(19) DANMARK

(10) DK/EP 2784075 T3

(12) Oversættelse af europæisk patentskrift

Patent- og Varemærkestyrelsen

(51) Int.Cl.: *C 07 D 401/14 (2006.01)* *A 61 K 31/4025 (2006.01)* *A 61 K 31/4178 (2006.01)*
A 61 P 31/12 (2006.01) *C 07 D 403/14 (2006.01)*

(45) Oversættelsen bekendtgjort den: **2016-07-04**

(80) Dato for Den Europæiske Patentmyndigheds bekendtgørelse om meddelelse af patentet: **2016-03-23**

(86) Europæisk ansøgning nr.: **14168065.2**

(86) Europæisk indleveringsdag: **2007-08-09**

(87) Den europæiske ansøgnings publiceringsdag: **2014-10-01**

(30) Prioritet: **2006-08-11 US 836996 P** **2007-08-08 US 835462**

(62) Stamansøgningsnr: **07800058.5**

(84) Designerede stater: **AT BE BG CH CY CZ DE DK EE ES FI FR GB GR HU IE IS IT LI LT LU LV MC MT NL PL PT RO SE SI SK TR**

(73) Patenthaver: **Bristol-Myers Squibb Holdings Ireland, Hinterbergstrasse 16, 6312 Steinhausen, Schweiz**

(72) Opfinder: **BACHAND, Carol, 119 Place Mercure, Candiac, Québec J5R 1B1, Canada**
Belema, Makonen, Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT Connecticut 06492, USA
Good, Andrew C., 52 High Hill Road, Wallingford, CT Connecticut 06492, USA
Goodrich, Jason, Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT Connecticut 06492, USA
Lopez, Omar D., Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT Connecticut 06492, USA
MARTEL, Alain, 29 Rue Monette, Delson, Quebec J5B 1G8, Canada
Meanwell, Nicholas A., Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT Connecticut 06492, USA
Nguyen, Van N., Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT Connecticut 06492, USA
Romine, Jeffrey Lee, Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT Connecticut 06492, USA
RUEDIGER, Edward H., 133 St. Charles, Greenfield Park, Quebec J4V 2K3, Canada
Snyder, Lawrence B., Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT Connecticut 06492, USA
St. Laurent, Denis R., Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT Connecticut 06492, USA
Yang, Fukang, Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT Connecticut 06492, USA
Langley, David R., Bristol-Myers Squibb Company, 5 Research Parkway, Walliingford, CT Connecticut 06492, USA
Wang, Gan, Bristol-Myers Squibb Company, 5 Research Parkway, Wallingford, CT Connecticut 06492, USA
Hamann, Lawrence G., c/o Novartis Institute for Biomedical Research, 250 Massachusetts Avenue, Cambridge, MA Massachusetts 02139, USA
Deon, Daniel H., 2122 Avenue Clifton, Montreal, Québec H4A 2N4, Canada
James, Clint A., 48 Danube, Candiac, Quebec J5R 6S4, Canada
LAVOIE, Rico, 16 Avenue de Bercy, , Candiac, Quebec J5R 4B7, USA

Fortsættes ...

(74) Fuldmægtig i Danmark: **Marks & Clerk (Luxembourg) LLP, 44 rue de la Vallée, B.P. 1775, L-1017 Luxembourg, Luxembourg**

(54) Benævnelse: **HEPATITIS C-VIRUSINHIBITORER**

(56) Fremdragne publikationer:
WO-A-2004/005264
WO-A-2006/133326
WO-A1-2005/082898
SCHUBERT, HERMANN ET AL: "Diimidazole. III. Synthese von aromatisch überbrückten 4(5),4'(5')-Diimidazolen", JOURNAL FUER PRAKTISCHE CHEMIE (LEIPZIG) , 22(3-4), 140-52 CODEN: JPCEAO; ISSN: 0021-8383, 1963, pages 140-152, XP002473762,

DESCRIPTION

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims the benefit of U.S. Provisional Application Serial Number 60/836,996 filed August 11, 2006.

[0002] The present disclosure is generally directed to an antiviral compound for use in therapy. The compound can inhibit the function of the NS5A protein encoded by Hepatitis C virus (HCV). HCV is a major human pathogen, infecting an estimated 170 million persons worldwide - roughly five times the number infected by human immunodeficiency virus type 1. A substantial fraction of these HCV infected individuals develop serious progressive liver disease, including cirrhosis and hepatocellular carcinoma.

[0003] Presently, the most effective HCV therapy employs a combination of alpha-interferon and ribavirin, leading to sustained efficacy in 40% of patients. Recent clinical results demonstrate that pegylated alpha-interferon is superior to unmodified alpha-interferon as monotherapy. However, even with experimental therapeutic regimens involving combinations of pegylated alpha-interferon and ribavirin, a substantial fraction of patients do not have a sustained reduction in viral load. Thus, there is a clear and long-felt need to develop effective therapeutics for treatment of HCV infection.

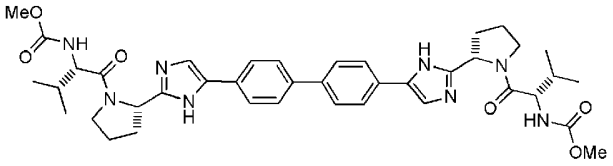
[0004] HCV is a positive-stranded RNA virus. Based on a comparison of the deduced amino acid sequence and the extensive similarity in the 5' untranslated region, HCV has been classified as a separate genus in the Flaviviridae family. All members of the Flaviviridae family have enveloped virions that contain a positive stranded RNA genome encoding all known virus-specific proteins *via* translation of a single, uninterrupted, open reading frame.

[0005] Considerable heterogeneity is found within the nucleotide and encoded amino acid sequence throughout the HCV genome. At least six major genotypes have been characterized, and more than 50 subtypes have been described. The major genotypes of HCV differ in their distribution worldwide, and the clinical significance of the genetic heterogeneity of HCV remains elusive despite numerous studies of the possible effect of genotypes on pathogenesis and therapy.

[0006] The single strand HCV RNA genome is approximately 9500 nucleotides in length and has a single open reading frame (ORF) encoding a single large polyprotein of about 3000 amino acids. In infected cells, this polyprotein is cleaved at multiple sites by cellular and viral proteases to produce the structural and non-structural (NS) proteins. In the case of HCV, the generation of mature non-structural proteins (NS2, NS3, NS4A, NS4B, NS5A, and NS5B) is effected by two viral proteases. The first one is believed to be a metalloprotease and cleaves at the NS2-NS3 junction; the second one is a serine protease contained within the N-terminal region of NS3 (also referred to herein as NS3 protease) and mediates all the subsequent cleavages downstream of NS3, both in cis, at the NS3-NS4A cleavage site, and in trans, for the remaining NS4A-NS4B, NS4B-NS5A, NS5A-NS5B sites. The NS4A protein appears to serve multiple functions, acting as a cofactor for the NS3 protease and possibly assisting in the membrane localization of NS3 and other viral replicase components. The complex formation of the NS3 protein with NS4A seems necessary to the processing events, enhancing the proteolytic efficiency at all of the sites. The NS3 protein also exhibits nucleoside triphosphatase and RNA helicase activities. NS5B (also referred to herein as HCV polymerase) is a RNA-dependent RNA polymerase that is involved in the replication of HCV.

[0007] Compounds useful for treating HCV-infected patients are desired which selectively inhibit HCV viral replication. In particular, compounds which are effective to inhibit the function of the NS5A protein are desired. The HCV NS5A protein is described, for example, in Tan, S.-L., Katzel, M.G. Virology 2001, 284, 1-12 ; and in Park, K.-J.; Choi, S.-H, J. Biological Chemistry 2003.

[0008] In a first aspect the present disclosure provides a compound which is methyl ((1S)-1-(((2S)-2-(5-(4'-(2-((2S)-1-((2S)-2-((methoxycarbonyl)amino)-3-methylbutanoyl)-2-pyrrolidinyl)-1H-imidazol-5-yl)-4-biphenylyl)-1H-imidazol-2-yl)-1-pyrrolidinyl)carbonyl)-2-methylpropyl)carbamate of Formula (I)



or a pharmaceutically acceptable salt thereof for use in therapy.

[0009] In a first embodiment of this aspect the pharmaceutically acceptable salt is a dihydrochloride salt.

[0010] In another embodiment of this aspect the use in therapy further comprises the use of one or two additional compounds having anti-HCV activity. In another embodiment at least one of the additional compounds is an interferon or a ribavirin. In another embodiment the interferon is selected from interferon alpha 2B, pegylated interferon alpha, consensus interferon, interferon alpha 2A, and lymphoblastoid interferon tau.

[0011] In another embodiment of this aspect the the use in therapy further comprises the use of one or two additional compounds having anti-HCV activity wherein at least one of the additional compounds is selected from interleukin 2, interleukin 6, interleukin 12, a compound that enhances the development of a type 1 helper T cell response, interfering RNA, anti-sense RNA, Imiqimod, ribavirin, an inosine 5'-monophospate dehydrogenase inhibitor, amantadine, and rimantadine.

[0012] In another embodiment of this aspect the the use in therapy further comprises the use of one or two additional compounds having anti-HCV activity wherein at least one of the additional compounds is effective to inhibit the function of a target selected from HCV metalloprotease, HCV serine protease, HCV polymerase, HCV helicase, HCV NS4B protein, HCV entry, HCV assembly, HCV egress, HCV NS5A protein, and IMPDH for the treatment of an HCV infection.

[0013] In another embodiment of this aspect the use in therapy further comprises administering one or two additional compounds having anti-HCV activity prior to, after or simultaneously with the compound of formula (I), or a pharmaceutically acceptable salt thereof.

[0014] Other embodiments of the present disclosure may comprise suitable combinations of two or more of embodiments disclosed herein.

[0015] Yet other embodiments and aspects of the disclosure will be apparent according to the description provided below.

[0016] The compounds of the present disclosure also exist as tautomers; therefore the present disclosure also encompasses all tautomeric forms.

[0017] The description of the present disclosure herein should be construed in congruity with the laws and principals of chemical bonding.

[0018] It should be understood that the compounds encompassed by the present disclosure are those that are suitably stable for use as pharmaceutical agent.

[0019] All patents, patent applications, and literature references cited in the specification are herein incorporated by reference in their entirety. In the case of inconsistencies, the present disclosure, including definitions, will prevail.

[0020] As used in the present specification, the following terms have the meanings indicated:

As used herein, the singular forms "a", "an", and "the" include plural reference unless the context clearly dictates otherwise.

[0021] Unless stated otherwise, all aryl, cycloalkyl, and heterocyclyl groups of the present disclosure may be substituted as described in each of their respective definitions. For example, the aryl part of an arylalkyl group may be substituted as described in the definition of the term 'aryl'.

[0022] Asymmetric centers exist in the compounds of the present disclosure. These centers are designated by the symbols "R" or "S", depending on the configuration of substituents around the chiral carbon atom. It should be understood that the disclosure encompasses all stereochemical isomeric forms, or mixtures thereof, which possess the ability to inhibit NS5A. Individual stereoisomers of compounds can be prepared synthetically from commercially available starting materials which contain chiral centers or by preparation of mixtures of enantiomeric products followed by separation such as conversion to a mixture of diastereomers followed by separation or recrystallization, chromatographic techniques, or direct separation of enantiomers on chiral chromatographic columns. Starting compounds of particular stereochemistry are either commercially available or can be made and resolved by techniques known in the art.

[0023] Certain compounds of the present disclosure may also exist in different stable conformational forms which may be separable. Torsional asymmetry due to restricted rotation about an asymmetric single bond, for example because of steric

hindrance or ring strain, may permit separation of different conformers. The present disclosure includes each conformational isomer of these compounds and mixtures thereof.

[0024] The term "compounds of the present disclosure", and equivalent expressions, are meant to embrace the compound of Formula (I), and pharmaceutically acceptable enantiomers, diastereomers, and salts thereof. Similarly, references to intermediates are meant to embrace their salts where the context so permits.

[0025] The compounds of the present disclosure can exist as pharmaceutically acceptable salts. The term "pharmaceutically acceptable salt," as used herein, represents salts or zwitterionic forms of the compounds of the present disclosure which are water or oil-soluble or dispersible, which are, within the scope of sound medical judgment, suitable for use in contact with the tissues of patients without excessive toxicity, irritation, allergic response, or other problem or complication commensurate with a reasonable benefit/risk ratio, and are effective for their intended use The salts can be prepared during the final isolation and purification of the compounds or separately by reacting a suitable nitrogen atom with a suitable acid. Representative acid addition salts include acetate, adipate, alginate, citrate, aspartate, benzoate, benzenesulfonate, bisulfate, butyrate, camphorate, camphorsulfonate; digluconate, dihydrobromide, diydrochloride, dihydroiodide, glycerophosphate, hemisulfate, heptanoate, hexanoate, formate, fumarate, hydrochloride, hydrobromide, hydro iodide, 2-hydroxyethanesulfonate, lactate, maleate, mesitylenesulfonate, methanesulfonate, naphthylenesulfonate, nicotinate, 2-naphthalenesulfonate, oxalate, palmoate, pectinate, persulfate, 3-phenylproprionate, picrate, pivalate, propionate, succinate, tartrate, trichloroacetate, trifluoroacetate, phosphate, glutamate, bicarbonate, para-toluenesulfonate, and undecanoate. Examples of acids which can be employed to form pharmaceutically acceptable addition salts include inorganic acids such as hydrochloric, hydrobromic, sulfuric, and phosphoric, and organic acids such as oxalic, maleic, succinic, and citric.

[0026] Basic addition salts can be prepared during the final isolation and purification of the compounds by reacting a carboxy group with a suitable base such as the hydroxide, carbonate, or bicarbonate of a metal cation or with ammonia or an organic primary, secondary, or tertiary amine. The cations of pharmaceutically acceptable salts include lithium, sodium, potassium, calcium, magnesium, and aluminum, as well as nontoxic quaternary amine cations such as ammonium, tetramethylammonium, tetraethylammonium, methylamine, dimethylamine, trimethylamine, triethylamine, diethylamine, ethylamine, tributylamine, pyridine, N,N-dimethylaniline, N-methylpiperidine, N-methylmorpholine, dicyclohexylamine, procaine, dibenzylamine, N,N-dibenzylphenethylamine, and N,N'-dibenzylethylenediamine. Other representative organic amines useful for the formation of base addition salts include ethylenediamine, ethanolamine, diethanolamine, piperidine, and piperazine.

[0027] When it is possible that, for use in therapy, therapeutically effective amounts of the compound of formula (I), as well as pharmaceutically acceptable salts thereof, may be administered as the raw chemical, it is possible to present the active ingredient as a pharmaceutical composition. Such pharmaceutical compositions include therapeutically effective amounts of the compound of formula (I) or pharmaceutically acceptable salts thereof, and one or more pharmaceutically acceptable carriers, diluents, or excipients. The term "therapeutically effective amount," as used herein, refers to the total amount of each active component that is sufficient to show a meaningful patient benefit, e.g., a reduction in viral load. When applied to an individual active ingredient, administered alone, the term refers to that ingredient alone. When applied to a combination, the term refers to combined amounts of the active ingredients that result in the therapeutic effect, whether administered in combination, serially, or simultaneously. The compound of formula (I) and pharmaceutically acceptable salts thereof, are as described above. The carrier(s), diluent(s), or excipient(s) must be acceptable in the sense of being compatible with the other ingredients of the formulation and not deleterious to the recipient thereof. The preparation of a pharmaceutical formulation includes admixing a compound of formula (I), or a pharmaceutically acceptable salt thereof, with one or more pharmaceutically acceptable carriers, diluents, or excipients. The term "pharmaceutically acceptable," as used herein, refers to those compounds, materials, compositions, and/or dosage forms which are, within the scope of sound medical judgment, suitable for use in contact with the tissues of patients without excessive toxicity, irritation, allergic response, or other problem or complication commensurate with a reasonable benefit/risk ratio, and are effective for their intended use.

[0028] Pharmaceutical formulations may be presented in unit dose forms containing a predetermined amount of active ingredient per unit dose. Dosage levels of between about 0.01 and about 250 milligram per kilogram ("mg/kg") body weight per day, preferably between about 0.05 and about 100 mg/kg body weight per day of the compounds of the present disclosure are typical in a monotherapy for the prevention and treatment of HCV mediated disease. Typically, the pharmaceutical compositions of this disclosure will be administered from about 1 to about 5 times per day or alternatively, as a continuous infusion. Such administration can be used as a chronic or acute therapy. The amount of active ingredient that may be combined with the carrier materials to produce a single dosage form will vary depending on the condition being treated, the severity of the condition, the time of administration, the route of administration, the rate of excretion of the compound employed, the duration of treatment, and the age, gender, weight, and condition of the patient. Preferred unit dosage formulations are those containing a daily dose or sub-dose, as herein above recited, or an appropriate fraction thereof, of an active ingredient. Treatment may be initiated with

small dosages substantially less than the optimum dose of the compound. Thereafter, the dosage is increased by small increments until the optimum effect under the circumstances is reached. In general, the compound is most desirably administered at a concentration level that will generally afford antivirally effective results without causing any harmful or deleterious side effects.

[0029] When the compositions of this disclosure comprise a combination of a compound of the present disclosure and one or more additional therapeutic or prophylactic agent, both the compound and the additional agent are usually present at dosage levels of between about 10 to 150%, and more preferably between about 10 and 80% of the dosage normally administered in a monotherapy regimen.

[0030] Pharmaceutical formulations may be adapted for administration by any appropriate route, for example by the oral (including buccal or sublingual), rectal, nasal, topical (including buccal, sublingual, or transdermal), vaginal, or parenteral (including subcutaneous, intracutaneous, intramuscular, intra-articular, intrasynovial, intrasternal, intrathecal, intralesional, intravenous, or intradermal injections or infusions) route. Such formulations may be prepared by any method known in the art of pharmacy, for example by bringing into association the active ingredient with the carrier(s) or excipient(s). Oral administration or administration by injection are preferred.

[0031] Pharmaceutical formulations adapted for oral administration may be presented as discrete units such as capsules or tablets; powders or granules; solutions or suspensions in aqueous or non-aqueous liquids; edible foams or whips; or oil-in-water liquid emulsions or water-in-oil emulsions.

[0032] For instance, for oral administration in the form of a tablet or capsule, the active drug component can be combined with an oral, non-toxic pharmaceutically acceptable inert carrier such as ethanol, glycerol, water, and the like. Powders are prepared by comminuting the compound to a suitable fine size and mixing with a similarly comminuted pharmaceutical carrier such as an edible carbohydrate, as, for example, starch or mannitol. Flavoring, preservative, dispersing, and coloring agent can also be present.

[0033] Capsules are made by preparing a powder mixture, as described above, and filling formed gelatin sheaths. Glidants and lubricants such as colloidal silica, talc, magnesium stearate, calcium stearate, or solid polyethylene glycol can be added to the powder mixture before the filling operation. A disintegrating or solubilizing agent such as agar-agar, calcium carbonate, or sodium carbonate can also be added to improve the availability of the medicament when the capsule is ingested.

[0034] Moreover, when desired or necessary, suitable binders, lubricants, disintegrating agents, and coloring agents can also be incorporated into the mixture. Suitable binders include starch, gelatin, natural sugars such as glucose or betalactose, corn sweeteners, natural and synthetic gums such as acacia, tragacanth or sodium alginate, carboxymethylcellulose, polyethylene glycol, and the like. Lubricants used in these dosage forms include sodium oleate, sodium chloride, and the like. Disintegrators include, without limitation, starch, methyl cellulose, agar, betonite, xanthan gum, and the like. Tablets are formulated, for example, by preparing a powder mixture, granulating or slugging, adding a lubricant and disintegrant, and pressing into tablets. A powder mixture is prepared by mixing the compound, suitable comminuted, with a diluent or base as described above, and optionally, with a binder such as carboxymethylcellulose, an aliginate, gelating, or polyvinyl pyrrolidone, a solution retardant such as paraffin, a resorption accelerator such as a quaternary salt and/or and absorption agent such as betonite, kaolin, or dicalcium phosphate. The powder mixture can be granulated by wetting with a binder such as syrup, starch paste, acadia mucilage, or solutions of cellulosic or polymeric materials and forcing through a screen. As an alternative to granulating, the powder mixture can be run through the tablet machine and the result is imperfectly formed slugs broken into granules. The granules can be lubricated to prevent sticking to the tablet forming dies by means of the addition of stearic acid, a stearate salt, talc, or mineral oil. The lubricated mixture is then compressed into tablets. The compounds of the present disclosure can also be combined with a free flowing inert carrier and compressed into tablets directly without going through the granulating or slugging steps. A clear or opaque protective coating consisting of a sealing coat of shellac, a coating of sugar or polymeric material, and a polish coating of wax can be provided. Dyestuffs can be added to these coatings to distinguish different unit dosages.

[0035] Oral fluids such as solution, syrups, and elixirs can be prepared in dosage unit form so that a given quantity contains a predetermined amount of the compound. Syrups can be prepared by dissolving the compound in a suitably flavored aqueous solution, while elixirs are prepared through the use of a non-toxic vehicle. Solubilizers and emulsifiers such as ethoxylated isostearyl alcohols and polyoxyethylene sorbitol ethers, preservatives, flavor additive such as peppermint oil or natural sweeteners, or saccharin or other artificial sweeteners, and the like can also be added.

[0036] Where appropriate, dosage unit formulations for oral administration can be microencapsulated. The formulation can also be prepared to prolong or sustain the release as for example by coating or embedding particulate material in polymers, wax, or

the like.

[0037] The compound of formula (I), and pharmaceutically acceptable salts thereof, can also be administered in the form of liposome delivery systems, such as small unilamellar vesicles, large unilamellar vesicles, and multilamellar vesicles. Liposomes can be formed from a variety of phopholipids, such as cholesterol, stearylamine, or phophatidylcholines.

[0038] The compound of formula (I) and pharmaceutically acceptable salts thereof may also be delivered by the use of monoclonal antibodies as individual carriers to which the compound molecules are coupled. The compound may also be coupled with soluble polymers as targetable drug carriers. Such polymers can include polyvinylpyrrolidone, pyran copolymer, polyhydroxypropylmethacrylamidephenol, polyhydroxyethylaspartamidephenol, or polyethyleneoxidepolylysine substituted with palitoyl residues. Furthermore, the compound may be coupled to a class of biodegradable polymers useful in achieving controlled release of a drug, for example, polylactic acid, polepsilon caprolactone, polyhydroxy butyric acid, polyorthoesters, polyacetals, polydihydropyrans, polycyanoacrylates, and cross-linked or amphipathic block copolymers of hydrogels.

[0039] Pharmaceutical formulations adapted for transdermal administration may be presented as discrete patches intended to remain in intimate contact with the epidermis of the recipient for a prolonged period of time. For example, the active ingredient may be delivered from the patch by iontophoresis as generally described in Pharmaceutical Research 1986, 3(6), 318.

[0040] Pharmaceutical formulations adapted for topical administration may be formulated as ointments, creams, suspensions, lotions, powders, solutions, pastes, gels, sprays, aerosols, or oils.

[0041] Pharmaceutical formulations adapted for rectal administration may be presented as suppositories or as enemas.

[0042] Pharmaceutical formulations adapted for nasal administration wherein the carrier is a solid include a course powder having a particle size for example in the range 20 to 500 microns which is administered in the manner in which snuff is taken, *i.e.,* by rapid inhalation through the nasal passage from a container of the powder held close up to the nose. Suitable formulations wherein the carrier is a liquid, for administration as a nasal spray or nasal drops, include aqueous or oil solutions of the active ingredient.

[0043] Pharmaceutical formulations adapted for administration by inhalation include fine particle dusts or mists, which may be generated by means of various types of metered, dose pressurized aerosols, nebulizers, or insufflators.

[0044] Pharmaceutical formulations adapted for vaginal administration may be presented as pessaries, tampons, creams, gels, pastes, foams, or spray formulations.

[0045] Pharmaceutical formulations adapted for parenteral administration include aqueous and non-aqueous sterile injection solutions which may contain anti-oxidants, buffers, bacteriostats, and soutes which render the formulation isotonic with the blood of the intended recipient; and aqueous and non-aqueous sterile suspensions which 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, and may be stored in a freeze-dried (lyophilized) condition requiring only the addition of the sterile liquid carrier, for example water for injections, immediately prior to use. Extemporaneous injection solutions and suspensions may be prepared from sterile powders, granules, and tablets.

[0046] It should be understood that in addition to the ingredients particularly mentioned above, the formulations may include other agents conventional in the art having regard to the type of formulation in question, for example those suitable for oral administration may include flavoring agents.

[0047] The term "patient" includes both human and other mammals.

[0048] The term "treating" refers to: (i) preventing a disease, disorder or condition from occurring in a patient that may be predisposed to the disease, disorder, and/or condition but has not yet been diagnosed as having it; (ii) inhibiting the disease, disorder, or condition, i.e., arresting its development; and (iii) relieving the disease, disorder, or condition, i.e., causing regression of the disease, disorder, and/or condition.

[0049] The compounds of the present disclosure can also be administered with a cyclosporin, for example, cyclosporin A. Cyclosporin A has been shown to be active against HCV in clinical trials (Hepatology 2003, 38, 1282; Biochem. Biophys. Res. Commun. 2004, 313, 42; J. Gastroenterol. 2003, 38, 567).

[0050] Table 1 below lists some illustrative examples of compounds that can be administered with the compounds of this disclosure. The compounds of the disclosure can be administered with other anti-HCV activity compounds in combination therapy, either jointly or separately, or by combining the compounds into a composition.

Table 1

Brand Name	*Physiological Class*	*Type of Inhibitor or Target*	*Source Company*
NIM811		Cyclophilin Inhibitor	Novartis
Zadaxin		Immunomodulator	Sciclone
Suvus		Methylene blue	Bioenvision
Actilon (CPG10101)		TLR9 agonist	Coley
Batabulin (T67)	Anticancer	β-tubulin inhibitor	Tularik Inc., South San Francisco, CA
ISIS 14803	Antiviral	antisense	ISIS Pharmaceutica ls Inc, Carlsbad, CA/Elan Phamaceutical s Inc., New York, NY
Summetrel	Antiviral	antiviral	Endo Pharmaceutica ls Holdings Inc., Chadds Ford, PA
GS-9132 (ACH-806)	Antiviral	HCV Inhibitor	Achillion / Gilead
Pyrazolopyrimidine compounds and salts From WO-2005047288 26 May 2005	Antiviral	HCV Inhibitors	Arrow Therapeutics Ltd.
Levovirin	Antiviral	IMPDH inhibitor	Ribapharm Inc., Costa Mesa, CA
Merimepodib (VX-497)	Antiviral	IMPDH inhibitor	Vertex Pharmaceutica ls Inc., Cambridge, MA
XTL-6865 (XTL-002)	Antiviral	monoclonal antibody	XTL Biopharmaceu ticals Ltd., Rehovot, Isreal
Telaprevir (VX-950, LY-570310)	Antiviral	NS3 serine protease inhibitor	Vertex Pharmaceutica ls Inc., Cambridge, MA/ Eli Lilly and Co. Inc., Indianapolis, IN
HCV-796	Antiviral	NS5B Replicase Inhibitor	Wyeth / Viropharma
NM-283	Antiviral	NS5B Replicase Inhibitor	Idenix / Novartis
GL-59728	Antiviral	NS5B Replicase Inhibitor	Gene Labs / Novartis
GL-60667	Antiviral	NS5B Replicase Inhibitor	Gene Labs / Novartis
2'C MeA	Antiviral	NS5B Replicase Inhibitor	Gilead
PSI 6130	Antiviral	NS5B Replicase Inhibitor	Roche
R1626	Antiviral	NS5B Replicase Inhibitor	Roche
2'C Methyl adenosine	Antiviral	NS5B Replicase Inhibitor	Merck
JTK-003	Antiviral	RdRp inhibitor	Japan Tobacco Inc., Tokyo, Japan
Levovirin	Antiviral	ribavirin	ICN Pharmaceutica ls, Costa Mesa, CA
Ribavirin	Antiviral	ribavirin	Schering-Plough Corporation, Kenilworth, NJ

Brand Name	*Physiological Class*	*Type of Inhibitor or Target*	*Source Company*
Viramidine	Antiviral	Ribavirin Prodrug	Ribapharm Inc., Costa Mesa, CA
Heptazyme	Antiviral	ribozyme	Ribozyme Pharmaceutica ls Inc., Boulder, CO
BILN-2061	Antiviral	serine protease inhibitor	Boehringer Ingelheim Pharma KG, Ingelheim, Germany
SCH 503034	Antiviral	serine protease inhibitor	Schering Plough
Zadazim	Immune modulator	Immune modulator	SciClone Pharmaceutica ls Inc., San Mateo, CA
Ceplene	Immunomodulator	immune modulator	Maxim Pharmaceutica ls Inc., San Diego, CA
CellCept	Immunosuppressant	HCV IgG immunosuppressant	F. Hoffmann-La Roche LTD, Basel, Switzerland
Civacir	Immunosuppressant	HCV IgG immunosuppressant	Nabi Biopharmaceu ticals Inc., Boca Raton, FL
Albuferon - a	Interferon	albumin IFN-α2b	Human Genome Sciences Inc., Rockville, MD
Infergen A	Interferon	IFN alfacon-1	InterMune Pharmaceutica ls Inc., Brisbane, CA
Omega IFN	Interferon	IFN-ω	Intarcia Therapeutics
IFN-β and EMZ701	Interferon	IFN-β and EMZ701	Transition Therapeutics Inc., Ontario, Canada
Rebif	Interferon	IFN-β1a	Serono, Geneva, Switzerland
Roferon A	Interferon	IFN-α2a	F. Hoffmann-La Roche LTD, Basel, Switzerland
Intron A	Interferon	IFN-α2b	Schering-Plough Corporation, Kenilworth, NJ
Intron A and Zadaxin	Interferon	IFN-α2b/α1-thymosin	RegeneRx Biopharmiceu ticals Inc., Bethesda, MD/ SciClone Pharmaceutica 1s Inc, San Mateo, CA
Rebetron	Interferon	IFN-α2b/ribavirin	Schering-Plough Corporation, Kenilworth, NJ
Actimmune	Interferon	INF-γ	InterMune Inc., Brisbane, CA
Interferon-β	Interferon	Interferon-β-1a	Serono
Multiferon	Interferon	Long lasting IFN	Viragen/Valen tis
Wellferon	Interferon	lymphoblastoid IFN-αn1	GlaxoSmithKl ine plc, Uxbridge, UK
Omniferon	Interferon	natural IFN-α	Viragen Inc., Plantation, FL
Pegasys	Interferon	PEGylated IFN-α2a	F. Hoffmann-La Roche LTD, Basel, Switzerland
Pegasys and Ceplene	Interferon	PEGylated IFN-α2a/ immune modulator	Maxim Pharmaceutica ls Inc., San Diego, CA
Pegasys and Ribavirin	Interferon	PEGylated IFN-α2a/ribavirin	F. Hoffmann-La Roche LTD, Basel, Switzerland
PEG-Intron	Interferon	PEGylated IFN-α2b	Schering-Plough Corporation, Kenilworth, NJ
PEG-Intron / Ribavirin	Interferon	PEGylated IFN-α2b/ribavirin	Schering-Plough Corporation, Kenilworth, NJ

Brand Name	*Physiological Class*	*Type of Inhibitor or Target*	*Source Company*
IP-501	Liver protection	antifibrotic	Indevus Pharmaceutica ls Inc., Lexington, MA
IDN-6556	Liver protection	caspase inhibitor	Idun Pharmaceutica ls Inc., San Diego, CA
ITMN-191 (R-7227)	Antiviral	serine protease inhibitor	InterMune Pharmaceutica ls Inc., Brisbane, CA
GL-59728	Antiviral	NS5B Replicase Inhibitor	Genelabs
ANA-971	Antiviral	TLR-7 agonist	Anadys

[0051] The compounds of the present disclosure may also be used as laboratory reagents. Compounds may be instrumental in providing research tools for designing of viral replication assays, validation of animal assay systems and structural biology studies to further enhance knowledge of the HCV disease mechanisms. Further, the compounds of the present disclosure are useful in establishing or determining the binding site of other antiviral compounds, for example, by competitive inhibition.

[0052] The compounds of this disclosure may also be used to treat or prevent viral contamination of materials and therefore reduce the risk of viral infection of laboratory or medical personnel or patients who come in contact with such materials, e.g., blood, tissue, surgical instruments and garments, laboratory instruments and garments, and blood collection or transfusion apparatuses and materials.

[0053] This disclosure is intended to encompass the compound having formula (I) when prepared by synthetic processes or by metabolic processes including those occurring in the human or animal body (*in vivo*) or processes occurring *in vitro*.

[0054] The abbreviations used in the present application, including particularly in the illustrative schemes and examples which follow, are well-known to those skilled in the art. Some of the abbreviations used are as follows: HATU for O-(7-azabenzotriazol-1-yl)-N,N,N',N'-tetramethyluronium hexafluorophosphate; Boc or BOC for tert-butoxycarbonyl; NBS for N-bromosuccinimide; tBu or t-Bu for tert-butyl; SEM for -(trimethylsilyl)ethoxymethyl; DMSO for dimethylsulfoxide; MeOH for methanol; TFA for trifluoroacetic acid; RT for room temperature or retention time (context will dictate); t_R for retention time; EDCI for 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride; DMAP for 4-dimethylaminopyridine; THF for tetrahydrofuran; DBU for 1,8-diazabicyclo[5.4.0]undec-7-ene; *t*-Bu; DEA for diethylamine; HMDS for hexamethyldisilazide; DMF for N,N-dimethylformamide; Bzl for benzyl; EtOH for ethanol; iPrOH or i-PrOH for isopropanol; Me_2S for dimethylsulfide; Et_3N or TEA for triethylamine; Ph for phenyl; OAc for acetate; EtOAc for ethyl acetate; dppf for 1,1'-bis(diphenylphosphino)ferrocene; iPr_2EtN or DIPEA for diisopropylethylamine; Cbz for carbobenzyloxy; n-BuLi for n-butyllithium; ACN for acetonitrile; h or hr for hours; m or min for minutes; s for seconds; LiHMDS for lithium hexamethyldisilazide; DIBAL for diisobutyl aluminum hydride; TBDMSCl for tert-butyldimethylsilyl chloride; Me for methyl; ca. for about; OAc for acetate; iPr for isopropyl; Et for ethyl; Bn for benzyl; and HOAT for 1-hydroxy-7-azabenzotriazole.

[0055] The abbreviations used in the present application, including particularly in the illustrative schemes and examples which follow, are well-known to those skilled in the art.

EXAMPLES

[0056] The present disclosure will now be described in connection with certain embodiments which are not intended to limit its scope. On the contrary, the present disclosure covers all alternatives, modifications, and equivalents as can be included within the scope of the claims. Thus, the following examples, which include specific embodiments, will illustrate one practice of the present disclosure, it being understood that the examples are for the purposes of illustration of certain embodiments and are presented to provide what is believed to be the most useful and readily understood description of its procedures and conceptual aspects.

[0057] Solution percentages express a weight to volume relationship, and solution ratios express a volume to volume relationship, unless stated otherwise. Nuclear magnetic resonance (NMR) spectra were recorded either on a Bruker 300, 400, or

500 MHz spectrometer; the chemical shifts (δ) are reported in parts per million. Flash chromatography was carried out on silica gel (SiO_2) according to Still's flash chromatography technique (J. Org. Chem. 1978, 43, 2923).

[0058] Purity assessment and low resolution mass analysis were conducted on a Shimadzu LC system coupled with Waters Micromass ZQ MS system. It should be noted that retention times may vary slightly between machines. The LC conditions employed in determining the retention time (RT) were:

Condition 1

Column	= Phenomenex-Luna 3.0X 50 mm S10
Start %B	= 0
Final %B	= 100
Gradient time	= 2 min
Stop time	= 3 min
Flow Rate	= 4 mL/min
Wavelength	= 220 nm
Solvent A	= 0.1% TFA in 10% methanol/90%H_2O
Solvent B	= 0.1% TFA in 90% methanol/10% H_2O

Condition 2

Column	= Phenomenex-Luna 4.6X50 mm S10
Start %B	= 0
Final %B	= 100
Gradient time	= 2 min
Stop time	= 3 min
Flow Rate	= 5 mL/min
Wavelength	= 220 nm
Solvent A	= 0.1% TFA in 10% methanol/90%H_2O
Solvent B	= 0.1% TFA in 90% methanol/10% H_2O

Condition 3

Column	= HPLC XTERRA C18 3.0 x 50mm S7
Start %B	= 0
Final %B	= 100
Gradient time	= 3 min
Stop time	= 4 min
Flow Rate	= 4 mL/min
Wavelength	= 220 nm
Solvent A	= 0.1% TFA in 10% methanol/90%H_2O
Solvent B	= 0.1% TFA in 90% methanol/10% H_2O

Method A: LCMS - Xterra MS C-18 3.0 x 50mm, 0 to 100% B over 30.0 minute gradient, 1 minute hold time, A = 5% acetonitrile, 95% water, 10mm ammonium acetate, B = 95% acetonitrile, 5% water, 10mm ammonium acetate.

Method B: HPLC - X-Terra C-18 4.6 x 50mm, 0 to 100% B over 10.0 minute gradient, 1 minute hold time, A = 10% methanol 90% water 0.1% TFA, B = 90% methanol 10% water 0.1% TFA

Method C: HPLC - YMC C-18 4.6 x 50mm, 0 to 100% B over 10.0 minute gradient, 1 minute hold time, A = 10% methanol 90% water 0.2% H_3PO_4, B = 90% methanol 10% water 0.2% H_3PO_4.

Method D: HPLC - Phenomenex C-18 4.6 x 150mm, 0 to 100% B over 10.0 minute gradient, 1 minute hold time, A = 10% methanol 90% water 0.2% H_3PO_4, B = 90% methanol 10% water 0.2% H_3PO_4

Method E: LCMS - Gemini C-18 4.6 x 50mm, 0 to 100% B over 10.0 minute gradient, 1 minute hold time, A = 5% acetonitrile, 95%

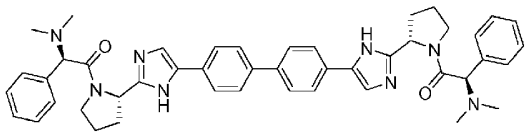
water, 10mm ammonium acetate, B = 95% acetonitrile, 5% water, 10mm ammonium acetate.

Method F: LCMS-Luna C-18 3.0 x 50mm, 0 to 100% B over 7.0 minute gradient, 1 minute hold time, A = 5% acetonitrile, 95% water, 10mm ammonium acetate, B = 95% acetonitrile, 5% water, 10mm ammonium acetate.

Example 1 (Reference Example)

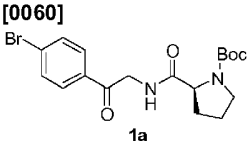
(1R,1'R)-2,2'-(4,4'-biphenyldiylbis(1H-imidazole-5,2-diyl(2S)-2,1-pyrrolidinediyl))bis(N,N-dimethyl-2-oxo-1-phenylethanamine)

[0059]



Example 1, Step a

[0060]

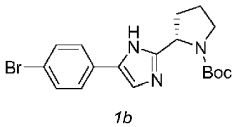


1a

[0061] *N,N*-Diisopropylethylamine (18 mL, 103.3 mmol) was added dropwise, over 15 minutes, to a heterogeneous mixture of *N*-Boc-L-proline (7.139 g, 33.17 mmol), HATU (13.324 g, 35.04 mmol), the HCl salt of 2-amino-1-(4-bromophenyl)ethanone (8.127 g, 32.44 mmol), and DMF (105 mL), and stirred at ambient condition for 55 minutes. Most of the volatile component was removed *in vacuo*, and the resulting residue was partitioned between ethyl acetate (300 mL) and water (200 mL). The organic layer was washed with water (200 mL) and brine, dried ($MgSO_4$), filtered, and concentrated *in vacuo*. A silica gel mesh was prepared from the residue and submitted to flash chromatography (silica gel; 50-60 % ethyl acetate/hexanes) to provide ketoamide 1a as a white solid (12.8 g). ^{1}H NMR (DMSO-d_6, δ = 2.5 ppm, 400 MHz): δ 8.25-8.1 (m, 1H), 7.92 (br d, *J* = 8.0, 2H), 7.75 (br d, *J* = 8.6, 2H), 4.61 (dd, *J* = 18.3,5.7, 1H), 4.53 (dd, *J* = 18.1, 5.6, 1H), 4.22-4.12 (m, 1H), 3.43-3.35 (m, 1H), 3.30-3.23 (m, 1H), 2.18-2.20 (m, 1H), 1.90-1.70 (m, 3H), 1.40/1.34 (two app br s, 9H). LC (Cond. 1): RT = 1.70 min; LC/MS: Anal. Calcd. for $[M+Na]^+$ $C_{18}H_{23}BrN_2NaO_4$: 433.07; found 433.09.

Example 1, Step b

[0062]



1b

[0063] A mixture of ketoamide 1a (12.8 g, 31.12 mmol) and NH_4OAc (12.0 g, 155.7 mmol) in xylenes (155 mL) was heated in a sealed tube at 140 °C for 2 hours. The volatile component was removed *in vacuo*, and the residue was partitioned carefully between ethyl acetate and water, whereby enough saturated $NaHCO_3$ solution was added so as to make the pH of the aqueous phase slightly basic after the shaking of the biphasic system. The layers were separated, and the aqueous layer was extracted

with an additional ethyl acetate. The combined organic phase was washed with brine, dried ($MgSO_4$), filtered, and concentrated *in vacuo*. The resulting material was recrystallized from ethyl acetate/hexanes to provide two crops of imidazole 1b as a light-yellow dense solid, weighing 5.85 g. The mother liquor was concentrated *in vacuo* and submitted to a flash chromatography (silica gel; 30% ethyl acetate/hexanes) to provide an additional 2.23 g of imidazole 1b. ^1H NMR (DMSO-d_6, δ = 2.5 ppm, 400 MHz): δ 12.17/11.92/11.86 (m, 1H), 7.72-7.46/7.28 (m, 5H), 4.86-4.70 (m, 1H), 3.52 (app br s, 1H), 3.36 (m, 1H), 2.30-1.75 (m, 4H), 1.40/1.15 (app br s, 9H). LC (Cond. 1): RT = 1.71 min; >98% homogeneity index; LC/MS: Anal. Calcd. for $[M+H]^+$ $C_{18}H_{23}BrN_3O_2$: 392.10; found 391.96; HRMS: Anal. Calcd. for $[M+H]^+$ $C_{18}H_{23}BrN_3O_2$: 392.0974; found 392.0959

[0064] The optical purity of the two samples of 1b were assessed using the chiral HPLC conditions noted below (ee > 99% for the combined crops; ee = 96.7% for the sample from flash chromatography):

Column: Chiralpak AD, 10 um, 4.6 x 50 mm

Solvent: 2% ethanol/heptane (isocratic)

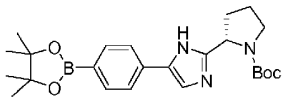
Flow rate: 1 mL/min

Wavelength: either 220 or 254 nm

Relative retention time: 2.83 minutes (*R*), 5.34 minutes (*S*)

Example 1, Step c

[0065]



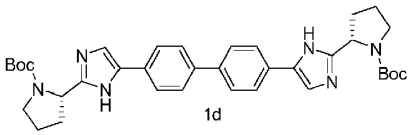
1c

[0066] $Pd(Ph_3P)_4$ (469 mg, 0.406 mmol) was added to a pressure tube containing a mixture of bromide 1b (4.008 g, 10.22 mmol), bis(pinacolato)diboron (5.422 g, 21.35 mmol), potassium acetate (2.573g, 26.21 mmol) and 1,4-dioxane (80 mL). The reaction flask was purged with nitrogen, capped and heated with an oil bath at 80 °C for 16.5 hours. The reaction mixture was filtered and the filtrate was concentrated *in vacuo*. The crude material was partitioned carefully between CH_2Cl_2 (150 mL) and an aqueous medium (50 mL water + 10 mL saturated $NaHCO_3$ solution). The aqueous layer was extracted with CH_2Cl_2, and the combined organic phase was dried ($MgSO_4$), filtered, and concentrated *in vacuo*. The resulting material was purified with flash chromatography (sample was loaded with eluting solvent; 20-35% ethyl acetate/CH_2Cl_2) to provide boronate 1c, contaminated with pinacol, as an off-white dense solid; the relative mole ratio of 1c to pinacol was about 10:1 (^1H NMR). The sample weighed 3.925 g after ~2.5 days exposure to high vacuum. ^1H NMR (DMSO-d_6, δ = 2.5 ppm, 400 MHz): 12.22/11.94/ 11.87 (m, 1H), 7.79-7.50/ 7.34-7.27 (m, 5H), 4.86-4.70 (m, 1H), 3.52 (app br s, 1H), 3.36 (m, 1H), 2.27-1.77 (m, 4H), 1.45-1.10 (m, 21H). LC (Cond. 1): RT = 1.64 min; LC/MS: Anal. Calcd. for $[M+H]^+$ $C_{24}H_{15}BN_3O_4$: 440.27; found 440.23.

Example 1, Step d

di-tert-butyl (2S,2'S)-2,2'-(4,4'-biphenyldiylbis(1H-imidazole-5,2-diyl))di(1-pyrrolidinecarboxylate)

[0067]



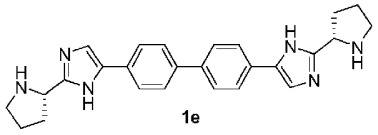
1d

[0068] $Pd(Ph_3P)_4$ (59.9 mg, 0.0518 mmol) was added to a mixture of bromide 1b (576.1 mg, 1.469 mmol), boronate 1c (621.8 mg, 1.415 mmol), $NaHCO_3$ (400.4 mg, 4.766 mmol) in 1,2-dimethoxyethane (12 mL) and water (4 mL). The reaction mixture was flushed with nitrogen, heated with an oil bath at 80 °C for 5.75 hours, and then the volatile component was removed *in vacuo*. The residue was partitioned between 20% methanol/ $CHCl_3$ (60 mL) and water (30 mL), and the aqueous phase was extracted with 20% methanol/$CHCl_3$ (30 mL). The combined organic phase was washed with brine, dried ($MgSO_4$), filtered, and concentrated *in vacuo*. A silica gel mesh was prepared from the resulting crude material and submitted to flash chromatography (ethyl acetate) to provide dimer 1d, contaminated with PH_3PO, as an off-white solid (563 mg). ^1H NMR (DMSO-d_6, δ = 2.5 ppm, 400 MHz): δ 12.21-12-16/11.95-11.78 (m, 2H), 7.85-7.48/7.32-7.25 (m, 10H), 4.90-4.71 (m, 2H), 3.60-3.32 (m, 4H), 2.30-1.79 (m, 8H), 1.46-1.10 (m, 18H). LC (Cond. 1b): RT = 1.77 min; LC/MS: Anal. Calcd. for $[M+H]^+$ $C_{36}H_{45}BN_6O_4$: 625.35; found 625.48.

Example 1, Step e

5,5'-(4,4'-biphenyldiyl)bis(2-((2S)-2-pyrrolidinyl)-1H-imidazole)

[0069]



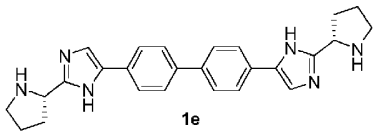
1e

[0070] A mixture of carbamate Id (560 mg) and 25% TFA/CH_2Cl_2 (9.0 mL) was stirred at ambient condition for 3.2 hours. The volatile component was removed *in vacuo*, and the resulting material was free based using an MCX column (methanol wash; 2.0 M NH_3/methanol elution) to provide pyrrolidine 1e as a dull yellow solid (340 mg). ^1H NMR (DMSO-d_6, δ = 2.5 ppm, 400 MHz): δ 11.83 (br s, 2H), 7.80 (d, *J* = 8.1, 4H), 7.66 (d, *J* = 8.3, 4H), 7.46 (br s, 2H), 4.16 (app t, *J* = 7.2, 2H), 2.99-2.69 (m, 6H), 2.09-2.00 (m, 2H), 1.94-1.66 (m, 6H). LC (Cond. 1): RT = 1.27 min; > 98% homogeneity index; LC/MS: Anal. Calcd. for $[M+H]^+$ $C_{26}H_{29}N_6$: 425.25; found 425.25; HRMS: Anal. Calcd. for $[M+H]^+$ $C_{26}H_{29}N_6$: 425.2454; found 425.2448

Alternative Synthesis of Example 1, Step e

5,5'-(4,4'-biphenyldiyl)bis(2-((2S)-2-pyrrolidinyl)-1H-imidazole)

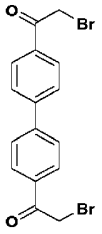
[0071]



1e

Example A-1e-1

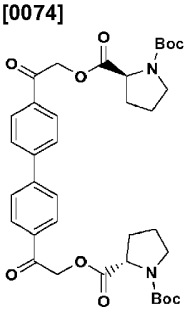
[0072]



[0073] AlL, 3-neck round bottom flask, fitted with a nitrogen line, overhead stirrer and thermocouple was charged with 20 g (83.9 mmol, 1 equiv) 1,1'-(biphenyl-4,4'-diyl)diethanone, 200 mL CH_2Cl_2 and 8.7 mL (27.1g, 169.3 mmol, 2.02 quiv) bromine. The mixture was allowed to stir under nitrogen for about 20h under ambient conditions. The resulting slurry was charged with 200 mL CH_2Cl_2 and concentrated down to about 150 mL *via* vacuum distillation. The slurry was then solvent exchanged into THF to a target volume of 200 mL *via* vacuum distillation. The slurry was cooled to 20-25 °C over 1h and allowed to stir at 20-25 °C for an additional hour. The off-white crystalline solids were filtered and washed with 150 mL CH_2Cl_2. The product was dried under vacuum at 60 °C to provide 27.4 g (69.2 mmol, 82%) of the desired product: ^{1}H NMR (400 MHz, $CDCl_3$) δ 7.95-7.85 (m, 4H), 7.60-7.50 (m, 4H), 4.26 (s, 4H); ^{13}C NMR (100 MHz, $CDCl_3$) δ 191.0, 145.1, 133.8, 129.9, 127.9, 30.8; IR (KBr, cm-1) 3007, 2950, 1691, 1599, 1199; Anal calcd for $C_{16}H_{12}Br_2O_2$: C, 48.52; H, 3.05; Br, 40.34. Found: C, 48.53; H, 3.03; Br, 40.53. HRMS calcd for $C_{16}H_{13}Br_2O_2$ (M+H; DCI$^+$): 394.9282. Found: 394.9292. mp 224-226 °C.

Example A-1e-2

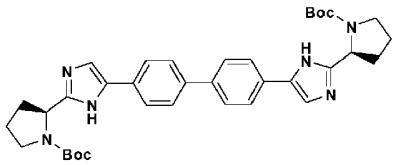
[0074]



[0075] A 500 mL jacketed flask, fitted with a nitrogen line, thermocouple and overhead stirrer, was charged with 20 g (50.5 mmol, 1 equiv) of Example A-1e-1, 22.8 g (105.9 moles, 2.10 equiv) 1-(*tert*-butoxycarbonyl)-L-proline, and 200 mL acetonitrile. The slurry was cooled to 20 °C followed by the addition of 18.2 mL (13.5 g, 104.4 mmol, 2.07 equiv) DIPEA. The slurry was warmed to 25 °C and allowed to stir for 3h. The resulting clear, organic solution was washed with 3 x 100 mL 13 wt% aqueous NaCl. The rich acetonitrile solution was solvent exchanged into toluene (target volume = 215 mL) by vacuum distillation until there was less than 0.5 vol% acetonitrile.

Example A-1e-3

[0076]

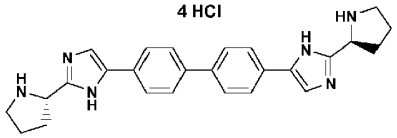


[0077] The above toluene solution of Example A-1e-2 was charged with 78 g (1.011 moles, 20 equiv) ammonium acetate and heated to 95-100 °C. The mixture was allowed to stir at 95-100 °C for 15h. After reaction completion, the mixture was cooled to

70-80 °C and charged with 7 mL acetic acid, 40 mL n-butanol, and 80 mL of 5 vol% aqueous acetic acid. The resulting biphasic solution was split while maintaining a temperature > 50 °C. The rich organic phase was charged with 80 mL of 5 vol% aqueous acetic acid, 30 mL acetic acid and 20 mL n-butanol while maintaining a temperature > 50 °C. The resulting biphasic solution was split while maintaining a temperature > 50 °C and the rich organic phase was washed with an additional 80 mL of 5 vol% aqueous acetic acid. The rich organic phase was then solvent exchanged into toluene to a target volume of 215 mL by vacuum distillation. While maintaining a temperature > 60 °C, 64 mL MeOH was charged. The resulting slurry was heated to 70-75 °C and aged for 1h. The slurry was cooled to 20-25 °C over 1h and aged at that temperature for an additional hour. The slurry was filtered and the cake was washed with 200 mL 10:3 toluene:MeOH. The product was dried under vacuum at 70 °C, resulting in 19.8 g (31.7 mmol, 63%) of the desired product: ^{1}H NMR (400 MHz, DMSO-d_6) δ 13.00-11.00 (s, 2H), 7.90-7.75 (m, 4H), 7.75-7.60 (m, 4H), 7.60-7.30 (s, 2H), 4.92-4.72 (m, 2H), 3.65-3.49 (m, 2H), 3.49-3.28 (m, 2H), 2.39-2.1 (m, 2H), 2.10-1.87 (m, 6H), 1.60-1.33 (s, 8H), 1.33-1.07 (s, 10H); ^{13}C NMR (100 MHz, DMSO-d_6) δ 154.1, 153.8, 137.5, 126.6, 125.0, 78.9, 78.5, 55.6, 55.0, 47.0, 46.7, 33.7, 32.2, 28.5, 28.2, 24.2, 23.5; IR (KBr, cm-1) 2975, 2876, 1663, 1407, 1156, 1125; HRMS calcd for $C_{36}H_{45}N_6O_4$ (M + H; ESI$^+$): 625.3502. Found: 625.3502. mp 190-195 °C (decomposed).

Example A-1e-4

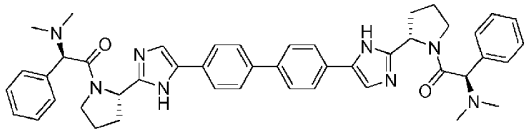
[0078]



[0079] To a 250 ml reactor equipped with a nitrogen line and overhead stirrer, 25.0 g of Example A-1e-3 (40.01 mmol, 1 equiv) was charged followed by 250 mL methanol and 32.85 mL (400.1 mmol, 10 equiv) 6M aqueous hydrogen chloride. The temperature was increased to 50 °C and agitated at 50 °C for 5h. The resulting slurry was cooled to 20-25 °C and held with agitation for ca. 18h. Filtration of the slurry afforded a solid which was washed successively with 100 ml 90% methanol/water (V/V) and 2x100 ml of methanol. The wet cake was dried in a vacuum oven at 50 °C overnight to give 18.12 g (31.8 mmol, 79.4%) of the desired product.

Recrystallization of Example A-1e-4

[0080] To a 250 ml reactor equipped with a nitrogen line and an overhead stirrer, 17.8g of crude Example A-1e-4 was charged followed by 72 mL methanol. The resulting slurry was agitated at 50 °C for 4h, cooled to 20-25 °C and held with agitation at 20-25 °C for 1h. Filtration of the slurry afforded a crystalline solid which was washed with 60 ml methanol. The resulting wet cake was dried in a vacuum oven at 50 °C for 4 days to yield 14.7 g (25.7 mmol, 82.6%) of the desired product: ^{1}H NMR (400 MHz, DMSO-d_6) δ 10.5-10.25 (br, 2H), 10.1-9.75 (br, 2H), 8.19 (s, 2H), 7.05 (d, J = 8.4, 4H), 7.92 (d, J = 8.5, 4H), 5.06 (m, 2H), 3.5-3.35 (m, 4H), 2.6-2.3 (m, 4H), 2.25-2.15 (m, 2H), 2.18-1.96 (m, 2H); ^{13}C NMR (100 MHz, DMSO- d_6) δ 156.6, 142.5, 139.3, 128.1, 127.5, 126.1, 116.9, 53.2, 45.8, 29.8, 24.3; IR (KBr, cm^{-1}) 3429, 2627, 1636, 1567, 1493, 1428, 1028. Anal calcd for $C_{26}H_{32}N_6Cl_4$: C, 54.75; H, 5.65; Cl, 24.86; Adjusted for 1.9% water: C, 53.71; H, 5.76; N, 14.46; Cl, 24.39. Found: C, 53.74; H, 5.72; N, 14.50; Cl, 24.49; KF = 1.9. mp 240 °C (decomposed)



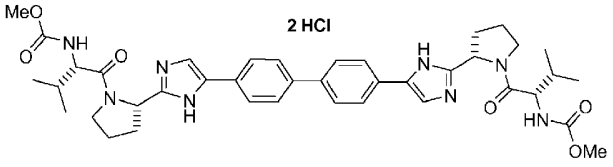
(1R,1'R)-2,2'-(4,4'-biphenyldiylbis(1H-imidazole-5,2-diyl(2S)-2,1-pyrrolidinediyl))bis(N,N-dimethyl-2-oxo-1-phenylethanamine)

[0081] HATU (44.6 mg, 0.117 mmol) was added to a mixture of pyrrolidine 1e (22.9 mg, 0.054 mmol), diisopropylethylamine (45

µL, 0.259 mmol) and *Cap*-1 (28.1 mg, 0.13 mmol) in DMF (1.5 mL), and the resulting mixture was stirred at ambient for 90 minutes. The volatile component was removed *in vacuo*, and the residue was purified first by MCX (methanol wash; 2.0 M NH_3/methanol elution) and then by a reverse phase HPLC system (H_2O/methanol/TFA) to provide the TFA salt of Example 1 as an off-white foam (44.1 mg). ^{1}H NMR (DMSO-d_6, δ = 2.5 ppm, 400 MHz): δ 10.25 (br s, 2H), 8.20-7.10 (m, 20H), 5.79-5.12 (m, 4H), 4.05-2.98 (m, 4H), 2.98-2.62 (m, 6H), 2.50-1.70 (m, 14H), [Note: the signal of the imidazole NH was too broad to assign a chemical shift]; LC (Cond. 1): RT = 1.40 min; > 98% homogeneity index; LC/MS: Anal. Calcd. for $[M+H]^+$ $C_{46}H_{51}N_8O_2$: 747.41; found 747.58
Solvent B = 0.1% TFA in 90% methanol/10% H_2O

Example 2

[0082]



methyl ((1S)-1-(((2S)-2-(5-(4'-(2-((2S)-1-((2S)-2-((methoxycarbonyl)amino)-3-methylbutanoyl)-2-pyrrolidinyl)-1H-imidazol-5-yl)-4-biphenylyl)-1H-imidazol-2-yl)-1-pyrrolidinyl)carbonyl)-2-methylpropyl)carbamate

[0083] A 50 mL flask equipped with a stir bar was sequentially charged with 2.5 mL acetonitrile, 0.344 g (2.25 mmol, 2.5 equiv) hydroxy benzotriazole hydrate, 0.374 g (2.13 mmol, 2.4 equiv) *N*-(methoxycarbonyl)-L-valine, 0.400 g (2.09 mmol, 2.4 equiv) 1-(3-dimethyaminopropyl)-3-ethylcarbodiimide hydrochloride and an additional 2.5 mL acetonitrile. The resulting solution was agitated at 20 °C for 1 hour and charged with 0.501 g (0.88 mmol, 1 equiv) Example A-1e-4. The slurry was cooled to about 0 °C and 0.45 g (3.48 mmol, 4 equiv) diisopropylethylamine was added over 30 minutes while maintaining a temperature below 10 °C. The solution was slowly heated to 15 °C over 3 hours and held at 15 °C for 16 hours. The temperature was increased to 20 °C and stirred for 3.25 hours. The resulting solution was charged with 3.3 g of 13 wt% aqueous NaCl and heated to 50 °C for 1 hour. After cooling to 20 °C, 2.5 mL of isopropyl acetate was added. The rich organic phase was washed with 2 x 6.9 g of a 0.5 N NaOH solution containing 13 wt% NaCl followed by 3.3 g of 13 wt% aqueous NaCl. The mixture was then solvent exchanged into isopropyl acetate by vacuum distillation to a target volume of 10 mL. The resulting hazy solution was cooled to 20 °C and filtered through a 0.45 µm filter. The clear solution was then solvent exchanged into ethanol by vacuum distillation with a target volume of 3 mL. 1.67 mL (2.02 mmol, 2.3 equiv) of 1.21 M HCl in ethanol was added. The mixture was then stirred at 25 °C for 15 hours. The resulting slurry was filtered and the wet cake was washed with 2.5 mL of 2:1 acetone:ethanol. The solids were dried in a vacuum oven at 50 °C to give 0.550 g (0.68 mmol, 77 %) of the desired product.

Recrystallization of the compound of Example 2

[0084] A solution of the compound of Example 2 prepared above was prepared by dissolving 0.520 g of the above product in 3.65 mL methanol. The solution was then charged with 0.078 g of type 3 Cuno Zeta loose carbon and allowed to stir for 0.25 hours. The mixture was then filtered and washed with 6 ml of methanol. The product rich solution was concentrated down to 2.6 mL by vacuum distillation. 7.8 mL acetone was added and allowed to stir at 25 °C for 15 h. The solids were filtered, washed with 2.5 mL 2:1 acetone:ethanol and dried in a vacuum oven at 70 °C to give 0.406 g (57.0%) of the desired product as white crystals: ^{1}H NMR (400 MHz, DMSO-d_6, 80 °C): 8.02 (d, *J*=8.34 Hz, 4 H), 7.97 (s, 2 H), 7.86 (d, *J*=8.34 Hz, 4 H), 6.75 (s, 2 H), 5.27 (t, *J*=6.44 Hz, 2 H), 4.17 (t, *J*=6.95 Hz, 2 H), 3.97 - 4.11 (m, 2 H), 3.74 - 3.90 (m, 2 H), 3.57 (s, 6 H), 2.32 - 2.46 (m, 2 H), 2.09 - 2.31 (m, 6 H), 1.91 - 2.07 (m, 2 H), 0.88 (d, *J*=6.57 Hz, 6 H), 0.79 (d, *J*=6.32 Hz, 6 H); ^{13}C NMR (75 MHz, DMSO-d_6): δ 170.9, 156.9, 149.3, 139.1, 131.7, 127.1, 126.5, 125.9, 115.0, 57.9, 52.8, 51.5, 47.2, 31.1, 28.9, 24.9, 19.6, 17.7; IR (neat, cm^{-1}): 3385, 2971, 2873, 2669, 1731, 1650. Anal. Calcd for $C_{40}H_{52}N_8O_6Cl_2$: C, 59.18; H, 6.45; N, 13.80; Cl, 8.73. Found C, 59.98; H, 6.80; N, 13.68; Cl, 8.77. mp 267 °C (decomposed). Characteristic diffraction peak positions (degrees 2θ ± 0.1) @ RT, based on a high quality pattern collected with a diffractometer (CuKα) with a spinning capillary with 2θ calibrated with a NIST other suitable standard are

as follows: 10.3, 12.4, 12.8, 13.3, 13.6, 15.5, 20.3, 21.2, 22.4, 22.7, 23.7.

BIOLOGICAL ACTIVITY

[0085] An HCV Replion assay was utilized in the present disclosure, and was prepared, conducted and validated as described in commonly owned PCT/US2006/022197 and in O'Boyle et. al. Antimicrob Agents Chemother. 2005 Apr;49(4): 1346-53.

[0086] HCV 1b-377-neo replicon cells were used to test the currently described compound series as well as cells resistant to compound A due to a Y2065H mutation in NS5A (described in application PCT/US2006/022197). The compounds tested were determined to have more than 10-fold less inhibitory activity on cells resistant to compound A than wild-type cells indicating a related mechanism of action between the two compound series. Thus, the compounds of the present disclosure can be effective to inhibit the function of the HCV NS5A protein and are understood to be as effective in combinations as previously described in application PCT/US2006/022197 and commonly owned WO/O4014852. Further, the compounds of the present disclosure can be effective against the HCV 1b genotype. It should also be understood that the compounds of the present disclosure can inhibit multiple genotypes of HCV. Table 2 shows the EC50 values of representative compounds of the present disclosure against the HCV 1b genotype. In one embodiment compounds of the present disclosure are active against the 1a, 1b, 2a, 2b, 3a, 4a, and 5a genotypes. EC50 ranges against HCV 1b are as follows: A = 1-10 μM; B = 100-999 nM; C = 1-99 nM; and D = 10-999 pM.

[0087] The compounds of the present disclosure may inhibit HCV by mechanisms in addition to or other than NS5A inhibition. In one embodiment the compounds of the present disclosure inhibit HCV replicon and in another embodiment the compounds of the present disclosure inhibit NS5A.

Table 2

Example	Range
1	D
2	D

[0088] It is desired that the examples be considered in all respects as illustrative and not restrictive, reference being made to the appended claims, rather than to the foregoing examples.

[0089] The compounds of the present disclosure may inhibit HCV by mechanisms in addition to or other than NS5A inhibition. In one embodiment the compounds of the of the present disclosure inhibit NS5A. Compounds of the present disclosure may inhibit multiple genotypes of HCV.

REFERENCES CITED IN THE DESCRIPTION

This list of references cited by the applicant is for the reader's convenience only. It does not form part of the European patent document. Even though great care has been taken in compiling the references, errors or omissions cannot be excluded and the EPO disclaims all liability in this regard.

Patent documents cited in the description

- US83699606P [0001]
- WO2005047288A [0050]
- US2006022197W [0085] [0086] [0086]
- WO4014852O [0086]

Non-patent literature cited in the description

- **TAN, S.-L.KATZEL, M.G.**Virology, 2001, vol. 284, 1-12 [0007]
- **PARK, K.-J.CHOI, S.-H**J. Biological Chemistry, 2003, [0007]
- Pharmaceutical Research, 1986, vol. 3, 6318- [0039]
- Hepatology, 2003, vol. 38, 1282- [0049]
- Biochem. Biophys. Res. Commun., 2004, vol. 313, 42- [0049]
- J. Gastroenterol., 2003, vol. 38, 567- [0049]
- J. Org. Chem., 1978, vol. 43, 2923- [0057]
- **O'BOYLE**Antimicrob Agents Chemother., 2005, vol. 49, 41346-53 [0085]

HEPATITIS C-VIRUSINHIBITORER

PATENTKRAV

1. Forbindelse, der er methyl ((1S)-1-(((2S)-2-(5-(4'-(2-((2S)-1-((2S)-2-((methoxycarbonyl)amino)-3-methylbutanoyl)-2-pyrrolidinyl)-1H-imidazol-5-yl)-4-biphenylyl)-1H-imidazol-2-yl)-1-pyrrolidinyl)carbonyl)-2-methylpropyl)carbamat; eller et farmaceutisk acceptabelt salt deraf til anvendelse i terapi.

2. Forbindelse til anvendelse ifølge krav 1, der er dihydrokloridet af methyl ((1S)-1-(((2S)-2-(5-(4'-(2-((2S)-1-((2S)-2-((methoxycarbonyl)amino)-3-methylbutanoyl)-2-pyrrolidinyl)-1H-imidazol-5-yl)-4-biphenylyl)-1H-imidazol-2-yl)-1-pyrrolidinyl)carbonyl)-2-methylpropyl)carbamat.

3. Forbindelse til anvendelse ifølge krav 1 eller 2, hvilken forbindelse yderligere omfatter anvendelse af en eller to yderligere forbindelser, der har anti-HCV-aktivitet før, efter eller samtidig med forbindelsen af krav 1, eller et farmaceutisk acceptabelt salt deraf eller med forbindelsen af krav 2.

4. Forbindelse til anvendelse ifølge krav 3, hvor mindst én af de yderligere forbindelser er interferon eller ribavirin.

5. Forbindelse til anvendelse ifølge krav 4, hvor interferonet er udvalgt fra interferon alpha 2B, pegyleret interferon-alpha, consensus interferon, interferon alpha 2A, og lymphoblastoid interferon tau.

6. Forbindelse til anvendelse ifølge krav 3, hvor mindst én af de yderligere forbindelser er udvalgt fra interleukin 2, interleukin 6, interleukin 12, en forbindelse, der fremmer udviklingen af et T-hjælpecellerepons type 1, interfererende RNA, antisense-RNA, Imiquimod, ribavirin, en inosin 5'-monophospat-dehydrogenaseinhibitor, amantadin og rimantadin.

7. Forbindelse til anvendelse ifølge krav 3, hvor mindst én af de yderligere forbindelser effektivt hæmmer funktionen af et target udvalgt fra HCV-metalloprotease, HCV-serinprotease, HCV-polymerase, HCV-helicase, HCV NS4B-protein, HCV-indgang, HCV-samling, HCV-udgang, HCV NS5A-protein og IMPDH til behandlingen af en HCV-infektion.