
RNAscope® VS Universal HRP and AP Automated Assay on the Ventana™ DISCOVERY™ ULTRA System

Doc. No. CHK 47-008

Note: This checklist includes consumables that are required for both the HRP-DAB assay and the AP-Red assay. If only one of these assays is ran, only the required combination of Ventana and ACD reagent kits needs to be ordered.



Reagents/Materials required from Ventana/Roche Diagnostics

- mRNA Amplification, Pretreatment, and Red/DAB Kits. Kits contain prefilled mRNA Detection Reagents and the user-fillable empty dispensers for all ACD reagents except probes.

mRNA DAB Reagent Kits

- mRNA DAB Detection Kit (Cat. No. 760-224; Ordering Code 06614353001)
- mRNA Probe Amplification Kit (Cat. No. 760-222; Ordering Code 06614337001)
- mRNA Sample Prep Kit (Cat. No. 760-248; Ordering Code 08127166001)

mRNA Red Reagent Kits

- mRNA RED Detection Kit (Cat. No. 760-234; Ordering Code 7099037001)
- mRNA RED Probe Amplification Kit (Cat. No. 760-236; Ordering Code 07095341001)
- mRNA Sample Prep Kit (Cat. No. 760-248; Ordering Code 08127166001)

- DISCOVERY™ Wash Buffer (Cat. No. 950-510; Ordering Code 07311079001). Use instead of EZ Prep Buffer.

- User-fillable probe dispensers for negative control probe, positive control probe, and the target probe. Additional dispensers can be ordered for m target probes. See the Ventana™ website for part numbers.

- Cat. No. 960-761; Ordering Code 05279828001
- Cat. No. 960-762; Ordering Code 05279836001
- Cat. No. 960-763; Ordering Code 05279844001

- 10X SSC Solution (Cat. No. 950-110; Ordering Code 05353947001). Required to run mRNA procedures on the DISCOVERY™ ULTRA.

- Ultra LCS (Predilute) Cat. No. 650-210; Ordering Code 05424534001.

- Reaction Buffer (10X) Cat. No. 950-300; Ordering Code 05353955001.
- Discovery CC1 (Cat. No.950-500; Ordering Code 06414575001).
- mRNA Universal staining procedure for DISCOVERY™ ULTRA (which can be installed by Roche or ACD FAS)
- Counterstaining reagents.
 - Hematoxylin II (Cat. No. 760-2208; Ordering Code 05277965001)
 - Bluing (Cat. No. 760-2037; Ordering Code 05266769001)

Note: If ACD's or one's own hematoxylin and bluing reagents are used, Counterstain 1 (Cat. No. 771-741) and Counterstain 2 (Cat. No. 771-742) user-fillable dispensers from Ventana™ will need to be ordered.



Reagents/Materials required from ACD for RNAscope

- RNAscope® VS Universal HRP Reagent Kit – BROWN (Cat. No. 323200). Contains the following:
 - RNAscope® VS Universal HRP Detection Reagents (Cat. No. 323210)
 - RNAscope® VS Accessory Kit (Cat. No. 320630)
 - RNAscope® VS Universal Sample Prep Reagents v2 (Cat. No. 323740)
- RNAscope® VS Universal Reagent Kit – RED (Cat. No. 323250). Contains the following:
 - RNAscope® VS Universal AP Detection Reagents (Cat. No. 323260)
 - RNAscope® VS Accessory Kit (Cat. No. 320630)
 - RNAscope® VS Universal Sample Prep Reagents v2 (Cat. No. 323740)
- RNAscope® 2.5 VS Positive Control Probe (choose species needed): Cat No. 312029 for Human *UBC*; Cat. No. 313909 for Human *PPIB*; Cat No. 310459 for Human *POLR2A*; Cat. No. 310779 for Mouse *Ubc*; Cat. No. 313919 for Mouse *Ppib*; Cat. No. 312479 for Mouse *Polr2a*).
- RNAscope® 2.5 VS Negative Control Probe (Cat. No. 312039). Targets the bacterial *dapB* gene to control for background noise, and to help interpret assay results.
- RNAscope® 2.5 VS Target Probes targeting specific RNAs of interest.
- RNAscope® Control Slides. Choose species needed (Cat. No. 310045 for Human Cell Pellet; Cat. No. 310023 for Mouse 3T3 Cell Pellet).
- Use RNAscope® Target Retrieval Reagents to boil samples offline (Cat No. 322000). This is included in the Reagent Kits.

Reagents/Materials provided by Researcher

- Superfrost™ Plus slides (Fisher Scientific): Superfrost Plus slides are required. Other slide types may result in tissue detachment.
- EcoMount or Cytoseal XYL mounting media:
 - Use EcoMount mounting media with the RNAscope® VS Universal AP Assay; no other mounting media can be used
 - Use a xylene-based mounting media (such as CytoSeal XYL) with the RNAscope® VS Universal HRP Assay.
- Fresh reagents (e.g. ethanol and xylene)
- Dawn detergent
- Fresh 10% NBF (neutral-buffered formalin)
- Tissue-Tek® slide rack and staining dishes
- 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 <https://acdbio.com/technical-support/user-manuals>.