Anti– human CK 8 Mouse Monoclonal Primary Antibody

Clone: 1B12

**Intended use**
Anti- human CK 8 (Clone: 1B12) Mouse Monoclonal Primary Antibody is intended for the detection of CK 8/Keratin 8 protein expression in frozen or formalin fixed human tissues and cells. 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 within the context of the patient's clinical history and other diagnostic tests by a qualified pathologist. The antibody is intended for in vitro diagnostic (IVD) use.

**Background**
CK 8/Keratin 8 is a member of the type II keratin family clustered on the long arm of chromosome 12. Type I and type II keratins heteropolymerize to form intermediate-sized filaments in the cytoplasm of epithelial cells. The product of this gene typically dimerizes with keratin 18 to form an intermediate filament in simple single-layered epithelial cells. This protein plays a role in maintaining cellular structural integrity and also functions in signal transduction and cellular differentiation. Mutations in this gene cause cryptogenic cirrhosis.

Alternative Names: CK 8/ Cytokeratin 8 / Keratin 8 CK-8, cytokeratin-8, keratin, type II cytoskeletal 8, keratin-8

**Reagent provided**
Anti- human CK 8 Mouse Monoclonal Primary Antibody (Clone: 1B12) is provided in liquid form in 20mM Sodium phosphate, 150mM Sodium chloride, 0.2% BSA, 0.09% Sodium azide, pH 7.4. The isotype of the antibody is IgG2b. The protein concentration is approximately 0.2 +/- 0.05 mg/mL.

For Immunohistochemistry the primary antibody may be used at a working dilution of 1:100 – 1:200 for formalin-fixed, paraffin-embedded human tissue and this can be dependent upon the detection system used. These are guidelines only and optimal dilutions should be determined by the individual laboratory.

**Immunogen**
Human recombinant protein fragment corresponding to amino acids 91-381 of human CK8 (NP_002264) produced in E.coli.
Specificity
The specificity of the anti-human CK 8 Mouse Monoclonal Primary Antibody was established on known positive breast cancer tissue and negative human tonsil tissue. The anti-CK8 presented no staining on formalin fixed negative human tonsil tissue and germinal and non-germinal center and positive staining on formalin fixed positive breast cancer tissue using immunohistochemical (IHC) test methods.

Materials Required but Not Supplied
Antibody diluent, HIER solution, Antibody detection kits, Chromogen, Staining reagents, negative and positive tissue control slides are not included.

Precautions
1. For use by trained professionals only.
2. This product contains sodium azide (NaN₃), a chemical highly toxic in pure form. At product concentrations, though not classified as hazardous, NaN₃ may react with lead and copper plumbing to form highly explosive build-ups of metal azides. Upon disposal, flush with large volumes of water to prevent metal azide build-up in plumbing.
3. Wear appropriate Personal Protective Equipment to avoid contact with eyes and skin.
4. Unused reagents should be disposed of according to local, State, and Federal regulations.

Storage
Store at 2-8°C. Do not use the product past the expiration date indicated on the label. If reagents are stored under any other conditions, the end user must verify the acceptability of those conditions. There are no obvious signs to indicate instability of this product therefore, positive and negative controls should be run simultaneously with patient specimens.

Specimen Preparation
Paraffin Sections
Anti-human CK 8 Mouse Monoclonal Primary Antibody can be used on formalin-fixed, paraffin-embedded tissue sections at a working dilution of 1:100 to 1:200. Anti-human CK 8 clone 1B12 Mouse Monoclonal Primary Antibody working dilution requires 20 minutes of pretreatment with Heat Induced Epitope Retrieval (HIER) for staining. We recommend using 10mM Citrate Buffer pH 6.0, which showed optimal staining at a dilution of 1:200 on positive breast cancer tissue and negative staining on negative tonsil germinal and non-germinal centers. The dilutions are estimates; the actual staining results may vary due to reagents and detection protocols used. Validation of antibody performance and final protocol are the responsibility of the end user.

Manual Staining Procedure
1. Deparaffinize slides.
2. Submerge slides in peroxidase quenching solution for ~10 minutes and rinse with PBS-T 3 times, 2 minutes each.
3. Heat Induced Epitope Retrieval is required for this antibody.
4. Apply serum blocking solution.[Optional]
5. Apply primary antibody and incubate for 30-60 minutes at room temperature. After incubation, wash with PBS-T 3 times, 2 minutes each.
6. Apply secondary antibody and incubate according to the data sheet of the detection system. Wash with PBS-T 3 times, 2 minutes each.
7. Apply enzyme conjugate and incubate according to data sheet of detection system. Wash with PBS-T 3 times, 2 minutes each.
8. Apply chromogen and incubate 5-10 minutes and rinse with distilled water.

Staining interpretation
The cellular staining pattern for Anti-human CK 8 Mouse Monoclonal Primary Antibody is cytoplasmic.

Performance Characteristics
Predicted Staining in Normal Tissue/Cells.
Human tonsil tissue was shown to be negative for this antibody.

Predictive Staining in Tumor
Anti-human CK 8 Mouse Monoclonal (Clone: 1B12) produced strong cytoplasmic staining when screened on human colon tumor.
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