

	Cat. No.
pET-44 Ek/LIC DNA	71143-3

Diagram illustrating the ligation reaction for creating a recombinant plasmid:

Top DNA Fragment (Synthetic):

5' ^{start} GACGACGACAAGATG...sense...TAACCGGGCTTCTCTCT 3'
 3' CTCTGCTGCTTTCTAC...anti...ATTGGCCCGAAGAGAG 5'

Enzyme/Condition: T4 DNA pol + dATP

Bottom DNA Fragment (Ekl/LIC vector):

GACGACGACAAGATG...sense...TAA
 AC...anti...ATTGGCCCGAAGAGAG

Reaction: +

Top DNA Fragment (Synthetic):

GAT CTACTGCTGCTTTCT

Bottom DNA Fragment (Ekl/LIC vector):

CCGGGGCTTCTCTCTCA

Enzyme/Condition: annealing

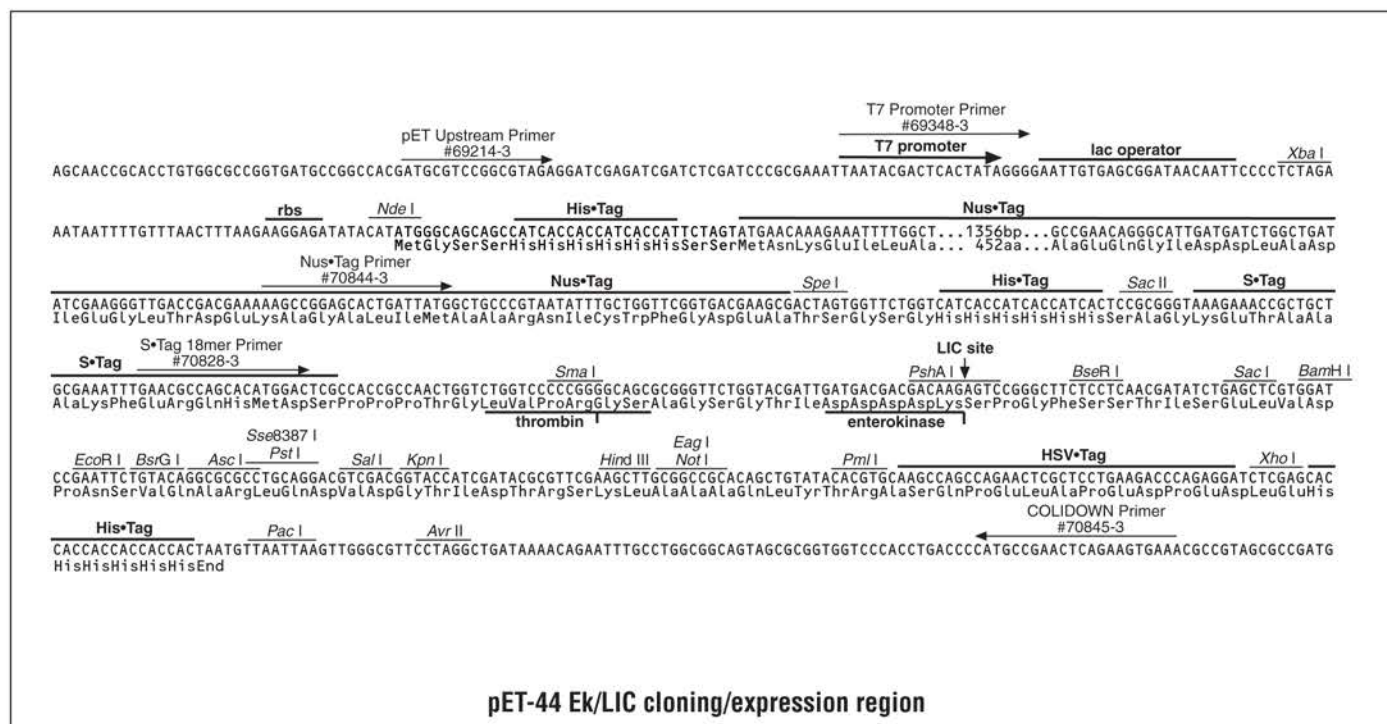
Top DNA Fragment (Synthetic):

AspAspAspAspLysMet ^{ek} insert stop
 GATGACGACGACAAGATG...sense...TAACCGGGCTTCTCTCTCA

Bottom DNA Fragment (Ekl/LIC vector):

CTACTGCTGCTTTCTAC...anti...ATTGGCCCGAAGAGAGAT

Result: plasmid recombinant



pET-44 Ek/LIC Restriction Sites

Enzyme	# Sites	Locations					Enzyme	# Sites	Locations					Enzyme	# Sites	Locations				
AatII	2	633	2208				DpnI	42					Sau96I	20						
AccI	4	580	627	1549	4837		DraI	3	6099	6791	6810		Scal	1	6196					
Acil	9						DraIII	1	7089				ScrFI	33						
AflIII	4	575	608	3203	5067		DrdI	3	4760	5175	7044		SfaNI	23						
AhdI	1	6677					Dsal	4	814	984	1423	2640	Sfcl	7	635	2314	2445	5332	5523	
AluI	27						EaeI	8	592	991	1093	1380	2511		6431	7308				
Alw26I	8	371	2900	3305	3431	3818			2643	3877	6284		SgrAI	2	1919	2522				
		4708	5834	6611			EagI	1	592				SmaI	1	741					
AlwI	20						EarI	3	2821	4951	5878		SpeI	1	848					
AlwNI	1	5483					Ecl136II	1	670				SphI	1	2678					
ApaI	1	3414					Eco47III	2	2608	4321			Sse8387I	1	639					
ApalI	4	3183	4881	5381	6005		Eco57I	4	532	1846	5615	6011	Sspl	2	877	6881				
ApoI	7	466	654	788	2323	3478	EcoNI	1	2738				StuI	1	1148					
		6891	6902				EcoO109I	3	53	2636	4073		StyI	4	57	483	1423	2119		
AscI	1	641					EcoRI	1	654				TaiI	25						
AvaI	2	530	739				EcoRII	13					TaqI	28						
Avall	7	440	745	3755	4073	4352	EcoRV	3	677	931	1522		TfiI	4	3882	4117	4621	5042		
		6313	6535				EheI	4	2527	2548	2662	3844	ThaI	52						
AvrII	1	483					FauI	20					TseI	30						
BamHI	1	660					Fnu4HI	54					Tsp45I	10	859	1745	2132	3384	4506	
BanI	10	620	1470	2525	2546	2660	FokI	14							4719	4814	6204	6415	7262	
		3123	3842	3972	6724	7126	FspI	1	6454				Tsp509I	23						
BanII	5	672	2587	2601	3414	7164	HaeII	15					TspRI	16						
BbsI	5	538	1228	3349	3688	4185	HaeIII	31					Tth111I	1	4812					
BbvI	30						HgaI	15					VspI	4	2460	3888	3947	6502		
BcgI	3	3529	4644	6139			HhaI	59					XbaI	1	2411					
BclI	2	1953	3217				HincII	4	628	920	1315	3709	XcmI	3	3059	3575	3593			
BfaI	11						HindIII	1	599				XhoI	1	530					
BglI	1	6559					HinfI	16					XmnI	3	2271	4625	6077			
BglII	1	963					HpaI	1	3709				Enzymes that do not cut pET-44 Ek/LIC:							
BpmI	4	3041	3530	4594	6608		HphI	31					AflII		Bsu36I	FseI	MunI	NheI	NsiI	
Bpu10I	1	4173					KpnI	1	624				PinAI		PmeI	RsrII	SanDI	SexAI	SfiI	
Bpu1102I	2	80	1298				MaeIII	22					Sgfi		SnaBI	SrfI	SunI	Swal		
BsaAI	3	576	4819	7089			MbolI	25												
BsaBI	3	2476	2486	4264			MluI	2	608	3203										
BsaHI	9	200	630	2205	2526	2547	MnlI	29												
		2661	3160	3843	6137		MscI	1	1095											
BsaI	2	371	6611				MseI	33												
BsaJI	17						MslI	11												
BsaWI	8	2	1596	3522	4025	4256	MspA1I	13												
		5273	5420	6381			MspI	42												
BseRI	1	699					MwoI	47												
BsgI	4	1976	3054	3254	4227		NarI	4	2526	2547	2661	3843								
BsiEI	10	595	1135	1273	1809	2190	NciI	20												
		3988	4983	5407	6159	6308	NcoI	1	1423											
BsiHKAI	11						NdeI	1	2373											
BslI	26						NgoAIV	2	2513	7190										
BsmBI	2	3818	4708				NlaIII	28												
BsmFI	5	453	758	2664	4338	7304	NlaIV	25												
BsmI	1	1007					NotI	1	592											
Bsp1286I	15						NruI	3	1230	1655	2279									
BspEI	2	2	4256				NspI	4	2678	4412	4704	5071								
BspLU11I	1	5067					NspV	2	604	2265										
BspMI	1	1297					PacI	1	503											
BsrBI	6	249	1343	2432	5000	5834	PfIMI	2	778	2785										
		7233					PleI	12												
BsrDI	6	1093	1474	3250	3616	6443	PmlI	1	576											
		6617					PshAI	1	701											
BsrFI	9	1047	1403	1467	1919	2513	Psp1406I	6	237	2865	4392	6075	6448							
		2522	2889	6592	7190				6874											
BsrGI	1	648					Psp5II	1	4073											
BsrI	29						PstI	1	639											
BssHII	5	641	1567	1759	2248	3614	PvuI	3	1273	2190	6308									
BssSI	4	665	1639	5240	6008		PvuII	4	586	3803	3896	4658								
Bst1107I	2	581	4838				RcaI	4	2601	5787	5836	5868								
BstEII	1	3384					RsaI	8	622	650	721	976	2198							
BstXI	3	3005	3134	3257					3350	4873	6196									
BstYI	13						SacI	1	672											
CacBI	52						SacII	1	817											
Clal	5	615	1524	1602	2190	2480	Sall	1	626											
CviJI	114						SapI	1	4951											
Ddel	15						Sau3AI	42												