

# LIPID PEROXIDE ( Malondialdehyde )

**Colorimetric Method  
For Research Only**

**25 Tests**

## PRINCIPLE :

Thiobarbituric acid (TBA) reacts with malondialdehyde (MDA ) in acidic medium at temperature of 95°C for 30 min to form thiobarbituric acid reactive product the absorbance of the resultant pink product can be measured at 534 nm.

## SAMPLE :

Serum, Urine and appropriate concentration of tissue homogenate.

## REAGENTS :

1.	Standard	10 nmol / mL
2.	Chromogen Thiobarbituric acid Detergent Stabilizer	25 mmol / L

## STABILITY:

Stable until the expiry date specified when stored at +4 to +8 °C.

## PROCEDURE:

	Sample ml	Standard ml	Blank ml
Sample	0.2	-	-
Standard	-	0.2	-
Chromogen	1.0	1.0	1.0

Mix well, cover the test tube with glass bead, heat in boiling water bath for 30 min, COOL , then add :

Sample	-	-	0.2
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Mix, read the absorbance of sample ( $A_{\text{Sample}}$ ) against blank and standard against d. water at 534 nm. Color stable for 6 hrs. Linearity up to 100 nmol/ ml .

## CALCULATION :

Malondialdehyde in sample :

$$\text{Serum} = \frac{A_{\text{Sample}}}{A_{\text{Standard}}} \times 10 \quad \text{nmol / ml}$$

$$\text{Tissue} = \frac{A_{\text{Sample}}}{A_{\text{Standard}}} \times \frac{10}{\text{g. tissue used}} \quad \text{nmol / g.tissue}$$

## REFERENCE :

- Satoh K. , Clinica Chimica Acta ( 1978 ) , 90 , 37 .
- Ohkawa, H. , Ohishi W, and Yagi K. Anal . Biochem ( 1979 ) 95 , 351
- EL-Aaser A.A, and Saleh T.R. (unpublished)

## SAMPLE PREPARATION

### Tissue Homogenate

1. Prior to dissection, perfuse tissue with a PBS ( phosphate buffered saline ) solution, pH 7.4 . containing 0.16 mg / ml heparin to remove any red blood cells and clots.
2. Homogenize the tissue in 5 – 10 ml cold buffer ( i , e , 50 mM potassium phosphate, pH 7.5.) per gram tissue.
3. Centrifuge at 4000 r.p.m for 15 minutes.
4. Remove the supernatant for assay and store on ice. If not assaying on the same day , freeze the sample at - 80°C. The sample will be stable for at least one month.

### Plasma

1. Collect blood using an anticoagulant such as heparin or citrate .
2. Centrifuge the blood at 3,000 rpm for 10 min. at 4°C. pipette off the top yellow plasma layer without disturbing the white buffy layer. Store plasma on ice until assaying or freeze at -80°C. The plasma sample will be stable for at least one month.

### Serum

1. Collect blood without using an anticoagulant. Allow blood to clot for 30 min. at 25°C.
2. Centrifuge the blood at 4,000 rpm for 15 min. at 4°C. pipette off the top yellow serum layer without disturbing the white buffy layer. Store serum on ice if not assaying the same day, freeze at -80°C. The sample will be stable for at least one month.

### Urine

Collect urine in a clear beaker or flask and store on ice. If not assaying the same day, freeze at -80°C.

**BIO DIAGNOSTIC**  
DIAGNOSTIC AND RESEARCH REAGENTS

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### Colorimetric Method

+4 to +8 °C

25 Tests

CAT. No.

MD 25 29

**FOR RESEARCH USE ONLY**

## REAGENTS

R1	Standard	5 ml
R2	Chromogen	50 ml

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