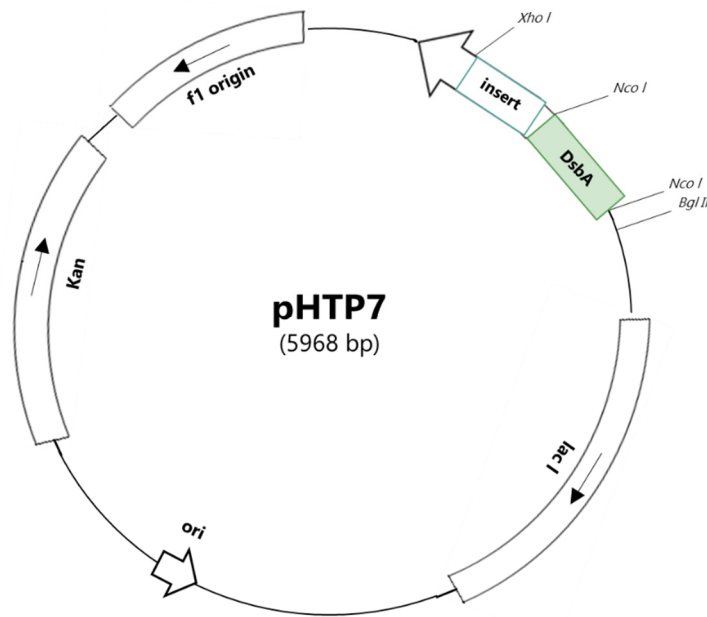


## pHTP7 Vector

pHTP7 was designed for the cloning and expression of high-levels of recombinant proteins in *Escherichia coli*. Recombinant proteins are expressed in fusion with the disulfide oxidoreductase (DsbA), which is able to promote solubility and folding of disulfide bond-containing partners. This vector, included in the portfolio of NZYTech pHTP expression vectors, is part of the NZYEasy Cloning & Expression System. pHTP7 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



#### pHTP7 Cloning/Expression Region

<i>Nco I</i>	<i>DsbA</i>	<i>Nco I</i>	His-Tag
<u>CCATGG</u> GAAAAAGATTGGCTGGCGCTGGCT . 621bp. TACTTAAGCGAGAAAAAGGATCAG <u>CCATGG</u> GCAGCAGCCATCATCATCATCACAGCAGCGGC			
MetGlyLysLysIleTrpLeuAlaLeuAla. 207aa. TyrLeuSerGluLysLysGlySerAlaMetGlySerSer			HisHisHisHisHisHisSerSerGly
CCTCAGCAAGGGCTGAGG / <del>⌘</del> / CCTCAGCTTCCGCTGAGGTCCGTCGACAAGCTTGC GGCCGCA		<i>Xho I</i>	His-Tag <i>STOP</i>
ProGlnGlnGlyLeuArg / <del>⌘</del> / ProGlnLeuProLeuArgSerValAspLysLeuAlaAlaLeuGlu			HisHisHisHisHisHis*

⌘ Represents the site where the gene will be inserted.

**Note:** For correct expression, inserted gene needs to be in frame with pHTP7 5' gene sequence. Inserts correctly cloned into pHTP7 will maintain reading frames starting on the ATG codon.

## 2. Vector Sequence (5968 bp)

TGGCGAATGGGACGCGCCCTGTAGCGGCGCATTAAAGCGCGGGGGTGTGGTGGTTACGCGCAGCGTGACCCTACACTTGCAGCGCCCTAGCGCCCGCTCCTTTTCGCTTTCTCCCT  
 TCCTTTCTCGCCAGCTTCGCGCGCTTTCCCGCTCAAGCTCTAAATCGGGGGCTCCCTTAGGGTTCCGATTTAGTGCTTTACGGCACCTCGACCCCAAAAACTTGATTAGGGTGATG  
 GTTACAGTAGTGGGCCATCGCCCTGATAGACGGTTTTTCGCCCTTTGACGTTGGAGTCCACGTTCTTTAATAGTGGACTCTTGTTCAAAATGGAACAACACTCAACCCATATCTCGGT  
 CTATCTTTTGATTATAAGGGATTTTCGCGATTTCGGCCATTGGTTAAAAATGAGCTGATTTAACAAAAATTTAACCGGAATTTAACAAAAATATAACGTTTACAAATTCAGGT  
 GGCACCTTTTCGGGAAATGTGCGCGGAACCCCTATTGTTTATTTTCTAAATACATTCAAAATATGATCCGCTCATGAATTAATTTAGAAAAACTCATCGAGCATCAAAATGAAAC  
 TGCATTTATTCATACAGATTAATCAATACCATATTTTGAAAAAGCCGTTTCTGTAATGAAGGAGAAACTCACCAGGCAGTCCATAGGATGGCAAGATCCCTGGTATCGGTCG  
 CGATTCCGACTCGTCCAACTAATCAACAACCTATTAATTTCCCGCTCGTCAAAAAAAGGTTATCAAGTGAGAAATCACCATGAGTGACGACTGAATCCGGTGAGAAATGGCAAAAGTTT  
 ATGCAATTTCTTCCAGACTGTTTCAACAGCCGACCCATTACGCTCGTATCAAAATCACTCGCATCAACCAACCGTTATTCATTCTGATTGCGCCTGAGCGAGACGAAATACCGGA  
 TCCTGTTAAAAAGGACAATTACAACAGGAATCGAATGCAACCCGGCGCAGGAACTGCCAGGCATCAACAATATTTTACCTGAATCAGGATATCTTCTAATACCTGGAATGCTG  
 TTTTCCGGGGATCGCAGTGGTGAATACCATGCATCATCAGGAGTACGGATAAAATGCTTGTGTTGCGGAAGAGGCATAAAATCCGTCAGCCAGTTTAGTCTGACCATCTCATCTGT  
 AACATCATTTGGCAACGCTACCTTTGCCATGTTTCAGAAAAAAGCTTGGCGCATCGGGCTTCCCATACAATCGATAGATTGTCGACCTGATTGCCCGACATTATCGCGAGCCCATTTA  
 TACCCATATAAATCAGCATCCATGTTGAATTTAATCGCGCCCTAGAGCAAGACGTTTCCCGTGAATATGGCTCATAACACCCCTTGTATTACTGTTTATGTAAGCAGACAGTTTAA  
 TTGTTTCATGACCAAAATCCCTTAACTGAGTTTTTCGTTTCACTGAGCGTCAAGCCCGTAGAAAAAGTCAAAGGATCTTCTTGTAGATCCTTTTTTCTGCGCGTAACTGCTGCTTGC  
 AAACAAAAAACCCCGCTACCAGCGTGGTTTGTGTCGGGATCAAGAGCTACCACTCTTTTCCGAAGTAACTGGCTTACGAGAGCGCAGATACCAATACTGCTCTTCTAGT  
 TGATCGCTGTTTAGCCGACACTCTGAGCAGCCGCTGAGCAGCCGCTGCTGCTCTGCTAATCCTTACCAGTGGCTGCTGCGCAGTGATAAGTGGCTGCTGCGCGTAACTGCTGCTG  
 GACTCAAGACGATAGTTACCGGATAAGCGCAGCGGTGCGGCTGAACGGGGGTTGCTGCACACAGCCGACTTGGAGCGAACGACCTACACCGAAGTGAATACCTACAGCGTGAGC  
 TATGAGAAAGCGCCACGCTTCCGGAAGGAGAAAGGCGGACAGGATCCGGAAGCGGCGAGGTCGGAACAGGAGAGCGCACGAGGGAGCTTCCAGGGGAAACCGCTGGTATCTTTA  
 TAGTCTGTGCGGTTTCGCCACCTTGTACTGAGCGTCGATTTTGTGATGCTCGTCAAGGGGGCGGAGCCTATGAAAAACCGCAGCAACCGGGCCTTTTTACGGTTCCTGGCCTTT  
 TGCTGCGCTTTTGGCTACATGTTTCTTCTGCTTATCCCGTATTCCTGCTGTTTACCGCTGATACCGCTGAGTACCGCTGAGTACCGCTGAGTACCGCTGAGTACCGCTGAGT  
 AGTCAAGTGAAGCGGAAGCGGACGCGCTGATGCGGTATTTTCTCTTACGCATCTGTGCGGTTTTCACACCGCATATATGGTGCCTCTCAGTACAATCTGCTGATGCGCGA  
 TAGTTAAGCCAGTATACCTCCGCTATCGCTACGTGACTGGGTGATGGCTGCGCCCGACACCCGCAACACCCGCTGACGCGCCTGACGGGCTTGTCTGCTCCCGCATCCGCTTA  
 CAGACAAGCTGTGACCGTCTCCGGGAGCTGCATGTGTCAGAGTTTCCACCGTCACTACCGAAACCGCGGAGGCGAGCTGCGGTAAGGCTCATCAGCGTGGTCTGAAGCGATTCACAG  
 ATGCTGCGCTGTTTCATCCGCGTCCGCTGTTGAGTTTCTCAGAAAGCTTAATGCTGCGCTCTGATAAAAGCGGGCAGTAAAGGGCGGTTTTTCTGTTTGGTCACTGATGCGCT  
 CCGTGAAGGGGATTTCTGTTTATGGGGTAAATGATACCGATGAAACGAGAGAGGATGCTCACGATACGGGTTACTGATGATGAACATGCCCGGTTACTGGAACGTTGTGAGGGTAA  
 ACAACTGGCGGTATGGATGCGGGGGGACAGAGAAAAATCACTCAGGGTCAATGCCAGCGCTCGTTAATACAGATGATAGTGTTCACAGGGTAGCCAGCAGCATCCGCGATGCGAG  
 ATCCGGAACATAATGTTGACAGGGCGCTGACTTCCCGCTTCCAGACTTACGAAACCGGAAACCGAAGACCATTATGTTGTTGCTCAGTTCGAGACGTTTTCGAGCAGCAGTCCG  
 TTCAGTTCGCTCGCGTATCGGTGATTCCTGCTAACCAAGTAAAGGCAACCCCGCAGCTAGCCGCTTCAACAGCAGGAGCAGCATGATCCGCAACCCGTTGGGGCCCGCATGCC  
 GCGGATAATGGCCTGCTTCTCGCCGAAACGTTTGGTGGCGGACAGTGCAGAAAGCTTGAAGCGGAGGCGTGAAGATTCCGGAATACCGCAAGCAGGCGGATCATCTGCGCGCTC  
 CAGCGAAAGCGGCTCCTCGCCGAAATGACCCAGAGCGCTGCGCGCACTGCTTACGAGTTGATGATAAAAGAGACAGTATAAGTGGCGGACGATGATGATGCCCGCGCCACC  
 GGAAGGAGTGCATGGTGAAGGCTCTCAAGGCGATCGGTCGAGATCCCGGTGCTTAATGAGTGAAGTAACTTACATTAATGCGTTCGCTCACTGCCCGCTTCCAGTCCGGAAA  
 CTTGCTGCGCAGCTGCATTAATGAAATCGGCAACCGCGGGGAGAGGCGGTTAATGCTGCGCTCTGATAAAAGCGGGCAGTAAAGGGCGGTTTTTCTTTTACCAGTGAAGCAGGCAACAGCTGATTGCCCTT  
 CACCGCTGGCCCTGAGAGAGTTGACAGAAAGCGTCCACGCTGGTTTGGCCAGCAGGCAAAATCCTGTTTGTGTTGGTTAACGGCGGGATATAACATGAGCTGTCTTCGTTATCG  
 TCGTATCCCACTACCGAGATATCCGCAACAGCGCAGCCCGACTCGGTAATGGCGCATGTCGCGCCAGCGCCATCTGATCTGGAACAGCAGTGCAGTGGGAACGATGCCCT  
 CATTGACATTTGCATGTTTGTGAAAAACCGGACATGGCACTCCAGTTCGCTTCCCGTTCGCTATCGGCTGAATTTGATTGCGAGTGAATATTTATGCCAGCCAGCCAGAGCGAG  
 ACGGCGGAGACAGAACTTAATGGCCCGCTAACAGCGCGATTTGCTGTTGACCCCAATGGCAACAGATGCTCAGCGCCAGTCCGCTTTCCTGGGAGAAAAATAACTGTTG  
 ATGGGTGCTGGTCAGAGACATCAAGAAATAACGCCGAACATTAGTGCAGGCAGTTCACAGCAATGGCATCCTGGTCAATCCAGCGGATAGTTAATGATCAGCCACTGACCGGTT  
 GCGGAGAAAGATTGTGACCGCGCTTTACAGGTTGACGCGCTTGGTTCTACCATCGACACCACCGCTGGCAGCCAGTTGATGGCGGAGATTTAATCGCCGCGACAATTTG  
 CGACGGCGGTCGAGGCGCAGACTGAGGTTGGCAACGCCAATCAGCAACGACTGTTTGGCCGCGAGTTGTTGTCGCGCGGTTGGGAATGTAATTCAGCTCCGCCATCGCGCTTCC  
 ACTTTTCCCGGTTTTTCGAGAAACGTTGGCTGGCTGGTTCCACCAGCGGGAAACGCTGCTGATAAGAGACACCGGCATACCTGCGACATCGTAAACGTTACTGTTTACATTTCA  
 CCACCTGAAATGACTCTCTTCCGGGCGCTATCATGCCATACCGCAAAAGGTTTTGCGCCATTCGATGTTGTCGCGGATCTCGACGCTTCCCTTATGCGACTCTGATTTAGGAAGC  
 AGCCAGTAGTAGTTGAGCGGTTGAGCACCAGCCGCGCAAGGAATGGTGCATGCAAGGAGATGGCGCCCAACAGTCCCGCGCCAGGGGCTGCCACCATCCACGCCGAAACA  
 AGCGCTCATGAGCCGAAGTGGCGAGCCGATCTTCCCATCGGTGATGTCGCGGATATAGCGCCAGCAACCCGACCTGTGGCGCGGTGATGCCGCCACGATGCTCCGCGGTAG  
 AGGATCCAGACTCGATCCCGGAAATTAATCAGACTCACTATAGGGGAATGTGAGCGGATAACAATCCCTCTAGAAATAATTTGTTTAACTTTAAGAGGAGATATA**CCATGG**  
 GTA AAAAGATTGGCTGGCGTGGCTGTTTAGTTTTAGCGTTTTAGCGCATTGCGCGCGCAGTATGAAGATTGGTAAACAGTACACTGGA AAAAACCGGTTAGCTGGCGCGCGCA  
 AGTGTGGAGTTTTTCTCTTCTCTGCGCCACTGCTATCAGTTTGAAGAAGTTCTGCATATTTCTGATAATGTGAAGAAAAACTGCCGAAGCGGTGAAGATGACTAAATACCAC  
 GTCAACTTCATGGTGGTGAACCTGGGCAAGATCTGACTCAGGCATGGGCTGTGGCGATGGCGCTGGCGTGAAGACAAAGTACTGTTCCGCTGTTGAAGCGGTACAGAAAAACC  
 AGACCATCTGTTCTGCTTCTGATATCCGCGATGATTTATCAACGCAGGTTAAAGGTGAAGAGTACGACGCGCGGTGAACAGCTTCGTTGGTGAATCTCTGTTGCTCAGCAGGA  
 AAAAGCTGACAGCTGACGTGCAATTTGCGTGGCGTTCCGCGGATGTTTTGTTAACGGTAAATATCAGCTGAATCCCGAGGGTATGGATACCAGCAATATGGATGTTTTTGTTCAGCAGTAT  
 GCTGATACAGTGAATACTTAAAGCGAGAAAAAGGATCAGCCATGGGCGAGCAGCCATCATCATCATCACAGCAGCGCCCTCAGCAAGGGCTGAGG/⌘/CCTCAGCTCCGCT  
 GAGGTCGCTGACAAGCTTCCGCGCGCAC**TCCAG**AGCACCCACCACCAC TGAGATCCGGCTGCTAACAAAGCCGAAAGGAAGCTGAGTTGGCTGCTGCCACCGCTGAGCAATA  
 CTAGCATAACCCCTTGGGGCCTTAAACGGGCTTGAAGGGTTTTTTGCTGAAAGGAGGAACATATCCGGAT

### pHTP7 sequence landmarks:

- **T7 promoter:** in gray
- **First ATG (methionine):** in yellow
- **DsbA 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 (25.43 kDa):

MGKKIWLALAGLVLAF SASAAQYEDGKQYTTLEKPVAGAPQVLEFFSFFCPHCYQFEEVLHISDNVKKKLPEGVKMTKYHVNFMGDLGKDLTQAWAVAMALG  
 VEDKVTVPLEFEGVQRTQTI RSASDIRDVF INAGIKGEEYDAAWN SFVVKLSVAQQEKAADVQLRQVPMFVNGKYQLNPQGMDSNMDFVQVQYADTVKYLS  
 EKKGSAMGSSHHHHHHSSGPPQQLR