

DATA SHEET

CELL LINE DESIGNATION
ORIGIN (PARENTAL CELL)
GENE INTRODUCED
RECEPTOR INTRODUCED:

Melanocortin 4 Receptor cell line (CB-80200-211)
HEK 293-CNG cell (CB-80200-200)
Genbank LocusID 4160
Human Melanocortin 4 receptor (NCBI protein database
XP_008716.1)

USAGE

- cAMP assay for Gs-coupled human Melanocortin 4 Receptor (MC4R).
- HEK293-CNG cells (CB-80200-200) without transfected Melanocortin 4 Receptor are used as a negative control.

QUALITY CONTROL

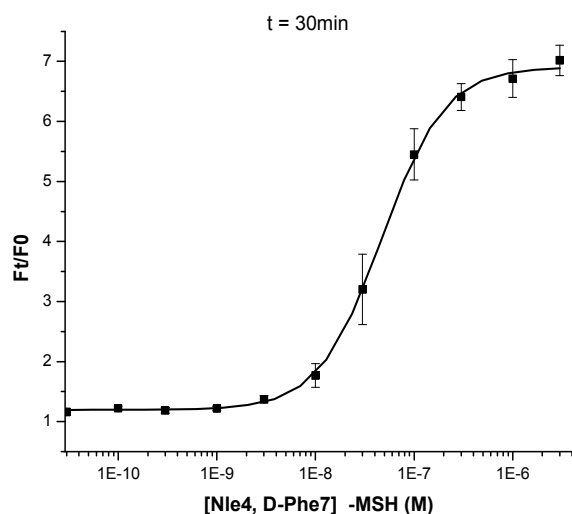
1. This cell line has been tested negative for *Mycoplasma sp.*
2. This cell line has been tested positive for Melanocortin 4 Receptor specific response.
3. Surviving rate: More than 2.5 million/vial on the second day after thawing.
4. The receptor specific activity is stable for 10 weeks continuous passage.

CELL CULTURE CONDITION

1. Growth medium: 90% DMEM, 10% FBS, 250 $\mu\text{g/ml}$ G418 and 1 $\mu\text{g/ml}$ puromycin
2. Freezing medium: 10% DMSO, 90% FBS

DATA EXAMPLE

A



B

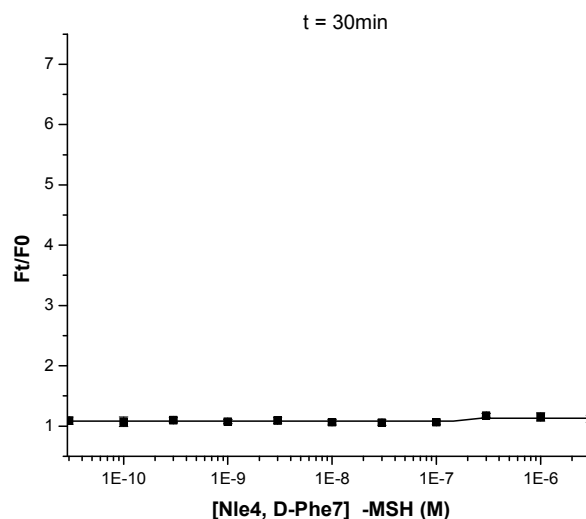


Figure 1. Response of ACTOne MC4R cell line & parental cell line to [Nle4, D-Phe7] α -MSH.

ACTOne MC4R cells and parental cells (CB-80200-200) were plated overnight in 20 μl culture medium on a BD Biocoat 384 well plate. The next day, cells were dye-loaded with 20 μl /well of 1X Dye-loading solution (ACTOne Membrane Potential Assay Kit). After 2 hours of incubation at room temperature, two readings were obtained prior to and 30 min after the addition of [Nle4, D-Phe7] α -MSH. Ratios of the two readings (F/F0) are plotted in the figure.

- A. Dose response curve of [Nle4, D-Phe7] α -MSH, in ACTOne MC4R cell line. EC₅₀ = 47 nM in the absence of PDE inhibitor, Ro 20-1724, and EC₅₀ = 1.7 nM in the presence of PDE inhibitor. (Data not shown)**
- B. Parental cells do not respond to [Nle4, D-Phe7] α -MSH.**