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Human Glucocorticoid Receptor Stable Cells (in mouse muscle C2C12)

Species: Cell Number: Homo Sapiens ~2x10⁶

Catalog Number: Storage/Shipping: CLHKGR03 Dry Ice/-80°C

Introduction:

Nuclear hormone receptors (NHRs) are a superfamily of transcription factors that function as powerful metabolic regulators to control a variety of systemic processes in physiology. They also play key roles in the pathophysiology of many major disease states, such as diabetes, obesity, inflammation, atherosclerosis and heart failure. To date, this superfamily has provided a rich source of drug design targets and it is continuing to be one of the hottest areas for pharmaceutical research.

PrimCells now provides a comprehensive set of NHR expressing cell lines to the research community. These high-quality, Flag and/or HA-tagged NHR-expressing cell lines aim to facilitate further biochemical and molecular studies of their functions and hence encourage new strategies for drug design.

Glucocorticoids (GCs) are steroid hormones that allow us to cope with environmental and physiological stresses. Although GCs are widely used as anti-inflammatory agents in several clinical areas, long-term use of those steroids causes problems instead. Full achievement of the clinical potential of GCs necessitates limiting the side effects of these drugs.

The lipophilic nature of GCs allows them to readily diffuse into the cells, where they bind to a cytosolic receptor, the Glucocorticoid Recetpor (GR) to exert their mechanism of action. Without ligand binding, GR is predominantly localized in cytoplasm, whereas upon ligand binding it rapidly translocates to nucleus where it can both positively and negatively regulate gene expression. GR belongs to the nuclear receptor superfamily of ligand-activated transcription factors that function as powerful metabolic regulators.

The GR stable cell line (Flag and HA tagged)/C2C12 cell line was created by stable transfection of C2C12 cells with a plasmid expressing Flag and HA tagged GR protein.

Thawing of Frozen Cells

1. Upon receipt of the frozen cells, it is recommended to thaw the cells and initiate the culture immediately in order to retain the highest cell viability.

2. To thaw the cells, put the vial in 37°C water bath with gentle agitation for ~1min. Keep the cap out of water to minimize the risk of contamination.

3. Pipette the cells into a 15ml conical tube with ~5ml fresh culture medium.

4. Centrifuge at 1000rpm (~220g) for 5min under room temp.

5. Remove the supernatant and resuspend the cells in fresh culture medium

6. Transfer the cells into new tissue culture flasks and move them to 37°C incubator (5% CO₂) for continuous culture.

Safety Precaution: it is highly recommended that protective gloves and clothing should be used when handling frozen vials.

Standard Culture Procedure

1. Cells should be maintained in the complete culture medium until reaching ~80-90% confluence. *Note: Never let the cells to become over confluent.*

2. Add ~2.5ml of 0.05% Trypsin-EDTA to the flask and incubate for 5min at 37°C.

3. Neutralize the enzyme activity by adding 2-3 volumes of fresh complete culture medium.

4. Centrifuge 1000rpm (~220g) for 5min and resuspend the cells in desired volume of medium.

5. Transfer the cells to a new tissue culture treated flask for subculture. *Note:* It is recommended that cells are passaged at the ratio of 1:10.

Complete Growth Medium

DMEM (Corning, Cat#10014CV): 450ml FBS: 50ml Total Volume: 500ml

Technical Support

For additional information regarding the product and technical questions, please contact <u>Supports@PrimCells.com</u>. You are guaranteed to receive a response within 24hrs from one of our scientists.

Disclaimers

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