

pRL-null Vector



Technical Bulletin No. 238

INSTRUCTIONS FOR USE OF PRODUCT E2271. PLEASE DISCARD PREVIOUS VERSIONS.

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I. Description

The pRL-null Vector^(a,b) (Figure 1) is intended for use in constructing a control reporter vector that may be used in combination with any experimental reporter vector to cotransfect mammalian cells. All of Promega's pRL Reporter Vectors contain a cDNA^(b) (*Rluc*) encoding *Renilla* luciferase, which was originally cloned from the marine organism *Renilla reniformis* (sea pansy; 1). As described below, the *Renilla* luciferase cDNA contained within the pRL Vectors has been modified slightly to provide greater utility.

The pRL-null Vector contains no enhancer or promoter elements. Rather, it contains a multiple cloning region upstream of *Rluc* to allow for the cloning of any desired regulatory element(s) to drive expression of *Renilla* luciferase. *Renilla* luciferase is a 36kDa monomeric protein that does not require post-translational modification for activity (2). Therefore, like firefly luciferase, the enzyme may function as a genetic reporter immediately following translation. For information about the use of this plasmid in conjunction with a reporter vector containing the firefly luciferase gene, refer to the Dual-Luciferase[®] Reporter Assay System^(c,d) Technical Manual (#TM040).

The pRL Vectors are isolated from a *dam*⁻/*dcm*⁻ *E. coli* K host strain, allowing digestion with restriction enzymes that are sensitive to *dam* and *dcm* methylation.

The GenBank[®]/EMBL Accession Number for the pRL-null Vector is AF025844.



II. Product Components

Product	Size	Cat.#
pRL-null Vector	20µg	E2271

All pRL Vectors are supplied in TE buffer (pH 7.4) and are provided with a glycerol stock of bacterial strain JM109. The JM109 cells do not contain vector and are not competent cells.

Storage Conditions: Store vector DNA at -20°C and the glycerol stock of JM109 cells at -70°C .

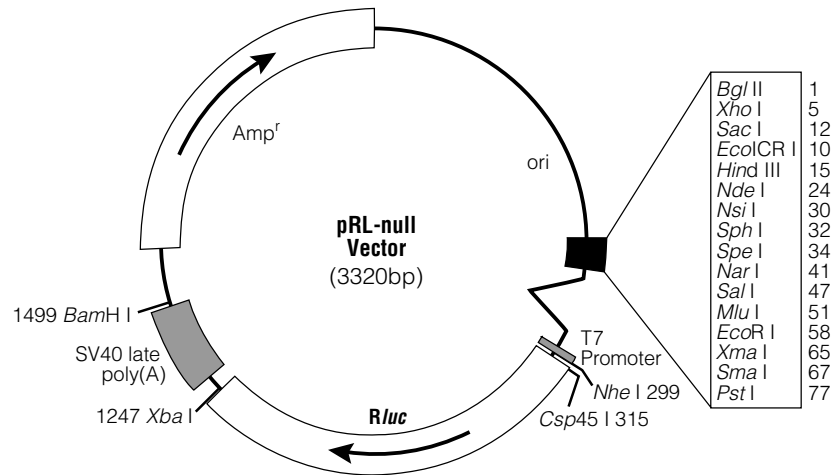


Figure 1. The pRL-null Vector circle map and sequence reference points.

Sequence reference points:

Multiple cloning region	1–78
Chimeric intron	101–237
T7 RNA polymerase promoter (–17 to +2)	281–299
T7 RNA polymerase promoter transcription initiation site	298
<i>R/luc</i> reporter gene	309–1244
SV40 late polyadenylation signal	1286–1487
β -lactamase (<i>Amp^r</i>) coding region	1634–2494

In addition:


-  indicates the position of the intron.
- *R/luc* is the cDNA encoding the *Renilla* luciferase enzyme.
- *Amp^r* indicates the gene encoding ampicillin resistance in *E. coli*.
- *ori* is the origin of replication in *E. coli*.
- The arrows within the *R/luc* and *Amp^r* genes indicate the direction of transcription.

Table 1. Restriction Enzyme Sites within the Multiple Cloning Region of the pRL-null Vector.

Enzyme Site	End Generated	Compatible Ends
<i>Bgl</i> I	5' overhang	<i>Bam</i> H I
<i>Xho</i> I	5' overhang	<i>Sal</i> I
<i>Eco</i> CR I	Blunt	<i>Sst</i> I
<i>Hind</i> III	5' overhang	None
<i>Nde</i> I	5' overhang	<i>Tru</i> 9 I, <i>Vsp</i> I
<i>Nsi</i> I	3' overhang	<i>Pst</i> I
<i>Sph</i> I	3' overhang	<i>Bbu</i> I, <i>Hsp</i> 92 II
<i>Spe</i> I	5' overhang	<i>Xba</i> I, <i>Nhe</i> I
<i>Nar</i> I	5' overhang	<i>Acy</i> I, <i>Cla</i> I, <i>Csp</i> 45 I, <i>Hpa</i> II, <i>Msp</i> I, <i>Taq</i> I
<i>Bbe</i> I	3' overhang	None
<i>Ehe</i> I	Blunt	Any blunt end
<i>Sal</i> I	5' overhang	<i>Xho</i> I
<i>Mlu</i> I	5' overhang	<i>Bss</i> H II
<i>Eco</i> R I	5' overhang	None
<i>Xma</i> I	5' overhang	<i>Acc</i> III
<i>Sma</i> I	Blunt	Any blunt end
<i>Pst</i> I	3' overhang	<i>Nsi</i> I

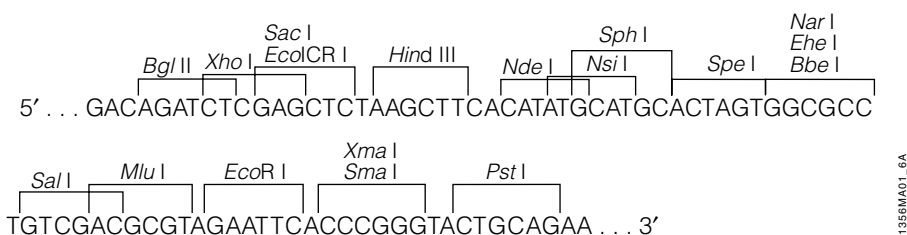


Figure 2. Multiple cloning region upstream of the intron and *Renilla* luciferase reporter gene in the pRL-null Vector.

III. Features of the pRL-null Vector

A. Multiple Cloning Region

The pRL-null Vector contains a multiple cloning region positioned immediately upstream of the chimeric intron and *Renilla* luciferase reporter gene (Figure 2). To aid in devising cloning strategies, Table 1 summarizes the types of DNA ends generated from restriction endonuclease digestion within the multiple cloning region as well as the compatibility of those ends with the ends of DNA fragments generated by heterologous restriction enzymes.

B. Chimeric Intron

Downstream of the multiple cloning region of the pRL-null Vector is a chimeric intron comprised of the 5'-donor splice site from the first intron of the human β -globin gene, and the branch and 3'-acceptor splice site from an intron preceding an immunoglobulin gene heavy chain variable region (3). The sequences of the donor and acceptor splice sites, along with the branchpoint site, have been modified to match the consensus sequences for optimal splicing (4).

Transfection studies have demonstrated that the presence of an intron flanking a cDNA insert frequently increases the level of gene expression (5–8). In the pRL-null Vector the intron is positioned 5' to *Rluc* to minimize the utilization of cryptic 5'-donor splice sites that may reside within the reporter gene sequence (9).

C. T7 Promoter

A T7 promoter is located downstream of the chimeric intron and immediately precedes the *Rluc* reporter gene. This T7 promoter can be used to synthesize RNA transcripts in vitro using T7 RNA Polymerase (Cat.# P2075). T7 RNA Polymerase can also be used to synthesize active *Renilla* luciferase in a cell-free coupled eukaryotic in vitro transcription/translation reaction (e.g., Promega's TNT® Reticulocyte Lysate(c,e,f,g) [Cat.# L4610], TNT® T7 Coupled Wheat Germ Extract(c,e,f,g) [Cat.# L4140] or TNT® T7 Quick Coupled Transcription/Translation (c,e,f,g,h) [Cat.# L1170] Systems).

D. *Renilla* Luciferase Reporter Gene (*Rluc*)

The *Renilla* luciferase cDNA inserted into all of the pRL Vectors is derived from the anthozoan coelenterate *Renilla reniformis* (1) but contains nucleotide changes that were engineered during the construction of the individual vectors. The following bases were altered in the pRL-null Vector: base 539 (T→C), to eliminate an internal *Bgl* II site; base 1082 (T→C), to eliminate an internal *Bam*H I site; base 1115 (C→T), to eliminate internal *Nar* I, *Kas* I, *Ban* I and *Acy* I sites. These nucleotide substitutions do not alter the amino acid sequence of the encoded *Renilla* luciferase reporter enzyme.

E. SV40 Late Polyadenylation Signal

Polyadenylation signals cause the termination of transcription by RNA polymerase II and signal the addition of approximately 200–250 adenosine residues to the 3'-end of the RNA transcript (10). Polyadenylation has been shown to enhance RNA stability and translation (11,12). The late SV40 polyadenylation signal, which is extremely efficient and has been shown to increase the steady-state level of RNA approximately 5-fold over the early SV40 polyadenylation signal (13), has been positioned 3' to the *Rluc* gene in the pRL-null Vector to increase the level of *Renilla* luciferase expression.



Due to sequence differences between the T7 Promoter Primer offered by Promega (Cat.# Q5021) and the T7 promoter used in the pRL family of *Renilla* luciferase co-reporter vectors, this primer cannot be used for sequencing with this vector.

IV. Transfection of Mammalian Cells with the pRL-null Vector

The pRL-null Vector, once it has been modified to contain appropriate genetic regulatory domains, may be used in combination with any experimental reporter vector to cotransfect mammalian cells. However, it is important to realize that trans effects between promoters on cotransfected plasmids can potentially affect reporter gene expression (14). Primarily this is of concern when either the control or experimental reporter vector, or both, contain very strong promoter/enhancer elements. The occurrence and magnitude of such effects will depend on several factors: i) the combination and activities of the genetic regulatory elements present on the cotransfected vectors; ii) the relative ratio of experimental vector to control vector introduced into the cells; and iii) the cell type transfected.

To help ensure independent genetic expression between experimental and control reporter genes, preliminary cotransfection experiments should be performed to optimize both the amount of vector DNA and the ratio of the coreporter vectors added to the transfection mixture. Similar to the firefly luciferase assay, the *Renilla* luciferase assay is extremely sensitive, providing accurate measurement of ≤ 10 femtograms of *Renilla* luciferase, with linearity over seven orders of enzyme concentration. Therefore, it is possible to use relatively small quantities of the pRL-null Vector to provide low-level, constitutive coexpression of *Renilla* luciferase control activity.

The pRL-null Vector, once genetic regulatory domains have been added, can be used for both transient and stable expression of *Renilla* luciferase. For stable expression, the pRL-null Vector must be cotransfected with an expression vector containing a selectable gene in mammalian cells. Transfection of DNA into mammalian cells may be mediated by cationic lipids (15,16), calcium phosphate (17,18), DEAE-Dextran (19–21), polybrene-DMSO (22,23), or electroporation (24,25).

Transfection systems based on cationic lipid compounds (TransFast™ Reagent⁽ⁱ⁾, Tfx™ Reagents⁽ⁱ⁾ and Transfectam® Reagent^(k)), calcium phosphate and DEAE-Dextran are available from Promega. For more information and a protocol for the Transfectam® Reagent, please request the *Transfectam® Reagent Technical Bulletin* (#TB116) and for the TransFast™ Reagent, please request the *TransFast™ Transfection Reagent Technical Bulletin* (#TB260). Protocols for the use of the Tfx™ Reagents can be found in the *Tfx™-10, Tfx™-20 and Tfx™-50 Reagents for the Transfection of Eukaryotic Cells Technical Bulletin* (#TB216). For transfection procedures using calcium phosphate or DEAE-Dextran, please request the *ProFection® Mammalian Transfection Systems Technical Manual* (#TM012).

V. pRL-null Vector Restriction Sites and Sequence

A. pRL-null 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 Office or Distributor. In the U.S., contact Promega Technical Services at 800-356-9526.

Table 2. Restriction Enzymes That Cut the pRL-null Vector Between 1 and 5 Times.

Enzyme	# of Sites	Location	Enzyme	# of Sites	Location
Acc I	1	48	Ecl HK I	1	2421
Acy I	2	41, 1881	Eco 52 I	1	1253
Afl II	1	264	Eco ICR I	1	10
Afl III	2	51, 491	Eco R I	1	5
Alw 26 I	4	129, 154, 1579, 2355	Ehe I	1	42
Alw 44 I	2	1749, 2995	Fsp I	1	2198
Alw N I	1	2900	Hae II	2	44, 3069
Asp H I	4	12, 1753, 1838, 2999	Hga I	4	59, 1889, 2619, 3197
Ava I	2	5, 65	Hinc II	2	49, 1396
Ava II	4	357, 1079, 2057, 2279	Hind II	2	49, 1396
Bam H I	1	1498	Hind III	1	15
Ban I	4	40, 190, 1113, 2468	Hpa I	1	1396
Ban II	1	12	Hsp 92 I	2	41, 1881
Bbe I	1	44	Kas I	1	40
Bbs I	2	175, 1149	Mae II	5	1003, 1040, 1819, 2192, 2608
Bbu I	1	32	Mlu I	1	51
Bcl I	2	593, 802	Msp A1 I	3	1785, 2726, 2971
Bgl I	1	2303	Nar I	1	41
Bgl II	1	1	Nci I	5	66, 67, 1885, 2236, 2932
Bsa I	2	129, 2355	Nde I	1	24
Bsa O I	4	1256, 1903, 2052, 2975	Nhe I	1	299
Bsa A I	1	1041	Not I	1	1253
Bsa B I	1	1497	Nsi I	1	30
Bsa H I	2	41, 1881	Nsp I	3	32, 435, 495
Bsa J I	3	65, 1082, 3149	Pae R7 I	2	157, 3315
Bsa M I	2	1317, 1410	Ple I	4	165, 281, 2430, 2933
Bsm I	2	1317, 1410	Ppu 10 I	1	26
Bsp 1286 I	4	12, 1753, 1838, 2999	Psp A I	1	65
Bsp H I	3	877, 1581, 2589	Pst I	1	77
Bsp M I	1	91	Pvu I	1	2052
Bsr BR I	1	1497	Rsa I	4	71, 277, 1009, 1940
Bsr G I	1	1007	Sac I	1	12
Bss S I	3	967, 1752, 3136	Sal I	1	47
Bst 98 I	1	264	Sca I	2	277, 1940
Bst O I	5	1084, 1503, 3150, 3163, 3284	Sin I	4	357, 1079, 2057, 2279
Bst Z I	1	1253	Sma I	1	67
Cfr 10 I	1	2336	Spe I	1	34
Cla I	1	1491	Sph I	1	32
Csp 45 I	1	315	Ssp I	1	1616
Dde I	5	13, 1920, 2460, 2626, 3035	Tfi I	5	399, 420, 759, 1074, 1087
Dra I	4	1457, 1843, 2535, 2554	Vsp I	2	409, 2246
Drd I	1	3207	Xba I	1	1246
Eae I	3	659, 1253, 2028	Xcm I	1	958
Eag I	1	1253	Xho I	1	5
Ear I	2	479, 1622	Xma I	1	65
			Xmn I	2	843, 1821

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

Table 3. Restriction Enzymes That Do Not Cut the pRL-null Vector.

<i>Aat</i> II	<i>Blp</i> I	<i>Dra</i> III	<i>Nae</i> I	<i>PshA</i> I	<i>Srf</i> I
<i>Acc</i> B7 I	<i>Bpu</i> 1102 I	<i>Dsa</i> I	<i>Nco</i> I	<i>Psp5</i> II	<i>Sse8387</i> I
<i>Acc</i> III	<i>Bsp</i> 120 I	<i>Eco47</i> III	<i>NgoM</i> IV	<i>Pvu</i> II	<i>Stu</i> I
<i>Acc65</i> I	<i>BssH</i> II	<i>Eco72</i> I	<i>Nru</i> I	<i>Rsr</i> II	<i>Sty</i> I
<i>Age</i> I	<i>Bst</i> 1107 I	<i>Eco81</i> I	<i>Pac</i> I	<i>Sac</i> II	<i>Swa</i> I
<i>Apa</i> I	<i>BstE</i> II	<i>EcoN</i> I	<i>PflM</i> I	<i>Sfi</i> I	<i>Tth111</i> I
<i>Asc</i> I	<i>BstX</i> I	<i>EcoR</i> V	<i>PinA</i> I	<i>Sgf</i> I (1)	
<i>Avr</i> II	<i>Bsu36</i> I	<i>Fse</i> I	<i>Pme</i> I	<i>SgrA</i> I	
<i>Bal</i> I	<i>Csp</i> I	<i>I-Ppo</i> I	<i>Pml</i> I	<i>SnaB</i> I	
<i>BbrP</i> I	<i>Dra</i> II	<i>Kpn</i> I	<i>PpuM</i> I	<i>Spl</i> I	

Table 4. Restriction Enzymes That Cut the pRL-null Vector 6 or More Times.

<i>Aci</i> I	<i>BstU</i> I	<i>Hae</i> III	<i>Mae</i> I	<i>Msp</i> I	<i>ScrF</i> I
<i>Alu</i> I	<i>Cfo</i> I	<i>Hha</i> I	<i>Mae</i> III	<i>Nde</i> II	<i>SfaN</i> I
<i>Bbv</i> I	<i>Dpn</i> I	<i>Hinf</i> I	<i>Mbo</i> I	<i>Nla</i> III	<i>Taq</i> I
<i>Bsr</i> I	<i>Dpn</i> II	<i>Hpa</i> II	<i>Mbo</i> II	<i>Nla</i> IV	<i>Tru9</i> I
<i>Bsr S</i> I	<i>Fnu4H</i> I	<i>Hph</i> I	<i>Mnl</i> I	<i>Sau3A</i> I	<i>Xho</i> II
<i>Bst71</i> I	<i>Fok</i> I	<i>Hsp92</i> II	<i>Mse</i> I	<i>Sau96</i> I	

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

B. pRL-null Vector Sequence

The sequence shown corresponds to the mRNA synthesized from the *Renilla* luciferase gene from a promoter positioned in the multiple cloning region. The vector sequence is also available on the Internet at www.promega.com/vectors/. The GenBank®/EMBL Accession number for the pRL-null Vector is AF025844.

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1  AGATCTCGAG CTCTAAGCTT CACATATGCA TGCCTAGTG GCGCCTGTCG
51  ACGCGTAGAA TTCACCCGGG TACTGCAGAA GTTGGTCGTG AGGCACTGGG
101 CAGGTAAGTA TCAAGTTAC AAGACAGGTT TAAGGAGACC AATAGAAACT
151 GGGCTTGTCT AGACAGAGAA GACTCTTGCG TTTCTGATAG GCACCTATTG
201 GTCTTACTGA CATCCACTTT GCCTTTCTCT CCACAGGTGT CCACTCCAG
251 TTCAATTACA GCTCTTAAGG CTAGAGTACT TAATACGACT CACTATAGGC
301 TAGCCACCAT GACTTCGAAA GTTTATGATC CAGAACAAAG GAAACGGATG
351 ATAAGTGGTC CGCAGTGGTG GGCCAGATGT AAACAAATGA ATGTTCTTGA
401 TTCATTTATT AATTATTATG ATTCAGAAAA ACATGCAGAA AATGCTGTTA
451 TTTTTTTTACA TGGTAACGCG GCCTCTTCTT ATTTATGGCG ACATGTTGTG
501 CCACATATTG AGCCAGTAGC GCGGTGTATT ATACCAGACC TTATTGGTAT
551 GGGCAAATCA GGCAAATCTG GTAATGGTTC TTATAGGTTA CTTGATCATT
601 ACAAATATCT TACTGCATGG TTTGAACTTC TTAATTTACC AAAGAAGATC
651 ATTTTTTGTCT GCCATGATTG GGGTGCTTGT TTGGCATTTC ATTATAGCTA
701 TGAGCATCAA GATAAGATCA AAGCAATAGT TCACGCTGAA AGTGTAGTAG
751 ATGTGATTGA ATCATGGGAT GAATGGCCTG ATATTGAAGA AGATATTGCG

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B. pRL-null Vector Sequence (continued)

801 TTGATCAAAT CTGAAGAAGG AGAAAAAATG GTTTTGGAGA ATAACTTCTT
851 CGTGGAAACC ATGTTGCCAT CAAAAATCAT GAGAAAGTTA GAACCAGAAG
901 AATTTGCAGC ATATCTTGAA CCATTCAAAG AGAAAGGTGA AGTTCGTCGT
951 CCAACATTAT CATGGCCTCG TGAAATCCCG TTAGTAAAAG GTGGTAAACC
1001 TGACGTTGTA CAAATTGTTA GGAATTATAA TGCTTATCTA CGTGCAAGTG
1051 ATGATTTACC AAAAATGTTT ATTGAATCGG ACCCAGGATT CTTTTCCAAT
1101 GCTATTGTTG AAGGTGCCAA GAAGTTTCCT AATACTGAAT TTGTCAAAGT
1151 AAAAGGTCTT CATTTTTTCGC AAGAAGATGC ACCTGATGAA ATGGGAAAAT
1201 ATATCAAATC GTTCGTTGAG CGAGTTCTCA AAAATGAACA ATAATTCTAG
1251 AGCGGCCGCT TCGAGCAGAC ATGATAAGAT ACATTGATGA GTTTGGACAA
1301 ACCACAATA GAATGCAGTG AAAAAAATGC TTTATTTGTG AAATTTGTGA
1351 TGCTATTGCT TTATTTGTAA CCATTATAAG CTGCAATAAA CAAGTTAACA
1401 ACAACAATTG CATTCAATTT ATGTTTCAGG TTCAGGGGGA GGTGTGGGAG
1451 GTTTTTTAAA GCAAGTAAAA CCTCTACAAA TGTGGTAAAA TCGATAAGGA
1501 TCCAGGTGGC ACTTTTCGGG GAAATGTGCG CGGAACCCCT ATTTGTTTAT
1551 TTTTCTAAAT ACATTCAAAT ATGTATCCGC TCATGAGACA ATAACCCTGA
1601 TAAATGCTTC AATAATATTG AAAAAGGAAG AGTATGAGTA TTCAACATTT
1651 CCGTGTCGCC CTTATTCCCT TTTTTCGGC ATTTTGCCTT CCTGTTTTTG
1701 CTCACCCAGA AACGCTGGTG AAAGTAAAAG ATGCTGAAGA TCAGTTGGGT
1751 GCACGAGTGG GTTACATCGA ACTGGATCTC AACAGCGGTA AGATCCTTGA
1801 GAGTTTTTCGC CCCGAAGAAC GTTTTCCAAT GATGAGCACT TTTAAAGTTC
1851 TGCTATGTGG CGCGGTATTA TCCCGTATTG ACGCCGGGCA AGAGCAACTC
1901 GGTCGCCGCA TACACTATTC TCAGAATGAC TTGGTTGAGT ACTCACCAGT
1951 CACAGAAAAG CATCTTACGG ATGGCATGAC AGTAAGAGAA TTATGCAGTG
2001 CTGCCATAAC CATGAGTGAT AACACTGCGG CCAACTTACT TCTGACAACG
2051 ATCGGAGGAC CGAAGGAGCT AACCGCTTTT TTGCACAACA TGGGGGATCA
2101 TGTAACTCGC CTTGATCGTT GGAACCGGA GCTGAATGAA GCCATACCAA
2151 ACGACGAGCG TGACACCACG ATGCCTGTAG CAATGGCAAC AACGTTGCGC
2201 AAACATATAA CTGGCGAACT ACTTACTCTA GCTTCCCGGC ACAATTAAT
2251 AGACTGGATG GAGGCGGATA AAGTTGCAGG ACCACTTCTG CGCTCGGCC
2301 TTCCGGCTGG CTGGTTTATT GCTGATAAAT CTGGAGCCGG TGAGCGTGGG
2351 TCTCGCGGTA TCATTGCAGC ACTGGGGCCA GATGGTAAGC CCTCCGTAT
2401 CGTAGTTATC TACACGACGG GGAGTCAGGC AACTATGGAT GAACGAAATA
2451 GACAGATCGC TGAGATAGGT GCCTCACTGA TTAAGCATTG GTAACGTCA
2501 GACCAAGTTT ACTCATATAT ACTTTAGATT GATTTAAAAC TTCATTTTTA
2551 ATTTAAAAGG ATCTAGGTGA AGATCCTTTT TGATAATCTC ATGACCAAAA
2601 TCCCTTAACG TGAGTTTTTCG TTTCACTGAG CGTCAGACCC CGTAGAAAAG
2651 ATCAAAGGAT CTTCTTGAGA TCCTTTTTTT CTGCGCGTAA TCTGCTGCTT

B. pRL-null Vector Sequence (continued)

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2701  GCAAACAAAA  AAACCACCGC  TACCAGCGGT  GGTTTGTTTG  CCGGATCAAG
2751  AGCTACCAAC  TCTTTTTCCG  AAGGTAAGTG  GCTTCAGCAG  AGCGCAGATA
2801  CCAAATACTG  TTCTTCTAGT  GTAGCCGTAG  TTAGGCCACC  ACTTCAAGAA
2851  CTCTGTAGCA  CCGCCTACAT  ACCTCGCTCT  GCTAATCCTG  TTACCAGTGG
2901  CTGCTGCCAG  TGGCGATAAG  TCGTGTCTTA  CCGGGTTGGA  CTCAAGACGA
2951  TAGTTACCGG  ATAAGGCGCA  GCGGTCGGGC  TGAACGGGGG  GTTCGTGCAC
3001  ACAGCCCAGC  TTGGAGCGAA  CGACCTACAC  CGAACTGAGA  TACCTACAGC
3051  GTGAGCTATG  AGAAAGCGCC  ACGCTTCCCG  AAGGGAGAAA  GGCGGACAGG
3101  TATCCGGTAA  GCGGCAGGGT  CGGAACAGGA  GAGCGCACGA  GGGAGCTTCC
3151  AGGGGGAAAC  GCCTGGTATC  TTTATAGTCC  TGTCGGGTTT  CGCCACCTCT
3201  GACTTGAGCG  TCGATTTTTG  TGATGCTCGT  CAGGGGGGCG  GAGCCTATGG
3251  AAAAACGCCA  GCAACGCGGC  CTTTTTACGG  TTCCTGGCCT  TTTGCTGGCC
3301  TTTTGCTCAC  ATGGCTCGAC

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VI. Related Products
pRL Family of *Renilla* Luciferase Vectors for Coreporter Applications

Product	Size	Cat.#
pRL-TK Vector ^(a,b)	20µg	E2241
pRL-CMV Vector ^(a,b,m)	20µg	E2261
pRL-SV40 Vector ^(a,b)	20µg	E2231

To inquire about the availability of bulk packaging and pricing for pRL Vectors, please contact Promega. For inquiries on the availability of new promoter variations within the pRL family of co-reporter vectors, contact Technical Services or visit our web site at www.promega.com.

pGL3 Vectors

Product	Size	Cat.#
pGL3 Control Vector ^(a,f,n)	20µg	E1741
pGL3 Basic Vector ^(a,f,n)	20µg	E1751
pGL3 Promoter Vector ^(a,f,n)	20µg	E1761
pGL3 Enhancer Vector ^(a,f,n)	20µg	E1771

Luciferase Assay Systems

Product	Size	Cat.#
Dual-Luciferase [®] Reporter Assay System	100 assays	E1910
Dual-Luciferase [®] Reporter Assay System 10-pack ^(c,d)	1,000 assays	E1960
Dual-Luciferase [®] Reporter 1000 Assay System ^(c,d)	1,000 assays	E1980

VI. Related Products (continued)

Transfection Systems

Product	Size	Cat.#
TransFast™ Transfection Reagent	1.2mg	E2431
Transfectam® Reagent for the Transfection of Eukaryotic Cells	1mg	E1231
	0.5mg	E1232
Tfx™-50 Reagent	2.1mg	E1811
Tfx™-10 Reagent	9.3mg	E2381
Tfx™-20 Reagent	4.8mg	E2391
Tfx™ Reagents Transfection Trio	5.4mg	E2400
ProFection® Mammalian Transfection System - Calcium Phosphate	80 transfections	E1200
ProFection® Mammalian Transfection System - DEAE-Dextran	80 transfections	E1210

VII. References

- Lorenz, W.W. *et al.* (1991) Isolation and expression of a cDNA encoding *Renilla reniformis* luciferase. *Proc. Natl. Acad. Sci. USA* **88**, 4438–42.
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