

Human Cytokeratin 19 Ready-To-Use IHC Kit

Cat.No: IHC0192H
Applications: **IHC-P**
Reactivity: Human
Size: 50T
Assay type: Immunohistochemistry
Sample type: FFPE tissue
General Information:

Number	Component	Size	Concentration	Storage
1	PBS Buffer (powder)	2 L×2	20x	RT
2	Antigen Retrieval Buffer	20 ml	100x	2-8°C
3	Endogenous Peroxidase Blocking Buffer	3 ml	RTU	2-8°C, protect from light
4	Blocking Buffer	3 ml	RTU	2-8°C
5	Primary Antibody (Human Cytokeratin 19 Mouse mAb)	6 ml	RTU	2-8°C
6	Secondary Antibody (AffiniPure Goat Anti-Mouse IgG H&L / HRP)	6 ml	RTU	2-8°C
7	Chromogen Component A	0.3 ml	RTU	-20°C, protect from light
8	Chromogen Component B	0.3 ml	RTU	-20°C
9	Counter Staining Reagent	5 ml	RTU	RT
10	Mounting Media	5 ml	RTU	RT
11	Control slide (Human colon cancer)	1 slide	RTU	RT
12	Datasheet	1 copy		

Storage and Stability: Please store components at the temperatures indicated on the individual tube labels. The kit is stable for 6 months from the date of receipt.

Immunohistochemistry Protocol:

1. Deparaffinization And Rehydration

Immerse slides in fresh xylene for 15 minutes and then repeat two more times using separate containers. Immerse slides sequentially in 100%, 95%, 90%, 80%, and 70% ethanol solutions for 5 minutes each. Rinse slides 3 times with distilled water for 5 minutes each.

2. Antigen Retrieval

Add 100× **Antigen Retrieval Buffer** into distilled water to prepare a 1× solution. Boil slides in 1× solution at 95°C-100°C for 15 minutes. Move the slides to 1× solution at room temperature (RT) and allow them to stand for 20 minutes. Rinse 3 times with **PBS Buffer** (dissolve the powder in 2L distilled water) for 5 minutes each.

3. **Block Endogenous Peroxidase**

Drain the liquid off the slides and then use a hydrophobic IHC pen to draw circles on the slides around tissue sections. Add 2-4 drops of **Endogenous Peroxidase Blocking Buffer** directly on slides, covering the whole tissue and block slides for 15 minutes at RT. Rinse 3 times with **PBS Buffer** for 5 minutes each.

4. **Serum Blocking**

Block with 2-4 drops of **Blocking Buffer** for 20 minutes at RT.

5. **Primary Antibody Incubation**

Drain blocking buffer from slides. Incubate slides with 2-4 drops of **Human Cytokeratin 19 Mouse mAb** overnight at 4°C or 1-2 hours at RT. Rinse 3 times with **PBS Buffer** for 5 minutes each.

6. **Secondary Antibody Incubation**

Incubate slides with 2-4 drops of **AffiniPure Goat Anti-Mouse IgG H&L / HRP** for 1-2 hours at RT. Rinse slides 3 times with **PBS Buffer** for 5 minutes each.

7. **Signal Development**

Remove residual liquid around the tissue section. Add 50ul fresh **DAB Buffer (Chromogen Component A : Chromogen Component B : PBS Buffer=1:1:18)** to cover the tissue. Monitor the reaction under the microscope until a brown color is visible (approximate 3-5 minutes at RT). Stop reaction immediately by rinsing with distilled water. Rinse slides 3 times with distilled water for 5 minutes each.

8. **Counterstain**

Counterstain with an appropriate amount of **Counter Staining Reagent** for 3-5 minutes at RT. Rinse slides with distilled water for 5 minutes. Use 2-4 drops of **Differentiation reagent** to cover the tissue for 30 seconds. Rinse slides twice with distilled water for 5 minutes each.

9. **Dehydration Sheet**

Immerse slides sequentially in 70%, 80%, 90%, 95%, and 100% ethanol for 5 minutes each at RT. Immerse slides in 2 changes of fresh xylene, 15 minutes each. Drop some **Mounting Media** on the tissue. Mount coverslips.

Notes:

1. The positive control slide provided in the kit allows you to be sure that the experimental set-up is working properly.
2. Do not allow slides to dry at any time during this procedure.

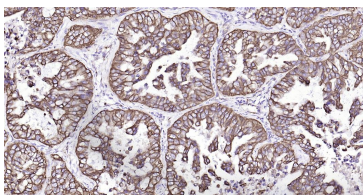
3. Please don't replace the matching reagents in this product with other manufacturers' products.
4. As DAB is a carcinogen, please take necessary precautions.
5. PBS (reagent 1) can be stored for one week at 4°C after preparation; The antigen retrieval buffer (1× reagent 2) and the chromogenic agent (the mixture of reagents 7 and 8) should be prepared right before each assay.

Please cite this product as "IHC0192H, Bioss Antibodies". Citation example: "Human Tissue sections using KRT19 IHC Kit (IHC0192H, Bioss Antibodies) were stained for KRT19 according to the manufacturer's instructions."

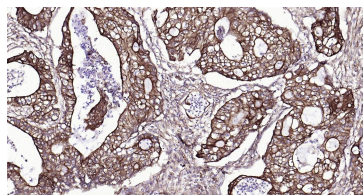
Introduction:

Cytokeratin 19 is part of a subfamily of intermediate filament proteins and are characterized by a remarkable biochemical diversity, represented in human epithelial tissues by at least 20 different polypeptides. Cytokeratins range in molecular weight from 40-68 kDa and isoelectric pH between 4.9 - 7.8. The individual human cytokeratins are numbered 1 to 20. The various epithelia in the human body usually express cytokeratins which are not only characteristic of the type of epithelium, but also related to the degree of maturation or differentiation within an epithelium. Cytokeratin subtype expression patterns are used to an increasing extent in the distinction of different types of epithelial malignancies. The cytokeratin antibodies are not only of assistance in the differential diagnosis of tumors using immunohistochemistry on tissue sections, but are also a useful tool in cytopathology and flow cytometric assays. For example, cytokeratin 19 is not expressed in hepatocytes, therefore, it is useful in the identification of liver metastasis.

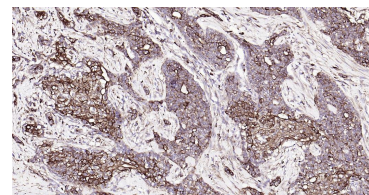
Validation Data



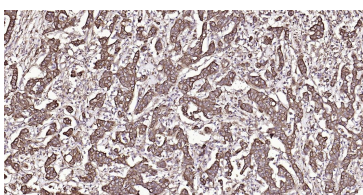
Immunohistochemical analysis of paraffin embedded human lung cancer tissue slide using IHC0192H (Human Cytokeratin 19 IHC Kit).



Immunohistochemical analysis of paraffin embedded human colon cancer tissue slide using IHC0192H (Human Cytokeratin 19 IHC Kit).



Immunohistochemical analysis of paraffin embedded human breast cancer tissue slide using IHC0192H (Human Cytokeratin 19 IHC Kit).



Immunohistochemical analysis of paraffin embedded human gastric cancer tissue slide using IHC0192H (Human Cytokeratin 19 IHC Kit).