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Lieferung & Zahlungsart

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Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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Data Sheet

pASK-IBA7C

Cat. No.: 2-1326-000

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 N-terminus of the recombinant protein and can be removed by cleavage with Factor Xa.
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.

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Multiple Cloning Site of pASK-IBA7C

```

1  CCATCGAATGGCCAGATGATTAATTCCTAATTTTGTGACACTCTATCATTGATAGAGTTATTTTACCACTCCCTATCA 80
                                     forward primer

                                     link Strep-tag®II
                                     M A S W S H P

81  GTGATAGAGAAAAGTGAAATGAATAGTTCGACAAAAATCTAGATAACGAGGGCAAAAAATGGCTAGCTGGAGCCACCCGC 160
                                     XbaI                               NheI

                                     R P R S R I R A R Y P G I P R G R P
                                     E T A V P N S S S V P G D P S R S T C
      factor Xa
      Q F E K I E G R R D R G P E F E L G T R G S L E V D L
161 AGTTCGAAAAATCGAAGGgcgCGAGACCGCGGTCCCGAATTCGAGCTCGGTACCCGGGGATCCCTCGAGGTCGACCTG 240
                                     BbeI BsaI BsmFI SstI KpnI BamHI SalI PstI
                                     EheI PshAI EcoRI SmaI XhoI
                                     KasI SacII
                                     NarI

      A G G P W S L I S N *
      R G T M V S D I *
      Q G D H G L *

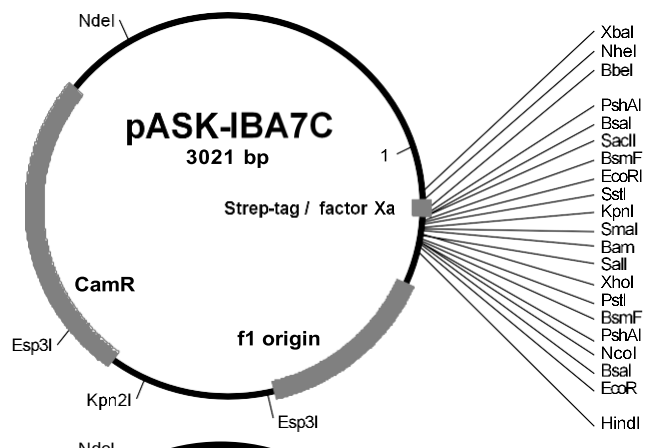
241 CAGGGGGACCATGGTCTCTgataTCTAACTAAGCTTGACCTGTGAAGTGAAAAATGGCGCACATTGTGCGACATTTTTTT 320
      BsmFI BsaI EcoRV HindIII
      PshAI
      NcoI

321 TGTCTGCCGTTTACCGTACTGCGTCACGGATCTCCACGCGCCTGTAGCGGCGCATTAAAGCGCGGCGGGTGTGGTGGTT 400
      reverse primer
  
```

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-IBA7C

	from bp	to bp
promoter	37	72
forward primer binding site	57	76
Strep-tag®II	139	171
Factor Xa cleavage site	172	183
multiple cloning site	184	260
reverse primer binding site	328	344
f1 origin	357	795
CamR resistance gene	917	1576
Tet-repressor	1589	2212
ColE1 origin	2365	2953



Cloning primers for the precise cloning using <i>BsaI</i> or <i>Eco31I</i>	Sequencing primers:
Forward: 5'- NNNNNNGGTCTCNGC GCC (N ₂₀) NNN NNN...	Forward: 5'- GAGTTATTTTACCACTCCCT -3'
Reverse: 5'- NNNNNNGGTCTCNTA TCA (N ₂₀) NNN NNN...	Reverse: 5'- CGCAGTAGCGGTAAACG -3'