p120 + E-cadherin

Prediluted Multiplex Antibody Reagent
901-3011DS-061919

BIOCARE MEDICAL

Catalog Number:	**API 3011DS AA**	**VLTMR 3011 G20**
Description:	6.0 mL, RTU	20 mL, RTU
Dilution:	Ready-to-use	Ready-to-use
Diluent:	N/A	N/A

Intended Use:
For In Vitro Diagnostic Use
p120 + E-cadherin is intended for laboratory use in the qualitative identification of p120 and E-cadherin proteins 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 patients clinical history and other diagnostic tests by a qualified pathologist.

Summary and Explanation:
Diagnostic reproducibility of lobular vs. ductal lesions, based on histology alone, is less than optimal. The proper distinction between atypical lobular hyperplasia, lobular carcinoma *in situ* and low-grade ductal carcinoma *in situ* is critical for patient management. Studies have shown E-cadherin, a negative membrane marker for lobular neoplasia, is useful in the distinction of ductal neoplasia vs. lobular; however as a negative marker for lobular carcinoma, it can be difficult to interpret, particularly in challenging cases (2). Studies have shown accurate categorization of ductal vs. lobular neoplasia in the breast with a cocktail of p120 + E-cadherin and helped give further clarification in the separation of low-grade ductal carcinoma *in situ* from lobular neoplasia. Of other tumors that may morphologically mimic lobular carcinoma, diffusely infiltrating variants of rectal and gastric carcinomas have shown diffuse cytoplasmic p120 immunostaining (1-2). This Multiplex IHC stain may enable the identification of the presence and the extent of the lobular lesions due to its bright pink color in lobular lesions, and therefore may aid in providing a more accurate diagnosis.

Principle of Procedure:
This product is a primary antibody cocktail of mouse and rabbit antibodies, which may be used in a Multiplex IHC staining procedure to produce a two-color stain. Following application of the primary antibody cocktail to the tissue sample, detection is performed by separate secondary antibodies specific for each species (i.e. mouse or rabbit) of the primary antibody cocktail, which are conjugated to horseradish peroxidase (HRP) or alkaline phosphatase (AP) enzymes. Visualization is accomplished by the application of chromogenic substrates (DAB and Red), which are enzymatically activated (by HRP or AP, respectively) to produce a colored reaction product at the antigen site. The specimen may be counterstained and coverslipped. Results are interpreted using a light microscope.

Reagent Provided:
p120 + E-cadherin is provided as a prediluted antibody cocktail of anti-p120 and anti-E-cadherin antibodies, in buffer with carrier protein and preservative.

Antibody	anti-p120	anti-E-cadherin
Clone	98/pp120	EP6*
Source	Mouse monoclonal	Rabbit monoclonal
Isotype	IgG1	IgG
Epitope/Antigen	p120 catenin	E-cadherin
Cellular Localization	Cytoplasmic	Membrane
Staining	Red	Brown (DAB)

*Previously known as EP700Y

Storage and Stability:
Store at 2ºC to 8ºC. The product is stable to the expiration date printed on the label, when stored under these conditions.

Known Applications:
Immunohistochemistry (formalin-fixed paraffin-embedded tissues)
Species Reactivity: Human; others not tested
Positive Tissue Control: Breast cancer

Protocol Recommendations (VALENT® Automated Slide Staining Platform):
VLTMR3011 is intended for use with the VALENT. Refer to the User Manual for specific instructions for use. Protocol parameters in the Protocol Manager should be programmed as follows:
Deparaffinization: Deparaffinize for 8 minutes with Val DePar.
Pretreatment: Perform heat retrieval at 98°C for 60 minutes using Val AR-Hi pH, 5X (use at 1X).
Peroxidase Block: Block for 5 minutes with Val Peroxidase Block.
Protein Block: Incubate for 10 minutes with Val Background Block.
Primary Antibody: Incubate for 45 minutes.
Double Stain Detection: Incubate for 30 minutes using Val Plex 1.
Chromogen (1): Incubate for 5 minutes with Val DAB.
Chromogen (2): Incubate for 15 minutes with Val Fast Red.
Counterstain: Counterstain for 5 minutes with Val Hematoxylin.

Protocol Recommendations (intelliPATH FLX® and manual use):
Peroxide Block: Block for 5 minutes with Peroxidazed 1.
Pretreatment: Perform heat retrieval using Diva Decloaker. Refer to the Diva Decloaker product data sheet for specific instructions.
Protein Block: Incubate for 10 minutes at RT with Background Punisher.
Primary Antibody: Incubate for 30 minutes at RT.
Double Stain Detection: Incubate for 30 minutes at RT using MACH 2 Double Stain 1.
Chromogen (1): Incubate for 5 minutes at RT with Betazoid DAB.
Chromogen (2): Incubate for 5-7 minutes at RT with Warp Red. Rinse in deionized water.
Counterstain: Counterstain with hematoxylin. Rinse with deionized water. Apply Tacha's Bluing Solution for 1 minute. Rinse with deionized water.

Technical Notes:
This antibody, for intelliPATH FLX and manual use, has been standardized with MACH 2 Double Stain 1. 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.

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

Biocare Medical
60 Berry Drive
Pacheco, CA 94553
USA

IVD CE

Rev: 062117

EC REP EMERGO EUROPE
Prinsessegracht 20
2514 AP The Hague
The Netherlands

Tel: 800-799-9499 | www.biocare.net | Fax: 925-603-8080

p120 + E-cadherin

Prediluted Multiplex Antibody Reagent
901-3011DS-061919

BIOCARE MEDICAL

Precautions:

1. 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 (NaN_3) 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 into 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 the expiration date printed on the vial.
6. The SDS is available upon request and is located at http://biocare.net.

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.

References:

1. Esposito NN, Chivukula M, Dabbs DJ. The ductal phenotypic expression of the E-cadherin/catenin complex in tubulolobular carcinoma of the breast: an immunohistochemical and clinicopathologic study. Mod Pathol. 2007 Jan; 20(1):130-8.
2. Dabbs DJ, Bhargava R, Chivukula M. Lobular versus ductal breast neoplasms: the diagnostic utility of p120 catenin. Am J Surg Pathol. 2007 Mar; 31(3):427-37.
3. Bellovin DI, *et al*. Altered localization of p120 catenin during epithelial to mesenchymal transition of colon carcinoma is prognostic for aggressive disease. Cancer Res. 2005 Dec 1; 65(23):10938-45.
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.

Produced using Abcam's RabMAb® technology. RabMAb® technology is covered by the following U.S. Patents, No. 5,675,063 and/or 7,429,487.

Biocare Medical
60 Berry Drive
Pacheco, CA 94553
USA

IVD CE

Rev: 062117

EC REP EMERGO EUROPE
Prinsessegracht 20
2514 AP The Hague
The Netherlands

Tel: 800-799-9499 | www.biocare.net | Fax: 925-603-8080