ELISA PRODUCT INFORMATION & MANUAL

MTS Cell Proliferation Assay Kit (Colorimetric)

NBP2-54884

Enzyme-linked Immunosorbent Assay for quantitative detection. For research use only.
Not for diagnostic or therapeutic procedures.

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NBP2-5488 MTS Cell Proliferation Assay Kit (Colorimetric)

For the rapid, sensitive and accurate measurement of cell proliferation.

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

Precautions

Please read these instructions carefully prior to beginning the assay.

All kit components have been formulated and quality control tested to function successfully as a kit. Modifications to the kit components or procedures may result in loss of performance.

Storage and Stability:

Store kit at -20°C in the dark immediately upon receipt. Briefly centrifuge small vials prior to opening. All kit components are supplied as ready to be used. Keep on ice while in use.

Refer to list of materials supplied for storage conditions of individual components.

Materials Supplied:

Item	100 Tests
Amount (250 tests)	5 mL
Amount (500 tests)	10 mL
Amount (2500 tests)	50 mL
Amount (5000 tests)	100 mL
Amount (10000 tests)	200 mL
Storage Condition	-20°C

For frequent use, kit can be stored at 4˚C for up to 6 weeks protected from light.

Materials Required, Not Supplied

These materials are not included in the kit, but will be required to successfully perform this assay:

- Multi-well spectrophotometer (ELISA reader)
- Colorimetric microplate reader – equipped with filter for OD 490 nm
- 96 well plate: clear, flat bottom plates for colorimetric assay
- Pipettes and pipette tips – preferably a multichannel pipette
- Plate shaker
- SDS

Limitations

- Assay kit intended for research use only. Not for use in diagnostic procedures
- Do not use kit or components if it has exceeded the expiration date on the kit labels
- Do not mix or substitute reagents or materials from other kit lots or vendors. Kits are QC tested as a set of components and performance cannot be guaranteed if utilized separately or substituted.

Technical Hints

- This kit is sold based on number of tests. A 'test' simply refers to a single assay well. The number of wells that contain sample, control or standard will vary by product. Review the protocol completely to confirm this kit meets your requirements. Please contact our Technical Support staff with any questions.
- Keep enzymes and heat labile components and samples on ice during the assay.
- Make sure all buffers and developing solutions are at room temperature before starting the experiment.
- Avoid cross contamination of samples or reagents by changing tips between sample, standard and reagent additions.
- Avoid foaming or bubbles when mixing or reconstituting components.
- Ensure plates are properly sealed or covered during incubation steps.
- Make sure you have the appropriate type of plate for the detection method of choice.
- Make sure the heat block/water bath and microplate reader are switched on before starting the experiment.

Reagent Preparation

Briefly centrifuge small vials at low speed prior to opening.

MTS Reagent: Ready to use as supplied. Equilibrate to room temperature before use. Store at -20°C.

Assay Procedure and detection

Equilibrate all materials and prepared reagents to room temperature prior to use.

It is recommended to assay all controls and samples in duplicate.

1. Culture cells (5 – 100 x 103/well) in a 96-well microtiter plate in a final volume of 200 µL/well. Cells can be cultured in the absence or presence of additional factors for viability/proliferation testing.
2. If cells are cultured in different volume of culture medium, adjust the amount of MTS Reagent accordingly.
3. Incubate cells for 20 – 48 hours. The appropriate incubation time will depend on the individual cell type and cell concentrations used. Therefore, it is recommended to determine the optimal incubation time for a particular experiment.
4. Add 20 µL/well MTS Reagent into each well and incubate for 0.5 – 4 hours at 37°C in standard culture conditions.
5. Shake the plate briefly on a shaker and measure absorbance of treated and untreated cells using a plate reader at OD=490 nm.

***NOTE**: To measure the absorbance at a later time, add 10 µL of 10% SDS to each well to stop the reaction. Store SDS-treated plates protected from light in a humidified chamber at room temperature for up to 18 hours.*

Troubleshooting

Problem	Cause	Solution
Assay not working	Use of ice-cold buffer	Buffers must be at room temperature
	Plate read at incorrect wavelength	Check the wavelength and filter settings of instrument
	Use of a different 96-well plate	Colorimetric: Clear plates Fluorometric: black wells/clear bottom plate
Sample with erratic readings	Cells/tissue samples not homogenized completely	Use Dounce homogenizer, increase number of strokes
	Samples used after multiple free/ thaw cycles	Aliquot and freeze samples if needed to use multiple times
	Use of old or inappropriately stored samples	Use fresh samples or store at - 80°C (after snap freeze in liquid nitrogen) till use
	Presence of interfering substance in the sample	Check protocol for interfering substances; deproteinize samples
Lower/ Higher readings in samples and Standards	Improperly thawed components	Thaw all components completely and mix gently before use
	Allowing reagents to sit for extended times on ice	Always thaw and prepare fresh reaction mix before use
	Incorrect incubation times or temperatures	Verify correct incubation times and temperatures in protocol
Unanticipated results	Measured at incorrect wavelength	Check equipment and filter setting
	Samples contain interfering substances	Troubleshoot if it interferes with the kit
	Sample readings above/ below the linear range	Concentrate/ Dilute sample so it is within the linear range