Molecular Devices/Arcturus
Guidelines for Preparing “RNase-free” Paraffin
Embedded Tissue Sections for LCM
Cleaning the Work Area and Equipment

To minimize nuclease contamination, clean the work area, rotary microtome, and tissue flotation water bath.

**Precautions**  
*Wear clean, disposable gloves throughout this entire procedure.*

**Work Area**  
Use RNase AWAY® and follow the manufacturer’s instructions to clean any surfaces that may come in contact with the sample.

**Rotary Microtome**  
To clean the rotary microtome:

1. Remove and discard the old disposable microtome blade.
2. Using a Kimwipe soaked with RNase AWAY, clean the knife holder.
3. Dry the knife holder with a clean Kimwipe.
4. Install a new disposable microtome blade into the knife holder.

**Tissue Flotation Water Bath**  
To clean the tissue flotation water bath:

1. Using a Kimwipe soaked with RNase AWAY, clean the interior of the water bath.
2. Rinse the interior of the water bath with Milli-Q or nuclease-free water.
3. Fill the water bath with Milli-Q or nuclease-free water.
4. Heat the water to an appropriate temperature for the paraffin used in your laboratory, typically 41–43°C.  
**IMPORTANT!** Do not add any additives (for example, antimicrobial solutions or chemicals) to the water bath.
Preparing the Tissue Scrapes

**Precautions**

Wear clean, disposable gloves throughout this entire procedure.

**Preparing the Slides**

IMPORTANT! For optimal performance, Arcturus recommends using tissue fixed in 10% Neutral Buffered Formalin for 14–24 hours. For complete tissue fixation guidelines, contact Arcturus Customer Support at www.arctur.com.

To prepare the slides:

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<tr>
<td>1.</td>
<td>On the rotary microtome, set the cutting thickness to 7 μm.</td>
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<td>2.</td>
<td>Prepare a paraffin block:</td>
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<td>a. Place a paraffin block into the specimen holder.</td>
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<td>b. Trim any excess paraffin from the block face.</td>
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<td>c. After trimming, cut and discard the first five sections.</td>
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<td>3.</td>
<td>From the fresh surface of the paraffin block, cut 7-μm section(s) from your specimen.</td>
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<td>If you are cutting more than one specimen, do one of the following to avoid cross-contamination:</td>
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<td>• Move to a new section of the blade, or</td>
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<td>• Use a new disposable blade for each specimen.</td>
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<td>4.</td>
<td>Remove the section(s) from the rotary microtome, then float them onto the heated tissue flotation water bath.</td>
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<td>Allow the section(s) to flatten, but do not leave them in the water bath for more than 2 minutes.</td>
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<td>5.</td>
<td>Mount each section on a room-temperature Superfrost Micro Slide (VWR, cat # 48311-600).</td>
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6. **Dry the slides:**
   a. Prop each slide on end to allow water to drain away from the section.
   b. Air-dry the slides for a minimum of 90 minutes at room temperature. Do not dry the slides in an oven.

   **IMPORTANT!** Depending on the humidity in the environment, it may take longer for the sections to dry. The sections must be dry before proceeding. Do not allow the sections to air-dry for longer than 3 hours.

   **Note:** Discard any slides that have wrinkles or folds in the section.

7. **When the slides are dry:**
   - Proceed immediately to “Performing De-paraffinization” on page 2-6, or
   - Store the slides in a micro slide box in a room-temperature desiccator for up to 2 weeks, or
   - Store the slides in a micro slide box at −65 to −80°C for up to 3 months.

8. **After completing the slide preparation process:**
   a. Remove any paraffin debris from the rotary microtome, clean the surfaces with a Kimwipe soaked with RNase AWAY, then dry all surfaces.
   b. Discard the water from the tissue flotation water bath, clean the interior with RNase AWAY, then dry all surfaces.