**p63**

Concentrated and Prediluted Monoclonal Antibody

901-163-031519

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<tbody>
<tr>
<td>Description:</td>
<td>0.1, 0.5, 1.0 mL, conc.</td>
<td>6.0, 25 mL, RTU</td>
<td>10 mL, RTU</td>
<td>60 tests, RTU</td>
<td>6.0, 25 mL, RTU</td>
<td>20 mL, RTU</td>
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<tr>
<td>Dilution:</td>
<td>1:100</td>
<td>Ready-to-use</td>
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<tr>
<td>Diluent:</td>
<td>Van Gogh Yellow</td>
<td>N/A</td>
<td>N/A</td>
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**Intended Use:**
For In Vitro Diagnostic Use

p63 [4A4] is a mouse monoclonal antibody that is intended for laboratory use in the qualitative identification of p63 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.

**Summary and Explanation:**
p63, a homolog of the tumor suppressor p53, has been identified in basal cells in the epithelial layers of a variety of tissues, including epidermis, cervix, urothelium, breast 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). As a result, p63 has been reported as a useful marker for differentiating benign from malignant lesions in the prostate, particularly when used in combination with markers of high molecular weight cytokeratins and the prostate-specific marker AMACR (P504S) (3-4).

p63 has also been shown to be a sensitive marker for lung squamous cell carcinomas (SqCC), with reported sensitivities of 80-100% (5-8). Specificity for lung SqCC vs. lung adenocarcinoma (LADC), has been reported to be approximately 70-90%, as positive staining with p63 has been typically observed in 10-30% of LADC cases (5-8).

In breast tissue, p63 has been identified in myoepithelial cells of normal ducts (9). Reports have described the utility of p63 in a panel of IHC markers for the assessment of breast lesions, due to the differential expression of the luminal vs. basal and myoepithelial markers (9-11).

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

**Source:** Mouse monoclonal

**Species Reactivity:** Human, mouse and rat

**Clone:** 4A4

**Isotype:** IgG2a/kappa

**Protein Concentration:** Call for lot specific Ig concentration

**Epitope/Antigen:** p63

**Cellular Localization:** Nuclear

**Positive Tissue Control:** Normal prostate

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

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

**Storage and Stability:** Store at 2°C to 8°C. The product is stable to the expiration date printed on the label, when stored under these conditions. Do not use after

**Storage and Stability Cont’d:** expiration date. Diluted reagents should be used promptly; any remaining reagent should be stored at 2°C to 8°C.

**Protocol Recommendations (VALENT® Automated Slide Staining Platform):**
VLTM163 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:

- **Deparaaffinization:** 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 (Optional):** Incubate for 10-20 minutes with Val Background Block.
- **Primary Antibody:** Incubate for 20 minutes.
- **Secondary:** Incubate for 10 minutes with Val Mouse Secondary.
- **Linker:** Incubate for 10 minutes with Val Universal Linker.
- **Polymer:** Incubate for 10 minutes with Val Universal Polymer.
- **Chromogen:** Incubate for 5 minutes with Val DAB.
- **Counterstain:** Counterstain for 5 minutes with Val Hematoxylin.

**Protocol Recommendations (intelliPATH FLX® and manual use):**
- **Peroxidase Block:** Block for 5 minutes with Peroxidazed 1.
- **Pretreatment:** Perform heat retrieval with Reveal Decloaker. Refer to the Reveal Decloaker product data sheet for specific instructions.
- **Protein Block (Optional):** Incubate for 5-10 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 Biocare’s DAB – OR – Incubate for 5-7 minutes at RT with Warp Red.
- **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:**
IP163 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 (IPBS000) 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.

**Protocol Recommendations (ONCORE™ Automated Slide Staining System):**
OA1163 is intended for use with the ONCORE. Refer to the User Manual for specific instructions for use. Protocol parameters in the Protocol Editor should be programmed as follows:

- **Protocol Name:** p63
- **Protocol Template (Description):** Ms HRP Template 1
- **Dewaxing (DS Option):** DS2
- **Antigen Retrieval (AR Option):** AR1, high pH; 101°C
- **Reagent Name, Time, Temp.:** p63, 30 min., 25°C
Protocol Recommendations (Ventana BenchMark XT / ULTRA):

VP163 is intended for use with the BenchMark XT / ULTRA. Refer to the User Manual for specific instructions for use. Recommended protocol parameters are as follows:

- Using ultraView on XT / ULTRA:
  - Template/Detection: ultraView DAB
  - Pretreatment Protocol: ULTRA 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.

- Using OptiView on XT:
  - Template/Detection: OptiView DAB IHC
  - Pretreatment Protocol: CC1 64 minutes
  - Peroxidase: Pre Primary Peroxidase Inhibitor
  - Primary Antibody: 32 minutes, 36°C

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:

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 (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) (12)
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. (13)
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:

References Cont’d:

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