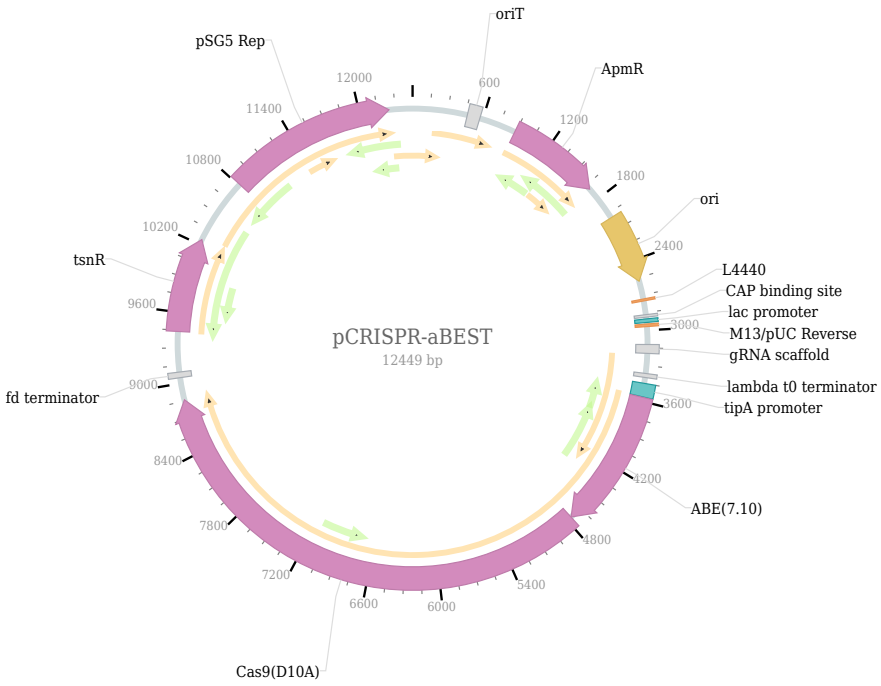
pCRISPR-aBEST

Vector ID	V000644
Antibiotic Resistance	Apramycin
Length	12449 bp
Vector Type	CRISPR, Synthetic Biology

Product Information

Replication Origin	ori
Copy Number	High Copy
Promoter	tipA
Cloning Method	Gibson Cloning
5' Primer	TCAGAGAAGGGAGCGGACAtatgagcgaggtcgagttctc
3' Primer	cgagccgccgctgctgccgc
Growth Strain(s)	DH10B
Growth Temperature	37℃
Comment	pCRISPR-aBEST is a pSG5-based shuttle plasmid for DSB-free adenine base editing in Streptomyces, fusing codon-optimized ecTadA deaminase with Cas9n(D10A) driven by PtipA promoter. Guided by sgRNA, it converts A:T to G:C within a 6-nucleotide editing window without DNA double-strand breaks. It shows lower editing efficiency than pCRISPR-cBEST but bears nearly negligible genome-wide off-target mutations. Equipped with Csy4 machinery, this all-in-one vector also supports multiplex parallel gene editing in single Streptomyces strains.

Vector Map



Plasmid Resuspension Protocol

1. Centrifuge at 5,000×g for 5 min.
2. Carefully open the tube and add sterile water to dissolve the DNA: add 20 µl for 5 µg plasmid, and 100 µl for 100 µg plasmid.
3. Close the tube and incubate for 10 minutes at room temperature.
4. Briefly vortex the tube and then do a quick spin to concentrate the liquid at the bottom. Speed is less than 5000×g.
5. Store the plasmid at -20 ℃.