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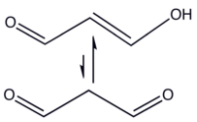
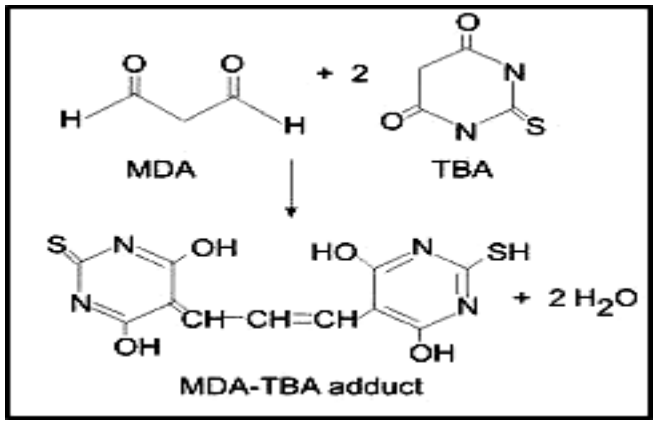


Malondialdehyde (MDA) Assay kit (48 Tests)

INTRODUCTION

Biocore Diagnostik (ZellBio) MDA assay Kit provides a simple, reproducible, and standardized tool for assessment of lipid peroxidation in biological sample e.g. **plasma, serum, urine, tissue homogenates, and cell lysates**. The MDA-TBA adduct formed by the reaction of MDA and TBA under high temperature. Malondialdehyde is measured in acidic media and heat (90-100°C) colorimetrically at 532 (530-540 nm).



Kit Contents (ZB-MDA96):

1. Reagent 1: BCD-R1 (contains TBA) 500mg (MDA31).
2. Reagent 2: BCD-R2 (contains 5X of HOAC) 10mL (MDA32).
3. Reagent 3: BCD-R3 (contains Standard 500 µM) 0.25mL (MDA33).
4. Reagent 4: BCD-R4 (contains10X Alkali) 5mL (MDA34).
5. Reagent 5: BCD-R5 (contains Detergent) 6mL (MDA35).
6. BCD-R6 (contains microplate) 1 plate (MDA36)

Assay Range:
Biocore (ZellBio) MDA assay kit can be use for MDA determination in range of 0.78 - 50µM.

Assay Sensitivity:
Biocore (ZellBio) MDA assay kit can determine MDA in biological samples with 0.1µM sensitivity.

Assay Precision:
Human serum sample with replication No.10 showed the intra and inter assay coefficient of variation 5.8% and 7.6% respectively.

Reagent preparation:

1) R2 ready reagent: Add 40mL distilled water to 10mL BCD-R2.
2) R4 ready reagent: Add 45mL distilled water to 5mL BCD-R4.
3) Chromogenic reagent: Mix 500mg of BCD-R1 with 50mL R2 ready reagent and 50mL R4 ready reagent. Warm slowly until powder dissolves completely. This reagent stable for a day. So, it can prepare the Chromogenic solution as it need with the same proportion. For example 250mg R1+25mL ready R2 and 25ml Ready R4.
4) Standard solutions: The stock standard is 500µM; by serial dilution prepare 50µM, 25µM, 12.5µM, 6.25µM, 3.12µM, 1.56µM and 0.78µM.

Assay Protocol:

1. All reagents (except samples) must be equilibrated to RT. Shake the unknown sample for homogenation well.
2. Add 100µL standards/samples to related name test tubes.
3. Add 100µL R4 reagent (if it is cloudy, warm until to become a clear solution).
4. Add 200µL ready Chromogenic solution.
5. Heat the above mixture for one hour at boiling water bath (pink color formation).
6. Cool the above tests tube in ice bath and centrifuge them 10min around 10,000 rpm.
7. Pipette 200µL of pink color supernatant into the microplate.
8. Read the absorbance at 535nm.
9. Calculate MDA level in unknown samples based on standard curve which drown using standard points' absorbance.

References

1) Dawn-Linsley, M., Ekinci, F.J., Ortiz, D., et al. Monitoring thiobarbituric acid- reactive substances (TBARs) as an assay for oxidative damage in neuronal cultures and central nervous system. J. Neurosci. Meth. 141, 219-222 (2005).
2) Yagi, K. Simple assay for the level of total lipid peroxides in serum or plasma. Methods in Molecular Biology 108, 101-106 (1998).
3) Ohkawa, H., Ohishi, N., and Yagi, K. Assay for lipid peroxides in animal tissues by thiobarbituric acid reaction. Anal. Biochem. 95, 351-358 (1979).