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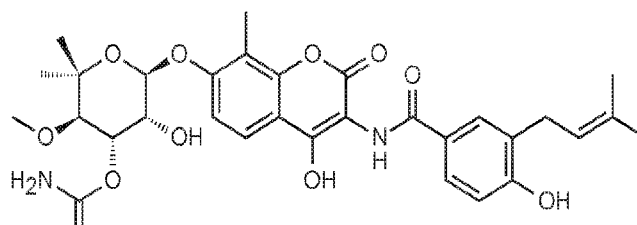


FIG. 1

(57) Abstract: Novobiocin and lopinavir-ritonavir have been found to have significant antiviral activity against a variety of members of the flaviviridae family, including dengue virus, Japanese encephalitis virus, and West Nile virus, and yellow fever virus. Synergistic antiviral effects are found when either novobiocin or lopinavir-ritonavir are used in combination with a variety of other antibiotic and antiviral compounds.



COMPOSITIONS AND METHODS FOR TREATING FLAVIVIRUS INFECTIONS

[0001] This application claims the benefit of United States Provisional Application No. 62/521178, filed on June 16, 2017. These and all other referenced extrinsic materials are incorporated herein by reference in their entirety. Where a definition or use of a term in a reference that is incorporated by reference is inconsistent or contrary to the definition of that term provided herein, the definition of that term provided herein is deemed to be controlling.

Field of the Invention

[0002] [0001] The field of the invention is compositions and methods for treating viral infections, more specifically dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus.

Background

[0003] The background description includes information that may be useful in understanding the present invention. It is not an admission that any of the information provided herein is prior art or relevant to the presently claimed invention, or that any publication specifically or implicitly referenced is prior art.

[0004] Flaviviridae are a family of over 100 species of single stranded RNA viruses, members of which are known to cause animal and human disease. They spread primarily by arthropod vectors (e.g. mosquitoes, ticks, etc.) Diseases caused by flaviviridae include hepatitis, encephalitis, and hemorrhagic fever/syndromes. The viral genome encodes a single polyprotein that is processed by viral and host proteases to provide structural and non-structural proteins. Several of the non-structural (“NS”) proteins include portions utilized in RNA or polyprotein processing that are highly conserved. Such highly conserved regions are useful in identification and phylogenetic analysis by nucleic acid amplification methods.

[0005] Treatment options for many diseases associated with flaviviridae are limited. For example current treatment for Japanese encephalitis, which has a case-fatality rate of approximately 30%, is essentially symptomatic and supportive. Similarly, no specific treatment

is available for West Nile virus, which has become widespread throughout the US with annual case-mortality rates ranging from 3% to 15%. Hepatitis C has been treated with some success using a combination of ribavarin and interferon, however such treatment provided a cure rate of only around 50% and is associated with unpleasant side effects. Highly effective antiviral drugs have recently become available for hepatitis C, however successful treatment is dependent on the genome of the infecting virus and is remarkably expensive (approximately \$1,000 USD per pill for a daily treatment regime that extends up to 6 months). Such costs render these drugs essentially inaccessible for treatment of hepatitis C on a public health scale.

[0006] Thus, there is still a need for safe and effective compositions and methods for treatment of flaviviridae infections.

Summary of The Invention

[0007] The inventive subject matter provides compositions and methods for treatment of flaviviridae, using novobiocin or lopinavir-ritonavir in combination with an additional antibiotic or antiviral compound.

[0008] One embodiment of the inventive concept is a composition for the treatment of flaviviridae infection that includes novobiocin (for example, at from about 1 μM to about 1 mM) and a secondary antiviral or antibiotic compound, where the novobiocin and the secondary antiviral or antibiotic compound exhibit a synergistic effect in inhibiting a virus of the flavivirus family. Suitable secondary antiviral or antibiotic compound include ribavarin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, doxycycline, amantadine, levodopa, and/or isatin. Viruses susceptible to such a drug combination include dengue virus, Japanese encephalitis, West Nile virus, yellow fever virus, and/or hepatitis C virus.

[0009] Another embodiment of the inventive concept is a composition for the treatment of flaviviridae infection that includes lopinavir-ritonavir (for example, at from about 1 μM to about 1 mM) and a secondary antiviral or antibiotic compound, where the lopinavir-ritonavir and the secondary antiviral or antibiotic compound exhibit a synergistic effect in inhibiting a virus of the flavivirus family. Suitable secondary antiviral or antibiotic compound include ribavarin, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, doxycycline,

amantadine, levodopa, and/or isatin. Viruses susceptible to such a drug combination include dengue virus, Japanese encephalitis, West Nile virus, yellow fever virus, and/or hepatitis C virus.

[0010] Another embodiment of the inventive concept is a method of treating an infection caused by a member of the flavivirus family (such as dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus), by providing to a person in need of treatment a formulation that includes novobiocin. In some of such embodiments the formulation also includes a secondary antiviral or antibiotic compound, such as ribavirin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and/or isatin. Such formulation can provide novobiocin in a concentration range of about 1 μ M to about 1 mM.

[0011] Another embodiment of the inventive concept is a method of treating an infection caused by a member of the flavivirus family (such as dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus), by providing to a person in need of treatment a formulation that includes lopinavir-ritonavir. In some of such embodiments the formulation also includes a secondary antiviral or antibiotic compound, such as ribavirin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and/or isatin. Such formulation can provide lopinavir-ritonavir in a concentration range of about 1 μ M to about 1 mM.

[0012] Another embodiment of the inventive concept is a kit for treating an infection caused by a member of the flavivirus family (such as dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus). The kit includes novobiocin and a secondary antiviral or antibiotic compound (such as ribavirin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and/or isatin). In some embodiments the novobiocin and the secondary antiviral or antibiotic compound are provided as a single formulation; in other embodiments they are provided as separate formulations. In some embodiments the kit is formulated to provide novobiocin in a concentration range of about 1 μ M to about 1 mM. Such a kit can include instructions for use, such as a dosing schedule effective in treating the infection.

[0013] Another embodiment of the inventive concept is a kit for treating an infection caused by a member of the flavivirus family (such as dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus). The kit includes lopinavir, ritonavir, and a secondary antiviral or antibiotic compound (such as ribavirin, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and/or isatin). In some embodiments the lopinavir, ritonavir, and the secondary antiviral or antibiotic compound are provided as a single formulation; in other embodiments they are provided as separate formulations. In some embodiments the kit is formulated to provide lopinavir-ritonavir in a concentration range of about 1 μ M to about 1 mM. Such a kit can include instructions for use, such as a dosing schedule effective in treating the infection.

[0014] 30. The kit of one of claims 26 to 29, wherein the secondary antiviral or antibiotic compound is selected from the group consisting of ribavirin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and isatin.

[0015] Various objects, features, aspects and advantages of the inventive subject matter will become more apparent from the following detailed description of preferred embodiments, along with the accompanying drawing figures in which like numerals represent like components.

Brief Description of The Drawings

[0016] FIG. 1: Structure of novobiocin.

[0017] FIG. 2: Structure of lopinavir.

[0018] FIG. 3: Structure of ritonavir.

Detailed Description

[0019] The following description includes information that may be useful in understanding the present invention. It is not an admission that any of the information provided herein is prior art or relevant to the presently claimed invention, or that any publication specifically or implicitly referenced is prior art.

[0020] The inventive subject matter is directed to compositions and methods that are useful in preventing and/or treating Japanese encephalitis, West Nile virus, hepatitis C virus, Dengue virus, and/or Yellow Fever virus infections. Surprisingly, novobiocin was identified as having this activity, as has lopinavir/ritonavir. Novobiocin is an aminocoumarin antibiotic produced by *Streptomyces niveus*, and has been known since the 1950's as an antibacterial drug. It is licensed for clinical use as an oral formulation under the trade name Albamycin™ and has been used as an antistaphylococcal agent effective against methicillin-resistant *streptococcus*. When used as an antibiotic it is well tolerated, with few and minor side effects. The structure of novobiocin is shown in FIG. 1. Lopinavir/ritonavir (sold commercially in combination as Kaletra™) is a drug combination used as a specific HIV protease inhibitor, and is currently approved for use in the treatment and prevention of HIV infections/AIDS. The structures of lopinavir and ritonavir are shown in FIGs. 2 and 3, respectively. Although this drug combination produces some notable side effects (*e.g.* fatigue, headache, nausea, diarrhea) it is considered safe and is approved for use in pregnancy.

[0021] Functional testing has shown that novobiocin and/or lopinavir/ritonavir is effective in inhibiting flaviviridae in culture at μM concentrations. Surprisingly, synergistic (*i.e.* greater than additive) effects are found when used in combination with known antiviral compounds and/or antibiotics such as ribavarin, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline.

[0022] Various objects, features, aspects and advantages of the inventive subject matter will become more apparent from the following detailed description of preferred embodiments, along with the accompanying drawing figures in which like numerals represent like components.

[0023] One should appreciate that the disclosed techniques provide many advantageous technical effects including provision of antiviral activity using a well tolerated, regulatory agency approved compound having a known safety profile, and provision of synergistic effects when such a compound is used in combination with antiviral compounds in current use.

[0024] The following discussion provides many example embodiments of the inventive subject matter. Although each embodiment represents a single combination of inventive elements, the inventive subject matter is considered to include all possible combinations of the disclosed

elements. Thus if one embodiment comprises elements A, B, and C, and a second embodiment comprises elements B and D, then the inventive subject matter is also considered to include other remaining combinations of A, B, C, or D, even if not explicitly disclosed.

[0025] Groupings of alternative elements or embodiments of the invention disclosed herein are not to be construed as limitations. Each group member can be referred to and claimed individually or in any combination with other members of the group or other elements found herein. One or more members of a group can be included in, or deleted from, a group for reasons of convenience and/or patentability. When any such inclusion or deletion occurs, the specification is herein deemed to contain the group as modified thus fulfilling the written description of all Markush groups used in the appended claims.

[0026] In some embodiments of the inventive concept, novobiocin and/or lopinavir/ritonavir is provided on a treatment schedule that is effective in treating an infection caused by a member of the flaviviridae family, particularly dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus. Novobiocin and/or lopinavir/ritonavir can be provided as an oral formulation (*e.g.* liquid suspension or solution, pill, tablet, capsule, powder, gel, *etc.*), but can also be provided as an injectable or infusible formulation, a topical formulation, an inhaled formulation, and/or a formulation suitable for mucosal (*e.g.* nasal, vaginal, rectal, *etc.*) administration. Such formulations can provide novobiocin and/or lopinavir/ritonavir in amounts ranging from 1 mg to 10 g, and can include additives such as bulking agents, flavorants, excipients, vehicles, transport enhancers, and/or stabilizing agents. In some embodiments novobiocin and/or lopinavir/ritonavir can be provided in a sustained-release formulation.

[0027] Suitable treatment schedules can provide novobiocin and/or lopinavir/ritonavir on a daily, 12 hour, 8 hour, 6 hour, 4 hour, or shorter interval. Alternatively, in some embodiments novobiocin and/or lopinavir/ritonavir can be provided every 2 days, every 3 days, weekly, every two weeks, monthly, or at longer intervals. Such treatment schedules can be provided for as short as two days or for as long as a year or more. In some embodiments treatment with novobiocin and/or lopinavir/ritonavir can extend for the remainder of the patient's life.

[0028] When used as an antiviral agent against flaviviridae unexpected synergistic effects were found when novobiocin and/or lopinavir/ritonavir was used in combination with other

antiviral/antibiotic compounds, including ribavirin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, doxycycline, amantadine, levodopa, and isatin. As such, in some embodiments novobiocin and/or lopinavir/ritonavir is provided in combination with such complementary antiviral/antibiotic agents. In such embodiments novobiocin and/or lopinavir/ritonavir and one or more complementary antiviral/antibiotic agents are provided as a single formulation. In other embodiments, novobiocin and/or lopinavir/ritonavir and one or more antiviral/antibiotic agents are provided as separate formulations. Such separate formulations can be provided on the same or different treatment schedules.

[0029] In still other embodiments, novobiocin and/or lopinavir/ritonavir can be provided with additional pharmaceutical compounds that provide symptomatic relief. Suitable additional pharmaceutical compounds include anti-inflammatory compounds, anti-emetic compounds, pain relieving compounds, and/or fever reducing compounds.

[0030] Without wishing to be bound by theory, the Applicants believe that novobiocin and/or lopinavir/ritonavir can complex with amino acids (*e.g.* His51) found in highly conserved regions of flaviviridae proteases (*e.g.* NSP2, NSP2A, NSP3) and/or other nonstructural gene products. Such complex formation inhibits protease/protein activity and provides antiviral activity, which is complementary to antiviral activity provided by other antiviral compounds in a synergistic fashion. Without wishing to be bound by theory, Applicants believe that the carboxamine group of novobiocin is at least partially responsible for this activity.

Examples

Viruses and cell lines

[0031] Clinical isolates of dengue virus (DENV) serotypes 1 to 4, Japanese encephalitis virus (JEV), and West Nile virus (WNV), and a vaccine strain of yellow fever virus (YFV) were used in the antiviral assays. Virus was amplified by passage in Vero cells (ATCC) cultured in minimum essential medium (MEM) supplemented with 1% fetal bovine serum (FBS, Gibco™, Life Technologies Corporation, Massachusetts, USA) and 100 units/ml penicillin plus 100 mg/ml streptomycin. Working stocks of virus were prepared at 4×10^6 tissue culture infectious

dose (TCID₅₀)/ml. For virus titration, aliquots of the working stock were applied on confluent Vero cells cultured in 96-well plates. Serial 10-fold dilutions of virus stock solution were inoculated (in quadruplicate) onto a Vero cell monolayer and subsequently cultured in penicillin/streptomycin-supplemented MEM and 1% FBS. Wells of the plate were observed for cytopathic effect (CPE) for from 3 to 7 days. Viral titer was calculated using the Reed and Munch endpoint method. One TCID₅₀ was interpreted as the amount of virus that resulting in CPE in 50% of inoculated wells.

Drug compounds and cytotoxicity assay

[0032] Amantadine hydrochloride (Sigma-Aldrich Chemie GmbH, Steinheim, Germany), ciprofloxacin (Sigma-Aldrich Chemie GmbH), desmopressin acetate (Ferring Pharmaceuticals, Saint Prex, Switzerland), doxycycline hydrochloride (Sigma-Aldrich Chemie GmbH), isatin (Sigma-Aldrich Chemie GmbH), levodopa (Sigma-Aldrich Chemie GmbH), lopinavir-ritonavir (Abbott Laboratories, Illinois, USA), minocycline (Pfizer, New York City, USA), montelukast (Merck & Co., Inc., New Jersey, USA), novobiocin sodium (Sigma-Aldrich Chemie GmbH), octreotide acetate (Novartis, Basel, Switzerland), ribavirin (Sigma-Aldrich Chemie GmbH), rifampicin (Gruppo Lepetit Srl, Milan, Italy), selegiline (Sigma-Aldrich Chemie GmbH), sirolimus (Pfizer, New York city, USA), and tacrolimus (Astellas Pharma, Tokyo, Japan), were used in this study. The 50% effective cytotoxic concentration (CC₅₀) of the selected drugs was characterized using a thiazolyl blue tetrazolium bromide (MTT) assay in accordance with the manufacturer's instructions.

CPE inhibition assay

[0033] For the CPE inhibition assay drug compounds were diluted with serum-free MEM and added to confluent Vero cells cultured in the wells of 96-well culture plates (4×10^4 cells/well) in triplicate, and incubated for 2 hours at 37° C. After incubation the drug-containing media were removed, and a flaviviridae virus (multiplicity of infection, MOI = 0.0001) was added to each well along with fresh drug-compound media. After an additional 1 hour at 37° C, the virus-drug compound mixture was removed and the cells were washed twice with MEM to remove unbound virus. Subsequently, drug-containing media were added to the cells for further incubation for 6 days at 37° C in 5% CO₂. CPE was determined by examination using inverted light microscopy,

and supernatant was collected for virus quantification. A 4:1 mixture of serum free MEM and 5 mg/ml MTT solution (prepared in 1 x PBS, filtered) was then added to the wells. The cell monolayers were incubated as above for 4 hours in the absence of light. 10% sodium dodecyl sulfate with 0.01M HCl was then added the wells, which were subsequently incubated at 37° C in 5% CO₂ overnight. Activity was characterized by measuring optical density at 570nm with reference absorbance read at 640nm. The half maximal inhibitory concentration (IC₅₀) and CC₅₀ were calculated using Sigma plot (SPSS) as Excel add-in ED50V10. Drug compounds were selected for combination studies with novobiocin or lopinavir-ritonavir using the CPE inhibition assay and the Loewe additivity index was calculated.

Virus yield reduction

[0034] Virus yield reduction was determined by quantitative reverse transcription-PCR (qRT-PCR) using total nucleic acid extracted from culture supernatants of the Vero cells infected by flaviviridae with different drugs or a dimethyl sulfoxide (DMSO) control at 6 days post-inoculation (dpi). β -actin gene was used as an internal control.

Plaque reduction

[0035] Plaque reduction (PRA) was determined by seeding Vero cells at 2×10^5 cells/well in 24-well tissue culture plates in MEM (Invitrogen, California, USA) with 10% FBS on the day prior to the assay. After 24 hours of incubation, 20-40 plaque-forming units of the virus were added to the cell monolayer with or without the addition of drug compounds and the plates further incubated for 1 hour at 37° C in 5% CO₂, after unbound viral particles were removed by aspiration of the media and washing with MEM. The monolayers were then overlaid with media containing 1% low melting agarose (Cambrex Corporation, New Jersey, USA) in MEM and appropriate concentrations of drug compounds and incubated as above for 96 hours. The wells were then fixed with 10% formaldehyde (BDH, Merck, Darmstadt, Germany) overnight. Following removal of the agarose plugs the cell monolayers were stained with 0.7% crystal violet (BDH, Merck) and the plaques counted. The percentage of plaque inhibition relative to the control (without compound addition) plates was determined for each drug compound concentration. Experiments were performed in triplicate and repeated twice for confirmation.

[0036] Surprisingly, novobiocin and lopinavir-ritonavir exhibited antiviral activity against all tested viruses, including DENV serotypes 1 to 4, JEV, WNV, and YFV. The CC_{50} of novobiocin and lopinavir-ritonavir was 850.5 $\mu\text{g/ml}$ (1,388.02 μM) and 30.0 $\mu\text{g/ml}$ in Vero cells, respectively, and was 1,103.18 $\mu\text{g/ml}$ (1,800.39 μM) and 32.12 $\mu\text{g/ml}$ in Huh-7 cells, respectively. Virus yield reduction studies showed a dose-dependent reduction in virus titer in novobiocin-treated ($IC_{50} = 26.12 \pm 0.33 \mu\text{g/ml}$ or 42.63 μM in Vero cells and $38.14 \pm 4.53 \mu\text{g/ml}$ or 62.24 μM in Huh-7 cells) and lopinavir-ritonavir-treated ($IC_{50} = 4.78 \pm 0.41 \mu\text{g/ml}$ in Vero cells and $3.31 \pm 0.36 \mu\text{g/ml}$ in Huh-7 cells) cells. The mean virus titer was significantly reduced ($P < 0.05$) at $\geq 25.0 \mu\text{g/ml}$ (40.8 μM) of novobiocin and at $\geq 50.0 \mu\text{g/ml}$ of lopinavir-ritonavir. Consistent with previous studies, amantadine, isatin, and levodopa exhibited antiviral activity against WNV, doxycycline and amantadine exhibited antiviral activity against DENV serotypes 1-4, and minocycline exhibited antiviral activity against JEV. Desmopressin, octreotide, rifampicin, selegiline, sirolimus, and tacrolimus did not exhibit apparent antiviral activity against any of the viruses.

[0037] Combination drug treatment studies of flavivirus-infected cells with novobiocin or lopinavir-ritonavir combined with secondary drugs such as amantadine, ciprofloxacin, doxycycline, isatin, levodopa, and/or ribavirin showed that such combinations provided an unexpected synergistic anti-flaviviral activity in vitro. Combination studies showed that the mean Loewe additivity indices of all of these combinations were < 1.0 , demonstrating that these drug combinations inhibited the in vitro replication of flaviviruses in a synergistic fashion.

[0038] It should be apparent to those skilled in the art that many more modifications besides those already described are possible without departing from the inventive concepts herein. The inventive subject matter, therefore, is not to be restricted except in the spirit of the appended claims. Moreover, in interpreting both the specification and the claims, all terms should be interpreted in the broadest possible manner consistent with the context. In particular, the terms “comprises” and “comprising” should be interpreted as referring to elements, components, or steps in a non-exclusive manner, indicating that the referenced elements, components, or steps may be present, or utilized, or combined with other elements, components, or steps that are not expressly referenced. Where the specification claims refers to at least one of something selected

from the group consisting of A, B, C and N, the text should be interpreted as requiring only one element from the group, not A plus N, or B plus N, etc.

CLAIMS

What is claimed is:

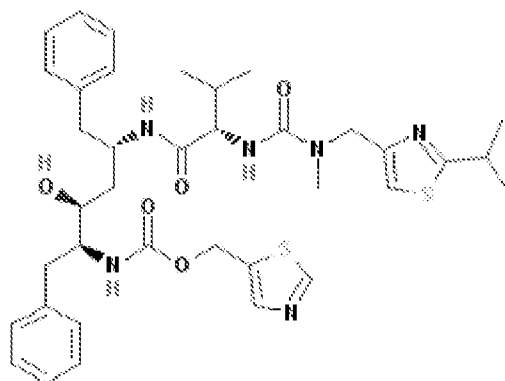
1. A composition for the treatment of flaviviridae infection, comprising:
novobiocin; and
a secondary antiviral or antibiotic compound,
wherein the novobiocin and the secondary antiviral or antibiotic compound exhibit a synergistic effect in inhibiting a virus of the flavivirus family.
2. The composition of claim 1, wherein the secondary antiviral or antibiotic compound is selected from the group consisting of ribavirin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, doxycycline, amantadine, levodopa, and isatin.
3. The composition of claim 1 or claim 2, wherein the virus of the flavivirus family is selected from the group consisting of dengue virus, Japanese encephalitis, West Nile virus, yellow fever virus, and hepatitis C virus.
4. The composition of one of claims 1 to 3, wherein the composition is formulated to provide novobiocin in a concentration range of about 1 μ M to about 1 mM.
5. A composition for the treatment of flaviviridae infection, comprising:
lopinavir-ritonavir; and
a secondary antiviral or antibiotic compound,
wherein the novobiocin and the secondary antiviral or antibiotic compound exhibit a synergistic effect in inhibiting a virus of the flavivirus family.
6. The composition of claim 5, wherein the secondary antiviral or antibiotic compound is selected from the group consisting of ribavirin, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and isatin.
7. The composition of claim 5 or claim 6, wherein the virus of the flavivirus family is selected from the group consisting of dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus.

8. The composition of one of claims 5 to 7, wherein the composition is formulated to provide lopinavir-ritonavir in a concentration range of about 1 μ M to about 1 mM.
9. A method of treating an infection caused by a member of the flavivirus family, comprising providing, to a person in need of treatment, a formulation comprising novobiocin.
10. The method of claim 9, wherein the formulation further comprises a secondary antiviral or antibiotic compound.
11. The method of claim 10, wherein the secondary antiviral or antibiotic compound is selected from the group consisting of ribavirin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and isatin.
12. The method of one of claims 9 to 11, wherein the formulation is configured to provide novobiocin in a concentration range of about 1 μ M to about 1 mM.
13. The method of one of claims 9 to 12, wherein the member of the flavivirus family is selected from the group consisting of dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus.
14. A method of treating an infection caused by a member of the flavivirus family, comprising providing, to a person in need of treatment, a formulation comprising lopinavir-ritonavir.
15. The method of claim 14, wherein the formulation further comprises a secondary antiviral or antibiotic compound.
16. The method of claim 15, wherein the secondary antiviral or antibiotic compound is selected from the group consisting of ribavirin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and isatin.
17. The method of one of claims 14 to 16, wherein the formulation is configured to provide lopinavir-ritonavir in a concentration range of about 1 μ M to about 1 mM.

18. The method of one of claims 14 to 17, wherein the member of the flavivirus family is selected from the group consisting of dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus.
19. A kit for treating an infection caused by a member of the flavivirus family, comprising:
novobiocin;
a secondary antiviral or antibiotic compound; and
instructions for use.
20. The kit of claim 19, wherein novobiocin and the secondary antiviral or antibiotic compound are provided as a single formulation.
21. The kit of claim 19, wherein novobiocin and the secondary antiviral or antibiotic compound are provided as separate formulations.
22. The kit of one of claims 19 to 21, wherein the secondary antiviral or antibiotic compound is selected from the group consisting of ribavirin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and isatin.
23. The kit of one of claims 19 to 22, wherein the kit is configured to provide novobiocin in a concentration range of about 1 μ M to about 1 mM.
24. The kit of one of claims 19 to 23, wherein the member of the flavivirus family is selected from the group consisting of dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus.
25. The kit of one of claims 19 to 24, wherein instructions for use comprise a dosing schedule effective in treating the infection.
26. A kit for treating an infection caused by a member of the flavivirus family, comprising:
lopinavir;
ritonavir;
a secondary antiviral or antibiotic compound; and
instructions for use.

27. The kit of claim 26, wherein lopinavir, ritonavir, and the secondary antiviral or antibiotic compound are provided as a single formulation.
28. The kit of claim 26, wherein lopinavir and ritonavir are provided as first formulation, and the secondary antiviral or antibiotic compound is provided as a second formulation.
29. The kit of claim 26, wherein lopinavir, ritonavir, and the secondary antiviral or antibiotic compound are provided as separate formulations.
30. The kit of one of claims 26 to 29, wherein the secondary antiviral or antibiotic compound is selected from the group consisting of ribavirin, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and isatin.
31. The kit of one of claims 26 to 30, wherein the kit is configured to provide lopinavir-ritonavir in a concentration range of about 1 μ M to about 1 mM.
32. The kit of one of claims 26 to 31, wherein the member of the flavivirus family is selected from the group consisting of dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus.
33. The kit of one of claims 26 to 32, wherein instructions for use comprise a dosing schedule effective in treating the infection.
34. Use of novobiocin for the manufacture of a medicament useful for treating an infection caused by a member of the flavivirus family.
35. The use of claim 34, wherein the medicament further comprises a secondary antiviral or antibiotic compound, and wherein novobiocin and the secondary antiviral or antibiotic compound provide a synergistic antiviral effect.
36. The use of claim 35, wherein the secondary antiviral or antibiotic compound is selected from the group consisting of ribavirin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and isatin.

37. The use of one of claims 34 to 36, wherein the medicament is formulated to provide novobiocin in a concentration range of about 1 μ M to about 1 mM.
38. The use of one of claims 34 to 37, wherein the member of the flavivirus family is selected from the group consisting of dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus.
39. Use of lopinavir-ritonavir for the manufacture of a medicament useful for treating an infection caused by a member of the flavivirus family.
40. The use of claim 40, wherein the medicament further comprises a secondary antiviral or antibiotic compound, and wherein lopinavir-ritonavir and the secondary antiviral or antibiotic compound provide a synergistic antiviral effect.
41. The use of claim 40, wherein the secondary antiviral or antibiotic compound is selected from the group consisting of ribavarin, ritonavir, ciprofloxacin, levofloxacin, moxifloxacin, tetracycline, tigecycline, minocycline, and doxycycline, amantadine, levodopa, and isatin.
42. The use of one of claims 39 to 41, wherein the medicament is formulated to provide lopinavir-ritonavir in a concentration range of about 1 μ M to about 1 mM.
43. The use of one of claims 39 to 42, wherein the member of the flavivirus family is selected from the group consisting of dengue virus, Japanese encephalitis virus, West Nile virus, yellow fever virus, and hepatitis C virus.

**FIG. 3**

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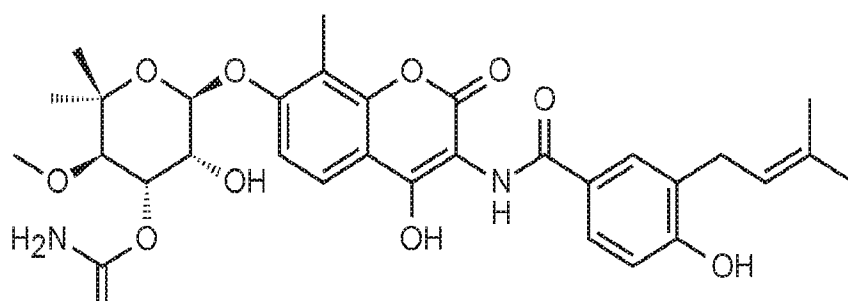


FIG. 1

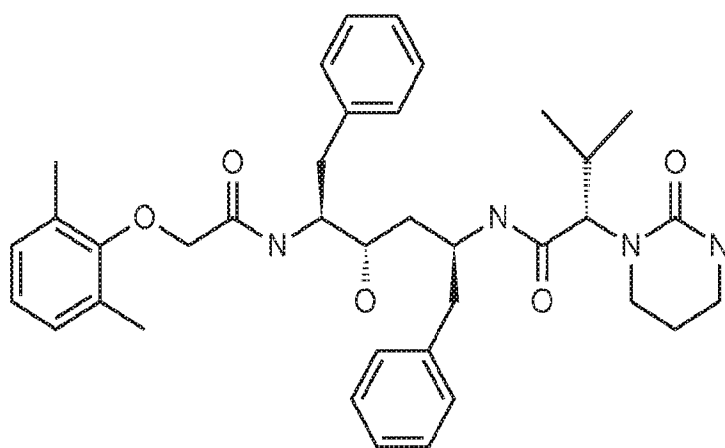


FIG. 2

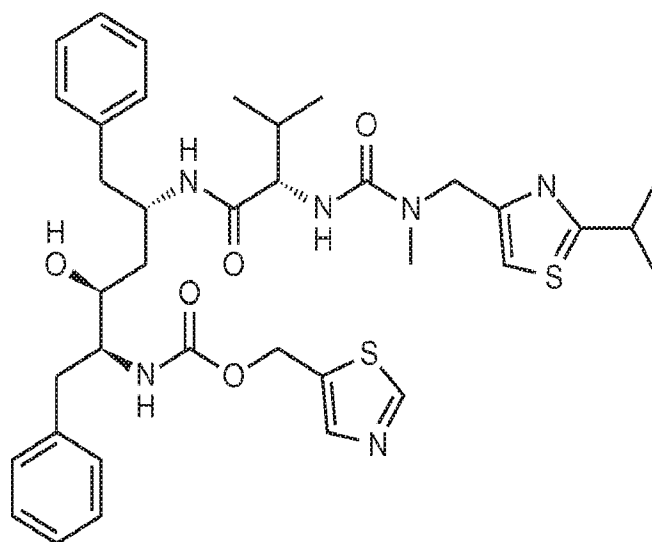


FIG. 3