



```

      830      840      850      860      870
C ACC ATG GCA TCA ATG CAG AAG CTG ATC TCA GAG GAG GAC CTG CTT
      Myc tag

      880      890      900      910      920
ATG GCC ATG GAG GCC CGA ATT CGG TCG ACC GAG ATC TCT CGA GGT ACC GCG GCC GC
      Sfi IA,B   EcoR I   Sal IB   Bgl II   Xho IA Kpn I   Not I
  
```

Restriction Map and Multiple Cloning Site (MCS) of pCMV-Myc. Unique restriction sites are in bold.

^A Sites are compatible with MATCHMAKER™ System 3 AD Vector.

^B Sites are compatible with MATCHMAKER™ System 3 BD Vector.

For older MATCHMAKER Systems, consult the Vector Information Packets provided with the vectors to determine compatibility.

Description:

The pCMV-Myc Mammalian Expression Vector expresses proteins containing the N-terminal c-Myc epitope tag. The c-Myc epitope tag is well-characterized and highly immunoreactive. High-level expression in mammalian cells is driven from the human cytomegalovirus immediate early promoter/enhancer ($P_{CMV IE}$). The vector contains an intron (splice donor/splice acceptor); the epitope tag; an MCS; and a polyadenylation signal from SV40. This vector also possesses the ampicillin resistance gene for selection in *E. coli*.

Use:

To create a fusion of a gene of interest and the Myc tag, insert the gene into the MCS in frame with the Myc coding sequence. The resulting Myc-tagged proteins can be identified with the c-Myc Monoclonal Antibody, provided with this vector, or another antibody raised against the Myc tag. The epitope tag is also useful for facilitating purification of the protein, identifying associated proteins, characterizing new proteins by immunoprecipitation, and determining subcellular localization.

The MCS in this vector is compatible with the MCSs in Clontech's MATCHMAKER™ Two-Hybrid System Vectors. Compatibility with System 3 Vectors is noted in the MCS diagram. Consult the Vector Information Packet provided with any MATCHMAKER vector for complete information.

After obtaining putative positive clones in your MATCHMAKER two-hybrid screen, use the pCMV-Myc and pCMV-HA Vectors to verify the interactions identified in yeast directly in mammalian cells. To accomplish this, subclone the selected inserts into the pCMV-Myc Vector and the "bait" insert into the pCMV-HA Vector. Alternatively, clone the "bait" insert into pCMV-Myc and the library inserts into pCMV-HA. To confirm predicted interactions *in vivo* via coimmunoprecipitation, cotransfect pCMV-Myc with the pCMV-HA Vector into mammalian cells and immunoprecipitate using the c-Myc Monoclonal or HA-Tag Polyclonal Antibody provided with the vectors.



Clontech

United States/Canada
800.662.2566

Asia Pacific
+1.650.919.7300

Europe
+33.(0)1.3904.6880

Japan
+81.(0)77.543.6116

Clontech Laboratories, Inc.
A Takara Bio Company
1290 Terra Bella Ave.
Mountain View, CA 94043
Technical Support (US)
E-mail: tech@clontech.com
www.clontech.com

(PR15767; published 23 May 2001)

Location of features:

- Immediate early cytomegalovirus promoter (P_{CMVIE}):
 - Enhancer region: 27–431
 - TATA Box: 520–526
 - Transcription start point: 549
- Intron (SV40 splice donor/splice acceptor):
 - SV40 late 19s mRNA intron: 672–702
 - Modified SV40 late 16s mRNA intron: 672–768
- Myc epitope tag with start codon (ATG): 829–867
- Multiple Cloning Site: 881–921
- SV40 polyadenylation signal:
 - Polyadenylation signal: 1053–1058
 - mRNA 3' end: 1072
- pUC plasmid replication region: 1545–2188
- Ampicillin resistance (β -lactamase) gene:
 - Promoter:
 - 35 region: 3266–3261
 - 10 region: 3243–3238
 - Transcription start point: 3231
 - Ribosome binding site: 3208–3204
 - β -lactamase coding sequences:
 - Start codon (ATG): 3196–3194
 - Stop codon (TAA): 2338–2336
 - β -lactamase signal peptide: 3196–3188
 - β -lactamase mature polypeptide: 3127–2339

Sequencing primer locations:

- pCMV Forward Sequencing Primer: 631–657
5'-GAT-CCG-GTA-CTA-GAG-GAA-CTG-AAA-AAC-3'

Propagation in *E. coli*:

- Suitable host strains: DH5 α , HB101, and other general purpose strains.
- Selectable marker: plasmid confers resistance to ampicillin (100 μ g/ml) to *E. coli* hosts.
- *E. coli* replication origin: pUC
- Copy number: ~500

Note: The attached sequence file has been compiled from information in the sequence databases, published literature, and other sources, together with partial sequences obtained by Clontech. This vector has not been completely sequenced.

Notice to Purchaser

Clontech products are to be used for research purposes only. They may not be used for any other purpose, including, but not limited to, use in drugs, *in vitro* diagnostic purposes, therapeutics, or in humans. Clontech products may not be transferred to third parties, resold, modified for resale, or used to manufacture commercial products or to provide a service to third parties without written approval of Clontech Laboratories, Inc.

Clontech, the Clontech logo and all other trademarks are the property of Clontech Laboratories, Inc.
Clontech is a Takara Bio Company. ©2006