



Estrogen Receptor

IHC of Estrogen Receptor on an FFPE Breast Tissue

Description Estrogen receptor (ER) is a nuclear receptor for estrogens such as estradiol (the main endogenous human estrogen). The two different estrogen receptor proteins produced from the ESR1 and ESR2 genes are usually called the alpha and beta receptors.

This ER antibody recognizes a protein of 67 kDa, which is identified as estrogen receptor (ER) alpha. The ER gene consists of more than 140 kb of genomic DNA divided into 8 exons, which are translated into a protein with six functionally discrete domains, labeled A through F. This antibody strongly stains the nucleus of epithelial cells in breast carcinomas. ER is an important regulator of growth and differentiation of cells in the mammary gland. Presence of ER in breast tumors indicates an increased likelihood of response to anti-estrogen (e.g., Tamoxifen) therapy.

Antibody Type	Rabbit Monoclonal	Clone	RBT11
Isotype	IgG	Reactivity	Paraffin, Frozen
Localization	Nuclear	Control	Breast Carcinoma
Storage	Store at 2°-8°C	Stability	2 years

For long-term storage of the concentrated antibody, it is recommended that aliquots of the antibody be frozen at -20°C in glycerol 50% (frost-free freezers are not recommended). Repeated freezing and thawing must be avoided. Dilute using an antibody diluent such as ImmunoDetector Protein Block/Antibody Diluent (BSB 0040 and BSB 0041) or ImmunoDNA Background Blocker (BSB 0103-BSB 0107).

Presentation Estrogen receptor clone RBT11 is a rabbit monoclonal antibody derived from cell culture supernatant that is concentrated, dialyzed, filter sterilized and diluted in buffer pH 7.5, containing BSA and sodium azide as a preservative.

Availability	Catalog No.	Antibody Type	Dilution	Volume/QTY
	BSB 5491	Prediluted	Ready-To-Use	3.0 ml
	BSB 5492	Prediluted	Ready-To-Use	7.0 ml
	BSB 5493	Prediluted	Ready-To-Use	15.0 ml
	BSB 5494	Concentrated	1:50-1:250	0.1 ml
	BSB 5495	Concentrated	1:50-1:250	0.5 ml
	BSB 5496	Concentrated	1:50-1:250	1.0 ml
	BSB 5497	Control Slides		5

Note: For concentrated antibodies, please centrifuge prior to use to ensure recovery of all product.

- References**
1. Dabbs DJ, et al. *Diagnostic Immunohistochemistry*. 2002
 2. Kell DL, et al. *Applied Immunohistochemistry*. 1993;1(4):275-81
 3. Leong ASY, et al. *Applied Immunohistochemistry*. 1993;1(4):282-288
 4. Tesch M, et al. *Am J Clin Pathol*. 1993;99:8-12
 5. Francesco AM, et al. *Applied Immunohistochemistry*. 1994;2(3):157-163
 6. Silvio M, et al. *Applied Immunohistochemistry*. 1995;3(2):85-90
 7. Bejar J, et al. *Arch Pathol Lab Med*. 1998;Apr;122(4):346-52

Protocol Suggested protocol on reverse

Recommended Immunohistochemical Protocol

- Pretreatment**
1. Cut and mount 3-4 micron formalin-fixed paraffin-embedded tissues on positive charged slides.
 2. Air dry for 2 hours at 58° C.
 3. Deparaffinize, dehydrate and rehydrate tissues.
 4. Subject tissues to heat epitope retrieval using a suitable retrieval solution such as **ImmunoDNA Retriever with Citrate** (BSB 0020-BSB 0023) or **EDTA** (BSB 0030-BSB 0033).
 5. Any of three heating methods may be used:
 - a. **Electric Pressure Cooker**
Place standoff rack at base of pressure cooker. Add 1-2 inches of distilled water to the pressure cooker and turn heat to high, and incubate for 15 minutes. Open and immediately transfer slides to room temperature.
 - b. **Water Bath Method**
Place tissues/slides in a pre-warmed staining dish or coplin jar containing the **ImmunoDNA Retriever with Citrate** or **EDTA** in a water bath set at 95°-99° C. Incubate for 30-60 minutes.
 - c. **Conventional Steamer Method**
Place tissues/slides in a pre-warmed staining dish or coplin jar containing the **ImmunoDNA Retriever with Citrate** or **EDTA** in a Steamer, cover and steam for 30-60 minutes.
 6. After heat treatment, transfer slides in **ImmunoDNA Retriever with Citrate** or **EDTA** to room temperature and let stand for 15-20 minutes.
 7. Wash slides with IHC wash buffer or DI water.
 8. Continue IHC staining protocol.

Immunohistochemical Protocol

Step	ImmunoDetector (AP or HRP)	PolyDetector (AP or HRP)
Peroxidase/AP Block	5 minutes	5 minutes
Primary Antibody	30 minutes	45 minutes
Secondary Biotinylated Link	10 minutes	Not Applicable
AP or HRP Label	10 minutes	45 minutes
Substrate-Chromogen	5-10 minutes	10 minutes
Counterstaining	Time varies with counterstain	Time varies with counterstain

