# VEGF

Concentrated Rabbit Monoclonal Antibody

Control Number: 901-356-081617

# Catalog Number:ACI 356 AK, BKDescription:0.1, 0.5 ml, concentrated

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Dilution:	1:100-1:200
Diluent:	Van Gogh Yellow

# Intended Use:

#### For In Vitro Diagnostic Use

VEGF [EP1176Y] is a rabbit monoclonal antibody that is intended for laboratory use in the qualitative identification of VEGF protein by immunohistochemistry (IHC) in formalin-fixed paraffin-embedded (FFPE) human tissues. The clinical interpretation of any staining or its absence should be complemented by morphological studies using proper controls and should be evaluated within the context of the patient's clinical history and other diagnostic tests by a qualified pathologist.

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#### **Summary and Explanation:**

Vascular endothelial growth factor (VEGF) is a sub-family of growth factors, more specifically of platelet-derived growth factor family of cystine-knot growth factors. They are important signaling proteins involved in both vasculogenesis (the formation of the embryonic circulatory system) and angiogenesis (the growth of blood vessels from pre-existing vasculature) (2-3). In certain cancers, high VEGF expression has been correlated with shorter survival and this provides a valuable prognostic marker (1).

# **Principle of Procedure:**

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, an enzyme labeled polymer is added to bind to the primary antibody. The detection of the bound antibody is evidenced by a colorimetric reaction.

Source: Rabbit monoclonal Species Reactivity: Human Clone: EP1176Y Isotype: IgG Total Protein Concentration: ~10 mg/ml. Call for lot specific Ig concentration. Epitope/Antigen: VEGF Cellular Localization: Membrane and / or cytoplasmic Positive Control: Tonsil, some breast and ovarian cancers Known Applications: Immunohistochemistry (formalin-fixed paraffin-embedded tissues) Supplied As: Buffer with protein carrier and preservative Van Gogh Yellow (PD902) Storage and Stability:

Store at 2°C to 8°C. 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 2°C to 8°C.

# **Protocol Recommendations:**

**Peroxide Block:** Block for 5 minutes with Biocare's Peroxidazed 1.

#### Pretreatment Solution (recommended): Diva

Pretreatment Protocol: N/A

## Heat Retrieval Method:

Preheat the retrieval solution to 95°C for 30 minutes in Biocare's Decloaking Chamber. Then, place slides into the preheated solution and retrieve under pressure at 95°C for 40 minutes; alternatively, steam tissue sections for 45-60 minutes or use a water bath at 95°C for 40 minutes. Allow solution to cool for 20 minutes then wash in distilled water.

# Protocol Recommendations Cont'd:

**Protein Block (Optional):** Incubate for 5-10 minutes at RT with Biocare's Background Punisher.

Primary Antibody: Incubate for 30-60 minutes at RT.

# Probe: N/A

Polymer: Incubate for 30 minutes at RT with a secondary-conjugated polymer.

## Chromogen:

Incubate for 5 minutes at RT with Biocare's DAB -OR- Incubate for 5-7 minutes at RT with Biocare's Warp Red.

#### **Counterstain:**

Counterstain with hematoxylin. Rinse with deionized water. Apply Tacha's Bluing Solution for 1 minute. Rinse with deionized water.

#### **Technical Note:**

This antibody has been standardized with Biocare's MACH 2 detection system. It can also be used on an automated staining system and with other Biocare polymer detection kits. Use TBS buffer for washing steps.

# Limitations:

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. The clinical interpretation of any positive or negative staining should be evaluated within the context of clinical pathologist. The clinical interpretation of any positive or negative staining should be complemented by morphological studies using proper positive and negative internal and external controls as well as other diagnostic tests.

## **Quality Control:**

Refer to CLSI Quality Standards for Design and Implementation of Immunohistochemistry Assays; Approved Guideline-Second edition (I/LA28-A2) CLSI Wayne, PA USA (www.clsi.org). 2011

#### **Precautions:**

1. This antibody contains less than 0.1% sodium azide. Concentrations less than 0.1% are not reportable hazardous material according to U.S. 29 CFR 1910.1200, OSHA Hazard communication and EC Directive 91/155/EC. Sodium azide (NaN<sub>3</sub>) 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) (4)

2. 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. (5)

3. Microbial contamination of reagents may result in an increase in nonspecific staining.

4. Incubation times or temperatures other than those specified may give erroneous results. The user must validate any such change.

5. Do not use reagent after expiration date printed on the vial.

6. The SDS is available upon request and is located at http://biocare.net/.

Biocare Medical 60 Berry Drive Pacheco, CA 94553 USA





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Control Number: 901-356-091014

# **References:**

1. Saad RS, *et al.* Lymphatic microvessel density as prognostic marker in colorectal cancer. Modern Pathol. 2006 Oct; 19(10):1317-23.

2. Lee AH, *et al.* Invasive lobular and invasive ductal carcinoma of the breast show distinct patterns of vascular endothelial growth factor expression and angiogenesis. J Pathol. 1998 Aug; 185(4):394-401.

3. Applanat MP, *et al.* Vascular endothelial growth factor is a target gene for estrogen receptor and contributes to breast cancer progression. Adv Exp Med Biol. 2008; 617:437-44.

4. Center for Disease Control Manual. Guide: Safety Management, NO. CDC-22, Atlanta, GA. April 30, 1976 "Decontamination of Laboratory Sink Drains to Remove Azide Salts."

5. Clinical and Laboratory Standards Institute (CLSI). Protection of Laboratory Workers from Occupationally Acquired Infections; Approved guideline-Fourth Edition CLSI document M29-A4 Wayne, PA 2014.

Manufactured incorporating RabMAb® technology under Epitomics US Patents, No 5,675,063 and 7,429,487, owned by Abcam.

#### **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.



