

CLAIMS

We Claim:

1. A method of preparing a diagnostic kit for ~~rapidly~~ diagnosing a dengue virus in 15 minutes or less, the method comprising:

~~designing~~ providing a first oligonucleotide comprising a sequence selected from the group consisting of sequences 1 as shown in Table 1 that is complementary to a target sequence of the dengue virus as a detection probe;

hybridising a plurality of the first oligonucleotide to a plurality of carbon nanotubes;

providing a second oligonucleotide comprising a sequence selected from the group consisting of sequences 2 as shown in Table 1 that is complimentary to the target sequence of the dengue virus for use as a capture probe; and

immobilizing a plurality of the second oligonucleotide to a plurality of solid supports.

~~providing a plurality of carbon nanotubes; and~~

~~providing a plurality of solid supports.~~

2. ~~The method of claim 1, further comprising:~~

~~designing a second oligonucleotide comprising a sequence that is complementary to the target sequence of the dengue virus.~~

3.2. ~~The method of claim 1 or 2, wherein the target sequence of the dengue virus comprises a target sequence of the NS1 region of the dengue virus.~~

4.3. ~~The method of any one of claims 1-32, wherein the carbon nanotubes are single walled carbon nanotubes.~~

5.4. ~~The method of any one of claims 1-43, wherein the solid supports are magnetic beads.~~

6.5. ~~The method of claim 54, wherein the plurality of magnetic beads are streptavidin functionalized magnetic beads or carboxylic acid functionalized magnetic beads.~~

~~7.6.~~ The method of any one of claims 4-~~65~~, further comprising:

~~designing the first oligonucleotide to function as a detection probe;~~
tagging the first oligonucleotide with a NH₂ functional group on the 5' end of the first oligonucleotide; ~~and prior to~~ hybridising a ~~the~~ plurality of the first oligonucleotide to the plurality of ~~single-walled~~ carbon nanotubes to form a plurality of carbon nanotube based labels.

~~87.~~ The method of any one of claims 5-~~76~~, further comprising:

~~designing the second oligonucleotide to function as a capture probe;~~
biotinylating the second oligonucleotide; ~~and~~
tagging the second oligonucleotide with a NH₂ functional group on the 3' end of the second oligonucleotide; ~~and prior to~~ immobilising a ~~the~~ plurality of the second oligonucleotide to the plurality of magnetic beads to form a plurality of conjugated magnetic beads.

~~98.~~ The method of any one of claims 1-~~87~~, further comprising:

~~designing~~ ~~providing~~ a third oligonucleotide comprising a sequence that is non-complementary to the target sequence of the dengue virus; ~~and~~
wherein the third oligonucleotide functions as a negative control sequence.

~~109.~~ The method of any one of claims ~~76-98~~, further comprising:

hybridising the plurality of carbon nanotube labels with the plurality of conjugated magnetic beads to form a mixture of the plurality of carbon nanotube based labels and the plurality of conjugated magnetic beads.

~~1110.~~ The method of claim ~~109~~, wherein a plurality of the third oligonucleotide and the mixture of the plurality of carbon nanotube based labels and the plurality of conjugated magnetic beads are provided in one test tube.

~~1211.~~ The method of any one of claims 1-~~1110~~, wherein the dengue virus is selected from the group consisting of dengue virus serotype 1 (DENV-1), dengue virus serotype 2 (DENV-2), dengue virus serotype 3 (DENV-3) and dengue virus serotype 4 (DENV-4).

~~13~~12. A diagnostic kit for ~~rapidly~~ diagnosing a dengue virus in 15 minutes or less, wherein the diagnostic kit is prepared ~~accorded~~ according to the method of any one of claims 1-~~12~~11.

~~14~~13. A diagnostic kit for ~~rapidly~~ diagnosing a dengue virus in 15 minutes or less, the diagnostic kit comprising:

a mixture comprising of:

(i) a plurality of carbon nanotube based labels consisting of a plurality of a first oligonucleotide hybridised to a plurality of single walled carbon nanotubes, wherein the first oligonucleotide comprising a sequence selected from the group consisting of sequences 1 as shown in Table 1 that is complementary to a target sequence of the dengue virus; and

(ii) a plurality of conjugated magnetic beads consisting of a plurality of a second oligonucleotide immobilised to a plurality of magnetic beads, wherein the second oligonucleotide comprising a sequence selected from the group consisting of sequences 2 as shown in Table 1 that is complementary to the target sequence of the dengue virus;

wherein the plurality of carbon nanotube labels are hybridised with the plurality of conjugated magnetic beads, and

wherein the mixture is provided in one test tube.

~~15.~~ The diagnostic kit of claim ~~14~~,

~~wherein the plurality of carbon nanotube based labels of the mixture comprises a plurality of a first oligonucleotide hybridised to a plurality of single walled carbon nanotubes, and~~

~~wherein the first oligonucleotide comprises a sequence that is complementary to a target sequence of the dengue virus.~~

~~16~~14. The diagnostic kit of claim ~~15~~13,

wherein the first oligonucleotide functions as a detection probe, and

wherein the first oligonucleotide is tagged with a NH₂ functional group on the 5' end of the first oligonucleotide to facilitate hybridisation of the plurality of the first

oligonucleotide to the plurality of single walled carbon nanotubes to form the plurality of carbon nanotube based labels.

~~17.~~ The diagnostic kit of any one of claims ~~15-16~~,

~~wherein the plurality of conjugated beads of the mixture comprises a plurality of a second oligonucleotide immobilised to a plurality of magnetic beads, and wherein the second oligonucleotide comprises a sequence that is complementary to the target sequence of the dengue virus.~~

~~1815.~~ The diagnostic kit of claim ~~1714~~,

wherein the second oligonucleotide functions as a capture probe,
wherein the second oligonucleotide is biotinylated, and

wherein the second oligonucleotide is tagged with a NH₂ functional group on the 3' end of the second oligonucleotide to facilitate the immobilisation of the plurality of the second oligonucleotide to the plurality of magnetic beads to form the plurality of conjugated magnetic beads.

~~1916.~~ The diagnostic kit of any one of claims ~~1513-1815~~, wherein the target sequence of the dengue virus comprises a target sequence of the NS1 region of the dengue virus.

~~2017.~~ The diagnostic kit of any one of claims ~~1713-1916~~, wherein the plurality of magnetic beads are streptavidin functionalized magnetic beads or carboxylic acid functionalized magnetic beads.

~~2118.~~ The diagnostic kit of any one of claims ~~1513-2017~~, further comprising:

a third oligonucleotide comprising a sequence that is non-complementary to the target sequence of the dengue virus;

wherein the third oligonucleotide functions as a negative control sequence; and
wherein a plurality of the third oligonucleotide is provided in the one test tube.

~~2219.~~ The diagnostic kit of any one of claims ~~1413-2118~~, wherein the dengue virus is selected from the group consisting of dengue virus serotype 1 (DENV-1), dengue virus

serotype 2 (DENV-2), dengue virus serotype 3 (DENV-3) and dengue virus serotype 4 (DENV-4).

2320. A method of ~~rapidly~~ diagnosing a dengue virus in 15 minutes or less, the method comprising:

providing the diagnostic kit of any one of claims ~~1312-2219~~;

providing target RNA of the dengue virus; and

adding the target RNA of the dengue virus to the mixture of the plurality of carbon nanotube-based labels and the plurality of conjugated magnetic beads in the one test tube.

2421. The method of claim 2320, wherein the plurality of carbon nanotube-based labels and the plurality of conjugated magnetic beads aggregate in the presence of the target RNA of the dengue virus.

2522. The method of claim 2421, wherein the plurality of carbon nanotube-based labels and the plurality of conjugated magnetic beads aggregate in the presence of the target RNA of the dengue virus to form dark coloured complexes.

2623. The method of claim 2522, wherein the formation of dark coloured complexes allows for visual detection of the target RNA of the dengue virus.

2724. The method of any one of claims 2421-2623, wherein the aggregation of the plurality of carbon nanotube-based labels and the plurality of conjugated magnetic beads is dependent on the concentration of the target RNA of the dengue virus added to the mixture.

2825. The method of any one of claims 2320-2724, wherein the dengue virus is selected from the group consisting of dengue virus serotype 1 (DENV-1), dengue virus serotype 2 (DENV-2), dengue virus serotype 3 (DENV-3) and dengue virus serotype 4 (DENV-4).

~~29~~26. The method of any one of claims ~~25~~22-~~28~~25, wherein the formation of the dark coloured complexes is generated in about 15 minutes or less, and wherein diagnosis of the dengue virus is achieved in about 15 minutes or less.

~~30~~27. The method of any one of claims ~~25~~22-~~29~~26, wherein the formation of the dark coloured complexes is generated in about 10 minutes or less, and wherein diagnosis of the dengue virus is achieved in about 10 minutes or less.

~~31~~28. The method of any one of claims ~~25~~22-~~30~~27, wherein the formation of the dark coloured complexes is generated in about 5 minutes or less, and wherein diagnosis of the dengue virus is achieved in about 5 minutes or less.

~~32~~29. The method of any one of claims ~~23~~20-~~31~~28, wherein the method has an accuracy of about 98% or greater for diagnosing the dengue virus.

~~33~~30. The method of any one of claims ~~23~~20-~~32~~29, wherein the method has an accuracy of about 99% or greater for diagnosing the dengue virus.

~~34~~31. The method of any one of claims ~~23~~20-~~33~~30, wherein the method has an accuracy of about 100% for diagnosing the dengue virus.

~~35~~32. The method of any one of claims ~~23~~20-~~34~~31, wherein the method has a specificity of about 98% or greater for diagnosing the dengue virus.

~~36~~33. The method of any one of claims ~~23~~20-~~35~~32, wherein the method has a specificity of about 99% or greater for diagnosing the dengue virus.

~~37~~34. The method of any one of claims ~~23~~20-~~36~~33, wherein the method has a specificity of about 100% for diagnosing the dengue virus.