

Rabbit monoclonal antibody against C-met (Clone ISR039)

Product identification

| ISC062-R3 | 3 ml ready-to-use (RTU) |
|-----------|-------------------------|
| ISC062-R7 | 7 ml ready-to-use (RTU) |
| ISC062-C1 | 1 ml concentrated |

Summary and explanation

MART-1/ Melan A is one of a group of melanocyte differentiation antigens that are expressed in melanoma cells and normal melanocytes. Peptide epitopes derived from MART-1/ Melan A antigen have been identified as targets for cytolytic Tlymphocytes (CTLs) and tumour-infiltrating lymphocytes (TILs) in the context of HLA-A2.1 and other MHC class I molecules. MART-1/ Melan A is expressed as a cytoplasmic protein in melanocytic and melanoma cells. MART-1/ Melan A is expressed in skin, retina and melanocyte cell lines but not in other normal tissues. Unlike the HMB-45 (gp100), which is a known melanosomal protein, MART-1/ Melan A is a much smaller molecule.

C-Met is a protein encoded by MET gene, which is a hepatocyte growth factor receptor (HGFR). It has tyrosine kinase activity and is related to a variety of oncogene products and regulatory proteins. It is involved in the regulation of cell information transmission and cytoskeleton rearrangement and is an important factor in cell proliferation, differentiation and movement [1]. This antibody gives membranous and cytoplasmic staining in positive cells. The normal expression of c-Met pathway can promote tissue differentiation and repair, while abnormal expression promotes the proliferation and metastasis of tumor cells. C-Met is not only expressed in normal human tissues, but also overexpressed in a variety of cancers. In breast cancer, overexpression of c-Met can identify patients with poor clinical results. In glioblastomas, overexpression of c-Met is associated with shorter survival time. In papillthyroid carcinoma, overexpression of c-Met is associated with a high risk of metastasis and recurrence of thyroid papilla carcinoma in children and adolescents. Also, c-Met is associated with poor prognosis of nonsmall cell lung cancer, colorectal cancer and gastric cancer [2,3].

Principle of the procedure

The stated primary antibody is suitable for immunohistochemical staining of FFPE tissue sections based on specific antigen-antibody reaction. Using a detection system linked to HRP or alkaline phosphatase the antigen visualization is performed via specific binding of the primary antibody. Secondary antibody is binding to the primary antibody, and the enzyme complex labels this complex. The enzymatic activation of the chromogen results in a visible reaction product at the antigen site. Each step is incubated for a precise time and temperature and requires interposed washing steps. The specimen may then be counterstained. Results are interpreted using a light microscope.

Materials provided

| Primary antibody | C-met |
|--------------------------|-------------------------|
| Host | Rabbit |
| Subclass | lgG |
| Clone | ISR039 |
| Species Cross-reactivity | Human. Others-not |
| | known |
| Applications | Immunohistochemistry |
| Epitope Retrieval | Heat-induced epitope |
| | retrieval |
| Ready-to-use antibody | Prediluted antibody in |
| | antibody diluent buffer |
| Recommended working | 1:50 to 1:100 |
| dilution range | |

Product label shows the specific lot number.

Prediluted antibody is ready-to-use and optimized for staining. No further dilution, reconstitution, mixing, or titration is needed.

Antibody concentrate is optimized for dilution within dilution range using InSituChem® Antibody Diluent for IHC (Cat. No. D005-80). Indicated dilution range should be considered as recommendation and depends on different facts (tissue, fixation, incubation conditions, etc.). Optimum dilution to be determined in user's own system.

Materials required but not provided

The following materials may be required for staining but are not provided with the primary antibody.

- Positive and negative controls
- Microscope slides (positively charged) and cover slips
- Water bath
- Humidified chamber
- Staining jars
- Stopwatch
- Xylene or xylene substitute
- Ethanol
- Deionized or distilled water
- Antigen retrieval reagent, e.g. Antigen Enhancer (HIER buffer), InSituChem® Cat. No. D004-500
- Detection system, e.g. IHC Complete Detection system (Goat anti mouse/rabbit HRP, DAB staining), InSituChem® Cat. No. D001-15
- Wash buffer: e.g. IHC Wash Buffer, InSituChem® Cat. No. D003-500
- Tap water/bluing reagent (e.g. ammonia water)
- Light microscope

Storage and handling

Store at 2 - 8 °C.



When stored correctly antibody is stable to the expiration date indicated on the vial. Do not use after expiration date.

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

Staining procedure

- Cut 3-4 μm section of formalin-fixed paraffinembedded tissue and place on positively charged slides.
- 2. Dry at 65°C for 2 hours.
- 3. Deparaffinize, rehydrate, and epitope retrieve. Upon completion, rinse with 3 changes of distilled or deionized water.
- 4. If using HRP detection system, place slides in peroxide block for 10 minutes; rinse. If using AP detection system, omit this step.
- 5. Apply the antibody and incubate for 60 minutes; rinse.
- 6. Apply the InSituVison[™] Polymer Rabbit/Mouse Detection System for 30 minutes; rinse.
- 7. Apply ample amount of DAB or AEC chromogen and incubate; rinse.
- 8. Dehydrate and coverslip.

Interpretation of results

The immunostaining procedure causes a colored reaction product to precipitate at the antigen sites localized by the primary antibody.

Cellular localization: Cytoplasm/membrane.

A qualified pathologist experienced in immunohistochemistry procedures must evaluate positive and negative tissue controls before interpreting patient specimens.

Positive staining intensity should be assessed within the context of any background staining of the negative reagent control.

Note: A negative result means that the antigen in question was not detected, but not that the antigen is not present in the cells/tissues tested. An antibody panel may be used to support the results in some circumstances. Additionally, the morphology of each tissue sample should be examined utilizing a hematoxylin and eosin stained section. A qualified pathologist must interpret the patient's morphologic findings and pertinent clinical data.

Warnings and precautions

- 1. Application only by qualified and trained personnel.
- 2. There are no estimated health risks, if the product is used as directed. MSDS is available on request.
- Product contains sodium azide as preservative. Pure sodium azide is toxic. The concentration of sodium azide in this reagent is < 0.1 % and is not classified hazardous. See MSDS.

- 4. As with any product derived from biological sources, proper handling procedures should be used.
- 5. Do not use reagents after expiration date.
- 6. Take reasonable precautions when handling reagents. Use protective clothing and gloves.
- 7. All hazardous materials should be disposed according to guidelines for hazardous waste disposal. Materials of human or animal origin should be handled as biohazardous materials and disposed of with proper precautions.
- 8. Avoid microbial contamination of reagents as it may cause incorrect results.

Literature

[1] Orosz Z. Melan-A/Mart-1 expression in various melanocytic lesions and in non-melanocytic soft tissue tumours[j]. Histopathology, 1999, 34(6): 517–525.

[2] Hofbauer GFL., Kamarashev J, Geertsen R, et al. Melan A/MART-1 immunoreactivity in formalinfixed paraffin-embedded primary and metastatic melanoma: frequency and distribution[J]. Melanoma Research, 1988, 8(4):337–343.

Date of publication or revision

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Explanation of symbols

