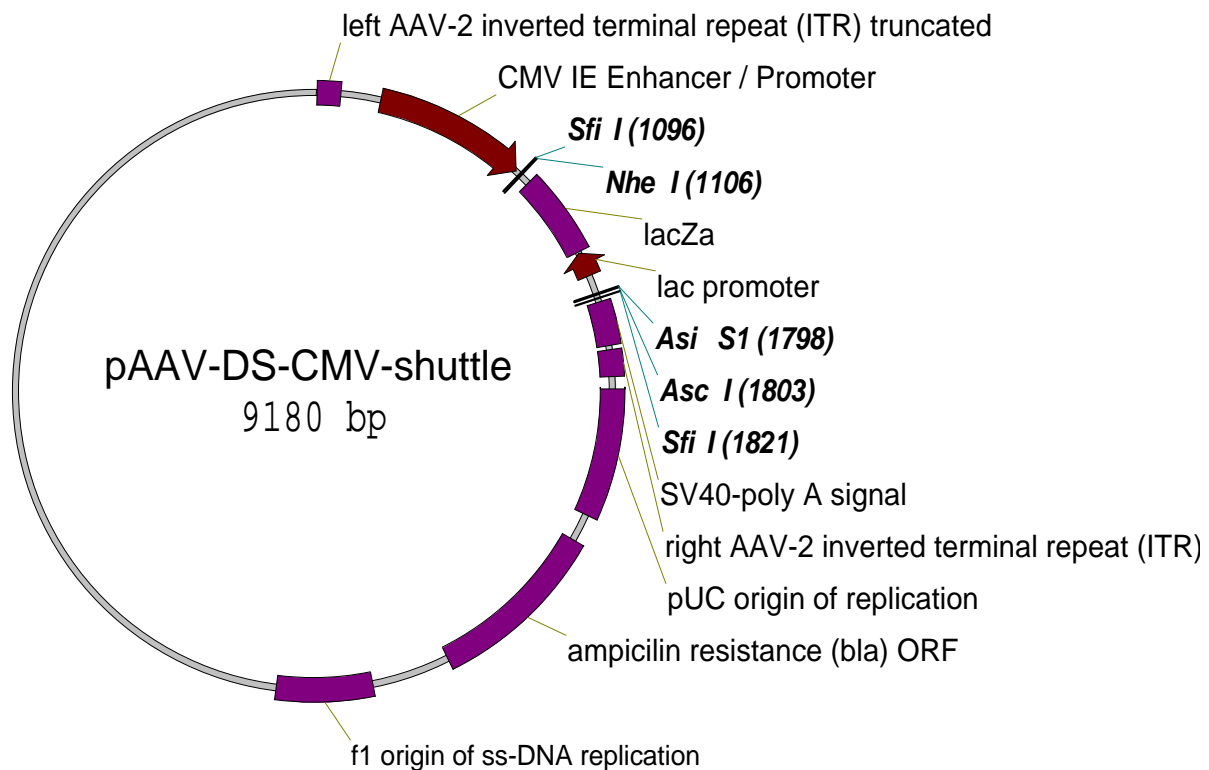


pAAV-DS-CMV-shuttle

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. lacZ α expression cassette provides convenient blue-white selection when subcloning transgene insert behind CMV promoter. Bacterial strains that complement lacZ α function such as DH5 α 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:

```
( 1085 )                ( 1793 )  
5' -AAGGGCCAGAG AGGCCGCTAGCG . . GCGATCGCGGCGCGCCGGCCTCTC TGGCTC  
3' -TTGCCGGT CTCTCCGGCGATCGC . . CGCTAGCGCCGCGCGGCCGGA GAGACCGAG  
      5'- Sfi I site                Asi SI site                3'- Sfi I site
```

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 8455bp fragment of the shuttle plasmid. Please contact Applied Viromics for detail.