General Information

Gene: neuropeptide Y

Official Symbol: NPY

Synonym: PYY

Source: Human

cDNA Size: 294bp

RefSeq: NM_000905.2

Description

Lot: Please refer to the label on the tube

Vector: pCMV3-SP-N-HA

Shipping carrier:
Each tube contains approximately 10 μg of lyophilized plasmid.

Storage:
The lyophilized plasmid can be stored at ambient temperature for three months.

Quality control:
The plasmid is confirmed by full-length sequencing with primers in the sequencing primer list.

Sequencing primer list:

<table>
<thead>
<tr>
<th></th>
<th>Primer Sequence</th>
</tr>
</thead>
<tbody>
<tr>
<td>pCMV3-F</td>
<td>5’ CAGGTGTCCACTCCAGGTCCAAG 3’</td>
</tr>
<tr>
<td>pcDNA3-R</td>
<td>5’ GGCAACTAGAAGGCACAGTCGAGG 3’</td>
</tr>
</tbody>
</table>

Or

<table>
<thead>
<tr>
<th>Primer</th>
<th>Primer Sequence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Forward T7</td>
<td>5’ TAATACGACTCCTATAGGG 3’</td>
</tr>
<tr>
<td>Reverse BGH</td>
<td>5’ TAGAAGGACACAGTCGAGG 3’</td>
</tr>
</tbody>
</table>

The plasmid is ready for:

• Restriction enzyme digestion
• PCR amplification
• E. coli transformation
• DNA sequencing

E. coli strains for transformation (recommended but not limited)

Most commercially available competent cells are appropriate for the plasmid, e.g. TOP10, DH5α and TOP10F’.

Plasmid Resuspension protocol

1. Centrifuge at 5,000 × g for 5 min.
2. Carefully open the tube and add 100 μl of sterile water to dissolve the DNA.
3. Close the tube and incubate for 10 minutes at room temperature.
4. Briefly vortex the tube and then do a quick spin to concentrate the liquid at the bottom. Speed is less than 5000 × g.
5. Store the plasmid at -20 °C.

Catalog Number: HG10946-NY
Vector Information

All of the pCMV vectors are designed for high-level stable and transient expression in mammalian hosts. High-level stable and non-replicative transient expression can be carried out in most mammalian cells. The vectors contain the following elements:

- Human enhanced cytomegalovirus immediate-early (CMV) promoter for high-level expression in a wide range of mammalian cells.
- Hygromycin resistance gene for selection of mammalian cell lines.
- A Kozak consensus sequence to enhance mammalian expression.

<table>
<thead>
<tr>
<th>Vector Name</th>
<th>pCMV3-SP-N-HA</th>
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<tbody>
<tr>
<td>Vector Size</td>
<td>6146bp</td>
</tr>
<tr>
<td>Vector Type</td>
<td>Mammalian Expression Vector</td>
</tr>
<tr>
<td>Expression Method</td>
<td>Constitutive, Stable / Transient</td>
</tr>
<tr>
<td>Promoter</td>
<td>CMV</td>
</tr>
<tr>
<td>Antibiotic Resistance</td>
<td>Kanamycin</td>
</tr>
<tr>
<td>Selection In Mammalian Cells</td>
<td>Hygromycin</td>
</tr>
<tr>
<td>Protein Tag</td>
<td>HA</td>
</tr>
</tbody>
</table>
**pCMV3-SP-N-HA** (suitable for secretory and membrane protein expression)

**Physical Map**

- CMV promoter
- enhancer
- T7 primer
- Kozak
- Signal Peptide
- N-HA
- linker
- MCS
- BGH reverse primer
- pCMV3-SP-N-HA
- SV40 early promoter
- Kan[R]
- Ori
- Hygro[R]

**Description**

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<td>HA</td>
</tr>
<tr>
<td><strong>Sequencing Primer</strong></td>
<td>Forward: T7(TAATACGACTCACTATAGGG) Reverse: BGH(TAGAAGGCACAGTCGAGG)</td>
</tr>
</tbody>
</table>

**Schematic of pCMV3-SP-N-HA Multiple Cloning Sites**

- **1415**: GGTGTCACCTCCACAGTCAAAGTTAACTTTATAGCAGCTACCTATAGGG GCCGCCACC
- **1475**: AAG CTC GGT ACC ATGCCACTGTCCTGTCCTGGCCCTCTTTGTGGATGACCTGGTGG
- **1535**: TAT CTC TAG GAC GTT CCT GAC TAC GCC GGA GGG GTT AGC GTA
- **1588**: GCT AGC GGA TCC GTT AAC CTT AGG ACC GTT CAT ATC GAT TAA A
- **1620**: CTC GAG TCT AGA GGG GCC GCC GAATTC GGA CCC GTTT AAG
- **1667**: CGCGTGATGAGCTGGCGTCGATGCTGGCTA GTTTGCCAGCCATCGTTGTTGC

**Comments for pCMV3-SP-N-HA:**
- CMV promoter: bases 250-837
- enhancer: bases 838-1445
- SV40 early promoter: bases 2387-2756
- Hygromycin ORF: bases 2774-3799
- pUC origin: bases 4442-5115
- Kanamycin ORF: bases 5189-6004

*pCMV3-SP-N-HA is recommended for constructing the N-HA tag secretory and membrane protein expression vector which containing a naïve signal peptide. An universal signal peptide is used to instead the naïve signal peptide.*