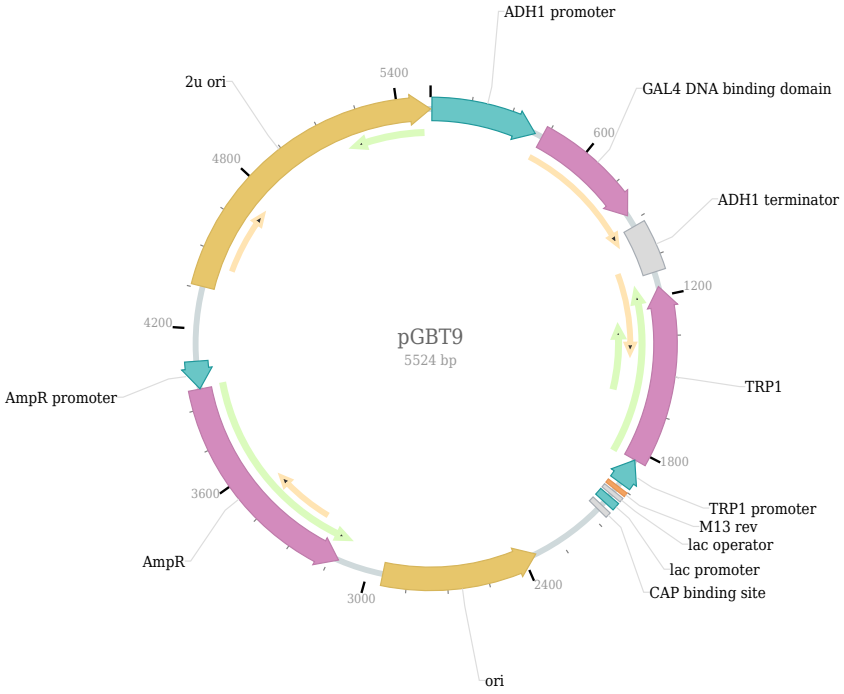
pGBT9

Vector ID	V001180
Antibiotic Resistance	Ampicillin
Length	5524 bp
Vector Type	Yeast Two-Hybrid System

Product Information

Replication Origin	ori
Promoter	ADH1
Comment	pGBT9 generates a hybrid protein that contains the sequences for the GAL4 DNA-binding domain (DNA-BD; a.a. 1–147). For the construction of a hybrid protein, the gene encoding the protein of interest is ligated into the MCS in the correct orientation and in the correct reading frame such that a fusion protein is generated. The fusion protein is expressed in yeast host cells from the constitutive ADH1 promoter; transcription is terminated at the ADH1 transcription termination signal. The hybrid protein is targeted to the yeast nucleus by nuclear localization sequences that are an intrinsic part of the GAL4 DNA-BD (2). pGBT9 is a shuttle vector that replicates autonomously in both E. coli and S. cerevisiae. It carries the bla gene (for ampicillin resistance in E. coli) and the TRP1 nutritional marker that allow yeast auxotrophs carrying pGBT9 to grow on limiting synthetic medium lacking Trp.

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 ℃.