Altogen Biosystems

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CHOK1 Transfection Protocol (Kit for Ovarian Adenocarcinoma Cells, CRL-9618)

Catalog No. 6450	Size: 0.5 ml
Catalog No. 6451	Size: 1.5 ml
Catalog No. 7030	Size: 8.0 ml

Contents and Shipping:

CHOK1 Transfection Kit includes CHOK1 Transfection Reagent (0.5 ml / 1.5 ml / 8.0 ml), Transfection Enhancer (0.5 ml), and Complex Condenser (0.5 ml). CHOK1 Transfection Reagent is supplied in liquid form at a concentration of 0.3 mg/ml, shipped at ambient temperature.

Description:

Lipid-based proprietary formulation optimized for transfection of CHOK1 cell line.

Product Qualification:

CHOK1 Transfection Reagent is tested functionally by transfection of CHOK1 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 (<u>https://altogen.com</u>) 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: <u>AltogenLabs.com</u>

Recommended Transfection Protocols (<u>for 24-well plate</u>):

CHOK1 Standard Transfection Protocol (24-well plate):	CHOK1 Reverse Transfection Protocol (24-well plate):				
1. Plate 10,000 - 15,000 CHOK1 cells per well in 0.5 ml of	1. Prepare CHOK1 cell suspension:				
complete growth medium 12–24 hours prior to transfection	a. Trypsinize cells (0.05% Trypsin) for 3-5 minutes at 37°C				
2. Wash with 1xPBS and add 0.5 ml of fresh growth medium	b. Dilute in complete growth medium to 5×10^4 cells/ml				
3. Prepare transfection complexes by mixing 40 µl of serum-	2. Prepare transfection complexes by mixing 40 µl of serum-				
free medium, 5 µl of transfection reagent, and	free medium, 5 µl of transfection reagent, and				
• 600 ng DNA (or mRNA), or	• 600 ng DNA (or mRNA), or				
• 30 nM - 50 nM of siRNA (or microRNA)	• 30 nM - 50 nM of siRNA (or microRNA)				
*Referred to a final volume including growth medium	<i>Referred to a final volume including growth medium</i>				
4. Incubate transfection complexes at RT for 15 - 30 minutes	3. Incubate transfection complexes at RT for 15 - 30 minutes				
5. Optional: Add 2 µl of Complex Condenser. This reagent	4. <u>Optional</u> : Add 2 µl of Complex Condenser. This reagent				
reduces the size of transfection complex, therefore increasing	reduces the size of transfection complex, therefore increasing				
transfection efficiency; however it may increase cell toxicity	transfection efficiency; however it may increase cell toxicity				
6. Add prepared transfection complexes to 0.5 ml of	5. Plate 20,000 - 30,000 cells per well in 0.5 ml of complete				
complete growth medium with CHOK1 cells (from step 2)	growth medium (from step #1) into culture plate				
7. Incubate cells at 37° C in a humidified CO ₂ incubator	6. Add prepared transfection complexes (from step 3 or 4)				
8. Assay for phenotype or target gene expression 48 - 72	7. Incubate cells at 37° C in a humidified CO ₂ incubator				
hours after transfection	8. Assay for phenotype or target gene expression 48 - 72				
	hours after transfection				
Optional: Transfection efficiency can be increased by	Optional: Transfection efficiency can be increased by				
addition of Transfection Enhancer reagent. Add 2 µl of	addition of Transfection Enhancer reagent. Add 2 µl of				
Transfection Enhancer reagent 12-24 hours after transfection	Transfection Enhancer reagent 12-24 hours after transfection				
If the viability of CHOK1 cells being transfected is affected	If the viability of CHOK1 cells being transfected is affected				
at 16 - 24 hours post-transfection, the level of cytotoxicity					
can be decreased by changing the growth medium and	can be decreased by changing the growth medium and				
eliminating redundant exposure of cells to transfectant	eliminating redundant exposure of cells to transfectant				

Scaling Up or Down Transfections:

Culture Vessel Surface Area (cm ²)	Volume of Growth Medium (ml)	Transfection Reagent (µl)	Complex Condenser (µl)	Transfection Enhancer (µl)
96-well, 0.3 cm x cm	0.12	1.4	0.3	0.3
24-well, 2 cm x cm	0.5	5	2	2
12-well, 4 cm x cm	1	10	4	4
6-well, 10 cm x cm	3	30	12	12
60-mm, 20 cm x cm	5	50	20	20
10-cm, 60 cm x cm	15	160	60	60

Optimizing Transfection:

To obtain the highest transfection efficiency, optimize transfection conditions by varying CHOK1 cell density and amount of transfection reagent. High passage of CHOK1 cells and use of antibiotics (or growth factors) may require using larger volumes of CHOK1 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 <u>orders@altogen.com</u>.

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