



## BCA-225

*IHC of BCA-225 on an FFPE Breast Carcinoma Tissue*

**Description** This antibody recognizes a human Breast Carcinoma-associated glycoprotein, BCA-225 (220-225 kDa). This protein differs in size and distribution from other Breast Carcinoma antigens. Unlike other carcinoma antibodies against Breast Carcinoma antigens, this antibody does not react with benign or malignant colonic tissues. Since this antigen is localized in malignancies of Breast Carcinomas and Carcinoma of the Uterine Cervix, it can be effectively used to identify metastatic Breast Carcinoma lesions.

Strong intracytoplasmic staining is seen in primary and metastatic Breast Carcinoma tissue, as well as in Cervical Carcinomas. Apical staining is seen in normal kidney, lung, Fallopian tube, liver, skin (eccrine sweat glands) and uterus. Similar staining patterns are observed in lung, ovarian, and endometrial cancers. Carcinomas of the colon, stomach, prostate, urinary bladder, liver, pancreas, thyroid, and parotid are negative, as are Sarcomas and Lymphoid Cancers.

<b>Antibody Type</b>	Mouse Monoclonal	<b>Clone</b>	Cu-18
<b>Isotype</b>	IgG1/K	<b>Reactivity</b>	Paraffin, Frozen
<b>Localization</b>	Cytoplasmic	<b>Control</b>	Breast Carcinoma
<b>Storage</b>	Store at 2°-8°C	<b>Stability</b>	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** BCA-225 is a mouse 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 5064	Prediluted	Ready-To-Use	3.0 ml
	BSB 5065	Prediluted	Ready-To-Use	7.0 ml
	BSB 5066	Prediluted	Ready-To-Use	15.0 ml
	BSB 5067	Concentrated	1:50-1:200	0.1 ml
	BSB 5068	Concentrated	1:50-1:200	0.5 ml
	BSB 5069	Concentrated	1:50-1:200	1.0 ml
	BSB 5070	Control Slides		5

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

**References**

1. Ceriani RL, *Monoclonal Ab's and Breast Cancer*. Boston, Martinus, Nijhoff. 1985
2. Mesa-Tejada R, et al. *Am J Pathol*. 1988;130:305-314

**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

