p21

CONTACT INFORMATION: Monoclonal Antibodies Unit. Centro Nacional de Investigaciones Oncológicas

STATUS: Validated

TYPE: rat anti mouse CLONE NAME: HUGO291

PROTEIN: mouse p21 (mouse cyclin-dependent kinase inhibitor 1)

PROTEIN WEB: http://www.ncbi.nlm.nih.gov/protein/6671726

ANTIGEN USED: HIS-GST-mp21 full length recombinant protein

FUSION PARTNER: NS1/Ag4-1 (NS1) cells

ISOTYPE: IgG2a SPECIES REACTIVITY: mouse

PREPARATION AND STORAGE: Aliquot and store at 4C. Do not freeze

APP RECOMMENDED: IHQ-paraffin, WB

APP NO TESTED: IHQ-frozen, Flow cytometry, IF, IP

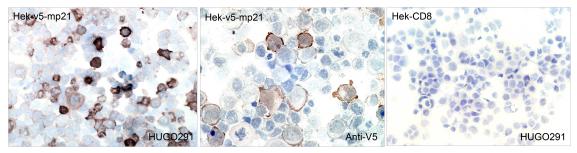
DESCRIPTION

This gene encodes a potent cyclin-dependent kinase inhibitor. The encoded protein binds to and inhibits the activity of cyclin-CDK2 or CDK4 complexes, and thus functions as a regulator of cell cycle progression at G1. The expression of this gene is tightly controlled by the tumor suppressor protein p53, through which this protein mediates the p53-dependent cell cycle G1 phase arrest in response to a variety of stress stimuli. This protein can interact with proliferating cell nuclear antigen (PCNA), a DNA polymerase accessory factor, and plays a regulatory role in S phase DNA replication and DNA damage repair. This protein was reported to be specifically cleaved by CASP3-like caspases, which thus leads to a dramatic activation of CDK2, and may be instrumental in the execution of apoptosis following caspase activation. Two alternatively spliced variants, which encode an identical protein, have been reported.

APPLICATIONS

| IHC Techniques | Clone | Dilution | Antibody concentration | Antigen retrieval method | Visualization kit | Positive control | Negative control | Protein localization | Positivity in other species |
|----------------|-------|----------|------------------------|--------------------------|-------------------|------------------|------------------|----------------------|-----------------------------|
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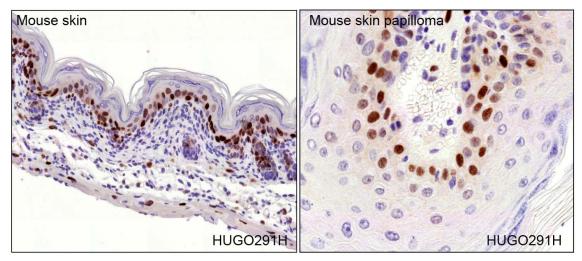
| Frozen tissue and cytospins | | | | | | | | | |
|-----------------------------|------|------|-------------|------------------|---------|------------|------------|---------|--|
| Paraffin tissue | | | | | | | | | |
| Recommended | HUGO | 1:10 | supernatant | Discovery Xt CC1 | Ventana | mouse skin | Lymph node | nuclear | |
| | 291 | | | OmniMap | | | | | |
| Immunofluorescence | | | | | | | | | |



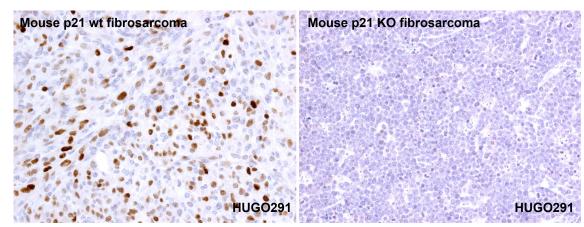
mp21 antibody (HUGO291) in transfected cells

Validation of HUGO291 monoclonal antibody in transfected cells.

Nuclear and cytoplasmic staining on frozen cytospin preparations of transfected HEK293T-V5-mp21 cells using antibody HUGO291. Anti-V5 was used as positive control. Hek-CD8 transfected cells were used as negative control.

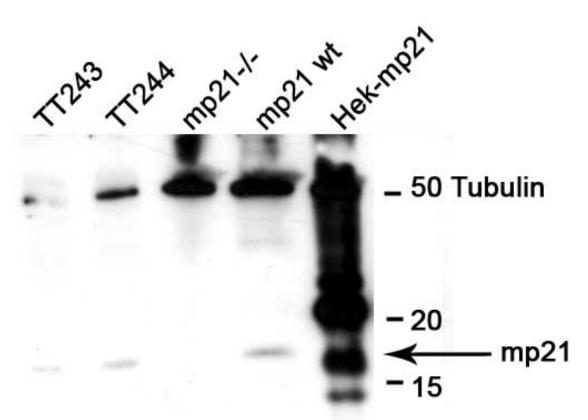


mp21 (HUGO291H) expression in mouse skin and mouse skin papilloma.



Anti-mp21 (HUGO291) IHQ in mouse fibrosarcoma paraffin sections.

| WB Techniques | Clone | Dilution | Antibody concentration | Positive control | Negative control | Expected MW | Observed Mw | Positivity in other species | |
|---------------------|--------|----------|------------------------|------------------|------------------|-------------|-------------|-----------------------------|--|
| Western Blotting | | | | | | | | | |
| Recommended | HUGO29 | 1:2 | supernatant | | | | | | |
| | 1 | | | | | | | | |
| Immunoprecipitation | | | | | | | | | |



HUGO 291H/A6 NEAT

Biochemical characterization of the anti-mp21 monoclonal antibody

Monoclonal Antibodies Catalogue

Western blot analysis of mp21 expression in total protein extracts from TT243 (no irradiated mefs), TT244 (10Gy 24h irradiated mefs), mouse mefs p21 ko, mouse mefs p21 wt and Hek-p21 transfected cells. TT244 mefs show a 18kDa band corresponding to mp21 protein. TT243 mefs were used as no irradiated control. p21 WT mouse cells also express mp21 while p21KO mouse cells does not express mp21 protein.

Lane 1 TT243 no irradiated mefs(100ug) (-)

Lane 2 TT244 10Gy irradiated mefs (100ug) (+)

Lane 3 p21 -/- KO mefs (60ug) (-)

Lane 4 p21 WT mefs (75ug) (+)

Lane 5 Hek-p21 transfected cells (20ug) (+)

Tubulin was used as loading control.