

PRIMARY ANTIBODY CARTRIDGE (PAC)

Cytokeratin AE1/AE3

Monoclonal Mouse, Anti-Cytokeratin AE1/AE3
Clone AE1/AE3

REF 014-1002-150

Ready-to-Use ■ 60 Tests

INSTRUCTIONS FOR USE

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INTENDED USE

IVD For *in vitro* diagnostic use.

Monoclonal mouse anti-cytokeratin AE1/AE3 is intended for laboratory use to identify an epitope located in cells that are of epithelial origin using light microscopy. The two clones identify human cytokeratins 56.5, 50, 50, 48, and 40kD of the acidic subfamily and 65 to 67, 64, 59, 58, 56, and 52kD of the basic subfamily. This cocktail of antibodies is one component of a panel of antibodies used to identify tissues of unknown origin.

Positive results aid in the classification of normal and abnormal cells/tissues, and serve as an adjunct to conventional histopathology. The clinical interpretation of any positive staining or its absence should be complemented by morphological and histological studies with proper controls. Evaluations should be made by a qualified individual in conjunction with the patient's clinical history and other diagnostic test results.

Refer to the *Celerus Wave*® RPD Operator's Manual for additional information about Materials Required but Not Provided; Storage; Staining Procedure; Troubleshooting; Interpretation of Staining; and General Limitations.

SUMMARY AND EXPLANATION

Cytokeratins are a family of water-soluble proteins with molecular weights between 40-70 kD; they are found in the microtubule structure which forms the epithelial cytoskeleton. A minimum of 19 different cytokeratins have been identified and can be divided into two subfamilies. Subfamily A contains relatively acidic cytokeratins (pI < 5.5), whereas subfamily B contains cytokeratins that are relatively basic (pI > 6).

AE1/AE3 is a cocktail of two monoclonal antibodies that were obtained by immunizing mice with a human epidermal cytokeratin preparation. AE1/AE3 has been shown to identify the majority of human cytokeratins and thus may be used as a tool for the positive IHC identification of simple and stratified epithelial tissues. Antibody AE1 binds with an antigenic determinant present on most of the subfamily A cytokeratins, including cytokeratins with Moll's designation 10, 13, 14, 15, 16, and 19 (MWs of 56.5, 54, 50, 50, 48, and 40 kD, respectively) but not on Nos. 12, 17 and 18 (55, 47 and 45 kD). Antibody AE3 binds with an antigenic determinant shared by the subfamily B cytokeratins including Nos. 1, 2, 3, 4, 5, 6, 7 and (MWs of 65-67, 64, 59, 58, 56 and 52 kD, respectively).

PRINCIPLE OF PROCEDURE

Immunohistochemistry is a multi-step process to identify specific cell markers within tissue biopsies or tumor specimens. The sequential steps include antigen retrieval (optional), antibody application, and antibody visualization followed by optional counter-staining. Specimens are then coverslipped and observed under light microscopy by trained personnel. Normally, multiple antibodies are tested to determine lineage and cell cycle markers. The *Celerus Wave*® RPD is an automated IHC system that produces stained tissue.

Refer to the *Celerus Wave*® RPD Operator's Manual for additional information about the staining procedure.

MATERIALS AND METHODS

Reagent Provided

CLONE	Ig CLASS	IMMUNOGEN
AE1 and AE3, mixed at a ratio of 20:1	AE1 / IgG1, kappa AE3 / IgG1, kappa	Human epidermal cytokeratin preparation

READY-TO-USE IN PRIMARY ANTIBODY CARTRIDGE

Anti-cytokeratin AE1/AE3 is provided with sodium azide as a preservative, in a Primary Antibody Cartridge (PAC), a self-contained dispenser of reagents. Each PAC contains sufficient reagent to complete 60 stained slides. PACs must remain upright to avoid spilling. PAC must be primed before first use.

Refer to the *Celerus Wave*® RPD Operator's Manual for additional information about the Primary Antibody Cartridge.

SPECIFICITY

Clone AE1 detects the 56.5, 50, 50, 48 and 40kD human cytokeratins of the acidic subfamily.

Clone AE3 detects 65 to 67, 64, 59, 58, 56 and 52kD of the basic subfamily.

PRECAUTIONS

For professional users.

Minimize microbial contamination of reagents or an increase in nonspecific staining may occur.

Proper handling procedures should be used.

A Material Safety Data Sheet is available upon request.

Sodium azide deposits in drainage pipes made of lead or copper can result in the formation of highly explosive metallic azides. To avoid such deposits in drainage pipes, sodium azide should be discarded in a large volume of running water.

Sodium azide in the concentration used is not classified as hazardous. The following are the appropriate Risk (R) and Safety (S) phrases.

R36	Irritating to eyes.
R43	May cause sensitization by skin contact.
S24	Avoid contact with skin.
S26	In case of contact with eyes, rinse immediately with plenty of water and seek medical advice.
S35	This material and its container must be disposed of in a safe way.
S37/39	Wear suitable gloves and eye/face protection.
S46	If swallowed, seek medical advice immediately and show the product container or label.



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WASTE DISPOSAL

Adhere to all local laws when disposing of the PAC.

PACKAGING DAMAGE

DO NOT USE this product if it is leaking, has leaked, has spilled, cannot be primed, or has visually apparent physical damage.

MATERIALS REQUIRED BUT NOT PROVIDED

Celerus Wave® RPD Instrument.

Refer to the Celerus Wave® RPD Operator's Manual for additional information about Materials Required but Not Provided.

STORAGE AND HANDLING

Store at the temperature indicated on the product label. Do not freeze. This product is stable up to the expiration date on the product label. Do not use this product after the expiration date.

To ensure a valid staining assay, the use of positive and negative tissue controls is recommended.

SPECIMEN COLLECTION AND HANDLING

Formalin-fixed paraffin embedded (FFPE) tissues, frozen tissues, or smears are suitable for use. Wave RPD detection kits have been optimized for tissues fixed with 10% formalin.

Slides should be baked overnight at 37 °C, or at 60 °C for one hour.

Use standard histochemical techniques to deparaffinize processed slides. Place slides in the Wave RPD Slide Rack according to the staining grid provided by the instrument software. Avoid drying tissue specimen during this process. After all slides to be stained have been inserted and reagents mounted on the instrument, start the staining run.

When the staining run is complete, remove the slide rack, then slides. Coverslip slides and view under light microscopy.

PRODUCT-SPECIFIC LIMITATIONS

Tissues of non-epithelial origin have been shown to react with antibodies to cytokeratin 8 including extrafollicular reticulum cells of lymph nodes, tonsil, and spleen. Smooth muscle cells of the uterus contain cytokeratin 19 and possibly 8.

When performing a tumor origin test, use of anti-epithelial membrane antigen (EMA) antibody in conjunction with Cytokeratin AE1/AE3 is recommended. Absence of Cytokeratin AE1/AE3 staining means that binding did not occur, not that cytokeratin or epithelial tissue is missing.

False positive staining has been reported on glial cells, leiomyosarcomas, or rare melanomas. Faint cytoplasmic staining of melanomas (50%, 2/4), lymphomas (29%, 2/7) or plasmacytomas (100%, 2/2) has been reported. This staining is considered to be nonspecific background staining.

RESULTS EXPECTED/PERFORMANCE CHARACTERISTICS

Positive AE1/AE3 staining will be observed in the cytoplasm of epithelial cells.

Normal Tissues

Clones AE1/AE3 staining has been observed in the cytoplasm of epithelial cells from a variety of tissues, including glandular epithelium of prostate, skin, breast and thyroid, and squamous and columnar epithelium of the skin, tonsil, cervix, stomach, small and large intestine. Staining has been noted in ovary, salivary gland, endometrium and myometrium of the uterus, and tubules and Bowman's capsule of the kidney. No staining has been observed in lymphoid cells or skeletal muscle.

Abnormal Tissues

Clones AE1/AE3 stained 49/91 cases of a variety of tumors evaluated. Positivity was noted in tumors of epithelial origin. No staining was observed in non-epithelial tumors, including melanomas, lymphomas and sarcomas.

REFERENCES

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


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Franke WW, Moll R. Cytoskeletal components of lymphoid organs. Differentiation 1987; 36:145.

Bacchi CA, Zarbo RJ, Jiang JJ, Gown AM. Do glioma cells express cytokeratin? Appl Immunohistol 1995; 3:45.

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EXPLANATION OF SYMBOLS

 Catalog Number	 Temperature Limitations	 Batch Code	 Sufficient for <n> Tests	 Harmful, Irritant	 Warning
 In Vitro Diagnostic Medical Device	 Use By	 European Conformity	 EC Representative	 Manufacturer	 Refer to Instructions for Use