

Supplementary Fig. S1 Nucleotide and amino acid sequences of CtPFD subunits.

CtPFD1

121 a.a. MW=13423.81

ATGTCACTAAGCACAGAGGCGCTACAAAAGCTTGTGCGGGAGATTGAGTCCCAGGCGATTGCGGCACAGCAGCAGATCAGCCTGGTGAGG
M S L S T E A L Q K L V R E I E S Q A I A A Q Q Q I S L V R

ACACAAATCGCAGCCAAGCAGCGGGAGATGCGGCTGGCGCAGTTGACGCGCAGCGAAATCTCCGACTGCCAGCGATACGGACGTATAT
T Q I A A K Q R E M R L A Q L T R S E I S A L P S D T D V Y

GAAGTGTTGGGAAGATGTTCTGTTGGCGGTGCCGGTCCGGGATGCAGGATAAGCTGGGGAGCAGATCAAGACTCTAGAGGGCGAGGTG
E G V G K M F V A V P V P A M Q D K L G E Q I K T L E G E V

GAGACGATGAACAAGAGGCTGCACTACCTGGAGACAACCTGCGAAGAACAGCCAGGAGCACATCGAGAAGATTATCAAGGGCGGTGGTCAG
E T M N K R L H Y L E T T A K N S Q E H I E K I I K G G G Q

TCATAA

S *

CtPFD2

117 a.a. MW=13449.85

ATGGCGACACAGTCTGCAGCAGCAAAGAGGCAGCAAGACCTCCAGCTGCAGTATACCACTTATAAGAACACCCTGCAGCAAATTGCCAG
M A T Q S A A A K R Q Q D L Q L Q Y T T Y K N T L Q Q I A Q

AAGATTGGAGATGTCGAGCAGGAGGCTGAGGAGCACAAGCTTGTCTTGGAGACTTTGGAGCCTCTCCAGGTGACCGCAAATGCTTCCGT
K I G D V E Q E A E E H K L V L E T L E P L P G D R K C F R

ATGATCAATGGCGTCTTGGTGGAAAGAACCGTCCAGGATGTGATTCTGCTCTTCGGACCAACGCTGAAGGGCTGAAGAAGTCTCGAA
M I N G V L V E R T V Q D V I P A L R T N A E G L K K V L E

GATCTTGTAACAATAACAAGCGAAACAGGATGAACCTTGAGCAGTGGAGAAGAAAAACAACGTTCCAGGTCGTGCAGTCATGA
D L V K Q Y K A K Q D E L E Q W K K K N N V Q V V Q S *

CtPFD3

180 a.a. MW=20558.89

ATGGCCAGTCAAGCAAAAGAAGCGGCGACAAAGGACGCTACCCCTCGAACCCCGCGGATTCCCTATGCGCCGTTCTGTCGACAAGGTC
M A S Q A K E A A T K D A T P S N P R G I P Y A P F V D K V

GAGGACTATGTGTCGTACGGGACGATGTGAGCCGACGCTGCGGCGATTCCAAGAGCTGATCGCGAAGTATCAGTTTCATGGAGCAGAAC
E D Y V S S R D D V E P T L R R F Q E L I A K Y Q F M E Q N

CTGCAGCGGAGGGTAGCCGGTCTGAAGGATAAGATGCCCGACATCCAAAAGACCTTGATACGGTTCGGTCTTGAAGCTCAGGACGTCC
L Q R R V A G L K D K M P D I Q K T L D T V R F L K L R T S

GAGTCGATCCGATCGAGACAACCTTTGAGCTGAACGACACGCTCTATGCGAAGGCCGAGATCCCGCCGACGGACGAGGTCTATCTCTGG
E S D P I E T T F E L N D T L Y A K A E I P P T D E V Y L W

CTTGGTGCCAACGTCATGCTAGCGTATCCTCTCGACGAAGCAGAGCAGCTCCTGGATTGAAACTCAAGGGCGCAAAGCAGAGTCTGGCT
L G A N V M L A Y P L D E A E Q L L D S K L K A A K Q S L A

AACTGCGAGGAGGACCTGGACTTTCTGCGGAGCAGATCACGACCATGGAGGTCGCCATCGCGAGAGTGACAACCTGGGACGTCAGCTCA
N C E E D L D F L R E Q I T T M E V A I A R V Y N W D V S S

TAA

*

CtPFD4

134 a.a. MW=15645.89

ATGAACATAAGTCGGCGAATGGAGGAGGAGGAGGCTATAGCGAGGATGTCGAGGTACGCCGAGGACCAGGACAAGATCAACAGGTTCC
M N I S R R M E E E E A I G E D V E V R R E D Q D K I N R F
AGCCGTCTGCATGGCCACGAATTGGCGATTGAGGAGGAGCTGAAGGCCAAGCAGAAGGAGAAGGAAGAGTTGGACGATATCACAAACGGAG
S R L H G H E L A I E E E L K A K Q K E K E E L D D I T T E
TTGGAGCTTGCCGACGAAGACGAGCTTGCCATACAAAGTCGGCGACGCATTCTTCCACATTCCCCTCCCCAAGCCCAAGAGATGCTC
L E L A D E D E L V P Y K V G D A F F H I P L P Q A Q E M L
GGTATTTTCGACGGCGAAGCTCGAGGAGGAAATCTCACAACCTGAGGAGAAGCTGAGCAATATCCGCGATGAGATGTCACAACCTAAAGGTG
G I S T A K L E E E I S Q L E E K L S N I R D E M S Q L K V
GAGTTGTACGCAAGATTTGGCAAGACCATCAATCTCGAGACATGA
E L Y A R F G K T I N L E T *

CtPFD5

165 a.a. MW=17724.67

ATGGCCAGCCGAGCGGTCAAGATAAACAAAGGACAAGGAGAGGAGGAGTTCGATCTTGCATCCCTCAGCGTACAACAACCTCAGTACAGTGAAG
M A S R S G Q D K Q G Q G E A V D L A S L S V Q Q L S T V K
AAGCAGCTTGACGGAGAAGTCGAGCACCTATCAACGTCGTACGCGCAGTTGGTGGCCGCTCAGGGGAAGTTTAAAGAATGTCTACGCATT
K Q L D G E V E H L S T S Y A Q L V A A Q G K F K E C L R I
GTCAAGGACAGAGCATCAACGCTTGAGAAGGATAAAACCGTTCTCGTTCCCCTTACCAACTCCCTCTACGTCAAGGGAAGCTGTCCGAC
V K D R A S T L E K D K T V L V P L T N S L Y V K G K L S D
CCGGAACACGTGCTCGTTGATGTCGGAACGGGCTTCTACGTGAAAAGGACACGACCAGCGCAGCTGAATTTTACGAAGCAAGATCAAG
P E H V L V D V G T G F Y V E K D T T S A A E F Y E G K I K
GAGCTAGCGACCAATATCCAGGGACTGGAGGGCATTGTGCAGGCAAAAACAGCAAATCTCCGAGTTGTGGAAGAAGTCCTTCGGCAAAA
E L A T N I Q G L E G I V Q A K T A N L R V V E E V L R Q K
GTACTCGCTTCAAACGCTGCTGGAACGGGAGCAAAGCAATCTTCATGA
V L A S N A A G T G A K Q S S *

CtPFD6

124 a.a. MW=14013.59

ATGGCTGAAATCCAGGCCCGCCTTCAAGCCCTCTCTGAGGAGTTTCAGCAAGCTCCAACAAGATCTCCAAGATGCTCTGCAATCAAGACAG
M A E I Q A R L Q A L S E E F S K L Q Q D L Q D A L Q S R Q
AAGCTCGAGGCGCAGAAGCAAGAAAACCTTGGGGTCCAAAAGGAGTTCAAGAAGCTCAAGGAGGGCGAAAACATCTACAAACTGATCGGC
K L E A Q K Q E N L G V Q K E F K K L K E G E N I Y K L I G
CCTGTTCTTCTCAAGCAAGACAAGGTTGATGCAGAGAGCATAGTCAATGGTCTAGACTTTATTGACAAGGAAATCAACCGAATAGAA
P V L L K Q D K V D A E S I V N G R L D F I D K E I N R I E
ACTCTCATCAAGGAACTCCAGGGCAAGATCGAGAAAAAGAAAGGGGAGATTATCCAGGTGCAAAGTGGTTTGAAGCAGCGGCAGTTCAA
T L I K E L Q G K I E K K K G E I I Q V Q S G L Q A A A V Q
GCCGTCAAGGCATGA
A V K A *

Supplementary Fig. S2 Amino acid sequence alignments of Prefoldin subunits

CtPFD1 MSLS-TEALQKLVREIESQAIAAQQQISLVRTQIAAKQREMRLAQLTRSEISALPSDTDV
 ScPFD1 -----MSQIAQEMTVSLRNARTQLDMVNQQLAYLDRQEKLAELTKKELESYPTD-KV
 HsPFD1 MAAPVDLELKKAFTELQAKVIDTQQKVKLADIQIEQLNRTKKHAHLTDTEIMTLVDETMM
 EnPFD1 MSIP-NEALQKLLQEIESRVITSQQQIGITKAQMTSKNRDIRMLELTSKELSSLPADTNV
 : . : * : : : : : : . * : : * : . * * . * : : : : . :

CtPFD1 YEGVGKMFVAVPVPAMQDKLGEQIKTLEGEVETMNKRLHYLETTAKNSQEHIEKIIKGGG
 ScPFD1 WRSCGKSFILQDKSKYVNDLSHDETVLLDQRKTLKIKKNYLETTVEKTIDNLKALMKN--
 HsPFD1 YEGVGRMFILQSKEAIHSOLLEKQKIAEEKIKELEQKKSYLERSVKEAEDNIREMLMARR
 EnPFD1 YEGVGKMFVAVPRATVDKRISTETGELKGEIENLEKRMNYLEMFTFKNSRENLEAILKSGR
 : . . * : * : . : . : : : : : * * * : : : : : : : . : :

CtPFD1 QS
 ScPFD1 --
 HsPFD1 --
 EnPFD1 A-

Ct: *Chaetomium thermophilum*
Sc: *Saccharomyces cerevisiae*
Hs: *Homo sapiens*
En: *Emericella nidulans*

CtPFD2 -----MATQSAAAKRQODLQLQYTTYKNTLQQIAQKIGDVEQEAEHKLVLLET
 ScPFD2 -----MEQRNNVFQAKYNEYKQILEELQTKIIELGHDKDEHTIVIKT
 HsPFD2 MAENSGRAGKSSGSGAGKGAVSAEQVIAGFNRLRQEQRLASKAAELEMELNEHSLVIDT
 EnPFD2 -----MASQPQIAPKKQQELQAQYTNFKNTLQQLAQKIGDIEQEAEHKLVIDT

: . :. :: . : * :: : : ** . : * : . *

CtPFD2 LEPLPGDRKCFRMINGVLVERTVQDVIPALRTNAEGLKKVLEDLVKQYKAKQDELEQWKK
 ScPFD2 LKDAEPTRKCYRMIGGALVESDVQTSLEPKKENIEGTISKMKETLIQTAKFEKWKK
 HsPFD2 LKEVDETRKCYRMVGGVLVERTVKEVLPALENNKEQIQKIIEITLQQLQAKGKELNEFRE
 EnPFD2 LEPLPQDRTCFRMVNGVLVERTVADVLP TLKTN SDGLKQVLEDMLKQYKSKQSELDNWKK

* : * . * : ** : . * . *** * : * * . . : : : : . . : : . * : : : : : :

CtPFD2 KNNVQVVQS
 ScPFD2 DNKIQVVKV
 HsPFD2 -----
 EnPFD2 KNNIQVVQP

CtPFD3 MA-----SQAKEAATKDATPSNPRGIPYAPFVDKVEDYVSSRDD--VEPTLRRFQELIAKY
 ScPFD3 -----MDTLFNSTENKARGIPQAPFIENVNEIIKDPD--FELCFNKFQERLSKY
 HsPFD3 MAAVKDSCGKGEMATGNRRLHLGIPEAVFVEDVDSFMKQPGNETADTVLKKLDEQYQKY
 EnPFD3 -----MADKKPSDPTQTNPRGIPVAPFVDNVSDYVSSRAD--VEPTMRSFQEMISKY

. *** * * : : . * . . : . . : : : . : : * **

CtPFD3 QFMEQNLQRRVAGLKDKMPDIQKTLDTVRFLKLRT----SESDPIETTFELNDTLYAKAE
 ScPFD3 KFMQESKLATIKQLKTRIPDLENTLKICQSLRNHSDEGDESDEPILLHYQLNDTLYTKAQ
 HsPFD3 KFMELNLAQKKRRLKGQIPEIKQTLLEILKYMQKKK----ESTNSMETRFLADNLYCKAS
 EnPFD3 QFMEVNTSRRAGLREKIPDIKKTLEMVTFKARKE--SSSATPLETNFELNDTLYARAT

: ** : . * : : : * : : : * * . : : : . . : : * * . * * : *

CtPFD3 IPP-----TDEVYLWLGANVMLAYPLDEAEQLLDSKLKAAKQSLANCEEDLDFLREQITT
 ScPFD3 VDIPEDRADLKVGLWLGADVMEYPIDEAIELLKKKLADSEQSLTVSTEDVEFLRENITT
 HsPFD3 VPP-----TDKMCLWLGANVMLEYDIDEAQALLEKNLSTATKNLDSLEEDLDFLRDQFTT
 EnPFD3 VDDPAH---TEEVYLWLGANVMLAYPIAEQAAMLEDKLAAAEQSLANCDEDLEFLREQITT

: : : * * * * * : * * * * * * : * * : * . . : * : : . * * * : : * * * * * : : * *

CtPFD3 MEVAIARVYNWDVSS-----
 ScPFD3 MEVNCARLYNWDVQRRQDLKQAQEGTKNLKI
 HsPFD3 TEVNMARVYNWDVKRRNKDDSTKNKA-----
 EnPFD3 LEVATARVYNWDVVQRRKERAEGKEETS---

** ** : * * * * *

CtPFD4 ----MNISRMEEEEAI GEDVEVRREDQDKINRFSRLHGHELAI EEELKAKQKEKEELDD
 ScPFD4 -----MELLPOGQRNNTQVTFEDQOKINEFSKLIMRKDAIAQELSLQREEKEYLDD
 HsPFD4 -----MAATMKKAAAEDVNVTFEDQOKINKFARNTSRITELKEEIEVKKKQLQNLED
 EnPFD4 MMNHRMLSKEDEAAIGNSEDTEVRREDQEKINRFSRLHQRETVLEEKLKKGKQKDKEDLEE
 . : : . : * ** : ** * . * : : : : : : : : : : : * : :

CtPFD4 ITTELELADEDE-LVPYKVGDAFFHIPLPQAQEMLGISTAKLEEEISQLEEKLSNIRDEM
 ScPFD4 VSLEIELIDEDE-PVQYKVGDLFIFMKQSKVTAQLEKDAERLDNKIETLEDKQRDIDSRL
 HsPFD4 ACDDIMLADDDCLMIPYQIGDVFI SHSQEETQEMLEEAKKNLQEEIDALESRVESIQRVL
 EnPFD4 ISTELELADEDE-LIPYKIGDSFVHLPLEEAQTLLASSTEQIDSEVAKLEETLSDLRDEM
 : : * * : * : * : : * * * . : . * . : : : : : * * . . : :

CtPFD4 SQLKVELYARFGKTINLET---
 ScPFD4 DALKAILYAKFGDNINLER---
 HsPFD4 ADLKVQLYAKFGSNINLEADES
 EnPFD4 QQLKVALYARFGRSINLET---
 ** . ** * : ** . *****

CtPFD5 --MASRSGQDKQGQGEAVDLASLSVQQLSTVKKQLDGEVEHLSTSYAQLVAAQGKFK---
 ScPFD5 -----MSSQKIDLTKLNPEQLNAVKKQDFDQELQHFQSLQALQALMAKGFQFT---
 HsPFD5 -----MAQSINITELNLPQLEMLKNQLDQEVFLSTSYAQLKVVQTKYVEAK
 EnPFD5 MPPQNTPASDSDAPPGAVNISSLSTPQLRALQTRLSTELEHLTSSHAKLRAAQSRFR---
 ::::.* ** :: ::. *::: * * .: ::

CtPFD5 ECLRIVKDRAS-----TLEKDKTVLVPLTNSLYVKGKLSDPEHVLVDVGTGFYVEKDTTS
 ScPFD5 ECIDDIKTVS-----QAGNEGQKLLVPASASLYIPGKIVDNKKFMVDIGTGYYVEKSAEA
 HsPFD5 DCLNVLNKS-----NEGKELLVPLTSSMYVPGKLHDVEHVLIDVGTGYYVEKTAED
 EnPFD5 DCVRSINIEGVIGSAKKGTEGKDEILVPLTSSLYVKGRLTDREKVLVDVGTGYYVEKTAAK
 :* : : : . :*** : *:* : *:: * :::: * :***:*** :

CtPFD5 AAEFYEGKIKELATNIQGLEGIVQAKTANLRVVEEVLKQKVLASNAAGTGAKQSS-----
 ScPFD5 AIAFYQKKVDKLNKESVQIQDIIKEKTQYSLSIEAQIRQAAIRQHEAMSKQQQQQKQKES
 HsPFD5 AKDFFKRKIDFLTKQMEKIQPALQEKHAMKQAVMEMMSQKIQQLTALGAAQATAKA-----
 EnPFD5 AIEFYEQKVKELETNLTELEKLVQTKSSQQLFEDALRQKLMSEGAASSAQAAG-----
 * *:: *:. * .: :: :: * . : * :

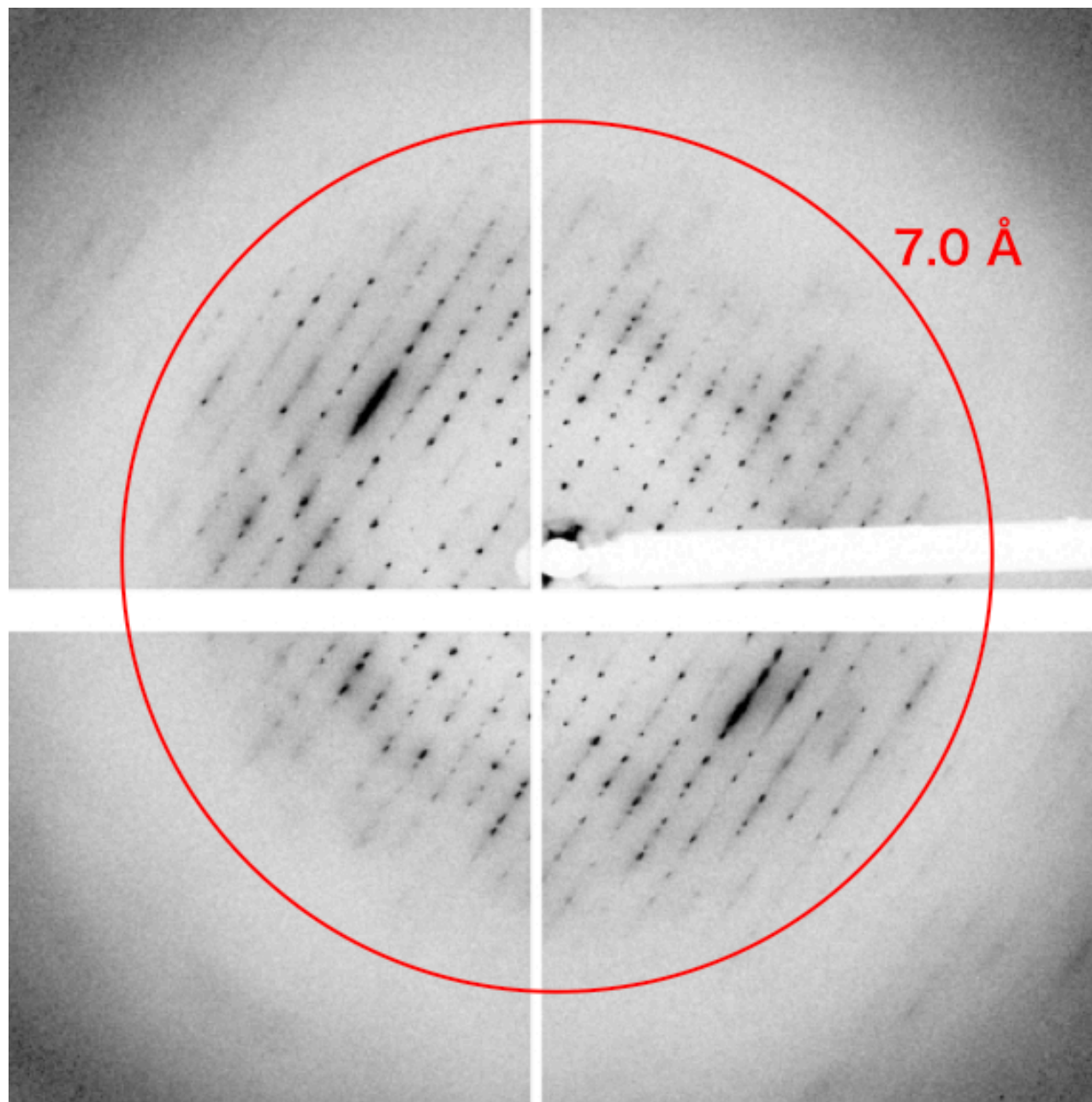
CtPFD5 ----
 ScPFD5 STA
 HsPFD5 ----
 EnPFD5 ----

CtPFD6 -MAEIQARLQALSEEF SKLQQDLQDALQSRQKLEAQKQENLGVQKEFKKLKEGENIYKLI
 ScPFD6 -----MSELGAKYQQLQNELEEFIVARQKLETQLQENKIVNEEFDQLEEDTPVYKLT
 HsPFD6 MAELIQKKLQGEVEKYQQLQKDL SKSMGRQKLEAQLTENNIVKEELALLDGSNVVFKLL
 EnPFD6 -MADPQKQMQUALSDEYQALQTELDGLVDARQKLESQQQENKSVQAEFNSLDDDANIFKLI
 :. :. ** :* . : .*****:* ** * : * : * . . : : **

CtPFD6 GPVLLKQDKVDAESIVNGRLDFIDKEINRIETLIKELQGKIEKKKGEIIQVQSGLQAAAV
 ScPFD6 GNVLLPVEQSEARTNVDKRLEFIEITEITRCEKNIRDKQEELEKMRSELIKLNNTAASTGP
 HsPFD6 GPVLVKQELGEARATVGKRLDYITAEIKRYESQLRDLERQSEQQRETLAQLQOEFORAQA
 EnPFD6 GPVLLKQDKTEALMAVNGRLEFIEKEIQRIEGQIKENQDKSDKKRAEIVQYQSQIQQQAA
 * ** : : : * * . ** : : * ** * * : : : : : : : : : .

CtPFD6 QAVKA-----
 ScPFD6 GR-----
 HsPFD6 AKAGAPGKA
 EnPFD6 AAAASA---

Supplementary Fig. S3 X-ray diffraction pattern from a crystal of CtPFD.



Supplementary Table S1 Primers, vectors, host strains for expression of PFD subunits

PFD1

Forward primer: GGAATTCCATATGTCCTAAGCACAGAGGCGC

Reverse primer: CGGGATCCATTATGACTGACCACCGCCCTT

Expression vector: pET-23b

Expression host: *E. coli*. BL21 star (DE3) pRARE

Complete amino-acid sequence of the construct produced:

MSLSTEALQKLVREIESQAIAAQQQISLVRTQIAAKQREMRLAQLTRSEISALPSDTDVYEGVGMFVAVPVPAMQDKLGE

QIKTLEGEVETMNRRLHYLETTAKNSQEHIKIKGGGQS

Molecular Mass: 13290.77 g/mol

PFD2_His

Forward primer: GGAATTCCATATGCACCACCACCACCACCACATCGAAGGGCGTGCGACACAGTCTGCAGC

Reverse primer: CGGGATCCTCATGACTGCACGACCTGAACG

Expression vector: pET-23b

Expression host: *E. coli*. BL21 star (DE3) pRARE

Complete amino-acid sequence of the construct produced:

MHHHHHHIEGRMATQSAAAKRQDDLQYTTYKNTLQQAQKIGDVEQEAEEHKLVLLETLEPLPGDRKCFRMINGVLVE

RTVQDVIPALRTNAEGLKKVLEDLVKQYKAKQDELEQWKKKNNVQVVQS

Molecular Mass: 14858.17 g/mol

PFD3_Strep

Forward primer: GGAATTCCATATGGCCAGTCAAGCAAAAGAAGC

Reverse primer: CTCGAGTTATTTTCAAACTGTGGATGCGACCAACGCCCTTCGATTGAGCTGACGTCCCAGTTGTACAC

Expression vector: pET-23b

Expression host: *E. coli*. Rosetta2 (DE3) pLysS

Complete amino-acid sequence of the construct produced:

MASQAKEAATKDATPSNPRGIPYAPFVDKVEDYVSSRDDVEPTLRRFQELIAKYQFMEQNLQRRVAGLKDKMPDIQKTLD

TVRFLKLRITSESDPIETTFELNDTLYAKAEIPPTDEVYLVWGANVMLAYPLDEAEQLLDSKLLKAAKQSLANCEEDLDFLRE

QITTEVAIARVYNWDVSSIEGRWSPQFEK

21918.95 g/mol

PFD4

Forward primer: GGAATTCCATATGAACATAAGTCGGCGAATGGAG

Reverse primer: CGGGATCCTCATGTCTCGAGATTGATGGTCTTG

Expression vector: pET-23b

Expression host: *E. coli*. Rosetta2 (DE3) pLysS

Complete amino-acid sequence of the construct produced:

MNISRRMEEEEAIGEDVEVRREDQDKINRFSRLHGHELAIEEELKAKQKEKEELDDITTELELADEDELVPYKVGDAFFHIP

LPQAQEMLGISTAKLEEEISQLEEKLSNIRDEMSQLKVELYARFGKTINLET

Molecular Mass: 15515.05 g/mol

PFD5

Forward primer: GGAATTCCATATGGCCAGCCGCGAGC

Reverse primer: CGGGATCCTCATGAAGATTGCTTTGCTCCCG

Expression vector: pET-23b

Expression host: *E. coli*. BL21 star (DE3) pRARE

Complete amino-acid sequence of the construct produced:

MASRSGQDKQGGEAVDLASLSVQQLSTVKKQLDGEVEHLSTSYAQLVAAQGFKECLRIVKDRASTLEKDKTVLVPLT

NSLYVKGKLSDEPHVLVDVGTGFYVEKDTTSAAEFYEGKIKELATNIQGLEGIVQAKTANLRVVEEVLVLRQKVLASNAAGT

GAKQSS

Molecular Mass: 17591.16 g/mol

PFD6

Forward primer: GGAATTCCATATGGCTGAAATCCAGGCC

Reverse primer: CGGGATCCTCATGCCTTGACGGCTTGAAC

Cloning vector: pET-23b

Expression vector: pET-23b

Expression host: *E. coli*. BL21 star (DE3) pRARE

Complete amino-acid sequence of the construct produced:

MAEIQARLQALSEEFSKLLQDLQDALQSRQKLEAQKQENLGVQKEFKLKEGENIYKLIQVLLKQDKVDAESIVNGRLD

FIDKEINRIETLIKELQKIEKKKGEIIVQVQSGLQAAAQVAVKA

Molecular Mass: 13879.64 g/mol