Mouse COCH ORF mammalian expression plasmid, N-His tag

Catalog Number:  MG50943-NH

General Information

Gene: coagulation factor C homolog (Limulus polyphemus)
Official Symbol: COCH
Synonym: AW122937, Coch-5B2, D12H14S564E, Coch
Source: Mouse
cDNA Size: 1659bp
RefSeq: NM_007728.5

Description

Lot: Please refer to the label on the tube
Vector: pCMV3-SP-N-His
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>Primer</th>
<th>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>
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</table>

Or

<table>
<thead>
<tr>
<th>Primer</th>
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<tbody>
<tr>
<td>Forward T7</td>
<td>5’ TAATACGACTCTATAGGG 3’</td>
</tr>
<tr>
<td>ReverseBGH</td>
<td>5’ TAGAAGGACAGTCGAGG 3’</td>
</tr>
</tbody>
</table>

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.

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’.

pCMV3-F and pcDNA3-R are designed by Sino Biological Inc. Customers can order the primer pair from any oligonucleotide supplier.
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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.

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<th>Vector Name</th>
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<tr>
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<td>6149bp</td>
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<tr>
<td>Vector Type</td>
<td>Mammalian Expression Vector</td>
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<td>Promoter</td>
<td>CMV</td>
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<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>His</td>
</tr>
</tbody>
</table>

Manufactured By Sino Biological Inc., FOR RESEARCH USE ONLY. NOT FOR USE IN HUMANS.
Fax :+86-10-51029969    Tel:+86- 400-890-9989    http://www.sinobiological.com
**pCMV3-SP-N-His** (suitable for secretary and membrane protein expression)

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**Sequencing Primer**

- Forward: T7:TAATACGACTCTATAGGG
- Reverse: BGH:TAGAAGGCAACATCGAGG

### Physical Map

- **CMV promoter**: bases 250-837
- **enhancer**: bases 838-1445
- **SV40 early promoter**: bases 2390-2759
- **Hygromycin ORF**: bases 2777-3802
- **pUC origin**: bases 4445-5118
- **Kanamycin ORF**: bases 5192-6007

**Comments for pCMV3-SP-N-His:**

- CMV promoter: bases 250-837
- enhancer: bases 838-1445
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**Schematic of pCMV3-SP-N-His Multiple Cloning Sites**

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