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Certificate of Analysis

p38/JTV-1

(Recombinant protein expressed in Sf21 insect cells)
Item # 23-049, 23-049-K, 23-049M
Parent Lot # D11HP022N

The data presented in this document apply to the parent lot shown above and to all pack sizes derived from subsequent vialling runs of this parent lot. An alphabetical suffix after the parent lot number is used to denote each vialling run.

Product Description: *N*-terminal cMyc, GST-tagged, recombinant human p38/JTV-1 full length, expressed by baculovirus in Sf21 insect cells. Purified using glutathione sepharose.

Purity 79% by SDS-PAGE and Coomassie blue staining. MW = 64kDa.

Formulation: 0.387mg/ml of enzyme in 50mM Tris/HCl pH7.5, 300mM NaCl, 0.1mM EGTA, 0.03% Brij-35, 270mM sucrose, 1mM benzamidine, 0.2mM PMSF, 0.1% 2-mercaptoethanol. Frozen solution.

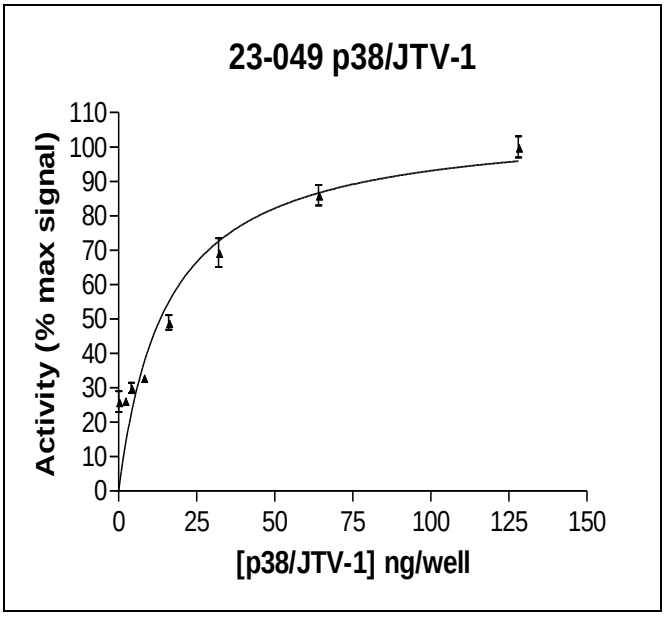
Storage and Stability: On receipt of material store at -70°C. Unopened reagent is stable for a minimum of 1 year from date of shipment when stored at recommended storage temperature. Avoid repeat freeze/thaw cycles. For maximum recovery of product, centrifuge original vial prior to removing the cap.

Handling Recommendations: Rapidly thaw the vial under cold water and immediately place on ice. Aliquot unused material into pre-chilled micro-centrifuge tubes and immediately snap-freeze the vials in liquid nitrogen prior to re-storage at -70°C.

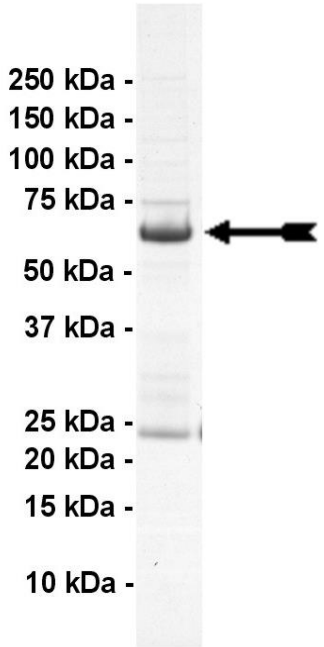
FOR IN VITRO RESEARCH USE ONLY
NOT FOR USE IN HUMANS OR ANIMALS

Quality Control Testing

Assay: This enzyme was titrated in a ubiquitination assay and the results normalised against the maximum signal.



Protein Identity: Confirmed identity as p38/JTV-1 by mass spectrometry.



SDS-PAGE and Coomassie Stain: Purity was assessed by SDS-PAGE and Coomassie blue staining using 3µg of p38/JTV-1.

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Assay Protocol

Reagents:

1. **UBE1, active (Item # 23-021)**
2. **UbcH7, active (Item # 23-047)**
3. **Parkin, active (Item # 23-048)**
4. **p38/JTV-1 (Item # 23-049)**
5. **1x Reaction Buffer**
6. **Biotinylated-Ubiquitin**
7. **Stop Solution**

Assay Outline:

All enzymes and reagents are diluted in the 1x reaction buffer (25mM MOPS pH 7.5, 0.01% Tween 20, 5mM $MgCl_2$).

p38/JTV-1 is incubated with 25mM MOPS pH 7.5, 0.01% Tween 20, 5mM $MgCl_2$, 10µM ATP, 10nM UBE1, 500nM UbcH7, 0.1µg Parkin, and 2µM biotinylated-ubiquitin. The reaction is initiated with the addition of biotinylated-ubiquitin. After 30 minutes at room temperature the reaction is terminated by the addition of 25mM MOPS pH 7.5 containing 125mM EDTA, 150mM NaCl, and 0.05% Tween 20. Reaction products are separated by capture onto a microplate coated with anti-c-Myc antibody and washing with PBS containing 0.05% Tween 20. Ubiquitination of p38/JTV-1 is measured by detection of bound ubiquitin via electrochemiluminescence

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p38/JTV-1 Information

Protein Human p38/JTV-1

Accession number GenBank BC002853

Alternative Names Aminoacyl tRNA synthase complex-interacting multifunctional protein 2, AIMP2, **Multisynthase** complex auxilliary component p38

Key Facts p38/JTV-1 is a scaffold required for the assembly and stability of the multi-tRNA synthetase complex, an essential enzyme that ligates specific amino acids to tRNAs before protein synthesis. It plays a pivotal role in the control of cell proliferation and has been shown to interact with FUSE binding protein, stimulating its ubquitination and degradation and leading to the down-regulation of the transcription factor c-Myc which is required for differentiation of functional alveolar type II cells. Genetic disruption of p38/JTV-1 in mice has been observed to cause neonatal lethality.
p38/JTV-1 is ubiquitinated by the E3 ligase Parkin, leading to its proteosomal degradation. It has been shown to accumulate in Parkin-deficient mice and in the brain tissue of patients with hereditary Parkinson's, suggesting that it may play a role in the pathogenicity of this disease.

Related Products Item # 23-021 UBE1, active, Item # 23-047 UbcH7, active, Item # 23-048 Parkin, active

Selected References

Corti O. *et al*., The p38 Subunit of the Aminoacyl-tRNA Synthetase Complex is a Parkin Substrate: Linking Protein Biosynthesis and Neurodegeneration. Hum Mol Genet., *12*: 1427–1437, 2003

Kim M. J *et al*., Downregulation of FUSE-binding Protein and c-Myc by tRNA Synthetase Cofactor p38 is Required for Lung Cell Differentiation. Nat Genet., *34*: 330-336, 2003

Kim M. J *et al*., p38 is Essential for the Assembly and Stability of Macromolecular tRNA Synthetase Complex: Implications for its Physiological Significance. Proc Natl Acad Sci USA. *99*: 7912-7916, 2002

Ko H. S., *et al*., Accumulation of the Authentic Parkin Substrate Aminoacyl tRNA Synthetase Cofactor, p38/JTV-1, Leads to Catecholaminergic Cell Death. J Neurosci., *25*: 7968-7978, 2005

Reviewed and approved by site quality representative.