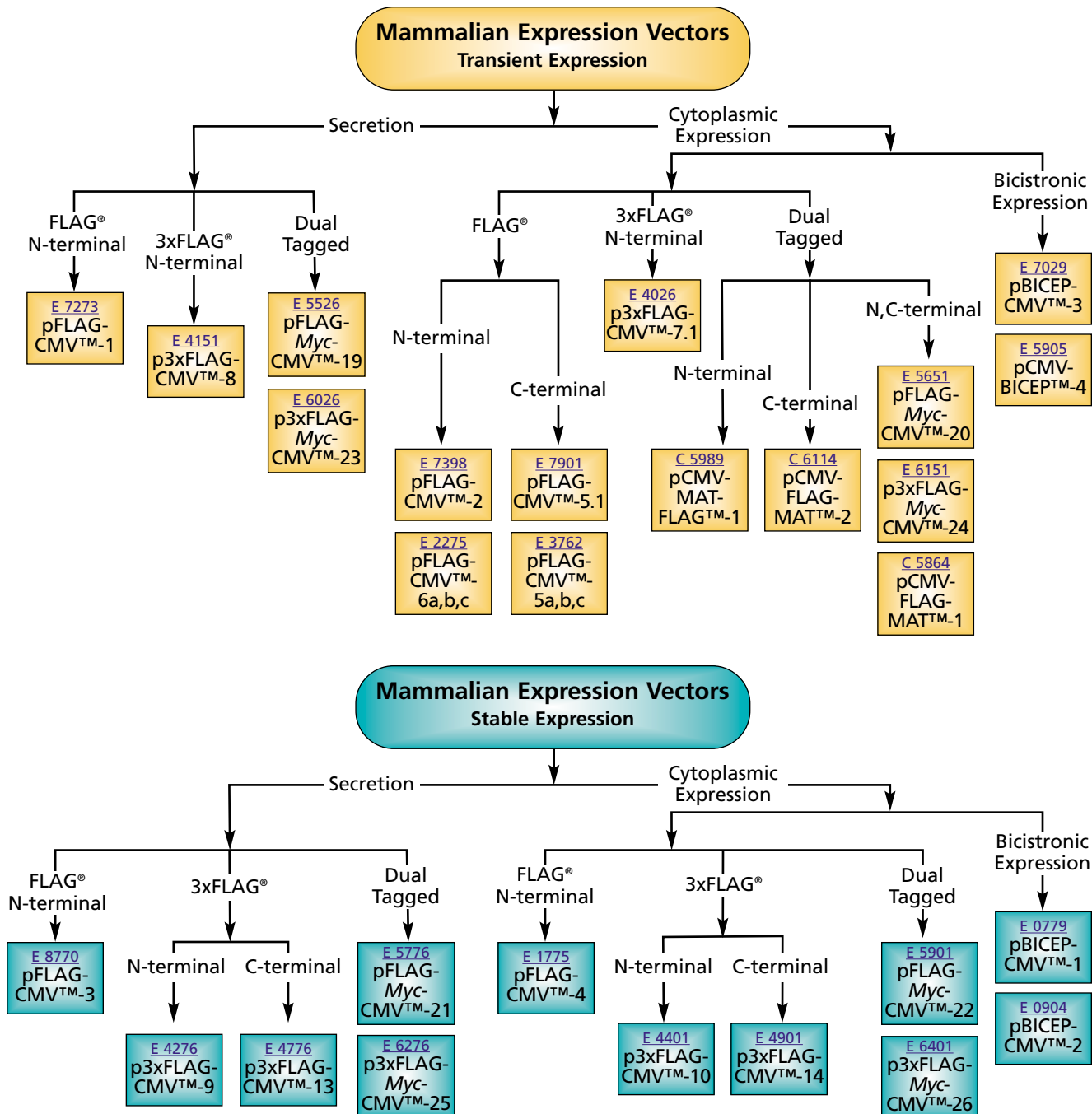


CLONING AND EXPRESSION

Mammalian Expression Vectors

Mammalian FLAG® expression vectors produce a variety of FLAG fusion proteins that allow easy detection, purification and analysis of recombinant protein for a wide range of applications. Sigma's offering of CMV promoter-based vectors provides flexibility in transient or stable expression, cytoplasmic expression or secretion, and N- or C-terminal tagging with FLAG or 3xFLAG®. A number of dual tag vectors contain the *c-myc* epitope tag or the MAT™ (Metal Affinity Tag) in addition to FLAG or 3xFLAG.

The strong human cytomegalovirus (CMV) promoter regulatory region drives constitutive protein expression levels as high as 1 mg/L in COS cells. For less potent cell lines, protein levels are typically ~0.1 mg/L. The presence of the SV40 replication origin will result in high levels of DNA replication in SV40 replication permissive COS cells. CMV vectors contain the pMB1 (derivative of pBR322) origin for replication in bacterial cells, the β-lactamase gene for ampicillin resistance selection in bacteria, hGH polyA, and the f1 origin. Vectors containing the preprotrypsin leader (PPT) sequence direct secretion of FLAG fusion proteins into the culture medium for purification using ANTI-FLAG® antibodies, resins, and plates.



Mammalian Expression Vector Selection Table

Transient Expression

Note: BICEP™ vectors can be found on p. 39-40.

Product	Product Name	PPT	FLAG	3x FLAG	c-myc	MAT	Ek Site	amp ^r
E 7273	pFLAG-CMV-1	√	N				√	√
E 7398	pFLAG-CMV-2		N				√	√
E 7901	pFLAG-CMV-5.1		C					√
E 3762	pFLAG-CMV-5a,b,c		C					√
E 2275	pFLAG-CMV-6a,b,c		N				√	√
E 4026	p3xFLAG-CMV-7.1			N			√	√
E 4151	p3xFLAG-CMV-8	√		N			√	√
E 5526	pFLAG-Myc-CMV-19	√	N		C		√	√
E 5651	pFLAG-Myc-CMV-20		N		C		√	√
E 6026	p3xFLAG-Myc-CMV-23	√		N	C		√	√
E 6151	p3xFLAG-Myc-CMV-24			N	C		√	√
C 5864	pCMV-FLAG-MAT-1		N			C	√	√
C 5989	pCMV-MAT-FLAG-1		N			N	√	√
C 6114	pCMV-FLAG-MAT-2		C			C		√
E 7029	pBICEP-CMV-3		N (MCS1)				√	√
E 5905	pCMV-BICEP-4		N (MCS1)		N (MCS2)		√	√

N = N-terminal tag C = C-terminal tag PPT = preprotrypsin leader for direct secretion

c-myc = c-myc epitope MAT = metal affinity tag amp^r = ampicillin resistance gene

Ek = Enterokinase cleavage site (cleavage of dual-tagged proteins may result in removal of one or more tags)



Mammalian Expression Vector Selection Table

Stable Expression

Note: BICEP vectors can be found on p. 39-40.

Product	Product Name	PPT	FLAG [®]	3x FLAG [®]	c-myc	Ek Site	amp ^r	neo ^r
E 8770	pFLAG-CMV-3	√	N			√	√	√
E 1775	pFLAG-CMV-4		N			√	√	√
E 4276	p3xFLAG-CMV-9	√		N		√	√	√
E 4401	p3xFLAG-CMV-10			N		√	√	√
E 4776	p3xFLAG-CMV-13	√		C			√	√
E 4901	p3xFLAG-CMV-14			C			√	√
E 5776	pFLAG-Myc-CMV-21	√	N		C	√	√	√
E 5901	pFLAG-Myc-CMV-22		N		C	√	√	√
E 6276	p3xFLAG-Myc-CMV-25	√		N	C	√	√	√
E 6401	p3xFLAG-Myc-CMV-26			N	C	√	√	√
E 0779	pBICEP-CMV-1		N (MCS1)			√	√	√
E 0904	pBICEP-CMV-2				N (MCS1)	√	√	√

N = N-terminal tag C = C-terminal tag neo^r = neomycin resistance for stable selection

PPT = preprotrypsin leader for direct secretion c-myc = c-myc epitope amp^r = ampicillin resistance gene

Ek = Enterokinase cleavage site (cleavage of dual-tagged proteins may result in removal of one or more tags)

