Certificate of Analysis

pGL4.14[*luc2*/Hygro] Vector:

Part No. E669A **Size** 20µg

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Instructions for use of this product can be found in the pGL4 Luciferase Reporter Vectors Technical Manual #TM259, available online at:

www.promega.com/protocols

Description: The pGL4.14[*luc2*/Hygro] Vector^(a-d) encodes the luciferase reporter gene *luc2* (*Photinus pyralis*) and is designed for high expression and reduced anomalous transcription. This vector also contains a mammalian selectable marker for hygromycin resistance in which the number of transcription factor binding sites has been reduced and mammalian codon usage optimized. The pGL4 Vectors are engineered with fewer consensus regulatory sequences than the pGL3 Vectors and a synthetic reporter gene, which has been codon optimized for mammalian expression.

The pGL4.14[*luc2*/Hygro] Vector is a basic vector with no promoter. However, it contains a multiple cloning region to allow cloning of a promoter of choice.

Concentration: 1µg/µl.

GenBank® Accession Number: AY864928.

Storage Buffer: The pGL4.14[*luc2*/Hygro] Vector is supplied in 10mM Tris-HCl (pH 7.4), 1mM EDTA.

Storage Conditions: See the product information label for storage temperature recommendations. Avoid multiple freezethaw cycles and exposure to frequent temperature changes. These fluctuations can greatly alter product stability. See the expiration date on the product information label.

Usage Notes:

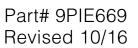
- For easy transfer from one pGL4 Vector to another, the multiple cloning region is consistent throughout the pGL4 Vector series. For easy transfer between pGL3 Vectors and pGL4 Vectors, many of the restriction enzyme sites present in the pGL3 Vectors are also present in the pGL4 Vectors.
- 2. Concentration gradients may form in frozen products and should be dispersed upon thawing. Mix well prior to use.

Quality Control Assays

Nuclease Assay: Following incubation of 1µg of pGL4.14[*luc2*/Hygro] Vector in standard restriction digest buffers at 37°C for 16–24 hours, no evidence of nuclease activity is detected by agarose gel electrophoresis.

Physical Purity: $A_{260}/A_{280} \ge 1.80$, $A_{260}/A_{250} \ge 1.05$ at pH 7.4.

Sequence: The pGL4.14[*luc2*/Hygro] Vector has been completely sequenced and has 100% identity with the published sequence, available at: www.promega.com/vectors





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Ken Wheeler

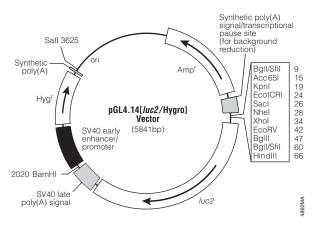
R. Wheeler, Quality Assurance

Signed by:



pGL4.14[luc2/Hygro] Vector Features List and Map

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Multiple cloning region	1–70
luc2 reporter gene	100-1752
SV40 late poly(A) signal	1787-2008
SV40 early enhancer/promoter	2056-2474
Synthetic hygromycin (Hyg ^r) coding region	2499-3536
Synthetic poly(A) signal	3560-3608
Reporter Vector primer 4 (RVprimer4) binding region	3675-3694
ColE1-derived plasmid replication origin	3932
Synthetic β-lactamase (Amp ^r) coding region	4723–5583
Synthetic poly(A) signal/transcriptional pause site	5688-5841
Reporter Vector primer 3 (RVprimer3) binding region	5790-5809



RVprimer3 5'...ACAAAACAAACTAGCAAAATAGGCTGTCCCCAGTGCAAGTGCAGGTGCCAGAACATTTCTCT Synthetic poly(A) signal/transcriptional pause site Sfil Acc 65I EcolCRI Bgll Kpnl Sacl Nhel Xhol EcoRV BgIII Sfil Bgll HindIII GGCCTCGGCGGCCAAGCTTGGCAATCCGGTACTGTTGGTAAAGCCACCATGG...3 173.6MA Luciferase start

Multiple cloning region of the pGL4.14[luc2/Hygro] Vector.

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^(b)U.S. Pat. No. 7,728,118.

(e)U.S. Pat. No. 8,008,006 and European Pat. No. 1341808. (d)Patents Pending.

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