Prostate Cocktail-2X (CK5 + CK14 + p63)

Prediluted Antibody Cocktail

Control Number: 902-364-081517

Catalog Number:	**APR 364 AAK, HK, JJK**	**IPR 364 G10**
Description:	6.0, 25, 50 ml, prediluted	10 ml, prediluted
Dilution:	Ready-to-use	Ready-to-use
Diluent:	N/A	N/A

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

Summary and Explanation:
CK5 and CK14 are high molecular weight cytokeratins expressed in a variety of normal and neoplastic epithelial tissues (1). In prostate tissue, mRNA for CK5 and CK14 has been detected in the basal cells of normal glands and prostatic intraepithelial neoplasia (PIN), a precursor lesion to prostatic adenocarcinoma; however, expression of CK5 or CK14 was not identified in invasive prostatic adenocarcinoma (2).

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 (3). p63 was detected in nuclei of the basal epithelium in normal prostate glands; however, it was not expressed in malignant tumors of the prostate (4).

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 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.

Reagent Provided:
Prostate Cocktail – 2X (CK5 + CK14 + p63) is provided as a prediluted antibody cocktail of anti-CK5, anti-CK14, and anti-p63 antibodies, in buffer with carrier protein and preservative, with additional dropper bottle (DB364).

Antibody	anti-CK5	anti-CK14	anti-p63
Clone	XM26	LL002	4A4
Source	Mouse monoclonal	Mouse monoclonal	Mouse monoclonal
Isotype	IgG1/kappa	IgG3	IgG2a/kappa
Epitope/ Antigen	CK5	CK14	p63
Cellular Localization	Cytoplasmic	Cytoplasmic	Nuclear

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.

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

Species Reactivity: Human

Positive Control: Normal prostate

Total Protein Concentration: ~10 mg/ml. Call for lot specific Ig concentration.

Staining Protocol Recommendations (intelliPATH and manual use):

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

Pretreatment: Perform heat retrieval using Biocare's Diva Decloaker. Refer to the Diva Decloaker product data sheet for specific instructions.

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

Primary Antibody: Incubate for 10-20 minutes at RT.

Probe: Incubate for 10 minutes at RT with a secondary probe.

Polymer: Incubate for 5-10 minutes at RT with a tertiary polymer.

Staining Protocol Recommendations (intelliPATH and manual use) Cont'd:

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.

intelliPATH™ Automated Slide Stainer:
IPR364 is intended for use on the intelliPATH™ Automated Slide Stainer. Refer to the intelliPATH Automated Slide Stainer manual for specific instructions on its use. When using the intelliPATH, peroxide block with intelliPATH Peroxidase Blocking Reagent (IPB5000) may be performed following heat retrieval.

Technical Note:
This antibody has been optomized for use with Biocare's MACH 4 Universal HRP-Polymer Detection and intelliPATH Universal HRP Detection Kit. Use TBS buffer for washing steps.

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) (11)
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. (12)
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. Moll R, *et al.* The catalog of human cytokeratins: patterns of expression in normal epithelia, tumors and cultures cells. Cell. 1982 Nov;31(1):11-24.
2. Yang Y, *et al.* differential expression of cytokeratin mRNA and protein in normal prostate, prostatic intraepithelial neoplasia, and invasive carcinoma. Am J Pathol. 1997 Feb;150(2):693-704.
3. 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.
4. 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.
5. Tacha DE, Miller RT. Use of p63/P504S monoclonal antibody cocktail in immunohistochemical staining of prostate tissue. Appl Immunohistochem Mol Morphol. 2004 Mar;12(1):75-8. Biocare Medical, Walnut Creek, California.
6. Beach R, *et al*. P504S immunohistochemical detection in 405 prostatic specimens including 376 18-gauge needle biopsies. Am J Surg Pathol. 2002 Dec;26 (12):1588-96.
7. Luo J, *et al.* Alpha-methylacyl-CoA racemase: a new molecular marker for prostate cancer. Cancer Res. 2002 Apr 15;62(8):2220-6.

Biocare Medical
60 Berry Drive
Pacheco, CA 94553
USA

Prostate Cocktail-2X (CK5 + CK14 + p63)

Prediluted Antibody Cocktail

Control Number: 902-364-081517

References cont'd:

8. Wang Y, *et al*. Cell differentiation lineage in the prostate. Differentiation. 2001 Oct;68(4-5):270-9.
9. Tokar EJ, *et al.* Stem/progenitor and intermediate cell types and the origin of human prostate cancer. Differentiation. 2005 Dec;73(9-10):463-73.
10. Collins AT, *et al.* Identification and isolation of human prostate epithelial stem cells based on alpha(2)beta(1)-integrin expression. J Cell Sci. 2001 Nov;114(Pt 21):3865-72.
11. 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."
12. 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.

Biocare Medical
60 Berry Drive
Pacheco, CA 94553
USA