Datasheet for MCF7/dCas9-VP64+MPH Cell Line

Catalog number: SL304

Product: MCF7 cell line stably expressing CRISPR dCas9-VP64 and MS2-P65-HSF1

Description: This product is a cell line stably expressing the catalytically inactive, HA-tagged dCas9 nuclease fused to the VP64 transcriptional activation domain and the activator helper complex MS2-P65-HSF1 (MPH). This cell line also contains blasticidin and hygromycin resistance genes. The target guide RNAs (with MS2-binding loops) can be transfected or transduced in the cell line to activate the gene transcription.

Quantity: 1 vial of 2 x 10^6 cells; frozen

Shipping conditions: Dry ice

Storage conditions: Liquid nitrogen vapor phase. Remove the item from the dry ice packaging and check all items for damage and leakage. Place immediately into storage at or below -140°C, preferably into the liquid nitrogen vapor phase, until use.

Source of parental line:

MCF7
Organism: Homo sapiens, human
Tissue: breast carcinoma
Cell type: epithelial

Safety instructions: To ensure safety, protective gloves, clothing, and a face mask should be worn when handling frozen vials. Some leakage of liquid nitrogen into the vial may occur during storage. The liquid nitrogen will be converted to gas upon thawing. Due to the nature of nitrogen gas, pressure may build within the vial and possibly result in the vial exploding or losing its cap. This may cause flying debris.

Thawing procedure: The vial of cells should be thawed in a 37°C water bath with gentle agitation. For optimal performance, the vial should be thawed in under two minutes. Ensure that the cap of the vial did not loosen upon thawing, and re-tighten as needed. Spray the vial with 70% EtOH and wipe off. Repeat. Using aseptic technique, add the contents of the vial to 9ml of complete growth medium (without selection). Centrifuge for 5min at 125xg. Aspirate the medium, being careful not to disturb the pellet. Re-suspend in 10mL of complete growth medium, and place into a culture vessel of your choice. Only add selection to the medium after 24 hours in
Culture conditions:

**Complete Growth Medium**

The base medium for this cell line is DMEM. For optimal growth and maintenance of selection, add the following components to the base medium: fetal bovine serum to a final concentration of 10% and human recombinant insulin 0.01 mg/ml.

**Selection**

Blasticidin to a final concentration of 8 µg/mL, and hygromycin to a final concentration of 200 µg/mL.

**Culture temperature:**

37°C with 5%CO₂

**Subculture:**

Rinse the cells with PBS without cations, digest cells with 0.25% (w/v) Trypsin-EDTA (0.53mM) solution and split at 1:6 to 1:10 ratio.

**Cryopreservation:**

Freeze slowly in complete growth medium supplemented with 5% (v/v) DMSO.

**Quality control:**

The genome integration, expression levels of dCas9-VP64 and MPH, and the transcription activation of target genes using the cell line have been verified by qPCR. All other QC specifications have also been met. For detailed information, please refer to the certificate of analysis.
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