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Data Sheet

pYSG-IBA62

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Description	StarGate Acceptor Vector for high-level expression of target proteins in yeast. It carries the the copper-inducible promoter (CUP1) for controlled high-level expression, the URA3 auxotrophy marker for selection after transformation (do not use URA3 for selection during expression), the LEU2d auxotrophy marker for selection to increase plasmid copy number for expression (do not use LEU2d for selection after transformation), and the 2 micron ori for episomal replication in yeast.			
Affinity tag	Strep-tag [®] II is fused to the C-terminus and FLAG-tag is fused to the N-terminus of the recombinant protein.			
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).			
Yeast Expression	Cultivate transformed yeast cells under LEU2d selection until OD ₆₀₀ reaches 0.8-1.2. Induce protein expression by addition of copper sulphate to a final concentration of 0.5 mM.			
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.			

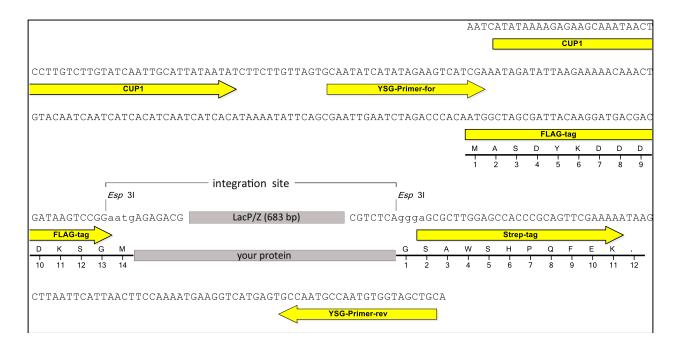
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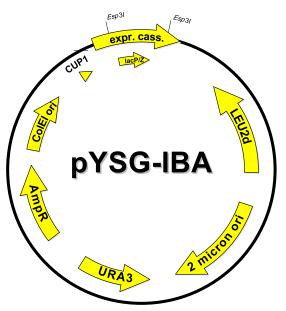
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 $\label{eq:LacP/Z cassette} \begin{array}{l} \mbox{contains LacZ alpha fragment under control} \\ \mbox{of a separate promoter, which allows alpha} \\ \mbox{complementation of } LacZ \, \mbox{mutations such as} \\ \mbox{LacZ} \Delta M15 \, \mbox{as in } E. \, \mbox{coli} \, DH5\alpha \, \mbox{or TOP10.} \\ \mbox{after StarGate cloning using } Esp3 \, \mbox{your} \\ \mbox{gene of interest will be located here} \end{array}$

Features	from bp	to bp	Sequencing primer
LEU2d	1668	574	YSG-Primer-for
2 micron ori	2032	3194	
URA3	4293	3490	5'- CAATATCATATAGAAGTCATCGA -3'
Ampicillin resistance gene	4725	5585	
ColEl ori	5756	6345	YSG-Primer-rev
CUP1 promoter	6873	6925	
forward primer binding site	6939	6961	5'- GCAGCTACCACATTGGCATTGGC -3'
FLAG-tag	7049	7087	
LacZ alpha fragment	7316	7717	
Strep-tag [®] II	7781	7813	
reverse primer binding site	7854	7876	
total vector length		7877	