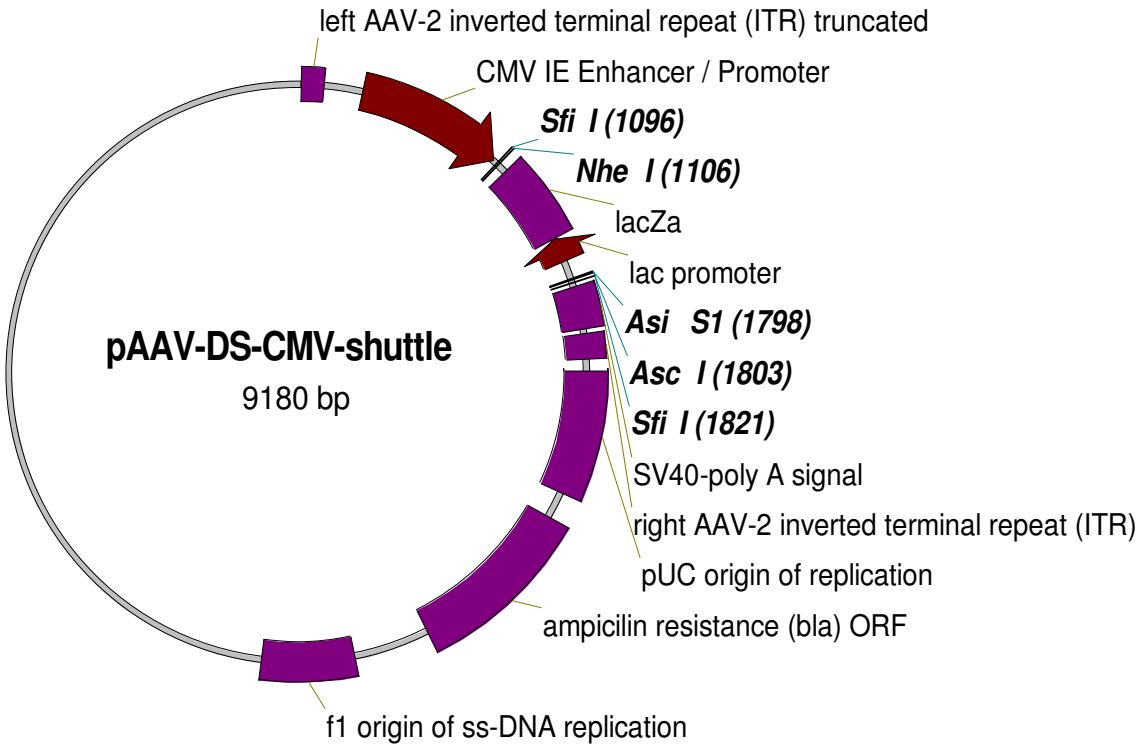
Applied Viromics

Plasmid Map

pAAV-DS-CMV-shuttle

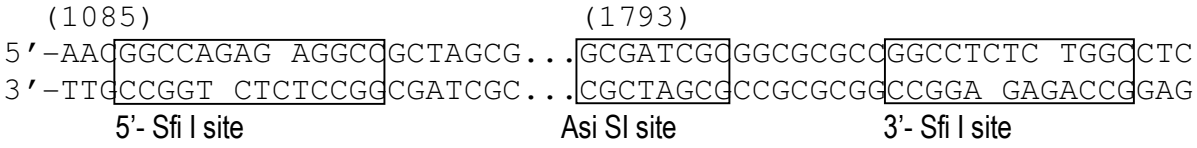
Catalog Number: 0915



Left ITR (truncated): 1-117
CMV IE enhancer/promoter: 316-1072
LacZα cassette (complementary): 1156-1703 (including lac promoter and lacZα)
SV40 poly-A: 1841-2059
Right ITR: 2082-2211
pUC origin of replication: 2273-2908
ampicillin resistance (bla) ORF: 3046-3903
f1 origin: 4294-4770

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.
2. lacZα expression cassette provides convenient blue-white selection when subcloning transgene insert behind CMV promoter. Bacterial strains that complement lacZα function such as DH5α or DH10B should be used.
3. Transgene can be cloned between Nhe I site at 5' and either Asc I or Asi SI site at 3'.
4. Detail of Sfi I subcloning sites:



5. Above Sfi I sites can be used to subclone blunt-ended fragment. Such fragment can be first ligated with Sfi I adaptors, then ligate with Sfi I-digested 5173bp fragment of the shuttle plasmid. Please contact Applied Viromics for detail.
6. This shuttle plasmid allows transgene as large as 1000bp to be packed into AAV vector.