Product Information

Monoclonal Anti-Microphthalmia, clone C5

Produced in Mouse, Purified Immunoglobulin

M6065

Product Description

Monoclonal Anti-Microphthalmia (mouse IgG1 isotype) is derived from the hybridoma produced by the fusion of splenocytes from RBF/DnJ mice immunized with an N–terminal fragment of human microphthalmia protein and mouse myeloma NS1 cells. The antibody is purified by Protein G chromatography.

Monoclonal Anti-Microphthalmia recognizes serine phosphorylated and non-phosphorylated melanocytic isoforms of microphthalmia from human, mouse or rat. This antibody has been used in immunoblotting (doublet of 52-56 kDa), immunoprecipitation, immunohistochemistry on formalin-fixed paraffin-embedded or frozen tissue sections, and gel shift.

Microphthalmia (Mi in mouse or MITF in human) is a basic helix-loop-helix-leucine zipper (BHLH–ZIP) transcription factor. Microphthalmia is involved in the differentiation, development and survival of melanocytes and cells of the retinal pigment epithelium, that is, cells responsible for hair, skin, and eye color.[1-4] It activates the expression of the melanocyte specific genes tyrosinase and TRP1 (tyrosinase-related protein 1) by binding as a homo or heterodimer to a symmetrical DNA sequence (E box) (5'-CATGTG-3') located within the M box found in their promoters.[5,6] Microphthalmia also appears to be involved in the differentiation of mast cells, osteoclasts, basophils and natural killer cells.[7,8]

Microphthalmia is expressed in a limited number of cell types including heart, mast cells, osteoclast precursors, and melanocytes. There are a number of different isoforms of microphthalmia resulting from alternative splicing and alternative promotors. These isoforms differ at their amino-termini and in their expression patterns.[9,10]

In humans, mutations of microphthalmia cause Waardenburg Syndrome Type II, a dominantly inherited hearing loss accompanied by pigmentary disturbances.[11] Mutations of Mi in mouse result in profound loss of pigmented cells in the skin, eye and inner ear, as well as osteoporosis and defects in natural killer and mast cells. An osteoporotic rat strain harbors a large genomic deletion encompassing the 3’ half of microphthalmia. Osteoclasts from those animals lack microphthalmia protein.[12]

Immunohistochemical detection of microphthalmia transcription factor in tissues using monoclonal antibodies is a valuable tool in the identification of melanocytic lesions in numerous sites. This technique may facilitate the detection of micrometastases in sentinel lymph nodes.[13]

Reagent

Supplied as a solution in phosphate buffered saline with 0.08% sodium azide as a preservative.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the safety data sheet for information regarding hazards and safe handling practices.

Storage/Stability

Store at −20 °C. Upon initial thawing freeze the solution in working aliquots for extended storage. Avoid repeated freezing and thawing to prevent denaturing the antibody. Do not store in a frost-free freezer. Working dilutions should be discarded if not used within 12 hours.

Product Profile

Immunoblotting

A 1:500 dilution recognizes the Mi protein in 10 µg of mouse brain lysates.

Immunohistochemistry

A recommended working concentration of 1 mg/mL is determined using formalin-fixed paraffin-embedded human melanoma tissue.

Immunoprecipitation and Gel Supershift Assays

- Use a working concentration of 2 µg/mg of protein lysate.
- 501 Mel human melanoma cells or wild-type human, rat or mouse osteoclast cells may be used for positive controls.

  Note: In order to obtain best results using different techniques and preparations we recommend determining optimal working concentration by titration.

References

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