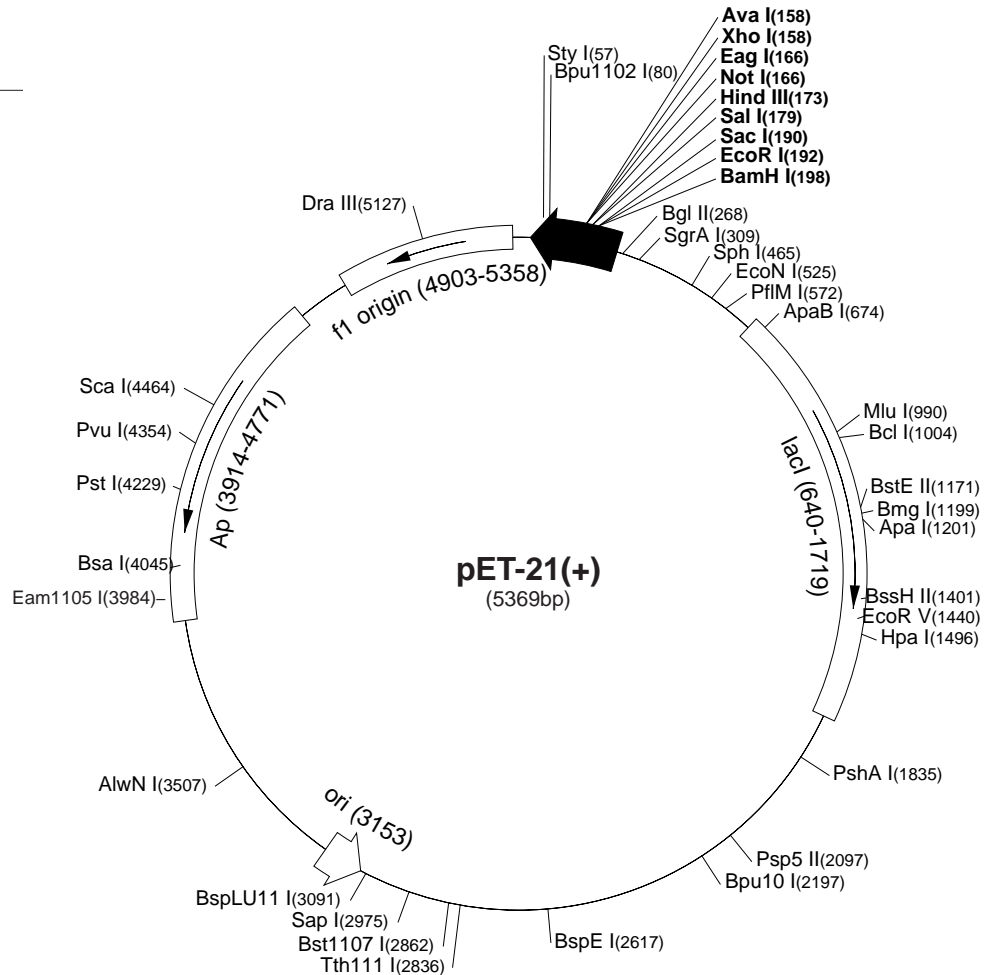


pET-21(+)⁺ Vector

pET-21(+)⁺ (Cat. No. 69770-3) is a transcription vector designed for expression from bacterial translation signals carried within a cloned insert. It therefore lacks the ribosome binding site and ATG start codon present on the pET translation vectors. A C-terminal His•Tag[®] sequence is available. Unique sites are shown on the circle map. Note that the sequence is numbered by the pBR322 convention, so the T7 expression region is reversed on the circular map. The cloning/expression region of the coding strand transcribed by T7 RNA polymerase is shown below. The f1 origin is oriented so that infection with helper phage will produce virions containing single-stranded DNA that corresponds to the coding strand. Therefore, single-stranded sequencing should be performed using the T7 terminator primer (Cat. No. 69337-3).

pET-21(+)⁺ sequence landmarks

T7 promoter	237-253
T7 transcription start	236
Multiple cloning sites (<i>Bam</i> H I - <i>Xho</i> I)	158-203
His•Tag [®] coding sequence	140-157
T7 terminator	26-72
<i>lacI</i> coding sequence	640-1719
pBR322 origin	3153
<i>bla</i> coding sequence	3914-4771
f1 origin	4903-5358



pET-21(+)⁺ cloning/expression region

pET-21(+) Restriction Sites

Enzyme	# Sites	Locations
AccI	2	180 2861
AccIII	8	757 1485 1816 2600 2741 3043 4283 4967
Acil	80	
AflIII	2	990 3091
AluI	25	
AlwI	16	
Alw21I	9	159 190 490 974 2085 2909 3409 4570 4655
Alw44I	4	970 2905 3405 4651
AlwNI	1	3507
Apal	1	1201
ApaBI	1	674
ApoI	4	192 1265 4929 4940
AvaI	1	158
Avall	7	1542 1918 2006 2097 2376 4122 4344
BamHI	1	198
BanI	9	312 333 447 910 1629 1759 1885 3932 5164
BanII	5	190 374 388 1201 5202
BbsI	4	1136 1475 1849 2209
BbvI	26	
BccI	13	
Bce83I	7	21 1804 1974 3182 3480 3721 4589
BceII	5	509 850 1477 3593 5153
BcgI	10	160 194 1282 1316 1816 1850 2668 2702 4489 4523
BclI	1	1004
Bfal	7	70 203 2105 3586 3839 4174 5278
BglI	2	2054 4104
BglII	1	268
BmgI	1	1199
BpmI	5	828 1317 1951 2618 4054
Bpu10I	1	2197
Bpu1102I	1	80
BsaI	1	4045
BsaAI	2	2843 5127
BsaBI	3	267 273 2288
BsaHI	6	313 334 448 947 1630 4521
BsaJI	6	57 427 433 1625 2063 3251
BsaWI	7	2 1309 1812 2280 3297 3444 4275
BsaXI	2	1649 5075
Bsbl	2	2807 5034
BscGI	13	
BsgI	3	841 1041 2251
Bsil	2	3264 4648
BsiEI	6	169 1775 3007 3431 4354 4503
BsII	20	
BsmAI	7	687 1092 1218 1605 2732 4045 4821
BsmBI	2	1605 2732
BsmFI	4	451 1992 2362 5342
BsoFI	46	
Bsp24I	10	280 312 831 863 1133 1165 3584 3616 3762 3794
Bsp1286I	13	
BspEI	2	2 2280
BspGI	1	2617
BspLU11I	1	3091
BsrI	25	
BsrBI	4	223 3024 4825 5271
BsrDI	4	1037 1403 4045 4219
BsrFI	7	300 309 676 1888 2048 4064 5228
BssHII	1	1401

Enzyme	# Sites	Locations
Bst1107I	1	2862
BstEII	1	1171
BstXI	3	792 921 1044
BstYI	12	
CacBI	38	
Cjel	24	
CjePI	18	
CviJI	85	
CviRI	24	
Ddel	11	
DpnI	27	
DraI	3	3850 3869 4561
DraIII	1	5127
DrdI	3	2784 3199 5082
DrdII	2	713 5132
Dsal	2	427 2063
EaeI	5	166 298 430 1664 4372
EagI	1	166
Eam1105I	1	3984
EarI	3	608 2975 4779
EciI	4	767 3165 3311 4139
Eco47III	3	395 1896 2345
Eco57I	2	3639 4651
EcoNI	1	525
EcoO109I	3	53 423 2097
EcoRI	1	192
EcoRII	7	713 1028 1568 1625 3117 3238 3251
EcoRV	1	1440
FauI	17	
FokI	10	1036 1045 2310 2372 2450 2636 2777 3950 4131 4418
Fspl	2	2072 4206
GdIII	5	166 298 430 1664 4372
HaeI	5	718 2039 3106 3117 3569
HaeII	14	
HaeIII	24	
Hgal	12	
HgiEII	2	588 3677
Hhal	45	
Hin4I	3	889 3983 4057
HincII	2	181 1496
HindIII	1	173
Hinfl	14	
HpaI	1	1496
HphI	16	
MaeI	15	
MaeIII	18	
MbolI	14	
MluI	1	990
MmeI	3	3306 3490 5104
MnlI	26	
MseI	26	
MsiI	9	1042 1330 1360 2078 2273 2664 4236 4395 4754
MspI	31	
MspA1I	9	84 1020 1590 1683 2682 2801 3433 3678 4619
Mwol	38	
NarI	4	313 334 448 1630
NciI	12	
NgoAIV	4	300 1888 2048 5228
NlaIII	24	
NlaIV	25	
Nott	1	166
NspI	4	465 2436 2728 3095
Pfi1108I	2	1877 4002
PfIMI	1	572
PleI	9	251 539 626 1422 2985 3470 3973 5062 5070
PshAI	1	1835
Psp5II	1	2097

Enzyme	# Sites	Locations
Psp1406I	6	652 2020 2416 4210 4583 4912
PstI	1	4229
PvuI	1	4354
PvuII	3	1590 1683 2682
RcaI	3	388 3811 4819
RsaI	3	1137 2897 4464
SacI	1	190
Sall	1	179
SapI	1	2975
Sau96I	18	
Sau3AI	27	
Scal	1	4464
ScrFI	19	
SfaNI	20	
Sfcl	5	236 3356 3547 4225 5346
SgrAI	1	309
SphI	1	465
Sspl	2	4788 4919
StyI	1	57
TaqI	13	
TaqII	9	898 1116 1789 2993 4332 4517 4670 4687 5031
TfiI	5	1669 1971 2141 2645 3066
Thal	34	
Tsel	26	
Tsp45I	8	1171 1999 2530 2743 2838 4240 4451 5300
Tsp509I	15	
Tth111I	1	2836
Tth111II	6	829 1522 2552 3681 3688 3720
UbaII	20	
VspI	4	251 1675 1734 4156
XcmI	3	846 1362 1380
XhoI	1	158
XmnI	2	2649 4583

Enzymes that do not cut pET-21(+):

AatII	AflII	AgeI	AscI	AvrII
BaeI	BseRI	BsmI	BspMI	BsrGI
Bsu36I	Clal	FseI	KpnI	MscI
MunI	NcoI	NdeI	NheI	NruI
NsiI	NspV	PacI	PmeI	PmlI
RleAI	RsrII	SacII	SexAI	SfiI
SgfI	Smal	SnaBI	SpeI	SrfI
Sse8387I	StuI	SunI	Swal	XbaI