Lamin Chromobody®-TagGFP plasmid

The vector sequence has been compiled using the information from sequence databases, published literature, and other sources, together with partial sequences obtained by ChromoTek. This vector has not been completely sequenced.

![Diagram of Lamin Chromobody®-TagGFP plasmid]

**Location of features**

**PCMV IE**: 1-589  
Enhancer region: 59-465  
TATA box: 554-560  
Transcription start point: 583  
Lamin-VH: 621-1004  
TagGFP2: 1077-1790  
Stop codon: 1788-1790  
SV40 early mRNA polyadenylation signal  
Polyadenylation signals: 1919-1924 & 1948-1953  
mRNA 3’ ends: 1957 & 1969  
f1 single-strand DNA origin: 2016-2471  
SV40 origin of replication: 2812-2947  
SV40 early promoter Enhancer (72-bp tandem repeats): 2648-2719 & 2720-2791  
21-bp repeats: 2795-2815, 2816-2836 & 2838-2858  
Early promoter element: 2871-2877  
Neomycin/neomycin resistance gene  
Neomycin phosphotransferase coding sequences:  
Start codon (ATG): 2999-3001; Stop codon: 3791-3793  
Herpes simplex virus (HSV) thymidine kinase (TK) polyadenylation signal  
Polyadenylation signals: 4029-4034 & 4042-4047  
ColE1 replication origin: 4325-5007

**Vector description**

Lamin Chromobody®-TagGFP plasmid (pLC-TagGFP) is a mammalian expression vector encoding the marker of the nuclear envelope Lamin-VH fused to green fluorescent protein TagGFP2 (from Evrogen). The vector allows expression Lamin Chromobody®, TagGFP fusion protein in eukaryotic (mammalian) cells.

Chromobody® codon usage is optimized for high expression in mammalian cells (humanized) [Haas et al. 1996].

The vector backbone contains immediate early promoter of cytomegalovirus (PCMV ie) for protein expression, SV40 origin for replication in mammalian cells expressing SV40 T-antigen, ColE1 origin of replication for propagation in E. coli and f1 origin for single-stranded DNA production. SV40 polyadenylation signals (SV40 poly A) direct proper processing of the 3’-end of the reporter mRNA.

SV40 early promoter (PSV40) provides neomycin resistance gene (Neo) expression to select stably transfected eukaryotic cells using G418. Bacterial promoter (P) provides kanamycin resistance gene expression (Kan®) in E. coli. Kan®/Neo® gene is linked with herpes simplex virus (HSV) thymidine kinase (TK) polyadenylation signals.

**Expression in mammalian cells**

pLC-TagGFP vector can be transfected into mammalian cells by any known transfection method. If required, stable transformants can be selected using G418 [Gorman 1985].

**Propagation in E. coli**

Suitable host strains for propagation in E. coli include DH5alpha, HB101, XL1-Blue, and other general purpose strains. Plasmid incompatibility group is pMB1/ColE1. The vector confers resistance to kanamycin (30 µg/ml) to E. coli hosts. Copy number in E. coli is about 500.

Note: The plasmid DNA was isolated from dam-methylated E.coli. Therefore some restriction sites are blocked by methylation. If you wish to digest the vector using such sites you will need to transform the vector into a dam® host and make fresh DNA.

**Notice to Purchaser:**

Chromobody®-related materials (the Products) are intended for research use only. The Products are covered by U.S. Pat. applications pending. By use of these Products, you accept the terms and conditions of the applicable End User License Agreement (EULA non-profit entities). The CMV promoter is covered under U.S. Patents 5,168,062 and 5,385,839, and its use is permitted for research purposes only. Any other use of the CMV promoter requires a license from the University of Iowa Research Foundation, 214 Technology Innovation Center, Iowa City, IA 52242.

MATERIAL SAFETY DATA SHEET INFORMATION: To the best of our knowledge, these products do not require a Material Safety Data Sheet. However, all the properties of these products (and, if applicable, each of their components) have not been thoroughly investigated. Therefore, we recommend that you use gloves and eye protection, and wear a laboratory coat when working with these products.

For plasmid sequence, please contact info@chromotek.com

**Product**

<table>
<thead>
<tr>
<th>Code</th>
<th>Size</th>
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<tr>
<td>pLC-TagGFP</td>
<td>20 µg</td>
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**Use**

Lamin Chromobody®-TagGFP expression in mammalian cells for non-invasive live cell visualization of endogenous nuclear lamina.

**Replication**

Promoter for mammalian expression vector encoding the marker of the nuclear envelope Lamin-VH fused to green fluorescent protein TagGFP2 (from Evrogen). The vector allows expression Lamin Chromobody®, TagGFP fusion protein in eukaryotic (mammalian) cells.

Chromobody® codon usage is optimized for high expression in mammalian cells (humanized) [Haas et al. 1996].