## Plasmid Map

## pAAV-DS-CMV-GatewayB

Catalog Number: 0922



Left ITR (truncated): 1-117 CMV IE Enhancer/Promoter: 316-1072 Gateway B cassette: 1111-2815 (including attR1, Cm(r), ccdB, and attR2) Human growth hormone poly-A: 2854-3072 Right ITR: 3095-3224 f1 origin: 3286-3921 ampicillin resistance (bla) ORF: 4059-4916 pUC origin of replication: 5307-5783

## Note:

- 1. Truncation of the left ITR allows the newly replicated single strand AAV genome to fold over and form double strand (DS) sequence. The DS genome can be packed to form AAV particle if its length is within 2500bp. Gene expression from DS-AAV starts earlier and reaches much higher level compared with AAV with SS genome.
- Gateway B cassette allows directional subcloning of gene of interest using Invitrogen's Gateway<sup>™</sup> system. The pAAV-CMV-GatewayB shuttle plasmid works as a "destination plasmid". The gene of interest needs to be in pENTR plasmid. Refer to user manual of Invitrogen's Gateway<sup>™</sup> system for detail of subcloning procedures.
- 3. Chloramphenicol resistant gene (Cm(r)) allows confirmation of the plasmid to contain the Gateway<sup>™</sup> cassette by adding chloramphenicol to agar plates or culture broth.
- 4. The plasmid contains ccdB toxic gene. Use One Shot ccdB Survival T1 Phage Resistant Cell from Invitrogen (Cat. No. C7510-03) for further expansion.
- 5. This shuttle plasmid allows transgene as large as 980bp (between two recombination sites) to be packed into AAV vector.