# JD633

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| **载体编号 Vector ID** | V012098 |
| **抗性 Antibiotic Resistance** | Kanamycin |
| **长度 Length** | 21667 bp |
| **载体类型 Vector Type** | Binary vector |

## 产品信息

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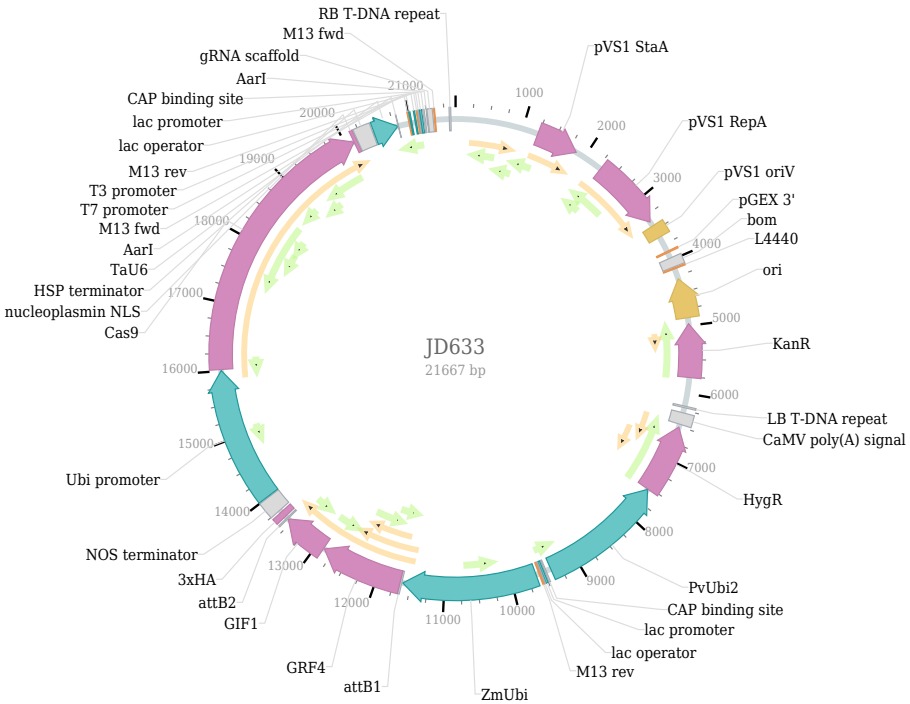
| **复制子 Replication Origin** | ori |
| **筛选标记 Selection Marker** | Hygromycin |
| **启动子 Promoter** | Ubi |
| **培养菌株 Growth Strain(s)** | stbl3 |
| **备注 Comment** | JD633 is an Agrobacterium binary vector optimized for high-efficiency CRISPR-Cas9 genome editing in wheat and other monocot crops. The chimeric GRF4-GIF1 fusion protein and wheat codon-optimized TaCas9 are both driven by the maize Zea mays ZmUbiquitin promoter. This fusion drastically boosts somatic regeneration efficiency, expands the range of transformable wheat genotypes, and enables cytokinin-free plant regeneration. Target gRNA is transcribed by the endogenous Triticum aestivum TaU6 Pol III promoter. Two flanking AarI type IIS restriction sites are integrated downstream of TaU6, enabling one-step Golden Gate assembly to rapidly insert custom gRNA spacer sequences via simultaneous AarI digestion and ligation. The hygromycin resistance gene (hptII/hygr) for transgenic cell selection is controlled by the Panicum virgatum PvUbi2 promoter with a CAT1 intron to enhance expression in monocot callus. All functional expression cassettes are assembled within a single T-DNA, combining regeneration enhancement and targeted gene editing in one construct. After genome editing, the GRF4-GIF1 and CRISPR-Cas9 components can be genetically segregated in $T_1$ progeny to generate transgene-free edited wheat plants. |

## 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 ℃.