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CDX-2 & Cytokeratin 7 Multiplex Cocktail; Clones EP25 & OV-TL12/30

Availability/Contents:

Item #	Volume
A00148-0002	2 ml
A00148-0007	7 ml
A00148-0025	25 ml

Description:

Species:	Rabbit and Mouse
Designation:	Cocktail of Rabbit and Mouse monoclonal antibodies to CDX-2 and CK7.
Immunogen:	Rabbits were injected with a synthetic peptide corresponding to residues near the C-terminus of human CDX-2. BALB/C mice were injected with OTN11-ovarian cell carcinoma cell line.
Mol. Weight:	Unknown
Clone:	Clones EP25 and OV-TL12/30
Isotype:	Rabbit IgG and Mouse IgG1, respectively.
Format:	This antibody has been pretitered and quality controlled to work on formalin-fixed paraffin-embedded as well as acetone fixed cryostat tissue sections. No further titration is required.
Specificity:	CDX-2 expression is restricted to nuclear staining in positive cells while CK7 expression is restricted to most glandular and transitional epithelia including lung, breast, bladder, and the female genital tract and their adenocarcinomas, but not in most gastrointestinal epithelium, prostate, hepatocyte, and squamous epithelium.
Background:	The caudal-related homeodomain protein 2 (CDX-2) which encodes an intestine-specific transcription factor is expressed in the nuclei of epithelial cells throughout the intestine, from duodenum to rectum. CDX-2 is thought to play an important role in the proliferation and differentiation of intestinal epithelial cells. The CDX-2 protein is expressed in primary and metastatic colorectal carcinomas, intestinal metaplasia of the stomach, and intestinal type gastric cancer. In human colorectal cancer, the expression of both CDX-2 and carbonic anhydrase 1, a gene regulated by CDX-2, is reduced or absent. However, CDX-2 is one of the important regulators in defining pathways seen in selected non-GI adenocarcinomas such as mucionous ovarian carcinomas and adenocarcinomas of the urinary bladder. CDX-2 is also used in diagnostic surgical pathology as a marker for gastrointestinal differentiation. Cytokeratin 7 (CK7) is a type II keratin which is a cytoplasmic intermediate filament protein (IFP) of low molecular weight (54 kDa). CK7 belongs to the neutral basic type B subfamily of cytokeratins. The genes encoding the type II cytokeratins are clustered in a region of chromosome 12q12-q13. CK7 is expressed in a tissue-specific manner which is generally restricted to the simple epithelium usually found in most glandular and transitional epithelia including lung, breast, bladder, and the female genital tract and their neoplasms, but not in most gastrointestinal epithelium, prostate, hepatyocyte and squamous epithelium. The predicted amino acid sequence of this keratin has revealed a striking difference between this keratin and the type II keratins expressed in epidermal cells.

Storage: 2° C  8° C


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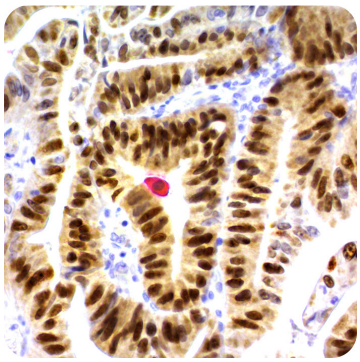
CK7 has been reported in the columnar and glandular epithelium of the lung, breast, endometrium, ovary, and thyroid, as well as in carcinomas of the bladder and renal cell carcinoma. CK7 expression has been reported to show characteristic patterns on primary and metastatic lung and colorectal adenocarcinomas. Cytokeratin 7 is reported to be expressed in abundance in cultured bronchial and mesothelial cells, but only at lower levels in cultured epidermal cells. CK7 can be used as a tool in order to distinguish ovarian and gastrointestinal carcinomas, or transitional cell carcinomas and prostate cancer. In hepatocytes, atypical expression of CK7 is a marker for primary biliary cirrhosis.

Species Reactivity:	Human
Positive Control:	CDX-2: Colon for normal tissue and colon adenocarcinoma for abnormal tissue.
	CK7: Carcinoma of ovary, lung, cervix, or breast.
Cellular Localization:	CDX-2: Nuclear. CK7: Cell surface / cytoplasm.
Titer/Working Dilution:	No further dilution is required.
Microbiological State:	This product is not sterile.

Uses/Limitations:

Not to be taken internally.
For research use only.
This product is intended for qualitative immunohistochemistry with normal and neoplastic formalin-fixed, paraffin-embedded tissue sections, to be viewed by light microscopy.
Do not use if reagent becomes cloudy.
Do not use past expiration date.
Non-Sterile.



Human colon adenocarcinoma stained with Ultra-Tek HRP using DAB Chromogen and UltraTek Alk-Phos using Permanent Red Chromogen.

Ordering Information and Current Pricing at www.scytek.com

Procedure:

1. Deparaffinize and rehydrate tissue section.
2. **Tissue Section Pretreatment (Highly Recommended):** Staining of formalin fixed, paraffin embedded tissue sections is enhanced by pretreatment with Citrate Plus (ScyTek catalog# CPL500).
3. Wash 2 times in DI/Distilled water.
4. To reduce nonspecific background staining due to endogenous peroxidase, incubate slide in hydrogen peroxide for 10-15 minutes.
5. Wash 2 times in buffer.
6. Apply Super Block (Catalog# AAA) and incubate for 5 minutes at room temperature to block nonspecific background staining. **Note:** Do not exceed 10 minutes or there may be a reduction in desired stain.
7. Wash 3 times in buffer.
8. Apply primary antibody cocktail and incubate for 30 minutes in a humid environment.
9. Wash 3 times in buffer.
10. Apply UltraTek Anti-Mouse (Catalog# ABJ) and incubate for 10 minutes at room temperature.
11. Wash 3 times in buffer.
12. Apply UltraTek HRP (Catalog# ABL) and incubate for 10 minutes at room temperature.
13. Rinse 3 times in buffer.

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14. Rinse 1 time in DI/Distilled water.
15. Apply mixed DAB Chromogen / DAB Substrate (Catalog# ACV) and incubate for 10-15 minutes, depending on the desired stain intensity.
    WARNING: DAB is a suspected carcinogen. Handle with care and dispose of according to all regulations.
16. Rinse 3 times in buffer.
17. Apply UltraTek Anti-Rabbit (Catalog# ABK) and incubate for 10 minutes at room temperature.
18. Rinse 3 times in buffer.
19. Rinse 1 time in DI/Distilled water.
20. Apply UltraTek Alk-Phos (Catalog# ABM) and incubate for 15 minutes at room temperature.
21. Rinse 3 times in buffer.
22. Rinse 1 time in DI/Distilled water.
23. Apply mixed Permanent Red Concentrate / Permanent Red Substrate (Catalog# PRD) and incubate for 10-15 minutes, depending on the desired stain intensity.
24. Rinse 2 times in DI/Distilled water.
25. Counterstain with Hematoxylin (Catalog# HMM) and coverslip using a permanent mounting media.

Precautions: Contains Sodium Azide as a preservative (0.09% w/v).
Do not pipette by mouth.
Avoid contact of reagents and specimens with skin and mucous membranes.
Avoid microbial contamination of reagents or increased nonspecific staining may occur.
This product contains no hazardous material at a reportable concentration according to U.S. 29 CFR 1910.1200, OSHA Hazardous Communication Standard and EC Directive 91/155/EC.

References:

1. Drummond F, *et al. Ann Hum Genet* 61(5): 393-400.
2. Gregory PA, *et al. Pharmacogenet Genomics* 16(7): 527-36, 2006.
3. Werling RW, Yaziji H, Bacchi CE, Gown AM. *Am J Surg Pathol.* 27(3): 303-10, 2003.
4. Ramaekers F, Huysmans A, Schaart G, *et al.* Tissue distribution of keratin 7 as monitored by a monoclonal antibody. *Exp Cell Res.* 170(1): 235-49, 1987.
5. Jovanovic I, Tzardi M, Mouzas IA, *et al.* Changing pattern of cytokeratin 7 and 20 expression from normal epithelium to intestinal metaplasia of the gastric mucosa and gastroesophageal junction." *Histol Histopathol.* 17(2): 445-54, 2002.
6. Ramaekers F, van Nierkerk C, Poels L, *et al.* Use of monoclonal antibodies to keratin 7 in the differential diagnosis of adenocarcinomas. *Am J Pathol.* 136: 641-55, 1990.

Note: CDX-2 bearing EP Clone EP25 is Manufactured using Epitomics's RabMAb® technology under U.S. Patent Nos. 5,675,063 and 7,402,409.

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