WHAT IS CLAIMED IS:

1. A method for spatial detection of RNA molecules in a biological sample, comprising:

(a) providing a substrate defined by an array of spots, wherein each spot comprises DNA oligomers immobilized thereto, wherein each of the DNA oligomers comprises:

(i) a spatial barcode, wherein all DNA oligomers in one spot share the same spatial barcode, which is different from the spatial barcodes in other spots; and

(ii) a poly(dT) sequence;

(b) placing a biological sample onto the substrate;

(c) contacting the substrate with a Poly(A) polymerase enzyme mix comprising a Poly(A) polymerase and a polymerase reaction buffer reagent to perform in situ polyadenylation;

(d) capturing RNA molecules from the biological sample;

(e) adding reverse transcription reagents to generate cDNA molecules from captured RNA molecules, wherein cDNA molecules generated from RNAs captured by DNA oligomers on a spot comprise a spatial barcode common to the spot; and

(f) obtaining a map of spatial gene expression based on the cDNA molecules generated.

2. The method of claim 1, wherein step (d) includes permeabilizing the cells in the biological sample to permit release and capture of the RNA molecules from the cells in the biological sample.

3. The method of claim 1 or 2, wherein each of the DNA oligomers further comprises a unique molecular identifier sequence.

4. The method according to any one of the proceeding claims, wherein step (b) further comprises fixing the biological sample (e.g., using formaldehyde, Formalin-fixed, parafin-embedded (FFPE), Acetone, Methanol+acetone, Glyoxal fixation);

optionally wherein step (b) further comprises staining the fixed biological sample;

further optionally wherein step (b) further comprises capturing an image of the fixed and stained biological sample.

5. The method according to any one of the preceding claims, wherein the sequences of the generated cDNA are obtained;

optionally wherein either the generated cDNAs with spatial barcodes and the cDNA sequences are used to map the spatial gene expression, or the generated cDNAs and the cDNA sequences are correlated with the captured image of the fixed and stained biological sample to map the spatial gene expression.

6. The method according to any one of the preceding claims, wherein:

(i) step (e) further comprises initiating second strand synthesis via the addition of a second strand primer; and/or

(ii) the cDNAs are denatured and transferred from the spots to a solution for amplification, optionally wherein the amplified cDNAs are further processed based on amplicon size; and/or

(iii) the Poly(A) polymerase enzyme is a yeast Poly(A) polymerase or an E. coli Poly(A) polymerase; and/or

(iv) the substrate is composed of a material selected from the group consisting of glass, silicon, poly-L-lysine coated material, nitrocellulose, polystyrene, cyclic olefin copolymers (COCs), cyclic olefin polymers (COPs), polypropylene, polyethylene and polycarbonate.

7. The method according to any one of the preceding claims, wherein step (c) further comprises, prior to the contacting step, equilibrating the substrate by adding a wash buffer comprising Poly(A) polymerase reaction buffer, an RNase inhibitor, and nuclease free water to the substrate;

optionally wherein step (c) comprises, after the equilibrating, adding a Poly(A) polymerase enzyme mix which comprises Poly(A) polymerase reaction buffer, a Poly(A) polymerase enzyme, adenosine triphosphate (ATP), RNase inhibitor, and nuclease-free water and incubating.

8. The method according to any one of the preceding claims, wherein:

(i) the array of spots comprises 10-100,000,000 spots, such as at least 10, at least 100, at least 1,000, at least 10,000, at least 50,000, at least 100,000, at least 200,000, at least 300,000, at

least 400,000, at least 500,000, at least 1,000,000, at least 2,000,000, at least 10,000,000, at least 20,000,000, at least 30,000,000, at least 40,000,000, at least 50,000,000, at least 75,000,000, or at least 100,000,000 spots; and/or

(ii) the array is placed within a capture area having a dimension of about 1 mm^2 to about 100 mm^2, optionally wherein the capture area has a dimension of up to 10 mm^2, 6.5 mm^2, or 3 mm^2; and/or

(iii) the spots are about 10 nm to about 1 mm in diameter; and/or

(iv) the spots are about 500 nm to about 125 μm apart as measured by center of spot to center of spot; and/or

(v) the spots are less than 60 μm in diameter and are no more than 100 μm apart as measured by center of spot to center of spot, optionally wherein the spatial resolution is about 60 microns; and/or

(vi) the spots are less than 220 nm in diameter and are no more than 750 nm apart as measured by center of spot to center of spot, optionally wherein the spatial resolution is about 10 microns; and/or

(vii) the spots are in an organized pattern in the array, or wherein the spots are randomly distributed in the array; and/or

(viii) the spatial resolution is from about 1 micron to about 100 microns, optionally wherein the spatial resolution is about 60 microns, or about 10 microns.

9. The method according to any one of the proceeding claims, wherein the length of the poly(A) tail is controlled in the in situ polyadenylation;

optionally wherein:

(i) the length of the poly(A) tail is about 10 base pairs to about 4,000 base pairs and/or

(ii) the length of the poly(A) tail is less than about 2,000,1,600, or 1,000 base pairs; and/or

(iii) the Poly(A) polymerase enzyme mix includes (i) ATP and (ii) biotin-11-ATP or dATP at a ratio of at least 5:1, optionally wherein the ratio of ATP to biotin-11-ATP or dATP is 1:1.

10. The method according to any one of the preceding claims, wherein the biological sample is a tissue;

optionally wherein:

(i) the tissue is connective tissue; further optionally wherein the connective tissue is connective tissue proper such as loose or dense tissue, comprising collagen, reticular, or elastic fibers; or wherein the connective tissue is specialized connective tissue, for example comprising adipose, cartilage, bone, blood, reticular, and lymphatic tissues;

(ii) the tissue is epithelial tissue, such as simple squamous, stratified squamous, simple cuboidal, stratified cuboidal, simple columnar, stratified columnar, pseudostratified columnar, or transitional epithelia;

(iii) the tissue is muscle tissue, such as a cardiac, skeletal, or smooth muscle tissue;

(iv) the tissue is nervous tissue; or

(v) the biological sample is a combination of tissue types which form an organ, such as wherein the biological sample is taken from a testis.

11. The method according to any one of the preceding claims, wherein:

(i) the poly(dT) sequence comprises a VN sequence at the 3' end wherein the V is any nucleotide base other than T and N is any nucleotide base; and/or

(ii) the biological sample is a histological section of tissue; and/or

(iii) the RNAs captured are ribonucleic acids (RNAs), RNA degradation products, RNAs comprising a poly(A) tail, messenger RNA (mRNA), long noncoding RNAs (lncRNAs), long intergenic noncoding RNAs (lincRNAs), cis- natural antisense transcripts (cisNATs), antisense RNAs, ribosomal RNAs (rRNAs), microRNAs (miRNAs), small interfering RNAs (siRNAs), short hairpin RNAs (shRNAs), guide RNAs (gRNAs), transfer RNAs (tRNAs), small nuclear RNAs (snRNAs), small nucleolar RNAs (snoRNAs), small Cajal body-specific RNA (scaRNAs), enhancer RNAs (eRNAs), piwi-interacting RNAs (piRNAs), Y RNAs, non-coding RNAs, vault RNA, viral RNA, microbial RNA such as bacterial RNA, archaeal RNA, or fungal RNA, or combinations thereof.

12. The method of claim 1, wherein the method further comprises isolating a subpopulation of cDNAs from the cDNAs generated in step (e);

optionally wherein the subpopulation of cDNAs is generated from viral RNAs, bacterial RNA, archaeal RNA, or fungal RNA, and/or

wherein the method further comprises obtaining the sequences of the cDNAs in the isolated subpopulation.

13. The method according to any one of the preceding claims, wherein the biological sample is a tissue sample of an injured tissue or an organ suspected to suffer an infection;

optionally wherein the tissue sample is a tumor section, gut microbiome, brain section, patient biopsy, or a plant sample, and/or

wherein the method further comprises comparing the spatial gene expression map of the tissue sample to (i) the spatial gene expression map of a control sample, or (ii) the spatial gene expression map of another sample of the same tissue taken at a different time point.

14. A kit comprising:

a substrate defined by an array of spots, wherein each spot comprises DNA oligomers immobilized on the substrate, wherein each of the DNA oligomers comprises:

(i) a spatial barcode, wherein all primers in one spot share the same spatial barcode, which is different from the spatial barcodes in other spots;

(ii) a poly(dT) sequence; and

(iii) a unique molecular identifier sequence;

at least one reagent comprising a Poly(A) polymerase enzyme mix; and

optionally instructions for use.

15. The kit of claim 14, wherein the Poly(A) polymerase enzyme mix comprises:

a polymerase reaction buffer reagent;

a poly(A) polymerase enzyme reagent; and optionally

nuclease free water reagent.

16. The kit of according to claim 14 or 15, wherein the Poly(A) polymerase enzyme mix further comprises adenosine triphosphate reagent and/or an RNase inhibitor reagent;

optionally wherein the Poly(A) polymerase enzyme mix comprises (i) ATP and (ii) biotin-11-ATP or dATP, further optionally at a ratio that is greater than about 5:1 ATP to biotin-11-ATP or dATP.

17. The kit according to any one of claims 14-16, wherein the kit further comprises a wash buffer reagent;

optionally wherein the wash buffer reagent comprises:

a polymerase reaction buffer reagent,

an RNase inhibitor reagent; and optionally

a nuclease-free water reagent.

18. The kit according to any one of claims 14-17, wherein:

(i) the poly(dT) sequence comprises a VN sequence at the 3' end wherein the V is any nucleotide base other than T and N is any nucleotide base; and/or

(ii) the reagents are provided in separate containers or provided in pre-mixed quantities of any combination of reagents.

19. The kit according to any one of claims 14-18, wherein:

(i) the array of spots comprises 10-100,000,000 spots, such as at least 10, at least 100, at least 1,000, at least 10,000, at least 50,000, at least 100,000, at least 200,000, at least 300,000, at least 400,000, at least 500,000, at least 1,000,000, at least 2,000,000, at least 5,000,000, at least 10,000,000, at least 20,000,000, at least 30,000,000, at least 40,000,000, at least 50,000,000, at least 75,000,000, or at least 100,000,000 spots; and/or

(ii) the array is placed within a capture area having a dimension of about 1 mm^2 to about 100 mm^2, optionally wherein the capture area has a dimension of up to 10 mm^2, 6.5 mm^2, or 3 mm^2; and/or

(iii) the spots are about 10 nm to about 1 mm in diameter; and/or

(iv) the spots are about 20 μm to about 125 μm apart as measured by center of spot to center of spot; and/or

(v) the spots are less than 60 μm in diameter and are no more than 100 μm apart as measured by center of spot to center of spot, optionally wherein the spatial resolution is about 60 microns; and/or

(vi) the spots are less than 220 nm in diameter and are not more than 750 nm apart as measured by center of spot to center of spot, optionally wherein the spatial resolution is about 10 microns; and/or

(vii) the spots are in an organized pattern in the array, or wherein the spots are randomly distributed in the array; and/or

(viii) the spatial resolution ranges from about 1 micron to about 100 microns, optionally wherein the spatial resolution is about 60 microns, or about 10 microns.