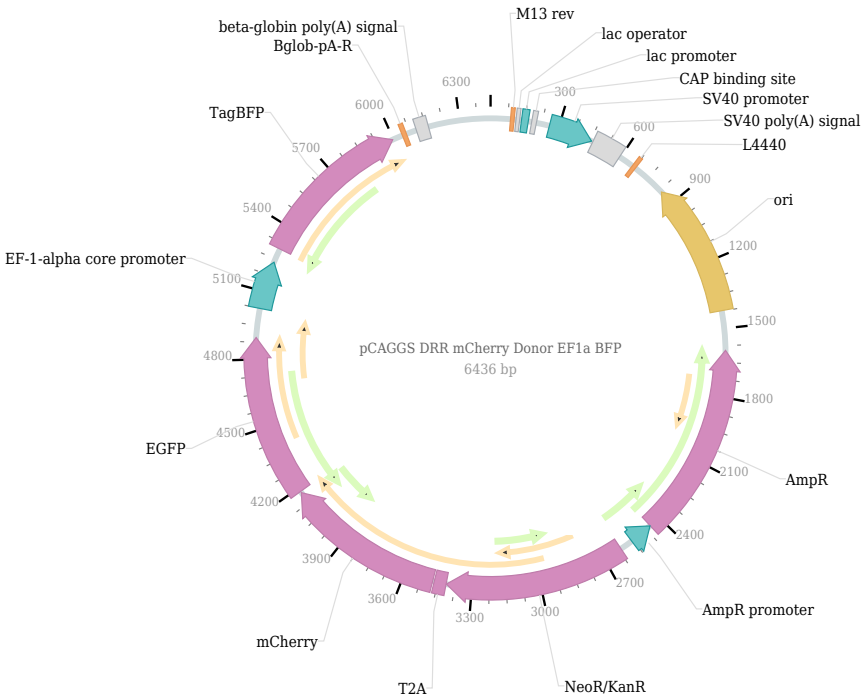
pCAGGS DRR mCherry Donor EF1a BFP

Vector ID	V012510
Antibiotic Resistance	Ampicillin
Length	6436 bp
Vector Type	Mammalian Expression, Synthetic Biology

Product Information

Replication Origin	ori
Selection Marker	BFP
Copy Number	High Copy
Growth Strain(s)	DH10B
Comment	This pCAGGS-based donor plasmid serves as homologous recombination template for the DRR DNA double-strand break reporter assay. Its core cassette consists of two homology arms flanking a T2A-mCherry expression fragment: the 5′ arm matches the 3′ end of NeoR-T2A from integrated pDRR, and the 3′ arm aligns with the 5′ region of EGFP in pDRR. Upon I-SceI-induced cleavage of genomic pDRR, homologous recombination swaps the stop/I-SceI segment in pDRR with mCherry from this plasmid, yielding mCherry-positive cells. An independent EF1α-BFP expression unit on the plasmid backbone acts as transfection control without participating in homologous pairing. Unlike integrated lentiviral pDRR, this donor is delivered via transient transfection and exists as episomal DNA in nuclei. Flow cytometry gates BFP-positive cells to quantify HR (mCherry+) versus cNHEJ (GFP+) repair efficiencies simultaneously.

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 ℃.