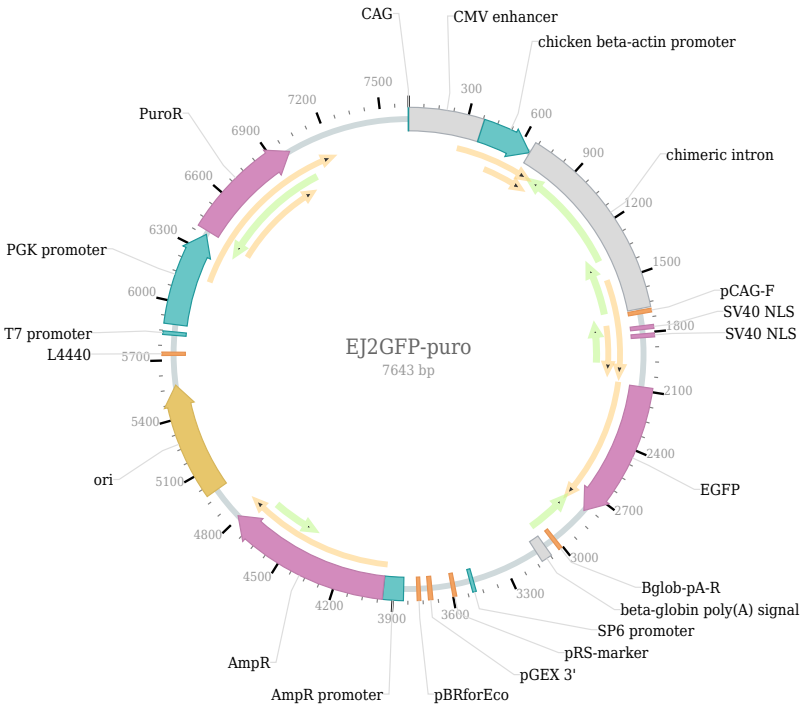
EJ2GFP-puro

载体编号 Vector ID	V001709
抗性 Antibiotic Resistance	Ampicillin
长度 Length	7643 bp
载体类型 Vector Type	Mammalian Expression

产品信息

复制子 Replication Origin	ori
筛选标记 Selection Marker	Puromycin
启动子 Promoter	CAG
5' 引物 5' Primer	ttcctacagctcctgggcaacg
3' 引物 3' Primer	ttttggcagagggaaaaaga
培养菌株 Growth Strain(s)	Stbl3
培养温度 Growth Temperature	37℃
备注 Comment	EJ2-GFP is a chromosome-integrated, GFP-based reporter specifically designed to monitor alternative non-homologous end joining (alt-NHEJ) of chromosomal double-strand breaks (DSBs) in mammalian cells. Its structure includes an N-terminal tag fused to GFP, with the coding sequence disrupted by an I-SceI site (for inducing DSBs) and stop codons in all three reading frames—flanked by 8-nucleotide microhomology regions. When DSBs are repaired via alt-NHEJ, the microhomology mediates annealing, removing the stop codons and causing a 35-nt deletion to restore functional GFP expression (with a unique XCM1 restriction site in the main product). This reporter primarily detects alt-NHEJ events (≈85% of GFP+ products are the 35-nt deletion type), with minor non-specific repair products (smaller/larger deletions) accounting for only ~15%. It is widely used to study alt-NHEJ regulation (e.g., Ku inhibits, CtIP promotes this pathway) and its role in cancer, such as BRCA2-mutant tumor resistance to PARP inhibitors.

质粒图谱



复溶方法

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 ℃.