

## Product Information

### pCMV-MAT-Tag®-FLAG®-1 Expression Vector

Catalog Number **C5989**  
Storage Temperature  $-20\text{ }^{\circ}\text{C}$

## TECHNICAL BULLETIN

### Product Description

pCMV-MAT-Tag-FLAG-1 is a 4714 bp mammalian expression vector used for transient production of dual tagged fusion proteins in mammalian cells. The vector is a derivative of the pCMV5 transient expression vector<sup>1</sup> for intracellular expression of a properly inserted open reading frame as an N-terminal MAT-Tag (Metal Affinity Tag)-FLAG fusion protein. The promoter-regulatory region of the human cytomegalovirus<sup>2</sup> drives transcription of the fusion containing the transition metal binding, e.g.  $\text{Ni}^{+2}$  and  $\text{Co}^{+2}$ , MAT tag (HNHRHKH) and the FLAG<sup>®</sup> epitope, DYKDDDDK<sup>3</sup>. The pCMV-MAT-Tag-FLAG-1 expression vector is a shuttle vector containing both bacterial and SV40 origins of replication for propagation in both *E. coli* and mammalian cells. Efficiency of replication in mammalian cells is optimal when using host cells that express the SV40 large T antigen, e.g. COS-7. Removal of the MAT-Tag-FLAG tag is possible using enterokinase which cleaves following the Asp-Asp-Asp-Asp-Lys recognition site at the C-terminal end of FLAG.

pCMV-MAT-Tag-FLAG-1 may be used in conjunction with the Director™ Universal PCR System, Catalog Number RDC1, for a simple, rapid, and universal method to directionally clone PCR products. The MCS has been optimized for use with the *Hind* III/*Bgl* II pair of 5' overhang producing restriction enzymes often used in the Director system. The N-terminal MAT-Tag-FLAG fusion protein may be detected using Monoclonal ANTI-FLAG<sup>®</sup> M2, Catalog Number F3165, and purified using ANTI-FLAG M2 Affinity Gel, Catalog Number A2220. The fusion protein may also be purified utilizing the metal affinity properties of the MAT tag by using HIS-Select<sup>®</sup> Nickel Affinity Gel, Catalog Number P6611. Sigma-Aldrich offers a wide selection of related ANTI-FLAG and HIS-Select products; please visit [www.sigma-aldrich.com](http://www.sigma-aldrich.com) for a complete listing of antibody conjugates, resins, and affinity capture plates.

### Reagents Provided

- pCMV-MAT-Tag-FLAG-1 Expression Vector, 20  $\mu\text{g}$ , Catalog Number E1280, 0.5mg/ml in 10 mM Tris-HCl, pH 8.0, 1 mM EDTA.
- pCMV-FLAG-MAT-Tag-1-MAPK1 Control Vector, 20  $\mu\text{g}$ , Catalog Number C0489, 0.5 mg/ml in 10 mM Tris-HCl, pH 8.0, 1 mM EDTA.

### Precautions/Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

### Storage

Store at  $-20\text{ }^{\circ}\text{C}$ .

### Vector Features

The following table provides map positions of the features in the pCMV-MAT-Tag-FLAG-1 vector. Sequence verification of the MCS can be performed using the N-CMV-30 Sequencing Primer, Catalog Number P5350, and C-CMV-24 Sequencing Primer, Catalog Number P5475.

Feature	Map Position
CMV Promoter	166-916
N-CMV30 Sequencing Primer	825-854
Binding Site	
MAT-Tag	938-958
FLAG	962-985
MCS	983-1048
hGH polyA	1058-1677
C-CMV-24 Sequencing Primer	1115-1138
Binding Site	
SV40 ori	1696-2040
pBR322 ori	2943-3062
Amp	3209-4127
f1 ori	4262-4714

**References**

1. Andersson, S., et al., *J. Biol. Chem.*, **264**, 8222-8229 (1989).
2. Thomsen, D.R., et al., *Proc. Natl. Acad. Sci. USA*, **81**, 659-663 (1984).
3. Hopp, T.V., et al., *Bio/Technology*, **6**, 1204-1210 (1988).

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