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Data Sheet

pASG-IBA168

Cat. No.: 5-4168-001
Lot No.: 4168-

Version: 2.3
Revision Date: 03.03.2020

Description	StarGate Acceptor Vector for bacterial expression. • The expression cassette is under transcriptional control of the tetracycline promoter/operator. • Compatible with any *E. coli* strain. The *tet*-promoter works independently from the genetic background of *E. coli*. • The expressed recombinant protein will be localized in the cytoplasm.
Cloning Strategy	Cloning into StarGate Acceptor Vectors has to be done with the restriction enzyme Esp3I. There is no Multiple Cloning Site (MCS) available that can be used for the integration of the gene of interest instead (see manual).
Bacterial Expression	Expression is induced upon addition of 200 µg anhydrotetracycline (order no.: 2-0401-001; 2-0401-002) per 1 liter *E. coli* shaking culture (A_{550} = 0.5).
Affinity tag	The recombinant protein will contain two affinity tags fused to the C-terminus: 1. Distal: FLAG-tag for the purification of recombinant protein via anti-FLAG M2 agarose column and a FLAG octapeptide for elution. These FLAG-products are not delivered by IBA but can be purchased from Sigma. 2. Proximal: Strep-Tactin affinity tag (Twin-Strep-tag®) for purification of recombinant protein via Strep-Tactin® resin. This combination of tags can be used to perform Two-TAP analysis (Two-tag Tandem Affinity Purification) as published by Gloeckner et al. (2007) Proteomics 7, 4228-4234
Resistance	Ampicillin
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.

Note: The sequences have been compiled from information in the sequence database, published literature, and other sources, together with partial sequences obtained by IBA, however, the vectors have not been completely sequenced.

For research use only

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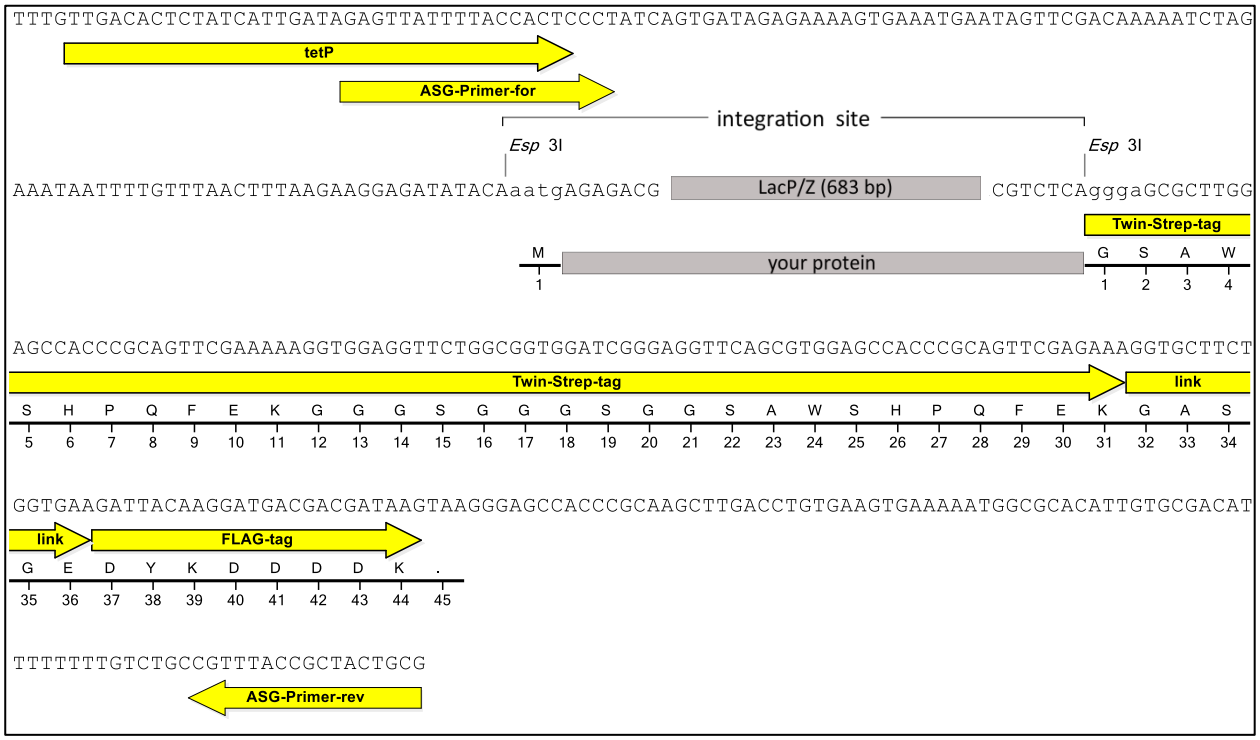
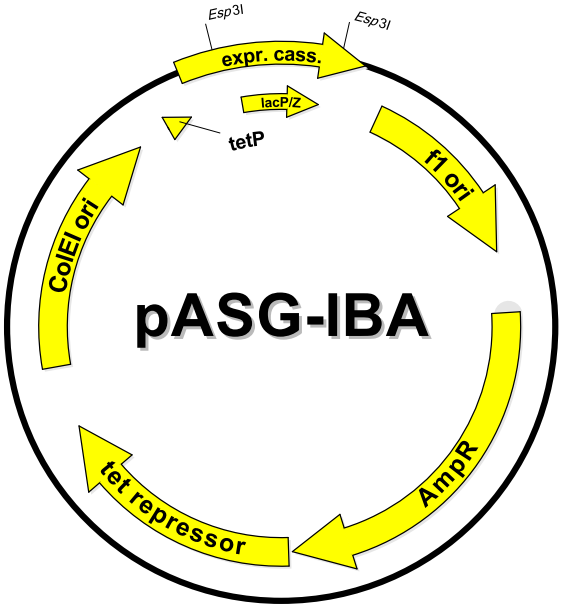
Expression cassette of pASG-IBA168



LacP/Z cassette = contains LacZ alpha fragment under control of a separate promoter, which allows alpha complementation of *LacZ* mutations such as *LacZΔM15* as in *E. coli* DH5α or TOP10.

your protein = after StarGate cloning using *Esp*3I your gene of interest will be located here



Features	from bp	to bp	Sequencing primer
f1 origin	13	451	**ASG-Primer-for (Cat. No. 5-0000-101)**
AmpR resistance gene	600	1460	
Tet-repressor	1470	2093	5’- GAGTTATTTTACCACTCCCT -3’
ColEIori	2246	2834	
Tet promoter	2939	2975	
forward primer binding site	2959	2978	**ASG-Primer-rev (Cat. No. 5-0000-102)**
LacZ alpha fragment	3290	3691	5’- CGCAGTAGCGGTAAACG -3’
Twin-Strep-tag	3755	3847	
FLAG-tag	3863	3886	
reverse primer binding site	3960	3976	
total vector length		3976	