



EpCAM/Epithelial Specific Antigen

IHC of EpCAM on an FFPE Breast Cancer Tissue

Description Epithelial Cell Adhesion Molecule (EpCAM) or Epithelial Specific Antigen is a 40kD cell surface antigen that is broadly distributed in epithelial cells and displays a highly conserved expression in carcinomas. These glycoproteins are located on the cell membrane surface and in the cytoplasm of virtually all epithelial cells, with the exception of most squamous epithelia, hepatocytes, renal proximal tubular cells, gastric parietal cells and myoepithelial cells. However, focal positivity may be seen in the basal layer of squamous cell epithelium of endoderm (e.g., palatine tonsils) and mesoderm (e.g., uterine cervix).

EpCAM expression has been reported to be a possible marker of early malignancy, with expression being increased in tumor cells, and de novo expression being seen in dysplastic squamous epithelium. Epithelial specific antigen has been known to play an important role as a tumor-cell marker in lymph nodes from patients with esophageal carcinoma. EpCAM can be used to distinguish among Basal Cell, Basosquamous Carcinomas and Squamous Cell Carcinomas of the skin.

| | | | |
|----------------------|------------------|-------------------|------------------|
| Antibody Type | Mouse Monoclonal | Clone | Ber-EP4 |
| Isotype | IgG1/K | Reactivity | Paraffin, Frozen |
| Localization | Cytoplasmic | Control | Adenocarcinomas |
| 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 EpCAM 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 6275 | Prediluted | Ready-To-Use | 3.0 ml |
| | BSB 6276 | Prediluted | Ready-To-Use | 7.0 ml |
| | BSB 6277 | Prediluted | Ready-To-Use | 15.0 ml |
| | BSB 6278 | Concentrated | 1:25-1:100 | 0.1 ml |
| | BSB 6279 | Concentrated | 1:25-1:100 | 0.5 ml |
| | BSB 6280 | Concentrated | 1:25-1:100 | 1.0 ml |
| | BSB 6281 | Control Slides | | 5 |

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

- References**
1. Beer TW, et al. *Histopathology*. 2000;37:218-223
 2. Carella R, et al. *Am J Surg Pathol*. 2001;25:43-50
 3. Hosch S, et al. *Cancer Res*. 2000; 60:6836-6840
 4. Latza U, et al. *J Clin Pathol*. 1990;43:213-219

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 |

