

# Estrogen Receptor (ER) [SP1]

Concentrated and Prediluted Rabbit Monoclonal Antibody  
902-301-071922

**BIOCARE**  
M E D I C A L

Available Product Formats				
Format	Catalog Number	Description	Dilution	Diluent
Concentrate	CRM 301 A, B, C	0.1, 0.5, 1.0 mL	1:100	Renoir Red
Predilute	PRM 301 AA, H	6.0, 25 mL	Ready-to-use	N/A
ONCORE	OAR 301 T60	60 tests	Ready-to-use	N/A
Q Series– For Leica BOND-III	ALR 301 G7	7.0 mL	Ready-to-use	N/A

## Intended Use:

For Research Use Only. Not for use in diagnostic procedures.

## Summary and Explanation:

Human estrogen receptor (ER) is a 66 kDa protein that acts as an estrogen-dependent, nuclear hormone receptor. Studies have shown ER is present in the nuclei of epithelial cells in normal breast and endometrial tissues, as well as a subset of breast carcinomas. The SP1 clone is a high affinity rabbit monoclonal antibody directed against an epitope of the C-terminus of the ER protein (1). SP1 has been shown to stain formalin-fixed paraffin-embedded tissues. The robustness of SP1 has been demonstrated by successful immunohistochemistry using lower temperatures for antigen retrieval (e.g. 80 or 95°C), and in some instances, staining can be obtained even without antigen retrieval (2-3).

## Principle of Procedure:

Antigen detection in tissues and cells is a multi-step immunohistochemical process. The initial step binds the primary antibody to its specific epitope. After labeling the antigen with a primary antibody, a one-step or two-step detection procedure can be applied. A one-step procedure will feature an enzyme labeled polymer that binds the primary antibody. A two-step procedure will feature a linker antibody added to bind to the primary antibody. An enzyme-labeled polymer is then added to bind the linker antibody. These detections of the bound antibodies are evidenced by a colorimetric reaction.

**Source:** Rabbit monoclonal

**Species Reactivity:** Human; others not tested

**Clone:** SP1

**Isotype:** IgG

**Protein Concentration:** Call for lot specific Ig concentration.

**Epitope/Antigen:** ER

**Cellular Localization:** Nuclear

**Positive Tissue Control:** Breast carcinoma

## Known Applications:

Immunohistochemistry (formalin-fixed paraffin-embedded tissues)

**Supplied As:** Buffer with protein carrier and preservative

## Storage and Stability:

Store at 2°C to 8°C. The product is stable to the expiration date printed on the label, when stored under these conditions. Do not use after expiration date. Diluted reagents should be used promptly; any remaining reagent should be stored at 2°C to 8°C.

## Staining Protocol Recommendations (intelliPATH FLX® and manual use):

**Peroxide Block:** Block for 5 minutes with Peroxidized 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 Decloaking Chamber Pro or Decloaking Chamber Plus. If using 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 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.

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

## Staining Protocol Recommendations (intelliPATH and manual use)

### Cont'd:

### Counterstain:

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

### Technical Note:

This antibody, for intelliPATH FLX and manual use, has been standardized with MACH 2 detection system. Use TBS for washing steps.

## Staining Protocol Recommendations (ONCORE™ Automated Slide Staining System):

OAR301 is intended for use with the ONCORE. Refer to the User Manual for specific instructions for use. Protocol parameters in the Protocol Editor should be programmed as follows:

**Protocol Name:** ER Rb

**Protocol Template (Description):** Rb HRP Template 1

**Dewaxing (DS Option):** DS2

**Antigen Retrieval (AR Option):** AR2, low pH; 95°C

**Reagent Name, Time, Temp.:** ER Rb, 30 min., 25°C

## Staining Protocol Recommendations (Q Series – For Leica BOND-III):

ALR301 is intended for use with the Leica BOND-III. Refer to the User Manual for specific instructions for use. Recommended protocol parameters are as follows:

**Protocol Name:** IHC Protocol F

**Detection:** Bond Polymer Refine

**HIER:** 10 min with ER1

**Peroxide Block:** 5 min

**Marker (Primary Antibody):** 15 min

**Post Primary:** 8 min

**Polymer:** 8 min

**Mixed DAB Refine:** 10 min

**Hematoxylin:** 5 min

## 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 (NaN<sub>3</sub>) 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 into 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.

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TP v1 (10/26/2021)

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### Precautions Cont'd:

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.

### References:

1. Cheang MC, *et al.* Immunohistochemical detection using the new rabbit monoclonal antibody SP1 of estrogen receptor in breast cancer is superior to mouse monoclonal antibody 1D5 in predicting survival. *J Clin Oncol.* 2006 Dec;24(36):5637-44.
2. Rossi S, *et al.* Rabbit monoclonal antibodies: a comparative study between a novel category of immunoreagents and the corresponding mouse monoclonal antibodies. *Am J Clin Pathol.* 2005 Aug;124(2):295-302.
3. Cano G, *et al.* Estimation of hormone receptor status in fine-needle aspirates and paraffin-embedded sections from breast cancer using the novel rabbit monoclonal antibodies SP1 and SP2. *Diagn Cytopathol.* 2003 Oct; 29(4):207-11.
4. Rocha R, *et al.* Rabbit monoclonal antibodies show higher sensitivity than mouse monoclonals for estrogen and progesterone receptor evaluation in breast cancer by immunohistochemistry. *Pathol Res Pract.* 2008; 204(9):655-62.
5. Center for Disease Control Manual. Guide: Safety Management, NO. CDC-22, Atlanta, GA. April 30, 1976 "Decontamination of Laboratory Sink Drains to Remove Azide Salts."
6. Clinical and Laboratory Standards Institute (CLSI). Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline-Fourth Edition CLSI document M29-A4 Wayne, PA 2014.

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