
Checklist for the RNAscope® Automated Assay on Leica Biosystems' BOND RX System RNAscope®2.5 LS Duplex Assay

Doc. No. CHK 47-001



Reagents/Materials required from Leica Biosystems

- BOND Polymer Refine Detection Kits (DAB kit, Cat. No. DS9800; Red kit, Cat. No. DS9390), and Hematoxylin. These kits contain prefilled DAB, Red, and Hematoxylin reagents.
- BOND Open Containers (Cat. No. Op309700). Fill these containers with reagents from the RNAscope LS Duplex Reagent Kit.
- BOND Universal Covertiles (Cat. No. S21.2001).
- BOND Dewax Solution (Cat. No. AR9222). Place solution in one of the large containers located in the bottom of the instrument.
- BOND Epitope Retrieval Solution 1 and 2 (Cat. Nos. AR9961 and AR9640).
- BOND Wash Solution 10X (Cat. No. AR9590). Dilute solution 1:10 and place in one of the large containers located in the bottom of the instrument.
- BOND Aspirating Probe Cleaning System (Cat. No. CS9100).
- BOND Mixing Stations (Cat. No. S21.1971). Replace every month.
- BOND DAB Enhancer 30ml (Cat. No. AR9432).
- BDZ 11.0 software.



Reagents/Materials required from ACD for RNAscope

- RNAscope 2.5 LS Duplex Reagent Kit (Cat. No. 322440). The kit contains all the detection and pretreatment reagents needed to run the RNAscope assay on the Leica BOND RX, except for the RNA-specific probes.

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- RNAscope 2.5 LS Duplex Positive Control Probe (Cat. No. 320748 for Human PPIB-C1/Polr2A-C2; Cat No. 320768 for Mouse PPIB-C1/POLR2A-C2). Probes target common housekeeping genes to help qualify your samples and interpret your results.
 - RNAscope 2.5 LS Duplex Negative Control Probe (Cat. No. 312758). The probe targets the bacterial dapB gene in two channels to control for background noise, and to help interpret assay results (DapB C1/DapB-C2).
 - RNAscope 2.5 LS Target Probes target specific RNAs of interest in Channel C1 and Channel C2.
 - RNAscope Control Slides (Cat. No. 310045 for Human Cell Pellet; Cat. No. 310023 for Mouse 3T3 Cell Pellet).
 - Software protocols created for each probe (workaround solution).



Reagents/Materials provided by Researcher

- Superfrost® Plus slides are required. Other slide types may result in tissue detachment.
- Fresh reagents (e.g. ethanol and xylene).
- Fresh 10% NBF (neutral-buffered formalin).
- Tissue-Tek® slide rack and staining dishes.
- EcoMount or PERTEX®.
- Hotplate, drying oven, water bath, thermometer, and microscope.
- Paraffin wax, microtome, and fume hood

Note: Always refer to the latest version of the user manuals for the complete set of materials required to run the RNAscope assay, available at <http://www.acdbio.com/technical-support/user-manuals>.

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