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ab233471
Lipid Peroxidation (MDA) Assay Kit (Colorimetric)

For the measurement of lipid peroxidation.

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

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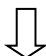
1. Overview

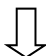
Lipid Peroxidation (MDA) Assay Kit (Colorimetric)(ab233471) offers the most rapid and convenient method to measure malondialdehyde (MDA) without TBARS heating steps. MDA Color Reagent reacts with MDA to generate a blue color product which is measured at 695 nm with absorbance microplate readers. This assay is very fast and specific to MDA with little interference from other aldehydes.

Prepare serially diluted MDA standards and test samples.

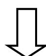
Add 10 µl of MDA Color Reagent stock solution into each well of MDA standard.



Incubate at room temperature for 10-30 minutes.



Add 40 µl of Reaction Solution and incubate at room temperature for 30-60 minutes.



Monitor absorbance increase at 695 nm.

2. Materials Supplied and Storage

Store kit at -20°C immediately upon receipt. Kit can be stored for 1 year from receipt, if components have not been reconstituted.

Aliquot components in working volumes before storing at the recommended temperature.

Avoid repeated freeze-thaws of reagents.

Item	Quantity	Storage temperature (before prep)	Storage temperature (after prep)
MDA Color Reagent	1 vial	-20°C	-20°C
Dilution Buffer	10 mL	-20°C	-20°C
MDA Standard	1 vial	-20°C	-20°C
Reaction Solution	10 mL	-20°C	-20°C

3. Materials Required, Not Supplied

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

- Ultra pure and sterile water (ddH_2O).
- 96-well clear bottom microplates.
- Microplate reader capable of reading absorbance at 695 nm.

4. General guidelines, precautions, and troubleshooting

Please observe safe laboratory practice and consult the safety datasheet.

For general guidelines, precautions, limitations on the use of our assay kits and general assay troubleshooting tips, particularly for first time users, please consult our guide:
www.abcam.com/assaykitguidelines

For typical data produced using the assay, please see the assay kit datasheet on our website.

5. Reagent Preparation

Briefly centrifuge small vials at low speed prior to opening.

5.1 MDA Color Reagent

Ready to use as supplied.

ΔNote: Aliquot into single use size and store at -20°C protected from light.

5.2 Dilution Buffer

Ready to use as supplied.

5.3 MDA Standard

Add 100 µL of ddH_2O into MDA Standard vial to make 100 mM MDA stock solution.

5.4 Reaction Solution

Ready to use as supplied.

6. Standard Preparation

Prepare serially diluted MDA standards

1. Prepare 100 mM MDA stock solution as described above (Reagent Preparation 5.3).
2. Prepare MDA standard dilutions: add 4 µL of 100 mM MDA stock solution (from step 6.1.1.) into 996 µL of Dilution Buffer to obtain 400 µM MDA solution.
3. Perform 1:2 serial dilutions in Dilution Buffer to obtain 200, 100, 50, 25, 12.5, 6.25 and 0 µM serially diluted MDA standards.

7. Sample Preparation

1. Prepare 1:2 serially diluted Test Samples in Dilution Buffer.

8. Assay Procedure

- Equilibrate all materials and prepared reagents to room temperature just prior to use and gently agitate.
- Assay all standards, controls and samples in duplicate.

8.1 Prepare assay plate

Add 50 µl of test samples and serially diluted MDA standards into a 96-well clear bottom microplate as described in the table, below.

Blank Control	Blank Control	Test Sample	Test Sample								
MDA 1	MDA 1	…	…								
MDA 2	MDA 2	…									
MDA 3	MDA 3										
MDA 4	MDA 4										
MDA 5	MDA 5										
MDA 6	MDA 6										
MDA 7	MDA 7										

Blank Control = Dilution Buffer.
MDA 1…MDA 7 = Serially diluted MDA Standard (6.25 to 400 µM, respectively).

8.2 Run MDA assay

1. Add 10 µL MDA Color Reagent solution into each well of MDA Standard, Blank Control, and Test Samples.

   ΔNote: For a 384-well plate, add 25 µL of MDA Standard /Test Sample and 5 µL of MDA Color Reagent stock solution into each well.

2. Incubate the reaction mixture at room temperature for 10-30 minutes.
3. Add 40 µL of Reaction Solution to make the total assay volume to 100 µL/well.

   ΔNote: For a 384-well plate, add 20 µL of Reaction Solution to make the total assay volume 50 µL/well.

4. Incubate the final reaction mixture at room temperature for 30-60 minutes.
5. Monitor absorbance increase with an absorbance plate reader with path-check correction at OD of 695~700 nm.

9. Data Analysis

The absorbance reading in blank wells (with Dilution Buffer only) is used as a control, and is subtracted from the value of those wells with the standards and test samples.

Calculate Test Sample MDA concentrations by reference to the MDA Standard Curve correcting for the Test Sample dilution.

10. FAQs / Troubleshooting

General troubleshooting points are found at
www.abcam.com/assaykitguidelines.

11. Typical Data

Data provided **for demonstration purposes only**.

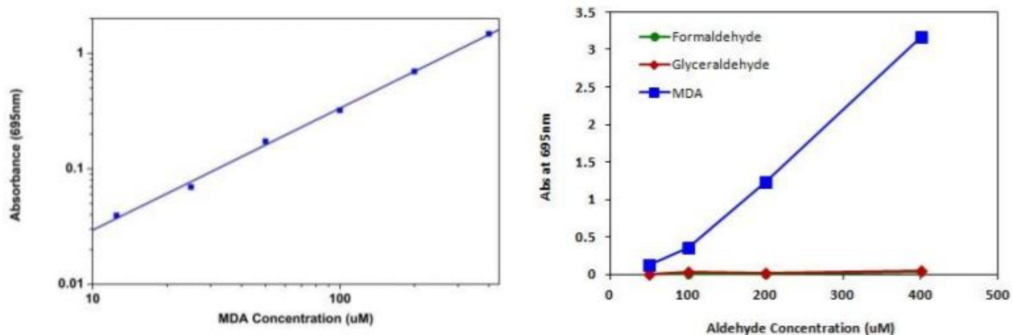


Figure 1. MDA dose response was measured using Lipid Peroxidation (MDA) Assay Kit (Colorimetric)(ab233471) in a 96-well clear bottom microplate. MDA standard curve (left). Signal comparison of MDA, Formaldehyde and Glyceraldehyde (right).

12. Notes

Technical Support

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