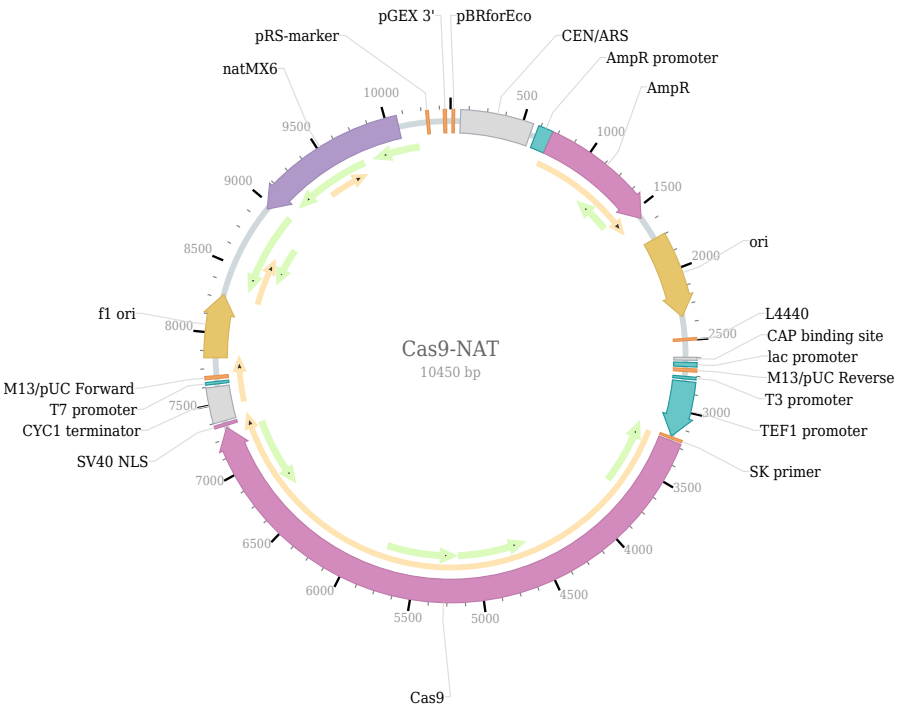
Cas9-NAT

载体编号 Vector ID	V000848
抗性 Antibiotic Resistance	Ampicillin
长度 Length	10450 bp
载体类型 Vector Type	Yeast Expression, CRISPR
宿主 Host	Yeast

产品信息

复制子 Replication Origin	ori
筛选标记 Selection Marker	Nourseothricin(clonNat)
拷贝数 Copy Number	Low Copy
启动子 Promoter	TEF1
克隆方法 Cloning Method	Restriction Enzyme
5' 引物 5' Primer	tctagagcggccgcTACGTAgatctgtttagcttgccttg
3' 引物 3' Primer	tctagagcggccgcCAATTGcgttttcgacactggatggc
培养菌株 Growth Strain(s)	DH10B
备注 Comment	Cas9-NAT is a modified yeast episomal plasmid built from p414-TEF1p-Cas9-CYC1t. This plasmid Cas9-NAT functions as the delivery vehicle for Cas9 endonuclease in polyploid industrial yeast: it constitutively expresses Cas9 via the TEF1 promoter and is selectable with nourseothricin due to the NAT1 resistance marker. The original backbone of Cas9-NAT is pRS414 (YCp centromeric plasmid), which relies on ARS (autonomously replicating sequence) for independent replication and CEN6 centromere sequence to segregate evenly during cell division, enabling it to exist as an extrachromosomal episome in yeast.

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 ℃.