

DATA SHEET

Catalog # Cell Line Designation Parental Cell Gene Introduced NCBI Accession #	AG-10200-211 Melanocortin 4 Receptor cell line HEK 293-CNG cell (AG-10200-200) Human Melanocortin 4 receptor (MC4R) XP_008716.1
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USAGE

- cAMP assay for Gs-coupled human Melanocortin 4 Receptor (MC4R).
- HEK293-CNG cells (AG-10200-200) without transfected Melanocortin 4 Receptor are used as a negative control.

QUALITY CONTROL

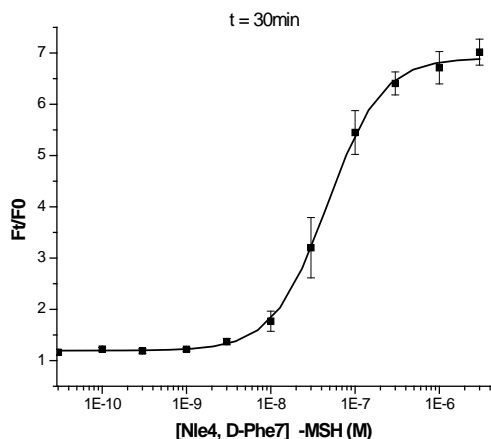
1. This cell line has been tested negative for *Mycoplasma sp.*
2. This cell line has been tested positive for Melanocortin 4 Receptor specific response.
3. Surviving rate: More than 2.5 million/vial on the second day after thawing.
4. The receptor specific activity is stable for 10 weeks continuous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

DATA EXAMPLE

A



B

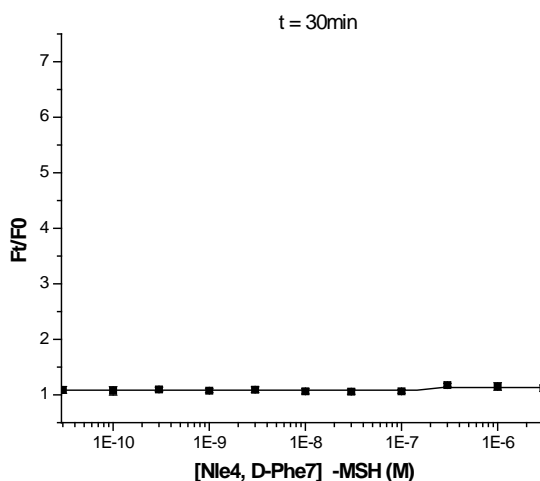


Figure 1. Response of ACTOne MC4R cell line & parental cell line to [Nle4, D-Phe7] a-MSH.

ACTOne MC4R cells and parental cells (AG-10200-200) were plated overnight in 20 ml culture medium on a BD Biocoat 384 well plate. The next day, cells were dye-loaded with 20 ml/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] a-MSH. Ratios of the two readings (F/F0) are plotted in the figure.

- A. Dose response curve of [Nle4, D-Phe7] a-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] a-MSH.**