Antigen detection in tissues and cells is a multi-step immunohistochemical process. The initial step binds the primary antibody to its specific epitope. After labeling the antigen with a primary antibody, a secondary antibody is added to bind to the primary antibody. An enzyme label is then added to bind to the secondary antibody; this detection of the bound antibody is evidenced by a colorimetric reaction.

**Principle of Procedure:**

Vimentin recognizes a 57-60 kDa protein, which is identified as vimentin. It shows no cross-reactivity with other closely related intermediate filament proteins such as desmin and GFAP. Vimentin is the main intermediate filament protein in mesenchymal cells, and therefore is of value in the differential diagnosis of undifferentiated neoplasms including melanoma and sarcoma.

**Source:** Mouse monoclonal

**Species Reactivity:** Human

**Clone:** V9

**Isotype:** IgG1/kappa

**Antibody Category:** Melanoma

**Epitope/Antigen:** Vimentin

**Total Protein Concentration:** Call for lot specific Ig Concentration.

**Cellular Localization:** Variable

**Positive Control:** Melanoma

**Normal Tissue:** Tonsil or Colon

**Abnormal Tissue:** Melanoma, sarcoma

**Known Applications:** Immunohistochemistry (formalin-fixed paraffin-embedded tissues)

**Supplied As:** Buffer with protein carrier and preservative.

**Storage and Stability:**
Store at 2ºC to 8ºC. Do not use after expiration date printed on vial. If reagents are stored under conditions other than those specified in the package insert, they must be verified by the user. Diluted reagents should be used promptly; any remaining reagent should be stored at 2ºC to 8ºC.

**Intended Use:**
For In Vitro Diagnostic Use

**Summary and Explanation:**

Biocare protocols have been standardized using in-house antibodies, detection and accessory reagents for use on the intelliPATH FLX automated stainer. Recommended staining protocols are specified in the datasheet of the antibody of interest. Pre-optimized intelliPATH FLX protocols with preset parameters can be displayed, printed and edited according to the procedure in the operator's manual. Refer to the operator's manual for additional instruction to navigate intelliPATH FLX software and stainer. Use TBS for washing steps unless otherwise specified.

**Protocol Recommendations:**

**Pretreatment Solution (recommended):** Reveal

**Pretreatment Protocol:**

Heat Retrieval Method:
Retrieve sections under pressure using Biocare's Decloaking Chamber, followed by a wash in distilled water. Alternatively, steam tissue sections for 45-60 minutes. Allow solution to cool for 10 minutes then wash in distilled water.

**Peroxide Block:** Block for 5 minutes at RT.

**Protein Block:**

Optional: Incubate for 5-10 minutes at RT.

**Primary Antibody:** Incubate for 15-30 minutes at RT.

**Secondary:** Incubate for 7-10 minutes at RT.

**Tertiary:** Incubate for 7-10 minutes at RT.

**Chromogen:** Incubate for 5 minutes with DAB at RT.

**Counterstain:**
1. Rinse with deionized water.
2. Incubate for 5 minutes with automated Hematoxylin.
3. Rinse with TBS Buffer for 1 minute followed by a rinse with deionized water.

**Quality Statement:**

Biocare protocols have been standardized using in-house antibodies, detection and accessory reagents for use on the intelliPATH FLX automated stainer. Recommended staining protocols are specified in the datasheet of the antibody of interest. Pre-optimized intelliPATH FLX protocols with preset parameters can be displayed, printed and edited according to the procedure in the operator's manual. Refer to the operator's manual for additional instruction to navigate intelliPATH FLX software and stainer. Use TBS for washing steps unless otherwise specified.

**Performance Characteristics:**

The optimum antibody dilution and protocols for a specific application can vary. These include, but are not limited to: fixation, heat-retrieval method, incubation times, tissue section thickness and detection kit used. Due to the superior sensitivity of these unique reagents, the recommended incubation times and titers listed are not applicable to other detection systems, as results may vary. The data sheet recommendations and protocols are based on exclusive use of Biocare products. Ultimately, it is the responsibility of the investigator to determine optimal conditions. These products are tools that can be used for interpretation of morphological findings in conjunction with other diagnostic tests and pertinent clinical data by a qualified pathologist.

**Quality Control:**

Refer to NCCLS Quality Assurance for Immunocytochemistry approved guidelines, December 1999 MM4-A Vol.19 No.26 for more information about tissue controls.

**Precautions:**

This antibody contains less than 0.1% sodium azide. Concentrations less than 0.1% are not reportable hazardous materials according to U.S. 29 CFR 1910.1200, OSHA Hazard communication and EC Directive 91/155/EC.

Sodium azide (NaN3) used as a preservative is toxic if ingested. Sodium azide may react with lead and copper plumbing to form highly explosive metal azides. Upon disposal, flush with large volumes of water to prevent azide build-up in plumbing. (Center for Disease Control, 1976, National Institute of Occupational Safety and Health, 1976)

Specimens, before and after fixation, and all materials exposed to them should be handled as if capable of transmitting infection and disposed of with proper precautions. Never pipette reagents by mouth and avoid contacting the skin and mucous membranes with reagents and specimens. If reagents or specimens come in contact with sensitive areas, wash with copious amounts of water.

Microbial contamination of reagents may result in an increase in nonspecific staining. Incubation times or temperatures other than those specified may give erroneous results. The user must validate any such change. The MSDS is available upon request.
Vimentin
Prediluted Mouse Monoclonal Antibody
Control Number: 901-048IP-082010

Troubleshooting:
Follow the antibody specific protocol recommendations according to data sheet provided. If atypical results occur, contact Biocare's Technical Support at 1-800-542-2002.

Limitations and Warranty:
There are no warranties, expressed or implied, which extend beyond this description. Biocare is not liable for property damage, personal injury, or economic loss caused by this product.

References: