Active Receptor Activator Of Nuclear Factor Kappa B Ligand (RANkL) Instruction Manual

SBPA196Mu61

Mus musculus (Mouse)

Buffer Formulation	PBS, pH7.4, containing 0.01% SKL, 1mM DTT, 5% Trehalose and Proclin300.
Traits	Freeze-dried powder
Purity	> 90%
Isoelectric Point	7.1
Applications	Cell culture; Activity Assays.

ACTIVITY TEST

Sample (cell supernatant of RAW264.7 cells)	O.D. value	Corrected	Concentration of TRAP (ng/mL)
stimulated with RANKL(1ng/mL)	0.665	0.609	2.36
stimulated with RANKL(10ng/mL)	0.631	0.574	2.21
unstimulated	0.209	0.152	0.86

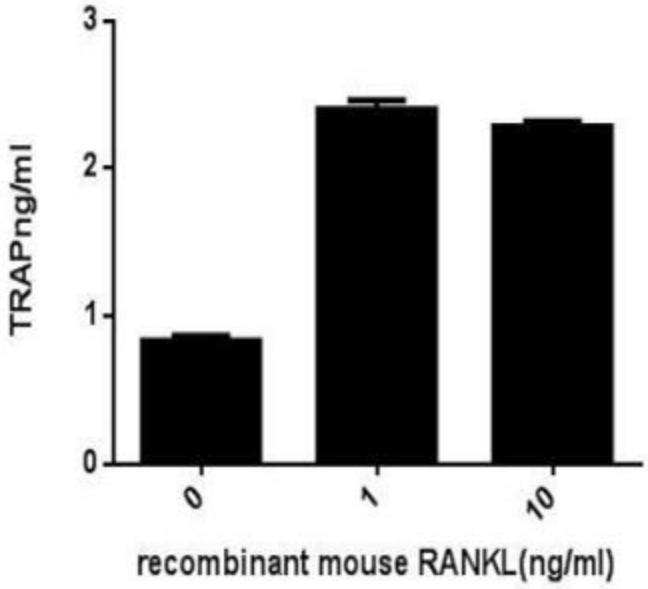

Figure1. TRAP levels in the cell of RAW264.7 cells induced by RANKL.

Receptor activator of nuclear factor kappa-B ligand (RANKL), also known as tumor necrosis factor ligand superfamily member 11 (TNFSF11), TNF-related activation-induced cytokine (TRANCE), osteoprotegerin ligand (OPGL), and osteoclast differentiation factor (ODF), is a member of the tumor necrosis factor (TNF) superfamily. RANKL has been identified to affect the immune system and control bone regeneration and remodeling. RANKL is an apoptosis regulator gene, a binding partner of osteoprotegerin (OPG), a ligand for the receptor RANK and controls cell proliferation by modifying protein levels of Id4, Id2 and cyclin D1. The protein can bind to RANK on cells of the myeloid lineage and functions as a key
factor for osteoclast differentiation and activation. Thus, we use recombinant RANKL to induce RAW264.7 mouse monocyte/macrophage cells to differentiate to osteoclast cell. RAW264.7 mouse monocyte/macrophage cells were seeded into 24-well plates at a density of 2×105 and allowed to attach overnight, then treated with RANKL (1ng/mL, 10ng/mL) and incubated for 72h. Then we assay TRAP (a kind of enzyme expressed by

osteoclast cell) by ELASA. Result: TRAP levels in the cell supernatant of RAW264.7 cells increased significantly after stimulated with RANK, the data was shown in Table 1 and Figure1.

USAGE

Reconstitute in 10mM PBS (pH7.6) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

STORAGE

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

STABILITY

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 37°C 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.

Image

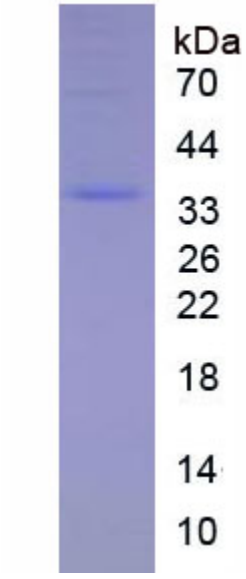

Figure. SDS-PAGE

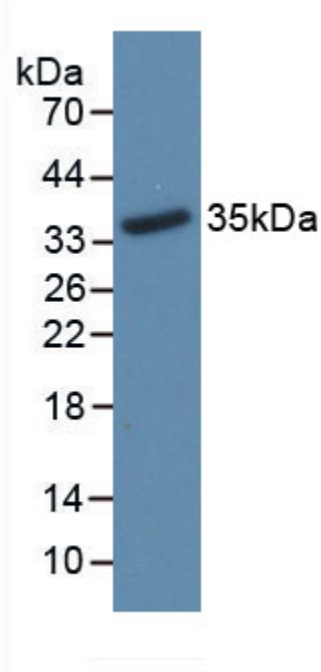

Figure. Western Blot

[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.