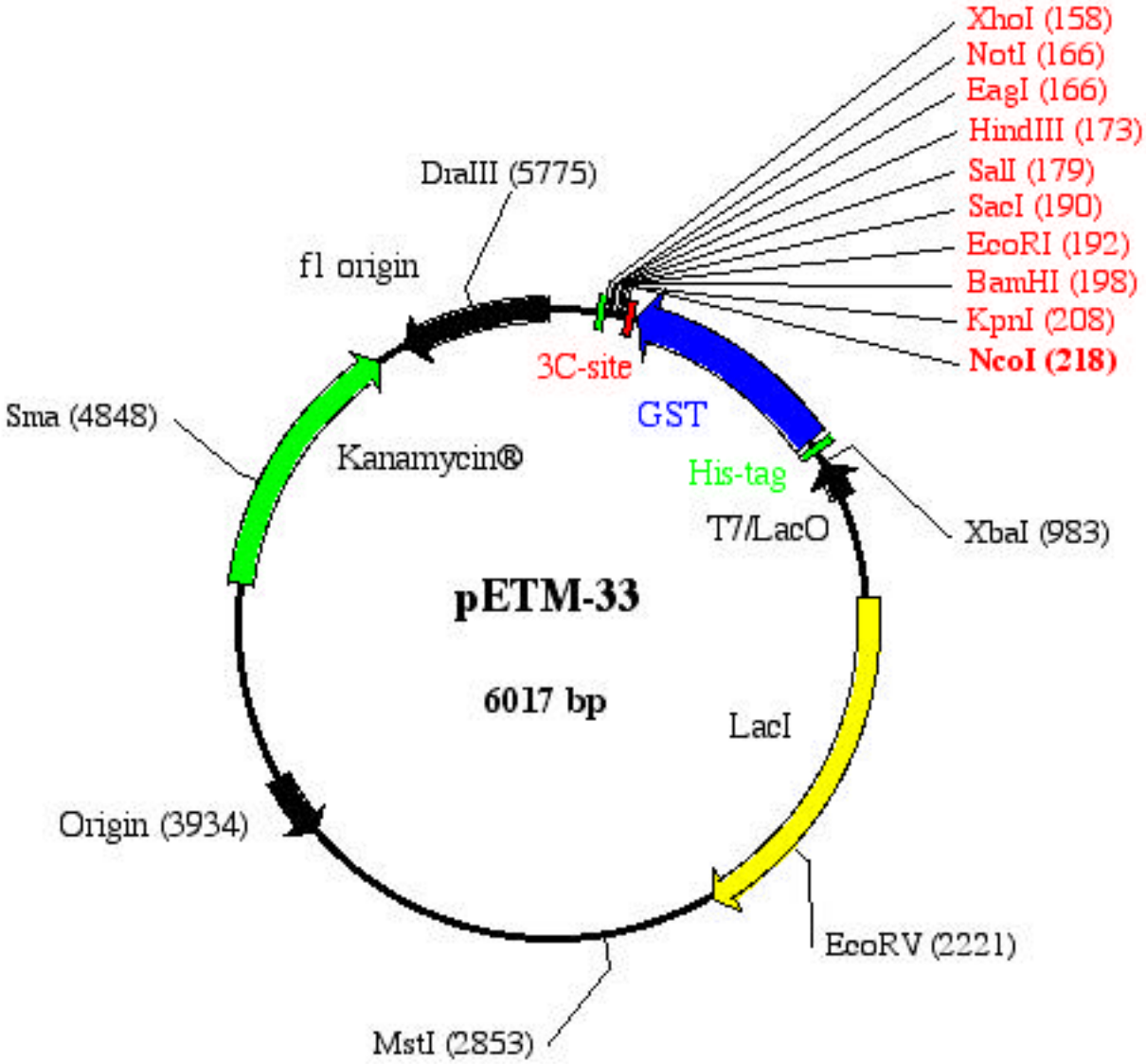
XhoI (158)
NotI (166)
EagI (166)
HindIII (173)
SalI (179)
SacI (190)
EcoRI (192)
BamHI (198)
KpnI (208)
NcoI (218)
DraIII (5775)
f1 origin
3C-site
GST
Sma (4848)
Kanamycin®
His-tag
T7/LacO
XbaI (983)
pETM-33
6017 bp
LacI
Origin (3934)
EcoRV (2221)
MstI (2853)

pETM-33

```
     T7 promoter  -->            Lac operator      XbaI
CGAAATTAATACGACTCACTATAGGGGAATTGTGAGCGGATAACAATTCCCCTCTAGAAA
GCTTTAATTATGCTGAGTGATATCCCCTTAACACTCGCCTACCGTTAAGGGGAGATCTTT

                        rbs                 His-tag
TAATTTTGTTTAACTTTAAGAAGGAGATATACCATGAAACATCACCATCACCATCACAGT
ATTAAAACAAATTGAAATTCTTCCTCTATATGGTACTTTGTAGTGGTAGTGGTAGTGTCA
                                   METLysHisHisHisHisHisHisAsn


ACACTAAGCAATTCCATGTCCCCTATACTA...621bp....GGCGACCATCCTCCAAAA
TGTGATTCGTTAAGGTACAGGGGATATGAT....GST.....CCGCTGGTAGGAGGTTTT
ThrSerSerAsnPheMetSerProIleLeu...207aa....GlyAspHisProProLys


                 3C-site  NcoI         KpnI  BamHI EcoRI
TCGGATCTGGAAGTTCTGTTCCAG GGGCCCATGGACTAATGAGGTACCGGATCCGAATTC
AGCCTAGACCTTCAAGACAAGGTC CCCGGGTACCTGATTACTCCATGGAATAGGCTTAAG
SerAspLeuGluValLeuPheGln|GlyProMETAsp******

                      NotI
SacI    SalI HundIII EagI    XhoI        C-His-tag
GAGCTCCGTCGACAAGCTTGCGGCCGCACTCGAGCACCACCACCACCACCACTGAGATCCG
CTCGAGGCAGCTGTTCGAACGCCGGCGTGAGCTCGTGGTGGTGGTGGTGGTGACTCTAGGC
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Single Cutters Listed by Site Order

80	Bpu1102I	190	SstI	697	MscI	2853	MstI
80	EspI	192	EcoRI	915	SpeI	3617	Tth111I
158	XhoI	198	BamHI	983	XbaI	4045	BsiI
160	SciI	204	Asp718I	1049	BglII	4288	AlwNI
166	Eco52I	204	Acc65I	1090	SgrAI	4420	Eco57I
166	EagI	208	KpnI	1246	SphI	4731	NruI
166	XmaIII	218	NcoI	1455	ApaBI	4731	SpoI
166	NotI	331	ScaI	1771	MluI	4765	ClaI
173	HindIII	333	RleAI	1952	BstEII	4946	XmaI
179	SalI	477	SwaI	2182	BssHII	4948	SmaI
188	Ecl136II	505	AsuII	2221	EcoRV	5074	PvuI
188	EcoICRI	505	Csp45I	2277	HpaI	5074	XorII
190	SacI	697	BalI	2835	BglI	5775	DraIII

Non Cutting Enzymes

AatII	AflII	AgeI	AscI	AvrII	Bsp1407I
BspMI	Bsu36I	CspI	CvnI	Eam1105I	Eco31I
Eco72I	FseI	I-PpoI	MfeI	Mlu113I	MstII
NdeI	NheI	PacI	PinAI	PmaCI	PmeI
PstI	SacII	SauI	SfiI	SnaBI	SplI
SrfI	SstII	StuI	SunI		