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(54) Title: SARS SPIKE PEPTIDES AND USES THEREOF

(57) Abstract: The present invention relates to a composition comprising at least one spike peptide species covering at least 60% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 60% identical thereto. The present invention also relates to a polynucleotide encoding said spike peptide and to host cells, methods and kits related thereto.



SARS spike peptides and uses thereof

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The new SARS-like coronavirus SARS-CoV-2 (also known as 2019-nCoV), the virus causing coronavirus disease 2019 (COVID-19) is spreading fast throughout the worldwide population. Currently, there is no specific therapy or vaccination known. Prevention of transmission is based on isolation quarantine of carriers. Equally, potential carriers are usually placed under
15 quarantine. However vast majority of infected people do not develop noticeable clinical symptoms. RT-PCR tests can identify subjects who are currently infected with SARS-CoV-2; however, subjects having successfully resolved infection cannot be identified by such tests.

There is, thus, a need in the art for improved means and methods for diagnosing the immune
20 status of subjects with regards to SARS-CoV-2, avoiding the drawbacks of the prior art. This problem is solved by the means and methods disclosed herein with the features of the independent claims. Advantageous embodiments which might be realized in an isolated fashion or in any arbitrary combinations are listed in the dependent claims.

25 In accordance, the present invention relates to a composition comprising at least one spike peptide species covering at least 60% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 60% identical thereto.

In general, terms used herein are to be given their ordinary and customary meaning to a
30 person of ordinary skill in the art and, unless indicated otherwise, are not to be limited to a special or customized meaning. As used in the following, the terms “have”, “comprise” or “include” or any arbitrary grammatical variations thereof are used in a non-exclusive way. Thus, these terms may both refer to a situation in which, besides the feature introduced by these terms, no further features are present in the entity described in this context and to a

situation in which one or more further features are present. As an example, the expressions “A has B”, “A comprises B” and “A includes B” may both refer to a situation in which, besides B, no other element is present in A (i.e. a situation in which A solely and exclusively consists of B) and to a situation in which, besides B, one or more further elements are present in entity A, such as element C, elements C and D or even further elements. Also, as is understood by the skilled person, the expressions "comprising a" and "comprising an" preferably refer to "comprising one or more", i.e. are equivalent to "comprising at least one".

Further, as used in the following, the terms "preferably", "more preferably", "most preferably", "particularly", "more particularly", "specifically", "more specifically" or similar terms are used in conjunction with optional features, without restricting further possibilities. Thus, features introduced by these terms are optional features and are not intended to restrict the scope of the claims in any way. The invention may, as the skilled person will recognize, be performed by using alternative features. Similarly, features introduced by "in an embodiment" or similar expressions are intended to be optional features, without any restriction regarding further embodiments of the invention, without any restrictions regarding the scope of the invention and without any restriction regarding the possibility of combining the features introduced in such way with other optional or non-optional features of the invention.

As used herein, the term "standard conditions", if not otherwise noted, relates to IUPAC standard ambient temperature and pressure (SATP) conditions, i.e. preferably, a temperature of 25°C and an absolute pressure of 100 kPa; also preferably, standard conditions include a pH of 7. Moreover, if not otherwise indicated, the term "about" relates to the indicated value with the commonly accepted technical precision in the relevant field, preferably relates to the indicated value $\pm 20\%$, more preferably $\pm 10\%$, most preferably $\pm 5\%$. Further, the term "essentially" indicates that deviations having influence on the indicated result or use are absent, i.e. potential deviations do not cause the indicated result to deviate by more than $\pm 20\%$, more preferably $\pm 10\%$, most preferably $\pm 5\%$. Thus, “consisting essentially of” means including the components specified but excluding other components except for materials present as impurities, unavoidable materials present as a result of processes used to provide the components, and components added for a purpose other than achieving the technical effect of the invention. For example, a composition defined using the phrase “consisting essentially of” encompasses any known acceptable additive, excipient, diluent, carrier, and the like.

Preferably, a composition consisting essentially of a set of components will comprise less than 5% by weight, more preferably less than 3% by weight, even more preferably less than 1% by weight, most preferably less than 0.1% by weight of non-specified component(s).

- 5 The degree of identity (e.g. expressed as "%identity") between two biological sequences, preferably DNA, RNA, or amino acid sequences, can be determined by algorithms well known in the art. Preferably, the degree of identity is determined by comparing two optimally aligned sequences over a comparison window, where the fragment of sequence in the comparison window may comprise additions or deletions (e.g., gaps or overhangs) as compared to the sequence it is compared to for optimal alignment. The percentage is calculated by determining, preferably over the whole length of the polynucleotide, peptide, or polypeptide, the number of positions at which the identical residue occurs in both sequences to yield the number of matched positions, dividing the number of matched positions by the total number of positions in the window of comparison and multiplying the result by 100 to yield the percentage of sequence identity. Optimal alignment of sequences for comparison may be conducted by the local homology algorithm of Smith and Waterman (1981), by the homology alignment algorithm of Needleman and Wunsch (1970), by the search for similarity method of Pearson and Lipman (1988), by computerized implementations of these algorithms (GAP, BESTFIT, BLAST, PASTA, and TFASTA in the Wisconsin Genetics Software Package, Genetics Computer Group (GCG), 575 Science Dr., Madison, WI), or by visual inspection. Given that two sequences have been identified for comparison, GAP and BESTFIT are preferably employed to determine their optimal alignment and, thus, the degree of identity. Preferably, the default values of 5.00 for gap weight and 0.30 for gap weight length are used. In the context of biological sequences referred to herein, the term "essentially identical" indicates a %identity value of at least 80%, preferably at least 90%, more preferably at least 98%, most preferably at least 99%. As will be understood, the term essentially identical includes 100% identity. The aforesaid applies to the term "essentially complementary" mutatis mutandis.
- 30 The term "fragment" of a biological macromolecule, preferably of a polynucleotide or polypeptide, is used herein in a wide sense relating to any sub-part, preferably subdomain, of the respective biological macromolecule comprising the indicated sequence, structure and/or function. Thus, the term includes sub-parts generated by actual fragmentation of a biological macromolecule, but also sub-parts derived from the respective biological macromolecule in

an abstract manner, e.g. in silico. Thus, as used herein, an Fc or Fab fragment, but also e.g. a single-chain antibody, a bispecific antibody, and a nanobody may be referred to as fragments of an immunoglobulin.

5 Unless specifically indicated otherwise herein, the compounds specified, in particular the spike peptide species, may be comprised in larger structures, e.g. may be covalently or non-covalently linked to carrier molecules, retardants, and other excipients. In particular, spike peptides as specified may be comprised in fusion polypeptides comprising further peptides, which may serve e.g. as a tag for purification and/or detection, as a linker, or to extend the in vivo half-life of a compound. The term “detectable tag” refers to a stretch of amino acids which are added to or introduced into the fusion polypeptide; preferably, the tag is added C- or N- terminally to the fusion polypeptide of the present invention. Said stretch of amino acids preferably allows for detection of the fusion polypeptide by an antibody which specifically recognizes the tag; or it preferably allows for forming a functional conformation, such as a chelator; or it preferably allows for visualization, e.g. in the case of fluorescent tags. Preferred detectable tags are the Myc-tag, FLAG-tag, 6-His-tag, HA-tag, GST-tag or a fluorescent protein tag, e.g. a GFP-tag. These tags are all well known in the art. Other further peptides preferably comprised in a fusion polypeptide comprise further amino acids or other modifications which may serve as mediators of secretion, as mediators of blood-brain-barrier passage, as cell-penetrating peptides, and/or as immune stimulants. Further polypeptides or peptides to which the peptides may be fused are signal sequences, transport sequences, and/or linker sequences.

The term “composition”, as used herein, relates to a composition of matter comprising the spike peptide(s) as specified and optionally one or more acceptable carrier(s). Preferably, the composition is a diagnostic composition and/or is a pharmaceutical composition; thus, the composition, preferably, comprises the spike peptide(s) as indicated as diagnostically and/or pharmaceutically active compound, and, preferably, the carrier is a diagnostically and/or pharmaceutically acceptable carrier. The diagnostically and/or pharmaceutically active compound can be formulated as, preferably pharmaceutically acceptable, salt. Preferred salts comprise acetate, methylester, HCl, sulfate, chloride and the like. The composition comprises at least one spike peptide; thus, the composition preferably comprises one spike peptide covering SEQ ID NO:1 or comprises a multitude of spike peptides covering SEQ ID NO:1, the term "multitude, preferably referring to at least two, more preferably referring to of from 2

to 50, in an embodiment of from 5 to 50 spike peptides. As specified herein below, the spike peptides may cover SEQ ID NO:1 in an overlapping manner.

Preferably, the composition is a diagnostic composition. The term "diagnostic composition" is understood by the skilled person. Preferably, the term relates to a composition comprising a spike peptide as specified herein as diagnostically active compound, i.e. as a compound enabling the diagnosis, preferably the diagnosis as specified herein below. The diagnostic composition may comprise further compounds, in particular one or more carrier(s). Preferably, the further compounds are the further compounds as specified herein below for the further compounds optionally comprised in a pharmaceutical composition. As will be understood, the carrier(s) in the diagnostic composition must be acceptable in the sense of being compatible with the other ingredients of the formulation and being not deleterious to establishing the diagnosis. Preferably, the carrier is water, phosphate-buffered saline, DMSO, acetonitrile, a pharmaceutical carrier, or any combination thereof.

Also preferably, the composition is a pharmaceutical composition, i.e., preferably, a medicament. The terms "medicament" and "pharmaceutical composition" are, in principle, known to the skilled person. As referred to herein, the term relate to any composition containing the spike peptide(s) or at least one expression construct encoding the same as pharmaceutically active compound and one or more other components such as one or more pharmaceutically acceptable carrier(s). The pharmaceutically active compound can be present in liquid or dry, e.g. lyophilized, form. For example, the pharmaceutically active compound can be present together with glycerol and/or protein stabilizers (e.g., human serum albumin). The medicament is, typically, administered systemically and, preferably, subcutaneously or intramuscularly. However, depending on the nature of the formulation and the desired therapeutic application, the medicament may be administered by other routes as well. The pharmaceutically active compound is the active ingredient or drug of the medicament, and is preferably administered in conventional dosage forms prepared by combining the drug with standard pharmaceutical carriers according to conventional procedures. These procedures may involve mixing, granulating, and compression, or dissolving the ingredients as appropriate to the desired preparation. It will be appreciated that the form and character of the pharmaceutical acceptable carrier or diluent is dictated by the amount of active ingredient with which it is to be combined, the route of administration, and other well-known variables. The carrier(s) must be acceptable in the sense of being compatible with the other ingredients

of the formulation and being not deleterious to the recipient thereof. The pharmaceutical carrier employed may include a solid, a gel, or a liquid. Exemplary of solid carriers are lactose, terra alba, sucrose, talc, gelatin, agar, pectin, acacia, magnesium stearate, stearic acid and the like. Exemplary of liquid carriers are phosphate buffered saline solution, syrup, oil, water, emulsions, various types of wetting agents, and the like. Similarly, the carrier or diluent may include time delay material well known to the art, such as glyceryl mono-stearate or glyceryl distearate alone or with a wax. Said suitable carriers comprise those mentioned above and others well known in the art, see, e.g., Remington's Pharmaceutical Sciences, Mack Publishing Company, Easton, Pennsylvania. The diluent(s) is/are selected so as not to affect the biological activity of the combination. Examples of such diluents are distilled water, physiological saline, Ringer's solutions, dextrose solution, and Hank's solution. In addition, the pharmaceutical composition or formulation may also include other carriers, adjuvants, or non-toxic, non-therapeutic, non-immunogenic stabilizers and the like. The medicament referred to herein is, preferably, administered at least once, e.g. as a bolus. However, the said medicament may be administered more than one time and, preferably, at least twice, e.g. permanently or periodically after defined time windows.

The term "peptide", as used herein, refers to a molecule consisting of several, typically at least 2 amino acids that are covalently linked to each other by peptide bonds. Molecules consisting of more than 100 amino acids covalently linked by peptide bonds are usually considered to be "polypeptides". The spike peptide(s) comprised in the composition cover the sequence of SEQ ID NO:1 (CNGVEGFNCYFPLQSYGFQPTNGVGYQPYPYR, corresponding to amino acids 480 to 509 of Chain A of the spike glycoprotein of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2, Genbank acc NO. YP_009724390.1) or a sequence at least 60% identical to SEQ ID NO:1. The term 60% identical is understood by the skilled person, in particular in view of the details provided herein above. Preferably, the spike peptide has a length of at least five amino acids, preferably at least ten amino acids, more preferably at least 20 amino acids, most preferably at least 25 amino acids. Also preferably, the spike peptide has a length of from 10 to 500 amino acids, preferably of from 15 to 400 amino acids, more preferably of from 20 to 300 amino acids, still more preferably of from 25 to 200 amino acids, still more preferably of from 27 to 100 amino acids, even more preferably of from 28 to 50 amino acids, most preferably of from 29 to 40 amino acids. More preferably, the spike peptide has a length of about 30 amino acids, more preferably of 30 amino acids. Preferably, the aforesaid amino acids correspond to the sequence of SEQ ID NO:1 as

specified herein above. Preferably, the at least one spike peptide species covers at least 60% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto. More preferably, the at least one spike peptide species covers at least 70% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto. Still more preferably, the at least one spike peptide species covers at least 80% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto. Even more preferably, the at least one spike peptide species covers at least 90% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto. Even more preferably, the at least one spike peptide species covers at least 94% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto. Most preferably, the at least one spike peptide species covers at least 96% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto. Preferably, the at least one spike peptide comprises, preferably consists of, the amino acid sequence of SEQ ID NO:1 or a variant comprising an exchange of the cysteine at position 9 to an amino acid having a size and charge similar to cysteine, preferably 2-amino butyric acid (Abu), serine (Ser), leucine (Leu), or isoleucine (Ile), more preferably Abu or ser, most preferably Abu. Preferably, the at least one spike peptide comprises, preferably consists of, the amino acid sequence of SEQ ID NO:1 or 2 (CNGVEGFNXYFPLQSYGFQPTNGVGYQPYPYR, with X = alpha aminobutyric acid); thus, preferably, the at least one spike peptide species is one spike peptide species.

The term "peptide species", as used herein, relates to a multitude of peptide molecules consisting, within the precision of the manufacturing process, essentially of the same amino acid sequence. Thus, the term "composition comprising a spike peptide species" relates to a composition comprising a multitude of spike peptides having essentially the same amino acid sequence.

The term "covering" a sequence is understood by the skilled person. Preferably, the term relates to the spike peptide(s) comprised in the composition corresponding, taken together, to the indicated fraction of the amino acids of SEQ ID NO:1. Whether a peptide corresponds to a given amino acid sequence can be established by the skilled person by aligning the sequence of the peptide(s) to the amino acid sequence. As used herein, a partial coverage of SEQ ID NO:1 by a spike peptide is taken into account, provided it corresponds to the indicated %identity and consists of at least five, preferably ten, contiguous amino acids. As will be understood, in case the composition comprises more than one spike peptide, the coverages of the respective spike peptides are summed up to provide the coverage as specified herein. As will also be understood, the share of SEQ ID NO:1 covered is determined based on the amino acid sequence of SEQ ID NO:1, so in case the spike peptides comprised in the composition cover SEQ ID NO:1 in an overlapping manner, double or several-fold coverage is not taken into account in determination of coverage. Preferably, determination of coverage with each spike peptide starts with the first identical amino acid at the N-terminus and ends after the last identical amino acid at the C-terminus.

Advantageously, it was found in the work underlying the present invention that the sequence of SEQ ID NO:1 is a sequence to which neutralizing antibodies are formed in subjects resolving a n SARS-2 infection. Thus, the spike peptide can be used for serologically identifying subjects afflicted or having been afflicted with a SARS-2 infection. Moreover, the spike peptide is useful for inducing neutralizing antibodies, i.e. for vaccination.

The definitions made above apply mutatis mutandis to the following. Additional definitions and explanations made further below also apply for all embodiments described in this specification mutatis mutandis.

The present invention further relates to a polynucleotide encoding the at least one spike peptide species of the present invention.

The term "polynucleotide" as used herein refers to single- or double-stranded DNA or RNA molecules. Encompassed by the term is genomic DNA, cDNA, hnRNA, mRNA as well as all naturally occurring or artificial derivatives of such molecular species, including fragments thereof. The polynucleotide may be, preferably, a linear or circular molecule. Moreover, in addition to the nucleic acid sequences encoding the aforementioned peptide(s), a

polynucleotide according to the present invention may comprise additional sequences required for proper transcription and/or translation such as 5'- or 3'-UTR sequences or sequences required for splicing or RNA stability. Preferably, the polynucleotide is comprised in an expression construct allowing for expression of the peptide(s) in a host cell or a subject.

5 The term "expression construct" as used herein refers to a heterologous polynucleotide comprising the aforementioned polynucleotide encoding the peptide(s) as well as nucleic acids required for expression of the polynucleotide encoding the fusion polypeptide. Typically, such additional nucleic acids, which preferably are heterologous to the polynucleotide encoding the peptide(s), may be promoter sequences, enhancer sequences
10 and/or transcription termination sequences such as terminators. Moreover, the expression construct may also comprise further nucleic acids required for introducing the expression construct into a host. For example, if expression in host cells is desired, the expression construct may comprise further nucleic acids required for transformation or transfection and for propagation of the introduced expression construct in the host cells.

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The term "encoding the at least one spike peptide", as used herein, is used to relate to a polynucleotide encoding the one or more spike peptide(s) required to cover the indicated fraction of SEQ ID NO:1. Thus, preferably, in case the spike peptide consists of SEQ ID NO:1, the polynucleotide comprises at least one open reading frame encoding the spike
20 peptide. Also preferably, in case the coverage of SEQ ID NO:1 is accomplished by a multitude of spike peptides, this multitude of spike peptides may be encoded by one open reading frame on the polynucleotide, e.g. as a, optionally self cleaving, fusion polypeptide, or as a multitude of open reading frames.

25 The present invention also relates to a host cell comprising the at least one spike peptide species as specified herein and/or the polynucleotide of the present invention.

As used herein, the term "host cell" relates to any cell capable of receiving and, preferably maintaining and/or expressing, the at least one spike peptide and/or the polynucleotide of the
30 present invention. More preferably, the host cell is capable of expressing a one or more spike peptide(s) as specified herein encoded on said polynucleotide. Preferably, the cell is a bacterial cell, more preferably a cell of a common laboratory bacterial strain known in the art, most preferably an Escherichia strain, in particular an E. coli strain. Also preferably, the host cell is an eukaryotic cell, preferably a yeast cell, e.g. a cell of a strain of baker's yeast, or is an

animal cell. More preferably, the host cell is an insect cell or a mammalian cell, preferably from a mammalian subject as specified herein above, in particular a mouse or rat cell. Most preferably, the host cell is a human cell. It is, however, also envisaged that the host cell is a plant cell.

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The present invention also relates to a composition according to the invention, the polynucleotide according to the invention, or the host cell according to the invention for use in diagnosis and/or medicine.

10 The present invention also relates to a composition according to the invention, the polynucleotide according to the invention, or the host cell according to the invention for use in diagnosis of coronavirus infection, preferably of infection with coronavirus SARS-CoV-2.

The term "diagnosis" and "diagnosing" as used herein relate to an assessment whether a
15 subject suffers from a disease referred to herein, or not. As will be understood by those skilled in the art, such an assessment is usually not intended to be correct for all (i.e. 100%) of the subjects to be identified. Preferably, the term, however, requires that a statistically significant portion of subjects can be identified (e.g., a cohort in a cohort study). Whether a portion is statistically significant can be determined without further ado by the person skilled in the art
20 using various well known statistic evaluation tools, e.g., determination of confidence intervals, p-value determination, Student's t-test, Mann-Whitney test etc. Details are found in Dowdy and Wearden, Statistics for Research, John Wiley & Sons, New York 1983. Preferred confidence intervals are at least 90%, at least 95%, at least 97%, at least 98% or at least 99%. The p-values are, preferably, 0.1, 0.05, 0.01, 0.005, or 0.0001. Preferably, the probability
25 envisaged by the present invention allows that the finding of coronavirus infection will be correct for at least 60%, at least 70%, at least 80%, or at least 90% of the subjects of a given cohort or population. Diagnosis according to the present invention includes applications of the method in monitoring, confirmation, and sub-classification of the relevant disease. Moreover, the establishment of a diagnosis as used herein may also include establishing a prognosis for a
30 subject. Such a prognosis is a predictive indicator for the further development of the disease in a future time window, i.e. the predictive window. Thus, a diagnosis as used herein, preferably, encompasses a prediction of whether a subject will improve with respect to the disease or diseases symptoms in the future or whether the disease or symptoms will become worse. Accordingly, the subject matter of the invention can be also applied for risk

stratification approaches and, thus, for determining the amount of intensive care and hospitalization which will be required for an individual subject suffering from a disease referred to herein. Preferably, diagnosis includes or is diagnosis of past infection and may include diagnosis at a stage where the infection has been cleared by the body of a subject.

5 Thus, the diagnosis, preferably, is diagnosis of immune status.

The term "coronavirus" is understood by the skilled person. Preferably the coronavirus is SARS-CoV-2. The terms "SARS-CoV-2" and "severe acute respiratory syndrome coronavirus 2" are understood by the skilled person. Preferably, the terms relate to the virus identified in
10 Genbank entry NCBI:txid2697049. Symptoms and diseases caused by coronavirus infection and in particular SARS-CoV-2 infection are known to the skilled person.

The term "subject" as used herein refers to any kind of animal encompassing, e.g., mammals, birds, fish or reptiles. Typically, the animal, however, is a mammal such as a mammals used
15 as pets including dogs, cats, horses, or rodents, laboratory animals, e.g., rats, mice or apes, or farming animals such as pigs, cows, goats, or sheep. More preferably, the mammal is a primate and, most preferably, a human. The subject according to the present invention shall preferably be known or suspected to suffer from coronavirus infection. In the context of diagnosis, the subject preferably is an animal infectable by a coronavirus, preferably a bat or a
20 human, more preferably a human. In the context of medical applications, the subject preferably is an animal suitable for antibody production, preferably a mouse or a rat, a rabbit, a sheep, a goat, a horse, a llama, or a donkey, or is a human.

The present invention also relates to a composition according to the present invention, the
25 polynucleotide according to the present invention, or the host cell according to the present invention for use in preventing and/or treating coronavirus infection, preferably infection with coronavirus SARS-CoV-2.

The term "preventing" as used herein refers to avoiding the onset of the disease or at least one
30 symptom associated therewith or to prevent the worsening of the disease or the said at least one symptom. The prevention as referred to herein can be typically achieved shortly after the peptide(s) is/are administered. If the administration stopped, however, the prevention may not persist for an unlimited time but may remain present for a certain preventive time window after application of the drug (time of immunity). Typically, a preventive time window in

accordance with the present invention may be at least 1 year, at least 2 years, at least 3 years, at least 4 years, at least 5 years, at least 10 years, or at least 20 years. However, the preventive time window may also depend on the dosage of a peptide(s) as well as the mode of administration, the kind of formulation, and/or the number of administrations. For example, if a high dosage is applied, usually longer preventive time windows can be achieved. The same holds true if repeated doses are administered. It will be understood that prevention might not be effective in all subjects. However, according to the present invention it is envisaged that prevention preferably will be effective in at least a statistically significant portion of subjects. It is well known to the skilled artisan how to determine a statistically significant portion of subjects that can be effectively prevented. Whether a portion is statistically significant can be determined without further ado by the person skilled in the art using various well known statistic evaluation tools as discussed above. In view of the above, prevention, preferably, is vaccination against coronavirus infection.

The term “treating”, as used herein, refers to ameliorating or curing a disease or at least one symptom associated therewith. Thus, if there is amelioration or cure of the disease or at least a symptom associated therewith, the treatment shall be deemed to be effective. It will be understood that treating might not be effective in all subjects. However, according to the present invention it is envisaged that treatment will preferably be effective in at least a statistically significant portion of subjects to be treated. It is well known to the skilled artisan how to determine a statistically significant portion of subjects that can be effectively treated. Whether a portion is statistically significant can be determined without further ado by the person skilled in the art using various well known statistic evaluation tools, discussed herein above.

The present invention also relates to a method of detecting a coronavirus infection in a subject comprising

- a) contacting a sample of said subject with a composition according to the present invention,
- b) determining binding of sample constituents to the at least one spike peptide comprised in said composition, and
- c) based on determining step b), detecting a coronavirus infection.

The method of the present invention, preferably, is an in vitro method. Moreover, it may comprise steps in addition to those explicitly mentioned above. For example, further steps

may relate, e.g., to providing a sample for step a), or providing treatment to said subject based on the result of step c). Moreover, one or more of said steps may be performed by automated equipment.

- 5 The term "sample" refers to a sample of separated cells or to a sample from a tissue or an organ. Tissue or organ samples may be obtained from any tissue or organ by, e.g., biopsy. Separated cells may be obtained from the body fluids, such as lymph, blood, plasma, serum, liquor and other, or from the tissues or organs by separating techniques such as centrifugation or cell sorting. Preferably, the sample is a tissue or body fluid sample which comprises
- 10 immunoglobulins, preferably antibodies. Preferably the sample is a sample of a body fluid, preferably a blood, plasma, or serum sample. The sample can be obtained from the subject by routine techniques which are well known to the person skilled in the art, e.g., venous or arterial puncture or open biopsy including aspiration of tissue or cellular material from a subject. For those areas which cannot be easily reached via an open biopsy, a surgery and,
- 15 preferably, minimal invasive surgery can be performed.

- The term "sample constituent" generally relates to any compound, preferably compound comprising or being at least one macromolecule, comprised in a sample. Preferably, the sample constituent is a macromolecule, more preferably a polypeptide, still more preferably
- 20 an immunoglobulin, even more preferably an antibody, most preferably an IgG or IgM. Also preferably, the sample constituent is a cell, preferably a B-cell or a T-cell. As is understood by the skilled person, binding of a cell to the spike peptide may require its presentation in the context of a biological macromolecule, such as a major histocompatibility complex (MHC).

- 25 The present invention also relates to a method of manufacturing a coronavirus diagnostic, comprising chemically synthesizing the spike peptide or spike peptides of a composition according to the present invention and formulating said spike peptide or spike peptides as a diagnostic composition.

- 30 The present invention further relates to a kit comprising a composition according to the present invention and a detection agent.

The term "kit", as used herein, refers to a collection of the aforementioned compounds, means or reagents of the present invention which may or may not be packaged together. The

components of the kit may be comprised by separate housings (i.e. as a kit of separate parts), or two or more components may be provided in a single housing. Moreover, it is to be understood that the kit of the present invention, in an embodiment, is to be used for practicing the methods referred to herein above. It is, in an embodiment, envisaged that components are provided in a ready-to-use manner for practicing the methods referred to above. In an embodiment, all or some of the chemical compounds of the kit are provided in dried, such as in lyophilized form, wherein the component is reconstituted using a liquid such as water or an aqueous buffered solution. In an embodiment, all or some of said compounds are provided in concentrated liquid form wherein the concentrated component is diluted using a liquid such as an aqueous buffered solution. Further, the kit, in an embodiment, contains instructions for carrying out said methods and, if applicable, said reconstitution of dried reagents. The instructions can be provided by a user's manual in paper- or electronic form. In addition, the manual may comprise instructions for interpreting the results obtained when carrying out the aforementioned methods using the kit of the present invention. In an embodiment, the kit further comprises water, a buffer, and/or a calibration reagent.

The term "detection agent", as used herein, relates to a chemical molecule binding, directly or indirectly, to the sample constituent of the present invention as specified herein above. In an embodiment, the detection agent is not bound to a solid surface and not adapted to be bound to a solid surface. As will be understood by the skilled person, the detection agent may also be an indirect detector compound, i.e. a detection agent not contacting the sample constituent directly, but by means of a further compound which itself binds to the sample constituent. In an embodiment, the detection agent is a direct detection agent, i.e. is an agent directly binding to the sample constituent. Preferably, the detection agent is an antibody, more preferably is an IgG. Preferably, the detection agent is a monoclonal antibody. Also preferably, the detection agent is an antibody, more preferably a monoclonal antibody bound to a bead, such as a latex bead. Preferably, the detection agent is an agent comprising at least two diagnostic epitopes, i.e. epitopes binding to a sample constituent. In a further embodiment, said diagnostic epitopes are identical, such as, e.g. in an IgG.

In view of the above, the following embodiments are particularly envisaged:

Embodiment 1. A composition comprising at least one spike peptide species covering at least 60% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 60% identical thereto.

- 5 Embodiment 2. The composition of embodiment 1, wherein the at least one spike peptide species has a length of at least five amino acids, preferably at least ten amino acids, more preferably at least 20 amino acids, most preferably at least 25 amino acids.

- 10 Embodiment 3. The composition of embodiment 1 or 2, wherein the at least one spike peptide species has a length of from 10 to 500 amino acids, preferably of from 15 to 400 amino acids, more preferably of from 20 to 300 amino acids, still more preferably of from 25 to 200 amino acids, still more preferably of from 27 to 100 amino acids, even more preferably of from 28 to 50 amino acids, most preferably of from 29 to 40 amino acids.

- 15 Embodiment 4. The composition of any one of embodiments 1 to 3, wherein the at least one spike peptide species has a length of about 30 amino acids.

- Embodiment 5. The composition of any one of embodiments 1 to 4, wherein the at least one spike peptide species covers at least 60% of the amino acid sequence of SEQ ID NO:1 or
20 an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto.

- Embodiment 6. The composition of any one of embodiments 1 to 5, wherein the at least one spike peptide species covers at least 70% of the amino acid sequence of SEQ ID NO:1 or
25 an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto.

- Embodiment 7. The composition of any one of embodiments 1 to 6, wherein the at least one spike peptide species covers at least 80% of the amino acid sequence of SEQ ID NO:1 or
30 an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto.

Embodiment 8. The composition of any one of embodiments 1 to 7, wherein the at least one spike peptide species covers at least 90% of the amino acid sequence of SEQ ID NO:1 or

an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto.

Embodiment 9. The composition of any one of embodiments 1 to 8, wherein the at least one spike peptide species covers at least 94% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto.

Embodiment 10. The composition of any one of embodiments 1 to 9, wherein the at least one spike peptide species covers at least 96% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto.

Embodiment 11. The composition of any one of embodiments 1 to 10, wherein said at least one spike peptide comprises, preferably consists of, the amino acid sequence of SEQ ID NO:1 or 2.

Embodiment 12. The composition of any one of embodiments 1 to 11, wherein said at least one spike peptide species is one spike peptide species.

Embodiment 13. The composition of any one of embodiments 1 to 12, wherein the composition is a diagnostic composition and/or is a pharmaceutical composition.

Embodiment 14. A polynucleotide encoding the at least one spike peptide species as specified in any one of embodiments 1 to 12.

Embodiment 15. A host cell comprising the at least one spike peptide species as specified in any one of embodiments 1 to 12 and/or the polynucleotide of embodiment 14.

Embodiment 16. The composition according to any one of embodiments 1 to 13, the polynucleotide according to embodiment 14, or the host cell according to embodiment 15 for use in diagnosis and/or medicine.

Embodiment 17. The composition according to any one of embodiments 1 to 13 , the polynucleotide according to embodiment 14 , or the host cell according to embodiment 15 for use in diagnosis of coronavirus infection, preferably of infection with coronavirus SARS-CoV-2.

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Embodiment 18. The composition according to any one of embodiments 1 to 13 , the polynucleotide according to embodiment 14 , or the host cell according to embodiment 15 for use in preventing and/or treating coronavirus infection, preferably infection with coronavirus SARS-CoV-2.

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Embodiment 19. A method of detecting a coronavirus infection in a subject comprising

a) contacting a sample of said subject with a composition according to any one of embodiment 1 to 13,

b) determining binding of sample constituents to the at least one spike peptide comprised in said composition, and

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c) based on determining step b), detecting a coronavirus infection.

Embodiment 20. The method of embodiment 19, wherein said sample is a sample of a bodily fluid, preferably a blood, serum, or plasma sample.

20

Embodiment 21. The method of embodiment 19 or 20, wherein said sample constituents are antibodies, preferably IgG and/or IgM.

Embodiment 22. A method of manufacturing a coronavirus diagnostic, comprising chemically synthesizing the spike peptide or spike peptides of a composition according to any one of embodiments 1 to 13 and formulating said spike peptide or spike peptides as a diagnostic composition.

25

Embodiment 23. A kit comprising a composition according to any one of embodiments 1 to 13 and a detection agent.

30

All references cited in this specification are herewith incorporated by reference with respect to their entire disclosure content and the disclosure content specifically mentioned in this specification.

Claims

1. A composition comprising at least one spike peptide species covering at least 60% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 60% identical thereto.
5
2. The composition of claim 1, wherein the at least one spike peptide species has a length of about 30 amino acids.
- 10 3. The composition of claim 1 or 2, wherein the at least one spike peptide species covers at least 60% of the amino acid sequence of SEQ ID NO:1 or an amino acid sequence at least 70%, preferably at least 80%, more preferably at least 90%, still more preferably at least 93%, most preferably at least 96%, identical thereto.
- 15 4. The composition of any one of claims 1 to 3, wherein said at least one spike peptide comprises, preferably consists of, the amino acid sequence of SEQ ID NO:1 or 2.
5. The composition of any one of claims 1 to 4, wherein said at least one spike peptide species is one spike peptide species.
20
6. The composition of any one of claims 1 to 5, wherein the composition is a diagnostic composition and/or is a pharmaceutical composition.
7. A polynucleotide encoding the at least one spike peptide species as specified in any one of claims 1 to 6.
25
8. A host cell comprising the at least one spike peptide species as specified in any one of claims 1 to 5 and/or the polynucleotide of claim 7.
- 30 9. The composition according to any one of claims 1 to 6, the polynucleotide according to claim 7, or the host cell according to claim 8 for use in diagnosis and/or medicine.
10. The composition according to any one of claims 1 to 6, the polynucleotide according to claim 7, or the host cell according to claim 8 for use in diagnosis of coronavirus

infection, preferably of infection with coronavirus SARS-CoV-2.

11. The composition according to any one of claims 1 to 6, the polynucleotide according
to claim 7, or the host cell according to claim 8 for use in preventing and/or treating
5 coronavirus infection, preferably infection with coronavirus SARS-CoV-2.
12. A method of detecting a coronavirus infection in a subject comprising
 - a) contacting a sample of said subject with a composition according to any one of
claim 1 to 6,
 - 10 b) determining binding of sample constituents to the at least one spike peptide
comprised in said composition, and
 - c) based on determining step b), detecting a coronavirus infection.
13. The method of claim 12, wherein said sample is a sample of a bodily fluid, preferably
15 a blood, plasma, or serum sample.
14. A method of manufacturing a coronavirus diagnostic, comprising chemically
synthesizing the spike peptide or spike peptides of a composition according to any one
of claims 1 to 6 and formulating said spike peptide or spike peptides as a diagnostic
20 composition.
15. A kit comprising a composition according to any one of claims 1 to 6 and a detection
agent.

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2021/057085

A. CLASSIFICATION OF SUBJECT MATTER
INV. G01N33/569
ADD.

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

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
G01N C07K C12N

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

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

EPO-Internal, Sequence Search, BIOSIS, CHEM ABS Data, EMBASE, WPI Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	TAI WANBO ET AL: "Characterization of the receptor-binding domain (RBD) of 2019 novel coronavirus: implication for development of RBD protein as a viral attachment inhibitor and vaccine", CELLULAR & MOLECULAR IMMUNOLOGY, CHINESE SOCIETY OF IMMUNOLOGY, CH, vol. 17, no. 6, 19 March 2020 (2020-03-19), pages 613-620, XP037153214, ISSN: 1672-7681, DOI: 10.1038/S41423-020-0400-4 [retrieved on 2020-03-19] abstract figure 1 ----- -/--	1-15



Further documents are listed in the continuation of Box C.



See patent family annex.

* Special categories of cited documents :

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

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

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

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

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

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

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

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

"&" document member of the same patent family

Date of the actual completion of the international search

28 June 2021

Date of mailing of the international search report

09/07/2021

Name and mailing address of the ISA/

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Authorized officer

Lewis, Birgit

INTERNATIONAL SEARCH REPORT

International application No
PCT/EP2021/057085

C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	DATABASE EMBL [Online] 3 March 2020 (2020-03-03), "Severe acute respiratory syndrome coronavirus 2 isolate SARS-CoV-2/human/CHN/IQTC04/2020, complete genome.", XP002803458, retrieved from EBI accession no. EM STD:MT123292 Database accession no. MT123292 nt. 23000-23089; sequence -----	7
X	DATABASE EMBL [Online] 11 March 2020 (2020-03-11), "Severe acute respiratory syndrome coronavirus 2 isolate SARS-CoV-2/human/CHN/SH01/2020, complete genome.", XP002803459, retrieved from EBI accession no. EM STD:MT121215 Database accession no. MT121215 nt. 23000-23089; sequence -----	7
A	WO 2005/118813 A2 (PASTEUR INSTITUT [FR]; HONG KONG PASTEUR RES CT LTD [CN] ET AL.) 15 December 2005 (2005-12-15) claims 1-3,13,14,32-37; sequence 7 -----	1-15
X,P	EP 3 715 847 A1 (EUROIMMUN MEDIZINISCHE LABORDIAGNOSTIKA AG [DE] ET AL.) 30 September 2020 (2020-09-30) page 6 - page 8; sequences 1-4 paragraph [0054] - paragraph [0064] -----	1-15
X,P	LAN JUN ET AL: "Structure of the SARS-CoV-2 spike receptor-binding domain bound to the ACE2 receptor", NATURE, MACMILLAN JOURNALS LTD., ETC, LONDON, vol. 581, no. 7807, 30 March 2020 (2020-03-30), pages 215-220, XP037182122, ISSN: 0028-0836, DOI: 10.1038/S41586-020-2180-5 [retrieved on 2020-03-30] abstract figures 1-4 -----	1-5

INTERNATIONAL SEARCH REPORT

International application No.

PCT/EP2021/057085

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

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing:
- a. ☒ forming part of the international application as filed:
- ☒ in the form of an Annex C/ST.25 text file.
- ☐ on paper or in the form of an image file.
- b. ☐ furnished together with the international application under PCT Rule 13~~ter~~.1(a) for the purposes of international search only in the form of an Annex C/ST.25 text file.
- c. ☐ furnished subsequent to the international filing date for the purposes of international search only:
- ☐ in the form of an Annex C/ST.25 text file (Rule 13~~ter~~.1(a)).
- ☐ on paper or in the form of an image file (Rule 13~~ter~~.1(b) and Administrative Instructions, Section 713).
2. ☐ In addition, in the case that more than one version or copy of a sequence listing has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that forming part of the application as filed or does not go beyond the application as filed, as appropriate, were furnished.
3. Additional comments:

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/EP2021/057085

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 2005118813 A2	15-12-2005	CA 2568838 A1	15-12-2005
		EP 1751178 A2	14-02-2007
		WO 2005118813 A2	15-12-2005

EP 3715847 A1	30-09-2020	EP 3715847 A1	30-09-2020
		ES 2788459 T1	21-10-2020
