

DATA SHEET

CELL LINE DESIGNATION

Corticotropin Releasing Hormone Receptor 1 cell line (CB-80200-249)

ORIGIN (PARENTAL CELL)

HEK 293-CNG cell (CB-80200-200)

GENE INTRODUCED

Genbank Locus ID 1394

RECEPTOR INTRODUCED:

Human corticotropin releasing hormone receptor 1 (NCBI protein database AAA35718)

USAGE

- cAMP assay for Gs-coupled human Corticotropin Releasing Hormone Receptor 1 (CRHR1).
- HEK293-CNG cells (CB-80200-200) without transfected Corticotropin Releasing Hormone Receptor 1 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 Corticotropin Releasing Hormone Receptor 1 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

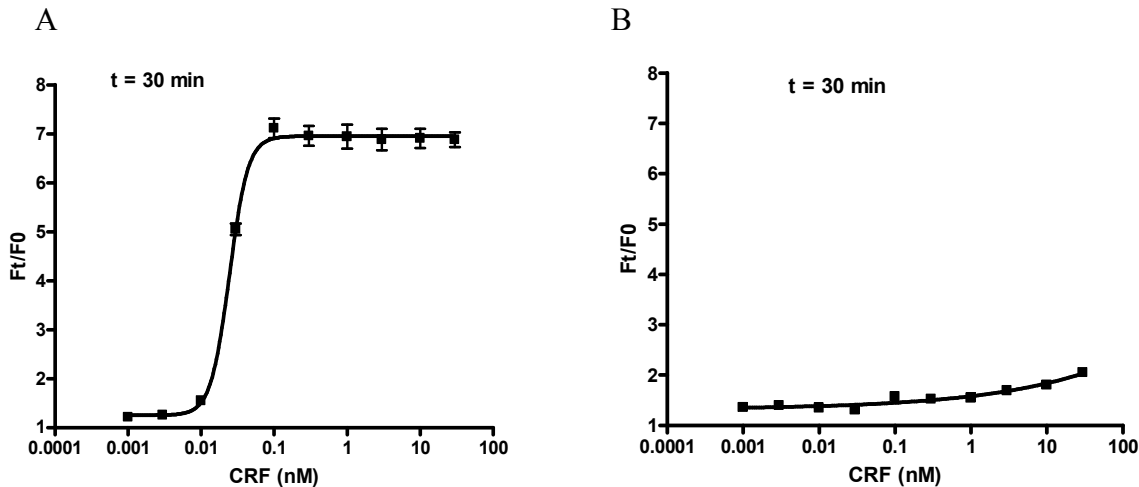


Figure 1. Response of ACTOne CRHR1 cell line & parental cell line to CRF.

ACTOne CRHR1 cells and parental cells (CB-80200-249) 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 CRF. Ratios of the two readings (Ft/F0) are plotted in the figure.

- A. Dose response curve of CRF in ACTOne CRHR1 cell line. EC50 = 24 pM in the presence of PDE inhibitor Ro20-1724, and EC50 = 148 pM in the absence of Ro20-1724 (data not shown).
- B. Parental cells have very small response to CRF.