### Staining Protocol Recommendations (intelliPATH and manual use) Cont’d:

#### Chromogen:
- Incubate for 5 minutes at RT with Biocare's DAB - OR - Incubate for 5-7 minutes at RT with Biocare's Warp Red.

#### Counterstain:
- Counterstain with hematoxylin. Rinse with deionized water. Apply Tacha's Bluing Solution for 1 minute. Rinse with deionized water.

#### Staining Protocol Recommendations (ONCORE Automated Slide Staining System):
- OAR301 is intended for use with the ONCORE Automated Slide Staining System. Refer to the ONCORE Automated Slide Staining System User Manual for specific instructions on its use. Protocol parameters in the ONCORE Automated Slide Staining Protocol Editor should be programmed as follows:

<table>
<thead>
<tr>
<th>Protocol Name</th>
<th>ER Rb</th>
</tr>
</thead>
<tbody>
<tr>
<td>Protocol Template (Description)</td>
<td>Rb HRP Template 1</td>
</tr>
<tr>
<td>Dewaxing (DS Option)</td>
<td>DS2</td>
</tr>
<tr>
<td>Antigen Retrieval (AR Option)</td>
<td>AR2, low pH; 95°C</td>
</tr>
<tr>
<td>Reagent Name, Time, Temp.</td>
<td>ER Rb, 30 min., 25°C</td>
</tr>
<tr>
<td>Technical Note</td>
<td>This antibody has been optimized for use with Biocare's MACH 2 Rabbit HRP-Polymer Detection and ONCORE Rabbit HRP Detection. Use TBS buffer for washing steps.</td>
</tr>
</tbody>
</table>

### Limitations:
- This product is provided for Research Use Only (RUO) and is not for use in diagnostic procedures. Suitability for specific applications may vary and it is the responsibility of the end user to determine the appropriate application for its use.

#### Precautions:
1. This antibody contains less than 0.1% sodium azide. Concentrations less than 0.1% are not reportable hazardous materials according to U.S. 29 CFR 1910.1200, OSHA Hazard communication and EC Directive 91/155/EC. Sodium azide (NaN₃) used as a preservative is toxic if ingested. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. Upon disposal, flush with large volumes of water to prevent azide build-up in plumbing. (Center for Disease Control, 1976, National Institute of Occupational Safety and Health, 1976) (5)
2. Specimens, before and after fixation, and all materials exposed to them should be handled as if capable of transmitting infection and disposed of with proper precautions. Never pipette reagents by mouth and avoid contacting the skin and mucous membranes with reagents and specimens. If reagents or specimens come in contact with sensitive areas, wash with copious amounts of water. (6)
3. Microbial contamination of reagents may result in an increase in nonspecific staining.
4. Incubation times or temperatures other than those specified may give erroneous results. The user must validate any such change.
5. Do not use reagent after the expiration date printed on the vial.
6. The SDS is available upon request and is located at http://biocare.net.

#### Technical Support:
- Contact Biocare's Technical Support at 1-800-542-2002 for questions regarding this product.

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### Estrogen Receptor (ER) [SP1]

#### Concentrated and Prediluted Rabbit Monoclonal Antibody

**Catalog Number:** CRM 301 A, B, C  
**Description:** 0.1, 0.5, 1.0 ml, concentrated  
**Dilution:** 1:100  
**Diluent:** Renoir Red  
**Isotype:** IgG  
**Total Protein Concentration:** ~10 mg/ml. Lot specific Ig concentration is not available.  
**Epitope/Antigen:** ER  
**Cellular Localization:** Nuclear  
**Positive Tissue Control:** Breast carcinoma

**Known Applications:**
- Immunohistochemistry (formalin-fixed paraffin-embedded tissues)
- Tissue engineering

**Storage and Stability:**
- Store at 2°C to 8°C. Do not use after expiration date printed on vial. If reagents are stored under conditions other than those specified in the package insert, they must be verified by the user. Diluted reagents should be used promptly; any remaining reagent should be stored at 2°C to 8°C.

**Staining Protocol Recommendations (intelliPATH and manual use):**

**Peroxide Block:** Block for 5 minutes with Biocare’s Peroxidazed 1.
**Pretreatment Solution (recommended):** Diva
**Pretreatment Protocol: Heat Retrieval Method:**
- Preheat the retrieval solution to 95°C for 30 minutes and then place slides into the preheated solution if using Biocare’s Decloaking Chamber Pro or Decloaking Chamber Plus. If using Biocare’s Decloaking Chamber NxGen, place slides into the retrieval solution without preheating. Retrieve at 95°C for 40 minutes. Allow solution to cool for 20 minutes and then wash in distilled water.
**Protein Block (Optional):** Incubate for 5-10 minutes at RT with Biocare’s Background Punisher.
**Primary Antibody:** Incubate for 30 minutes at RT.
**Probe:** N/A
**Polymer:** Incubate for 30 minutes at RT with a secondary-conjugated polymer.
References: