

Human P38 γ ProtéGene™ Set

Catalog# P1072
Lot# Labeled on vial

Materials Provided

1. pMEV2HA-P38 γ -WT (P1072a): 20 μ g in 40 μ l TE, 0.5mg/ml.
2. pMEV2HA-P38 γ -K56M (P1072b): 20 μ g in 40 μ l TE, 0.5mg/ml.
3. pMEV2HA-P38 γ -EE (P1072c): 20 μ g in 40 μ l TE, 0.5mg/ml.
4. pMEV2HA-P38 γ -AA (P1072d): 20 μ g in 40 μ l TE, 0.5mg/ml.
5. Product Information Sheet.

Note: Individual plasmids can be ordered separately. Some plasmids are shipped as lyophilized pellet.

Receiving and Storage:

If received in lyophilized form, add 40 μ l sterile DI water to the vial, mix thoroughly by vortex and then collect the contents by centrifuging the vials briefly in a microcentrifuge. If received in liquid form, spin the vials briefly in a microcentrifuge to collect the contents. Store the products at 2-8 $^{\circ}$ C if used immediately or, store at -20 $^{\circ}$ C for extended storage.

Prokaryotic selection:

The kanamycin-resistance gene (aminoglycoside 3' phospho-transferase) expression cassette in the plasmids confers Kanamycin resistance to bacteria cells. Bacterial cells transformed with the plasmids should be maintained and grown in media containing 25-50 μ g/ml Kanamycin (e.g. cat#LK-1100, Pre-Poured LB Agar plates, Biomyx, San Diego, California).

Eukaryotic selection:

The neomycin resistance gene, driven by SV40 early promoter, confers G418 resistance to eukaryotic cells. Stable mammalian cell lines can be selected with G418.

Description of P38 and Mutants

Mitogen-activated protein kinases (MAPKs) cascade relays extracellular signals from cell membrane to the nucleus to induce intracellular responses and to regulate many aspects of cell physiology. These cascades, including JNK, ERK and p38 pathways, consist of distinct members of regulatory enzymes that serially activate one another in response to growth factors, cytokines and other mitogenic stimuli, and lead to activation or inactivation of transcription factors. Proinflammatory cytokine and microbial products activate p38 (SAPK2, MAPK12) gene regulating pathway and cause the expression of multiple cytokine genes including IL-1, IL-6 and TNF alpha. There're four known p38 isoforms (α , β , γ and δ) that share about 60% homology. Please refer to the reference section for key references and the links at the end of this document for further reading.

Molecular Features of the inserts:

Gene: *Homo sapiens mitogen-activated protein kinase 12 (MAPK12)*, (Nicknames: p38 gamma, p38r)
GenBank Reference Sequence: NM_002969
5'-Cloning Site: Xho I
5'-Junction Sequence: 5'...GCTAGC CTC GAG ATG (P38G)...3'
3'-Cloning Site: Xba I
3'-Junction Sequence: 5'...ATG (P38G). TGA TCTAGAGTC-3'

Human P38- γ Nucleotide and Protein Sequence

(1104 bps encoding 367 amino acid residues, with mutation locations marked in red)

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1  ATGAGCTCTC  CGCCGCCCGC  CGGCAGTGGC  TTTTACCACC  AGGAGGTGAC  CAAGACGGCC
   m s s p p p a r s g f y r q e v t k t a
61  TGGGAGGTGC  GCGCCGTGTA  CCGGGACCTG  CAGCCCGTGG  GCTCGGGCGC  CTACGGCGCG
   w e v r a v y r d l q p v g s g a y g a
121  GTGTGCTCGG  CCGTGGACGG  CCGCACCGGC  GCTAAGTGGT  CCATCAAGAA  GCTGTATTCG
   v c s a v d g r t g a k v a i k k l y r
181  CCCTTCCAGT  CCGAGCTGTT  CGCCAAAGGC  GCTACTACGG  AGCTGCGSCT  GCTCAAGCAC
   p f q s e l f a k r a y r e l r i l k h
241  ATCGCCACAG  AGAAGCTGAT  CGGCTGCTGT  GACGTATTCA  CTCTGTATGA  GACCTGTGAT
   m r h e n v i g l l d v f t p d e t l d
301  GACTTCACGG  ACTTTTACCT  GGTGTGCCGC  TTCATGGCCA  CCGACTGGG  CAAGCTCATG
   d f t d f y l v m p f m g t d l g k l m
361  AAACATGAGA  AGCTAGGCGA  GGACCGGATC  CAGTTCCTCG  TGTACCAGAT  GCTGAAGGGG
   k h e k l g e d r i g f l v y g m l k g
421  CTGAGGTATA  TCCACGCTGC  CCGCATCATC  CACAGAGACC  TGAAGCCCGG  CAACCTGGCT
   l r y i h a a g i i h r d l k p g n l a
481  GTGAACGAAG  ACTGTGAGCT  GAAGATCCTG  GACTTGGCCC  TGCCAGGCA  GGCAGACAGT
   v n e d c e l k i l d f g l a r q a d s
541  GAGATGACTG  GGTACGTGGT  GACCCGGTGG  TACCCGGCTC  CCGAGGTGAT  CTTGAATTGG
   e m t g y v v t r w y r a p e v i l n w
601  ATGCGCTACA  CCGACACGGT  GGACATCTGG  TCTGTGGCTG  GCATCATGGC  GGAGATGATC
   m r y t q t v d i w s v g c i m a e e m i
661  ACAGGCAAGA  CGCTGTTCAA  GGGCAGCGAC  CACCTGGACC  AGCTGAAGGA  GATCATGAAG
   t g k t l f k g s d h l d g l k e l m k
721  GTGACGGGGA  CGCCTCCGGC  TGAGTTTGTG  CAGCGCTGTC  AGAGCATGTA  GGCCAGAAGC
   v t g t p p a e f v g r l g s d e a k n
781  TACATGAAGG  GCCTCCCGGA  ATTGGAGAAG  AAGGATTTTG  CCTTATCTCT  GACCAATGCA
   y m k g l p e l e k k d f a a s i l t n a
841  AGCCCTCTGG  CTGTGAACCT  CCTGGAGAAG  ATGCTGGTGC  TGGACGGCGA  GCAGCGGGTG
   s p l a v n l l e k m l v l d a e q r v
901  ACGGCAGGCG  AGCGCTGGC  CCATCCCTAC  TTCCAGTCCC  TGCAAGCAC  GGAAGATGAG
   t a g e a l a h p y f e s l h d t e d e
961  CCCCAGGTCC  AGAAGTATGA  TGACTCCTTT  GACGAGTTTG  ACCGCACACT  GGATGATGG
   p q v q k y d d s f d d v d r t l d e w
1021  AAGCCTGTTA  CTTACAAGA  GGTGCTCAGC  TTCAAGCTAC  CCCGCAGCT  GGGGCTCAGG
   k r v t y k e v l s f k p p r q l g a r
1081  GCTCCCAAGG  AGACGCCCTT  GTCA
   v s k e t p l -
    
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Mutations:

pMEV-P38-WT (P1072a): No mutation
 pMEV-P38-K56M (P1072b): K56M: AAG > ATG
 pMEV-P38-EE (P1072c): T183E: ACT > GAA, Y185E: TAC>GAA
 pMEV-P38-AA (P1072d): T183A: ACT > GCT, Y185A: TAC>GCC

Selected References:

- Li Z, et al, The primary structure of p38 γ : A new member of p38 group of MAP kinase. *Biochem Biophys Res Comm* 228(2):334-340, 1996
- Han J, et al, Molecular cloning of human p38 MAP kinase. *Biochim Biophys Acta* 1265(2-3):224-227, 1995
- Roux PP, Blenis J, ERK and p38 MAPK-activated protein kinases: a family of protein kinases with diverse biological functions. *Microbiol Mol Biol Rev* 68(2):320-344, 2004
- Saccani S, Pantano S and Natoli G, p38-dependent marking of inflammatory genes for increased NF- κ B recruitment. *Nature Immunol* 3:69-75, 2002

Web Resources:

For sequence, references and a comprehensive description, please click the links below or copy and paste the link to your browser address bar:
http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=Nucleotide&list_uids=48255969&dopt=GenBank
<http://www.ncbi.nlm.nih.gov/entrez/dispomim.cgi?id=602399>