anti-Cytokeratin (CAM 5.2) Mouse Monoclonal Primary Antibody

REF 790-4555

06478425001

IVD Σ 50

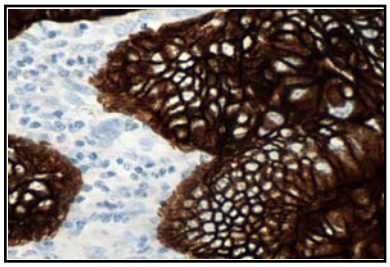

Figure 1. anti-Cytokeratin (CAM 5.2) staining colon adenocarcinoma

INTENDED USE

Anti-Cytokeratin (CAM 5.2) Mouse Monoclonal Primary Antibody (anti-Cytokeratin (CAM 5.2)) is directed against cytokeratin 7 and cytokeratin 8 present on secretory epithelia but not on stratified squamous epithelia. This antibody may be used to aid in the identification of tumors of epithelial origin such as colorectal, pancreatic, and non-small cell lung cancer and in distinguishing carcinomas from other malignant tumors of non-epithelial origin. The antibody is intended for qualitative staining in sections of formalin-fixed, paraffin-embedded tissue.

This product should be interpreted by a qualified pathologist in conjunction with histological examination, relevant clinical information, and proper controls.

This antibody is intended for *in vitro* diagnostic (IVD) use.

SUMMARY AND EXPLANATION

Anti-Cytokeratin (CAM 5.2) is a mouse monoclonal antibody produced against the colon carcinoma cell line HT29, and recognizes lower molecular weight intracellular cytokeratin proteins (cytokeratins 7 and 8).[1] Anti-Cytokeratin (CAM 5.2) Mouse Monoclonal Primary Antibody is directed against cytokeratin 7 and cytokeratin 8 present on secretory epithelia but not on stratified squamous epithelia. The antibody can be used to aid in the identification of neuroendocrine carcinomas in various tissues including the thyroid, pancreas, non-small cell lung, and breast carcinomas.[2,3]

REAGENT PROVIDED

Anti-Cytokeratin (CAM 5.2) contains sufficient reagent for 50 tests.

One 5 mL dispenser of anti-Cytokeratin (CAM 5.2) contains approximately 57 µg of a mouse monoclonal antibody.

The antibody is diluted in 0.05 M Tris-HCL with 1% carrier protein and ProClin 300, a preservative

Total protein concentration of the reagent is approximately 10 mg/mL. Specific antibody concentration is approximately 11 µg/mL. There is no known non-specific antibody reactivity observed in this product.

Anti-Cytokeratin (CAM 5.2) is a monoclonal antibody produced as cell culture supernatant.

Refer to the appropriate Ventana detection kit package insert for detailed descriptions of: (1) Principles of the Procedure, (2) Materials and Reagents Needed but Not Provided, (3) Specimen Collection and Preparation for Analysis, (4) Quality Control Procedures, (5) Troubleshooting, (6) Interpretation of Results, and (7) General Limitations.

MATERIALS REQUIRED BUT NOT PROVIDED

Staining reagents, such as Ventana detection kits (*ultra*View Universal DAB Detection Kit), and ancillary components, including negative and positive tissue control slides, are not provided.

STORAGE

Store at 2-8°C. Do not freeze.

To ensure proper reagent delivery and the stability of the antibody, replace the dispenser cap after every use and immediately place the dispenser in the refrigerator in an upright position.

Every antibody dispenser is expiration dated. When properly stored, the reagent is stable to the date indicated on the label. Do not use reagent beyond the expiration date.

SPECIMEN PREPARATION

Routinely processed, formalin-fixed, paraffin-embedded tissues are suitable for use with this primary antibody when used with Ventana detection kits and a Ventana BenchMark Series automated slide stainer. The recommended tissue fixative is 10% neutral buffered formalin.[4] Slides should be stained immediately, as antigenicity of cut tissue sections may diminish over time.

It is recommended that positive and negative controls be run simultaneously with unknown specimens.

WARNINGS AND PRECAUTIONS

1. For *in vitro* diagnostic (IVD) use.
2. ProClin 300 is used as a preservative in this solution. It is classified as an irritant and may cause sensitization through skin contact. Take reasonable precautions when handling. Avoid contact of reagents with eyes, skin, and mucous membranes. Use protective clothing and gloves.
3. Materials of human or animal origin should be handled as biohazardous materials and disposed of with proper precautions.
4. Avoid contact of reagents with eyes and mucous membranes. If reagents come in contact with sensitive areas, wash with copious amounts of water.
5. Avoid microbial contamination of reagents as it may cause incorrect results.
6. Consult local and/or state authorities with regard to recommended method of disposal.
7. Refer to the MSDS for additional information.

STAINING PROCEDURE

Ventana primary antibodies have been developed for use on a Ventana BenchMark Series automated slide stainer in combination with Ventana detection kits and accessories. A recommended staining protocol for the BenchMark XT and BenchMark ULTRA instruments with *ultra*View Universal DAB Detection Kit is listed in Table 1.

The parameters for the automated procedures can be displayed, printed and edited according to the procedure in the instrument's Operator's Manual. Refer to the appropriate Ventana detection kit package insert for more details regarding immunohistochemistry staining procedures.

Table 1. Recommended Staining Protocol for anti-Cytokeratin (CAM 5.2) with *ultra*View Universal DAB Detection Kit on a BenchMark XT and BenchMark ULTRA instrument.

Procedure Type	Method
Deparaffinization	Selected
Cell Conditioning (Antigen Unmasking)	None required
Enzyme (Protease)	Protease 1, 8 Minutes
Antibody (Primary)	BenchMark XT instrument 16 Minutes, 37°C BenchMark ULTRA Instrument 16 minutes, 36°C
Counterstain	Hematoxylin II, 4 Minutes
Post Counterstain	Bluing, 4 Minutes

Table 2. Recommended Staining Protocol for anti-Cytokeratin (CAM 5.2) with *i*VIEW DAB Detection Kit on a BenchMark XT instrument.

Procedure Type	Method
Deparaffinization	Selected
Cell Conditioning (Antigen Unmasking)	None required
Enzyme (Protease)	Protease 1, 8 minutes
Antibody (Primary)	BenchMark XT instrument 32 Minutes, 37°C
Counterstain	Hematoxylin II, 4 Minutes
Post Counterstain	Bluing, 4 Minutes

Due to variation in tissue fixation and processing, as well as general lab instrument and environmental conditions, it may be necessary to increase or decrease the primary antibody incubation, cell conditioning or protease pretreatment based on individual specimens, detection used, and reader preference. For further information on fixation variables, refer to "Immunohistochemistry Principles and Advances".[5]

POSITIVE TISSUE CONTROL

Examples of positive control tissues for this antibody are pancreatic carcinoma and colon.

STAINING INTERPRETATION

The cellular staining pattern for anti-Cytokeratin (CAM 5.2) is cytoplasmic.

SPECIFIC LIMITATIONS

This antibody has been optimized for specific incubation times but the user must validate results obtained with this reagent.

PERFORMANCE CHARACTERISTICS

Staining tests for specificity, sensitivity, and reproducibility were conducted using anti-Cytokeratin (CAM 5.2) with *ultra*View Universal DAB Detection Kit on the BenchMark XT instrument.

Specificity

Table 3. Specificity of anti-Cytokeratin (CAM 5.2) was determined by testing formalin-fixed, paraffin-embedded normal tissues.

Tissue	# positive / total cases	Tissue	# positive / total cases
Cerebrum	0/3	Thymus	0/3
Cerebellum	0/3	Myeloid (bone marrow)	0/3
Adrenal gland	3/3	Lung	2/3
Ovary	0/3	Heart	0/3
Pancreas	3/3	Esophagus	2/3
Parathyroid gland	2/3	Stomach	3/3
Hypophysis	3/3	Small intestine	3/3
Testis	0/3	Colon	3/3
Thyroid	3/3	Liver	3/3
Breast	3/3	Salivary gland	3/3
Spleen	2/3	Kidney	3/3
Tonsil	0/3	Prostate	3/3
Endometrium	0/3	Cervix	0/3
Skeletal muscle	0/3	Skin	0/3
Nerve (sparse)	0/3	Mesothelium and lung	3/3

Sensitivity

Table 4. Sensitivity of anti-Cytokeratin (CAM 5.2) was determined by testing a variety of formalin-fixed, paraffin-embedded neoplastic tissues.

Pathology	# positive / total cases
Glioblastoma	1/1
Atypical meningioma	0/1
Malignant ependymoma	0/1
Malignant oligodendroglioma	0/1
Serous papillary adenocarcinoma	1/1
Mucinous papillary adenocarcinoma	1/1
Islet cell carcinoma	0/1
Pancreatic adenocarcinoma	135/140
Seminoma	1/1
Embryonal carcinoma	1/1
Medullary carcinoma	1/1
Papillary carcinoma	1/1
Breast intraductal carcinoma	1/1
Breast lobular carcinoma *in situ*	0/1
Breast invasive ductal carcinoma	1/1
Diffuse B-cell lymphoma	0/1
Lung small cell undifferentiated carcinoma	3/9
Lung squamous cell carcinoma	20/23
Lung adenocarcinoma	23/23
Lung atypical carcinoid	0/2
Bronchioloalveolar carcinoma	2/2
Giant cell carcinoma	2/2
Large cell carcinoma	2/2
Lung papillary adenocarcinoma	8/8
Esophageal squamous cell carcinoma	0/1
Esophageal adenocarcinoma	1/1
Gastric mucinous adenocarcinomas	14/14
Gastrointestinal adenocarcinoma	56/57
GIST	0/3
Hepatocellular carcinoma	0/1
Hepatoblastoma	0/1
Renal clear cell carcinoma	1/1

Pathology	# positive / total cases
Prostatic adenocarcinoma	1/1
Prostatic transitional cell carcinoma	1/1
Leiomyoma	0/1
Endometrial adenocarcinoma	1/1
Endometrial clear cell carcinoma	1/1
Uterine squamous cell carcinoma	1/2
Embryonal rhabdomyosarcoma	0/1
Anal malignant melanoma	0/1
Basal cell carcinoma	1/1
Squamous cell carcinoma	1/1
Neurofibroma	0/1
Retroperitoneal neuroblastoma	0/1
Epithelial malignant mesothelioma	1/1
Diffuse malignant lymphoma	0/1
Hodgkin's lymphoma	0/1
Diffuse malignant lymphoma	0/1
Bladder transitional cell carcinoma	1/1
Low grade leiomyosarcoma	0/1
Osteosarcoma	0/1
Spindle cell rhabdomyosarcoma	0/1
Intermediate grade leiomyosarcoma	0/1
Malignant melanoma	0/1

Repeatability

Repeatability studies for anti-Cytokeratin (CAM 5.2) were completed to demonstrate:

- Inter-lot reproducibility of the antibody.
- Intra-run and Inter-run reproducibility on a BenchMark XT instrument.
- Intra-platform reproducibility on the BenchMark XT instrument and the BenchMark ULTRA instrument.
- Inter-platform reproducibility between the BenchMark XT and BenchMark ULTRA instrument.

All studies met their acceptance criteria.

REFERENCES

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