PC-12 Transfection Kit (for Rat Pheochromocytoma Cells, CRL-1721)

Catalog No. 3229  Size: 0.5 ml
Catalog No. 3230  Size: 1.5 ml
Catalog No. 7077  Size: 8.0 ml

Contents and Shipping:
PC-12 Transfection Kit includes PC-12 Transfection Reagent (0.5 ml / 1.5 ml / 8.0 ml), Transfection Enhancer (0.5 ml), and Complex Condenser (0.5 ml). PC-12 Transfection Reagent is supplied in liquid form at a concentration of 0.3 mg/ml, shipped at ambient temperature.

Description:
Two component lipid-based proprietary formulation optimized for PC-12 cells transfection.

Product Qualification:
PC-12 Transfection Reagent is tested functionally by transfection of PC-12 cells with a small interfering RNAs targeting 3 different genes (Lamin A/C, GAPDH, Cyclophilin B). Transfection Reagent is tested for absence of nuclease contamination and microbial contamination.

Storage:
Store reagent at 4°C upon receipt. If stored properly, reagent is stable for 6 months.

Intended Use:
For \textit{in vitro} use only.

MSDS:
MSDS documents are available online at \url{www.altogen.com}

\textbf{In Vivo Transfection Kits (for compound testing in rodents)}:
- Catalog #5010 / 5011 / 5012 - Lipid \textit{In Vivo} Transfection Kit
- Catalog #5020 / 5021 / 5022 - Polymer \textit{In Vivo} Transfection Kit
- Catalog #5030 / 5031 / 5032 - Nanoparticle \textit{In Vivo} Transfection Kit
- Catalog #5040 / 5041 / 5042 - PEG-Liposome \textit{In Vivo} Transfection Kit
- Catalog #5050 / 5051 / 5052 - Pancreas \textit{In Vivo} Transfection Kit
- Catalog #5060 / 5061 / 5062 - Liver \textit{In Vivo} Transfection Kit
- Catalog #5070 / 5071 / 5072 - Kidney \textit{In Vivo} Transfection Kit

Transfection Controls and Recommended Products:
- Catalog #4060 - GFP-expressing plasmid DNA (25 ug)
- Catalog #4061 - Cell Cycle Arrest siRNA (5 nmol)
- Catalog #4062 - Apoptosis Inducing siRNA (5 nmol)

To Place an Order:
Both domestic (USA) and international orders can be placed online (\url{www.altogen.com}) using credit card payment. Purchase Order (PO) can be faxed at (702) 989-0841 (for USA only).

Transfection Resource: \url{www.altogen.com/transfection-resource}

Altogen Labs. GLP Compliant Pre-clinical CRO Laboratory Services: \url{www.altogenlabs.com}
**Recommended Transfection Protocols (for 24-well plate):**

<table>
<thead>
<tr>
<th>PC-12 Standard Transfection Protocol (24-well plate):</th>
<th>PC-12 Reverse Transfection Protocol (24-well plate):</th>
</tr>
</thead>
</table>
| 1. Plate 10,000 - 15,000 PC-12 cells per well in 0.5 ml of complete growth medium 12-24 hours prior to transfection | 1. Prepare PC-12 cell suspension:  
   a. Trypsinize cells (0.05% Trypsin) for 3-5 minutes at 37°C  
   b. Dilute in complete growth medium to 5 x 10^4 cells/ml |
| 2. Wash with 1xPBS and add 0.5 ml of fresh growth medium | 2. Prepare transfection complexes by mixing 40 µl of serum-free medium, 5.5 µl of transfection reagent, and  
   • 750 ng DNA (or mRNA), or  
   • 30 nM - 50 nM of siRNA (or microRNA)  
   *Referred to a final volume including growth medium |
| 3. Prepare transfection complexes by mixing 40 µl of serum-free medium, 5.5 µl of transfection reagent, and  
   • 750 ng DNA (or mRNA), or  
   • 30 nM - 50 nM of siRNA (or microRNA)  
   *Referred to a final volume including growth medium | 3. Incubate transfection complexes at RT for 15 - 30 minutes |
| 4. Optional: Add 2 µl of Complex Condenser. This reagent reduces the size of transfection complex, therefore increasing transfection efficiency; however it may increase cell toxicity | 4. Optional: Add 2 µl of Complex Condenser. This reagent reduces the size of transfection complex, therefore increasing transfection efficiency; however it may increase cell toxicity |
| 5. Optional: Add 2 µl of Complex Condenser. This reagent reduces the size of transfection complex, therefore increasing transfection efficiency; however it may increase cell toxicity | 5. Plate 20,000 - 30,000 cells per well in 0.5 ml of complete growth medium (from step #1) into culture plate |
| 6. Incubate cells at 37°C in a humidified CO2 incubator | 6. Add prepared transfection complexes (from step 3 or 4) |
| 7. Incubate cells at 37°C in a humidified CO2 incubator | 7. Incubate cells at 37°C in a humidified CO2 incubator |
| 8. Assay for phenotype or target gene expression 48 - 72 hours after transfection | 8. Assay for phenotype or target gene expression 48 - 72 hours after transfection |

**Scaling Up or Down Transfections:**

<table>
<thead>
<tr>
<th>Culture Vessel Surface Area (cm²)</th>
<th>Volume of Growth Medium (ml)</th>
<th>Transfection Reagent (µl)</th>
<th>Complex Condenser (µl)</th>
<th>Transfection Enhancer (µl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>96-well, 0.3 cm x cm</td>
<td>0.12</td>
<td>1.5</td>
<td>0.3</td>
<td>0.3</td>
</tr>
<tr>
<td>24-well, 2 cm x cm</td>
<td>0.5</td>
<td>5.5</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>12-well, 4 cm x cm</td>
<td>1</td>
<td>12</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>6-well, 10 cm x cm</td>
<td>3</td>
<td>35</td>
<td>12</td>
<td>12</td>
</tr>
<tr>
<td>60-mm, 20 cm x cm</td>
<td>5</td>
<td>60</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>10-cm, 60 cm x cm</td>
<td>15</td>
<td>180</td>
<td>60</td>
<td>60</td>
</tr>
</tbody>
</table>

**Optimizing Transfection:**

To obtain the highest transfection efficiency, optimize transfection conditions by varying PC-12 cell density and amount of transfection reagent. High passage of PC-12 cells and use of antibiotics (or growth factors) may require using larger volumes of PC-12 transfection reagent per reaction.

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