USER GUIDE

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# pMT/V5-His A, B, and C

**Catalog number** V4120-20

**Revision date** 20 February 2012
**Publication Part number** 28-0177

MAN0000658

**For Research Use Only. Not intended for any animal or human therapeutic or diagnostic use.**

life technologies™

# Contents

# Kit Contents and Storage

| | |
|---|---|
| **Kit Contents** | 20 µg each of pMT/V5-His A, B, and C are supplied at 0.5 µg/µL in 10 mM Tris-HCl, 1 mM EDTA, pH 8.0 in a total volume of 40 µL.<br>20 µg of pMT/V5-His/*lac*Z is supplied at 0.5 µg/µL in 10 mM Tris-HCl, 1 mM EDTA, pH 8.0 in a total volume of 40 µL. |
| **Shipping/Storage** | Vectors are shipped at room temperature. Upon receipt, store at −30°C to −10°C. |
| **Product Use** | For research use only. Not intended for any human or animal therapeutic or diagnostic use. |

# Methods

## Product Overview

### Introduction

pMT/V5-His is a 3.5 kb expression vector designed for use with the *Drosophila* Inducible Expression System (DES®; Catalog nos. K4120-01, K5120-01). Upon transfection, the vector allows transient, inducible expression of your protein of interest in *Drosophila* cells. When cotransfected with the selection vectors, pCoHygro or pCoBlast, included with the appropriate DES® Inducible Kit, pMT/V5-His allows selection of stable cell lines exhibiting inducible expression of the protein of interest. The pMT/V5-His vector contains the following elements:

- The *Drosophila* metallothionein (MT) promoter for high-level, metal-inducible expression of the gene of interest in S2 cells (Angelichio *et al.*, 1991; Bunch *et al.*, 1988; Maroni *et al.*, 1986; Olsen, 1992)
- Multiple cloning site to facilitate cloning the gene of interest
- C-terminal peptide containing the V5 epitope and polyhistidine (6xHis) tag for detection and purification of your protein of interest (if desired)
- Three reading frames to facilitate in-frame cloning with the C-terminal peptide
- Ampicillin resistance gene for selection of transformants in *E. coli*

The control plasmid, pMT/V5-His/*lacZ*, is included for use as a positive control for transfection and expression.

For more information about the DES® Inducible Kits, pCoHygro, and pCoBlast, refer to the *Drosophila* Expression System manual. The *Drosophila* Expression System manual is supplied with each DES® Inducible Kit, but is also available from **www.lifetechnologies.com** or by contacting Technical Support (see page 15).

### Description of MT Promoter

The *Drosophila* MT promoter allows high-level, inducible expression of the gene of interest in *Drosophila* S2 (or D.Mel-2) cells. When used to express heterologous proteins, the promoter is extremely efficient and tightly regulated, even at high copy number (Johansen *et al.*, 1989). The MT promoter is well characterized (Angelichio *et al.*, 1991; Bunch *et al.*, 1988; Maroni *et al.*, 1986; Olsen, 1992), with regulatory elements and the start of transcription well defined.

The MT promoter is inducible by addition of copper sulfate or cadmium chloride to the culture medium (Bunch *et al.*, 1988). Copper sulfate is generally the preferred inducer due to its reduced toxicity as compared to cadmium. While cadmium is an effective inducer, it also induces a heat-shock response in S2 cells.

*Continued on next page*

## Experimental Outline

The table below describes the general steps needed to clone and express your gene of interest. For more details, refer to the manual and pages indicated.

| Step | Action | Source |
|---|---|---|
| 1 | Develop a cloning strategy to ligate your gene of interest into pMT/V5-His A, B, or C in frame with the C-terminal peptide encoding the V5 epitope and the polyhistidine tag (if desired). | Pages 3–7, this manual |
| 2 | Transform your ligation reactions into a *rec*A, *end*A *E. coli* strain (e.g. TOP10). Select on LB agar plates containing 50–100 µg/mL ampicillin. | Page 8, this manual |
| 3 | Analyze your transformants for the presence of insert. | Page 8, this manual |
| 4 | Select a transformant with the correct restriction pattern and sequence it to confirm that your gene is cloned in frame with the C-terminal peptide. | Page 8, this manual |
| 5 | Transfect your pMT/V5-His construct into S2 cells and induce expression of the gene of interest with copper sulfate. | Page 9, this manual and DES® manual |
| 6 | Assay for transient expression of your recombinant protein. | Page 9, this manual and DES® manual |
| 7 | To generate stable cell lines, cotransfect your pMT/V5-His construct and pCoHygro or pCoBlast into S2 cells and select for hygromycin or blasticidin resistant clones, as appropriate. | DES® manual |
| 8 | Scale up expression for purification. | DES® manual |
| 9 | Purify your recombinant protein by chromatography on metal-chelating resin (i.e. ProBond™). | DES® manual |

# Cloning into pMT/V5-His A, B, and C

## Introduction

Diagrams are provided on pages 5–7 to help you clone your gene of interest into pMT/V5-His. General considerations for cloning and transformation are discussed below.

## General Molecular Biology Techniques

For help with DNA ligations, *E. coli* transformations, restriction enzyme analysis, DNA sequencing, and DNA biochemistry, refer to *Molecular Cloning: A Laboratory Manual* (Sambrook *et al.*, 1989) or *Current Protocols in Molecular Biology* (Ausubel *et al.*, 1994).

## *E. coli* Strain

Many *E. coli* strains are suitable for the propagation and maintenance of the pMT/V5-His vectors including TOP10, DH5α™-T1$^R$, and JM109. We recommend that you propagate the vectors in *E. coli* strains that are recombination deficient (*rec*A) and endonuclease A deficient (*end*A).

For your convenience, the TOP10 and DH5α™-T1$^R$ strains are available as chemically competent cells (see page 13 for ordering information). TOP10 cells are also available as electrocompetent cells.

## Transformation Method

You may use any method of your choice for transformation. Chemical transformation is the most convenient method for many researchers. Electroporation is the most efficient and the method of choice for large plasmids.

## Maintenance of Plasmids

To propagate and maintain the pMT/V5-His and pMT/V5-His/*lacZ* vectors, we recommend that you use the supplied 0.5 µg/µL stock solution in TE buffer to transform a *rec*A, *end*A *E. coli* strain like TOP10, DH5α™-T1$^R$, JM109, or equivalent. Select transformants on LB agar plates containing 50 to 100 µg/mL ampicillin. Be sure to prepare a glycerol stock of each plasmid for long-term storage (see page 8).

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# Cloning into pMT/V5-His A, B, and C, Continued

## Cloning Considerations

Consider the following points when designing a strategy to clone your gene of interest into pMT/V5-His.

- Your insert should contain a Kozak translation initiation sequence with an ATG initiation codon for proper initiation of translation (Kozak, 1987; Kozak, 1991; Kozak, 1990). An example of a Kozak consensus sequence is provided below. Note that other sequences are possible, but the G or A at position –3 and the G at position +4 are the most critical for function (shown in bold). The ATG initiation codon is shown underlined.

  **(G/A)**NN<u>ATG</u>**G**

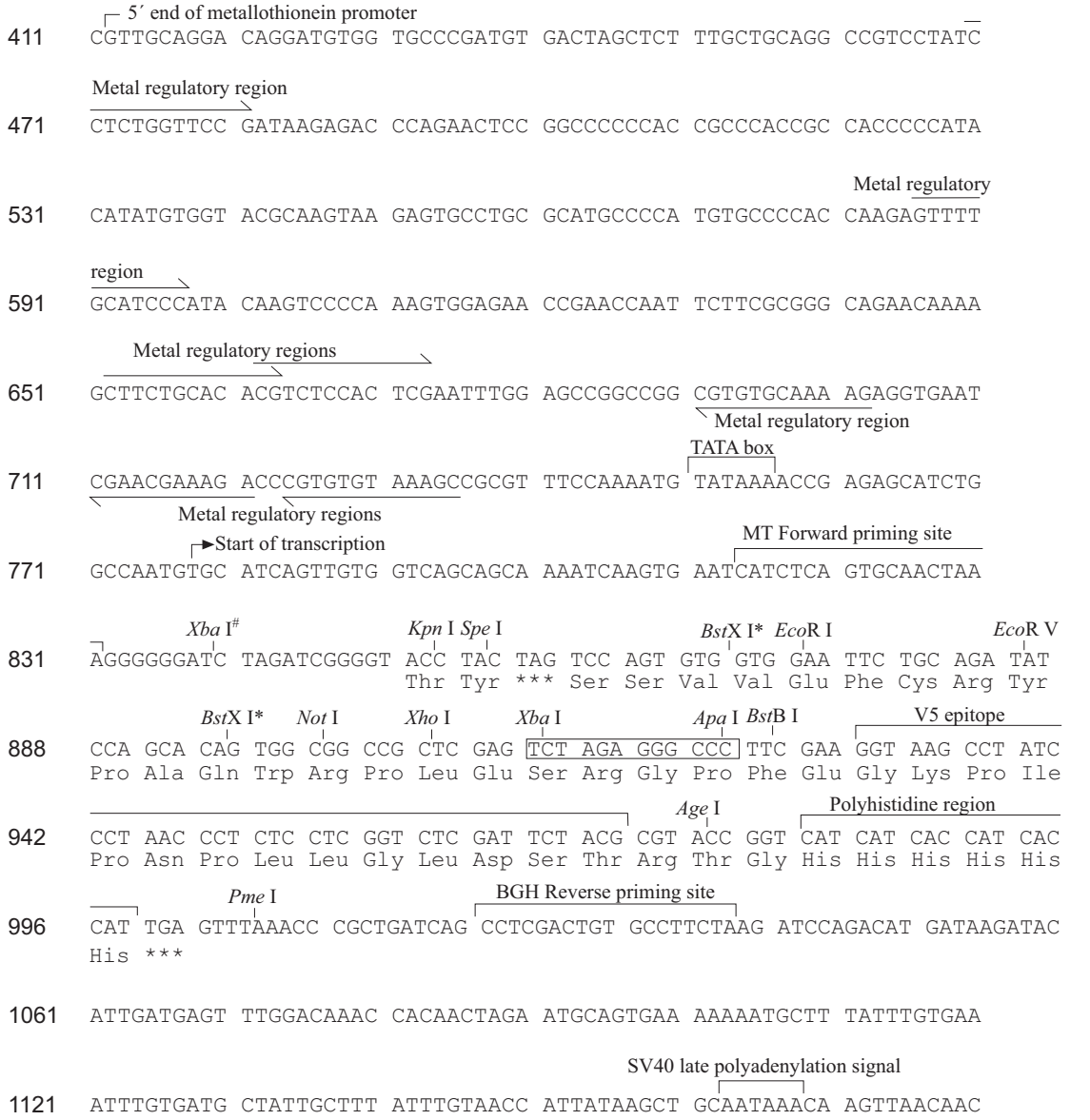
- It is possible to clone and express a secreted protein in pMT/V5-His if your protein includes a native signal sequence. If your protein does not have a secretion signal, you may wish to consider using the pMT/BiP/V5-His vector for secreted expression. For more information, refer to the DES® manual.
- If you wish to use the V5 epitope and the polyhistidine (6xHis) tag for detection and purification of your recombinant protein, you must clone your gene in frame with the C-terminal peptide. The vector is supplied in three reading frames to facilitate cloning. Refer to the diagrams on pages 5–7 to develop a cloning strategy. **Be sure that your gene does not contain a stop codon upstream of the C-terminal peptide.**
- If you do not wish to include the C-terminal peptide, include the native stop codon for your gene of interest.

*Continued on next page*

# Cloning into pMT/V5-His A, B, and C, Continued

## Multiple Cloning Site of pMT/V5-His A

Below is the multiple cloning site for pMT/V5-His A. The metal regulatory regions are marked as per Maroni, *et al.*, 1986. The start of transcription is at nucleotide 778. Restriction sites are labeled to indicate the actual cleavage site. The boxed nucleotides indicate the variable region. The multiple cloning site has been confirmed by sequencing and functional testing. The nucleotide sequence of pMT/V5-His A is available from **www.lifetechnologies.com** or from Technical Support (see page 15). For a map and a description of the features of pMT/V5-His A, refer to pages 10–11.




# This *Xba* I site is *dam* methylated in certain strains of *E. coli*.

* Note that there are two *Bst*X I sites in the polylinker.

*Continued on next page*

# Cloning into pMT/V5-His A, B, and C, Continued

## Multiple Cloning Site of pMT/V5-His B

Below is the multiple cloning site for pMT/V5-His B. The metal regulatory regions are marked as per Maroni, *et al.*, 1986. The start of transcription is at nucleotide 778. Restriction sites are labeled to indicate the actual cleavage site. The boxed nucleotides indicate the variable region. The multiple cloning site has been confirmed by sequencing and functional testing. The nucleotide sequence of pMT/V5-His B is available from **www.lifetechnologies.com** or from Technical Support (see page 15). For a map and a description of the features of pMT/V5-His B, refer to pages 10–11.

```
                5′ end of metallothionein promoter
411   CGTTGCAGGA CAGGATGTGG TGCCCGATGT GACTAGCTCT TTGCTGCAGG CCGTCCTATC

                Metal regulatory region
471   CTCTGGTTCC GATAAGAGAC CCAGAACTCC GGCCCCCCAC CGCCCACCGC CACCCCCATA

                                                          Metal regulatory
531   CATATGTGGT ACGCAAGTAA GAGTGCCTGC GCATGCCCCA TGTGCCCCAC CAAGAGTTTT

              region
591   GCATCCCATA CAAGTCCCCA AAGTGGAGAA CCGAACCAAT TCTTCGCGGG CAGAACAAAA

                Metal regulatory regions
651   GCTTCTGCAC ACGTCTCCAC TCGAATTTGG AGCCGGCCGG CGTGTGCAAA AGAGGTGAAT
                                                  Metal regulatory region
                                                  TATA box
711   CGAACGAAAG ACCCGTGTGT AAAGCCGCGT TTCCAAAATG TATAAAACCG AGAGCATCTG
            Metal regulatory regions
              Start of transcription                  MT Forward priming site
771   GCCAATGTGC ATCAGTTGTG GTCAGCAGCA AAATCAAGTG AATCATCTCA GTGCAACTAA

          Xba I#              Kpn I  Spe I                BstX I*  EcoR I              EcoR V
831   AGGGGGGATC TAGATCGGGG TA CCT ACT AGT CCA GTG TGG TGG AAT TCT GCA GAT ATC
                               Pro Thr Ser Pro Val Trp Trp Asn Ser Ala Asp Ile
          BstX I*  Not I      Xho I     Xba I              Apa I  Sac II  BstB I
889   CAG CAC AGT GGC GGC CGC TCG AGT CTA GAG GGC CCG CGG TTC GAA GGT AAG CCT
      Gln His Ser Gly Gly Arg Ser Ser Leu Glu Gly Pro Arg Phe Glu Gly Lys Pro
                V5 epitope                                  Age I   Polyhistidine region
943   ATC CCT AAC CCT CTC CTC GGT CTC GAT TCT ACG CGT ACC GGT CAT CAT CAC CAT
      Ile Pro Asn Pro Leu Leu Gly Leu Asp Ser Thr Arg Thr Gly His His His His
                Pme I                BGH Reverse priming site
997   CAC CAT TGA GTTTA AACCCGCTGA TCAGCCTCGA CTGTGCCTTC TAAGATCCAG ACATGATAAG
      His His ***

1061  ATACATTGAT GAGTTTGGAC AAACCACAAC TAGAATGCAG TGAAAAAAAT GCTTTATTTG

                                                  SV40 late polyadenylation signal
1121  TGAAATTTGT GATGCTATTG CTTTATTTGT AACCATTATA AGCTGCAATA AACAAGTTAA
```

# This *Xba* I site is *dam* methylated in certain strains of *E. coli*.

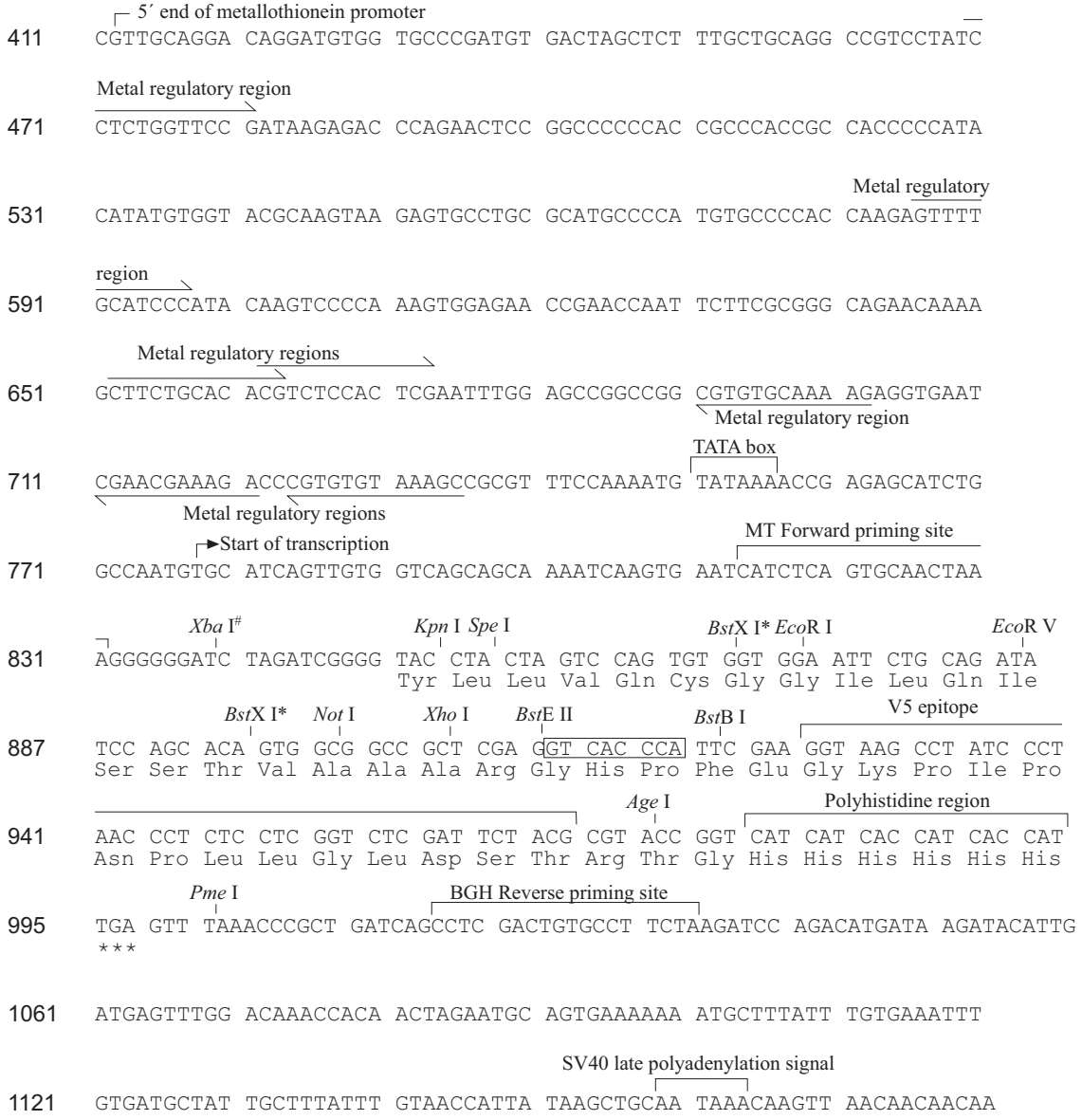
* Note that there are two *Bst*X I sites in the polylinker.

*Continued on next page*

# Cloning into pMT/V5-His A, B, and C, Continued

## Multiple Cloning Site of pMT/V5-His C

Below is the multiple cloning site for pMT/V5-His C. The metal regulatory regions are marked as per Maroni, *et al.*, 1986. The start of transcription is at nucleotide 778. Restriction sites are labeled to indicate the actual cleavage site. The boxed nucleotides indicate the variable region. The multiple cloning site has been confirmed by sequencing and functional testing. The nucleotide sequence of pMT/V5-His C is available from **www.lifetechnologies.com** or from Technical Support (see page 15). For a map and a description of the features of pMT/V5-His C, refer to pages 10–11.




# This *Xba* I site is *dam* methylated in certain strains of *E. coli*.

* Note that there are two *Bst*X I sites in the polylinker.

*Continued on next page*

### *E. coli* Transformation

Transform your ligation mixtures into a competent *rec*A, *end*A *E. coli* strain (e.g. TOP10, DH5α™-T1$^R$) and select on LB agar plates containing 50–100 µg/mL ampicillin. Select 10–20 clones and analyze for the presence and orientation of your insert.


We recommend that you sequence your construct with the MT Forward and BGH Reverse primers to confirm that your gene is in the correct orientation for expression and is cloned in frame with the C-terminal peptide. The MT Forward and BGH Reverse primers are included in each DES® Inducible Kit. Refer to the diagrams on pages 5–7 for the sequences and location of the priming sites.

**Note:** For your convenience, we offer a custom primer synthesis service. For more information, see **www.lifetechnologies.com** or call Technical Support (see page 15).

### Preparing a Glycerol Stock

Once you have identified the correct clone, purify the colony and make a glycerol stock for long-term storage. You should keep a DNA stock of your plasmid at –20°C.

1. Streak the original colony out on an LB plate containing 50 µg/mL ampicillin. Incubate the plate at 37°C overnight.
2. Isolate a single colony and inoculate into 1–2 mL of LB containing 50 µg/mL ampicillin.
3. Grow the culture to mid-log phase ($OD_{600}$ = 0.5–0.7).
4. Mix 0.85 mL of culture with 0.15 mL of sterile glycerol and transfer to a cryovial.
5. Store at –80°C.

# Transfection and Analysis

## Introduction

Once you have cloned your gene of interest into pMT/V5-His and have prepared purified plasmid DNA, you are ready to transfect your construct into S2 cells. If you are assaying for transient, inducible expression of your gene of interest, you may transfect your pMT/V5-His construct alone into S2 cells. If you wish to generate stable cell lines, you **must** cotransfect your pMT/V5-His construct with pCoHygro or pCoBlast into S2 cells. Note that the pMT/V5-His vector does not contain a resistance marker for selection in *Drosophila* cells. We recommend that you include the pMT/V5-His/*lacZ* positive control vector and a mock transfection (negative control) in your experiments to evaluate your results. Specific guidelines and protocols for transient transfection and generation of stable cell lines can be found in the DES® manual.

**Note:** The pCoHygro or pCoBlast selection vector is supplied with the appropriate DES® Inducible Kit. For more information about each vector, refer to the DES® manual.

## Plasmid Preparation

Plasmid DNA for transfection into eukaryotic cells must be pure and free from phenol and sodium chloride. Contaminants will kill the cells, decreasing transfection efficiency. We recommend isolating plasmid DNA using the PureLink® HiPure MiniPrep Kit (up to 30 µg DNA), the PureLink® HiPure MidiPrep Kit (up to 150 µg DNA) (see page 13 for ordering), or CsCl gradient centrifugation.

## Positive Control

pMT/V5-His/*lacZ* is provided as a positive control vector for *Drosophila* cell transfection and expression (see page 12 for a map) and may be used to optimize transfection conditions for S2 cells. Transfection of pMT/V5-His/*lacZ* results in induction of β-galactosidase expression upon addition of copper sulfate. A successful transfection will result in β-galactosidase expression that can be easily assayed by staining with X-gal.

## Assay for β-galactosidase Activity

You may assay for β-galactosidase expression by activity assay using cell-free lysates (Miller, 1972) or by staining the cells for activity. The β-Gal Assay Kit and the β-Gal Staining Kit are available for fast and easy detection of β-galactosidase expression (see page 13 for ordering information).

## Induction of Recombinant Protein Expression

Once you have transfected your pMT/V5-His construct into S2 cells, you will induce expression of recombinant protein using copper sulfate. In general, we recommend that you add copper sulfate directly to the culture medium to a final concentration of 500 µM and incubate the cells for 24 hours to obtain maximal induction of your protein of interest. Refer to the DES® manual for more details. Copper sulfate is provided in each DES® Inducible Kit.

## Detection and Purification of Recombinant Fusion Proteins

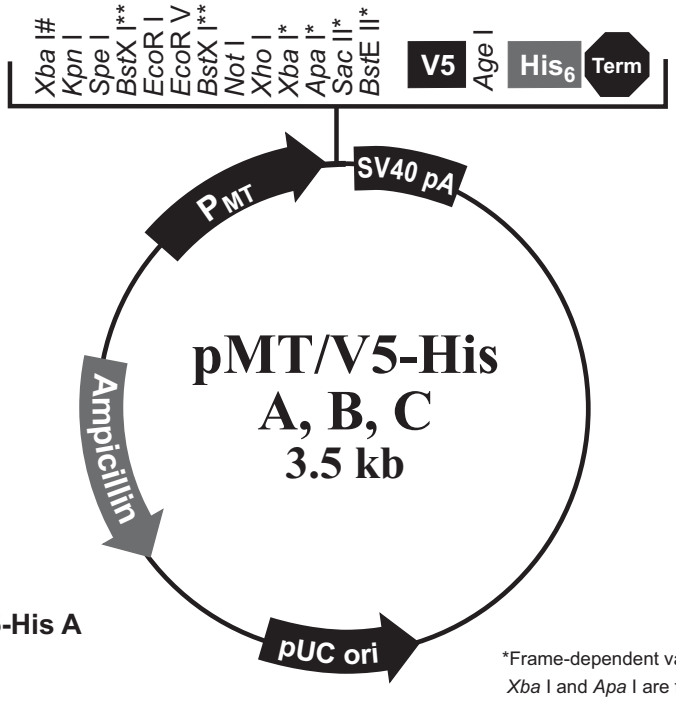
If you have cloned your gene of interest in frame with the C-terminal peptide containing the V5 epitope and the polyhistidine (6xHis) tag, you may use the Anti-V5 antibodies or Anti-His(C-term) antibodies to detect expression of your recombinant fusion protein by western blot analysis (see page 13 for ordering). The 6xHis tag also allows purification of recombinant protein using metal-chelating resins including ProBond™. Refer to the DES® manual for more detailed guidelines and instructions to detect and purify your recombinant fusion protein.

# Appendix

## pMT/V5-His Vector

### Map of pMT/V5-His

The figure below summarizes the features of the pMT/V5-His A, B, and C vectors. For a more detailed description of each feature, see page 11. The nucleotide sequences of pMT/V5-His A, B, and C are available from **www.lifetechnologies.com** or from Technical Support (see page 15).




*Frame-dependent variations
*Xba* I and *Apa* I are found only in versions A and B.
*Sac* II is found only in version B.
*Bst*E II is found only in version C.
#This *Xba* I site is *dam* methylated.
**There are two *Bst*X I sites in the polylinker.

**Comments for pMT/V5-His A**

3538 nucleotides

Metallothionein promoter: bases 412-778
Start of transcription: base 778
MT Forward priming site: bases 814-831
Multiple cloning site: bases 854-923
V5 epitope tag: bases 930-971
Polyhistidine region: bases 981-1001
BGH Reverse priming site: bases 1021-1038
SV40 late polyadenylation signal: bases 1163-1168
pUC origin: bases 1601-2334 (complementary strand)
*bla* promoter: bases 3340-3438 (complementary strand)
Ampicillin (*bla*) resistance gene ORF: bases 2479-3339 (complementary strand)

*Continued on next page*

### Features of pMT/V5-His

The features of pMT/V5-His A (3538 bp), pMT/V5-His B (3542 bp), and pMT/V5-His C (3534 bp) are described below. All features have been functionally tested. The multiple cloning site has been tested by restriction enzyme analysis.

| Feature | Benefit |
|---|---|
| *Drosophila* metallothionein (MT) promoter | Permits high-level, inducible expression of heterologous proteins (Bunch *et al.*, 1988; Maroni *et al.*, 1986) |
| MT Forward priming site | Allows sequencing in the sense orientation |
| Multiple cloning site | Allows insertion of your gene of interest |
| V5 epitope<br>(Gly-Lys-Pro-Ile-Pro-Asn-Pro-Leu-Leu-Gly-Leu-Asp-Ser-Thr) | Allows detection of your recombinant protein with the Anti-V5, Anti-V5-HRP, or Anti-V5-AP antibodies (Southern *et al.*, 1991) |
| Polyhistidine (6xHis) tag | Permits purification of your recombinant protein on metal-chelating resin such as ProBond™.<br>In addition, the C-terminal 6xHis tag is the epitope for the Anti-His(C-term) Antibody and the Anti-His(C-term)-HRP Antibody (Lindner *et al.*, 1997) (see page 13 for ordering) |
| BGH Reverse priming site | Permits sequencing of the non-coding strand |
| SV40 late polyadenylation signal | Allows efficient transcription termination and polyadenylation of mRNA (Angelichio *et al.*, 1991) |
| pUC origin | Allows high-copy number replication and growth in *E. coli* |
| *bla* promoter | Allows expression of the ampicillin (*bla*) resistance gene |
| Ampicillin (*bla*) resistance gene<br>(β-lactamase) | Permits selection of transformants in *E. coli* |

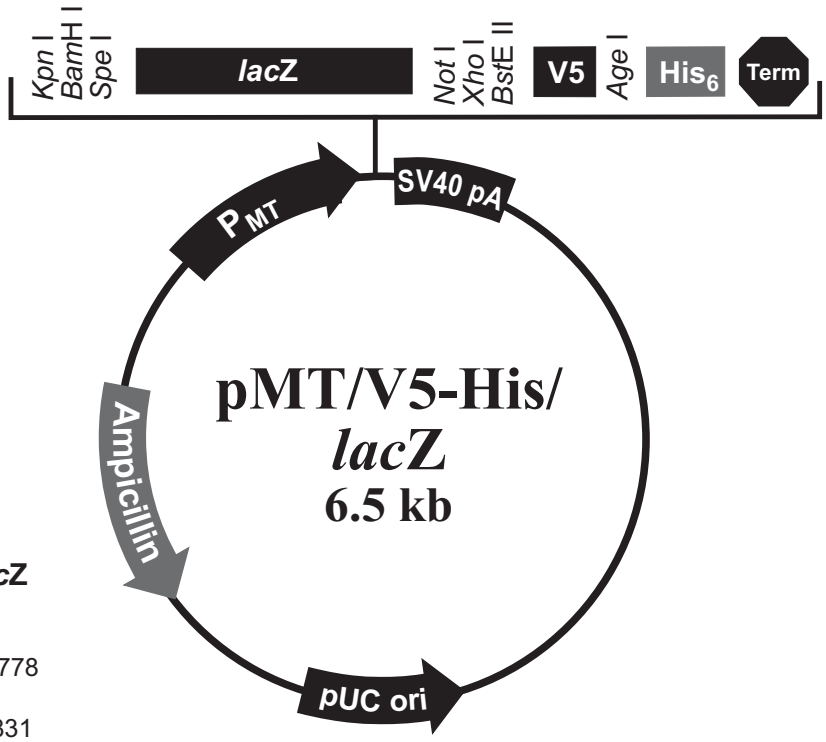
# pMT/V5-His/*lacZ* Vector

## Description

pMT/V5-His/*lacZ* is a 6596 bp control vector expressing β-galactosidase. The plasmid was constructed by digesting pMT/V5-His B with *Kpn* I and *Age* I and ligating a 3.2 kb *Kpn* I-*Age* I fragment containing the *lacZ* gene and the V5 epitope in frame with the polyhistidine tag.

## Map of pMT/V5-His/*lacZ*

The figure below summarizes the features of the pMT/V5-His/*lacZ* vector. The nucleotide sequence for pMT/V5-His/*lacZ* is available for downloading from **www.lifetechnologies.com** or by contacting Technical Support (see page 15).




### Comments for pMT/V5-His/*lacZ*

6596 nucleotides

Metallothionein promoter: bases 412-778
Start of transcription: base 778
MT Forward priming site: bases 814-831
LacZ portion of fusion: bases 904-3987
V5 epitope tag: bases 3988-4029
Polyhistidine region: bases 4039-4059
BGH Reverse priming site: bases 4079-4096
SV40 late polyadenylation signal: bases 4221-4226
pUC origin: bases 4719-5392 (complementary strand)
*bla* promoter: bases 6398-6496 (complementary strand)
Ampicillin (*bla*) resistance gene ORF: bases 5537-6397 (complementary strand)

# Accessory Products

## Additional Products

Many of the reagents supplied with the pMT/V5-His vector and the DES® Inducible Kits, and other reagents suitable for use with the kits are available separately. Ordering information for these reagents is provided below. For more information, refer to **www.lifetechnologies.com** or call Technical Support (see page 15).

| Item | Quantity | Catalog no. |
|---|---|---|
| *Drosophila* Inducible Expression System (DES) | 1 kit | K4120-01 |
| | 1 kit | K5120-01 |
| One Shot® TOP10 (chemically competent cells) | 20 reactions | C4040-03 |
| One Shot® TOP10 Electrocomp™ (electrocompetent cells) | 20 reactions | C4040-52 |
| One Shot® DH5α™-T1$^R$ Max Efficiency® Chemically Competent *E. coli* | 20 × 50 µL | 12297-016 |
| β-Gal Assay Kit | 80 mL | K1455-01 |
| β-Gal Staining Kit | 1 kit | K1465-01 |
| PureLink® HiPure MiniPrep Kit | 25 preps | K2100-02 |
| PureLink® HiPure MidiPrep Kit | 25 preps | K2100-04 |
| BGH Reverse Primer | 2 µg, lyophilized in TE | N575-02 |
| Hygromycin B | 20 mL | 10687-010 |
| Blasticidin S HCl | 50 mg | R210-01 |
| Schneider's Drosophila Medium | 500 mL | 21720-024 |
| Calcium Phosphate Transfection Kit | 75 reactions | K2780-01 |

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# Accessory Products, Continued

## Detection of Recombinant Proteins

Expression of your recombinant fusion protein can be detected using an antibody to the appropriate epitope. The table below describes the antibodies available for detection of C-terminal fusion proteins expressed using pMT/V5-His. Horseradish peroxidase (HRP)- and alkaline phosphatase (AP)-conjugated antibodies allow one-step detection using colorimetric or chemiluminescent detection methods.

The amount of antibody supplied is sufficient for 25 western blots.

| Product | Epitope | Catalog no. |
|---|---|---|
| Anti-V5 Antibody | Detects 14 amino acid epitope derived from the P and V proteins of the paramyxovirus, SV5 (Southern *et al.*, 1991) GKPIPNPLLGLDST | R960-25 |
| Anti-V5-HRP Antibody | | R961-25 |
| Anti-V5-AP Antibody | | R962-25 |
| Anti-His (C-term) Antibody | Detects the C-terminal polyhistidine (6xHis) tag (requires the free carboxyl group for detection (Lindner *et al.*, 1997) HHHHHH-COOH | R930-25 |
| Anti-His(C-term)-HRP Antibody | | R931-25 |
| Anti-His(C-term)-AP Antibody | | R932-25 |

## Purification of Recombinant Protein

The metal binding domain encoded by the polyhistidine tag allows simple, easy purification of your recombinant protein by Immobilized Metal Affinity Chromatography (IMAC) using the ProBond™ Resin. To purify proteins expressed from pMT/V5-His, the Xpress™ Purification System or the ProBond™ resin in bulk are available separately. See the table below for ordering.

| Product | Quantity | Catalog no. |
|---|---|---|
| ProBond™ Metal-Binding Resin (precharged resin provided as a 50% slurry in 20% ethanol) | 50 mL | R801-01 |
| | 150 mL | R801-15 |
| Xpress™ Purification System (includes six 2 mL precharged, prepacked ProBond™ resin columns and buffers for native and denaturing purification) | 6 purifications | K850-01 |
| Xpress™ Purification System with Anti-V5-HRP Antibody | 1 kit | K854-01 |
| Xpress™ Purification System with Anti-His(C-term)-HRP Antibody | 1 kit | K853-01 |
| Purification Columns (10 mL polypropylene columns) | 50 | R640-50 |

# Technical Support

## Obtaining support

For the latest services and support information for all locations, go to **www.lifetechnologies.com/support**.

At the website, you can:

- Access worldwide telephone and fax numbers to contact Technical Support and Sales facilities
- Search through frequently asked questions (FAQs)
- Submit a question directly to Technical Support (**techsupport@lifetech.com**)
- Search for user documents, SDSs, vector maps and sequences, application notes, formulations, handbooks, certificates of analysis, citations, and other product support documents
- Obtain information about customer training
- Download software updates and patches

## Safety Data Sheets (SDS)

Safety Data Sheets (SDSs) are available at **www.lifetechnologies.com/support**.

## Certificate of Analysis

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**Limited Use Label License: 6X His Tag**

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# Notes

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