

Original Research Article

## Association of Glycemic Indices and Hs-CRP with Dietary Energy and Fat Consumption in Patients with Type 2 Diabetes Mellitus

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### ABSTRACT

Nutritional etiology of metabolic stress in diabetes has been well explored and documented. However, influence of dietary habits and glycemic control on severity of inflammation among type 2 diabetes patients is sparse. This study investigated the association between dietary nutrient intake, dietary habits, glycemic control and serum hs-CRP in type 2 diabetes mellitus patients. A total of 160 uncontrolled type 2 diabetic patients [81 (50.6%) vegetarians, 79 (49.4%) non-vegetarians] were enrolled in a cross-sectional observational study and accessed for biochemical glycemic markers [(Fasting Blood Sugar (FBS), Post Prandial Blood Sugar (PPBS), Glycosylated Hemoglobin (HbA1c) and High Sensitive C Reactive Protein (hs-CRP)]. Information on the dietary patterns and dietary nutrient intake was recorded using 3 day diet recall and FFQ. Results showed higher intake of total dietary fat, SFA, percentage energy contribution from fat and lower intake of omega 3 fatty acid in vegetarians compared to non vegetarians ( $p < 0.05$ ). Higher degree of inflammation (hs CRP) was observed in vegetarians compared to non vegetarians ( $\chi^2 = 9.371$ ,  $p = 0.003$ ). No other significant difference in glycemic indices (FBS, PPBS, HbA1C) was observed between them ( $p > 0.05$ ). HbA1C levels negatively correlated with n-3 fatty acid consumption while pro-inflammatory food intake was significant positively correlated with hs-CRP and negatively correlated with anti-inflammatory food intake ( $p < 0.05$ ). Thus the study shows higher level of nutrient related metabolic stress in vegetarian uncontrolled type 2 diabetic patients than non vegetarians, indicating a need for suitable dietary strategies with respect to pro and anti inflammatory foods along with omega-3 fatty acids.

**Key words:** Type 2 diabetes mellitus, hs-CRP, Omega 3 fatty acids, pro inflammatory and anti inflammatory foods.

### INTRODUCTION

Diabetes is a widespread metabolic and inflammatory disease with multiple pathological abnormalities worldwide as well as in India. [1] The major reason for the current endemic of diabetes is a consequence of faulty nutrition, predominantly imbalance in macro nutrients. Dietary practices appear to influence the macronutrient intake. Lower intake of saturated fat as well as cholesterol in vegetarians has been reported to be the

reason for lower risk of chronic diseases among them than non vegetarians. However, vegetarian diets are low in n-3 polyunsaturated fatty acids [particularly eicosapentaenoic (EPA) and docosahexaenoic acids (DHA)] and high in carbohydrates as well as n-6 polyunsaturated fatty acids than non-vegetarian diets. [2]

Omega-3 fatty acids play multiple roles in the body i.e. improve the fluidity and responsiveness of cell membranes,

enzyme activity, insulin signaling, glucose metabolism and gene expression [3,4] thereby, reduce the vascular derived oxidative stress associated with diabetes. These fatty acids have also been reported to inhibit inflammation by down regulating the transcription of chief inflammatory genes by competing with the n-6 fatty acid, arachidonic acid (ARA), involved in pro-inflammatory eicosanoid biosynthesis. [5] Hence the ratio of dietary n-3 to n-6 fatty acids is important in the aetiology of insulin resistance as well inflammation in diabetes.

Furthermore, there is a greater concern about the EFA status of vegetarians and vegans due to their diverse dietary practices i.e. relatively high intakes of n-6 FA combined with low intakes of EPA and DHA. Thus the present study has been proposed with the aim to investigate the dietary nutrient consumption and its association with glycemic control and serum hs-CRP in type 2 diabetes mellitus (T2DM).

## **MATERIALS AND METHODS**

The present study is a cross-sectional observational study of 160 patients suffering from type 2 diabetes mellitus including vegetarian and non vegetarian males and females in the age group of 40-60 years. All the participants were on oral hypoglycaemic agents, regularly visiting diabetes clinics and were enrolled purely based on their willingness to participate in the study. Subjects with type 1 DM or on insulin therapy, and suffering from any critical clinical condition like serious hepatic disease, renal failure, cardiac failure, cancer and haematological disorders, smokers and alcoholics were excluded from the study in order to curtail potential unadjusted confounders. The study protocol was approved by the Ethics Committee, Inter System Biomedica Ethics Committee (ISBEC), Mumbai, India (DCGI Registration NO. – ECR/108/Indt/MH/2013). An interviewer-administered pre-tested questionnaire was used to obtain information on age, gender, medical history, family history of diabetes,

symptoms and other clinical parameters related to diabetes and nutritional status of the participants.

## **Assessment of glycemic and inflammatory response:**

Serum glucose (Fasting and Post Prandial blood sugar) was measured by using glucose oxidase-peroxidase (GOD POD) enzymatic end point method, HbA1c was estimated by high pressure liquid chromatography (HPLC) and High Sensitive C Reactive Protein (hs-CRP) by immunoturbidimetric methodology. For categorical analysis of hs-CRP levels, participants were classified as per the criteria given by the American Heart Association and US Centers for Disease Control and Prevention [6] i.e. Low risk: <1.0 mg/L Average risk: 1.0 to 3.0 mg/L High risk: >3.0 mg/L. The reports were obtained from subject's case files.

## **Assessment of dietary nutrient consumption and dietary Patterns**

Information on the daily dietary energy and macronutrient intake was obtained by using food frequency questionnaire and 24-hour recall method applied on 3 different days (2 week days and 1 weekend). The dietary energy and macronutrient intakes of all subjects were calculated using DietCal software (DietCal' version 5.0; Profound Tech Solution; <http://dietcal.in/> based on values from the Nutritive Value of Indian Foods). Omega 3 nutrient content was calculated using the data from Fish processing- Advances in Fish Processing Technology. [7] A specified FFQ was designed to assess intake of omega-3 PUFAs rich, pro-inflammatory and anti-inflammatory rich foods. Food models, household measuring utensils, printed visuals and open-ended questions were applied to assist the participants.

## **STATISTICAL ANALYSIS**

Data were analysed using SPSS software for Windows (Version 16.0, 2007, SPSS Inc, Chicago IL) and presented as Mean±SD. Independent sample T test was used to

analyze the difference in the blood parameters between vegetarians and non-vegetarians (both for entire group and when classified according to gender). Cross tabulations were done for gender, food frequency intake and n3 fatty acids, pro-inflammatory foods and anti-inflammatory foods according to type of diet and analyzed using Chi-square test for association. P value at 5% was used to declare the level of significance.

## RESULTS

In the current study glycemic indices (FBS, PPBS, HbA1C) were found to be lower among the vegetarians (n=81) than non-vegetarians (n=79). However, the difference was not significant (p>0.05). And in spite of having lower glycemic indices, significantly higher levels of hs-CRP were found in vegetarian participants when compared with non-vegetarians ( $\chi^2=9.371$ , p=0.003) (Table 1).

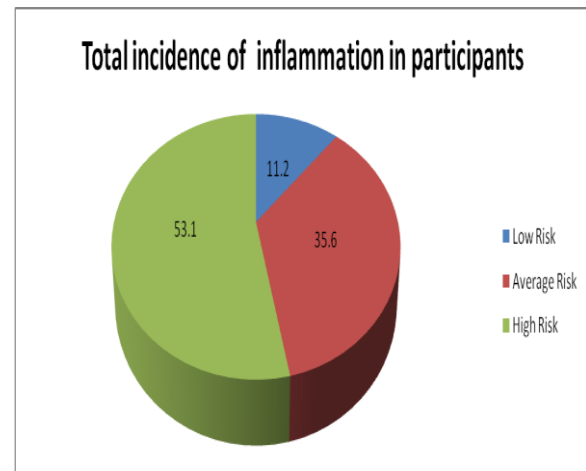
**Table 1: Glycemic markers and hs-CRP levels of the participants**

	Vegetarians (n=81)	Non-Vegetarians (n=79)	P value
FBS (mg/dL)	182.8±69.9	185.2±69.1	0.826
PPBS (mg/dL)	256.7±90.6	266.2±101.5	0.534
HbA1C	8.9±1.8	9.2±1.9	0.223
Hs-CRP (mg/L)	3.7±2.0	2.8±1.8	0.004

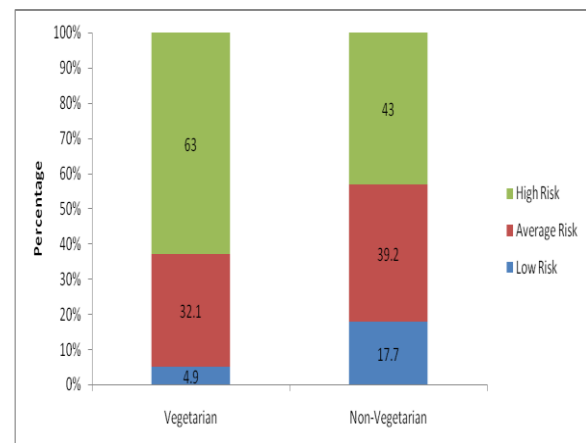
Data presented as Mean±SD

When participants were further classified according to hs-CRP levels, 18 (11.2%) patients had low risk, 57 (35.6%) patients had average risk and 85 (53.1%) patients had high risk of inflammation thus confirming higher incidence of inflammation among diabetics especially

among vegetarians than non vegetarians ( $\chi^2=9.371$ , p=0.003) (Figure 1, Figure 2).



**Figure 1: Distribution of participants according to the Level of inflammation**



**Figure 2: Distribution of participants according to the Level of inflammation and type of diet**

### Dietary energy and fat intake:

As seen in Table 2A & Table 2B, vegetarians (males and females) as well non vegetarians (males and females) consumed lower amounts of energy and greater amounts of fat (p <0.05).

**Table 2A: Dietary energy and fat intake of male participants and type of diet (n=83)**

Parameters	RDA-India	Vegetarians (n=39) Mean±SD	Non-Vegetarians(n=44) Mean±SD	P value
Energy (kcal/day)	2320	1308±173	1316±18	0.851
Percent energy from total fat	15-30	40.3±6.5	36.1±6.3	<b>0.004</b>
Percent energy from SFA	<10	5.4±2.2	5.4±2.8	0.861
Percent energy from PUFA	<8	9.1±4	9.6±3.6	0.958
ratio N6 : N3	1:4-5	01:09.5	01:09.5	0.471
Ratio PUFA:MUFA: SFA	1:5:1.5	1:2:2	1:2:2	0.492
Total fat (g/day)	50	58.6±12.1	53.1±12.2	<b>0.045</b>
Saturated fats (g/day)	up to 25	7.9±3.1	7.9±4.1	0.985
MUFA (g/day)	up to 25	16.2±6.6	14±6.5	0.131
PUFA (g/day)	not more than 21	13.2±5.9	13.3±5.3	0.936
N-6 fatty acid (g/day)	8 to 18	10.9±4.9	11.2±5.4	0.741
N-3 fatty acid (g/day)	more than 1.2	1.3±1.7	1.2±0.2	0.738
EPA + DHA (g/day)	0.5 -1**	--	0.3±0.4	--

\* ICMR 2011\*\* AHA guidelines Prevention of Cardiovascular Disease and Stroke

**Table 2B: Dietary energy and fat intake of female participants and type of diet (n=77)**

Parameters	RDA-India	Vegetarians (n=42) Mean±SD	Non-Vegetarians(n=35) Mean±SD	P value
Energy (kcal/day)	1900	1163±174	1169±145	0.873
Percent energy from total fat	15-30	42.4±8	36.9±5.4	<b>0.001</b>
Percent energy from saturated fat	<10	6.6±3.5	5.6±2.4	0.18
Percent energy from PUFA	up to 8	12.1±4.3	11.7±4	0.716
Percent energy from MUFA	up to 10			
ratio N6 : N3	1:4-5	01:09.4	01:09.6	0.994
Ratio PUFA:MUFA: SFA	1: 5.5	1:2:2	1:2:2	0.543
Total fat (g/day)	40	54.9±13.1	48.0±9.5	<b>0.012</b>
Saturated fats (g/day)	Up to 25	8.5±4.7	7.4±3.6	0.274
MUFA (g/day)	up to 21	13.8±6.5	11.06±4.1	<b>0.037</b>
PUFA (g/day)	not more than 16	15.3±4.4	15.1±5.3	0.891
N-6 fatty acid (g/day)	6 to 14	12.8±4.1	13.3±4.9	0.671
N-3 fatty acid (g/day)	more than 1.0	1.3±1.4	1.2±1.4	0.615
EPA + DHA (g/day)	0.5 -1**	--	0.3±0.4	

\* ICMR 2011\*\* AHA guidelines Prevention of Cardiovascular Disease and Stroke

Dietary fatty acids more importantly, the relative amounts of dietary saturated and unsaturated fatty acids have been shown to influence insulin sensitivity and also regulate inflammatory gene expression. Hence these are often associated with insulin resistance and inflammation. In the current study, the dietary fatty acid intake of the participants indicated that vegetarians (males-5.4±2.2, females-6.6±3.5) had higher percentage energy intake from SFA than their recommended intake as well as that of non vegetarians (males-5.4±2.8, females-5.6±2.4). The percentage energy intake from PUFA as well as the ratio of N3: N6 and SFA:MUFA:PUFA were also found to be higher than RDA in all the participants, indicating absolute disproportion in the fatty acid consumption of participants with respect to quality as well as quantity (Table 2A & Table 2B).

### Correlation of biochemical parameters with Omega 3 Fatty acids

**Table 3 : Correlation of biochemical parameters with omega 3 fatty acid intake**

	FBS	PPBS	HbA1C	Hs-CRP
Energy (kcal/day)	-0.043	0.006	0.035	0.03
Total fat (g/day)	-0.027	-0.081	-0.014	0.087
Saturated fats (g/day)	-0.136	-0.089	-0.139	-0.008
MUFA (g/day)	-0.095	-0.083	-0.144	0.114
PUFA (g/day)	-0.014	-0.027	0.032	-0.085
N 6 fatty acid (g/day)	0.013	-0.001	0.061	-0.12
N 3 fatty acid (g/day)	-0.131	-0.126	-0.169*	0.08
N6 to N3 fatty acid ratio	0.122	0.043	0.114	-0.006
Ratio PUFA to Saturated fats	0.066	0.008	0.084	-0.119
EPA to DHA Ratio	-0.004	0.161	-0.028	0.158

Data presented as Spearman's correlation value \*p <0.05 for significant correlation

As seen in Table-3, HbA1C levels were found to be significantly negatively correlated with n 3 fatty acid consumption (p<0.05) where as no significant correlation was found between FBS, PPBS, hs-CRP levels and n-3 fatty acid consumption (p>0.05).

### Correlation of blood parameters with food intake:

**Table-4** presents Spearman's correlation of blood parameters with frequency of pro-inflammatory and anti-inflammatory food intake in all diabetic patients (n=160). Overall, frequency of consumption of walnut, almond, whole grains & legumes of participants was found to be negatively correlated with glycemic indices (p<0.05) and significant positively correlated with fried foods, carbonated beverages and red meat (p<0.05) (**Table 4**). While hs-CRP levels were positively correlated with frequency of consumption of carbonated beverages, junk food, butter/cheese and it has negatively correlated with frequency of consumption spices and condiments (p<0.05).

**Table 4: Correlation of biochemical parameters with pro-inflammatory and anti-inflammatory food consumption**

Foods	FBS	PPBS	HbA1C	Hs-CRP
Walnuts	-0.17*	-0.198*	-0.153	-0.07
Almonds	-0.137	-0.238*	-0.171*	-0.108
Fried foods	0.088	0.144	0.231*	0.001
Carbonated beverages	0.155	0.176*	0.222*	0.161*
Junk foods	0.048	0.074	0.116	0.212*
Butter/cheese	0.127	0.128	0.066	0.254*
Spices condiments	-0.106	-0.04	-0.091	-0.167*
Whole grains/legumes	-0.212*	-0.119	-0.067	0.03

Data presented as Spearman's correlation value \*p <0.05 for significant correlation

## DISCUSSION

Globally, vegetarians comprise only a small proportion (< 5%) of the population, but in India, a large proportion of the population (approximately 35%) follows vegetarian diet with varied dietary practices.<sup>[8]</sup> It is widely assumed that higher intake of fiber and lower intake of saturated fat and cholesterol are the reasons for the health benefits of vegetarian diets. When compared between vegetarian diets and non vegetarian diets, vegetarian diets were also reported to offer an advantage over non vegetarian diets with respect to prevention and management of diabetes and also to resolve inflammation in hyperglycemic diabetes. This study was therefore designed to investigate the association between dietary nutrient intake, dietary habits, glycemic control and serum hs-CRP in type 2 diabetes mellitus patients.

No significant difference in glycemic indices (FBS, PPBS, HbA1C) was observed between the vegetarian (n=81) and non-vegetarian (n=79) participants of the study ( $p>0.05$ ). However there was a trend towards lower glycemic indices in vegetarians than non vegetarians which was in line with the results of a number of studies that have confirmed significantly lower levels of FBS, HbA1c and hs-CRP levels in vegans/vegetarians than non vegetarians patients with diabetes.<sup>[9,10]</sup> Similar trends were also seen in an Indian cross sectional population based study conducted by Aggarwal et al (2014) where lower occurrence of diabetes was observed in vegetarians than non-vegetarians after adjusting for a number of socioeconomic and lifestyle factors.<sup>[8]</sup>

Increased hs-CRP levels are associated with hyperglycemia in patients with diabetes as shown in few cross sectional or case-control studies. The probable mechanism has been an upregulation of duodenal cells' gene expression of pro-inflammatory cytokines<sup>[11]</sup> along with altered activities of erythrocyte antioxidant enzymes<sup>[12]</sup> and imbalances in pro and anti-inflammatory adipokines.<sup>[13]</sup> In the current study,

vegetarians had significantly higher hs-CRP levels as compared to non-vegetarians ( $p<0.05$ ) and this trend is in line with the results of a cross-sectional study.<sup>[14]</sup>

Faulty nutritional practices are found to be one of the major contributory factors for the present epidemic of diabetes. From the last 3 decades consumption of total dietary fat was found to be increasing in India as reviewed by A Laxmaiah et al. (2014).<sup>[15]</sup> Surprisingly, the dietary fat consumption was different between vegetarian and non vegetarian patients in the current study i.e. vegetarians had significantly greater intake of total fat & percentage energy from fat as compared to their non-vegetarian counterparts ( $p<0.05$ ). Similar results were found in an urban survey conducted in Maharashtra<sup>[16]</sup> as well as in Starch study.<sup>[17]</sup> These studies demonstrated higher intake of energy from fat (>30% to 50% of energy) in both vegetarians and non vegetarians against their respective RDA (upto 30% from total energy) and also higher fat intake (55.30 g/day) in Indian participants.

Omega-3 FA consumption has been associated with improved insulin sensitivity in type 2 diabetes patients and these fatty acids also have been related to low prevalence of diabetes in few cohort studies. Various interventional trials have also documented positive role of omega 3 fatty acids in DM and inflammation.<sup>[18,19]</sup> In the present study, HbA1C levels were found to be negatively correlated with n 3 fatty acid consumption ( $p<0.05$ ). In line with these findings, a recent study conducted in China revealed significantly lower level of serum omega-3PUFA in patients with type 2 diabetes mellitus ( $7.17\pm 2.38$ ) than healthy control subjects ( $10.08 \pm 2.76$ )<sup>[20]</sup> and they have also found a negative correlation of dietary omega-3 PUFA<sup>[21]</sup> with HOMA-IR, blood glucose level and also impaired glucose tolerance (IGT).

The health benefits of plant food based diets could be related to integrated antioxidant and anti-inflammatory mechanisms exerted by a wide range of

phytochemicals present in these foods which modulate oxidative and inflammatory stress efficiently and also help to prevent diet related diseases. In the present study, glycemic indices and hs-CRP levels were found to be negatively correlated with anti-inflammatory food intake (walnut, almond, whole grains & legumes and spices & condiments) ( $p < 0.05$ ) and positively correlated with pro-inflammatory food intake (fried foods, carbonated beverages, junk food, butter/ cheese and red meat) ( $p < 0.05$ ). These results were similar to a parallel randomized controlled trial conducted by Tapsell et al (2009) [22] and Mary et al (2017) [23] wherein greater reductions in fasting insulin levels ( $P = 0.046$ ), CRP levels (36.2%) and significant improvement in mean glycosylated hemoglobin level ( $P < 0.04$ ) were found in DM patients with 30 gms walnut and almond consumption per day, suggesting addition of walnuts and almonds as one of the healthy options for diabetics. Several studies have also reported significant positive association of pro-inflammatory food consumption [24,25] with inflammatory markers (CRP, IL6, TNFr1, TNFr2); and consumption of anti-inflammatory foods with decreased inflammatory markers. [26]

## CONCLUSION

Poor glycemic control and higher level of inflammation were observed among vegetarian uncontrolled type 2 diabetic patients than non-vegetarians. This higher degree of nutrient related metabolic stress among vegetarian patients demands management of the disease beyond glycemic control with special emphasis on their dietary practices. These findings are valuable for community specific health interventions in diabetes care in India.

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