# pCMV-LacZ Vector Information

PT2004-5 Cat. No. 631719

GenBank Accession No. U02451



Restriction Map of pCMV-LacZ. Unique restriction sites are in bold.

## Description

pCMV-LacZ is a mammalian reporter vector designed to expression  $\beta$ -galactosidase in mammalian cells from the human cytomegalovirus immediate early gene promoter (1). pCMV-LacZ contains an intron (splice donor/splice acceptor; 2) and polyadenylation signal from SV40, and the full-length *E. coli*  $\beta$ -galactosidase gene with eukaryotic translation initiation signals (3). pCMV-LacZ expresses high levels of  $\beta$ -galactosidase and can be used as a reference (control) plasmid when transfecting other reporter gene constructs and can be used to optimize transfection protocols by employing standard assays or stains to assay  $\beta$ -galactosidase activity. Alternatively, the  $\beta$ -galactosidase gene can be excised using the *Not* l sites at each end to allow other genes to be inserted into the pCMV-LacZ vector backbone for expression in mammalian cells or to insert the  $\beta$ -galactosidase fragment into another expression vector.

#### Location of features

- Immediate early cytomegalovirus promoter (P<sub>CMV IE</sub>) 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 (2): 672–768
- β-galactosidase gene with eukaryotic initiation signals (3) Eukaryotic translation initiation signal: 867–876
  β-galactosidase fusion protein: start codon (ATG): 873–875; stop codon: 4014–4016
  Amino acids from *D. melanogaster* alcohol dehydrogenase: 873–965
  Amino acids from *E. coli* β-galactosidase: 969–4013
  C→A (Phe→Leu) mutation removing *Eco*R I site: 3965
- SV40 polyadenylation signal Polyadenylation signal: 4426–4431 mRNA 3' end: 4445
- pUC origin of replication: 4918–5561

(032712)



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## Location of features (continued)

 Ampicillin resistance (β-lactamase) gene Promoter: -35 region: 6639–6634; -10 region: 6616–6611 Transcription start point: 6604 Ribosome binding site: 6581–6577 β-lactamase coding sequences: start codon (ATG): 6569–6567; stop codon: 5711–5709 β-lactamase signal peptide: 6569–6501 β-lactamase mature polypeptide: 6500–5712

## Propagation in E. coli

- Suitable host strains: DH5 $\alpha$ , HB101, and other general purpose strains.
- Selectable marker: plasmid confers resistance to ampicillin (100 µg/ml) to E. coli hosts.
- E. coli replication origin: pUC
- Copy number: ~500
- Plasmid incompatibility group: pMB1/Col E1

## References

- 1. MacGregor, G. R. & Caskey, C.T. (1989) Nucleic Acids Res. 17:2365.
- 2. Okayama, H. & Berg, P. (1983) Mol. Cell Biol. 3:280-289.
- 3. MacGregor, G. R., et al. (1987) Somat. Cell Mol. Genet. 13:253-265.

**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.

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