

WHAT IS CLAIMED IS:

1. A method of determining if a first organism having a first genome or a second organism having a second genome is present in a test sample or an initial sample from which the test sample was prepared, comprising:

(a) probing the test sample with a virtual probe comprising two or more probe molecules, wherein each probe molecule is capable of specifically hybridizing to one or more target nucleic acids corresponding to the first genome and/or one or more homologous target nucleic acids corresponding to the second genome, and wherein the probe molecules hybridize non-identically to the target nucleic acids corresponding to the first and second genomes, such that the hybridizing of the probe molecules to the one or more target nucleic acids corresponding to the first genome and the one or more target nucleic acids corresponding to the second genome can distinguish between the target nucleic acids corresponding to the first genome and the target nucleic acids corresponding to the second genome; and

(b) detecting and/or quantifying signals from hybridization of the probe molecules in the virtual probe to nucleic acids, if any, in the test sample,

thereby determining if the first organism or second organism is present in the test sample or initial sample.

2. The method of claim 1, wherein the one or more target nucleic acids corresponding to the first genome are a first amplicon set and the one or more target nucleic acids corresponding to the second genome are a second amplicon set, and wherein each probe molecule in the virtual probe is capable of specifically hybridizing to one or more amplicons in the first amplicon set and/or the second amplicon set, and wherein the probe molecules hybridize non-identically to the amplicons in the first amplicon set and the amplicons in the second amplicon set, such that the hybridizing of the probe molecules to the amplicons in the first amplicon set and the second amplicon set can distinguish between the first amplicon set and the second amplicon set.

3. The method of claim 2, which further comprises preparing the test sample by performing a PCR amplification reaction on the initial sample using PCR primers capable of hybridizing to, and initiating a PCR amplification from, both the first genome and the second genome, resulting in the first amplicon set and a second amplicon set, respectively, when the first genome and second genome are present in the initial sample.

4. The method of claim 3, wherein the PCR primers comprise more than one primer pair and wherein the first amplicon set comprises a plurality of first amplicons and/or the second amplicon set comprises a plurality of second amplicons.

5. The method of claim 4, wherein the plurality of first amplicons corresponds to different regions in the first genome and/or the plurality of second amplicons corresponds to different regions in the second genome.

6. The method of claim 3, wherein the PCR primers comprise a single primer pair and the first amplicon set consists of a single first amplicon and the second amplicon set consists of a single second amplicon.

7. The method of any one of claims 1 to 6, wherein the probe molecules of the virtual probe are positionally addressable probe molecules present on an array, each at a discrete location on the array.

8. The method of any one of claims 2 to 7, wherein the first amplicon set and the second amplicon set each comprise a nucleotide sequence corresponding to a gene encoding rRNA.

9. The method of any one of claims 2 to 7, wherein the first amplicon set and the second amplicon set each comprise a nucleotide sequence corresponding to an intergenic spacer region between rRNA genes.

10. The method of any one of claims 1 to 9, wherein the first organism and second organism are microorganisms.

11. The method of claim 10, wherein the microorganisms are members of the same order, family, genus, or group.

12. The method of claim 10 or claim 11, wherein the microorganisms are bacteria.

13. The method of claim 2, wherein the first organism and second organism are bacteria, and wherein the first amplicon set and the second amplicon set each comprise a nucleotide sequence corresponding to a 16S rRNA gene and/or a nucleotide sequence corresponding to a 23S rRNA gene.

14. The method of claim 13, wherein the first amplicon set and the second amplicon set each comprise a nucleotide sequence corresponding to a 16S rRNA gene.

15. The method of claim 13 or claim 14, wherein the first amplicon set and the second amplicon set each comprise a nucleotide sequence corresponding to a 23S rRNA gene.

16. The method of any one of claims 13 to 15, wherein the first amplicon set and the second amplicon set each comprise a nucleotide sequence corresponding to a 16S-23S intergenic spacer region.

17. The method of any one of claims 12 to 16, wherein:

(a) the first microorganism is a coagulase negative *Staphylococcus sp.* and the second microorganism is a coagulase positive *Staphylococcus sp.*;

(b) the first microorganism is *Streptococcus gordonii* and the second microorganism is *Streptococcus anginosus*; or

(c) the first microorganism is *Streptococcus mitis* and the second microorganism is *Streptococcus pneumoniae*.

18. An addressable array, comprising:

(a) one or more virtual probes for distinguishing a first genomic sequence from a second, homologous genomic sequence, each virtual probe comprising a group of positionally addressable oligonucleotide probe molecules, each at a discrete location on the array, wherein each probe molecule in the one or more virtual probes comprises a nucleotide sequence that is 90% to 100% complementary to 15 to 40 consecutive nucleotides in the first genomic sequence or second genomic sequence; and

(b) optionally, one or more control probe molecules.

19. The addressable array of claim 18, in which at least one virtual probe comprises a probe molecule whose nucleotide sequence comprises CCAGTCTTATAGGTAGGTTAYCCACG (SEQ ID NO:1), GCTTCTCGTCCGTTTCGCTCG (SEQ ID NO:2), CAGTCTATGGTGTAGCAAGCTACGGTAT (SEQ ID NO:3), TATCCCCCTCTAATAGGCAGGTTA (SEQ ID NO:4), AGCTAATACAACGCAGGTCCATCT (SEQ ID NO:5), GATGCAAGTGACCTTTTAAGCAA (SEQ ID NO:6), or GATGCAAGTGACCTTTTAAGTAA (SEQ ID NO:7).

20. A method of determining if a first organism having a first genome or a second organism having a second genome is present in a test sample or an initial sample from which the test sample is derived, comprising:

(a) probing the test sample with an array according to claim 18 or claim 19 which comprises a virtual probe comprising two or more probe molecules, wherein each probe molecule is capable of specifically hybridizing to one or more target nucleic acids corresponding to the first genome and/or one or more homologous target nucleic acids corresponding to the second genome, and wherein the probe molecules hybridize non-identically to the target nucleic acids corresponding to the first and second genomes, such that the hybridizing of the probe molecules to the one or more target nucleic acids corresponding to the first genome and the one or more target nucleic acids corresponding to the second genome can distinguish between the target nucleic acids corresponding to the first genome and the target nucleic acids corresponding to the second genome; and

(b) washing unbound nucleic acid molecules from the array;

(c) detecting and/or quantifying the signal at each probe molecule location on the array; and

(d) if the signals indicate that:

(i) target nucleic acids that hybridize to the probe molecules of the array are present in the test sample, analyzing the signals to determine if target nucleic acids corresponding to the first genome or target nucleic acids corresponding to the second genome are present in the sample, thereby determining if the first organism or second organism are present in the initial sample or the test sample; or

(ii) no target products that hybridize to the probe molecules of the virtual probe are produced in step (a), determining that that initial sample or test sample does not contain the first organism or the second organism,

thereby determining if the first organism or second organism is present in the initial sample or the test sample.

21. A system for determining if an organism is present in a sample, comprising :

(a) an optical reader for generating signal data for each probe molecule location of the array of claim 18 or claim 19; and

(b) at least one processor which:

(i) is configured to receive signal data from the optical reader;

(ii) is configured to analyze the signal data for the one or more virtual probes; and

(iii) has an interface to a storage or display device or network for outputting a result of the analysis.

22. An oligonucleotide probe molecule whose nucleotide sequence comprises CCAGTCTTATAGGTAGGTTAYCCACG (SEQ ID NO:1), GCTTCTCGTCCGTTTCGCTCG (SEQ ID NO:2), CAGTCTATGGTGTAGCAAGCTACGGTAT (SEQ ID NO:3), TATCCCCCTCTAATAGGCAGGTTA (SEQ ID NO:4), AGCTAATACAACGCAGGTCCATCT (SEQ ID NO:5), GATGCAAGTGACCTTTTAAGCAA (SEQ ID NO:6), or GATGCAAGTGACCTTTTAAGTAA (SEQ ID NO:7) .

23. A virtual probe comprising a plurality of oligonucleotide probe molecules, wherein:

(a) at least one oligonucleotide probe molecule in the virtual probe has a nucleotide sequence comprising CCAGTCTTATAGGTAGGTTAYCCACG (SEQ ID NO:1) and another oligonucleotide molecule in the virtual probe has a nucleotide sequence comprising GCTTCTCGTCCGTTTCGCTCG (SEQ ID NO:2);

(b) at least one oligonucleotide probe molecule in the virtual probe has a nucleotide sequence comprising CAGTCTATGGTGTAGCAAGCTACGGTAT (SEQ ID NO:3) and another oligonucleotide molecule in the virtual probe has a nucleotide sequence comprising TATCCCCCTCTAATAGGCAGGTTA (SEQ ID NO:4); or

(c) at least one oligonucleotide probe molecule in the virtual probe has a nucleotide sequence comprising AGCTAATACAACGCAGGTCCATCT (SEQ ID NO:5), another oligonucleotide molecule in the virtual probe has a nucleotide sequence comprising GATGCAAGTGACCTTTTAAGCAA (SEQ ID NO:6) and another oligonucleotide molecule in the virtual probe has a nucleotide sequence comprising GATGCAAGTGACCTTTTAAGTAA (SEQ ID NO:7).

24. An addressable array comprising a group of positionally addressable probe molecules, each at a discrete location on the array, wherein the group of probe molecules comprises the oligonucleotide probe molecule of claim 22.

25. An addressable array comprising the virtual probe of claim 23, wherein each probe molecule in the virtual probe is at a discrete location on the array.