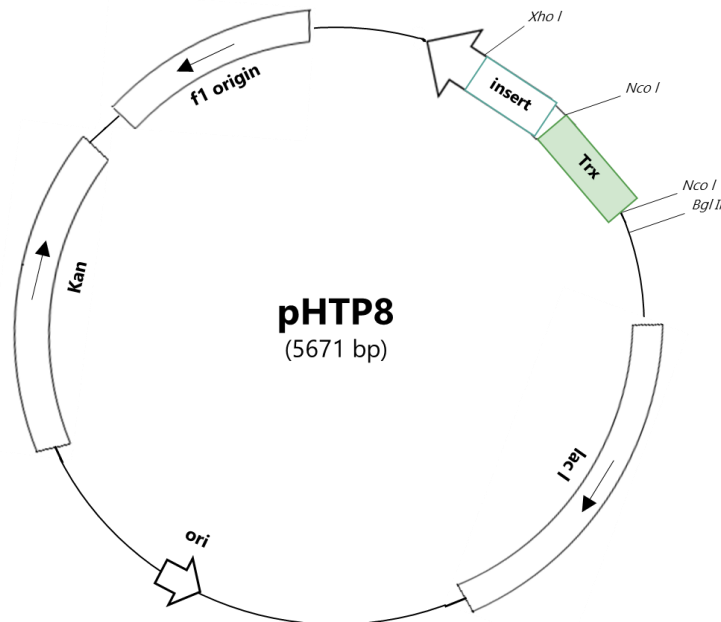


pHTP8 Expression Vector

pHTP8 was designed for the cloning and expression of high-levels of recombinant proteins in *Escherichia coli*. Recombinant proteins are expressed in fusion with the thioredoxin (Trx) protein, which is commonly used to promote solubility and folding of its fusion partners. This vector, included in the portfolio of NZYTech pHTP expression vectors, is part of the NZYEasy Cloning & Expression System. pHTP8 contains two poly-histidine (6xHis) sequences (N- and C-terminal) which allow subsequent recombinant protein purification by immobilized metal ion affinity chromatography (IMAC).

1. Vector Map



pHTP8 Cloning/Expression Region

<i>Nco I</i>	Trx	<i>Nco I</i>	His-Tag
CCATGG GTAGCGATAAAATTATTCACCTGACT . 330bp . CTCGACGCTAACCTGGCCGGTTCTG CCATGG GCAGCAGCC CATCATCATCATCAC AGCAGCGGC			
MetGlySerAspLysIleIleHisLeuThr . 110aa . LeuAspAlaAsnLeuAlaGlySerAlaMetGlySerSerHisHisHisHisHisHisSerSerGly			
CCTCAGCAAGGGCTGAGG / ✂ / CCTCAGCTTCCGCTGAGGTCGTCGACAAGCTTGCGGCCGCA	<i>Xho I</i>	His-Tag	<i>STOP</i>
ProGlnGlnGlyLeuArg / ✂ / ProGlnLeuProLeuArgSerValAspLysLeuAlaAlaAlaLeuGluHisHisHisHisHisHis*	CTCGAGCACCACCACCACCACCAC		TGAGATCCGGCTGCT

✂ Represents the site where the gene will be inserted.

Note: For correct expression, inserted gene needs to be in frame with pHTP8 5' gene sequence. Inserts correctly cloned into pHTP8 will maintain reading frames starting on the ATG codon.

2. Vector Sequence (5671 bp)

TGGCGAATGGGACGCGCCCTGTAGCGGGCGCATTAAAGCGCGGGGGTGTGGTGGTTACGCGCAGCGTGACCCTGACACTTGCCAGCGCCCTAGCGCCCGCTCCCTTCGCTTCT
 TCCCTTCCTTCTCGCCACGTTTCGGCGGCTTTCCCGCTCAAGCTCAAATCGGGGGCTCCCTTAGGGTTCGGATTTAGTGTCTTACGGCACCTCGACCCCAAAAACTTGAT
 TAGGGTATGGTTACGTTAGTGGGCGATCGCCCTGATAGACGGTTTTTCGCCCTTGACGTTGGAGTCCACGTTCTTTAATAGTGGACTTGTTCCAAACCTGGAAACAACACT
 CAACCCTATCTCGGTCTATTCTTTGATTTATAAGGGATTTTCCGGATTTTCGGCTATTGGTTAAAAAATGAGCTGATTTAACAAAAATTTAACCGGAATTTTAAACAAAATAT
 TAAGTTTACAATTTACAGTGGCACTTTTCGGGGAAATGTGCGCGGAACCCCTATTGTTTATTTTCTAAATACATTCAAATATGATCCGCTCATGAATTAATCTTAGAA
 AAACATCATCGAGCATCAAATGAAACTGCAATTTATTCATATCAGGATTATCAATACCATTATTTTGAAGAAAGCCGTTTCTGTAATGAAGGAGAAAACTCACCGAGGCAGTTCC
 ATAGGATGGCAAGATCCTGGTATCGGTTCGGATTCCGACTCGTCCAACATCAATACAACCTAATTAATTTCCCTCGTCAAAAAAAGGTTATCAAGTGAAGAAATCACCATGA
 GTGACGACTGAATCCGGTGAAGTGGCAAAAGTTTATGCATTTCTTCCAGACTTGTTCACAGCCGACGCTTACGCTCGTCAATCAAATCACTCGCATCAACCAAAACCGTT
 ATTCATTGCTGATTGCGCTGAGCGAGACGAAATACGGGATCGCTGTAAAAGGACAATTACAAACAGGAATCGAATGCAACCGGCGCAGGAACACTGCGAGCGCATCAACAA
 TATTTTACCTGAATCAGGATATCTTCTAATACCTGGAATGCTGTTTTCCCGGGGATCGCAGTGGTGAATACCATGCATCATCAGGAGTACGGATAAAATGCTTGATGGT
 GGAAGAGGCATAAAATCCGTCAGCCAGTTAGTCTGACCATCTCATCTGTAACATCAATGGCAACGCTACCTTTGCCATGTTTCAGAAACAACCTGCGGCATCGGGCTTCCC
 ATACAATCGATAGATTTGTCGACATTTGCCGACATTTATCCCGACATTTATACCATATAAATCAGCATGTCATGTTGAATTTAATCCGCGCCTAGAGCAAGACGTTTT
 CCCGTTGAATATGGCTCATAACCCCTTGTATTACTGTTTATGTAAGCAGACAGTTTTATTGTTTATGACCAAAATCCCTTAACGTGAGTTTTGCTTCCACTGAGCGTCAGA
 CCCCGTAGAAAAGATCAAAGGATCTTCTGAGATCCTTTTTTTCGCGGTAATCTGCTGCTGCAAAACAAAAAACCCCGCTACACGCGGTGGTTTTGTTGCCGGATCAAG
 AGCTACCAACTCTTTTTCCGAAGGTAACCTGGCTTCAGCAGAGCCAGATACCAAACTGTCCTTCTAGTGTAGCCGTAAGTTAGGCCACCCTCAAGAACTCTGTAGCACCG
 CCTACATACCTCGCTCTGTAATCTGTACCAGTGGCTGCTGCCAGTGGCGATAAGTCTGCTCTTACCAGGTTGGACTCAAGACGAAATACCGGATAAGCGCAAGCCGTC
 GGGCTGAACGGGGGGTTCGTGCACACAGCCAGCTTGGAGCGAACGACCTACACCGAAGTACGATACCTACAGCGTGAAGTATGAGAAAGCGCCACGCTCCCGAAGGGAGAA
 AGGGCGACAGGATATCCGTAAGCGCGCAGGGTCGGAACAGGAGAGCGCAGAGGGAGCTTCCAGGGGGAAACGCTGGTATCTTTATAGTCTCGGGTTTCGCCACCTCTGA
 CTTGAGCGTCGATTTTGTGATGCTCGTCAGGGGGGGGAGCCATGGAAGAAACGCGCAGCAACGCGGCCCTTTTACGGTTCTGCGCTTTTGTGCGCTTTTGTCTCACATGTT
 CTTTCCGTCAGTATCCCTGATCTGTTGATGTAACCGTATTACCGGCTTTGAGTGAAGTATACCGCTCCGCCGACGCGCAACGCGCAGCCAGCCAGCGAGTGAAGGAG
 CGGAAGAGCGCTGATGCGGATTTTCTCCTTACGCATCTGTGCGGATTTTACACCGCATATATGGTGCACCTCTCAGTACAATCTGCTCTGATGCCGATAGTTAAGCCAGT
 ATACACTCCGCTATCGTACGTGACTGGGTGATGGCTGCGCCCGACACCCGCCAACCCGCTGACCGCCCTGACGGCTTGTCTGCTCCCGGATCCGCTTACAGACAAG
 CTGTGACCGCTCCCGGAGTGCATGTGTGACAGGTTTTTACCGTATCAGCAACCGCGCAGGAGCTGCGGTAAGGCTCATCAGCGTGGTCTGTAAGCGATTACAGATG
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 CTCGCTGTAAGGGGATTTCTGTTTATGGGGTAAATGATACCGATGAAACGAGAGAGGATGCTCACGATACGGGTTACTGATGATGAACATGCCCGGTTACTGGAACGTTGTG
 AGGGTAAACAACCTGCGGATGATGATGCGCGGGGACAGAGAAAATCACTCAGGGTCAATGCCAGCGCTTCGTTAATACAGATGTAGGTGTTCCACAGGATAGCCAGCAGCAT
 CCTGCGATGCAGATCCGGAACATAATGGTGCAGGGCGCTGACTTCCCGGTTTCCAGACTTACGAAACCGGAAACCGAAGACCATTCTATGTTGTTGCTCAGGTCGAGACGCT
 TTTGCGAGCAGCTGCTTCAGCTTCGCTCGGTATCCGTTGATTCAGTGAAGCAACCGCCAGCCTAGCCGCGGCTCAACGACGAGCAGCAGCATGATGAGCGAGGATGCG
 GCACCCGTTGGGGCCGCTATGCCGCGATAATGGCTGCTTTCGCGCAACGTTTGGTGGCGGGACAGTGAACGAGGCTTGAAGGCTTCAAGGCAATCGGTCGAGATCCCGGTGCCTAATGAGTGAAGTAACTTACATTA
 TTGGTTGCGCTCACTGCCGCTTTCCAGTGGGAAACCTGTGTCAGCTGATTAATGAATCGGCAACGCGCGGGGAGAGGCGGTTTGGTATTTGGCGCCGAGGTTGGT
 TTTTCTTTTACCAGTGAAGCGGGCAACAGCTGATTGCCCTTACCGCTTGGCCCTGAGAGAGTTGACGCAAGCGGTCACCGCTGGTTTGGCCAGCAGCGGAAATCCTGTT
 TGATGGTGGTTAAGCGCGGATATAACATGAGCTGCTTCGATGATCGTATCCACTACCGAGATATCCGACCAACGCGCAGCCCGGACTCGGTAATGGCGCGCATTGGC
 CCCAGCGCATCTGATCGTTGGCAACAGCATCGCAGTGGGAACGATGCCCTATTGACATTTGATGGTTTGGTGAAGAAACCGGACATGGCACTCCAGTCCGCTTCCCGTTC
 CGCTACCGCTGAATTTGATGGCAGTGAATATTTATGCCAGCCAGCAGCAGACGCGCCGAGCAAGACTTAAATGGGCGGCTAACAGCGCGGATTTGCTGGTGACCCA
 ATGCGACAGCATGCTCCACCGCCAGTCCGCTACCGTCTTTCATGGGAGAAAATAACTGTTGATGGGTGCTGGTTCAGAGACATCAAGAAATAACCGCGGAAATAGTGCAG
 GCAGTTCCACAGCAATGGCATCCTGGTTCATCCAGCGGATAGTTAATGATCAGCCACTGAGCGGTTGCGCGAGAAGATGTCACCGCGCTTTACAGGCTTTCGAGCGCGCT
 TCGTTCTACCATCGACACCACCGCTGGCAGCCAGTTGATCGCGCGAGATTTAATCGCCCGCAAAATTTGCGCAGCGCGCTGCAGGGCCAGACTGGAGGTGGCAACGCCAA
 TCAGCAACGAGTTTTTGGCCCGAGTTGTTGTCACCGCGGTTGATGTAATTCAGCTCCGCCATCGCCGCTTTCCAGCTTTTTCGCAAGAAACGTTGGCTGGCC
 TGGTTACACCGCGGGGAAACGCTGATGAAGAGACACCGGCATACTCTGCGACATCGTATAACGTTACTGGTTTACATTTACCAACCCCTGAATTTGACTCTCTTCCGGGCGCTA
 TCATGCCATACCGCGAAAGGTTTTGCGCCATTGATGGTGTCCGGGATCTCGACGCTCTCCCTTATGCGACTCCTGCATTAGGAAGCAGCCAGTAGTAGGTTGAGGCGGTTG
 AGCACCGCGCGCGCAAGGAAATGGTGCATGCAAGGAGATGGCGCCCAACAGTCCCGCGCCAGGGGCTGCCACCATAACCCAGCGGAAACAGCCGCTCATGAGCCGAAGT
 GCGAGCCGATCTTCCCATCGGTATGTCGGGATATAGCGCGCAGCAACCGCAGCTGTCGGCCGCGGATGATGCCGCGCAGATGCGTCCGGCTGAGGATCGAGATCTCGA
 TCCCGCGAAATTAATACGACTCACTATAGGGGAAATGTGAGCGGATAACAATTTCCCTCTAGAAAATAAATTTGTTTAACTTTAAGAAAGGAGATATA**CCATGGGTAAGAA**
 ATTATTCACCTGACTGACGACAGTTTTGACACGGATGTAACAAAGCGGACGGGGCGATCCTCGTCGATTTCTGGGAGAGTGGTGGGTCGGTCCGTCAAAATGATCGCCCGAT
 TCTGGATGAAATCGTGCAGAAATATCAGGGCAAACTGACCGTTGCAAAACTGAACATCGATCAAAACCTGGCACTGCGCCGAAATATGGCATCCCGTGGTATCCCGACTCTGC
 TGCTGTTCAAAAACGGTGAAGTGGCGGCAACCAAGTGGGTGCACTGCTAAAGGTCAGTTGAAAGAGTTCCTCGACGCTAACTGGCCGTTCTGCCATGGGCAGCAGCCAT
 CATCATCATCATCAGCAGCGCCCTCAGCAAGGGCTGAGG/↗/CCTCAGCTTCCGCTGAGGTCCTCGACAAGCTTGGCGCCGACT**CTCGAGCACCAACCAACCAACCAAC**
 TGAGATCCGGCTGTAACAAAGCCGAAAGGAGCTGAGTTGGTGTCTGCC**CCGCTGAGCAATAAC**TAGCATAACCCCTTGGGGCTCTAAACGGGCTTGTAGGGGTTTTT
 CTGAAAGGAGAACTATATCCGGAT

pHTP8 sequence landmarks:

- **T7 promoter:** in gray
- **First ATG (methionine):** in yellow
- **Thioredoxin (Trx) gene:** in green
- **His-Tag coding sequences:** in purple
- **Cloning region:** ↗
- **T7 terminator:** in dark gray
- **Sequencing primers (T7 universal and T7 terminator):** underlined
- **BglII, NcoI & XhoI recognition sites:** in bold

Sequence added to the final recombinant protein (14.52 KDa):

MGSDKI IHLTDDSFDTDLVKADGAILVDFWAEWCGPCKMIAPILDEIADEYQKLTVAKLNIDQNPGTAPKYGIRGIPTLLLFKNGEVAATKVGALSK
 GQLKEFLDANLAGSAMGSSHHHHHHSSGPQQGLR

Revised 10/16