

## Cytokeratin (HMW) (Clone: K903P) Rabbit Monoclonal Antibody

### PRODUCT INFORMATION:

#### REF

MR1336	6ml Ready to use
MR1336	3ml Ready to use
MRC1336	1ml Concentrated
MRC1336	0.5ml Concentrated
MRC1336	0.1ml Concentrated
MRH1336	6ml Ready to use
MRH1336	3ml Ready to use

### PERFORMANCE CHARACTERISTICS:

**Localization:** Cytoplasm  
**Retrieval Buffer:** Tris-EDTA, pH 9.0  
**Incubation:** 30-60 minutes  
**Positive control:** Prostate Ca, Lung SCC

### INTENDED USE

#### For research use only

This antibody is intended for use in qualitatively identifying Cytokeratin (HMW) antigen by light microscopy in formalin-fixed, paraffin-embedded (FFPE) tissue sections using immunohistochemical (IHC) detection methodology. Interpretation of any positive or negative staining must be complemented with the evaluation of proper known controls (Positive and Negative) and must be made within the context of the patient's clinical history and other diagnostic tests. A qualified and trained pathologist must perform an evaluation of the test. This antibody is intended to be used after the primary diagnosis of the tumour has been made by conventional histopathology using nonimmunologic histochemical stains

### SUMMARY AND EXPLANATION

In normal tissues, cytokeratin (high molecular weight) is mainly expressed in normal squamous epithelium, ductal epithelium and other laminated epithelium. Positive expression was found in lung squamous cell carcinoma, breast cancer, pancreatic cancer, cholangiocarcinoma and bladder transitional cell carcinoma.

Anti-cytokeratin, high molecular weight is capable of recognizing all basic keratins; therefore, it is a broadly reactive antibody staining most epithelia and their neoplasms. Members of the acidic and basic subfamilies are found together in pairs. Since each epithelium contains at least one acidic and one basic keratin, this antibody is used to observe the distribution of keratin-containing cells in normal epithelia and to identify neoplasms derived from such epithelium.

### PRINCIPLE OF THE PROCEDURE

The identification of the antigen on the FFPE tissues is carried out using the above-stated antibody. The antigen and antibody complex is visualized using an enzyme-coupled (HRP/AP) secondary antibody with specific binding to the primary antibody, this complex is visualized by the enzymatic activation of the chromogen resulting in a visible reaction production of the antigenic site. Each and every step involves precise time and optimal temperature and the results are interpreted using a light microscope by a qualified and trained pathologist.

### REAGENT PROVIDED

**Concentrated format:** Antibody to Cytokeratin (HMW) is affinity purified and diluted in antibody diluent with 1% bovine serum albumin (BSA) and 0.05% sodium azide (NaN<sub>3</sub>).

**Recommended dilutions:** 1:50 – 1:100

The antibody dilution and protocol may vary depending on the specimen preparation and specific application. Optimal conditions should be determined by individual laboratories.

**Pre-diluted format:** PathnSitu's ready-to-use antibodies are pre-titrated to optimal staining conditions. Further dilution will affect the efficacy of the antibody and may yield to sub-optimal staining.

**Immunogen:** Synthetic peptide corresponding to residues within aa200-300 of Cytokeratin 1 was used as an immunogen.

Synthetic peptide according to the aa490-590(C-terminal) of Cytokeratin 5 was used as an immunogen.

**Host, Isotype:** Rabbit, IgG

### STORAGE AND HANDLING

**Storage Recommendations:** Store at 2-8°C. When stored at the appropriate conditions, the antibody is stable until expiry. Do not use the antibody after the expiration date provided on the vial in any condition.

To ensure proper reagent delivery and stability, replace the dispenser cap after every use and immediately place the vial into the refrigerated conditions in an

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upright position. The contents of the vial should be used within 9 months from the opening of the vial.

### SPECIMEN PREPARATION

#### Staining Recommendations:

Routinely processed, FFPE tissues are suitable for use with this primary antibody, when using PathnSitu's Poly Excel HRP/DAB detection system. The recommended tissue fixative is 10% neutral buffered formalin. Variable results may occur as a result of prolonged fixation or special processes such as decalcification. The thickness of the sections should be 2-5µm. Slides should be stained once the sections are made as the antigenicity of the cut sections may diminish over a period of time. It is recommended to stain known positive and negative controls simultaneously with unknown specimens.

### PRECAUTIONS

1. This product should be used by qualified and trained professional users only
2. The product contains < 0.1% of sodium azide as a preservative and is not classified as hazardous, refer to MSDS for further details
3. As with any product derived from biological sources, proper handling procedures should be used
4. Do not use reagents after the expiration date
5. Use protective clothing and gloves, while handling reagents
6. All hazardous materials should be disposed of according to local state and federal regulations
7. Avoid microbial contamination of reagents as it may lead to incorrect results

### STAINING PROCEDURE

**Antigen Retrieval Solution:** Use Tris-EDTA Buffer (Cat#PS009) as an antigen retrieval solution.

**Heat Retrieval Method:** Retrieve sections under steam pressure for 15 minutes using PathnSitu's MERS (Multi Epitope Retrieval System) for optimal retrieval of the epitopes, allow solution to cool at room temperature, transfer the tissue sections/slides to the distilled water prior to the primary antibody application.

**Primary Antibody:** Cover the tissue sections with primary antibody and incubate for 30-60 min at room temperature when using PathnSitu's PolyExcel Detection System.

**Detection System:** Refer to PathnSitu's PolyExcel HRP/ DAB detection system protocol for optimal staining results.

### QUALITY CONTROL

The recommended positive tissue control for Cytokeratin (HMW) are Prostate Carcinoma, Lung SCC and DCIS. A positive and negative tissue control must be run with every staining procedure performed to monitor the correct performance of processed tissue and test reagents. A negative tissue control provides an indication of non-specific background staining. If the results are not expected in positive and negative controls the test must be considered invalid and the entire procedure must be cross-verified. The individual laboratory must establish its own quality control to validate the process and antibody when opening a vial.

### INTERPRETATION OF RESULTS

Cytokeratin (HMW) stains the Cytoplasm. A qualified experienced/trained pathologist must interpret the results in the patient's sample along with the positive and negative controls.

### PERFORMANCE CHARACTERISTICS

PathnSitu products will undergo a thorough quality control check before it is released to the market. The antibody showed consistent specific and sensitive staining on the multiple positive tissue controls tested, by inter-run, intra run and lot-based studies. The antibody is stable for the expiry mentioned on the labels which is determined by real-time or accelerated methods.

### TROUBLESHOOTING

1. Follow the antibody-specific protocol recommendations according to the datasheet provided
2. Tissue staining is dependent on the handling and processing of the tissue prior to staining. Improper fixation, tissue processing, antibody freezing and thawing, washing, drying, heating, sectioning or contamination with other tissues or fluids may produce artefacts, antibody trapping or inaccurate

results

3. Do not allow the section to dry out during the entire IHC process
4. Excessive or incomplete counterstaining may compromise the interpretation of the results
5. If unusual results occur, contact PathnSitu's Technical Support at +91-40-2701 5544 or E-mail: [techsupport@pathnsitu.com](mailto:techsupport@pathnsitu.com)

#### LIMITATIONS AND WARRANTY

Authorized and skilled/trained personnel only may use the product. The clinical interpretation of any test results should be evaluated within the context of the patient's medical history and other diagnostic test results. A qualified trained pathologist must perform the evaluation of the test results. There are no warranties, expressed or implied, which extend beyond the description. PathnSitu is not liable for property damage, personal injury, time or effort or economic loss caused by this product.

#### BIBLIOGRAPHY

1. Kriho UK, et al. Virehows. Arch. 1997; 431:139-47.
2. D Oh, RA Prayson. Arch Pathol Lab Med- Vol 123, October 1999:917-920.

#### EXPLANATION OF SYMBOLS

**LOT**

Lot number / Batch number



Expiry

**RUO** *Research use only*



Storage limitation



Date of manufacture

**REF**

Catalogue number