

CLAIMS:

1. An *in vitro* method of detecting a plurality of targets in a biological sample, comprising:

(a) contacting the biological sample with a plurality of target-binding probes to form a plurality of target-bound probes;

(b) covalently attaching at least one of the target-bound probes to the biological sample;

(c) ~~observing~~ detecting a first set of signals from a first set of the plurality of target-bound probes;

(d) modifying the ~~observed~~ detected -signals;

(e) generating a second set of signals from a second set of the plurality of target-bound probes; and

(f) ~~observing~~ detecting the second set of signals.

2. The method of claim 1, further comprising repeating steps (d) to (f) multiple times with a third, a fourth, and an n^{th} set of the plurality of target-bound probes to ~~observe~~ detect a third, a fourth, and an n^{th} set of signals.

3. The method of claim 1, further comprising correlating the ~~detected~~ observed signals to the detection of the plurality of targets.

4. The method of claim 1, wherein all of the plurality of target-bound probes are covalently attached to the biological sample.

5. The method of claim 1, wherein the target-bound probe is covalently attached to the biological sample via the target itself or via a molecule that is located in the vicinity of the target.

6. The method of claim 1, wherein the target-binding probe comprises a unit derived from a target-binding moiety and an independently detectable moiety.

16. The method of claim 1, further comprising imaging the biological sample to obtain a background signal signature prior to contacting the biological sample with a plurality of target-binding probes.

17. The method of claim 1, wherein the detection of the plurality of targets comprises identifying the presence, absence, location, or amount of the plurality of targets.

18. The method of claim 1, wherein the plurality of targets is selected from the group consisting of an oligonucleotide, a nucleic acid, a peptide, a protein, a hormone, a receptor, a polysaccharide, a lipid, and a combination thereof.

19. The method of claim 1, wherein the plurality of targets consist essentially of a plurality of ribonucleic acids.

20. The method of claim 1, wherein the plurality of targets consist essentially of a plurality of proteins.

21. An *in vitro* method of detecting a plurality of targets in a biological sample, comprising:

(a) contacting the biological sample with a plurality of target-binding probes to form a plurality of target-bound probes;

(b) covalently attaching at least one of the target-bound probes to the biological sample;

(c) generating a first set of signals from a first set of the plurality of target-bound probes;

(d) ~~observing~~ detecting the first set of signals;

(e) modifying the ~~observed~~ detected signals;

(f) generating a second set of signals from a second set of the plurality of target-bound probes; and

(g) detecting ~~observing~~ the second set of signals.

22. The method of claim 21, further comprising repeating steps (e) to (g) with a third, a fourth, and an n^{th} set of the plurality of target-bound probes to ~~observe~~detect a third, a fourth, and an n^{th} set of signals.

23. The method of claim 22, wherein integer value of n ranges from 5 to 100.

24. The method of claim 21, further comprising correlating the ~~observed~~detected signals to the detection of the plurality of targets.

25. The method of claim 21, further comprising imaging the biological sample to obtain a background signal signature prior to generating the first set of signals.

26. The method claim 25, further comprising subtracting the background signal signature from the signals ~~observed~~detected in the subsequent signal ~~observing~~detecting steps prior to the correlating step.

27. An *in vitro* method of detecting a plurality of targets in a biological sample, comprising:

(a) contacting the sample with a plurality of target-binding probes to form a plurality of target-bound probes;

(b) covalently attaching at least one of the target-bound probes to the biological sample;

(c) contacting the plurality of target-bound probes with a first set signal-generating probes that are capable of binding a first set of the plurality of target-bound probes, and are capable of generating a first set of signals;

(d) detecting the first set of the plurality of targets via ~~detecting~~observing the first set of signals;

(e) modifying the ~~detected~~observed first set of signals; and

(f) repeating steps (c) to (e) multiple times using a subsequent set of signal-generating probes to detect a subsequent set of the plurality of targets, wherein the subsequent set of signal-generating probes are capable of binding a subsequent set of the plurality of target-bound probes, and are capable of generating a subsequent set of signals.

28. The method of claim 27, wherein the ~~observed~~-detected first set of signals is modified by applying a chemical agent.

29. The method of claim 27, further comprising steps for removing the target-binding probes that are not bound to the plurality of targets prior to contacting the target-bound probes with the first set of signal-generating probes.

30. The method of claim 29, further comprising steps for removing the first set of signal-generating probes that are not bound to the first set of the plurality of target-bound probes, prior to detecting the first set of the plurality of targets.

31. The method of claim 27, wherein all of the plurality of target-bound probes are covalently coupled to the target itself or to a molecule that is located in the vicinity of the target.

32. The method of claim 27, wherein the steps (c) to (f) are repeated up to 100 times.

33. The method of claim 27, wherein the plurality of targets is selected from the group consisting of a deoxyribonucleic acid, a ribonucleic acid, a protein, and a combination thereof.

34. The method of claim 27, wherein the plurality of targets consist essentially of a plurality of ribonucleic acids.

35. The method of claim 1, claim 21 or claim 27, wherein modifying the ~~detected~~observed signals comprises a method selected from the group consisting of decreasing the intensity of the signals, shifting the signal peaks, changing the resonant frequencies, and removing the signals.

36. A kit for detection of a plurality of multiple targets in a biological sample, comprising:

a plurality of target-binding probes, wherein each target-binding probe is capable of generating a signal and at least one of the target-binding probe is capable of cross-linking to a target or to a molecule that is located in the vicinity of the target; and

at least one chemical agent, capable of modifying the generated signal from at least one target-binding probe; and

a cross-linking agent capable of cross-linking the probes to said target or molecule.

37. A device for detection of a plurality of targets in a biological sample, comprising:

a sample handling system;

a reagent dispensing system;

a probe cross-linking system; and

a signal detection system.

38. The device of claim 37, wherein the reagent dispensing system dispenses at least one reagent solution comprising at least one target-binding probe.

39. The device of claim 37, wherein at least one of the sample handling system, the reagent dispensing system, the probe cross-linking system, or the signal detection system is operable without an operator intervention.

40. The device of claim 37, wherein the device is operable to detect the plurality of targets in the biological sample using a method comprising the steps of:

(a) contacting the biological sample with a plurality of target-binding probes to form a plurality of target-bound probes;

(b) ~~detecting~~observing a first set of signals from a first set of the plurality of target-bound probes;

(c) modifying the ~~observed~~detected signals;

(d) generating a second set of signals from a second set of the plurality of target-bound probes; and

(e) ~~detecting~~observing the second set of signals.

41. The device of claim 37, wherein the device is operable to detect the plurality of targets in the biological sample using a method comprising the steps of:

(a) contacting the biological sample with a plurality of target-binding probes to form a plurality of target-bound probes;

(b) covalently attaching at least one of the target bound probe to the biological sample;

(c) ~~detecting~~observing a first set of signals from a first set of the plurality of target-bound probes;

(d) modifying the ~~observed~~~~detected~~ signals;

(e) generating a second set of signals from a second set of the plurality of target-bound probes; and

(f) ~~detecting~~observing the second set of signals.