

Technical Data Certificate of Analysis

Caution: For Laboratory Use. A research chemical for research purposes only.

Membrane Target Systems™ human Opioid delta Receptor

PRODUCT No.: RBHODM400UA

LOT No.: 492-911-A

MATERIAL PROVIDED

MEMBRANES: 1 x 400 units / 1000 µl frozen aliquot.
PACKAGING BUFFER: 50 mM Tris-HCl (pH 7.4), 0.5 mM EDTA, 10 mM MgCl₂, 10% sucrose.

PRODUCT INFORMATION

CELLULAR BACKGROUND: CHO-K1
PROTEIN ACCESSION NUMBER: not available
UNIT SIZE: 15 µg protein / unit
STORAGE CONDITIONS: Store at -80°C. **Freeze-thaw is not recommended** as it can affect product performance and homogeneity. In order to minimize negative impact of freeze-thawing, flash freeze in liquid nitrogen for 30 seconds prior to transferring to -80°C.
STABILITY: This product is stable for at least 3 years from reception if used and stored under recommended conditions.

QUALITY CONTROL

EXPRESSION LEVEL (B_{Max}): 3.9 pmol/mg membrane protein.
K_D FOR [³H]-NALTRINDOLE : 0.39 nM
PROTEIN CONCENTRATION: 6 mg/ml

RECOMMENDED ASSAY CONDITIONS

ASSAY BUFFER: 50 mM Tris-HCl pH 7.4, 5 mM MgCl₂
WASH BUFFER: 50 mM Tris-HCl pH 7.4, 5 mM MgCl₂
BINDING PROTOCOL: Binding assays are performed in 550 µl total volume according to the following conditions:

- 1 - Membrane dilution: 0.125 ml of membranes + 24.875 ml assay buffer (1:200 dilution)
- 2 - Incubation: 25 µl of incubation buffer or unlabeled ligand (Naltrexone, 100 µM final for non specific binding)
25 µl of radioligand at the appropriate concentration (see graph on page 2)
500 µl of diluted membranes
- 3 - Incubation time: 60 minutes at RT °C
- 4 - Filtration: Over GF/C filter presoaked in 0.5 % PEI then washed 9x with 500 µl of ice cold wash buffer.

LOT SPECIFIC DATA

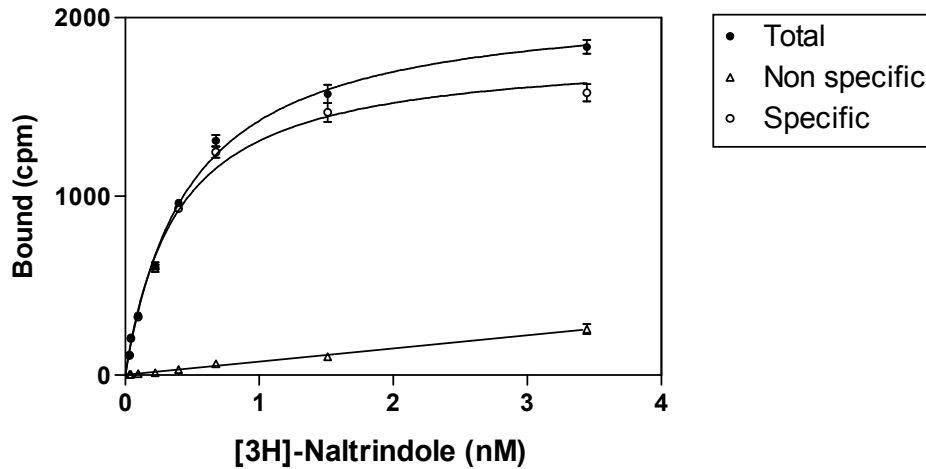


Figure 1: SATURATION BINDING ASSAY CURVE (FILTRATION)
96-well saturation binding assay curve (15 μ g membranes/well, TopCount™).

TYPICAL PRODUCT DATA

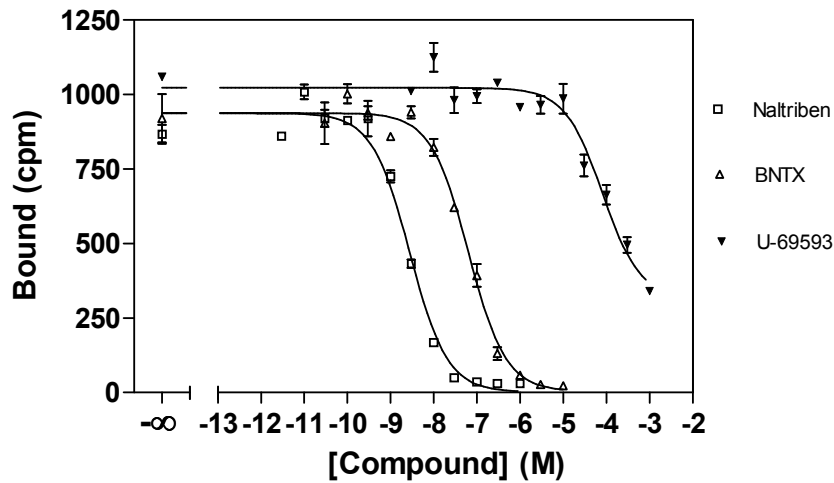


Figure 2: COMPETITION BINDING ASSAY CURVE (FILTRATION)
96-well competition binding assay curve (15 μ g membranes/well, TopCount™). Recommended radioligand concentration = 0.4 nM.

*Even though two sites can be observed occasionally with some ligands, the data presented is derived from single site fitting.

| Reference Compounds | Ki (nM) |
|---------------------|---------|
| Naltriben | 1.6 |
| BNTX | 37 |
| U-69593 | 47145 |

SUGGESTED MATERIALS AND INSTRUMENTATION

| | SUPPLIER | CATALOG # |
|------------------------------------|-------------|---|
| PHARMACOLOGY ASSAYS | | |
| • [³ H]-Naltrindole | PerkinElmer | NET1065 |
| • Naltrexone | Sigma | N3136 |
| • Unifilter-96 GF/C | PerkinElmer | 6005174 |
| • Unifilter-96 GF/B | PerkinElmer | 6005177 |
| • Filtermat A GF/C | PerkinElmer | 1450-421 |
| INSTRUMENTS | | |
| • TopCount™ | PerkinElmer | visit the web site or contact your local sales office |
| • Microbeta® | PerkinElmer | visit the web site or contact your local sales office |
| • FilterMate Unifiter 96-Harvester | PerkinElmer | C961962 |

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