

Pan-Basal Cell Cocktail

Mouse anti-p63, Mouse anti-Cytokeratin 5 and Mouse anti-Cytokeratin 14

Cat. No.: CO005K (1 ml Concentrate); CO005K-05 (0.5 ml Concentrate);
COG005 (6 ml Ready-to-use)

Instructions for use

Intended use

This antibody cocktail is designed for the specific localisation of basal cell layers in certain human epithelia in formalin-fixed, paraffin-embedded tissue sections. Pan-Basal Cell Cocktail is intended for research use only.

Specifications

Specificity:	p63, Cytokeratin 5 and Cytokeratin 14
Immunogens:	p63: recombinant protein according to the N-terminus of human p63 Cytokeratin 5: recombinant protein according to the C-terminus of human CK5 Cytokeratin 14: synthetic peptide according to the C-terminus of human CK14
Clone:	4A4 (p63), XM26 (Cytokeratin 5) and LL002 (Cytokeratin 14)
Isotypes:	Mouse IgG2a (p63), mouse IgG1 (Cytokeratin 5) and mouse IgG3 (Cytokeratin 14)
Species reactivity:	human+, others not tested

Summary and description

p63 is a reliable marker for detection of basal cell layers in benign prostate glands. Like the cytokeratins of high molecular weight (34 β E12, CK 5/6, CK 5/14) p63 is only detectable in benign glands and not in areas of the carcinoma. Anti-p63 antibody stains positive in nuclei of benign glands and shows a sometimes higher sensitivity than i.e. 34 β E12 (Weinstein et al. 2002, Shah et al. 2002).

Cytokeratins 5 and 14 are, like cytokeratin 6, very reliable markers for squamous cell epithelia. Using antibody cocktails against these cytokeratins is helpful for the detection of basal cell layers and myoepithelia. Their use is well established for differential diagnosis of atypical proliferations and DCIS of the breast (Böcker et al. 1997, Bánkfalvi et al. 2004).

The detection of basal cell layers with a combination of antibodies against p63 and high molecular cytokeratins is particularly reliable (Zhou et al. 2003).

Reagent provided

Mixture of mouse monoclonal antibodies in buffer with carrier protein and preservative for stabilisation in the formats:

Concentrate:	1 ml	(Cat. No. CO005K)
Concentrate:	0.5 ml	(Cat. No. CO005K-05)
Ready-to-use:	6 ml	(Cat. No. COG005)

Dilution of primary antibody

Dilution of Zytomed Systems' concentrated antibody depends on the detection system used. The final working dilution must always be determined by the user. The elaboration of staining protocol should be done by an experienced specialist. For Zytomed Systems' recommendations see chapter 'Staining procedure'.

Storage and handling

The antibody cocktail should be stored at 2-8°C without further dilution.

If necessary, dilutions of the antibody should be done in a suitable antibody dilution buffer (e.g. ZUC025 from Zytomed Systems). The diluted antibody should be stored at 2-8°C after use. Stability of this working solution depends on various parameters and has to be confirmed by appropriate controls.

The antibody cocktail provided is suitable for use until the expiry date indicated on the label, if stored at 2-8°C. Do not use product after the expiry date. Positive and negative controls should be run simultaneously with all specimens. If unexpected staining is observed which cannot be explained by variations in laboratory procedures and a problem with the antibody is suspected, contact Zytomed Systems' technical support or your local distributor.

Precautions

Use through qualified personnel only. Wear protective clothing to avoid contact of reagents and specimens with eye, skin and mucous membranes. If reagents or specimens come in contact with sensitive area, wash with large amounts of water.

Microbial contamination of the reagent must be avoided, since otherwise non-specific staining may occur. ProClin300 and sodium azide (NaN₃) are used for stabilisation. Reaction of sodium azide with lead or copper in drainage pipes can result in the formation of highly explosive metallic azides. Discard the antibody solution in a

large volume of running water to avoid formation of deposits. A material safety data sheet (MSDS) for the pure substances is available upon request.

Staining procedure

Refer to the following table for conditions specifically recommended for this antibody. Also refer to detection system data sheets for guidance on specific staining protocols or other requirements.

Parameters	Zytomed Systems recommendations
*Pre-treatment:	Heat Induced Epitope Retrieval (for example in Citrate Buffer pH 6.0 ZUC028)
*Control tissue:	Prostate
*Working dilution:	1:50 (for concentrated antibodies only)
*Incubation time:	30 - 60 minutes

Quality control

The recommended positive control tissue for this antibody is prostate. We recommend carrying out a positive and a negative control with every staining run. Please refer to the instructions of the detection system for guidance on general quality control procedures.

Troubleshooting

If you observe unusual staining or other deviations from the expected results please read these instructions carefully, refer to the instructions of the detection system for relevant information or contact your local distributor.

Expected results

The antibody against p63 stains positive in nuclei of basal cells of various epithelial tissues. The antibodies against Cytokeratins 5 and 14 stain positive in the cytoplasm of these basal cells. Interpretation of the staining results is solely the responsibility of the user. Any experimental result should be confirmed by a medically established diagnostic procedure.

Limitations of the Procedure

Immunohistochemistry is a complex technique involving both histological and immunological detection methods. Tissue processing and handling prior to immunostaining, for example variations in fixation and embedding or the inherent nature of the tissue can cause inconsistent results (Nadji and Morales, 1983). Endogenous peroxidase, alkaline phosphatase or biotin may cause non-specific staining depending on the detection system used. Tissues containing Hepatitis B Surface Antigen (HBsAg) may give false positive results with HRP (horse radish peroxidase) detection systems (Omata *et al*, 1980). Inadequate counterstaining and mounting can influence the interpretation of the results.

Zytomed Systems warrants that the product will meet all requirements described from its shipping date until the expiry date is reached, if the product is stored and utilised as recommended. No additional guarantees can be given. Under no circumstances shall Zytomed System be liable for any damages arising out of the use of the reagent provided.

Performance characteristics

Zytomed Systems has conducted studies to evaluate the performance of the antibody for use with a standard detection system. The product has been found to be sensitive and specific to the antigen of interest with minimal or no cross-reactivity.

Bibliography:

Bánkfalvi A *et al* (2004) *Mod Pathol* 17:1051 ff
 Böcker W *et al* (1997) *Pathologie* 18:3 ff
 Hungermann D *et al* (2005) *BMC Cancer* 5
 Shah RB *et al* (2002) *Am J Surg Pathol* 26:1161 ff

Weinstein MH *et al* (2002) *Mod Pathol* 15:1302 ff
 Zhou M *et al* (2003) *Am J Surg Pathol* 27:365 ff
 Nadji M and Morales AR *Ann N.Y. Acad Sci* 420:134-9, 1983
 Omata M *et al*. *Am J Clin Pathol* 73(5): 626-32, 1980

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Explanations of the symbols on the product label:

REF	Bestellnummer Catalog Number Reference du catalogue		Verwendbar bis Use By Utiliser jusque		Gebrauchsanweisung beachten Consult Instructions for use Consulter les instructions d'utilisation
LOT	Chargenbezeichnung Batch Code Code du lot		Lagerungstemperatur Temperature Limitation Limites de température	RUO	Nur für Forschungszwecke For Research Use Only Pour la recherche uniquement
IVD	In vitro Diagnostikum In Vitro Diagnostic Medical Device Dispositif médical de diagnostic in vitro		Achtung Warning Attention		Hersteller / Manufacturer / Fabricant Zytomed Systems GmbH • Anhaltinerstraße 16 14163 Berlin, Germany • Tel: (+49) 30-804 984 990 www.zytomed-systems.com