Altogen Biosystems

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HCC1937 Transfection Protocol (Kit for Breast Carcinoma Cells, CRL-2336)

Catalog No. 6807 Size: 0.5 ml Catalog No. 6808 Size: 1.5 ml Catalog No. 7042 Size: 8.0 ml

Contents and Shipping:

HCC1937 Transfection Kit includes HCC1937 Transfection Reagent (0.5 ml / 1.5 ml / 8.0 ml), Transfection Enhancer (0.5 ml), and Complex Condenser (0.5 ml). HCC1937 Transfection Reagent is supplied in liquid form at a concentration of 0.25 mg/ml, shipped at ambient temperature.

Description:

Polymer-based transfection reagent is a proprietary formulation optimized for transfection of DNA and RNA into HCC1937 cells.

Product Qualification:

HCC1937 Transfection Reagent is tested functionally by transfection of HCC1937 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 12 months.

Intended Use:

For *in vitro* use only.

Safety Data Sheet (SDS):

SDS documents are available online at https://altogen.com

In Vivo Transfection Kits (for compound testing in rodents):

- Catalog #5010 / 5011 / 5012 Lipid In Vivo Transfection Kit
- Catalog #5020 / 5021 / 5022 Polymer In Vivo Transfection Kit
- Catalog #5030 / 5031 / 5032 Nanoparticle In Vivo Transfection Kit
- Catalog #5040 / 5041 / 5042 PEG-Liposome In Vivo Transfection Kit
- Catalog #5050 / 5051 / 5052 Pancreas In Vivo Transfection Kit
- Catalog #5060 / 5061 / 5062 Liver In Vivo Transfection Kit
- Catalog #5070 / 5071 / 5072 Kidney 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 (https://altogen.com) using credit card payment. Purchase Order (PO) can be faxed at (702) 989-0841 (for USA only).

Transfection Resource: altogen.com/transfection-resource

Altogen Labs. GLP Compliant Pre-clinical CRO Laboratory Services:

AltogenLabs.com

Recommended Transfection Protocols (for 24-well plate):

HCC1937 Standard Transfection Protocol (24-well plate):

- **1.** Plate 10,000 15,000 HCC1937 cells per well in 0.5 ml of complete growth medium 12–24 hours prior to transfection
- 2. Wash with 1xPBS and add 0.5 ml of fresh growth medium
- 3. Prepare transfection complexes by mixing 40 μ l of serum-free medium, 5.5 μ l 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
- **4.** Incubate transfection complexes at RT for 15 30 minutes
- **5.** Optional: Add $2 \mu l$ of Complex Condenser. This reagent reduces the size of transfection complex, therefore increasing transfection efficiency; however it may increase cell toxicity
- $\bf 6.$ Add prepared transfection complexes to 0.5 ml of complete growth medium with HCC1937 cells (from step 2)
- **7.** Incubate cells at 37°C in a humidified CO₂ incubator
- **8.** Assay for phenotype or target gene expression 48 72 hours after transfection

Optional: Transfection efficiency can be increased by addition of Transfection Enhancer reagent. Add 2 μl of Transfection Enhancer reagent 12-24 hours after transfection If the viability of HCC1937 cells being transfected is affected at 16 - 24 hours post-transfection, the level of cytotoxicity can be decreased by changing the growth medium and eliminating redundant exposure of cells to transfectant

HCC1937 Reverse Transfection Protocol (24-well plate):

- 1. Prepare HCC1937 cell suspension:
 - a. Trypsinize cells (0.05% Trypsin) for 3-5 minutes at 37°C
- **b.** Dilute in complete growth medium to 5×10^4 cells/ml
- **2.** Prepare transfection complexes by mixing 40 μ l of serum-free medium, 5.5 μ l 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 $2 \mu l$ 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.** Add prepared transfection complexes (from step 3 or 4)
- 7. Incubate cells at 37°C in a humidified CO₂ incubator
- **8.** Assay for phenotype or target gene expression 48 72 hours after transfection

Optional: Transfection efficiency can be increased by addition of Transfection Enhancer reagent. Add 2 μl of Transfection Enhancer reagent 12-24 hours after transfection If the viability of HCC1937 cells being transfected is affected at 16 - 24 hours post-transfection, the level of cytotoxicity can be decreased by changing the growth medium and eliminating redundant exposure of cells to transfectant

Scaling Up or Down Transfections:

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

Optimizing Transfection:

To obtain the highest transfection efficiency, optimize transfection conditions by varying HCC1937 cell density and amount of transfection reagent. High passage of HCC1937 cells and use of antibiotics (or growth factors) may require using larger volumes of HCC1937 transfection reagent per reaction.

Limited Use Label License:

The purchase of this product conveys to the purchaser the limited right to use the purchased amount of the product only to perform internal research for the sole benefit of the purchaser. This product is for research purposes only and is not for use in commercial applications of any kind. For information on obtaining additional rights, please contact Altogen Biosystems at orders@altogen.com.

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