Technical Bulletin

pSP72 Vector

INSTRUCTIONS FOR USE OF PRODUCT P2191.

www.promega.com

PRINTED IN USA.
Revised 9/06

AF9TB040 0906TB040

Part# TB040

pSP72 Vector

All technical literature is available on the Internet at: www.promega.com/tbs/
Please visit the web site to verify that you are using the most current version of this Technical Bulletin. Please contact Promega Technical Services if you have questions on use of this system. E-mail: techserv@promega.com

I. Description

The pSP72 Vector (1) can be used as a standard cloning vector and can also be used for transcription of RNA in vitro. The pSP72 Vector contains the SP6 and T7 RNA polymerase promoters flanking a unique multiple cloning region, which includes restriction sites for XhoI, PvuII, HindIII, SphI, PstI, SalI, AccI, XbaI, BamHI, SmaI, KpnI, SacI, EcoRI, ClaI, EcoRV and BglII.

The sequences of Promega vectors are available online at: **www.promega.com/vectors/** and are also available from the GenBank® database.

II. Product Components and Storage Conditions

Product	Size	Cat.#
pSP72 Vector	20µg	P2191

Storage Conditions: Store the pSP72 Vector at –20°C.

III. pSP72 Vector Multiple Cloning Region and Circle Map

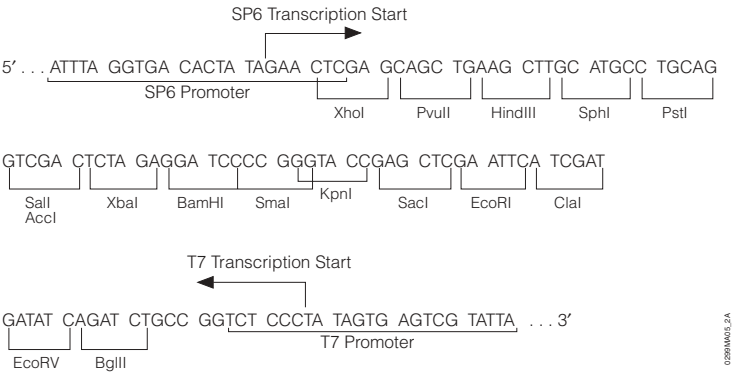

Figure 1. pSP72 Vector promoter and multiple cloning region sequence. The sequence shown corresponds to RNA synthesized by SP6 RNA polymerase and is complementary to RNA synthesized by T7 RNA polymerase.

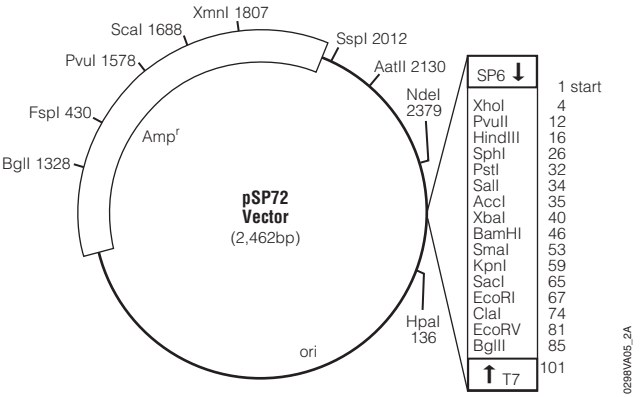

Figure 2. pSP72 Vector circle map and sequence reference points.

pSP72 Vector sequence reference points.

SP6 RNA polymerase transcription initiation site	1
T7 RNA polymerase transcription initiation site	101
SP6 RNA polymerase promoter (–17 to +3)	2446–3
T7 RNA polymerase promoter (–17 to +3)	99–118
multiple cloning region	4–90
β-lactamase coding region	1135–1995

The pSP72 and pSP73 Vectors are identical except for the orientation of the multiple cloning region.

Note: Blue/white screening of recombinants is **not** possible with the pSP72 Vector.

Specialized application of the pSP72 Vector.

- Transcription in vitro from dual-opposed promoters (For protocol information, please request the Riboprobe® in vitro Transcription Systems Technical Manual, #TM016) available at: **www.promega.com/tbs/**

IV. pSP72 Vector Restriction Sites

The following restriction enzyme tables were constructed using DNASTAR® sequence analysis software. Please note that we have not verified this information by restriction digestion with each enzyme listed. The location given specifies the 3´ end of the cut DNA (the base to the left of the cut site). For more information on the cut sites of these enzymes, or if you identify a discrepancy, please contact your local Promega Branch or Distributor. In the U.S., contact Promega Technical Services at 800-356-9526. Vector sequences are also available in the GenBank® database (GenBank®/EMBL Accession Number **X65332**) and on the Internet at: **www.promega.com/vectors/**

Table 1. Restriction Enzymes That Cut the pSP72 Vector Between 1 and 5 Times.

Enzyme	# of Sites	Location
AatII	1	2130
AccI	1	35
Acc65I	1	55
AcyI	2	1745, 2127
AflIII	1	315
Alw26I	5	100, 1269, 2045, 2198, 2240
Alw44I	3	629, 1875, 2372
AlwNI	1	731
AspHI	5	65, 633, 1794, 1879, 2376
AvaI	2	4, 51
AvaII	2	1346, 1568
BamHI	1	46
BanI	2	55, 1156
BanII	1	65
BbuI	1	26
BglI	1	1328
BglII	1	85
BsaI	2	100, 1269
BsaOI	4	231, 655, 1578, 1727
BsaHI	2	1745, 2127
BsaJI	3	50, 51, 475
Bsp1286I	5	65, 633, 1794, 1879, 2376
BspHI	3	1035, 2043, 2148
BspMI	2	21, 147
BssSI	3	488, 1872, 2179
BstOI	4	131, 343, 464, 477
Cfr10I	2	91, 1288
ClaI	1	74
DraI	3	1074, 1093, 1785
DraII	1	2184
DrdI	2	423, 2292
EaeI	2	154, 1596
EarI	2	199, 2003
EclHKI	2	1208, 2389
EcoICRI	1	63
EcoRI	1	67
EcoRV	1	81
FokI	4	1174, 1355, 1642, 2285
FspI	1	1430
HaeII	2	193, 563
HgaI	4	426, 1004, 1734, 2292
HincII	2	36, 136
HindII	2	36, 136
HindIII	1	16
HpaI	1	136
Hsp92I	2	1745, 2127
KpnI	1	59
MaeI	4	41, 810, 1063, 1398
MaeII	4	1018, 1434, 1807, 2127
MspA1I	5	12, 657, 902, 1843, 2309
NdeI	1	2379
NspI	3	26, 319, 2236
PaeR7I	1	4
PleI	5	31, 116, 209, 694, 1197

Table 1. Restriction Enzymes That Cut the pSP72 Vector Between 1 and 5 Times (continued).

Enzyme	# of Sites	Location	Enzyme	# of Sites	Location
PspAI	1	51	**SmaI**	1	53
PstI	1	32	**SphI**	1	26
PvuI	1	1578	Sse8387I	1	32
PvuII	1	12	**SspI**	1	2012
RsaI	3	57, 1688, 2364	TfiI	2	150, 290
SacI	1	65	**VspI**	4	116, 145, 1380, 2413
SalI	1	34			
Sau96I	5	1250, 1329, 1346, 1568, 2184	**XbaI**	1	40
			XhoI	1	4
ScaI	1	1688	**XmaI**	1	51
SinI	2	1346, 1568	**XmnI**	1	1807

Table 2. Restriction Enzymes That Do Not Cut the pSP72 Vector.

AccB7I	BsaBI	DraIII	**NcoI**	RsrII
AccIII	**BsaMI**	DsaI	**NgoMIV**	**SacII**
AflII	BsmI	EagI	**NheI**	**SfiI**
AgeI	Bsp120I	**Eco47III**	**NotI**	**SgfI**
ApaI	BsrBRI	Eco52I	**NruI**	SgrAI
AscI	BsrGI	Eco72I	**NsiI**	**SnaBI**
AvrII	**BssHII**	Eco81I	PacI	**SpeI**
BalI	Bst1107I	EcoNI	PflMI	SplI
BbeI	**Bst98I**	EheI	PinAI	SrfI
BbrPI	**BstEII**	FseI	PmeI	**StuI**
BbsI	**BstXI**	**I-PpoI**	PmlI	**StyI**
BclI	**BstZI**	KasI	Ppu10I	SwaI
BlpI	**Bsu36I**	**MluI**	PpuMI	**Tth111I**
Bpu1102I	**CspI**	**NaeI**	PshAI	XcmI
BsaAI	**Csp45I**	**NarI**	Psp5II	

Table 3. Restriction Enzymes That Cut the pSP72 Vector 6 or More Times.

AciI	**CfoI**	**HinfI**	MnlI	**Sau3AI**
AluI	**DdeI**	**HpaII**	MseI	ScrFI
BbvI	**DpnI**	HphI	**MspI**	SfaNI
BsrI	DpnII	**Hsp92II**	**NciI**	**TaqI**
BsrSI	Fnu4HI	MaeIII	**NdeII**	**Tru9I**
Bst71I	**HaeIII**	MboI	NlaIII	**XhoII**
BstUI	**HhaI**	**MboII**	NlaIV	

Note: The enzymes listed in boldface type are available from Promega.

V. Related Products

Product	Size	Cat.#
pSP64 Poly(A) Vector	20µg	P1241
pSP73 Vector	20µg	P2221

Product	Size	Cat.#
pGEM®-3Z Vector	20µg	P2151
pGEM®-4Z Vector	20µg	P2161
pGEM®-3Zf(+) Vector	20µg	P2271
pGEM®-3Zf(-) Vector	20µg	P2261
pGEM®-5Zf(+) Vector	20µg	P2241
pGEM®-5Zf(-) Vector	20µg	P2351
pGEM®-7Zf(+) Vector	20µg	P2251
pGEM®-7Zf(-) Vector	20µg	P2371
pGEM®-9Zf(-) Vector	20µg	P2391
pGEM®-11Zf(+) Vector	20µg	P2411
pGEM®-11Zf(-) Vector	20µg	P2421
pGEM®-13Zf(+) Vector	20µg	P2541

All pGEM® Vectors are provided with a glycerol stock of bacterial strain JM109. The JM109 cells do not contain vector and are not competent.

Sequencing Primers

Product	Size	Cat.#
SP6 Promoter Primer	2µg	Q5011
T7 Promoter Primer	2µg	Q5021

Riboprobe® in vitro Transcription Systems

Product	Cat.#
Riboprobe® System—SP6	P1420
Riboprobe® System—T7	P1440

For Laboratory Use.

VI. Reference

1. Krieg, P.A. and Melton, D.A. (1987) In vitro RNA synthesis with SP6 RNA polymerase. *Methods Enzymol.* **155**, 397–415.

Promega Corporation · 2800 Woods Hollow Road · Madison, WI 53711-5399 USA
Toll Free in USA 800-356-9526 · Phone 608-274-4330 · Fax 608-277-2516 · **www.promega.com**
Printed in USA. Part# TB040
Revised 9/06