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Data Sheet

pASK-IBA3C

Cat. No.: 2-1322-000

Version: 11.0
Revision Date: 11.06.2021

Description	Expression plasmid. The expression cassette is under transcriptional control of the tetracycline promoter/operator. The expressed recombinant protein will be localized in the cytoplasm.
Affinity tag	Strep-tag®II fused to the C-terminus of the recombinant protein.
Bacterial Expression	Expression is induced upon addition of 200 µg anhydrotetracycline per 1 liter <i>E. coli</i> shaking culture ($A_{550} = 0.5$).
Expression strain	Any <i>E. coli</i> strain. The <i>tet</i> -promoter works independently from the genetic background of <i>E. coli</i> .
Resistance	Chloramphenicol Note: The Cam ^R resistance gene codes for homotetrameric chloramphenicol acetyltransferase (MW of the monomer = 26.6 kDa) which is predominantly expressed in the cytosol of <i>E. coli</i> transformed with this plasmid.
Form	5 µg, dissolved in 20 µl TE buffer, pH 8.0: 10 mM Tris/HCl, 1 mM EDTA
Concentration	250 ng/µl
Stability	12 months after shipping
Storage	recommended: 2-8 °C for frequent usage, -20 °C for long-term storage
Shipping	room temperature
Hazards	Product is not classified as hazardous according to (EC) No 1272/2008 [CLP]. A Material Safety Data Sheet is provided.

For research use only

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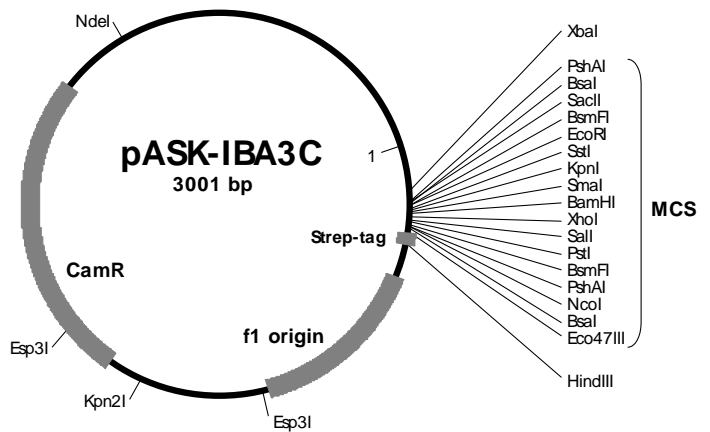
Multiple Cloning Site of pASK-IBA3C

1	CCATCGAATGGCCAGATGATTAATTCTAATTTTTGTTGACACTCTATCATTGATAGAGTTATTTTACCACTCCCTATCA	80
	forward primer	
	M G D R G P E	
81	GTGATAGAGAAAAGTGAATGAATAGTTTCGACAAAAATCTAGATAACGAGGGCAAAAatgGGAGACCGCGTCCCGAAT	160
	XbaI BsaI BsmFI	
	PshAI EcoRI	
	SacII	
	link Strep-tag [®] II	
	F E L G T R G S L E V D L Q G D H G L S A W S H P Q F	
161	TCGAGCTCGGTACCCGGGATCCCTCGAGGTCGACCTGCAGGGGACCATGGTCTCagcgtTTGGAGCCACCCGAGTTC	240
	SstI KpnI BamHI SalI PstI BsmFI BsaI Eco47III	
	SmaI XhoI PshAI	
	NcoI	
	E K *	
241	GAAAAATAATAAGCTTGACCTGTGAAGTGA AAAATGGCGCACATTGTGCGACATTTTTTTGTCTGCCGTTTACCGCTAC	320
	HindIII reverse primer	
321	TGCGTCACGGATCTCCACGCGCCCTGTAGCGGCATTAAAGCGCGCGGGTGTGGTTACGCGCAGCGTGACCCTAC	400

Please note: Restriction enzymes in bold cut twice. The *BsaI* sites (isoschizomer of *Eco31I*) at each end of the multiple cloning site are useful for precise and oriented insertion of the recombinant gene by one cleavage reaction only. The “link” contains a restriction site which can be used for subcloning.

Features of pASK-IBA3C

	from bp	to bp
promoter	37	72
forward primer binding site	57	76
multiple cloning site	139	222
Strep-tag [®] II	223	252
reverse primer binding site	308	324
f1 origin	337	775
CamR resistance gene	897	1556
Tet-repressor	1569	2192
ColE1 origin	2345	2933



Cloning primers for the precise cloning using <i>BsaI</i> or <i>Eco31I</i>	Sequencing primers:
Forward: 5'- NNNNNNGGTCTCNA ATG ^(N₁₇) * NNN NNN...	Forward: 5'- GAGTTATTTTACCACTCCCT -3'
Reverse: 5'- NNNNNNGGTCTCNGC ^(N₂₀) GCT NNN NNN...	Reverse: 5'- CGCAGTAGCGGTAAACG -3'

* The ATG start codon is already included