SUMMARY

【0007】 Therefore it is an object of the present invention to provide monoclonal antibody that binds specifically to dengue virus NS1 antigen. It can be effectively employed to detect dengue virus infection in a subject, such as a human. Such assays can be employed to detect the presence of dengue virus infection using biological samples including blood, serum, plasma, saliva, cerebrospinal fluid.(Monique da Rocha Queiroz Lima et al., PLOSNTDS, v5 (5), e1147, 2011).In one example, the disclosed assays employ a first antibody specific for dengue NS1 as a capture antibody, with a second antibody specific for dengue NS1 in monomeric and/or oligomeric (primarily dimeric) forms being employed as a detection antibody.

【0008】 Another object of the present invention is to provide an antibody composition having at least one monoclonal antibody that binds specifically to dengue antigen for detection of dengue NS1 antigen. Thus a monoclonal antibody of the antibody composition can be mass-produced and applied to detect dengue pathogens.

【0009】 In order to achieve the above objects, an antibody composition of the present invention includes at least one monoclonal antibody. In one aspect, there is provided an antibody composition comprising a first monoclonal antibody, a second monoclonal antibody, a third monoclonal antibody, and a fourth monoclonal antibody; wherein the first monoclonal antibody includes heavy chain complementarity determining region 1(HCDR1) of sequence ID number (SEQ ID NO):26, heavy chain complementarity determining region 2(HCDR2) of SEQ ID NO:28, heavy chain complementarity determining region 3(HCDR3) of SEQ ID NO:30, light chain complementarity determining region 1(LCDR1) of SEQ ID NO:32, light chaincomplementarity determining region 2(LCDR2) of SEQ ID NO:34, light chain complementarity determining region

3(LCDR3) of SEQ ID NO:36; the second monoclonal antibody includes HCDR1 of SEQ ID NO:62, HCDR2 of SEQ ID NO:64, HCDR3 of SEQ ID NO:66, LCDR1 of SEQ ID NO:68, LCDR2 of SEQ ID NO:70, LCDR3 of SEQ ID NO:72; the third monoclonal antibody includes HCDR1 of SEQ ID NO:74, HCDR2 of SEQ ID NO:76, HCDR3 of SEQ ID NO:78, LCDR1 of SEQ ID NO:80, LCDR2 of SEQ ID NO:82, LCDR3 of SEQ ID NO:84; the fourth monoclonal antibody includes HCDR1 of SEQ ID NO:50, HCDR2 of SEQ ID NO:52, HCRD3 of SEQ ID NO:54, LCDR1 of SEQ ID NO:56, LCDR2 of SEQ ID NO:58, LCDR3 of SEQ ID NO:60; . The monoclonal antibody specifically binds to a

At last produce a monoclonal antibody by the hybridoma cell. The dengue fever can be type 1 dengue fever, type 2 dengue fever, type 3 dengue fever, or type 4 dengue fever. The monoclonal antibody produced can be used to detect the dengue fever.

In another aspect, there is also provided a detection method for detecting dengue virus including the steps of: coating at least one capture monoclonal antibody at the bottom of plurality of sample wells, wherein the at least one capture monoclonal antibody comprises an antibody which includes HCDR1 of SEQ ID NO:38, HCDR2 of SEQ ID NO:40, HCDR3 of SEQ ID NO:42, LCDR1 of SEQ ID NO:44, LCDR2 of SEQ ID NO:46, LCDR3 of SEQ ID NO:48;

adding a sample, a positive control or a negative control in the sample wells respectively and then a dengue antigen in the samples and the positive control bind to the capture monoclonal antibody at the bottom of the sample wells;

adding at least one labeled monoclonal antibody in the sample wells respectively and the labeled monoclonal antibody bind to the dengue antigens captured by the capture monoclonal antibodies, wherein the labeled monoclonal antibody are an antibody which includes HCDR1 of SEQ ID NO:50, HCDR2 of SEQ ID NO:52, HCRD3 of SEQ ID NO:54, LCDR1 of SEQ ID NO:56, LCDR2 of SEQ ID NO:58, LCDR3 of SEQ ID NO:60, an antibody which includes HCDR1 of SEQ ID NO:26, HCDR2 of SEQ ID NO:28, HCDR3 of SEQ ID NO:30, LCDR1 of SEQ ID NO:32,

LCDR2 of SEQ ID NO:34, LCDR3 of SEQ ID NO:36, an antibody which includes HCDR1 of SEQ ID NO:62, HCDR2 of SEQ ID NO:64, HCDR3 of SEQ ID NO:66, LCDR1 of SEQ ID NO:68, LCDR2 of SEQ ID NO:70, LCDR3 of SEQ ID NO:72 or an antibody which includes HCDR1 of SEQ ID NO:74, HCDR2 of SEQ ID NO:76, HCDR3 of SEQ ID NO:78, LCDR1 of SEQ ID NO:80, LCDR2 of SEQ ID NO:82, LCDR3 of SEQ ID NO:84; and

detecting the absorbance of the samples;

wherein the detection method is used to detect type 1 dengue antigens, type 2 dengue antigens, type 3 dengue antigens or type 4 dengue antigens.

In another aspect, there is provided a kit comprising a first monoclonal antibody, a second monoclonal antibody, a third monoclonal antibody, a fourth monoclonal antibody, a fifth monoclonal antibody, and a sixth monoclonal antibody, when used according to the method of claim 2 or 3, wherein the first monoclonal antibody includes HCDR1 of sequence ID number (SEQ ID NO):26, HCDR2 of SEQ ID NO:28, HCDR3 of SEQ ID NO:30, LCDR1 of SEQ ID NO:32, LCDR2 of SEQ ID NO:34, LCDR3 of SEQ ID NO:36; the second monoclonal antibody includes HCDR1 of SEQ ID NO:62, HCDR2 of SEQ ID NO:64, HCDR3 of SEQ ID NO:66, LCDR1 of SEQ ID NO:68, LCDR2 of SEQ ID NO:70, LCDR3 of SEQ ID NO:72; the third monoclonal antibody includes HCDR1 of SEQ ID NO:74, HCDR2 of SEQ ID NO:76, HCDR3 of SEQ ID NO:78, LCDR1 of SEQ ID NO:80, LCDR2 of SEQ ID NO:82, LCDR3 of SEQ ID NO:84; the fourth monoclonal antibody includes HCDR1 of SEQ ID NO:50, HCDR2 of SEQ ID NO:52, HCRD3 of SEQ ID NO:54, LCDR1 of SEQ ID NO:56, LCDR2 of SEQ ID NO:58, LCDR3 of SEQ ID NO:60; the fifth monoclonal antibody includes HCDR1 of SEQ ID NO:2, HCDR2 of SEQ ID NO:4, HCDR3 of SEQ ID NO:6, LCDR1 of SEQ ID NO:8, LCDR2

of SEQ ID NO:10, LCDR3 of SEQ ID NO:12; and the sixth monoclonal antibody specifically includes HCDR1 of SEQ ID NO:38, HCDR2 of SEQ ID NO:40, HCDR3 of SEQ ID NO:42, LCDR1 of SEQ ID NO:44, LCDR2 of SEQ ID NO:46, LCDR3 of SEQ ID NO:48.

In a further aspect, there is provided a kit comprising a first monoclonal antibody, a second monoclonal antibody, a third monoclonal antibody and a fourth monoclonal antibody, when used according to the method of claim 2 or 3, wherein the first monoclonal antibody includes HCDR1 of sequence ID number (SEQ ID NO):14, HCDR2 of SEQ ID NO:16, HCDR3 of SEQ ID NO:18, LCDR1 of SEQ ID NO:20, LCDR2 of SEQ ID NO:22, LCDR3 of SEQ ID NO:24; the second monoclonal antibody includes HCDR1 of SEQ ID NO:50, HCDR2 of SEQ ID NO:52, HCRD3 of SEQ ID NO:54, LCDR1 of SEQ ID NO:56, LCDR2 of SEQ ID NO:58, LCDR3 of SEQ ID NO:60; the third monoclonal antibody includes HCDR1 of SEQ ID NO:2, HCDR2 of SEQ ID NO:4, HCDR3 of SEQ ID NO:6, LCDR1 of SEQ ID NO:8, LCDR2 of SEQ ID NO:10, LCDR3 of SEQ ID NO:12; and the fourth monoclonal antibody specifically includes HCDR1 of SEQ ID NO:38, HCDR2 of SEQ ID NO:40, HCDR3 of SEQ ID NO:42, LCDR1 of SEQ ID NO:44, LCDR2 of SEQ ID NO:46, LCDR3 of SEQ ID NO:48.

【0011】 The type 1 dengue fever, type 2 dengue fever, type 3 dengue fever, and type 4 dengue fever mentioned above represents each of four different serotypes of the dengue virus-being infected by dengue virus type 1, dengue virus type 2, dengue virus type 3, and dengue virus type 4 respectively, and each of which is able to be detected by at least one monoclonal antibody.

【0012】 The dengue antigen can be selected from the following: a virus particle of dengue virus type 1, a virus particle of dengue virus type 2, a virus particle of dengue virus type 3, a virus particle of dengue virus type 4, a dengue virus type 1 NS1 antigen, a dengue virus type 2 NS1

antigen, a dengue virus type 3 NS1 antigen, a dengue virus type 4 NS1 antigen, recombinant dengue virus type 1 NS1 protein, recombinant dengue virus type 2 NS1 protein, recombinant dengue virus type 3 NS1 protein, and recombinant dengue virus type 4 NS1 protein.

BRIEF DESCRIPTION OF THE DRAWINGS

【0013】 The structure and the technical means adopted by the present invention to achieve the above and other objects can be best understood by referring to the following detailed description of the preferred embodiments and the accompanying drawings, wherein:

Fig. 1 is a flow chart showing steps of an embodiment according to the present invention;

Fig. 2 shows test results of a dengue fever enzyme-linked immunoassay kit produced according to the present invention;

Fig. 3 shows an immunochromatography kit produced by an

Claims

What is claimed is:

1. An antibody composition comprising a first monoclonal antibody, a second monoclonal antibody, a third monoclonal antibody, and a fourth monoclonal antibody; wherein the first monoclonal antibody includes heavy chain complementarity determining region 1(HCDR1) of sequence ID number (SEQ ID NO):26, heavy chain complementarity determining region 2(HCDR2) of SEQ ID NO:28, heavy chain complementarity determining region 3(HCDR3) of SEQ ID NO:30, light chain complementarity determining region 1(LCDR1) of SEQ ID NO:32, light chain complementarity determining region 2(LCDR2) of SEQ ID NO:34, light chain complementarity determining region 3(LCDR3) of SEQ ID NO:36; the second monoclonal antibody includes HCDR1 of SEQ ID NO:62, HCDR2 of SEQ ID NO:64, HCDR3 of SEQ ID NO:66, LCDR1 of SEQ ID NO:68, LCDR2 of SEQ ID NO:70, LCDR3 of SEQ ID NO:72; the third monoclonal antibody includes HCDR1 of SEQ ID NO:74, HCDR2 of SEQ ID NO:76, HCDR3 of SEQ ID NO:78, LCDR1 of SEQ ID NO:80, LCDR2 of SEQ ID NO:82, LCDR3 of SEQ ID NO:84; the fourth monoclonal antibody includes HCDR1 of SEQ ID NO:50, HCDR2 of SEQ ID NO:52, HCRD3 of SEQ ID NO:54, LCDR1 of SEQ ID NO:56, LCDR2 of SEQ ID NO:58, LCDR3 of SEQ ID NO:60.

2. A detection method for detecting dengue virus comprising the steps of:

coating at least one capture monoclonal antibody

at the bottom of plurality of sample wells, wherein the at least one capture monoclonal antibody comprises an antibody which includes HCDR1 of SEQ ID NO:38, HCDR2 of SEQ ID NO:40, HCDR3 of SEQ ID NO:42, LCDR1 of SEQ ID NO:44, LCDR2 of SEQ ID NO:46, LCDR3 of SEQ ID NO:48;

adding a sample, a positive control or a negative control in the sample wells respectively and then a dengue antigen in the samples and the positive control bind to the capture monoclonal antibody at the bottom of the sample wells;

adding at least one labeled monoclonal antibody in the sample wells respectively and the labeled monoclonal antibody bind to the dengue antigens captured by the capture monoclonal antibodies, wherein the labeled monoclonal antibody are an antibody which includes HCDR1 of SEQ ID NO:50, HCDR2 of SEQ ID NO:52, HCRD3 of SEQ ID NO:54, LCDR1 of SEQ ID NO:56, LCDR2 of SEQ ID NO:58, LCDR3 of SEQ ID NO:60, an antibody which includes HCDR1 of SEQ ID NO:26, HCDR2 of SEQ ID NO:28, HCDR3 of SEQ ID NO:30, LCDR1 of SEQ ID NO:32, LCDR2 of SEQ ID NO:34, LCDR3 of SEQ ID NO:36, an antibody which includes HCDR1 of SEQ ID NO:62, HCDR2 of SEQ ID NO:64, HCDR3 of SEQ ID NO:66, LCDR1 of SEQ ID NO:68, LCDR2 of SEQ ID NO:70, LCDR3 of SEQ ID NO:72 or an antibody which includes HCDR1 of SEQ ID NO:74, HCDR2 of SEQ ID NO:76, HCDR3 of SEQ ID NO:78, LCDR1 of SEQ ID NO:80, LCDR2 of SEQ ID NO:82, LCDR3 of SEQ ID NO:84; and

detecting the absorbance of the samples;

wherein the detection method is used to detect

type 1 dengue antigens, type 2 dengue antigens, type 3 dengue antigens or type 4 dengue antigens.

3. The method of claim wherein the at least one capture monoclonal antibody further comprises an antibody which includes HCDR1 of SEQ ID NO:2, HCDR2 of SEQ ID NO:4, HCDR3 of SEQ ID NO:6, LCDR1 of SEQ ID NO:8, LCDR2 of SEQ ID NO:10, LCDR3 of SEQ ID NO:12.

4. A kit comprising a first monoclonal antibody, a second monoclonal antibody, a third monoclonal antibody, a fourth monoclonal antibody, a fifth monoclonal antibody, and a sixth monoclonal antibody, when used according to the method of claim 2 or 3, wherein the first monoclonal antibody includes HCDR1 of sequence ID number (SEQ ID NO):26, HCDR2 of SEQ ID NO:28, HCDR3 of SEQ ID NO:30, LCDR1 of SEQ ID NO:32, LCDR2 of SEQ ID NO:34, LCDR3 of SEQ ID NO:36; the second monoclonal antibody includes HCDR1 of SEQ ID NO:62, HCDR2 of SEQ ID NO:64, HCDR3 of SEQ ID NO:66, LCDR1 of SEQ ID NO:68, LCDR2 of SEQ ID NO:70, LCDR3 of SEQ ID NO:72; the third monoclonal antibody includes HCDR1 of SEQ ID NO:74, HCDR2 of SEQ ID NO:76, HCDR3 of SEQ ID NO:78, LCDR1 of SEQ ID NO:80, LCDR2 of SEQ ID NO:82, LCDR3 of SEQ ID NO:84; the fourth monoclonal antibody includes HCDR1 of SEQ ID NO:50, HCDR2 of SEQ ID NO:52, HCRD3 of SEQ ID NO:54, LCDR1 of SEQ ID NO:56, LCDR2 of SEQ ID NO:58, LCDR3 of SEQ ID NO:60; the fifth monoclonal antibody includes HCDR1 of SEQ ID NO:2, HCDR2 of SEQ ID NO:4, HCDR3 of SEQ ID NO:6, LCDR1 of SEQ

ID NO:8, LCDR2 of SEQ ID NO:10, LCDR3 of SEQ ID NO:12; and the sixth monoclonal antibody specifically includes HCDR1 of SEQ ID NO:38, HCDR2 of SEQ ID NO:40, HCDR3 of SEQ ID NO:42, LCDR1 of SEQ ID NO:44, LCDR2 of SEQ ID NO:46, LCDR3 of SEQ ID NO:48.

5. A kit comprising a first monoclonal antibody, a second monoclonal antibody, a third monoclonal antibody and a fourth monoclonal antibody, when used according to the method of claim 2 or 3, wherein the first monoclonal antibody includes HCDR1 of sequence ID number (SEQ ID NO):14, HCDR2 of SEQ ID NO:16, HCDR3 of SEQ ID NO:18, LCDR1 of SEQ ID NO:20, LCDR2 of SEQ ID NO:22, LCDR3 of SEQ ID NO:24; the second monoclonal antibody includes HCDR1 of SEQ ID NO:50, HCDR2 of SEQ ID NO:52, HCRD3 of SEQ ID NO:54, LCDR1 of SEQ ID NO:56, LCDR2 of SEQ ID NO:58, LCDR3 of SEQ ID NO:60; the third monoclonal antibody includes HCDR1 of SEQ ID NO:2, HCDR2 of SEQ ID NO:4, HCDR3 of SEQ ID NO:6, LCDR1 of SEQ ID NO:8, LCDR2 of SEQ ID NO:10, LCDR3 of SEQ ID NO:12; and the fourth monoclonal antibody specifically includes HCDR1 of SEQ ID NO:38, HCDR2 of SEQ ID NO:40, HCDR3 of SEQ ID NO:42, LCDR1 of SEQ ID NO:44, LCDR2 of SEQ ID NO:46, LCDR3 of SEQ ID NO:48.