p63 + P504S

Prediluted Antibody Cocktail
902-201-072122

BIOCARE MEDICAL

Available Product Formats				
Format	Catalog Number	Description	Dilution	Diluent
Predilute	PPM 201 AA, H	6.0, 25 mL	Ready-to-use	N/A
intelliPATH FLX	IPR 201 G10	10 mL	Ready-to-use	N/A
ONCORE Pro	OPAR 201 T60	60 tests	Ready-to-use	N/A
UltraLine – For BenchMark	VP 201 G, G25	6.0, 25 mL	Ready-to-use	N/A
Q Series– For Leica BOND-III	ALR 201 G7	7.0 mL	Ready-to-use	N/A

Intended Use:
For Research Use Only. Not for use in diagnostic procedures.

Summary and Explanation:
p63, a homolog of the tumor suppressor p53, has been identified in proliferating basal cells in the epithelial layers of a variety of tissues, including epidermis, cervix, urothelium and prostate (1). p63 was detected in nuclei of the basal epithelium in normal prostate glands; however, it was not expressed in malignant tumors of the prostate (2). P504S, also known as α-methylacyl coenzyme A racemase (AMACR), is a peroxisomal and mitochondrial enzyme that plays a role in bile acid synthesis and β-oxidation of branched chain fatty acids (3). P504S was initially identified from a cDNA library as a gene that is overexpressed in human prostate cancer; with little or no expression in normal prostate (4,5).

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, a one-step or two-step detection procedure can be applied. A one-step procedure will feature an enzyme labeled polymer that binds the primary antibody. A two-step procedure will feature a linker antibody added to bind to the primary antibody. An enzyme-labeled polymer is then added to bind the linker antibody. These detections of the bound antibodies are evidenced by a colorimetric reaction.

Reagent Provided:
p63 + P504S is provided as a prediluted antibody cocktail of anti-p63 and anti-P504S antibodies, in buffer with carrier protein and preservative.

Antibody	anti-p63	anti-P504S
Clone	4A4	N/A
Source	Mouse Monoclonal	Rabbit Polyclonal
Isotype	IgG2a/kappa	IgG
Epitope/ Antigen	p63	P504S
Cellular Localization	Nuclear	Granular Cytoplasm
Staining	Brown (DAB)	Brown (DAB)

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. Do not use after expiration date.

Known Applications:
Immunohistochemistry (formalin-fixed paraffin-embedded tissues)
Species Reactivity: Human, others not tested
Positive Tissue Control: Normal prostate and prostate adenocarcinoma

Staining Protocol Recommendations (intelliPATH FLX® and manual use):
Peroxide Block: Block for 5 minutes with Peroxidazed 1.
Pretreatment: Perform heat retrieval using Reveal Decloaker. Refer to the Reveal 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.
Probe: Incubate for 10 minutes at RT with a secondary probe.
Polymer: Incubate for 10-20 minutes at RT with a tertiary polymer.
Chromogen: Incubate for 5 minutes at RT with Betazoid DAB -OR- 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.

intelliPATH FLX Automated Slide Stainer:
IPR201 is intended for use with the intelliPATH FLX. Refer to the User Manual for specific instructions for use. When using the intelliPATH FLX, peroxide block with intelliPATH FLX Peroxidase Blocking Reagent (IPB5000) may be performed following heat retrieval.

Technical Note:
This antibody, for intelliPATH FLX and manual use, has been standardized with MACH 4 detection system. Use TBS for washing steps.

Staining Protocol Recommendations (ONCORE™ Pro Automated Slide Staining System):
OPAR201 is intended for use with the ONCORE Pro. Refer to the User Manual for specific instructions for use. Protocol parameters in the Protocol Editor should be programmed as follows:
Protocol Name: p63 + P504S
Protocol Template (Description): Special (Univ. HRP Detection Cat# OPRI6062 required)
Dewaxing (DS Buffer Option): DS2-50
Antigen Retrieval (AR Option): AR2, low pH; 101°C
Block Option: Buffer
Reagent Name, Time, Temp.: p63+P504S, 30 min., 25°C

Staining Protocol Recommendations (Ventana BenchMark ULTRA):
VP201 is intended for use with the BenchMark ULTRA. Refer to the User Manual for specific instructions for use. Recommended protocol parameters are as follows:
Template/Detection: ultraView DAB
Pretreatment Protocol: CC1 Standard
Primary Antibody: 32 minutes, 37°C
ultraBlock (V-Blocker BRI4001): Incubate for 4 minutes (with appropriate Option # registered by user)
V-Blocker is recommended to be applied prior to any detection system. Protocol Recommendation for Ventana BenchMark XT available upon request.

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Staining Protocol Recommendations (Q Series – For Leica BOND-III):

ALR201 is intended for use with the Leica BOND-III. Refer to the User Manual for specific instructions for use. Recommended protocol parameters are as follows:

Protocol Name: IHC Protocol F
Detection: Bond Polymer Refine
HIER: 10 min with ER1
Peroxide Block: 5 min
Marker (Primary Antibody): 15 min
Post Primary: 8 min
Polymer: 8 min
Mixed DAB Refine: 10 min
Hematoxylin: 5 min

Limitations:

This product is provided for Research Use Only (RUO) and is not for use in diagnostic procedures. Suitability for specific applications may vary and it is the responsibility of the end user to determine the appropriate application for its use.

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) (6)
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. (7)
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.

Technical Support:

Contact Biocare's Technical Support at 1-800-542-2002 for questions regarding this product.

References:

1. Yang A, *et al*. p63, a p53 Homolog at 3q27–29, Encodes Multiple Products with Transactivating, Death-Inducing, and Dominant- Negative Activities. Mol Cell. 1998 Sep; 2(3):305-16.
2. Signoretti S, *et al*. p63 Is a Prostate Basal Cell Marker and Is Required for Prostate Development. Am J Pathol. 2000 Dec; 157(6):1769-75.
3. Ferdinandusse S, *et al*. Subcellular localization and physiological role of α-methylacyl-CoA racemase. J Lipid Res. 2000 Nov; 41(11):1890-6.
4. Xu J, et al. Identification of Differentially Expressed Genes in Human Prostate Cancer Using Subtraction and Microarray. Cancer Res. 2000 Mar 15; 60(6):1677-82.
5. Rubin MA, *et al*. α-Methylacyl Coenzyme A Racemase as a Tissue Biomarker for Prostate Cancer. JAMA. 2002 Apr 3; 287(13):1662-70.
6. 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."

References Cont'd:

7. 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.

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