



User's Manual

NMDAR2B Cell-Based ELISA kit



DEIAPY2710



96T



This product is for research use only and is not intended for diagnostic use.

For illustrative purposes only. To perform the assay the instructions for use provided with the kit have to be used.

Creative Diagnostics

 Address: 45-1 Ramsey Road, Shirley, NY 11967, USA

 Tel: 1-631-624-4882 (USA) 44-161-818-6441 (Europe)  Fax: 1-631-938-8221

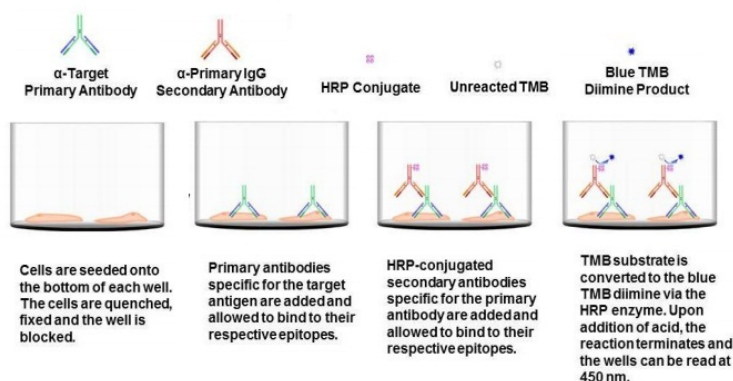
 Email: info@creative-diagnostics.com  Web: www.creative-diagnostics.com

PRODUCT INFORMATION

Intended Use

The NMDAR2B Cell-Based ELISA kit is intended for the qualitative measuring of the relative amounts of NMDAR2B in cultured cells.

Principles of Testing



Materials Required But Not Supplied

1. Microplate reader able to measure absorbance at 450 nm and/or 595 nm for Crystal Violet Cell Staining (Optional)
2. Micropipettes with capability of measuring volumes ranging from 1 μ L to 1 ml
3. 7% formaldehyde or formaldehyde from other sources
4. Deionized or sterile water
5. Squirt bottle, manifold dispenser, multichannel pipette reservoir or automated microplate washer
6. Graph paper or computer software capable of generating or displaying logarithmic functions
7. Absorbent papers or vacuum aspirator
8. Test tubes or microfuge tubes capable of storing ≥ 1 ml
9. Orbital shaker
10. Poly-L-Lysine

Storage

Upon receipt, the kit should be stored at 4°C. The un-opened kit will be stable for up to 6 months from the date of shipment if stored at 4°C.

Plate Preparation

1. **Cell Line:** The cell line must express the target protein. This protocol can be used directly for adherent cells. For suspension cells and loosely attached cells, two steps are required: 1.Coat the plates with 100 µL of 10 µg/ml Poly-L-Lysine to each well of the 96-well plate for 30 minutes at 37°C before proceeding to Step 1 of Assay Protocol. Use 8% formaldehyde to fix the cells on Step 5 of Assay Protocol.
2. **Cell Number and Sensitivity:** The number of cells plated onto the 96-well plates depends on the expression level of NMDAR2B protein in the cells, cell size, treatment conditions and incubation time. The cells used for testing should be around 75-90% confluent. Typically for HeLa cells, seed 30,000 cells per well overnight for treatment the following day. The NMDAR2B Colorimetric Cell-Based ELISA Kit can detect NMDAR2B expression in as little as 5,000 HeLa cells.
3. **Cell Treatment:** The cells can be treated with inhibitors, activators, stimulators (ie. chemicals, proteins/peptides.or a combination of the substances listed above. The cells can be treated with UV and serum starvation to meet the needs of the end-user.
4. **Positive and Negative Controls:** Mouse Anti-GAPDH Antibody (included. should be used to detect the internal positive controls for normalization of OD values of the target protein. The negative controls are HRP-Conjugated Anti-Rabbit IgG Antibody and HRP-Conjugated Anti-Mouse IgG Antibody alone in different wells (without the primary antibodies). Both positive and negative controls should be performed in the same plate with the NMDAR2B target experiments.
5. **Accuracy and Precision:** Each condition should be performed in duplicate or in triplicate.

Precautions

1. Reagents provided in this kit may be harmful if ingested, inhaled or absorbed through the skin.
2. Fixing Solution contains formaldehyde. Formaldehyde is known to be a highly toxic reagent. Personal protection is strongly recommended while working with this chemical.
3. Stop Solution contains 2 N Sulfuric Acid (H₂SO₄) and is an extremely corrosive agent. Please wear proper eye, hand and face protection when handling this material. When the experiment is finished, be sure to rinse the plate with copious amounts of running water to dilute the Stop Solution prior to disposing the plate or strips.
4. Crystal Violet is an intense stain reagent. Avoid contact stain and clothing.

