

Preparation of RNAProtect®-preserved tissues for histological studies

This procedure has been adapted by customers for use with the RNAProtect Tissue Reagent. **It has not been thoroughly tested and optimized by QIAGEN.**

The procedure can be used to prepare RNAProtect-stabilized biopsies for histological and immunohistochemical studies.

Please be sure to read carefully the *RNeasy® Mini Handbook*, the *RNeasy Midi/Maxi Handbook*, or the *RNeasy Protect and RNAProtect Handbook*, including the detailed Protocol for the “Stabilization of RNA in Harvested Animal Tissues”, before beginning this procedure.

Procedure

- 1. Remove stabilized tissue from the RNAProtect Tissue Reagent using forceps.**
- 2. If only a portion of the stored tissue is to be used, place the tissue on a clean surface for cutting, and cut the tissue. Transfer the remaining tissue back into the RNAProtect Tissue Reagent for further storage, or store it at –90 to –65°C without the reagent.**

RNA in the RNAProtect-treated tissue is still protected while the tissue is processed at 18 to 25°C. This allows cutting and weighing of tissues at ambient temperatures. It is not necessary to cut the tissue on ice or dry ice or in a refrigerated room.

The remaining tissue can be stored in RNAProtect Tissue Reagent for subsequent RNA isolation. The reagent preserves RNA for up to 1 day at 37°C, 7 days at 18 to 25°C, or 4 weeks at 2 to 8°C, allowing transportation, storage, and shipping of samples without ice or dry ice. Alternatively, the samples can be placed at –30 to –15°C or –90 to –65°C for archival storage.

- 3. Wash the tissue twice for 15 min each time in PBS.**

Once the RNAProtect Tissue Reagent is washed out, RNA in the tissue is no longer protected. However, this does not affect subsequent histological and immunohistochemical analyses.

- 4. Remove the tissue from the PBS, and fix for 24 h in 10% buffered formalin.**
- 5. Embed the tissue in paraffin or freeze in Tissue-Tek® OCT™ Compound following standard procedures.**

Paraffin-embedded RNAProtect-stabilized tissues can be used for immunohistochemical and routine hematoxylin and eosin staining.

RNAProtect-stabilized tissues frozen in Tissue-Tek OCT Compound can be used for immunohistochemical and frozen-section hematoxylin and eosin staining.

Note: After RNAProtect treatment, S-100 immunostain can appropriately stain nerve fibers but not melanocytes or Langerhan’s cells. In addition to S-100 antigen, other soluble antigens may not stain appropriately after treatment with RNAProtect Tissue Reagent.

For up-to-date licensing information and product-specific disclaimers, see the respective QIAGEN kit handbook or user manual. QIAGEN kit handbooks and user manuals are available at or can be requested from QIAGEN Technical Services or your local distributor.