Deciphering the pGL4 Vector Code

The pGL4 Vectors[a–e] are the most advanced luciferase reporter gene vectors that maximize expression, minimize off-target responses such as anomalous expression, and respond rapidly to transcriptional dynamics. pGL4 Vectors that express firefly luciferase (Table 1) or *Renilla* luciferase (Table 2) are available. The pGL4 Vectors offer a variety of choices for the three key components shown in Figure 1: The luciferase reporter gene, the upstream elements, including any promoters and response elements, and the selectable marker. The vector nomenclature for the pGL4 Vectors logically reflects these features. As an example, we will decipher the name of the pGL4.31[*luc2P*/CRE/Hygro] Vector.
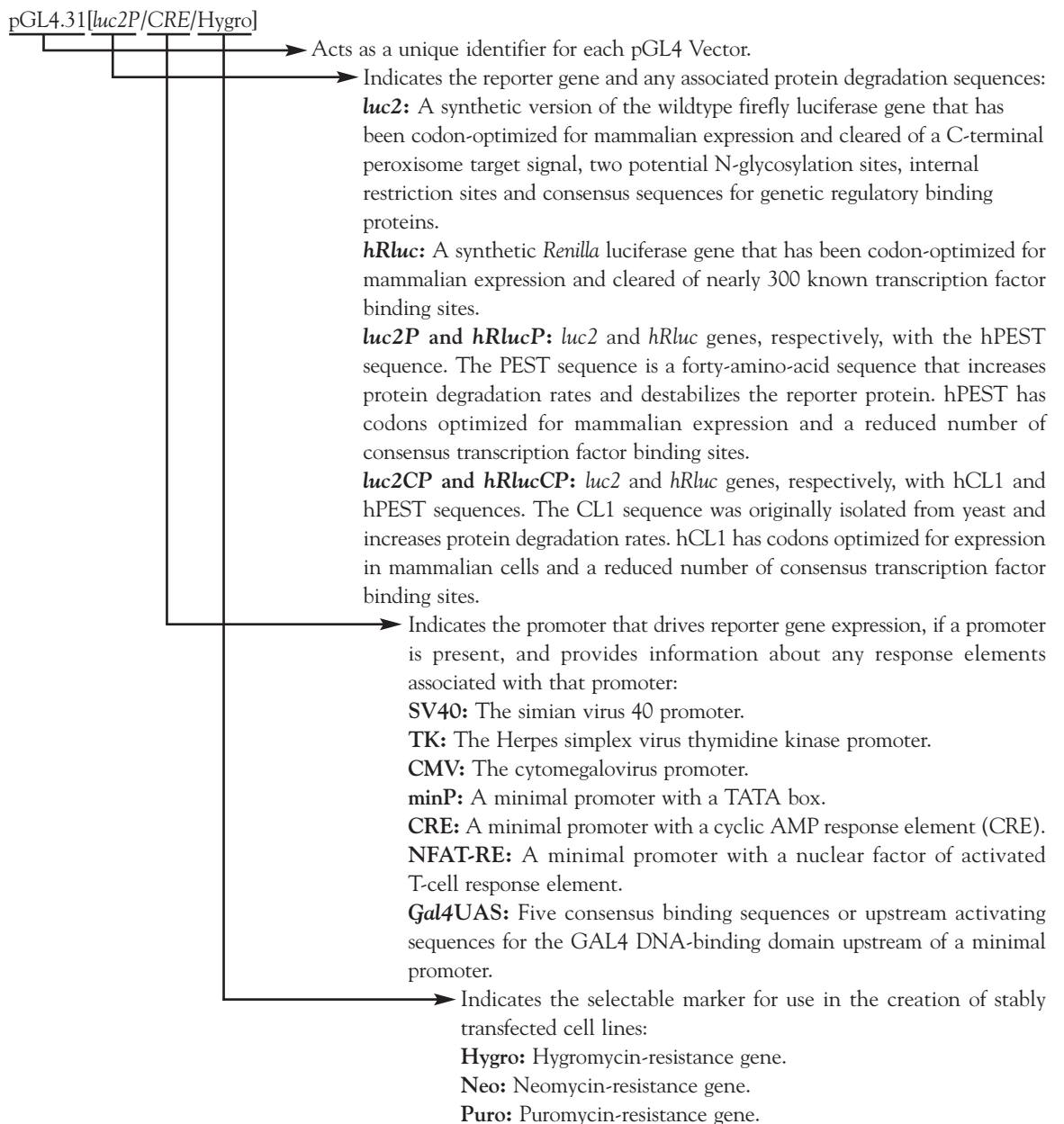
pGL4.31[*luc2P*/CRE/Hygro]

Acts as a unique identifier for each pGL4 Vector.

Indicates the reporter gene and any associated protein degradation sequences:

luc2: A synthetic version of the wildtype firefly luciferase gene that has been codon-optimized for mammalian expression and cleared of a C-terminal peroxisome target signal, two potential N-glycosylation sites, internal restriction sites and consensus sequences for genetic regulatory binding proteins.

hRluc: A synthetic *Renilla* luciferase gene that has been codon-optimized for mammalian expression and cleared of nearly 300 known transcription factor binding sites.

luc2P and **hRlucP:** *luc2* and *hRluc* genes, respectively, with the hPEST sequence. The PEST sequence is a forty-amino-acid sequence that increases protein degradation rates and destabilizes the reporter protein. hPEST has codons optimized for mammalian expression and a reduced number of consensus transcription factor binding sites.

luc2CP and **hRlucCP:** *luc2* and *hRluc* genes, respectively, with hCL1 and hPEST sequences. The CL1 sequence was originally isolated from yeast and increases protein degradation rates. hCL1 has codons optimized for expression in mammalian cells and a reduced number of consensus transcription factor binding sites.

Indicates the promoter that drives reporter gene expression, if a promoter is present, and provides information about any response elements associated with that promoter:

SV40: The simian virus 40 promoter.

TK: The Herpes simplex virus thymidine kinase promoter.

CMV: The cytomegalovirus promoter.

minP: A minimal promoter with a TATA box.

CRE: A minimal promoter with a cyclic AMP response element (CRE).

NFAT-RE: A minimal promoter with a nuclear factor of activated T-cell response element.

Gal4UAS: Five consensus binding sequences or upstream activating sequences for the GAL4 DNA-binding domain upstream of a minimal promoter.

Indicates the selectable marker for use in the creation of stably transfected cell lines:

Hygro: Hygromycin-resistance gene.

Neo: Neomycin-resistance gene.

Puro: Puromycin-resistance gene.

Table 1. pGL4 Vectors With the Synthetic Firefly Luciferase *luc2* gene.

Vector Name	Multiple Cloning Region	Catalog Number
pGL4.10[*luc2*]	Yes	E6651
pGL4.11[*luc2P*]	Yes	E6661
pGL4.12[*luc2CP*]	Yes	E6671
pGL4.13[*luc2/SV40*]	No	E6681
pGL4.14[*luc2*/Hygro]	Yes	E6691
pGL4.15[*luc2P*/Hygro]	Yes	E6701
pGL4.16[*luc2CP*/Hygro]	Yes	E6711
pGL4.17[*luc2*/Neo]	Yes	E6721
pGL4.18[*luc2P*/Neo]	Yes	E6731
pGL4.19[*luc2CP*/Neo]	Yes	E6741
pGL4.20[*luc2*/Puro]	Yes	E6751
pGL4.21[*luc2P*/Puro]	Yes	E6761
pGL4.22[*luc2CP*/Puro]	Yes	E6771
pGL4.23[*luc2*/minP]	Yes	E8411
pGL4.24[*luc2P*/minP]	Yes	E8421
pGL4.25[*luc2CP*/minP]	Yes	E8431
pGL4.26[*luc2*/minP/Hygro]	Yes	E8441
pGL4.27[*luc2P*/minP/Hygro]	Yes	E8451
pGL4.28[*luc2CP*/minP/Hygro]	Yes	E8461
pGL4.29[*luc2P*/CRE/Hygro]	No	E8471
pGL4.30[*luc2P*/NFAT-RE/Hygro]	No	E8481
pGL4.31[*luc2P*/*Gal*4UAS/Hygro]	No	C9351

Table 2. pGL4 Vectors with the Synthetic *Renilla* Luciferase *hRluc* gene.

Vector Name	Multiple Cloning Region	Catalog Number
pGL4.70[*hRluc*]	Yes	E6881
pGL4.71[*hRlucP*]	Yes	E6891
pGL4.72[*hRlucCP*]	Yes	E6901
pGL4.73[*hRluc*/SV40]	No	E6911
pGL4.74[*hRluc*/TK]	No	E6921
pGL4.75[*hRluc*/CMV]	No	E6931
pGL4.76[*hRluc*/Hygro]	Yes	E6941
pGL4.77[*hRlucP*/Hygro]	Yes	E6951
pGL4.78[*hRlucCP*/Hygro]	Yes	E6961
pGL4.79[*hRluc*/Neo]	Yes	E6971
pGL4.80[*hRlucP*/Neo]	Yes	E6981
pGL4.81[*hRlucCP*/Neo]	Yes	E6991
pGL4.82[*hRluc*/Puro]	Yes	E7501
pGL4.83[*hRlucP*/Puro]	Yes	E7511
pGL4.84[*hRlucCP*/Puro]	Yes	E7521

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[b] Cat.# C9351, E6651, E6661, E6671, E6681, E6691, E6701, E6711, E6721, E6731, E6741, E6751, E6761, E6771, E8411, E8421, E8431, E8441, E8451, E8461, E8471, E8481: The method of recombinant expression of *Coleoptera* luciferase is covered by U.S. Pat. Nos. 5,583,024, 5,674,713 and 5,700,673. A license (from Promega for research reagent products and from The Regents of the University of California for all other fields) is needed for any commercial sale of nucleic acid contained within or derived from this product.

[c] Cat.# E6881, E6891, E6901, E6911, E6921, E6931, E6941, E6951, E6961, E6971, E6981, E6991, E7501, E7511, E7521: Licensed from University of Georgia Research Foundation, Inc., under U.S. Pat. Nos. 5,292,658, 5,418,155, Canadian Pat. No. 2,105,984 and related patents.

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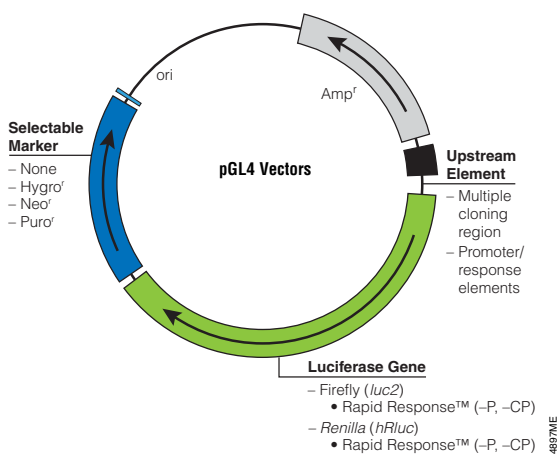

Figure 1. Schematic diagram of the pGL4 Vectors.