
MANUAL ASSAYS: RNAscope® 2.5 HD RED Assay

CHK 47-003



Reagents/Materials required from ACD

- RNAscope Target Probes C1 Channel probe
- RNAscope Positive Control Probes targeting common housekeeping genes to help qualify samples and interpret assay results.
- RNAscope Negative Control Probe targeting the bacterial gene *dapB* to control for background noise and to help interpret assay results.
- RNAscope® 2.5 HD Reagent Kit RED Cat No. **32235**) consisting of a pretreatment kit, a detection kit and wash buffer.
- HybEZ™ Hybridization System with ACD EZ-Batch Slide System (321461, 321462) is required for RNAscope hybridization to maintain optimum humidity and temperature at 40°C.
- RNAscope Introductory Pack designed for first-time users of the RNAscope assay (Cat. No. 322370 Human or Cat. No. 322371 Mouse), and providing a complete set of reagents including controls probes, slides, the ImmEdge® pen and the RNAscope 2.5 HD Reagent Kit – BROWN.
- RNAscope Control Slides – Human Cell Pellet Cat. No. 310045, or Mouse 3T3 Cell Pellet Cat. No. 310023.
- ImmEdge Hydrophobic Barrier Pen (Cat. No. 310018) is the only pen that will maintain a hydrophobic barrier throughout the RNAscope procedure. No other pen should be used.
- Ecomount*
 - . Ecomount Mounting media is a must-have reagent for RNAscope 2.5 Red assay. No other mounting media can be used.

**Reagents/Materials provided by Researcher**

- Superfrost® Plus slides (Fisher Scientific): Superfrost Plus slides are required. Other slide types may result in tissue detachment.
 - Counter staining: diluting Gill's Hematoxylin 1:2 with distilled water is required for counter staining.
- Fresh reagents (e.g. ethanol and xylene).
- Fresh 10% NBF (neutral-buffered formalin).
- Ammonium Hydroxide, 28–30%.
- Hotplate, drying oven, water bath, thermometer, microscope, etc.
- Paraffin wax, microtome, 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>.

© 2015 Advanced Cell Diagnostics. All rights reserved. RNAscope® and HybEZ™ are trademarks of Advanced Cell Diagnostics, Inc. All other trademarks belong to their respective owners.