

pTriEx-3 Neo Vector

Baculovirus Locus	polh
Promoters	CMV immediate early p10 T7lac
C-terminal fusion options	HSV•Tag His•Tag
Cloning options	polylinker
Selectable marker (vertebrate cells)	Neomycin ^R

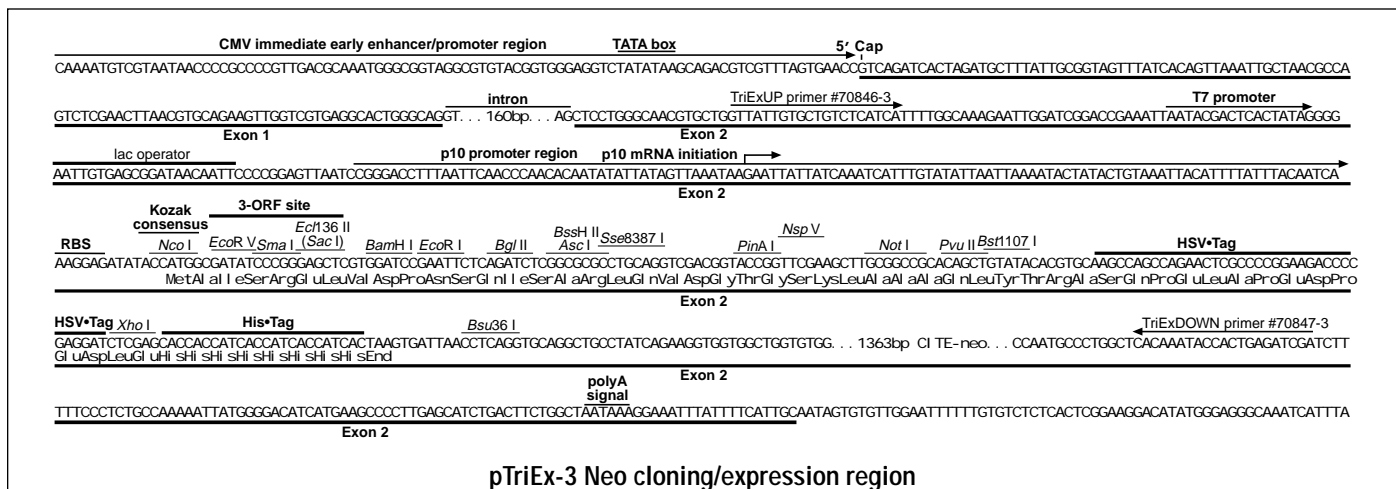
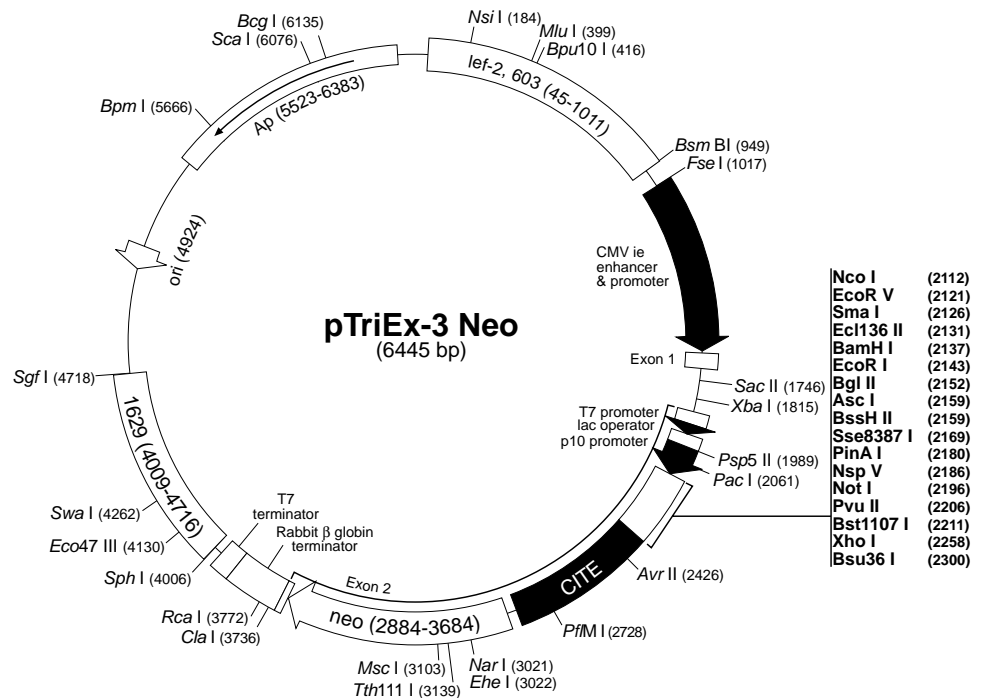
The pTriEx™-3 Neo vector¹ (Cat. No. 70931-3) is designed to allow rapid characterization of target genes in *E. coli*, insect and vertebrate cells, and to allow rapid selection of stable transfected vertebrate cells expressing high levels of target gene. Expression in vertebrate cells is mediated by the CMV immediate early enhancer and promoter². The drug selection marker is expressed under the control of the EMC virus Cap-Independent Translation Enhancer (CITE) sequence (or IRES), allowing rapid selection of transfected vertebrate cells using the drug G 418 or neomycin sulfate. For expression in insect cells, pTriEx-3 Neo contains flanking baculovirus sequences to permit the generation of recombinant baculoviruses using the BacVector® System. In baculovirus-infected insect cells, expression is driven by the very late p10 promoter. Expression in *E. coli* is regulated by the tightly controlled T7lac promoter. Expression can be induced in hosts such as NovaBlue by infecting with λCE6, a phage that constitutively expresses T7 RNA polymerase from the λp_L and λp_L promoters. Alternatively, pTriEx recombinant plasmids can be transferred into a (DE3)pLacI host that allows induction with IPTG.

¹ patent pending

² The CMV promoter is covered by U.S. Patent nos. 5,168,062 and 5,385,839 issued to the University of Iowa Research Foundation and is licensed for research use only.

pTriEx-3 Neo sequence landmarks

CMV ie enhancer/promoter	1021–1597
Vertebrate transcription start	1598
T7 promoter	1931–1947
T7 transcription start	1948
lac operator	1952–1972
p10 promoter region	1986–2099
p10 transcription start	2030–2031
Multiple cloning sites (Nco I–Bsu36 I)	2112–2300
HSV•Tag® coding sequence	2222–2257
His•Tag® coding sequence	2264–2287
CITE sequence	2349–2848
neo	2884–3684
Rabbit globin terminator region	3705–3947
T7 terminator	3948–3995
pUC origin	4924
bla coding sequence	5523–6383



pTriEx-3 Neo cloning/expression region

pTriEx-3 Neo Restriction Sites

Enzyme	# Sites	Locations	Enzyme	# Sites	Locations	Enzyme	# Sites	Locations		
AatII	6	1140 1193 1276 1462 1583 4453	DraI	4	429 3892 4262 6173	RsrII	2	1921 3537		
AccI	3	244 2172 2210	DraIII	2	2290 2638	SacI	1	2133		
AcI	59		DrdI	2	3048 4969	SacII	1	1746		
AflIII	7	399 2213 2590 2765 4523 4673 4862	DsaI	3	1743 2112 2809	SalI	2	243 2171		
AhdI	2	499 5596	EaeI	9	1011 2196 2578 2891 2927 3101 3492 3519 5984	SapI	2	3365 3575		
AluI	21		EagI	2	2196 2927	Sau3AI	29			
Alw26I	8	285 949 1449 1659 1897 3858 5657 6433	EarI	5	51 547 3365 3575 6391	Sau96I	18			
AlwI	16		Ecl136II	1	2131	Scal	1	6076		
AlwNI	2	1865 5277	Eco47III	1	4130	ScrFI	22			
Apal	2	1712 2392	Eco57I	5	2720 3167 3599 5409 6263	SfaNI	16			
ApaLI	3	2752 5175 6263	EcoO109I	6	1709 1989 2388 2744 2798 3964	Sfcl	9	1857 1943 2165 2523 2873 3070 5126 5317 5837		
ApoI	11		EcoRI	1	2143	Sgfl	1	4718		
Ascl	1	2159	EcoRII	11		SmaI	1	2126		
Aval	4	1732 2124 2249 2258	EcoRV	1	2121	SnaBI	2	1355 4172		
Avall	6	1921 1989 3537 4081 5734 5956	EheI	1	3022	SphI	1	4006		
AvrII	1	2426	FauI	9	1108 1134 1301 1529 1712 1738 1775 2868 3144	Sse8387I	1	2169		
BamHI	1	2137	Fnu4HI	42		SspI	4	425 4266 4463 6400		
BanI	7	1480 2177 2568 2716 3020 3055 5544	FokI	6	2717 3345 3370 5562 5743 6030	StyI	3	2112 2426 3959		
BanII	4	1712 2133 2392 3386	FseI	1	1017	Swal	1	4262		
BbsI	5	498 2250 2401 2515 4631	FspI	3	659 3123 5818	Tal	25			
BbvI	22		HaeII	3	3024 4132 5109	TaqI	18			
Bcgl	1	6135	HaeIII	25		TfiI	5	446 3506 3640 4135 4837		
Bfal	12		Hgal	9	146 503 966 1541 4072 4232 4658 4972 6122	Thal	16			
BglI	5	1105 1227 1298 2577 5716	Hhal	23		TseI	22			
BglII	1	2151	HincII	3	245 1531 2173	Tsp45I	5	1802 3141 3447 5852 6063		
Bpml	1	5666	HindIII	3	2189 2499 2847	Tsp509I	41			
Bpu10I	1	416	Hinfl	13		TspRI	14			
BsaAI	5	1355 2216 2591 3325 4172	HphI	10	183 879 2264 2270 2665 3199 5666 6082 6288 6323	Tth111I	1	3139		
BsaHI	10	495 1137 1190 1273 1459 1580 3021 4450 4650 6133	KpnI	2	2181 2720	VspI	5	1022 1930 2057 4657 5768		
Bsal	2	285 5657	MaeIII	15		XbaI	1	1815		
BsaJI	15		MboII	20		XhoI	1	2258		
BsaWI	7	833 2180 2346 3052 5067 5214 5887	MluI	1	399	XmnI	2	2487 6195		
BsgI	2	1689 2326	MnlI	34		Enzymes that do not cut pTriEx-3 Neo:				
BsiEI	7	2199 2930 4718 4778 5201 5966 6115	MscI	1	3103	AflIII	BclI	Bpu1102I	BsaBI	BseRI
BsiHKAI	8	2133 2265 2756 3134 3324 5179 6182 6267	MseI	38		BspEI	BstEII	EcoNI	HpaI	NheI
BsII	17		MslI	8	950 1380 1837 3458 3901 5848 6007 6366	NruI	PmeI	PshAI	SanDI	SexAI
BsmBI	1	949	MspA1I	7	655 1745 2206 2545 5203 5448 6231	SfiI	SgrAI	SpeI	SrfI	StuI
BsmFI	10	1190 1341 1509 1764 1783 2002 2828 3172 3780 4578	MspI	30		SunI	XcmII			
Bsml	2	2421 2460	MunI	2	4161 4506					
Bsp1286I	13		Mwol	29						
BspLU11I	2	2765 4862	NarI	1	3021					
BspMI	5	1686 2158 2614 2908 3289	NciI	11						
BsrBI	4	1728 1960 3634 4795	NcoI	1	2112					
BsrDI	6	79 2386 3254 3823 5657 5831	NdeI	4	1249 3872 3932 3940					
BsrFI	8	42 781 1013 1806 2180 3340 3521 5676	NgoAIV	3	781 1013 3521					
BsrGI	3	49 768 4502	NlaIII	19						
Bsrl	16		NlaIV	20						
BssHII	1	2159	NotI	1	2196					
BssSI	4	2132 3613 5034 6260	Nsil	1	184					
Bst1107I	1	2211	NspI	4	2760 2769 4006 4866					
BstXI	2	167 3712	NspV	1	2186					
BstYI	9	2137 2151 2254 2732 3192 5502 5513 6221 6238	Pacl	1	2061					
Bsu36I	1	2300	PfIMI	1	2728					
CacBI	28		PinAI	1	2180					
Clal	1	3736	PleI	8	150 1419 1931 2669 3444 4505 5240 5585					
CviJI	100		PmII	2	2216 2591					
Ddel	9	416 2148 2287 2300 3684 3730 5136 5553 6093	Psp1406I	2	5822 6195					
Dpnl	29		Psp5II	1	1989					
			PstI	3	2169 2877 3074					
			PvuI	2	4718 5966					
			PvuII	1	2206					
			Rcal	1	3772					
			Rsal	17						