the above documents that the combination of tenofovir alafenamide (GS-7340) and emtricitabine is both chemically stable and synergistic and/or reduces the side effects of one or both of GS-7340 and emtricitabine.

36. Moreover, the documents discussed above teach that the combination of tenofovir alafenamide hemifumarate and emtricitabine can be formulated as a polymer coated tablet comprising of excipients microcrystalline cellulose, croscarmellose, magnesium stearate, and that it is most preferred to have tenofovir alafenamide hemifumarate in the unit dosage form like tablet in 3±1 mg or 8±mg weight of the composition (tablet) whereas the

dose of tenofovir alafenamide hemifumarate would be 11mg or 28 mg equivalent to 10mg or 25mg of tenofovir.

37. Thus, the Applicant has simply followed the teachings of the prior art and has observed minor differences in the lower limit of the amount of tenofovir alafenamide to be used in the tablet composition which has been projected in the impugned specification to be a technical contribution in the field whereas the purported "contribution" is nothing but a observation projected as a superficial advantage.

38. Opponent submits that the Applicant, in its written submission pursuant to hearing under section 14 (submitted on April 29, 2022), has put forth the following objective of the alleged invention of impugned patent:
"*The present invention relates to a tablet which aims to provide stable formulations containing tenofovir alafenamide hemifumarate and emtricitabine*." (internal page 2, written submission)

39. However, it can be seen that US'350 already provides solution to the problem of stability resulting from combined formulation of tenofovir alafenamide and emtricitabine in a single dosage form including tablet since it provides for a unit dosage form in tenofovir alafenamide and emtricitabine can be combined together.

40. Furthermore, Sax et al. also discloses that a tablet containing both tenofovir alafenamide and emtricitabine was successfully used in clinical trials. The use in clinical trial can be done only when the formulation is stable and the degradation impurities are below or within the acceptable limits.

49. It is submitted that Gilead press release entitled "*European Medicines Agency Validates Gilead's Marketing Application for Fixed-Dose Combination of Emtricitabine and Tenofovir Alafenamide for HIV Treatment*", discloses (paragraph 1, page 1) the announcement for validation of Marketing Authorization Application for two doses of Gilead's investigational fixed-dose combination of emtricitabine and tenofovir alafenamide (200/10 mg and 200/25 mg) (F/TAF).

50. It is further disclosed (paragraph 4, page 1) that F/TAF is Gilead's second F/TAF-based regimen to be validated by the EMA. An MAA for an investigational once-daily single

tablet regimen containing elvitegravir 150 mg, cobicistat 150 mg, emtricitabine 200 mg and tenofovir alafenamide 10 mg (E/C/F/TAF) was fully validated on December 23, 2014. Thus, it is evident that the regimen utilizing the combination of F/TAF is separate from that of the earlier validated combination of E/C/F/TAF.

51. It is further disclosed (paragraph 5, page 1) that bioequivalence studies demonstrated that the formulation of the fixed-dose combinations of F/TAF achieved the same drug levels in the blood as in E/C/F/TAF.

52. Thus, it is evident that a fixed-dose combination of 200 mg emtricitabine with 10 mg of tenofovir alafenamide as well as a fixed-dose combination of 200 mg emtricitabine with 25 mg of tenofovir alafenamide was already known before the priority date of the impugned patent and it was within the purview of a person skilled in the art that 11 mg of tenofovir alafenamide hemifumarate is equivalent to 10 mg tenofovir alafenamide and 28mg tenofovir alafenamide hemifumarate is equivalent to 25 mg tenofovir alafenamide.

53. Thus, the subject matter claimed in impugned patent is obvious and lacks inventive step. In view of the above submissions, impugned patent should be rejected on this ground alone.

GROUND 3: Claims not patentable under Section 25(2)(f)

The claimed subject matter is not patentable under Section 3(d) of the Act

54. It is submitted that the impugned patent falls within the purview of section 3(d) of the Patents Act, 1970 which states that *"the mere discovery of a new form of a known substance which does not result in the enhancement of the known efficacy of that substance or the mere discovery of any new property or new use for a known substance or of the mere use of a known process, machine or apparatus unless such known process results in a new product or employs at least one new reactant.*

Explanation -For the purposes of this clause, salts, esters, ethers, polymorphs, metabolites, pure form, particle size, isomers, mixtures of isomers, complexes, combinations and other derivatives of known substance shall be considered to be the same substance, unless they differ significantly in properties with regard to efficacy."

55. It is submitted that the claims 1-17 of the impugned patent are all directed towards a fixed-dose combination of emtricitabine and tenofovir alafenamide fumarate salt (hemifumarate form). As previously elucidated, the fixed-dose combination of tenofovir alafenamide and emtricitabine, in tablet form, was already disclosed in Annexure 2. The claimed subject matter is therefore nothing but a new form of the aforementioned known substance already known in Annexure 2. The complete specification of the impugned patent does not demonstrate any enhanced therapeutic efficacy data of the claimed compositions over those already disclosed in the closest prior art.

56. Moreover, the Applicant in its written submission pursuant to the hearing u/s 14 has contended that:

"*The claimed invention addressed the problem of degradation and formulated tablets containing tenofovir alafenamide hemifumarate and emtricitabine that are intended to be pharmaceutically acceptable (i.e., pharmacologically efficacious and physically acceptable)*"

Thus, by the Applicant's own admission, the inventive merit of impugned patent lies in solving the problem of degradation of tablets containing emtricitabine and tenofovir alafenamide. Nowhere does the complete specification demonstrate any kind of biological activity data comparing the compositions of the invention with those of the prior art. Thus, impugned patent miserably fails to demonstrate any kind of enhanced therapeutic efficacy data against the closest prior art.

57. Therefore, it is humbly submitted that in light of the above, present impugned patent falls squarely within the purview of Section 3(d) of the Patents Act 1970 and ought to be rejected.

The claimed subject matter is not patentable under Section 3(e) of the Act

58. It is submitted that the impugned patent falls within the purview of section 3(e) of the Patents Act, 1970 which states that "*a substance obtained by a mere admixture resulting only in the aggregation of the properties of the components thereof or a process for producing such substance*".

59. It is submitted that the claims (1-17) of impugned patent are directed towards fixed-dose tablet compositions of emtricitabine and tenofovir alafenamide hemifumarate. The complete specification of the impugned patent is devoid of any data demonstrating synergistic effect of compositions claimed in the impugned patent. Nowhere does the complete specification demonstrate an effect of the claimed compositions which is more than additive effect of both active ingredients formulated along with inactive excipients. The claimed compositions, therefore, are nothing but mere admixtures resulting only in the aggregation of the properties of the active and inactive components.

60. It is humbly submitted that, in light of the above reasons, the present impugned patent falls squarely within the purview of Section 3(e) of the Patents Act 1970 and ought to be rejected.

GROUND 4: THE INVENTION WAS PUBLICLY KNOWN BEFORE THE PRIORITY DATE

61. It is submitted that the claimed invention was publicly known before the priority date of the impugned patent.

62. It is submitted that Gilead press release entitled "*European Medicines Agency Validates Gilead's Marketing Application for Fixed-Dose Combination of Emtricitabine and Tenofovir Alafenamide for HIV Treatment*", discloses (paragraph 1, page 1) the announcement for validation of Marketing Authorization Application for two doses of Gilead's investigational fixed-dose combination of emtricitabine and tenofovir alafenamide (200/10 mg and 200/25 mg) (F/TAF).

63. It is further disclosed (paragraph 4 and 5, page 1) that F/TAF is Gilead's second F/TAF-based regimen to be validated by the EMA based on results of Phase III clinical trial and that Gilead has already obtained US FDA approval for F/TAF combination on April 07, 2015.

64. Thus, it is evident that a fixed-dose combination of 200 mg emtricitabine with 10 mg of tenofovir alafenamide as well as a fixed-dose combination of 200 mg emtricitabine with 25 mg of tenofovir alafenamide was already known and were in public domain before the priority date of the impugned patent.

65. It is submitted that once the composition of F/TAF i.e. a composition of emtricitabine 200mg and tenofovir alafenamide 10 or 25mg was in public domain, the knowledge of weight of tenofovir alafenamide in such a composition was a mere exercise of weighing said composition without use of any special equipment or exercise of any special methodology.

66. The tablets in above mentioned clinical trial were already presented to the members of public without any restriction on how to handle the tablet i.e. without putting any express restriction that no one can weigh the tablets being used in the trial. As discussed in earlier paragraphs under the Ground 2 Lack of Inventive Step standard dose of emtricitabine and tenofovir alafenamide were also public knowledge. Therefore, a simple act of weighing the tablet reveals the percentage of tenofovir alafenamide in the tablet unless the members of public were expressly restricted from doing such an act.

67. The Patent Act provides an exemption to this ground on basis of reasonable trial. However, the legislative intent for proving an exemption in case of reasonable trial pertains to providing an opportunity to an inventor to establish the working of his invention if such working cannot be performed without bring the invention in front of the public.

68. In present instance, a clinical trial is not necessary to establish the working of the claimed invention as the same can be established by in vitro or in vivo data generated in laboratory under secure conditions without access to public.

69. Therefore, a clinical trial of the claimed invention before priority date amounts to the invention being publicly known and the impugned patent ought to be rejected on this ground alone.

GROUND 5: INSUFFICIENCY OF DISCLOSURE

70. It is submitted that complete specification does not sufficiently and clearly describe the invention or the method by which it is to be performed.

71. It is submitted that it is a well settled law that the specification should clearly and fairly describe the invention and disclose the best mode of working the invention so that the

person skilled in the art could perform the invention without any undue efforts. Further, it is submitted that claims of impugned application are not fairly based on the specification and the complete specification does not fairly describe the invention and the method by which it is to be performed.

72. Claim1 and Claim 11 of the impugned patent recite that the % by weight of Tenofovir alafenamide hemifumarate in the claimed composition have to be within 3 to 4% by weight or 7 to 9%by weight of the tablet. However, the dose of the both the drugs is fixed in the composition as per their known therapeutically effective dose i.e. emtricitabine 200mg and tenofovir alafenamide hemifumarate 11 or 28 mg equivalent to tenofovir alafenamide 10mg or 25mg, respectively. Therefore, the percentage by weight of tenofovir has to be adjusted by varying the amount of excipients. But neither the claims nor the specification provide support of the many variations that can be done in the amount of the excipients. In absence of this information claim 1 and 11 are too broad, lack enablement in the specification and have comprise of non-working embodiments.

73. Absence of abovementioned information also implies that a person skilled in the art has to undergo undue experimentation to ascertain the precise variation in amount of the excipients to be used in the composition to arrive at the claimed invention.

74. It is submitted that the complete specification of impugned patent shows statistical comparison of emtricitabine and TAF pharmacokinetic parameters between F/TAF 200/25 mg (Tablet D) and E/C/F/TAF. The corresponding table (internal page 67) shows data for parameters AUC_{last} (h.ng/ml), AUC_{inf} (h.ng/ml) and C_{max} (ng/ml) for both TAF and emtricitabine. It can be seen from the aforementioned table that the test mean CV% for emtricitabine are actually lower than the reference mean CV % for all the parameters mentioned above.

75. Therefore, the impugned patent does not enable a person of average skill in the art to make or use the invention as claimed without undue experimentation. Consequently, the specification as filed does not provide enabling disclosure of the claims at hand. Considering above, impugned application does not sufficiently and clearly describe the invention. Therefore, the impugned application should be refused on this ground alone.

GROUND 6: INFORMATION RELATING TO CORRESPONDING APPLICATIONS UNDER SECTION 8 [SECTION 25(2)(h)]

76. It is submitted that the Patentee has filed number of corresponding patent applications in many countries outside India. However, the Form 3 details submitted by the Patentee do not disclose the details of all corresponding patent applications at Indian Patent Office. It is pertinent to note that many of these corresponding patent applications and the details of the proceedings have significant bearing on the prosecution of impugned patent and are instrumental to the Learned Controller in adjudicating the patentability of impugned invention.

77. The opponent submits that the Patentee has purposefully and with malafide intention refrained from disclosing details of all the corresponding patents filed outside India and the prosecution details of same to the Learned Controller and therefore, on this ground alone the patent patents should be rejected.

78. The opponents crave leave to file further submissions and evidence with respect to this ground.

CONCLUSION

79. In view of the above, the claims are not novel, inventive and not patentable and insufficient. The pre-grant opposition as filed may be allowed and the subject patent application may be refused.

HEARING REQUESTED

80. The Opponent hereby requests a hearing under section 25(2) of the Patents Act, 1970 (hereinafter referred to as “the Patents Act”) and Rule 55A of the Patents Rules (hereinafter referred to as “the Rules”).

P R A Y E R

In the fact and circumstances of the case, the Opponent prays as follows:

i. that the Ld. Controller take the present Opposition on record; that the Indian patent 397784 (formerly patent application number 201817001590), be revoked under Section 25(2) of the Patents (Amendment) Act, 2005;

ii. that the Opponent may be allowed to file further documents and evidence if necessary to support their averments;

iii. that the Opponent may be granted an opportunity of being heard in the matter before any final orders are passed;

iv. that the Opponent may be allowed to make further submissions in case the Patentee makes any amendments in the claims;

v. any other reliefs considering the facts and circumstances may be granted in favour of the Opponent in the interest of justice.

Dated this 02nd day of June, 2023

PRAGYA SINGH THAKUR (IN /PA – 3329)
OF RAJESHWARI AND ASSOCIATES
AGENT FOR THE OPPONENT

TO
THE CONTROLLER OF PATENTS
THE PATENT OFFICE, NEW DELHI

We Claim:

1. A tablet consisting of:
   (a) 28 mg tenofovir alafenamide hemifumarate, and
   (b) 200 mg emtricitabine as the only active ingredients, and
   (c) one or more excipients;
   wherein the tablet comprises 7% to 9% by weight tenofovir alafenamide hemifumarate, and
   wherein the tablet is uncoated or coated by a polymeric film coating.

2. The tablet as claimed in claim 1 comprising 8% by weight tenofovir alafenamide hemifumarate and at least 55% by weight emtricitabine.

3. The tablet as claimed in any one of claims 1-2, wherein the one or more excipients comprise croscarmellose sodium, microcrystalline cellulose, and magnesium stearate.

4. The tablet as claimed in any one of claims 1-3, wherein the one or more excipients comprise 20-35 mg croscarmellose sodium, 70-120 mg microcrystalline cellulose and 1-7 mg magnesium stearate.

5. The tablet as claimed in claim 1, wherein the tablet consists of 200 mg emtricitabine, 28 mg tenofovir alafenamide hemifumarate, 28 mg croscarmellose sodium, 88.70 mg microcrystalline cellulose, 5.25 mg magnesium stearate, and a polymeric film coating.

6. The tablet as claimed in claim 1, wherein the tablet consists of 28 mg tenofovir alafenamide hemifumarate, 200 mg emtricitabine, croscarmellose sodium, microcrystalline cellulose, magnesium stearate, and a polymeric film coating.

7. The tablet as claimed in claim 1, wherein the tablet has a total weight of 350 mg ± 25 mg.

8. The tablet as claimed in claim 1 consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±3
Microcrystalline cellulose	89 ±9
Magnesium stearate	5.2 ±1.1.

9. The tablet as claimed in claim 1, consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±1.4
Microcrystalline cellulose	89 ±4
Magnesium stearate	5.2 ±0.5.

10. The tablet as claimed in claim 1, consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28
Microcrystalline cellulose	89
Magnesium stearate	5.3.

11. The tablet as claimed in claim 1, consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±3
Microcrystalline cellulose	89 ±9
Magnesium stearate	5.2 ±1.1

and a polymeric film coating.

12. The tablet as claimed in claim 1, consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±1.4
Microcrystalline cellulose	89 ±4
Magnesium stearate	5.2 ±0.5

and a polymeric film coating.

13. The tablet as claimed in claim 1, consisting of:

Ingredient	**Mass (mg)**
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28
Microcrystalline cellulose	89
Magnesium stearate	5.3

and a polymeric film coating.

14. A tablet consisting of:

(a) 11 mg tenofovir alafenamide hemifumarate, and

(b) 200 mg emtricitabine as the only active ingredients; and

(c) one or more excipients;

wherein the tablet comprises 3% to 4% by weight tenofovir alafenamide hemifumarate, and

wherein the tablet is uncoated or coated by a polymeric film coating.

15. The tablet as claimed in claim 14, wherein the tablet comprises between 100 mg and 150 mg of excipients.

16. The tablet as claimed in claim 14 or 15, wherein the tablet comprises 3% by weight tenofovir alafenamide hemifumarate.

17. The tablet as claimed in claim 14, wherein the total weight of the tablet is 360.5 mg.

Dated this: 15th day of January, 2018

Digitally signed and filed through e-filing

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OF K&S PARTNERS
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IN/PA-2560

US 20150105350A1

(19) **United States**

(12) **Patent Application Publication**

Ramanathan

(10) **Pub. No.: US 2015/0105350 A1**

(43) **Pub. Date: Apr. 16, 2015**

(54) **COMBINATION THERAPY COMPRISING TENOFOVIR ALAFENAMIDE HEMIFUMARATE AND COBICISTAT FOR USE IN THE TREATMENT OF VIRAL INFECTIONS**

(71) Applicant: **Gilead Sciences, Inc.**, Foster City, CA (US)

(72) Inventor: **Srinivasan Ramanathan**, San Carlos, CA (US)

(73) Assignee: **Gilead Sciences, Inc.**, Foster City, CA (US)

(21) Appl. No.: **14/376,116**

(22) PCT Filed: **Feb. 1, 2013**

(86) PCT No.: **PCT/US13/24438**

§ 371 (c)(1),
(2) Date: **Dec. 12, 2014**

Related U.S. Application Data

(60) Provisional application No. 61/594,894, filed on Feb. 3, 2012, provisional application No. 61/618,411, filed on Mar. 30, 2012, provisional application No. 61/624,676, filed on Apr. 16, 2012, provisional application No. 61/692,392, filed on Aug. 23, 2012, provisional application No. 61/737,493, filed on Dec. 14, 2012.

Publication Classification

(51) **Int. Cl.**

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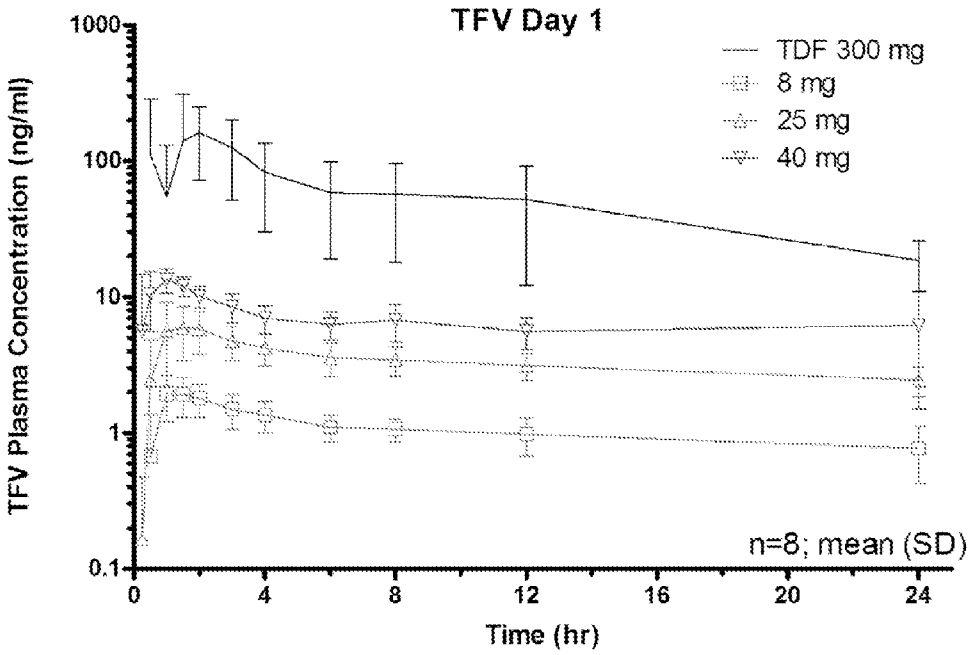
A61K 31/513	(2006.01)
A61K 31/47	(2006.01)
A61K 31/5377	(2006.01)

(52) **U.S. Cl.**

CPC ***A61K 31/675*** (2013.01); ***A61K 31/5377*** (2013.01); ***A61K 31/513*** (2013.01); ***A61K 31/47*** (2013.01)

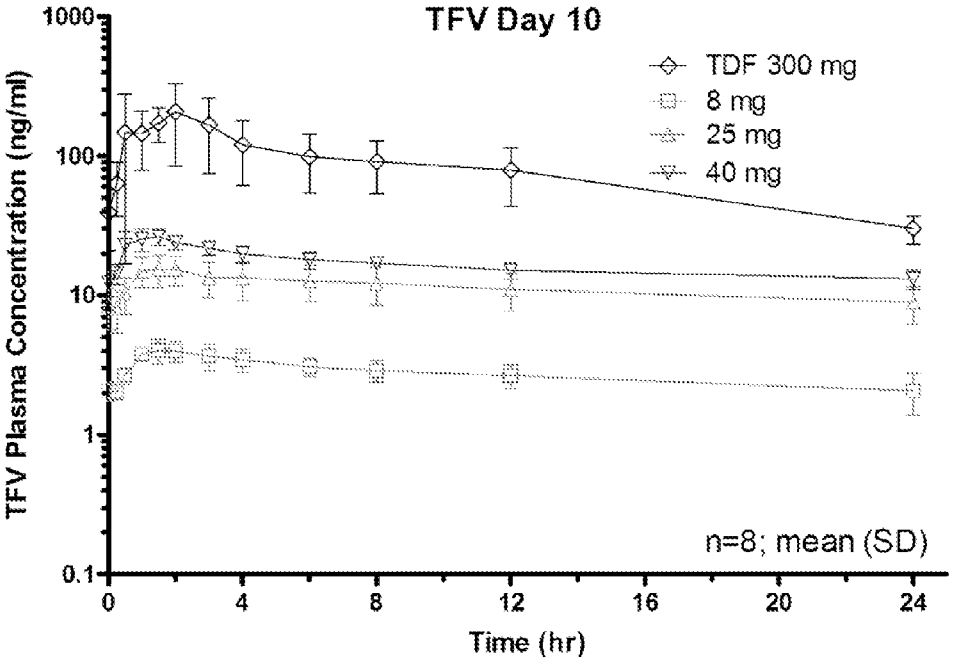
(57) **ABSTRACT**

The use of the hemifumarate form of {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine} (tenofovir alafenamide hemifumarate) in combination with cobicistat is disclosed. In addition, the combination of tenofovir alafenamide hemifumarate, cobicistat, emtricitabine, and elvitegravir, and the combination of tenofovir alafenamide hemifumarate, cobicistat, emtricitabine, and darunavir, are disclosed.



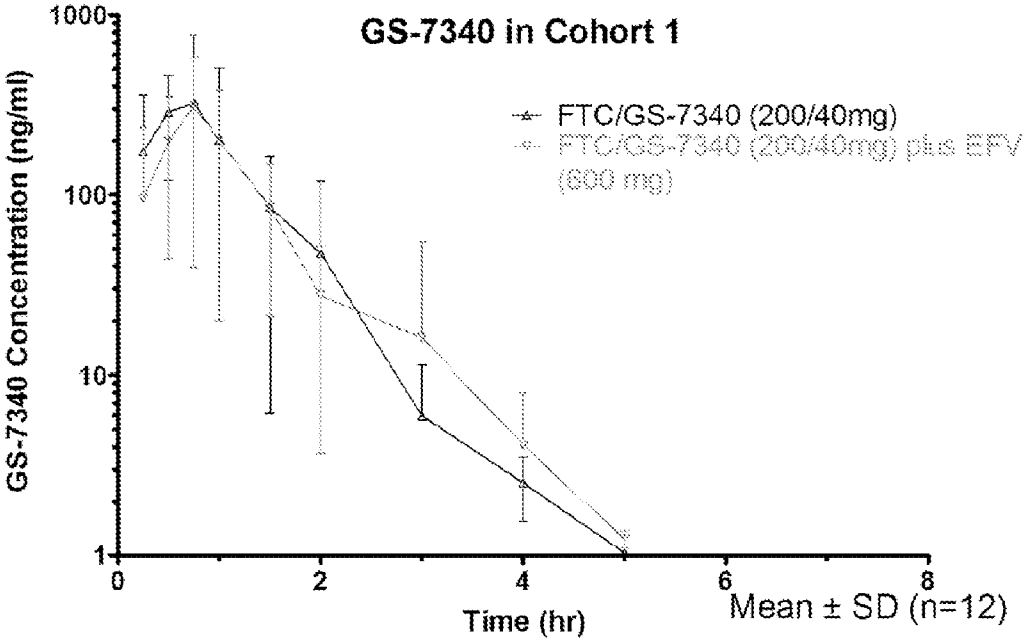
Day 1 PK Mean (%CV)	GS-7340 8 mg	GS-7340 25 mg	GS-7340 40 mg	TDF 300 mg
Cmax (ng/ml)	2 (30)	6.5 (40)	15 (36)	210 (52)
AUClast (ng.hr/ml)	25 (27)	70 (37)	143 (40)	1132 (48)

Figure 1



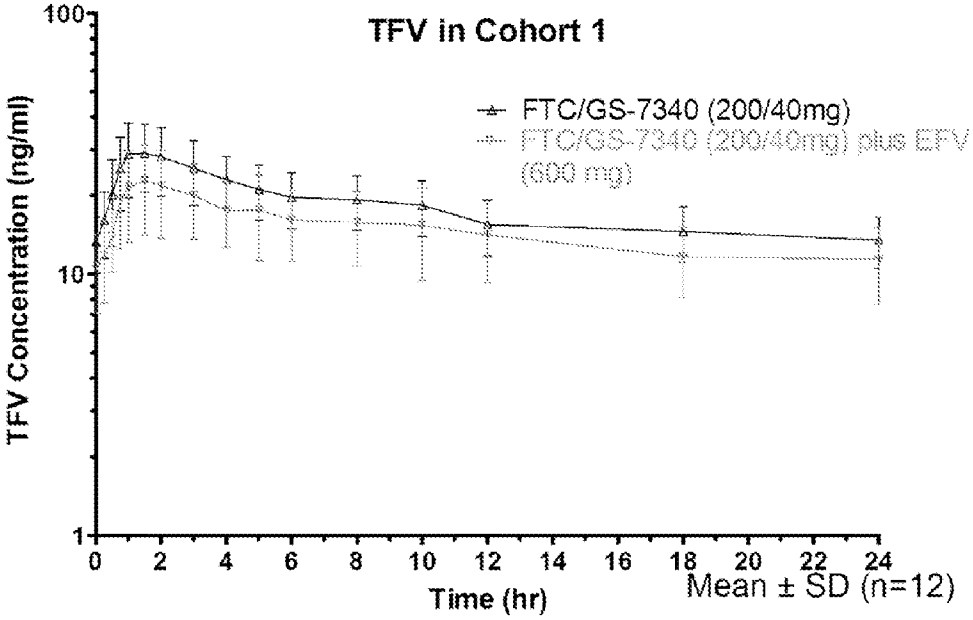
Day 10 PK Mean (%CV)	GS-7340 8 mg	GS-7340 25 mg	GS-7340 40 mg	TDF 300 mg
Cmax (ng/ml)	3.9 (28)	16 (22)	28 (9)	260 (43)
AUCtau (ng.hr/ml)	66 (19)	268 (29)	389 (11)	2090 (44)

Figure 2



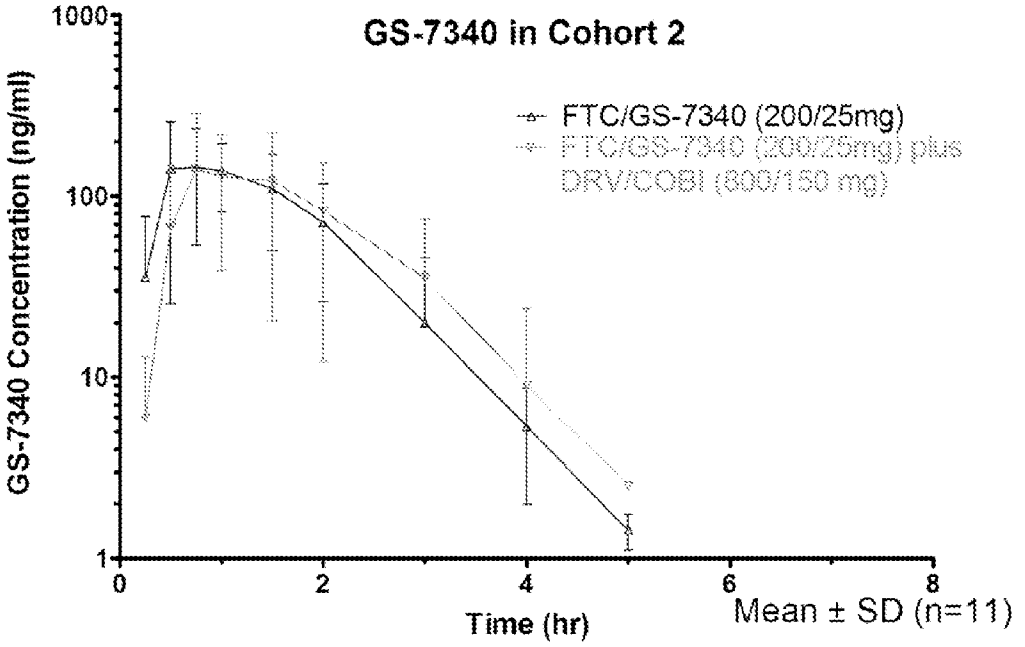
	GS-7340 PK in Cohort 1	
Mean (%CV)	FTC/GS-7340 (200/40 mg)	FTC/GS-7340 plus EFV 600 mg
AUC_{last} (ng.hr/ml)	330.5 (63)	285.5 (47)
C_{max} (ng/ml)	481.3 (83)	390.8 (62)

Figure 3A



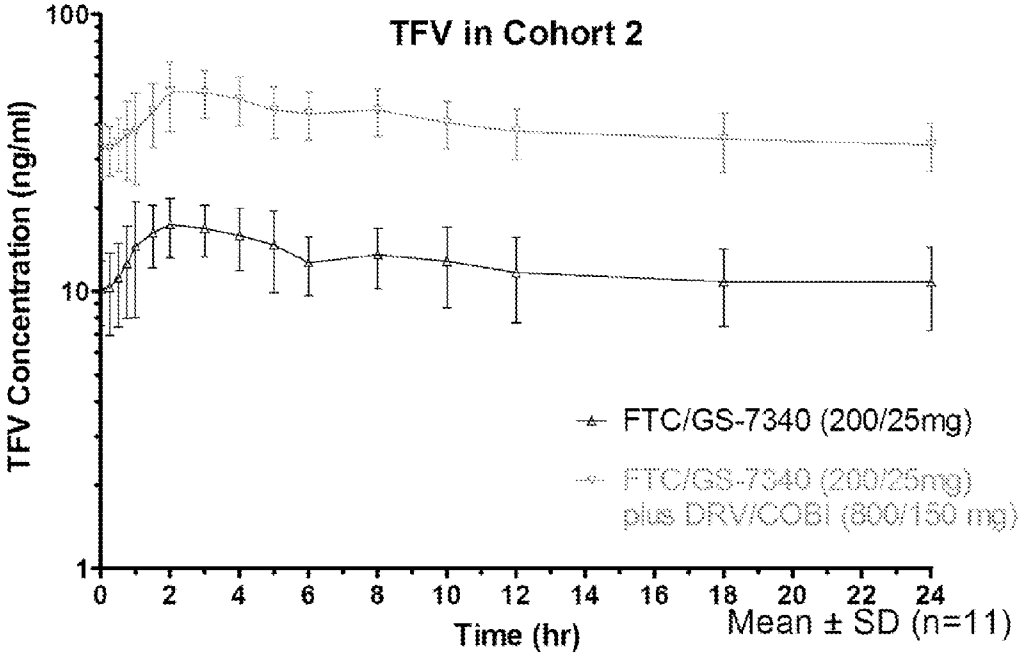
	TFV PK in Cohort 1	
Mean (%CV)	FTC/GS-7340 (200/40 mg)	FTC/GS-7340 plus EFV 600 mg
AUC_{0-24} (ng.hr/ml)	427.5 (23)	350.2 (32)
C_{max} (ng/ml)	31.4 (25)	24.0 (35)
C_{tau} (ng/ml)	13.5 (22)	11.3 (32)

Figure 3B



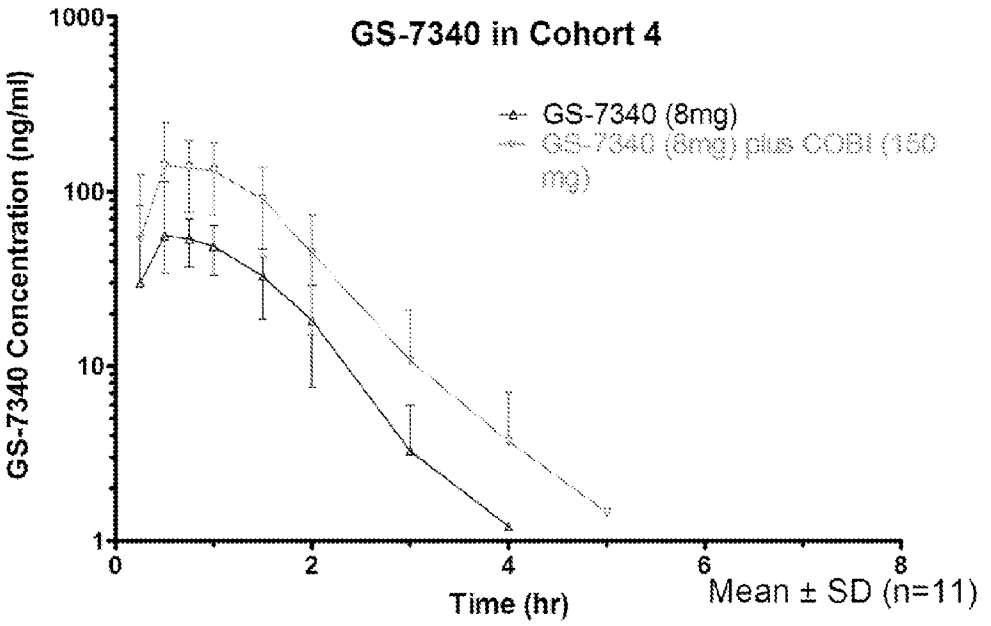
GS-7340 PK	Cohort 2		Cohort 3
Mean (%CV)	FTC/GS-7340 (200/25 mg)	FTC/GS-7340 plus DRV/COBI (800/150 mg)	FTC/GS-7340 (200/25 mg) plus DRV/COBI (800/150 mg)
AUC_{last} (ng.hr/ml)	245.6 (42)	243.9 (41)	271.0 (39)
C_{max} (ng/ml)	208.3 (40)	215.0 (59)	287.4 (73)

Figure 4A



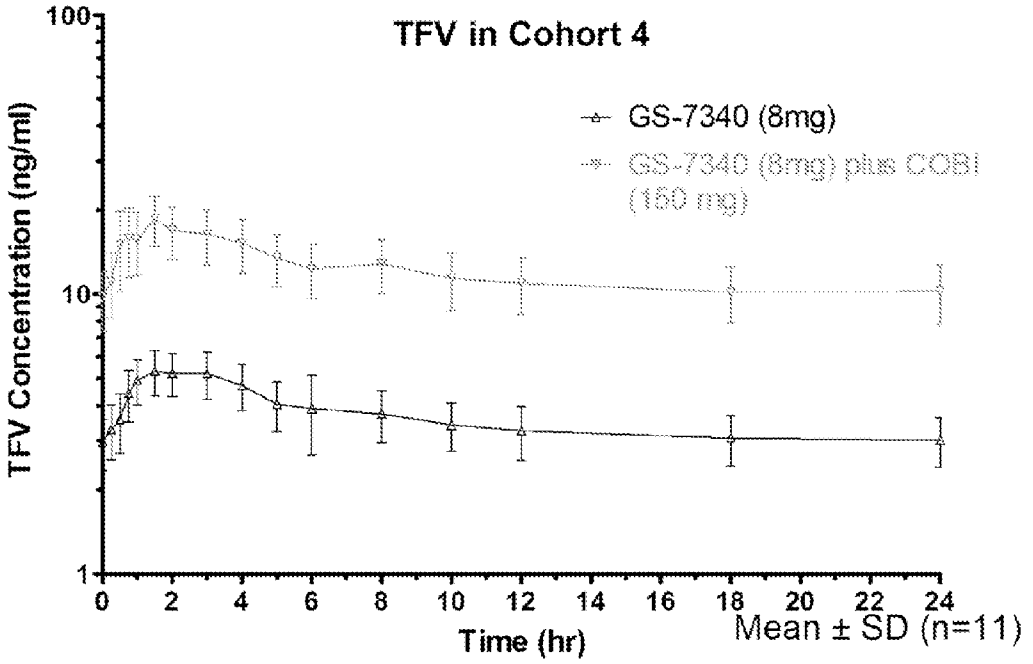
TFV PK	Cohort 2		Cohort 3
Mean (%CV)	FTC/GS-7340 (200/25 mg)	FTC/GS-7340 plus DRV/COBI (800/150 mg)	FTC/GS-7340 (200/25 mg) plus DRV/COBI (800/150 mg)
AUC_{tau} (ng.hr/ml)	299.2 (29)	953.4 (20)	967.6 (13)
C_{max} (ng/ml)	18.3 (28)	57.4 (23.2)	57.7 (15)
C_{tau} (ng/ml)	10.8 (33)	33.7 (20)	36.2 (13)

Figure 4B



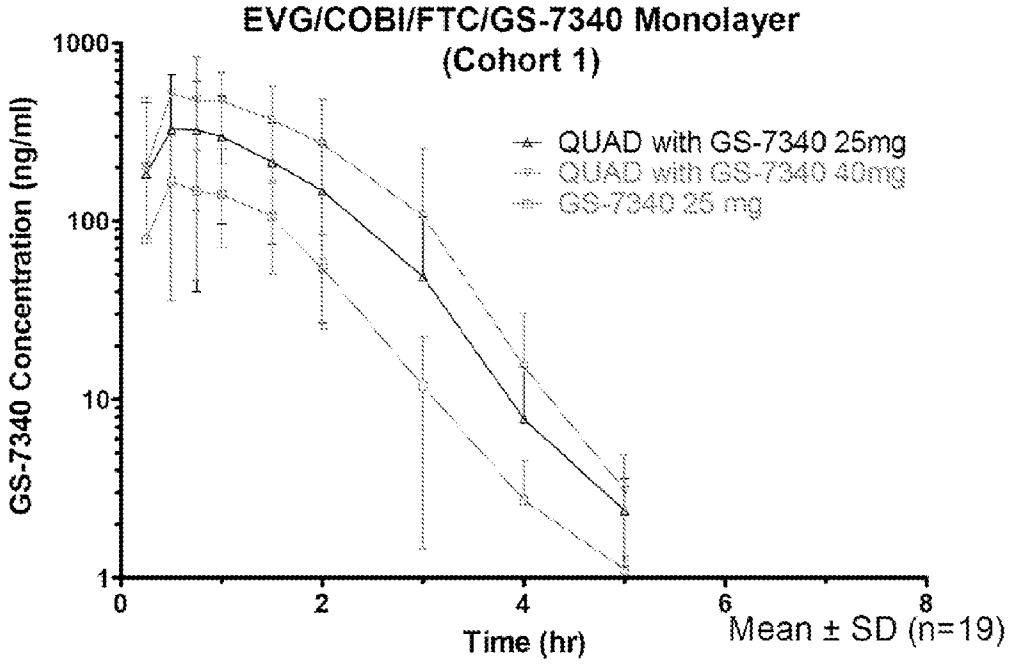
	Cohort 4			
	Steady State PK		Single Dose PK	
Mean (%CV)	GS-7340 (8 mg)	GS-7340 (8 mg) plus COBI (150 mg)	GS-7340 (8 mg)	GS-7340 (8 mg) plus COBI (150 mg)
AUC_{last} (ng.hr/ml)	81.2 (44)	213.3 (38)	64.7 (34)	188.0 (27)
C_{max} (ng/ml)	71.0 (73)	189.9 (46)	49.9 (38)	141.5 (33)

Figure 5A



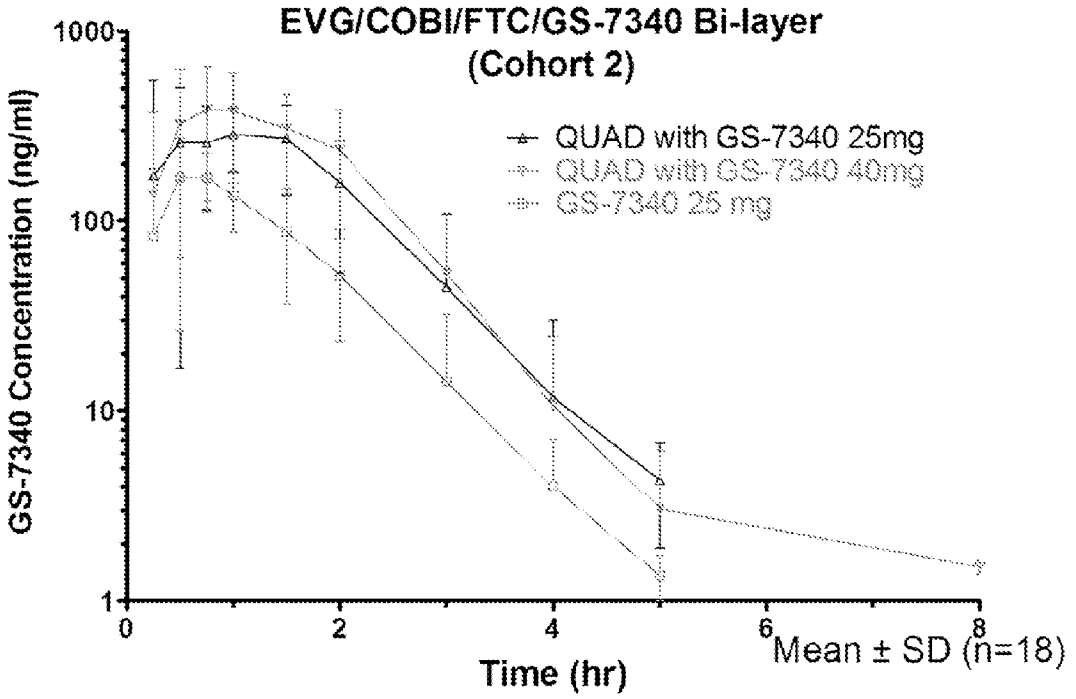
	TFV PK Multiple Dose PK	
Mean (%CV)	GS-7340 (8 mg)	GS-7340 (8 mg) plus COBI (150 mg)
AUC_{tau} (ng.hr/ml)	86.1 (19)	286.9 (22)
C_{max} (ng/ml)	5.8 (19)	19.3 (20)
C_{tau} (ng/ml)	3.0 (20)	10.2 (24)

Figure 5B



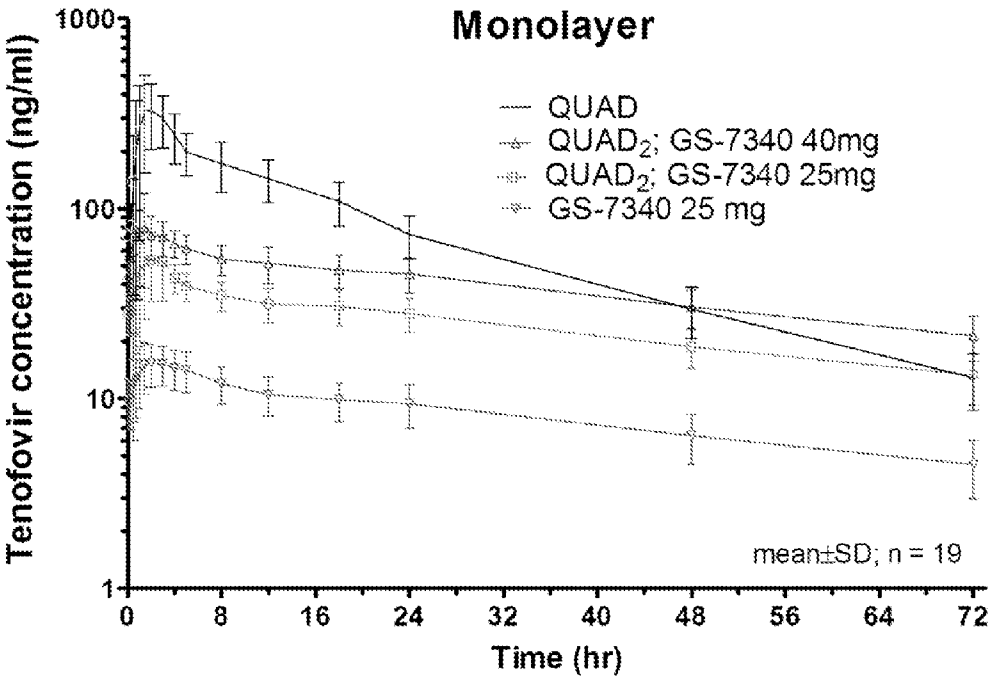
Monolayer Mean (%CV)	$QUAD_2$ with GS-7340 25 mg	$QUAD_2$ with GS-7340 40 mg	GS-7340 25 mg
C_{max} (ng/ml)	506 (54)	793 (52)	215 (55)
AUC_{tau} (ng.hr/ml)	552 (41)	929 (34)	243 (42)

Figure 6



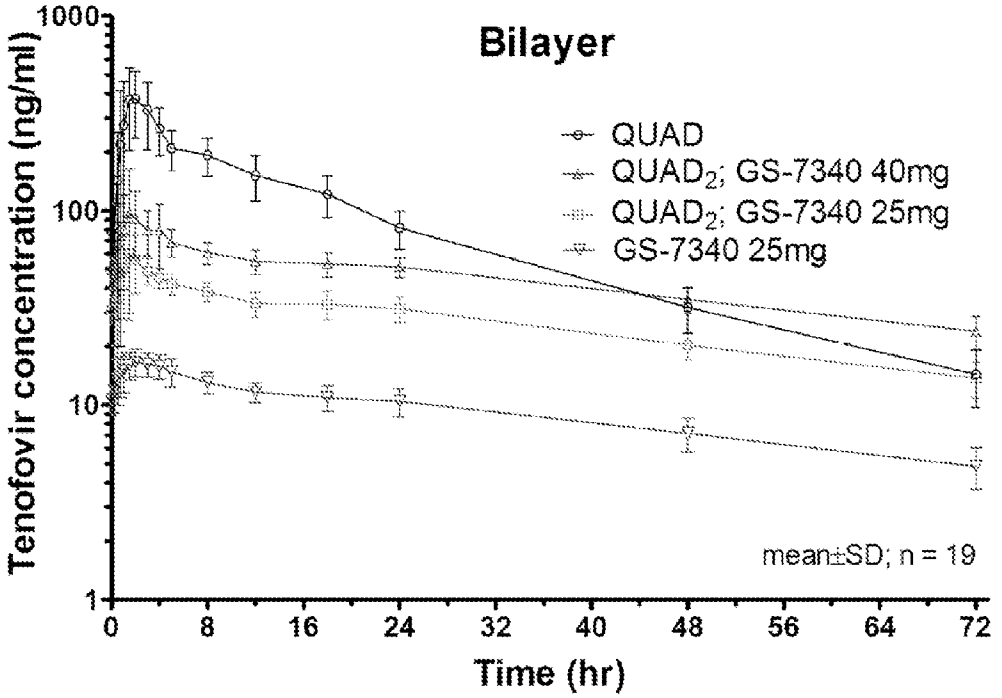
Bi-layer Mean (%CV)	$QUAD_2$ with GS-7340 25 mg	$QUAD_2$ with GS-7340 40 mg	GS-7340 25 mg
C_{max} (ng/ml)	472 (58)	587 (33)	211 (44)
AUC_{tau} (ng.hr/ml)	559 (29)	760 (27)	245 (34)

Figure 7



Monolayer Mean (%CV)	$QUAD_2$ 25 mg	$QUAD_2$ 40 mg	GS-7340 25 mg	QUAD
C_{max} (ng/ml)	66 (51)	103 (64)	16 (25)	445 (29)
C_{tau} (ng/ml)	28.1 (20)	45.4 (21)	9.40 (26)	73.1 (25)
AUC_{tau} (ng.hr/ml)	837 (18)	1310 (21)	274 (24)	3760 (22)

Figure 8



Bi-layer Mean (%CV)	$QUAD_2$ 25 mg	$QUAD_2$ 40 mg	GS-7340 25 mg	QUAD
C_{max} (ng/ml)	71.9 (57)	117 (60)	17.5 (15)	505 (27)
C_{tau} (ng/ml)	31.3 (15)	51.4 (12)	10.5 (17)	81.6 (22)
AUC_{tau} (ng.hr/ml)	899 (13)	1460 (16)	301 (13)	4120 (22)

Figure 9

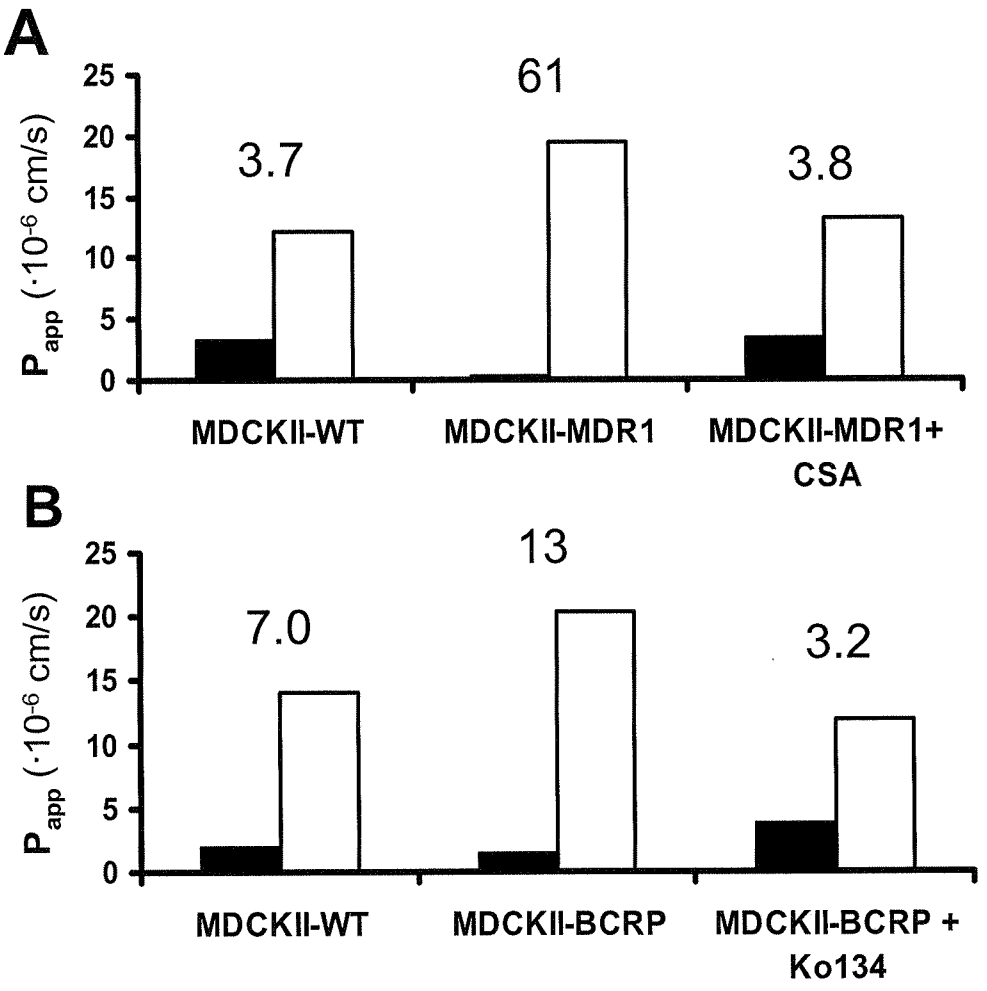

A
25
20
15
10
5
0
P_{app} ($\cdot 10^{-6}$ cm/s)
3.7
61
3.8
MDCKII-WT
MDCKII-MDR1
MDCKII-MDR1+ CSA
B
25
20
15
10
5
0
P_{app} ($\cdot 10^{-6}$ cm/s)
7.0
13
3.2
MDCKII-WT
MDCKII-BCRP
MDCKII-BCRP + Ko134

Figure 10

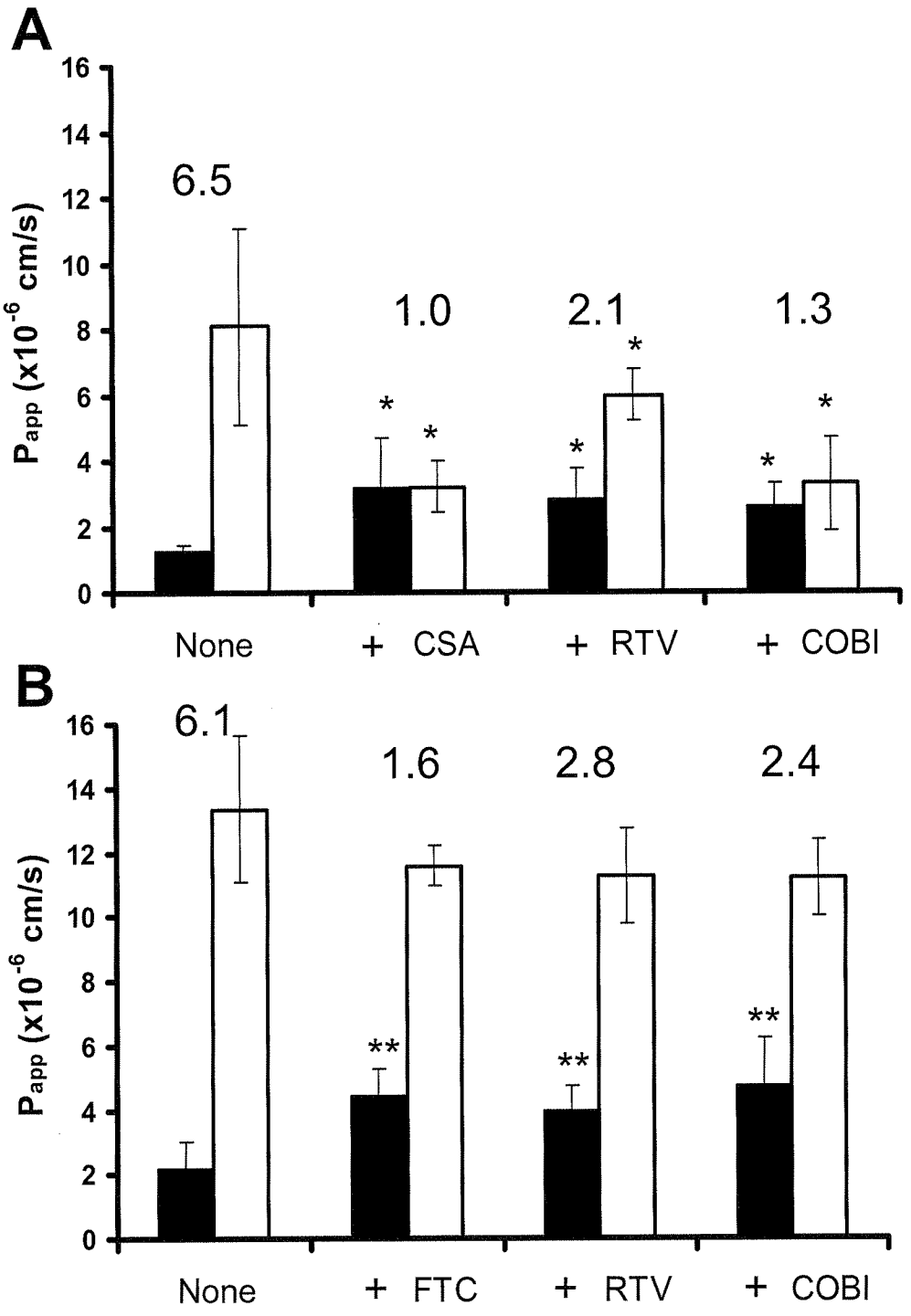

A
16
14
12
10
8
6
4
2
0
P_{app} ($x10^{-6}$ cm/s)
6.5
1.0
2.1
1.3
*
None
+ CSA
+ RTV
+ COBI
B
6.1
1.6
2.8
2.4
**
None
+ FTC
+ RTV
+ COBI

Figure 11

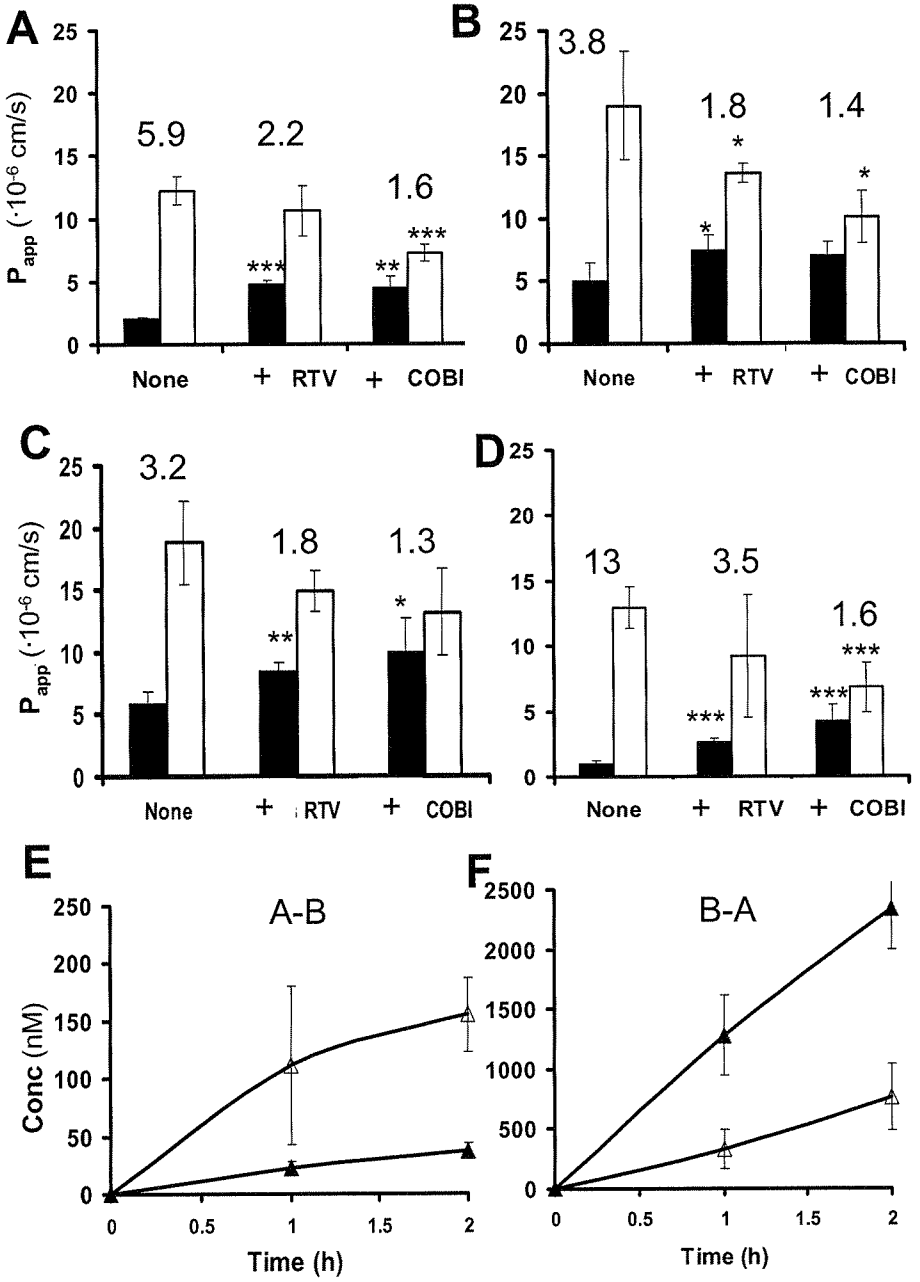

A
B
C
D
E
F
P_{app} ($\cdot 10^{-6}$ cm/s)
5.9
2.2
1.6
3.8
1.8
1.4
3.2
1.8
1.3
13
3.5
1.6
None
+ RTV
+ COBI
A-B
B-A
Conc (nM)
Time (h)

Figure 12

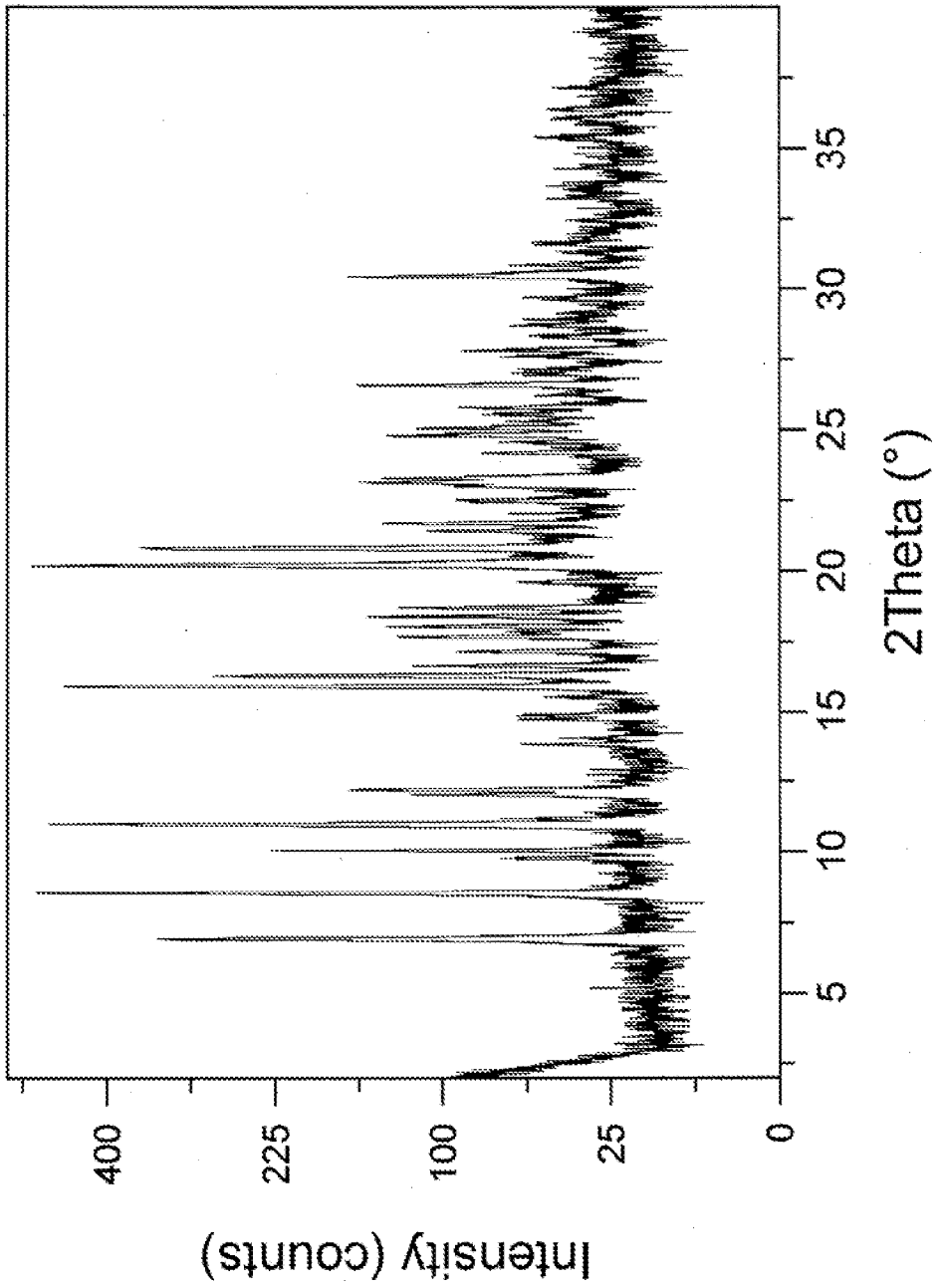


Figure 13

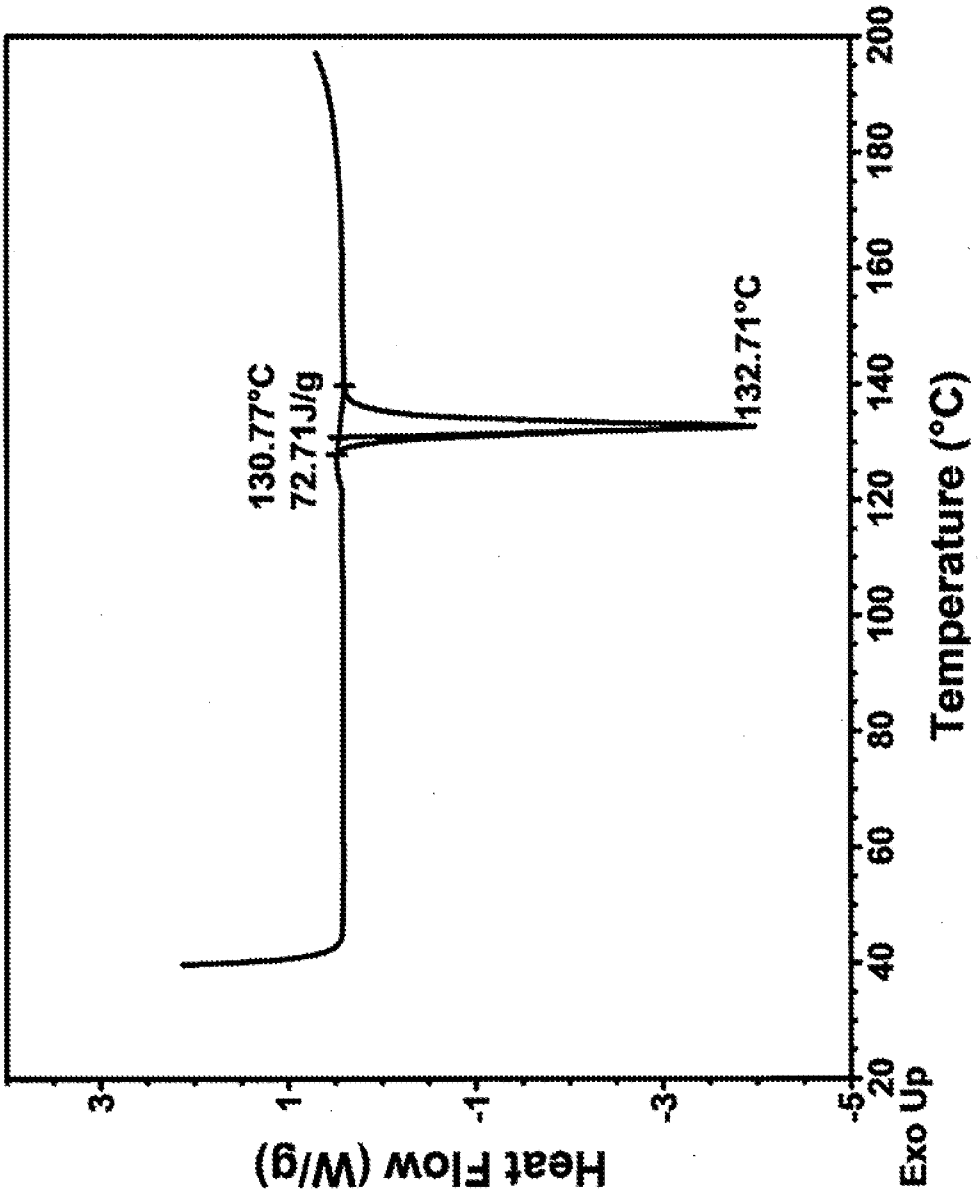

130.77°C
72.71J/g
132.71°C
Heat Flow (W/g)
Exo Up
Temperature (°C)
3
1
-1
-3
-5
20
40
60
80
100
120
140
160
180
200

Figure 14

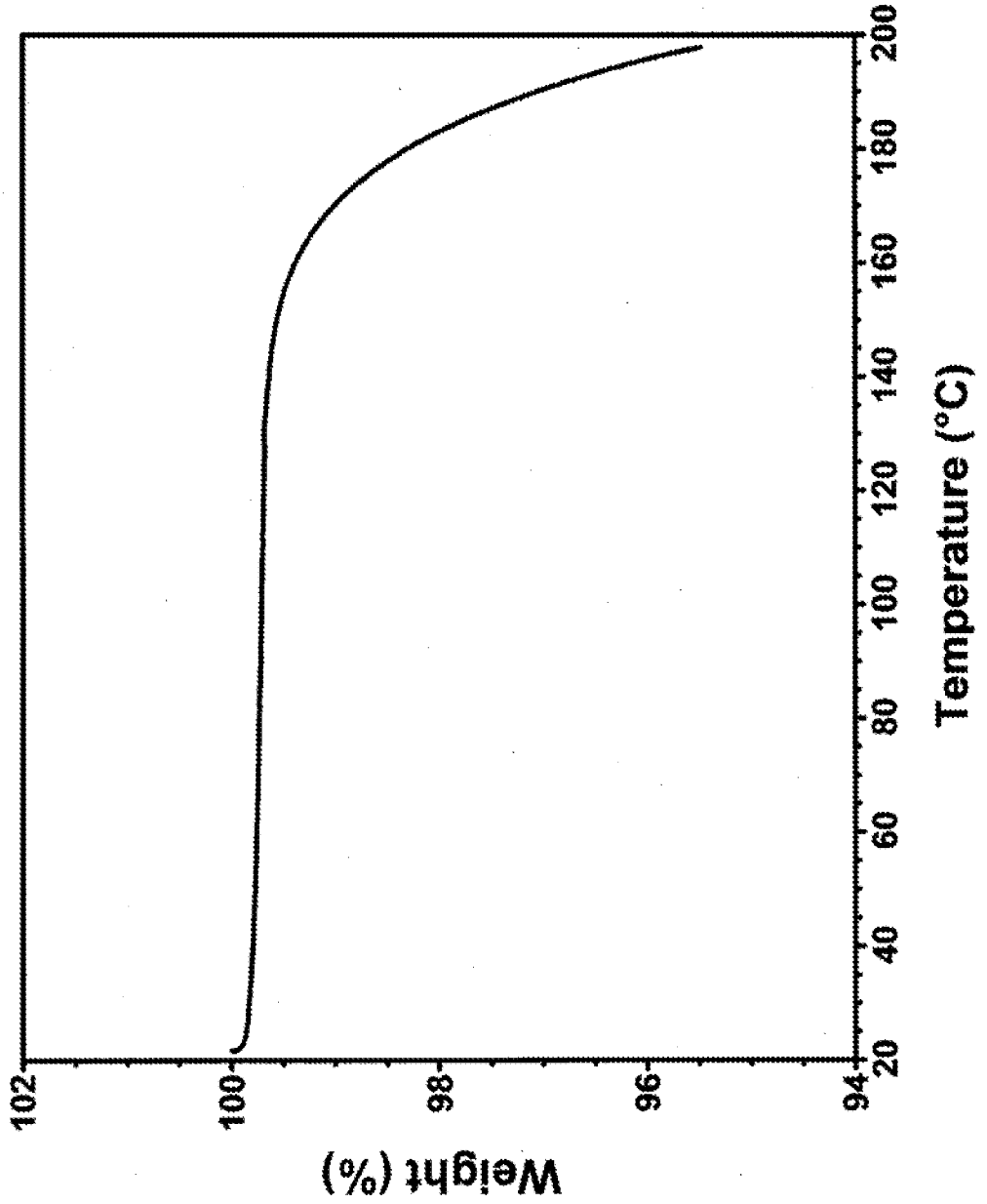

Weight (%)
102
100
98
96
94
Temperature (°C)
20
40
60
80
100
120
140
160
180
200

Figure 15

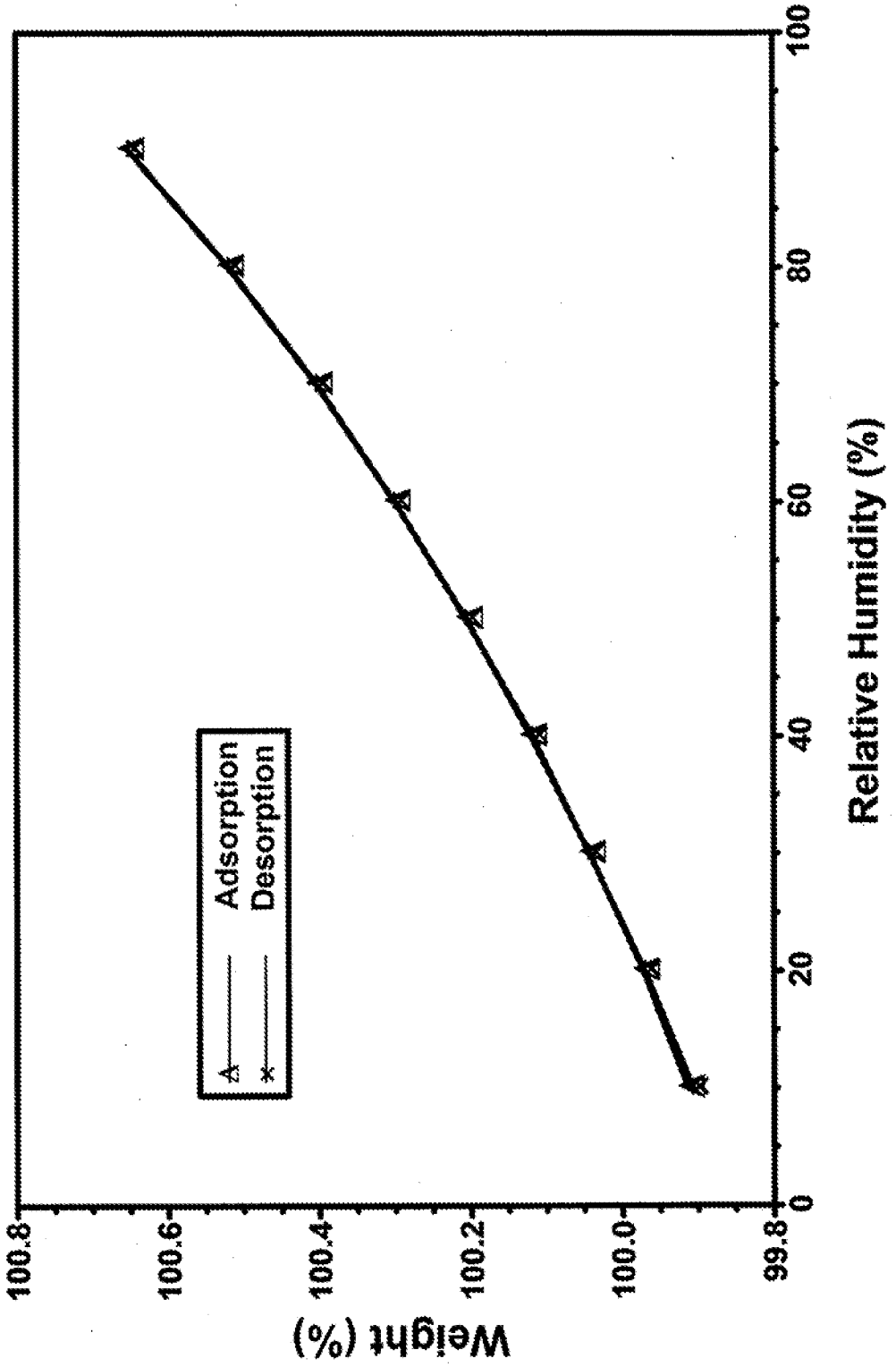

Weight (%)
100.8
100.6
100.4
100.2
100.0
99.8
Relative Humidity (%)
0
20
40
60
80
100
Adsorption
Desorption

Figure 16

COMBINATION THERAPY COMPRISING TENOFOVIR ALAFENAMIDE HEMIFUMARATE AND COBICISTAT FOR USE IN THE TREATMENT OF VIRAL INFECTIONS

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims the benefit of priority from U.S. Provisional Patent Application No. 61/594,894, filed Feb. 3, 2012; U.S. Provisional Patent Application No. 61/618,411, filed Mar. 30, 2012; U.S. Provisional Patent Application No. 61/624,676, filed Apr. 16, 2012; U.S. Provisional Patent Application No. 61/692,392, filed Aug. 23, 2012; and U.S. Provisional Patent Application No. 61/737,493, filed Dec. 14, 2012, the content of each of which is hereby incorporated by reference herein in its entirety.

BACKGROUND OF THE INVENTION

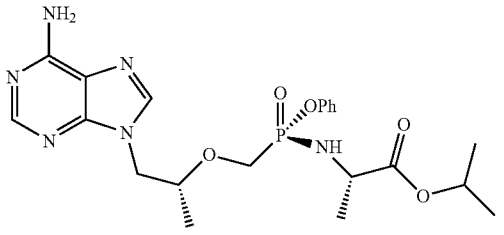
Field of the Invention

[0002] Tenofovir {9-R-[(2-phosphonomethoxy)propyl]adenine}, an acyclic nucleotide analog of dAMP, is a potent in vitro and in vivo inhibitor of human immunodeficiency virus type 1 (HIV-1) replication. Tenofovir is sequentially phosphorylated in the cell by AMP kinase and nucleoside diphosphate kinase to the active species, tenofovir diphosphate, which acts as a competitive inhibitor of HIV-1 reverse transcriptase that terminates the growing viral DNA chain. The presence of a nonhydrolyzable phosphonic acid moiety in tenofovir circumvents an initial phosphorylation step that can be rate limiting for the activation of nucleoside analog inhibitors of HIV reverse transcriptase. Due to the presence of a phosphonate group, tenofovir is negatively charged at neutral pH, thus limiting its oral bioavailability.

[0003] Tenofovir disoproxil fumarate (TDF; VIREAD®), the first generation oral prodrug of tenofovir, has been extensively studied in clinical trials and has received marketing authorization in many countries as a once-daily tablet (300 mg) in combination with other antiretroviral agents for the treatment of HIV-1 infection.

[0004] U.S. Pat. No. 7,390,791 describes certain prodrugs of phosphonate nucleotide analogs that are useful in therapy. One such prodrug is 9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]-methoxy]propyl] adenine 16:

16



[0005] GS-7340 {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl] adenine} is an isopropylalaninyl phenyl ester prodrug of tenofovir (9-[(2-phosphonomethoxy) propyl]adenine). GS-7340 exhibits potent anti-HIV activity 500- to 1000-fold enhanced activity relative to tenofovir against HIV-1 in T cells, activated peripheral blood mononuclear lymphocytes (PBMCs), and macrophages. GS-7340 also has enhanced ability to deliver and increase the accumulation of the parent tenofovir into PBMCs and other lymphatic tissues in vivo. It is also a potent inhibitor of hepatitis B virus.

[0006] GS-7340 is metabolized to tenofovir, which is not dependent on an intracellular nucleoside kinase activity for the first step in the conversion to the active metabolite, tenofovir diphosphate (PMPApp). The cellular enzymes responsible for tenofovir metabolism to the active diphosphorylated form are adenylate kinase and nucleotide diphosphate kinase, which are highly active and ubiquitous. Adenylate kinase exists as multiple isozymes (AK1 to AK4), with the phosphorylation of tenofovir mediated most efficiently by AK2.

[0007] Tenofovir does not interact significantly with human drug metabolizing cytochrome P450 enzymes or UDP-glucuronosyltransferases as a substrate, inhibitor, or inducer, in vitro or in vivo in humans. GS-7340 has limited potential to alter cytochrome P450 enzyme activity through inhibition (IC_{50}>7 μM compared to all isoforms tested). Similarly GS-7340 does not inhibit UGT1A1 function at concentrations up to 50 μM. In addition, GS-7340 is not an activator of either the aryl hydrocarbon receptor or human pregnane X receptor.

[0008] Although tenofovir and GS-7340 show desirable activities, the treatment cost and the potential for unwanted side effects can both increase as the required dose of a drug increases. Therefore, there is a need for methods and compositions that are useful for achieving an acceptable anti-viral effect using a reduced dose of tenofovir or GS-7340.

[0009] Along with U.S. Pat. No. 7,390,791, U.S. Pat. No. 7,803,788 (the content of each of which is incorporated by reference herein in its entirety) also describes certain prodrugs of phosphonate nucleotide analogs that are useful in therapy. As noted above, one such prodrug is 9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine. This compound is also known by the Chemical Abstract name L-alanine, N—[(S)-[[(1R)-2-(6-amino-9H-purin-9-yl)-1-methylethoxy]methyl]phenoxyphosphinyl]-, 1-methylethyl ester. U.S. Pat. Nos. 7,390,791 and 7,803,788 disclose a monofumarate form of this compound and its preparation method (see, e.g., Example 4).

SUMMARY OF THE INVENTION

[0010] It has been determined that the systemic exposure to GS-7340 in humans improves when GS-7340 is administered with cobicistat (1,3-thiazol-5-ylmethyl (2R,5R)-(5-{[(2S)-2-[(methyl{[2-(propan-2-yl)-1,3-thiazol-4-yl] methyl}carbamoyl)amino]]-4-(morpholin-4-yl)butanamido}-1,6-diphenylhexan-2-yl)carbamate). When administered with cobicistat, GS-7340 was calculated to have a systemic exposure equivalent 2.2 fold higher than a dose of GS-7340 alone. In another case, GS-7340 administered with cobicistat was calculated to have a systemic exposure equivalent 3-4 fold higher than a dose of GS-7340 alone. In another case, GS-7340 administered with cobicistat was calculated to have a systemic exposure equivalent 1.3 fold higher than a dose of GS-7340 alone.

[0011] In one embodiment, the invention provides for the use of the compound GS-7340 or a pharmaceutically acceptable salt thereof and cobicistat, or a pharmaceutically acceptable salt thereof, for the prophylactic or therapeutic treatment

of a viral infection in a human. The cobicistat may be coadministered with GS-7340. GS-7340 or a pharmaceutically acceptable salt thereof, may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg, or other ranges as set forth below. Cobicistat or a pharmaceutically acceptable salt thereof may be used in an amount of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. GS-7340, or a pharmaceutically acceptable salt thereof, and cobicistat or a pharmaceutically acceptable salt thereof, may be coadministered. A unit dosage form comprising a daily amount of GS-7340 or a pharmaceutically acceptable salt thereof, and a daily amount of cobicistat or pharmaceutically acceptable salt thereof, may be used. The virus of the viral infection may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0012] In one embodiment, the invention provides for the use of the compound GS-7340, or a pharmaceutically acceptable salt thereof, and cobicistat, or a pharmaceutically acceptable salt thereof, for improving the pharmacokinetics of GS-7340. The cobicistat may be coadministered with GS-7340. GS-7340, or a pharmaceutically acceptable salt thereof, may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg, or other ranges as set forth below. Cobicistat or a pharmaceutically acceptable salt thereof may be used in an amount of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. GS-7340, or a pharmaceutically acceptable salt thereof, and the cobicistat or pharmaceutically acceptable salt thereof may be coadministered. A unit dosage form comprising a daily amount GS-7340 or a pharmaceutically acceptable salt thereof, and a daily amount cobicistat or pharmaceutically acceptable salt thereof may be used. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0013] In one embodiment, the invention provides for the use of the compound GS-7340 or a pharmaceutically acceptable salt thereof and cobicistat, or a pharmaceutically acceptable salt thereof, for improving the $C_{max\ of\ GS\text{-}}$7340. The cobicistat may be coadministered with GS-7340. GS-7340 or a pharmaceutically acceptable salt thereof, may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat or a pharmaceutically acceptable salt thereof may be used in an amount of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. GS-7340, or a pharmaceutically acceptable salt thereof, and the cobicistat or pharmaceutically acceptable salt thereof may be coadministered. A unit dosage form comprising a daily amount of GS-7340 or a pharmaceutically acceptable salt thereof, and a daily amount of cobicistat or pharmaceutically acceptable salt thereof may be used. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0014] In one embodiment, the invention provides for the use of the compound GS-7340 or a pharmaceutically acceptable salt thereof and cobicistat, or a pharmaceutically acceptable salt thereof, for improving blood levels of GS-7340. The cobicistat may be coadministered with GS-7340. GS-7340 or a pharmaceutically acceptable salt thereof, may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat or a pharmaceutically acceptable salt thereof may be used in an amount of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. GS-7340, or a pharmaceutically acceptable salt thereof, and the cobicistat or pharmaceutically acceptable salt thereof may be coadministered. A unit dosage form comprising a daily amount GS-7340 or a pharmaceutically acceptable salt thereof, and a daily amount cobicistat or pharmaceutically acceptable salt thereof may be used. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0015] In one embodiment, the invention provides for a composition comprising a unit-dosage form of GS-7340 or a pharmaceutically acceptable salt thereof; a unit-dosage form of cobicistat, or a pharmaceutically acceptable salt thereof; and a pharmaceutically acceptable carrier or diluent. The composition may include GS-7340 or a pharmaceutically acceptable salt thereof in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. The composition may include cobicistat in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. The unit dosage form may be a single daily dosage.

[0016] In one embodiment, the invention provides for a kit comprising: (1) GS-7340, or a pharmaceutically acceptable salt thereof; (2) cobicistat, or a pharmaceutically acceptable salt thereof; (3) one or more containers; and (4) prescribing information regarding administering the GS-7340 or a pharmaceutically acceptable salt thereof with the cobicistat or the pharmaceutically acceptable salt thereof. The kit may include GS-7340 or a pharmaceutically acceptable salt thereof in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. The kit may include cobicistat in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg.

[0017] In one embodiment, the invention provides for a method of treating a viral infection in a human comprising coadministering GS-7340 with cobicistat, or a pharmaceutically acceptable salt thereof, wherein the dose of cobicistat coadministered with the GS-7340 provides a systemic exposure of GS-7340 comparable to the systemic exposure obtainable by administration of a greater dose of GS-7340 in the absence of cobicistat. GS-7340 or a pharmaceutically acceptable salt thereof in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below may be coadministered with cobicistat. Cobicistat in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg may be coadministered with GS-7340. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0018] In one embodiment, the invention provides for a method for inhibiting activity of a retroviral reverse transcriptase in a human comprising coadministering GS-7340 with cobicistat, or a pharmaceutically acceptable salt thereof, wherein the dose of cobicistat coadministered with the GS-7340 provides a systemic exposure of GS-7340 comparable to the systemic exposure obtainable by administration of a greater dose of GS-7340 in the absence of cobicistat. GS-7340 or a pharmaceutically acceptable salt thereof in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below may be coadministered with cobicistat. Cobicistat in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg may be coadministered with GS-7340. The virus may be human immunodeficiency virus (HIV).

[0019] In one embodiment, the invention provides for the use of the compound GS-7340 or a pharmaceutically acceptable salt thereof coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for treating a viral infection. The invention further provides for the use of the compound GS-7340 or a

pharmaceutically acceptable salt thereof coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for treating a viral infection in a human. GS-7340 or a pharmaceutically acceptable salt thereof may be used in a subtherapeutic amount (or, in some embodiments throughout, in a therapeutic amount). GS-7340 or a pharmaceutically acceptable salt thereof may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat may be used in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. Cobicistat may be used in an amount that provides a systemic exposure of GS-7340 comparable to the systemic exposure obtainable by administration of a greater dose of GS-7340 in the absence of cobicistat is used in the manufacture of the medicament. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0020] In one embodiment, the invention provides for the use of the compound GS-7340 or a pharmaceutically acceptable salt thereof coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for inhibiting activity of a retroviral reverse transcriptase. The invention further provides for the use of the compound GS-7340 or a pharmaceutically acceptable salt thereof coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for inhibiting activity of a retroviral reverse transcriptase in a human. GS-7340 or a pharmaceutically acceptable salt thereof may be used in a subtherapeutic amount. GS-7340 or a pharmaceutically acceptable salt thereof may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat may be used in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. The cobicistat may be used in an amount that provides a systemic exposure of GS-7340 comparable to the systemic exposure obtainable by administration of a greater dose of GS-7340 in the absence of cobicistat is used in the manufacture of the medicament. The virus may be human immunodeficiency virus (HIV).

[0021] In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof, to prepare a medicament useful for improving the pharmacokinetics of GS-7340, or a pharmaceutically acceptable salt thereof, following administration to a human. GS-7340 or a pharmaceutically acceptable salt thereof may be used in a subtherapeutic amount. GS-7340 or a pharmaceutically acceptable salt thereof may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat may be used in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. The cobicistat may be used in an amount that provides a systemic exposure of GS-7340 comparable to the systemic exposure obtainable by administration of a greater dose of GS-7340 in the absence of cobicistat is used in the manufacture of the medicament. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0022] In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof, to prepare a medicament useful for improving the pharmacokinetics of {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl] adenine}, or a pharmaceutically acceptable salt thereof, following administration to a human. GS-7340 or a pharmaceutically acceptable salt thereof may be used in a subtherapeutic amount. {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl] adenine}, or a pharmaceutically acceptable salt thereof, may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth herein. Cobicistat may be used in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. The cobicistat may be used in an amount that provides a systemic exposure of GS-7340 comparable to the systemic exposure obtainable by administration of a greater dose of GS-7340 in the absence of cobicistat is used in the manufacture of the medicament. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0023] In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament for a human useful for reducing a dose of GS-7340 by about 30-70%, or a pharmaceutically acceptable salt thereof, upon administration of the cobicistat. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0024] In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament for a human useful for reducing a dose of GS-7340 by about 2-4 fold, or a pharmaceutically acceptable salt thereof, upon administration of the cobicistat. In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament for a human useful for reducing a dose of GS-7340 by about 3 fold, or a pharmaceutically acceptable salt thereof, upon administration of the cobicistat. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0025] In one embodiment, the invention provides for a method of treating a viral infection in a human comprising coadministering 1) GS-7340 or a pharmaceutically acceptable salt thereof; and 2) cobicistat, or a pharmaceutically acceptable salt thereof to the human. GS-7340 or a pharmaceutically acceptable salt thereof is administered in a subtherapeutic amount. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0026] In one embodiment, the invention provides for a use of a subtherapeutic dose of GS-7340 coadministered with cobicistat for treating a viral infection. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0027] In one embodiment, the invention provides for the use of a subtherapeutic dose of GS-7340 coadministered with cobicistat for inhibiting retroviral reverse transcriptase. The virus may be human immunodeficiency virus (HIV).

[0028] In one embodiment, the invention provides for an anti-virus agent(s) comprising (a) a compound GS-7340 or a pharmaceutically acceptable salt thereof and (b) cobicistat, or a pharmaceutically acceptable salt thereof. The anti-virus agent(s) may include GS-7340 or a pharmaceutically acceptable salt thereof may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. The anti-virus agent(s) may include cobicistat in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg.

The cobicistat may be used in an amount that provides a systemic exposure of GS-7340 comparable to the systemic exposure obtainable by administration of a greater dose of GS-7340 in the absence of cobicistat is used in the manufacture of the medicament. The anti-virus agent may further include 200 mg of emtricitabine and 150 mg of elvitegravir. The anti-virus agent may further include 150 mg cobicistat, 8 or less mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may further include 150 mg cobicistat, 25 or less mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may further include 150 mg cobicistat, 10 or less mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may include 150 mg cobicistat, 8 mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may include 150 mg cobicistat, 10 mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine.

[0029] In one embodiment, the invention provides for a unit-dosage of GS-7340 or a pharmaceutically acceptable salt thereof and cobicistat, or a pharmaceutically acceptable salt thereof, wherein the unit-dosage is a daily dose. GS-7340 may be present in a subtherapeutic amount. The unit-dosage may further include 150 mg cobicistat, 8 or less mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine. The unit-dosage may further include 150 mg cobicistat, 25 or less mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine. The unit-dosage may further include 150 mg cobicistat, 10 or less mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine. The unit-dosage may include 150 mg cobicistat, 10 mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine.

[0030] In one embodiment, the invention provides the use of cobicistat, or a pharmaceutically acceptable salt thereof, to prepare a medicament useful for improving the pharmacokinetics of {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine}, or a pharmaceutically acceptable salt thereof, following administration to a human. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be, e.g., human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0031] In one embodiment, the invention provides cobicistat for use in improving the pharmacokinetics of {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine} or a pharmaceutically acceptable salt thereof, following administration to a human. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0032] In one embodiment, the invention provides a kit comprising: (1) {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine}, or a pharmaceutically acceptable salt thereof; (2) cobicistat, or a pharmaceutically acceptable salt thereof; (3) one or more containers; and (4) prescribing information regarding administering the {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine} or a pharmaceutically acceptable salt thereof with the cobicistat or a pharmaceutically acceptable salt thereof.

[0033] In one embodiment, the invention provides a kit comprising: (1) a unit dosage form comprising 5-100 mg of {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine}, or a pharmaceutically acceptable salt thereof; (2) a unit dosage form comprising 150 mg cobicistat, or a pharmaceutically acceptable salt thereof; (3) one or more containers; and (4) prescribing information regarding administering the {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine} or a pharmaceutically acceptable salt thereof with cobicistat or a pharmaceutically acceptable salt thereof.

[0034] In one embodiment, the invention provides a use of {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine} or its pharmaceutically acceptable salt for the manufacture of a medicament for inhibiting activity of a retroviral reverse transcriptase in a human, comprising administering GS-7340 or a pharmaceutically acceptable salt thereof, and cobicistat, or a pharmaceutically acceptable salt thereof to the human. The virus may be human immunodeficiency virus (HIV).

[0035] In one embodiment, the invention provides {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine} or its pharmaceutically acceptable salt; and cobicistat, or a pharmaceutically acceptable salt thereof; for use in inhibiting activity of a retroviral reverse transcriptase in a human.

[0036] In one embodiment, the invention provides a use of cobicistat, or a pharmaceutically acceptable salt thereof, to prepare a medicament for a human useful for reducing a dose between about 30-70% of {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine} or a pharmaceutically acceptable salt thereof, upon administration of the cobicistat. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0037] In one embodiment, the invention provides the use of {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine} or a pharmaceutically acceptable salt thereof; and cobicistat or a pharmaceutically acceptable salt thereof for the prophylactic or therapeutic treatment of a viral infection in a human. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0038] In one embodiment, the invention provides an antiviral agent(s) comprising (a) {9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine} or a pharmaceutically acceptable salt thereof, which is used in combination with (b) cobicistat or a pharmaceutically acceptable salt thereof for use in the prophylactic or therapeutic treatment of a viral infection in a human.

[0039] It has also been determined that the systemic exposure to tenofovir in humans improves when tenofovir is administered with cobicistat. When administered with cobicistat, tenofovir was calculated to have a systemic exposure equivalent 3 to 4 fold higher than a dose of tenofovir alone.

[0040] In one embodiment, the invention provides for the use of the compound tenofovir or a pharmaceutically acceptable salt thereof and cobicistat, or a pharmaceutically acceptable salt thereof, for the prophylactic or therapeutic treatment of a viral infection in a human. Tenofovir may be used in amounts of less than 300 mg, 200 mg or less and 100 mg or less. Cobicistat may be used in amounts of 50-500 mg, 100-400 mg, 100-300 mg, and 150 mg. The tenofovir or a pharmaceutically acceptable salt thereof, and the cobicistat or pharmaceutically acceptable salt thereof may be coadminis-

tered. The use may provide a unit dosage form comprising a daily amount tenofovir or a pharmaceutically acceptable salt thereof, and a daily amount cobicistat or pharmaceutically acceptable salt thereof is administered. The virus may be human immunodeficiency virus (HIV).

[0041] In one embodiment, the invention provides for a composition comprising a unit-dosage form of tenofovir or a pharmaceutically acceptable salt thereof; a unit-dosage form of cobicistat, or a pharmaceutically acceptable salt thereof; and a pharmaceutically acceptable carrier or diluent. Tenofovir may be present in the composition in amounts of less than 300 mg, 200 mg or less and 100 mg or less. Cobicistat may be used in amounts of 50-500 mg, 100-400 mg, 100-300 mg, and 150 mg.

[0042] In one embodiment, the invention provides for a kit that includes (1) tenofovir, or a pharmaceutically acceptable salt thereof; (2) cobicistat, or a pharmaceutically acceptable salt thereof; (3) one or more containers; and (4) prescribing information regarding administering the tenofovir or a pharmaceutically acceptable salt thereof with the cobicistat or the pharmaceutically acceptable salt thereof. Tenofovir may be present in the kit in amounts of less than 300 mg, 200 mg or less and 100 mg or less. Cobicistat may be used in amounts of 50-500 mg, 100-400 mg, 100-300 mg, and 150 mg.

[0043] In one embodiment, the invention provides for a method of treating a viral infection in a human that includes coadministering tenofovir with cobicistat, or a pharmaceutically acceptable salt thereof, wherein the dose of cobicistat coadministered with the tenofovir provides a systemic exposure of tenofovir comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir in the absence of cobicistat. Tenofovir may be administered in amounts of less than 300 mg, 200 mg or less and 100 mg or less. Cobicistat may be administered in amounts of 50-500 mg, 100-400 mg, 100-300 mg, and 150 mg. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0044] In one embodiment, the invention provides for a method for inhibiting activity of a retroviral reverse transcriptase in a human comprising coadministering tenofovir with cobicistat, or a pharmaceutically acceptable salt thereof, wherein the dose of tenofovir coadministered with the cobicistat provides a systemic exposure of tenofovir comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir in the absence of cobicistat. Tenofovir may be coadministered in amounts of less than 300 mg, 200 mg or less and 100 mg or less. Cobicistat may be coadministered in amounts of 50-500 mg, 100-400 mg, 100-300 mg, and 150 mg. The virus may be human immunodeficiency virus (HIV)

[0045] In one embodiment, the invention provides for the use of the compound tenofovir or a pharmaceutically acceptable salt thereof coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for treating a viral infection. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0046] In one embodiment, the invention provides for the use of the compound tenofovir or a pharmaceutically acceptable salt thereof coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for treating a viral infection in a human. The tenofovir or a pharmaceutically acceptable salt thereof may be used in a subtherapeutic amount (or, in some embodiments throughout, in a therapeutic amount). Tenofovir may be administered in amounts of less than 300 mg, 200 mg or less and 100 mg or less. The cobicistat may be administered in an amount that provides a systemic exposure of tenofovir comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir in the absence of cobicistat is used in the manufacture of the medicament. Cobicistat in an amount of 150 mg may be used in the manufacture of the medicament. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0047] In one embodiment, the invention provides for the use of the compound tenofovir or a pharmaceutically acceptable salt thereof coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for inhibiting activity of a retroviral reverse transcriptase. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0048] In one embodiment, the invention provides for use of the compound tenofovir or a pharmaceutically acceptable salt thereof coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for inhibiting activity of a retroviral reverse transcriptase in a human. The tenofovir or a pharmaceutically acceptable salt thereof may be used in a subtherapeutic amount. Tenofovir may be used in amounts of less than 300 mg, 200 mg or less and 100 mg or less. The cobicistat may be coadministered in an amount that provides a systemic exposure of tenofovir comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir in the absence of cobicistat is used in the manufacture of the medicament. Cobicistat in an amount of 150 mg may be coadministered. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0049] In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament useful for improving the pharmacokinetics of tenofovir, or a pharmaceutically acceptable salt thereof, following administration to a human. The tenofovir or a pharmaceutically acceptable salt thereof may be used in a subtherapeutic amount. Tenofovir or a pharmaceutically acceptable salt thereof, may be coadministered to the human in an amount of 100 mg or less, 200 mg or less or in amount less than 300 mg. Cobicistat may be used in an amount that provides a systemic exposure of tenofovir comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir in the absence of cobicistat is used in the manufacture of the medicament. Cobicistat in an amount 150 mg may be used to prepare the medicament. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0050] In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament for a human useful for reducing a dose of tenofovir by about 30-70%, or a pharmaceutically acceptable salt thereof, upon administration of the

cobicistat. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0051] In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament for a human useful for reducing a dose of tenofovir by about 2 to 4 fold, or a pharmaceutically acceptable salt thereof, upon administration of the cobicistat. In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament for a human useful for reducing a dose of tenofovir by about 3-fold, or a pharmaceutically acceptable salt thereof, upon administration of the cobicistat. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0052] In one embodiment, the invention provides for a method of treating a viral infection in a human comprising coadministering 1) tenofovir or a pharmaceutically acceptable salt thereof; and 2) cobicistat, or a pharmaceutically acceptable salt thereof to the human. The tenofovir or a pharmaceutically acceptable salt thereof may be administered in a subtherapeutic amount. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0053] In one embodiment, the invention provides for a use of a subtherapeutic dose of tenofovir coadministered with cobicistat for treating a viral infection. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0054] In one embodiment, the invention provides for a use of a subtherapeutic dose of tenofovir coadministered with cobicistat for inhibiting retroviral reverse transcriptase. The virus may be human immunodeficiency virus (HIV).

[0055] In one embodiment, the invention provides for an anti-virus agent(s) comprising (a) a compound tenofovir or a pharmaceutically acceptable salt thereof and (b) cobicistat, or a pharmaceutically acceptable salt thereof. The tenofovir may be present in the anti-virus agent(s) in a subtherapeutic amount. The tenofovir may be present in the anti-virus agent (s) in an amount of 100 mg or less, 200 mg or less or less than 300 mg. The cobicistat coadministered with the tenofovir may be present in the anti-virus agent(s) in an amount that provides a systemic exposure of tenofovir comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir in the absence of cobicistat. The anti-virus agent may further include cobicistat in an amount of 150 mg. The anti-virus agent may further include 200 mg of emtricitabine and 150 mg of elvitegravir. The anti-virus agent may include 150 mg cobicistat, 100 or less mg tenofovir, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may include 150 mg cobicistat, 200 or less mg tenofovir, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may include 150 mg cobicistat, less than 300 mg tenofovir, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may include 150 mg cobicistat, 50 mg tenofovir, 150 mg elvitegravir, and 200 mg emtricitabine. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0056] In one embodiment, the invention provides for a unit-dosage of tenofovir or a pharmaceutically acceptable salt thereof and cobicistat, or a pharmaceutically acceptable salt thereof, wherein the unit-dosage is a daily dose. Tenofovir may be present in a subtherapeutic amount. The unit-dosage may include 100 mg or less, 200 mg or less or less than 300 mg of tenofovir. The unit-dosage may include an amount of cobicistat that provides a systemic exposure of tenofovir comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir in the absence of cobicistat. The unit-dosage may include 150 mg of cobicistat. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0057] Also described is a hemifumarate form of 9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine. The name for 9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino] phenoxyphosphinyl]methoxy]propyl]adenine (GS-7340) is tenofovir alafenamide. The hemifumarate form of tenofovir alafenamide is also referred to herein as tenofovir alafenamide hemifumarate.

[0058] In one embodiment of the invention is provided tenofovir alafenamide hemifumarate, especially in combination with cobicistat and/or with other an additional therapeutic agent or agents.

[0059] In another embodiment is provided tenofovir alafenamide hemifumarate, wherein the ratio of fumaric acid to tenofovir alafenamide is 0.5±0.1, or 0.5±0.05, or 0.5±0.01, or about 0.5.

[0060] In one embodiment is provided tenofovir alafenamide hemifumarate in a solid form.

[0061] In one embodiment is provided tenofovir alafenamide hemifumarate that has an X-ray powder diffraction (XRPD) pattern having 2theta values of 6.9±0.2° and 8.6±0.2°. In another embodiment is provided tenofovir alafenamide hemifumarate wherein the XRPD pattern comprises 2theta values of 6.9±0.2°, 8.6±0.2°, 11.0±0.2°, 15.9±0.2°, and 20.2±0.2°.

[0062] In one embodiment is provided tenofovir alafenamide hemifumarate that has a differential scanning calorimetry (DSC) onset endotherm of 131±2° C., or 131±1° C.

[0063] In one embodiment is provided a pharmaceutical composition comprising tenofovir alafenamide hemifumarate and a pharmaceutically acceptable excipient. In another embodiment is provided the pharmaceutical composition, further comprising an additional therapeutic agent. In a further embodiment, the additional therapeutic agent is selected from the group consisting of human immunodeficiency virus (HIV) protease inhibiting compounds, HIV nonnucleoside inhibitors of reverse transcriptase, HIV nucleoside inhibitors of reverse transcriptase, HIV nucleotide inhibitors of reverse transcriptase, HIV integrase inhibitors, CCR5 inhibitors, and additional protease inhibiting compounds.

[0064] In one embodiment is provided a method for treating a human immunodeficiency virus (HIV) infection comprising administering to a subject in need thereof a therapeutically effective amount of tenofovir alafenamide hemifumarate. In another embodiment is provided a method for treating an HIV infection comprising administering to a subject in need thereof a therapeutically effective amount of a pharmaceutical composition comprising tenofovir alafenamide hemifumarate. In a further embodiment, the method comprises administering to the subject one or more additional therapeutic agents selected from the group consisting of HIV protease inhibiting compounds, HIV nonnucleoside inhibitors of reverse transcriptase, HIV nucleoside inhibitors of reverse transcriptase, HIV nucleotide inhibitors of reverse

transcriptase, HIV integrase inhibitors, CCR5 inhibitors, and additional protease inhibiting compounds.

[0065] In one embodiment is provided a method for treating a hepatitis B virus (HBV) infection comprising administering to a subject in need thereof a therapeutically effective amount of tenofovir alafenamide hemifumarate. In another embodiment is provided a method for treating an HBV infection comprising administering to a subject in need thereof a therapeutically effective amount of the pharmaceutical composition comprising tenofovir alafenamide hemifumarate.

[0066] In one embodiment is provided a method for preparing a pharmaceutical composition comprising combining tenofovir alafenamide hemifumarate and a pharmaceutically acceptable excipient to provide the pharmaceutical composition.

[0067] In one embodiment is provided tenofovir alafenamide hemifumarate for use in medical therapy.

[0068] In one embodiment is provided the use of tenofovir alafenamide hemifumarate for the prophylactic or therapeutic treatment of an HIV infection. In another embodiment is provided the use of tenofovir alafenamide hemifumarate to treat an HIV infection. In a further embodiment is provided the use of tenofovir alafenamide hemifumarate for the preparation or manufacture of a medicament for the treatment of an HIV infection. In another further embodiment is provided tenofovir alafenamide hemifumarate for use in treating an HIV infection.

[0069] In one embodiment is provided the use of tenofovir alafenamide hemifumarate for the prophylactic or therapeutic treatment of an HBV infection. In another embodiment is provided the use of tenofovir alafenamide hemifumarate to treat an HBV infection. In a further embodiment is provided the use of tenofovir alafenamide hemifumarate for the preparation or manufacture of a medicament for the treatment of an HBV infection. In another further embodiment is provided tenofovir alafenamide hemifumarate for use in treating an HBV infection.

[0070] In some embodiments of the invention, the methods of treating and the like comprise administration of multiple daily doses. In other embodiments, the methods of treating and the like comprise administration of a single daily dose.

[0071] In one embodiment, the invention provides for the use of tenofovir alafenamide hemifumarate and cobicistat, or a pharmaceutically acceptable salt thereof, for the prophylactic or therapeutic treatment of a viral infection in a human. The cobicistat may be coadministered with tenofovir alafenamide hemifumarate. Tenofovir alafenamide hemifumarate may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat or a pharmaceutically acceptable salt thereof may be used in an amount of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. Tenofovir alafenamide hemifumarate and cobicistat or pharmaceutically acceptable salt thereof may be coadministered. A unit dosage form comprising a daily amount of tenofovir alafenamide hemifumarate, and a daily amount of cobicistat or pharmaceutically acceptable salt thereof may be used. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0072] In one embodiment, the invention provides for the use tenofovir alafenamide hemifumarate and cobicistat, or a pharmaceutically acceptable salt thereof, for improving the pharmacokinetics of tenofovir alafenamide hemifumarate. Cobicistat may be coadministered with tenofovir alafenamide hemifumarate. Tenofovir alafenamide hemifumarate may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat or a pharmaceutically acceptable salt thereof may be used in an amount of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. Tenofovir alafenamide hemifumarate and cobicistat, or pharmaceutically acceptable salt thereof, may be coadministered. A unit dosage form comprising a daily amount of tenofovir alafenamide hemifumarate, and a daily amount of cobicistat or pharmaceutically acceptable salt thereof may be used. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0073] In one embodiment, the invention provides for the use of tenofovir alafenamide hemifumarate and cobicistat, or a pharmaceutically acceptable salt thereof, for improving the C_{max} of tenofovir alafenamide hemifumarate. The cobicistat may be coadministered with tenofovir alafenamide hemifumarate. Tenofovir alafenamide hemifumarate may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat or a pharmaceutically acceptable salt thereof may be used in an amount of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. Tenofovir alafenamide hemifumarate and cobicistat, or pharmaceutically acceptable salt thereof, may be coadministered. A unit dosage form comprising a daily amount of tenofovir alafenamide hemifumarate, and a daily amount of cobicistat, or a pharmaceutically acceptable salt thereof, may be used. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0074] In one embodiment, the invention provides for the use of tenofovir alafenamide hemifumarate and cobicistat, or a pharmaceutically acceptable salt thereof, for improving blood levels of tenofovir alafenamide hemifumarate. The cobicistat may be coadministered with tenofovir alafenamide hemifumarate. Tenofovir alafenamide hemifumarate may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat, or a pharmaceutically acceptable salt thereof, may be used in an amount of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. Tenofovir alafenamide hemifumarate and cobicistat, or a pharmaceutically acceptable salt thereof, may be coadministered. A unit dosage form comprising a daily amount tenofovir alafenamide hemifumarate, and a daily amount cobicistat, or a pharmaceutically acceptable salt thereof, may be used. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0075] In one embodiment, the invention provides for a composition comprising a unit-dosage form of tenofovir alafenamide hemifumarate; a unit-dosage form of cobicistat, or a pharmaceutically acceptable salt thereof; and a pharmaceutically acceptable carrier or diluent. The composition may include tenofovir alafenamide hemifumarate in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. The composition may include cobicistat in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. The unit-dosage form may be a single daily dosage.

[0076] In one embodiment, the invention provides for a kit comprising: (1) tenofovir alafenamide hemifumarate; (2) cobicistat, or a pharmaceutically acceptable salt thereof; (3) one or more containers; and (4) prescribing information regarding administering the tenofovir alafenamide hemifu-

marate with the cobicistat, or the pharmaceutically acceptable salt thereof. The kit may include tenofovir alafenamide hemifumarate in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. The kit may include cobicistat in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg.

[0077] In one embodiment, the invention provides for a method of treating a viral infection in a human comprising coadministering tenofovir alafenamide hemifumarate with cobicistat, or a pharmaceutically acceptable salt thereof, wherein the dose of cobicistat coadministered with the tenofovir alafenamide hemifumarate provides a systemic exposure of tenofovir alafenamide hemifumarate comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir alafenamide hemifumarate in the absence of cobicistat. Tenofovir alafenamide hemifumarate in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below may be coadministered with cobicistat. Cobicistat in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg may be coadministered with tenofovir alafenamide hemifumarate. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0078] In one embodiment, the invention provides for a method for inhibiting activity of a retroviral reverse transcriptase in a human comprising coadministering tenofovir alafenamide hemifumarate with cobicistat, or a pharmaceutically acceptable salt thereof, wherein the dose of cobicistat coadministered with the tenofovir alafenamide hemifumarate provides a systemic exposure of tenofovir alafenamide hemifumarate comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir alafenamide hemifumarate in the absence of cobicistat. Tenofovir alafenamide hemifumarate or a pharmaceutically acceptable salt thereof in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below may be coadministered with cobicistat. Cobicistat in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg may be coadministered with tenofovir alafenamide hemifumarate. The virus may be human immunodeficiency virus (HIV).

[0079] In one embodiment, the invention provides for the use of tenofovir alafenamide hemifumarate coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for treating a viral infection. The invention further provides for the use of tenofovir alafenamide hemifumarate coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for treating a viral infection in a human. Tenofovir alafenamide hemifumarate may be used in a subtherapeutic amount (or, in some embodiments throughout, in a therapeutic amount). Tenofovir alafenamide hemifumarate may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat may be used in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. Cobicistat may be used in an amount that provides a systemic exposure of tenofovir alafenamide hemifumarate comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir alafenamide hemifumarate in the absence of cobicistat in the manufacture of the medicament. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0080] In one embodiment, the invention provides for the use of tenofovir alafenamide hemifumarate coadministered with cobicistat, or a pharmaceutically acceptable salt thereof for the manufacture of a medicament for inhibiting activity of a retroviral reverse transcriptase. The invention further provides for the use of tenofovir alafenamide hemifumarate coadministered with cobicistat, or a pharmaceutically acceptable salt thereof, for the manufacture of a medicament for inhibiting activity of a retroviral reverse transcriptase in a human. Tenofovir alafenamide hemifumarate may be used in a subtherapeutic amount. Tenofovir alafenamide hemifumarate may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat may be used in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. Cobicistat may be used in an amount that provides a systemic exposure of tenofovir alafenamide hemifumarate comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir alafenamide hemifumarate in the absence of cobicistat in the manufacture of the medicament. The virus may be human immunodeficiency virus (HIV).

[0081] In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof, to prepare a medicament useful for improving the pharmacokinetics of tenofovir alafenamide hemifumarate following administration to a human. Tenofovir alafenamide hemifumarate may be used in a subtherapeutic amount. Tenofovir alafenamide hemifumarate may be used in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. Cobicistat may be used in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. Cobicistat may be used in an amount that provides a systemic exposure of tenofovir alafenamide hemifumarate comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir alafenamide hemifumarate in the absence of cobicistat in the manufacture of the medicament. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0082] In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament for a human useful for reducing a dose of tenofovir alafenamide hemifumarate by about 30-70% upon administration of the cobicistat. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0083] In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament for a human useful for reducing a dose of tenofovir alafenamide hemifumarate by about 2-4 fold upon administration of the cobicistat. In one embodiment, the invention provides for the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament for a human useful for reducing a dose of tenofovir alafenamide hemifumarate by about 3 fold upon administration of the cobicistat. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0084] In one embodiment, the invention provides for a method of treating a viral infection in a human comprising coadministering 1) tenofovir alafenamide hemifumarate; and 2) cobicistat, or a pharmaceutically acceptable salt thereof, to the human. Tenofovir alafenamide hemifumarate is administered in a subtherapeutic amount. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0085] In one embodiment, the invention provides for a use of a subtherapeutic dose of tenofovir alafenamide hemifumarate coadministered with cobicistat for treating a viral infection. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0086] In one embodiment, the invention provides for the use of a subtherapeutic dose of tenofovir alafenamide hemifumarate coadministered with cobicistat for inhibiting retroviral reverse transcriptase. The virus may be human immunodeficiency virus (HIV)

[0087] In one embodiment, the invention provides for an anti-virus agent(s) comprising (a) tenofovir alafenamide hemifumarate and (b) cobicistat, or a pharmaceutically acceptable salt thereof. The anti-virus agent(s) may include tenofovir alafenamide hemifumarate in amounts of 3 mg, 8±3 mg, 10±5 mg, 25±5 mg, or 40±10 mg or other ranges as set forth below. The anti-virus agent(s) may include cobicistat in amounts of 50-500 mg, 100-400 mg, 100-300 mg or 150 mg. The cobicistat may be used in an amount that provides a systemic exposure of tenofovir alafenamide hemifumarate comparable to the systemic exposure obtainable by administration of a greater dose of tenofovir alafenamide hemifumarate in the absence of cobicistat in the manufacture of the medicament. The anti-virus agent may further include 200 mg of emtricitabine and 150 mg of elvitegravir. The anti-virus agent may further include 150 mg cobicistat, 8 or less mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may further include 150 mg cobicistat, 25 or less mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may further include 150 mg cobicistat, 10 or less mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may include 150 mg cobicistat, 8 mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine. The anti-virus agent may include 150 mg cobicistat, 10 mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine.

[0088] In one embodiment, the invention provides for a unit-dosage of tenofovir alafenamide hemifumarate and cobicistat, or a pharmaceutically acceptable salt thereof, wherein the unit-dosage is a daily dose. Tenofovir alafenamide hemifumarate may be present in a subtherapeutic amount. The unit-dosage may further include 150 mg cobicistat, 8 or less mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine. The unit-dosage may further include 150 mg cobicistat, 25 or less mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine. The unit-dosage may further include 150 mg cobicistat, 10 or less mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine. The unit-dosage may include 150 mg cobicistat, 10 mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine.

[0089] In one embodiment, the invention provides the use of cobicistat, or a pharmaceutically acceptable salt thereof; to prepare a medicament useful for improving the pharmacokinetics of tenofovir alafenamide hemifumarate following administration to a human. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0090] In one embodiment, the invention provides cobicistat for use in improving the pharmacokinetics of tenofovir alafenamide hemifumarate following administration to a human. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0091] In one embodiment, the invention provides a kit comprising: (1) tenofovir alafenamide hemifumarate; (2) cobicistat, or a pharmaceutically acceptable salt thereof; (3) one or more containers; and (4) prescribing information regarding administering the tenofovir alafenamide hemifumarate with the cobicistat or a pharmaceutically acceptable salt thereof.

[0092] In one embodiment, the invention provides a kit comprising: (1) a unit dosage form comprising 5-100 mg of tenofovir alafenamide hemifumarate; (2) a unit dosage form comprising 150 mg cobicistat, or a pharmaceutically acceptable salt thereof; (3) one or more containers; and (4) prescribing information regarding administering the tenofovir alafenamide hemifumarate with cobicistat or a pharmaceutically acceptable salt thereof.

[0093] In one embodiment, the invention provides a use of tenofovir alafenamide hemifumarate for the manufacture of a medicament for inhibiting activity of a retroviral reverse transcriptase in a human, comprising administering tenofovir alafenamide hemifumarate and cobicistat, or a pharmaceutically acceptable salt thereof, to the human. The virus may be human immunodeficiency virus (HIV).

[0094] In one embodiment, the invention provides tenofovir alafenamide hemifumarate and cobicistat, or a pharmaceutically acceptable salt thereof, for use in inhibiting activity of a retroviral reverse transcriptase in a human.

[0095] In one embodiment, the invention provides a use of cobicistat, or a pharmaceutically acceptable salt thereof, to prepare a medicament for a human useful for reducing a dose between about 30-70% of tenofovir alafenamide hemifumarate upon administration of the cobicistat. The medicament may be used for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0096] In one embodiment, the invention provides the use of tenofovir alafenamide hemifumarate and cobicistat, or a pharmaceutically acceptable salt thereof, for the prophylactic or therapeutic treatment of a viral infection in a human. The use may be for the prophylactic or therapeutic treatment of a viral infection in a human. The virus may be human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0097] In one embodiment, the invention provides an antiviral agent(s) comprising (a) tenofovir alafenamide hemifumarate, which is used in combination with (b) cobicistat, or a pharmaceutically acceptable salt thereof, for use in the prophylactic or therapeutic treatment of a viral infection in a human.

[0098] In one embodiment, the invention provides for the use of ritonavir in the compositions, kits, unit-dosages and uses set forth above in place of cobicistat.

[0099] In one embodiment, the invention provides a method for inhibiting Pgp-mediated intestinal secretion of GS-7340, or a pharmaceutically acceptable salt thereof, in a human by coadministration of cobicistat, or a pharmaceutically acceptable salt thereof, with GS-7340, or a pharmaceutically acceptable salt thereof. In one embodiment, 150 mg of cobicistat, or a pharmaceutically acceptable salt thereof, is coadministered with 10 mg of GS-7340, or a pharmaceutically acceptable salt thereof.

[0100] In one embodiment, the invention provides a method for inhibiting Pgp-mediated intestinal secretion of tenofovir alafenamide hemifumarate in a human by coadministration of cobicistat, or a pharmaceutically acceptable salt thereof, with tenofovir alafenamide hemifumarate. In one embodiment, 150 mg of cobicistat, or a pharmaceutically acceptable salt thereof, is coadministered with 10 mg of tenofovir alafenamide hemifumarate.

[0101] In one embodiment, the invention provides the use of an anti-virus agent for the prophylactic or therapeutic treatment of a viral infection in a human, wherein the anti-virus agent comprises 150 mg cobicistat, 10 or less mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine.

[0102] In one embodiment, the invention provides a method of treating a viral infection in a human comprising coadministering 150 mg cobicistat, 10 or less mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine to the human.

[0103] In one embodiment, the invention provides the use of 150 mg cobicistat, 10 or less mg GS-7340, 150 mg elvitegravir, and 200 mg emtricitabine for the manufacture of a medicament for treating a viral infection in a human.

[0104] In one embodiment, the invention provides the use of an anti-virus agent for the prophylactic or therapeutic treatment of a viral infection in a human, wherein the anti-virus agent comprises 150 mg cobicistat, 10 or less mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine.

[0105] In one embodiment, the invention provides a method of treating a viral infection in a human comprising coadministering 150 mg cobicistat, 10 or less mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine to the human.

[0106] In one embodiment, the invention provides the use of 150 mg cobicistat, 10 or less mg tenofovir alafenamide hemifumarate, 150 mg elvitegravir, and 200 mg emtricitabine for the manufacture of a medicament for treating a viral infection in a human.

[0107] In one embodiment, the invention provides an anti-virus agent(s) comprising (a) tenofovir alafenamide hemifumarate, (b) cobicistat, or a pharmaceutically acceptable salt thereof, (c) emtricitabine, and (d) darunavir.

[0108] In one embodiment, the invention provides an anti-virus agent(s) comprising (a) 8 or less mg of tenofovir alafenamide hemifumarate, (b) 150 mg of cobicistat, or a pharmaceutically acceptable salt thereof, (c) 200 mg of emtricitabine, and (d) 800 mg of darunavir.

[0109] In one embodiment, the invention provides an anti-virus agent(s) comprising (a) 25 or less mg of tenofovir alafenamide hemifumarate, (b) 150 mg of cobicistat, or a pharmaceutically acceptable salt thereof, (c) 200 mg of emtricitabine, and (d) 800 mg of darunavir.

[0110] In one embodiment, the invention provides an anti-virus agent(s) comprising (a) 10 mg of tenofovir alafenamide hemifumarate, (b) 150 mg of cobicistat, or a pharmaceutically acceptable salt thereof, (c) 200 mg of emtricitabine, and (d) 800 mg of darunavir.

[0111] In one embodiment, the invention provides an anti-virus agent(s) comprising (a) GS-7340, or a pharmaceutically acceptable salt thereof, (b) cobicistat, or a pharmaceutically acceptable salt thereof, (c) emtricitabine, and (d) darunavir.

[0112] In one embodiment, the invention provides an anti-virus agent(s) comprising (a) 8 or less mg of GS-7340, or a pharmaceutically acceptable salt thereof, (b) 150 mg of cobicistat, or a pharmaceutically acceptable salt thereof, (c) 200 mg of emtricitabine, and (d) 800 mg of darunavir.

[0113] In one embodiment, the invention provides an anti-virus agent(s) comprising (a) 25 or less mg of GS-7340, or a pharmaceutically acceptable salt thereof, (b) 150 mg of cobicistat, or a pharmaceutically acceptable salt thereof, (c) 200 mg of emtricitabine, and (d) 800 mg of darunavir.

[0114] In one embodiment, the invention provides an anti-virus agent(s) comprising (a) 10 mg of GS-7340, or a pharmaceutically acceptable salt thereof, (b) 150 mg of cobicistat, or a pharmaceutically acceptable salt thereof, (c) 200 mg of emtricitabine, and (d) 800 mg of darunavir.

[0115] In one embodiment, the invention provides the use of an anti-virus agent for the prophylactic or therapeutic treatment of a viral infection in a human, wherein the anti-virus agent comprises 150 mg cobicistat, 10 or less mg GS-7340, 800 mg of darunavir, and 200 mg emtricitabine.

[0116] In one embodiment, the invention provides a method of treating a viral infection in a human comprising coadministering 150 mg cobicistat, 10 or less mg GS-7340, 800 mg of darunavir, and 200 mg emtricitabine to the human.

[0117] In one embodiment, the invention provides the use of 150 mg cobicistat, 10 or less mg GS-7340, 800 mg of darunavir, and 200 mg emtricitabine for the manufacture of a medicament for treating a viral infection in a human.

[0118] In one embodiment, the invention provides the use of an anti-virus agent for the prophylactic or therapeutic treatment of a viral infection in a human, wherein the anti-virus agent comprises 150 mg cobicistat, 10 or less mg tenofovir alafenamide hemifumarate, 800 mg of darunavir, and 200 mg emtricitabine.

[0119] In one embodiment, the invention provides a method of treating a viral infection in a human comprising coadministering 150 mg cobicistat, 10 or less mg tenofovir alafenamide hemifumarate, 800 mg of darunavir, and 200 mg emtricitabine to the human.

[0120] In one embodiment, the invention provides the use of 150 mg cobicistat, 10 or less mg tenofovir alafenamide hemifumarate, 800 mg of darunavir, and 200 mg emtricitabine for the manufacture of a medicament for treating a viral infection in a human.

[0121] In one embodiment, the invention provides the use of a dose of a cytochrome p450 inhibitor, or a pharmaceutically acceptable salt thereof, to boost a dose GS-7340, or a pharmaceutically acceptable salt thereof, for the prophylactic or therapeutic treatment of a viral infection in a human. In one embodiment, the cytochrome p450 inhibitor is cobicistat, or a pharmaceutically acceptable salt thereof. In one further embodiment, the dose of GS-7340 would be a subtherapeutic amount absent the dose of cobicistat.

[0122] In one embodiment, the invention provides a composition comprising: a unit-dosage form of GS-7340, or a pharmaceutically acceptable salt thereof; a unit-dosage form of cobicistat, or a pharmaceutically acceptable salt thereof; and a pharmaceutically acceptable carrier or diluent, wherein the amount of GS-7340 in the unit-dosage form is a subtherapeutic amount.

[0123] In one embodiment, the invention provides the use of a dose of a cytochrome p450 inhibitor, or a pharmaceutically acceptable salt thereof, to boost a dose tenofovir alafenamide hemifumarate for the prophylactic or therapeutic treatment of a viral infection in a human. In one embodiment, the cytochrome p450 inhibitor is cobicistat, or a pharmaceutically acceptable salt thereof. In one further embodiment, the

dose of tenofovir alafenamide hemifumarate would be a subtherapeutic amount absent the dose of cobicistat.

[0124] In one embodiment, the invention provides a composition comprising: a unit-dosage form of tenofovir alafenamide hemifumarate; a unit-dosage form of cobicistat, or a pharmaceutically acceptable salt thereof; and a pharmaceutically acceptable carrier or diluent, wherein the amount of tenofovir alafenamide hemifumarate in the unit-dosage form is a subtherapeutic amount.

[0125] In one embodiment, the invention provides the uses and methods related to treating a viral infection, as noted herein, wherein the viral infection is human immunodeficiency virus (HIV).

[0126] In one embodiment, the invention provides the uses and methods related to treating a viral infection, as noted herein, wherein the viral infection is Hepatitis B virus (HBV).

[0127] In one embodiment, the invention provides a method of treating a viral infection in a human, comprising administering to the human a composition comprising cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, wherein the composition contains an amount of cobicistat, or a pharmaceutically acceptable salt thereof, sufficient for an amount of tenofovir alafenamide hemifumarate in the composition to provide an effect on the viral infection that is greater than the effect of the amount of tenofovir alafenamide hemifumarate in the absence of cobicistat, or a pharmaceutically acceptable salt thereof, and wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0128] In one embodiment, the invention provides a method of treating a viral infection in a human, comprising administering to the human a composition comprising cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, wherein an effect on the viral infection of an amount of tenofovir alafenamide hemifumarate in the composition is greater than the effect of the same amount of tenofovir alafenamide hemifumarate in the absence of cobicistat, or a pharmaceutically acceptable salt thereof, and wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0129] In one embodiment, the invention provides an anti-viral treatment method on a viral infection in a human, comprising administering to the human a composition comprising cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, wherein the composition contains an amount of cobicistat, or a pharmaceutically acceptable salt thereof, sufficient for an amount of tenofovir alafenamide hemifumarate in the composition to provide an anti-viral effect that is greater than the anti-viral effect of the amount of tenofovir alafenamide hemifumarate in the absence of cobicistat, or a pharmaceutically acceptable salt thereof, and wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0130] In one embodiment, the invention provides an anti-viral treatment method on a viral infection in a human, comprising administering to the human a composition comprising cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, wherein an anti-viral effect of an amount of tenofovir alafenamide hemifumarate in the composition is greater than the anti-viral effect of the same amount of tenofovir alafenamide hemifumarate in the absence of cobicistat, or a pharmaceutically acceptable salt thereof, and wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0131] In one embodiment, the invention provides a composition comprising: cobicistat, or a pharmaceutically acceptable salt thereof; and tenofovir alafenamide hemifumarate. In a further embodiment, the composition comprises: 50-500 mg of cobicistat, or a pharmaceutically acceptable salt thereof; and 3-40 mg of tenofovir alafenamide hemifumarate. In another embodiment, the composition further comprises a pharmaceutically acceptable carrier or diluent.

[0132] In one embodiment, the invention provides a method of treating a viral infection in a human comprising administering a composition comprising: cobicistat, or a pharmaceutically acceptable salt thereof; and tenofovir alafenamide hemifumarate, to the human.

[0133] In one embodiment, the invention provides a method of treating a viral infection in a human comprising coadministering cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, to the human.

[0134] In one embodiment, the invention provides a method of inhibiting activity of a retroviral reverse transcriptase comprising coadministering cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate. In a further embodiment, the coadministering of cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, is in a human.

[0135] In one embodiment, the invention provides use of cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, for the prophylactic or therapeutic treatment of a viral infection in a human.

[0136] In one embodiment, the invention provides use of cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, for the manufacture of a medicament for treating a viral infection in a human.

[0137] In one embodiment, the invention provides use of cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, for the manufacture of a medicament for inhibiting activity of a retroviral reverse transcriptase. In a further embodiment, the medicament is for inhibiting activity of a retroviral reverse transcriptase in a human.

[0138] In one embodiment, the invention provides a method of boosting an anti-viral effect of tenofovir alafenamide hemifumarate in a human comprising administering a composition comprising: cobicistat, or a pharmaceutically acceptable salt thereof; and tenofovir alafenamide hemifumarate, to the human.

[0139] In one embodiment, the invention provides a method of boosting an anti-viral effect of tenofovir alafenamide hemifumarate in a human comprising coadministering cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate to the human. In a further embodiment, 50-500 mg of cobicistat, or a pharmaceutically acceptable salt thereof, is coadministered with 3-40 mg of tenofovir alafenamide hemifumarate.

[0140] In one embodiment, the invention provides a method of inhibiting Pgp-mediated intestinal secretion of tenofovir alafenamide hemifumarate in a human comprising administering a composition comprising: cobicistat, or a pharmaceutically acceptable salt thereof; and tenofovir alafenamide hemifumarate, to the human.

[0141] In one embodiment, the invention provides a method of inhibiting Pgp-mediated intestinal secretion of tenofovir alafenamide hemifumarate in a human by coadmin-

istration of cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate. In a further embodiment, 50-500 mg of cobicistat, or a pharmaceutically acceptable salt thereof, is coadministered with 3-40 mg of tenofovir alafenamide hemifumarate.

[0142] In additional embodiments, the invention provides the methods and uses disclosed wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0143] In one embodiment, the invention provides a composition comprising: (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) elvitegravir. In a further embodiment, the composition comprises: (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 50-500 mg cobicistat, or a pharmaceutically acceptable salt thereof; (c) 50-500 mg emtricitabine; and (d) 50-500 mg elvitegravir. In a further embodiment, the invention provides a method of treating a viral infection in a human comprising administering such a composition to the human.

[0144] In one embodiment, the invention provides a method of treating a viral infection in a human comprising coadministering (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) elvitegravir to the human. In a further embodiment, the method comprises coadministering (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 50-500 mg cobicistat, or a pharmaceutically acceptable salt thereof; (c) 50-500 mg emtricitabine; and (d) 50-500 mg elvitegravir to the human.

[0145] In one embodiment, the invention provides use of a composition comprising: (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) elvitegravir, for the prophylactic or therapeutic treatment of a viral infection in a human.

[0146] In one embodiment, the invention provides use of (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) elvitegravir for the manufacture of a medicament for treating a viral infection in a human. In a further embodiment, the invention provides use of (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 50-500 mg cobicistat, or a pharmaceutically acceptable salt thereof; (c) 50-500 mg emtricitabine; and (d) 50-500 mg elvitegravir for the manufacture of a medicament for treating a viral infection in a human.

[0147] In one embodiment, the invention provides a composition comprising: (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) elvitegravir for the treatment of a viral infection, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0148] In one embodiment, the invention provides a composition comprising: (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 50-500 mg cobicistat, or a pharmaceutically acceptable salt thereof; (c) 50-500 mg emtricitabine; and (d) 50-500 mg elvitegravir for the treatment of a viral infection, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0149] In additional embodiments, the invention provides the methods and uses disclosed wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0150] In one embodiment, the invention provides a composition comprising: (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) darunavir. In a further embodiment, the composition comprises: (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 50-500 mg cobicistat, or a pharmaceutically acceptable salt thereof; (c) 50-500 mg emtricitabine; and (d) 400-1600 mg darunavir. In a further embodiment, the invention provides a method of treating a viral infection in a human comprising administering such a composition to the human.

[0151] In one embodiment, the invention provides a method of treating a viral infection in a human comprising coadministering (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) darunavir to the human. In a further embodiment, the method comprises coadministering (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 50-500 mg cobicistat, or a pharmaceutically acceptable salt thereof; (c) 50-500 mg emtricitabine; and (d) 400-1600 mg darunavir to the human.

[0152] In one embodiment, the invention provides use of a composition comprising: (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) darunavir, for the prophylactic or therapeutic treatment of a viral infection in a human.

[0153] In one embodiment, the invention provides use of (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) darunavir for the manufacture of a medicament for treating a viral infection in a human. In a further embodiment, the invention provides use of (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 50-500 mg cobicistat, or a pharmaceutically acceptable salt thereof; (c) 50-500 mg emtricitabine; and (d) 400-1600 mg darunavir for the manufacture of a medicament for treating a viral infection in a human.

[0154] In one embodiment, the invention provides a composition comprising: (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) darunavir for the treatment of a viral infection, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0155] In one embodiment, the invention provides a composition comprising: (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 50-500 mg cobicistat, or a pharmaceutically acceptable salt thereof; (c) 50-500 mg emtricitabine; and (d) 400-1600 mg darunavir for the treatment of a viral infection, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0156] In additional embodiments, the invention provides the methods and uses disclosed wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0157] In one embodiment, the invention provides a composition comprising: tenofovir alafenamide hemifumarate and emtricitabine. In a further embodiment, the composition comprises: 3-40 mg tenofovir alafenamide hemifumarate and 50-500 mg emtricitabine. In a further embodiment, the invention provides a method of treating a viral infection in a human comprising administering such a composition to the human.

[0158] In one embodiment, the invention provides a method of treating a viral infection in a human comprising coadministering tenofovir alafenamide hemifumarate and emtricitabine to the human. In a further embodiment, the method comprises coadministering 3-40 mg tenofovir alafenamide hemifumarate and 50-500 mg emtricitabine to the human.

[0159] In one embodiment, the invention provides use of a composition comprising: tenofovir alafenamide hemifumarate and emtricitabine for the prophylactic or therapeutic treatment of a viral infection in a human.

[0160] In one embodiment, the invention provides use of tenofovir alafenamide hemifumarate and emtricitabine for the manufacture of a medicament for treating a viral infection in a human. In a further embodiment, the invention provides use of 3-40 mg tenofovir alafenamide hemifumarate and 50-500 mg emtricitabine for the manufacture of a medicament for treating a viral infection in a human.

[0161] In one embodiment, the invention provides a composition comprising: tenofovir alafenamide hemifumarate and emtricitabine for the treatment of a viral infection, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0162] In one embodiment, the invention provides a composition comprising: 3-40 mg tenofovir alafenamide hemifumarate and 50-500 mg emtricitabine for the treatment of a viral infection, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0163] In additional embodiments, the invention provides the methods and uses disclosed wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0164] In one embodiment, the invention provides a composition comprising: (a) tenofovir alafenamide hemifumarate; (b) rilpivirine; and (c) emtricitabine. In a further embodiment, the composition comprises: (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 10-80 mg rilpivirine; and (c) 50-500 mg emtricitabine. In a further embodiment, the invention provides a method of treating a viral infection in a human comprising administering such a composition to the human.

[0165] In one embodiment, the invention provides a method of treating a viral infection in a human comprising coadministering (a) tenofovir alafenamide hemifumarate; (b) rilpivirine; and (c) emtricitabine to the human. In a further embodiment, the method comprises coadministering (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 10-80 mg rilpivirine; and (c) 50-500 mg emtricitabine to the human.

[0166] In one embodiment, the invention provides use of a composition comprising: (a) tenofovir alafenamide hemifumarate; (b) rilpivirine; and (c) emtricitabine, for the prophylactic or therapeutic treatment of a viral infection in a human.

[0167] In one embodiment, the invention provides use of (a) tenofovir alafenamide hemifumarate; (b) rilpivirine; and (c) emtricitabine for the manufacture of a medicament for treating a viral infection in a human. In a further embodiment, the invention provides use of (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 10-80 mg rilpivirine; and (c) 50-500 mg emtricitabine for the manufacture of a medicament for treating a viral infection in a human.

[0168] In one embodiment, the invention provides a composition comprising: (a) tenofovir alafenamide hemifumarate; (b) rilpivirine; and (c) emtricitabine for the treatment of a viral infection, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0169] In one embodiment, the invention provides a composition comprising: (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 10-80 mg rilpivirine; and (c) 50-500 mg emtricitabine for the treatment of a viral infection, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0170] In additional embodiments, the invention provides the methods and uses disclosed wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0171] In one embodiment, the invention provides a composition comprising: tenofovir alafenamide hemifumarate and GS-9441. In a further embodiment, the composition comprises: 3-40 mg tenofovir alafenamide hemifumarate and 5-1500 mg GS-9441. In a further embodiment, the invention provides a method of treating a viral infection in a human comprising administering such a composition to the human.

[0172] In one embodiment, the invention provides a method of treating a viral infection in a human comprising coadministering tenofovir alafenamide hemifumarate and GS-9441 to the human. In a further embodiment, the method comprises coadministering 3-40 mg tenofovir alafenamide hemifumarate and 5-1500 mg GS-9441 to the human.

[0173] In one embodiment, the invention provides use of a composition comprising: tenofovir alafenamide hemifumarate and GS-9441 for the prophylactic or therapeutic treatment of a viral infection in a human.

[0174] In one embodiment, the invention provides use of tenofovir alafenamide hemifumarate and GS-9441 for the manufacture of a medicament for treating a viral infection in a human. In a further embodiment, the invention provides use of 3-40 mg tenofovir alafenamide hemifumarate and 5-1500 mg GS-9441 for the manufacture of a medicament for treating a viral infection in a human.

[0175] In one embodiment, the invention provides a composition comprising: tenofovir alafenamide hemifumarate and GS-9441 for the treatment of a viral infection, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0176] In one embodiment, the invention provides a composition comprising: 3-40 mg tenofovir alafenamide hemifumarate and 5-1500 mg GS-9441 for the treatment of a viral infection, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

[0177] In additional embodiments, the invention provides the methods and uses disclosed wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

BRIEF DESCRIPTION OF THE DRAWINGS

[0178] FIG. **1** shows pharmacokinetic data from patients dosed with various doses of GS-7340 and TDF.

[0179] FIG. **2** shows pharmacokinetic data from patients dosed with various doses of GS-7340 and TDF.

[0180] FIG. **3**A-B shows pharmacokinetic data from patients dosed with various formulations of GS-7340.

[0181] FIG. **4**A-B shows pharmacokinetic data from patients dosed with various formulations of GS-7340.

[0182] FIG. **5**A-B shows pharmacokinetic data from patients dosed with various formulations of GS-7340.

[0183] FIG. **6** shows pharmacokinetic data from patients dosed with various formulations of GS-7340.

[0184] FIG. **7** shows pharmacokinetic data from patients dosed with various formulations of GS-7340.

[0185] FIG. **8** shows pharmacokinetic data from patients dosed with various formulations of GS-7340.

[0186] FIG. **9** shows pharmacokinetic data from patients dosed with various formulations of GS-7340.

[0187] FIG. **10**A-B shows results of substrate assays in cells transfected with the genes for human P-glycoprotein (Pgp; MDR1) and breast cancer resistance protein (BCRP) genes.

[0188] FIG. **11**A-B shows results of bidirectional permeability assays in cells transfected with the genes for human Pgp and BCRP.

[0189] FIG. **12**A-F shows results of bidirectional permeability assays in cells transfected with the genes for human Pgp and BCRP.

[0190] FIG. **13** shows the X-ray powder diffraction (XRPD) pattern of tenofovir alafenamide hemifumarate.

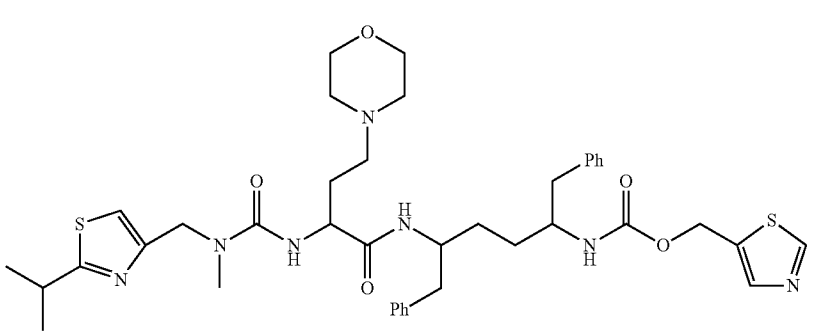
[0191] FIG. **14** shows a graph of the DSC analysis of tenofovir alafenamide hemifumarate.

[0192] FIG. **15** shows a graph of the thermogravimetric analysis (TGA) data for tenofovir alafenamide hemifumarate.

[0193] FIG. **16** shows a graph of the dynamic vapor sorption (DVS) analysis of tenofovir alafenamide hemifumarate.

DETAILED DESCRIPTION OF THE INVENTION

[0194]

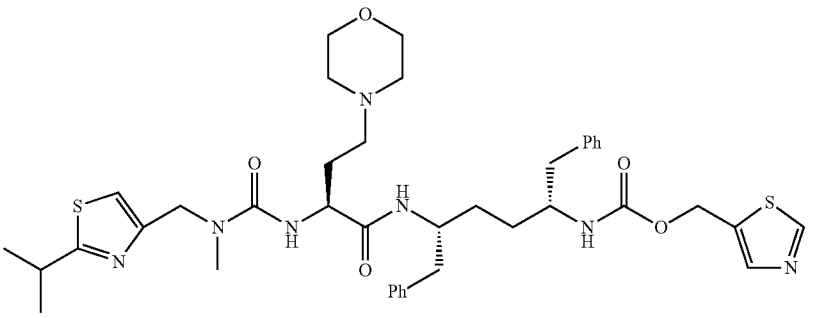


(I)

[0195] Cobicistat (chemical name 1,3-thiazol-5-ylmethyl (2R,5R)-(5-{[(2S)-2-[(methyl {[2-(propan-2-yl)-1,3-thiazol-4-yl]methyl}carbamoyl)amino]]-4-(morpholin-4-yl)butanamido}-1,6-diphenylhexan-2-yl)carbamate) is a chemical entity that has been shown to be a mechanism-based inhibitor that irreversibly inhibits CYP3A enzymes.

[0196] Detailed enzyme inactivation kinetic studies were performed comparing cobicistat with ritonavir. Cobicistat was found to be an efficient inactivator of human hepatic microsomal CYP3A activity with kinetic parameters similar to those of ritonavir. In addition, cobicistat is a moderate inhibitor of CYP2B6 (similar potency to ritonavir), a weak inhibitor of CYP2D6, and does not appreciably inhibit CYP1A2, CYP2C8, CYP2C9, CYP2C19, or uridine glucuronosyltransferase 1A1. In xenobiotic receptor transactivation and human hepatocyte studies, cobicistat displayed no/weak potential as an inducer of cytochrome P450, UGT1A1, or P-glycoprotein (at up to 30 μM). Permeability assays suggest that cobicistat is not a strong substrate or inhibitor of transporters including P-glycoprotein, MRP1, and MRP2. Inhibition of intestinal P-glycoprotein by cobicistat is only possible during absorption due to its high aqueous solubility, but it is not potent enough to inhibit transporters at systemic concentrations. These data indicate that, compared to ritonavir, cobicistat is a more selective inhibitor of CYP3A in vitro and a weaker inducer of CYP enzymes, which may potentially result in fewer clinically significant interactions with substrates of other CYP enzymes.

[0197] Cobicistat may also be present in compositions enriched with a stereoisomer of formula (Ia):



(Ia)

which is thiazol-5-ylmethyl (2R,5R)-5-((S)-2-(3-((2-isopropylthiazol-5-yl)methyl)-3-methylureido)-4-morpholinobutanamido)-1,6-diphenylhexan-2-ylcarbamate.

[0198] In one embodiment, the cobicistat has an enriched concentration of 85±5% of the stereoisomer of formula (Ia). In another embodiment, the cobicistat has an enriched concentration of 90±5% of the stereoisomer of formula (Ia). In another embodiment, the cobicistat has an enriched concentration of 95±2% of the stereoisomer of formula (Ia). In another embodiment, the cobicistat has an enriched concentration of 99±1% of the stereoisomer of formula (Ia). In another embodiment, the cobicistat is present as the pure stereoisomer of formula (Ia).

[0199] Coadministration of cobicistat with GS-7340 or tenofovir alafenamide hemifumarate boosts systemic exposure to GS-7340 or tenofovir alafenamide hemifumarate in humans, improves the pharmacokinetics of GS-7340 or tenofovir alafenamide hemifumarate (including, but not limited to, C_{max} increases), and increases blood levels of GS-7340/tenofovir alafenamide hemifumarate/tenofovir. Therefore, GS-7340 or tenofovir alafenamide hemifumarate coadministered with cobicistat may be administered in lower amounts than previously thought to achieve a therapeutic effect. Such

lower amounts may be amounts that would be subtherapeutic in the absence of coadministration of cobicistat.

[0200] Without being bound by any theory of the invention, it is believed that cobicistat may be acting to inhibit intestinal Pgp-mediated intestinal secretion of GS-7340 or tenofovir alafenamide hemifumarate. In in vitro studies, cobicistat and ritonavir significantly increased the accumulation of probe substrates (such as calcein AM and Hoechst 33342) in cells transfected with P-glycoprotein (Pgp) and breast cancer resistance protein (BCRP), and cobicistat was found to be a substrate for these transporters. Cobicistat appears to be a substrate of Pgp and BCRP and likely has a competitive mode of inhibition with coadministered agents. Cobicistat appears to be a relatively weak inhibitor of Pgp and BCRP and may only have a transient effect on these transporters during intestinal absorption, facilitated by high solubility of, and resulting high concentrations of, cobicistat achievable in the gastrointestinal tract. Combined, these results suggest that cobicistat can effectively inhibit intestinal transporters and increase the absorption of coadministered substrates, including HIV protease inhibitors and GS-7340 or tenofovir alafenamide hemifumarate, contributing to its effectiveness as a pharmacoenhancer.

[0201] As used herein, the term "coadminister" (or "coadministration") refers to administration of two or more agents within a 24-hour period of each other, for example, as part of a clinical treatment regimen. In other embodiments, "coadminister" refers to administration of two or more agents within 2 hours of each other. In other embodiments, "coadminister" refers to administration of two or more agents within 30 minutes of each other. In other embodiments, "coadminister" refers to administration of two or more agents within 15 minutes of each other. In other embodiments, "coadminister" refers to administration of two or more agents at the same time, either as part of a single formulation or as multiple formulations that are administered by the same or different routes.

[0202] The term "unit dosage form" refers to a physically discrete unit, such as a capsule, tablet, or solution, that is suitable as a unitary dosage for a human patient, each unit containing a predetermined quantity of one or more active ingredient(s) calculated to produce a therapeutic effect, in association with at least one pharmaceutically acceptable diluent or carrier, or combination thereof. Unit dosage formulations contain a daily dose or unit daily subdose or an appropriate fraction thereof, of the active ingredient(s).

[0203] The term "subtherapeutic amount" of a compound is any amount of the compound that upon dosing is insufficient to achieve the desired therapeutic benefit.

[0204] The term "boosting amount" or "boosting dose" is the amount of a compound needed to improve the pharmacokinetics of a second compound (or increase availability or exposure). The boosting amount or boosting dose may improve the pharmacokinetics (or increase availability or exposure) of the second compound to a level that is therapeutic in a subject. In other words, a subtherapeutic amount of the second compound (i.e., subtherapeutic when administered without coadministration of the boosting amount) reaches a therapeutic level(s) in a subject due to improved pharmacokinetics (or increased availability or exposure) upon coadministration of the boosting amount.

[0205] The present invention also provides a method for the treatment or prophylaxis of diseases, disorders, and conditions. An example of a disease, disorder, or condition includes, but is not limited to, a retrovirus infection, or a disease, disorder, or condition associated with a retrovirus infection. Retroviruses are RNA viruses and are generally classified into the alpharetrovirus, betaretrovirus, deltaretrovirus, epsilonretrovirus, gammaretrovirus, lentivirus, and spumavirus families. Examples of retroviruses include, but are not limited to, human immunodeficiency virus (HIV), human T-lymphotrophic virus (HTLV), rous sarcoma virus (RSV), and the avian leukosis virus. In general, three genes of the retrovirus genome code for the proteins of the mature virus: gag (group-specific antigen) gene, which codes for the core and structural proteins of the virus; pol (polymerase) gene, which codes for the enzymes of the virus, including reverse transcriptase, protease, and integrase; and env (envelope) gene, which codes for the retrovirus surface proteins.

[0206] Retroviruses attach to and invade a host cell by releasing a complex of RNA and the pol products, among other things, into the host cell. The reverse transcriptase then produces double-stranded DNA from the viral RNA. The double-stranded DNA is imported into the nucleus of the host cell and integrated into the host cell genome by the viral integrase. A nascent virus from the integrated DNA is formed when the integrated viral DNA is converted into mRNA by the host cell polymerase, and the proteins necessary for virus formation are produced by the action of the virus protease. The virus particle undergoes budding and is released from the host cell to form a mature virus.

[0207] The active agents may be administered to a human in any conventional manner. While it is possible for the active agents to be administered as raw compounds, they are preferably administered as a pharmaceutical composition. The salt, carrier, or diluent should be acceptable in the sense of being compatible with the other ingredients and not deleterious to the recipient thereof. Examples of carriers or diluents for oral administration include cornstarch, lactose, magnesium stearate, talc, microcrystalline cellulose, stearic acid, povidone, crospovidone, dibasic calcium phosphate, sodium starch glycolate, hydroxypropyl cellulose (e.g., low substituted hydroxypropyl cellulose), hydroxypropylmethyl cellulose (e.g., hydroxypropylmethyl cellulose 2910), and sodium lauryl sulfate.

[0208] The pharmaceutical compositions may be prepared by any suitable method, such as those methods well known in the art of pharmacy, for example, methods such as those described in Gennaro et al., Remington's Pharmaceutical Sciences (18th ed., Mack Publishing Co., 1990), especially Part 8: Pharmaceutical Preparations and their Manufacture. Such methods include the step of bringing into association GS-7340 or tenofovir alafenamide hemifumarate with the carrier or diluent and optionally one or more accessory ingredients. Such accessory ingredients include those conventional in the art, such as, fillers, binders, excipients, disintegrants, lubricants, colorants, flavoring agents, sweeteners, preservatives (e.g., antimicrobial preservatives), suspending agents, thickening agents, emulsifying agents, and/or wetting agents.

[0209] The term "GS-7340, or pharmaceutically acceptable salt thereof" or the like includes any amorphous, crystalline, co-crystalline, complex, or other physical form thereof. In one embodiment, a composition comprising a pharmaceutically acceptable coformer and GS-7340 is administered. The pharmaceutically acceptable coformer can be any pharmaceutically acceptable compound that is capable of forming a "pharmaceutically acceptable salt" with

GS-7340. For example, the pharmaceutically acceptable coformer can be a pharmaceutically acceptable acid (e.g. adipic acid, L-aspartic acid, citric acid, fumaric acid, maleic acid, malic acid, malonic acid, succinic acid, tartaric acid, or oxalic acid). In one embodiment of the invention, the pharmaceutically acceptable coformer is a bis-acid. In another embodiment, the pharmaceutically acceptable coformer is fumaric acid. In another embodiment, a composition comprising a coformer and GS-7340 in a ratio of about 0.5±0.05 can be administered. One form of GS-7340 is a hemifumarate form (tenofovir alafenamide hemifumarate), as described further herein.

[0210] The pharmaceutical compositions may provide controlled, slow release or sustained release of the agents (e.g., GS-7340 or tenofovir alafenamide hemifumarate) over a period of time. The controlled, slow release or sustained release of the agents (e.g., GS-7340 or tenofovir alafenamide hemifumarate) may maintain the agents in the bloodstream of the human for a longer period of time than with conventional formulations. Pharmaceutical compositions include, but are not limited to, coated tablets, pellets, solutions, powders, capsules, and dispersions of GS-7340 or tenofovir alafenamide hemifumarate in a medium that is insoluble in physiologic fluids, or where the release of the therapeutic compound follows degradation of the pharmaceutical composition due to mechanical, chemical, or enzymatic activity.

[0211] The pharmaceutical compositions of the invention may be, for example, in the form of a pill, capsule, solution, powder, or tablet, each containing a predetermined amount of GS-7340 or tenofovir alafenamide hemifumarate. In an embodiment of the invention, the pharmaceutical composition is in the form of a tablet comprising GS-7340 or tenofovir alafenamide hemifumarate. In another embodiment of the invention, the pharmaceutical composition is in the form of a tablet comprising GS-7340 and the components of the tablet utilized and described in the Examples provided herein.

[0212] For oral administration, fine powders or granules may contain diluting, dispersing, and or surface active agents and may be present, for example, in water or in a syrup, in capsules or sachets in the dry state, or in a nonaqueous solution or suspension wherein suspending agents may be included, or in tablets wherein binders and lubricants may be included.

[0213] When administered in the form of a liquid solution or suspension, the formulation may contain GS-7340 or tenofovir alafenamide hemifumarate and purified water. Optional components in the liquid solution or suspension include suitable sweeteners, flavoring agents, preservatives (e.g., antimicrobial preservatives), buffering agents, solvents, and mixtures thereof. A component of the formulation may serve more than one function. For example, a suitable buffering agent also may act as a flavoring agent as well as a sweetener.

[0214] Suitable sweeteners include, for example, saccharin sodium, sucrose, and mannitol. A mixture of two or more sweeteners may be used. The sweetener or mixtures thereof are typically present in an amount of from about 0.001% to about 70% by weight of the total composition. Suitable flavoring agents may be present in the pharmaceutical composition to provide a cherry flavor, cotton candy flavor, or other suitable flavor to make the pharmaceutical composition easier for a human to ingest. The flavoring agent or mixtures thereof are typically present in an amount of about 0.0001% to about 5% by weight of the total composition.

[0215] Suitable preservatives include, for example, methylparaben, propylparaben, sodium benzoate, and benzalkonium chloride. A mixture of two or more preservatives may be used. The preservative or mixtures thereof are typically present in an amount of about 0.0001% to about 2% by weight of the total composition.

[0216] Suitable buffering agents include, for example, citric acid, sodium citrate, phosphoric acid, potassium phosphate, and various other acids and salts. A mixture of two or more buffering agents may be used. The buffering agent or mixtures thereof are typically present in an amount of about 0.001% to about 4% by weight of the total composition.

[0217] Suitable solvents for a liquid solution or suspension include, for example, sorbitol, glycerin, propylene glycol, and water. A mixture of two or more solvents may be used. The solvent or solvent system is typically present in an amount of about 1% to about 90% by weight of the total composition.

[0218] The pharmaceutical composition may be coadministered with adjuvants. For example, nonionic surfactants such as polyoxyethylene oleyl ether and n-hexadecyl polyethylene ether may be administered with or incorporated into the pharmaceutical composition to artificially increase the permeability of the intestinal walls. Enzymatic inhibitors may also be administered with or incorporated into the pharmaceutical composition.

GS-7340

[0219] In one embodiment of the invention, a dose of 3 mg, 3±2 mg, or 3±1 mg of GS-7340, or a pharmaceutically acceptable salt thereof, is administered.

[0220] In one embodiment of the invention, a dose of 8±3 mg, 8±2 mg or 8±1 mg of GS-7340, or a pharmaceutically acceptable salt thereof, is administered.

[0221] In one embodiment of the invention, a unit dosage form comprises a dose of 8±2 mg of GS-7340, or a pharmaceutically acceptable salt thereof.

[0222] In various embodiments of the invention, a dose of 8±3 mg; 25±10 mg; 10±5 mg; 25±5 mg; 25±2 mg; 40±10 mg; 40±5 mg; 40±2 mg; 60±20 mg; 60±10 mg; 100±20 mg; 100±10 mg; 125±20 mg; 125±10 mg; 150±20 mg; 150±10 mg; 200±40 mg; or 200±15 mg of GS-7340, or a pharmaceutically acceptable salt thereof, is administered.

[0223] The desired daily dose of GS-7340 also may be administered as two, three, four, five, six, or more subdoses administered separately at appropriate intervals throughout the day, optionally, in unit dosage forms.

[0224] The concentration of tenofovir/GS-7340 in the bloodstream may be measured as the plasma concentration (e.g., ng/mL). Pharmacokinetic parameters for determining the plasma concentration include, but are not limited to, the maximum observed plasma concentration (C_{max}), observed plasma concentration at the end of the dosing interval or "trough" concentration (C_{tau} or C_{min}), area under the plasma concentration time curve (AUC) from time zero up to the last quantifiable time point ($AUC_{0\text{-}last}$), AUC from time zero to infinity ($AUC_{0\text{-}inf}$), AUC over the dosing interval (AUC_{tau}), time of maximum observed plasma concentration after administration (t_{max}), and half-life of GS-7340 in plasma ($t_{1/2}$).

[0225] Administration of GS-7340 with food according to the methods of the invention may also increase absorption of GS-7340. Absorption of GS-7340 may be measured by the concentration attained in the bloodstream over time after

administration of GS-7340. An increase in absorption by administration of GS-7340 with food may also be evidenced by an increase in C_{max} and/or AUC of GS-7340 as compared to the values if GS-7340 was administered without food. Typically protease inhibitors are administered with food.

Tenofovir Alafenamide Hemifumarate

[0226] In one embodiment, there is provided a hemifumarate form of tenofovir alafenamide (i.e., tenofovir alafenamide hemifumarate). This form may have a ratio (i.e., a stoichiometric ratio or mole ratio) of fumaric acid to tenofovir alafenamide of 0.5±0.1, 0.5±0.05, 0.5±0.01, or about 0.5, or the like.

[0227] In one embodiment, tenofovir alafenamide hemifumarate consists of fumaric acid and tenofovir alafenamide in a ratio of 0.5±0.1.

[0228] In one embodiment, tenofovir alafenamide hemifumarate consists essentially of fumaric acid and tenofovir alafenamide in a ratio of 0.5±0.1.

[0229] In one embodiment, tenofovir alafenamide hemifumarate has an XRPD pattern comprising 2theta values of 6.9±0.2°, 8.6±0.2°, 10.0±0.2°, 11.0±0.2°, 12.2±0.2°, 15.9±0.2°, 16.3±0.2°, 20.2±0.2°, and 20.8±0.2°.

[0230] In one embodiment, tenofovir alafenamide hemifumarate has an XRPD pattern comprising at least four 2theta values selected from 6.9±0.2°, 8.6±0.2°, 10.0±0.2°, 11.0±0.2°, 12.2±0.2°, 15.9±0.2°, 16.3±0.2°, 20.2±0.2°, and 20.8±0.2°.

[0231] In one embodiment, tenofovir alafenamide hemifumarate has a DSC onset endotherm of 131±2° C., or 131±1° C.

[0232] In various embodiments, a tenofovir alafenamide hemifumarate composition comprises less than about 5%; 1%; or 0.5% by weight of tenofovir alafenamide monofumarate.

[0233] In one embodiment, a tenofovir alafenamide hemifumarate composition comprises no detectable tenofovir alafenamide monofumarate.

[0234] Tenofovir alafenamide (i.e., the compound 9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine) can be prepared as described in U.S. Pat. No. 7,390,791.

[0235] In various embodiments of the invention, a dose of 3 mg; 3±2 mg; 3±1 mg; 8±3 mg; 8±2 mg; 8±1 mg;

[0236] In one embodiment of the invention, a unit dosage form comprises a dose of 8±2 mg of tenofovir alafenamide hemifumarate.

[0237] 25±10 mg; 10±5 mg; 10 mg; 25±5 mg; 25±2 mg; 40±10 mg; 40±5 mg; 40±2 mg; 60±20 mg; 60±10 mg; 100±20 mg; 100±10 mg; 125±20 mg; 125±10 mg; 150±20 mg; 150±10 mg; 200±40 mg; or 200±15 mg of tenofovir alafenamide hemifumarate is administered.

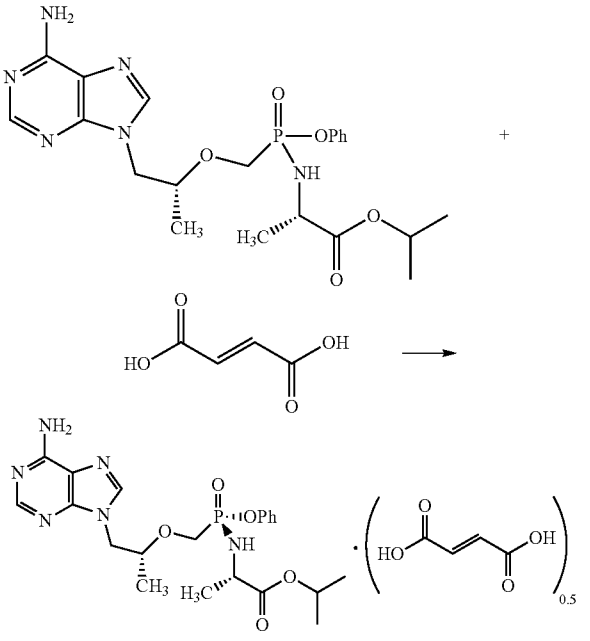
[0238] The desired daily dose of tenofovir alafenamide hemifumarate also may be administered as two, three, four, five, six, or more subdoses administered separately at appropriate intervals throughout the day, optionally, in unit dosage forms.

[0239] The concentration of tenofovir, GS-7340, or tenofovir alafenamide hemifumarate in the bloodstream may be measured as the plasma concentration (e.g., ng/mL). Pharmacokinetic parameters for determining the plasma concentration include, but are not limited to, the maximum observed plasma concentration (C_{max}), observed plasma concentration at the end of the dosing interval or "trough" concentration (C_{tau} or C_{min}), area under the plasma concentration time curve (AUC) from time zero up to the last quantifiable time point ($AUC_{0\text{-}last}$), AUC from time zero to infinity ($AUC_{0\text{-}inf}$), AUC over the dosing interval (AUC_{tau}), time of maximum observed plasma concentration after administration (t_{max}), and half-life of tenofovir, GS-7340, or tenofovir alafenamide hemifumarate in plasma ($t_{1/2}$).

[0240] Administration of GS-7340 or tenofovir alafenamide hemifumarate with food according to the methods of the invention may also increase absorption of GS-7340 or tenofovir alafenamide hemifumarate. Absorption of GS-7340 or tenofovir alafenamide hemifumarate may be measured by the concentration attained in the bloodstream over time after administration of GS-7340 or tenofovir alafenamide hemifumarate. An increase in absorption by administration of GS-7340 or tenofovir alafenamide hemifumarate with food may also be evidenced by an increase in C_{max} and/or AUC of GS-7340 or tenofovir alafenamide hemifumarate as compared to the values if GS-7340 or tenofovir alafenamide hemifumarate was administered without food. Typically protease inhibitors are administered with food.

Selective Crystallization—Tenofovir Alafenamide Hemifumarate

[0241] In one embodiment, tenofovir alafenamide hemifumarate can be prepared using selective crystallization. An example of a scheme for this preparation method is as follows.



[0242] The method can be carried out by subjecting a solution comprising: a) a suitable solvent; b) fumaric acid; c) tenofovir alafenamide; and, optionally, d) one or more seeds comprising tenofovir alafenamide hemifumarate, to conditions that provide for the crystallization of fumaric acid and tenofovir alafenamide. The starting solution can contain the single diastereomer of tenofovir alafenamide or a mixture of

tenofovir alafenamide and one or more of its other diastereomers (e.g., GS-7339, as described in U.S. Pat. No. 7,390,791).

[0243] The selective crystallization can be carried out in any suitable solvent. For example, it can be carried out in a protic solvent or in an aprotic organic solvent, or in a mixture thereof. In one embodiment, the solvent comprises a protic solvent (e.g., water or isopropyl alcohol). In another embodiment, the solvent comprises an aprotic organic solvent (e.g., acetone, acetonitrile (ACN), toluene, ethyl acetate, isopropyl acetate, heptane, tetrahydrofuran (THF), 2-methyl THF, methyl ethyl ketone, or methyl isobutyl ketone, or a mixture thereof). In one embodiment, the solvent comprises ACN or a mixture of ACN and up to about 50% methylene chloride (by volume). The selective crystallization also can be carried out at any suitable temperature, for example, a temperature in the range of from about 0° C. to about 70° C. In one specific embodiment, the resolution is carried out at a temperature of about 0° C.

[0244] One major advantage of the hemifumarate form of tenofovir alafenamide over the monofumarate form is its exceptional capability to purge GS-7339 (i.e., 9-[(R)-2-[[(R)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine; described in, e.g., U.S. Pat. No. 7,390,791), which is the major diastereomeric impurity in the active pharmaceutical ingredient. Thus, the hemifumarate form of tenofovir alafenamide can be more readily and easily separated from impurities than the monofumarate form. Other major advantages of tenofovir alafenamide hemifumarate over the monofumarate form include improved thermodynamic and chemical stability (including long-term storage stability), superior process reproducibility, superior drug product content uniformity, and a higher melting point.

[0245] Tenofovir alafenamide hemifumarate is useful in the treatment and/or prophylaxis of one or more viral infections in man or animals, including infections caused by DNA viruses. RNA viruses, herpesviruses (e.g., CMV, HSV 1, HSV 2, VZV), retroviruses, hepadnaviruses (e.g., HBV), papillomavirus, hantavirus, adenoviruses and HIV. U.S. Pat. No. 6,043,230 (incorporated by reference herein in its entirety) and other publications describe the anti-viral specificity of nucleotide analogs, such as tenofovir disoproxil. Like tenofovir disoproxil, tenofovir alafenamide is another prodrug form of tenofovir, and can be used in the treatment and/or prophylaxis of the same conditions.

[0246] Tenofovir alafenamide hemifumarate can be administered by any route appropriate to the condition to be treated. Suitable routes include oral, rectal, nasal, topical (including ocular, buccal, and sublingual), vaginal, and parenteral (including subcutaneous, intramuscular, intravenous, intradermal, intrathecal, and epidural). Generally, tenofovir alafenamide hemifumarate is administered orally, but it can be administered by any of the other routes noted herein.

[0247] Accordingly, pharmaceutical compositions include those suitable for topical or systemic administration, including oral, rectal, nasal, buccal, sublingual, vaginal, or parenteral (including subcutaneous, intramuscular, intravenous, intradermal, intrathecal, and epidural) administration. The formulations are in unit dosage form and are prepared by any of the methods well known in the art of pharmacy.

[0248] For oral therapeutic administration, the tenofovir alafenamide hemifumarate may be combined with one or more excipients and used in the form of ingestible tablets, buccal tablets, troches, capsules, elixirs, suspensions, syrups, wafers, and the like. Such pharmaceutical compositions and preparations will typically contain at least 0.1% of tenofovir alafenamide hemifumarate. The percentage of this active compound in the compositions and preparations may, of course, be varied and may conveniently be between about 2% to about 60% or more of the weight of a given unit dosage form. The amount of active compound in such therapeutically useful pharmaceutical compositions is preferably such that an effective dosage level will be obtained upon administration of a single-unit dosage (e.g., tablet). Other dosage formulations may provide therapeutically effective amounts of tenofovir alafenamide hemifumarate upon repeated administration of subclinically effective amounts of the same. Preferred unit dosage formulations include those containing a daily dose (e.g., a single daily dose), as well as those containing a unit daily subclinical dose, or an appropriate fraction thereof (e.g., multiple daily doses), of tenofovir alafenamide hemifumarate.

[0249] Pharmaceutical compositions suitable for oral administration may be presented as discrete units such as capsules, cachets, or tablets, each containing a predetermined amount of tenofovir alafenamide hemifumarate; as a powder or granules; as a solution or a suspension in an aqueous liquid or a nonaqueous liquid; or as an oil-in-water liquid emulsion or a water-in-oil liquid emulsion. Tenofovir alafenamide hemifumarate may also be presented as a bolus, electuary, or paste.

[0250] Tenofovir alafenamide hemifumarate is preferably administered as part of a pharmaceutical composition or formulation. Such pharmaceutical composition or formulation comprises tenofovir alafenamide hemifumarate together with one or more pharmaceutically acceptable carriers/excipients, and optionally other therapeutic ingredients. The excipient (s)/carrier(s) must be "acceptable" in the sense of being compatible with the other ingredients of the formulation and not deleterious to the patient. Excipients include, but are not limited to, substances that can serve as a vehicle or medium for tenofovir alafenamide hemifumarate (e.g., a diluent carrier). They may be enclosed in hard or soft shell gelatin capsules, may be compressed into tablets, or may be incorporated directly with the food of the patient's diet.

[0251] Accordingly, the tablets, troches, pills, capsules, and the like may also contain, without limitation, the following: a binder(s), such as hydroxypropyl cellulose, povidone, or hydroxypropyl methylcellulose; a filler(s), such as microcrystalline cellulose, pregelatinized starch, starch, mannitol, or lactose monohydrate; a disintegrating agent(s), such as croscarmellose sodium, cross-linked povidone, or sodium starch glycolate; a lubricant(s), such as magnesium stearate, stearic acid, or other metallic stearates; a sweetening agent(s), such as sucrose, fructose, lactose, or aspartame; and/or a flavoring agent(s), such as peppermint, oil of wintergreen, or a cherry flavoring. When the unit dosage form is a capsule, it may contain, in addition to materials of the above types, a liquid carrier, such as a vegetable oil or a polyethylene glycol. Various other materials may be present as coatings or to otherwise modify the physical form of the solid unit dosage form. For instance, tablets, pills, or capsules may be coated with gelatin, polymers, wax, shellac, or sugar and the like. Of course, any material used in preparing any unit dosage form typically will be pharmaceutically acceptable and substantially nontoxic in the amounts employed. In addition, tenofovir alafenamide hemifumarate may be incorporated into sustained-release preparations and devices.

[0252] For infections of the eye or other external tissues, e.g., mouth and skin, the pharmaceutical compositions are preferably applied as a topical ointment or cream containing tenofovir alafenamide hemifumarate in an amount of, for example, 0.01 to 10% w/w (including active ingredient in a range between 0.1% and 5% in increments of 0.1% w/w such as 0.6% w/w, 0.7% w/w, etc.), preferably 0.2 to 3% w/w and most preferably 0.5 to 2% w/w. When formulated in an ointment, the active ingredient may be employed with either a paraffinic or a water-miscible ointment base. Alternatively, the active ingredient may be formulated in a cream with an oil-in-water cream base.

[0253] Pharmaceutical compositions suitable for topical administration in the mouth include lozenges comprising tenofovir alafenamide hemifumarate in a flavored basis, for example, sucrose and acacia or tragacanth; pastilles comprising the active ingredient in an inert basis such as gelatin and glycerin, or sucrose and acacia; and mouthwashes comprising the active ingredient in a suitable liquid carrier.

[0254] Formulations for rectal administration may be presented as a suppository with a suitable base comprising, for example, cocoa butter or a salicylate.

[0255] Pharmaceutical formulations suitable for parenteral administration are sterile and include aqueous and nonaqueous injection solutions that may contain antioxidants, buffers, bacteriostats, and solutes that render the formulation isotonic with the blood of the intended recipient; and aqueous and nonaqueous sterile suspensions that may include suspending agents and thickening agents. The formulations may be presented in unit-dose or multi-dose containers, for example, sealed ampoules and vials with elastomeric stoppers, and may be stored in a freeze-dried (lyophilized) condition requiring only the addition of the sterile liquid carrier (e.g., water for injections) immediately prior to use. Injection solutions and suspensions may be prepared from sterile powders, granules, and tablets of the kind previously described.

[0256] In addition to the ingredients particularly mentioned above, the pharmaceutical compositions/formulations may include other ingredients conventional in the art, having regard to the type of formulation in question.

[0257] In another embodiment, there is provided veterinary compositions comprising tenofovir alafenamide hemifumarate together with a veterinary carrier therefor. Veterinary carriers are materials useful for the purpose of administering the composition to cats, dogs, horses, rabbits, and other animals, and may be solid, liquid, or gaseous materials that are otherwise inert or acceptable in the veterinary art and are compatible with the active ingredient. These veterinary compositions may be administered orally, parenterally, or by any other desired route.

[0258] The tenofovir alafenamide hemifumarate can be used to provide controlled release pharmaceutical formulations containing a matrix or absorbent material and an active ingredient of the invention, in which the release of the active ingredient can be controlled and regulated to allow less frequent dosing or to improve the pharmacokinetic or toxicity profile of the compound. Controlled release formulations adapted for oral administration, in which discrete units comprising a compounds of the invention, can be prepared according to conventional methods.

[0259] Useful dosages of tenofovir alafenamide hemifumarate can be determined by comparing in vitro activities, and the in vivo activities in animal models. Methods for the extrapolation of effective amounts/dosages in mice and other animals to therapeutically effective amounts/dosages in humans are known in the art.

[0260] The amount of tenofovir alafenamide hemifumarate required for use in treatment will vary with several factors, including but not limited to the route of administration, the nature of the condition being treated, and the age and condition of the patient; ultimately, the amount administered will be at the discretion of the attendant physician or clinician. The therapeutically effective amount/dose of tenofovir alafenamide hemifumarate depends, at least, on the nature of the condition being treated, any toxicity or drug interaction issues, whether the compound is being used prophylactically (e.g., sometimes requiring lower doses) or against an active disease or condition, the method of delivery, and the pharmaceutical formulation, and will be determined by the clinician using conventional dose escalation studies.

[0261] In one embodiment, the oral dose of tenofovir alafenamide hemifumarate may be in the range from about 0.0001 to about 100 mg/kg body weight per day, for example, from about 0.01 to about 10 mg/kg body weight per day, from about 0.01 to about 5 mg/kg body weight per day, from about 0.5 to about 50 mg/kg body weight per day, from about 1 to about 30 mg/kg body weight per day, from about 1.5 to about 10 mg/kg body weight per day, or from about 0.05 to about 0.5 mg/kg body weight per day. As a nonlimiting example, the daily candidate dose for an adult human of about 70 kg body weight will range from about 0.1 mg to about 1000 mg, or from about 1 mg to about 1000 mg, or from about 5 mg to about 500 mg, or from about 1 mg to about 150 mg, or from about 5 mg to about 150 mg, or from about 5 mg to about 100 mg, or about 10 mg, and may take the form of single or multiple doses. In one embodiment, the oral dose of tenofovir alafenamide hemifumarate may be in the form of a combination of agents (e.g., tenofovir alafenamide hemifumarate/emtricitabine/elvitegravir/cobicistat).

[0262] The pharmaceutical compositions described herein may further include one or more therapeutic agents in addition to tenofovir alafenamide hemifumarate. In one specific embodiment of the invention, the additional therapeutic agent can be selected from the group consisting of HIV protease inhibiting compounds, HIV nonnucleoside inhibitors of reverse transcriptase, HIV nucleoside inhibitors of reverse transcriptase, HIV nucleotide inhibitors of reverse transcriptase, HIV integrase inhibitors, and CCR5 inhibitors.

[0263] Therapeutic methods include administering tenofovir alafenamide hemifumarate to a subject/patient in need of the same as a therapeutic or preventative treatment. Thus, tenofovir alafenamide hemifumarate may be administered to a subject/patient having a medical disorder or to a subject who may acquire the disorder. One of ordinary skill will appreciate that such treatment is given in order to ameliorate, prevent, delay, cure, and/or reduce the severity of a symptom or set of symptoms of a disorder (including a recurring disorder). The treatment may also be given to prolong the survival of a subject, e.g., beyond the survival time expected in the absence of such treatment. The medical disorders that may be treated with tenofovir alafenamide hemifumarate include those discussed herein, including without limitation, HIV infection (including, without limitation, HIV-1 and HIV-2 infections; preferably HIV-1 infection) and HBV infection.

Formulation of Cobicistat

[0264] When cobicistat or a pharmaceutically acceptable salt thereof is combined with certain specific solid carrier particles (e.g. silica derivatives), the resulting combination possesses improved physical properties. Even though cobicistat is hygroscopic in nature, the resulting combination has comparatively low hygroscopicity. Additionally, the resulting combination is a free-flowing powder, with high loading values for cobicistat, acceptable physical and chemical stability, rapid drug release properties, and excellent compressibility. Thus, the resulting combination can readily be processed into solid dosage forms (e.g. tablets), which possess good drug release properties, low tablet friability, good chemical and physical stability, and a low amount of residual solvents. The compositions of the invention represent a significant advance that facilitates the commercial development of cobicistat for use in treating viral infections such as HIV.

[0265] Cobicistat can be combined with any suitable solid carrier, provided the resulting combination has physical properties that allow it to be more easily formulated than the parent compound. For example, suitable solid carriers include kaolin, bentonite, hectorite, colloidal magnesium-aluminum silicate, silicon dioxide, magnesium trisilicate, aluminum hydroxide, magnesium hydroxide, magnesium oxide and talc. In one embodiment of the invention, the solid carrier can comprise calcium silicate (such as ZEOPHARM), or magnesium aluminometasilicate (such as NEUSILIN). As used herein, "loaded" on a solid carrier includes, but is not limited to a compound being coated in the pores and on the surface of a solid carrier.

[0266] Suitable silica derivatives for use in the compositions of the invention and methods for preparing such silica derivatives include those that are described in international patent application publication number WO 03/037379 and the references cited therein. A specific silica material that is particularly useful in the compositions and methods of the invention is AEROPERL® 300 (fumed silica), which is available from Evonik Degussa AG, Dusseldorf, Germany. Other materials having physical and chemical properties similar to the silica materials described herein can also be used.

Ritonavir

[0267] Ritonavir (1,3-thiazol-5-ylmethyl N-[(2S,3S,5S)-3-hydroxy-5-[(2S)-3-methyl-2-{[methyl({[2-(propan-2-yl)-1,3-thiazol-4-yl]methyl})carbamoyl]amino}butanamido]-1,6-diphenylhexan-2-yl]carbamate) was developed as an inhibitor of retroviral (HIV) protease; however, it is now used in a manner similar to cobicistat to inhibit the action of certain cytochrome P450 proteases (specifically Cyp3A4) thereby allowing greater circulating levels of drugs for treatment of HIV than would be obtained by administration of the drugs alone. Although none of GS-7340, tenofovir, or tenofovir alafenamide hemifumarate apparently is metabolized by cytochrome P450 proteases, it is contemplated that ritonavir may be used in the manner that cobicistat is used to boost the circulating levels of GS-7340, tenofovir, or tenofovir alafenamide hemifumarate, to improve the pharmacokinetics of GS-7340, tenofovir, or tenofovir alafenamide hemifumarate and achieve the other advantages of the use of cobicistat as disclosed herein.

Combination Treatment

[0268] The compounds and methods of the invention may also be used with any of the following compounds:

[0269] 1) amprenavir, atazanavir, fosamprenavir, indinavir, lopinavir, ritonavir, nelfinavir, saquinavir, tipranavir, brecanavir, darunavir, TMC-126, TMC-114, mozenavir (DMP-450), JE-2147 (AG1776), L-756423, RO0334649, KNI-272, DPC-681, DPC-684, GW640385X, DG17, GS-8374, PPL-100, DG35, and AG 1859;

[0270] 2) an HIV nonnucleoside inhibitor of reverse transcriptase, e.g., capravirine, emivirine, delaviridine, efavirenz, nevirapine, (+) calanolide A, etravirine, GW5634, DPC-083, DPC-961, DPC-963, MIV-150, and TMC-120, TMC-278 (rilpivirine), BILR 355 BS, VRX 840773, UK-453061, and RDEA806;

[0271] 3) an HIV nucleoside inhibitor of reverse transcriptase, e.g., zidovudine, emtricitabine, didanosine, stavudine, zalcitabine, lamivudine, abacavir, amdoxovir, elvucitabine, alovudine, MIV-210, racivir (±-emtricitabine), D-d4FC, phosphazide, fozivudine tidoxil, apricitibine (AVX754), GS-7340, KP-1461, and fosalvudine tidoxil (formerly HDP 99.0003);

[0272] 4) an HIV nucleotide inhibitor of reverse transcriptase, e.g., tenofovir disoproxil fumarate and adefovir dipivoxil;

[0273] 5) an HIV integrase inhibitor, e.g., curcumin, derivatives of curcumin, chicoric acid, derivatives of chicoric acid, 3,5-dicaffeoylquinic acid, derivatives of 3,5-dicaffeoylquinic acid, aurintricarboxylic acid, derivatives of aurintricarboxylic acid, caffeic acid phenethyl ester, derivatives of caffeic acid phenethyl ester, tyrphostin, derivatives of tyrphostin, quercetin, derivatives of quercetin, S-1360, zintevir (AR-177), L-870812, and L-870810, MK-0518 (raltegravir), elvitegravir, BMS-538158, GSK364735C, BMS-707035, MK-2048, and BA 011;

[0274] 6) a gp41 inhibitor, e.g., enfuvirtide, sifuvirtide, FB006M, and TRI-1144;

[0275] 7) a CXCR4 inhibitor, e.g., AMD-070;

[0276] 8) an entry inhibitor, e.g., SP01A;

[0277] 9) a gp120 inhibitor, e.g., BMS-488043 or BlockAide/CR;

[0278] 10) a G6PD and NADH-oxidase inhibitor, e.g., immunitin;

[0279] 11) a CCR5 inhibitor, e.g., aplaviroc, vicriviroc, maraviroc, PRO-140, INCB15050, PF-232798 (Pfizer), and CCR5 mAb004;

[0280] 12) other drugs for treating HIV, e.g., BAS-100, SPI-452, REP 9, SP-01A, TNX-355, DES6, ODN-93, ODN-112, VGV-1, PA-457 (bevirimat), Ampligen, HRG214, Cytolin, VGX-410, KD-247, AMZ 0026, CYT 99007A-221 HIV, DEBIO-025, BAY 50-4798, MDX010 (ipilimumab), PBS 119, ALG 889, and PA-1050040 (PA-040);

[0281] 13) an interferon, e.g., pegylated rIFN-alpha 2b, pegylated rIFN-alpha 2a, rIFN-alpha 2b, rIFN-alpha 2a, consensus IFN alpha (infergen), feron, reaferon, intermax alpha, r-IFN-beta, infergen+actimmune, IFN-omega with DUROS, albuferon, locteron, Albuferon, Rebif, oral interferon alpha, IFNalpha-2b XL, AVI-005, PEG-Infergen, and pegylated IFN-beta;

[0282] 14) a ribavirin analog, e.g., rebetol, copegus, viramidine (taribavirin);

[0283] 15) an NS5b polymerase inhibitor, e.g., NM-283, valopicitabine, R1626, PSI-6130 (R1656), HCV-796, BILB 1941, XTL-2125, MK-0608, NM-107, R7128 (R4048), VCH-759, PF-868554, and GSK625433;

[0284] 16) an NS3 protease inhibitor, e.g., SCH-503034 (SCH-7), VX-950 (telaprevir), BILN-2065, BMS-605339, and ITMN-191;

[0285] 17) an alpha-glucosidase 1 inhibitor, e.g., MX-3253 (celgosivir), UT-231B;

[0286] 18) hepatoprotectants, e.g., IDN-6556, ME 3738, LB-84451, and MitoQ;

[0287] 19) a nonnucleoside inhibitor of HCV, e.g., benzimidazole derivatives, benzo-1,2,4-thiadiazine derivatives, phenylalanine derivatives, A-831, GS-9190, and A-689; and

[0288] 20) other drugs for treating HCV, e.g., zadaxin, nitazoxanide (alinea), BIVN-401 (virostat), PYN-17 (altirex), KPE02003002, actilon (CPG-10101), KRN-7000, civacir, GI-5005, ANA-975, XTL-6865, ANA 971, NOV-205, tarvacin, EHC-18, NIM811, DEBIO-025, VGX-410C, EMZ-702, AVI 4065, Bavituximab, Oglufanide, and VX-497 (merimepodib).

[0289] Exemplary combinations (including, but not limited to, single tablet regimens) include (a) emtricitabine/darunavir/cobicistat/GS-7340; (b) emtricitabine/darunavir/cobicistat/tenofovir alafenamide hemifumarate; (c) emtricitabine/darunavir/cobicistat/tenofovir disoproxil fumarate (TDF); (d) emtricitabine/elvitegravir/cobicistat/GS-7340; (e) emtricitabine/elvitegravir/cobicistat/tenofovir alafenamide hemifumarate; (f) emtricitabine/elvitegravir/cobicistat/TDF; (g) cobicistat/GS-7340; (h) cobicistat/tenofovir alafenamide hemifumarate; and (i) cobicistat/TDF. The combinations listed above may contain various dosages of the component agents; as nonlimiting examples, combination (b) above can include 200 mg of emtricitabine, 800 mg of darunavir, 150 mg of cobicistat, and 10 mg of tenofovir alafenamide hemifumarate, and combination (e) above can include 200 mg of emtricitabine, 150 mg of elvitegravir, 150 mg of cobicistat, and 10 mg of tenofovir alafenamide hemifumarate.

[0290] An alternative exemplary combination is emtricitabine and tenofovir alafenamide hemifumarate. The combination of emtricitabine and TDF is currently marketed as TRUVADA®. See also U.S. Patent Application Publication No. 2004/0224916, the content of which is hereby incorporated by reference herein in its entirety. The present invention provides the combination of emtricitabine and tenofovir alafenamide hemifumarate. This combination may contain various dosages of the two component agents; as a nonlimiting example, this combination can include 200 mg of emtricitabine and 10 mg of tenofovir alafenamide hemifumarate.

[0291] An additional alternative exemplary combination is emtricitabine, rilpivirine, and tenofovir alafenamide hemifumarate. The combination of emtricitabine, rilpivirine (a nonnucleoside reverse transcriptase inhibitor), and TDF is currently marketed as COMPLERA®. The present invention provides the combination of emtricitabine, rilpivirine, and tenofovir alafenamide hemifumarate. This combination may contain various dosages of the three component agents; as a nonlimiting example, this combination can include 200 mg of emtricitabine, 25 mg of rilpivirine, and 10 mg of tenofovir alafenamide hemifumarate.

[0292] A further additional alternative exemplary combination is GS-9441 and tenofovir alafenamide hemifumarate. The combination of GS-9441 (a reverse transcriptase inhibitor) and GS-7340 is disclosed in U.S. Patent Application Publication No. 2009/0075939 and U.S. Pat. No. 8,354,421, the content of each of which is hereby incorporated by reference herein in its entirety. The present invention provides the combination of GS-9441 and tenofovir alafenamide hemifumarate. This combination may contain various dosages of the two component agents; as a nonlimiting example, this combination can include 5-1500 mg of GS-9441 and 10 mg of tenofovir alafenamide hemifumarate.

[0293] Exemplary amounts of agents in various combinations include, but are not limited to, the following: (1) cobicistat: 10-500 mg, 50-500 mg, 75-300 mg, 100-200 mg, or 150 mg; (2) tenofovir alafenamide hemifumarate: 1-60 mg, 3-40 mg, 5-30 mg, 8-20 mg, or 10 mg; (3) emtricitabine: 10-500 mg, 50-500 mg, 75-300 mg, 150-250 mg, or 200 mg; (4) elvitegravir: 10-500 mg, 50-500 mg, 75-300 mg, 100-200 mg, or 150 mg; (5) darunavir: 300-1800 mg, 400-1600 mg, 500-1200 mg, 600-1000 mg, or 800 mg; and (6) rilpivirine: 5-100 mg, 10-80 mg, 15-60 mg, 20-40 mg, or 25 mg. One of skill in the art will know that, in the case of administering a pharmaceutically acceptable salt or complex of an agent, the amount administered will be adjusted relative to the weight of the component added to produce the salt or complex.

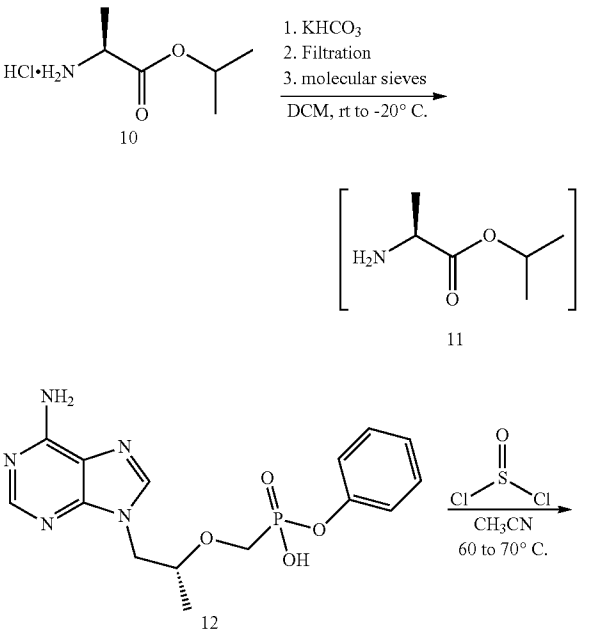
[0294] The invention will now be illustrated by the following nonlimiting Examples. The Synthetic Examples provided herein describe the synthesis of compounds of the invention as well as intermediates used to prepare compounds of the invention.

Synthetic Examples

Synthetic Example 1

Preparation of Diastereomeric Mixture of 9-[(R)-2-[[(R,S)-1-[[(S)-(isopropoxycarbonyl)ethyl]amino] phenoxyphosphinyl]methoxy]propyl]adenine (15)

[0295]



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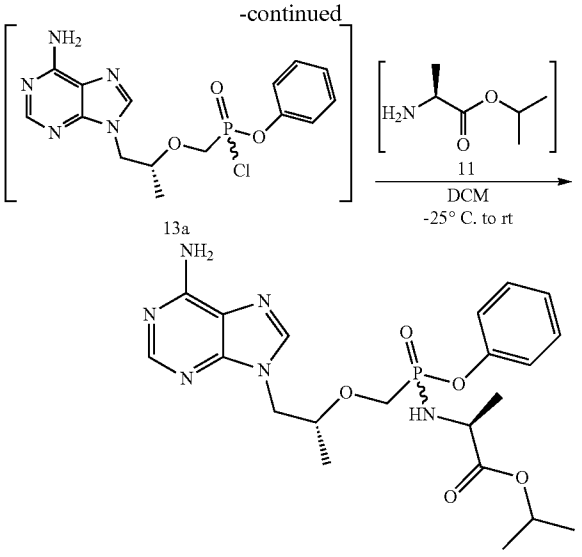


15

a. Preparation of Compound 11

[0296] Isopropyl L-alanine ester hydrochloride 10 (1 kg, 5.97 mol, 1.0 equiv) and potassium bicarbonate (1.45 kg, 14.5 mol, 2.43 equiv) were agitated in DCM (4 kg) for 10-14 hours with maximum agitation, maintaining the pot temperature between 19 and 25° C. The mixture was then filtered and rinsed forward with DCM (2 kg). The filtrate was dried over a bed of 4 Å molecular sieves until the water content of the solution was ≤0.05%. The resultant stock solution containing compound 11 was then cooled to a pot temperature of −20° C. and held for further use.

b. Preparation of Compound 13a

[0297] To a solution of thionyl chloride (0.72 kg, 6.02 mol, 2.19 equiv) in acetonitrile (5.5 kg) at 60° C. was added compound 12 (1 kg, 2.75 mol, 1.00 equiv) in 10 equal portions over 2 hours. The pot temperature was then adjusted to 70° C. and stirred for 1-3 hours until deemed complete by ^{31}P NMR analysis (Target: >97.0% conversion of starting material signal at 12.6 ppm to product signal at 22.0 ppm). The pot temperature was then adjusted to 40° C. and vacuum applied. The mixture was distilled to dryness, maintaining a maximum jacket temperature of 40° C. The dry residue was then taken up in dichloromethane (30 kg) and the pot temperature adjusted to 19-25° C. The resultant slurry containing compound 13a was held for further use.

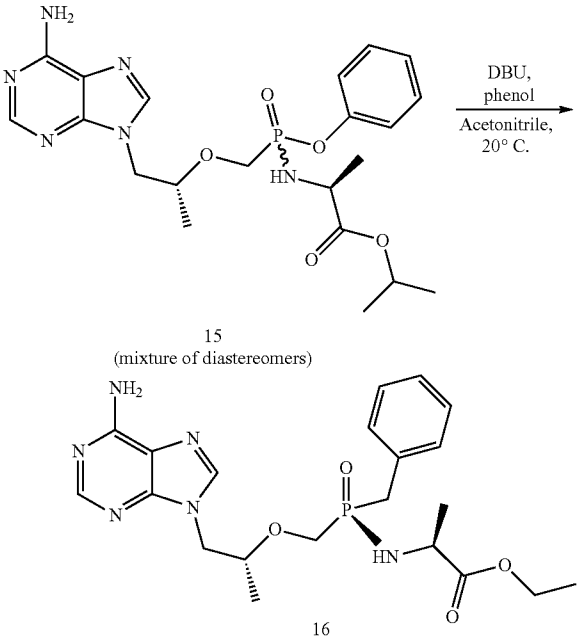
c. Preparation of Compound 15

[0298] To the stock solution of isopropyl L-alanine ester 11 (4.82 equiv) at −25° C. was added slurry containing compound 13a (1.0 equiv) over a minimum of 2 hours, maintaining the pot temperature≤−10° C. The mixture was then held at a temperature ≤−10° C. for at least 30 minutes, then the pH checked using water wet pH paper. If the pH was <4, adjustment with triethylamine to pH 4-7 was performed. The pot temperature was then adjusted to room temperature (19-25° C.). In a separate vessel, a solution of sodium phosphate monobasic (2.2 kg, 18 mol, 6.90 equiv) in water (16 kg) was prepared. Half of the sodium phosphate monobasic solution was charged to the phosphonamidate reactor, and vigorously stirred. The layers were settled and partitioned. The organic layer was washed again with the remaining half of sodium phosphate monobasic solution. In a separate vessel, a solution of potassium bicarbonate (1.1 kg, 11 mol, 4.22 equiv) in water (5.5 kg) was prepared. Half of the potassium bicarbonate solution was charged to the organic phase, and vigorously stirred. The layers were settled and partitioned. The organic layer was washed again with the remaining half of the potassium bicarbonate solution followed by a final water (3.3 kg) wash. The organic phase was then retained and distilled to a volume of ca. 6 L. The resultant solution was analyzed for water content. If the water content was >1.0%, DCM could be charged and the distillation to ca. 6 L repeated. When the solution water content was less than or about 1.0%, the pot temperature was adjusted to 19-25° C. prior to discharge of the stock solution in DCM to provide the diastereomeric mixture of 9-[(R)-2-[[(R,S)-1-[[(S)-(isopropoxycarbonyl) ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine (15). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.20-1.33 (m, 12H), 3.62-3.74 (m, 1H), 3.86-4.22 (m, 5H), 4.30-4.44 (m, 1H), 4.83-5.10 (m, 1H), 6.02 (br s, 3H), 7.18-7.34 (m, 5H), 7.98-8.02 (m, 1H), 8.32-8.36 (m, 1H); ^{31}P NMR (162 MHz, $CDCl_3$): δ. 21.5, 22.9.

Synthetic Example 2

Crystallization-Induced Dynamic Resolution of Diastereomeric Mixture of 9-[(R)-2-[[(R,S)-1-[[(S)-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine (15) to provide 9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino] phenoxyphosphinyl]methoxy]propyl]adenine (16)

[0299]



15
(mixture of diastereomers)

16

[0300] A 22 wt % solution of diastereomeric mixture of 9-[(R)-2-[[(R,S)-1-[[(S)-(isopropoxycarbonyl)ethyl]amino] phenoxyphosphinyl]methoxy]propyl]adenine (15) in acetonitrile (2.3 kg solution, 0.51 kg 15, 1.1 mol, 1 equiv) was charged to a vessel equipped with an overhead stirrer, distillation apparatus, and nitrogen inlet. The mixture was concentrated by distillation at 100-300 mbar over a temperature range of 45-55° C. to a final concentration of 30-35 wt %. The distillation apparatus was then removed and the solution was cooled to 20° C. The solution was seeded with 2.0% compound 16 and allowed to stir for one hour at 20° C. Phenol (9.9 g, 0.11 mol, 0.1 equiv) and DBU (16 g, 0.11 mol, 0.1 equiv) were added and the mixture was stirred for an additional 24 hours or until the weight percent of compound 16 remaining in solution was less than 12%. The slurry was then cooled to 0° C. and stirred for an additional 18 hours at 0° C. The slurry was filtered and washed with a 1:1 solution of isopropyl acetate:acetonitrile (1.5 L) at 0° C. The solids were dried in a vacuum oven at 50° C. to give 0.40 kg of compound 16 (80% yield) as a white solid. ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.21 (m, 9H), 1.28 (d, J=7.0 Hz, 3H), 3.65 (dd, J=13.1, 10.7, 1H) 4.00 (m, 4H), 4.33 (dd, J=14.4, 3.1 Hz, 1H), 5.00 (m, 1H) 6.00 (bs, 2H), 6.99 (m, 2H), 7.07 (m, 1H), 7.19 (m, 2H), 7.97 (s, 1H), 8.33 (s, 1H). ^{31}P NMR (162 MHz, $CDCl_3$): δ. 20.8.

Synthetic Example 3

Preparation of Compound 13a in High Diastereomeric Purity

[0301] To a slurry of compound 12 (10.0 g, 27.5 mmol, 1.00 equiv) in toluene (60 mL) at ambient temperature was added thionyl chloride (3.0 mL, 41 mmol, 1.5 equiv). The slurry was heated to 70° C. and agitated for 48-96 hours until reaction and diastereomeric enrichment were deemed complete by HPLC (Target: >97.0% conversion of compound 12 to compound 13a and >90:10 diastereomeric ratio of compound 13a). The mixture was concentrated to dryness by vacuum distillation, and the dry residue was taken up in toluene (50 mL). The resultant slurry containing compound 13a was held at ambient temperature for further use.

Synthetic Example 4

Preparation of 9-[(R)-2-[[(R,S)-1-[[(S)-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy] propyl]adenine (15) in High Diastereomeric Purity

[0302] To a solution of isopropyl L-alanine ester 11 (4.50 equiv) in DCM (80 mL) at −25° C. was added a slurry containing compound 13a (1.00 equiv) that is at least 90% diastereomerically pure in toluene (50 mL) over a minimum of 45 minutes, maintaining the internal temperature ≤−20° C. The mixture was then held at a temperature ≤−20° C. for at least 30 minutes, and the pH checked using water wet pH paper. If the pH was <4, it was adjusted with triethylamine to pH 4-7. The pot temperature was adjusted to room temperature (19-25° C.). The mixture was transferred to a separatory funnel and washed sequentially with 10% w/v aqueous solution of sodium phosphate monobasic (2×50 mL), 15% w/v aqueous solution of potassium bicarbonate (2×20 mL), and water (50 mL). The final organic layer was dried over anhydrous sodium sulfate, filtered, and concentrated in vacuo to a viscous amber oil. The oil was dissolved in toluene/acetonitrile (4:1) (50 mL), and the solution was seeded with 9-[(R)-2-[[(R,S)-1-[[(S)-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine (about 1 mg, 99:1 diastereomeric ratio) and stirred for 2 hours at ambient temperature. The resultant slurry was filtered and the filter cake was washed with toluene/acetonitrile (4:1) (15 mL) and dried in a vacuum oven at 40° C. for 16 hours to give the product, 9-[(R)-2-[[(R,S)-1-[[(S)-(isopropoxycarbonyl) ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine (15), as a white solid (10.0 g, 76.4%, 97.5:2.5 diastereomeric ratio). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.20-1.33 (m, 12H), 3.62-3.74 (m, 1H), 3.86-4.22 (m, 5H), 4.30-4.44 (m, 1H), 4.83-5.10 (m, 1H), 6.02 (br s, 3H), 7.18-7.34 (m, 5H), 7.98-8.02 (m, 1H), 8.32-8.36 (m, 1H); ^{31}P NMR (162 MHz, $CDCl_3$): δ. 21.5, 22.9.

Synthetic Example 5

Preparation of Compound 12

[0303] PMPA (100.0 g, 0.35 mol, 1 equiv) was charged to a vessel equipped with an overhead stirrer, reflux condenser and nitrogen inlet followed by acetonitrile (800 mL). To the vessel was added triethylamine (71.0 g, 0.70 mol, 2 equiv) followed by DMAP (42.6 g, 0.35 mol, 1 equiv) and triphenylphosphite (162.1 g, 0.52 mol, 1.5 equiv). The mixture was heated to 80° C. and agitated for ≥48 hours at 80° C. or until the reaction was complete by ^{31}P NMR. (A sample directly from the reaction is taken and an insert containing 10% H_3PO_2 in D_2O is added. The intermediate formed is the PMPA anhydride and is at 6 ppm; the product is at 11 ppm. The reaction is deemed complete when less than 5% anhydride is present). The reaction mixture was distilled to ~1.5 volumes of acetonitrile and diluted with ethyl acetate (200 mL) and water (300 mL). The aqueous layer was separated and washed with ethyl acetate (200 mL) twice. The aqueous layer was recharged to the vessel and pH adjusted to pH 3 using 12.1 M HCl (21.0 mL). The reaction was then seeded with 0.05% of compound 12 seed and allowed to stir at 25° C. Additional 12.1 M HCl was added over 20 minutes (7.0 mL) until pH 2 was achieved. The crystallization was allowed to stir at ambient temperature for 30 minutes and then cooled to 10° C. over 2 hours. Once at 10° C. the crystallization was allowed to stir for 2.5 hours at 10° C. The slurry was filtered and washed with pH 1.5 water (200 g). After drying in the vacuum oven, 102.2 g of compound 12 (81% yield) was obtained as a white solid. ^{1}H NMR (400 MHz, D_2O): δ 1.31 (d, J=6.1 Hz, 3H), 3.59 (dd, J=14.0, 9.0 Hz, 1H), 3.85 (dd, J=14.0, 9.0 Hz, 1H), 4.1 (m, 1H), 4.3 (dd, J=15.0, 9.0 Hz, 1H), 4.5 (dd, J=15.0, 2 Hz, 1H), 6.75 (d, J=7 Hz, 2H), 7.15 (t, J=7 Hz, 1H), 7.25 (t, J=7 Hz, 2H), 8.26 (s, 1H), 8.35 (s, 1H). ^{31}P NMR (162 MHz, D_2O): δ. 14.8.

Synthetic Examples

Tenofovir Alafenamide Hemifumarate

Synthetic Example 6

[0304] Tenofovir alafenamide monofumarate solids (5.0 g) and 9-[(R)-2-[[(R)-[[(S)-1-(isopropoxycarbonyl)ethyl] amino]phenoxyphosphinyl]methoxy]propyl]adenine (GS-7339) monofumarate solids (0.75 g) were charged into 35 g MTBE at 22° C. and the mixture was stirred for 1 hour. A slurry was formed and was dried in a rotary evaporator. 58 g acetonitrile (ACN) was charged into the solids and the mixture was heated to reflux to dissolve the solids. The resulting solution was allowed to cool naturally while agitated. A slurry

was formed, and the slurry was further cooled by an ice-water bath. The solids were isolated by filtration and washed with 5 g ACN. The solids were dried in a vacuum oven at 40° C. overnight. 5.52 g off-white solids were obtained. The solids were analyzed by XRPD and found to contain tenofovir alafenamide monofumarate, GS-7339 monofumarate, and tenofovir alafenamide hemifumarate.

Synthetic Example 7

Preparation of Tenofovir Alafenamide Hemifumarate Via Selective Crystallization

[0305] 9-[(R)-2-[[[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine as a slurry in ACN (9.7 kg slurry, 13.8 wt %, a diastereomeric mixture of 1.0 kg (2.10 mol, 1 mol equiv) of 9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine and 0.35 kg of 9-[(R)-2-[[(R)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine was charged into a reactor and rinsed forward with dichloromethane (5 kg). The mixture was concentrated under vacuum to about 3 L with jacket temperature below 40° C. The concentrate was then coevaporated with ACN (6 kg) under vacuum to about 3 L with jacket temperature below 40° C. The concentrate was diluted with ACN (8.5 kg) and warmed to 40-46° C. The warm mixture was filtered into a second reactor and the filtrate was cooled to 19-25° C.

[0306] To the above solution was charged fumaric acid (0.13 kg, 1.12 mol, 0.542 mole equiv) followed by ACN (1 kg), and the mixture was heated to 67-73° C. The hot mixture was transferred into a reactor via a polishing filter, and then adjusted to 54-60° C. Seed crystals (5 g) of the hemifumarate form of tenofovir alafenamide were charged (for example, the mixture can be seeded with tenofovir alafenamide hemifumarate formed in Synthetic Example 6 or a subsequent production), and the resulting mixture was agitated at 54-60° C. for about 30 minutes. The mixture was cooled over a minimum of 4 hours to 0-6° C., and then agitated at 0-6° C. for a minimum of 1 hour. The resulting slurry was filtered and rinsed with chilled (0-6° C.) ACN (2 kg). The product was dried under vacuum below 45° C. until loss on drying (LOD) and organic volatile impurities (OVI) limits were met (LOD ≤1.0%, dichloromethane content ≤0.19%, acetonitrile content ≤0.19%) to afford the final compound of the hemifumarate form of tenofovir alafenamide as a white to off-white powder (typical yield is about 0.95 kg). ^{1}H NMR (400 MHz, d6 DMSO): δ 1.06 (d, J=5.6 Hz, 3H), 1.12-1.16 (m, 9H), 3.77 (dd, J=10.4, 11.6 Hz, 1H), 3.84-3.90 (m, 2H), 3.94 (m, 1H), 4.14 (dd, J=6.8, 14.8 Hz, 1H), 4.27 (m, 1H), 4.85 (heptet, J=6.0 Hz, 1H), 5.65 (t, J=11.2 Hz, 1H), 6.63 (s, 1H), 7.05 (d. J=7.6 Hz, 2H), 7.13 (t, J=7.2 Hz, 1H), 7.24 (s, 2H), 7.29 (t, J=7.6 Hz, 2H), 8.13 (t, J=13.6 Hz, 2H), ^{31}P NMR (162 MHz, d6 DMSO): δ 23.3.

Synthetic Example 8

Preparation of Tenofovir Alafenamide Hemifumarate

[0307] To a jacketed reactor equipped with overhead agitator, was charged 9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine (10 g), fumaric acid (1.22 g), and ACN (100 mL). The mixture was heated to 70-75° C. to dissolve the solids. Any undissolved particulates were removed by filtration through a cartridge filter. The filtered solution was cooled to 60-65° C., and seeded with 1% (by weight) of tenofovir alafenamide hemifumarate. The slurry was aged for 30 minutes and cooled to 0-5° C. over 2 hours. The temperature was maintained for 1-18 hours, and the resulting slurry was filtered and washed with 2 ml of cold ACN (0-5° C.). The solids were dried under vacuum at 50° C. to provide the hemifumarate form of tenofovir alafenamide, which was characterized as described below.

Characterization of Tenofovir Alafenamide Hemifumarate from Synthetic Example 8

[0308] Tenofovir alafenamide hemifumarate from Synthetic Example 8 consists of 9-[(R)-2-[[(S)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine and one-half an equivalent of fumaric acid. Tenofovir alafenamide hemifumarate is anhydrous, nonhygroscopic, and has a DSC onset endotherm of about 131° C.

X-ray Powder Diffraction

[0309] The XRPD pattern of tenofovir alafenamide hemifumarate was obtained in the following experimental setting: 45 KV, 45 mA, Kα1=1.5406 Å, scan range 2-40°, step size 0.0084°, counting time: 8.25 s. The XRPD pattern for tenofovir alafenamide hemifumarate is shown in FIG. **13**. The characteristic peaks include: 6.9±0.2°, 8.6±0.2°, 10.0±0.2°, 11.0±0.2°, 12.2±0.2°, 15.9±0.2°, 16.3±0.2°, 20.2±0.2°, and 20.8±0.2°.

Single-Crystal X-Ray Diffraction

[0310] The crystal size was 0.32×0.30×0.20 mm^3. The sample was held at 123 K and the data was collected using a radiation source with a wavelength of 0.71073 Å in the theta range of 1.59 to 25.39°. Conditions of and data collected from the single-crystal X-ray diffraction are shown in Table 1.

TABLE 1

Single-Crystal X-ray Diffraction

Empirical formula	$C_{23}H_{31}N_6O_7P$	
Formula weight	534.50	
Temperature	123(2) K	
Crystal size	0.32 × 0.30 × 0.20 mm^3	
Theta range for data collection	1.59 to 25.39°	
Wavelength	0.71073 Å	
Crystal system	Tetragonal	
Space group	P4(2)2(1)2	
Unit cell dimensions	a = 18.1185(12) Å	α = 90°
	b = 18.1185(12) Å	β = 90°
	c = 17.5747(11) Å	γ = 90°
Volume	5769.4(6) Å^3	
Z	8	
Density (calculated)	1.231 g/cm^3	

DSC Analysis

[0311] The DSC analysis was conducted using 2.517 mg of tenofovir alafenamide hemifumarate. It was heated at 10° C./min over the range of 40-200° C. The onset endotherm was found to be about 131° C. (FIG. **14**).

TGA Data

[0312] The TGA data were obtained using 4.161 mg of tenofovir alafenamide hemifumarate. It was heated at 10°

C./min over the range of 25-200° C. The sample lost 0.3% weight before melting (FIG. **15**). It was determined to be an anhydrous form.

DVS Analysis

[0313] DVS analysis was conducted using 4.951 mg of tenofovir alafenamide hemifumarate. The material was kept at 25° C. in nitrogen at humidities ranging from 10% to 90% relative humidity; each step was equilibrated for 120 minutes. The sorption isotherm is shown at FIG. **16**. The material was found to be nonhygroscopic, and to absorb 0.65% water at a relative humidity of 90%.

Purging of Diastereomeric Impurity

[0314] In the prior syntheses of tenofovir alafenamide, one of the major impurities is typically the diastereomer 9-[(R)-2-[[(R)-[[(S)-1-(isopropoxycarbonyl)ethyl]amino]phenoxyphosphinyl]methoxy]propyl]adenine. The hemifumarate form of tenofovir alafenamide from Synthetic Example 8 has an exceptional capability to purge this diastereomeric impurity, as compared with the capability of the monofumarate form (described in, e.g., U.S. Pat. No. 7,390,791). The data in Table 2 (below) demonstrates that tenofovir alafenamide hemifumarate (Batch 2) purged the diastereomeric impurity to less than one-tenth of the starting concentration, whereas the monofumarate form of tenofovir alafenamide (Batch 1) only slightly purged the diastereomeric impurity.

TABLE 2

Purging Capability Comparison

Batch	Diastereomeric Impurity in Starting Material	Solvent	Fumaric acid charge (mole equivalent)	Product obtained	Diastereomeric Impurity in Product
1	9.3%	ACN	0.9	Monofumarate form	7.6%
2	10.0%	ACN	0.5	Hemifumarate form	0.65%

Chemical Stability

[0315] Chemical stability of the hemifumarate form of tenofovir alafenamide was compared with the monofumarate form. As shown in Table 3 (below), under identical conditions, the hemifumarate form of tenofovir alafenamide was chemically more stable and exhibited better long-term storage stability, with significantly less degradation (% Total Deg. Products) than the monofumarate form. Conditions evaluated include temperature, relative humidity (RH), and the open or closed state of the container cap.

TABLE 3

Chemical Stability Comparison

		Monofumarate form		Hemifumarate form	
Storage Condition	Time Points (weeks)	% TA* Area Normalized	% Total Deg. Products	% TA Area Normalized	% Total Deg. Products
40° C./75% RH Cap Closed	0	97.1	0.69	98.4	0.05
	1	97.0	0.87	98.4	0.14
	2	96.6	1.18	98.5	0.14
	4	96.4	1.49	98.4	0.25
	8	95.4	2.36	98.0	0.49

TABLE 3-continued

Chemical Stability Comparison

		Monofumarate form		Hemifumarate form	
Storage Condition	Time Points (weeks)	% TA* Area Normalized	% Total Deg. Products	% TA Area Normalized	% Total Deg. Products
40° C./75% RH Cap Open	0	97.1	0.69	98.4	0.05
	1	96.9	0.90	98.5	0.15
	2	96.6	1.10	98.5	0.14
	4	96.2	1.67	98.4	0.26
	8	95.0	2.74	98.1	0.50
70° C. Cap Closed	0	97.1	0.69	98.4	0.05
	2	96.2	1.83	98.5	0.22
	4	93.3	4.78	98.4	0.33

*TA is tenofovir alafenamide

Thermodynamic Stability

[0316] Stable form screening of tenofovir alafenamide hemifumarate showed that it is thermodynamically stable in most solvents, such as ACN, toluene, ethyl acetate, methyl tert-butyl ether (MTBE), acetone, THF, and 2-methyl THF. A similar stable form screening of the monofumarate form showed that this form is not thermodynamically stable in the above-listed solvents. When suspended in these solvents, the monofumarate form of tenofovir alafenamide fully converts to the hemifumarate form in THF and 2-methyl THF, and partially converts to the hemifumarate form in ACN, ethyl acetate, MTBE, and acetone, as well as at ambient temperatures.

Thermal Stability

[0317] As shown by the DSC data, the hemifumarate form of tenofovir alafenamide has a melting point that is about 10° C. higher than that of the monofumarate form, indicating that the hemifumarate form has improved thermal stability as compared with the monofumarate form.

Biological Example 1

Transport Studies

[0318] Caco-2 transepithelial transport studies: Caco-2 cells between passage 43 and 69 were grown to confluence over at least 21 days on 24-well polyethylene-terephthalate (PET) transwell plates (BD Biosciences, Bedford, Mass.). Experiments were conducted using Hank's Buffered Salt Solution (HBSS) containing 10 mM HEPES and 15 mM Glucose obtained from Life Technologies (Grand Island, N.Y.). Donor and receiver buffers had their pH adjusted to pH 6.5 and 7.4, respectively. The receiver well used HBSS buffer supplemented with 1% bovine serum albumin. In studies done to determine transport inhibition, monolayers were preincubated for 60 minutes in the presence of assay buffer and inhibitor in order to saturate any transporter binding sites. Following preincubation, fresh assay buffer containing inhibitor and the test compound were added. Test compound concentrations in assay chambers were analyzed by liquid chromatography coupled to tandem mass spectrometry (LC/MS/MS). Transepithelial electrical resistance (TEER) and lucifer yellow permeability were determined to assure membrane integrity. Each individual experiment was done in duplicate and the permeation of control compounds atenolol

(low permeability), propranolol (high permeability), and vinblastine (efflux transport) were determined to meet acceptance criteria for each batch of assay plates.

[0319] Pgp and BCRP inhibition assays in transfected Madin-Darby canine kidney (MDCKII) cells: Inhibition of Pgp-mediated transport was studied using the Pgp substrate calcein AM and MDCKII cells transfected with the human MDR1 (ABCB1) gene (encoding Pgp). Similarly, inhibition of BCRP-mediated transport was studied using the BCRP substrate Hoechst 33342 and MDCKII cells transfected with the human ABCG2 gene (encoding BCRP). Briefly, MDCKII cells were seeded in 96-well black cell culture plates with clear bottoms at a density of 5×10^4 cells/well and grown to confluence overnight. Test compounds were diluted in cell culture medium containing 10 μM Hoechst 33342 and incubated for 3 hours with MDCKII-BCRP and nontransfected cells. Following removal of media containing Hoechst 33342 and test compound, cells were washed twice with warm medium and lysed at room temperature for 5-10 minutes in a buffer containing 20 mM Tris-HCl pH 9.0 and 0.4% Triton X-100. Wells were analyzed for Hoechst 33342 fluorescence at an excitation of 353 nm and an emission of 460 nm.

[0320] Pgp and BCRP substrate assays in transfected MDCKII cells: MDCKII cells were grown to confluence over 4-6 days on 24-well PET transwell plates (BD Biosciences). The same buffers were used in the donor and receiver wells as described above for caco-2 studies. Experiments were conducted as described above for caco-2 transepithelial transport studies and samples analyzed by LC/MS/MS. Similar quality control and acceptance criteria were used as those described above for caco-2 studies. TEER values and the permeability of lucifer yellow, atenolol, and propranolol were determined to meet acceptance criteria for each batch of assay plates. Efflux ratios were determined to be at least 3-fold higher in transfected versus nontransfected monolayers for the model Pgp substrate vinblastine and BCRP substrate prazosin.

[0321] Data analysis: The 50% inhibition constants (IC_{50}) values for transporters in the fluorescent accumulation studies done in MDCKII cells, defined as the test article concentration needed to inhibit the maximal transporter specific transport by 50%, were calculated using nonlinear curve fitting of inhibition versus concentration to a sigmoidal curve with a variable Hill coefficient using GraphPad Prism 5 (GraphPad Software Inc., San Diego, Calif.). Apparent permeability coefficients and efflux ratios (ER) from transcellular experiments in caco-2 or MDCKII cells were calculated as previously described (Tong et al. (2007) *Antimicrob Agents Chemother* 51:3498-504). Where appropriate, the statistical significance of differences observed between test conditions was assessed using paired two-tailed Student's t tests.

[0322] Inhibition of Pgp and BCRP in transfected MDCKII cells: The inhibition of Pgp and BCRP by cobicistat relative to ritonavir and the known transport inhibitors cyclosporin A (CSA) and fumitremorgin C was studied by monitoring the effects of coincubation on the Pgp- and BCRP-dependent accumulation of the fluorescent probe substrates calcein AM and Hoechst 33342 in MDCKII-MDR1 and MDCKII-ABCG2 cells, respectively. Cobicistat inhibited Pgp and BCRP with IC_{50} values of 36±10 μM and 59±28 μM, respectively. Ritonavir, when incubated at its approximate solubility limit in assay buffers (20 μM) showed 35% inhibition of Pgp and 21% inhibition of BCRP. Higher concentrations of cobicistat were achievable in assays because of its >35-fold higher aqueous solubility at neutral pH. Greater differences in the concentrations of cobicistat and ritonavir may exist in the gastrointestinal (GI) tract based on their respective solubility under acidic conditions. Taken together, the solubility and inhibition results indicate that cobicistat should have similar inhibition of Pgp and BCRP in the GI tract relative to ritonavir.

[0323] Pgp and BCRP substrate assays in transfected MDCKII cells: To further characterize the mechanism interaction of cobicistat with Pgp (multidrug resistance protein 1; MDR1) and BCRP, bidirectional permeability assays were completed in cells transfected with the genes for the human transport proteins to determine if cobicistat is a substrate for these efflux transporters (FIG. **10**). Bidirectional permeability of cobicistat (10 μM) was assessed in MDCKII-WT, MDCKII-MDR1 (FIG. **10**A) and MDCKII-BCRP cells (FIG. **10**B). The black bars show apical to basolateral (A-B) permeability, and the open bars show basolateral to apical (B-A) permeability. Efflux ratios are indicated above graphs for each experimental condition. CSA (10 μM) and Ko134 (10 μM) were used as known inhibitors of Pgp and BCRP, respectively. Results are the average of duplicate wells from a representative side by side experiment done comparing wild type MDCKII (MDCKII-WT) to MDCKII-MDR1 or MDCKII-BCRP cells in the presence or absence of respective inhibitors. The overexpression of Pgp or BCRP in MDCKII cells increased the efflux ratios of cobicistat. These increased efflux ratios reflected a decrease in the forward permeability and an increase in the reverse permeability of cobicistat. Consistent with Pgp- and BCRP-dependent transport, cobicistat efflux was decreased in the presence of the Pgp inhibitor CSA and the BCRP inhibitor Ko 134. These results illustrate that cobicistat is a substrate for both Pgp and BCRP, suggesting that the observed inhibition may be due to competition for the binding sites of the respective transporters.

[0324] Effect of cobicistat on the bidirectional permeability of model Pgp and BCRP substrates through caco-2 cell monolayers: Caco-2 cells have been reported as a physiologically relevant model system of GI absorption that supports the polarized expression of intestinal transporters including Pgp and BCRP. The effect of cobicistat (COBI; 90 μM) and ritonavir (RTV; 20 μM) on the bidirectional permeability through monolayers of caco-2 cells of 10 μM of the Pgp substrate digoxin (FIG. **11**A) and BCRP substrate prazosin (FIG. **11**B) were studied. Digoxin and prazosin were chosen as model substrates of Pgp and BCRP, respectively, based on recommendations from the FDA and by the International Transporter Consortium. The known Pgp inhibitor CSA (10 μM) and BCRP inhibitor fumitremorgin C (2 μM; noted in FIG. **11**B as "FTC") were used as positive controls. The black bars show apical to basolateral (A-B) and the open bars basolateral to apical (B-A) permeability, and efflux ratios are indicated above graphs for each experimental condition. Results are the mean±standard deviation of at least four independent experiments done in duplicate, and statistical significance was assessed by comparing results to no cotreatment wells using paired two-tailed Student's t tests (*, $P<0.05$; **, $P<0.01$). Similar to the known Pgp inhibitor CSA, cobicistat and ritonavir markedly reduced the efflux ratio and significantly increased the apical to basolateral (A-B) permeability of digoxin (FIG. **11**A). Similar effects were observed in experiments studying the effect of cobicistat and ritonavir relative to the known BCRP inhibitor fumitremorgin C on the permeability of the BCRP substrate prazosin (FIG. **11**B). These data suggest similar inhibitory effects of cobicistat and

ritonavir on the Pgp-mediated transport of digoxin- and BCRP-mediated transport of prazosin.

[0325] Effect of cobicistat on the bidirectional permeability of HIV protease inhibitors and GS-7340 through caco-2 cell monolayers: The effect of cobicistat (90 μM) and ritonavir (20 μM) on the bidirectional permeability of the HIV protease inhibitors (PIs) atazanavir, darunavir, lopinavir, and GS-8374, an experimental HIV PI, through caco-2 cell monolayers was assessed. The effect of RTV and COBI was assessed with 10 μM of the HIV PIs atazanavir (FIG. **12**A), darunavir (FIG. **12**B), lopinavir (FIG. **12**C) and GS-8374 (FIG. **12**D). The black bars show apical to basolateral (A-B) and the open bars basolateral to apical (B-A) permeability, and efflux ratios are indicated above graphs for each experimental condition. Results are the mean±standard deviation of at least four independent experiments done in duplicate, and statistical significance was assessed comparing directional results to no cotreatment wells by using paired two-tailed Student's t tests (*, $P<0.05$; **, $P<0.01$; ***, $P<0.001$). The effect of COBI (90 μM) was assessed on the bidirection permeability of GS-7340 (10 μM) through caco-2 monolayers over a 2 hour time course in the A-B (FIG. **12**E) and B-A (FIG. **12**F) directions. Open symbols depict presence and solid symbols depict absence of COBI. Results are the mean±standard deviation of duplicate measurements from two independent experiments. Consistent with previous studies reporting these compounds as Pgp substrates, significant efflux was observed for each of the protease inhibitors. Coadministration of cobicistat and ritonavir comparably reduced the efflux ratios by increasing the A-B flux and decreasing the B-A flux of the protease inhibitors (FIG. **12**A-D). The effect of cobicistat on GS-7340 permeability across caco-2 monolayers was monitored over 2 hours, and cobicistat increased the A-B flux of GS-7340 while concomitantly reducing B-A flux (FIG. **12**E-F).

[0326] These results support the hypothesis that cobicistat may be acting to inhibit Pgp-mediated intestinal secretion of GS-7340.

Biological Example 2

[0327] Pharmacokinetic studies were done in humans to determine exposure to GS-7340 at three dose levels. Eligible subjects were randomized to receive either GS-7340 dose of 8 mg, GS-7340 dose of 25 mg, GS-7340 dose of 40 mg, tenofovir (as TDF) 300 mg or placebo-to-match GS-7340 for 10 days. (Note: Doses of GS-7340 are given as the mass of free base of GS-7340, even where other forms of GS-7340 were dosed.) GS-7340 was administered in a blinded fashion, unless a subject was randomized to receive tenofovir which was given on an open-label basis.

[0328] FIG. **1** shows tenofovir plasma concentrations in patients on Day 1 of the study. The top line (no symbol) shows the concentration of tenofovir in patients dosed with 300 mg tenofovir (as TDF). The next line down (triangles pointed down) shows the concentration of tenofovir in patients dosed with 40 mg GS-7340. The next line down (triangles pointed up) shows the concentration of tenofovir in patients dosed with 25 mg GS-7340. The bottom line (squares) shows the concentration of tenofovir in patients dosed with 8 mg GS-7340. The table below the graph shows Cmax and AUC values obtained.

[0329] FIG. **2** shows tenofovir plasma concentrations in patients on Day 10 of the study. The top line (diamonds) shows the concentration of tenofovir in patients dosed with 300 mg tenofovir. The next line down (triangles pointed down) shows the concentration of tenofovir in patients dosed with 40 mg GS-7340. The next line down (triangles pointed up) shows the concentration of tenofovir in patients dosed with 25 mg GS-7340. The bottom line (squares) shows the concentration of tenofovir in patients dosed with 8 mg GS-7340. The table below the graph shows Cmax and AUC values obtained.

Biological Example 3

[0330] Drug interaction potential between once-daily emtricitabine (FTC)/GS-7340 fixed dose combination, cobicistat boosted darunavir plus GS-7340 as a single agent, and efavirenz or cobicistat-boosted darunavir was evaluated in an open-label, crossover, single-center, multiple-dose, multiple-cohort study.

[0331] Table 4 shows the dosing regimen and schedule for the study.

TABLE 4

Cohort 1 (n = 12)		
Cohort	Day 1-12	Day 13-26
	Treatment A: FTC/GS-7340 FDC (200/40 mg) administered once-daily in the morning under fasted condition	Treatment B: FTC/GS-7340 FDC (200/40 mg) plus efavirenz (EFV) 600 mg administered once-daily in the morning under fasted condition
Cohort 2 (n = 12)		
Cohort	Day 1-12	Day 13-22
	Treatment C: FTC/GS-7340 FDC (200/25 mg) administered once-daily in the morning under fed condition	Treatment D: FTC/GS-7340 FDC (200/25 mg) plus cobicistat-boosted darunavir (DRV/co; 800/150 mg) administered once-daily in the morning under fed condition
Cohort 3 (n = 14)		
Cohort	Day 1-10	Day 11-22
	Treatment E: Cobicistat boosted darunavir (DRV/co; 800/150 mg)	Treatment F: FTC/GS-7340 FDC (200/25 mg) plus cobicistat boosted darunavir (DRV/co, 800/150 mg)

TABLE 4-continued

	administered once-daily in the morning under fed condtion	adminstered once daily in the morning under fed condition
	Cohort 4 (n = 12)	
Cohort	Day 1-12	Day 13-22
	Treatment G: GS-7340 (8 mg) single agent administered once daily in the morning under fed conditions	Treatment H: GS-7340 (8 mg) single agent PLUS cobicistat (150 mg) administered once daily in the morning under fed Conditions

[0332] Results of the pharmacokinetic analysis in this study are shown in FIGS. **3-5**. (Note: Doses of GS-7340 are given as the mass of free base of GS-7340, even where other forms of GS-7340 were dosed.)

[0333] FIG. **3**A shows GS-7340 (tenofovir alafenamide) concentrations (ng/ml) for doses of emtricitabine and GS-7340 (triangles pointed up) and emtricitabine, GS-7340 and efavirenz ((initial value=100 ng/ml); triangles pointed down) in patients from Cohort 1. C_{max} and AUC results are displayed in the table below for GS-7340 exposure. Tenofovir (TFV) concentrations are shown in FIG. **3**B for doses of emtricitabine and GS-7340 (upper line; triangles pointed up) and emtricitabine, GS-7340 and efavirenz (lower line: triangles pointed down). C_{max} and AUC results are displayed in the table below for tenofovir exposure.

[0334] FIG. **4**A shows GS-7340 concentrations (ng/ml) for doses of emtricitabine and GS-7340 (triangles pointed up) and emtricitabine, GS-7340, darunavir, and cobicistat (triangles pointed down) in patients from Cohort 2. C_{max} and AUC results are displayed in the table below for GS-7340 exposure. Tenofovir (TFV) concentrations are shown in FIG. **4**B for doses of emtricitabine and GS-7340 (triangles pointed up) and emtricitabine, GS-7340, darunavir, and cobicistat (triangles pointed down). C_{max} and AUC results are displayed in the table below for tenofovir exposure.

[0335] FIG. **5**A shows GS-7340 concentrations (ng/ml) for doses of GS-7340 alone and GS-7340 and cobicistat (triangles pointed up). C_{max} and AUC results are displayed in the table below for GS-7340 exposure. Tenofovir (TFV) concentrations are shown in FIG. **5**B for doses of GS-7340 alone (triangles pointed up) and GS-7340 and cobicistat (triangles pointed down). C_{max} and AUC results are displayed in the table below for tenofovir exposure.

[0336] Increases in exposures were observed for GS-7340 (tenofovir alafenamide) and TFV when dosed as GS-7340 (8 mg) plus COBI (150 mg) versus GS-7340 (8 mg) as a standalone agent. GS-7340 AUC_{last} and C_{max} were ~2.7- and 2.8-fold higher, respectively, whereas TFV AUC_{tau} and C_{max} were ~3.3- and 3.3-fold higher, respectively. These data suggest that the interaction is COBI-mediated, likely due to inhibition of Pgp-mediated intestinal secretion of tenofovir alafenamide (GS-7340).

Biological Example 4

[0337] GS-7340 and cobicistat were administered in conjunction with elvitegravir and emtricitabine in a clinical trial to determine the relative bioavailability of these compounds. The compounds were administered using a 25 mg or 40 mg dose of GS-7340 (test) relative to exposures (elvitegravir, cobicistat, emtricitabine) from elvitegravir/cobicistat/emtricitabine/tenofovir (reference) or GS-7340 (TFV) (reference). A second cohort with a similar design evaluated an alternate formulation of elvitegravir/cobicistat/emtricitabine/GS-7340 STR. (Note: Doses of Compound are given as the mass of free base of GS-7340, even where other forms of GS-7340 were dosed.) Elvitegravir/cobicistat/emtricitabine/GS-7340 (monolayer) tablets were manufactured by blending of emtricitabine/GS-7340 granulation with elvitegravir granulation and cobicistat, tablet compression, tablet film-coating, and packaging. Elvitegravir/cobicistat/emtricitabine/GS-7340 bilayer tablets are manufactured by compression of the elvitegravir/cobicistat layer and emtricitabine/GS-7340 layer, tablet film-coating, and packaging. In order to provide a robust assessment of pharmacokinetic comparisons between test versus reference treatments, a balanced Williams 4×4 design was used in each cohort.

[0338] The dose of elvitegravir (150 mg), the boosting dose of cobicistat (150 mg), and dosage of emtricitabine (200 mg) in elvitegravir/cobicistat/emtricitabine/GS-7340 represent current investigational doses (elvitegravir, cobicistat) or marketed dose (emtricitabine) with demonstrated durable efficacy and long-term safety in HIV-infected patients.

[0339] The evaluation used two cohorts of twenty patients. In Cohort 1, the following study treatments were administered.

[0340] Treatment A: 1×Single Tablet Regimen (STR) of Formulation 1 (150 mg elvitegravir plus 150 mg cobicistat plus 200 mg emtricitabine plus 25 mg GS-7340 (as 31.1 mg of the fumarate salt GS-7340-02)) QD, administered in A.M. for 12 days.

[0341] Treatment B: 1×STR Formulation 1 (150 mg elvitegravir plus 150 mg cobicistat plus 200 mg emtricitabine plus 40 mg GS-7340 (as 49.7 mg of the fumarate salt GS-7340-02)) QD, administered in A.M. for 12 days.

[0342] Treatment C: 1×STR (150 mg elvitegravir plus 150 mg cobicistat plus 200 mg emtricitabine plus 300 mg tenofovir (as tenofovir disoproxil fumarate) QD, administered in A.M. for 12 days.

[0343] Treatment D: 1×25 mg GS-7340 tablet QD, administered in A.M. for 12 days.

[0344] Patients were randomized to one of four sequences (I, II, III, or IV).

	Day 1-12	Day 15-26	Day 29-40	Day 43-54
Sequence I	A	B	C	D
Sequence II	B	D	A	C
Sequence III	C	A	D	B
Sequence IV	D	C	B	A

[0345] Formulation 1 (monolayer) was prepared by blending of emtricitabine/GS-7340 granulation with elvitegravir granulation and cobicistat, tablet compression, tablet film-coating, and packaging. The EVG/COBI/FTC/GS-7340 STR

tablet cores contain colloidal silicon dioxide, croscarmellose sodium, hydroxypropyl cellulose, lactose monohydrate, microcrystalline cellulose, sodium lauryl sulfate, and magnesium stearate as inactive ingredients and are film-coated with polyvinyl alcohol, polyethylene glycol, talc, and titanium dioxide.

[0346] In Cohort 2, the following study treatments were administered:

[0347] Treatment E: 1×STR Formulation 2 (150 mg elvitegravir plus 150 mg cobicistat plus 200 mg emtricitabine plus 25 mg GS-7340 (as 31.1 mg of the fumarate salt GS-7340-02)) QD, administered in A.M. for 12 days.

[0348] Treatment F: 1×STR Formulation 2 (150 mg elvitegravir plus 150 mg cobicistat plus 200 mg emtricitabine plus 40 mg GS-7340 (as 49.7 mg of the fumarate salt GS-7340-02)) QD, administered in A.M. for 12 days.

[0349] Treatment C: 1×STR (150 mg elvitegravir plus 150 mg cobicistat plus 200 mg emtricitabine plus 300 mg tenofovir) QD, administered in A.M. for 12 days.

[0350] Treatment D: 1×25 mg GS-7340 tablet QD, administered in A.M. for 12 days.

[0351] Patients were randomized to one of four sequences (I, II, III, or IV).

	Day 1-12	Day 15-26	Day 29-40	Day 43-54
Sequence I	E	F	C	D
Sequence II	F	D	E	C
Sequence III	C	E	D	F
Sequence IV	D	C	F	E

[0352] Formulation 2 was prepared as bilayer tablets that were manufactured by compression of the elvitegravir/cobicistat layer and emtricitabine/GS-7340 layer, tablet film-coating, and packaging. The EVG/COBI/FTC/GS-7340 STR tablet cores contain colloidal silicon dioxide, croscarmellose sodium, hydroxypropyl cellulose, lactose monohydrate, microcrystalline cellulose, sodium lauryl sulfate, and magnesium stearate as inactive ingredients and are film-coated with polyvinyl alcohol, polyethylene glycol, talc, and titanium dioxide.

[0353] FIG. **6** shows pharmacokinetic data for GS-7340 from patients treated in Cohort 1 (Formulation 1, monolayer). The top line (triangles pointed down) shows GS-7340 concentration (ng/ml) when 40 mg GS-7340 is administered with cobicistat. The middle line (triangles pointed up) shows GS-7340 concentration (ng/ml) when 25 mg GS-7340 is administered with cobicistat. The bottom line (squares) shows GS-7340 concentration (ng/ml) when 25 mg GS-7340 is administered alone. These results show GS-7340 levels that are 2.2-fold higher for dosing at the 25 mg level when GS-7340 is administered with cobicistat.

[0354] FIG. **7** shows pharmacokinetic data for GS-7340 from patients treated in Cohort 2 (Formulation 2, bilayer). The top line (triangles pointed down) shows GS-7340 concentration (ng/ml) when 40 mg GS-7340 is administered with cobicistat. The middle line (triangles pointed up) shows GS-7340 concentration (ng/ml) when 25 mg GS-7340 is administered with cobicistat. The bottom line (squares) shows GS-7340 concentration (ng/ml) when 25 mg GS-7340 is administered alone. These results also show GS-7340 levels that are 2.2-fold higher for dosing at the 25 mg level when GS-7340 is administered with cobicistat.

[0355] FIG. **8** shows pharmacokinetic data for tenofovir from patients treated in Cohort 1 (Formulation 1, monolayer). The top line (no symbol) shows tenofovir concentration (ng/ml) when 300 mg tenofovir is administered with cobicistat. The next line down (triangles pointed up) shows tenofovir concentration (ng/ml) when 40 mg GS-7340 is administered with cobicistat. The next line down (squares) shows tenofovir concentration (ng/ml) when 25 mg GS-7340 is administered with cobicistat. The bottom line (triangles pointed down) shows tenofovir concentration (ng/ml) when 25 mg GS-7340 is administered alone. These results also show tenofovir levels that are 3-4 fold higher for dosing at the 25 mg level when tenofovir or GS-7340 is administered with cobicistat.

[0356] FIG. **9** shows pharmacokinetic data for tenofovir from patients treated in Cohort 2 (Formulation 2, bilayer). The top line (circles) shows tenofovir concentration (ng/ml) when 300 mg tenofovir is administered with cobicistat. The next line down (triangles pointed up) shows tenofovir concentration (ng/ml) when 40 mg GS-7340 is administered with cobicistat. The next line down (squares) shows tenofovir concentration (ng/ml) when 25 mg GS-7340 is administered with cobicistat. The bottom line (triangles pointed down) shows tenofovir concentration (ng/ml) when 25 mg GS-7340 is administered alone. These results also show GS-7340 levels that are 3-4 fold higher for dosing at the 25 mg level when tenofovir or GS-7340 is administered with cobicistat.

[0357] Following administration of EVG/COBI/FTC/GS-7340 (25 mg) Formulations 1 and 2, geometric mean GS-7340 and TFV exposures were substantially higher, relative to GS-7340 (25 mg) as a stand-alone agent. With both formulations of EVG/COBI/FTC/GS-7340 (25 mg), GS-7340 AUC_{last} and C_{max} were ~2.2- and 2.3-fold higher, respectively, whereas TFV AUC_{tau} and C_{max} were ~3.1- and 3.7-fold higher, respectively. GS-7340 and TFV exposures were generally dose-proportional following EVG/COBI/FTC/GS-7340 (40 mg) versus EVG/COBI/FTC/GS-7340 (25 mg).

Biological Example 5

[0358] GS-7340 was coformulated with elvitegravir (EVG), cobicistat (COBI), and emtricitabine (FTC) into a single tablet regimen (STR). Across three healthy subject studies, the multiple dose pharmacokinetics (PK) of EVG/COBI/FTC/GS-7340 STR and/or interaction potential between GS-7340 and COBI were evaluated to facilitate GS-7340 dose selection for STR clinical development.

[0359] In Study 1 (n=20), subjects received EVG/COBI/FTC/GS-7340 (150/150/200/40 or 150/150/200/25 mg), EVG/COBI/FTC/TDF (150/150/200/300 mg) or GS-7340 25 mg stand alone (SA), 12 days/treatment in a balanced Williams 4×4 design. In Study 2 (n=12), subjects sequentially received GS-7340 (8 mg) SA (Reference) for 12 days and GS-7340 plus COBI (8/150 mg) (Test) for 10 days. In Study 3 (n=34), across two cohorts (each 2×2 crossover design), subjects received EVG/COBI/FTC/GS-7340 (150/150/200/10 mg) (Test, both cohorts), EVG plus COBI (150/150 mg) (Reference, Cohort 1), and FTC plus GS-7340 (200/25 mg) (Reference, Cohort 2), each treatment dosed for 12 days. Statistical comparisons of GS-7340 and TFV were made using geometric mean ratios (GMR), with 90% confidence intervals (CI) of 70-143% (Study 1: Test=EVG/COBI/FTC/GS-7340, Reference=GS-7340 SA). Safety assessments were performed throughout dosing and follow up.

[0360] All treatments were generally well tolerated. Study 1 entailed 19/20 completers with one discontinuation from adverse events (AEs) (rhabdomyolysis (Grade 2) while receiving GS-7340 SA). All subjects completed Study 2, while 33 of 34 subjects completed Study 3. No Grade 3 or 4 AE was observed in the studies. In Study 1, when dosed as EVG/COBI/FTC/GS-7340, GS-7340 (25 mg) and resulting TFV exposures were substantially higher versus GS-7340 SA (GMR (90% CI) GS-7340 AUC_{last}: 222 (200, 246) and C_{max}: 223 (187, 265); TFV AUC_{tau}: 307 (290, 324), C_{max}: 368 (320, 423)). In Study 2, when dosed as GS-7340 plus COBI versus GS-7340 SA, GS-7340 exposures were similarly high, suggesting that the interaction observed in Study 1 was COBI-mediated (GMR (90% CI) GS-7340 AUC_{last}: 265 (229, 307) and C_{max}: 283 (220, 365, TFV AUC_{tau}: 331 (310, 353), C_{max}: 334 (302, 370), and C_{tau}: 335 (312, 359)). In Study 3, upon dose adjustment of GS-7340 to 10 mg, EVG/COBI/FTC/GS-7340 (150/150/200/10 mg) versus Reference resulted in comparable GS-7340 and TFV exposures. (GMR (90% CI) GS-7340 AUC_{last}: 89.0 (76.7, 103) and C_{max}: 97.3 (82.1, 115), TFV AUC_{last}: 124 (113, 136), C_{max}: 113 (98.8, 129), and C_{tau}: 120 (103, 140)). EVG/COBI/FTC/GS-7340 STR provided similar EVG, COBI, and FTC exposures versus reference treatments and historical data.

[0361] GS-7340 and TFV exposures increase ~2-3 fold following coadministration with COBI or as EVG/COBI/FTC/GS-7340 dosing, which may be due to COBI inhibition of Pgp-mediated intestinal secretion of GS-7340. With a 10 mg dose of GS-7340, EVG/COBI/FTC/GS-7340 provided comparable GS-7340 and TFV exposures as GS-7340 at 25 mg and ~90% lower TFV exposure versus EVG/COBI/FTC/TDF.

Biological Example 6

[0362] EVG/COBI/FTC/TDF and EVG/COBI/FTC/tenofovir alafenamide hemifumarate were administered as single tablet regimens (STR) in a Phase 2 clinical trial evaluating safety and efficacy in HIV+treatment-naïve adults. All subjects had HIV-1 RNA >5000 c/ml. Week 24 data indicated that treatment with the two STRs resulted in 87% of subjects on EVG/COBI/FTC/tenofovir alafenamide hemifumarate and 90% of subjects on EVG/COBI/FTC/TDF having HIV-1 RNA <50 c/ml. The EVG/COBI/FTC/tenofovir alafenamide hemifumarate STR was well tolerated, and relative to the known safety profile of EVG/COBI/FTC/TDF, no new or unexpected adverse drug reactions were identified.

[0363] Renal function was assessed in the subjects at week 24. When compared with subjects taking EVG/COBI/FTC/TDF, subjects taking EVG/COBI/FTC/tenofovir alafenamide hemifumarate had significantly less reduction in the estimated glomerular filtration rate (eGFR), a trend towards less proteinuria, and statistically less tubular proteinuria. These differences may represent a reduction in subclinical tenofovir-associated nephrotoxicity.

[0364] To assess bone mineral density, dual-energy X-ray absorptiometry scans were performed at baseline and week 24. Subjects taking EVG/COBI/FTC/tenofovir alafenamide hemifumarate experienced a significantly smaller reduction in bone mineral density at both spine and hip after 24 weeks, compared with subjects taking EVG/COBI/FTC/TDF. Importantly, the proportion of subjects with >3% decrease from baseline in hip bone mineral density was 10-fold lower in the EVG/COBI/FTC/tenofovir alafenamide hemifumarate group than the EVG/COBI/FTC/TDF group (3.0% vs. 31.6%).

[0365] Together, these data support the hypothesis that TDF-associated renal and bone toxicity is driven by circulating tenofovir, as tenofovir levels are reduced by 90% in subjects administered EVG/COBI/FTC/tenofovir alafenamide hemifumarate.

[0366] All references, publications, patents, and patent documents cited herein are incorporated by reference herein, as though individually incorporated by reference. The invention has been described with reference to various specific and preferred embodiments and techniques. However, it should be understood that many variations and modifications may be made while remaining within the spirit and scope of the invention.

[0367] The use of the terms "a," "an," "the," and similar articles and the like in the context of describing the invention (including the following claims) are to be construed to cover both the singular and the plural, unless otherwise indicated herein or clearly contradicted by context. The terms "comprising," "having," "including," and "containing" are to be construed as open-ended terms (i.e., meaning "including, but not limited to"), unless otherwise noted. Recitation of ranges of values herein are merely intended to serve as a shorthand method of referring individually to each separate value falling within the range, unless otherwise indicated herein, and each separate value is incorporated into the specification as if it were individually recited herein. All methods described herein may be performed in any suitable order unless otherwise indicated herein or otherwise clearly contradicted by context. The use of any and all examples, or exemplary language (e.g., "such as") provided herein, is intended merely to better illuminate the invention and does not pose a limitation on the scope of the invention, unless otherwise claimed. No language in the specification should be construed as indicating any nonclaimed element as essential to the practice of the invention.

[0368] The embodiments within the specification provide an illustration of embodiments of the invention and should not be construed to limit the scope of the invention. The skilled artisan recognizes that many other embodiments are encompassed by the claimed invention and that it is intended that the specification and examples be considered as exemplary only, with the true scope and spirit of the invention being indicated by the following claims.

1. A composition comprising: cobicistat, or a pharmaceutically acceptable salt thereof; and tenofovir alafenamide hemifumarate.

2. The composition of claim **1** comprising: 50-500 mg of cobicistat, or a pharmaceutically acceptable salt thereof; and 3-40 mg of tenofovir alafenamide hemifumarate.

3. The composition of claim **1**, further comprising a pharmaceutically acceptable carrier or diluent.

4. A method of treating a viral infection in a human comprising administering a composition of claim **1** to the human.

5. A method of treating a viral infection in a human comprising coadministering cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, to the human.

6. A method of inhibiting activity of a retroviral reverse transcriptase comprising coadministering cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate.

7. The method of claim **6**, wherein the coadministering of cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate, is in a human.

8-11. (canceled)

12. A method of boosting an anti-viral effect of tenofovir alafenamide hemifumarate in a human comprising administering a composition of claim **1** to the human.

13. A method of boosting an anti-viral effect of tenofovir alafenamide hemifumarate in a human comprising coadministering cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate to the human.

14. The method of claim **13**, wherein 50-500 mg of cobicistat, or a pharmaceutically acceptable salt thereof, is coadministered with 3-40 mg of tenofovir alafenamide hemifumarate.

15. A method of inhibiting Pgp-mediated intestinal secretion of tenofovir alafenamide hemifumarate in a human comprising administering a composition of claim **1** to the human.

16. A method of inhibiting Pgp-mediated intestinal secretion of tenofovir alafenamide hemifumarate in a human by coadministration of cobicistat, or a pharmaceutically acceptable salt thereof, and tenofovir alafenamide hemifumarate.

17. The method of claim **16**, wherein 50-500 mg of cobicistat, or a pharmaceutically acceptable salt thereof, is coadministered with 3-40 mg of tenofovir alafenamide hemifumarate.

18. The method of claim **5**, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

19. (canceled)

20. The method of claim **13**, wherein the virus is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

21. A composition comprising: (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) elvitegravir.

22. A composition comprising: (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 50-500 mg cobicistat, or a pharmaceutically acceptable salt thereof; (c) 50-500 mg emtricitabine; and (d) 50-500 mg elvitegravir.

23. A method of treating a viral infection in a human comprising administering a composition of claim **21** to the human.

24. A method of treating a viral infection in a human comprising coadministering (a) tenofovir alafenamide hemifumarate; (b) cobicistat, or a pharmaceutically acceptable salt thereof; (c) emtricitabine; and (d) elvitegravir to the human.

25. The method of claim **24** comprising coadministering (a) 3-40 mg tenofovir alafenamide hemifumarate; (b) 50-500 mg cobicistat, or a pharmaceutically acceptable salt thereof; (c) 50-500 mg emtricitabine; and (d) 50-500 mg elvitegravir to the human.

26-30. (canceled)

31. The method of claim **24**, wherein the viral infection is human immunodeficiency virus (HIV) or Hepatitis B virus (HBV).

32. (canceled)

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(54) Title: PHARMACEUTICAL ANTIRETROVIRAL COMPOSITIONS

(57) Abstract: The present invention relates to pharmaceutical antiretroviral compositions comprising a combination of antiretroviral agents, the manufacturing process thereof and use of the said compositions for the prevention, treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection.

PHARMACEUTICAL ANTIRETROVIRAL COMPOSITIONS

FIELD OF INVENTION

The present invention relates to pharmaceutical antiretroviral compositions comprising a combination of antiretroviral agents, the manufacturing process thereof and use of the said compositions for the prevention, treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection.

BACKGROUND AND PRIOR ART

Demographically the second largest country in the world, India also has the third largest number of people living with HIV/AIDS. The total number of people living with HIV (PLHIV) in India is estimated at 2.4 million with uncertainty bounds of 1.93 to 3.04 million in 2009. Children under 15 years of age account for 4.4% of all infections, whilst people aged 15 to 49 years account for 82.4% of all infections. Thirty-nine percent of all HIV infections are estimated to be among women. This amounts to 0.93 million women with HIV in India.

Acquired Immune Deficiency Syndrome (AIDS) causes a gradual breakdown of the body's immune system as well as progressive deterioration of the central and peripheral nervous systems. Since its initial recognition in the early 1980's, AIDS has spread rapidly and has now reached epidemic proportions within a relatively limited segment of the population. Intensive research has led to the discovery of the responsible agent, human T-lymphotropic retrovirus 111 (HTLV-111), now more commonly referred to as the human immunodeficiency viruses or HIV.

Human immunodeficiency virus (HIV) is the etiological agent of Acquired Immune Deficiency Syndrome (AIDS) that has created a major health care problem not only in India but also globally.

HIV is a member of the class of viruses known as retroviruses. The retroviral genome is composed of RNA, which is converted to DNA by reverse transcription. This retroviral DNA is then stably integrated into a host cell's chromosome and, employing the replicative processes of the host cells, produces new retroviral particles and advances the infection to other cells. HIV appears to have a particular affinity for the human T- 4 lymphocyte cell which plays a vital role in the body's immune system. HIV infection of these white blood cells depletes this white cell population. Eventually, the immune system is rendered inoperative and ineffective against various opportunistic diseases.

The current strategy recommended for the treatment of HIV infection is Highly Active Antiretroviral Therapy (HAART). HAART normally consists of a combination of three or more antiretroviral drugs (ARV) taken together.

Currently available antiretroviral drugs for the treatment of HIV include nucleoside reverse transcriptase inhibitors (NRTI) or approved single pill combinations: zidovudine or AZT (Retrovir®), didanosine or DDI (Videx®), stavudine or D4T (Zenith®), lamivudine or 3TC (Epivir®), zalcitabine or DDC (Hivid®), abacavir sulphate (Ziagen®), tenofovir disoproxil fumarate salt (Viread®), emtricitabine (Emtriva®), Combivir® (contains 3TC and AZT), Trizivir® (contains abacavir, 3TC and AZT); non-nucleoside reverse transcriptase inhibitors (NNRTI): nevirapine (Viramune®), delavirdine (Rescriptor®) and efavirenz (Sustiva®), peptidomimetic protease inhibitors or approved formulations: saquinavir (Invirase®, Fortovase®), indinavir (Crixivan®), ritonavir (Norvir®), nelfinavir (Viracept®), amprenavir (Agenerase®), atazanavir (Reyataz®), fosamprenavir (Lexiva®), Kaletra® (contains lopinavir and ritonavir), one fusion inhibitor enfuvirtide (T-20, Fuzeon®), Truvada® (contains Tenofovir and Emtricitabine) and Atripla® (contains fixed-dose triple combination of tenofovir, emtricitabine and efavirenz).

The goal of HAART therapy is to maximize viral suppression thus limiting and reversing damage to the immune system, leading to decline of opportunistic infections. The durability of response depends on various factors such as viral, drug and patient related factors. However, the most important patient related factor is adherence, to ensure the success of HAART therapy.

The HIV therapy is a life-long therapy coupled with high levels of adherence to the same. This is rather a demanding task for HIV infected patients due to various reasons such as low morale, social stigma, low immunity attributed to the disease.

Further, the therapy may involve use of different drug combinations, which are difficult to adhere, because of the different dosage forms for administering each such as antiretroviral drug separately. This is particularly of importance in case of elderly patients.

Further some studies have shown that adherence to prescribed drugs over long treatment periods is generally poor. (Jintanat A. et al. Swiss HIV Cohort Study. Failures of 1 week on, 1 week off antiretroviral therapies in a randomized trial AIDS, 2003; 17:F33-F37).

Hence, such non-adherence to HAART can lead to rebound in viral replication and, in presence of sub-optimal drug concentration may lead to rapid development of drug resistance. This development of drug resistance can be disastrous because of the complexity and cost associated with second line regimens and the potential for transmission of drug resistant virus in the community.

For most of the therapeutic agents, to produce systemic effects, the oral route still represents the preferred way of administration, owing to its several advantages and high patient compliance as compared to any other routes of administration. Tablets and hard gelatin capsules still constitute a major portion of drug delivery systems that are currently available.

However, many patient groups such as the elderly, children, and patients who are mentally retarded, uncooperative, nauseated, or on reduced liquid-intake/diets have difficulties swallowing the dosage forms such as tablets and hard gelatin capsules. Further, those who are traveling or have little access to water are similarly affected.

Also, the route of drug administration, appearance, color, taste, tablet size and dosing regimen are most important parameters that govern patient compliance.

Especially, the geriatric and pediatric patients experience difficulty in swallowing larger sized tablets wherein large size tablet may result in esophageal damage due to its physical characteristics if it is not swallowed properly, which ultimately leads to poor patient compliance.

Also, oral administration of bitter drugs with an acceptable degree of palatability is a key issue for health care providers, especially for pediatric patients.

Further, there has been an enhanced demand for dosage forms that are more patient-friendly and patient compliant. Since the development cost of a new drug molecule is very high, efforts are now being made to focus on the development of new drug dosage forms for existing drugs with improved safety and efficacy together with reduced dosing frequency as well as which are cost-effective.

Although, different treatment methods and dosage regimens have been framed in order to increase the patient adherence for treatment of HIV, there still remains a critical need for developing improved dosage forms such as a kit composition or dosage form by which a patient is encouraged to adhere to his daily dosage regimen. Keeping in mind the patient compliance and the urge to treat HIV to achieve the desired positive results avoiding repetition or change in the line of therapy due to non- adherence to the dosing regimen, the inventors of the present invention have been successful in designing kits comprising compositions of HIV drugs in line of the therapy or dosing regimens generally recommended. In particular, the present invention attempts to overcome the problems of patient adherence for the treatment of HIV.

OBJECTS OF THE INVENTION

The object of the present invention is to provide a pharmaceutical antiretroviral composition comprising at least one reverse transcriptase inhibitor or at least one integrase inhibitor and at least one protease inhibitor, preferably wherein said composition is in the form of a kit.

Another object of the present invention is to provide a pharmaceutical antiretroviral composition comprising at least one reverse transcriptase inhibitor or at least one integrase inhibitor and at least one protease inhibitor, optionally with one or more pharmaceutically acceptable excipients, preferably wherein said composition is in the form of a kit comprising a plurality of antiretroviral agents which have been co-formulated, or a plurality of dosage forms containing one or more antiretroviral agents.

Yet another object of the present invention is to provide a pharmaceutical antiretroviral composition for once or twice a day administration comprising at least one reverse transcriptase inhibitor or at least one integrase inhibitor and at least one protease inhibitor, optionally with one or more pharmaceutically acceptable excipients, preferably wherein said composition is in the form of a kit comprising a plurality of antiretroviral agents which have been co-formulated, or a plurality of dosage forms containing one or more antiretroviral agents.

Yet another object of the present invention is to provide a process for manufacturing a pharmaceutical antiretroviral composition for once or twice daily administration comprising at least one reverse transcriptase inhibitor or at least one integrase inhibitor and at least one protease inhibitor, optionally with one or more pharmaceutically acceptable excipients, preferably wherein said composition is in the form of a kit.

Another object of the present invention is to provide a method of prevention, treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection, which method comprises administering a pharmaceutical antiretroviral composition comprising at least one reverse transcriptase inhibitor or at least one integrase inhibitor and at least one protease inhibitor to a patient in need thereof, preferably wherein said composition is in the form of a kit.

Yet another object of the present invention is to provide the use of a pharmaceutical antiretroviral composition comprising at least one reverse transcriptase inhibitor or at least one integrase inhibitor and at least one protease inhibitor for the treatment or

prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection, wherein said composition is for once or twice a day administration and is preferably in the form of a kit.

SUMMARY OF THE INVENTION

In a first aspect, the present invention provides a pharmaceutical antiretroviral composition comprising:

(i) at least one reverse transcriptase inhibitor comprising: zidovudine; didanosine; stavudine; lamivudine; abacavir; adefovir; lobucavir; entecavir; apricitabine; emtricitabine; zalcitabine; dexelvucitabine; alovudine; amdoxovir; elvucitabine; tenofovir; festinavir; racivir; lersivirine; rilpivirine; etravirine; SP1093V and/or stampidine, and/or

(ii) at least one integrase inhibitor comprising: raltegravir; dolutegravir and/or elvitegravir, and

(iii) at least one protease inhibitor comprising: saquinavir; ritonavir; nelfinavir; amprenavir; lopinavir, indinavir; nelfinavir; atazanavir; lasinavir; palinavir; fosamprenavir; darunavir; and/or tiprinavir,

wherein the pharmaceutical antiretroviral composition optionally further comprises one or more pharmaceutically acceptable excipients.

The zidovudine; didanosine; stavudine; lamivudine; abacavir; adefovir; lobucavir; entecavir; apricitabine; emtricitabine; zalcitabine; dexelvucitabine; alovudine; amdoxovir; elvucitabine; tenofovir; festinavir; racivir; lersivirine; rilpivirine; etravirine; SP1093V; stampidine; raltegravir elvitegravir; dolutegravir; saquinavir; ritonavir; nelfinavir; amprenavir; lopinavir; indinavir; nelfinavir; atazanavir; lasinavir; palinavir; fosamprenavir; darunavir; and/or tiprinavir may be in the form of a pharmaceutically acceptable derivative thereof. The pharmaceutically acceptable derivative may be a salt, solvate, complex, hydrate, isomer, ester, tautomer, anhydrate, enantiomer, polymorph or prodrug. The reverse transcriptase inhibitor may comprise: lamivudine; zidovudine; tenofovir; emtricitabine and/or abacavir. The integrase inhibitor may comprise: raltegravir and/or dolutegravir. The protease inhibitor may comprise: ritonavir and/or

darunavir. The pharmaceutical antiretroviral composition may comprise lamivudine, zidovudine, darunavir and ritonavir. The pharmaceutical antiretroviral composition may comprise lamivudine, tenofovir, darunavir and ritonavir. The pharmaceutical antiretroviral composition may comprise tenofovir, emtricitabine, darunavir and ritonavir. The pharmaceutical antiretroviral composition may comprise abacavir, lamivudine, darunavir and ritonavir. The pharmaceutical antiretroviral composition may comprise raltegravir or dolutegravir, darunavir and ritonavir. The pharmaceutical antiretroviral composition may comprise: at least two reverse transcriptase inhibitors, wherein the at least two reverse transcriptase inhibitors are provided in a separate single unit dosage form; at least two protease inhibitors, wherein the at least two protease inhibitors are provided in a separate single unit dosage form; and/or at least one integrase inhibitor, wherein the at least one integrase inhibitor is provided in a separate single unit dosage form. The pharmaceutical antiretroviral composition may comprise darunavir and ritonavir provided in a separate single unit dosage form. The pharmaceutical antiretroviral composition may comprise raltegravir or dolutegravir provided in a separate single unit dosage form. The pharmaceutical antiretroviral composition may comprise lamivudine and zidovudine provided in a separate single unit dosage form. The pharmaceutical antiretroviral composition may comprise tenofovir and emtricitabine provided in a separate single unit dosage form. The pharmaceutical antiretroviral composition may comprise abacavir and lamivudine provided in a separate single unit dosage form. The pharmaceutical antiretroviral composition may comprise lamivudine and tenofovir provided in a separate single unit dosage form. The composition may be provided as a kit comprising instructions for administration. The pharmaceutical antiretroviral composition may be for once or twice daily administration. The at least one reverse transcriptase inhibitor, the at least one integrase inhibitor and the at least one protease inhibitor may be provided in a dosage form selected from: a tablet, a mini-tablet, sprinkles comprising a plurality of particles, a capsule, or a liquid. The tablet may be a disintegrating tablet, a dissolving tablet, a dispersible tablet, a mouth-dissolving tablet, a tablet for oral suspension, an immediate release tablet, an extended release tablet, an immediate and extended release tablet, or a matrix tablet. The plurality of particles of the sprinkles may be provide in the form of granules, powders, powders for reconstitution, beads, pellets, mini-tablets, film-coated tablets, film coated tablets MUPS, orally

disintegrating MUPS, pills, micro-pellets, small tablet units, MUPS, disintegrating tablets, dispersible tablets, granules, effervescent granules, or microspheres. The sprinkles may be provided in a sachet, a packet or a capsule. The one or more pharmaceutically acceptable excipient may comprise: a diluent, filler, bulking agent, disintegrant, binder, glidant, anti-adherent, lubricant, water soluble polymer, water insoluble polymer, water swellable polymer, plasticizer, and any mixture thereof.

In a second aspect, the present invention provides a process for preparing the pharmaceutical antiretroviral composition according to any preceding claim, comprising: admixing the at least one reverse transcriptase inhibitor or the at least one integrase inhibitor and the at least one protease inhibitor, optionally with the one or more pharmaceutically acceptable excipient.

In a third aspect, the present invention provides a method of preventing, treating or prophylaxis of a disease caused by a retrovirus, specifically acquired immune deficiency syndrome or an HIV infection, which method comprises administering to a patient in need thereof a pharmaceutical antiretroviral composition of the present invention.

In a fourth aspect, the present invention provides a pharmaceutical composition of the invention for use in the treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection, in a patient in need thereof.

In a fifth aspect, the present invention provides a use of the pharmaceutical antiretroviral composition of the invention for the treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection.

According to one aspect of the present invention, there is provided a pharmaceutical antiretroviral composition comprising:

(i) at least one reverse transcriptase inhibitor comprising zidovudine; didanosine; stavudine; lamivudine; abacavir; adefovir; lobucavir; entecavir; apricitabine; emtricitabine; zalcitabine; dexelvucitabine; alovudine; amdoxovir;

elvucitabine; tenofovir; festinavir; racivir; lersivirine; rilpivirine; etravirine; SP1093V and stampidine, and/or

(ii) at least one integrase inhibitor comprising raltegravir; elvitegravir; and/or dolutegravir and

(iii) at least one protease inhibitor comprising saquinavir; ritonavir; nelfinavir; amprenavir; lopinavir, indinavir; nelfinavir; atazanavir; lasinavir; palinavir;; fosamprenavir; darunavir; and/or tipranavir, and optionally

one or more pharmaceutically acceptable excipients.

Preferably, said composition is for once or twice a day administration and may be in the form of a kit comprising a plurality of antiretroviral agents have been co-formulated, or a plurality of dosage forms containing one or more antiretroviral agents.

According to another aspect of the present invention, there is provided a pharmaceutical antiretroviral composition comprising:

(i) at least one reverse transcriptase inhibitor comprising zidovudine, lamivudine, tenofovir, abacavir and emtricitabine, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; or

at least one integrase inhibitor comprising raltegravir or dolutegravir or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof and

(ii) at least one protease inhibitor comprising darunavir and ritonavir, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; and

(iii) optionally one or more pharmaceutically acceptable excipients;

preferably, said composition is for once or twice a day administration and may be in the form of a kit comprising a plurality of antiretroviral agents have been co-formulated, or a plurality of dosage forms containing one or more antiretroviral agents.

According to another aspect of the present invention, there is provided a process of manufacturing a pharmaceutical antiretroviral composition comprising:

(i) at least one reverse transcriptase inhibitor comprising zidovudine; didanosine; stavudine; lamivudine; abacavir; adefovir; lobucavir; entecavir; apricitabine; emtricitabine; zalcitabine; dexelvucitabine; alovudine; amdoxovir; elvucitabine; tenofovir; festinavir; racivir; lersivirine; rilpivirine; etravirine; SP1093V and stampidine, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; or

at least one integrase inhibitor comprising raltegravir; elvitegravir; dolutegravir or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof and

(ii) and at least one protease inhibitor comprising saquinavir; ritonavir; nelfinavir; amprenavir; lopinavir, indinavir; nelfinavir; atazanavir; lasinavir; palinavir; fosamprenavir; darunavir; and tipranavir, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; and optionally

(iii) one or more pharmaceutically acceptable excipients;

wherein said composition is for once or twice a day administration and is preferably in the form of a kit.

According to another aspect of the present invention there is provided a process of manufacturing a pharmaceutical antiretroviral composition comprising:

(i) at least one reverse transcriptase inhibitor comprising zidovudine, lamivudine, tenofovir, abacavir and emtricitabine, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; or

at least one integrase inhibitor comprising raltegravir or dolutegravir or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof and

(ii) at least one protease inhibitor comprising darunavir and ritonavir, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; and optionally

(iii) one or more pharmaceutically acceptable excipients;

wherein said composition is for once or twice a day administration and is preferably in the form of a kit.

According to yet another aspect of the present invention, there is provided a method of preventing, treating or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection, which method comprises administering to a patient in need thereof, a pharmaceutical antiretroviral composition comprising:

(i) at least one reverse transcriptase inhibitor comprising zidovudine; didanosine; stavudine; lamivudine; abacavir; adefovir; lobucavir; entecavir; apricitabine; emtricitabine; zalcitabine; dexelvucitabine; alovudine; amdoxovir; elvucitabine; tenofovir; festinavir; racivir; lersivirine; rilpivirine; etravirine; SP1093V and stampidine, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; or
at least one integrase inhibitor comprising raltegravir; elvitegravir; dolutegravir or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof and

(ii) at least one protease inhibitor comprising saquinavir; ritonavir; nelfinavir; amprenavir; lopinavir, indinavir; nelfinavir; atazanavir; lasinavir; palinavir; fosamprenavir; darunavir; and tipranavir, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; and optionally

(iii) one or more pharmaceutically acceptable excipients;

wherein said composition is in the form of a kit.

According to yet another aspect of the present invention, there is provided a method of preventing, treating or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection, which method comprises administering to a patient in need thereof, a pharmaceutical antiretroviral composition comprising:

(i) at least one reverse transcriptase comprising zidovudine, lamivudine, tenofovir, abacavir and emtricitabine, or a pharmaceutically acceptable salts, solvates,

hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; or

at least one integrase inhibitor comprising raltegravir dolutegravir or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof and

(ii) at least one protease inhibitor comprising darunavir and ritonavir, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; and optionally

(iii) one or more pharmaceutically acceptable excipients; wherein said composition is for once or twice a day administration and is preferably in the form of a kit.

According to another aspect of the present invention, there is provided the use of a pharmaceutical antiretroviral composition comprising:

(i) at least one reverse transcriptase inhibitor comprising zidovudine; didanosine; stavudine; lamivudine; abacavir; adefovir; lobucavir; entecavir; apricitabine; emtricitabine; zalcitabine; dexelvucitabine; alovudine; amdoxovir; elvucitabine; tenofovir; festinavir; racivir; lersivirine; rilpivirine; etravirine; SP1093V and stampidine, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; or

at least one integrase inhibitor comprising raltegravir; elvitegravir; dolutegravir or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof and

(ii) and at least one protease inhibitor comprising saquinavir; ritonavir; nelfinavir; amprenavir; lopinavir, indinavir; nelfinavir; atazanavir; lasinavir; palinavir; fosamprenavir; darunavir; and tipranavir, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; and optionally

(iii) one or more pharmaceutically acceptable excipients;

for the treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection, wherein said composition is for once or twice a day administration and is preferably in the form of a kit.

According to another aspect of the present invention, there is provided the use of a pharmaceutical antiretroviral composition comprising:

(i) at least one reverse transcriptase inhibitor comprising zidovudine, lamivudine, tenofovir, abacavir and emtricitabine, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; or

at least one integrase inhibitor comprising raltegravir dolutegravir or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof and

(ii) at least one protease inhibitor comprising darunavir and ritonavir, or a pharmaceutically acceptable salts, solvates, hydrates, esters, enantiomers, polymorphs, prodrugs, complexes, derivatives thereof; and

(iii) optionally one or more pharmaceutically acceptable excipients;

for the treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection, wherein said composition is for once or twice a day administration and is preferably in the form of a kit.

DETAILED DESCRIPTION OF THE INVENTION

Many forms of dispensing containers and other holders for pharmaceutical compositions have been proposed and commercially introduced, especially those in the dosage form of tablets, pills, and capsules, are generally dispensed in vials, bottles, or blister packs. One deficiency with such packaging is that the user is responsible for maintaining an independent record by human memory or other means as to whether or not the proper dosage has actually been administered. This deficiency is particularly problematic for users who suffer from impaired memory performance or who may be taking multiple medications. Some patients may find it difficult to continuously maintain such records, especially over the long term, such that some dosages are missed and thus leading to reduced patient compliance.

The lack of patient compliance with adhering to a drug administration program is a problem. Aside from the fact that the patient may not be receiving the intended therapeutic benefit from noncompliance, there are potentially more serious issues that can arise from noncompliance. Studies documenting patient noncompliance have shown rates ranging from about 15% to about 93%, depending upon the population studied and the medical regimen involved ["Overview of Patient Compliance with Medication Dosing: A Literature Review", Clinical Therapeutics, Vol. 6, No. 5, pp. 592-599 (1984)].
Evidently, there is a continuing unmet need to make patient compliance in the administration of pharmaceutical compositions easier. Many forms of dispensing kits, containers, and other holders for pharmaceutical compositions have been proposed and commercially introduced. Many of these containers are intended for dispensing dosages of the pharmaceutical product on a daily basis.

Even though such once-daily therapies represent a significant advantage, it would be highly desirable to foster the ease of administration and help ensure patient compliance with such once daily therapies in the form of a kit.

Therefore, there is a need to develop and formulate a suitable pharmaceutical antiretroviral composition, in the form of a once or twice a day composition, comprising at least one reverse transcriptase inhibitor or at least one integrase inhibitor and at least one protease inhibitor which would not only be convenient for patient administration, but would also maintain or improve patient adherence to the therapy.

In general, the therapy for the treatment of HIV infection comprises a combination of actives such as nucleoside reverse transcriptase inhibitors (NRTIs), nucleotide reverse transcriptase inhibitors (NtRTIs), non-nucleoside reverse transcriptase inhibitors (NNRTIs), integrase inhibitors and protease inhibitors (PIs). The dose regimen of these drugs is such that the patient needs to administer several drugs throughout the day and at different time intervals. Further, this dosage regimen has to be followed throughout the patient's lifetime. This long-term therapy may generally cause great inconvenience to the patient. Hence, there is a precise need that the patient is provided with the means that

allows the patient to eliminate the inconvenience caused to him, such as remembering the administration of the medication, as well as the time at which it is to be administered.

Thus, the present invention provides a kit comprising a combination of NRTIs, NtRTIs, integrase inhibitors and PIs that provides the patient with his daily regimen of drugs in a single package. This further facilitates the patient in getting the drug regimen of the entire day in a single package, which also enables the patient to avoid carrying of numerous medications, and also confirms if the same are administered. It will be appreciated that the kit of the present invention may comprise a single dosage form in which a plurality of antiretroviral agents have been co-formulated, or a plurality of dosage forms containing one or more antiretroviral agents.

In an aspect, the present invention thus provides a pharmaceutical antiretroviral composition comprising at least one reverse transcriptase inhibitor or at least one integrase inhibitor and at least one protease inhibitor as a combined preparation in a kit form, for simultaneous or separate use in the treatment of an HIV infection.

It will be appreciated from the above that the respective therapeutic agents of the combined preparation can be administered simultaneously, either in the same or different pharmaceutical composition or dosage form, or separately. If there is separate administration, it will also be appreciated that the subsequently administered therapeutic agents should be administered to a patient within a time-scale so as to achieve, or more particularly optimize, a synergistic therapeutic effect of such a combined preparation.

Preferred protease inhibitors (PIs) that may be employed in a pharmaceutical antiretroviral composition of the present invention are darunavir and ritonavir.

Preferred nucleoside reverse transcriptase inhibitors (NRTIs) that may be employed in a pharmaceutical antiretroviral composition of the present invention include lamivudine, emtricitabine and/or zidovudine. In a preferred aspect, the NRTI may be lamivudine and/or zidovudine. In an alternative preferred aspect the NRTI may be lamivudine and/or emtricitabine.

A preferred nucleotide reverse transcriptase inhibitor (NtRTI) that may be employed in a pharmaceutical antiretroviral composition of the present invention is tenofovir.

A preferred integrase inhibitor that may be employed in a pharmaceutical antiretroviral composition of the present invention is raltegravir or dolutegravir.

A preferred reverse transcriptase inhibitor that may be employed in any pharmaceutical antiretroviral composition of the present invention is lamivudine.

A preferred reverse transcriptase inhibitor that may be employed in any pharmaceutical antiretroviral composition of the present invention is zidovudine.

A preferred reverse transcriptase inhibitor that may be employed in any pharmaceutical antiretroviral composition of the present invention is emtricitabine.

A preferred reverse transcriptase inhibitor that may be employed in any pharmaceutical antiretroviral composition of the present invention is tenofovir.

A preferred reverse transcriptase inhibitor that may be employed in any pharmaceutical antiretroviral composition of the present invention is abacavir.

A preferred integrase inhibitor that may be employed in any pharmaceutical antiretroviral composition of the present invention is raltegravir.

A preferred integrase inhibitor that may be employed in any pharmaceutical antiretroviral composition of the present invention is dolutegravir.

A preferred protease inhibitor that may be employed in any pharmaceutical antiretroviral composition of the present invention is ritonavir.

In an aspect, the present invention may provide a pharmaceutical antiretroviral composition comprising lamivudine, zidovudine, darunavir and ritonavir. Preferably, said composition is formulated for, or suitable for, once or twice a day administration. Said composition may be in the form of a kit.

In an aspect, the present invention may provide a pharmaceutical antiretroviral composition comprising lamivudine, tenofovir, darunavir and ritonavir. Preferably, said composition is formulated for, or suitable for, once or twice a day administration. Said composition may be in the form of a kit.

In an aspect, the present invention may provide a pharmaceutical antiretroviral composition comprising tenofovir, emtricitabine, darunavir and ritonavir. Preferably, said composition is formulated for, or is suitable for, once or twice a day administration. Said composition may be in the form of a kit.

In another aspect, the present invention may provide a pharmaceutical antiretroviral composition comprising raltegravir or dolutegravir, darunavir and ritonavir. Preferably, said composition is formulated for, or is suitable for once or twice a day administration. Said composition may be in the form of a kit.

In another aspect, the pharmaceutical antiretroviral composition in accordance with the present invention may be provided for use in the treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection, in a patient in need thereof.

In the context of the present invention, any references herein to an antiretroviral agent, for example, “zidovudine”; “didanosine”; “stavudine”; “lamivudine”; “abacavir”; “adefovir”; “lobucavir”; “entecavir”; “apricitabine”; “emtricitabine”; “zalcitabine”; “dexelvucitabine”; “alovudine”; “amdoxovir”; “elvucitabine”; “tenofovir”; “festinavir”; “racivir”; “lersivirine”; “rilpivirine”; “etravirine”; “SP1093V”, “stampidine”, “raltegravir”, “elvitegravir”, “dolutegravir”, “saquinavir”; “ritonavir”; “nelfinavir”; “amprenavir”; “lopinavir”, “indinavir”; “nelfinavir”; “atazanavir”; “lasinavir”;

"palinavir"; "fosamprenavir"; "darunavir"; "tiprinavir" includes by definition not only "zidovudine"; "didanosine"; "stavudine"; "lamivudine"; "abacavir"; "adefovir"; "lobucavir"; "entecavir"; "apricitabine"; "emtricitabine"; "zalcitabine"; "dexelvucitabine"; "alovudine"; "amdoxovir"; "elvucitabine"; "tenofovir"; "festinavir"; "racivir"; "lersivirine"; "rilpivirine"; "etravirine"; "SP1093V", "stampidine", "raltegravir", "elvitegravir", "dolutegravir". "saquinavir"; "ritonavir"; "nelfinavir"; "amprenavir"; "lopinavir", "indinavir"; "nelfinavir"; "atazanavir"; "lasinavir"; "palinavir"; "fosamprenavir"; "darunavir"; "tiprinavir" *per se*, but also its pharmaceutically acceptable derivatives thereof. Suitable derivatives include pharmaceutically acceptable salts, pharmaceutically acceptable solvates, pharmaceutically acceptable hydrates, pharmaceutically acceptable anhydrates, pharmaceutically acceptable enantiomers, pharmaceutically acceptable esters, pharmaceutically acceptable isomers, pharmaceutically acceptable polymorphs, pharmaceutically acceptable prodrugs, pharmaceutically acceptable tautomers, pharmaceutically acceptable complexes, and the like.

For instance, "tenofovir" as used herein by definition includes not only the phosphonic acid form, but also its prodrug form tenofovir disoproxil fumarate, tenofovir alafenamide fumarate, tenofovir alafenamide hemifumarate. The term "darunavir" as used herein by edition includes not darunavir *per se*, but also its solvates, such as darunavir ethanolate, and its hydrates, such as darunavir hydrate.

Tenofovir disoproxil fumarate is also known as PMPA. Tenofovir DF is a fumaric acid salt of bis-isopropoxycarbonyloxymethyl ester derivative of tenofovir. Tenofovir disoproxil fumarate is 9-[(R)-2-[[bis[[(isopropoxycarbonyl) oxy] methoxy] phosphinyl] methoxy] propyl] adenine fumarate (1:1). Tenofovir disoproxil fumarate requires initial diester hydrolysis for conversion to tenofovir and subsequent phosphorylations by cellular enzymes to form tenofovir diphosphate. Tenofovir diphosphate inhibits the activity of HTV reverse transcriptase by competing with the natural substrate deoxyadenosine 5 '-triphosphate and, after incorporation into DNA, by DNA chain termination. Tenofovir diphosphate is a weak inhibitor of mammalian DNA polymerases alpha & beta and of mitochondrial DNA polymerase.

Tenofovir disoproxil fumarate is an analog of adefovir and is classified as a nucleotide reverse transcriptase inhibitor (NtRTI). Tenofovir DF is a competitive inhibitor of other naturally occurring nucleotides, and its ultimate biological activity is viral DNA chain termination. Tenofovir DF is a novel nucleotide analog with antiviral activity against both HIV and HBV. The mechanism of tenofovir DF is similar to that of nucleoside analogs, which interferes with reverse transcriptase and prevents translation of viral genetic material into viral DNA. Unlike the nucleoside analogs, the nucleotide reverse transcriptase inhibitors are chemically pre-activated with the presence of phosphate group. Since the phosphorylation step is not necessary, nucleotide analogs can incorporate into viral DNA chain more rapidly than nucleoside analogs. More importantly, this will bypass a viral mechanism of nucleoside resistance. A preferred dosage of tenofovir disoproxil for use in a pharmaceutical antiretroviral composition of the present invention is in an amount from about 75mg to 300 mg.

Emtricitabine, is chemically known as 4-amino-5-fluoro-l- [2- (hydroxymethyl) - l, 3-oxathiolan-5-yl] - pyrimidin-2-one, belongs to a category of nucleoside reverse transcriptase inhibitor (NRTI) which is used to treat infection by HIV-I. Specifically, emtricitabine inhibits HBV DNA polymerase and HIV-1 reverse transcriptase (RT) both *in vivo* and *in vitro*. Emtricitabine is anabolized to its triphosphate form which is the active moiety that inhibits the polymerase. A preferred dosage of emtricitabine for use in a pharmaceutical antiretroviral composition of the present invention is in an amount form 9 mg to 300 mg.

Zidovudine, chemically known as 3'-azido-3'deoxythymidine, is a pyrimidine nucleoside analogue, which is well established as an important and useful chemotherapeutic agent for the treatment and / or prophylaxis of HIV infections including related clinical conditions such as AIDS, AIDS-related complex (ARC), AIDS dementia complex (ADC) and also for the treatment of patients who have an asymptomatic HIV infection and who are anti-HIV antibody positive. In addition to lamivudine's proven antiviral activity against HIV as referred to above, lamivudine also exhibits antiviral activity against other viruses such as HBV. A preferred dosage of zidovudine for use in a pharmaceutical

antiretroviral composition of the present invention is in an amount from about 60 mg to about 600 mg.

Lamivudine (also known as 3TC) is a synthetic nucleoside analogue, chemically known as (2R, cis)-4-amino-l-(2-hydroxymethyl-l, 3-oxathiolan-5-yl)-(lH)-pyrimidin-2- one. Intracellularly, lamivudine is phosphorylated to its active 5'-triphosphate metabolite, lamivudine triphosphate (L-TP). The principal mode of action of L-TP is the inhibition of HIV-I reverse transcriptase (RT) via DNA chain termination after incorporation of the nucleoside analogue into viral DNA. L-TP is a weak inhibitor of mammalian DNA polymerases (alpha) and (beta), and mitochondrial DNA polymerase (gamma). Lamivudine has also been referred to as (-)-l-[(2R, 5S) 2-(Hydroxymethyl)-1,3-oxathiolan-5-yl] cytosine, (Hydroxymethyl)-l,3-oxathiolan-5-yl] cytosine and it has proven antiviral activity against human immunodeficiency virus (HIV) and other viruses such as hepatitis B. A preferred dosage of lamivudine for use in a pharmaceutical antiretroviral composition of the present invention is in an amount from about 30 mg to about 300 mg.

Darunavir (TMC-1 14, UIC-94017) is chemically known as [(3R,3aS,6aR)-2,3,3a,4,5,6a-Hexahydrofuro[5,4-b]furan-3-yl]N-[(2S,3R)-4-[(4-aminophenyl)sulfonyl-(2-methylpropyl) amino]-3-hydroxy-l-phenylbutan-2 yl]carbamate A preferred dosage of darunavir for use in a pharmaceutical antiretroviral composition of the present invention is in an amount from about 350 mg to about 1200 mg.

Ritonavir is chemically designated as 10-Hydroxy-2-methyl-5-(1-methylethyl)-1- [2-(1-methylethyl)-4thiazolyl]- 3,6-dioxo-8,11-bis(phenylmethyl)-2,4,7,12- tetraazatridecan-13-oic acid, 5-thiazolylmethyl ester, [5S-(5R*,8R*,10R*,11R*)]. Ritonavir is a peptidomimetic inhibitor of the HIV-1 protease. Inhibition of HIV protease renders the enzyme incapable of processing the gag-pol polyprotein precursor which leads to production of noninfectious immature HIV particles. A preferred dosage of ritonavir for use in a pharmaceutical antiretroviral composition of the present invention is in an amount from about 33 mg to about 100 mg.

Abacavir is a synthetic carbocyclic nucleoside analog and reverse transcriptase inhibitor which is used typically in combination with other agents in the therapy of the human immunodeficiency virus (HIV) infection. In vivo, the activated triphosphate metabolite of abacavir is incorporated into the viral DNA instead of the natural substrate deoxyguanosine, thereby inhibiting human immunodeficiency virus (HIV) reverse transcriptase (RT) and the replication of the viral DNA and infectious viral particles. This agent decreases HIV viral loads, retards or prevents the damage to the immune system, and reduces the risk of developing AIDS. A preferred dosage of abacavir is from about 60 mg to about 600 mg.

Raltegravir is a human immunodeficiency virus integrase strand transfer inhibitor. Raltegravir inhibits the catalytic activity of HIV-1 integrase, an HIV-1 encoded enzyme that is required for viral replication. Inhibition of integrase prevents the covalent insertion, or integration, of unintegrated linear HIV-1 DNA into the host cell genome preventing the formation of the HIV-1 provirus. The provirus is required to direct the production of progeny virus, so inhibiting integration prevents propagation of the viral infection. Raltegravir did not significantly inhibit human phosphoryltransferases including DNA polymerases α, β, and γ. A preferred dosage of raltegravir is from about 40 mg to about 1200 mg.

Dolutegravir is an orally active integrase inhibitor, and has been approved for the treatment of HIV infections. It is a HIV-1 integrase strand transfer inhibitor which inhibits HIV integrase by binding to the integrase active site and blocking the strand transfer step of retroviral deoxyribonucleic acid integration which is essential for the HIV replication cycle. A preferred dosage of dolutegravir is from about 1 mg to about 100 mg.

In another aspect, the present invention provide a pharmaceutical antiretroviral composition comprising lamivudine, zidovudine, darunavir and ritonavir in a kit form. Preferably, said composition is for once or twice a day administration.

The pharmaceutical antiretroviral composition in kit form may comprise a separate unit dosage form of lamivudine, a separate unit dosage form of zidovudine, a separate unit dosage form of darunavir and a separate unit dosage form of ritonavir.

In another aspect, the present invention may provide a pharmaceutical antiretroviral composition comprising lamivudine, tenofovir, darunavir and ritonavir in a kit form. Preferably, said composition is for once or twice a day administration.

The pharmaceutical antiretroviral composition in kit form may comprise a separate unit dosage form of lamivudine, a separate unit dosage form of tenofovir, a separate unit dosage form of darunavir and a separate unit dosage form of ritonavir.

In another aspect, the present invention may provide a pharmaceutical antiretroviral composition comprising emtricitabine, tenofovir, darunavir and ritonavir in a kit form. Preferably, said composition is for once or twice a day administration.

The pharmaceutical antiretroviral composition in kit form may comprise a separate unit dosage form of emtricitabine, a separate unit dosage form of tenofovir, a separate unit dosage form of darunavir and a separate unit dosage form of ritonavir.

In another aspect, the present invention may provide a pharmaceutical antiretroviral composition comprising abacavir, lamivudine, darunavir and ritonavir in a kit form. Preferably, said composition is for once or twice a day administration.

The pharmaceutical antiretroviral composition in kit form may comprise a separate unit dosage form of abacavir, a separate unit dosage form of lamivudine, a separate unit dosage form of darunavir and a separate unit dosage form of ritonavir.

In another aspect, the present invention may provide a pharmaceutical antiretroviral composition comprising raltegravir or dolutegravir, darunavir and ritonavir in a kit form. Preferably, said composition is for once or twice a day administration.

The pharmaceutical antiretroviral composition in kit form may comprise a separate unit dosage form of raltegravir a separate unit dosage form of darunavir and a separate unit dosage form of ritonavir.

The pharmaceutical antiretroviral composition in kit form may comprise a separate unit dosage form of dolutegravir a separate unit dosage form of darunavir and a separate unit dosage form of ritonavir.

In any of the above compositions the pharmaceutical antiretroviral composition of the composition of the present invention may comprise darunavir and ritonavir in a single unit dosage form.

In any of the above compositions, where appropriate, the pharmaceutical antiretroviral composition of the present invention may comprise lamivudine and tenofovir in a single unit dosage form.

In any of the above compositions, where appropriate, the pharmaceutical antiretroviral composition of the present invention may comprise lamivudine and zidovudine in a single unit dosage form.

In any of the above compositions, where appropriate, the pharmaceutical antiretroviral composition of the present invention comprises tenofovir and emtricitabine in a single unit dosage form.

In any of the above compositions, where appropriate, the pharmaceutical antiretroviral composition of the present invention comprises abacavir and lamivudine in a single unit dosage form.

In any of the above compositions, where appropriate, the pharmaceutical antiretroviral composition of the present invention comprises raltegravir in a single unit dosage form.

In any of the above compositions, where appropriate, the pharmaceutical antiretroviral composition of the present invention comprises dolutegravir in a single unit dosage form.

In any of the above compositions, where appropriate, the pharmaceutical antiretroviral composition of the present invention comprises darunavir and ritonavir, raltegravir dolutegravir, lamivudine and tenofovir, lamivudine and zidovudine, tenofovir and emtricitabine, abacavir and lamivudine are each provided in a separate single unit dosage forms.

In any of the above compositions, where appropriate, any two of the at least one reverse transcriptase inhibitor, the at least one integrase inhibitor and the at least one protease inhibitor may be provided in a single unit dosage form.

In any of the above compositions, where appropriate, any three of the at least one reverse transcriptase inhibitor, the at least one integrase inhibitor and the at least one protease inhibitor are provided in a single unit dosage form.

In any of the above compositions, where appropriate, all of the at least one reverse transcriptase inhibitor, the at least one integrase inhibitor and the at least one protease inhibitor are provided in a single unit dosage form.

When formulated as a single unit dose, the antiretroviral agents may initially be co-formulated with one or more pharmaceutically acceptable excipients to provide a single uniform composition or they may initially be formulated as individual compositions. When formulated individually, the individual compositions may subsequently be co-formulated as a single unit dosage form, where the unit dosage form may comprise two or more layers, each layer comprising a composition of at least one antiretroviral agent (i.e. the individually formulated compositions). Preferably, the single unit dosage form is suitable for once or twice daily administration.

Suitably, the pharmaceutical antiretroviral composition according to the present invention are presented in solid dosage form, conveniently in unit dosage form, and include dosage

form suitable for oral and buccal administration. However, other dosage forms, such as liquid dosage forms and the like, may be envisaged under the ambit of the present invention.

Unit dosage forms, according to the present invention, are preferably in the form of a tablet (disintegrating tablet, dissolving tablet, dispersible tablets, mouth dissolving tablets, tablets for oral suspension immediate release tablets, extended release tablet, immediate and extended release tablets, matrix tablets), mini-tablet, granules, sprinkles (filled with powders, powders for reconstitution; beads; pellets; mini-tablets; film coated tablets; film coated tablets MUPS (multiple unit pellet system); orally disintegrating MUPS; pills; micro-pellets; small tablet units; MUPS; disintegrating tablets; dispersible tablets; granules; effervescent granules; microspheres) or capsules filled with (powders, powders for reconstitution; beads; pellets; mini-tablets; film coated tablets; film coated tablets MUPS; orally disintegrating MUPS; pills; micro-pellets; small tablet units; MUPS; disintegrating tablets; dispersible tablets; granules; effervescent granules; microspheres), liquids such as suspension, emulsions, solutions, syrups, elixirs but other dosage forms may also fall within the scope of this invention.

Preferably, the pharmaceutical antiretroviral composition, according to the present invention, may be administered orally through known solid unit dosage forms including capsule and sachets or packets (filled with powders, powders for reconstitution; beads; pellets; mini-tablets; film coated tablets; film coated tablets MUPS; orally disintegrating MUPS; pills; micro-pellets; small tablet units; MUPS; disintegrating tablets; dispersible tablets; granules; effervescent granules; microspheres). The capsules may be hard gelatin capsules. Sachets or packets may be filled with powders, powders for reconstitution; beads; pellets; mini-tablets; film coated tablets; film coated tablets MUPS; orally disintegrating MUPS; pills; micro-pellets; small tablet units; MUPS; disintegrating tablets; dispersible tablets; granules; effervescent granules; microsphere that are suitable for direct administration. Preferably, the present invention may be administered as mini-tablets or granules filled in hard gelatin capsules, sachets or packets.

Preferably, the mini-tablets or granules filled in such hard gelatin capsules, sachets or packets are directly administered or by sprinkling the mini-tablet or granules on regular meals. Alternatively, the mini-tablets or granules filled in hard gelatin capsules, sachets or packets may be administered with liquid or semi-solid beverages such as but not limited to, fruit juices, water, milk, baby formulas, soft foods, apple sauce or yogurt and the like.

The mini-tablets or granules, according to the present invention, may also optionally be coated. Preferably, mini-tablets or granules, according to the present invention, may be film coated. More preferably, the mini-tablets or granules may be seal coated and then film coated and further filled in hard gelatin capsules, sachets or packets.

It is further well known in the art that a tablet formulation is the preferred solid dosage form due to its greater stability, less risk of chemical interaction between different medicaments, smaller bulk, accurate dosage, and ease of production.

Solid unit dosage forms, according to the present invention, are preferably in the form of tablets but other conventional dosages such as powders, pellets, capsules, sachets or packets may fall within the scope of this invention.

Kit compositions of the type disclosed herein have an advantage over other packaged dosage forms since the patient always has access to the set of instructions for administration contained in the kit. The inclusion of a set of instructions for administration has been shown to improve patient compliance.

It will be understood that the administration of the pharmaceutical antiretroviral composition of the invention by means of a kit, with a set of instructions for administration diverting the patient to the correct use of the invention is a desirable additional feature of this invention.

According to a preferred aspect, the pharmaceutical antiretroviral composition may be administered simultaneously, separately or sequentially in a single unit dosage form

wherein the drugs and excipients are present in one or more single layer tablets (such as a tablet or mini tablet in a capsule or sprinkle).

According to another preferred aspect, the pharmaceutical antiretroviral composition may be in the form of one or more bilayered or multilayered unit dosage forms.

In a preferred aspect, the pharmaceutical antiretroviral composition in a kit form may comprise a separate unit dosage form comprising lamivudine and zidovudine and a further separate unit dosage form comprising darunavir and ritonavir.

In a further preferred aspect, the pharmaceutical antiretroviral composition in a kit form may comprise a separate unit dosage form comprising lamivudine and tenofovir and a further separate unit dosage form comprising darunavir and ritonavir.

In another preferred aspect, the pharmaceutical antiretroviral composition in a kit form may comprise a separate unit dosage form comprising emtricitabine and tenofovir and a further separate unit dosage form comprising darunavir and ritonavir.
In another preferred aspect, the pharmaceutical antiretroviral composition in a kit form may comprise a separate unit dosage form comprising abacavir and lamivudine and a further separate unit dosage form comprising darunavir and ritonavir.

In another preferred aspect, the pharmaceutical antiretroviral composition in a kit form may comprise a separate unit dosage form comprising raltegravir and a further separate unit dosage form comprising darunavir and ritonavir.

In another preferred aspect, the pharmaceutical antiretroviral composition in a kit form may comprise a separate unit dosage form comprising dolutegravir and a further separate unit dosage form comprising darunavir and ritonavir.

Suitable excipients may be used for formulating the various dosage forms according to the present invention.

According to the present invention, pharmaceutically acceptable diluents or fillers for use in the pharmaceutical antiretroviral composition of the present invention may comprise one or more, but not limited to lactose (for example, spray-dried lactose, α-lactose, β-lactose) lactitol, saccharose, sorbitol, mannitol, dextrates, dextrins, dextrose, maltodextrin, croscarmellose sodium, microcrystalline cellulose, silicified microcrystalline cellulose, hydroxypropylcellulose, L-hydroxypropylcellulose (low substituted), hydroxypropyl methylcellulose (HPMC), methylcellulose polymers, hydroxyethylcellulose, sodium carboxymethylcellulose, carboxymethylene, carboxymethyl hydroxyethylcellulose and other cellulose derivatives, starches or modified starches (including potato starch, corn starch, maize starch and rice starch) and combinations thereof.

The amount of diluents or fillers that may be present in the pharmaceutical antiretroviral composition can range from about 20% to about 70%.

According to the present invention, glidants, anti-adherents and lubricants may also be incorporated in the pharmaceutical antiretroviral composition of the present invention, which may comprise one or more, but not limited to stearic acid and pharmaceutically acceptable salts or esters thereof (for example, magnesium stearate, calcium stearate, sodium stearyl fumarate or other metallic stearate), talc, waxes (for example, microcrystalline waxes), glycerides, light mineral oil, PEG, silica acid or a derivative or salt thereof (for example, silicates, silicon dioxide, colloidal silicon dioxide and polymers thereof, magnesium aluminosilicate and/ or magnesium alumino metasilicate), sucrose ester of fatty acids, hydrogenated vegetable oils (for example, hydrogenated castor oil) and combinations thereof.

The amount of glidants, anti-adherents and lubricants that may be present in the pharmaceutical antiretroviral composition can range from about 0.3% to about 2%.

According to the present invention, suitable binders may also be present in the in the pharmaceutical antiretroviral composition of the present invention, which may comprise one or more, but not limited to polyvinyl pyrrolidone (also known as povidone),

polyethylene glycol(s), acacia, alginic acid, agar, calcium carragenan, cellulose derivatives such as ethyl cellulose, methyl cellulose, hydroxypropyl cellulose, hydroxypropyl methyl cellulose, sodium carboxymethylcellulose, dextrin, gelatin, gum arabic, guar gum, tragacanth, sodium alginate, anhydrous dibasic calcium phosphate and combinations thereof or any other suitable binder.

The amount of binders that may be present in the pharmaceutical antiretroviral composition can range from about 1% to about 7%.

According to the present invention, suitable disintegrants may also be present in the pharmaceutical antiretroviral composition of the present invention, which may comprise one or more, but not limited to hydroxylpropyl cellulose (HPC), low density HPC, carboxymethylcellulose (CMC), crospovidone, sodium CMC, calcium CMC, croscarmellose sodium; starches exemplified under examples of fillers and carboxymethyl starch, hydroxylpropyl starch, modified starch, pregelatinized starch, crystalline cellulose, sodium starch glycolate; alginic acid or a salt thereof, such as sodium alginate or their equivalents and combinations thereof.

The amount of disintegrants that may be present in the pharmaceutical antiretroviral composition can range from about 0.7% to about 5%.

The present invention also provides a hot melt extruded pharmaceutical antiretroviral composition comprising antiretroviral drug/drugs and at least one water soluble and/or water swellable and/or water insoluble polymer or combination thereof and one or more optional pharmaceutically acceptable excipients.

Water soluble polymers which may be used in the pharmaceutical antiretroviral composition of the present invention, include, but are not limited to, homopolymers and co-polymers of N-vinyl lactams, especially homopolymers and co-polymers of N- vinyl pyrrolidone e.g. polyvinylpyrrolidone (PVP), co-polymers of PVP and vinyl acetate, co-polymers of N-vinyl pyrrolidone and vinyl acetate (Copovidone) or vinyl propionate, dextrins such as grades of maltodextrin, cellulose esters and cellulose ethers, high

molecular polyalkylene oxides such as polyethylene oxide and polypropylene oxide and co-polymers of ethylene oxide, propylene oxide and combinations thereof.

The amount of water soluble polymers that may be present in the pharmaceutical antiretroviral composition can range from about 10% to about 50%.

Water insoluble polymers which may be used in the pharmaceutical antiretroviral composition of the present invention, include, but are not limited to, acrylic copolymers e.g. Eudragit E100 or Eudragit EPO; Eudragit L30D-55, Eudragit FS30D, Eudragit RL30D, Eudragit RS30D, Eudragit NE30D, Acryl-Eze (Colorcon Co.); polyvinylacetate, for example, Kollicoat SR 30D (BASF Co.); cellulose derivatives such as ethylcellulose, cellulose acetate e.g. Surelease (Colorcon Co.), Aquacoat ECD and Aquacoat CPD (FMC Co.) and combinations thereof.

The amount of water insoluble polymers that may be present in the pharmaceutical antiretroviral composition can range from about 3% to about 15%.

Water swellable polymers that may be used, according to the present invention include, but are not limited to polyethylene oxide; poly (hydroxy alkyl methacrylate); poly (vinyl) alcohol, having a low acetal residue, which is cross-linked with glyoxal, formaldehyde or glutaraldehyde and having a degree of polymerization of from 200 to 30,000; a mixture of methyl cellulose, cross- linked agar and carboxymethyl cellulose; Carbopol® carbomer which is an acidic carboxy polymer; Cyanamer® polyacrylamides; cross-linked water swellable indene- maleic anhydride polymers; Goodrich® polyacrylic acid; starch graft copolymers; Aqua Keeps® acrylate polymer polysaccharides composed of condensed glucose units such as diester cross-linked polyglucan, and the like; Amberlite® ion exchange resins; Explotab® sodium starch glycolate; Ac-Di-Sol® croscarmellose sodium or combinations thereof.

The amount of water swellable polymers that may be present in the pharmaceutical antiretroviral composition can range from about 1% to about 10%.

One or more optional pharmaceutically acceptable excipients may include plasticizer.

Plasticizers reduce the viscosity of the polymer melt and thereby allow for lower processing temperature and extruder torque during hot melt extrusion. They further decrease the glass transition temperature of the polymer.

Plasticizers which may be used in the pharmaceutical antiretroviral composition of the present invention, include, but are not limited to, polysorbates such as sorbitan monolaurate (Span 20), sorbitan monopalmitate, sorbitan monostearate, sorbitan monoisostearate; citrate ester type plasticizers like triethyl citrate, citrate phthalate; propylene glycol; glycerin; polyethylene glycol (low & high molecular weight); triacetin; dibutyl sebacate, tributyl sebacate; dibutyltartrate, dibutyl phthalate, glycerol palmitosterate and combinations thereof.

The amount of plasticizers that may be present in the pharmaceutical antiretroviral composition can range from about 0.4% to about 3%.

The pharmaceutical antiretroviral composition, according to the present invention, may be prepared through various techniques or processes known in the art which includes, but are not limited to direct compression, wet granulation, dry granulation, melt granulation, melt extrusion, spray drying, solution evaporation or combinations thereof.

It will be appreciated that the above mentioned techniques may be used either singly or in combination with other above mentioned techniques to provide unit dosage form according to the present invention in the form of single layered, bilayered or multilayered tablets, mini tablets or sprinkles.

Suitable processes may be used for formulating the various dosage forms according to the present invention.

In one aspect, the dosage form of the present invention may be prepared by hot melt extrusion. The process of hot melt extrusion is carried out in the conventional extruders as known to a person having a skill in the art.

Typically, the melt-extrusion process comprises the steps of preparing a homogeneous melt of one or more drugs, the polymer and the excipients, and cooling the melt until it solidifies.

Melting usually involves heating above the softening point of the polymer. The preparation of the melt can take place in a variety of ways. The mixing of the components can take place before, during or after the formation of the melt.

Usually, the melt temperature is in the range of about 50° C to about 200° C.

Suitable extruders include single screw extruders, intermeshing screw extruders or else multiscrew extruders, preferably twin screw extruders, which can be co - rotating or counter - rotating and, optionally, be equipped with kneading disks.

The extrudates can be in the form of beads, granulates, tube, strand or cylinder and this can be further processed into any desired shape.
In an alternative process, the present invention may further be allowed to form granules which may be compressed to form tablets, or the granules may be filled into capsules, sachets, pellets in capsules or in a similar dosage form.

This process involves heating the polymer(s) to soften it, without melting it, and mixing the active ingredient(s) with polymer(s), to form granules.

The process can be carried out in the same type of extrusion apparatus as the hot melt extrusion process, except that the product is not extruded through the extrusion nozzle of the apparatus.

The extrudates/granules so obtained according to the present invention may then be admixed with other suitable one or more pharmaceutically acceptable excipients.

According to a preferred aspect, the pharmaceutical antiretroviral composition of the present invention may processed by wet granulation of lamivudine and zidovudine wherein the diluent, the disintegrant along with the actives lamivudine and zidovudine are treated with the binder solution to form granules. Granules are lubricated and compressed to provide a single layered tablet or compressed separately to provide a bilayered tablet which may optionally be coated. Alternatively, the granules so obtained are filled into hard gelatin capsules or sachets or by compressing the granules to form mini-tablets which may also be filled into capsules or sachets and can be sprinkled onto food.

According to yet another preferred aspect, the pharmaceutical antiretroviral composition of the present invention may be processed by mixing darunavir with intragranular excipients such as diluents, disintegrants to form granules. Ritonavir, polymers (i.e. either water soluble and/or water swellable or/and water insoluble or mixture thereof), one or more plasticizer, one or more disintegrants, one or more lubricants and glidants are extruded through hot melt extrusion technique wherein extrudates are obtained which can be molded into granules. Granules are lubricated and compressed to provide a single layered tablet or compressed separately to provide a bilayered tablet which may optionally be coated. Alternatively, the granules so obtained are filled into hard gelatin capsules or sachets or by compressing the granules to form mini-tablets which may also be filled into capsules or sachets and can be sprinkled onto food.

Further, the granules comprising darunavir and ritonavir as obtained above may be further mixed, sieved, sifted and compressed into a single tablet. Alternatively, the tablet may be seal coated and finally film coated or the tablet may be film coated and then seal coated.

Alternatively, the granules comprising darunavir and ritonavir as obtained above may be individually compressed into two tablets and finally compacted and compressed into a bilayer tablet. Alternatively, the tablet may be seal coated and finally film coated or the tablet may be film coated and then seal coated.

According to a further preferred aspect, the pharmaceutical antiretroviral composition of the present invention may be processed by wet granulation of tenofovir and emtricitabine wherein the diluent, the disintegrant along with the actives tenofovir and emtricitabine are sifted and dried. Then, binder solution is prepared by first dissolving the binder in purified water. Granulation is carried out by spraying of the binder solution to the above dry mixture of the ingredients, after which the formed granules are dried, sifted through the specified mesh. After unloading, the granules of tenofovir, emtricitabine were lubricated. The granules as obtained above are compressed to provide a single layered tablet or compressed separately to provide a bilayered tablet. The tablets thus obtained via the process are then sprayed with a coating suspension.

According to another preferred aspect, the pharmaceutical antiretroviral composition of the present invention may be processed by wet granulation of tenofovir and lamivudine wherein the diluent, the disintegrant along with the actives tenofovir and lamivudine are sifted and dried. Then, binder solution is prepared by first dissolving the binder in purified water. Granulation is carried out by spraying of the binder solution to the above dry mixture of the ingredients, after which the formed granules are dried, sifted through the specified mesh. After unloading, the granules of tenofovir, lamivudine were lubricated. The granules as obtained above are compressed to provide a single layered tablet or compressed separately to provide a bilayered tablet. The tablets thus obtained via the process are then sprayed with a coating suspension.

Alternatively, after compression into tablets, they can be further seal coated and then sprayed with a coating suspension.

Additional excipients such as film forming polymers, solvents, plasticizers, anti-adherents, opacifiers, colorants, pigments, antifoaming agents, and polishing agents can be used in coatings.

Suitable seal forming material may comprise: hydroxypropylmethylcellulose (optionally HPMC 6 CPS, or HPMC 6 CPS to HPMC 15CPS grade); hydroxypropylcellulose;

polyvinylpyrrolidone; methylcellulose; carboxymethylcellulose; hypromellose; acacia; gelatin; or any combination thereof, to increase adherence and coherence of the seal coat. Preferably the seal coat comprises hydroxypropylmethylcellulose.

The amount of seal forming materials that may be present in the pharmaceutical antiretroviral composition can range from about 0.2% to about 10%.

The HPMC component of the seal coating, if present, may be mixed with a solvent, wherein said solvent may comprise: acetone; methylene chloride; isopropyl alcohol; or any combination thereof. The seal coating may also comprise talc.

Suitable film-forming agents include, but are not limited to, cellulose derivatives, such as, soluble alkyl- or hydroalkyl-cellulose derivatives such as methylcelluloses, hydroxymethyl celluloses, hydroxyethyl celluloses, hydroxypropyl celluloses, hydroxymethylethyl celluloses, hydroxypropyl methylcelluloses, sodium carboxymethyl celluloses, insoluble cellulose derivatives such as ethylcelluloses and the like, dextrins, starches and starch derivatives, polymers based on carbohydrates and derivatives thereof, natural gums such as gum Arabic, xanthans, alginates, polyacrylic acids, polyvinyl alcohols, polyvinyl acetates, polyvinylpyrrolidones, polymethacrylates and derivatives thereof, chitosan and derivatives thereof, shellac and derivatives thereof, waxes, fat substances and any combinations or combinations thereof.

The amount of film forming agents that may be present in the pharmaceutical antiretroviral composition can range from about 0.2% to about 10%.

Suitable enteric coating materials, include, but are not limited to, cellulosic polymers like cellulose acetate phthalates, cellulose acetate trimellitates, hydroxypropyl methylcellulose phthalates, polyvinyl acetate phthalates, methacrylic acid polymers, any copolymer thereof, any mixture thereof, or combination thereof.

The amount of enteric coating materials that may be present in the pharmaceutical antiretroviral composition can range from about 1% to about 15%.

Some of the excipients are used as adjuvant to the coating process, including excipients such as plasticizers, opacifiers, antiadhesives, polishing agents, and the like.

Suitable plasticizers include, but are not limited to, stearic acid, castor oil, diacetylated monoglycerides, dibutyl sebacate, diethyl phthalate, glycerin, polyethylene glycols, propylene glycols, triacetin, triethyl citrate, or combinations thereof.

Suitable opacifiers include, but are not limited to, titanium dioxide.

Suitable anti-adhesives include, but are not limited to, talc.

Suitable polishing agents include, but are not limited to, polyethylene glycols of various molecular weights or combinations thereof, talc, surfactants (glycerol monostearate and poloxamers), fatty alcohols (stearyl alcohol, cetyl alcohol, lauryl alcohol and myristyl alcohol) and waxes (carnauba wax, candelilla wax and white wax), or combinations thereof.

The amount of polishing agents that may be present in the pharmaceutical antiretroviral composition can range from about 0.2% to about 1%.

Suitable solvents used in the processes of preparing the pharmaceutical solid oral composition of the present invention, include, but are not limited to, water, methanol, ethanol, acidified ethanol, acetone, diacetone, polyols, polyethers, oils, esters, alkyl ketones, methylene chloride, isopropyl alcohol, butyl alcohol, methyl acetate, ethyl acetate, isopropyl acetate, castor oil, ethylene glycol monoethyl ether, diethylene glycol monobutyl ether, diethylene glycol monoethyl ether, dimethylsulphoxide, N,N-dimethylformamide, tetrahydrofuran, or combinations thereof.

Further, the inventors have surprisingly found that when, by a process comprising hot melt extrusion of one or more drugs with at least one or more water insoluble polymers, with at least one or more water soluble polymers, with at least one or more water swellable polymers or a combination of at least one or more water soluble polymers

and/or water swellable and/or water insoluble polymer, the resulting product acquires taste masking property wherein the ratio of drug: polymer is 1:1 to 1: 6.

It was surprisingly found that while carrying out the melt extrusion process an in-situ reaction occurred between the drug and polymer. This in-situ reaction led to ionic interaction between the drug and polymer eventually leading to taste masked product.

According to a preferred aspect, the present invention may be formulated for pediatric patients and from the point of view of pediatric patient acceptability suitable bulking agents may be incorporated, in the pharmaceutical antiretroviral composition comprising saccharides, including monosaccharides, disaccharides, polysaccharides and sugar alcohols but not limited to arabinose, lactose, dextrose, sucrose, fructose, maltose, mannitol, erythritol, sorbitol, xylitol, lactitol, powdered cellulose, microcrystalline cellulose, purified sugar and their derivatives and combination thereof.

Accordingly, the present invention may further incorporate suitable pharmaceutically acceptable flavourants, such as but not limited to citric acid, tartaric acid, lactic acid, orange permaseal, strawberry cream flavour or other natural flavourants and sweeteners such as but not limited to aspartame or combination thereof.

Alternatively, the pharmaceutical antiretroviral composition according to the present invention may also comprise the actives in nano size form. Preferably, the active pharmaceutical ingredients have an average particle size less than about 2000 nm, preferably less than about 1000 nm.

Nanonization of hydrophobic or poorly water-soluble drugs generally involves the production of drug nanocrystals through either chemical precipitation (bottom-up technology) or disintegration (top-down technology). Different methods may be utilized to reduce the particle size of the hydrophobic or poorly water soluble drugs. [Huabing Chen et al., discusses the various methods to develop nano-formulations in “Nanonization strategies for poorly water-soluble drugs,” Drug Discovery Today, Volume 00, Number 00, March 2010].

Nano-sizing leads to increase in the exposure of surface area of particles leading to an increase in the rate of dissolution.

The nanoparticles of the present invention can be obtained by any of the process such as but not limited to milling, precipitation, homogenization, high pressure homogenization, spray-freeze drying, supercritical fluid technology, emulsion/solvent evaporation, PRINT, thermal condensation, ultrasonication and spray drying.

The present invention provides method of prevention, treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection, which method comprises administering a pharmaceutical antiretroviral composition of the type hereinbefore described.

In preferred aspects, the pharmaceutical antiretroviral composition of the present invention may comprise: (i) lamivudine, zidovudine, darunavir and ritonavir; (ii) lamivudine, tenofovir, darunavir and ritonavir; (iii) emtricitabine, tenofovir, darunavir and ritonavir (iv) lamivudine, abacavir, darunavir and ritonavir; (v) raltegravir, darunavir and ritonavir or (vi) dolutegravir, darunavir and ritonavir.

The present invention also provides use of the pharmaceutical antiretroviral composition of the type hereinbefore described for the treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection. In preferred aspects, the pharmaceutical antiretroviral composition of the present invention may comprise: (i) lamivudine, zidovudine, darunavir and ritonavir; (ii) lamivudine, tenofovir, darunavir and ritonavir; (iii) emtricitabine, tenofovir, darunavir and ritonavir; (iv) lamivudine, abacavir, darunavir and ritonavir; (v) raltegravir, darunavir and ritonavir or (vi) dolutegravir, darunavir and ritonavir.

The present invention further provides pharmaceutical antiretroviral composition of the type hereinbefore described for simultaneous, separate or sequential use in the prevention, treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune

deficiency syndrome or an HIV infection. In, preferred aspects, the pharmaceutical antiretroviral composition may comprise: (i) lamivudine, zidovudine, darunavir and ritonavir; (ii) lamivudine, tenofovir, darunavir and ritonavir; (iii) emtricitabine, tenofovir, darunavir and ritonavir; (iv) lamivudine, abacavir, darunavir and ritonavir; (v) raltegravir, darunavir and ritonavir or (vi) dolutegravir, darunavir and ritonavir.

The following examples are for the purpose of illustration of the invention only and are not intended in any way to limit the scope of the present invention.

EXAMPLE 1

Emtricitabine & Tenofovir Disoproxil Bilayered Tablets:

Sr. No	Name of Ingredients	Quantity/tab (mg)
Tenofovir Layer		
I	**Dry Mix**	
1.	Tenofovir Disoproxil Fumarate	300.00
2.	Lactose Monohydrate	159.00
3.	Croscarmellose sodium	20.00
4.	Corn Starch	30.00
II	**Binder Preparation**	
5.	Corn Starch	15.00
6.	Polysorbate	3.00
7.	Purified Water	q. s.
III.	**Lubrication**	
8.	Microcrystalline Cellulose	100.00
9.	Croscarmellose sodium	20.00
10.	Magnesium Stearate	12.50
	Total	**659.50**

Sr. No	Name of Ingredients	Quantity/tab (mg)
Emtricitabine Layer		
I	**Dry Mix**	
1.	Emtricitabine	200.00
2.	Microcrystalline cellulose	106.00
3.	Crospovidone	20.00
II	**Binder Preparation**	
4.	Povidone	10.00
5.	Purified water	q.s.
III.	**Lubrication**	
6.	Microcrystalline Cellulose	86.00
7.	Colloidal silicon dioxide	15.00
8.	Crospovidone	4.00
9.	Magnesium Stearate	4.00
	Total	**445.00**

Film Coating:

Sr. No	Name of Ingredients	Quantity/tab (mg)
1.	Opadry	18.00
2.	Purified water	q.s.

Process:

Preparation of Layer I

A) Granulation

1) Tenofovir, lactose, croscarmellose, corn starch were sifted.

2) The sifted ingredients were dry mixed.

3) Binder solution was prepared using corn starch, polysorbate 80 and purified water.

4) Binder solution obtained in step (3) was sprayed on the mixture obtained in step (2) to form granules.

5) Granules obtained in step (4) were dried, sized and lubricated.

Preparation of Layer II

1) Emtricitabine, microcrystalline cellulose, crospovidone were sifted.

2) The sifted ingredients were dry mixed.

3) Binder solution was prepared using povidone and purified water.

4) Binder solution obtained in step (3) was sprayed on the mixture obtained in step (2) to form granules.

5) Granules obtained in step (4) were dried, sized and lubricated.

B) Compression

1) Lubricated granules of Layer I and Layer II was compressed to produce a bilayer tablets.

C) Coating

1) Tablets so obtained were coated with Opadry solution.

EXAMPLE 2

Emtricitabine & Tenofovir Disoproxil Single layer Tablets:

Sr. No.	Ingredients	Quantity/Unit (mg)
I.	**Tenofovir Disoproxil Fumarate Layer**	
Dry Mix		
1.	Tenofovir Disoproxil Fumarate	300.00
2.	Emtricitabine	200.00
3.	Lactose Monohydrate	80.00
4.	Croscarmellose Sodium	30.00
5.	Microcrystalline Cellulose	300.00
	Pregelatinized Starch	25.00
Binder		
6.	Pregelatinized Starch	25.00
7.	Purified Water	q.s
Blending and Lubrication		
8.	Croscarmellose Sodium	30.00
9.	Magnesium Stearate	10.00
	Total	**1000.00**

	Film Coating	
1.	Opadry Blue	15.00
2.	Purified Water	q.s
	Final Tablet Weight	**1115.00**

Process:

Preparation of Layer I

A) Granulation

1) Tenofovir, emtricitabine, lactose, croscarmellose, microcrystalline cellulose and pregelatinized starch were sifted.

2) The sifted ingredients were dry mixed.

3) Binder solution was prepared using pregelatinized starch and purified water.

4) Binder solution obtained in step (3) was sprayed on the mixture obtained in step (2) to form granules.

5) Granules obtained in step (4) were dried, sized and lubricated.

B) Compression

1) Lubricated granules was compressed to produce a single layer tablets.

C) Coating

1) Tablets so obtained were coated with Opadry solution.

EXAMPLE 3

Ritonavir and Darunavir Bilayered Tablet

Sr. No.	Ingredients	Quantity/Unit (mg)
	Drug Premix	
1	Ritonavir	100.00
2	Colliodal Silicon Dioxide	5.00
	Polymer Premix	
3	Kollidon	400.00
4	Span 20	40.00
	Blending	
5	Crospovidone	50.00
6	Colliodal Silicon Dioxide	5.00
7	Microcrystalline Cellulose	50.00
	Lubrication	
8	Sodium Stearyl Fumarate	10.00
	Total	**660.00**
	Darunavir Layer	

9	Darunavir	325.24
10	Crospovidone	10.00
	Binder	
11	PVP	15.00
12	Purified water	q.s.
	Blending & Lubrication	
13	Crospovidone	10.00
14	Yellow iron oxide	0.50
15	Microcrystalline Cellulose	254.50
16	Colliodal Silicon Dioxide	4.00
17	Magnesium Stearate	6.00
	Total	**625.24**
	Seal Coating	
19	Opadry	5.00
20	Purified water	q.s.
	Film Coating	
21	Opadry 04F 52201 Yellow	15.00
22	Purified water	q.s.
	Total	**1305.24**

Process:

(1) Darunavir was mixed with pre-sieved and pre-sifted amounts of crospovidone, yellow iron oxide, polyvinyl pyrrolidone, microcrystalline cellulose and colloidal silicon dioxide.

(2) The blend obtained in step (1) was granulated with purified water.

(3) Ritonavir with small amount of colloidal silicon dioxide was sifted and mixed with Kollidon and Span 20 in a mixer.

(4) The blend obtained in (3) was mixed and subjected to hot melt extrusion (HME) and the molten mass thus obtained was collected on a conveyor where it was cooled to form extrudates and these extrudates on further milling were converted into granules.

(5) Crospovidone, colloidal silicon dioxide and microcrystalline cellulose were added to the granules obtained in step (4) and further lubricated with sodium stearyl monostearate and magnesium stearate.

(6) The granules obtained in (2) and (5) were compressed together to form a bilayer tablet which was then seal coated and finally film coated.

EXAMPLE 4

Ritonavir and Darunavir Bilayered Tablet

Sr. No.	Ingredients	Quantity/Unit (mg)

	Darunavir Part	
1	Darunavir ethanolate	325.24
2	Microcrystalline cellulose	145.00
3	Crospovidone	10.00
4	Binder	
5	Povidone (PVP)	15.00
6	Purified water	q. s.
	Extragranular	
7	Microcrystalline cellulose	110.00
8	Crospovidone	10.00
	Lubrication	
9	Colliodal Silicon dioxide	4.00
10	Magnesium stearate	6.00
	Total	**625.24**
	Ritonavir Part	
11	Ritonavir	50.00
12	Colloidal silicon dioxide	3.45
	Polymer Part	
13	Kollidon	246.50
14	Polyoxyl hydrogenated castor oil	33.35
	Blending & Lubrication	
15	Colloidal silicon dioxide	6.95
16	Dibasic calcium phosphate (anhydrous)	84.70
	Total	**1050.19**

Process:

(1) Darunavir Ethanolate was mixed with pre-sieved and presifted quantities of crospovidone and microcrystalline cellulose.

(2) The mixture obtained in step (1) was granulated with PVP followed by mixing and lubrication with crospovidone, microcrystalline cellulose, colloidal silicon dioxide and magnesium stearate.

(3) Ritonavir with small amount of colloidal silicon dioxide was sifted and mixed together with Kollidon and polyoxyl 40 hydrogenated castor oil in a mixer.

(4) The blend obtained in (3) was mixed and subjected to hot melt extrusion (HME) and the molten mass thus obtained was collected on a conveyor where it was cooled to form extrudates and these extrudates on further milling were converted into granules.

(5) Colloidal silicon dioxide and anhydrous dibasic calcium phosphate was added to the granules obtained in step (4).

(6) The granules obtained in (2) and (5) were compressed together to form a bilayer tablet which was then finally film coated.

EXAMPLE 5

Ritonavir and Darunavir Bilayered Tablet

Darunavir Layer:

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Dry Mix for Compaction**	
1	Darunavir Ethanolate	325.24
2	Silicified Microcrystalline Cellulose	275.36
3	Crospovidone	12.48
	Colloidal Silicon Dioxide	6.24
II	**Lubrication**	
4	Magnesium Stearate	4.68
	Total of Layer I	**624.00**

Ritonavir Layer:

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Drug Premix**	
1	Ritonavir	50.00
2	Colloidal Silicon Dioxide	3.45
3	Dibasic Calcium Phosphate	22.50
II	**Polymer Premix**	
4	Copovidone	246.55
5	Sorbitan Monolaureate	33.35
III	**Blending**	
6	Colloidal Silicon Dioxide	5.00
7	Dibasic Calcium Phosphate	22.20
IV	**Lubricant**	
8	Sodium Stearyl Fumarate	1.95
	Total of Layer II	**385.00**
	Total (Layer I + Layer II)	**1009.00**
	Film Coating	
9	Opadry Orange	31.00
10	Purified Water	q. s.
	Total	**1040.00**

Process

Manufacturing Process for Darunavir Layer:

1. Darunavir, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide and were sifted and mixed to form granules.
2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide & dibasic calcium phosphate were sifted and granulated with copovidone and sorbitan monolaureate to form granules
2. The granules obtained in step (1) were extruded and lubricated with sodium stearyl fumarate
3. The lubricated granules obtained in step (2) of the darunavir layer and step (2) of the ritonavir layer were compressed to produce a bilayer tablet and coated with opadry orange.

EXAMPLE 6

Darunavir and Ritonavir Bilayered Tablet

Darunavir Layer:

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Dry Mix for Compaction**	
1	Darunavir Ethanolate	325.24
2	Silicified Microcrystalline Cellulose	275.36
3	Crospovidone	12.48
4	Colloidal Silicon Dioxide	6.24
II	**Lubrication**	
5	Magnesium Stearate	4.68
	Total of Layer I	**624.00**

Ritonavir Layer:

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Drug Premix**	
1	Ritonavir	100.00
2	Colloidal Silicon Dioxide	6.90
3	Dibasic Calcium Phosphate	45.00
II	**Polymer Premix**	
4	Copovidone	493.10
5	Sorbitan Monolaureate	66.70
III	**Blending**	
6	Colloidal Silicon Dioxide	10.00
7	Dibasic Calcium Phosphate	44.40
IV	**Lubricant**	
8	Sodium Stearyl Fumarate	3.90
	Total of Layer II	**770.00**
	Total (Layer I + Layer II)	**1394.00**
	Film Coating	
9	Opadry Orange	42.00
10	Purified Water	q. s.
	Total	**1436.00**

Process

Manufacturing Process for Darunavir Layer:

1. Darunavir, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide and were sifted and mixed to form granules.
2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide & dibasic calcium phosphate were sifted and granulated with copovidone and sorbitan monolaureate to form granules
2. The granules obtained in step (1) were extruded and lubricated with sodium stearyl fumarate
3. The lubricated granules obtained in step (2) of the darunavir layer and step (2) of the ritonavir layer were compressed to produce a bilayer tablet and coated with opadry orange.

EXAMPLE 7

Darunavir and Ritonavir Bilayered Tablet

Sr. No.	Ingredients	Quantity / Unit (mg)
I	**Dry Mix**	
1	Darunavir Ethanolate	432.87
II	**Binder**	
2	Hydroxy propyl methyl cellulose	6.60
3	Purified water	q.s.
III	**Blending and lubrication**	
4	Silicified microcrystalline cellulose	122.50
5	Colloidal silicon dioxide	1.65
6	Crospovidone	16.50
7	Magnesium Stearate	2.75
	Total weight of Layer I	**582.87**
Ritonavir Part		
I	**Drug premix**	
1	Ritonavir	50.00
2	Colloidal silicon dioxide	3.45
3	Dibasic calcium phosphate anhydrous	22.50
II	**Polymer premix**	
4	Copovidone	246.55
5	Sorbitan monolaurate	33.35

III	**Blending and lubrication**	
6	Colloidal silicon dioxide	5.00
7	Dibasic calcium phosphate anhydrous	22.20
8	Sodium stearyl fumarate	1.95
	Total weight of Layer II	**385.00**
	Total weight of core tablet	**967.87**
	Film Coating	
9	Opadry Red	37.00
10	Purified water	-
	Total weight of film coated tablet	**1004.87**

Process

Manufacturing Process for Darunavir Layer:

1. Darunavir, HPMC, silicified microcrystalline cellulose, colloidal silicon dioxide and crospovidone were sifted and granulated to form granules.
2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted and granulated with copovidone, sorbitan monolaureate.
2. The granules obtained in step (1) were extruded and lubricated by sodium stearyl fumarate.
3. The lubricated granules obtained in step (2) of the darunavir layer and step (2) of the ritonavir layer were compressed to produce a bilayer tablet and coated with opadry red.

EXAMPLE 8

Darunavir and Ritonavir Bilayered Tablet

Sr. No.	Ingredients	Quantity / Unit (mg)
I	**Dry Mix**	
1	Darunavir Ethanolate	432.87
II	**Binder**	
2	Hydroxy propyl methyl cellulose	6.60
3	Purified water	q.s.
III	**Blending and lubrication**	
4	Silicified microcrystalline cellulose	122.50
5	Colloidal silicon dioxide	1.65
6	Crospovidone	16.50
7	Magnesium Stearate	2.75

	Total weight of Layer I	**582.87**
Ritonavir Part		
I	**Drug premix**	
1	Ritonavir	100.000
2	Colloidal silicon dioxide	6.900
3	Dibasic calcium phosphate anhydrous	45.000
II	**Polymer premix**	
4	Copovidone	493.100
5	Sorbitan monolaurate	66.700
III	**Blending and lubrication**	
6	Colloidal silicon dioxide	10.000
7	Dibasic calcium phosphate anhydrous	44.400
8	Sodium stearyl fumarate	3.900
	Total weight of Layer II	**770.00**
	Total weight of core tablet	**1352.87**
	Film Coating	
9	Opadry Red	**40.00**
10	Purified water	-
	Total weight of film coated tablet	**1392.87**

Process

Manufacturing Process for Darunavir Layer:

1. Darunavir, HPMC, silicified microcrystalline cellulose, colloidal silicon dioxide and crospovidone were sifted and granulated to form granules.
2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted and granulated with copovidone, sorbitan monolaureate.
2. The granules obtained in step (1) were extruded and lubricated with sodium stearyl fumarate.
3. The lubricated granules obtained in step (2) of the darunavir layer and step (2) of the ritonavir layer were compressed to produce a bilayer tablet and coated with opadry red.

EXAMPLE 9

Darunavir Ritonavir and Dolutegravir Bilayered Tablet

Darunavir Layer:

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Dry Mix for Compaction**	
1	Darunavir Ethanolate	325.24
2	Dolutegravir	50.00
3	Silicified Microcrystalline Cellulose	275.36
4	Crospovidone	12.48
	Colloidal Silicon Dioxide	6.24
II	**Lubrication**	
5	Magnesium Stearate	4.680
	Total of Layer I	**674.000**

Ritonavir Layer:

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Drug Premix**	
1	Ritonavir	50.00
2	Colloidal Silicon Dioxide	3.450
3	Dibasic Calcium Phosphate	22.50
II	**Polymer Premix**	
4	Copovidone	246.55
5	Sorbitan Monolaureate	33.35
III	**Blending**	
6	Colloidal Silicon Dioxide	5.00
7	Dibasic Calcium Phosphate	22.20
IV	**Lubricant**	
8	Sodium Stearyl Fumarate	1.95
	Total of Layer II	**385.00**
	Total (Layer I + Layer II)	**1059.00**
	Film Coating	
9	Opadry Orange	31.00
10	Purified Water	q. s.
	Total	**1090.00**

Process

Manufacturing Process for Darunavir Layer:

1. Darunavir, dolutegravir, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide were sifted and granulated to form granules.

2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted and granulated with copovidone, sorbitan monolaureate and water to form granules.

2. The granules obtained in step (1) were extruded and lubricated with sodium stearyl fumarate.

3. The lubricated granules obtained in step (2) of the darunavir layer and step (2) of the ritonavir layer were compressed to produce a bilayer tablet and coated with opadry orange.

EXAMPLE 10

Darunavir Ritonavir and Dolutegravir Bilayered Tablet

Darunavir Layer:

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Dry Mix for Compaction**	
1	Darunavir Ethanolate	325.24
2	Dolutegravir	50.00
3	Silicified Microcrystalline Cellulose	275.364
4	Crospovidone	12.48
	Colloidal Silicon Dioxide	6.24
II	**Lubrication**	
5	Magnesium Stearate	4.68
	Total of Layer I	**674.00**

Ritonavir Layer:

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Drug Premix**	
1	Ritonavir	100.00
2	Colloidal Silicon Dioxide	6.90
3	Dibasic Calcium Phosphate	45.00
II	**Polymer Premix**	
4	Copovidone	493.10
5	Sorbitan Monolaureate	66.70
III	**Blending**	
6	Colloidal Silicon Dioxide	10.00
7	Dibasic Calcium Phosphate	44.40
IV	**Lubricant**	
8	Sodium Stearyl Fumarate	3.90
	Total of Layer II	**770.0**
	Total (Layer I + Layer II)	**1444.0**
	Film Coating	
9	Opadry Orange	42.00
10	Purified Water	q. s.
	Total	**1486.00**

Process

Manufacturing Process for Darunavir Layer:

1. Darunavir, dolutegravir, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide were sifted and granulated to form granules.

2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted and granulated with copovidone, sorbitan monolaureate and water to form granules.

2. The granules obtained in step (1) were extruded and lubricated with sodium stearyl fumarate.

3. The lubricated granules obtained in step (2) of the darunavir layer and step (2) of the ritonavir layer were compressed to produce a bilayer tablet and coated with opadry orange.

EXAMPLE 11

Darunavir Ritonavir and Dolutegravir Bilayered Tablet

Sr. No.	Ingredients	Quantity / Unit (mg)
I	**Dry Mix**	
1	Darunavir Ethanolate	432.87
II	**Binder**	
2	Hydroxy propyl methyl cellulose	6.60
3	Purified water	q.s.
III	**Blending and lubrication**	
4	Dolutegravir	50.00
5	Silicified microcrystalline cellulose	122.50
6	Colloidal silicon dioxide	1.65
7	Crospovidone	16.50
8	Magnesium Stearate	2.75
	Total weight of Layer I	**632.87**
Ritonavir Part		
I	**Drug premix**	
1	Ritonavir	50.00
2	Colloidal silicon dioxide	3.45
3	Dibasic calcium phosphate anhydrous	22.50
II	**Polymer premix**	
4	Copovidone	246.55
5	Sorbitan monolaurate	33.35
III	**Blending and lubrication**	

6	Colloidal silicon dioxide	5.00
7	Dibasic calcium phosphate anhydrous	22.20
8	Sodium stearyl fumarate	1.95
	Total weight of Layer II	**385.00**
	Total weight of core tablet	**1017.87**
	Film Coating	
9	Opadry Red	37.00
10	Purified water	-
	Total weight of film coated tablet	**1054.87**

Process

Manufacturing Process for Darunavir Layer:

1. Hypromellose was sprayed onto darunavir to produce granules.
2. The granulates obtained in step (1) were dried, sized and mixed with dolutegravir, microcrystalline cellulose, colloidal Silicon Dioxide and crospovidone to produce a mixed blend.
3. The mixed blend obtained in step (2) was lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted, dry mixed and granulated with copovidone, sorbitan monolaureate and water to produce granules.
2. The granules obtained in step (1) were extruded and lubricated with sodium stearyl fumarate.
3. The lubricated granules obtained in step (3) of the darunavir layer and step (2) of the ritonavir layer were compress to form a bilayer tablet and coated with opadry red.

EXAMPLE 12

Darunavir Ritonavir and Dolutegravir Bilayered Tablet

Sr. No.	Ingredients	Quantity / Unit (mg)
I	**Dry Mix**	
1	Darunavir Ethanolate	432.87
II	**Binder**	
2	Hydroxy propyl methyl cellulose	6.60
3	Purified water	q.s.
III	**Blending and lubrication**	
4	Dolutegravir	50.000
5	Silicified microcrystalline cellulose	122.50

6	Colloidal silicon dioxide	1.65
7	Crospovidone	16.50
8	Magnesium Stearate	2.75
	Total weight of Layer I	**632.87**
Ritonavir Part		
I	**Drug premix**	
1	Ritonavir	100.000
2	Colloidal silicon dioxide	6.900
3	Dibasic calcium phosphate anhydrous	45.000
II	**Polymer premix**	
4	Copovidone	493.100
5	Sorbitan monolaurate	66.700
III	**Blending and lubrication**	
6	Colloidal silicon dioxide	10.000
7	Dibasic calcium phosphate anhydrous	44.400
8	Sodium stearyl fumarate	3.900
	Total weight of Layer II	**770.00**
	Total weight of core tablet	**1402.87**
	Film Coating	
9	Opadry Red	**40.00**
10	Purified water	-
	Total weight of film coated tablet	**1442.87**

Process

Manufacturing Process for Darunavir Layer:

1. Hypromellose was sprayed onto darunavir to produce granules.

2. The granulates obtained in step (1) were dried, sized and mixed with dolutegravir, microcrystalline cellulose, colloidal Silicon Dioxide and crospovidone to produce a mixed blend.

3. The mixed blend obtained in step (2) was lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted, dry mixed and granulated with copovidone, sorbitan monolaureate and water to produce granules.

2. The granules obtained in step (1) were extruded and lubricated with sodium stearyl fumarate.

3. The lubricated granules obtained in step (3) of the darunavir layer and step (2) of the ritonavir layer were compress to form a bilayer tablet and coated with opadry red.

EXAMPLE 13

Darunavir Ritonavir and Dolutegravir Trilayered Tablet

Dolutegravir layer

Sr. No.	Ingredients	Quantity / Unit (mg)
	Dry Mixing	
1	Dolutegravir	50.00
2	Mannitol	60.40
3	Microcrystalline Cellulose	162.85
4	Sodium Starch Glycolate	6.00
5	Polyvinyl Pyrollidone	5.00
	Binder	
6	Purified Water	q.s
	Lubrication	
7	Sodium Starch Glycolate	15.00
8	Sodium Stearyl Fumarate	0.75
	Total of Layer I	**300.00**

Darunavir Layer

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Dry Mix for Compaction**	
1	Darunavir Ethanolate	325.24
2	Silicified Microcrystalline Cellulose	275.36
3	Crospovidone	12.48
	Colloidal Silicon Dioxide	6.24
II	**Lubrication**	
4	Magnesium Stearate	4.68
	Total of Layer II	**624.00**

Ritonavir Layer:

Sr. No.	Ingredients	Qty/Unit (mg)
I	**Drug Premix**	
1	Ritonavir	50.00
2	Colloidal Silicon Dioxide	3.45
3	Dibasic Calcium Phosphate	22.50
II	**Polymer Premix**	
4	Copovidone	246.55
5	Sorbitan Monolaureate	33.35
III	**Blending**	
6	Colloidal Silicon Dioxide	5.00
7	Dibasic Calcium Phosphate	22.20
IV	**Lubricant**	
8	Sodium Stearyl Fumarate	1.95
	Total of Layer III	**385.000**

	Total (Layer I + Layer II + Layer III)	**1309.00**
	Film Coating	
9	Opadry Orange	41.00
10	Purified Water	q. s.
	Total	**1350.00**

Process

Manufacturing Process for Dolutegravir Layer:

1. Dolutegravir, mannitol, microcrystalline cellulose and polyvinyl pyrollidone were sifted, granulated, dried and sized to form granules

2. The granules obtained in step (1) were lubricated with sodium starch glycolate and sodium stearyl fumarate.

Manufacturing Process for Darunavir Layer:

1. Darunavir, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide were sifted, mixed and granulated.

2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted, dry mixed and granulated with copovidone, sorbitan monolaureate and water to form granules.

2. The granules were extruded and colloidal silicon dioxide, dibasic calcium phosphate were further added to the granules which were then lubricated with sodium stearyl fumarate.

3. The lubricated granules obtained in step (2) of the dolutegravir layer and the step (2) of the darunavir layer and the step (2) of the ritonavir layer were compressed to form a trilayer tablet and coated with opadry orange.

EXAMPLE 14

Darunavir Ritonavir and Dolutegravir Trilayered Tablet

Dolutegravir layer

Sr. No.	Ingredients	Quantity / Unit (mg)
	Dry Mixing	
1.	Dolutegravir	50.00
2.	Mannitol	60.40

3.	Microcrystalline Cellulose	162.853
4.	Sodium Starch Glycolate	6.00
5.	Polyvinyl Pyrollidone	5.00
	Binder	
6.	Purified Water	q.s
	Lubrication	
7.	Sodium Starch Glycolate	15.00
8.	Sodium Stearyl Fumarate	0.75
	Total of Layer I	**300.00**

Darunavir Layer

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Dry Mix for Compaction**	
1	Darunavir Ethanolate	325.236
2	Silicified Microcrystalline Cellulose	275.364
3	Crospovidone	12.480
	Colloidal Silicon Dioxide	6.240
II	**Lubrication**	
4	Magnesium Stearate	4.680
	Total of Layer II	**624.000**

Ritonavir Layer:

Sr. No.	Ingredients	Quantity/Unit (mg)
I	**Drug Premix**	
1	Ritonavir	100.000
2	Colloidal Silicon Dioxide	6.900
3	Dibasic Calcium Phosphate	45.000
II	**Polymer Premix**	
4	Copovidone	493.100
5	Sorbitan Monolaureate	66.700
III	**Blending**	
6	Colloidal Silicon Dioxide	10.000
7	Dibasic Calcium Phosphate	44.400
IV	**Lubricant**	
8	Sodium Stearyl Fumarate	3.900
	Total of Layer III	**770.00**
	Total (Layer I + Layer II + Layer III)	**1694.00**
	Film Coating	
9	Opadry Orange	41.000
10	Purified Water	q. s.
	Total	**1735.00**

Process

Manufacturing Process for Dolutegravir Layer:

1. Dolutegravir, mannitol, microcrystalline cellulose and polyvinyl pyrollidone were sifted, granulated, dried and sized to form granules

2. The granules obtained in step (2) were lubricated with sodium starch glycolate and sodium stearyl fumarate.

Manufacturing Process for Darunavir Layer:

1. Darunavir, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide were sifted, mixed and granulated.

2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted, dry mixed and granulated with copovidone, sorbitan monolaureate and water to form granules.

2. The granules were extruded and lubricated with sodium stearyl fumarate.

3. The lubricated granules obtained in step (2) of the dolutegravir layer and the step (2) of the darunavir layer and the step (2) of the ritonavir layer were compressed to form a trilayer tablet and coated with opadry orange.

EXAMPLE 15

Darunavir Ritonavir and Dolutegravir Trilayered Tablet

Dolutegravir Layer

Sr. No.	Ingredients	Quantity / Unit (mg)
	Dry Mixing	
1.	Dolutegravir	50.00
2.	Mannitol 25 (Plain)	60.40
3.	Microcrystalline Cellulose	162.853
4.	Sodium Starch Glycolate	6.00
5.	Polyvinyl Pyrollidone	5.00
	Binder	
6.	Purified Water	q.s
	Lubrication	
7.	Sodium Starch Glycolate	15.00
8.	Sodium Stearyl Fumarate	0.75

	Total of Layer I	**300.00**

Darunavir Layer

Sr. No.	Ingredients	Quantity / Unit (mg)
I	**Dry Mix**	
1	Darunavir Ethanolate	432.87
II	**Binder**	
2	Hydroxy propyl methyl cellulose	6.60
3	Purified water	q.s.
III	**Blending and lubrication**	
4	Silicified microcrystalline cellulose	122.50
5	Colloidal silicon dioxide	1.65
6	Crospovidone	16.50
7	Magnesium Stearate	2.75
	Total weight of Layer II	**582.87**
Ritonavir Part		
I	**Drug premix**	
1	Ritonavir	50.00
2	Colloidal silicon dioxide	3.45
3	Dibasic calcium phosphate anhydrous	22.50
II	**Polymer premix**	
4	Copovidone	246.55
5	Sorbitan monolaurate	33.35
III	**Blending and lubrication**	
6	Colloidal silicon dioxide	5.00
7	Dibasic calcium phosphate anhydrous	22.20
8	Sodium stearyl fumarate	1.95
	Total weight of Layer III	**385.00**
	Total weight of core tablet	**1267.87**
	Film Coating	
9	Opadry Red	37.00
10	Purified water	Q s
	Total weight of film coated tablet	**1304.87**

Process

Manufacturing Process for Dolutegravir Layer:

1. Dolutegravir, mannitol, microcrystalline cellulose and polyvinyl pyrollidone were sifted, granulated, dried and sized to form granules

2. The granules obtained in step (2) were lubricated with sodium starch glycolate and sodium stearyl fumarate.

Manufacturing Process for Darunavir Layer:

1. Darunavir, HPMC, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide were sifted, mixed and granulated.

2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted, dry mixed and granulated with copovidone and sorbitan monolaureate water to form granules.

2. The granules were extruded and lubricated with sodium stearyl fumarate.

3. The lubricated granules obtained in step (2) of the dolutegravir layer and the step (2) of the darunavir layer and the step (2) of the ritonavir layer were compressed to form a trilayer tablet and coated with opadry red.

EXAMPLE 16

Darunavir Ritonavir and Dolutegravir Trilayered Tablet

Dolutegravir Layer

Sr. No.	Ingredients	Quantity / Unit (mg)
	Dry Mixing	
1	Dolutegravir	50.00
2	Mannitol	60.40
3	Microcrystalline Cellulose	162.853
4	Sodium Starch Glycolate	6.00
5	Polyvinyl Pyrollidone	5.00
	Binder	
6	Purified Water	q.s
	Lubrication	
7	Sodium Starch Glycolate	15.00
8	Sodium Stearyl Fumarate	0.75
	Total of Layer I	**300.00**

Darunavir Layer

Sr. No.	Ingredients	Quantity / Unit (mg)
I	**Dry Mix**	
1	Darunavir Ethanolate	432.87
II	**Binder**	
2	Hydroxy propyl methyl cellulose	6.60
3	Purified water	q.s.
III	**Blending and lubrication**	
4	Silicified microcrystalline cellulose	122.50

5	Colloidal silicon dioxide	1.65
6	Crospovidone	16.50
7	Magnesium Stearate	2.75
	Total weight of Layer II	**582.87**
Ritonavir Part		
I	**Drug premix**	
1	Ritonavir	100.000
2	Colloidal silicon dioxide	6.900
3	Dibasic calcium phosphate anhydrous	45.000
II	**Polymer premix**	
4	Copovidone	493.100
5	Sorbitan monolaurate	66.700
III	**Blending and lubrication**	
6	Colloidal silicon dioxide	10.000
7	Dibasic calcium phosphate anhydrous	44.400
8	Sodium stearyl fumarate	3.900
	Total weight of Layer III	**770.00**
	Total weight of core tablet (Layer I + Layer II + Layer III)	**1652.87**
	Film Coating	
9	Opadry Red	**40.00**
10	Purified water	-
	Total weight of film coated tablet	**1692.87**

Process

Manufacturing Process for Dolutegravir Layer:

1. Dolutegravir, mannitol, microcrystalline cellulose and polyvinyl pyrollidone were sifted, granulated, dried and sized to form granules

2. The granules obtained in step (2) were lubricated with sodium starch glycolate and sodium stearyl fumarate.

Manufacturing Process for Darunavir Layer:

1. Darunavir, HPMC, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide were sifted, mixed and granulated.

2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted, dry mixed and granulated with copovidone and sorbitan monolaureate water to form granules.

2. The granules were extruded and lubricated with sodium stearyl fumarate.

3. The lubricated granules obtained in step (2) of the dolutegravir layer and the step (2) of the darunavir layer and the step (2) of the ritonavir layer were compressed to form a trilayer tablet and coated with opadry red.

EXAMPLE 17

Darunavir Ritonavir Raltegravir trilayer tablets

Raltegravir Layer

Sr. No.	Ingredients	Quantity / Unit (mg)
	Dry Mixing	
1	Raltegravir Potassium	434.40
2	Lactose Monohydrate	26.06
3	Calcium Phosphate	69.50
4	Magnesium Stearate	13.03
	Binder	
5.	Polaxamer	104.48
6.	Hypromellose	43.44
7.	Purified Water	q.s
	Blending and Lubrication	
8.	Microcrystalline Cellulose	50.40
9.	Sodium Stearyl Fumarate	8.69
	Total of Layer I	**750.0**

Darunavir Layer

Sr. No.	Ingredients	Quantity / Unit (mg)
I	**Dry Mix**	
1	Darunavir Ethanolate	400.00
II	**Binder**	
2	Hydroxy propyl methyl cellulose	6.60
3	Purified water	q.s.
III	**Blending and lubrication**	
4	Silicified microcrystalline cellulose	52.50
5	Colloidal silicon dioxide	1.65
6	Crospovidone	16.50
7	Magnesium Stearate	2.75
	Total weight of Layer II	**480.0**
Ritonavir Part		
I	**Drug premix**	
1	Ritonavir	100.00
2	Colloidal silicon dioxide	6.90
3	Dibasic calcium phosphate	45.00
II	**Polymer premix**	

4	Copovidone	293.10
5	Sorbitan monolaurate	66.70
III	**Blending and lubrication**	
6	Colloidal silicon dioxide	10.00
7	Dibasic calcium phosphate anhydrous	44.40
8	Sodium stearyl fumarate	3.90
	Total weight of Layer III	**570.00**
	Total weight of core tablet (Layer I + Layer II + Layer III)	**1800.00**
	Film Coating	
9	Opadry Red	**36.00**
10	Purified water	-
	Total weight of film coated tablet	**1836.00**

Process

Manufacturing Process for Raltegravir Layer:

1. Raltegravir, lactose, calcium phosphate, polaxamer, hypromellose, and microcrystalline cellulose were sifted, granulated, dried and sized to form granules

2. The granules obtained in step (2) were lubricated with magnesium stearate and sodium stearyl fumarate.

Manufacturing Process for Darunavir Layer:

1. Darunavir, HPMC, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide were sifted, mixed and granulated.

2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted, dry mixed and granulated with copovidone and sorbitan monolaureate water to form granules.

2. The granules were extruded and lubricated with sodium stearyl fumarate.

3. The lubricated granules obtained in step (2) of the dolutegravir layer and the step (2) of the darunavir layer and the step (2) of the ritonavir layer were compressed to form a bilayer tablet and coated with opadry red.

EXAMPLE 18

Darunavir Ritonavir Raltegravir bilayer tablets

Raltegravir Layer

Sr. No.	Ingredients	Quantity / Unit (mg)

	Dry Mixing	
1	Raltegravir Potassium	434.40
2	Lactose Monohydrate	26.06
3	Calcium Phosphate	69.50
4	Magnesium Stearate	13.03
	Binder	
5.	Polaxamer	104.48
6.	Hypromellose	43.44
7.	Purified Water	q.s
	Blending and Lubrication	
8.	Microcrystalline Cellulose	50.40
9.	Sodium Stearyl Fumarate	8.69
	Total Weight of Layer I	**750.00**

Darunavir Layer

Sr. No.	Ingredients	Quantity / Unit (mg)
I	**Dry Mix**	
1	Darunavir Ethanolate	400.00
II	**Binder**	
2	Hydroxy propyl methyl cellulose	6.60
3	Purified water	q.s.
III	**Blending and lubrication**	
4	Silicified microcrystalline cellulose	52.50
5	Colloidal silicon dioxide	1.65
6	Crospovidone	16.50
7	Magnesium Stearate	2.75
	Total weight of Layer II	**480.0**
Ritonavir Part		
I	**Drug premix**	
1	Ritonavir	50.00
2	Colloidal silicon dioxide	6.90
3	Dibasic calcium phosphate	45.00
II	**Polymer premix**	
4	Copovidone	293.10
5	Sorbitan monolaurate	66.70
III	**Blending and lubrication**	
6	Colloidal silicon dioxide	10.00
7	Dibasic calcium phosphate anhydrous	94.40
8	Sodium stearyl fumarate	3.90
	Total weight of Layer III	**570.00**
	Total weight of core tablet (Layer I + Layer II + Layer III)	**1800.0**
	Film Coating	
9	Opadry Red	**36.00**

10	Purified water	-
	Total weight of film coated tablet	**1836.00**

Process

Manufacturing Process for Raltegravir Layer:

1. Raltegravir, lactose, calcium phosphate, polaxamer, hypromellose, and microcrystalline cellulose were sifted, granulated, dried and sized to form granules

2. The granules obtained in step (2) were lubricated with magnesium stearate and sodium stearyl fumarate.

Manufacturing Process for Darunavir Layer:

1. Darunavir, HPMC, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide were sifted, mixed and granulated.

2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted, dry mixed and granulated with copovidone and sorbitan monolaureate to form granules.

2. The granules were extruded and lubricated with sodium stearyl fumarate.

3. The lubricated granules obtained in step (2) of the dolutegravir layer and the step (2) of the darunavir layer and the step (2) of the ritonavir layer were compressed to form a bilayer tablet and coated with opadry red.

EXAMPLE 19

Lamivudine and Tenofovir Disoproxil Bilayer Tablets:

Sr. No	Name of Ingredients	Quantity/tab (mg)
Tenofovir Layer		
I	**Dry Mix**	
1.	Tenofovir Disoproxil Fumarate	300.00
2.	Lactose Monohydrate	159.00
3.	Croscarmellose sodium	20.00
4.	Corn Starch	30.00
II	**Binder Preparation**	
5.	Corn Starch	15.00
6.	Polysorbate 80	3.00
7.	Purified Water	q. s.
III.	**Lubrication**	
8.	Microcrystalline Cellulose	100.00
9.	Croscarmellose sodium	20.00

10.	Magnesium Stearate	12.50
	Total	**659.50**

Sr. No	**Name of Ingredients**	**Quantity/tab (mg)**
Lamivudine Layer		
I	**Dry Mix**	
1.	Lamivudine	300.00
2.	Microcrystalline cellulose	103.20
3.	Sodium starch glycolate	30.00
II	**Binder Preparation**	
4.	Corn starch	10.20
5.	Purified water	q.s.
III.	**Lubrication**	
6.	Sodium starch glycolate	20.00
7.	Magnesium Stearate	6.00
	Total	**469.40**

Film Coating:

Sr. No	**Name of Ingredients**	**Quantity/tab (mg)**
1.	Opadry	18.00
2.	Purified water	q.s.

Process:

Preparation of Layer I

A) Granulation

1) Tenofovir, lactose, croscarmellose, corn starch were sifted.

2) The sifted ingredients were dry mixed.

3) Binder solution was prepared using corn starch, polysorbate 80 and purified water.

4) Binder solution obtained in step (3) was sprayed on the mixture obtained in step (2) to form granules.

5) Granules obtained in step (4) were dried, sized and lubricated.

Preparation of Layer II

1) Lamivudine, microcrystalline cellulose, sodium starch glycolate were sifted.

2) The sifted ingredients were dry mixed.

3) Binder solution was prepared using corn starch and purified water.

4) Binder solution obtained in step (3) was sprayed on the mixture obtained in step (2) to form granules.

5) Granules obtained in step (4) were dried, sized and lubricated.

B) Compression

1) Lubricated granules of Layer I and Layer II was compressed to produce a bilayer tablets.

C) Coating

1) Tablets so obtained were coated with Opadry solution.

EXAMPLE 20

Lamivudine and Zidovudine Monolayer Tablet for Oral Suspension:

Sr. No.	Ingredients	Quantity / tablet (mg)
1.	Lamivudine	30.00
2.	Zidovudine	60.00
3.	Microcrystalline Cellulose	52.38
4.	Sodium starch glycolate	6.00
5.	Starch	4.00
6.	Purified water	q.s.
7.	Colloidal silicon dioxide	0.50
8.	Aspartame	3.00
9.	Flavour	3.00
10.	Magnesium Stearate	1.12
	Total	**160.0**

Process:

(1) Dry mix of lamivudine, zidovudine with microcrystalline cellulose, sodium starch glycolate, starch and colloidal silicon dioxide was prepared.

(2) Binder solution was prepared and dry mix obtained from step (1) was granulated.

(3) Granules obtained from step (2) were blended and lubricated and were compressed to form tablet.

EXAMPLE 21

Lamivudine and Zidovudine Monolayer Tablets

Sr No.	Name of Ingredients	Quantity/tab (mg)
Dry Mix		
1.	Lamivudine	150.00
2.	Zidovudine	300.00
3.	Microcrystalline Cellulose	269.62
4.	Sodium Starch Glycolate	22.50
5.	Colloidal Silicon Dioxide	2.25
Lubrication		
6.	Magnesium Stearate	5.63

	Total	**750.00**
Film Coating		
7.	Hypromellose	8.83
8.	Titanium Dioxide	2.94
9.	Talc	1.77
10.	Propylene Glycol	1.46
11.	Isopropyl Alcohol	q.s.
12.	Purified Water	q.s.
	Total	**765.00**

Process

(1) Lamivudine, zidovudine, microcrystalline cellulose, sodium starch glycolate and colloidal silicon dioxide were mixed in a blender.

(2) Magnesium stearate was added to the mixture obtained in step (1) and the mixture was compressed into tablets.

(3) The tablets obtained in step (2) were film coated.

EXAMPLE 22

Tenofovir Disoproxil Fumarate & Lamivudine Bilayer Tablets

Sr. No.	**Name of Ingredients**	**Quantity/tab (mg)**
Tenofovir Disoproxil Fumarate Layer		
I. Dry Mix		
1.	Tenofovir Disoproxil Fumarate	300.00
2.	Lactose Monohydrate	159.50
3.	Croscarmellose Sodium	20.00
4.	Corn starch	30.00
II. Binder Preparation		
5.	Corn Starch	15.00
6.	Polysorabte 80	3.00
7.	Purified Water	
III. Lubrication		
8.	Microcrystalline Cellulose	100.00
9.	Croscarmellose Sodium	20.00
10.	Magnesium Stearate	12.50
	Total	**660.00**
Lamivudine Layer		
I. Dry Mix		
11.	Lamivudine	300.00
12.	Microcrystalline Cellulose	103.20
13.	Sodium Starch Glycolate	30.00

14.	FD& C Yellow No.6 INH	0.60
II. Binder Preparation		
15.	Corn Starch	10.20
16.	Purified water	q.s.
III. Lubrication		
17.	Sodium Starch Glycolate	20.00
18.	Magnesium Staerate	6.00
	Total	**470.00**
Film Coating		
19	Opadry AMB OY-B 29000 Translucent	18.00
20.	Purified Water	q.s.
	Total	**1148.00**

Process:

Tenofovir Disoproxil Fumarate Layer:

(1) Tenofovir Disoproxil Fumarate, lactose monohydrate, croscarmellose sodium and corn starch were mixed to produce a dry mix.

(2) Corn starch and polysorabte 80 were mixed to form a binder solution.

(3) The binder solution obtained in step (2) was sprayed onto the dry mix obtained in step (1) to form granules.

(4) The granules obtained in step (3) were dried and sized and mixed with microcrystalline cellulose and croscarmellose sodium and lubricated with magnesium stearate.

Lamivudine Layer:

(1) Lamivudine, microcrystalline cellulose, sodium starch glycolate were mixed to form a dry mix.

(2) Corn starch and purified water were mixed to form a binder solution.

(3) The binder solution obtained in step (2) was sprayed onto the dry mix obtained in step (1) to form granules.

(4) The granules obtained in step (3) were dried and sized and mixed with sodium starch glycolate and lubricated with magnesium stearate.

Compression and Coating:

(1) Tenofovir Disoproxil Fumarate granules and Lamivudine granules were compressed into a bilayer tablet and coated with Opadry solution.

EXAMPLE 23

Abacavir Sulfate & Lamivudine Monolayer Tablets

Formula

Sr. No.	Name of Ingredients	Quantity/tab (mg)
Abacavir Granules		
1.	Abacavir Sulfate	702.78
2.	Microcrystalline Cellulose	145.22
3.	Sodium Starch Glycolate	25.00
4.	Hypromellose	10.00
5.	Purified water	q.s.
Lamivudine Granules		
6.	Lamivudine	300.00
7.	Sodium starch glycolate	25.00
8.	Microcrystalline Cellulose	138.00
9.	Corn Starch	10.00
10.	Purified Water	q.s.
Lubrication		
11.	Colliodal Silicon Dioxide	7.00
12.	Sodium starch glycolate	50.00
13.	Magnesium Stearate	12.00
	Total	**1425.00**
Film Coating		
14.	Opadry Orange 14B 53805	20.00
15.	Purified Water	q.s.
	Total	**1445.00**

Process:

Abacavir granules

(1) Abacavir Sulfate, microcrystalline cellulose, sodium starch glycolate were mixed to form a dry mix.

(2) Hypromellose and purified water were mixed to form a binder solution.

(3) The binder solution obtained in step (2) was sprayed onto the dry mix obtained in step (1) to form granules.

(4) The granules obtained in step (3) were dried and sized

Lamivudine Granules

(1) Lamivudine, sodium starch glycolate microcrystalline cellulose were mixed to form a dry mix.

(2) Corn Starch and purified water were mixed to form a binder solution.

(3) The binder solution obtained in step (2) was sprayed onto the dry mix obtained in step (1) to form granules.

(4) The granules obtained in step (3) were dried and sized

Blending and Lubrication

(1) Abacavir granules and lamivudine granules were blended with colloidal silicon dioxide and sodium starch glycolate and lubricated with magnesium stearate.

Compression and coating

(1) The granules were compressed into the monolayer tablet and coated with Opadry.

EXAMPLE 24

Darunavir Ritonavir Raltegravir bilayer tablets

Darunavir Layer

Sr. No.	Ingredients	Quantity / Unit (mg)
I	**Dry Mix**	
1	Darunavir Ethanolate	432.87
2	Raltegravir Potassium	434.40
II	**Binder**	
3	Hydroxy propyl methyl cellulose	6.60
4	Purified water	q.s.
III	**Blending and lubrication**	
5	Silicified microcrystalline cellulose	52.50
6	Colloidal silicon dioxide	1.65
7	Crospovidone	16.50
8	Magnesium Stearate	2.75
	Total weight of Layer I	**947.27**
Ritonavir Part		
I	**Drug premix**	
1	Ritonavir	100.00
2	Colloidal silicon dioxide	6.90
3	Dibasic calcium phosphate	45.00
II	**Polymer premix**	
4	Copovidone	293.10
5	Sorbitan monolaurate	66.70
III	**Blending and lubrication**	
6	Colloidal silicon dioxide	10.00
7	Dibasic calcium phosphate anhydrous	44.40
8	Sodium stearyl fumarate	3.90
	Total weight of Layer II	**570.00**
	Total weight of core tablet (Layer I + Layer II)	**1517.27**

	Film Coating	
9	Opadry Red	**36.00**
10	Purified water	-
	Total weight of film coated tablet	**1553.27**

Process

Manufacturing Process for Darunavir Layer:

1. Darunavir, Raltegravir, HPMC, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide were sifted, mixed and granulated.
2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted, dry mixed and granulated with copovidone and sorbitan monolaureate water to form granules.
2. The granules were extruded and lubricated with sodium stearyl fumarate.
3. The lubricated granules obtained in step (2) of the dolutegravir layer and the step (2) of the darunavir layer and the step (2) of the ritonavir layer were compressed to form a bilayer tablet and coated with opadry red.

EXAMPLE 25

Darunavir Ritonavir Raltegravir bilayer tablets

Darunavir Layer

Sr. No.	Ingredients	Quantity / Unit (mg)
I	**Dry Mix**	
1	Darunavir Ethanolate	432.87
2	Raltegravir Potassium	434.40
II	**Binder**	
3	Hydroxy propyl methyl cellulose	6.60
4	Purified water	q.s.
III	**Blending and lubrication**	
5	Silicified microcrystalline cellulose	52.50
6	Colloidal silicon dioxide	1.65
7	Crospovidone	16.50
8	Magnesium Stearate	2.75
	Total weight of Layer I	**947.27**
Ritonavir Part		
I	**Drug premix**	
1	Ritonavir	50.00
2	Colloidal silicon dioxide	6.90

3	Dibasic calcium phosphate	45.00
II	**Polymer premix**	
4	Copovidone	293.10
5	Sorbitan monolaurate	66.70
III	**Blending and lubrication**	
6	Colloidal silicon dioxide	10.00
7	Dibasic calcium phosphate anhydrous	94.40
8	Sodium stearyl fumarate	3.90
	Total weight of Layer II	**570.00**
	Total weight of core tablet (Layer I + Layer II)	**1517.27**
	Film Coating	
9	Opadry Red	**36.00**
10	Purified water	-
	Total weight of film coated tablet	**1553.27**

Process

Manufacturing Process for Darunavir Layer:

1. Darunavir, Raltegravir, HPMC, silicified microcrystalline cellulose, crospovidone and colloidal silicon dioxide were sifted, mixed and granulated.
2. The granules obtained in step (1) were lubricated with magnesium stearate.

Manufacturing Process for Ritonavir Layer:

1. Ritonavir, colloidal silicon dioxide and dibasic calcium phosphate were sifted, dry mixed and granulated with copovidone and sorbitan monolaureate water to form granules.
2. The granules were extruded and lubricated with sodium stearyl fumarate.
3. The lubricated granules obtained in step (2) of the dolutegravir layer and the step (2) of the darunavir layer and the step (2) of the ritonavir layer were compressed to form a bilayer tablet and coated with opadry red.

It will be readily apparent to one skilled in the art that varying substitutions and modifications may be made to the invention disclosed herein without departing from the spirit of the invention. Thus, it should be understood that although the present invention has been specifically disclosed by the preferred embodiments and optional features, modification and variation of the concepts herein disclosed may be resorted to by those skilled in the art, and such modifications and variations are considered to be falling within the scope of the invention.

It is to be understood that the phraseology and terminology used herein is for the purpose of description and should not be regarded as limiting. The use of “including,” “comprising,” or “having” and variations thereof herein is meant to encompass the items listed thereafter and equivalents thereof as well as additional items.

It must be noted that, as used in this specification and the appended claims, the singular forms "a," "an" and "the" include plural references unless the context clearly dictates otherwise. Thus, for example, reference to "a propellant" includes a single propellant as well as two or more different propellants; reference to a "cosolvent" refers to a single cosolvent or to combinations of two or more cosolvents, and the like.

It will be appreciated that the invention may be modified within the scope of the appended claims.

CLAIMS:

1. A pharmaceutical antiretroviral composition comprising:

(i) at least one reverse transcriptase inhibitor comprising: zidovudine; didanosine; stavudine; lamivudine; abacavir; adefovir; lobucavir; entecavir; apricitabine; emtricitabine; zalcitabine; dexelvucitabine; alovudine; amdoxovir; elvucitabine; tenofovir; festinavir; racivir; lersivirine; rilpivirine; etravirine; SP1093V and/or stampidine, and/or

(ii) at least one integrase inhibitor comprising: raltegravir; dolutegravir and/or elvitegravir, and

(iii) at least one protease inhibitor comprising: saquinavir; ritonavir; nelfinavir; amprenavir; lopinavir, indinavir; nelfinavir; atazanavir; lasinavir; palinavir; fosamprenavir; darunavir and/or tiprinavir,

wherein the pharmaceutical antiretroviral composition optionally further comprises one or more pharmaceutically acceptable excipients.

2. The pharmaceutical antiretroviral composition according claim 1, wherein the zidovudine; didanosine; stavudine; lamivudine; abacavir; adefovir; lobucavir; entecavir; apricitabine; emtricitabine; zalcitabine; dexelvucitabine; alovudine; amdoxovir; elvucitabine; tenofovir; festinavir; racivir; lersivirine; rilpivirine; etravirine; SP1093V; stampidine; raltegravir elvitegravir; dolutegravir; saquinavir; ritonavir; nelfinavir; amprenavir; lopinavir; indinavir; nelfinavir; atazanavir; lasinavir; palinavir; fosamprenavir; darunavir; and/or tiprinavir is in the form of a pharmaceutically acceptable derivative thereof.

3. The pharmaceutical antiretroviral composition according to claim 2, wherein the pharmaceutically acceptable derivative is a salt, solvate, complex, hydrate, isomer, ester, tautomer, anhydrate, enantiomer, polymorph or prodrug.

4. The pharmaceutical antiretroviral composition according to any preceding claim wherein the reverse transcriptase inhibitor comprises: lamivudine; zidovudine; tenofovir; emtricitabine and/or abacavir.

5. The pharmaceutical antiretroviral composition according to any preceding claim wherein the integrase inhibitor comprises: raltegravir and/or dolutegravir.

6. The pharmaceutical antiretroviral composition according to any preceding claim wherein the protease inhibitor comprises: ritonavir and/or darunavir.

7. The pharmaceutical antiretroviral composition according to any preceding claim comprising lamivudine, zidovudine, darunavir and ritonavir.

8. The pharmaceutical antiretroviral composition according to any preceding claim comprising lamivudine, tenofovir, darunavir and ritonavir.

9. The pharmaceutical antiretroviral composition according to any preceding claim comprising tenofovir, emtricitabine, darunavir and ritonavir.

10. The pharmaceutical antiretroviral composition according to any preceding claim comprising abacavir, lamivudine, darunavir and ritonavir.

11. The pharmaceutical antiretroviral composition according to any preceding claim comprising raltegravir or dolutegravir, darunavir and ritonavir.

12. The pharmaceutical antiretroviral composition according to any preceding claim, comprising:

at least two reverse transcriptase inhibitors, wherein the at least two reverse transcriptase inhibitors are provided in a separate single unit dosage form;

at least two protease inhibitors, wherein the at least two protease inhibitors are provided in a separate single unit dosage form; and/or

at least one integrase inhibitor, wherein the at least one integrase inhibitor is provided in a separate single unit dosage form.

13. The pharmaceutical antiretroviral composition according to any preceding claim comprising darunavir and ritonavir provided in a separate single unit dosage form.

14. The pharmaceutical antiretroviral composition according to any preceding claim comprising raltegravir or dolutegravir provided in a separate single unit dosage form.

15. The pharmaceutical antiretroviral composition according to any preceding claim comprising lamivudine and zidovudine provided in a separate single unit dosage form.

16. The pharmaceutical antiretroviral composition according to any preceding claim comprising tenofovir and emtricitabine provided in a separate single unit dosage form.

17. The pharmaceutical antiretroviral composition according to any preceding claim comprising abacavir and lamivudine provided in a separate single unit dosage form.

18. The pharmaceutical antiretroviral composition according to any preceding claim comprising lamivudine and tenofovir provided in a separate single unit dosage form.

19. The pharmaceutical antiretroviral composition according to any preceding claim, wherein the composition is provided as a kit comprising instructions for administration.

20. The pharmaceutical antiretroviral composition according to any preceding claim for once or twice daily administration.

21. The pharmaceutical antiretroviral composition according to any preceding claim, wherein the at least one reverse transcriptase inhibitor, the at least one integrase inhibitor and the at least one protease inhibitor are provided in a dosage form selected from: a tablet, a mini-tablet, sprinkles comprising a plurality of particles, a capsule, or a liquid.

22. The pharmaceutical antiretroviral composition according to claim 21, wherein the tablet is a disintegrating tablet, a dissolving tablet, a dispersible tablet, a mouth-dissolving tablet, a tablet for oral suspension, an immediate release tablet, an extended release tablet, an immediate and extended release tablet, or a matrix tablet.

23. The pharmaceutical antiretroviral composition according to claim 21, wherein the plurality of particles of the sprinkles are provide in the form of granules, powders, powders for reconstitution, beads, pellets, mini-tablets, film-coated tablets, film coated tablets MUPS, orally disintegrating MUPS, pills, micro-pellets, small tablet units, MUPS, disintegrating tablets, dispersible tablets, granules, effervescent granules, or microspheres.

24. The pharmaceutical antiretroviral composition according to claim 21 or 23, wherein the sprinkles are provided in a sachet, a packet or a capsule.

25. The pharmaceutical antiretroviral composition according to any preceding claim, wherein the one or more pharmaceutically acceptable excipient comprises: a diluent, filler, bulking agent, disintegrant, binder, glidant, anti-adherent, lubricant, water soluble polymer, water insoluble polymer, water swellable polymer, plasticizer, and any mixture thereof.

26. A process for preparing the pharmaceutical antiretroviral composition according to any preceding claim, comprising: admixing the at least one reverse transcriptase inhibitor or the at least one integrase inhibitor and the at least one protease inhibitor, optionally with the one or more pharmaceutically acceptable excipient.

27. A method of preventing, treating or prophylaxis of a disease caused by a retrovirus, specifically acquired immune deficiency syndrome or an HIV infection, which method comprises administering to a patient in need thereof a pharmaceutical antiretroviral composition according to any one of claims 1 to 25.

28. A pharmaceutical composition according to any one of claims 1 to 25 for use in the treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection, in a patient in need thereof.

29. A use of the pharmaceutical antiretroviral composition according to any one of claims 1 to 25 for the treatment or prophylaxis of diseases caused by retroviruses, specifically acquired immune deficiency syndrome or an HIV infection.

30. The pharmaceutical antiretroviral composition as substantially described herein with reference to the examples.

31. The process of manufacturing a pharmaceutical antiretroviral composition as substantially described herein with reference to the examples.

INTERNATIONAL SEARCH REPORT

International application No
PCT/GB2014/051478

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61K31/34 A61K31/427 A61K31/513 A61K31/535 A61P31/18
ADD.

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)
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 practicable, search terms used)
EPO-Internal, BIOSIS, EMBASE, FSTA, INSPEC, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2004/087139 A1 (BOEHRINGER INGELHEIM INT [DE]; KLAES HEINZ-GERD [DE]; MAYERS DOUGLAS L) 14 October 2004 (2004-10-14) tables 1-7	1-4,6,7, 12,19-31
X	WO 2013/057469 A1 (CIPLA LTD [IN]; MALHOTRA GEENA [IN]; PURANDARE SHRIVINAS [IN]; KING LA) 25 April 2013 (2013-04-25) claims 1-5	1-4,6, 12,15, 19-31
X	WO 2006/005720 A1 (TIBOTEC PHARM LTD [IE]; HOETELMANS RICHARD MARINUS WIL [NL]) 19 January 2006 (2006-01-19) claims 1,8	1-4,6, 13,19-31

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[X] Further documents are listed in the continuation of Box C.

[X] See patent family 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 application or patent 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
9 July 2014	09/10/2014

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, Fax: (+31-70) 340-3016	Baurand, Petra

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Form PCT/ISA/210 (second sheet) (April 2005)

INTERNATIONAL SEARCH REPORT

International application No
PCT/GB2014/051478

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	MATTIOLI BENEDETTA ET AL: "Effect of indinavir used alone or in double or triple combination with AZT and ddC on human immune functions.", LIFE SCIENCES, vol. 74, no. 18, 19 March 2004 (2004-03-19), pages 2291-2300, XP002726856, ISSN: 0024-3205 page 2291, abstract	1-4,21, 25,26, 30,31
X	GHOSN JADE ET AL: "Absence of HIV-1 shedding in male genital tract after 1 year of first-line lopinavir/ritonavir alone or in combination with zidovudine/lamivudine", JOURNAL OF ANTIMICROBIAL CHEMOTHERAPY, vol. 61, no. 6, June 2008 (2008-06), pages 1344-1347, XP002726857, ISSN: 0305-7453 page 1344, abstract	1-4,6, 12,15, 19-31
X	VITHAYASAI VICHARN ET AL: "Safety and efficacy of saquinavir soft-gelatin capsules + zidovudine + optional lamivudine in pregnancy and prevention of vertical HIV transmission", JAIDS JOURNAL OF ACQUIRED IMMUNE DEFICIENCY SYNDROMES, vol. 30, no. 4, 1 August 2002 (2002-08-01) , pages 410-412, XP002726858, ISSN: 1525-4135 page 410, abstract	1-4,15, 19-31
X	MOOLASART PIKUL ET AL: "The efficacy of combined zidovudine and lamivudine compared with that of combined zidovudine, lamivudine and nelfinavir in asymptomatic and early symptomatic HIV-infected children", SOUTHEAST ASIAN JOURNAL OF TROPICAL MEDICINE AND PUBLIC HEALTH, vol. 33, no. 2, June 2002 (2002-06), pages 280-287, XP002726859, ISSN: 0125-1562 page 280, abstract	1-4, 19-31

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INTERNATIONAL SEARCH REPORT

International application No.
PCT/GB2014/051478

Box No. II Observations where certain claims were found unsearchable (Continuation of item 2 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☐ Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

2. ☐ Claims Nos.:
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 No. III Observations where unity of invention is lacking (Continuation of item 3 of first sheet)

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

see additional sheet

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 fees, this Authority did not invite payment of additional fees.

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.:

1-4, 6, 7, 12, 13, 15, 19-31(all partially)

Remark on Protest

☐ The additional search fees were accompanied by the applicant's protest and, where applicable, the payment of a protest fee.

☐ The additional search fees were accompanied by the applicant's protest but the applicable protest fee was not paid within the time limit specified in the invitation.

☐ No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. claims: 1-4, 6, 7, 12, 13, 15, 19-31(all partially)

   Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor zidovudine and (iii) at least one protease inhibitor

   ---

2. claims: 1-3, 6, 12, 13, 19-31(all partially)

   Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor didanosine and (iii) at least one protease inhibitor

   ---

3. claims: 1-3, 6, 12, 13, 19-31(all partially)

   Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor stavudine and (iii) at least one protease inhibitor

   ---

4. claims: 1-4, 6-8, 10, 12, 13, 15, 17-31(all partially)

   Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor lamivudine and (iii) at least one protease inhibitor

   ---

5. claims: 1-4, 6, 10, 12, 13, 17, 19-31(all partially)

   Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor abacavir and (iii) at least one protease inhibitor

   ---

6. claims: 1-3, 6, 12, 13, 19-31(all partially)

   Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor adefovir and (iii) at least one protease inhibitor

   ---

7. claims: 1-3, 6, 12, 13, 19-31(all partially)

   Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor lobucavir and (iii) at least one protease inhibitor

   ---

8. claims: 1-3, 6, 12, 13, 19-31(all partially)

   Pharmaceutical composition comprising (i) the reverse

transcriptase inhibitor entecavir and (iii) at least one protease inhibitor

---

9. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor apricitabine and (iii) at least one protease inhibitor

---

10. claims: 1-4, 6, 9, 12, 13, 16, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor emtricitabine (racivir) and (iii) at least one protease inhibitor

---

11. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor zalcitabine and (iii) at least one protease inhibitor

---

12. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor dexelvucitabine (elvucitabine) and (iii) at least one protease inhibitor

---

13. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor alovudine and (iii) at least one protease inhibitor

---

14. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor amdoxovir and (iii) at least one protease inhibitor

---

15. claims: 1-4, 6, 8, 9, 12, 13, 16, 18-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor tenofovir and (iii) at least one protease inhibitor

---

16. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor festinavir and (iii) at least one protease inhibitor

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17. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor lersivirine and (iii) at least one protease inhibitor

---

18. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor rilpivirine and (iii) at least one protease inhibitor

---

19. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor etravirine and (iii) at least one protease inhibitor

---

20. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor SP1093V and (iii) at least one protease inhibitor

---

21. claims: 1-3, 6, 12, 13, 19-31(all partially)

Pharmaceutical composition comprising (i) the reverse transcriptase inhibitor stampidine and (iii) at least one protease inhibitor

---

22. claims: 5, 11, 14(completely); 1-3, 6, 12, 13, 19-31(partially)

Pharmaceutical composition comprising (ii) at least one integrase inhibitor and (iii) at least one protease inhibitor

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INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No
PCT/GB2014/051478

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 2004087139 A1	14-10-2004	US 2004235779 A1	25-11-2004
		WO 2004087139 A1	14-10-2004
WO 2013057469 A1	25-04-2013	NONE	
WO 2006005720 A1	19-01-2006	AT 406161 T	15-09-2008
		AU 2005261701 A1	19-01-2006
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		SI 1765337 T1	28-02-2009
		US 2007208009 A1	06-09-2007
		WO 2006005720 A1	19-01-2006
		ZA 200700176 A	30-04-2008

Tenofovir Alafenamide Vs. Tenofovir Disoproxil Fumarate in Single Tablet Regimens for Initial HIV-1 Therapy: A Randomized Phase 2 Study

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Objectives: To evaluate the safety and efficacy of the novel tenofovir prodrug, tenofovir alafenamide (TAF), as part of a single-tablet regimen (STR) for the initial treatment of HIV-1 infection.

Design: Phase 2, randomized, double-blind, double-dummy, multicenter, active-controlled study.

Methods: Antiretroviral naive adults with HIV-1 RNA ≥5000 copies per milliliter and a CD4 count ≥50 cells per microliter were randomized 2:1 to receive an STR of elvitegravir/cobicistat/emtricitabine/tenofovir alafenamide (E/C/F/TAF) or elvitegravir/cobicistat/emtricitabine/tenofovir disoproxil fumarate (E/C/F/TDF), plus placebo for 48 weeks.

Results: Patients on both E/C/F/TAF (n = 112) and E/C/F/TDF (n = 58) had high rates of virologic suppression (<50 HIV copies per milliliter) at week 24 (86.6%; 89.7%) and at week 48 (88.4%; 87.9%), and had similar improvements in CD4 at week 48 (177; 204), respectively. Both treatments were well tolerated, and most adverse events were self-limiting and of mild to moderate severity. Compared with patients on E/C/F/TDF, patients on E/C/F/TAF had smaller reductions in estimated creatinine clearance (−5.5 vs. −10.1 mL/min, $P = 0.041$), significantly less renal tubular proteinuria, and smaller changes in bone mineral density for hip (−0.62% vs. −2.39%, $P < 0.001$) and spine (−1.00% vs. −3.37%, $P < 0.001$). Patients on E/C/F/TAF had higher increases in total cholesterol, low-density lipoprotein, and high-density lipoprotein, but the total cholesterol/high-density lipoprotein ratio was unchanged for both.

Conclusions: Treatment-naive patients given the STR that contained either TAF or TDF achieved a high rate of virologic success. Compared with those receiving TDF, patients on E/C/F/TAF experienced significantly smaller changes in estimated creatinine clearance, renal tubular proteinuria, and bone mineral density.

Key Words: tenofovir alafenamide, GS-7340, elvitegravir, stribild, clinical trials

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INTRODUCTION

Currently available antiretroviral regimens have led to marked declines in the morbidity and mortality of patients living with HIV-1[1–3] and decreased risk of HIV-1 transmission.[4–6] This success has shifted clinical attention toward antiretroviral drug regimens that optimize tolerability, long-term safety, and durable efficacy. Morbidity and mortality are increasingly driven by non-AIDS associated comorbidities, which are observed earlier than in age-matched controls, despite durable suppression with the best available antiretroviral therapy (ART).[1,2,7–10] Current guidelines recommend that patients begin ART earlier and stay on it continuously,[11] so the contribution of specific antiretroviral agents to long-term morbidity and mortality is increasingly important. In regimens of comparable efficacy, pill burden, dose frequency, safety, and tolerability are significant factors affecting maximal adherence over the long term.[12–14] Single-tablet regimens (STRs) represent a simple and convenient way for patients to maximize adherence and to control their HIV for many years.

Current Department of Health and Human Services guidelines recommend tenofovir disoproxil fumarate (TDF) as a preferred component of the nucleotide reverse-

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P.E.S., A.Z., I.B., R.E., and R.O. enrolled patients and reviewed and interpreted analyses of data. S.M., M.W.F., C.C., and H.W. designed the study. Data collection was overseen by H.W., C.C., H.M., M.W.F., and S.M. H.W. analyzed data, which were reviewed and interpreted by C.C., H.M., M.F., and S.M. The first draft of this report was written by P.E.S., H.M., M.F., and S.M. The draft report was edited by P.E.S., A.Z., I.B., R.E., and R.O.

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Abstract presented by Dr Paul Sax, during 53rd Annual ICAAC Conference, September 10–13, 2013, Denver, CO.

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transcriptase inhibitor (NRTI) backbone for HIV-1–positive treatment-naive patients.[11] Despite a favorable safety and tolerability profile, TDF has been associated with nephrotoxicity,[15,16] requires dose adjustment as creatinine clearance falls <50 mL/min,[17] and has been shown to result in a greater decline in bone mineral density (BMD) relative to some other NRTIs.[18,19]

Tenofovir (TFV) is a nucleotide analog HIV-1 reverse transcriptase inhibitor. TDF, the first-generation prodrug of TFV, undergoes rapid metabolism in the plasma after oral administration.[20] TFV is then distributed intracellularly, where it is phosphorylated to the active moiety TFV diphosphate (TFV-DP). TFV alafenamide (TAF, formerly GS-7340) is a next-generation oral prodrug of TFV that may offer improved safety and efficacy. Relative to TDF, TAF is more stable in plasma and is predominantly metabolized intracellularly to TFV by cathepsin A.[20–22] This intracellular drug metabolism results in higher intracellular levels of the active metabolite TFV-DP and lower plasma levels of TFV, relative to TDF.[20,21]

Both nonhuman primate studies and human clinical studies have shown a relationship between plasma TFV levels and renal toxicity.[23–25] Because TFV (but not TAF) actively enters renal tubular cells via organic anion transporters 1 and 3, the reduced TFV levels that occur with TAF may be clinically manifest as reduced nephrotoxicity.[26,27] The higher intracellular TFV-DP levels may result in improved antiviral potency. In a 10-day monotherapy study in HIV-1–positive patients, those who received 25 mg of TAF had an approximately 0.5 $\log_{10}$ greater decline in plasma HIV-1 RNA than did patients who received the standard 300-mg dose of TDF.[28] In vitro, higher intracellular TFV-DP levels enable TAF to retain activity against viruses that have reduced susceptibility to TDF,[29] suggesting the potential use of TAF in a broader range of patients.

Because cobicistat increases the bioavailability of TAF by approximately 2.2-fold via the inhibition of P-glycoprotein intestinal secretion, the 10-mg dose of TAF delivered by the E/C/F/TAF STR is equivalent to the 25-mg dose of TAF.[28,30,31] To confirm the antiviral activity and safety profile of TAF compared with that of TDF, we conducted a randomized, double-blind Phase 2 clinical trial of 2 STRs—elvitegravir 150 mg, cobicistat 150 mg, emtricitabine 200 mg, and TAF 10 mg (E/C/F/TAF) compared with elvitegravir 150 mg, cobicistat 150 mg, emtricitabine 200 mg, and TDF 300 mg (E/C/F/TDF), licensed as Stribild (Gilead Sciences, Foster City, CA). The primary objective of this study, GS-292-0102, was to evaluate the efficacy and safety of TAF relative to TDF, both as part of an elvitegravir-based STR in treatment-naive patients at 24 weeks, with a particular focus on virologic, renal, and bone endpoints. Here, we report the efficacy and safety data from this study through 48 weeks.

METHODS

Study Design

Study 292-0102 is an ongoing randomized, double-blind, double dummy, active controlled, Phase 2 study being conducted in the United States that was approved by the US Food and Drug Administration and by institutional review boards at all sites. HIV-positive treatment-naive adults were considered eligible if they were ≥18 years of age with a plasma HIV-1 RNA ≥5000 copies per milliliter, a $CD4^+$ cell count >50 cells per microliter, an HIV-1 genotype showing sensitivity to TFV and emtricitabine (FTC), and an estimated glomerular filtration rate (eGFR; Cockcroft–Gault) of ≥70 mL/min. Patients were excluded if they were hepatitis B or C coinfected, had a new AIDS-defining condition within 30 days of screening, or were pregnant. The study was conducted from December 2011 through April 2013 (week 48 endpoint) and is posted on clinicaltrials.gov (NCT01497899).

Eligible participants were randomized centrally by a third party interactive voice/web response system, stratified by screening HIV-1 RNA (≤ or >,100,000 copies per milliliter), in a 2:1 fashion to receive treatment with either E/C/F/TAF or E/C/F/TDF administered once daily with food; all patients also received matching placebo tablets.

Randomized patients were seen at screening, baseline, and at weeks 2, 4, 8, 12, 16, and then every 8 weeks through week 48. Laboratory analyses (hematology, serum chemistries, $CD4^+$ cell count, and urinalysis; Covance Laboratories, Indianapolis, IN), HIV-1 RNA (TaqMan 2.0; Roche Diagnostics, Indianapolis, IN), and physical examinations were performed at all visits. HIV-1 genotype (reverse transcriptase and protease) was tested at screening (GenoSure MG, Monogram Biosciences, South San Francisco, CA). Any patient with confirmed virologic failure (2 consecutive viral load samples >50 copies/mL) and an HIV RNA >400 copies/mL at week 8 or later had the second, confirmatory, sample sent for resistance analysis by GeneSeq Integrase, PhenoSense GT, and PhenoSense Integrase (Monogram Biosciences).

Trough pharmacokinetic (PK) samples were collected at weeks 8, 24, and 48 visits, and population PK samples were collected at weeks 2, 4, 12, 16, 24, and 40. An intensive PK substudy was performed on a subset of patients at week 4 or 8 and included peripheral blood mononuclear cell (PBMC) sampling for intracellular TFV-DP levels.

Dual energy x-ray absorptiometry (DEXA) was used to measure BMD only at the hip and lumbar spine before study drug administration at baseline and every 24 weeks. These were read centrally by BioClinica (Newtown, PA), with investigators and patients blinded to the results. Patients were scanned with the same machine throughout the study, and phantom scans were used for quality assurance across sites. Blood and urine for selected bone and renal biomarkers were collected and analyzed at baseline and weeks 24 and 48.

Statistical Methods

The primary objective was to determine the efficacy of a regimen containing E/C/F/TAF vs. E/C/F/TDF in HIV-1 treatment-naive adults at week 24 (primary endpoint) and week 48 (secondary endpoint) according to US Food and Drug Administration snapshot analysis (the proportion of patients with HIV-1 RNA <50 copies per milliliter).[32] The sample size of 150 patients (100 in the E/C/F/TAF arm) was chosen to estimate the response rate of HIV-1 RNA <50

copies per milliliter at week 24 and to have 76% power to detect a 1.5% (standard deviation of 3.2%) difference in hip BMD in the E/C/F/TAF arm relative to the E/C/F/TDF treatment group. The safety and tolerability of the 2 treatment arms through 48 weeks of treatment were assessed as secondary endpoints.

An individual patient was considered a treatment success if he or she had HIV-1 RNA <50 copies per milliliter at week 24 without virologic failure (confirmed >50 copies per milliliter in week 24 window). These criteria were also used for the week 48 secondary analysis. Safety analyses included available data from all participants who consented to participate, and who received at least 1 dose of study medication; patients who discontinued were followed up for 30 days after drug discontinuation. Demographic and baseline characteristics were summarized using standard descriptive methods.

RESULTS

Of 233 patients screened, 171 were randomized, and 170 received at least 1 dose of study drug (E/C/F/TAF, n = 112, E/C/F/TDF, n = 58). Baseline characteristics are outlined in Table 1. Patients were primarily male, and approximately 30% were black or African American; most had no prior AIDS-defining condition. The median viral load at baseline was 4.6 $\log_{10}$ copies per milliliter, and the median $CD4^+$ cell count was 391 cells per microliter. Overall, 21% had baseline HIV-1 RNA >100,000 copies per milliliter, and 15% had a baseline CD4 count <200 cells per microliter (E/C/F/TAF, 12.5%; E/C/F/TDF, 18.9%). The median eGFR at baseline was 114 mL/min. Through week 48, 4 patients in the E/C/F/TAF arm and 0 in the E/C/F/TDF arm discontinued due to adverse events; 3 of the discontinuations due to unrelated illness (coxsackie infection, dual cytomegalovirus/*Mycobacterium avium* infection on day 13, acute promyelocytic leukemia) and 1 due to flushing that was considered drug related.

At week 24, virologic success was attained in 87.5% of those on the E/C/F/TAF arm and 89.7% for those receiving E/C/F/TDF [weighted difference −3.7%, 95% confidence interval −14.4 to 7.0, $P = 0.48$]. At week 48, there was virologic success in 88.4% (99/112) of patients who received E/C/F/TAF and 87.9% (51/58) who received E/C/F/TDF (weighted difference −1.0%, 95% confidence interval −12.1 to 10.0, $P = 0.84$). An analysis of virologic response using different virologic endpoints (missing = failure) demonstrated that 79% of patients in the E/C/F/TAF arm had HIV-1 RNA <20 copies per milliliter compared with 74% on E/C/F/TDF ($P = 0.56$), whereas no RNA signal was detectable in 59% of those on E/C/F/TDF vs. 53% on E/C/F/TDF. Median adherence to study treatment was equivalent in the 2 treatment arms (98%). Two patients in the E/C/F/TDF arm and none in the E/C/F/TAF arm discontinued their treatment due to the loss of efficacy. The mean $CD4^+$ cell count increase from baseline was 177 cells per microliter in the E/C/FTAF arm vs. 204 cells per microliter in the E/C/F/TDF arm ($P = 0.41$).

Six patients, 3 in each treatment arm (3/112 = 2.7% for E/C/F/TAF, 3/58 = 5.2% for E/C/F/TDF), met the criteria for virologic resistance testing. No resistance was detected in the E/C/F/TAF arm.

Resistance was detected in 2 patients in the E/C/F/TDF arm: 1 developed NRTI resistance with M184V and K70E and 1 developed both NRTI and Integrase Strand Transfer Inhibitor (INSTI) resistance with M184V and E92Q.

In patients in the intensive PK substudy (n = 26), plasma TFV exposure was 91% lower for patients taking E/C/F/TAF than for patients taking E/C/F/TDF, as measured by AUC_{tau}. Conversely, intracellular TFV-DP levels in PBMCs were 5.3-fold higher for patients in the E/C/F/TAF arm.

The 48-week safety profile of E/C/F/TAF was generally similar to that of E/C/F/TDF, with 94.6% (106) vs. 94.8% (55) patients reporting any treatment-emergent adverse event, and 9.8% (11) vs. 5.2% (3) reporting a grade 3 or 4 adverse event. The most common treatment-emergent adverse events were nausea and diarrhea, where nausea was reported in 21% on E/C/F/TAF and 12% of those on E/C/F/TDF. Of the 23 patients reporting nausea in the E/C/F/TAF treatment arm, 18 of these were grade 1, and 5 were grade 2, 15 resolved within 2 weeks, and none led to treatment discontinuation. A total of 7% in each arm reported vomiting. Diarrhea was reported in 16% in each treatment arm. No patient in either arm discontinued treatment due to any of these gastrointestinal events. Adverse events reported in at least 5% of participants in either arm are shown in Table 2.

All postbaseline grade 3 or 4 laboratory abnormalities are shown in Table 3. Grade 3 or 4 low-density lipoprotein (LDL) cholesterol elevations were more common in the E/C/F/TAF than in the E/C/F/TDF arm (9% vs. 3%). Fasting metabolic assessments showed that the median increase in LDL cholesterol was 17 mg/dL for E/C/F/TAF vs. 11 mg/dL for E/C/F/TDF ($P = 0.11$). There were statistically significant differences between groups in the median changes in total cholesterol (30 vs. 17 mg/dL, $P = 0.007$) and high-density lipoprotein (HDL) cholesterol (7 vs. 3 mg/dL, $P = 0.023$), whereas total cholesterol/HDL cholesterol ratio remained unchanged (median increase 0.2 vs. 0.1, $P = 0.34$). However, categorical analysis at week 48 by National Cholesterol Education Program Adult Treatment Panel III[33] classification showed no differences between the 2 treatment arms for total cholesterol ($P = 0.54$) or for LDL cholesterol

TABLE 1. Baseline Characteristics

Characteristic	E/C/F/TAF (n = 112)	E/C/F/TDF (n = 58)
Age (yrs), median	34	38
Male (%)	96	98
Race (%)		
White	67	69
Black/African descent	30	28
Hispanic/Latino	22	19
Asymptomatic HIV infection (%)	88	91
HIV-1 RNA ($\log_{10}$ copies/mL), median	4.55	4.58
>100,000 copies/mL	17%	28%
CD4 cell count (cells/mm^3), median	385	397
≤ 200 cells/mm^3	13%	19%
Estimated GFR, median, (Cockcroft–Gault), mL/min	115	113

TABLE 2. Adverse Events

Adverse Event (Any Grade) Occurring in At Least 5% of Patients, % (n)	E/C/F/TAF (n = 112)	E/C/F/TDF (n = 58)
Nausea	21% (23)	12% (7)
Diarrhea	16% (18)	16% (9)
Upper respiratory tract infection	15% (17)	21% (12)
Fatigue	14% (16)	9% (5)
Headache	10% (11)	14% (8)
Cough	10% (11)	10% (6)
Pharyngitis	8% (9)	3% (2)
Rash	8% (9)	5% (3)
Vomiting	7% (8)	7% (4)
Influenza	7% (8)	0
Bronchitis	6% (7)	5% (3)
Nasopharyngitis	6% (7)	3% (2)
Depression	6% (7)	3% (2)
Conjunctivitis	6% (7)	0
Anogenital warts	5% (6)	5% (3)
Abnormal dreams	5% (6)	2% (1)
Flatulence	5% (6)	3% (2)
Insomnia	4% (4)	7% (4)
Sinusitis	4% (5)	5% (3)
Seasonal allergies	3% (3)	5% (3)
Back pain	3% (3)	10% (6)
Paresthesia	3% (3)	7% (4)
Neck pain	3% (3)	5% (3)
Syphilis	3% (3)	5% (3)
Anxiety	2% (2)	9% (5)
Pain in extremities	1% (1)	5% (3)
Skin papilloma	0	5% (3)

($P = 0.37$), and there were no differences in change in triglycerides or serum glucose between treatment arms.

There was a rise in serum creatinine and consequent decline in creatinine clearance in both arms. Compared with baseline, the median change in serum creatinine at week 48 was 0.07 mg/dL for E/C/F/TAF vs. 0.10 mg/dL for E/C/F/TDF ($P = 0.077$), and the median change in eGFR by Cockcroft–Gault was −5.5 mL/min for E/C/F/TAF vs. −10.1 mL/min for E/C/F/TDF ($P = 0.041$). The median changes in serum creatinine generally occurred by week 4 for both treatment arms and then stabilized for the duration of the study.

TABLE 3. Grade 3 or 4 Laboratory Abnormalities

Maximum Toxicity Grade Postbaseline % (n)	E/C/F/TAF (n = 112)	E/C/F/TDF (n = 58)
Any grade 3 or grade 4 abnormality	25% (28)	17% (10)
LDL	9% (10)	3% (2)
Creatine phosphokinase	6% (7)	3% (2)
Neutropenia	5% (6)	2% (1)
Amylase	3% (3)	3% (2)
Urine RBC	2% (2)	0
Total cholesterol	2% (2)	0
ALT	1% (1)	2% (1)
AST	1% (1)	0
GGT	1% (1)	2% (1)
White blood cells	1% (1)	0
Hypophosphatemia	1% (1)	0
Urine protein	1% (1)	0
Glucose	1% (1)	2% (1)
Triglycerides	1% (1)	2%(1)

ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma glutamyl transpeptidase; RBC, red blood cells.

There were no clinically defined cases of proximal renal tubulopathy in either arm, and there were no treatment discontinuations due to laboratory or clinical renal events. Less proteinuria (urine protein/creatinine ratio) and albuminuria (urine albumin/creatinine ratio) were observed with E/C/F/TAF, but the differences were not statistically significant (Fig. 1). Renal tubular proteinuria [urine retinol-binding protein (RBP)/creatinine ratio and urine β-2 microglobulin/creatinine ratio] was significantly lower in patients who received E/C/F/TAF (Fig. 1).

Changes in BMD, expressed as the median percent change from baseline, are shown in Figure 2. There was significantly less change in the E/C/F/TAF arm in BMD as measured by using DEXA at both the hip (−0.62% vs. −2.39%, $P < 0.001$) and lumbar spine (−1.00% vs. −3.37%, $P < 0.001$) at week 48, which were also significant at week 24. In the E/C/F/TAF arm, 32% of the patients had no decrease seen in hip BMD vs. 7% in the E/C/F/TDF arm ($P < 0.001$), and no decrease at the lumbar spine was seen in 37% of the patients who received E/C/F/TAF vs. 11% who received E/C/F/TDF ($P < 0.001$). Conversely, a change in BMD >3% from baseline at the hip was observed 11.5% vs. 40.0% and at the lumbar spine in 24.8% vs. 55.3% (E/C/F/TAF vs. E/C/F/TDF, respectively, $P < 0.001$ for both). At weeks 24 and 48, markers of bone turnover were lower in patients on E/C/F/TAF than on E/C/F/TDF. At week 48, procollagen type 1 N-terminal propeptide, a marker of bone formation, increased 9% from baseline for E/C/F/TAF vs. 69% for E/C/F/TDF ($P < 0.001$), whereas C-terminal telopeptide (CTx), a marker of bone resorption, increased 19% from baseline for E/C/F/TAF vs. 78% for E/C/F/TDF ($P < 0.001$). There were no fragility fractures in either arm of the study.

DISCUSSION

TDF is a preferred NRTI in the initial therapy based on its favorable efficacy and safety data in randomized clinical trials and widespread use in clinical practice.[11] However, TDF may be associated with renal toxicity,[15,16] and comparative studies demonstrate that TDF treatment is linked to a greater loss in bone density as compared with other NRTI options.[18,34] Given the prolonged survival of patients with HIV with effective therapy, and the need for indefinite treatment, there is a need for an NRTI option that provides antiviral activity comparable with TDF with an improved safety profile.

In this Phase 2, randomized clinical trial, HIV-positive treatment-naive adults received STRs of E/C/F/TAF or E/C/F/TDF. Both E/C/F/TAF and E/C/F/TDF demonstrated high and comparable rates of virologic suppression, with expected rises in the $CD4^+$ cell count through 48 weeks of therapy. Both regimens were well tolerated, with few discontinuations due to adverse events. Although nausea occurred more

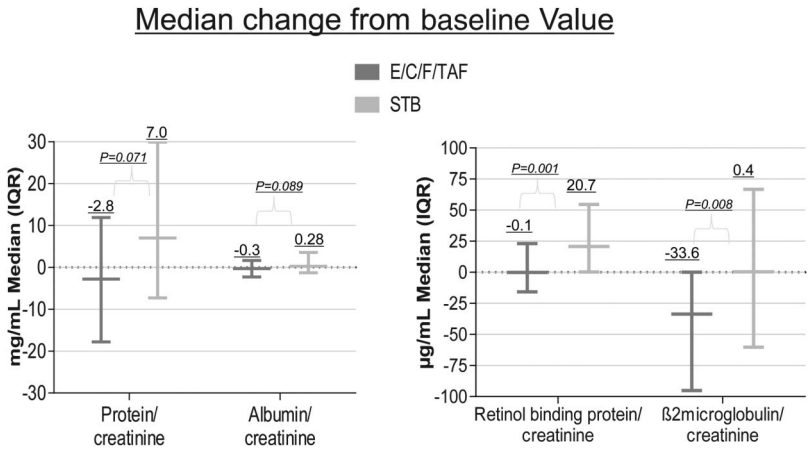
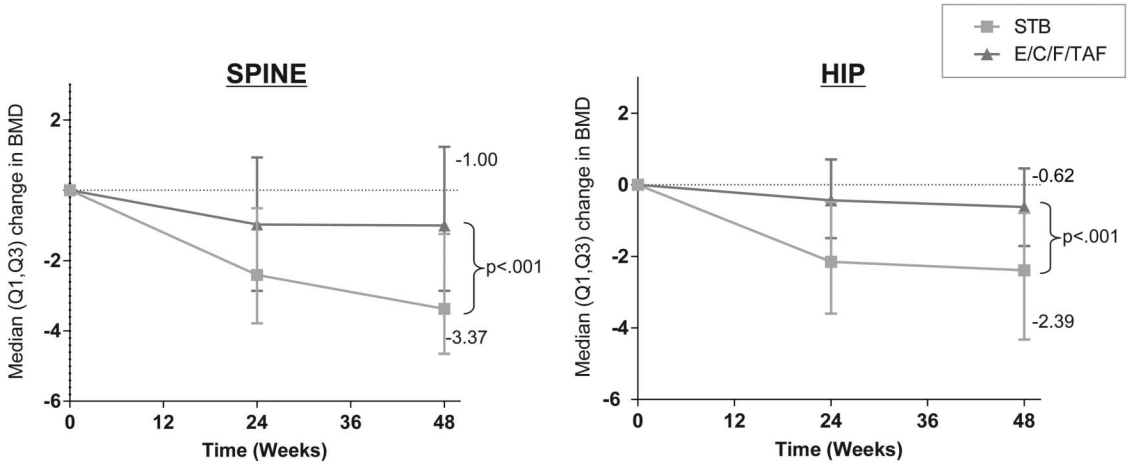


FIGURE 1. Urine tubular proteins: median change from the baseline value.

frequently in the E/C/F/TAF than in the E/C/F/TDF arm, the nausea reported was all grade 1 and grade 2 in severity, did not lead to drug discontinuation in any patient, and was comparable with the rate of nausea in the phase 3 studies of E/C/F/TDF.[35,36]

Because the 2 treatment regimens in this study differed only in whether patients received TAF or TDF, the study offered an opportunity to compare the pharmacokinetics, renal, and bone effects of the 2 prodrugs. Plasma concentrations of TFV were substantially (91%) lower with E/C/F/TAF than with E/C/F/TDF, and the TAF regimen delivered 5.3 times the intracellular, physiologically active metabolite, TFV-DP, to PBMCs, which could translate into less end-organ toxicity and/or improved virologic control. Although the effect of these differences did not translate into an observed significant difference in antiviral activity between the 2 regimens, the study had not been powered to demonstrate differences in virologic endpoints, and this is being further explored in the E/C/F/TAF Phase 3 program. In addition, no genotypic resistance emerged in the E/C/F/TAF group for the 3 patients with virologic failure and who met criteria for resistance analysis.

There were significant differences between the E/C/F/TAF and E/C/F/TDF treatment groups in specified renal, bone, and lipid endpoints. Early small increases in creatinine were seen in both arms, which were expected to be due to the known nonpathologic inhibitory effect of cobicistat on tubular creatinine secretion.[37] However, after week 2, patients on TAF had a lower magnitude increase in serum creatinine than did patients on TDF despite receiving the same other components of combination ART. The mechanism for this



W48 Median Value of Bone Biomarkers as % of Baseline: E/C/F/TAF vs. STB	
Procollagen Type 1 N-terminal propeptide (P1NP):	109% vs 169% (p<0.001)
C-terminal telopeptide (CTx):	119% vs. 178% (p<0.001)

FIGURE 2. Percent change in the spine and hip BMD as determined using DEXA.

difference is currently unknown but potentially may be related to the 91% lower plasma TFV exposure observed when TFV is delivered as TAF compared with TDF, because higher plasma TFV levels have been associated with an increased risk of renal impairment in other studies.[24,25] TFV in plasma is actively transported into the proximal renal tubular cell via organic anion transporter (OAT) 1 and OAT 3, but TAF is not a substrate for these transporters.[27] Thus, the intracellular concentration of TFV within proximal tubules is lower in patients treated with TAF compared with those treated with TDF, and decreased cytotoxicity of TAF in isolated human renal cells was recently demonstrated.[27]

The effect of TAF vs. TDF on proximal renal function was assessed using standard clinical measures of proteinuria and albuminuria, and markers of proximal renal tubular cell dysfunction, RBP, and β-2 microglobulin. These low molecular weight proteins are freely filtered at the glomerulus, and hence are almost entirely removed from the ultrafiltrate and catabolized in the proximal renal tubules.[38,39] The presence of these proteins in increased amounts in the urine may indicate subclinical renal tubular cell dysfunction.[38,39] In this study, urinary RBP/creatinine and β-2 microglobulin/creatinine ratios were significantly lower in the E/C/F/TAF arm, which suggests that TAF has a lesser effect than TDF on the proximal renal tubular cell. Whether this translates into long-term clinical benefit in renal function must be explored in larger studies with a longer follow-up. Nonetheless, these data are encouraging, as they demonstrate that TAF has a reduced effect on serum creatinine and is associated with reduced tubular proteinuria, both of which are important clinical markers of chronic kidney disease.

Patients with HIV have a lower BMD than age-matched HIV-uninfected controls, and they also experience higher fracture rates.[10,19] In addition, several studies have shown that TDF-containing regimens lead to a greater decline in bone density than in the case of comparator drugs.[18,34] In this study, patients who received E/C/F/TAF had smaller decreases in BMD through 48 weeks than those receiving E/C/F/TDF. It is noteworthy that the magnitude of BMD change that was observed for patients on the E/C/F/TAF arm was the lowest magnitude BMD change reported to date for treatment-naive study patients receiving NRTIs who had bone density assessed with bone DEXA scans. For example, the ASSERT study compared patients treated with TDF/FTC vs. ABC/3TC, each combined with efavirenz, and found a loss of BMD in both groups after the initiation of ART. However, there was a statistically greater loss of BMD at the hip and spine in patients treated with TDF/FTC than in those given ABC/3TC.[18] Although cross-study comparisons should only be made cautiously, the STR E/C/F/TAF demonstrated less loss of BMD than seen with ABC/3TC + EFV demonstrated in the ASSERT study (hip: −0.62% vs. −1.9%; spine: −1.0% vs. −1.6% for E/C/F/TAF and ABC/3TC + EFV, respectively). The DEXA results in this study were supported by changes in markers of bone turnover, with significantly less change in markers of bone formation (procollagen type 1 N-terminal propeptide) and bone resorption (CTX) among patients on E/C/F/TAF compared with those on E/C/F/TDF.

There were significantly greater increases in total and HDL cholesterol in the E/C/F/TAF than in the E/C/F/TDF study arm. By contrast, the total cholesterol/HDL ratio, triglycerides, and glucose were not significantly different, and there were no differences in National Cholesterol Education Program risk classification. The likely cause of these differences may relate to the previously reported lipid-lowering effect of TFV,[40,41] and the markedly lower plasma concentrations of TFV in the E/C/F/TAF compared with that in the E/C/F/TDF arm.

CONCLUSIONS

Treatment-naive patients given either TAF or TDF as part of an STR containing emtricitabine, cobicistat, and elvitegravir achieved a high rate of virologic suppression, with comparably low rates of adverse events and adverse event-related drug discontinuation in both arms. Nausea was more common in those receiving E/C/F/TAF than in those receiving E/C/F/TDF in this study, though it was mild and did not lead to study drug discontinuation. Pharmacokinetic data demonstrated that TAF delivers the parent drug TFV into PBMCs, where the active, phosphorylated metabolite, TFV-DP, achieves a concentration 5- to 7-fold higher than TDF, with 91% lower plasma TFV levels. The E/C/F/TAF-treatment group had a significantly higher eGFR and significantly less tubular proteinuria than E/C/F/TDF; further, changes in BMD significantly favored E/C/F/TAF. These promising results await confirmation in fully powered Phase 3 randomized controlled clinical trials comparing TAF with TDF, which are underway.

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GILEAD

European Medicines Agency Validates Gilead's Marketing Application for Fixed-Dose Combination of Emtricitabine and Tenofovir Alafenamide for HIV Treatment

– If Approved, Would Provide Potential New Backbone for Future HIV Therapy Combinations –

May 28, 2015 08:30 AM Eastern Daylight Time

FOSTER CITY, Calif.--(BUSINESS WIRE)--Gilead Sciences, Inc. (Nasdaq:GILD) today announced that the company's Marketing Authorization Application (MAA) for two doses of an investigational fixed-dose combination of emtricitabine and tenofovir alafenamide (200/10 mg and 200/25 mg) (F/TAF) has been fully validated and is now under evaluation by the European Medicines Agency (EMA). The data included in the application support the use of F/TAF for the treatment of HIV-1 infection in adults in combination with other HIV antiretroviral agents.

TAF is a novel investigational nucleotide reverse transcriptase inhibitor (NRTI) that has demonstrated high antiviral efficacy at a dose less than one-tenth that of Gilead's Viread® (tenofovir disoproxil fumarate, TDF), as well as improved renal and bone laboratory parameters as compared to TDF in clinical trials.

"Therapy innovations have transformed HIV into a chronic condition and people with HIV are living longer, necessitating new treatment options that deliver on both high efficacy and long-term safety," said Norbert Bischofberger, PhD, Executive Vice President, Research and Development and Chief Scientific Officer, Gilead Sciences. "F/TAF is the latest advance in Gilead's long history of innovating in HIV therapy and has the potential to become the backbone for the next generation of HIV regimens."

F/TAF is Gilead's second F/TAF-based regimen to be validated by the EMA. An MAA for an investigational once-daily single tablet regimen containing elvitegravir 150 mg, cobicistat 150 mg, emtricitabine 200 mg and tenofovir alafenamide 10 mg (E/C/F/TAF) was fully validated on December 23, 2014. In addition, Gilead filed New Drug Applications to the U.S. Food and Drug Administration for E/C/F/TAF and F/TAF on November 5, 2014, and April 7, 2015, respectively.

The MAA for F/TAF is supported by data from Phase 3 clinical studies evaluating the safety and efficacy of E/C/F/TAF for the treatment of HIV-1 infection among treatment-naïve adults, in which the F/TAF-based regimen (administered as E/C/F/TAF) resulted in non-inferior efficacy and improved renal and bone laboratory parameters as compared to F/TDF-based therapy (administered as E/C/F/TDF or Stribild®). The MAA is also supported by data from additional Phase 3 studies evaluating the F/TAF-based regimen (administered as E/C/F/TAF) among virologically suppressed adults who switched regimens and adults with mild-to-moderate renal impairment. Lastly, bioequivalence studies demonstrated that the formulation of the fixed-dose combinations of F/TAF achieved the same drug levels in the blood as in E/C/F/TAF.

Review of the MAA will be conducted by the EMA under the centralized procedure, which, when finalized, may lead to the grant of marketing authorization by the European Commission, which is valid in all 28 member states of the European Union.

F/TAF and TAF are investigational products and have not been determined to be safe or efficacious.

About Gilead

Gilead Sciences is a biopharmaceutical company that discovers, develops and commercializes innovative therapeutics in areas of unmet medical need. The company's mission is to advance the care of patients suffering from life-threatening diseases. Gilead has operations in more than 30 countries worldwide, with headquarters in Foster City, California.

Forward-Looking Statement

This press release includes forward-looking statements within the meaning of the Private Securities Litigation Reform Act of 1995 that are subject to risks, uncertainties and other factors, including the possibility that the EMA may not adopt a positive opinion in its evaluation and the European Commission may not grant marketing authorization. Further, the FDA and other regulatory authorities may not approve F/TAF, E/C/F/TAF and other F/TAF-based regimens in the currently anticipated timelines or at all, and marketing approvals, if granted, may have significant limitations on their use. As a result, F/TAF, E/C/F/TAF and other F/TAF-based regimens may never be successfully commercialized. In addition, Gilead may be unable to file for regulatory approval for F/TAF with other regulatory authorities in the currently anticipated timelines. These risks, uncertainties and other factors could cause actual results to differ materially from those referred to in the forward-looking statements. The reader is cautioned not to rely on these forward-looking statements. These and other risks are described in detail in Gilead's Quarterly Report on Form 10-Q for the quarter ended March 31, 2015, as filed with the U.S. Securities and Exchange Commission. All forward-looking statements are based on information currently available to Gilead, and Gilead assumes no obligation to update any such forward-looking statements.

The European SmPCs for Stribild and Viread are available from the EMA website at www.ema.europa.eu.

For more information on Gilead Sciences, please visit the company's website at www.gilead.com, follow Gilead on Twitter (@GileadSciences) or call Gilead Public Affairs at 1-800-GILEAD-5 or 1-650-574-3000.

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BEFORE THE OFFICE OF THE CONTROLLER OF PATENTS, NEW DELHI

IN THE MATTER OF:

The Patents Act, 1970 as amended by the Patents (Amendment) Act 2005, and The Patent Rules, 2003, as amended by The Patents (Amendment) Rules, 2016

AND

IN THE MATTER OF:

Indian Patent Number 397784 (formerly patent application number 201817001590) filed by GILEAD SCIENCES INC. having the principal business place at 333 Lakeside Drive Foster City CA 94404

AND

REPRESENTAION BY:

LOW COST STANDARD THERAPEUTICS. ..... **OPPONENT**

vs

GILEAD SCIENCES INC. .....**PATENTEE**

NOTARY SANYOGITA Tis Hazari Courts DELHI Regn. No. 7517 GOVT. OF INDIA

AFFIDAVIT

[A] INTRODUCTION:

I, Dr. Foziyah Zakir, age 38 years, D/o Mr. Zakir Hussain, residing at House no. 598, Sushant Lok, Block C, Phase 1, Gurugram, Haryana and working for gain at School of Pharmaceutical Sciences, Delhi Pharmaceutical Sciences and Research university (DPSRU), presently at Delhi do hereby state as under:

1. I am serving as Assistant Professor with the School of Pharmaceutical Sciences, Delhi Pharmaceutical Sciences and Research University (DPSRU). I hold PhD in the field of pharmaceutics with extensive experience in the field of pharmaceutical sciences including formulation research and development. I have over 10 years of experience in research on pharmaceutical formulations. I also have 7 years of experience in teaching and training students in pharmaceutical formulation development.

2. I have been approached by LOW COST STANDARD THERAPEUTICS (herein after referred as the "Opponent") regarding an Indian Patent Number 397784 (formerly patent application number 201817001590) (herein after referred as "impugned patent") in the name of GILEAD SCIENCES INC., (hereinafter referred as "Patentee"). The relevant documents like specification and claims, was provided by Opponent. The impugned patent claims fixed-dose compositions of two antiviral active ingredients. The claims that I referred are given below:

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1. A tablet consisting of:
(a) 28 mg tenofovir alafenamide hemifumarate, and
(b) 200 mg emtricitabine as the only active ingredients, and
(c) one or more excipients;
wherein the tablet comprises 7% to 9% by weight tenofovir alafenamide hemifumarate, and
wherein the tablet is uncoated or coated by a polymeric film coating.

2. The tablet as claimed in claim 1 comprising 8% by weight tenofovir alafenamide hemifumarate and at least 55% by weight emtricitabine.

3. The tablet as claimed in any one of claims 1-2, wherein the one or more excipients comprise croscarmellose sodium, microcrystalline cellulose, and magnesium stearate.

4. The tablet as claimed in any one of claims 1-3, wherein the one or more excipients comprise 20-35 mg croscarmellose sodium, 70-120 mg microcrystalline cellulose and 1-7 mg magnesium stearate.

5. The tablet as claimed in claim 1, wherein the tablet consists of 200 mg emtricitabine, 28 mg tenofovir alafenamide hemifumarate, 28 mg croscarmellose sodium, 88.70 mg microcrystalline cellulose, 5.25 mg magnesium stearate, and a polymeric film coating.

6. The tablet as claimed in claim 1, wherein the tablet consists of 28 mg tenofovir alafenamide hemifumarate, 200 mg emtricitabine, croscarmellose sodium, microcrystalline cellulose, magnesium stearate, and a polymeric film coating.

7. The tablet as claimed in claim 1, wherein the tablet has a total weight of 350 mg ± 25 mg.

8. The tablet as claimed in claim 1 consisting of:

Ingredient	Mass (mg)
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±3
Microcrystalline cellulose	89 ±9
Magnesium stearate	5.2 ±1.1.

9. The tablet as claimed in claim 1, consisting of:

Ingredient	Mass (mg)
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±1.4
Microcrystalline cellulose	89 ±4
Magnesium stearate	5.2 ±0.5.

10. The tablet as claimed in claim 1, consisting of:

Ingredient	Mass (mg)
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28
Microcrystalline cellulose	89
Magnesium stearate	5.3.

11. The tablet as claimed in claim 1, consisting of:

Ingredient	Mass (mg)
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±3
Microcrystalline cellulose	89 ±9
Magnesium stearate	5.2 ±1.1

and a polymeric film coating.

12. The tablet as claimed in claim 1, consisting of:

Ingredient	Mass (mg)
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28 ±1.4
Microcrystalline cellulose	89 ±4
Magnesium stearate	5.2 ±0.5

and a polymeric film coating.

13. The tablet as claimed in claim 1, consisting of:

Ingredient	Mass (mg)
Emtricitabine	200
Tenofovir alafenamide hemifumarate	28
Croscarmellose sodium	28
Microcrystalline cellulose	89
Magnesium stearate	5.3

and a polymeric film coating.

14. A tablet consisting of:
(a) 11 mg tenofovir alafenamide hemifumarate, and
(b) 200 mg emtricitabine as the only active ingredients; and
(c) one or more excipients;
wherein the tablet comprises 3% to 4% by weight tenofovir alafenamide hemifumarate, and
wherein the tablet is uncoated or coated by a polymeric film coating.

15. The tablet as claimed in claim 14, wherein the tablet comprises between 100 mg and 150 mg of excipients.

16. The tablet as claimed in claim 14 or 15, wherein the tablet comprises 3% by weight tenofovir alafenamide hemifumarate.

17. The tablet as claimed in claim 14, wherein the total weight of the tablet is 360.5 mg.

3. I have been apprised that a case of lack of novelty as well as inventive stepis made out where the subject matterof IN 397784is not novel and devoid of any inventive step.

4. I would like to make it clear that my opinion is purely based on my education, knowledge, training and experiences in the relevant scientific field.

5. I also want to declare that for consulting the matter described herein on this case, I am being compensated but neither my compensation is related to outcome of this proceeding nor I have personal interest with bad faith.

6. Importantly, I am well aware that hindsight approach is not permissible in these deliberations.

7. In addition to above all, I firmly believe that the inventions filed without merits should loudly be opposed.

8. I am no stranger to the requirements of an invention as per Indian Patents Act, 1970 (herein after referred to as "The Patents Act") that to be patentable, invention must be novel, inventive/non-obvious and have industrial applicability. More importantly, the invention shall also cross the barrier of non-patentable subject matters that are described in the Patents Act.

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9. I am quite acquainted with the definition of inventive step which is also known as non-obviousness that it is a feature of an invention that involves technical advance as compared to the existing knowledge or having economic significance or both and that makes the invention not obvious to a person skilled in the art.

10. Thus, from above information, it may be clear to the Learned Controller that I am well mindful of Indian patent law also.

11. I am advised that this scrutiny is also to be done by a person skilled in the art who may be one with Ph.D. degree in Pharmacy and experience in pharmaceutical formulation development.

12. I have also been exposed to patents and I am familiar with reading and understanding pharmaceutical patents as I have gone through the same in my career.

13. Any 'skilled person' as described above is one who would be able to understand patent and its working. I believe I would meet these criteria.

[B] EDUCATIONAL AND PROFESSIONAL BACKGROUND:

14. My educational qualifications, work experience, associations are more particularly stated in Exhibit-1, hereto. In view thereof, I am the person skilled in the art who is the addressee of the invention disclosed and claimed in the opposed patent application and I am competent to make this Affidavit inter alia expressing an opinion, as an expert in the field of pharmaceutical Sciences on the issue stated herein below.

[C] TECHNICAL:

15. In view of my position, qualifications, experience and knowledge in the field of Pharmaceutical Sciences, I am competent to depose this Affidavit by expressing my opinions on the discussed matter.

16. I say that the claimed subject matteris not novel, devoid of any inventive step in light of prior arts and sufficiently not disclosed the present invention due to following reasons.

17. The impugned patent lacks novelty in view of the disclosure of US2015/0105350, hereinafter referred to as US'350.

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18. US'350 discloses hemifumarate form of 9-(R)-2-(S)-(S)- 1-(isopropoxycarbonyl)ethylaminophenoxyphosphinylmethoxypropylladenine (tenofovir alafenamide hemifumarate, TAF, GS-7340) in combination with Emtricitabine.

19. US'350 further disclose the composition comprising 3-40 mg tenofoviralafenamide hemifumarate (TAF) and 50-500 mg Emitricitabinefor treating a viral infection in a human.(Paragraph[0157] to [0162]).Particularly, as seen in Figure 4A and 4b, TAF can be combined with Emtricitabine in the dose of TAF 25mg + Emtricitabine 200mg.

20. Paragraph [0290] of US'350 also discloses that the combination can include 200 mg of emitricitabine and 10 mg oftenofoviralafenamide hemifumarate.

21. It is disclosed in US'350 that the unit dosage form can be a tablet form comprising excipient like microcrystalline cellulose, croscarmellose sodium and magnesium stearate with a polymeric coating.

22. I say that para[0251] of US'350 further discloses that the composition could be a polymer coated tablet composition and the excipients could be filler (diluents): micro crystalline cellulose, a disintegrating agent: croscarmellose sodium, a lubricant: magnesium stearate.

23. Para [0219 and 0220] ofUS' 350 also discloses that though the drugs TAF and Emtricitabine are used in their standard known doses of 25 or 10mg TAF and 200mg Emtricitabine, as aforementioned, it is preferred to have the amount of tenofovir alafenamide in a unit composition to be 3±1mg weight or 8±2mg weight of the composition.

24. I say that para [0293] of US' 350 further discloses that one of skill in the art will know that, in the case of administering a pharmaceutically acceptable salt or complex of an agent, the amount administered will be adjusted relative to the weight of the component added to produce the salt or complex. In light of this, the dose of 10 mg and 25mg tenofovir alafenamide comes to be 11mg and 28mg of tenofovir alafenamide hemifumarate.

25. In light of the above discussed disclosure of US'350, in my opinion,the impugned patent lacks novelty.

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26. I further say that the claimed subject matter of the impugned patent is also devoid of any inventive step in light of documents available in public domain in view of following reasons.

27. US'350 discloses the combination of hemifumarate salt of tenofovir alfenamide (also known as TAF) and emtricitabinein 25/200 mg and 10/200 mg dosage.

28. US' 350 also discloses that the unit dosage form can be a tablet form comprising excipient like microcrystalline cellulose, croscarmellose sodium and magnesium stearate with a polymeric coating.

29. I observe that even though US'350 specifically teaches the dose of tenofovir alafenamide to be used in combination with Emtricitabine to be 25 mg or 10m which is equivalent to 11mg or 28mg of tenofovir alafenamide hemifumarate, at the same time the document also teaches that the preferred weight of tenofovir in the composition should 3±1mg or 8±2mg.

30. From the above disclosure of US'350 a person skilled in the art gets the teaching that when a unit dosage form of tenofovir alafenamide and emtricitabine is prepared the dose of tenofovir in the combination tablet should be 25mg or 10mg, equivalent to 28mg or 11mg of tenofovir alafenamide hemifumarate while the weight of the tenofovir component in said unit dosage form should be 3±1mg or 8±2mg of the composition.

31. However, I find that apart from tenofovir alafenamide and emtricitabine combination US'350 also teaches combination of elvitegravir, cobicistat, emtricitabine and tenofovir alafenamide and particularly teaches the combination of tenofovir alafenamide and cobicistat.

32. In order to ascertain whether the combination of tenofovir alafenamide and emtricitabine alone without other drugs taught in US'350 especially without cobicistat will be therapeutically effective or not, I did further study on the combinations known in the field at the time before the contested patent was first filed.

33. I found validation for use of combination of tenofovir alafenamide and emtricitabine alone without other drugs taught in US'350, especially without cobicistat in a Gilead press release entitled "*European Medicines Agency Validates Gilead's Marketing*

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Application for Fixed-Dose Combination of Emtricitabine and Tenofovir Alafenamide for HIV Treatment" which was published on May 28, 2015

34. I note that the press release discloses that two different fixed-dose combinations regimens of emtricitabine and tenofovir alafenamide incorporating 200/10 mg and 200/25 mg dose strength were validated by the EMA (European Medicinal Agency which is the drug regulatory authority of European Union). It is further disclosed in this press release that bioequivalence studies demonstrated that the formulation of fixed-dose combinations of emtricitabine and tenofovir alafenamide achieved the same drug levels in the blood as in another regimen incorporating elvitegravir, cobicistat, emtricitabine and tenofovir alafenamide (E/C/F/TAF).

35. I also find further support for the maintenance of standard dose of Tenofovir alafenamide and emtricitabine in a composition to be 10mg/200mg or 25mg/200mg, respectively from thisGilead press release.

36. Therefore, a person skilled in the art is taught that the combination of Tenofovir alafenamide and emtricitabine in a composition in dose of 10mg/200mg or 25mg/200mg is therapeutically effective and bioequivalent to combination of elvitegravir, cobicistat, emtricitabine and tenofovir alafenamide.

37. I found yet another document WO2014184553 (WO'553)which relates to pharmaceutical antiretroviral compositions comprising a combination of antiretroviral agents, said compositionsincluding tablets.

38. The tablet compositions of antiviral agents disclosed in WO'553 include the tablets composed of tenofovir disoproxil fumarate and emtricitabine in example 1 and example 2. The compositions of tenofovir and emtricitabine comprising microcrystalline cellulose, croscarmellose sodium and also have a polymer coating.

39. Another document Paul E. Sax et al., Tenofovir Alafenamide Vs. Tenofovir Disoproxil Fumarate in Single Tablet Regimens for Initial HIV-1 Therapy: A Randomized Phase 2 Study, J.Acquir. Immune.Defic.Syndr. 2014;67:52–58discloses that a 10-day monotherapy as well as combination therapy study was conducted in patients in HIV-1–positive patients.

40. In the study conducted by Paul Sax et al it was found that those who received 25 mg of TAF had an approximately 0.5 log10 greater decline in plasma HIV-1 RNA than did patients who received the standard 300-mg dose of TDF (tenofovir disoproxil fumarate). In vitro, higher intracellular TFV-DP levels enable TAF to retain activity against viruses that have reduced susceptibility to TDF suggesting the potential use of TAF in a broader range of patients.

41. In the combination therapy study by same group a randomized, double-blind Phase 2 clinical trial of 2 combinations was conducted. One combination was elvitegravir 150 mg, cobicistat 150 mg, emtricitabine 200 mg, and TAF 10 mg (E/C/F/TAF) compared with another combination of elvitegravir 150 mg, cobicistat 150 mg, emtricitabine 200 mg, and TDF 300 mg (E/C/F/TDF). It was concluded in the study that the combination containing TAF fared better than the combination containing TDF.

42. Thus, the disclosure of Paul Sax et al teaches that TAF can effectively replace TDF in any combination. This teaching of Sax et al when read with disclosure of WO'553, a person skilled the art is taught that combination TAF and emtricitabine without use of any other drug and with use of TAF in dose of 10mg or 25mg can provide a therapeutically effective combination.

43. The combined disclosure of WO'553 and Sax et al reinforces the teaching of US'350 and Gilead press release document that tenofovir alafenamide combined with emtricitabine is a therapeutically effective combination.

44. Further, the combined reading of the documents discussed above also teaches a person skilled in the art that the when tenofovir alafenamide is combined with emtricitabine the dose of tenofovir alafenamide should be 10mg or 25mg, equivalent to 11mg or 28mg tenofovir alafenamide hemifumarate and that the weight of the tenofovir alafenamide present in a composition containing said combination should be 3±1mg or 8±2mg of the composition i.e. the percent weight of tenofovir in such a composition should be 3±1 or 8±2weight % of the composition.

45. In view of above, I find the invention claimed in the patent 397784 to be obvious to a person skilled in the art.

46. Further, I have gone through the specification of the patent 397784 and I have also been told by the Opponent that the Patentee had submitted before the patent office that the presence of particular weight % of tenofovir alafenamide in the claimed composition provides better stability i.e. lesser degradation of tenofovir in the claimed composition.

47. However, I find that the published documents prevailing before the filing of the application for said patent clearly teach not only the standard dose of tenofovir alafenamide and emtricitabine to be used but also teach the preferred weight of tenofovir alafenamide to be present in a unit composition. The values of dose and weight of tenofovir alafenamide in a composition claimed in said patent are same as that taught by these published documents.

48. Therefore, any effect on stability of tenofovir alafenamide in a composition prepared in accordance with the teachings of aforesaid published documents is merely an observation on part of the formulator rather than an application of the formulator's own ingenious faculty and science.

49. In view of above, I am of the opinion that the composition claimed in patent 397784 provides no technical contribution in the field over and above what was already known at the time of the invention.

50. I also observe in the claims of said patent, particularly in Claim1 and Claim 14 that the claims reflect % by weight of Tenofovir alafenamide hemifumarate in the claimed composition which has to be in range of7 to 9% or 3 to 4% by weight of the tablet. However, the doses of the both drugs is fixed as per known therapeutically effective dose i.e. emtricitabine 200mg and tenofovir alafenamide hemifumarate28 or 11 mg which is also equivalent to tenofovir alafenamide 25 or 10 mg respectively. Therefore, the percentage by weight of tenofoviralfenamide has to be adjusted by varying the amount of excipient. The specification does notprovide any teaching or direction about which excipients to vary and the variation in weight of the excipients has to be done to what extent or range in order to arrive at the required weight % of tenofovir alafenamide. Lack of this information results in a lot of experimentation to be done by a person of ordinary skill in the field to modulate the weight of excipients in the tablet so as to achieve the tablet as claimed in the said patent.

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51. I also note that before the filing date of the application for patent 397784a document is published which is Gilead press release "*European Medicines Agency Validates Gilead's Marketing Application for Fixed-Dose Combination of Emtricitabine and Tenofovir Alafenamide for HIV Treatment*".

52. In this press release Gilead has announced that marketing authorization has been obtained for Gilead's investigational fixed-dose combination of emtricitabine and tenofovir alafenamide (200/10 mg and 200/25 mg) (F/TAF) from European Medicinal Agency as well as US Food and Drug Authority on the basis of successful Phase III clinical trial conducted by Gilead.

53. Thus, the fixed-dose combination of 200 mg emtricitabine with 10 mg of tenofovir alafenamide as well as a fixed-dose combination of 200 mg emtricitabine with 25 mg of tenofovir alafenamide was already known and were in public domain before the priority date of the impugned patent.

54. I say that once the composition of F/TAF i.e. a composition of emtricitabine 200mg and tenofovir alafenamide 10 or 25mg was in public domain, the knowledge of weight of tenofovir alafenamide in such a composition was a mere exercise of weighing said composition without use of any special equipment or exercise of any special methodology.

55. Therefore, in my opinion the composition claimed in the patent 397784 was already known to public before the date of filing of application for said patent.

56. I say that in light of preceding paragraphs it is clear that the invention claimed in the patent 397784 lacks novelty, is obvious and isdevoid of any technical advancement and was publicly known before the date of filing of the application for said patent. In addition, I say that the patent does not sufficiently and clearly describe the invention or the method by which it is to be performed.

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DEPONENT

VERIFICATION

I, Dr. Foziyah Zakir, the Deponent do hereby verify that the contents of my affidavit at para 1 to 56 are true and correct to the best of my knowledge, my experience and records. No part of it is false and nothing material has been concealed there from.

Verified at New Delhi on this 2nd day of June, 2023

DEPONENT

ATTESTED

NOTARY PUBLIC
DELHI (INDIA)

2 JUN 2023

NOTARY
MAYOGITA
Tis Hazari Courts
Delhi
Regn. No. 7517
GOVT OF INDIA

Annexure - 7

Dr. FOZIYAH ZAKIR

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Orcid id: https://orcid.org/0000-0002-5315-1190
Google scholar: https://scholar.google.com/citations?user=YNn1-7gAAAAJ&hl=en

Career Snapshot

Degrees/Qualification	**D.Pharm, B.Pharm, M.Pharm (Pharmaceutics), Ph.D (Pharmaceutics)**
Total Teaching Experience	**7.5 years**
International teaching experience (Pharm.D curriculum)	**3.5 years**
Total Research Experience	**10 years**
Total publications	**29**
Books/Book chapters authored	**08**
Cumulative Impact factor	**67.88**
Google scholar h-index	**12**
Google scholar i-10 index	**14**
Scopus h-index	**9**
Total API (Academic Performance Indicator) score	**691**
Awards	**07**
DST-DISHA Woman Scientist project	**01**
M.Pharm theses co-supervised	**07**
Conferences/workshops attended	**40**
Presentations delivered at International/National platform	**27**
Workshops/Seminars organized	**04**
FDP/QIP attended	**08**
e-PG pathshala modules contributed (under UGC scheme)	**07**
Invited lectures delivered	**03**

Work Experience

Aug 2022 –Till date **Assistant Professor (Regular)**

School of Pharmaceutical Sciences, Delhi Pharmaceutical Sciences and Research University (DPSRU)

- Working as sessional exam coordinator and mentor-mentee coordinator
- Actively involved in establishment of departmental laboratory
- Engaged in revision and planning of Course curriculum
- Website In-charge
- Conduct lectures and practicals

Nov 2021 –Till date **Assistant Professor (Full time contractual)**

Department of Pharmaceutics, School of Pharmaceutical Sciences, Delhi Pharmaceutical Sciences and Research University (DPSRU)

- Involved in teaching M.Pharm (Cosmeceutics), M.Pharm (Industrial Pharmacy), B.Pharm and Certificate in Beauty and Wellness Consultant (World Class Skill Centre)
- Supervised projects in areas of dermal diseases and oral health with focused approach of harnessing the therapeutic potential of traditional medicines through modern drug delivery tools
- Worked as subject and course coordinator

Sep 2019 – Oct 2021 **Assistant Professor (Full time visiting)**

Department of Pharmaceutics, School of Pharmaceutical Sciences, Delhi Pharmaceutical Sciences and Research University (DPSRU)

- Involved in teaching M.Pharm (DRA), M.Pharm (Cosmeceutics), M.Pharm (Industrial Pharmacy), B.Pharm and Certificate in Beauty and Wellness Consultant (World Class Skill Centre, New Delhi)
- Supervised projects in areas of dermatological drug delivery through transdermal, topical for wound healing
- Conducted lectures and practicals
- Prepared sessional examination question papers, conducting exams, grading the answer sheets

June 2016 – June 2019 **Woman Scientist (Sponsored by DST-KIRAN scheme)**

Jamia Hamdard, New Delhi

- Completed the prestigious independent DST Research project based on "Design and development of nanodelivery system for the treatment of postmenopausal osteoporosis". The project yielded a pathbreaking tool for treating a debilitating women issue of osteoporosis associated with menopause.
- Developed skills in QbD, PK/PD software tools
- Acquired proficiency in project management, design and execution of experiments and troubleshooting

Jan 2013 – June 2016 **Lecturer**

College of Pharmacy, Jazan University, Saudi Arabia

- Prepared study material and course material for Pharm.D curriculum

- Mentored project works of the students (Annual student conference) and helped them design novel delivery systems such as polymeric nanoparticles, nanosuspension, liposome etc.
- Wrote research, review papers and book chapters
- Conducted lectures and practicals
- Held the prestigious post of Academic advisor

July 2010 – July 2011 **Lecturer**

Rayat Insitute of Pharmacy, Railmajra, Ropar, Punjab

- Taught B.Pharm curriculum, courses taught included Pharmaceutical Technology, Pharmaceutical Jurisprudence, Physical Pharmacy etc.
- Taught M.Pharm (Pharmaceutics) subjects such as Novel Drug Delivery Systems
- Co-supervised M.Pharm post-graduate thesis

Nov 2009 – July 2010 **Lecturer**

Global College of Pharmacy, AnandpurSahib, Punjab

- Conducted lectures and practicals
- Organized industrial tours for Bachelor students

Education

2016 - 2020 PhD (Pharmaceutics)
Department of Pharmaceutics, School of Pharmaceutical Education and Research, Jamia Hamdard, New Delhi

2007 - 2009 M.Pharm (Pharmaceutics)
ISF College of Pharmacy, Moga
Punjab Technical University

2004 - 2007 B.Pharm
Department of Pharmaceutical Sciences
Maharishi Dayanand University, Rohtak

2002 – 2004 D.Pharm
Hindu College of Pharmacy, Sonepat

Professional Summary

Publications

Journal publications:	29
e –PG Pathshala Modules :	07
Books:	01
Book Chapters:	08
h–index:	12
i-10 index:	14
Cumulative IF:	67.88
Citations:	640

Conference/Seminars/ Workshops

- Participated in 40 National and International Conferences
- 24 oral and poster presentations
- 10 Workshops/Seminars

Awards

- **Distinguished Research Award** during 2nd Research Promotion Award Ceremony held at Delhi Pharmaceutical Sciences and Research University, New Delhi on 28 Feb 2023.
- **Notable Research Award** during 1st Research Promotion Award Ceremony held at Delhi Pharmaceutical Sciences and Research University, New Delhi on 05 April 2022.
- **Best poster award** at 6th World Congress on Nanomedical Sciences, 7-9 Jan 2019, New Delhi, (awarded by ACS publishers)
- **2nd prize** in poster presentation at Pharma Ratan, Nov 2017
- **3rd prize** in oral presentation (as a supervisor) at 6th annual student conference, Jazan University, Jizan, Saudia Arabia, Jan 2015
- **Best oral presentation award** for delivering presentation on "Surface Engineered nanoparticles for oral immunization" held at **International Symposium on the safe use of Nanomaterials,** Feb 2011, CSIR, Lucknow, India
- **Award for academic excellence** during M.Pharm, Punjab Technical University, 2010
- **5th position** in university during **Masters in Pharmacy**, Punjab Technical University, 2010

Projects

Development of Raloxifene nanodelivery system for the prevention and treatment of postmenopausal osteoporosis

Amount sanctioned: 27 lacs

Duration: 3 yrs

Sponsoring Agency: DST (under DISHA scheme)

Status: Completed

Skills

- QbD Approaches learnt and softwares handled
- Highly trained in formulation development and worked in areas such as nanosuspension, polymeric nanoparticles, nanoemulsions, fatty acid vesicles, solid dispersion etc.
- Ability to conduct research independently with minimum supervision
- Excellent writing skills with publications in peer reviewed international journals with a cumulative impact factor of 67.88

Extra-curricular and Co-curricular activities

- Co-coordinated preconference workshop on "**Role of Pharmacognosy and Regulations for the Development of Traditional Medicine",** held on 03 Nov 2022.
- Co-Coordinated one week symposium on "**Har Din Har Ghar Ayurveda**", 5-8th Oct, 2022
- Coordinated workshop on "**International Women's Day**", organized by DPSRU in collaboration with DIIF, 07 March 2022.
- Coordinated webinar on "**Overview of Indian Medical Device Regulations**", organized by Dept. Of Pharmaceutics, SPS, DPSRU, 17 Aug, 2021
- Organizing committee member, **58th pharmacy week celebration**, Oct 21-24, 2019 at School of Pharmaceutical Sciences, DPSRU.
- Content writer in e-PG pathshala modules
- Volunteer in ICMR sponsored workshop on "**Nanomedical Sciences and its impact on healthcare: Challenges, Opportunities and Future Directions**", organized by Jamia Hamdard and University of Delhi, 7 Jan 2019, Vigyan Bhawan.

Design and

FDPs attended	05
Quality Improvement Programs attended	03

Instruments handled

- Differential Scanning Calorimeter(DSC)
- Fourier-transform infrared spectroscopy (FTIR)
- UV-visible spectrophotometer
- Rheometer
- Confocal Laser Scanning Microscope
- Zeta sizer
- X-ray powder diffraction (XRD)
- HPLC
- Micro-CT scanner

Core Competencies

- Teaching , learning and Evaluation (UG and PG)
- Research in Pharmaceutical Sciences :Design, Development and Evaluation of Novel Drug Delivery Systems

List of journal publications

1. R Khan, MA Mirza, M Aqil, N Hassan, **F Zakir**, MJ Ansari, Z Iqbal. A pharmaco-technical investigation of Thymoquinone and Peat-sourced Fulvic acid nanoemulgel: A combination therapy. Gels, Nov 2022.
2. A Sah, PP Naseef, MS Kuruniyan, GK Jain, **F Zakir**, G Aggarwal. A comprehensive study of therapeutic applications of chamomile. Pharmaceuticals, Oct 2022; 15(10): 1284.
3. R Khan, P Jain, **F Zakir**, M Aqil, S Alshehri, MA Mirza, Z Iqbal. Quality and in vivo assessment of a Fulvic Acid complex: A validation study. Scientica Pharmaceutica, May 2022; 90 (2): 33.
4. A Choudhary, AC Rana, G Aggarwal, V Kumar, **F Zakir**. Development and characterization of an atorvastatin solid dispersion formulation using skimmed milk for improved oral bioavailability. Acta Pharmaceutica Sinica B, Aug 2012; 2(4): 421-428. **IF 11.41**
5. S Maddheshiya, A Ahmad, W Ahmad, G Aggarwal, **F Zakir**. Essential oils for the treatment of skin anomalies: Scope and potential. South African Journal of Botany, In press, Jan 2022. **IF 2.3**
6. S Bharti, **F Zakir**, MA Mirza, G Aggarwal. Antifungal biofilm strategies: a less explored area in wound management. Current Pharmaceutical Biotechnology, Accepted, Jan 2022. **IF 2.83**
7. R Kumar, MA Mirza, PP Naseef, MS Kurunian, G Aggarwal, **F Zakir**. Exploring the potential of natural products based nanomedicine for maintaining oral health. Molecules, Accepted, Jan 2022. **IF 4.4**

8. N Kashyap, A Kumari, N Raina, **F Zakir**, M Gupta. Prospects of essential oil loaded nanosystems for skincare. Phytomedicine plus, Feb 2022; 2(1): 100198.

9. S Mohapatra, MA Mirza, AR Hilles, **F Zakir**, AC Gomes, MJ Ansari, Z Iqbal, S Mahmood. Biomedical application, Patent repository, Clinical trial and Regulatory updates on Hydrogel: An extensive review. Gels, Nov 2021;7(4): 207. **IF 3.22**

10. **F Zakir**, A Ahmad, MA Mirza, K Kohli, FJ Ahmad. Exploration of a transdermal nanoemulgel as an alternative therapy for postmenopausal osteoporosis. Drug Delivery Science and Technology. July 2021; 65, 102745 . **IF-3.98**

11. **F Zakir**, A Ahmad, U Farooq, MA, Mirza, A Tripathi, D Singh, F Shakeel, S Mohapatra, FJ Ahmad, K Kohli. Design and development of a commercially viable in situ nanoemulgel for the treatment of postmenopausal osteoporosis. Nanomedicine (Lond), 2020 May;15(12):1167- 1187. **IF 4.71**

12. **F Zakir**, FJ Ahmad, A Ahmad, K Kohli. Insights into transdermal drug delivery: Approaches for redressal of a burgeoning issue of Osteoporosis. Endocr Metab Immune Disord Drug Target. 2020;20(10):1682-1695. **IF 2.89**

13. F Anjum, **F Zakir**, D Verma, M Aqil, M Singh, P Jain, MA Mirza, MK Anwer, Z Iqbal. Exploration of nanoethosomal transgel of naproxen sodium for the treatment of Arthritis. Curr Drug Deliv. 2020;17(10):885-897. **IF 1.58**

14. **F Zakir**, Farah Islam , A Jabeen , SS Moni. Vaccine development: A historical perspective. Biomedical Research, April 2019; 30(3):452-455. **IF-0.219**

15. L Thomas, **F Zakir**, MA Mirza, MK Anwer , FJ Ahmad, Z Iqbal. Development of curcumin loaded chitosan polymer based nanoemulsion gel: In vitro, ex vivo evaluation and in vivo wound healing studies. Int J Biol Macromol. March 2017; 16(101), 569-579. **IF 6.95**

16. **F Zakir**, S Manvi, Z Iqbal. Ocular drug delivery: Recent Updates. International Journal of Drug Regulatory Affairs. Dec 2017; 4(4): 29-32.

17. MD Sarfaraz Alam, MD Sajid Ali, **F Zakir**, N Alam, MD Intekhab Alam, MR Siddiqui, MF Ahmad, MD Daud Ali, MD Salahuddin Ansari. Enhancement of anti-dermatitis potential of Clobetasol propionate by DHA [docosahexaenoic acid] rich algal oil nanoemulsion gel. Iran J Pharm Res, Dec 2016;15(1):35–52. **IF- 1.5**

18. MM Safhi, SM Sivakumar, A Jabeen, **F Zakir**, F Islam, US Bagul, S Sanobar, T Anwer, R Siddiqui, MA Hakeem Siddiqui, ME Elmobark, BB Barik, MF Alam. Therapeutic potential of chitosan nanoparticles as antibiotic delivery system: challenges to treat multiple drug resistance. Asian J Pharm, Apr 2016; 10(2):S61-S66. **IF- 0.46**

19. S M Sivakumar, MM Safhi, MF Alam, T Anwer, G Khan, A Jabeen, **F Zakir**, F Islam. Insight on Hepatitis B vaccine adjuvanticity “Future Perspectives”. SM J Hepat Res Treat. April 2016; 2(1):1007-1009.

20. B Malik, AK Goyal, TS Markandeywar, G Rath, **F Zakir**, SP Vyas. Microfold-cell targeted surface engineered polymeric nanoparticles for oral immunization. Journal of Drug Targeting, Jan 2012; 20(1): 76-84. IF 3.408

21. **F Zakir**, Himanshu S, Karandeep K, Basant M, B Vaidya, AK Goyal, SP Vyas. Nanocrystallization of poorly water soluble drugs for parenteral administration. Journal of Biomedical Nanotechnology, Jan 2011; 7(1):127-129. **IF 5.068**

22. B Malik, AK Goyal, **F Zakir**, SP Vyas. Surface engineered nanoparticles for oral immunization. Journal of Biomedical Nanotechnology, Jan 2011; 7(1):132-134. **IF 5.068**

23. **F Zakir**, B Vaidya, AK Goyal, B Malik, SP Vyas. Development and characterization of oleic acid vesicles for the topical delivery of fluconazole. Drug

Delivery, 2010; 17(4):238- 248. **IF 3.09**

24. B Malik, AK Goyal, S Mangal, **F Zakir**, SP Vyas. Implication of gut immunology in the design of oral vaccines. Current Molecular Medicine, Feb 2010; 10(1):47-70. **IF- 2.22**

25. M Safhi, SM Sivakumar, **F Zakir**, A Jabeen, F Islam. Chitosan nanoparticles as a sustained Penicillin G delivery prepared by ionic gelatin technique. Journal of Pharmacy Research, Oct 2014; 8(10):1352-1354.

26. V Kumar, **F Zakir**, G Agarwal, A Choudhary. Formulation and evaluation of buccal patches of venlafaxine. International Journal of Pharmacy and Biological Sciences. Sept 2011; 1(3):170-182. **IF- 0.88**

27. V Kumar, A Geeta, **F Zakir**, A Choudhary. Buccal bioadhesive drug delivery—a novel technique. International Journal of Pharmacy and Biological Sciences. July 2011; 1(3): 89- 102. **IF- 0.88**

28. A Choudhary, G Aggarwal, **F Zakir**, V Kumar. Mini Review: Journey of solid dispersion technique from bench to scale. Int Res J Pharm, Aug 2011; 2(8):46-51. **IF-0.76**

29. R Khan, P Jain, **F Zakir**, M Aqil, S Alshehri, MA Mirza, Z Iqbal. Quality and In Vivo Assessment of a Fulvic Acid Complex: A Validation Study. Scientica Pharmaceutica. May 2022; 90(2): 33.

Books

F Zakir, G Aggarwal. Pharmaceutical Engineering Lab Manual. New Delhi: SR Health Sciences Pvt. Ltd, 2021.

Book Chapters

1. T Alex, DK Lang, **F Zakir**, A Mirza, Z Iqbal. The Tumor Microenvironment. In: Padhi, S., Behera, A., Lichtfouse, E. (eds) Polymeric nanoparticles for the treatment of solid tumors. Environmental Chemistry for a Sustainable World, vol 71. Springer, Cham, Pages 1-49.
2. **F Zakir**, S Mohapatra, U Farooq, MA Mirza, Z Iqbal. Introduction to metabolic disorders. In: H Dureja, SN Murthy, PR Wich, K Dua (eds). Drug Delivery Systems for Metabolic Disorders. Elsevier Inc. publications USA, 2022, Pages 1-20.
3. **F Zakir**, S Mohapatra, B Aftab, Z Iqbal, G Aggarwal. Clinical trials, future prospectsand challenges of drug delivery in combating metabolic disorders. In: H Dureja, SN Murthy, PR Wich, K Dua (eds). Drug Delivery Systems for Metabolic Disorders. Elsevier Inc. publications USA, 2022, Pages 481-489.
4. **F Zakir**, H Mishra, M Azharuddin, MA Mirza, G Aggarwal, Z Iqbal. Gastrointestinal abnormalities and Nigella sativa: A Narrative Review of Preclinical and Clinical Studies. In: Andaleep Khan (ed). Black seeds (Nigella Sativa): Pharmacological and Therapeutic Applications. 1st edition. Elsevier Inc. publications USA, 2021, Pages

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5. **F Zakir**, MA Mirza, Rahmuddin, Z Iqbal. Role of Nanomedicine in the Diagnosis of Cardiovascular Diseases. In: Iqbal Z, Dilnawaz F (eds). Nanomedicinal Approaches towards Cardiovascular Disease. 1st edition. Bentham Sciences publishers, UAE, 2021, Pages 17-25.
6. S Padhi, M Azharuddin, A Behera, **F Zakir**, MA Mirza, AA Chyad, Z Iqbal, S Mansoor. Nanopotentiation of Repurposed Drugs for Amelioration of COVID-19: Insights and Perspectives. In: Chukwuebuka Egbuna (ed). Coronavirus Drug Discovery, Elsevier Publications USA, 2021, 1st edition.
7. Sivakumar SM, Anwer T, Jabeen A, **F Zakir**, Bagul U, Eltaib M, Khan G, Siddiqui R, Abouelhag H, Alam F. Nanoparticle System for Anticancer Drug Delivery: Targeting to Overcome Multidrug Resistance. In: Alexandru Grumezescu (ed). Multifunctional Systems for Combined Delivery, Biosensing and Diagnostics, Elsevier publications, 2st edition, 2017, Pages 159–169.
8. **F Zakir**, B Vaidya, AK Goyal, B Malik, SP Vyas. Fatty acid vesicles: promising drift in colloidal drug carriers. In: SP Vyas, RSR Murthy, RK Narang (eds). Nanocolloidal carriers: site specific and controlled drug delivery, CBS publishers, 1st edition, 2010, Pages 201-218.

Personal Information

Date of Birth	15 Oct 1984
Marital Status	Married
IT skills	MS office (MS word, MS powerpoint, MS excel), QbD softwares, PK/PD modeling
Hobbies	Scientific reading and writing

सत्यमेव जयते

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FORM 26
THE PATENTS ACT, 1970
(39 of 1970)
&
The Patent Rules, 2003
FORM OF AUTHORISATION OF A PATENT AGENT/OR ANY PERSON IN A MATTER OR PROCEEDING UNDER THE ACT
[See sections 127 and 132 and rule 135]

We, **LOW COST STANDARD THERAPEUTICS**, I Floor, Premananda Sahitya Bhavan, Opposite Lakadipul, Dandia Bazar, Vadodara, 390 001, Gujarat, India; hereby authorize Rajeshwari H. (IN/PA – 358), Gopalan Deepak Sriniwas (IN/PA – 508), S. Hariharan (D/5892/2004), Pragya Singh Thakur (IN/PA – 3329), Mariam Sadaf (IN/PA – 5231), Rachita Singh (IN/PA - 4617), Ashish Rai (IN/PA - 4254), Swapnil Gaur (D/6242/2018), Nupur Goswami (D/1706/2012), Tahir Abdul Zabbar, Deepanshu Nagar (D/5323/2019), Sugandh Shahi (D/5129/2021), all are Indian citizens, Advocates / Patent Agents of RAJESHWARI & ASSOCIATES, A – 202, FIRST FLOOR, SHIVALIK ENCLAVE, MALVIYA NAGAR, NEW DELHI - 110017, INDIA, and Also at: S – 357, FIRST FLOOR, NEAR HDFC BANK, PANCHSHEEL PARK, NEW DELHI – 110017, INDIA, jointly or severally to act on our behalf for filing an opposition and/or representation by the way of opposition against the Indian Patent No. 397784 (Formerly Indian Patent Application No. 201817001590) dated 29/06/2016 by **GILEAD SCIENCES INC.** entitled: **PHARMACEUTICAL FORMULATIONS COMPRISING TENOFOVIR AND EMTRICITABINE"** is a National Phase of PCT Application No. PCT/US2016/040158 dated 29/06/2016 under the above mentioned Act and in all matters and proceedings relating to the Opposition against patent application before the Controller of Patents or the Government of India in connection therewith or incidental thereto and in general to do all acts or things including filing of representation, statements, replies, extensions, fees, evidence and any or all documents or pleadings, attending hearings and appointment of a substitute or substitutes as the said Agent(s) may deem necessary or expedient and request that all notices, requisitions and communication relating thereto may be sent to such Agent(s) at Rajeshwari & Associates, India.

We hereby revoke all previous authorization, if any made, in respect of same matter or proceeding.

We hereby assent to the action already taken by the said person in the above matter.

Dated this 02nd day of June, 2023

S. SRINIVASAN
MANAGING TRUSTEE

TO
THE CONTROLLER OF PATENTS
THE PATENT OFFICE
DELHI, MUMBAI, KOLKATA AND CHENNAI