



Cytokeratin 8 & 18

IHC of CK 8 and 18 on an FFPE Colon Carcinoma Tissue

Description Cytokeratin 8 belongs to the Type II (basic) subfamily of high molecular-weight keratins and exists in combination with Cytokeratin 18 (Type I [acidic] subfamily of low molecular weight keratins). They are perhaps the most commonly found products of the intermediate filament gene family, and are expressed in single-layer epithelial tissues of the body.

Cytokeratins 8 and 18 can be found in most simple epithelium (e.g., thyroid, female breast, gastrointestinal tract, and respiratory tract). Adenocarcinomas and most Non-keratinizing Squamous Carcinomas will stain, but Keratinizing Squamous Carcinomas will not. This antibody is used when attempting to demonstrate the presence of Paget cells; there is very little keratin 18 in the normal epidermis so only Paget cells will stain. This approach facilitates the interpretation using immunostains and is more sensitive than mucin histochemistry.

Antibody Type	Mouse Monoclonal	Clone	B22.1 & B23.1
Isotype	IgG1	Reactivity	Paraffin, Frozen
Localization	Cytoplasmic	Control	Prostate, Pancreas, Salivary Gland
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 Anti-Cytokeratin 8 & 18 is a cocktail of mouse monoclonal antibodies 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 5414	Prediluted	Ready-To-Use	3.0 ml
	BSB 5415	Prediluted	Ready-To-Use	7.0 ml
	BSB 5416	Prediluted	Ready-To-Use	15.0 ml
	BSB 5417	Concentrated	1:100-1:400	0.1 ml
	BSB 5418	Concentrated	1:100-1:400	0.5 ml
	BSB 5419	Concentrated	1:100-1:400	1.0 ml
	BSB 5420	Control Slides		5

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

References

1. Angus B, Purvis J, et al. *Journal of Pathology*. 1987;155:377-384
2. Corson JM, *Pathol Annual*. 1986; 21 (part 2):47-81
3. Sasaki M, et al. *Histopathology*. 1998;Mar;32(3):199-208

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

