

Human Vimentin Ready-To-Use IHC Kit

Cat. No.: IHC0113H

Size: 50T (including a control slide)

Sample Type: FFPE tissue

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.

General Information

Number	Component	50T	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
4	Blocking Buffer	3 ml	RTU	2-8°C
5	Primary Antibody (Human Vimentin Mouse mAb)	6 ml	RTU	2-8°C
6	Secondary Antibody (HRP-Goat anti-Mouse IgG pAb)	6 ml	RTU	2-8°C
7	Chromogen Component A	0.3 ml	RTU	-20°C
8	Chromogen Component B	0.3 ml	RTU	-20°C
9	Counter Staining Reagent	5 ml	RTU	RT
10	Differentiation Reagent	6 ml	RTU	RT
11	Mounting Media	5 ml	RTU	RT
12	Control slide (Human cervical carcinoma)	1 slide	RTU	RT
13	Datasheet	1 copy		

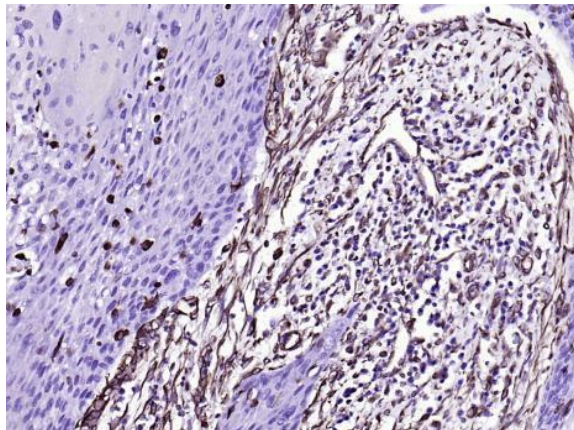
Background

Vimentin is a developmentally regulated intermediate filament protein (IFP) found in cells of mesenchymal origin. It is believed to be involved with the intracellular transport of proteins between the nucleus and plasma membrane. Unlike other IFP proteins, vimentin is expressed, along with desmin, during the early stages of cellular development. During the development process, vimentin is exchanged for new, tissue-specific IFPs. Vimentin has been implicated to be involved in the rate of steroid synthesis via its role as a storage network for steroidogenic cholesterol containing lipid droplets. Vimentin phosphorylation by a protein kinase causes the breakdown of intermediate filaments and activation of an ATP and myosin light chain dependent contractile event. This results in cytoskeletal changes that facilitate the interaction of the lipid droplets within mitochondria, and subsequent transport of cholesterol to the organelles leading to an increase in steroid synthesis.

Synonyms

FLJ36605; OTTHUMP00000019224; VIM; VIME_HUMAN; Vimentin.

Validation Data



Immunohistochemical analysis of paraffin embedded human cervical carcinoma tissue slide using IHC0113H (Human Vimentin IHC Kit).

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 Vimentin 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 **HRP-Goat anti-Mouse IgG pAb** 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.

Important Note: This product as supplied is intended for research use only, not for use in human, therapeutic or diagnostic applications.

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 "IHC0113H, Bioss Antibodies". Citation example: "Human tissue sections using Human Vimentin IHC Kit (IHC0113H, Bioss Antibodies) were stained for Vimentin according to the manufacturer's instructions."