CLAIMS:

1. A method of probing multiple targets in a biological sample comprising the steps of :

(a) subjecting the sample to an in situ hybridization reaction using a labeled nucleic acid probe that directly or indirectly binds an RNA target ;

(b) observing a signal from the labeled probe bound to the RNA target;

(c) optionally removing the signal from the labeled probe;

(d) subjecting the sample to an antigen retrieval protocol to retrieve the sample's protein epitopes ;

(e) subjecting the sample to an in situ hybridization reaction using an antibody-based method and attaching one or more antibody probe to antigens on the sample;

(f) observing a signal from the one or more antibody probes;

(g) removing the signal from the antibody probes;

(h) optionally applying a protease treatment to access the sample's DNA targets;

(i) subjecting the sample to an in situ hybridization reaction using a labeled nucleic acid probe to directly or indirectly label one or more of the sample's DNA targets ;

(j) observing a signal from the labeled DNA targets;

(k) optionally removing the signal from the one or more labeled DNA targets.

~~2. The method of claim 1 wherein the *in situ* hybridization reaction using a labeled nucleic acid probe in step (a) comprises hybridization with multiple probes targeting the same RNA target .~~

~~2~~3. The method of claim 1 wherein step a further comprises a prehybridization step to block DNA, the use of a blocking agent in the hybridization reaction, or a combination thereof.

34. The method of claim 1 wherein after observing a signal from the labeled probe bound to the RNA target the sample is subjected to step c, and steps a, b, and c are repeated one or more times with other labeled nucleic acid probes for different RNA targets.

45. The method of claim 1 wherein an *in situ* hybridization reaction using an antibody-based method comprises immunohistochemistry (IHC) or immunofluorescence (IF) techniques.

56. The method of claim 45 wherein antibody probe comprises a mixture of more than one probe.

~~7. The method of claim 6 wherein the mixture comprises 2 to 10 probes.~~

~~8. The method of claim 5 wherein at least one antibody probe is conjugated to an enzymatic label, a fluorescent signal generator, or a combination thereof.~~

69. The method of claim 45 wherein after observing and removing a signal from the one or more antibody probes, steps e, f, and g are repeated one or more times with another antibody probe for a different antigen.

710. The method of claim 1 further comprising after protease treatment, the step of preserving tissue morphology, prehybirdization , or a combination thereof.

~~11. The method of claim 1 wherein *in situ* hybridization in step (i) comprises treatment with a nucleic acid based binder to form a Watson-Crick bond, a Hoogsteen bond, or a combination thereof.~~

~~12. The method of claim 11 wherein the nucleic-acid binder length in range of from about 4 nucleotides to about 1000 nucleotides.~~

~~13. The method of claim 12 wherein the nucleic-acid binder length is in range of from about 12 nucleotides to about 400 nucleotides.~~

8~~14~~. The method of claim 1 wherein after observing a signal from the labeled probe bound to the DNA target, steps I, j, and k are repeated one or more times with another labeled nucleic acid probe for a different DNA target.

9~~15~~. The method of claim 1 wherein the removing the signals in steps c, g, and i comprises a signal inactivation agent, photoreaction, photactivated chemical bleaching, probe stripping, oxidation, electron transfer, or a combination thereof.

10~~16~~. The method of claim 1 further comprising staining the sample with one or more control probes to allow for registration of multiple images of the sample.

11~~17~~. The method of claim 10~~16~~ wherein the control probe is a morphological stain.

12~~18~~. The method of claim 10~~16~~ further comprising registering multiple images of the sample wherein registering comprises:

obtaining multiple images of the samples from steps b, f, j, or a combination thereof;

aligning and overlaying the multiple images according the the signals detected from the control probe.

13~~19~~. The method of claim 12~~18~~ further comprising the step of analyzing the expression of the protein, RNA, and DNA from the overlaid images.

1420. The method of claim 1, further comprising measuring one or more intensity values of the signal observed in observing steps b, f, j, or a combination thereof.

1521. The method of claim 1420 wherein measuring one or more intensity values of the signal comprises a signal amplification technique.

~~22. The method of claim 21 wherein the signal comprises a fluorescent signal, a chromogenic probe, or a combination thereof.~~

~~23. The method of claim 22 further comprising correlating the intensity value with an amount of target present in the sample.~~

~~24. The method of claim 1, wherein said sample comprises a cell or a tissue sample.~~

~~25. The method of claim 24 wherein said sample comprises a Formalin-Fixed, Paraffin-Embedded (FFPE) tissue sample.~~

~~26. The method of claim 25 wherein the paraffin-embedded tissue sample is dewaxed prior to step a.~~

~~27. The method of claim 1, wherein said sample comprises a cellular suspension, such as a hematapoetic cell or circulating tumor cell.~~

~~28. The method of claim 27 wherein step a comprises subjecting the sample in suspension to an in-situ hypridization reaction in solution.~~

~~29. The method of claim 1 comprising detecting three or more targets in a single tissue section wherein said targets include at least one DNA, one RNA and one protein target.~~

~~30. The method of claim 29 further comprising detection of a morphological target.~~