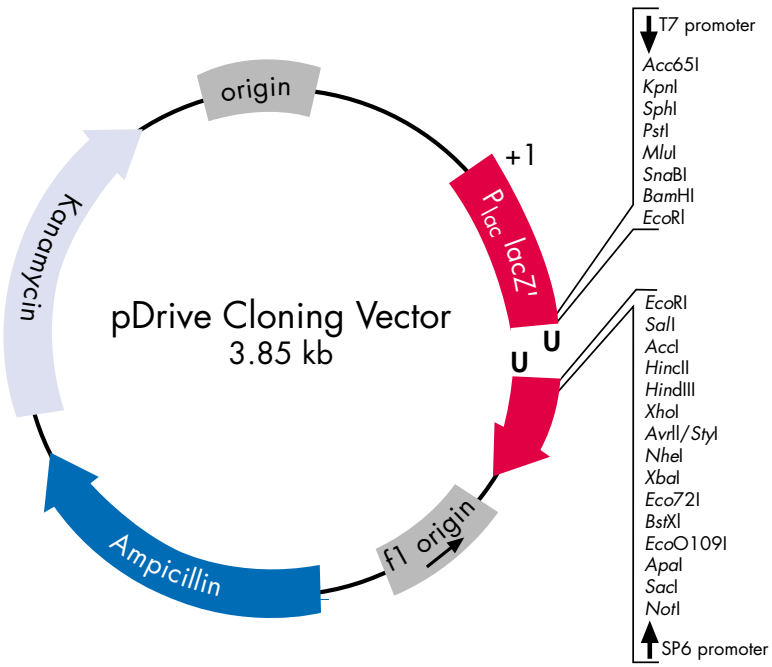
pDrive Cloning Vector

Positions of various elements

Vector size (bp)	3851
Multiple cloning site	266–393
LacZ α-peptide	216–593
T7 RNA polymerase promoter	239–258
T7 transcription start	256
SP6 RNA polymerase promoter	398–417
SP6 transcription start	400
Ampicillin resistance gene	1175–2032
Kanamycin resistance gene	2181–2993
pUC origin	3668
Phage f1 origin	588–1043
Primer binding sites:*	
M13 forward (–20)	431–447
M13 forward (–40)	451–467
M13 reverse	209–224
T7 promoter primer	239–258
SP6 promoter primer	400–418

** Primer sequences are provided below*



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     M13 reverse primer
     binding site                                          T7 promoter
5' GAA ACA GCT ATG ACC ATG ATT ACG CCA AGC TCT AAT ACG ACT CAC TAT AGG GAA      260
3' CTT TGT CGA TAC TGG TAC TAA TGC GGT TCG AGA TTA TGC TGA GTG ATA TCC CTT
                M   T   M   I   T   P   S   S   N   T   T   H   Y   R   E
                LacZ α-petide coding sequence

          Acc65I                      MluI
            |     KpnI       SphI  PstI|          SnaBI    BamHI     EcoRI
            |      |          |     |  |            |         |         |
AGC TCG GTA CCA CGC ATG CTG CAG ACG CGT TAC GTA TCG GAT CCA GAA TTC GTG      314
TCG AGC CAT GGT GCG TAC GAC GTC TGC GCA ATG CAT AGC CTA GGT CTT AAG CAC
 S   S   V   P   R   M   L   Q   T   R   Y   V   S   D   P   E   F   V

PCR product
                          SalI
                          |AccI   HindIII          AvrII
                EcoRI     | |HincII|       XhoI    StyI   NheI     XbaI
                  |       | | |    |         |      |      |        |
ATU   A TCT GAA TTC GTC GAC AAG CTT CTC GAG CCT AGG CTA GCT CTA GAC CAC      365
TA   UT AGA CTT AAG CAG CTG TTC GAA GAG CTC GGA TCC GAT CGA GAT CTG GTG
          S   E   F   V   D   K   L   L   E   P   R   L   A   L   D   H

Eco72I      EcoO109I      SacI
  |  BstXI    |     ApaI    |    NotI                 ←————————————————
  |   |       |      |      |     |                        SP6 promoter
ACG TGT GGG GGC CCG AGC TCG CGG CCG CTG TAT TCT ATA GTG TCA CCT AAA TGG      419
TGC ACA CCC CCG GGC TCG AGC GCC GGC GAC ATA AGA TAT CAC AGT GGA TTT ACC
 T   C   G   G   P   S   S   R   P   L   Y   S   I   V   S   P   K   W

                M13 forward (-20) primer   M13 forward (-40) primer
                binding site               binding site
                ←————————————————          ←————————————————
CCG CAC AAT TCA CTG GCC GTC GTT TTA CAA CGT CGT GAC TGG GAA AAC 3'           467
GGC GTG TTA AGT GAC CGG CAG CAA AAT GTT GCA GCA CTG ACC CTT TTG 5'
 P   H   N   S   L   A   V   V   L   Q   R   R   D   W   E   N
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Primer sequences

M13 forward (–20): 5' GTAAAACGACGGCCAGT 3'

M13 forward (–40): 5' GTTTTCCCAGTCACGAC 3'

M13 reverse: 5' AACAGCTATGACCATG 3'

SP6 promoter: 5' CATTTAGGTGACACTATAG 3'

T7 promoter: 5' GTAATACGACTCACTATAG 3'