pET30 a+

Location: Expresión 02
Resistance: Kanamycin 25 µg/mL

General description for pET-30a-c vectors:
The pET-30a-c(+) vectors carry an N-terminal His•Tag®/thrombin/S•Tag™/enterokinase configuration plus an optional C-terminal His•Tag sequence. 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.
The maps for pET-30b+ and pET-30c+ are the same as pET30a+ with the following exceptions: pET-30b+ is a 5421 bp plasmid; subtract 1 bp from each site beyond BamHI at 198. pET-30c+ is a 5423 bp plasmid; add 1 bp to each site beyond BamHI at 198.

Complete sequence: http://sil.biochem.uiowa.edu/vectors/pET/pET-30a_seq.html

Genotype of *E coli* strain BL21(DE3) pLysS : F– *ompT hsdS*(rB– mB–) *gal dcm* λ(DE3) pLysS (Camr)(λ(DE3): *lac*I, *lac*UV5-T7 gene 1, *ind*1, *sam*7, *nin*5)

Landmarks and maps

pET-30a(+) sequence landmarks	
T7 promoter	419-435
T7 transcription start	418
His•Tag coding sequence	327-344
S•Tag coding sequence	249-293
Multiple cloning sites	
(*Nco* I - *Xho* I)	158-217
His•Tag coding sequence	140-157
T7 terminator	26-72
lacI coding sequence	826-1905
pBR322 origin	3339
Kan coding sequence	4048-4860
f1 origin	4956-5411

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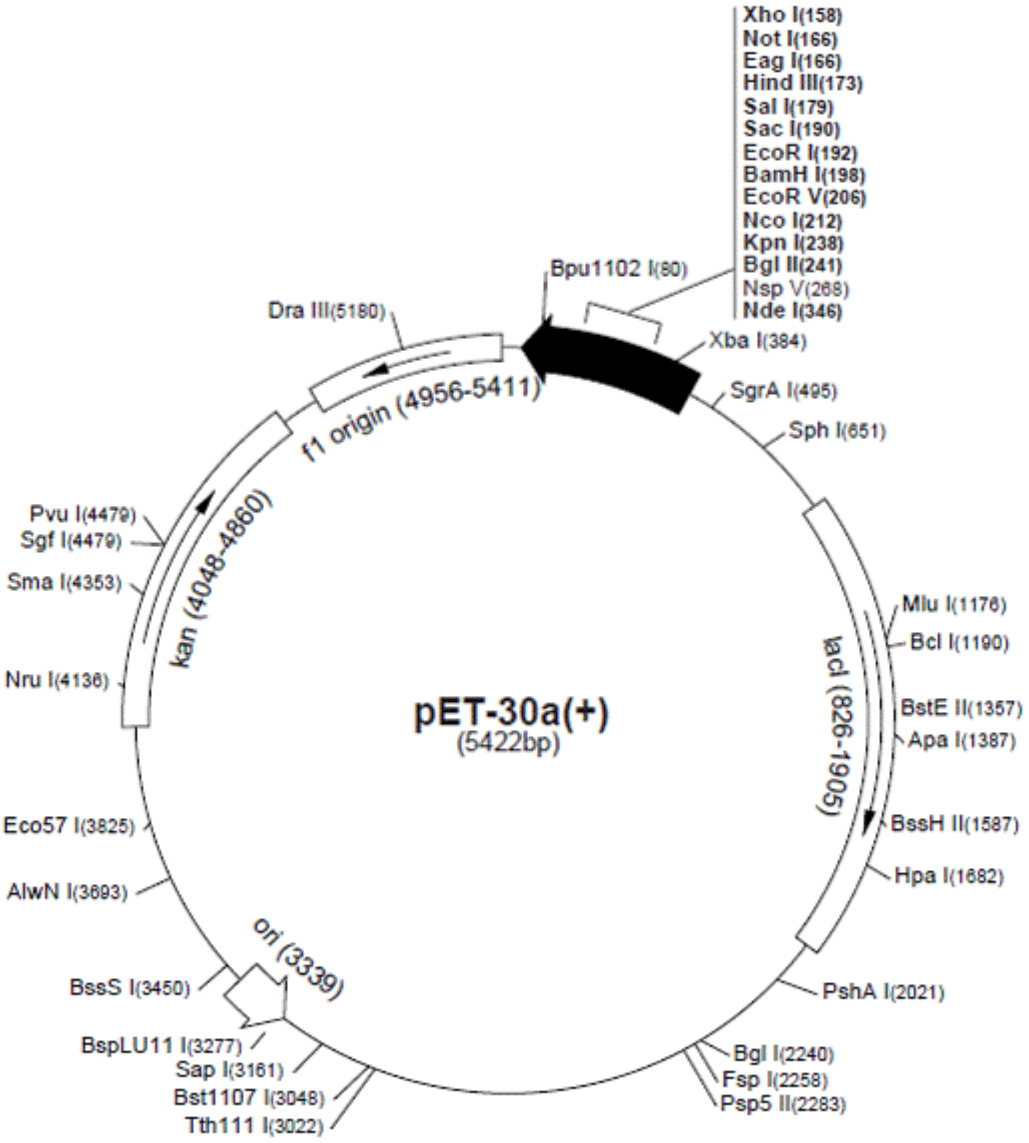

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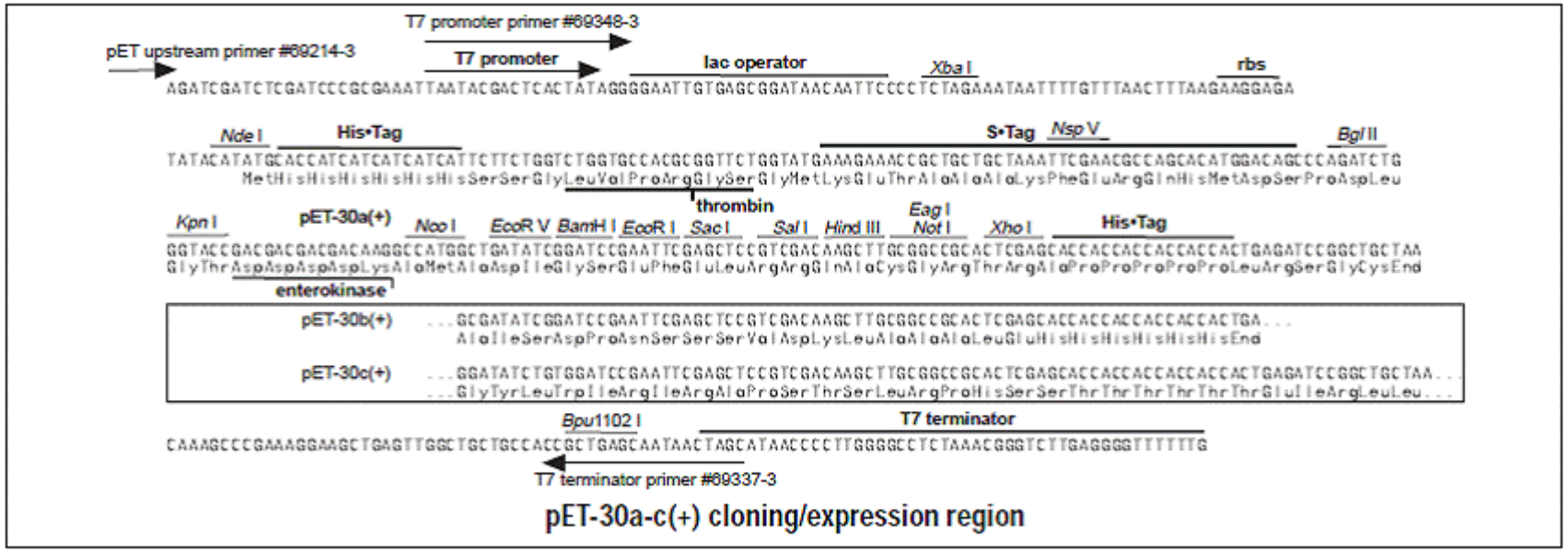

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pET-30a(+) Restriction Sites

TB095 12/

Enzyme	# Sites	Locations					
AccI	2	180	3047				
AceIII	7	943	1671	2002	2786	2927	
		3229	5020				
AciI	75						
AflIII	2	1176	3277				
AluI	22						
AlwI	13						
Alw21I	7	159	190	676	1160	2271	
		3095	3585				
Alw44I	3	1156	3091	3591			
AlwNI	1	3693					
ApaI	1	1387					
ApaBI	1	860					
ApoI	7	192	270	1451	4092	4276	
		4982	4993				
AvaI	2	158	4351				
AvaII	5	1728	2104	2192	2283	2562	
BamHI	1	198					
BanI	10	234	310	498	519	633	
		1096	1815	1945	2071	5217	
BanII	6	190	560	574	1387	4134	
		5255					
BbsI	4	1322	1661	2035	2395		
BbvI	25						
BccI	14						
Bce83I	6	21	1990	2160	3368	3666	
		3907					
BcefI	6	695	1036	1663	3779	4798	
		5206					
BcgI	8	160	194	1468	1502	2002	
		2036	2854	2888			
BclI	1	1190					
BfaI	6	70	385	2291	3772	4079	
		5331					
BglI	1	2240					
BglII	1	241					
BmgI	1	1385					
BpmI	4	1014	1503	2137	2804		
Bpu10I	2	2383	4496				
Bpu1102I	1	80					
BsaAI	2	3029	5180				
BsaBI	3	449	459	2474			
BsaHI	5	499	520	634	1133	1816	
BsaJI	10	57	212	613	619	1811	
		2249	3437	4350	4351	4752	
BsaWI	7	2	1495	1998	2466	3483	
		3630	4614				
BsaXI	2	1835	5128				
BsbI	2	2993	5087				
BscGI	11						
BsgI	3	1027	1227	2437			
BsiI	1	3450					
BsiEI	5	169	1961	3193	3617	4479	
BslI	26						
BsmI	2	4363	4440				
BsmAI	6	873	1278	1404	1791	2918	
		4495					
BsmBI	3	1791	2918	4495			
BsmFI	4	637	2178	2548	5395		
BsoFI	43						
Bsp24I	10	466	498	1017	1049	1319	
		1351	3770	3802	3948	3980	
Bsp1286I	12						
BspEI	2	2	2466				
BspGI	1	2803					
BspLU11I	1	3277					
BsrI	21						
BsrBI	4	405	3210	4878	5324		
BsrDI	2	1223	1589				
BsrFI	7	486	495	862	2074	2234	
		4433	5281				
BssHII	1	1587					

Enzyme	# Sites	Locations				
Bst1107I	1	3048				
BstEII	1	1357				
BstXI	3	978	1107	1230		
BstYI	9	132	198	241	740	1952
		2469	3918	3929	4728	
Cac8I	40					
CjeI	24					
CjePI	18					
ClaI	2	453	4170			
CviJI	85					
CviRI	31					
DdeI	11					
DpnI	23					
DraIII	1	5180				
DrdI	3	2970	3385	5135		
DrdII	2	899	5185			
DsaI	3	212	613	2249		
EaeI	4	166	484	616	1850	
EagI	1	166				
EarI	3	794	3161	4292		
EciI	3	953	3351	3497		
Eco47III	3	581	2082	2531		
Eco57I	1	3825				
EcoNI	2	711	4391			
EcoO109I	3	53	609	2283		
EcoRI	1	192				
EcoRII	9	899	1214	1754	1811	3303
		3424	3437	4367	4724	
EcoRV	1	206				
FauI	17					
FokI	9	1222	1231	2496	2558	2636
		2822	2963	4117	4723	
FspI	1	2258				
GdiII	4	166	484	616	1850	
HaeI	7	217	904	2225	3292	3303
		3755	4566			
HaeII	14					
HaeIII	24					
HgaI	11					
HgiEII	2	774	3863			
HhaI	46					
Hin4I	4	203	1075	4165	4707	
HincII	2	181	1682			
HindIII	1	173				
HinfI	18					
HpaI	1	1682				
HphI	16					
KpnI	1	238				
MaeII	14					
MaeIII	16					
MboII	13					
MluI	1	1176				
MmeI	7	3492	3676	4121	4315	4677
		4686	5157			
MnlI	25					
MseI	25					
MslI	6	1228	1516	1546	2264	2459
		2850				
MspI	29					
MspA1I	9	84	283	1206	1776	1869
		2868	2987	3619	3864	
MwoI	39					
NarI	4	499	520	634	1816	
NciI	12					
NcoI	1	212				
NdeI	1	346				
NgoAIV	4	486	2074	2234	5281	
NlaIII	26					
NlaIV	23					
NotI	1	166				
NruI	1	4136				
NsiI	2	4329	4595			

Enzyme	# Sites	Locations				
NspI	4	651	2622	2914	3281	
NspV	1	268				
Pfl1108I	1	2063				
PflMI	3	260	758	4742		
PleI	9	433	725	812	1608	3171
		3656	4711	5115	5123	
PshAI	1	2021				
Psp5II	1	2283				
Psp1406I	4	838	2206	2602	4965	
PvuI	1	4479				
PvuII	3	1776	1869	2868		
RcaI	3	574	3997	4872		
RsaI	4	236	1323	3083	4314	
SacI	1	190				
SalI	1	179				
SapI	1	3161				
Sau96I	14					
Sau3AI	23					
ScrFI	21					
SfaNI	23					
SfcI	4	418	3542	3733	5399	
SgfI	1	4479				
SgrAI	1	495				
SmaI	1	4353				
SphI	1	651				
SspI	2	4404	4972			
StyI	2	57	212			
TaqI	17					
TaqII	6	1084	1302	1975	3179	4733
		5084				
TfiI	9	1855	2157	2327	2831	3252
		4390	4446	4618	4709	
ThaI	36					
TseI	25					
Tsp45I	7	1357	2185	2716	2929	3024
		4626	5353			
Tsp509I	21					
Tth111I	1	3022				
Tth111II	8	1015	1708	2738	3867	3874
		3906	4315	4442		
UbaJI	18					
VspI	5	433	1861	1920	4678	4867
XbaI	1	384				
XcmI	3	1032	1548	1566		
XhoI	1	158				
XmnI	2	2835	4868			

Enzymes that do not cut pET-30a(+):

AatII	AflII	AgeI	AscI	AvrII
BaeI	BsaI	BseRI	BspMI	BsrGI
Bsu36I	DraI	Eam1105I	FseI	MscI
MunI	NheI	PacI	PmeI	PmlI
PstI	RleAI	RsrII	SacII	ScaI
SexAI	SfiI	SnaBI	SpeI	SrfI
Sse8387I	StuI	SunI	SwaI	