

Claims :

1. A lateral flow assay device (100) comprising:

porous membrane (20) mounted on a solid support (10); said porous membrane having a sample pad (14) for receiving a liquid sample (12) comprising a target analyte at a first end and an absorbent pad (28) at the second end, said solid support permits capillary flow of the liquid sample comprising the target analyte from the sample pad (14) to the absorbent pad (28);

characterized in that:

 - a. the porous membrane (20) comprises a conjugate pad (16) comprising gold nanoparticle sensor conjugate (18), said conjugate comprises gold nanoparticles having a particle size of 10nm to 20nm conjugated with a peptide that specifically binds with a protein in the target analyte or said conjugate comprises gold nanoparticles having a particle size of 10nm to 20nm conjugated with an antibody against the protein in the target analyte;
 - b. a test region (22) comprising an immobilized capture molecule (24); said capture molecule is a peptide capable of specifically binding to the protein in the target analyte or an antibody against protein in the target analyte; and
 - c. optionally a control region (26) comprising the protein in the target analyte immobilized on the porous membrane (20).
2. The lateral flow assay device (100) as claimed in claim 1, wherein the gold nanoparticle is conjugated with the peptide capable of specifically binding with the protein in the liquid sample (12) and the capture molecule comprises the immobilized antibody against the protein in the liquid sample (12).
3. The lateral flow assay device (100) as claimed in claim 1, wherein the gold nanoparticle is conjugated with the antibody against the protein in the liquid sample (12) and the capture molecule comprises the peptide capable of specifically binding with the protein in the liquid sample (12).
4. The lateral flow assay device (100) as claimed in claim 1, wherein the gold nanoparticle is conjugated with the peptide capable of specifically binding with a

spike protein or a protein of the target analyte or with the antibody against the nucleocapsid protein of the target analyte.

5 5. The lateral flow assay device (100) as claimed in claims 4, wherein the gold nanoparticles comprise 30µg-50µg of the peptide or 0.5µg-1µg of the antibody.

10 6. The lateral flow assay device(100) as claimed in claim 1, wherein the capture molecule (24) is the peptide capable of specifically binding with a spike protein or a protein of the target analyte or the antibody against the nucleocapsid protein of the target analyte.

15 7. The lateral flow assay device (100) as claimed in claim 1 to 6, wherein the capture molecule (24) comprises 0.75-1 µg of the peptide or 0.75-1µg of the antibody.

8. The lateral flow assay device (100) as claimed in claim 1, wherein the control region (26) comprises 0.5µg to 1µg of the spike protein or the nucleocapsid protein of the target analyte.

20 9. The lateral flow assay device(100) as claimed in claims 1 to 8, wherein the target analyte is an enveloped virus selected from SARS CoV1, SARS CoV2, MERS CoV, influenza virus, Hepatitis B and C, and Ebola virus.

25 10. The lateral flow assay device (100) as claimed in claim 1 comprising:
a porous membrane (20) mounted on a solid support (10); said porous membrane having a sample pad (14) for receiving a liquid sample (12) comprising a SARS CoV2 virus at a first end and an absorbent pad (28) at the second end, said solid support permits capillary flow of the liquid sample comprising the target analyte from the sample pad (14) to the absorbent pad (28);
30

characterized in that:

- a. the porous membrane (20) comprises a conjugate pad (16) comprising gold nanoparticle sensor conjugate (18), said conjugate comprises gold nanoparticles having a particle size of 10nm to 20nm conjugated with a

- peptide capable of binding with the S1 spike protein of SARS CoV2 and has a SEQ ID NO: 1 or said conjugate comprises gold nanoparticles having a particle size of 10nm to 20nm conjugated with an anti-S1mAB or anti-NmAB of the SARS CoV2;
- 5 b. a test region (22) comprising an immobilized capture molecule (24); said capture molecule is a peptide capable of binding with the S1 spike protein of SARS CoV2 and has a SEQ ID NO: 1 or is an anti-S1mAB or anti-NmAB of the SARS CoV2; and
- 10 c. optionally a control region (26) comprising the S1 spike protein in the SARS CoV 2 virus immobilized on the porous membrane (20).
11. A lateral flow assay method for detecting a target analyte in a sample comprising:
- 15 a. applying a sample (12) containing the target analyte on the sample pad (14) of the device (100) as claimed in claims 1 to 10;
- b. allowing the sample to flow from the sample pad (14) to the test region (22) through the conjugate pad(16); and
- c. detecting the presence or absence of the target analyte in the test region (22) with a change in colour from red to purple in about 60 seconds to 300 seconds.
- 20
12. The method as claimed in claim 11, wherein further comprises allowing the sample to flow further to the control region (26); observing the colour change from red to purple in the control region in about 60 seconds to about 300 seconds; and confirming the presence or absence of the target analyte in the test region (22).
- 25
13. The method as claimed in claims 11 to 12, wherein the sample (12) is oral swab, nasal swab, sputum or saliva.
14. The method as claimed in claims 11 to 13, wherein the sample (12) is diluted in a
- 30 buffer selected from phosphate buffered saline.
15. The method as claimed in claims 11 to 14, wherein the target analyte is an enveloped virus selected from SARS CoV 1, SARS CoV2, MERS CoV, influenza virus, Hepatitis B and C, and Ebola virus.

16. The method for detecting analyte in a sample as claimed in claim 11, wherein said analyte is a SARS CoV-2 , said method comprising:
- a. applying a sample (12) containing the target analyte on the sample pad (14) of the device (100) as claimed in claim 10;
 - b. allowing the sample to flow from the sample pad(14) to the test region(22) through the conjugate pad(16); and
 - c. detecting the presence of the SARS CoV2 virus in the test region with a change in colour from red to purple in about 60 seconds to about 300 seconds.
17. The method as claimed in claim 16, further comprises allowing the sample to flow further to the control region(26); observing the colour change from red to purple in the control region in about 60 seconds to about 300 seconds; and confirming the presence of the SARS CoV 2 virus in the test region.
18. The method as claimed in claims 16 to 17, wherein detecting the presence of the SARS CoV 2 virus in the test region with a change in colour from red to purple in about 60 seconds to about 180 seconds.
19. The method as claimed in claim 16, wherein the method detects SARS CoV2 virus up to 192TCID50.
20. The method as claimed in claims 16 to 19, wherein the detection has 90%-92% sensitivity and 98%-100% specificity for SARS CoV2 virus.
21. A kit for detecting SARS CoV2 virus in a sample comprising:
- a. a lateral flow assay device(100) as claimed in claim 16 and
 - b. dilution buffer selected from phosphate buffer saline.
22. A method for quantitatively detecting a target analyte in a sample comprising the step of:
- a. measuring absorbance of gold nanoparticles having particle size of 10nm-20nm conjugated with a peptide that specifically binds with a protein in the target analyte or of said gold nanoparticle having particle size of 10nm-20nm

conjugated with an antibody against the protein in the target analyte at 525 nm;

b. mixing 50-200µl of the sample with 50-100 µl said gold nanoparticles, observing the change in colour from red to purple; and measuring the absorbance at 700nm; and

c. calculating the absorbance ratio of the 525nm and the 700nm;

wherein the absorbance ratio of 525nm to 700nm is inversely proportional to the amount of target analyte in the sample.

23. The method as claimed in claim 22, wherein the target analyte is an enveloped virus selected from SARS CoV 1, SARS CoV2, MERS CoV, influenza virus, Hepatitis B and C, and Ebola virus.

24. A method for quantitatively detecting a target analyte in a sample, wherein said analyte is SARS CoV2 virus comprising the step of:

a. measuring absorbance of gold nanoparticles having particle size of 10nm-20nm conjugated with a peptide is capable of binding with the S1 spike protein of SARS CoV2 and has a SEQ ID NO: 1 or of said gold nanoparticle having particle size of 10nm-20nm conjugated with an anti-S1mAB or anti-NmAB of the SARS CoV2 at 525 nm;

b. mixing 50-200µl of the sample with 50-100 µl of said gold nanoparticles, observing the change in colour from red to purple; and measuring the absorbance at 700nm; and

c. calculating the absorbance ratio of the 525nm and the 700nm;

wherein the absorbance ratio of 525nm to 700nm is inversely proportional to the amount of SARS CoV 2 in the sample.

25. The method as claimed in claim 24, wherein the absorbance ratio of 525nm to 700nm is 1.4 to 7.6.

26. A conjugate comprising 10nm to 20nm gold nanoparticle and a peptide is capable of binding with the S1 spike protein of SARS CoV2 and has a SEQ ID NO: 1.

27. A method of diagnosing COVID-19 in a patient sample comprising:

- a. Diluting the sample in a buffer;
- b. Applying the diluted sample to the lateral flow device claimed in claim 16;
- c. Change in colour from red to purple from about 60 seconds to about 300 seconds indicates the presence of SARS CoV2 virus in the sample.

5

28. The method as claimed in claim 27, wherein the change in colour from red to purple is within 60 to 180 seconds.

29. The method as claimed in claim 28, wherein the sample is from symptomatic or asymptomatic patient.

10

30. The method as claimed in claim 27-29, wherein the sample is oral swab, nasal swab, sputum or saliva.

15