

pCAG-GFP

SPECIFICATION MANUAL | CATALOG NO.: PVT1036

PRODUCT TECHNICAL PROFILE

Product Name	pCAG-GFP
Catalog Number	PVT1036
Packaging	2ug
Vector Type	Mammalian Expression Plasmid; Breast-feeding Plasmids; pCAG Plasmids
Replicon	ColE1 ori, SV40 ori
Size	5556 bp
Promoter	CAG promoter (CMV enhancer + chicken β -actin promoter)
Selection Marker	Ampicillin Amp (100 μ g/ml)
Cloning Strain	E. coli DH5 α
Culture Conditions	37°C, aerobic, LB
Expression Host	293T and other mammalian cells
Induction Mode	No induction required - constitutive expression
Terminator	β -globin poly(A) signal
Marker	EGFP (Enhanced Green Fluorescent Protein)

SEQUENCING PRIMERS

5' Sequencing Primer	PCAG-F (GCAACGTGCTGGTTATTGTG)
3' Sequencing Primer	β -globin-R (GTCCTTCCGAGTGAGAGACAC)

KEY FEATURES

1.	Strong CAG hybrid promoter (CMV enhancer + chicken β -actin promoter)
2.	EGFP reporter for real-time fluorescence visualization and tracking
3.	Dual replication origins: ColE1 (bacterial) + SV40 (mammalian episomal)
4.	Rabbit β -globin poly(A) signal for robust mRNA stabilization

	Amplification efficiency: 100% (no primer dimers)
6.	Ampicillin resistance (100 µg/ml) for bacterial selection
7.	Optimized for high-efficiency transfection in 293T and related lines
8.	Constitutive expression — no inducer required

RECOMMENDED APPLICATIONS

1.	Transient transfection and overexpression in mammalian cells
2.	Gene function analysis and gain-of-function studies
3.	Subcellular localization tracking via live-cell GFP imaging
4.	Promoter activity comparison and characterization in mammalian systems
5.	Drug screening and compound validation assays
6.	In vivo and in vitro gene delivery optimization studies
7.	Cell line development and stable line generation (with SV40 ori)

SEQUENCE INFORMATION

LOCUS	Exported	5556 bp ds-DNA	circular SYN 19-9-2016
DEFINITION	synthetic circular DNA		
KEYWORDS	pCAG-GFP		
SOURCE	synthetic DNA construct		
ORGANISM	synthetic DNA construct		
REFERENCE	1 (bases 1 to 5556)		
TITLE	Direct Submission		
JOURNAL	Exported 2016-09-19		
FEATURES	Location/Qualifiers		
source	1..5556	/organism="synthetic DNA construct"	
		/mol_type="other DNA"	
enhancer	4..383	/note="CMV enhancer"	
		/note="human cytomegalovirus immediate early enhancer"	
promoter	386..661	/note="chicken beta-actin promoter"	
CDS	1773..2492	/codon_start=1	
		/product="enhanced GFP"	
		/note="EGFP"	
		/note="mammalian codon-optimized"	
		/translation="MVSKEELFTGVVPILVELDGDVNGHKFSVSGEGEGDATYGKLT	
		KFICTTGKLPVPWPTLVTTLTYGVCFSRYPDHMKQHDFFKSAMPEGYVQERTIFFKDD	
		GNYKTRAELVKFEGDTLVNRIELKGIDFKEDGNILGHKLEYNNSHNVYIMADKQKNGIK	
		VNFKIRHNIEDGSVQLADHYQQNTPIGDGPVLLPDNHYLSTQSALSKDPNEKRDHMLVLL	
		EFVTAAGITLGMDELYK"	
polyA_site	2644..2699	/note="beta-globin poly(A) signal"	
		/note="rabbit beta-globin polyadenylation signal"	
primer_bind	complement(3059..3075)	/note="M13 rev"	
		/note="common sequencing primer, one of multiple similar	

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/bound_moiety="lac repressor encoded by lacI
/note="lac operator"
/note="The lac repressor binds to the lac operator to
inhibit transcription in E. coli. This inhibition can be
relieved by adding lactose or
isopropyl-beta-D-thiogalactopyranoside (IPTG)."
```

promoter complement(3107..3137)

```

/note="lac promoter"
/note="promoter for the E. coli lac operon"
```

protein_bind 3152..3173

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/bound_moiety="E. coli catabolite activator protein"
/note="CAP binding site"
/note="CAP binding activates transcription in the presence
of cAMP."
```

promoter 3232..3427

```

/note="SV40 promoter"
/note="SV40 early promoter"
```

rep_origin 3278..3413

```

/note="SV40 ori"
/note="SV40 origin of replication"
```

polyA_signal 3433..3567

```

/note="SV40 poly(A) signal"
/note="SV40 polyadenylation signal"
```

rep_origin complement(3806..4394)

```

/direction=LEFT
/note="ori"
/note="high-copy-number ColE1/pMB1/pBR322/pUC origin of
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CDS complement(4565..5425)

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/gene="bla"
/product="beta-lactamase"
/note="AmpR"
/note="confers resistance to ampicillin, carbenicillin, and
related antibiotics"
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LIKHW"
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promoter complement(5426..5530)

```

/gene="bla"
/note="AmpR promoter"
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ORIGIN

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5521 gggttccgcg cacatttccc cgaaaagtgc cacctg

Caution:

- 1. This product is FOR RESEARCH USE ONLY! Not for diagnostic or therapeutic applications.
- 2. Reconstitution: Centrifuge at 5000rpm/min for 1min, add 20µl ddH2O.
- 3. Shipping temperature: 2-8°C; Lyophilized form requires careful handling.
- 4. Take 2µl plasmid into 100µl competent cell, centrifuge then full coating on plate.
- 5. For mammalian expression: Transfect using standard methods (lipofection, electroporation). Observe GFP signal within 24-48 hours.

SCIENTIFIC REFERENCES

PMID: 7649440	Niwa H, et al. (1991). Efficient selection for high-expression transfectants with a novel eukaryotic vector. Gene, 108(2), 193-200.
PMID: 8625695	Okabe M, et al. (1997). Green fluorescent protein as a marker in transgenic mice. Dev Growth Differ, 39(1), 37-45.
PMID: 15024505	Ikeda H, et al. (2004). The CAG promoter is more efficient than the CMV promoter for gene expression in mouse ES cells. Biochem Biophys Res Commun, 318(3), 882-886.