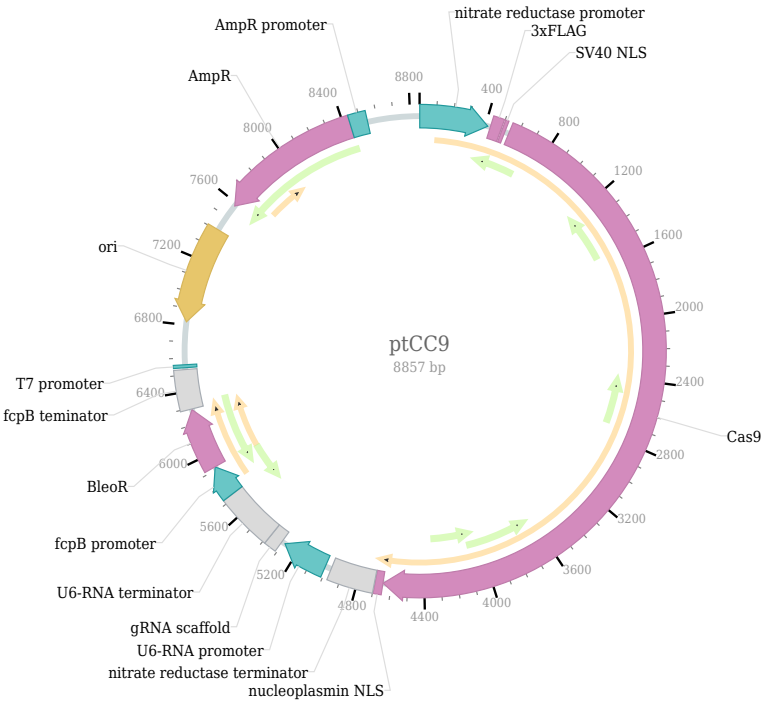
ptCC9

Vector ID	V002803
Antibiotic Resistance	Ampicillin
Length	8857 bp
Vector Type	Shuttle vector

Product Information

Replication Origin	ori
Source/Author	Stukenberg D, Zauner S, Dell'Aquila G, Maier UG.
Growth Strain(s)	DH10B
Growth Temperature	37℃
Comment	ptCC9 (GenBank: MH143578) is an all-in-one CRISPR-Cas9 vector for genome editing in Phaeodactylum tricornutum (Stukenberg et al., 2018). Derived from pPha-DUAL-[2xNR], it expresses diatom-codon-optimized Cas9 (diaCas9) under the nitrate-reductase (NR) promoter, which can be induced by nitrate-only nitrogen source. The sgRNA is transcribed from the endogenous P. tricornutum U6 promoter-terminator cassette; target spacers are inserted via BsaI-mediated cloning. The ble gene, driven by fcpB promoter, confers zeocin resistance for selection of diatom transformants. For propagation in E. coli, the plasmid carries pMB1 origin and ampicillin-resistance marker. Upon biolistic transformation, the vector integrates randomly into the diatom nuclear genome.

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 ℃.