



## Original Research Article

## Effect of yoga practicing on Ischemia modified albumin, lipid profile and antioxidant enzymes in Type 2 Diabetes Mellitus

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## ARTICLE INFO

## Article history:

Received 11-08-2019

Accepted 31-08-2019

Available online 14-12-2019

## Keywords:

T2DM

Ischemia modified albumin

Lipid profile

Oxidative stress

## ABSTRACT

**Introduction:** Type 2 diabetes mellitus (T2DM) is the leading cause of death among non-communicable diseases. The cause for T2DM is multi-factorial and the consequence of hyperglycaemia are many like altered lipid profile, increased oxidative stress, increased amount of altered albumin in circulation i.e., ischemia modified albumin (IMA). All these alterations would lead to microvascular and macrovascular complications. Therefore treating T2DM with hypoglycaemic drugs and insulin alone is failing to achieve the long term glycaemic control. Hence there is need for supportive therapy like yoga. The aim of the study was to determine the effect of yoga practicing on IMA, lipid profile and antioxidant enzymes in T2DM patients on oral hypoglycaemic drugs (OHD).

**Materials and Methods:** This was a prospective cohort study, conducted from June 2017 to October 2018. Two hundred T2DM patients were included in the study. In group I 100 T2DM patients on Metformin and Glimipride (500 mg BD + 2 mg OD, respectively) along with yoga practicing were included and in group II there were 100 T2DM patients of age and duration of T2DM matched with out practicing yoga. The levels of Fasting plasma glucose (FPG), Post prandial plasma glucose (PPPG), Glycated hemoglobin (HbA1c), Ischemia modified albumin (IMA), Lipid profile (TC, TG, LDL-C, VLDL-C, HDL-C), lipid peroxidation (Malondialdehyde levels) and activity of enzymatic antioxidants (Superoxide dismutase, catalase, Glutathione peroxidase) were measured in both groups.

Glycemic profile (FPG, PPPG, HbA1C, IMA), Lipid profile and oxidative stress were comparable in patients of both groups. Patients on oral hypoglycemic drugs (OHD) and yoga practicing showed significantly lower levels of FPG, PPPG, HbA1C, IMA, TC, TG, LDL-C and MDA and significantly increased levels of HDL-C, SOD, Catalase and GPx in comparison with T2DM patients those who were only on OHD.

Reduced Oxidative stress, decreased lipids profile and Glycemic control was seen in yoga practicing T2DM patients than in those who were only on OHD. Practicing of yoga with OHD in T2DM patients will improve the glycemic index, reduce risk of MI and help to prevent the CVD complications. Hence this study suggests to practice yoga with OHD to reduce further complications of T2DM.

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## 1. Introduction

Diabetes mellitus (DM) is fast becoming a potential epidemic in India with more than 62 millions of diabetic individuals.<sup>1</sup> Currently, India has the highest number of people with diabetes mellitus followed by China and United State.<sup>1</sup> In India, the epidemic of type 2 diabetes

mellitus (T2DM) is increasing and there exists a shift in diabetes prevalence from urban to rural areas, older to younger people and from the affluent to the less privileged.<sup>2</sup> After seeing the increase in the incidence of diabetes WHO has declared India as diabetic capital. The cause for T2DM is multi factorial and includes both genetic and environmental factors. The non modifiable factors like, obesity, age, lifestyle factors, particularly physical

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inactivity, over nutrition are primarily responsible for the current global diabetes epidemic.<sup>3</sup> In diabetic complications there exists the relationship between hyperglycemia, dyslipidemia and oxidative stress.<sup>4</sup> Dyslipidemia is the major risk factor for cardiovascular complications in T2DM.<sup>5–7</sup> The lipid change that occur in T2DM is because of the increased flux of free fatty acids caused by insulin resistance.<sup>5,8</sup>

Hyperglycaemia cause oxidative stress and this leads to structural modification of the circulating albumin in serum. This modified albumin is known as ischemia modified albumin (IMA). It is considered as new biomarker for T2DM.<sup>9</sup> It is good indicator for severity of diabetic complications.<sup>10</sup>

The effect of yoga practicing on IMA in T2DM has been not yet clear.

Oxidative stress plays an important role in the development of both microvascular and macrovascular complications of T2DM.<sup>10</sup> The oxidative stress causes glucose intolerance and insulin resistance leading to more atherosclerotic complications.<sup>5</sup>

Yoga is the popular form of exercise all around the world. It is considered as an exercise to mind and body. Physical exercise has been considered as one of the corner stone in the treatment of diabetes mellitus along with nutrition and medications from the past 100 years. Many clinical studies are there which tells us the benefits of yoga. Practising of yoga regularly will delay the progression of type 2DM by decreasing the oxidative stress and increasing the levels of antioxidants.<sup>11,12</sup> It increases insulin sensitivity.<sup>13</sup> As per the data published, the exercise based research for T2DM patients are less in India.

## 2. Aims and Objectives

To study the effect of yoga practicing on IMA, lipid profile and antioxidant enzymes in T2DM patients.

### 2.1. Objectives

1. To estimate the levels of lipid profile and IMA in T2DM patients with and without yoga practicing.
2. To measure the Malondialdehyde (MDA) or lipid peroxidation (LPO) and activity of antioxidant enzymes in T2DM patients with and without yoga practicing.

## 3. Materials and Methods

### 3.1. Study design

This was an interventional study.

### 3.2. Setting

Department of Biochemistry at BIMS, Belagavi from June 2017 to October 2018.

### 3.3. Selection of participants

The study subjects were selected from the medicine Out Patient Department (OPD), BIMS hospital, Belagavi attending for Diabetic check up. The guidelines of the National Diabetes Data Group and the third set of the Adult Treatment Panel of the National Cholesterol Education Program (NCEP ATP III) were used to recruit 20 0 patients with T2DM and dyslipidemia.

### 3.4. Interventions

The study subjects were divided into 2 groups,

Group 1 : 100 T2DM on OHD + Yoga practicing (6months).

Group 2 : 100 T2DM on OHD (oral hypoglycaemic drugs- Metformin 500mg BD + Glimipride 2mg OD).

### 3.5. Inclusion criteria

T2DM patients included in the study were diabetic  $\leq$  10 years and without microvascular and macrovascular complications. All these patients were in the age group 45-60 years.

### 3.6. Exclusion criteria

T2DM patients who were smokers, alcoholics, known neuropathy, retinopathy, nephropathy, hypothyroidism, cerebrovascular disease and coronary vascular disease were excluded. The study was approved from the institutional ethical committee and informed consent taken from all participants.

The detailed history was taken from all patients including age, gender, duration of T2DM, complications of diabetes, treatment and BMI was measured. Baseline Biochemical investigations were done for all patients. T2DM patients were divided into 2 groups. Group I was T2DM on OHD and yoga practicing daily one hour in the morning for 6months and Group II was T2DM on OHD alone.

The yoga class was conducted by Mr. Nagaraj Hosamani, a certified yoga teacher at Shree jaya yoga center, Belagavi.

### 3.7. Biochemical analysis

The biochemical analysis was done in both groups after 6 months of yoga practicing and OHD treatment. 15 hours fasting blood samples used for all investigations. 5ml of blood samples collected in plain and EDTA vacutainers from every patient under strict aseptic conditions. The EDTA sample was analysed for Glycated Haemoglobin (HbA1c) and plain sample for Fasting plasma glucose (FPG) and lipid profile. FPG was estimated by Glucose oxidase peroxidase method, HbA1c by HPLC method, total Cholesterol was estimated by modified Roeschlav's method<sup>14</sup> and Triacylglycerol by the method of Wako modified by Mc Gowan et al.<sup>15</sup> HDL cholesterol

**Table 1:** Details of yoga practicing in T2DM patients

Each yoga session consists of	
Bhastrika Pranayam	3 minutes per day
Anuloma viloma	3-5 minutes per day
Kapalbhati Pranayam	10-15 minutes per day
Brahmari Pranayam	5 times per day
Suryanamaskaram (sun salutation)	5 turns per day
Yoga Mudrasana(psychic union pose)	5 minutes per day
Vajrasana (thunderbolt pose)	
Vakrasana (twisted pose)	
Paschimottasana (seated forward bend)	
Pavanamuktasana (wind relieving pose)	
Sashankasana (hare pose),	
Ushtrasana (camel pose)	Each asana practiced for 2 times per day
Bhujangasan (cobra pose)	
Dhanurasana (bow pose)	
Arthakatchakrasana (lateral arc pose)	
Parivatha trilokasana (revolved triangle pose)	
Shavasana (corpse pose)	5 minutes per day
Meditation (one-one meditation/ breath counting meditation)	10 minutes per day

was assessed by phosphotungstic acid method.<sup>16</sup> LDL cholesterol was calculated from the formula:  $\text{LDL cholesterol} = \text{Total cholesterol} - [\text{HDL cholesterol} + \text{TG}/5]$ . The 2 hour postprandial blood sample was collected for analysis of postprandial plasma glucose (PPPG). For serum IMA estimation approximately 0.5ml of the serum separated from the blood in plain vacutainer was transferred into small Eppendorf tube and stored at  $-20^{\circ}\text{C}$  until analysis for IMA estimation. Serum IMA was determined manually by the albumin cobalt binding assay, the method described by Bar-Or et al.<sup>9</sup> The results were given in absorbance units (ABSU). The packed cells were used for analysis of MDA, SOD, GPx and catalase. MDA was determined as the measure of thiobarbituric acid reactive substances (TBARS).<sup>17</sup> SOD activity was determined in the hemolysate by the method of Mishra and Fridovich based on the inhibition of autoxidation of epinephrine to adrenochrome at pH 10.2.<sup>18</sup> Catalase activity was measured by the method of Beer and Seazer<sup>19</sup> and GPx activity by Paglia and Valentine in erythrocytes.<sup>20</sup>

### 3.8. Statistical analysis

Data collected were entered in Microsoft Excel and SPSS Version 20 was used for analysis. Paired and unpaired t tests were employed to compare measures. A p value of  $<0.05$  as significant and p value of  $<0.01$  was considered highly significant.

## 4. Results

The Mean age of all the study subjects was  $50 \pm 6$  years. The mean BMI of Group 1 was  $23.81 \pm 2.01$  and in Group 2 was  $24.25 \pm 2.14$ . The BMI in the yoga practicing group is

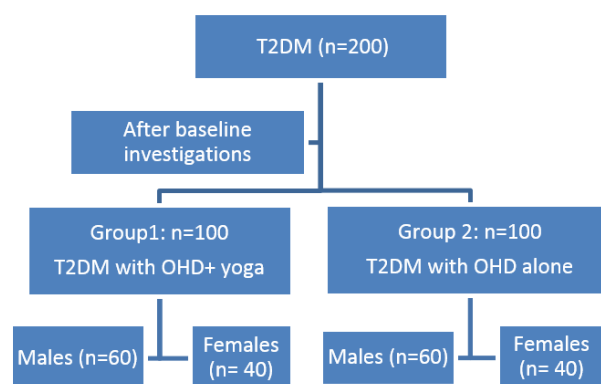


Chart 1:

decreased when compared to T2DM patients on OHD alone. This shows that regular practice of yoga helps in reducing BMI in the T2DM patients.

Table no II shows that the values of HbA1c, IMA, total cholesterol, triacylglycerides, serum LDL levels were significantly decreased in the Group I when compared to Group II. The serum HDL levels were significantly increased in Group I than in Group II. The fasting plasma glucose, 2-hr postprandial plasma glucose and serum VLDL were also decreased in T2DM with yoga practicing group than in the T2DM without yoga practice group. This indicates that lipid profile and glycaemic status of T2DM improve on practicing yoga regularly along with taking OHD medications.

**Table 2:** Demographic profile of T2DM patients

Parameters (mean $\pm$ SD)	T2DM with yoga practicing(OHD +yoga intervention)Group I (n=100)	T2DM without yoga practicing (OHD only)GROUP II (n= 100)
	Males (60)	Females( 40)
Age (years)	45.42 $\pm$ 10.32	45.02 $\pm$ 9.4
BMI ((kg/m <sup>2</sup> )	25.03 $\pm$ 2.14	22.9 $\pm$ 2.15

**Table 3:** Levels of Glucose, IMA and lipid profile in T2DM

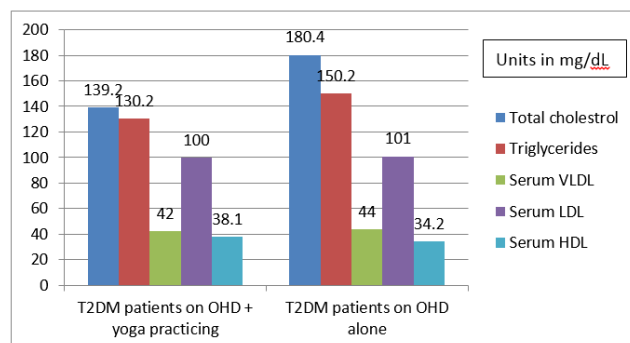
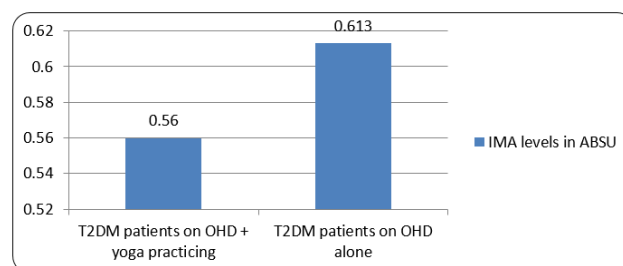
Parameters Mean $\pm$ SD	T2DM with yoga practicing(OHD +yoga intervention)Group I (n=100)	T2DM without yoga practicing (OHD only) Group II (n= 100)
Fasting plasma glucose (FPG) (mg/dl)	100.19 $\pm$ 6.05	110 $\pm$ 10.2*
Postprandial plasma glucose (mg/dl)	210.4 $\pm$ 21.3	230.4 $\pm$ 62.4*
HbA1c (%)	5.5 $\pm$ 0.2	6.3 $\pm$ 0.1*
IMA (ABSU)	0.56 $\pm$ 0.2	0.613 $\pm$ 0.7*
Serum Total Cholesterol (mg/dl)	129.2 $\pm$ 0.6	180.4 $\pm$ 10.5*
Serum Triacylglycerides (mg/dl)	130.2 $\pm$ 20.7	150.2 $\pm$ 16.7*
Serum VLDL – C (mg/dl)	42 $\pm$ 1	44.2 $\pm$ 3
Serum LDL-C (mg/dl)	100 $\pm$ 0.8	105 $\pm$ 23*
Serum HDL-C (mg/dl)	38.1 $\pm$ 0.7	34.2 $\pm$ 9.3*

\*p Value &lt;0.001

**Table 4:** Lipid peroxidation and antioxidant enzymes in T2DM

Parameters Mean $\pm$ SD	T2DM with yoga practicing(OHD +yoga intervention) Group I (n=100)	T2DM without yoga practicing(OHD only) Group II (n= 100)
MDA (n moles/gm of Hb)	3.30 $\pm$ 0.30	4.99 $\pm$ 0.45*
SOD (IU/gm of Hb)	1705 $\pm$ 250	1560.00 $\pm$ 328.32*
Catalase (nmole/H <sub>2</sub> O 2 decomposed mg protein/min)	550 $\pm$ 95	333 $\pm$ 85.74*
GPx (IU/gm of Hb)	51 $\pm$ 4.95	41.24 $\pm$ 4.09*

\*p Value &lt;0.001

**Fig. 1:** Bar diagram of lipid profile in T2DM patients**Fig. 2:** IMA levels in T2DM

## 5. Discussion

Diabetes mellitus being the major health problem in India, many studies are going on to understand the pathophysiology and ways to prevent the diabetic complications.<sup>21–23</sup> The estimated expenditure of diabetic person on health is nearly 4 times higher than that of healthy person. So, now

a day the treatment modalities of T2DM includes not only antidiabetic medications but also supportive exercise, yoga practicing, dietary modifications, life style changes will be helpful to control T2DM. The present study was done to know the effect of yoga practicing on lipid profile, IMA and antioxidants in patients with T2DM. The lipid abnormalities that occur in T2DM is because of insulin resistance or deficiency that affects key enzymes and pathways in lipid metabolism.<sup>5</sup> It has been found that the lipid particles

in diabetic dyslipidemia are more atherogenic causing cardiovascular complications.

Present study found that there was significant decrease d level of total cholesterol, LDL cholesterol, Triacylglycerol and significant increase d levels of HDL cholesterol in T2DM patients practicing yoga with OHD on comparison with T2DM taking only OHD. Yoga practicing reduce lipid profile levels by improving the hepatic lipoprotein lipase enzyme activity. Thus there is increased triacylglyceride uptake by adipose tissue.<sup>24,25</sup> Practicing of yoga in T2DM patients may increase activity of enzyme LCAT or influence the synthesis of apo AI which might be responsible for raised levels of HDL-C in T2DM patients those who were practicing yoga with OHD medications. Hence there may be reduction of LDL-C levels in T2DM patients practicing yoga with OHD medications than in T2DM patients with OHD alone. Arati et al<sup>26</sup> showed that yogasana and pranayama could be used as a supportive therapy to improve lipid profile, cardiovascular parameters and respiratory functions. Similar observations were also noted by Sahay and Bijlani,<sup>27,28</sup> Savita S,<sup>29</sup> Malhotra<sup>30</sup> and Avnish K Upadhyay<sup>31</sup> who reported a significant reduction in free fatty acids(FFA), VLDL, LDL and increase in HDL.

Yoga is an effective therapy to reduce levels of plasma glucose, HbA1C and improvement in hemoglobin in T2DM.<sup>32</sup> Present study also found that regular practicing of yoga significantly reduce d BMI, plasma Glucose, HbA1c and IMA in T2DM patients taking OHD and practicing yoga regularly than only OHD T2DM patients. These findings were similar to Cerranque et al who tested biochemical and haematological profile in T2DM patients and found out that there was a significant decrease in Plasma glucose levels in yoga practitioners than in ordinary subjects.<sup>33</sup>

Hegde et al<sup>11</sup> found that there was significant improvement in the levels of plasma glucose after practising yoga for 3 months. This low glucose causes reduction in lipid profile and reduce the risk for diabetic complications. The mechanisms by which yogasana decreases plasma glucose levels is not yet clarified. Yoga practicing can causes the regeneration of pancreatic cells which will increase utilization and peripheral uptake of glucose. Yoga can increase expression of insulin receptors on the muscles which leads to increase peripheral uptake of glucose by muscles. Thus there may be reduction of glucose levels.

Present study found that IMA was significantly reduced in OHD with yoga in comparison with OHD T2DM patients. In T2DM there is continuous exposure of albumin to hyperglycemia and oxidative stress which may increase IMA in T2DM patients. This indicates that there is reduction in risk of diabetic complications. Our findings goes in accordance with Piwowar et al who showed that IMA levels were significantly higher in T2DM patients with complication and was positively related to HbA1c levels.<sup>34</sup> Ukinc et al,<sup>35</sup> Nehal Hamdy El Saida et al<sup>36</sup>

also reported that IMA was a significant discriminator for diabetic complications with high 100% sensitivity and 100% specificity.

In present study there was reduction in levels of MDA in T2DM with OHD and yoga practicing than only OHD T2DM. The activity of erythrocyte antioxidant enzymes SOD, GPx and catalase were significantly increased in yoga practicing T2DM than on OHD only. Our findings were in agreement with Manna I, who reported a significant reduction in MDA and significant elevation in activity of SOD, catalase and GPx in yoga practicing individuals.<sup>37</sup>

Also Singh S et al,<sup>38</sup> Mahapure HH et al,<sup>39</sup> Gordon LA et al,<sup>40</sup> Hegde SV et al<sup>11</sup> have reported that yoga could be used as a effective therapy to control hyperglycemia and oxidative stress in T2DM patients along with routine treatment. Present study indicates that there was significant reduction in plasma glucose, IMA, HbA1C and increased activity of antioxidant enzymes which may reduce lipid peroxidation in T2DM patients those who were practicing yoga with OHD.

## 6. Conclusions

In present study there was reduction in levels of TAG, LDL-C, TC, IMA, HbA1C and lipid peroxidation in T2DM practicing yoga with OHD in comparison with T2DM with OHD only. The study also found that increased levels of HDL-and activity of enzymatic antioxidants in T2DM practicing yoga with OHD in comparison with T2DM with OHD alone. The study suggests that practicing of yoga with OHD is more beneficial to reduce the diabetic complications and reduce risk of CVD in T2DM patients.

## 7. Limitations

Diet modifications were not considered.

## 8. Acknowledgement

I would like to thank the yoga teacher Mr Nagaraj Hosamani, the institution and all the participants involved in the study.

## 9. Author Contributions

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by Dr Priya Patil, Dr Padmaja Nikam, Dr Shashikant Nikam. The diagnosis, treatment and counselling of T2DM subjects was done by Dr Naren Nimbal. The first draft of the manuscript was written by Dr Priya Patil and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

## 10. Ethical approval

All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional (Belagavi Institute of Medical Sciences, Belagavi) and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

This article does not contain any studies with animals performed by any of the authors.

## 11. Informed consent

Informed consent was obtained from all individual participants included in the study

## 12. Conflict of interest

We (Authors) declare that we have no conflict of interest.

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**Cite this article:** Patil P, Nikam P, Nimbal N, Nikam S. Effect of yoga practicing on Ischemia modified albumin, lipid profile and antioxidant enzymes in Type 2 Diabetes Mellitus. *Int J Clin Biochem Res* 2019;6(4):546-552.