

pTWIN1 DNA: Location of Sites

AatII	1	6688		
AfeI	1	3194		
AgeI	1	7048		
AlwNI	1	1896		
ApaI	1	3891		
BamHI	1	7239		
BlpI	1	7291		
BmtI	1	5971		
BsaBI	1	5618		
BseRI	1	6243		
BsiWI	1	6628		
BsmI	1	6769		
BssHII	1	3687		
BstEII	1	3916		
BstZ17I	1	2480		
DraIII	1	1319		
EcoO109I	1	7318		
EcoRI	1	6424		
EcoRV	1	3650		
FspI	1	702		
HpaI	1	3594		
KasI	1	3459		
NarI	1	3459		
NcoI	1	6410		
NdeI	1	5722		
NheI	1	5971		
NotI	1	6416		
NruI	1	6377		
PaeR7I	1	6430		
PciI	1	2310		
PflFI	1	2504		
PmeI	1	5606		
PsiI	1	1194		
PspOMI	1	3891		
PspXI	1	6429		
PstI	1	7228		
PvuI	1	555		
SacII	1	6798		
ScaI	1	444		
SfoI	1	3459		
SgrAI	1	6957		
SpeI	1	6462		
StuI	1	7062		
SwaI	1	1096		
ThiI	1	6430		
Thl111I	1	2504		
XbaI	1	5683		
XhoI	1	6430		
ZraI	1	6688		
AcuI	2	240	1783	
BclI	2	4084	5960	
BglI	2	803	3035	
BsaAI	2	1322	2499	
BsmBI	2	2602	3489	
BspEI	2	5935	7369	
BspHI	2	87	1590	
BspQI	2	6404	6437	
BsrGI	2	6393	6617	
BssSI	2	258	2137	
BtgI	2	6410	6798	
EagI	2	3030	6417	
EcoNI	2	4562	6471	
MluI	2	4098	7004	
NmeAIII	2	799	3873	
PfIMI	2	4517	5576	
PshAI	2	3253	6324	
SapI	2	6404	6437	
SphI	2	4627	4635	
AflIII	3	2310	4098	7004
AhdI	3	922	5798	7121
AvaI	3	1213	6430	6475
BanII	3	1395	3891	6435
BfuAI	3	5772	6787	7095
BsaI	3	855	5917	6878
BsaXI	3	1268	3438	6296
BsoBI	3	1213	6430	6475
BspMI	3	5772	6787	7095
BstAPI	3	4416	6460	6787
MfeI	3	5800	6253	7123
PvuII	3	2660	3407	3500
SalI	3	6685	6778	6988
SspI	3	120	1093	1114
XcmI	3	3706	3724	4240

There are no restriction sites for the following enzymes:
AarI(x), Acc65I, AflII, AleI, AscI, AsiSI, AvrII, BaeI, BbvCI, BglIII, BmgBI, Bpu10I, BspDI, BstBI, Bsu36I, ClaI, CspCI, FseI, FspAI(x), HindIII, I-CeuI, I-SceI, KpnI, MscI, NsiI, PI-PspI, PI-SceI, PacI, PmlI, PpuMI, RsrII, SacI, SanDI(x), SbfI, SexAI, SfiI, SmaI, SnaBI, SrfI(x), TspMI, XmaI
(x) = enzyme not available from NEB

Ssp DnaB Intein Forward Primer →

...ACTGGGACTCCATCGTTTCTATTACGGAGACTGGAGTCGAAGAGGTTTTTGATTGACTGTGCCAGGACCACATAACTTTGTCGCGAAT
NruI
Ssp DnaB Intein ... V A N

Ssp DnaB Intein

GACATCATTGTACACAAC GGAAGAGCCATGGGCGCCGCGAATTCTCGAGGGCTCTCC
D I I V H N G R A M G G R E F L E G S S

pTWIN1
SpeI
TGC ATCACGGGAGATGCACTAGTTGCCCTACCCGAGGGCGAGTCGGTACGCATCGCCGACATCGTCC...
C I T G D A ... Mxe GyrA Intein
Mxe GyrA Intein

pTWIN2
SpeI
TGC GTATCCGGTGACACCATTGTAATGACTAGTGGCGGTCCGCGCACTGTGGCTGAAGTGGAGGGCAAACCGTTCCACC...
C V S G D T ... Mth RIR1 Intein
Mth RIR1 Intein