

Certificate of Analysis

PTP β , active

(Recombinant protein expressed in *E. coli*)

Item # 14-948, 14-948-K, 14-948M

Parent Lot # D14EP006N

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 GST-tagged, recombinant, human PTP β , amino acids 1643-end, expressed in *E. coli*. Purified using glutathione agarose. Purity 87% by SDS-PAGE and Coomassie blue staining. MW = 69 kDa.

Specific Activity (Parent lot# D14EP006N): 12044U/mg, where one unit of PTP β , active activity is defined as the release of 1nmol of phosphate per minute from the phosphorylated substrate 6,8-difluoro-4-methylumbelliferyl phosphate (DiFMUP) at room temperature.

Formulation: 0.538mg/ml of protein in 25mM Tris/HCl pH 8.0, 75mM NaCl, 10mM GSH, 50% glycerol, 0.5mM DTT. Frozen solution.

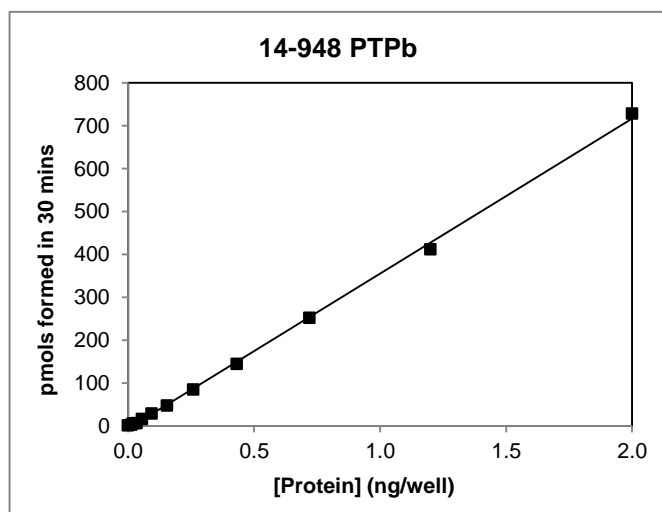
Storage and Stability: On receipt of material store at -70°C. Unopened reagent is stable for a minimum of 1 year from date of shipment when stored at recommended storage temperature. Avoid repeat freeze/thaw cycles. For maximum recovery of product, centrifuge original vial prior to removing the cap.

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

Phosphatase Assay: 0–2ng of this lot of enzyme dephosphorylated 200 μ M DiFMUP in the assay described on page two. Assay background was subtracted from the actual Fluorescence Intensity (FI) to yield the results shown below. Quantification of FI was against a 6,8-difluoro-7-hydroxy-4-methylcoumarin (DiFMU) standard curve.



MS Tryptic Fingerprint: Confirmed identity as PTP β with the translated sequence listed on page three



Certificate of Analysis

Phosphatase Assay Protocol

Stock Solutions:

1. **Reaction Buffer:** 60mM Hepes pH 7.2, 150mM NaCl, 1mM EDTA, 0.17mM DTT, 0.83(v/v)% glycerol, 0.017(w/v)% BSA, 0.002% Brij-35
2. 500 μ M DiFMUP (Molecular Probes Catalogue # D6567) in water.
3. 100mM sodium orthovanadate.
4. 500 μ M DiFMU (Molecular Probes Catalogue # D6566) in water for the calibration curve.

Assay Procedure:

1. Dilute PTP β , active in reaction buffer and use 0–2ng in 15 μ l per assay point.
2. Add 10 μ l DiFMUP 500 μ M stock solution (200 μ M final assay concentration).
3. Incubate for 30 minutes at room temperature.
4. Stop the reaction by adding 5 μ l of 100mM sodium orthovanadate.
5. Read FI using an appropriate reader (Excitation 340 nm; Emission 450 nm).

Subtract the zero enzyme values from each FI reading and calculate the enzyme activity by conversion to nmoles product formed using a DiFMU standard calibration curve.

Certificate of Analysis

PTPβ Sequence Information

Protein	human PTPβ
Tags	N-terminal GST
Native sequence	R243 of the recombinant protein is equivalent to R1643 of human PTPβ
Accession number	GenBank NM_002837.2

Recombinant PTPβ amino acid sequence:

```

1  MSPILGYWKI  KGLVQPTRL  LEYLEEKYEE  HLYERDEGDK  WRNKKFELGL  EFPNLPYYID
61  GDVKLTQ SMA  IIRYIADKHN  MLGGCPKERA  EISMLEGAVL  DIRYGVSRIA  YSKDFETLKV
121  DFLSKLPEML  KMFEDRLCHK  TYLNGDHVTH  PDFMLYDALD  VVLYMDPMCL  DAFPKLVCFK
181  KRIEAI PQID  KYLKSSKYIA  WPLQGWQATF  GGDHPPKSD  LVPRGSPGYP  YDVPDYASPG
241  ASRQKVSHGR  ERPSARLSIR  RDRPLSVHLN  LGQKGNR KTS  CPIKINQFEG  HFMKLQADSN
301  YLLSKEYEEL  KDVGRNQSCD  IALLPENR GK  NRYNNILPYD  ATRVKLSNVD  DDPCSDYINA
361  SYIPGNNFRR  EYIVTQGPLP  GTKDDFWKMV  WEQNVHNIVM  VTQCVEKGRV  KCDHYWPADQ
421  DSLYYGDLIL  QMLSESVLPE  WTIREFKICG  EEQLDAHRLI  RHFHYTVWPD  HGPETTQSL
481  IQFVRTVRDY  INRSPGAGPT  VVHCSAGVGR  TGTFIALDRI  LQQLD SKDSV  DIYGAVHDLR
541  LHRVHMVQTE  CQYVYLHQCV  RDVLRARKLR  SEQENPLFPI  YENVNPEYHR  DPVYSRH
  
```

Recombinant PTPβ nucleotide sequence:

```

1  atgtccccta  tactaggtta  ttggaaaatt  aagggccttg  tgcaaccac  tcgacttctt
61  ttggaatata  ttgaagaaaa  atatgaagag  catttगतग  agcgcgatga  aggtgataaa
121  tggcgaaaca  aaaagtttga  attgggtttg  gagtttccca  atcttcctta  ttatattgat
181  ggtगतगtta  aattaacaca  gtctatggcc  atcatacgtt  atatagctga  caagcacaac
241  atgttgggtg  gttgtccaaa  agagcgtgca  gagatttcaa  tgcttgaagg  agcggttttg
301  gatattगतग  acggtगतग  gagaattgca  tataगतााग  actttgaaac  tctcaaagtt
361  gattttctta  gcaagctacc  tgaaatगतग  aaaatगतग  aagatगतग  atgtcataaa
421  acataattta  atggtगतग  tgtaaccat  cctgacttca  tgttगतग  cgctctगतग
481  gttgttttat  acatggaccc  aatगतग  gatगतग  caaaattगतग  ttgttttaaa
541  aaacगतग  aagctatccc  acaaattगतग  aagtactगता  aatccagcaa  गतातागता
601  tggcctttgc  agggctगता  agccacगतग  ggtगतग  accatcctcc  aaaatcgगतग
661  ctggtttccg  gtगतग  cgggtaaccg  tacgacगतग  cggactacgc  atccccggg
721  gctगतग  agaaaगतग  ccatगतग  gaaagaccct  ctgcccगतग  gagcattगतग
781  agggगतग  cattatctgt  ccacttaaac  ctgggccaगता  aaggtaaccg  gaaaacttct
841  tgtccaataa  aaataaatca  gtttगागग  catttगतग  agctacaggc  tgactccaac
901  taccttctat  ccaaggaata  cgaggगतग  aaagacगतग  gccgaaacca  गतागतग
961  attgactct  tgccggगता  tagagggaaa  aatcगता  acaatata  gccctगतग
1021  gccacgगतग  tgaagctct  caatगतग  gatगतग  gctctgacta  catcaatgcc
1081  agctacatcc  ctggcaacaa  cttcagaaga  gaatacगतग  tcactcaggg  accgcttct
1141  ggcaccaagg  atgacttctg  gaaaatगतग  tgggaacaaa  acgttcaca  catcगतग
1201  gtgaccगतग  gtगतग  gggccगतग  aagtगतग  attactगतग  agcggaccगता
1261  gattcccct  actatgggga  cctcatcctg  cagatगतग  cagagtcctg  cctgcctगता
1321  tggaccatcc  gggगतग  gatatgगतग  gaggaacagc  ttगतग  cagactcatc
1381  cgccactttc  actatacggg  gtggccaगता  catggगतग  cagaaaccac  ccagtctctg
1441  atccगतग  tgagaactगतग  cagggactac  atcaacगता  gccggगतग  tgggcccact
1501  gtगतग  gcagtगतग  tgtggगतग  actggaacct  ttattगतग  ggaccgaa  tc
1561  ctccagगतग  tagactcca  agactctगतग  gacattगतग  gagcगतग  cgacctaga
1621  cttcacaggg  ttcacatगतग  ccagactगतग  tgtगतग  tctacctaca  tcagtगतग
1681  agगतग  tcagagcaa  aaगतग  agtगागा  aaaaccct  gttccaatc
  
```

Certificate of Analysis

1741 tatgaaaatg tgaatccaga gtatcacaga gatccagtct attcaaggca ttga

Reviewed and approved by site quality representative.

Unless otherwise stated in our catalogue or other company documentation accompanying the product(s), our products are intended for research use only and are not to be used for any other purpose, which includes but is not limited to, unauthorized commercial uses, in vitro diagnostic uses, ex vivo or in vivo therapeutic uses or any type of consumption or application to humans or animals.

© 2014 **Eurofins Pharma Discovery Services UK Limited** is an independent member of Eurofins Discovery Services