APH650Mu01 100μg
Active Melanoma Inhibitory Activity Protein 1 (MIA1)
Organism Species: *Mus musculus (Mouse)*
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Prokaryotic expression.
Host: *E. coli*
Residues: Asp23~Gln130
Tags: N-terminal His-tag
Purity: >95%
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 5%Trehalose .
Original Concentration: 200μg/mL
Applications: Cell culture; Activity Assays.
(May be suitable for use in other assays to be determined by the end user.)
Predicted isoelectric point: 7.9
Predicted Molecular Mass: 16.0kDa
Accurate Molecular Mass: 16kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

Reconstitute in 10mM PBS (pH7.4) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

[STORAGE AND STABILITY]

Storage: Avoid repeated freeze/thaw cycles.
Store at 2-8ºC for one month.
Aliquot and store at -80ºC for 12 months.

Stability Test: The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37ºC for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date under appropriate storage condition.

[SEQUENCE]

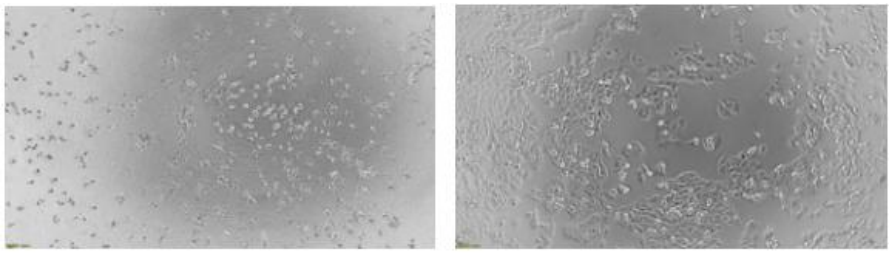
DRAMPKLA DWKLCADEEC SHPISMAVAL
QDYVAPDCRF LTIYRGQVVY VFSKLKGRGR LFWGGSVQGG YYGDLAARLG
YFPSSIVRED LTLKPGKIDM KTDQWDFYCQ

[ACTIVITY]

Melanoma inhibitory activity (MIA), also known as cartilage-derived retinoic acid-sensitive protein (CD-RAP), is a 12-kDa protein that is secreted from both chondrocytes and malignant melanoma cells. MIA has been reported to have effects on cell growth and adhesion, and it may play a role in melanoma metastasis and cartilage development. In the presence of MIA, we observed enhanced migratory ability of melanocytic cells, induction of melanoma-associated genes as well as inhibition of apoptosis due to anoikis.

To test the effect of MIA1 on cell apoptosis, A375 cells were seeded into triplicate wells of 96-well plates at a density of 4,000 cells/well and allowed to attach overnight, then the medium was replaced with various concentrations of recombinant mouse MIA1 diluted with 5% serum standard DMEM. After incubated for 48h, cells were observed by inverted microscope and cell viability was measured by Cell Counting Kit-8 (CCK-8). Briefly, 10 μl of CCK-8 solution was added to each well of the plate, then the absorbance at 450 nm was measured using a microplate reader after incubating the plate for 1-4 hours at 37 ℃. Apoptosis of A375 cells after incubation with MIA1 for 48h observed by inverted microscope was shown in Figure 1. The result of cell viability assessed by CCK-8 was shown in Figure 2. It was obvious that MIA1 significantly decreased cell

viability of A375 cells. The ED50 of recombinant mouse MIA1 is 5.94 ug/ml.



A B

Figure 1. Inhibition of A375 cells proliferation after stimulated with recombinant mouse MIA1

(A) A375 cells cultured in DMEM, stimulated with 10 μg/mL MIA1 for 48h;

(B) Unstimulated A375 cells cultured in DMEM for 48h.

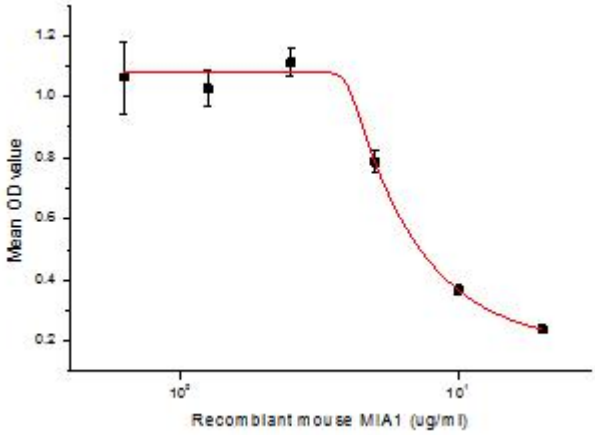


Figure 2. Inhibition of A375 cells proliferation after stimulated with recombinant human MIA1

[IDENTIFICATION]

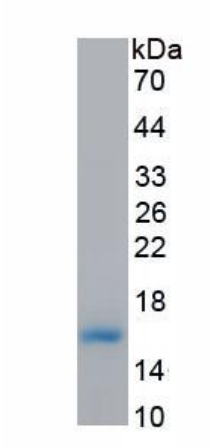


Figure 3. SDS-PAGE

Sample: Active recombinant MIA1, Mouse

[IMPORTANT NOTE]

The kit is designed for research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.