

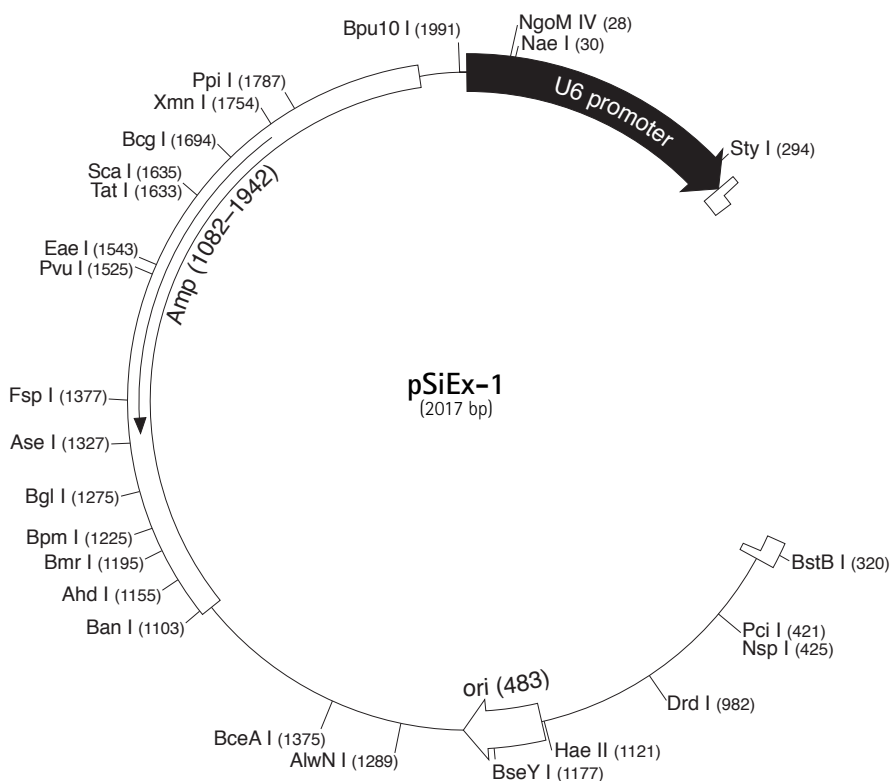
pSiEx™-1 siRNA Expression Vector (linearized vector)

TB409 0304

	Cat No.
pSiEx-1 Cloning Kit	71407-3
pSiEx-1 sequence landmarks	
U6 promoter	1-313
U6 transcription start	314
pUC ori	483
<i>bla</i> (Amp ^R)	1082-1942

The pSiEx™-1 vector is designed for DNA-directed gene silencing by the expression of small interfering RNA (siRNA) driven by a mouse U6 RNA polymerase III promoter. The use of a Pol III promoter, and a stop signal of five consecutive T residues, provides a mechanism to produce an siRNA with a defined 3'-terminus (1, 2). The pSiEx-1 Cloning Kit includes a linearized form of the vector (shown below) to facilitate directional cloning. Oligonucleotides encoding siRNA sequence with appropriate overhangs can be readily ligated into the vector and used to transform *E. coli* to generate plasmids for siRNA studies.

1. Yu, J., DeRuiter, S. L., and Turner, D. L. (2002) *Proc. Natl. Acad. Sci. USA* 9, 6505-6525.
2. Paddison, P. J., Caudy, A. A., Bernstein, E., Hannon, G. J., and Conklin, D. S. (2002) *Genes Dev.* 16, 948-958.



Mouse U6 promoter Sty I

GATACAAATACTAAATTATATTTTAAAAACAGCACA AAAAGGAAACTCACCCTAACGTAAAGTAATTGTGTGTTTTGAGACTATAAATATCCCTTGGA
 CTATGTTTATGATTTAATAAATAAAATTTTTGTCGTCGTTTTCTTTGAGTGGGATTGACATTTTCATTAACACACAAAACCTCGATATTTATAGGGAACCT

transcription start →

GAAAAGCCTTGT BstB I TATCCACAGAATCAGGGGATAACGCAGGAAAGAACATGTGAGCAAAGGCCAGCAAAGGCCAGGAAACCGTAAAAAGCCGCGTTGCTGGCGTTTTTCCA
 CTTTTCGGAACAAAC AAGCTTGCAGCGCAGCCAGCAAGCCGACGCGCTCGCCATAGTCGAGTGAGTTTCCGCATTATGCCA

Pci I
Nsp I

TATCCACAGAATCAGGGGATAACGCAGGAAAGAACATGTGAGCAAAGGCCAGCAAAGGCCAGGAAACCGTAAAAAGCCGCGTTGCTGGCGTTTTTCCA
 ATAGGTGCTTAGTCCCTATTGCGTCCTTTCTTGACACTCGTTTTCCGGTCGTTTTCCGGTCCTTGCCATTTTTCCGGCGCAACGACCGCAAAAAGGT

Drd I

TAGGCTCCGCCCCCTGACGAGCATCACAAAATCGACGCTCAAGTCAGAGGTGGCGAAACCGACAGGACTATAAAGATACAGGCGTTTTCCCTGGAA
 ATCCGAGCCGGGGACTGCTCGTAGTGTTTTTAGCTGCGAGTTCAGTCTCCACCCTTTGGGCTGCTCGATATTTCTATGGTCCGCAAAGGGGACCTT

← SiExDOWN primer

pSiEx-1 cloning/expression regions

pSiEx™-1 Restriction Sites

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Enzyme	# Sites	Locations			
AclI	2	1381	1754		
AfillI	1	421			
AhdI	1	1155			
AlwNI	1	836			
ApaLI	2	734	1822		
Asel	1	1327			
BanI	1	1103			
BceAI	1	922			
BcgI	1	1694			
BciVI	2	623	1991		
BglI	1	1275			
Bme1580I	2	738	1826		
Bmrl	1	1195			
Bpml	1	1225			
Bpu10I	1	1991			
BpuEI	4	511	809	1050	1760
BsaHI	2	6	1692		
BsaWI	3	626	773	1446	
BseYI	1	724			
BsiEI	4	337	760	1525	1674
BsiHKAI	3	738	1741	1826	
BsmAI	2	273	1991		
Bsp1286I	3	738	1741	1826	
BspCNI	4	708	1125	1644	1983
BspLU11I	1	421			
BsrBI	2	354	1995		
BsrDI	2	1216	1390		
BsrFI	2	28	1235		
BssSI	2	593	1819		
BstBI	1	320			
BstYI	5	1061	1072	1780	1797 2012
BtsI	2	1555	1575		
DraI	2	225	1732		
DrdI	1	528			
EaeI	1	1543			
Ecil	3	483	628	1298	
Eco57I	2	968	1822		
Eco57MI	3	968	1225	1822	
FspI	1	1377			
HaeII	1	668			
MslI	3	1407	1566	1925	
NaeI	1	30			
NgoMIV	1	28			
NspI	1	425			
NspV	1	320			
Pcil	1	421			
Ppil	1	1787			
PvuI	1	1525			
Scal	1	1635			
Sfcl	4	110	685	876	1396
SmlI	4	526	788	1065	1775
Sspl	2	97	1958		
Styl	1	294			
TaqII	3	323	1503	1688	
TatI	1	1633			
TspGWI	2	1595	1937		
XmnI	1	1754			

Enzymes that do not cut pSiEx-1:					
AarI	AatII	Acc65I	AccI	AfeI	AfilI
AgeI	AleI	Alol	Apal	AscI	AsiSI
AvaI	AvrII	BaeI	BamHI	BanII	BbeI
BbsI	BbvCI	BclI	BfrBI	BglIII	BlpI
BmgBI	BmtI	BplI	BsaAI	BsaBI	BsaI
BsaXI	BseRI	BsgI	BsiWI	BsmBI	BsmFI
BsmI	BspEI	BspHI	BspMI	BsrGI	BssHII
Bst1107I	BstAPI	BstEII	BstXI	BstZ17I	Bsu36I
BtgI	BtrI	Clal	DrallI	EagI	EarI
Ecl136III	EcoCRI	EcoNI	EcoO109I	EcoRI	EcoRV
FalI	FseI	FspAI	HincII	HindIII	HpaI
KasI	KpnI	MfeI	MluI	MscI	NarI
NcoI	NdeI	NheI	NotI	NruI	NsiI
Pacl	PfIMI	PfoI	PinAI	PmeI	PmlI
PpuMI	PshAI	PsiI	PspOMI	PsrI	PstI
PvuII	RsrII	SacI	SacII	SalI	SanDI
SapI	SbfI	SexAI	SfiI	SfoI	SgrAI
SmaI	SnaBI	SpeI	SphI	SrfI	
Sse8387I	StuI	Swal	Tth111I	XbaI	XcmI
XhoI	XmaI	ZraI			