

Vector pTREX-3flag-ddFKBP-EYFP for Regulated Gene Expression in *Trypanosoma cruzi*

Catalog No. NR-36603

Product Description: Vector pTREX-3flag-ddFKBP-EYFP is a modified pTREX vector which uses an N-terminal fusion of a ligand controlled destabilization domain (ddFKBP) to Enhanced Yellow Fluorescent Protein (EYFP) to regulate gene expression in *Trypanosoma cruzi*.

Lot¹: 61277424

Manufacturing Date: 02NOV2012

TEST	SPECIFICATIONS	RESULTS
Confirmation of Insert by Restriction Enzyme Digestion Digestion with <i>HindIII</i> and <i>XhoI</i> ²	~ 1.1 kb (insert); ~ 6 kb (vector)	~ 1.1 kb (insert); ~ 6 kb (vector)
Confirmation of Insert by PCR ³	~ 1.6 kb amplicon	~ 1.6 kb amplicon
Confirmation of Insert by Sequencing Insert sequence 1-397 (Figure 1) Insert sequence 394-1130 (Figure 1)	Consistent with FKBP domain Consistent with EYFP gene	Consistent with FKBP domain ⁴ Consistent with EYFP gene ⁵
DNA Content by PicoGreen[®] Measurement	0.7 µg to 1.5 µg per vial	0.8 µg per vial
OD₂₆₀/OD₂₈₀ Ratio	1.8 to 2.1	1.8
Bacterial Transformability (Post-Vial) ⁶	≥ 100 colonies	≥ 100 colonies

¹Produced in One Shot[®] TOP10 chemically competent *Escherichia coli* (Invitrogen™) and extracted using a QIAGEN[®] EndoFree[®] Plasmid Maxi Kit.

²New England Biolabs R0104S (*HindIII*) and R0146S (*XhoI*).

³PCR was performed with primers designed to amplify 281 bases upstream and 177 bases downstream of the 3flag-ddFKBP-EYFP region. PCR primers used for amplification were pTREX-281 Forward- 5'GTCAAAGCGCCGTGTGGATG 3' and pTREX-177 Reverse- 5'GAAGTCATCGCCACGCCTAC 3'. Conditions used for PCR are available upon request.

⁴GenBank: NM_054014 bases 181 to 498

⁵GenBank: AY818380 bases 1326 to 2058

⁶One Shot[®] TOP10 *Escherichia coli* were transformed with 13 ng of plasmid DNA.

Figure 1. Vector pTREX-3flag-ddFKBP-EYFP Insert Sequence

```

ddFKBP (1, 397)
|
1   ATGGACTACAAAGACCATGACGGTGATTATAAAGATCATGACATCGACTACAAGGATGAC 60
61  GATGACAAGGTGCAGGTGGA AACCATCTCCCCAGGAGACGGGCGCACCTTCCCCAAGCGC 120
121 GGCCAGACCTGTGTGGTGC ACTACACCGGGATGCTTGAAGATGGAAAGAAAGTCGATTCC 180
181 TCCCGGGACAGAAACAAGCCCTTTAAGTTTATGCTAGGCAAGCAGGAGGTGATCCGAGGC 240
241 TGGGAAGAAGGGGTTGCC CAGATGAGTGTGGGTGAGAGAGCCAAACTGACTATATCTCCA 300
301 GATTATGCCTATGGTGCCACTGGGCACCCAGGCATCATCCCACCACATGCCACTCTCGTC 360
      EYFP (394, 1130)
      |
361 TTCGATGTGGAGCTTCTAAAACCGGAAGAATTCGTGAGCAAGGGCGAGGAGCTGTTCCACC 420
421 GGGGTGGTGCCCATCTGGTGCAGCTGGACGGCGACGTAAACGGCCACAAGTTCAGCGTG 480
481 TCCGGCGAGGGCGAGGGCGATGCCACCTACGGCAAGCTGACCCTGAAGTTCATCTGCACC 540
541 ACCGGCAAGCTGCCCGTGCCCTGGCCCACCCTCGTGACCACCTTCGGCTACGGCGTGCAG 600
601 TGCTTCGCCCCTACCCCGACCACATGAAGCAGCACGACTTCTTCAAGTCCGCCATGCC 660
661 GAAGGCTACGTCCAGGAGCGCACCATCTTCTTCAAGGACGACGGCAACTACAAGACCCGC 720
721 GCCGAGGTGAAGTTCGAGGGCGACACCCTGGTGAACCGCATCGAGCTGAAGGGCATCGAC 780
781 TTCAAGGAGGACGGCAACATCCTGGGGCACAAGCTGGAGTACAACAGCCACAAC 840
    
```

Certificate of Analysis for NR-36603

841 GTCTATATCATGGCCGACAAGCAGAAGAACGGCATCAAGGTGAACTTCAAGATCCGCCAC 900
901 AACATCGAGGACGGCAGCGTGCAGCTCGCCGACCACTACCAGCAGAACACCCCATCGGC 960
961 GACGGCCCCGTGCTGCTGCCCCACAACCACTACCTGAGCTACCAGTCCGCCCTGAGTAAA 1020
1021 GACCCCAACGAGAAGCGCGATCACATGGTCTGCTGGAGTTCGTGACCGCCGCCGGGATC 1080
1081 ACTCTCGGCATGGACGAGCTGTACAAGTCCGGCCGGACTCAGATCTCGAG 1130

Date: 15 OCT 2013

Signature: 

Title: Technical Manager, BEI Authentication or designee

ATCC[®], on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC[®]'s knowledge.

ATCC[®] is a trademark of the American Type Culture Collection.

You are authorized to use this product for research use only. It is not intended for human use.

