

pET-17b Vector

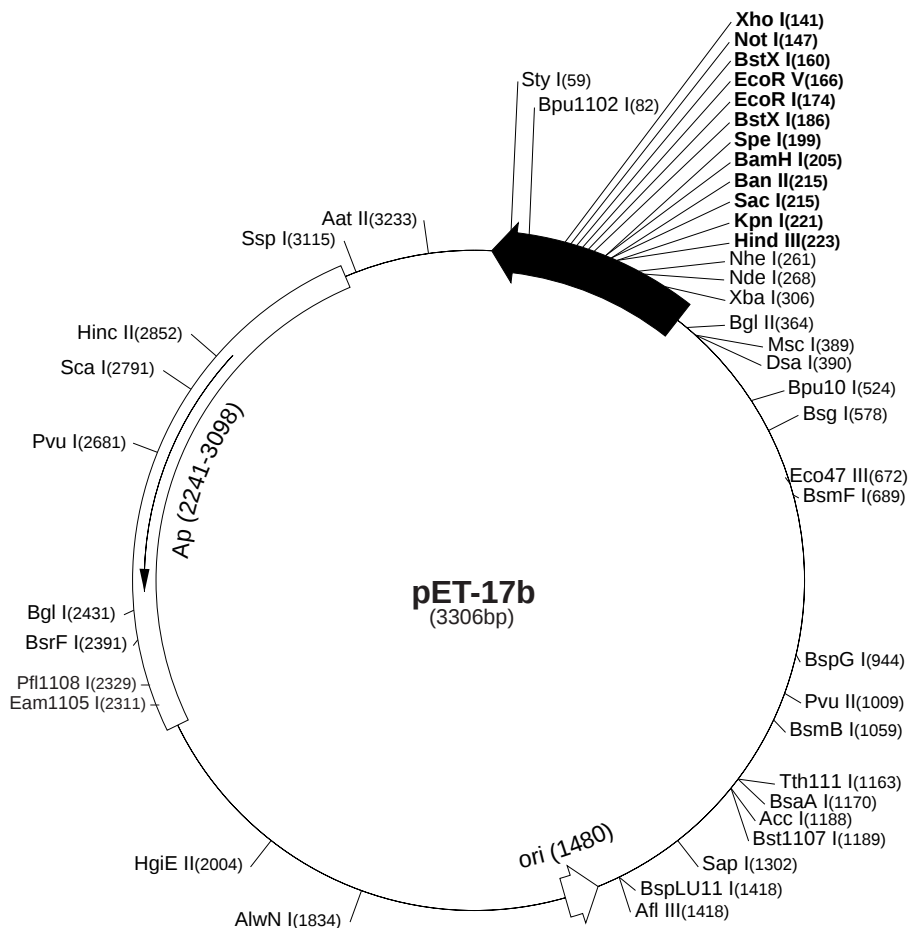
TB047 12/98

The pET-17b vector (Cat. No. 69663-3) carries an N-terminal 11aa T7•Tag[®] sequence followed by a region of useful cloning sites. Included in the multiple cloning region are dual *Bst*X I sites, which allow efficient cloning using an asymmetric linker (1). Unique sites (except for the two *Bst*X I 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.

1. Seed, B. (1987) *Nature* **329**, 840.

pET-17b sequence landmarks

T7 promoter	333-349
T7 transcription start	332
T7•Tag coding sequence	237-269
Multiple cloning sites	
(<i>Hind</i> III - <i>Xho</i> I)	141-228
T7 terminator	28-74
pBR322 origin	1480
<i>bla</i> coding sequence	2241-3098



T7 promoter primer #69348-3

Bgl II T7 promoter T7•Tag

AGATCTCGATCCCGGAAATTAATACGACTCCTATAGGGAGACCACACGGTTTCCCTCTAGAAATAATTTGTTTAACTTTAAGAAGGAGA

Nde I *Nhe* I *Hind* III *Kpn* I *Sac* I *Bam* HI *Spe* I *Bst*X I *Eco*R I *Eco*R V *Bst*X I *Not* I *Xho* I

TATACATATGGCTAGCATGACTGGTGGACAGCAATGGGTCCGGATTCAAGCTTGGTACCGAGCTCGGATCCACTAGTAACGGCCGCGCAGTGTGCTGGAATTCGAGATATCCATCACACTGGCGGCGCTCGAG

MetAlaSerMetThrGlyGlyGlnGlnMetGlyArgAspSerSerLeuValProSerSerAspProLeuValThrAlaAlaSerValLeuGluPheCysArgTyrProSerHisTrpArgProLeuGlu

*Bpu*1102 I T7 terminator

CAGATCCGCTGCTAACAAGCCGGAAGGAAAGCTGAGTTGGCTGCTGCCACCGCTGAGCAATAACTAGCATAACCCCTTGGGGCCTCTAAACGGGCTTGGAGGGGTTTTTGG

GlnIleArgLeuLeuThrLysProGluArgLysLeuSerTrpLeuLeuProProLeuSerAsnAsnEnd

T7 terminator primer #69337-3

pET-17b cloning/expression region

pET-17b Restriction Sites

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Enzyme	# Sites	Locations	Enzyme	# Sites	Locations	Enzyme	# Sites	Locations
AatII	1	3233	DpnI	19		SalI	1	2791
AccI	1	1188	DraI	3	2177 2196 2888	ScrFI	11	
AceIII	4	927 1068 1370 2610	DrdI	2	1111 1526	SfaNI	13	
Acil	44		Dsal	1	390	Sfcl	5	169 332 1683 1874 2552
AflIII	1	1418	EaeI	4	147 191 387 2699	SpeI	1	199
AluI	16		EagI	2	147 191	SspI	1	3115
AlwI	13		Eam1105I	1	2311	StyI	1	59
Alw21I	6	215 412 1236 1736 2897	EarI	2	1302 3106	TaqI	4	142 361 1518 2962
		2982	Ecil	3	1492 1638 2466	TaqII	5	1320 2659 2844 2997 3014
Alw44I	3	1232 1732 2978	Eco47III	1	672	TfiI	4	229 468 972 1393
AlwNI	1	1834	Eco57I	2	1966 2978	ThaI	12	
ApoI	1	174	EcoO109I	4	55 382 424 3287	TseI	20	
AvaI	2	141 368	EcoRI	1	174	Tsp45I	5	857 1070 1165 2567 2778
Avall	5	382 424 703 2449 2671	EcoRII	4	384 1444 1565 1578	Tsp509I	7	174 298 348 2178 2484
BamHI	1	205	EcoRV	1	166			2739 3304
BanI	2	217 2259	FauI	6	348 433 714 900 1121			
BanII	1	215			1131	Tth111I	1	1163
BbsI	2	536 3289	FokI	8	637 699 777 963 1104	Tth111II	4	879 2008 2015 2047
BbvI	20				2277 2458 2745	UbaII	11	
BccI	4	162 2348 2472 2759	FspI	2	399 2533	VspI	2	347 2483
Bce83I	5	23 1509 1807 2048 2916	GdiII	3	147 191 2699	XbaI	1	306
BceII	2	179 1920	HaeI	4	389 1433 1444 1896	XhoI	1	141
BcgI	6	224 258 995 1029 2816	HaeII	4	591 674 1296 1666	XmnI	2	976 2910
		2850	HaeIII	13				
Bfal	8	72 200 262 307 432	HgaI	5	954 1111 1529 2107 2837	Enzymes that do not cut pET-17b:		
		1913 2166 2501	HgiEI	1	2004	AflII	Agel	Apal
BglI	1	2431	HhaI	17		AvrII	Bael	BclI
BglII	1	364	HinAI	3	210 2310 2384	BseRI	BsmI	BspMI
BpmI	2	945 2381	HincII	1	2852	BstEII	Bsu36I	Clal
Bpu10I	1	524	HindIII	1	223	EcoNI	FseI	Hpal
Bpu1102I	1	82	HinfI	8	229 339 468 972 1318	NarI	NcoI	NgoAIV
BsaI	2	331 2372			1393 1789 2306	NspV	PacI	PflMI
BsaAI	1	1170	HphI	8	463 1038 1047 2154 2381	PshAI	RleAI	RsrII
BsaBI	3	164 363 615			2797 3003 3038	SexAI	SfiI	SgfI
BsaHI	2	2848 3230	KpnI	1	221	SnaBI	SphI	SrfI
BsaJI	3	59 390 1578	Maell	8	489 513 743 1169 2121	SunI	Swal	XcmI
BsaWI	5	4 607 1624 1771 2602			2537 2910 3230			
Bsbl	2	1134 2854	MaeIII	14				
BscGI	9	46 394 779 1112 1745	MbolI	8	536 1289 2080 2151 2906			
		2091 2312 2336 2858			2984 3093 3289			
BsgI	1	578	MmeI	2	1633 1817			
BsiI	3	1591 2975 3282	MnlI	19				
BsiEI	6	150 194 1334 1758 2681	MscI	1	389			
		2830	MseI	16				
BsII	10	52 391 754 845 887	MslI	6	405 600 991 2563 2722			
		1067 1440 1458 1624 1903			3081			
BsmAI	4	331 1059 2372 3148	MspI	16				
BsmBI	1	1059	MspAII	6	86 1009 1128 1760 2005			
BsmFI	1	689			2946			
BsoFI	31		MwoI	15				
Bsp24I	8	231 263 1911 1943 2089	NciI	7	428 756 1062 1097 1798			
		2121 3215 3247			2494 2845			
Bsp1286I	6	215 412 1236 1736 2897	NdeI	1	268			
		2982	NheI	1	261			
BspEI	2	4 607	NlaIII	16				
BspGI	1	944	NlaIV	13				
BspLU11I	1	1418	NottI	1	147			
BsrI	16		Nspl	3	763 1055 1422			
BsrBI	3	146 1351 3152	Pfl1108I	1	2329			
BsrDI	2	2372 2546	PleI	4	347 1312 1797 2300			
BsrFI	1	2391	Psp5II	2	382 424			
Bst1107I	1	1189	Psp1406I	3	743 2537 2910			
BstXI	2	160 186	PstI	2	173 2556			
BstYI	10	134 205 364 610 2059	PvuI	1	2681			
		2070 2156 2168 2936 2953	PvuII	1	1009			
Cac8I	12		RcaI	3	2138 3146 3251			
CjeI	14		RsaI	3	219 1224 2791			
CjePI	14		SacI	1	215			
CviJI	52		SapI	1	1302			
CviRI	14		Sau96I	10	55 382 424 703 890			
Ddel	10	82 103 524 686 1226			2353 2432 2449 2671 3287			
		1693 2102 2268 2808 3234	Sau3AI	19				