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(54) **Title:** DOSING REGIMENS FOR SARS-COV-2 BINDING MOLECULES

(57) **Abstract:** The instant disclosure relates to compositions comprising SARS-CoV-2 binding molecules and methods of use thereof. Provided herein are dosing regimens for administration of SARS-CoV-2 binding molecules, including the DARPin® protein ensovibep.

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DOSING REGIMENS FOR SARS-COV-2 BINDING MOLECULES

FIELD

[0001] The instant disclosure relates to methods for treating coronavirus infections in a subject in need thereof. For example, the instant disclosure provides dosing regimens for SARS-CoV-2 binding molecules, including the DARPin® protein ensovibep.

SEQUENCE LISTING

[0002] The instant application contains a Sequence Listing which has been submitted electronically and is hereby incorporated by reference in its entirety. The Sequence Listing was created on August 19, 2022, is named PAT059375_SL.XML and is about 12,288 bytes in size.

BACKGROUND

[0003] COVID-19 is a disease caused by infection with the SARS-CoV-2 virus, and it escalated into a global pandemic in 2020. The rapid development of vaccines has been crucial in fighting the pandemic. However, vaccination is not suitable for all individuals, and despite the wide availability of vaccines since mid-2021, the pandemic still poses ongoing challenges via breakthrough infections and emerging variants. Therefore, therapeutics that are effective, widely available, and can be routinely administered are urgently needed.

[0004] DARPin® proteins are genetically engineered ankyrin repeat proteins (or designed ankyrin repeat proteins) that typically exhibit highly specific and high-affinity target protein binding. They are derived from natural ankyrin repeat motifs from proteins, which are responsible for diverse intra-cellular functions such as cell signaling, regulation and structural integrity of the cell. DARPin® proteins constitute a new class of potent, specific and versatile protein therapeutics, and are used as investigational tools in various research, diagnostic and therapeutic applications.

[0005] Ensovibep is a DARPin® molecule being explored for its therapeutic benefit in treating COVID-19 patients. There is an urgent need to develop methods for administering ensovibep to patients in need thereof. Specifically, there is an urgent

need for a safe and effective dosing regimen for ensovibep to treat patients infected with SARS-CoV-2.

BRIEF SUMMARY

[0006] Provided herein are methods for treating coronavirus infections in a subject in need thereof. In some embodiments, the instant disclosure provides dosing regimens for administration of SARS-CoV-2 binding molecules, including the DARPin® protein ensovibep.

[0007] In some embodiments, a method for treating a coronavirus infection in a subject in need thereof comprises administering to the subject about 1 to about 700 mg of ensovibep, such as about 75 mg to about 600 mg of ensovibep. In some embodiments, about 75 mg, about 225 mg, or about 600 mg of ensovibep are administered to the subject.

[0008] In some embodiments, ensovibep comprises an amino acid sequence of SEQ ID NO: 1. In some embodiments, ensovibep comprises an amino acid sequence of SEQ ID NO: 2. In some embodiments, ensovibep is encoded by a nucleic acid having a sequence of SEQ ID NO: 8.

[0009] In some embodiments, ensovibep is administered once or more than once to the subject.

[0010] In some embodiments, the coronavirus infection is caused by SARS-CoV-2 or a variant thereof.

[0011] In some embodiments, ensovibep is administered by intravenous infusion.

[0012] In some embodiments, the subject is a human.

[0013] These and other embodiments are addressed further in the detailed description set forth below.

BRIEF DESCRIPTION OF THE DRAWINGS

[0014] **Figure 1** shows a QSP (Quantitative Systems Pharmacology) model to predict antiviral effects for either an antibody (Ab) or a DARPin®.

[0015] **Figure 2** shows simulated viral kinetics across individuals and optimization runs. The graphs show viral load (RNA copies/throat swab, y-axis) at various days post-symptom onset (x-axis).

[0016] **Figure 3** shows model prediction of anti-viral response for bamlanivimab using the QSP model described herein. This data was used to calibrate the QSP model, and

it was demonstrated that the model successfully predicted clinically observed results for bamlanivimab.

[0017] **Figure 4** provides a comparison of day 1-7 free spike protein trimer area under the curve (AUC) reduction between ensovibep and antibody (Ab) treatment.

[0018] **Figure 5** provides a visual predictive check to compare model prediction with observed clinical data for total viral load (top) and change from baseline (bottom).

[0019] **Figure 6** is an illustration of ensovibep binding to a SARS-CoV-2 spike protein via three of its DARPin® domains.

DETAILED DESCRIPTION

[0020] Computational models of in-host viral kinetics have been used for decades to describe the pathogenesis of HIV-1, influenza and hepatitis B virus, where evolving data and knowledge of viral function inform parameter values and assumptions. In the last few years, with limited biological information about SARS-CoV-2 pathophysiology, pharmacokinetic and pharmacodynamics (PK/PD) models that vary widely in mechanistic detail have been published to guide dose selection for repurposed drugs or novel therapies to fight SARS-CoV-2. Models range in complexity from more empirical PK/PD Emax models to quantitative systems pharmacology (QSP) of the entire immune system, in the middle of which are adaptations of the in-host viral kinetics models developed for other viruses. In general, these in-host models consist of uninfected and infected host cells and viral particles with various descriptions of an immune component. With model size comes the tradeoff of mechanistic interpretability and parameter identifiability, relying either on estimating model parameters or making assumptions from limited clinical data, which may have impacts on predictions in the context of therapeutic intervention.

[0021] Described herein is research applying one such developed in-host viral kinetic model in the context of a PK/PD model to aid the development of novel therapeutic approaches for treating SARS-CoV-2 infections, in particular to explore the predicted effect of a DARPin® therapeutic. The data provided herein demonstrates how a mathematical model that represents the current understanding of biological interactions can provide nonintuitive insights and guide drug development. Using the approach described herein (see Example 1), a dosing regimen for administration of the SARS-CoV-2 binding molecule ensovibep was established and tested in the

EMPATHY clinical trial, which was a multicenter, randomized, double-blind, placebo-controlled investigation (see Example 2).

Definitions

[0022] Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs. The terminology used in the detailed description herein is for the purpose of describing particular embodiments only and is not intended to be limiting.

[0023] All publications, patent applications, patents, GenBank or other accession numbers and other references mentioned herein are incorporated by reference in their entirety. To the extent the material incorporated by reference contradicts or is inconsistent with this specification, the specification will supersede any such material. The citation of any references herein is not an admission that such references are prior art to the present invention.

[0024] The singular forms "a," "an" and "the" are intended to include the plural forms as well, unless the context clearly indicates otherwise.

[0025] Furthermore, the term "about" as used herein when referring to a measurable value such as an amount of the length of a polynucleotide or polypeptide sequence, dose, time, temperature, and the like, is meant to encompass variations of ± 20%, ± 10%, ± 5%, ± 1%, ± 0.5%, or even ± 0.1 % of the specified amount.

[0026] Also as used herein, "and/or" refers to and encompasses any and all possible combinations of one or more of the associated listed items, as well as the lack of combinations when interpreted in the alternative ("or").

[0027] As used herein, "administering" and "administration" means a method of giving a dosage of a protein as described herein or a pharmaceutical composition (*e.g.* a pharmaceutical composition including the protein) to a subject (*e.g.* a patient).

[0028] Unless the context indicates otherwise, it is specifically intended that the various features described herein can be used in any combination.

[0029] Various aspects of the disclosure are described in further detail below. Additional definitions are set out throughout the specification.

SARS-CoV-2

[0030] Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2; also called 2019-nCoV) is the virus that causes COVID-19. COVID-19 patients exhibit a spectrum of illness from mild to moderate flu-like symptoms to severe responses that may lead to prolonged hospitalizations and death.

[0031] SARS-CoV-2 belongs to the large Coronaviridae family and possesses an unsegmented single-stranded RNA structure with four major structural proteins: nucleocapsid, spike, membrane and envelope. The first step of the SARS-CoV-2 infection cycle starts with direct binding of the viral spike protein (also called the S-glycoprotein) to the host angiotensin-covering enzyme 2 (ACE2) receptor. Following the fusion of viral and host membranes, endocytosis leads to the release of the viral nucleocapsid into the cytosol of the infected cell, followed by viral replication and exocytosis of new viruses. Each step of SARS-CoV-2 infection life-cycle is a potential target for anti-viral agents.

[0032] As used herein, the term “SARS-CoV-2” may include both wild-type virus (such as SARS-CoV-2 found in infected humans at the beginning of the COVID-19 pandemic, e.g., Wuhan HU-1 virus) and mutated forms or variants thereof. SARS-CoV-2 variants include, but are not limited to, Alpha, Beta, Gamma, Delta, Epsilon, Eta, Iota, Kappa, Omicron, Mu, and Zeta.

SARS-CoV-2 Binding Molecules

[0033] Described herein are SARS-CoV-2 binding molecules, including recombinant binding proteins with binding specificity for coronavirus spike proteins. The recombinant binding proteins described herein bind to the coronavirus spike protein at one or more binding sites, thereby neutralizing the virus.

[0034] An illustrative class of SARS-CoV-2 binding molecules is DARPin® proteins. DARPin® proteins are designed ankyrin repeat proteins. The term "designed" refers to the property that such repeat proteins are man-made and do not occur in nature. The term "ankyrin repeat domain" refers to a repeat domain comprising two or more consecutive ankyrin repeat modules as structural units, wherein said ankyrin repeat modules have structural and sequence homology. An “ankyrin repeat protein” comprises an ankyrin repeat domain. Ankyrin repeat proteins are described in, for example, International Patent Publication Nos. WO 2002/020565, WO 2010/060748, WO 2011/135067, WO 2012/069654, WO 2012/069655, WO 2014/001442, WO

2014/191574, WO 2014/083208, WO 2016/156596, and WO 2018/054971, all of which are incorporated by reference in their entireties. Ankyrin repeat domains optionally comprise appropriate capping modules.

[0035] DARPin® proteins typically exhibit highly specific and high-affinity target binding. DARPin® proteins comprise one or more designed ankyrin repeat domains, such as 1, 2, 3, 4, 5, or more designed ankyrin repeat domains. Due to their high specificity, stability, potency and affinity and due to their flexibility in formatting to generate mono-, bi- or multi-specific proteins, DARPin® proteins are attractive therapeutic agents. WO 2021/224686 (incorporated by reference herein its entirety) describes DARPin® proteins for use in the treatment of coronavirus infections. DARPin® is a registered trademark owned by Molecular Partners AG.

[0036] Without wishing to be bound by any theory, the designed ankyrin repeat proteins of instant disclosure are believed to act by (i) inhibiting receptor binding; (ii) providing allosteric inhibition of spike protein conformational change; and/or (iii) blocking protease sites needed for spike protein activation. These designed ankyrin repeat domains can bind to the spike protein and/or inhibit association with the ACE2 receptor as individual proteins. However, multi-epitope targeting by multi-domain, multi-specific DARPin® proteins is believed to provide even more potent neutralization of the spike proteins through cooperative binding interactions, and to minimize the likelihood of escape mutations through multi-specificity.

[0037] The DARPin® domains described herein generally comprise a core scaffold that provides structure and target binding residues that bind to a target. The structural core includes conserved amino acid residues, and the target binding surface includes amino acid residues that differ depending on the target. International Patent Publication No. WO 2002/020565 and Binz et al., Nature Biotech. 22(5): 575-582 (2004) describe libraries of ankyrin repeat proteins that can be used for the selection/screening of a protein that binds specifically to a target. Methods of making such libraries are also provided. Multiple DARPin® domains can be linked to form bispecific or multi-specific molecules.

[0038] In some embodiments, the recombinant binding proteins described herein comprise a DARPin® domain that specifically binds to coronavirus spike protein, such as a SARS-CoV-2 spike protein. In some embodiments, the recombinant binding protein comprises two, three or more ankyrin repeat domains that specifically bind to coronavirus spike protein. In some embodiments, the recombinant binding protein

comprises one, two, three or more DARPin® domains that specifically bind to SARS-CoV-2 spike protein. The target domains of interest on the coronavirus spike protein include, but are not limited to, the receptor binding domain (RBD domain); the S1 NTD domain; and the S2 domain. These domains are known in the art (see, e.g. Wrapp et al., Science 367, 1260-1263 (2020)).

[0039] In some embodiments, the DARPin® protein ensovibep may be used in the compositions and methods described herein. Ensovibep (Cas Registry Number 2451126-06-8; also called MP0420) is a first-in-class multi-specific antiviral therapeutic that comprises five DARPin® domains: two bind to human serum albumin (providing half-life extension) and three bind to the receptor-binding domain (RBD) of the SARS-CoV-2 spike protein trimer (**Figure 6**). The mechanism of action of ensovibep is based on cooperative binding of the three distinct RBD binding DARPin® domains to the spike protein trimer of SAR-CoV-2, thereby inhibiting association with the human ACE2 receptor, and preventing viral entry and replication. Ensovibep has a high potency against many SARS-CoV-2 variants of concern.

[0040] The amino acid sequence of Ensovibep is shown below:

(M)GSDLGKKLLEAARAGQDDEVRELLKAGADVNAKDYFSHTPLHLAARNGHLKIVEVLLKA
GADVNAKDFAGKTPLHLAANEGHLEIVEVLLKAGADVNAQDIFGKTPADIAADAGHEDIAEV
LQKAAGSPTPTPTTPTPTPTTPTPTPTGSDLGKKLLEAARAGQDDEVRELLKAGADVNAKDY
FSHTPLHLAARNGHLKIVEVLLKAGADVNAKDFAGKTPLHLAANEGHLEIVEVLLKAGADVN
AQDIFGKTPADIAADAGHEDIAEVLQKAAGSPTPTPTTPTPTPTTPTPTPTGSDLGKKLLQA
ARAGQLDEVRELLKAGADVNAKDREGITPLHLAAQHGHLEIVEVLLKAGADVNAKDVWGRTP
LHLAAWQGHLEIVEVLLKAGADVNAKDLAGATPLHVAALYGHLEIVEVLLKAGADVNAQDKS
GKTPADLAARAGHQDIAEVLQKAAGSPTPTPTTPTPTPTTPTPTPTGSDLGKKLLQAARAGQ
LDEVRELLKAGADVNAKDREGKTPLHVAAQEGHLEIVEVLLKAGADVNAKDVWGRTPLHLAA
WIGHLEIVEVLLKAGADVNAKDVSGATPLHAAALHGHLEIVEVLLNAGADVNAQDKSGKTPA
DLAARAGHQDIAEVLQKAAGSPTPTPTTPTPTPTTPTPTPTGSDLGKKLLQAARAGQLDEVR
ELLKAGADVNAKDQEGITPLHVAAHQGHLEIVEVLLKAGADVNAKDVWGRTPLHLAAWRGHL
EIVEVLLKAGADVNAKDHAGATPLHAAALSGHLEIVEVLLKAGADVNAQDKSGKTPADLAAR
AGHQDIAEVLQKAA(SEQ ID NO: 1).

[0041] In some embodiments, the Methionine (M) at the N-terminus of the ensovibep protein is removed during the protein maturation process (see SEQ ID NO: 2).

[0042] The three DARPin® domains of ensovibep that bind to the SARS-CoV-2 spike protein are shown in SEQ ID NOs: 3, 4, and 5. The two DARPin® domains that bind to human serum albumin (HSA) are shown in SEQ ID NO: 6, and function to increase the half-life of the protein *in vivo*. Each of these domains are separated by a linker (see SEQ ID NO: 7).

[0043] In some embodiments, a variant of ensovibep may be used in the compositions and/or methods of the instant disclosure. In some embodiments, the variant of ensovibep comprises at least one amino acid substitution relative to SEQ ID NO: 1 or SEQ ID NO: 2. For example, in some embodiments, the variant of ensovibep comprises 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, or 16 amino acid substitutions relative to SEQ ID NO: 1 or SEQ ID NO: 2. In some embodiments, the variant of ensovibep may comprise one or more amino acid substitutions in a DARPin® domain that binds to SARS-CoV-2 (i.e., within the DARPin® domain sequence of SEQ ID NO: 3, 4, or 5). In some embodiments, the variant of ensovibep may comprise one or more amino acid substitutions in a DARPin® domain that binds to HSA (i.e., within the DARPin® domain of SEQ ID NO: 6). In some embodiments, the variant of ensovibep may comprise one or more amino acid substitutions in a linker (i.e., within the linker of SEQ ID NO: 7). In some embodiments, the variant of ensovibep comprises at least 98% sequence identity to SEQ ID NO: 1 or SEQ ID NO: 2. In some embodiments, the variant of ensovibep comprises at least 99% sequence identity to SEQ ID NO: 1 or SEQ ID NO: 2. Sequence identity can be determined using a publicly available alignment program, such as the BLAST® program available from the National Library of Medicine (see blast.ncbi.nlm.nih.gov).

[0044] In some embodiments, Ensovibep may be encoded by the following nucleic acid sequence:

ggatccgatctgggcaaaaagttgctggaagcggcacgtgcaggccaggacgatgaagttcg
cgagctgttaaaagcaggcgcagatgttaatgcgaaggactatttcagccacacgccgttgc
atttggcggcacgtaacggccacctgaaaatcgtggaagtgctgttaaaggctggtgccgat
gtgaatgccaaagattttgcgggtaaaaccccgttacacttggcggcgaacgagggtcattt
agagatcgtcgaggtcctgcttaaggctggcgcggacgtaaatgcgcaagacatcttcggca
aaaccccggctgacattgcggcggatgcgggccacgaagatatcgccgaagtgttgcaaaag
gcagcgggcagcccgaccccaaccctacgaccccgactcctacgcctacgactccgactcc
gaccctacgggtagcgatctgggtaagaagctgctggaagccgcacgcgcaggccaagacg
acgaagtccgtgagctcctgaaggcaggcgccgatgtcaatgcaaaagactacttttctcat

acgccactgcacctggcagcccgtaacggtcatctgaagattgtagaagtgttgctgaaggc
gggcgcagatgttaatgcaaaggactttgcgggcaagaccccgttgcaccttgcggcgaatg
agggtcacttagagattgtggaggtcctgttgaaggccggtgccgacgtgaatgcacaagac
attttcggcaagacgccggcagacatcgcagcggacgcgggtcacgaggacatcgctgaagt
tctgcagaaagctgcgggttccccgacgccaacgcctaccacgcctacgccaactcctacca
ccccgaccccgacgccgaccggcagcgatttgggtaaaaagctgctgcaggccgcgagagcg
ggccagttggacgaggtgcgtgagctgttgaaagcgggtgcagacgttaatgctaaagaccg
cgagggtatcactccgttgcatctggcggcacagcacggtcatcttgagatcgtagaagtct
tgttgaaagctggtgctgacgtcaatgccaaggatgtttggggtcgtactccactgcatctc
gcggcgtggcagggtcaccttgagattgtcgaggttttgctgaaagcgggcgccgacgtcaa
tgcgaaagatctggcaggcgcgacccctctgcatgttgcggcgctgtacggtcacctggaaa
tcgttgaagtcctcttgaaggcgggtgcggatgtaaacgcgcaggacaagagcggtaaaacg
ccggccgatctggcagcacgcgccggtcaccaagatatcgcagaagtgctgcaaaaggctgc
gggtagcccaaccccaacgccgaccactcctacccctaccccaaccaccccaactccaactc
cgactggtagcgacctgggcaagaaactgctgcaagccgcacgtgccggtcagctggacgaa
gtgcgtgagttgttgaaggctggcgctgacgtgaacgcaaaagaccgtgagggtaagacgcc
gttacacgtggcagcgcaagaaggccacctggagattgttgaagtgctgctcaaagctggcg
cagacgtcaacgctaaagatgtgtggggtcgcactccgctgcatctggccgcgtggattggt
catttggaaattgtggaagttctgctcaaggccggcgctgatgtcaacgccaaagatgtgag
cggtgcgaccccgctgcacgcagcggcgctgcacggtcacttggaaatcgtggaggttctgt
tgaacgccggtgctgatgttaacgcgcaagataaatcgggtaagactccggcggatctggcg
gctcgtgcgggtcaccaggatattgccgaagttttgcagaaagccgctggttctcctacgcc
gaccccgacgacgccgactccaaccccgaccacgccgacgcctaccccgaccggtagcgact
tgggtaagaaactgttgcaggcagcgcgcgcgggtcaactggacgaggttcgtgagcttttg
aaagccggtgcggacgttaacgcgaaggatcaagaaggcattaccccactgcacgtggcggc
acatcagggtcatctggagatcgttgaggttctgctgaaggccggagcggatgtcaacgcga
aagacgtttggggccgtaccccattgcacctggcggcgtggcgcggtcacctcgaaatcgtc
gaagtgttactgaaagctggggcagatgtgaacgccaaggaccacgcgggtgcgacgccgct
gcatgcggcagcgctgagcggccatctggaaattgtcgaagtcctgctgaaagccggcgcag
atgttaatgcccaggataaatccggtaagaccccggcagacctggcagcgcgtgcgggccac
caagacattgccgaggttctgcaaaaagccgcg (SEQ ID NO: 8)

[0045] Ensovibep binds to SARS-CoV-2, and variants thereof. In some embodiments, ensovibep binds to a SARS-CoV-2 variant such as Alpha (B.1.1.7 and Q lineages),

Beta (B.1.351 and descendent lineages), Gamma (P.1 and descendent lineages), Delta (B.1.617.2 and AY lineages), Epsilon (B.1.427 and B.1.429), Eta (B.1.525), Iota (B.1.526), Kappa (B.1.617.1), 1.617.3, Mu (B.1.621, B.1.621.1), or Zeta (P.2). In some embodiments, ensovibep binds to an omicron strain of SARS-CoV-2, such as B.1.1.529, BA.1, BA.1.1, BA.2, BA.2.12.1, BA.3 lineages. In some embodiments, ensovibep binds to omicron BA.4 and BA.5 lineages with reduced potency (i.e., with reduced potency compared to the Wuhan HU-1 virus or other omicron sub-lineages such as BA.2 or BA.3).

Pharmaceutical Compositions

[0046] In some embodiments, the present disclosure provides a pharmaceutical composition comprising a SARS-CoV-2 binding protein (e.g., ensovibep) and a pharmaceutically acceptable carrier or excipient. The pharmaceutical composition may comprise a pharmaceutically acceptable carrier, diluent, or excipient. Standard pharmaceutical carriers include a phosphate buffered saline solution, water, emulsions such as an oil/water or water/oil emulsion, and various types of wetting agents.

[0047] The pharmaceutical composition can comprise one or more pharmaceutically acceptable ingredients, including, for example, acidifying agents, additives, adsorbents, aerosol propellants, air displacement agents, alkalizing agents, anticaking agents, anticoagulants, antimicrobial preservatives, antioxidants, antiseptics, bases, binders, buffering agents, chelating agents, coating agents, coloring agents, desiccants, detergents, diluents, disinfectants, disintegrants, dispersing agents, dissolution enhancing agents, dyes, emollients, emulsifying agents, emulsion stabilizers, fillers, film forming agents, flavor enhancers, flavoring agents, flow enhancers, gelling agents, granulating agents, humectants, lubricants, mucoadhesives, ointment bases, ointments, oleaginous vehicles, organic bases, pastille bases, pigments, plasticizers, polishing agents, preservatives, sequestering agents, skin penetrants, solubilizing agents, solvents, stabilizing agents, suppository bases, surface active agents, surfactants, suspending agents, sweetening agents, therapeutic agents, thickening agents, tonicity agents, toxicity agents, viscosity-increasing agents, water-absorbing agents, water-miscible cosolvents, water softeners, or wetting agents. See, e.g., the Handbook of Pharmaceutical Excipients, Third Edition, A. H. Kibbe (Pharmaceutical Press, London, UK, 2000); and

Remington's Pharmaceutical Sciences, Sixteenth Edition, E. W. Martin (Mack Publishing Co., Easton, Pa., 1980), each of which is incorporated by reference in its entirety.

[0048] The pharmaceutical compositions can be formulated to achieve a physiologically compatible pH. In some embodiments, the pH of the pharmaceutical composition can be, for example, between about 4.0 and about 8.0 or about 5.0 and about 8.0, or between about 4.5 and about 7.5, or between about 5.0 and about 7.5. In some embodiments, the pH of the pharmaceutical composition is between 5.5 and 7.5.

Methods of Treatment

[0049] The SARS-CoV-2 binding proteins described herein can be used to treat a subject in need thereof. The term "treat," as well as words related thereto, does not necessarily imply 100% or complete cure. Rather, there are varying degrees of treatment of which one of ordinary skill in the art recognizes as having a potential benefit or therapeutic effect. In this respect, the methods of treating coronavirus infections described herein can provide any amount or any level of treatment. Furthermore, the treatment provided by the methods of the present disclosure can include treatment of (i.e., relief from) one or more conditions or symptoms. In some embodiments, the methods treat by increasing the likelihood of survival of the subject.

[0050] Therapeutic responses in any given disease or condition can be determined by standardized response criteria specific to that disease or condition. The subject undergoing therapy may experience the beneficial effect of an improvement in the symptoms associated with the disease.

[0051] In some embodiments, a method of treating a coronavirus infection in a subject comprises the step of administering an effective amount of at least one SARS-CoV-2 binding protein or a pharmaceutical composition comprising the same to a subject in need thereof. In some embodiments, the subject is a human.

[0052] An "effective amount" or an "effective dosage" preferably reduces a measurable parameter, e.g., viral load by at least about 20%, at least about 40%, at least about 60%, at least about 80%, or more, relative to untreated subjects. The ability of a protein described herein to reduce a measurable parameter, e.g., a symptom of SARS-CoV-2, can be evaluated in an animal model system predictive of efficacy in humans.

Alternatively, this property of a composition can be evaluated by examining the ability of the compound to inhibit, such inhibition in vitro by assays known to the skilled practitioner.

[0053] The subject may be exhibiting any of the symptoms associated with a coronavirus infection, with differing degrees of severity, when the method of treating is administered. In some embodiments, the subject is treated within 7 days of the onset of symptoms. For example, the subject may be treated 7 days, 6, days, 5, days, 4 days, 3 days, 2 days, or 1 day after the onset of symptoms. In some embodiments, the subject is treated on the same day as the onset of symptoms. In some embodiments, the subject is treated within 7 days after testing positive for infection with SARS-CoV-2 or a variant thereof. For example, the subject may be treated 10 days, 9 days, 8 days, 7 days, 6, days, 5, days, 4 days, 3 days, 2 days, or 1 day after testing positive. In some embodiments, the subject is treated on the same day as testing positive. SARS-CoV-2 diagnostic tests, including PCR-based and antigen-based tests, are known in the art.

[0054] In some embodiments, a method of treating a coronavirus infection in a subject comprises the step of administering an effective amount of ensovibep, or a pharmaceutical composition comprising the same, to a subject in need thereof. In some embodiments, a method of treating a coronavirus infection in subject in need thereof comprises administering to the subject about 1 mg to about 700 mg of ensovibep. In some embodiments, a method of treating a coronavirus infection in subject in need thereof comprises administering to the subject about 75 mg to about 600 mg of ensovibep. In some embodiments, the method comprises administering to the subject about 1 mg to about 10 mg, about 10 mg to about 25 mg, about 25 mg to about 50 mg, about 50 mg to about 75 mg, about 75 mg to about 100 mg, about 100 mg to about 125 mg, about 125 mg to about 150 mg, about 150 mg to about 175 mg, about 175 mg to about 200 mg, about 200 mg to about 225 mg, about 225 mg to about 250 mg, about 250 mg to about 275 mg, about 275 mg to about 300 mg, about 300 mg to about 325 mg, about 325 mg to about 350 mg, about 350 mg to about 375 mg, about 375 mg to about 400 mg, about 400 mg to about 425 mg, about 425 mg to about 450 mg, about 450 mg to about 475 mg, about 475 mg to about 500 mg, about 500 mg to about 525 mg, about 525 mg to about 550 mg, about 550 mg to about 575 mg, about 575 mg to about 600 mg, about 600 mg to about 625 mg, about 625 mg to about 650 mg, about 650 mg to about 675 mg, or about 675 mg to about 700 mg of

ensovibep. In some embodiments, about 1 mg of ensovibep is administered to the subject. In some embodiments, about 10 mg of ensovibep is administered to the subject. In some embodiments, about 25 mg of ensovibep is administered to the subject. In some embodiments, about 75 mg of ensovibep is administered to the subject. In some embodiments, about 225 mg of ensovibep is administered to the subject. In some embodiments, about 600 mg of ensovibep is administered to the subject. In some embodiments, about 700 mg of ensovibep is administered to the subject. As will be understood by those in the art, the dosage regimen may be adjusted to provide an optimum therapeutic response, e.g., to optimize safety and efficacy.

[0055] In some embodiments, a method of treating a coronavirus infection in subject in need thereof comprises administering to the subject about 3 mg/kg to about 20 mg/kg of ensovibep. As used herein, the term "mg/kg" refers to milligrams of ensovibep per kilogram of bodyweight of the subject. In some embodiments, the method comprises administering to the subject about 3 mg/kg to about 6 mg/kg, about 6 mg/kg to about 9 mg/kg, about 9 mg/kg to about 12 mg/kg, about 12 mg/kg to about 15 mg/kg, about 15 mg/kg to about 18 mg/kg, or about 18 mg/kg to about 20 mg/kg of ensovibep. In some embodiments, the method comprises administering to the subject about 3 mg/kg of ensovibep. In some embodiments, the method comprises administering to the subject about 9 mg/kg of ensovibep. In some embodiments, the method comprises administering to the subject about 20 mg/kg of ensovibep.

[0056] In some embodiments, a method of treating a coronavirus infection in subject in need thereof comprises administering to the subject about 1 mg of ensovibep. In some embodiments, a method of treating a coronavirus infection in subject in need thereof comprises administering to the subject about 10 mg of ensovibep. In some embodiments, a method of treating a coronavirus infection in subject in need thereof comprises administering to the subject about 25 mg of ensovibep. In some embodiments, a method of treating a coronavirus infection in subject in need thereof comprises administering to the subject about 75 mg of ensovibep. In some embodiments, a method of treating a coronavirus infection in subject in need thereof comprises administering to the subject about 225 mg of ensovibep. In some embodiments, a method of treating a coronavirus infection in subject in need thereof comprises administering to the subject about 600 mg of ensovibep. In some embodiments, a method of treating a coronavirus infection in subject in need thereof comprises administering to the subject about 700 mg of ensovibep.

[0057] Also provided herein is ensovibep for use in the treatment of a coronavirus infection in a subject, wherein about 1 mg to about 700 mg of ensovibep is administered to the subject. In some embodiments, about 75 to about 600 mg of ensovibep is administered to the subject. In some embodiments, about 75 mg of ensovibep is administered to the subject. In some embodiments, about 225 mg of ensovibep is administered to the subject. In some embodiments, about 600 mg of ensovibep is administered to the subject. In some embodiments, about 700 mg of ensovibep is administered to the subject.

[0058] Also provided herein is the use of ensovibep for the manufacture of a medicament for use in the treatment of a coronavirus infection in a subject, wherein about 1 mg to about 700 mg of ensovibep is administered to the subject. In some embodiments, about 75 mg to about 600 mg of ensovibep is administered to the subject. In some embodiments, about 1 mg of ensovibep is administered to the subject. In some embodiments, about 10 mg of ensovibep is administered to the subject. In some embodiments, about 25 mg of ensovibep is administered to the subject. In some embodiments, about 75 mg of ensovibep is administered to the subject. In some embodiments, about 225 mg of ensovibep is administered to the subject. In some embodiments, about 600 mg of ensovibep is administered to the subject. In some embodiments, about 700 mg of ensovibep is administered to the subject.

[0059] In some embodiments, the coronavirus infection is caused by SARS-CoV-2. In some embodiments, the coronavirus infection is caused by a variant of SARS-CoV-2. In some embodiments, the coronavirus infection is caused by variant of SARS-CoV-2 selected from Alpha (B.1.1.7 and Q lineages), Beta (B.1.351 and descendent lineages), Gamma (P.1 and descendent lineages), Delta (B.1.617.2 and AY lineages), Epsilon (B.1.427 and B.1.429), Eta (B.1.525), Iota (B.1.526), Kappa (B.1.617.1), 1.617.3, Mu (B.1.621, B.1.621.1), and Zeta (P.2). In some embodiments, the coronavirus infection is caused by an omicron strain of SARS-CoV-2, such as B.1.1.529, BA.1, BA.1.1, BA.2, BA.2.12.1, or BA.3 variants. In some embodiments, the coronavirus infection is not caused by an omicron BA.4 or BA.5 variant.

[0060] In some embodiments, the subject is treated with ensovibep, which has a sequence of SEQ ID NO: 1 or SEQ ID NO: 2. In some embodiments, the subject is treated with a variant of ensovibep, such as a variant that has at least 98% or at least 99% sequence identity to SEQ ID NO: 1 or SEQ ID NO: 2.

[0061] In some embodiments, a single administration of ensovibep is used to treat the subject. In some embodiments, ensovibep is administered more than once to the subject. In some embodiments, ensovibep is administered to the subject one time, two times, three times, four times, five times, or more. Various factors will impact the number and frequency of administrations, such as the age and general health of the subject, as well as the state of the subject's coronavirus infection and the severity of the symptoms associated with coronavirus infection.

[0062] In some embodiments, the method is a prophylactic method, i.e. a method of preventing a coronavirus infection in a subject. In such methods, an effective amount of at least one binding protein as defined herein, or the nucleic acid as defined herein, or of the pharmaceutical composition as defined herein is administered to a subject. Typically, the subject will not be exhibiting any of the symptoms associated with a coronavirus infection when the prophylactic method is administered. In some embodiments, a single administration of the prophylactic method may be sufficient. In other embodiments, repeated administration may be necessary. Various factors will impact on the number and frequency of administrations, such as the age and general health of the subject, as well as the subject's risk of exposure to a coronavirus.

[0063] In some embodiments, the ensovibep is administered by intravenous infusion. The intravenous infusion may last for about 15 minutes, about 30 minutes, about 45 minutes, about 60 minutes, about 75 minutes, about 90 minutes, about 105 minutes, or about 130 minutes. In some embodiments, the intravenous infusion lasts for about 60 minutes.

[0064] The embodiments within the specification provide an illustration of embodiments of the invention and should not be construed to limit the scope of the invention. The skilled artisan readily recognizes that many other embodiments are encompassed by the invention.

EXAMPLES

[0065] The following examples, which are included herein for illustration purposes only, are not intended to be limiting.

Example 1: Viral Kinetics Model of SARS-CoV-2 Infection Shows Benefits of Viral Neutralization Mechanism in Reducing Viral Loads

[0066] This example describes the pharmacokinetics (PK) and pharmacodynamics (PD) modeling and simulation approach used for dose selection of ensovibep in the EMPATHY study in ambulatory adult patients with symptomatic COVID-19 (See Example 2).

[0067] Briefly, the PK/PD model was constructed from publicly available data describing viral kinetics from SARS-CoV-2 patient throat swabs from untreated COVID19 patients (Wölfel, R., Corman, V. M., Guggemos, W., et al (2020), Nature; 581(7809), 465-469. doi:10.1038/s41586-020-2196-x; Pan, Y., Zhang, D., Yang, P., et al (2020). Lancet Infect Dis; 20(4), 411-412. doi:10.1016/s1473-3099(20)30113-4; and Kim, K. S., Ejima, K., Ito, Y., et al (2020). medRxiv, 2020.2003.2023.20040493. doi:10.1101/2020.03.23. 20040493) and calibrated to describe reported viral load data from patients treated with the neutralizing antibody (Ab) therapeutic bamlanivimab (Gottlieb RL, et al. JAMA. 2021;325(7):632–644), and then applied to ensovibep to predict response and inform dose selection. Preliminary PK parameters for ensovibep were determined using an exploratory population PK model derived from the first two cohorts of a Phase 1 study with ensovibep (MP0420-CP101). The PK/PD model predicts anti-viral effects of either Ab or ensovibep in the lung.

[0068] Construction and analysis using the PK/PD model will be described in further detail below.

Modeling and simulation approach and assumptions

[0069] A Quantitative Systems Pharmacology (QSP) model was used to predict the human PK/PD response of ensovibep and inform dose selection.

[0070] To construct the QSP model, the topology of a “one-state immune model” (Figure 1) was generated by adapting a published influenza model (Canini L and Carrat F. J Virol. 2011;85(6):2764–2770) using various assumptions, as follows: (i) infected cells can rapidly produce new viral particles, (ii) the timescale of immune cell

recruitment is rapid such that the immune response can be represented by a one-state variable (X) that accounts for both inhibiting production of viral particles and promoting clearance of infected cells, (iii) the immune response is persistent.

[0071] The QSP model for an untreated condition was constructed by fitting to viral kinetics from untreated SARS-CoV-2 patients (throat swabs) and the modeled anti-viral drug effect was calibrated by comparing model predictions for a generic neutralizing antibody (Ab) monotherapy with reported viral load from patients treated with bamlanivimab (Gottlieb RL, et al. JAMA. 2021;325(7):632–644). The QSP model was then tailored to predict ensovibep PK/PD response by incorporating binding affinity and preliminary human PK data.

[0072] Anti-viral effects of neutralizing biologics, either Ab or DARPin® (**Figure 1**) were performed under the following key assumptions:

- 10% of systemic Ab or DARPin® rapidly distributes from blood to lungs
- Lung viral kinetics are approximated by throat swab viral kinetics
- Sixty spike protein trimers are present on the surface of each virus, and either one Ab or one DARPin® molecule binds to one spike protein trimer
- Binding neutralizes interaction with the ACE-2 receptor, blocking infection, and all spike protein trimers must be bound to completely block infection capacity
- Ensovibep binds to the RBD on spike protein trimers with Kd = 1 pM (off-rate immeasurable by surface plasmon resonance) and Ab binds with Kd = 100pM
- Clearance of Ab-virus or DARPin®-virus complex is equal to clearance of free virus
- Treatment is initiated 5 days after infection
- There is a sustained immune response over the duration of viral infection
- Binding of free spike protein trimers is expected to correlate with clinical effect

Viral kinetics model parameterization using data from untreated COVID-19 patients

[0073] **Figure 2** shows the simulated viral kinetics curves for all parameter optimization runs for each individual, reflecting the range of behavior considered to address model non-identifiability, or the inability for a single parameter set to uniquely describe the data. There was considerable variability in the initial slope of the viral kinetics curve as

well as in the time to peak viral load (T_{max}) due to the unavailability of early time-point data.

QSP model calibration by comparing predicted viral load change for Ab monotherapy with published data for bamlanivimab

[0074] The model-predicted change from baseline in log viral load for placebo and treatment with Ab monotherapy (**Figure 3**) were in good quantitative agreement with those reported for bamlanivimab monotherapy (Gottlieb RL, et al. JAMA. 2021;325(7):632–644).

[0075] Clinically, bamlanivimab as monotherapy at 700 mg, 2800 mg, and 7000 mg doses did not lead to a statistically significant reduction in the primary endpoint of viral load change from baseline compared with placebo on Day 11, whereas exploratory analysis at 7 days post-dose were more favorable; importantly, the secondary endpoint of reduced hospitalization and death was met.

EMPATHY dose and regimen selection driven by model predictions, including benchmarking to bamlanivimab

[0076] Leveraging the PK/PD model built to describe viral kinetics in response to neutralizing biologic therapeutics, simulations were performed for single, IV administrations of SKO136 over a dose range from 1 to 7000 mg initiated on Day 5 post-infection. A key finding was that model predictions for total viral load exhibited a largely flat predicted dose-response across all output metrics. The prediction of a flat dose-response in mean log10 viral load change from baseline in the dose range tested was consistent with results reported for doses of 700, 2800, and 7000 mg bamlanivimab when used as a monotherapy. While total viral load is a relevant prediction metric due to its ability to be measured clinically, At the time of QSP model construction, viral load data were limited, highly variable, and were not consistently associated with a dose-response in clinical trials. Along this line, clinical response was reported for bamlanivimab in the absence of statistically significant reductions in viral load.

[0077] Free spike protein trimers represent the fraction of free and infectious viruses whereas the clinical assessment of total viral load is a composite of free and infectious virus, partially or fully neutralized virus (less or no longer infectious), and dead virus.

[0078] Predictions for free spike protein trimer showed dose-dependent behavior for mean percent reduction in Day 0-7 free spike trimer AUC, allowing for an interpretable dynamic range for dose selection. Although free spike protein trimer AUC is not a

measurable quantity in the clinic, free spike protein trimer was hypothesized to be the relevant species to drive disease pathophysiology and by extension, inhibition thereof reasoned to better correlate with clinical efficacy than total viral load. Therefore, the QSP model was leveraged to simulate free spike protein trimer area under curve (AUC) reduction as a metric for reducing free infectious virus. The model predictions for a single, IV administration of ensovibep or Ab (**Figure 4**) were as follows:

- Ensovibep 75 mg IV was predicted to achieve 95% reduction in free spike protein trimer AUC. In comparison, Ab treatment was predicted to achieve 95% reduction with a 225 mg dose.
- Ensovibep 225 mg IV was predicted to achieve a similar level of reduction in mean percent reduction in free spike protein trimer AUC as a 700 mg Ab dose. Bamlanivimab 700 mg dose reduced COVID-19 related hospitalizations and emergency room visits compared with placebo.
- Ensovibep 600 mg IV was additionally selected to account for uncertainty in the modeling projections.
- The approximately 3-fold lower dose for ensovibep compared with Ab treatment was mainly attributed to the following differences: (i) lower molecular weight for ensovibep (84.5 kDa), and (ii) higher affinity of ensovibep to spike protein RBD.
- Moreover, uncertainty in modeling predictions was attributed to: (i) lack of data informing the viral kinetics from infection to peak load, and consequent numerical uncertainty in parameters, (ii) interindividual variability of viral kinetics.

Comparison between QSP model predicted viral kinetics and observed data from EMPATHY

[0079] Model predictions for viral load, total and change from baseline, were in good agreement with observed clinical data (**Figure 5**). 80% of the mean viral load clinical data was within 2-fold of the model predictions and 100% was within 3-fold of model predictions.

[0080] 85% of the mean viral load change from baseline clinical data was within 2-fold of the model predictions and 95% was within 3-fold of model predictions.

Conclusions

[0081] The QSP model was developed by integrating a human SARS-CoV-2 viral kinetics model with ensovibep-binding measurements and preliminary PK data to provide a rationale for the dose regimen in the EMPATHY study (see Example 2). Specifically, doses of 75 mg, 225 mg, and 600 mg were selected.

[0082] The QSP model was calibrated by describing the reported viral load data from COVID-19 patients treated with bamlanivimab.

[0083] Dose selection was based on model outputs for the reduction of free spike protein trimer AUC (achieved by ensovibep binding), a metric proposed to be more sensitive and predictive of clinical efficacy than viral load reduction.

Example 2: A Dose Finding, Efficacy and Safety Study of Ensovibep (MP420) in Ambulatory Patients with Symptomatic COVID-19 (EMPATHY)

[0084] The global EMPATHY clinical trial (ClinicalTrials.gov Identifier: NCT04828161) is a randomized, double-blind, placebo-controlled study in ambulatory (non-hospitalized) adult patients with COVID-19. The purpose of this study is to establish the antiviral efficacy of ensovibep against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in humans, identify the optimal dose, and demonstrate its clinical value for treating COVID-19 in adult and adolescent ambulatory patients.

[0085] This Phase II, dose-ranging study recruited patients in four arms randomized 1:1:1:1 to receive ensovibep (75 mg, 225 mg, or 600 mg) or placebo, administered as a single intravenous (IV) infusion over 60 minutes.

[0086] Primary objectives were as follows:

- Demonstrate superiority of ensovibep, compared to placebo, in reducing SARS-CoV-2 viral load through Day 8.

[0087] Secondary objectives were as follows:

- To assess the effect of ensovibep, compared to placebo, in reducing the occurrence of hospitalizations (≥ 24 hours of acute care) and/or emergency room visits related to COVID-19 or death from any cause up to Day 29
- To assess the effect of ensovibep, compared to placebo, in reducing COVID-19 symptoms through Day 29
- To evaluate safety and tolerability of ensovibep

- To characterize the pharmacokinetics (PK) of ensovibep

Study Design

[0088] Participants were assigned to one of two or more groups in parallel for the duration of the study. There were four treatment arms.

Arms and Interventions

Arm	Intervention/treatment
Experimental: ensovibep active treatment arm 1 **Phase 2: ensovibep active treatment arm 1**	Drug: ensovibep Phase 2: 3 active treatment arms vs placebo, IV on day 1 only.
Experimental: ensovibep active treatment arm 2 **Phase 2: ensovibep active treatment arm 2**	Drug: ensovibep Phase 2: 3 active treatment arms vs placebo, IV on day 1 only.
Experimental: ensovibep active treatment arm 3 **Phase 2: ensovibep active treatment arm 3**	Drug: ensovibep Phase 2: 3 active treatment arms vs placebo, IV on day 1 only.
Placebo Comparator: Placebo **Phase 2: Placebo**	Drug: Placebo Phase 2: 3 active treatment arms vs placebo, IV on day 1 only.

Primary Outcome Measures

[0089] Primary outcome measures were as follows:

1. SARS-CoV-2 viral load [Time Frame: 8 days (days 0, 3, 5 and 8)]
   Time-weighted change from baseline (measured at Day 3, Day 5, and Day 8) in SARS-CoV-2 viral load in nasopharyngeal swabs through Day 8.

Secondary Outcome Measures

[0090] Secondary outcome measures were as follows:

1. Occurrence of hospitalizations, emergency room visits or death
   [Time Frame: up to day 29]

Proportion of patients experiencing hospitalizations (≥ 24 hours of acute care) and/or emergency room visits related to COVID-19 or death from any cause up to Day 29.

2. Time to sustained clinical recovery [Time Frame: up to day 29]
   Time to sustained clinical recovery, defined as (a) all symptoms from the modified FDA COVID-19 symptom list scored as moderate or severe at baseline are subsequently scored as mild or absent, AND (b) all symptoms from the modified FDA COVID-19 symptom list scored as mild or absent at baseline are subsequently scored as absent, with no subsequent worsening up to Day 29.
3. Serious adverse events (SAEs), AEs of Special Interest (AESIs), vital signs and clinical laboratory measurements [Time Frame: up to day 91]
   In order to evaluate the safety and tolerability of ensovibep, the proportion of patients up to end of study with: (a) SAEs, including death from any cause and (b) AESIs, including infusion-related reactions (IRRs) CTCAE grade 2 or higher, were assessed. Vital signs and clinical laboratory measurements were assessed too.
4. Ensovibep Maximum Plasma Concentration [Cmax]). [Time Frame: up to day 91]
   Cmax is the observed maximum concentration.
5. Ensovibep PK parameter - AUClast [Time Frame: up to day 91]
   AUClast is the area under the concentration-time curve from time zero to the time of the last quantifiable concentration.

Inclusion/Exclusion Criteria

[0091] Inclusion criteria were as follows:

1. Males or females ≥ 18 years of age on the day of inclusion (no upper limit).
2. Presence of two or more COVID-19 symptoms and onset within 7 days prior to dosing: Feeling hot or feverish, cough, sore throat, low energy or tiredness, headache, muscle or body aches, chills or shivering, and shortness of breath.
3. Positive test for SARS-CoV-2 in upper respiratory swab on the day of dosing (rapid antigen test).
4. Understands and agrees to comply with the planned study procedures.

5. The patient or legally authorized representative gives signed informed consent.

[0092] Exclusion criteria were as follows:

1. Requiring hospitalization at time of screening, or at time of study drug administration.
2. Oxygen saturation (SpO2) ≤ 93% on room air at sea level or ratio of arterial oxygen partial pressure (PaO2 in mmHg) to fractional inspired oxygen (FiO2) < 300, respiratory rate ≥ 30 per minute, and heart rate ≥ 125 per minute.
3. Known allergies to any of the components used in the formulation of the ensovibep or placebo.
4. Suspected or proven serious, active bacterial, fungal, viral, or other infection (besides SARS-CoV-2) that in the opinion of the investigator could constitute a risk when taking intervention.
5. Any serious concomitant systemic disease, condition, or disorder that, in the opinion of the investigator, should preclude participation in this study.
6. Any co-morbidity requiring surgery within 7 days of dosing, or that is considered life-threatening within 29 days of dosing.
7. Prior or concurrent use of any medication for treatment of COVID-19, including antiviral agents, convalescent serum, or anti-viral antibodies. Purely symptomatic therapies (e.g., over-the-counter [OTC] cough medications, acetaminophen, and nonsteroidal anti-inflammatories [NSAIDs]) are permitted. Prior use of steroids for management of COVID-19 symptoms may be permitted, provided they can be stopped at study entry based on investigator judgement. Prior vaccination for COVID-19 is permitted.

Results

[0093] Results from the study showed that the primary endpoint was met with a statistically significant reduction in viral load over eight days, compared to placebo, for all three dosing arms. The secondary endpoint of hospitalization and/or ER visits related to COVID-19, or death showed an overall 78% reduction in risk of events across ensovibep arms compared to placebo. Treatment arms were generally

balanced in terms of demographic, baseline and disease characteristics. The placebo arm with 99 patients had a total of six events (event rate of 6.0%); five patients were hospitalized, two of whom died due to worsening of COVID-19 and one patient had an ER visit only. In the 301 patients treated with ensovibep, there were four events, hospitalizations occurred in two patients and two needed to visit ER (event rate of 1.3%). No deaths occurred in any of the patients treated with ensovibep.

[0094] All doses were well-tolerated and no unexpected safety issues were identified for any of the doses. The lowest dose of 75mg was the planned dose for further development.

[0095] The foregoing is illustrative of the present invention, and is not to be construed as limiting thereof. The invention is defined by the following claims, with equivalents of the claims to be included therein.

CLAIMS

What is claimed is:

1. A method of treating a coronavirus infection in subject in need thereof, the method comprising administering to the subject about 1 to about 700 mg of ensovibep.

2. The method of claim 1, wherein about 75 mg of ensovibep is administered to the subject.

3. The method of claim 1, wherein about 225 mg of ensovibep is administered to the subject.

4. The method of claim 1, wherein about 600 mg of ensovibep is administered to the subject.

5. The method of any one of claims 1-4, wherein ensovibep comprises the amino acid sequence of SEQ ID NO: 1.

6. The method of any one of claims 1-4, wherein ensovibep comprises the amino acid sequence of SEQ ID NO: 2.

7. The method of any one of claims 1-4, wherein ensovibep is encoded by a nucleic acid comprising the sequence of SEQ ID NO: 8.

8. The method of any one of claims 1-7, wherein ensovibep is administered more than once to the subject.

9. The method of any one of claims 1-8, wherein the coronavirus infection is caused by SARS-CoV-2 or a variant thereof.

10. The method of any one of claims 1-9, wherein the ensovibep is administered by intravenous infusion.

11. The method of any one of claims 1-10, wherein the subject is a human.

12. A method of treating a coronavirus infection in subject in need thereof, the method comprising administering to the subject about 75 mg of ensovibep.

13. The method of claim 12, wherein ensovibep is administered more than once to the subject.

14. The method of any one of claims 12-13, wherein ensovibep comprises the sequence of SEQ ID NO: 1.

15. The method of any one of claims 12-13, wherein ensovibep comprises the sequence of SEQ ID NO: 2.

16. The method of any one of claims 12-13, wherein the ensovibep is encoded by a nucleic acid comprising the sequence of SEQ ID NO: 8.

17. The method of any one of claims 12-16, wherein the coronavirus infection is caused by SARS-CoV-2 or a variant thereof.

18. The method of any one of claims 12-17, wherein the ensovibep is administered by intravenous infusion.

19. The method of any one of claims 12-18, wherein the subject is a human.

Figure 1

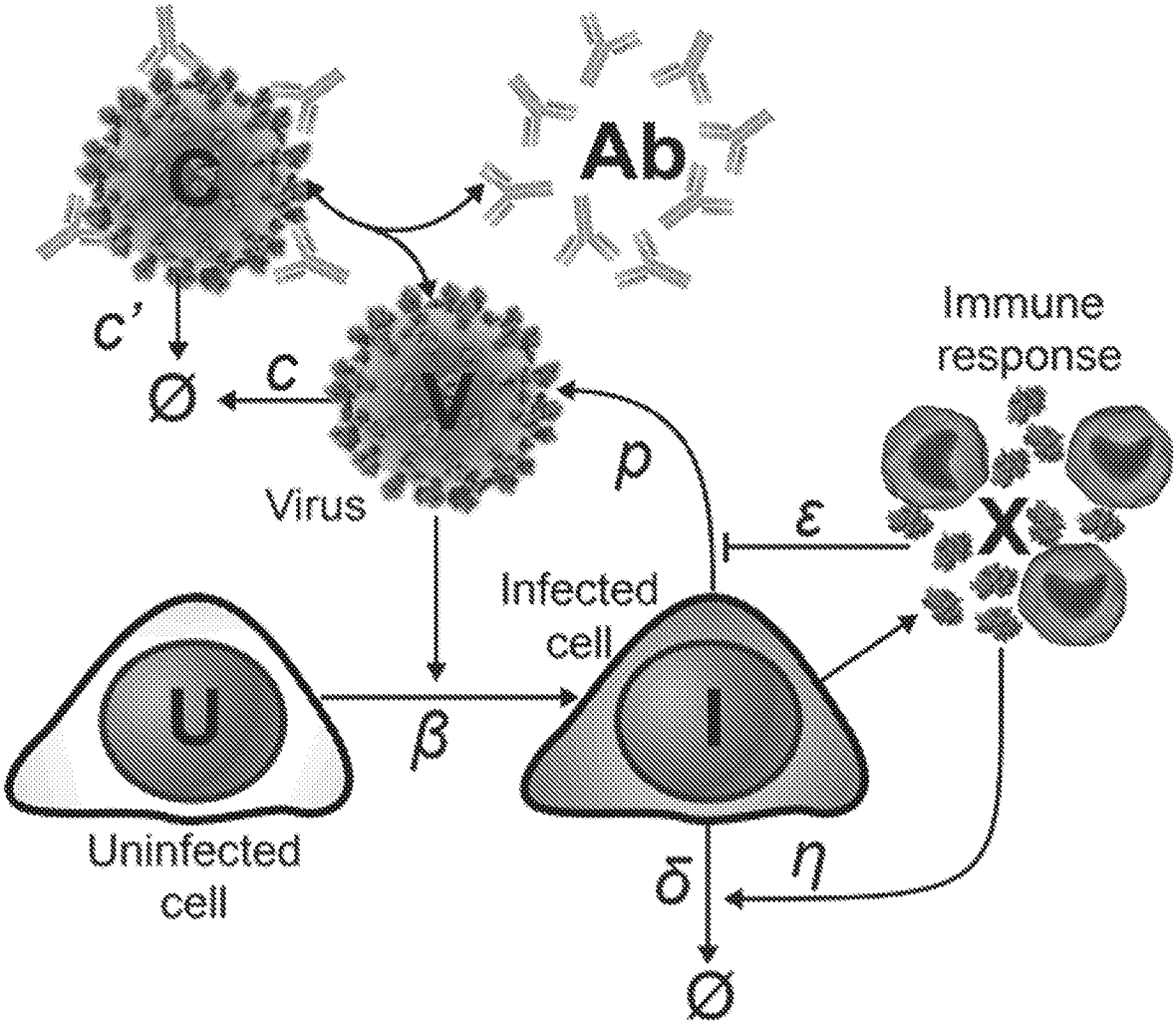

C
Ab
c'
Ø
c
V
Virus
ρ
Immune
response
ε
X
Infected
cell
U
β
I
Uninfected
cell
δ
η
Ø

Figure 2

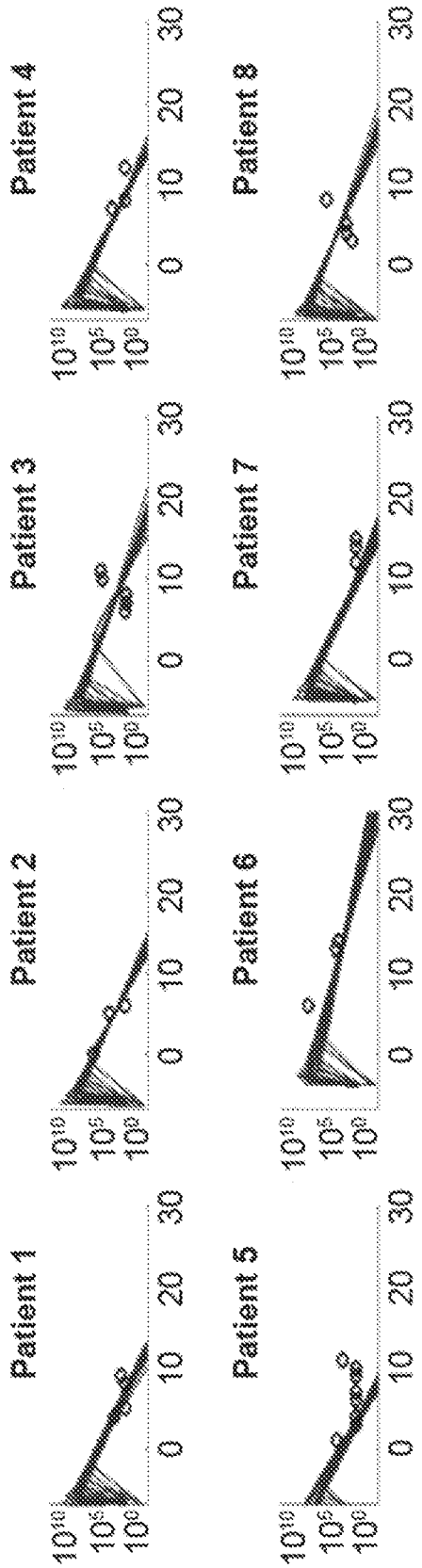

Patient 1
Patient 2
Patient 3
Patient 4
Patient 5
Patient 6
Patient 7
Patient 8
10^{10}
10^{5}
10^{0}
0
10
20
30

Figure 2 (continued)

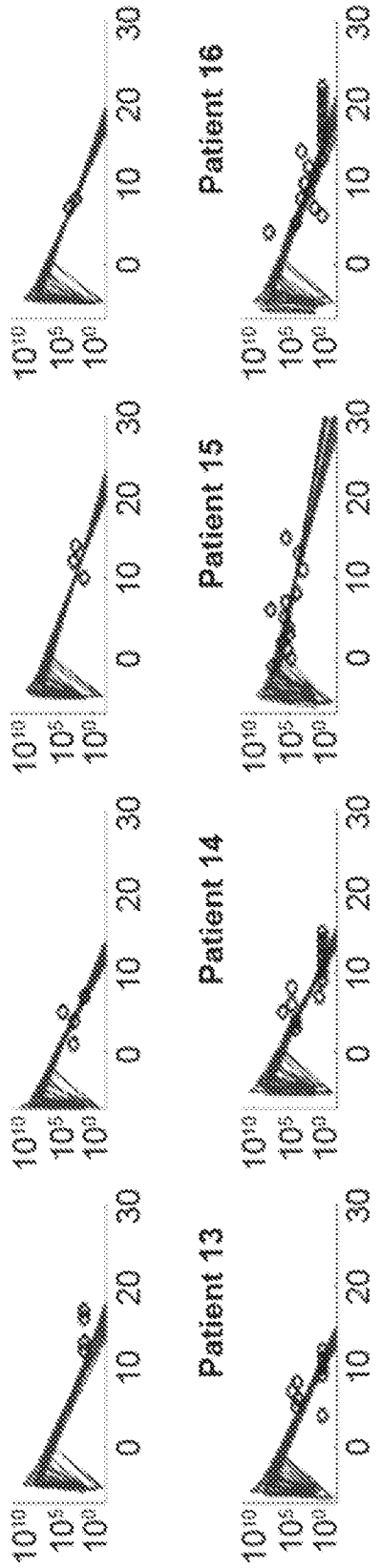


Figure 2 (continued)

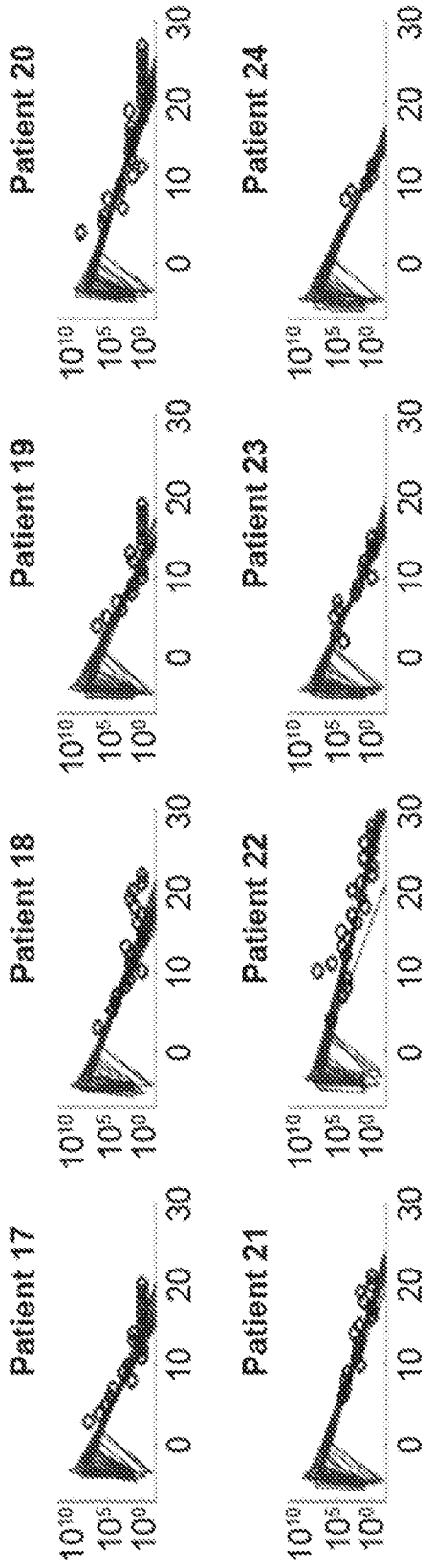
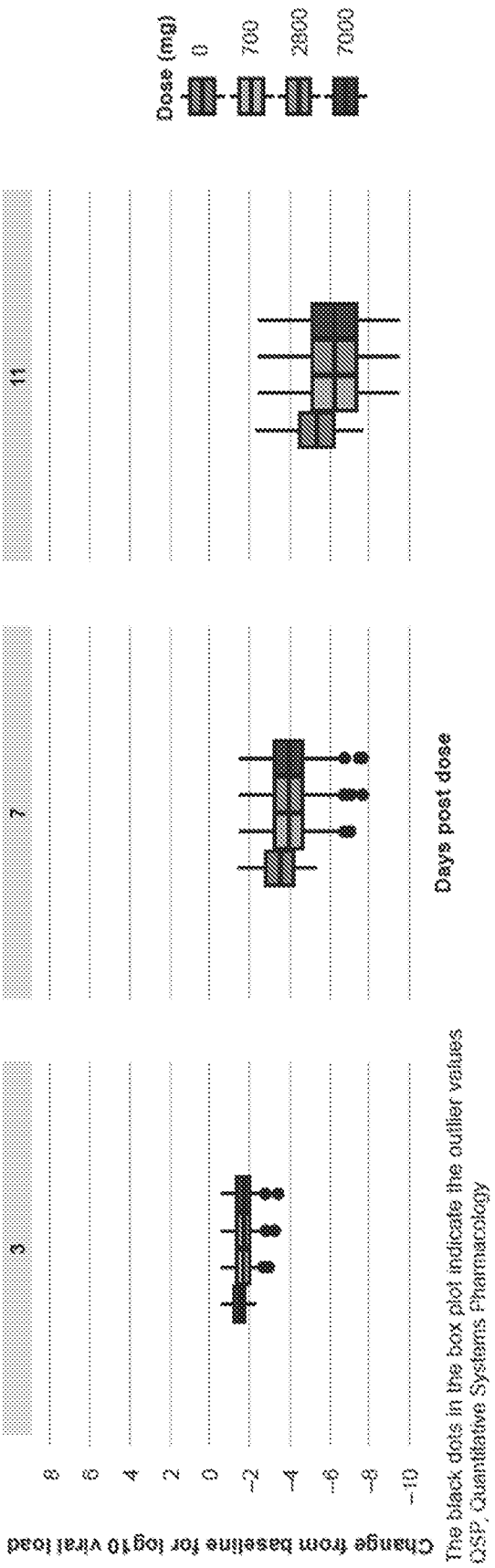


Figure 3



The black dots in the box plot indicate the outlier values
QSP, Quantitative Systems Pharmacology

Figure 4

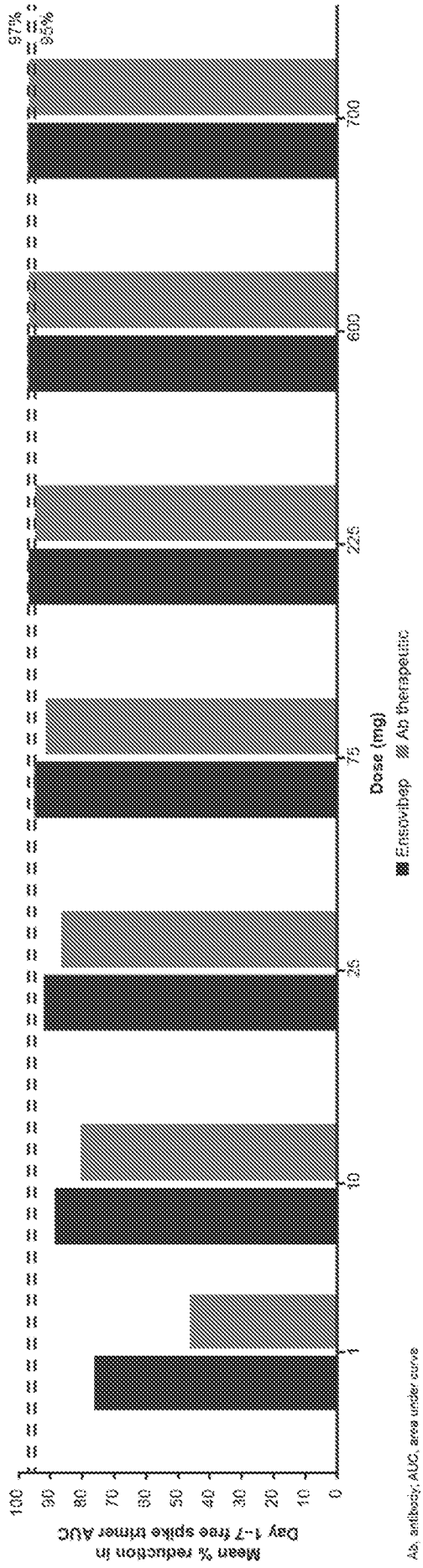
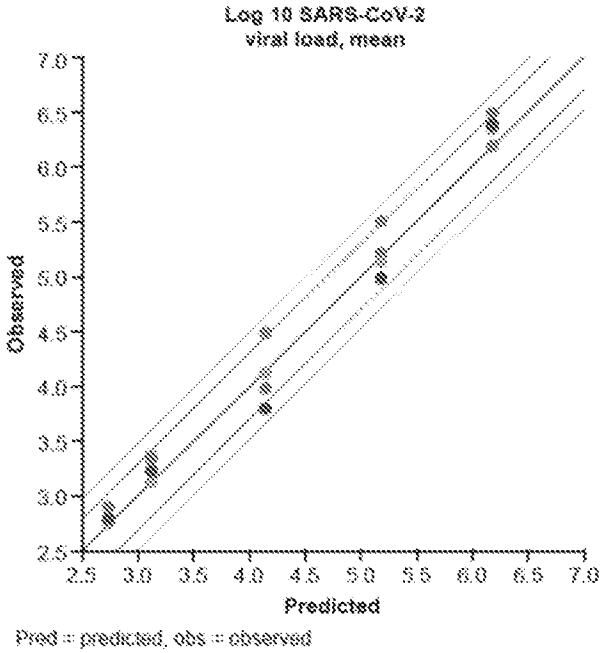

97%
95%
Mean % reduction in
Day 1–7 free spike trimer AUC
100
90
80
70
60
50
40
30
20
10
0
1
10
25
75
225
600
700
Dose (mg)
Ensovibep
Ab therapeutic
Ab, antibody; AUC, area under curve

Figure 5



Pred = predicted, obs = observed

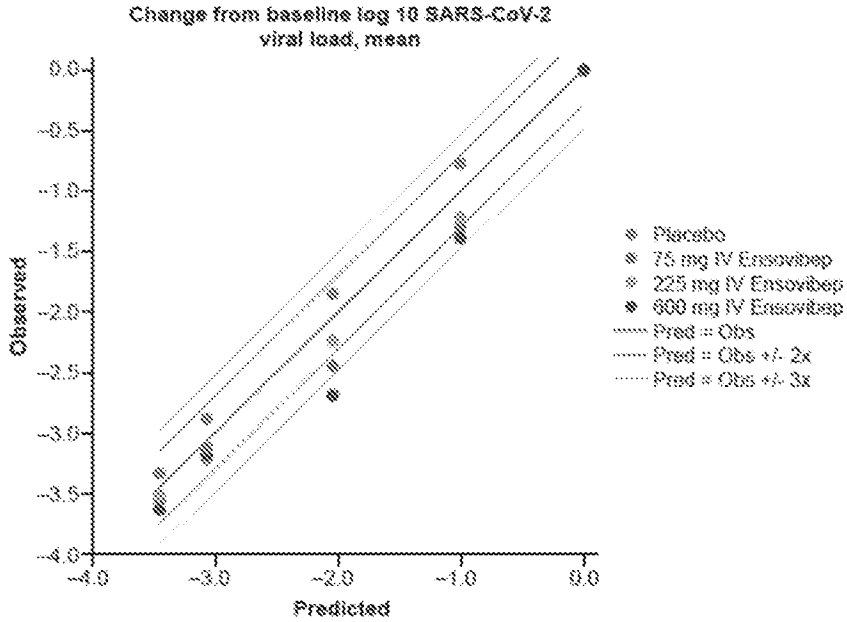
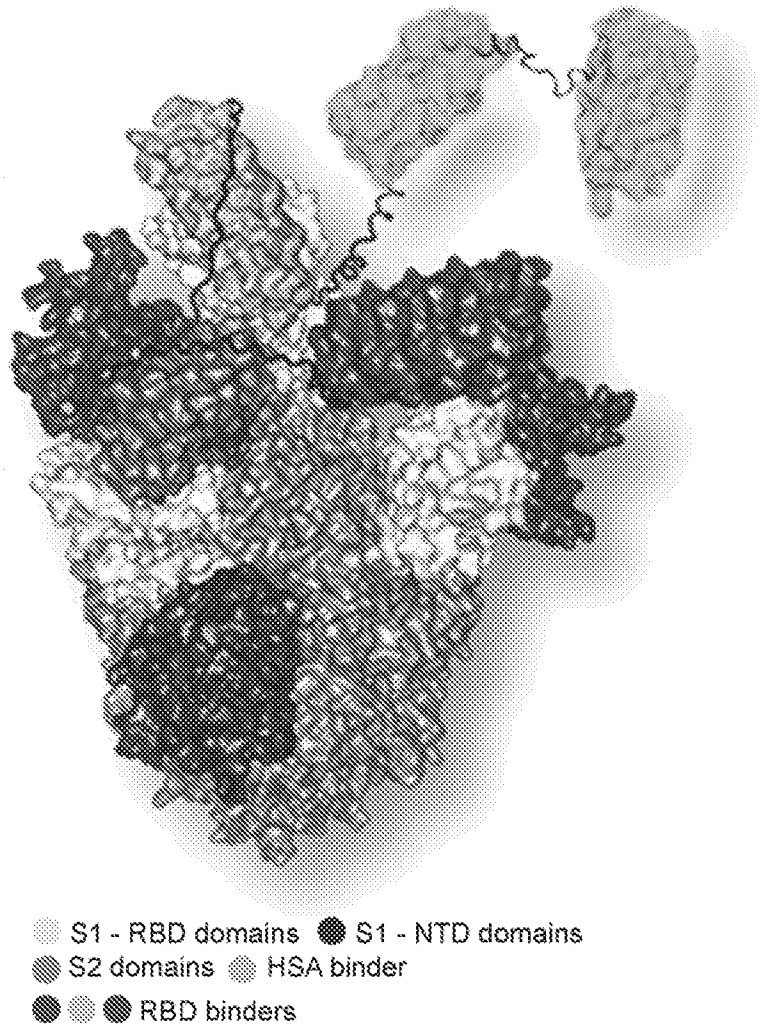


Figure 6


S1 - RBD domains
S1 - NTD domains
S2 domains
HSA binder
RBD binders

INTERNATIONAL SEARCH REPORT

International application No
PCT/IB2022/057822

A. CLASSIFICATION OF SUBJECT MATTER
INV. A61K38/16 A61P31/14 A61P11/00
ADD.

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

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
A61K A61P

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

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
EPO-Internal

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	CHRISTINA BARKAUSKAS: "Efficacy and Safety of Ensovibep for Adults Hospitalized With COVID-19", ANNALS OF INTERNAL MEDICINE, 9 August 2022 (2022-08-09), pages 1-10, XP002808431, Retrieved from the Internet: URL:https://www.ncbi.nlm.nih.gov/pmc/articles/PMC9384272/ [retrieved on 2023-01-16]	1,4-7, 9-11
Y	the whole document	1-19

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[x] Further documents are listed in the continuation of Box C.

[ ] See patent family annex.

* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance;; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance;; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
16 January 2023	31/01/2023

Name and mailing address of the ISA/	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL - 2280 HV Rijswijk Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016	Weisser, Dagmar

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INTERNATIONAL SEARCH REPORT

International application No
PCT/IB2022/057822

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	Rothenberger Sylvia ET AL: "Ensovibep, a novel trispecific DARPin candidate that protects against SARS-CoV-2 variants", bioRxiv, 26 February 2022 (2022-02-26), XP093014154, DOI: 10.1101/2021.02.03.429164 Retrieved from the Internet: URL:https://www.biorxiv.org/content/10.1101/2021.02.03.429164v4 [retrieved on 2023-01-16]	1,5-7,9
Y	pages 1,10	1-19

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INTERNATIONAL SEARCH REPORT

International application No.

PCT/IB2022/057822

Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.c of the first sheet)

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

   a. [X] forming part of the international application as filed.

   b. [ ] furnished subsequent to the international filing date for the purposes of international search (Rule 13*ter*.1(a)).

      [ ] accompanied by a statement to the effect that the sequence listing does not go beyond the disclosure in the international application as filed.

2. [ ] With regard to any nucleotide and/or amino acid sequence disclosed in the international application, this report has been established to the extent that a meaningful search could be carried out without a WIPO Standard ST.26 compliant sequence listing.

3. Additional comments: