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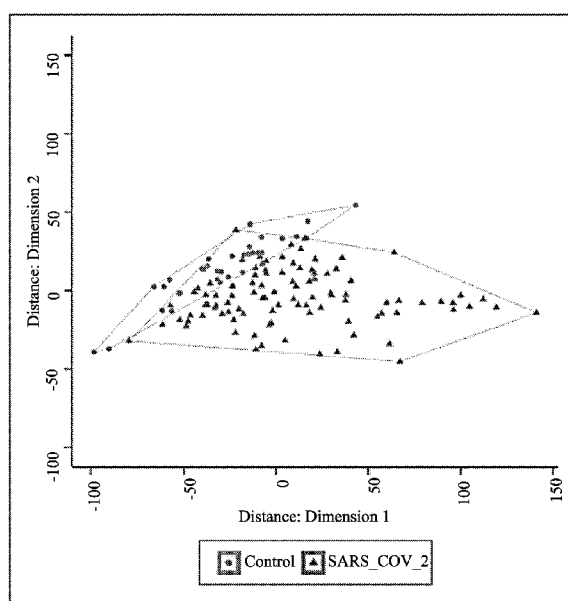


Fig. 1

(57) Abstract: The current disclosure provides compositions, methods, and kits for detecting the exposure to and infection by certain viruses. Specifically, the current disclosure allows for the rapid differential serological detection of exposure to, and infection by viruses. In particular, the current disclosure allows for the rapid differential serological detection of exposure to, and infection by corona virus (SARS-CoV-2). The current disclosure provides for peptides for the differential detection of SARS-CoV-2.



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**PEPTIDES FOR DETECTION AND DIFFERENTIATION OF ANTIBODY
RESPONSES TO SARS-CoV-2 AND OTHER HUMAN CORONA VIRUSES**

CROSS-REFERENCE TO OTHER APPLICATIONS

5 The present application claims priority to U.S. Patent Applications Serial Nos. 63/005,865 filed April 6, 2020 and 63/064,074 filed August 11, 2020, both of which are hereby incorporated by reference in its entirety.

STATEMENT OF GOVERNMENT SUPPORT

10 This invention was made with government support under AI109761 awarded by the National Institutes of Health. As such, the United States government has certain rights in this invention.

FIELD

15 This disclosure herein relates to the field of detection of antibody responses and exposure to viruses, in particular the detection of an antibody response and exposure to corona virus (SARS-CoV-2).

BACKGROUND

20 In December 2019, a cluster of cases were found positive for severe pneumonia-like illness in Wuhan, Hubei Province, China. A cluster of 41 patients that developed severe pneumonia like illness were shown to be infected with a novel coronavirus (nCoV) (Coronaviridae Study Group 2020). Sequence analyses of nCoV indicated it was a beta coronavirus, with high similarity to severe acute respiratory syndrome coronavirus (SARS-CoV). Subsequently, the International
25 Committee on Taxonomy of Viruses (ICTV) /WHO named this virus severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) and its disease coronavirus disease 2019 (COVID-19) (Zhou *et al.* 2020). On January 30, 2020, the WHO declared COVID-19 a global health emergency. Within 12 weeks after identification of first cluster, there were greater than 790,000 positive cases of COVID-19 globally, with 38,000 fatalities (19%). A COVID-19 outbreak
30 began in the United States with an evacuated patient from Wuhan on January 15, 2020. On February 4, 2020, the Secretary of the Department of Health and Human Services (HHS), USA also declared a public health emergency with significant potential to affect national security or the health and security of United States citizens living abroad. As of April 2020, the virus had resulted in over a million cases worldwide, with tens of thousand deaths.

The severity and rapid progression of COVID-19 has impacted the majority of the population through physical, social, and financial distress. The mitigation and containment measures forced global lockdowns, curfews and home isolations, that have already affected global economy to a greater extent than the terrorist attacks of September 11, 2001, or the 2008 financial crisis. It has been estimated the pandemic may cost approximately \$3 trillion and result in a loss of 5-10% global GDP

Diagnostic tests are critical for clinical and public health management of COVID-19. These tests fall into two broad categories: molecular assays used to identify infected individuals and to detect environmental contamination; and serological assays used to determine who has been exposed.

Molecular assays for detection of SARS-CoV-2 are useful in diagnosis of active infection but cannot provide insight into historical infection, especially in those individuals with mild or non-existence symptoms.

Additionally, vaccines are at the forefront of COVID-19 research. As these vaccines are developed, there is a need for rapid reliable assays for testing the efficacy of such vaccines.

Thus, there is a need for a sensitive, specific, and inexpensive high throughput serological assay for the diagnosis of SARS-CoV-2 exposure.

SUMMARY

The current disclosure provides compositions, methods, devices, and kits for detecting the exposure to, and infection by, certain viruses. Specifically, the current disclosure allows for the rapid differential serological detection of exposure to, and infection by viruses. In particular, the current disclosure allows for the rapid serological differential detection of exposure to, and infection by corona virus (SARS-CoV-2), and discrimination of antibody responses to linear epitopes specific to seven human coronaviruses: SARS-CoV-2; SARS; MERS; NL-63; OC-43; 229E; and HKU1.

Described herein is the identification of peptides that will enable specific and sensitive serological differential diagnosis of coronavirus infections on a wide range of platforms including but not limited to microarrays, ELISA, RIA, lateral flow, western blot, and bead-based assays. These assays will allow identification of individuals who are immune to future infection, capable of deployment to the workforce and who may serve as donors of antibodies for control of COVID-19. Cross-reactivity between SARS-CoV-2, SARS-CoV, MERS-CoV and seasonal coronaviruses (*e.g.*, OC43, 229E, HKU1, and NL63) complicate differential serodiagnosis and efforts to investigate the epidemiology of infection and linkage to disease.

Serology by plaque reduction neutralization test (PRNT) is an option but PRNTs are expensive, labor-intensive and require live virus and BSL3 facility. Furthermore, antibodies to conserved coronaviruses domains may confound assay specificity, particularly in the acute and early convalescent phase.

5 Described herein is a sensitive, highly multiplexed, microarray-based assay that enabled the discrimination of antibody responses to linear epitopes specific to seven human coronaviruses: SARS-CoV-2: SARS: MERS: NL-63: OC-43: 229E and HKU1. Also disclosed herein is the identification of 29 highly specific IgG epitopes for SARS-CoV-2 (Table 2) as well as additional 163 IgG epitopes for SARS-CoV-2 (Table 4) and 16 highly specific IgM
10 epitopes for SARS-CoV-2 (Table 3). Also disclosed herein are highly specific epitopes for a human coronavirus other than SARS-CoV-2, listed in Table 5.

The compositions, methods, devices, and kits for detection of exposure to, and infection by SARS-CoV-2 comprise specific peptides, isolated and non-isolated, which are strongly reactive with, and specific for SARS-CoV-2, *i.e.*, reactive and specific epitopes of antibodies
15 to SARS-CoV-2. Additionally, the compositions, methods, devices, and kits for differential detection of exposure to, and infection by SARS-CoV-2 comprise specific peptides, isolated and non-isolated, which are strongly reactive with, and specific for SARS-CoV-2, *i.e.*, reactive and specific epitopes of antibodies to SARS-CoV-2, as well as specific peptides, isolated and non-isolated, which are strongly reactive with, and specific for a human coronavirus other than
20 SARS-CoV-2.

Thus, one embodiment of the present disclosure is a peptide, which is reactive with, and specific for SARS-CoV-2 antibodies, listed in Table 2 (SEQ ID NOs: 1-26), Table 3 (SEQ ID NOs: 30-45), and Table 4 (SEQ ID NOs: 46-208). A further embodiment is a peptide, which is reactive with, and specific for a human coronavirus other than SARS-CoV-2, listed in Table
25 5 (SEQ ID NOs: 209-261).

A further embodiment is a collection or set of peptides comprising at least one peptide which is reactive with, and specific for SARS-CoV-2 antibodies, listed in Table 2 (SEQ ID NOs: 1-26), Table 3 (SEQ ID NOs: 30-45), and Table 4 (SEQ ID NOs: 46-208). A further embodiment is a collection or set of peptides comprising at least one peptide which is reactive
30 with, and specific for a human coronavirus other than SARS-CoV-2, listed in Table 5 (SEQ ID NOs: 209-261).

Yet a further embodiment of the present disclosure are collections or sets of peptides comprising amino acid sequences shifted one residue across one or more peptides chosen from the group consisting of the peptides that are reactive with, and specific for SARS-CoV-2

antibodies listed in listed in Table 2 (SEQ ID NOs: 1-26), Table 3 (SEQ ID NOs: 30-45), and Table 4 (SEQ ID NOs: 46-208). Yet a further embodiment of the present disclosure are collections or sets of peptides comprising amino acid sequences shifted one residue across one or more peptides chosen from the group consisting of the peptides which is reactive with, and
5 specific for a human coronavirus other than SARS-CoV-2, listed in Table 5 (SEQ ID NOs: 209-261).

These various peptides, or collections or sets of peptides can comprise or consist of peptides, that comprise or consist of 6 amino acids in length, 7 amino acids in length, 8 amino acids in length, 9 amino acids in length, 10 amino acids in length, 11 amino acids in length, 12
10 amino acids in length, 13 amino acids in length, 14 amino acids in length, 15 amino acids in length, 16 amino acids in length, 17 amino acids in length, 18 amino acids in length, 19 amino acids in length, 20 amino acids in length, up to 25 amino acids in length, up to 30 amino acids in length, up to 35 amino acids in length, up to 40 amino acids in length, and up to 50 amino acids in length.

15 The number of peptides in a used in a collection or set can range from 2 peptides to a number in the thousands to tens of thousands to hundreds of thousands.

In another aspect, the disclosure provides compositions comprising two or more disclosed peptides.

In another aspect, the disclosure provides nucleic acids comprising a sequence encoding
20 a disclosed peptide. In addition, the disclosure provides vectors comprising such nucleic acids, and host cells comprising such vectors. In certain embodiments, the vector is a shuttle vector. In other embodiments, the vector is an expression vector (*e.g.*, a bacterial or eukaryotic expression vector). In certain embodiments, the host cell is a bacterial cell. In other embodiments, the host cell is a eukaryotic cell.

25 In non-limiting examples, the antibodies to SARS-CoV-2 and/or other human coronaviruses can be detected using any number of immunodetection techniques, which include but are not necessarily limited to microarrays, ELISA, RIA, lateral flow, western blot, bead-based assays, dipstick type of assay or a SNAP test, multiplex antibody detection techniques of various kinds, or any modification of such assays that are suitable for detecting
30 antibodies of interest.

In one embodiment, the immunodetection technique is in the form of a programmable peptide array.

In certain embodiments, peptides are attached to or immobilized on a solid support. In one embodiment, the peptides are attached to a solid support through a metallic nanolayer. In

certain embodiments, the solid support is a bead (*e.g.*, a colloidal particle, metallic nanoparticle or nanoshell, or latex bead), a flow path in a lateral flow immunoassay device (*e.g.*, a porous membrane), a blot (*e.g.*, Western blot, a slot blot, or dot blot), a flow path in an analytical or centrifugal rotor, or a tube or well (*e.g.*, in a plate suitable for an ELISA assay or microarray).

5 In certain embodiments, peptides are isolated (*e.g.*, synthetic and/or purified) peptides. In certain embodiments, peptides are conjugated to a ligand. For example, in certain embodiments, the peptides are biotinylated. In other embodiments, the peptides are conjugated to streptavidin, avidin, or neutravidin. In other embodiments, the peptides are conjugated to a carrier protein (*e.g.*, serum albumin, keyhole limpet hemocyanin (KLH), or an immunoglobulin
10 Fc domain).

In another aspect, the disclosure provides devices. In certain embodiments, the devices are useful for performing an immunoassay. For example, in certain embodiments, the device is a lateral flow immunoassay device. In other embodiments, the device is an analytical or centrifugal rotor. In other embodiments, the device is a tube or a well, *e.g.*, in a plate suitable
15 for an ELISA assay or a microarray. In still other embodiments, the device is an electrochemical, optical, or opto-electronic sensor.

In certain embodiments, the device comprises at least one disclosed peptide. In other embodiments, the device comprises a collection or set of the disclosed peptides as described herein. In certain embodiments, the peptides are attached to or immobilized upon the device.

20 In another aspect, the disclosure provides methods of detecting in a sample an antibody to an epitope of SARS-CoV-2. In certain embodiments, the methods comprise contacting a sample with one or more disclosed peptides and detecting formation of an antibody-peptide complex comprising said peptide, wherein formation of said complex is indicative of the presence of an antibody to an epitope of SARS-CoV-2 antigen in said sample. In certain
25 embodiments, the methods comprise contacting the sample with a collection or set of different peptides described herein. In certain embodiments, the methods comprise contacting the sample with a collection or set of all of the peptides described herein.

In one embodiment, the disclosure provides a method for the serological detection of exposure to and/or infection by SARS-CoV-2, comprising the use of a peptide or peptides
30 which are reactive with, and specific for SARS-CoV-2 antibodies comprising: a peptide or peptides chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides

shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); or combinations thereof.

In further embodiments, the method for the serological detection of exposure to and/or infection by SARS-CoV-2 in a sample, comprises: contacting the sample with a peptide or peptides which are reactive with, and specific for SARS-CoV-2 antibodies comprising: a peptide or peptides chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); or combinations thereof; and detecting the binding between the anti-SARS-CoV-2 antibodies in the sample and the peptide or peptides.

Yet a further embodiment is a method for the differential serological detection of exposure to and/or infection by SARS-CoV-2, comprising the use of a peptide or peptides which are reactive with, and specific for SARS-CoV-2 antibodies and/or other human coronaviruses comprising: a peptide or peptides chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208);

and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations thereof.

In further embodiments, the method for the differential serological detection of exposure to and/or infection by SARS-CoV-2 in a sample, comprises: contacting the sample with a peptide or peptides which are reactive with, and specific for SARS-CoV-2 antibodies and/or other human coronaviruses comprising: a peptide or peptides chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations thereof; and detecting the binding between the anti-SARS-CoV-2 and/or anti-human coronavirus antibodies in the sample and the peptide or peptides.

In certain embodiments, the peptide or each peptide in the collection or set is an isolated (*e.g.*, synthetic and/or purified) peptide. In certain embodiments, the peptide, peptides or collection or set of peptides is attached to or immobilized upon a solid support. In one embodiment, the peptide, peptides or collection or set of peptides is attached to the solid support through a metallic (*e.g.*, gold) nanolayer. In certain embodiments, the solid support is a bead or plurality of beads (*e.g.*, a colloidal particle, a metallic nanoparticle or nanoshell, or a latex bead), a flow path in a lateral flow immunoassay device (*e.g.*, a porous membrane), a flow path in an analytical or centrifugal rotor, a blot (*e.g.*, Western blot, a slot blot, or dot blot), or a tube or a well (*e.g.*, in a plate suitable for an ELISA assay or microarray). In certain embodiments, the solid support comprises metal, glass, a cellulose-based material (*e.g.*, nitrocellulose), or a polymer (*e.g.*, polystyrene, polyethylene, polypropylene, polyester, nylon, or polysulfone).

A further embodiment is a method for the serological detection of exposure to and/or infection by SARS-CoV-2, comprising the use of a peptide microarray comprising peptide or peptides which are reactive with, and specific for SARS-CoV-2 antibodies comprising: a peptide or peptides chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); or combinations thereof.

In further embodiments, the method for the serological detection of exposure to and/or infection by SARS-CoV-2 in a sample, comprises: contacting the sample with a peptide microarray comprising a peptide or peptides which are reactive with, and specific for SARS-CoV-2 antibodies comprising: a peptide or peptides chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); or combinations thereof; and detecting the binding between the anti-SARS-CoV-2 antibodies in the sample and the peptide or peptides in the microarray.

Yet a further embodiment is a method for the differential serological detection of exposure to and/or infection by SARS-CoV-2, comprising the use of a peptide microarray comprising a peptide or peptides which are reactive with, and specific for SARS-CoV-2 antibodies and/or other human coronaviruses comprising: a peptide or peptides chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-

45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations thereof.

In further embodiments, the method for the differential serological detection of exposure to and/or infection by SARS-CoV-2 in a sample, comprises: contacting the sample with a peptide microarray comprising a peptide or peptides which are reactive with, and specific for SARS-CoV-2 antibodies and/or other human coronaviruses comprising: a peptide or peptides chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations thereof; and detecting the binding between the anti-SARS-CoV-2 and/or anti-human coronavirus antibodies in the sample and the peptide or peptides in the microarray.

In certain embodiments, the detecting step comprises performing an ELISA assay. In other embodiments, the detecting step comprises performing a lateral flow immunoassay. In other embodiments, the detecting step comprises performing an agglutination assay. In other embodiments, the detecting step comprises spinning the sample in an analytical or centrifugal

rotor. In other embodiments, the detecting step comprises analyzing the sample using a Western blot, a slot blot, or a dot blot. In still other embodiments, the detecting step comprises analyzing the sample with an electrochemical sensor, an optical sensor, or an opto-electronic sensor. In certain embodiments, the detecting step comprises performing a wavelength shift assay.

5 In some embodiments, the peptides consist of the amino acid sequences SEQ ID NOs: 1-261.

The present disclosure also includes systems and kits for the differential serological detection of exposure to and/or infection by SARS-COV-2.

10 **BRIEF DESCRIPTION OF THE FIGURES**

For the purpose of illustrating the invention, there are depicted in drawings certain embodiments of the invention. However, the invention is not limited to the precise arrangements and instrumentalities of the embodiments depicted in the drawings.

Fig. 1 shows the multidimensional scaling (MDS) of differential IgM peptide signals in assays of sera from subjects with a history of infection with SARS-CoV-2 (n=100) or without historical exposure to SARS-CoV-2 (controls) (n=32). Based on MDS analysis, samples with exposure of SARS-CoV-2 samples (green triangles) versus controls (red circles) clustered in overlapping groups.

Fig. 2 shows the multidimensional scaling (MDS) of differential IgG peptide signals in assays of sera from subjects with a history of infection with SARS-CoV-2 (n = 100) or without historical exposure to SARS-CoV-2 (controls) (n = 32). Based on MDS analysis, samples with exposure of SARS-CoV-2 samples (green triangles) versus controls (red circles) clustered into two separate groups.

Fig. 3 is a heatmap showing the proteome-wide linear epitope mapping of SARS-CoV-2-specific IgG antibodies by a HCoV peptide array. X axis represents 981 peptides from SARS-CoV-2 proteins, Y axis represents 132 samples tested using the HCoV peptide array. The heatmap is plotted with normalized values of individual peptide intensity in AU for each of the 132 plasma samples. Panel grids outlined in dotted lines show highly reactive areas in S and N proteins. *ORF1ab protein is large so divided partly in lower panel owing to larger size.

Fig. 4 are graphs of the reactivity intensities of each peptide from 29 immunogenic linear IgG epitopes are plotted on log scale (y-axis). Reactivity to selected peptides is plotted for corresponding groups shown in different colors (x-axis).

DETAILED DESCRIPTION

Definitions

The terms used in this specification generally have their ordinary meanings in the art, within the context of this invention and the specific context where each term is used. Certain
5 terms are discussed below, or elsewhere in the specification, to provide additional guidance to the practitioner in describing the methods of the invention and how to use them. Moreover, it will be appreciated that the same thing can be said in more than one way. Consequently, alternative language and synonyms may be used for any one or more of the terms discussed herein, nor is any special significance to be placed upon whether or not a term is elaborated or
10 discussed herein. Synonyms for certain terms are provided. A recital of one or more synonyms does not exclude the use of the other synonyms. The use of examples anywhere in the specification, including examples of any terms discussed herein, is illustrative only, and in no way limits the scope and meaning of the invention or any exemplified term. Likewise, the invention is not limited to its preferred embodiments.

15 As used herein, the term “sample” means any substance containing or presumed to contain antibodies, in particular those to SARS-CoV-2. The sample can be of natural or synthetic origin and can be obtained by any means known to those of skill in the art. The sample can be a sample of tissue or fluid isolated from a subject including but not limited to, plasma, serum, whole blood, spinal fluid, semen, amniotic fluid, lymph fluid, synovial fluid,
20 urine, tears, blood cells, organs, and tissue. Samples can be research, clinical, or environmental. Sample can also be blood products used to transfuse or treat. Samples can also be synthetic and include but are not limited to *in vitro* cell culture constituents including but not limited to conditioned medium, recombinant cells, and cell components.

As used herein, the term “subject” means any organism including, without limitation, a
25 mammal such as a mouse, a rat, a dog, a guinea pig, a ferret, a rabbit and a primate. In the preferred embodiment, the subject is a human being, a pet or livestock animal.

The term “patient” as used in this application means a human subject.

The term “detection”, “detect”, “detecting” and the like as used herein means as used herein means to discover the presence or existence of.

30 The terms “identification”, “identify”, “identifying” and the like as used herein means to recognize exposure to a specific virus or viruses in sample from a subject.

The term “peptide” includes any sequence of two or more amino acids. Peptide sequences specifically recited herein are written with the amino terminus on the left and the carboxy terminus on the right.

The term "amino acid," includes the residues of the natural amino acids (*e.g.* Ala, Arg, Asn, Asp, Cys, Glu, Gln, Gly, His, Hyl, Hyp, Ile, Leu, Lys, Met, Phe, Pro, Ser, Thr, Trp, Tyr, and Val) in D or L form, as well as unnatural amino acids (*e.g.* phosphoserine, phosphothreonine, phosphotyrosine, hydroxyproline, gamma-carboxyglutamate, hippuric acid, octahydroindole-2-carboxylic acid, statine, 1,2,3,4,-tetrahydroisoquinoline-3-carboxylic acid, penicillamine, omithine, citruline, alpha-methylalanine, para-benzoylphenylalanine, phenylglycine, propargylglycine, sarcosine, and tert-butylglycine). The term also includes natural and unnatural amino acids bearing a conventional amino protecting group (*e.g.* acetyl or benzyloxycarbonyl), as well as natural and unnatural amino acids protected at the carboxy terminus (*e.g.* as a (C₁-C₆) alkyl, phenyl or benzyl ester or amide).

An "antigen" (from antibody-generating) or "immunogen" is a substance that prompts the generation of antibodies and can cause an immune response. They may also be used for diagnostic or patient selection or characterization purposes.

Antibodies (also known as immunoglobulins (Ig)) are globulin proteins that are found in blood or other bodily fluids of vertebrates and are used by the immune system to identify and neutralize foreign objects, such as bacteria and viruses. They are typically made of basic structural units--each with two large heavy chains and two small light chains--to form, for example, monomers with one unit, dimers with two units or pentamers with five units. Antibodies are produced by B cells. There are several different types of antibody heavy chains, and several different kinds of antibodies, which are grouped into different isotypes based on which heavy chain they possess. Five different antibody isotypes are known in mammals, which perform different roles, and help direct the appropriate immune response for each different type of foreign object they encounter.

Although the general structure of all antibodies is very similar, a small region at the tip of the protein is extremely variable, allowing millions of antibodies with slightly different tip structures to exist. This region is known as the hypervariable region. Each of these variants can bind to a different target, known as an antigen. This huge diversity of antibodies allows the immune system to recognize an equally wide diversity of antigens. The part of the antigen recognized by an antibody is termed an "epitope." These epitopes bind with their antibody in a highly specific interaction, called induced fit, which allows antibodies to identify and bind only their specific epitope in the matching antigen(s) in the midst of the millions of different molecules that make up an organism. Recognition of an antigen by an antibody tags it for attack by other parts of the immune system. Antibodies can also neutralize targets directly by, for

example, binding to a part of a pathogen that it needs to cause an infection. Production of antibodies is the main function of the humoral immune system.

As used herein, the term “isolated” and the like means that the referenced material is free of components found in the natural environment in which the material is normally found.

5 In particular, isolated biological material is free of cellular components. An isolated peptide or protein may be associated with other proteins or nucleic acids, or both, with which it associates in the cell, or with cellular membranes if it is a membrane-associated protein. An isolated material may be, but need not be, purified.

The term “substantially purified,” as used herein, refers to a molecule, such as a peptide, 10 that is substantially free of cellular material (proteins, lipids, carbohydrates, nucleic acids), culture medium, chemical precursors, chemicals used in synthesis of the peptide, or combinations thereof. A peptide that is substantially purified has less than about 40%, 30%, 25%, 20%, 15%, 10%, 5%, 2%, 1% or less of the cellular material, culture medium, other polypeptides, chemical precursors, and/or chemicals used in synthesis of the peptide. 15 Accordingly, a substantially pure molecule, such as a peptide, can be at least about 60%, 70%, 75%, 80%, 85%, 90%, 95%, 98%, or 99%, by dry weight, the molecule of interest.

The term “about” or “approximately” means within an acceptable error range for the particular value as determined by one of ordinary skill in the art, which will depend in part on how the value is measured or determined, *i.e.*, the limitations of the measurement system, *i.e.*, 20 the degree of precision required for a particular purpose, such as a pharmaceutical formulation. For example, “about” can mean within 1 or more than 1 standard deviations, per the practice in the art. Alternatively, “about” can mean a range of up to 20%, preferably up to 10%, more preferably up to 5%, and more preferably still up to 1% of a given value. Alternatively, particularly with respect to biological systems or processes, the term can mean within an order 25 of magnitude, preferably within 5-fold, and more preferably within 2-fold, of a value. Where particular values are described in the application and claims, unless otherwise stated, the term “about” meaning within an acceptable error range for the particular value should be assumed.

The current disclosure enhances the differential diagnosis and management of SARS-CoV-2 by establishing a new serologic assay platform for profiling a subject’s pathogen 30 exposure history, as well as providing peptides which are reactive with and sensitive to antibodies to SARS-CoV-2.

In contrast to molecular diagnostics where advances in technology such as polymerase chain reaction and high-throughput screening have dramatically improved sensitivity, specificity and breadth over the past 20 years, serologic methods remained largely unchanged.

This lag is important given the role of serology in establishing the distribution and frequency of infection, testing the significance of association between the finding of an agent and disease, and in focusing efforts in pathogen discovery. Described herein is a sensitive, unbiased, highly multiplexed platform for diagnostic serology of SARS-CoV-2 in particular. This peptide array-based platform will enable new strategies for investigating the epidemiology and pathogenesis of acute and chronic diseases due to infection and for monitoring humoral responses to vaccines and immunomodulatory drugs. It will also serve as a screening tool for rapid selection of key informative peptides that can be used in established, inexpensive, alternative platforms including lateral flow immunoassays. Such applications will have practical utility for both clinical medicine and public health by enabling retrospective differential diagnosis of an infectious illness (when genetic footprints of the agent may no longer be present), and in facilitating outbreak investigation and surveillance.

This is particularly useful in infection by SARS-CoV-2 as a patient could no longer have the evidence of acute infection, *e.g.*, viral nucleic acids, but have had a SARS-CoV-2 infection in the past. These assays will allow identification of individuals who are immune to future infection, capable of deployment to the workforce and who may serve as donors of antibodies for control of COVID-19, as well as for testing the effectiveness of vaccines for SARS-CoV-2.

Additionally, serodiagnosis of SARS-CoV-2 infection can be impeded by immunological cross-reactivity among the human coronaviruses (HCoVs): SARS-CoV-2; SARS-CoV-1; MERS-CoV; OC43; 229E; HKU1; and NL63. Disclosed herein are SARS-CoV-2 peptides that enable discrimination between exposure to SARS-CoV-2 and other HCoVs.

A high-density peptide microarray and plasma samples collected at two time points from 50 subjects with SARS-CoV-2 infection confirmed by qPCR, samples collected in 2004–2005 from 11 subjects with IgG antibodies to SARS-CoV-1, 11 subjects with IgG antibodies to other seasonal human coronaviruses (HCoV), and 10 healthy human subjects were used. Through statistical modeling with linear regression and multidimensional scaling specific peptides were identified that were reassembled to identify 29 linear IgG SARS-CoV-2 epitopes that were immunoreactive with plasma from individuals who had asymptomatic, mild or severe SARS-CoV-2 infections, as well as 16 IgM SARS-CoV-2 epitopes and an additional 163 IgG epitopes for SARS-CoV-2. Also disclosed herein are highly specific epitopes for a human coronavirus other than SARS-CoV-2.

Thus disclosed herein is a sensitive, highly multiplexed, microarray-based assay that enabled the discrimination of antibody responses to linear epitopes specific to seven human coronaviruses: SARS-CoV-2; SARS; MERS; NL-63; OC-43; 229E; and HKU1 and the identification of 29 highly specific IgG epitopes, 16 IgM epitopes for SARS-CoV-2, and additional 163 IgG epitopes for SARS-CoV-2 and 53 highly specific epitopes for a human coronavirus other than SARS-CoV-2.

The work reported herein has been published in recently (see Mishra *et al.* Immunoreactive peptide maps of SARS-CoV-2. *Commun Biol* 4, 225 (2021) (herein incorporated by reference in its entirety)).

Isolated Peptides Specific and Sensitive for SARS-COV-2 Antibodies (SARS-COV-2 Reactive Peptides) and other Human Coronavirus Antibodies (Human Coronavirus Reactive Peptides)

The present disclosure includes isolated peptides which are strongly reactive with, and sensitive to antibodies to SARS-CoV-2 in a patient sample. These peptides can be used in any type of serological assay or platform, now known or later developed, to screen for the presence of antibodies to SARS-CoV-2 and to determine if a subject has had an infection by and/or exposure to SARS-COV-2. These peptides can also be used to test for and monitor humoral responses to vaccines and immunomodulatory drugs, thus, being useful for the development of treatment and preventative agents for SARS-CoV-2.

One embodiment is an isolated peptide highly specific for SARS-CoV-2 listed in Table 2 (SEQ ID NOs: 1-29). A further embodiment is an isolated peptide highly specific for SARS-CoV-2 listed in Table 3 (SEQ ID NOs: 30-46). A further embodiment is an isolated peptide highly specific for SARS-CoV-2 listed in Table 4 (SEQ ID NOs: 46-208).

In further embodiments, isolated peptides highly specific for other human coronaviruses are also provided for herein. Isolated peptides highly specific for human coronavirus HKU1 are listed in Table 5 (SEQ ID NOs: 209-223). Isolated peptides highly specific for human coronavirus NL63 are listed in Table 5 (SEQ ID NOs: 224-233). Isolated peptides highly specific for human coronavirus OC43 are listed in Table 5 (SEQ ID NOs: 234-247). Isolated peptides highly specific for human coronavirus 229E are listed in Table 5 (SEQ ID NOs: 248-252). Isolated peptides highly specific for human coronavirus SARS-CoV-1 are listed in Table 5 (SEQ ID NOs: 253-261).

A further embodiment comprises a collection or set of peptides comprising amino acid sequences shifted one residue across any of the peptides listed in Table 2 (SEQ ID NOs: 1-26), Table 3 (SEQ ID NOs: 30-45), Table 4 (SEQ ID NOs: 46-208), and Table 5 (SEQ ID NOs:

209-261). This collection or set can contain peptides that are 6 amino acids in length, 7 amino acids in length, 8 amino acids in length, 9 amino acids in length, 10 amino acids in length, 11 amino acids in length, and up to 12 amino acids in length.

In one embodiment, each peptide in the set or collection is 12 amino acids in length.

5 Other embodiments include a collection or set of isolated peptides comprising or consisting of amino acid sequences shifted one residue across one, two, three, four, five, six, seven, eight, nine, ten, eleven or all twelve of these twelve peptides. This collection or set can comprise or consist of isolated peptides comprising or consisting of 6 amino acids in length, 7 amino acids in length, 8 amino acids in length, 9 amino acids in length, 10 amino acids in length, 11 amino acids in length, and up to 12 amino acids in length.

Peptides of 12 amino acids were constructed based upon work that shows that antibodies bind to linear peptide sequences ranging from 5 to 9 amino acids in length and bind most efficiently when targets are flanked by additional amino acids (Buus *et al.* 2012). However, peptides containing less than 12 amino acids in length and more than 12 amino acids in length can be used. Peptides 13 amino acids in length, 14 amino acids in length, 15 amino acids in length, 16 amino acids in length, 17 amino acids in length, 18 amino acids in length, 19 amino acids in length, 20 amino acids in length, up to 25 amino acids in length, up to 30 amino acids in length, up to 35 amino acids in length, up to 40 amino acids in length, and up to 50 amino acids in length can be used.

20 In certain embodiments, the peptides described herein are produced by synthetic chemistry (*i.e.*, a "synthetic peptide"). In other embodiments, peptides of the invention are produced biologically. An isolated peptide of the invention can be in water, a buffer, or in a dry form awaiting reconstitution, *e.g.*, as part of a kit. An isolated peptide of the present invention can be in the form of a pharmaceutically acceptable salt. Suitable acids and bases that are capable of forming salts with the peptides of the present invention are well known to those of skill in the art and include inorganic and organic acids and bases.

In certain embodiments, the peptides described herein are modified. The peptides may be modified by a variety of techniques, such as by denaturation with heat and/or a detergent (*e.g.*, SDS). Alternatively, peptides may be modified by association with one or more further moieties. The association can be covalent or non-covalent, and can be, for example, via a terminal amino acid linker, such as lysine or cysteine, a chemical coupling agent, or a peptide bond. The additional moiety can be, for example, a ligand, a ligand receptor, a fusion partner, a detectable label, an enzyme, or a substrate that immobilizes the peptide.

The disclosed peptides can be conjugated to a ligand, such as biotin (*e.g.*, via a cysteine or lysine residue), a lipid molecule (*e.g.*, via a cysteine residue), or a carrier protein (*e.g.*, serum albumin, immunoglobulin Fc domain, keyhole limpet hemocyanin (KLH) via *e.g.*, a cysteine or lysine residue). Attachment to ligands, such as biotin, can be useful for associating the peptide with ligand receptors, such as avidin, streptavidin, polymeric streptavidin, or neutravidin. Avidin, streptavidin, polymeric streptavidin, or neutravidin, in turn, can be linked to a signaling moiety (*e.g.*, an enzyme, such as horse radish peroxidase (HRP) or alkaline phosphatase (ALP), or other moiety that can be visualized, such as a metallic nanoparticle or nanoshell (*e.g.*, colloidal gold) or a fluorescent moiety), or a solid substrate (*e.g.*, nitrocellulose membrane). Alternatively, the peptides of the invention can be fused or linked to a ligand receptor, such as avidin, streptavidin, polymeric streptavidin, or neutravidin, thereby facilitating the association of the peptides with the corresponding ligand, such as biotin and any moiety (*e.g.*, signaling moiety) or solid substrate attached thereto. Examples of other ligand-receptor pairs are well-known in the art and can similarly be used.

The peptides can be fused to a fusion partner (*e.g.*, a peptide or other moiety) that can be used to improve purification, to enhance expression of the peptide in a host cell, to aid in detection, and to stabilize the peptide. Examples of suitable compounds for fusion partners include carrier proteins (*e.g.*, serum albumin, immunoglobulin Fc domain, KLH), and enzymes (*e.g.*, horse radish peroxidase (HRP), beta-galactosidase, glutathione-S-transferase, alkaline phosphatase). The fusion can be achieved by means of a peptide bond. For example, peptides of the invention and fusion partners can be fusion proteins and can be directly fused in-frame or can comprise a peptide linker.

In addition, the disclosed peptides may be modified to include any of a variety of known chemical groups or molecules. Such modifications include, but are not limited to, glycosylation, acetylation, acylation, ADP-ribosylation, amidation, covalent attachment to polyethylene glycol (*e.g.*, PEGylation), covalent attachment of flavin, covalent attachment of a heme moiety, covalent attachment of a nucleotide or nucleotide derivative, covalent attachment of a lipid or lipid derivative, covalent attachment of phosphatidylinositol, cross-linking, cyclization, disulfide bond formation, demethylation, formation of covalent cross-links, formation of cystine, formation of pyroglutamate, formylation, gamma carboxylation, glycosylation, GPI anchor formation, hydroxylation, iodination, methylation, myristoylation, oxidation, proteolytic processing, phosphorylation, prenylation, racemization, selenoylation, sulfation, ubiquitination, modifications with fatty acids, and transfer-RNA mediated addition

of amino acids to proteins such as arginylation. Analogues of an amino acid (including unnatural amino acids) and peptides with substituted linkages are also included.

The disclosed peptides that consist of any of the sequences discussed herein may be modified by any of the discussed modifications. Such peptides still “comprise” or “consist of”
5 the amino acids.

Modifications as set forth above are well-known to those of skill in the art and have been described in great detail in the scientific literature.

Nucleic Acids Comprising a Sequence Encoding the SARS-CoV-2 Reactive Peptides and 10 Other Human Coronavirus Reactive Peptides

In another aspect, the disclosure provides nucleic acids comprising a sequence encoding any disclosed peptide. Nucleic acids of the invention can be single- or double-stranded. A nucleic acid can be RNA, DNA, cDNA, genomic DNA, chemically synthesized RNA or DNA or combinations thereof. The nucleic acids can be purified free of other components, such as
15 proteins, lipids and other polynucleotides. For example, the nucleic acids can be 50%, 75%, 90%, 95%, 96%, 97%, 98%, 99%, or 100% purified. The nucleic acids encode the peptides described herein. In certain embodiments, the nucleic acids encode a peptide having the sequence listed in Table 2 (SEQ ID NOs: 1-26), Table 3 (SEQ ID NOs: 30-45), Table 4 (SEQ ID NOs: 46-208) and/or Table 5 (SEQ ID NOs: 209-261). Nucleic acids can comprise other
20 nucleotide sequences, such as sequences coding for linkers, signal sequences, TMR stop transfer sequences, transmembrane domains, or ligands useful in protein purification such as glutathione-S-transferase, histidine tag, and staphylococcal protein A.

Methods for preparing polynucleotides operably linked to an expression control sequence and expressing them in a host cell are well-known in the art. See, *e.g.*, U.S. Pat. No.
25 4,366,246. A nucleic acid of the invention is operably linked when it is positioned adjacent to or close to one or more expression control elements, which direct transcription and/or translation of the polynucleotide.

Thus, for example, the peptides described herein can be produced recombinantly following conventional genetic engineering techniques. To produce a recombinant peptide, a
30 nucleic acid encoding the peptide is inserted into a suitable expression system. Generally, a recombinant molecule or vector is constructed in which the polynucleotide sequence encoding the selected peptide is operably linked to an expression control sequence permitting expression of the peptide. Numerous types of appropriate expression vectors are known in the art, including, *e.g.*, vectors containing bacterial, viral, yeast, fungal, insect or mammalian

expression systems. Methods for obtaining and using such expression vectors are well-known. For guidance in this and other molecular biology techniques used for compositions or methods of the invention, see, *e.g.*, Sambrook *et al.*, Molecular Cloning, A Laboratory Manual, current edition, Cold Spring Harbor Laboratory, New York; Miller *et al.*, Genetic Engineering, 8:277-298 (Plenum Press, current edition), Wu *et al.*, Methods in Gene Biotechnology (CRC Press, New York, N.Y., current edition), Recombinant Gene Expression Protocols, in Methods in Molecular Biology, Vol. 62, (Tuan, ed., Humana Press, Totowa, N.J., current edition), and Current Protocols in Molecular Biology, (Ausabel *et al.*, Eds.,) John Wiley & Sons, NY (current edition), and references cited therein.

Accordingly, the disclosure also provides vectors comprising nucleic acids described herein, and host cells comprising such vectors. In certain embodiments, the vector is a shuttle vector. In other embodiments, the vector is an expression vector (*e.g.*, a bacterial or eukaryotic expression vector). In certain embodiments, the host cell is a bacterial cell. In other embodiments, the host cell is a eukaryotic cell.

Platforms, Assays and Device for Using the SARS-CoV-2 Reactive Peptides and Other Human Coronavirus Reactive Peptides

There are a number of different conventional assays for detecting formation of an antibody-peptide complex comprising a peptide or peptides of the invention. For example, the detecting step can comprise performing a microarray assay, an ELISA assay, performing an immunofluorescence assay, performing a lateral flow immunoassay, performing an agglutination assay, performing a wavelength shift assay, performing a Western blot, slot blot, or dot blot, analyzing the sample in an analytical or centrifugal rotor, or analyzing the sample with an electrochemical, optical, or opto-electronic sensor. These different assays are described herein and/or are well-known to those skilled in the art.

Thus, the peptides disclosed herein can be used in any assay, format or platform for antibody detection including but not limited to microarrays, ELISA, RIA, lateral flow, western blot, and bead-based assays, as well as those platforms that are later developed.

In certain embodiments, the assay comprises: immobilizing the antibody(s) in the sample; adding a peptide disclosed herein; and detecting the degree of antibody bound to the peptide, *e.g.*, by the peptide being labeled or by adding a labeled substance, such as a labeled binding partner (*e.g.*, streptavidin-HRP or streptavidin-colloidal gold complex) or a labeled antibody which specifically recognizes the peptide.

In other embodiments, the assay comprises: immobilizing a peptide disclosed herein; adding the sample containing antibodies; and detecting the amount of antibody bound to the peptide, *e.g.*, by adding another peptide disclosed herein conjugated, directly or indirectly, to a label (*e.g.*, metallic nanoparticle or metallic nanoshell, fluorescent label, or enzyme (*e.g.*, horseradish peroxidase or alkaline phosphatase)) or by adding a labeled substance, such as a binding partner or a labeled antibody which specifically recognizes the sample antibodies (*e.g.*, anti-human IgG antibodies, or anti-human IgM antibodies).

In other embodiments, the assay comprises: immobilizing a peptide disclosed herein; adding the sample containing antibodies; and detecting the amount of antibody bound to the peptide, *e.g.*, by adding a first binding partner which specifically recognizes the sample antibodies (*e.g.*, anti-human IgG antibodies, or anti-human IgM antibodies), and further adding a second binding partner, wherein the second binding partner is labeled and recognizes said first binding partner.

In still other embodiments, the assay comprises: reacting the peptide and the sample containing antibodies without any of the reactants being immobilized, and then detecting the amount of complexes of antibody and peptide, *e.g.*, by the peptide being labeled or by adding a labeled substance, such as a labeled binding partner (*e.g.*, streptavidin-HRP or streptavidin-colloidal gold complex) or a labeled antibody which specifically recognizes the peptide.

Immobilization of a peptide can be either covalent or non-covalent, and the non-covalent immobilization can be non-specific (*e.g.*, non-specific binding to a polystyrene surface in a microtiter well). Specific or semi-specific binding to a solid or semi-solid carrier, support or surface, can be achieved by the peptide having, associated with it, a moiety which enables its covalent or non-covalent binding to the solid or semi-solid carrier, support or surface. For example, the moiety can have affinity to a component attached to the carrier, support or surface. In this case, the moiety may be, for example, a biotin or biotinyl group or an analogue thereof bound to an amino acid group of the peptide, and the component is then avidin, streptavidin, neutravidin, or an analogue thereof.

Suitable carriers, supports, and surfaces include, but are not limited to, metallic nanolayers, beads (*e.g.*, magnetic beads, colloidal particles or metallic nanoparticles or nanoshells, such as colloidal gold, or particles or nanoparticles comprising silica, latex, polystyrene, polycarbonate, or PDVF), latex of co-polymers such as styrene-divinyl benzene, hydroxylated styrene-divinyl benzene, polystyrene, carboxylated polystyrene, beads of carbon black, non-activated or polystyrene or polyvinyl chloride activated glass, epoxy-activated

porous magnetic glass, gelatin or polysaccharide particles or other protein particles, red blood cells, mono- or polyclonal antibodies or Fab fragments of such antibodies.

The protocols for immunoassays using antigens for detection of specific antibodies are well known in art. For example, a conventional sandwich assay can be used, or a conventional competitive assay format can be used.

Devices for performing specific binding assays, especially immunoassays, are known and can be readily adapted for use in the present methods. Solid-phase assay devices include microtiter plates, flow-through assay devices (*e.g.*, lateral flow immunoassay devices), dipsticks, and immunocapillary or immunochromatographic immunoassay devices.

In embodiments, the solid or semi-solid surface or carrier is the floor or wall in a microtiter well, a filter surface or membrane (*e.g.*, a nitrocellulose membrane or a PVDF (polyvinylidene fluoride) membrane), a hollow fiber, a beaded chromatographic medium (*e.g.*, an agarose or polyacrylamide gel), a magnetic bead, a fibrous cellulose matrix, an HPLC matrix, an FPLC matrix, a substance having molecules of such a size that the molecules with the peptide bound thereto, when dissolved or dispersed in a liquid phase, can be retained by means of a filter, a substance capable of forming micelles or participating in the formation of micelles allowing a liquid phase to be changed or exchanged without entraining the micelles, a water-soluble polymer, or any other suitable carrier, support or surface.

In some embodiments, the peptide is provided with a suitable label which enables detection. Conventional labels may be used which are capable, alone or in concert with other compositions or compounds, of providing a detectable signal. Suitable labels include, but are not limited to, enzymes (*e.g.*, HRP, beta-galactosidase, or alkaline phosphatase), fluorescent labels, radioactive labels, colored latex particles, and metal-conjugated labels (*e.g.*, metallic nanolayers, metallic nanoparticle- or metallic nanoshell-conjugated labels). Suitable metallic nanoparticle or metallic nanoshell labels include, but are not limited to, gold particles, silver particles, copper particles, platinum particles, cadmium particles, composite particles, gold hollow spheres, gold-coated silica nanoshells, and silica-coated gold shells. Metallic nanolayers suitable for detectable layers include nanolayers comprised of cadmium, zinc, mercury, and noble metals, such as gold, silver, copper, and platinum.

Suitable detection methods include, but are not limited to, detection of an agent which is tagged, directly or indirectly, with a colorimetric assay (*e.g.*, for detection of HRP or beta-galactosidase activity), visual inspection using light microscopy, immunofluorescence microscopy, including confocal microscopy, or by flow cytometry (FACS), autoradiography (*e.g.*, for detection of a radioactively labeled agent), electron microscopy, immunostaining,

subcellular fractionation, or the like. In one embodiment, a radioactive element (*e.g.*, a radioactive amino acid) is incorporated directly into a peptide chain. In another embodiment, a fluorescent label is associated with a peptide via biotin/avidin interaction, association with a fluorescein conjugated antibody, or the like. In one embodiment, a detectable specific binding partner for the antibody is added to the mixture. For example, the binding partner can be a detectable secondary antibody or other binding agent (*e.g.*, protein A, protein G, protein L or combinations thereof) which binds to the first antibody. This secondary antibody or other binding agent can be labeled with, for example, a radioactive, enzymatic, fluorescent, luminescent, metallic nanoparticle or metallic nanoshell (*e.g.* colloidal gold), or other detectable label, such as an avidin/biotin system. In another embodiment, the binding partner is a peptide disclosed herein, which can be conjugated directly or indirectly to an enzyme, such as horseradish peroxidase or alkaline phosphatase or other signaling moiety. In such embodiments, the detectable signal is produced by adding a substrate of the enzyme that produces a detectable signal, such as a chromogenic, fluorogenic, or chemiluminescent substrate.

In some embodiments, the detection procedure comprises visibly inspecting the antibody-peptide complex for a color change or inspecting the antibody-peptide complex for a physical-chemical change. Physical-chemical changes may occur with oxidation reactions or other chemical reactions. They may be detected by eye, using a spectrophotometer, or the like.

One assay format is a lateral flow immunoassay format. Antibodies to human or animal immunoglobulins, can be labeled with a signal generator or reporter (*e.g.*, colloidal gold) that is dried and placed on a glass fiber pad (sample application pad or conjugate pad). The diagnostic peptide is immobilized on membrane, such as nitrocellulose or a PVDF (polyvinylidene fluoride) membrane. When a sample is applied to the sample application pad (or flows through the conjugate pad), it dissolves the labeled reporter, which then binds to all antibodies in the sample. The resulting complexes are then transported into the next membrane (PVDF or nitrocellulose containing the diagnostic peptide) by capillary action. If antibodies against the diagnostic peptide are present, they bind to the diagnostic peptide striped on the membrane, thereby generating a signal (*e.g.*, a band that can be seen or visualized). An additional antibody specific to the labeled antibody or a second labeled antibody can be used to produce a control signal.

An alternative format for the lateral flow immunoassay comprises the peptides or compositions being conjugated to a ligand (*e.g.*, biotin) and complexed with labeled ligand receptor (*e.g.*, streptavidin-colloidal gold). The labeled peptide complexes can be placed on the

sample application pad or conjugate pad. Anti-human IgG/IgM or anti-animal IgG/IgM antibodies are immobilized on a membrane, such as nitrocellulose or PVDF, at a test site. When sample is added to the sample application pad, antibodies in the sample react with the labeled peptide complexes such that antibodies that bind to peptides become indirectly labeled. The antibodies in the sample are then transported into the next membrane (PVDF or nitrocellulose containing the diagnostic peptide) by capillary action and bind to the immobilized anti-human IgG/IgM or anti-animal IgG/IgM antibodies. If any of the sample antibodies are bound to the labeled peptides, the label associated with the peptides can be seen or visualized at the test site.

Another assay for the screening of blood products or other physiological or biological fluids is an enzyme linked immunosorbent assay, *i.e.*, an ELISA. Typically in an ELISA, isolated peptides or collection or set of peptides disclosed herein, are adsorbed to the surface of a microtiter well directly or through a capture matrix (*e.g.*, an antibody). Residual, non-specific protein-binding sites on the surface are then blocked with an appropriate agent, such as bovine serum albumin (BSA), heat-inactivated normal goat serum (NGS), or BLOTTO (a buffered solution of nonfat dry milk which also contains a preservative, salts, and an antifoaming agent). The well is then incubated with a biological sample suspected of containing specific antibody. The sample can be applied neat, or more often it can be diluted, usually in a buffered solution which contains a small amount (0.1-5.0% by weight) of protein, such as BSA, NGS, or BLOTTO. After incubating for a sufficient length of time to allow specific binding to occur, the well is washed to remove unbound protein and then incubated with an optimal concentration of an appropriate anti-immunoglobulin antibody that is conjugated to an enzyme or other label by standard procedures and is dissolved in blocking buffer. The label can be chosen from a variety of enzymes, including horseradish peroxidase (HRP), beta-galactosidase, alkaline phosphatase (ALP), and glucose oxidase. Sufficient time is allowed for specific binding to occur again, then the well is washed again to remove unbound conjugate, and a suitable substrate for the enzyme is added. Color is allowed to develop and the optical density of the contents of the well is determined visually or instrumentally (measured at an appropriate wave length). Conditions for performing ELISA assays are well-known in the art.

In another embodiment of an ELISA, a peptide or a collection or set of peptides disclosed herein is immobilized on a surface, such as a ninety-six-well ELISA plate. A sample is then added and the assay proceeds as above.

In still other embodiments, a peptide or collection or set of peptides disclosed herein are electro- or dot-blotted onto nitrocellulose paper. Subsequently, a sample, such as a biological fluid (*e.g.*, serum or plasma) is incubated with the blotted antigen, and antibody in

the biological fluid is allowed to bind to the antigen(s). The bound antibody can then be detected, *e.g.*, by standard immunoenzymatic methods or by visualization using metallic nanoparticles or nanoshells coupled to secondary antibodies or other antibody binding agents or combinations thereof.

5 It should be understood by one of skill in the art that any number of conventional protein assay formats, particularly immunoassay formats, may be designed to utilize the isolated peptides, and collections and sets of peptides disclosed herein, for the detection of SARS-CoV-2 and/or other human coronavirus antibodies in a subject. Thus, the use of the peptides is thus not limited by the selection of the particular assay format, and is believed to encompass assay
10 formats that are known to those of skill in the art. To date most serology has been performed using singleplex ELISA, complement fixation or neutralization assays. More recently, Luminex-based systems have been employed that can address up to 100 antigenic targets simultaneously (*i.e.*, 100 individual pathogens, 100 individual antigenic targets for one pathogen, or some variation thereof). Additionally, arrays are established that comprise spotted
15 recombinant proteins expressed *in vitro* in *E. coli*, *S. cerevisiae*, baculoviruses, or cell-free, coupled transcription-translation.

One goal of the present disclosure is to automate the process of SARS-CoV-2 antibody detection and make it inexpensive, quick and accurate as well as detect exposure *per se* rather than to rigorously characterize humoral responses to specific pathogens.

20 One assay that meets these requirements is a programmable peptide array.

The peptide array capacity can be exploited to print multiple arrays per glass slide (configurations of 1, 3, 8, or 12 arrays can be printed).

Thus, one embodiment is a peptide microarray comprising peptides that are reactive with, and specific for SARS-CoV-2 antibodies. In some embodiments the peptide microarray
25 comprises: a peptide or peptides chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group
30 consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); or combinations thereof.

A further embodiment is a peptide microarray comprising peptide or peptides which are reactive with, and specific for SARS-CoV-2 antibodies and/or other human coronaviruses comprising: a peptide or peptides chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations thereof.

In some embodiments the peptide array comprises a set or collection of peptides, comprising amino acid sequences shifted one residue across all of the peptides comprising or consisting of the amino acid sequences of SEQ ID NOs: 1-29 and/or SEQ ID NOs: 30-45 and/or SEQ ID NOs: 46-208 and/or SEQ ID NOs: 209-261.

20

Methods and Systems for Serological Detection of Exposure to SARS-CoV-2

The present disclosure also includes methods and systems for the detection of exposure to antigens of SARS-CoV-2, *i.e.*, antibodies to SARS-CoV-2, in any sample utilizing the various peptides, isolated and non-isolated, and peptide microarrays disclosed herein.

Suitable methods typically include: receiving or obtaining (*e.g.*, from a patient) a sample of bodily fluid or tissue likely to contain antibodies; contacting (*e.g.*, incubating or reacting) a sample to be assayed with a peptide or peptides of the invention, under conditions effective for the formation of a specific peptide-antibody complex (*e.g.*, for specific binding of the peptide to the antibody); and assaying the contacted (reacted) sample for the presence of an antibody-peptide reaction (*e.g.*, determining the amount of an antibody-peptide complex). The presence of an elevated amount of the antibody-peptide complex indicates that the subject was exposed to and infected by SARS-CoV-2. A peptide, including a modified form thereof, which "binds specifically" to an antibody against a SARS-CoV-2 antigen interacts with the antibody,

30

or forms or undergoes a physical association with it, in an amount and for a sufficient time to allow detection of the antibody.

Conditions for reacting peptides and antibodies so that they react specifically are well-known to those of skill in the art. See, *e.g.*, Current Protocols in Immunology (Coligan et al.,
5 editors, John Wiley & Sons, Inc).

Once the disclosed peptide or peptides and sample antibody are permitted to react in a suitable medium, an assay is performed to determine the presence or absence of an antibody-peptide reaction. Any of the assays discussed herein can be used.

The methods and systems of the present disclosure may be used to detect exposure to
10 antigens of SARS-CoV-2 in research and clinical settings.

One sample for use in the methods is a biological sample. A biological sample may be obtained from a tissue of a subject or bodily fluid from a subject including but not limited to nasopharyngeal aspirate, blood, cerebrospinal fluid, saliva, serum, plasma, urine, sputum, bronchial lavage, pericardial fluid, or peritoneal fluid, or a solid such as feces. The subject
15 may be any animal, particularly a vertebrate and more particularly a mammal, including, without limitation, a cow, dog, human, monkey, mouse, pig, or rat. In one embodiment, the subject is a human.

A sample may also be a research, clinical, or environmental sample, such as cells, cell culture, cell culture medium, and compositions for use as, or the development of
20 pharmaceutical and therapeutic agents.

Additional applications include, without limitation, detection of the screening of blood products (*e.g.*, screening blood products for infectious agents), biodefense, food safety, environmental contamination, forensics, and genetic-comparability studies. The present disclosure also provides methods and systems for detecting viral antibodies in cells, cell
25 culture, cell culture medium and other compositions used for the development of pharmaceutical and therapeutic and immunomodulatory agents.

The subject may have been exposed to antigens of SARS-CoV-2, suspected of having exposure to antigens of SARS-CoV-2 or believed not to have had exposure to antigens of SARS-CoV-2.

30 In one embodiment, the subject is being tested for use of their plasma for treatment of COVID-19 patients.

In one embodiment, the subject may be a test subject, which has been administered a SARS-CoV-2 vaccine or immunomodulatory agent.

The systems and methods described herein support the detection and measure of a humoral immune response to SARS-CoV-2.

One embodiment provides a system for the detection of exposure to antigens of SARS-CoV-2, *i.e.*, antibodies to SARS-CoV-2, in any sample. The system includes at least one subsystem, wherein the subsystem includes a peptide or peptides or a collection or set of peptides disclosed herein, which are reactive with, and specific for SARS-CoV-2 antibodies, comprising: a peptide or peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); or combinations thereof. The system can also include additional subsystems for the purpose of: preparation of the sample; binding of any SARS-CoV-2 antibody in the sample with the peptide(s); washing the unbound sample; and visualization and/or quantification of bound antibody or antibodies.

A further embodiment provides a system for the detection of exposure to antigens of SARS-CoV-2, *i.e.*, antibodies to SARS-CoV-2, in any sample. The system includes at least one subsystem, wherein the subsystem includes a peptide microarray comprising a peptide or peptides or a collection or set of peptides disclosed herein, which are reactive with, and specific for SARS-CoV-2 antibodies comprising: a peptide or peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); or combinations thereof. The system can also include additional subsystems for the purpose of: preparation of the sample; binding

of any SARS-CoV-2 antibody in the sample with the peptides(s); washing the unbound sample; and visualization and/or quantification of bound antibody or antibodies.

Another embodiment provides a system for the differential detection of exposure to antigens of SARS-CoV-2, *i.e.*, antibodies to SARS-CoV-2 and other human coronaviruses, in any sample. The system includes at least one subsystem, wherein the subsystem includes a peptide or peptides or a collection or set of peptides disclosed herein, which are reactive with, and specific for SARS-CoV-2 and other human coronavirus antibodies, comprising: a peptide or peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a peptide or peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations thereof. The system can also include additional subsystems for the purpose of: preparation of the sample; binding of any antibody in the sample with the peptides(s); washing the unbound sample; and visualization and/or quantification of bound antibody or antibodies.

A further embodiment provides a system for the differential detection of exposure to antigens of SARS-CoV-2, *i.e.*, antibodies to SARS-CoV-2 and other human coronaviruses, in any sample. The system includes at least one subsystem, wherein the subsystem includes a peptide microarray comprising a peptide or peptides or a collection or set of peptides disclosed herein, which are reactive with, and specific for SARS-CoV-2 and other human coronavirus antibodies comprising: peptide or peptides or a collection or set of peptides disclosed herein, which are reactive with, and specific for SARS-CoV-2 and other human coronavirus antibodies, comprising: a peptide or peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208);

and/or a peptide or peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide
5 chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations thereof.
10 The system can also include additional subsystems for the purpose of: preparation of the sample; binding of any antibody in the sample with the peptides(s); washing the unbound sample; and visualization and/or quantification of bound antibody or antibodies.

The present disclosure provides a method for detecting the exposure to antigens of SARS-CoV-2, *i.e.*, antibodies to SARS-CoV-2, in any sample, including the steps of:
15 contacting the sample with a peptide or peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen
20 from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); or combinations thereof, under
25 conditions sufficient for any antibodies to SARS-CoV-2 in the sample and the peptides to bind; and visualizing and/or quantifying any bound antibody or antibodies to the peptides. The method may optionally include a step for washing any unbound sample.

The present disclosure provides a method for differentially detecting the exposure to antigens of SARS-CoV-2 and other human coronaviruses, *i.e.*, antibodies to SARS-CoV-2 and
30 other human coronaviruses, in any sample, including the steps of: contacting the sample with a peptide or peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a peptide or peptide

chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations thereof, under conditions sufficient for any antibodies to SARS-CoV-2 and/or other human coronaviruses in the sample and the peptides to bind; and visualizing and/or quantifying any bound antibody or antibodies to the peptides. The method may optionally include a step for washing any unbound sample.

The present disclosure further provides a method for detecting the exposure to antigens of SARS-CoV-2, *i.e.*, antibodies to SARS-CoV-2, in any sample, including the steps of: contacting the sample with a peptide microarray comprising a peptide or peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); or combinations thereof, under conditions sufficient for any antibodies to SARS-CoV-2 in the sample and the peptides to bind; and visualizing and/or quantifying any bound antibody or antibodies to the peptides. The method may optionally include a step for washing any unbound sample.

The present disclosure further provides a method for differentially detecting the exposure to antigens of SARS-CoV-2 and other human coronaviruses, *i.e.*, antibodies to SARS-CoV-2 and other human coronaviruses, in any sample, including the steps of: contacting the sample with a peptide microarray comprising a peptide or peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ

ID NOs: 46-208); and/or a peptide or peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across
5 at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations
10 thereof, under conditions sufficient for any antibodies to SARS-CoV-2 and/or other human coronaviruses in the sample and the peptides to bind; and visualizing and/or quantifying any bound antibody or antibodies to the peptides. The method may optionally include a step for washing any unbound sample.

Any method of detection discussed herein or known in the art can be used for
15 visualizing and/or quantifying the bound antibodies.

Kits

The present disclosure also includes reagents and kits for practicing the methods of the invention. These reagents and kits may vary.

20 One reagent of the kit would be a peptide or peptides or a collection or set of peptides disclosed herein, which are reactive with, and specific for SARS-CoV-2 and/or other human coronavirus antibodies, comprising: a peptide or peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide
25 or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a peptide or peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least
30 one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from

the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations thereof.

In certain embodiments, the peptides are attached to or immobilized on a solid support. In some embodiments, the peptides are attached to or immobilized on a solid support through a metallic nanolayer (*e.g.*, cadmium, zinc, mercury, gold, silver, copper, or platinum nanolayer). In certain embodiments, the solid support is a bead (*e.g.*, a colloidal particle or a metallic nanoparticle or nanoshell), a flow path in a lateral flow immunoassay device, a flow path in an analytical or centrifugal rotor, a tube or a well (*e.g.*, in a plate), or a sensor (*e.g.*, an electrochemical, optical, or opto-electronic sensor).

Reagents for particular types of assays can also be provided in kits disclosed herein. Thus, the kits can include a population of beads (*e.g.*, suitable for an agglutination assay or a lateral flow assay), or a plate (*e.g.*, a plate suitable for an ELISA assay). In other embodiments, the kits comprise a device, such as a lateral flow immunoassay device, an analytical or centrifugal rotor, a Western blot, a dot blot, a slot blot, or an electrochemical, optical, or opto-electronic sensor.

In one particular embodiment, the kit would comprise peptide microarrays comprising a peptide or peptides or a collection or set of peptides, which are reactive with, and specific for SARS-CoV-2 and/or other human coronavirus antibodies, comprising: a peptide or peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a peptide or peptides chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a peptide or peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 2 (SEQ ID NOs: 1-29); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 3 (SEQ ID NOs: 30-45); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 4 (SEQ ID NOs: 46-208); and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of the peptides listed in Table 5 (SEQ ID NOs: 209-261); or combinations thereof.

In addition, the kits can include various diluents and buffers, labeled conjugates or other agents for the detection of specifically bound antigens or antibodies (*e.g.* labeling reagents), and other signal-generating reagents, such as enzyme substrates, cofactors and chromogens. In

some embodiments, the kit comprises an anti-human IgG/IgM antibody conjugated to a detectable label (*e.g.*, a metallic nanoparticle, metallic nanoshell, metallic nanolayer, fluorophore, colored latex particle, or enzyme) as a labeling reagent. In other embodiments, the kit comprises protein A, protein G, protein A/G fusion proteins, protein L, or combinations thereof conjugated to a detectable label (*e.g.*, a metallic nanoparticle, metallic nanoshell, metallic nanolayer, fluorophore, colored latex particle, or enzyme) as a labeling reagent. In still other embodiments, the labeling reagents of the kit are a second collection or set of peptides of the invention conjugated to a detectable label (*e.g.*, a metallic nanoparticle, metallic nanoshell, metallic nanolayer, fluorophore, colored latex particle, or enzyme). The second collection or set of peptides can be the same as or different than the collection or set of peptides, which may optionally be attached to or immobilized upon a solid support.

Other components of a kit can easily be determined by one of skill in the art. Such components may include coating reagents, polyclonal or monoclonal capture antibodies specific for a peptide of the invention, or a cocktail of two or more of the antibodies, purified or semi-purified extracts of these antigens as standards, monoclonal antibody detector antibodies, an anti-mouse, anti-dog, anti-cat, anti-chicken, or anti-human antibody conjugated to a detectable label, indicator charts for colorimetric comparisons, disposable gloves, decontamination instructions, applicator sticks or containers, a sample preparatory cup and buffers or other reagents appropriate for constituting a reaction medium allowing the formation of a peptide-antibody complex.

Such kits provide a convenient, efficient way for a clinical laboratory to diagnose infection by corona virus. Thus, in certain embodiments, the kits further comprise instructions.

EXAMPLES

The present invention may be better understood by reference to the following non-limiting examples, which are presented in order to more fully illustrate the preferred embodiments of the invention. They should in no way be construed to limit the broad scope of the invention.

Example 1 – Materials and Methods

Generation of Human Peptide Array- HCoV SeroChip

The platform for epitope discovery was a programmable peptide microarray that can accommodate up to 3 million distinct linear peptides on a 75mm x 26mm slide (Nimblegen-Roche). The array can also be divided into 12 subarrays, each containing approximately

173,000 “12-mer peptides”. The 12-mer format was based on the observation that serum antibodies bind linear peptide sequences ranging from 5 to 9 amino acid (aa) and bind most efficiently when targets are flanked by additional amino acids (Buus *et al.* 2012). To enable differential detection of antibodies specific for SARS-CoV-2 infections, a custom NCBI
5 GenBank and VIPR database for full proteomes of seven human coronaviruses: SARS-CoV-2; SARS; MERS; NL-63; OC-43; 229E; and HKU1, was created (Table 1). Two bat coronavirus proteomes similar to SARS-CoV-2 were also included (Zhou *et al.* 2020). 1000 randomly selected 12 amino acid long scrambled peptides were added for background correction and nonspecific binding of peptides.

10 For each virus selected, all available protein sequences available before January 2020 from the NCBI and Virus Pathogen Database and Analysis Resource (VIPR) protein databases were downloaded and then a peptide database created comprising overlapping 12-mer peptides that tiled the whole proteome of each of these agents with 11 amino acid (aa) overlap in a sliding window pattern (Tokarz *et al.* 2020; Tokarz *et al.* 2018; Mishra *et al.* 2019; Mishra *et al.* 2018). The selected peptide sequences were passed through a redundancy filter to yield
15 283,630 unique peptide sequences and 24,779,921 non-unique (present in more than one virus) for a total of 172,665 peptides. Redundant peptides were excluded prior to synthesis. The individual peptides in the library were printed in random positions on the peptide array to minimize the impact of locational bias.

20

Table 1- Design and Characteristics of HCoV-peptide Array

	Virus database	No. of protein sequences	Total redundant "12-aa" peptides	Total unique "12-aa" peptides
1	<i>SARS-CoV-2-MN908947-CDS</i>	10	9591	9591
2	<i>Human-229E-CoV-PDB</i>	662	576999	15756
3	<i>Human-HKU1-CoV-PDB</i>	721	554447	16390
4	<i>Human-NC63-CoV-PDB</i>	1320	1000287	16392
5	<i>Human-OC43-CoV-PDB</i>	2667	2679272	29635
6	<i>MERS-CoV-PDB</i>	7253	8604059	54188
7	<i>SARS-CoV-PDB</i>	1996	1484140	29791
8	<i>Human-CoV-variants-VIPR-PDB</i>	12131	9880717	101145
9	<i>Bat-CoV-ZC45</i>	11	9678	9678
10	<i>Bat-CoV-ZXC21</i>	11	9655	9655
11	<i>Redundant-non-specific-peptides</i>	NA	NA	1000
Total number of unique and redundant peptides			24779921	283630
Total peptides in HCOV-SeroChip			172665	

Samples and experimental design

The study was approved by the Medical Ethical Committee of Sun Yat-Sen University (approval number 2020-060). An informed and written consent was obtained by patients.

A total 132 plasma samples were tested and analyzed using HCoV peptide arrays. Samples were divided into six groups: group 1] COVID-19 patients with non-severe (mild) disease (n=22); group 2] COVID-19 patients with severe disease (n=22); group 3] patients with SARS-CoV-2 infections but no-symptoms (asymptomatic COVID-19) (n=6); group 4] SARS-2003 IgG positive cases (n=11); group 5] other banked HCoV IgG positive controls (n=11); and group 6] healthy controls (n=10). The average age was 44.0±16.73 years for the mild COVID-19 group, 60.1±12.37 years for the severe COVID-19 group, and 43.5±15.08 years for the asymptomatic COVID-19 group. The asymptomatic group subjects were family members or close contacts of the mild or severely ill patients. The average age for SARS-CoV-1 IgG

positive control group was 24.4 ± 5.0 years. The average age for healthy control group was 46.3 ± 7.3 years. Other HCoV controls included banked samples for which age and sex information was not available.

Plasma samples were collected at two different time points, a minimum of two weeks
5 apart, from group COVID-19 patients (groups 1, 2 and 3). The first time point (early) was collected at 12.9 ± 5.9 post onset of disease (POD) for the mild disease group, and at 9.6 ± 3.5 days POD for the severe disease group. The second time point (late) was collected at 34.7 ± 8.3 days POD for the mild disease group, and at 24.8 ± 6.8 days POD for the severe disease group. For asymptomatic group, the first time point was collected on the day of hospitalization and
10 the second time point was collected at 14.5 ± 4.6 days after the day of hospitalization. Non COVID-19 samples from control groups (group 4, group 5 and group 6), were from adults without any evidence or history of infection with SARS-CoV-2.

All COVID-19 patients were tested for SARS-CoV-2 RNA in respiratory specimens using the China FDA approved Novel Coronavirus (2019-nCoV) Real Time RT-PCR kit from
15 LifeRiver Ltd. (Catalog #: RR-0479-02) real-time RT-PCR (CDC 2003). The diagnosis of COVID-19 pneumonia, and severity criteria were assessed at Guangdong CDC based on the New Coronavirus Pneumonia Prevention and Control Program (6th edition) published by the National Health Commission of China (Wu et al. 2020).

HCOV peptide array synthesis, sample binding and processing

20 Peptide synthesis was accomplished by light-directed array synthesis in a Nimble therapeutics Maskless Array Synthesizer (MAS) using an amino-functionalized substrate coupled with 6-amino hexanoic acid as a spacer and amino acid derivatives carrying a photosensitive 2-(2-Nitrophenyl) propyl-oxy-carbonyl-group (NPPOC) protection group (Orgentis Chemicals). Amino acids (final concentration 20 mM) were pre-mixed for 10
25 minutes in N,N-Dimethylformamide (DMF, Sigma Aldrich) with N,N,N',N'-Tetramethyl-O-(1H-benzotriazol-1-yl) uroniumhexafluorophosphate (HBTU, Protein Technologies, Inc.; final concentration 20 mM) as an activator, 6-Chloro-1-hydroxybenzotriazole (6-Cl-HOBt, Protein Technologies, Inc.; final concentration 20 mM) to suppress racemization, and N,N-Diisopropylethylamine (DIPEA, Sigma Aldrich; final concentration 31 mM) as a base.
30 Activated amino acids were then coupled to the array surface for 3 minutes. Following each coupling step, the microarray was washed with N-methyl-2-pyrrolidone (VWR International), and site-specific cleavage of the NPPOC protection group was accomplished by irradiation of an image created by a Digital Micro- Mirror Device (Texas Instruments), projecting 365 nm

wavelength light. Coupling cycles were repeated to synthesize the full in silico-generated peptide library.

Eleven arrays were used to test 132 plasma samples. Before loading, plasma samples were heat inactivated at 56°C for 30 minutes. Plasma samples were diluted (1:50) with binding
5 buffer (0.1M Tris-Cl, 1% alkali soluble casein, 0.05% Tween-20, and water). The peptide arrays were incubated overnight at 4°C on a flat surface with individual sample/sub-array. Overnight sample incubation was followed by three 10-minute washes with 1X TBS-T (0.05% Tween-20) at room temperature (RT). Secondary antibodies IgG (cat no. 109-605-098, Alexa
10 Fluor 647-AffiniPure Goat Anti-Human IgG, Fcγ fragment specific, Jackson ImmunoResearch Labs) and IgM (cat no. 109-165-129, Cy™3 AffiniPure Goat Anti-Human IgM, Jackson ImmunoResearch Labs) were diluted in 1X PBS at a concentration of 0.1μg/ml, and arrays were incubated in Plastic Coplin Jar (cat no. S90130, Fisher Scientific) for 3 hours at RT with gentle shaking. Secondary antibody incubation was also followed by three 10-minute washes with 1X TBST at RT. After a final wash, the arrays were dried and scanned on a microarray
15 scanner at 2-μm resolution, with an excitation wavelength of 635 nm (IgG) and 532 nm (IgM). The images were analyzed using the PepArray analysis program. The fluorescent signals were converted into arbitrary unit (AU) intensity plots ranging minimum to maximum intensity 0-65,000 AU.

Quality and reproducibility of the array data

20 In the array synthesis process, Nimble Therapeutics uses a quality control step that builds thousands of peptides with known binding epitopes to streptavidin, which have been confirmed through SPR and crystallization method (Lyamichev et al. 2017). Each subarray on the entire synthesized array slide contained these quality control peptides in addition to the customized targeted experimental peptides. Additionally, every array slide was QC analyzed
25 and the signal AU from these control peptides for each slide was correlated to the banked data from previous QC analyses. Arrays that did not meet standard cutoff thresholds were deemed as failed and removed from the further experimental process. Random nonadjacent tiling of overlapping peptides also enhanced confidence in the probe quality and supported the reactivity of an individual epitope by having multiple, overlapping peptides per epitope. In our previous
30 studies, epitope reactivity of 12 aa overlapping peptides with an 11 amino acid overlap (single aa tiling) has been shown.

The reproducibility of data and inter-array reproducibility analysis of peptide arrays have been reported in previous studies by testing technical replicates on two separate

microarrays. The two technical replicates for the target epitopes showed almost identical results with distinct epitopes (Tokarz et al.2018; Mishra et al. 2018; Forsstrom et al. 2014).

All analysis similar to IgG was also performed to generate data from IgM reactive peptides. For IgM analysis, >10,000 AU filtration step reduced the initial number of peptides from 172,665 to 24,728 for further analysis. A total of 10,816 peptides (7144 from the COVID-19 group and 3672 from the control group) peptides yielded group-specific differences ($p < 0.05$) in signal intensity. MDS efforts did not separate samples collected from patients with COVID-19 and controls (Fig. 1). However, the presence of three continuous peptides that were reactive in samples from SARS-CoV-2-infected subjects (irrespective of disease status) but not in control groups, allowed the identification of 16 SARS-CoV-2-specific IgM epitopes (Table 3).

Statistics and reproducibility

Reactivity values for all 132 samples were pooled together and peptides showing >10,000 AU for any samples were retained for further analysis. Regression analysis, fold-changes, and standard errors were estimated by fitting a linear model for signal intensities generated by each peptide, applying empirical Bayesian smoothing to the standard errors, and then determining those peptides that yielded statistically significant signal by contrasting linear models for each peptide between SARS-CoV-2 and Control samples at a significance value of <0.0520 . edgeR package in R was used for this purpose. It implements quantile-adjusted conditional maximum-likelihood method for estimating dispersions followed by fitting negative binomial generalized linear models. This was followed by a quasi-likelihood F-test to determine those peptides that yielded statistically significant signal by contrasting linear models for each peptide between SARS-CoV-2 and Control samples. MDS plots (Fig. 2) shows the discriminative ability of these peptides. These peptides were then used to reassemble longer epitope sequences (Table 2, Table 3). The normalization, background correction was performed using preprocessCore R package (Bolstad et al. 2003) and statistical comparison of peptide microarray intensities between groups were performed using the edgeR package (Robinson et al. 2010).

The analysis was performed to differentiate peptides that were immunoreactive with COVID-19 patients (groups 1, 2, and 3) and versus control groups (groups 4, 5, and 6) samples. MDS plots were generated using signal data for these significant peptides. The code for reassembly and plots was prepared using Rstudio v 1.2.5019. The plots were generated using ggplot2 package. A custom color-blind friendly color pallette was used to make the plots.

Alignment of reactive epitopes on SARS-CoV-2 proteome was performed using Geneious version 10.0.9.

Example 2 - Results

5 As described in Example 1, plasma samples were heat-inactivated at 56°C for 30 minutes, diluted (1:50), added to HCoV peptide arrays, incubated with Alexa Fluor 647 labeled goat anti-human IgG, CyTM3 labeled goat anti-human IgM antibodies, and scanned on a microarray scanner.

10 Fluorescence signal data for all the peptides from IgG and IgM scanned images of all HCoV peptide arrays was converted to arbitrary units (AU), pooled, background corrected, and normalized to avoid any inter-experimental variations (Tokarz et al. 2018; Mishra et al. 2018; Buus et al. 2012). A peptide signal was considered reactive if the intensity reading (AU) was above the threshold (mean $\pm$ 2 SD readings of random peptides, >10,000 AU for IgG and IgM analysis). A cutoff threshold for peptide recognition was defined as mean $\pm$ 2 times the standard deviation of the mean intensity value of all negative controls (Valentini et al. 2017). The regression analysis was performed to measure fold-changes on normalized and background corrected data for filtered peptides and followed by multidimensional scaling (MDS) analysis.

For IgG analysis, the >10,000 AU filtration step reduced the initial number of peptides from 172,665 to 79,714 for further analysis. A total of 37,237 peptides (18,533 from the COVID-19 group and 18,704 from the control group) were identified by regression analysis. This yielded group specific differences ($p < 0.05$) in signal intensity. MDS analysis was performed for IgG and IgM antibodies to differentiate peptides that were immunoreactive with COVID-19 patients (Groups 1–3) versus control groups (Group 4–6). MDS analysis of these peptides confirmed separation of patients with COVID-19 (Groups 1–3) and controls (Groups 20 4–6) into separate clusters with minimal overlap (Fig. 2). Out of 18,533 reactive 12-mer peptides associated with the COVID-19 group, 981 peptides were specific for the SARS-CoV-2 polyprotein.

The 981 SARS-CoV-2 peptides with IgG signal intensity >10,000 AU, were used to assemble a heatmap. The signal data was pooled from all the arrays and then reactivity compared across different sample groups. The pooled data were normalized; background corrected, and filtered peptides with immunoreactivity above the 10,000AU to generate the heatmap shown in Fig. 3. Immunoreactive peptides included 566 from ORF1ab, three from ORF10, 243 from surface glycoprotein (S protein), 20 from ORF3a, 20 from membrane glycoprotein (mGP), four from ORF7a, 21 from ORF8, and 104 from nucleocapsid 30

phosphoprotein (N). Peptides from “S” and “N” proteins had higher reactive intensity (higher AU) and rate of reactivity in comparison to peptides from other proteins (Fig. 3). Immunoreactivity was higher in COVID-19 subjects with severe versus mild disease, and in asymptomatic SARS-CoV-2-infected subjects versus subjects with mild disease. Samples from second time point collections were more reactive than first time point collections in subjects with severe, mild and asymptomatic SARSCoV-2 infection (Fig. 3).

The presence of a minimum of three continuous reactive peptides in samples with SARS-CoV-2 infection but non-reactive in control groups, were used to identify SARS-CoV-2 reactive IgG epitopes. Analysis of these 981 peptide sequences led to the identification of 163 epitopes (Table 4). The 29 epitopes with the strongest and most specific reactivity with SARS-CoV-2 are shown in Table 2. Table 2 also indicates the location of each epitope on the SARS-CoV-2 proteome, its length, aa sequence, and the percentages of plasma samples that were immunoreactive in Groups 1–6.

These 29 epitopes included 11 epitopes (37.9%) in S protein (SP1-SP11), 8 (27.5%) epitopes in N protein (NP1-NP8), 6 (20.7%) epitopes in ORF1ab polyprotein (OP1-OP6), 2 (6.9%) in mGP protein (MP1 and MP2), one (3.4%) each from ORF3, and ORF8 proteins. Immunoreactivity was higher in second time point plasma samples (Fig. 4) and in patients with more severe disease. In samples from patients with severe disease, 7–22 epitopes (mean of 13) were reactive in second time point samples (24.8 ± 6.8 days POD) versus 0–15 (mean of 7) epitopes in first time point samples (9.6 ± 3.5 days POD). In patients with mild disease, 3–22 epitopes (mean of eight) were reactive in second time point samples (34.7 ± 8.3 POD) versus 0–12 epitopes (mean of 4) in first time point samples (12.9 ± 5.9 POD). In asymptomatic subjects 1–9 epitopes (mean of four) were reactive at either time point (day of hospitalization and 14.5 ± 4.6 days after the hospitalization). Plasma samples from 19 of 22 patients (86%) with mild disease were reactive with at least 1 of 29 epitopes at the first time point. All 22 (100%) were reactive with at least 3 of 29 epitopes at the second time point. Plasma from 21 of 22 patients (95%) with severe disease were reactive with at least one epitope at first time point. All 22 (100%) were reactive with at least six epitopes at the second time point. All six (100%) asymptomatic cases were reactive with at least 2 of 29 epitopes in first and 3 of 29 in second timepoint collections.

Plasma from none of the asymptomatic cases was reactive with any ORF1ab, ORF3, ORF8, or mGP epitope (Table 4). Plasma from asymptomatic cases were reactive only with S and N epitopes. Plasma collected at both time points from asymptomatic case AS1 was reactive with five epitopes in the S protein (SP1, SP2, SP4, SP5, and SP10) and three epitopes in the N

protein (NP2, NP7, and NP8). Case AS1 plasma was reactive with SP9 at only first time point. Plasma from asymptomatic case AS2 was reactive with three epitopes in the S protein (SP7, SP8, and SP10) and five epitopes in the N protein (NP2, NP3, NP4, NP5, and NP8) at both time points. Plasma from asymptomatic cases AS1, AS2, and AS4 were reactive with SP10 and NP8 epitopes at both time points. Plasma from AS3 was reactive with SP10 at both time points and with NP8 only at the second time point. Plasma from AS5 and AS6 were immunoreactive with the NP1 epitope and the epitope NP6, respectively, at both time points. Plasma from mild case M7 was reactive only with the SP10 epitope at first time point but was reactive to 22/29 epitopes at second time point of collection. All mild and severe COVID-19 cases were reactive to a greater number of epitopes at second time points versus the first time point.

The 29 immunoreactive epitopes were mapped to the proteome of SARS-CoV-2 (acc. number MN908947). ORF1ab epitopes (OP1–OP6) were dispersed throughout the protein. Eleven linear epitopes from “S” protein (SP1–SP11) mapped outside of the RBD (Receptor Binding Domain). Four (SP1–SP4) epitopes were located in the SD1/SD2. Four (SP5–SP8) epitopes were located between SD1/SD2 and fusion peptide region. The SP9 epitope overlapped the fusion peptide. The SP10 epitope is located between CD (Connector Domain) region and HR2 (Heptad Repeat 2). The SP11 epitope was located at the beginning of the HR2 region of the S2 subunit of the surface glycoprotein. The SP1 and SP9 peptides identified in this study were recently reported as potentially linked to neutralization (Poh et al. 2020).

Ten of eleven spike epitopes (SP1–SP10) were located in regions of high immunoreactivity reported in a recent pre-print from Li et al. 2020. The SP10 epitope was the most reactive spike epitope in subjects with severe (69% first time point, 96% second timepoint), mild (41% first timepoint, 82% second timepoint), and asymptomatic SARS-CoV-2 (67% either timepoint) infections.

The NP2 and NP8 epitopes were the most reactive N protein epitopes. In severe disease, NP2 reactivity was found in 55% of subjects at the first timepoint, and 82% at the second timepoint. NP8 reactivity was found in 64% of subjects at first timepoint, and 100% at second timepoint. In mild disease, NP2 reactivity was found in 37% of subjects at the first timepoint, and 69% at the second timepoint. NP8 reactivity was found in 28% of subjects at the first timepoint, and 82% at the second timepoint. In asymptomatic cases, reactivity was 59% for either NP2 or NP8. In severe disease SP11 reactivity was found in 46% of subjects at the first time point, and 91% at the second time point. In mild disease SP11 reactivity was found in 28% of subjects at the first time point, and 60% at the second time point. No asymptomatic

cases were reactive with SP11. Epitopes for ORF1ab (OP1–OP6) showed reactivity in cases with mild and severe disease at both timepoints but not in asymptomatic infections. Only second timepoint samples from severe and mild diseases were reactive to epitopes for mGP, MP1, and MP2 (37 and 26% for severe disease and 28 and 19% for mild disease).

5 IgM analysis revealed one linear epitope in the membrane glycoprotein (MADSNGTITVEELKKLLEQWN) (SEQ ID NO: 42), that was reactive in 4/22 (19%) COVID-19 patients with mild disease and 8/22 (37%) patients with severe disease at late time point collection (Fig. 1, Table 3).

10 Through use of a limited number of plasma samples from subjects with known exposure to other HCoV, IgG epitopes were identified in HKU1 (n = 15), NL63 (n = 10), OC43 (n = 14), 229E (n = 5), and SARS-CoV-1 (n = 9) (Table 5). More than 90% of samples from patients exposed to SARS-CoV-2 and controls showed a wide range of reactivity to epitopes from seasonal coronaviruses (HKU1, NL63, OC43, and 229E).

15 In summary, the HCoV peptide array and plasma from 50 patients with asymptomatic, mild, or severe SARS-CoV-2 infection was used to identify immunoreactive IgG epitopes for SARSCoV-2. Immunoreactivity profiles differed with severity of illness and over the time course of infection. Two subjects with a history of SARS-CoV-1 infection had reactivity to two of 29 IgG SARSCoV-2 epitopes. Their plasma was collected in 2004 or 2005; thus, this presumably reflects cross-reactivity due to proteome homology (Lv et al. 2020; Chia et al. 2020).
20 Two healthy controls with immunoreactivity to SP11 (one of 29 epitopes of SARS-CoV-2) may have had an asymptomatic infection with either SARS-CoV-1 or SARS-CoV-2 (Xu et al. 2006; CDC 2003).

Table 2-IgG linear epitopes for detection of SARS-CoV-2 infection

S. No	Epitope ID	Epitope Sequence	Epitope length	AA (start)	AA (end)	Protein name (Accession: MN908947)	Severe-COV ID-19 Early (N=22)	Severe-COV ID-19 Late (N=22)	Mild-COV ID-19 Early (N=22)	Mild-COV ID-19 Late (N=22)	Asymptomatic COV ID-19 (N=12)	SARS-CoV-1 (N=11)	Healthy Controls (N=10)	Other-HCoV (N=11)
1	OP 1	TAYN GYLTS SSKTP (SEQ ID NO:1)	13	1,484	1,497	ORF1ab polyprotein (pp1ab)	5/22 (23%)	4/22 (19%)	5/22 (23%)	5/22 (23%)	0/12 (0%)	1/11 (10%)	0/10 (0%)	1/11 (10%)
2	OP 2	LVPNQ PYPNA SFDNF (SEQ ID NO:2)	14	1,914	1,928		8/22 (37%)	12/22 (55%)	3/22 (14%)	3/22 (14%)	0/12 (0%)	0/11 (0%)	0/10 (0%)	0/11 (0%)
3	OP 3	NRFTT TLNDF NLVAMK (SEQ ID NO:3)	15	3,484	3,499		1/22 (5%)	1/22 (5%)	4/22 (19%)	3/22 (14%)	0/12 (0%)	0/11 (0%)	0/10 (0%)	0/11 (0%)
4	OP 4	QGLLP PKNSI DAFKL NIK (SEQ ID NO:4)	17	3,826	3,843		5/22 (23%)	5/22 (23%)	4/22 (19%)	6/22 (28%)	0/12 (0%)	0/11 (0%)	0/10 (0%)	2/11 (19%)

5	OP 5	NTCVG SDNVT DFNAI ATCD W (SEQ ID NO:5)	19	5,419	5,438		5/22 (23 %)	13/22 (60%)	5/22 (23 %)	6/22 (28 %)	0/12 (0%)	0/11 (0%)	0/10 (0%)	0/11 (0%)
6	OP 6	FYSYA THSDK FTDGV (SEQ ID NO:6)	14	6,292	6,306		7/22 (32 %)	11/22 (50%)	0/22 (0%)	2/22 (10 %)	0/12 (0%)	0/11 (0%)	0/10 (0%)	0/11 (0%)
7	SP1	TESNK KFLPF QQFGR DIAD (SEQ ID NO:7)	18	553	571	Surface glycoprotein	4/22 (19 %)	15/22 (69%)	1/22 (5%)	9/22 (41 %)	3/12 (25 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)
8	SP2	TDAVR DPQTL EILDIT PCS (SEQ ID NO:8)	18	573	591		6/22 (28 %)	17/22 (78%)	1/22 (5%)	11/22 (50 %)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)
9	SP3	QVAV LYQD VNCTE VPVAI HADQ LTPTW (SEQ ID NO:9)	26	607	633		0/22 (0%)	9/22 (41%)	2/22 (10 %)	4/22 (19 %)	0/12 (0%)	0/11 (0%)	0/10 (0%)	0/11 (0%)
10	SP4	CASYQ TQTNS PRRAR SV	16	671	687		2/22 (10 %)	4/22 (19%)	0/22 (0%)	1/22 (5%)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)

		(SEQ ID NO:10)												
11	SP5	ARSVA SQSIH YTMSL GAENSV (SEQ ID NO:11)	22	684	706	2/22 (10 %)	6/22 (28%)	4/22 (19 %)	4/22 (19 %)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)	
12	SP6	RALTG IAVEQ DKNT QEVFA QV (SEQ ID NO:12)	20	765	785	11/22 (50 %)	14/22 (64%)	2/22 (10 %)	5/22 (23 %)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)	
13	SP7	FAQVK QIYKT PPIKDF GGFNF (SEQ ID NO:13)	20	782	802	5/22 (23 %)	8/22 (37%)	1/22 (5%)	3/22 (14 %)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)	
14	SP8	NFSQI LPDPS KPSKRS (SEQ ID NO:14)	15	801	816	1/22 (5%)	4/22 (19%)	0/22 (0%)	4/22 (19 %)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)	
15	SP9	KRSFIE DLLFN KVT (SEQ ID NO:15)	13	814	827	5/22 (23 %)	8/22 (37%)	4/22 (19 %)	5/22 (23 %)	1/12 (9%)	0/11 (0%)	0/10 (0%)	0/11 (0%)	

16	SP1 0	PELDS FKEEL DKYFK NHTSP DVD (SEQ ID NO:16)	22	1,143	1,165		15/22 (69%)	21/22 (96%)	9/22 (41%)	18/22 (82%)	8/12 (67%)	1/11 (10%)	0/10 (0%)	1/11 (10%)
17	SP1 1	SPDVD LGDIS GINAS VV (SEQ ID NO:17)	16	1,161	1,177		10/22 (46%)	20/22 (91%)	6/22 (28%)	13/22 (60%)	0/12 (0%)	1/11 (10%)	2/10 (20%)	1/11 (10%)
18	ORF3- 1	EHDY QIGGY TEKW ESGVK DCVVL (SEQ ID NO:18)	22	181	203	ORF3a protein	4/22 (19%)	7/22 (32%)	1/22 (5%)	2/22 (10%)	0/12 (0%)	0/11 (0%)	0/10 (0%)	0/11 (0%)
19	MP 1	CDIKD LPKEIT VATS (SEQ ID NO:19)	14	159	173	Membrane glycoprotein	0/22 (0%)	8/22 (37%)	0/22 (0%)	6/22 (28%)	0/12 (0%)	0/11 (0%)	0/10 (0%)	0/11 (0%)
20	MP 2	QRVA GDSGF AAYSR Y (SEQ ID NO:20)	14	185	199		0/22 (0%)	10/22 (46%)	0/22 (0%)	4/22 (19%)	0/12 (0%)	0/11 (0%)	0/10 (0%)	0/11 (0%)
21	ORF8- 1	GSKSPI QYIDI GNYT VSCLP F (SEQ	20	66	86	ORF8 protein	3/22 (14%)	12/22 (55%)	4/22 (19%)	3/22 (14%)	0/12 (0%)	0/11 (0%)	0/10 (0%)	1/11 (10%)

		ID NO:21)												
22	NP 1	ALLLL DRLNQ LESKM SG (SEQ ID NO:22)	16	220	236	Nucleocapsid phosphoprotein	3/22 (14 %)	5/22 (23%)	2/22 (10 %)	5/22 (23 %)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)
23	NP 2	NQLES KMSG KGQQ QQGQ TVTKK SA (SEQ ID NO:23)	23	228	251		12/22 2 (55 %)	18/22 (82%)	8/22 (37 %)	15/22 2 (69 %)	7/12 (59 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)
24	NP 3	QTVTK KSAAE ASKKP RQK (SEQ ID NO:24)	17	244	261		2/22 (10 %)	7/22 (32%)	1/22 (5%)	4/22 (19 %)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)
25	NP 4	RGPEQ TQGNF GDQEL I (SEQ ID NO:25)	15	277	292		6/22 (28 %)	3/22 (14%)	2/22 (10 %)	3/22 (14 %)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)
26	NP 5	AYKTF PPTEP KKDK KKK (SEQ ID NO:26)	16	359	375		2/22 (10 %)	5/22 (23%)	0/22 (0%)	2/22 (10 %)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)

27	NP 6	KDKK KKAD ETQAL PQR (SEQ ID NO:27)	15	370	385		3/22 (14 %)	7/22 (32%)	0/22 (0%)	5/22 (23 %)	3/12 (25 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)
28	NP 7	ADETQ ALPQR QKKQ QTVTL LPAAD L (SEQ ID NO:28)	24	376	400		4/22 (19 %)	17/22 (78%)	4/22 (19 %)	9/22 (41 %)	2/12 (17 %)	0/11 (0%)	0/10 (0%)	0/11 (0%)
29	NP 8	TLLPA ADLD DFSKQ LQQS MSSAD S (SEQ ID NO:29)	23	393	416		14/2 2 (64 %)	22/22 (100 %)	6/22 (28 %)	18/2 2 (82 %)	7/12 (59 %)	1/11 (10 %)	0/10 (0%)	0/11 (0%)

Table 3-IgM linear epitopes for detection of SARS-CoV-2 infection

		Asymptomatic	Healthy- Controls_China	Mild- COVID_Early	Mild- COVID_Late	Other_CoV
1	LGDELGTDPPYEDFQ	SEQ ID NO: 30 0/12 (0%)	0/10 (0%)	0/22 (0%)	0/22 (0%)	1/12 (9%)
2	LYIDINGNLHPDSAT	SEQ ID NO: 31 0/12 (0%)	0/10 (0%)	2/22 (10%)	0/22 (0%)	0/12 (0%)
3	ATKYLVOQESPFVMM	SEQ ID NO: 32 0/12 (0%)	0/10 (0%)	0/22 (0%)	0/22 (0%)	1/12 (9%)
4	IVFDGKSKCEESSA	SEQ ID NO: 33 0/12 (0%)	0/10 (0%)	1/22 (5%)	1/22 (5%)	0/12 (0%)
5	LNDFSNSGSDVLYQP	SEQ ID NO: 34 0/12 (0%)	1/10 (10%)	0/22 (0%)	0/22 (0%)	0/12 (0%)
6	LSFCAFADVAAKAY	SEQ ID NO: 35 0/12 (0%)	0/10 (0%)	0/22 (0%)	0/22 (0%)	0/12 (0%)
7	YDFGDFIQTTGSG	SEQ ID NO: 36 0/12 (0%)	1/10 (10%)	0/22 (0%)	0/22 (0%)	0/12 (0%)
8	DFIQTTGSGVPVV	SEQ ID NO: 37 0/12 (0%)	1/10 (10%)	0/22 (0%)	0/22 (0%)	0/12 (0%)
9	FLLKYNENGTTDAV	SEQ ID NO: 38 0/12 (0%)	0/10 (0%)	0/22 (0%)	0/22 (0%)	0/12 (0%)
10	GVLTESNKKFLPFQ	SEQ ID NO: 39 0/12 (0%)	0/10 (0%)	0/22 (0%)	0/22 (0%)	0/12 (0%)
11	YQTQTNSPRRARSA	SEQ ID NO: 40 0/12 (0%)	0/10 (0%)	0/22 (0%)	0/22 (0%)	0/12 (0%)
12	NTQEVFAQVKQIYK	SEQ ID NO: 41 0/12 (0%)	0/10 (0%)	0/22 (0%)	0/22 (0%)	0/12 (0%)
13	MADSNGTITVEELKKLLEQWN	SEQ ID NO: 42 1/12 (9%)	0/10 (0%)	0/22 (0%)	4/22 (19%)	0/12 (0%)

14	KEPCSSGTYEGNSPFHP	SEQ ID NO: 43	0/12 (0%)	0/10 (0%)	0/22 (0%)	1/22 (5%)	1/12 (9%)
15	TLLPAADLDDFSKQ	SEQ ID NO: 44	1/12 (9%)	0/10 (0%)	0/22 (0%)	0/22 (0%)	0/12 (0%)
16	AADLDDFSKQLQQS	SEQ ID NO: 45	0/12 (0%)	0/10 (0%)	0/22 (0%)	0/22 (0%)	0/12 (0%)

Table 3 (continued)

			SARS_2003	Severe- COVID_Early	Severe- COVID_Late
1	LGDELGTDPYEDFQ	SEQ ID NO: 30	0/10 (0%)	0/22 (0%)	0/22 (0%)
2	LYIDINGNLHPDSAT	SEQ ID NO: 31	0/10 (0%)	0/22 (0%)	2/22 (10%)
3	ATKYLQQESPFFVMM	SEQ ID NO: 32	0/10 (0%)	0/22 (0%)	0/22 (0%)
4	IVFDGKSKCESSA	SEQ ID NO: 33	0/10 (0%)	0/22 (0%)	0/22 (0%)
5	LNDFSNSGSDVLYQP	SEQ ID NO: 34	0/10 (0%)	0/22 (0%)	0/22 (0%)
6	LSFCFAFVDAKAY	SEQ ID NO: 35	0/10 (0%)	1/22 (5%)	0/22 (0%)
7	YDFGDFIQITTPGSG	SEQ ID NO: 36	0/10 (0%)	0/22 (0%)	0/22 (0%)
8	DFIQITTPGSGVPV	SEQ ID NO: 37	0/10 (0%)	0/22 (0%)	0/22 (0%)
9	FLLKYNENGTITDAV	SEQ ID NO: 38	0/10 (0%)	0/22 (0%)	1/22 (5%)
10	GVLTESNKKFLPFQ	SEQ ID NO: 39	1/10 (10%)	0/22 (0%)	0/22 (0%)
11	YQTQTNSPRRARSA	SEQ ID NO: 40	0/10 (0%)	0/22 (0%)	1/22 (5%)
12	NTQEVFAQVKQIYK	SEQ ID NO: 41	0/10 (0%)	1/22 (5%)	0/22 (0%)
13	MADSNGTITVEELKKLLEQWN	SEQ ID NO: 42	0/10 (0%)	1/22 (5%)	8/22 (37%)
14	KEPCSSGTYEGNSPFHP	SEQ ID NO: 43	0/10 (0%)	0/22 (0%)	0/22 (0%)
15	TLLPAADLDDFSKQ	SEQ ID NO: 44	0/10 (0%)	1/22 (5%)	1/22 (5%)
16	AADLDDFSKQLQQS	SEQ ID NO: 45	0/10 (0%)	1/22 (5%)	0/22 (0%)

Table 4 – Additional IgG linear epitopes for detection of SARS-CoV-2 infection. Sequences (aa) are based on proteome of Severe acute respiratory syndrome coronavirus 2 isolate Wuhan-Hu-1 (Accession no. MN908947)

Epitope sequence	Protein Info (MN908947_CDS_proteome)	Severe- COVID_ Early	Severe- COVID_ Late	Mild- COVID_ Early	Mild- COVID_ Late
YRKVLLRKNKGKGA	SEQ ID NO: 46 orf1ab polyprotein (pp1ab)	0/22 (0%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
LLRKNKGKAGGHSY	SEQ ID NO: 47 orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
GNKGAGGHSYGADLKSF DL	SEQ ID NO: 48 orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
DELGTDPYEDFQEN	SEQ ID NO: 49 orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
WNTKHSSGVIRELM	SEQ ID NO: 50 orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)
GVYCCREHEHEIAW	SEQ ID NO: 51 orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
HEIAWYTERSEKSY	SEQ ID NO: 52 orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
SYELQTPFEIKLAK	SEQ ID NO: 53 orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
AKKFDTFNGECPNF	SEQ ID NO: 54 orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
CDHCGETSWQTGDF	SEQ ID NO: 55 orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)
LTNIFGTVEKLPVLD	SEQ ID NO: 56 orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
GVEFLRDGWEIFVKF	SEQ ID NO: 57 orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	0/22 (0%)	0/22 (0%)

TGDLQPLEQPTSEAVEAPL V	SEQ ID NO: 58	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)
KGGAPTKVTFGDDTVIEV	SEQ ID NO: 59	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)
NITFELDERIDKVL	SEQ ID NO: 60	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	0/22 (0%)
EFACVVADAVIKTLQPV	SEQ ID NO: 61	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	1/22 (5%)
NKGEDIQLLKSAYENF	SEQ ID NO: 62	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	0/22 (0%)
YDKLVSSFLEMKSEK	SEQ ID NO: 63	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	0/22 (0%)
AFYILPSIISNEKQEI	SEQ ID NO: 64	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	0/22 (0%)
LGYVTHGLNLEEAA	SEQ ID NO: 65	orf1ab polyprotein (pp1ab)	2/22 (10%)	0/22 (0%)	0/22 (0%)
THGLNLEEAAARYMRS	SEQ ID NO: 66	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	1/22 (5%)
TAYNGYLTSSSKTP	SEQ ID NO: 67	orf1ab polyprotein (pp1ab)	5/22 (23%)	5/22 (23%)	5/22 (23%)
GEVITFDNLKTL	SEQ ID NO: 68	orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	1/22 (5%)
GRYMSALNHTKKWKYPQ V	SEQ ID NO: 69	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	0/22 (0%)
TVGELGDVRETMSYL	SEQ ID NO: 70	orf1ab polyprotein (pp1ab)	0/22 (0%)	1/22 (5%)	0/22 (0%)
MGTLSYEQFKKGVPQIPC	SEQ ID NO: 71	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	0/22 (0%)
FTCASEYTGNYQCGHY	SEQ ID NO: 72	orf1ab polyprotein (pp1ab)	2/22 (10%)	3/22 (14%)	3/22 (14%)
YQCGHYKHITSKET	SEQ ID NO: 73	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)

LVPNQYPNASFDFNF	SEQ ID NO: 74	orf1ab polyprotein (pp1ab)	8/22 (37%)	12/22 (55%)	3/22 (14%)	3/22 (14%)
FPDLNGDVVAIDYK	SEQ ID NO: 75	orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
NSFDVVKSEDAQGMNDNL	SEQ ID NO: 76	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
SEEVVENPTIQKDVLECN	SEQ ID NO: 77	orf1ab polyprotein (pp1ab)	0/22 (0%)	2/22 (10%)	0/22 (0%)	1/22 (5%)
GLAAVNSVPWDTIA	SEQ ID NO: 78	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	1/22 (5%)	0/22 (0%)
TIANYAKPFLNKKVVSTTT	SEQ ID NO: 79	orf1ab polyprotein (pp1ab)	1/22 (5%)	2/22 (10%)	1/22 (5%)	1/22 (5%)
FLNKVVSTTTNIVTRC	SEQ ID NO: 80	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
VKMFDA YVNTFSST	SEQ ID NO: 81	orf1ab polyprotein (pp1ab)	2/22 (10%)	3/22 (14%)	1/22 (5%)	0/22 (0%)
FSSTFNVPMEKLKT	SEQ ID NO: 82	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
TLVATAEAEELAKNVSL	SEQ ID NO: 83	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
SAAKKNNLPFKLTCA	SEQ ID NO: 84	orf1ab polyprotein (pp1ab)	0/22 (0%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
VHVMSKHTDFSSEIIG	SEQ ID NO: 85	orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
DFDFTWFSQRGGSYT	SEQ ID NO: 86	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
TFDSEYCRHGTCER	SEQ ID NO: 87	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
DVLYQPPQTTSITSAVL	SEQ ID NO: 88	orf1ab polyprotein (pp1ab)	2/22 (10%)	1/22 (5%)	1/22 (5%)	0/22 (0%)
NRFTTTLNDFNLVAMK	SEQ ID NO: 89	orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	4/22 (19%)	3/22 (14%)

LTQDHVDILGPLSA	SEQ ID NO: 90	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	1/22 (5%)
GMNGRTILGSALLED	SEQ ID NO: 91	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	1/22 (5%)
QGLLPKNSIDAFKLNK	SEQ ID NO: 92	orf1ab polyprotein (pp1ab)	5/22 (23%)	5/22 (23%)	4/22 (19%)	6/22 (28%)
KLKKSILNVAKSEFDRD	SEQ ID NO: 93	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
LRANSVAVKLQNNELS	SEQ ID NO: 94	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
AVKLQNNELSPVALR	SEQ ID NO: 95	orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
CAAGTTQACTDDN	SEQ ID NO: 96	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)
LALLSDLQDLKWARF	SEQ ID NO: 97	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
ATVRLQAGNATEVPA	SEQ ID NO: 98	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
VKMLCTHTGTGQAITV	SEQ ID NO: 99	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	2/22 (10%)	2/22 (10%)
KYVQIPTTCANDPV	SEQ ID NO: 100	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
RVCVSAARLTTPCGT	SEQ ID NO: 101	orf1ab polyprotein (pp1ab)	2/22 (10%)	2/22 (10%)	0/22 (0%)	0/22 (0%)
AVAKHDFEFKFRIDG	SEQ ID NO: 102	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
GDFIQITTPGSGVPV	SEQ ID NO: 103	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
QTYHPNCVNCCLDDR	SEQ ID NO: 104	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	1/22 (5%)	2/22 (10%)
ALTNNVAFQTVKPGNF	SEQ ID NO: 105	orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	0/22 (0%)	0/22 (0%)

GFFKEGSSVELKHF	SEQ ID NO: 106	orf1ab polyprotein (pp1ab)	0/22 (0%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
QLLFVVEVDKYFDCYD GGC	SEQ ID NO: 107	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)
SLYVKPGTSSGDATTAY A	SEQ ID NO: 108	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
NRDVDTDVNEFYAYL	SEQ ID NO: 109	orf1ab polyprotein (pp1ab)	0/22 (0%)	1/22 (5%)	2/22 (10%)	2/22 (10%)
SQGLVASIKNFKSV	SEQ ID NO: 110	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
QHTMLVKQGGDDYVY	SEQ ID NO: 111	orf1ab polyprotein (pp1ab)	0/22 (0%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
VDDIVKTDGTLME	SEQ ID NO: 112	orf1ab polyprotein (pp1ab)	0/22 (0%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
AIDAYPLTKHPNQE	SEQ ID NO: 113	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
GCDVTDVTQLYLG	SEQ ID NO: 114	orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
GLYKNTCVGSNDVT	SEQ ID NO: 115	orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
NTCVGSDNVTDFNAIATC DW	SEQ ID NO: 116	orf1ab polyprotein (pp1ab)	5/22 (23%)	13/22 (60%)	5/22 (23%)	6/22 (28%)
YQKVGMQKYSTLQG	SEQ ID NO: 117	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
STLQPPPGTGKSHF	SEQ ID NO: 118	orf1ab polyprotein (pp1ab)	2/22 (10%)	0/22 (0%)	1/22 (5%)	2/22 (10%)
ARARVECFDKFKVNS	SEQ ID NO: 119	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)
DPAQLPAPRTLLTK	SEQ ID NO: 120	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
PRTLLTKGTLEPEY	SEQ ID NO: 121	orf1ab polyprotein (pp1ab)	2/22 (10%)	2/22 (10%)	0/22 (0%)	0/22 (0%)

IFTQTETAHSCNV	SEQ ID NO: 122	orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	0/22 (0%)	1/22 (5%)
GCHATREAVGTNLP	SEQ ID NO: 123	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)
VQMLSDTLKNLSDR	SEQ ID NO: 124	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
FDYVYNPFMIDVQQWG	SEQ ID NO: 125	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
MIDVQQWGFTGNLQ	SEQ ID NO: 126	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
TGNLQSNHDLQCQVH	SEQ ID NO: 127	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
LADKFPVLHDIGNPKAIKC VP	SEQ ID NO: 128	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
FYSYATHSDKFTDGV	SEQ ID NO: 129	orf1ab polyprotein (pp1ab)	7/22 (32%)	11/22 (50%)	0/22 (0%)	2/22 (10%)
FDTRVLSNLPGCCD	SEQ ID NO: 130	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
QVVSDIDYVPLKSATC	SEQ ID NO: 131	orf1ab polyprotein (pp1ab)	3/22 (14%)	3/22 (14%)	1/22 (5%)	1/22 (5%)
PSVGPQKQASLNGVTL	SEQ ID NO: 132	orf1ab polyprotein (pp1ab)	1/22 (5%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
GVVQQLPETYFTQSRN	SEQ ID NO: 133	orf1ab polyprotein (pp1ab)	1/22 (5%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
AMDEFIERYKLEGY	SEQ ID NO: 134	orf1ab polyprotein (pp1ab)	3/22 (14%)	6/22 (28%)	0/22 (0%)	1/22 (5%)
LAKRFKESPFLED	SEQ ID NO: 135	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
FITDAQTGSSKCVCSVID	SEQ ID NO: 136	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
IKSQDLSVVSKVVKV	SEQ ID NO: 137	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)

YPKLQSSQAWQPGV	SEQ ID NO: 138	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	1/22 (5%)	0/22 (0%)
QRMILLEKCDLQNYG	SEQ ID NO: 139	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
HFGAGSDKGVAPGTA	SEQ ID NO: 140	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
ITEHSWNADLYKLM	SEQ ID NO: 141	orf1ab polyprotein (pp1ab)	0/22 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)
LSSYSLFDMSKFPLKL	SEQ ID NO: 142	orf1ab polyprotein (pp1ab)	2/22 (10%)	3/22 (14%)	0/22 (0%)	0/22 (0%)
SLLLCRMNSRNYIA	SEQ ID NO: 143	ORF10 protein	1/22 (5%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
QCVNLTTRTQLPPA	SEQ ID NO: 144	surface glycoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
TQLPPAYTNSFTRGVYYP D	SEQ ID NO: 145	surface glycoprotein	4/22 (19%)	5/22 (23%)	2/22 (10%)	2/22 (10%)
FHAIHVSNGTHKRF	SEQ ID NO: 146	surface glycoprotein	0/22 (0%)	0/22 (0%)	1/22 (5%)	0/22 (0%)
NLVRDLPQGFSALEPLV	SEQ ID NO: 147	surface glycoprotein	0/22 (0%)	1/22 (5%)	1/22 (5%)	1/22 (5%)
LALHRSYLTSGDSSSGWT	SEQ ID NO: 148	surface glycoprotein	1/22 (5%)	3/22 (14%)	0/22 (0%)	2/22 (10%)
KYNENGTTTDAVDCALDP LS	SEQ ID NO: 149	surface glycoprotein	1/22 (5%)	1/22 (5%)	1/22 (5%)	1/22 (5%)
IYQAGSTPCNGVEGFNCY	SEQ ID NO: 150	surface glycoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
VLSFELLHAPATVC	SEQ ID NO: 151	surface glycoprotein	1/22 (5%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
LLHAPATVCGPKKST	SEQ ID NO: 152	surface glycoprotein	0/22 (0%)	2/22 (10%)	0/22 (0%)	1/22 (5%)
VKNKCVNFNFNGLT	SEQ ID NO: 153	surface glycoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)

TESNKKFLPFQQFGRDIAD	SEQ ID NO: 154	surface glycoprotein	4/22 (19%)	15/22 (69%)	1/22 (5%)	9/22 (41%)
TDAVRDPQTLLEILDITPCS	SEQ ID NO: 155	surface glycoprotein	6/22 (28%)	17/22 (78%)	1/22 (5%)	11/22 (50%)
QVAVLYQDVNCTEVPVAI HADQLTPTW	SEQ ID NO: 156	surface glycoprotein	0/22 (0%)	9/22 (41%)	2/22 (10%)	4/22 (19%)
IHADQLTPTWRYSTGSN	SEQ ID NO: 157	surface glycoprotein	0/22 (0%)	3/22 (14%)	0/22 (0%)	2/22 (10%)
YSTGSNVFQTRAGCL	SEQ ID NO: 158	surface glycoprotein	2/22 (10%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
CASYQTQTNSPRRARSV	SEQ ID NO: 159	surface glycoprotein	2/22 (10%)	4/22 (19%)	0/22 (0%)	1/22 (5%)
ARSVASQSHIAYTMSLGAE NSVA	SEQ ID NO: 160	surface glycoprotein	2/22 (10%)	6/22 (28%)	4/22 (19%)	4/22 (19%)
PVSMTKTSVDCTMY	SEQ ID NO: 161	surface glycoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
RALTGIAVEQDKNTQEVF AQV	SEQ ID NO: 162	surface glycoprotein	11/22 (50%)	14/22 (64%)	2/22 (10%)	5/22 (23%)
FAQVKQIYKTPPIKDFGGF NF	SEQ ID NO: 163	surface glycoprotein	5/22 (23%)	8/22 (37%)	1/22 (5%)	3/22 (14%)
NFSQILPDPSKPSKRS	SEQ ID NO: 164	surface glycoprotein	1/22 (5%)	4/22 (19%)	0/22 (0%)	4/22 (19%)
KRSFIEDLLFNKVT	SEQ ID NO: 165	surface glycoprotein	5/22 (23%)	8/22 (37%)	4/22 (19%)	5/22 (23%)
EDLLFNKVTLADAGF	SEQ ID NO: 166	surface glycoprotein	1/22 (5%)	1/22 (5%)	1/22 (5%)	0/22 (0%)
YGDCLGDIAARDLIC	SEQ ID NO: 167	surface glycoprotein	1/22 (5%)	3/22 (14%)	1/22 (5%)	0/22 (0%)
ICAQKFENGLTVLPPLL	SEQ ID NO: 168	surface glycoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
FGAGAAALQIPFAMQM	SEQ ID NO: 169	surface glycoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)

NGIGVTQNVLYENQK	SEQ ID NO: 170	surface glycoprotein	1/22 (5%)	0/22 (0%)	1/22 (5%)	1/22 (5%)
IGKIQDSLSSTASA	SEQ ID NO: 171	surface glycoprotein	0/22 (0%)	1/22 (5%)	1/22 (5%)	0/22 (0%)
VLNDILSRDLKVEAE	SEQ ID NO: 172	surface glycoprotein	0/22 (0%)	2/22 (10%)	0/22 (0%)	0/22 (0%)
LSRLDKVEAEVQID	SEQ ID NO: 173	surface glycoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
AICHDGKAHFPREGVF	SEQ ID NO: 174	surface glycoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
HWFVTQRNFYEPQIITT	SEQ ID NO: 175	surface glycoprotein	0/22 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)
VIGIVNNTVYDPLQPEL	SEQ ID NO: 176	surface glycoprotein	2/22 (10%)	2/22 (10%)	0/22 (0%)	1/22 (5%)
TVYDPLQPELDSFK	SEQ ID NO: 177	surface glycoprotein	1/22 (5%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
PELDSFKEELDKYFKNHT SPDVD	SEQ ID NO: 178	surface glycoprotein	15/22 (69%)	21/22 (96%)	9/22 (41%)	18/22 (82%)
SPDVDLGDIGINASVV	SEQ ID NO: 179	surface glycoprotein	10/22 (46%)	20/22 (91%)	6/22 (28%)	13/22 (60%)
EIDRLNEVAKNLNESL	SEQ ID NO: 180	surface glycoprotein	0/22 (0%)	5/22 (23%)	0/22 (0%)	1/22 (5%)
EVAKNLNESLIDLQ	SEQ ID NO: 181	surface glycoprotein	1/22 (5%)	1/22 (5%)	2/22 (10%)	1/22 (5%)
SCGSCCKFDEDDSE	SEQ ID NO: 182	surface glycoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
FDEDDSEPVKGVKLH	SEQ ID NO: 183	surface glycoprotein	0/22 (0%)	3/22 (14%)	0/22 (0%)	1/22 (5%)
TIGTVTLKQGEIKDAT	SEQ ID NO: 184	ORF3a protein	0/22 (0%)	2/22 (10%)	0/22 (0%)	1/22 (5%)
EHDYQIGGYTEKWESGV KDCVVVL	SEQ ID NO: 185	ORF3a protein	4/22 (19%)	7/22 (32%)	1/22 (5%)	2/22 (10%)

MADSNGTITVEELK	SEQ ID NO: 186	membrane glycoprotein	0/22 (0%)	0/22 (0%)	2/22 (10%)	2/22 (10%)
RSMWTFNPETNILLNV	SEQ ID NO: 187	membrane glycoprotein	0/22 (0%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
CDIKDLPKEITVATS	SEQ ID NO: 188	membrane glycoprotein	0/22 (0%)	8/22 (37%)	0/22 (0%)	6/22 (28%)
KLGAQRVAGDSGFA	SEQ ID NO: 189	membrane glycoprotein	0/22 (0%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
QRVAGDSGFAAYSRY	SEQ ID NO: 190	membrane glycoprotein	0/22 (0%)	10/22 (46%)	0/22 (0%)	4/22 (19%)
SSGTYEGRNSPFHPLA	SEQ ID NO: 191	ORF7a protein	1/22 (5%)	1/22 (5%)	1/22 (5%)	1/22 (5%)
LIELCVDEAGSKSPIQYID	SEQ ID NO: 192	ORF8 protein	0/22 (0%)	3/22 (14%)	2/22 (10%)	1/22 (5%)
GSKSPIQYIDIGNYTVSCLP F	SEQ ID NO: 193	ORF8 protein	3/22 (14%)	12/22 (55%)	4/22 (19%)	3/22 (14%)
RSKQRRPQGLPNNT	SEQ ID NO: 194	nucleocapsid phosphoprotein	1/22 (5%)	5/22 (23%)	0/22 (0%)	1/22 (5%)
DLKFPRGGVPINT	SEQ ID NO: 195	nucleocapsid phosphoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
NTNSSPDDQIGYYRR	SEQ ID NO: 196	nucleocapsid phosphoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
KDHIGTRNPANNAA	SEQ ID NO: 197	nucleocapsid phosphoprotein	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
RGTSPARMAGNGGDA	SEQ ID NO: 198	nucleocapsid phosphoprotein	1/22 (5%)	3/22 (14%)	0/22 (0%)	0/22 (0%)
MAGNGGDAALALLLDR	SEQ ID NO: 199	nucleocapsid phosphoprotein	0/22 (0%)	2/22 (10%)	2/22 (10%)	1/22 (5%)
ALLLLDRNLQLESKMSG	SEQ ID NO: 200	nucleocapsid phosphoprotein	3/22 (14%)	5/22 (23%)	2/22 (10%)	5/22 (23%)
NQLESKMSGKGQQQQGQ TVTKKSA	SEQ ID NO: 201	nucleocapsid phosphoprotein	12/22 (55%)	18/22 (82%)	8/22 (37%)	15/22 (69%)

QTVTKKSAEASKKPRQK	SEQ ID NO: 202	nucleocapsid phosphoprotein	2/22 (10%)	7/22 (32%)	1/22 (5%)	4/22 (19%)
RGPEQTQGNFGDQELI	SEQ ID NO: 203	nucleocapsid phosphoprotein	6/22 (28%)	3/22 (14%)	2/22 (10%)	3/22 (14%)
IKLDDKDPNFKDQVIL	SEQ ID NO: 204	nucleocapsid phosphoprotein	4/22 (19%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
AYKTFPPTEPKDKKKK	SEQ ID NO: 205	nucleocapsid phosphoprotein	2/22 (10%)	5/22 (23%)	0/22 (0%)	2/22 (10%)
KDKKKKADETQALPQR	SEQ ID NO: 206	nucleocapsid phosphoprotein	3/22 (14%)	7/22 (32%)	0/22 (0%)	5/22 (23%)
ADETQALPQRQKKQQT V TLLPAADL	SEQ ID NO: 207	nucleocapsid phosphoprotein	4/22 (19%)	17/22 (78%)	4/22 (19%)	9/22 (41%)
TLLPAADLDDFSKQLQQS MSSADS	SEQ ID NO: 208	nucleocapsid phosphoprotein	14/22 (64%)	22/22 (100%)	6/22 (28%)	18/22 (82%)

Table 4 (Continued)

Epitope sequence	Asymptomatic	Other_CoV	Healthy- Controls_China	SARS-CoV- 1 (2003)
YRKVLLRKNKGKGA	SEQ ID NO: 46	0/12 (0%)	0/10 (0%)	0/10 (0%)
	SEQ ID NO: 47	1/12 (9%)	0/10 (0%)	0/10 (0%)
GNKGAGGHSYGADLKSFDL	SEQ ID NO: 48	2/12 (17%)	0/10 (0%)	0/10 (0%)
	SEQ ID NO: 49	0/12 (0%)	1/10 (10%)	0/10 (0%)
DELGTDPPYEDFQEN	SEQ ID NO: 50	0/12 (0%)	0/10 (0%)	0/10 (0%)
	SEQ ID NO: 51	0/12 (0%)	1/10 (10%)	0/10 (0%)
WNTKHSSGVTTRELM	SEQ ID NO: 51	0/12 (0%)	1/10 (10%)	0/10 (0%)
	SEQ ID NO: 51	0/12 (0%)	1/10 (10%)	0/10 (0%)

HEIAWYTERSEKSY	SEQ ID NO: 52	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
SYELQTPFEIKLAK	SEQ ID NO: 53	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
AKKFDTFENGECPNF	SEQ ID NO: 54	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
CDHCGETSWQTGDF	SEQ ID NO: 55	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
LTNIFGTVYEKLKPVLD	SEQ ID NO: 56	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
GVEFLRDGWEIFVKF	SEQ ID NO: 57	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
TGDLQPLEQPTSEAVEAPLV	SEQ ID NO: 58	0/12 (0%)	1/12 (9%)	1/10 (10%)	1/10 (10%)
KGGAPTKVTFGDDTVIEV	SEQ ID NO: 59	0/12 (0%)	0/12 (0%)	1/10 (10%)	1/10 (10%)
NITFELDERIDKVL	SEQ ID NO: 60	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
EFACVVADAVIKTLQPV/S	SEQ ID NO: 61	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
NKGEDIQLLKSAYENF	SEQ ID NO: 62	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
YDKLVSSFLEMKSEK	SEQ ID NO: 63	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
AFYILPSHSNEKQEI	SEQ ID NO: 64	0/12 (0%)	1/12 (9%)	1/10 (10%)	0/10 (0%)
LGYVTHGLNLEEA	SEQ ID NO: 65	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
THGLNLEEAARYMRS	SEQ ID NO: 66	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
TAYNGYLTSSSKTP	SEQ ID NO: 67	0/12 (0%)	1/12 (9%)	0/10 (0%)	1/10 (10%)

GEVITFDNLKTLSS	SEQ ID NO: 68	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
GRYMSALNHTKKWKYPQV	SEQ ID NO: 69	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
TVGELGDVRETMSYL	SEQ ID NO: 70	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
MGTLSYEQFKKGVQIPC	SEQ ID NO: 71	0/12 (0%)	0/12 (0%)	2/10 (20%)	0/10 (0%)
FTCASEYTGNYQCGHY	SEQ ID NO: 72	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
YQCGHYKHITSKET	SEQ ID NO: 73	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
LVPNQPYPNASFDNF	SEQ ID NO: 74	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
FPDLNGDVVAIDYK	SEQ ID NO: 75	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
NSFDVLKSEDAQGMNDL	SEQ ID NO: 76	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
SEEVVENPTIQKDVLECN	SEQ ID NO: 77	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
GLAAVNSVPWDITIA	SEQ ID NO: 78	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
TIANYAKPFLNKVVSTTT	SEQ ID NO: 79	0/12 (0%)	3/12 (25%)	2/10 (20%)	0/10 (0%)
FLNKVVSTTTNIVTRC	SEQ ID NO: 80	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
VKMFDAYVNTFSST	SEQ ID NO: 81	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
FSSTFNVPMKLLKT	SEQ ID NO: 82	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
TLVATAEAEELAKNVSL	SEQ ID NO: 83	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)

SAAKNNLPFKLTCA	SEQ ID NO: 84	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
VHVMSKHTDFSSEIG	SEQ ID NO: 85	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
DFDTWFSQRGGSYT	SEQ ID NO: 86	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
TFDSEYCRHGTCE	SEQ ID NO: 87	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
DVLYQPPQTSITS AVL	SEQ ID NO: 88	1/12 (9%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
NRFTTTLNDFNLVAMK	SEQ ID NO: 89	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
LTQDHVDILGPLSA	SEQ ID NO: 90	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
GMNGRTILGSALLED	SEQ ID NO: 91	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
QGLLPPKNSIDAFKLNK	SEQ ID NO: 92	0/12 (0%)	2/12 (17%)	0/10 (0%)	0/10 (0%)
KLKKS LNVAKSEFDRD	SEQ ID NO: 93	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
LRANS AVKLQNNELS	SEQ ID NO: 94	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
AVKLQNNELSPVALR	SEQ ID NO: 95	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
CAAGTTQTACTDDN	SEQ ID NO: 96	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
LALLSDLQDLKWARF	SEQ ID NO: 97	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
ATVRLQAGNATEVPA	SEQ ID NO: 98	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
VKMLCTHTGTGQAITY	SEQ ID NO: 99	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)

KYVQIPTTCANDPV	SEQ ID NO: 100	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
RVCGVSAARLTTCGT	SEQ ID NO: 101	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
AVAKHDFKFRIDG	SEQ ID NO: 102	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
GDFIQTPGSGVPV	SEQ ID NO: 103	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
QTYHPNCVNCLDDR	SEQ ID NO: 104	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
ALTNNVAFQTVKPGNF	SEQ ID NO: 105	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
GFFKEGSSVELKHF	SEQ ID NO: 106	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
QLLFVVEVDKYFDCYDGGC	SEQ ID NO: 107	0/12 (0%)	3/12 (25%)	1/10 (10%)	0/10 (0%)
SLYVKPGGTSSGDATTAYA	SEQ ID NO: 108	0/12 (0%)	0/12 (0%)	2/10 (20%)	0/10 (0%)
NRDVDTDVNEFYAYL	SEQ ID NO: 109	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
SQGLVASIKNFKSV	SEQ ID NO: 110	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
QHTMLVKQGDDYVY	SEQ ID NO: 111	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
VDDIVKTDGTLME	SEQ ID NO: 112	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
AIDAYPLTKHPNQE	SEQ ID NO: 113	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
GCDVTDVTVQLYLG	SEQ ID NO: 114	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
GLYKNTCVGSDNVT	SEQ ID NO: 115	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)

NTCVGSDNVTDNFNAIATCDW	SEQ ID NO: 116	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
YQKVGMQKYSTLQG	SEQ ID NO: 117	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
STLQGGPGTGKSHF	SEQ ID NO: 118	1/12 (9%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
ARARVECFDKFKVNS	SEQ ID NO: 119	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
DPAQLPAPRTLTK	SEQ ID NO: 120	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
PRTLTKGTLEPEY	SEQ ID NO: 121	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
IFTQTETAHSCNV	SEQ ID NO: 122	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
GCHATREAVGTNLP	SEQ ID NO: 123	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
VQMLSDTLKNLSDR	SEQ ID NO: 124	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
FDYVYNPFMIDVQQWG	SEQ ID NO: 125	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
MIDVQQWGTGNLQ	SEQ ID NO: 126	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
TGNLQSNHDL YCQVH	SEQ ID NO: 127	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
LADKFPVLHDIGNPKAICVP	SEQ ID NO: 128	0/12 (0%)	1/12 (9%)	0/10 (0%)	1/10 (10%)
FYSYATHSDKFTDGV	SEQ ID NO: 129	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
FDTRVLSNLSLPGCD	SEQ ID NO: 130	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
QVVSDIDYVPLKSATC	SEQ ID NO: 131	1/12 (9%)	0/12 (0%)	0/10 (0%)	0/10 (0%)

PSVGPQKQASLNGVTL	SEQ ID NO: 132	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
GVVQQLPETYFTQSRN	SEQ ID NO: 133	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
AMDEFIERYKLEGY	SEQ ID NO: 134	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
LAKRFKESPELED	SEQ ID NO: 135	0/12 (0%)	0/12 (0%)	1/10 (10%)	1/10 (10%)
FITDAQTGSSKVCVCSVID	SEQ ID NO: 136	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
IKSQDLSVVSKVVKV	SEQ ID NO: 137	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
YPKLQSSQAWQPGV	SEQ ID NO: 138	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
QRMILLEKCDLQNYG	SEQ ID NO: 139	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
HFGAGSDKGVAPGTA	SEQ ID NO: 140	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
ITEHSWNADLYKLM	SEQ ID NO: 141	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
LSSYSLFDMSKFPLKL	SEQ ID NO: 142	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
SLLLCRMNSRNYIA	SEQ ID NO: 143	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
QCVNLTTRTQLPPA	SEQ ID NO: 144	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
TQLPPAYTNSFTRGVYYPD	SEQ ID NO: 145	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
FHAHVSGTNGTKRF	SEQ ID NO: 146	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
NLVRDLPQGFSALEPLV	SEQ ID NO: 147	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)

LALHRSYLTPGDSSSGWT	SEQ ID NO: 148	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
KYNENGTITDAVDCALDPLS	SEQ ID NO: 149	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
IYQAGSTPCNGVEGFCY	SEQ ID NO: 150	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
VLSFELLHAPATVC	SEQ ID NO: 151	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
LLHAPATVCGPKKST	SEQ ID NO: 152	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
VKNKCVNFNFNGLT	SEQ ID NO: 153	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
TESNKKFLPFQFGRDIAD	SEQ ID NO: 154	3/12 (25%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
TDAVRDPQTLEILDITPCS	SEQ ID NO: 155	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
QVAVLYQDVNCTEVPVAIHADQLTPTW	SEQ ID NO: 156	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
IHADQLTPTWRVYSTGSN	SEQ ID NO: 157	1/12 (9%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
YSTGSNVFQTRAGCL	SEQ ID NO: 158	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
CASYQTQTNPRRARSV	SEQ ID NO: 159	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
ARSVASQSIAYTMSLGAENSA	SEQ ID NO: 160	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
PVSMTKTSVDCCTMY	SEQ ID NO: 161	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
RALTGIAVEQDKNTQEVFAQV	SEQ ID NO: 162	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
FAQVKQIYKTPPIKDFGGFNF	SEQ ID NO: 163	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)

NFSQILPDPSKPSKRS	SEQ ID NO: 164	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
KRSFIEDLLFNKVT	SEQ ID NO: 165	1/12 (9%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
EDLLFNKVTLADAGF	SEQ ID NO: 166	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
YGDCLGDIARDLIC	SEQ ID NO: 167	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
ICAQKFNGLTVLPPLL	SEQ ID NO: 168	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
FGAGAAIQIPFAMQM	SEQ ID NO: 169	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
NGIGVTQNVLYENQK	SEQ ID NO: 170	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
IGKIQDSLSTASA	SEQ ID NO: 171	0/12 (0%)	1/12 (9%)	1/10 (10%)	0/10 (0%)
VLNDILSRLDKVEAE	SEQ ID NO: 172	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
LSRLDKVEAEVQID	SEQ ID NO: 173	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
AICHDGKAHFPREGVF	SEQ ID NO: 174	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
HWFVTQRNFYEPQIITT	SEQ ID NO: 175	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
VIGIVNNTVYDPLQPEL	SEQ ID NO: 176	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
TVYDPLQPELDSFK	SEQ ID NO: 177	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
PELDSFKEELDKYFKNHTSPDVD	SEQ ID NO: 178	8/12 (67%)	1/12 (9%)	0/10 (0%)	1/10 (10%)
SPDVDLGDISGINASVV	SEQ ID NO: 179	0/12 (0%)	1/12 (9%)	2/10 (20%)	1/10 (10%)

EIDRLNEVAKNLNLSL	SEQ ID NO: 180	1/12 (9%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
EVAKNLNLSLIDLQ	SEQ ID NO: 181	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
SCGSCCKFDEDDSE	SEQ ID NO: 182	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
FDEDDSEPVKGVKLH	SEQ ID NO: 183	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
TIGTVTLKQGEIKDAT	SEQ ID NO: 184	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
EHDYQIGGYTEKWESGVKDCVVL	SEQ ID NO: 185	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
MADSNGTITVEELK	SEQ ID NO: 186	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
RSMWSFNPETNILLNV	SEQ ID NO: 187	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
CDIKDLPKEITVATS	SEQ ID NO: 188	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
KLGASQRVAGDSGFA	SEQ ID NO: 189	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
QRVAGDSGFAAYSRY	SEQ ID NO: 190	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
SSGTYEGNSPFHPLA	SEQ ID NO: 191	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
LIELCVDEAGSKSPIQYID	SEQ ID NO: 192	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
GSKSPIQYIDIGNYTVSCLPF	SEQ ID NO: 193	0/12 (0%)	1/12 (9%)	0/10 (0%)	0/10 (0%)
RSKQRRPQGLPNNT	SEQ ID NO: 194	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
DLKFPRGQGVPIINT	SEQ ID NO: 195	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)

NTNSSPDDQIGYYRR	SEQ ID NO: 196	0/12 (0%)	0/12 (0%)	0/10 (0%)	1/10 (10%)
KDHIGTRNPANAA	SEQ ID NO: 197	0/12 (0%)	0/12 (0%)	1/10 (10%)	0/10 (0%)
RGTSPARMAGNGGDA	SEQ ID NO: 198	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
MAGNGGDAALALLLDR	SEQ ID NO: 199	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
ALLLLDRNLNLESKMSG	SEQ ID NO: 200	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
NQLESKMSGKGQQQQGQTVTKKSA	SEQ ID NO: 201	7/12 (59%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
QTVTKKSAAEASKKPRQK	SEQ ID NO: 202	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
RGPEQTQGNFGDQELI	SEQ ID NO: 203	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
IKLDDKDPNFKDQVIL	SEQ ID NO: 204	0/12 (0%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
AYKTFPPTEPKKDKKKK	SEQ ID NO: 205	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
KDKKKKADETQALPQR	SEQ ID NO: 206	3/12 (25%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
ADETQALPQRQKKQQTVTLPAADL	SEQ ID NO: 207	2/12 (17%)	0/12 (0%)	0/10 (0%)	0/10 (0%)
TLLPAADLDDFSKQLQQSMSSADS	SEQ ID NO: 208	7/12 (59%)	1/12 (9%)	0/10 (0%)	0/10 (0%)

Table 5 - IgG linear IgG epitopes for detection of HKU-1, NL63, OC43, 229E, SARS-CoV-1. Sequences (aa) are based on proteome of reference sequences from NCBI

			Controls				
	Virus name		Epitope sequences	Protein name	SARS-CoV-1	Other Human CoV	Healthy
1.	Human coronavirus HKU1	SEQ ID NO: 209	MVNPSKGSSGLLIT KLT	Nucleocapsid phosphoprotein	0/10 (0%)	4/12 (34%)	0/10 (0%)
2.	Human coronavirus HKU1	SEQ ID NO: 210	VKQLPEQFDSL NLS AGTQ	Nucleocapsid phosphoprotein	0/10 (0%)	2/12 (17%)	1/10 (10%)
3.	Human coronavirus HKU1	SEQ ID NO: 211	SLLATLDDPYVEDS VA	Nucleocapsid phosphoprotein	1/10 (10%)	2/12 (17%)	0/10 (0%)
4.	Human coronavirus HKU1	SEQ ID NO: 212	MNKSFLPQFTSDQA V	Membrane glycoprotein	5/10 (50%)	4/12 (34%)	6/10 (60%)
5.	Human coronavirus HKU1	SEQ ID NO: 213	WLN NFNVP SPLNW ERKI	Spike glycoprotein	0/10 (0%)	2/12 (17%)	0/10 (0%)
6.	Human coronavirus HKU1	SEQ ID NO: 214	ISASYRFVTFEPFNV S	Spike glycoprotein	2/10 (20%)	4/12 (34%)	2/10 (20%)
7.	Human coronavirus HKU1	SEQ ID NO: 215	SRSFFEDLLFDKVK LS	Spike glycoprotein	0/10 (0%)	1/12 (9%)	1/10 (10%)
8.	Human coronavirus HKU1	SEQ ID NO: 216	VDKQVILVSN NKE DFDIIQ	orf1ab polyprotein	0/10 (0%)	1/12 (9%)	1/10 (10%)
9.	Human coronavirus HKU1	SEQ ID NO: 217	LLHSKVKVVP NLY VVV	orf1ab polyprotein	1/10 (10%)	3/12 (25%)	2/10 (20%)
10.	Human coronavirus HKU1	SEQ ID NO: 218	FDLVQYDFTDYKL EL	orf1ab polyprotein	1/10 (10%)	1/12 (9%)	0/10 (0%)
11.	Human coronavirus HKU1	SEQ ID NO: 219	PASQVIVNNYDKSA GY	orf1ab polyprotein	0/10 (0%)	2/12 (17%)	1/10 (10%)
12.	Human coronavirus HKU1	SEQ ID NO: 220	CTGSPYIDDFNKIAS	orf1ab polyprotein	0/10 (0%)	1/12 (9%)	0/10 (0%)
13.	Human coronavirus HKU1	SEQ ID NO: 221	CWETGKVKPPLNK NYVFTG	orf1ab polyprotein	1/10 (10%)	1/12 (9%)	2/10 (20%)

14.	Human coronavirus HKU1	SEQ ID NO: 222	AKVRVDCYDKFKI NDTTCKY	orf1 ab polyprotein	1/10 (10%)	2/12 (17%)	0/10 (0%)
15.	Human coronavirus HKU1	SEQ ID NO: 223	ISPYN SQNYVAKRV LGVQ	orf1 ab polyprotein	0/10 (0%)	1/12 (9%)	1/10 (10%)
16.	Human coronavirus NL63	SEQ ID NO: 224	EFRDYFNNTDSIV	orf1 ab polyprotein	1/10 (10%)	2/12 (17%)	2/10 (20%)
17.	Human coronavirus NL63	SEQ ID NO: 225	RKASGLFDAIWDAF VAAI	orf1 ab polyprotein	0/10 (0%)	1/12 (9%)	2/10 (20%)
18.	Human coronavirus NL63	SEQ ID NO: 226	NVGEPIISQPSKLLN G	orf1 ab polyprotein	2/10 (20%)	1/12 (9%)	0/10 (0%)
19.	Human coronavirus NL63	SEQ ID NO: 227	ISYLN RARGSSAAR LEPC	orf1 ab polyprotein	1/10 (10%)	2/12 (17%)	0/10 (0%)
20.	Human coronavirus NL63	SEQ ID NO: 228	CIKACEVVVTNLNK SAGWPLNKFKA	orf1 ab polyprotein	1/10 (10%)	3/12 (25%)	1/10 (10%)
21.	Human coronavirus NL63	SEQ ID NO: 229	VFINKYGLEDFNFE H	orf1 ab polyprotein	0/10 (0%)	1/12 (9%)	0/10 (0%)
22.	Human coronavirus NL63	SEQ ID NO: 230	TVLNASIPESKPLA DDDSA	Nucleocapsid phosphoprotein	7/10 (70%)	3/12 (25%)	6/10 (60%)
23.	Human coronavirus NL63	SEQ ID NO: 231	ASQEGVLSEREKLL HLI	Membrane glycoprotein	0/10 (0%)	2/12 (17%)	0/10 (0%)
24.	Human coronavirus NL63	SEQ ID NO: 232	QFGLQDGFYSANFL DDNVL	Spike glycoprotein	3/10 (30%)	4/12 (34%)	6/10 (60%)
25.	Human coronavirus NL63	SEQ ID NO: 233	CDCGSTKLPPYEF KVHVQ	Spike glycoprotein	0/10 (0%)	3/12 (25%)	7/10 (70%)
26.	Human coronavirus OC43	SEQ ID NO: 234	FDLVQYDFTDYKL EL	nsp9	1/10 (10%)	1/12 (9%)	0/10 (0%)
27.	Human coronavirus OC43	SEQ ID NO: 235	SQVIVNNYDKSAG YPF	nsp9	0/10 (0%)	2/12 (17%)	0/10 (0%)
28.	Human coronavirus OC43	SEQ ID NO: 236	LSTCDGQKFTDES YKNMY	nsp9	0/10 (0%)	2/12 (17%)	1/10 (10%)
29.	Human coronavirus OC43	SEQ ID NO: 237	FPLKLAGTAVINLR AD	nsp13	0/10 (0%)	2/12 (17%)	0/10 (0%)

30.	Human coronavirus OC43	SEQ ID NO: 238	HATRDSIGTNTFPLQ L	nsp11	0/10 (0%)	1/12 (9%)	1/10 (10%)
31.	Human coronavirus OC43	SEQ ID NO: 239	CTGSPYIDDFENRIAS	nsp10	0/10 (0%)	1/12 (9%)	0/10 (0%)
32.	Human coronavirus OC43	SEQ ID NO: 240	AKVRVECYDKFKI NDTTRKYV	nsp10	1/10 (10%)	3/12 (25%)	0/10 (0%)
33.	Human coronavirus OC43	SEQ ID NO: 241	ASSRSAIEDLLFDK VKLS	Spike glycoprotein	5/10 (50%)	3/12 (25%)	5/10 (50%)
34.	Human coronavirus OC43	SEQ ID NO: 242	SQVIVNNYDKSAG YPF	orf1ab polyprotein	0/10 (0%)	2/12 (17%)	0/10 (0%)
35.	Human coronavirus OC43	SEQ ID NO: 243	LSTCDGQKFTDES YKNMY	orf1ab polyprotein	0/10 (0%)	2/12 (17%)	1/10 (10%)
36.	Human coronavirus OC43	SEQ ID NO: 244	AETQKATEEAFKQS YA	orf1ab polyprotein	0/10 (0%)	2/12 (17%)	0/10 (0%)
37.	Human coronavirus OC43	SEQ ID NO: 245	SWEIGKVKPPLNKN YVFTG	orf1ab polyprotein	1/10 (10%)	1/12 (9%)	3/10 (30%)
38.	Human coronavirus OC43	SEQ ID NO: 246	AKVRVECYDKFKI NDTTRKYV	orf1ab polyprotein	2/10 (20%)	3/12 (25%)	1/10 (10%)
39.	Human coronavirus OC43	SEQ ID NO: 247	HATRDSIGTNTFPLQ L	orf1ab polyprotein	0/10 (0%)	2/12 (17%)	1/10 (10%)
40.	Human coronavirus 229E	SEQ ID NO: 248	DVIDNEIIVKPNISL CVPL	orf1ab polyprotein	3/10 (30%)	4/12 (34%)	3/10 (30%)
41.	Human coronavirus 229E	SEQ ID NO: 249	KHSDFGGLGDLVDS YFEND	orf1ab polyprotein	0/10 (0%)	1/12 (9%)	2/10 (20%)
42.	Human coronavirus 229E	SEQ ID NO: 250	CITSREVVVTNLNK SA	orf1ab polyprotein	0/10 (0%)	3/12 (25%)	0/10 (0%)
43.	Human coronavirus 229E	SEQ ID NO: 251	DVIDNEIIVKPNISL CVPL	orf1ab polyprotein	3/10 (30%)	4/12 (34%)	3/10 (30%)
44.	Human coronavirus 229E	SEQ ID NO: 252	KHSDFGGLGDLVDS YFEND	orf1ab polyprotein	0/10 (0%)	1/12 (9%)	2/10 (20%)

45.	SARS-CoV-1	SEQ ID NO: 253	LYEQVVMIDYLDNL KPRVEAPKQEEPPN TED	orf1ab polyprotein	7/10 (70%)	3/12 (25%)	4/10 (40%)
46.	SARS-CoV-1	SEQ ID NO: 254	VSVSSPDADVTTYNG YLTSSSKTSEEHF	nsp3	3/10 (30%)	0/12 (0%)	2/10 (20%)
47.	SARS-CoV-1	SEQ ID NO: 255	LGDELGTDPIDYE QNWNTKHGSGALR	orf1ab polyprotein	3/10 (30%)	2/12 (17%)	3/10 (30%)
48.	SARS-CoV-1	SEQ ID NO: 256	FTLRSITAQPVKIDN ASPASTVHATA	Orf3	4/10 (40%)	0/12 (0%)	0/10 (0%)
49.	SARS-CoV-1	SEQ ID NO: 257	TIVVTEGDGISTPKL KEDYQIGGYSEDR	Orf3	5/10 (50%)	1/12 (9%)	0/10 (0%)
50.	SARS-CoV-1	SEQ ID NO: 258	SSGVANPAMDPIYD EPTTTTSVPL	Orf3	4/10 (40%)	1/12 (9%)	1/10 (10%)
51.	SARS-CoV-1	SEQ ID NO: 259	MADNGTITVEELK QLLEQWNLVIG	membrane glycoprotein	6/10 (60%)	1/12 (9%)	0/10 (0%)
52.	SARS-CoV-1	SEQ ID NO: 260	TPKDHIGTRNPNN AATVLQLPQGTTLP KGFYAEGRGGS	Nucleocapsid phosphoprotein	5/10 (50%)	2/12 (17%)	0/10 (0%)
53.	SARS-CoV-1	SEQ ID NO: 261	FSLDVSSEKSGNFKH LREFVF	spike glycoprotein	4/10 (40%)	0/12 (0%)	1/10 (10%)

Table 5 (continued)

	Epitope sequences	Protein name	Asymptomatic (first)	Asymptomatic (Second)	Mild disease (first)	Mild disease (second)	Severe disease (first)	Severe disease (second)
1.	MVNPSKGSGLLIT KLT	Nucleocapsid phosphoprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	1/22 (5%)	0/22 (0%)	0/22 (0%)
2.	VKQLPEQFDSLNL AGTQ	Nucleocapsid phosphoprotein	2/6 (34%)	0/6 (0%)	6/22 (28%)	6/22 (28%)	10/22 (46%)	11/22 (50%)

3.	SLLATLDDPYVEDS V/A	SEQ ID NO: 211	Nucleocapsid phosphoprotein	0/6 (0%)	0/6 (0%)	4/22 (19%)	1/22 (5%)	5/22 (23%)	10/22 (46%)
4.	MNKSFLPQFTSDQA V	SEQ ID NO: 212	Membrane glycoprotein	6/6 (100%)	4/6 (67%)	19/22 (87%)	15/22 (69%)	18/22 (82%)	21/22 (96%)
5.	WLNFNVPSPLNWE RKI	SEQ ID NO: 213	Spike glycoprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	0/22 (0%)	1/22 (5%)	0/22 (0%)
6.	ISASYRFVTFEPFNV S	SEQ ID NO: 214	Spike glycoprotein	2/6 (34%)	0/6 (0%)	11/22 (50%)	10/22 (46%)	11/22 (50%)	10/22 (46%)
7.	SRSFFEDLLFDKVKL S	SEQ ID NO: 215	Spike glycoprotein	0/6 (0%)	1/6 (17%)	5/22 (23%)	3/22 (14%)	3/22 (14%)	4/22 (19%)
8.	VDKQVILVSNKED FDIIQ	SEQ ID NO: 216	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	2/22 (10%)	7/22 (32%)	11/22 (50%)
9.	LLHSKVKVVPNLYV VV	SEQ ID NO: 217	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	1/22 (5%)	0/22 (0%)	2/22 (10%)	2/22 (10%)
10.	FDLVQYDFTDYKLE L	SEQ ID NO: 218	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	4/22 (19%)	2/22 (10%)	7/22 (32%)	11/22 (50%)
11.	PASQVIVNNYDKSA GY	SEQ ID NO: 219	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	0/22 (0%)	4/22 (19%)	3/22 (14%)
12.	CTGSPYIDDFNKIAS	SEQ ID NO: 220	orf1ab polyprotein	0/6 (0%)	1/6 (17%)	6/22 (28%)	10/22 (46%)	11/22 (50%)	14/22 (64%)
13.	CWETGKVKPPLNK NYVFTG	SEQ ID NO: 221	orf1ab polyprotein	1/6 (17%)	2/6 (34%)	4/22 (19%)	5/22 (23%)	4/22 (19%)	5/22 (23%)
14.	AKVRVDCYDKFKIN DTTCKY	SEQ ID NO: 222	orf1ab polyprotein	2/6 (34%)	2/6 (34%)	2/22 (10%)	1/22 (5%)	4/22 (19%)	4/22 (19%)
15.	ISPYN SQNYVAKRV LGVQ	SEQ ID NO: 223	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	2/22 (10%)	2/22 (10%)	5/22 (23%)	6/22 (28%)
16.	EFRDYFNNTDSIV	SEQ ID NO: 224	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	3/22 (14%)	3/22 (14%)	6/22 (28%)	8/22 (37%)
17.	RKASGLFDAIWDAF VA AI	SEQ ID NO: 225	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	2/22 (10%)	1/22 (5%)	0/22 (0%)	1/22 (5%)
18.	NVGEPIISQPSKLLN G	SEQ ID NO: 226	orf1ab polyprotein	1/6 (17%)	1/6 (17%)	0/22 (0%)	0/22 (0%)	2/22 (10%)	1/22 (5%)

19.	ISYLNRRARGSSAARL EPC	SEQ ID NO: 227	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
20.	CIKACEVVVTNLNK SAGWPLNKFKA	SEQ ID NO: 228	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	1/22 (5%)	2/22 (10%)	1/22 (5%)	3/22 (14%)
21.	VFINKYGLEDFENFE H	SEQ ID NO: 229	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	0/22 (0%)	5/22 (23%)	5/22 (23%)
22.	TVLNASIPESKPLAD DDSA	SEQ ID NO: 230	Nucleocapsid phosphoprotein	3/6 (50%)	3/6 (50%)	15/22 (69%)	17/22 (78%)	15/22 (69%)	21/22 (96%)
23.	ASQEGVLSEREKLL HLI	SEQ ID NO: 231	Membrane glycoprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)	2/22 (10%)
24.	QFGLQDGFYSANFL DDNVL	SEQ ID NO: 232	Spike glycoprotein	1/6 (17%)	1/6 (17%)	6/22 (28%)	7/22 (32%)	8/22 (37%)	13/22 (60%)
25.	CDCGSTKLPPYYEFE KVHVQ	SEQ ID NO: 233	Spike glycoprotein	1/6 (17%)	2/6 (34%)	8/22 (37%)	6/22 (28%)	12/22 (55%)	15/22 (69%)
26.	FDLVQYDFTDYKLE L	SEQ ID NO: 234	nsp9	0/6 (0%)	0/6 (0%)	4/22 (19%)	2/22 (10%)	7/22 (32%)	12/22 (55%)
27.	SQVIVNNYDKSAGY PF	SEQ ID NO: 235	nsp9	0/6 (0%)	0/6 (0%)	0/22 (0%)	0/22 (0%)	6/22 (28%)	9/22 (41%)
28.	LSTCDGQKFTDES YKNMY	SEQ ID NO: 236	nsp9	0/6 (0%)	0/6 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)	6/22 (28%)
29.	FPLKLAGTAVINLR AD	SEQ ID NO: 237	nsp13	0/6 (0%)	0/6 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
30.	HATRDSIGTNFPLQL	SEQ ID NO: 238	nsp11	0/6 (0%)	0/6 (0%)	5/22 (23%)	6/22 (28%)	2/22 (10%)	6/22 (28%)
31.	CTGSPYIDDFNRIAS	SEQ ID NO: 239	nsp10	0/6 (0%)	0/6 (0%)	2/22 (10%)	2/22 (10%)	3/22 (14%)	5/22 (23%)
32.	AKVRVECYDKFKIN DTRKYY	SEQ ID NO: 240	nsp10	1/6 (17%)	2/6 (34%)	1/22 (5%)	3/22 (14%)	2/22 (10%)	5/22 (23%)
33.	ASSRSAIEDLLFDKV KLS	SEQ ID NO: 241	Spike glycoprotein	1/6 (17%)	3/6 (50%)	13/22 (60%)	14/22 (64%)	13/22 (60%)	16/22 (73%)
34.	SQVIVNNYDKSAGY PF	SEQ ID NO: 242	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)

35.	LSTCDGQKFTDES YKNMY	SEQ ID NO: 243	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	1/22 (5%)	1/22 (5%)	6/22 (28%)
36.	AETQKATEEAFKQ YA	SEQ ID NO: 244	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)	0/22 (0%)
37.	SWEIGKVKPPLNKN YVFTG	SEQ ID NO: 245	orf1ab polyprotein	2/6 (34%)	2/6 (34%)	5/22 (23%)	4/22 (19%)	5/22 (23%)	6/22 (28%)
38.	AKVRVECYDKFKIN DTTRKYV	SEQ ID NO: 246	orf1ab polyprotein	1/6 (17%)	2/6 (34%)	1/22 (5%)	3/22 (14%)	5/22 (23%)	8/22 (37%)
39.	HATRDSIGTNFPLQL	SEQ ID NO: 247	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	6/22 (28%)	7/22 (32%)	2/22 (10%)	6/22 (28%)
40.	DVIDNEIIVKPNISLC VPL	SEQ ID NO: 248	orf1ab polyprotein	1/6 (17%)	0/6 (0%)	9/22 (41%)	11/22 (50%)	16/22 (73%)	19/22 (87%)
41.	KHSDFGGLGDLVDSY FEND	SEQ ID NO: 249	orf1ab polyprotein	0/6 (0%)	1/6 (17%)	10/22 (46%)	7/22 (32%)	15/22 (69%)	19/22 (87%)
42.	CITSREVVVNTLNKS A	SEQ ID NO: 250	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	0/22 (0%)	0/22 (0%)	2/22 (10%)	1/22 (5%)
43.	DVIDNEIIVKPNISLC VPL	SEQ ID NO: 251	orf1ab polyprotein	1/6 (17%)	0/6 (0%)	10/22 (46%)	11/22 (50%)	16/22 (73%)	19/22 (87%)
44.	KHSDFGGLGDLVDSY FEND	SEQ ID NO: 252	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	12/22 (55%)	7/22 (32%)	15/22 (69%)	19/22 (87%)
45.	LYEQVVM DYLDNL KPRVEAPKQEEPPN TED	SEQ ID NO: 253	orf1ab polyprotein	3/6 (50%)	4/6 (67%)	12/22 (55%)	14/22 (64%)	13/22 (60%)	14/22 (64%)
46.	VSVSSPDVAVTTYNG YLTSSSKTSEEHF	SEQ ID NO: 254	nsp3	0/6 (0%)	0/6 (0%)	10/22 (46%)	10/22 (46%)	7/22 (32%)	12/22 (55%)
47.	LGDELGTDPIDYE QNWNTKHGSGALR	SEQ ID NO: 255	orf1ab polyprotein	0/6 (0%)	0/6 (0%)	10/22 (46%)	13/22 (60%)	17/22 (78%)	18/22 (82%)
48.	FTLSITAQPVKIDN ASPASTVHATA	SEQ ID NO: 256	Orf3	1/6 (17%)	1/6 (17%)	0/22 (0%)	1/22 (5%)	0/22 (0%)	1/22 (5%)
49.	TIVVTEGDGISTPKL KEDYQIGGYSEDR	SEQ ID NO: 257	Orf3	0/6 (0%)	0/6 (0%)	0/22 (0%)	0/22 (0%)	2/22 (10%)	1/22 (5%)

50.	SSGVANPAMDPIYD EPTTTSVPL	SEQ ID NO: 258	Orf3	1/6 (17%)	0/6 (0%)	0/22 (0%)	2/22 (10%)	4/22 (19%)	8/22 (37%)
51.	MADNGTTTVEELKQ LLEQWNLVIG	SEQ ID NO: 259	membrane glycoprotein	0/6 (0%)	1/6 (17%)	1/22 (5%)	15/22 (69%)	4/22 (19%)	13/22 (60%)
52.	TPKDHIGTRNPNN AATVLQLPQGTTL KGFYAEGRGGS	SEQ ID NO: 260	Nucleocapsid phosphoprotein	2/6 (34%)	3/6 (50%)	10/22 (46%)	16/22 (73%)	15/22 (69%)	22/22 (100%)
53.	FSLDVSEKSGNFKH LREFVF	SEQ ID NO: 261	spike glycoprotein	0/6 (0%)	0/6 (0%)	1/22 (5%)	2/22 (10%)	1/22 (5%)	4/22 (19%)

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CLAIMS

1. An isolated peptide that is strongly reactive with, and specific for antibodies to SARS-CoV-2, wherein the peptide comprises the amino acid sequence chosen from the group consisting of SEQ ID NOs: 1-45.
- 5 2. A collection of isolated peptides comprising at least one peptide of claim 1.
3. A collection of isolated peptides that are strongly reactive with, and specific for antibodies to SARS-CoV-2, wherein the peptides comprise amino acid sequences shifted one residue across at least one peptide of claim 1.
4. The collection of isolated peptides of claims 2 or 3, wherein the peptides range in
10 length from about 6 amino acids to 50 amino acids.
5. The collection of isolated peptides of claims 2 or 3, wherein one or more of the isolated peptides is conjugated to a ligand, avidin, streptavidin, neutravidin, serum albumin, keyhole limpet hemocyanin (KLH), an enzyme, or a metallic nanoparticle or nanoshell or is biotinylated.
- 15 6. The collection of isolated peptides of claims 2 or 3 wherein one or more of the isolated peptides is immobilized to a solid support.
7. A collection of isolated peptides comprising: a peptide or peptide chosen from the group consisting of SEQ ID NOs: 1-45; and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of SEQ ID
20 NOs: 1-45; or combinations thereof.
8. A peptide microarray comprising the collection of isolated peptides of any of claims 2 - 7.
9. A method for the differential serological detection of serological detection of exposure to and/or infection by SARS-CoV-2 in a sample, comprising:
25 a. contacting the sample with the collection of peptides of any of claims 2- 7, under conditions sufficient to allow binding of antibody(ies) to the peptide(s); and
b. detecting formation of an antibody-peptide complex comprising said one or more peptides in the collection,
wherein formation of the complex is indicative of an antibody to an epitope of a
30 SARS-CoV-2 antigen being present in the sample.
10. The method of claim 10, wherein the sample is from a subject.
11. The method of claim 10, wherein the subject is a test subject who has been administered a corona virus vaccine or immunomodulatory agent.

12. The method of claim 10, wherein the sample is chosen from the group consisting of nasopharyngeal aspirate, blood, cerebrospinal fluid, saliva, serum, plasma, urine, sputum, bronchial lavage, pericardial fluid, and peritoneal fluid.
13. The method of claim 10, wherein the sample is chosen from the group consisting of cells, cell culture, cell culture medium and compositions used for the development of pharmaceutical and therapeutic agents.
14. The method of claim 10, wherein the collection of peptides is immobilized to a solid support.
15. The method of claim 9, further comprising contacting the sample with a collection of peptides comprising a peptide or peptides chosen from the group consisting of SEQ ID NOs: 209-261; and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of SEQ ID NOs: 209-261; or combinations thereof, and further detecting the formation of an antibody-peptide complex comprising said one or more peptides in the collection, wherein formation of the complex is indicative of an antibody to an epitope of another human coronavirus other than SARS-CoV-2 being present in the sample
16. A method for the differential serological detection of exposure to and/or infection by SARS-CoV-2 in a sample, comprising:
 - a. contacting the sample with at least one peptide comprising the amino acid sequence chosen from the group consisting of SEQ ID NOs: 1-45, under conditions sufficient to allow binding of antibody(ies) in the sample to the peptide(s); and
 - b. detecting formation of an antibody-peptide complex comprising said one or more peptides,
 wherein formation of the complex is indicative of an antibody to an epitope of SARS-CoV-2 being present in the sample.
17. The method of claim 16, wherein the sample is from a subject.
18. The method of claim 17, wherein the subject is a test subject which has been administered a SARS-CoV-2 vaccine or immunomodulatory agent.
19. The method of claim 16, wherein the sample is chosen from the group consisting of nasopharyngeal aspirate, blood, cerebrospinal fluid, saliva, serum, plasma, urine, sputum, bronchial lavage, pericardial fluid, and peritoneal fluid.
20. The method of claim 16, wherein the sample is chosen from the group consisting of cells, cell culture, cell culture medium and compositions used for the development of pharmaceutical and therapeutic agents.

21. The method of claim 16, further comprising contacting the sample with at least one peptide comprising the amino acid sequence chosen from the group consisting of SEQ ID NOs: 209-261 and further detecting the formation of an antibody-peptide complex comprising said peptide(s), wherein formation of the complex is indicative of an antibody to an epitope of another human coronavirus other than SARS-CoV-2 being present in the sample
22. The method of any of claims 9-21, wherein the method is performed using an ELISA assay, lateral flow immunoassay, an agglutination assay, Western blot, a slot blot, dot blot or a programmable peptide array.
23. A kit comprising the collection of isolated peptides of any of claims 2-7.
24. The kit of claim 22, further comprising one or more additional components chosen from the group consisting of diluents and buffers, labeled conjugates, other reagents for the detection of specifically bound antigens or antibodies, other signal-generating reagents, coating reagents, capture reagents, washing reagents, and instructions for use.
25. The kit of claim 23, further comprising a collection of peptides comprising a peptide or peptides chosen from the group consisting of SEQ ID NOs: 209-261; and/or a collection or set of peptides shifted one residue across at least one peptide chosen from the group consisting of SEQ ID NOs: 209-261; or combinations thereof.

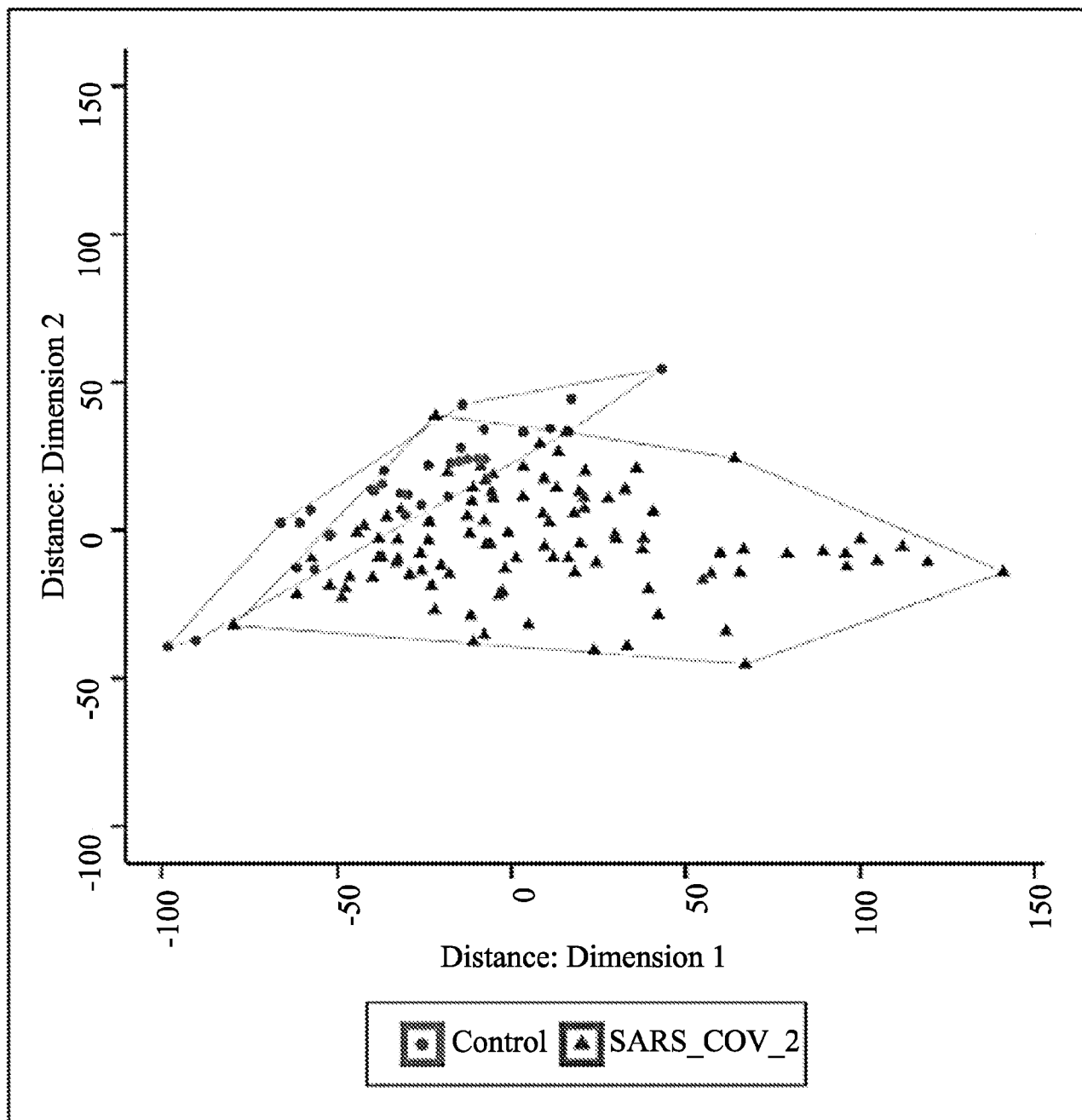


Fig. 1

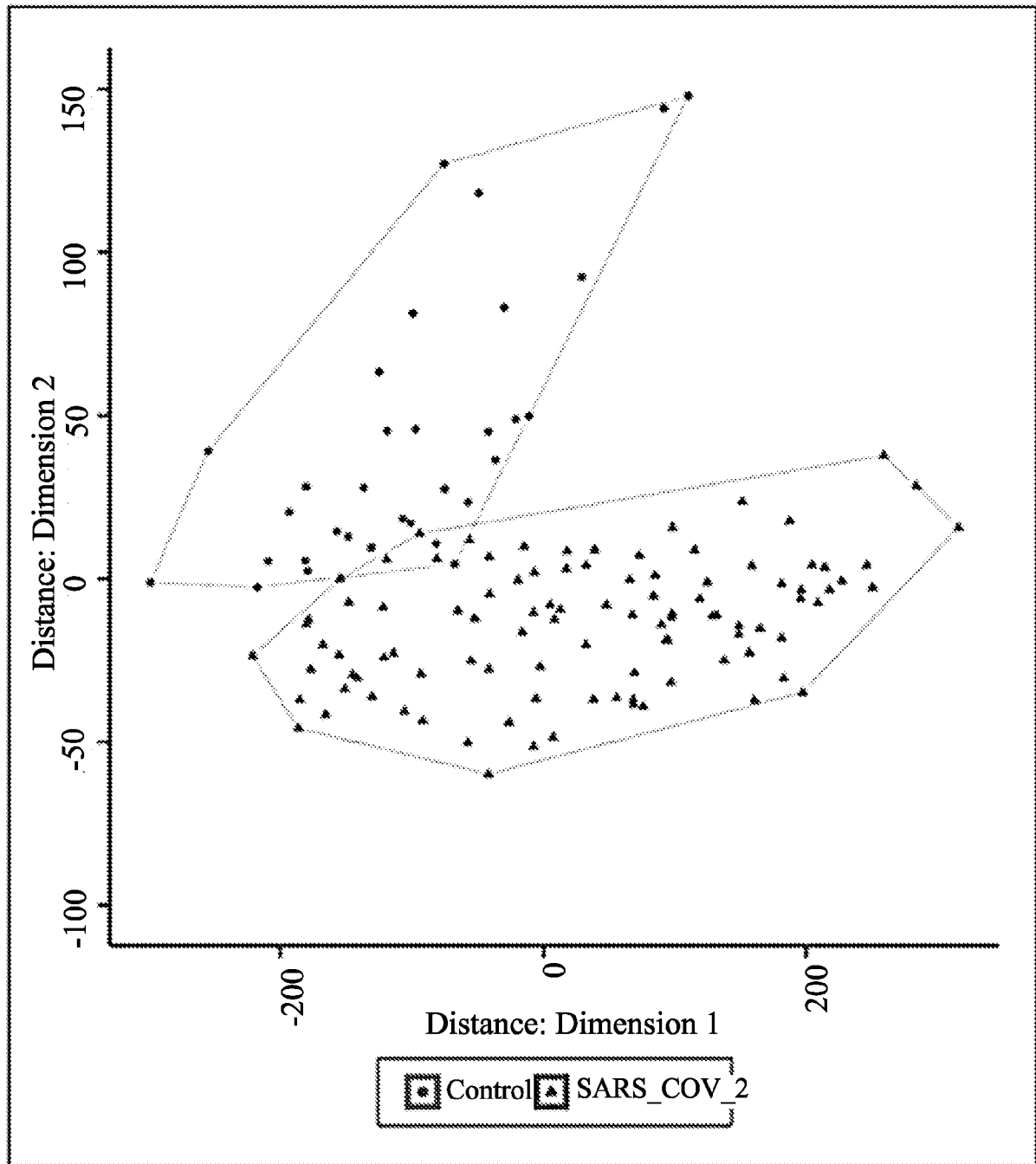


Fig. 2

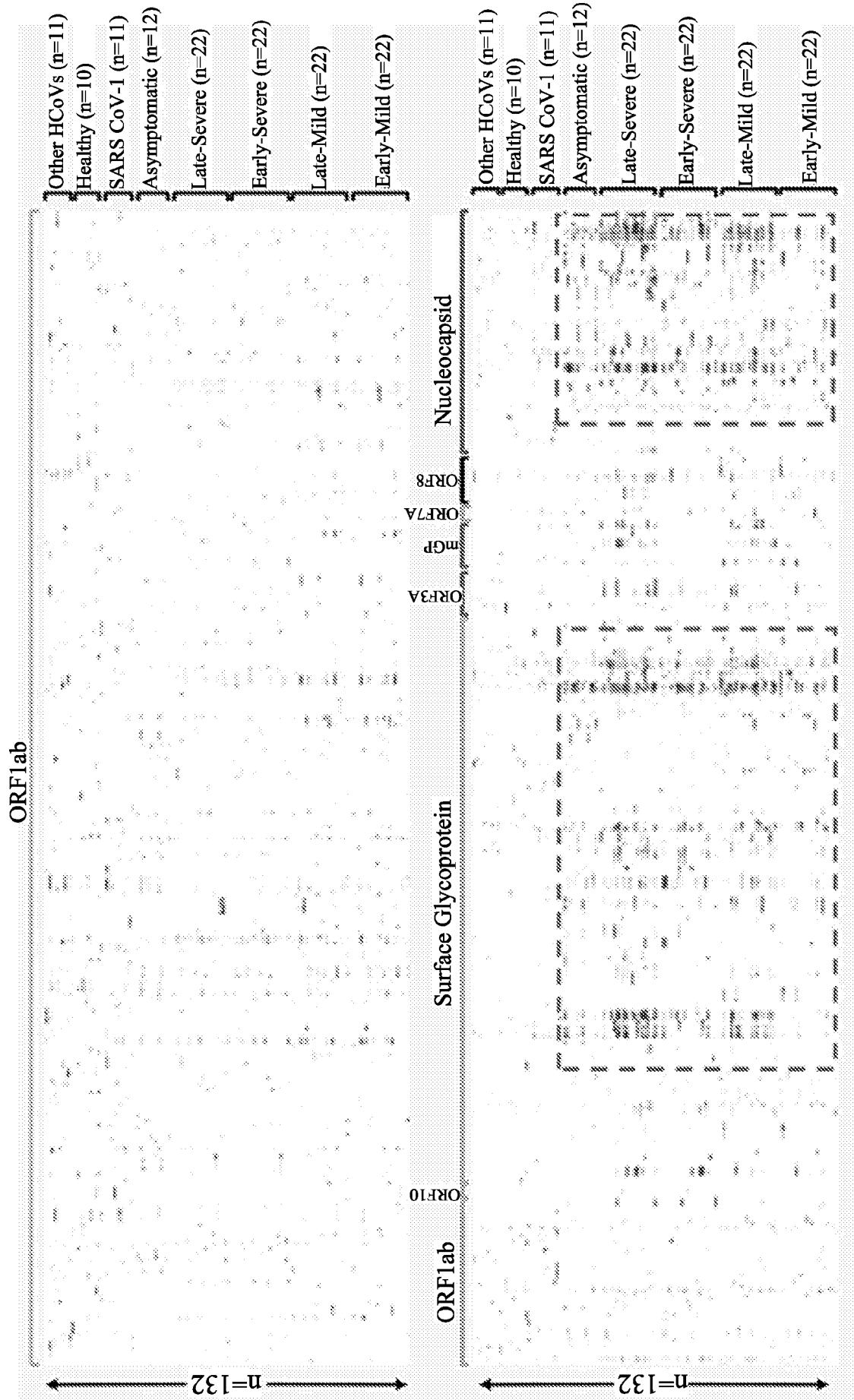


Fig. 3

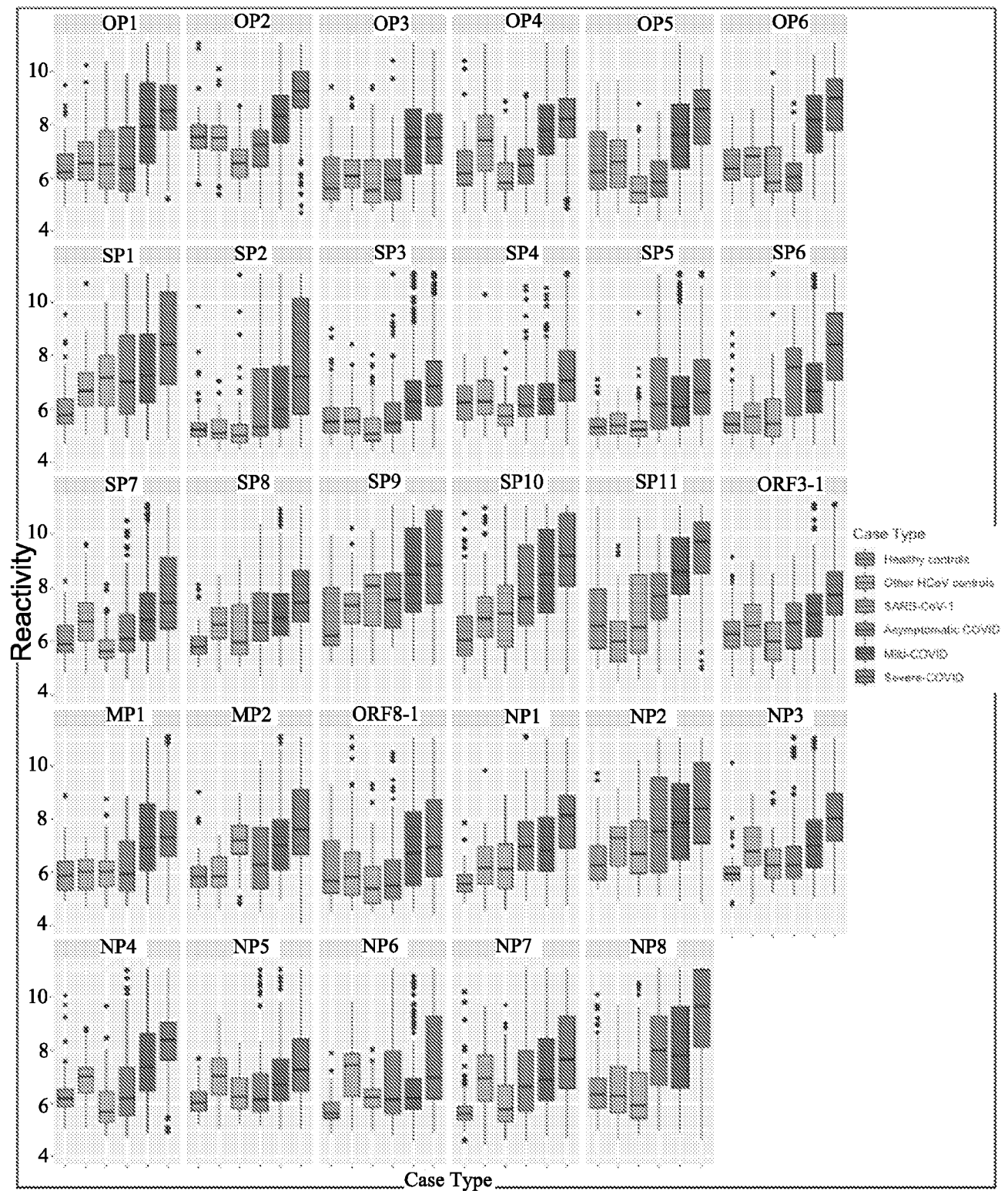


Fig. 4