

## Original Research Article

# OXIDATIVE STRESS MARKERS IN TYPE II DIABETES: MDA LEVELS AND CLINICAL ASSOCIATIONS

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### ABSTRACT

Diabetes mellitus (DM) is expected to affect 550 million people globally. Chronic hyperglycemia in DM leads to complications such as atherosclerosis, strokes, nephropathy, neuropathy, and retinopathy. It induces oxidative stress, increasing free radicals and decreasing antioxidants. Malondialdehyde (MDA) is a marker of this oxidative stress. This study examines the association between MDA, fasting blood glucose (FBG), and glycated hemoglobin (HbA1C) levels in diabetic patients, and the correlation with diabetes duration and complications. The study was conducted at RDJM Medical College, Turki, Muzaffarpur, India, from March 2023 to April 2024, the study involved 200 subjects. These were divided into two groups: 100 healthy controls and 100 Type II diabetics, the latter group further split into 50 non-complicated diabetics and 50 diabetics with nephropathy. Result showed significantly higher levels of FBG, HbA1C, and MDA in diabetics compared to healthy controls, with the highest levels in those with nephropathy. Microalbuminuria was also elevated in complicated diabetics.

The study concludes that diabetic patients, particularly those with nephropathy, experience increased oxidative stress. This stress worsens with the duration of diabetes, emphasizing the need for early intervention and continuous monitoring.

**Keywords:** Diabetes, complications, oxidative stress, MDA

## INTRODUCTION

Diabetes mellitus (DM) is expected to affect around 550 million people all over the world according to global estimates of the prevalence of diabetes.<sup>[1]</sup> DM is characterized by constant hyperglycemia that damages various organs and manifests in macro vascular complications like premature atherosclerosis resulting in strokes, peripheral vascular disease, and myocardial infarctions and micro vascular complications such as nephropathy, neuropathy, and retinopathy.<sup>[2]</sup> Hyperglycemia causes oxidative stress in T2DM through several mechanisms, including glucose autoxidation, nonenzymatic protein replication, polyol pathway activation, glycolysis pathway and pentose phosphate pathway.<sup>[3-5]</sup> Oxidative stress occurs due to decreased concentration or antioxidant activity and increased production of free radicals reactive

oxygen species.<sup>[6]</sup> Malondialdehyde (MDA) is a stable end product of lipid peroxidation.<sup>[7]</sup> It is a three-carbon aldehyde that can exist in various forms in an aqueous solution. Serum MDA has been used as a biomarker of lipid peroxidation and has served as an indicator of free radical damage.<sup>[8]</sup>

Diabetic nephropathy is the most common cause of end-stage renal disease. If untreated, 80% of people who have type-1 diabetes and microalbuminuria will progress to overt nephropathy (i.e. proteinuria characterized by > 300 mg albumin excreted daily, whereas only 20-40% of those with type 2 diabetes over a period of 15 years will progress. Diabetic nephropathy has several distinct phases of development. Kavas GL (2009), concluded that functional changes occur in the nephron at the level of glomerulus, including podocyte foot process effacement, decrease in podocyte number, thickening of the glomerular basement membrane

and mesangial expansion, all occur with the early changes.<sup>[9]</sup>

**Aims and objectives**

The present study was planned to study the association between Malondialdehyde (MDA), fasting blood glucose and glycated hemoglobin (HbA1C) levels among diabetic patients attending medicine and to determine the correlation, if any between MDA levels with duration and complications of diabetes in patients with diabetes mellitus.

## MATERIAL AND METHODS

**Study Area:** The study was conducted in the Department of Biochemistry at RDJM Medical college, Turki, Muzaffarpur.

**Study period:** Study was conducted from March 2023 to April 2024.

**Study Population:** Subjects included for the study were categorised into 2 groups.

100 age-matched healthy controls were taken with respect to Type II Diabetics were included in group 1. Total 100 type-II diabetic patients were taken in group 2 and further divided into two sub groups. 50 noncomplicated Type-II diabetic patients in subgroup 2A and 50 complicated Type-II diabetic patients with nephropathy as a complication in subgroup 2B.

Blood Glucose was estimated by GOD- POD method<sup>10</sup>, Glycosylated haemoglobin by Ion exchange resin method,<sup>[11]</sup> Microalbuminuria by Pyrogallol red method (end point),<sup>[12]</sup> and MDA by method of *Ohkawa et al.*,<sup>[13]</sup> Since MDA is not stable, MDA standard was prepared from 1,1,3,3-Tetramethoxypropane (TMOP). It is hydrolyzed during the acid incubation step at 45°C, which will generate MDA. To each test tube, 0.5 ml of plasma, 0.5 ml of normal saline, 1ml of 20% Trichloroacetic acid (TCA) and 0.25 ml of TBA reagent (200 mg of Thiobarbituric acid in 30 ml distilled water and 30

ml of Acetic acid) were added. The test tubes were kept boiling at 95°C for 1 hour. To each of the test tubes 3 ml of n- Butanol was added and mixed well. The tubes were centrifuged at 3000 rpm for 10 minutes. The separated Butanol layer was collected and read in a colorimeter against reagent blank at 540 nm. The MDA concentration was expressed in terms of µmol/L.

## RESULTS AND DISCUSSION

The study showed the significant increase in the fasting blood glucose (FBG), glycated haemoglobin (HbA1c) and Malondialdehyde (MDA) levels in noncomplicated and complicated diabetics with nephropathy as compared to healthy controls. The microalbuminuria levels were also significantly higher in complicated diabetics with nephropathy compared to noncomplicated cases. The present study is in agreement with the various authors in their studies like Bhatia S et al,<sup>[14]</sup> Apakkan AS et al,<sup>[15]</sup> and Kornelia Z et al,<sup>[16]</sup> who all have shown the significant higher levels of MDA in complicated diabetics with nephropathy and noncomplicated diabetics compared to healthy controls. MDA levels have also been reported to be elevated in other complications of diabetics like neuropathy, retinopathy, coronary heart disease, hypertension etc by various authors like Sawant MJ et al,<sup>[17]</sup> Vivian ST et al.<sup>[18]</sup>

Further detailed analysis of the data revealed that the Pearson's correlation coefficient increased from noncomplicated to complicated diabetics as far as HbA1C, MDA and Microalbuminuria were concerned. The same held for the disease duration. Therefore, the present study has shown that the MDA level is also increased with duration of diabetes which is also observed in the studies of various authors like Vivian ST et al,<sup>18</sup> Bhatia et al.<sup>[14]</sup>

**Table 1: Mean value, standard deviation and p value of age, duration, FBG, HbA1C, MDA, and microalbuminuria between the healthy control & noncomplicated and complicated Type-II diabetic patients**

GROUP	N	Mean±SD	P Value
<b>AGE</b>			
<b>Controls</b>	100	43.32±13.20	
<b>Non-complicated</b>	50	52.00±11.25	<0.001
<b>Complicated</b>	50	58.02± 10.80	
<b>FBG</b>			
<b>Controls</b>	100	79.40±6.97	
<b>Non-complicated</b>	50	178.03±71.77	<0.001
<b>Complicated</b>	50	198.78± 68.41	
<b>HbA<sub>1c</sub></b>			
<b>Controls</b>	100	4.73±0.44	
<b>Non-complicated</b>	50	7.05±0.40	<0.001
<b>Complicated</b>	50	8.36±1.27	
<b>MDA</b>			
<b>Controls</b>	100	1.23±0.25	
<b>Non-complicated</b>	50	2.35±0.23	<0.001
<b>Complicated</b>	50	4.49±1.27	
<b>Microalbuminuria</b>			
<b>Controls</b>	-	-	-
<b>Non-complicated</b>	50	19.78±5.09	<0.001
<b>Complicated</b>	50	766.87±694.24	
<b>Duration</b>			
<b>Controls</b>	-	-	-
<b>Non-complicated</b>	50	1.85±1.63	<0.001
<b>Complicated</b>	50	8.96±5.38	

## CONCLUSION

The study concludes that diabetic patients suffer more from oxidative stress compared to healthy control. Oxidative stress is still higher in diabetic patients with nephropathy than diabetics without nephropathy.

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