ijlpr 2022; doi 10.22376/ijpbs/lpr.2022.12.1.L170-174

International Journal of Life science and Pharma Research

ISSN 2250-0480

Research Article

Clinical Bio chemsitry

Significance Of Interleukin-6 In Diabetes Mellitus And Its Complications

A. Mary Chandrika[1], Dr. V.S.Kalai Selvi[2*]

[1] *Assistant Professor, Department of Biochemistry, Sree Balaji Medical College and Hospital, Chrompet, Chennai-600044.*
[*2] *Professor, Department of Biochemistry, Sree Balaji Medical College and Hospital, Chrompet, Chennai-600044*

Abstract: Low grade inflammation is one of the causes in the pathogenesis of type 2 diabetes. Interleukin – 6 (IL-6) a pro-inflammatory cytokine is found to be increased in type 2 diabetic patients. This study was aimed to assess the role of Interleukin -6 in type 2 diabetes mellitus. This was a cross sectional study done among diabetics and non- diabetics. The total number of subjects involved in the study was 120 within the age group of 20-50 years attending the outpatient department and diabetic clinic of Sree Balaji Medical College and Hospital. The study group comprised of Group- I – 40, age and sex matched non diabetic subjects. Group- II – 40, type II diabetic patients with good control of blood glucose, Group III – 40, type II diabetic patients with poor glycemic control. Blood samples were taken after 8 hours of fasting period, centrifuged at 3000 rpm for 10 min and serum, plasma separated. Plasma glucose levels (FBG, PPBG) were estimated by GOD/POD method, IL- 6 levels were measured by ELISA method and HbA1c by immunoturbidimetric method. Data was statistically analyzed using the Statistical Package for the Social Sciences (SPSS) version 18. Results showed that the circulating Interleukin- 6 (IL-6) levels were found to be elevated in type 2 diabetics than non diabetic subjects ($p < 0.0001$). Raised level of Interleukin-6 was found in diabetics compared to non diabetics that showed the presence of inflammatory mechanisms in diabetes patients. Also uncontrolled diabetics have increased Interleukin-6 levels. In addition to periodic checking of HbA1c, it is paramount important to check the inflammatory status in high risk subjects for diabetes and also in diabetics to prevent diabetic complications.

Keywords: Interleukin- 6; glycated hemoglobin, type 2 diabetes mellitus; HbA1C, diabetic complications.

***Corresponding Author**

Dr. V.S.Kalai Selvi , Professor, Department of Biochemistry, Sree Balaji Medical College and Hospital, Chrompet, Chennai-600044

Received On 7 August, 2021
Revised On 30 December, 2021
Accepted On 6 January, 2022
Published On 11 January, 2022

Funding This research did not receive any specific grant from any funding agencies in the public, commercial or not for profit sectors.

Citation A. Mary Chandrika, Dr. V.S.Kalai Selvi , Significance Of Interleukin-6 In Diabetes Mellitus And Its Complications.(2022).Int. J. Life Sci. Pharma Res.12(1), L170-174 http://dx.doi.org/10.22376/ijpbs/lpr.2022.12.1.L170-174

This article is under the CC BY- NC-ND Licence (https://creativecommons.org/licenses/by-nc-nd/4.0)

Copyright @ International Journal of Life Science and Pharma Research, available at www.ijlpr.com

1. INTRODUCTION

Diabetes mellitus is a group of metabolic disorders characterized by hyperglycemia caused by a defect in insulin secretion, insulin action, or both. Several pathophysiology theories have been proposed to explain the onset of diabetes mellitus. Several studies have found that low-grade inflammation is a risk factor for the development of type 2 diabetes mellitus in the future. The precise mechanism is not known. Diabetes type 2 is linked to ageing, obesity, and inactivity. It is caused by the progressive failure of beta cells in the pancreatic islets to compensate for insulin resistance[1]. Various mechanisms, including oxidative stress, ectopic lipid deposition in muscles, liver, and pancreas, lipotoxicity, and glucotoxicity, have been proposed to explain impaired insulin resistance and sensitivity in type 2 diabetes. All of these stresses have the potential to cause an inflammatory response. Other mechanisms include hypoxia, nuclear factor activation, and interleukin activation. Immunological research examines the potential role of inflammation in the pathogenesis of type 2 diabetes. Interleukin-6 (IL-6), a pro-inflammatory cytokine is responsible for the pathology of type 2 diabetes mellitus through insulin resistance by controlling differentiation, migration, proliferation, and cell apoptosis. IL-6, apart from its immuno-regulatory actions, has been suggested to affect glucose homeostasis and metabolism directly and indirectly by the action on skeletal muscle, adipocytes, hepatocytes, pancreatic beta cells and neuro-endocrine cells. Immuno-regulatory mediators not only cause metabolic aberrances but also cause beta cell dysfunction and insulin resistance. IL-6 associated with obesity is due to accumulation of macrophages in adipose tissue which release inflammatory mediators promoting inflammation. A low grade inflammation is one of the causes in the pathogenesis of type 2 diabetes mellitus[2].IL-6 is a multifunctional pro-inflammatory cytokine produced by adipocytes, endothelial cells, smooth muscle cells, fibroblasts, lymphocytes and macrophages[3,4]. It affects insulin sensitivity and regulation, lipoprotein lipase action, adipocyte function and thus decreases islet cell secretory function [5,6]. Certain studies emphasize that endothelium is the target tissue of insulin and therefore Insulin Resistance also exists at the level of endothelium. Insulin promote vasodilatation by activating the signaling pathway involving IR, IRS 1 and PI3 kinase which is similar tone that stimulate insulin mediated glucose uptake in tissues Few studies have addressed the effect of IL6 on Insulin signaling pathway causing impairment of insulin signal transduction in hepatocytes and adipocytes[7]. The potential role of IL6 in vascular insulin is still not yet clear. Previous studies highlight the role of Interleukin6 as a pleiotropic cytokine involved in inflammation. Diabetes Mellitus is one of the major endocrine disorders for which low grade inflammation forms the pathogenesis. This study was planned to confirm the same finding in our population which might pave the way for development of Anti IL6 monoclonal antibodies as a treatment modality in type 2 Diabetes Mellitus. This study was aimed to evaluate the role of Interleukin -6 in type 2 diabetes mellitus and its complications. The objectives of the study were to evaluate the association of IL-6 in type 2 diabetes mellitus and to evaluate the significance of IL- 6 in patients uncontrolled diabetes mellitus.

2. MATERIALS AND METHODS

This was a cross sectional study done among diabetics and non- diabetics. The total numbers of subjects involved in the study were 120 within the age group of 20-50 years attending the outpatient department and diabetic clinic of Sree Balaji Medical College and Hospital. The study group comprised of Group- I – 40 age and sex matched non diabetic subjects. Group- II –40 type II diabetic patients with good control of blood glucose, Group III -40 type II diabetic patients with poor glycemic control.

All the study participants were given a proforma to collect the detailed history. Clearance obtained from the Institutional Ethical Committee (REF NO.002/SBMC/IHEC/2017/930) and informed consent obtained from all those who have been involved in the study. Under aseptic precaution blood samples were collected for analysis

2.1 Inclusion criteria

- Type 2 diabetes mellitus patients and Non diabetic subjects of 20-50 years of age.
- Willing to participate in the study.

2.2 Exclusion criteria

- Patients with acute infection, chronic inflammatory diseases, obesity, cancer, elevated liver indices
- Patients with high serum creatinine levels of >1.5mg/dl, pregnancy, arthritis and those on corticosteroids.

2.3 Biochemical analysis

Blood samples were taken after 8 hours of fasting period, centrifuged at 3000 rpm for 10 min and serum, plasma separated. Plasma glucose levels (FBG, PPBG) were estimated by GOD/POD method, IL- 6 levels were measured by ELISA method[8] and HbA1c by Immunoturbidimetric method[9].

3. STATISTICAL ANALYSIS

The characteristics of study participants are summarized using mean and standard deviation. The outcome variables namely, fasting plasma glucose, postprandial plasma glucose, glycated hemoglobin (HbA1c), serum IL6 were compared among three groups by **Bonferroni method.** A two sided p-value less than 0.05 was considered to be statistically significant. All the results data were statistically analyzed using SPSS version 18.

4. RESULTS

Forty Non diabetic persons were included in Group I with mean age 30.4 years and SD 7.4. Forty Type 2 Diabetic patients with good Glycemic control as Group II with mean age 32.8 years and SD 7.9. Forty Type II Diabetic patients without Glycemic control as Group III with mean age 35.4 years and SD 9.6were included.In Group I the mean Fasting Blood Glucose 95.3 mg/dl with SD 8.5 and Postprandial Blood Sugar103 mg/dl with SD 7.4 In Group II the mean Fasting Blood Glucose 114.2 mg/dl with SD 6.9 and Postprandial Blood Sugar 118.6mg/dl with SD 8.6 In Group III the mean Fasting Blood Glucose 137.2 mg/dl with SD 17.0 and Postprandial Blood Sugar 190.7 mg/dl with SD 44.5. Mean HbA1C in Group I -5.5% with SD 0.3 Mean HbA1C in Group II -6.3% with SD 0.4. Mean HbA1C in Group III -7.2% with SD 0.7.Mean Interleukin 6 in Group I 4.6pg/ml with SD 0.3. Mean Interleukin 6 in Group II 7.3pg/ml with SD 1.3.

Mean Interleukin 6 in Group III 12.8pg/ml with SD 2.0.All the results were tabulated in Table I

Table I: Characteristics of study participants

	Group I		Group II		Group III	
N	40		40		40	
Parameters	Mean	SD	Mean	SD	Mean	SD
Age (years)	30.4	7.4	32.8	7.9	35.4	9.6
FPG (mg/dl)	95.3	8.5	114.2	6.9	137.2	17.0
PPPG (mg/dl)	103	7.4	118.6	8.6	190.7	44.5
HbA1C (%)	5.5	0.3	6.3	0.4	7.2	0.7
IL 6 (pg/ml)	4.6	0.3	7.3	1.3	12.8	2.0

SD- standard deviation; FPG- fasting plasma glucose; PPPG- postprandial plasma glucose; HbA1C – glycated hemoglobin; IL 6 – interleukin 6.

Group I – Non diabetic subjects
Group II - type II diabetic patients with good control of blood glucose
Group III – type II diabetic patients without blood glucose control.
The mean age of participants was 33± 8.3 years. The mean FPG, PPPG, HbA1c and IL 6 levels were higher in group III compared to the other groups.
Comparison of Glycated hemoglobin between the groups I and II show a p value of less than 0.0001
Comparison of Glycated hemoglobin between the groups I and III show a p value of less than 0.0001.
Comparison of Glycated hemoglobin between the groups II and III show a p value of less than 0.0001

Table II: Comparison of glycatedhaemoglobin between the groups (by Bonferroni)

GROUP	I	II
II	<0.0001	-
III	<0.0001	<0.0001

Data in the table are p values

Comparison of Glycated hemoglobin between the groups I and II show a p value of less than 0.0001.
Comparison of Glycated hemoglobin between the groups I and III show a p value of less than 0.0001.
Comparison of Glycated hemoglobin between the groups II and III show a p value of less than 0.0001
The mean of glycated hemoglobin was significantly different between groups I and II, groups II and III, groups I and III (all p values <0.05) as in table II.

Table III: Comparison of Interleukin 6 between the groups (by Bonferroni)

GROUP	I	II
II	<0.0001	-
III	<0.0001	<0.0001

Data in the table are p values

Comparison of Interleukin 6 between the groups I and II show a p value of less than 0.0001
Comparison of Interleukin 6 between the groups I and III show a p value of less than 0.0001.
Comparison of Interleukin 6 between the groups II and III show a p value of less than 0.0001.
The mean of interleukin 6 level was significantly different between groups I and II, groups I and III, groups II and III (all p values <0.05) as in table III.
The effects of Hba1c and fasting blood glucose of our study is consistent with the study by He Q et. Al[10].
The findings of a meta analysis by Nicholas Bowker et. al., is consistent with the findings of this study[11]. Studies by Pradhan et al ha.ve demonstrated an increase of CRP and IL-6 in individuals with features of insulin resistance and clinically overt type 2 diabetes mellitus. The study also revealed that CRP is a more powerful inflammatory marker than IL-6. In this CRP was not done.

5. DISCUSSION

The pathogenesis of type 2 Diabetes is characterized by a combination of Insulin Resistance and failure of pancreatic beta cells to compensate for the increased insulin demand. During the past decades, much evidence supports the concept that the insulin resistance and type 2 Diabetes Mellitus are related to chronic low grade inflammation. Circulating IL 6 levels have been reported to be elevated in patients with type 2 Diabetes and correlate with the direct and indirect measures of Insulin Resistance. IL-6 is one of the major inflammatory cytokines produced by a variety of tissues like activated leukocytes, adipocytes & endothelial cells. IL 6 is one of the mediators of acute phase response, and also enhances the release of hepatic C- reactive protein in the liver. The result obtained from this study shows that HbA1c in uncontrolled diabetic patients was significantly correlated with IL-6 but this observation was not seen in

non-diabetic subjects. Persistent hyperglycemia contributes to the formation of advanced glycation end products, up-regulation of the innate immune system resulting in chronic inflammation[12]. This association among glycated hemoglobin (HbA1c), IL-6 suggests the role of a prominent inflammatory mechanism in type 2 diabetes mellitus. This study shows that IL 6 is elevated in diabetic patients than the diabetic subjects. Zahran et al and Spranger J et al have also mentioned similar results in their studies[13,14]. Elevated levels of IL-6 were observed in both poorly and well- controlled diabetes in many independent clinical studies. Pradhan et al[15]. in their study stated that type 2 diabetes mellitus may be a manifestation of an ongoing cytokine- mediated acute phase response initiated by the body's innate immune system. Nalysnyk et al, indicated that not only poor glycemic control[16] but also enhanced production of IL-6 leads to microvascular complications. IL-6 participates in pathogenesis of endothelial dysfunction by stimulation of monocyte chemotactic protein-1 and cell adhesion molecules[17]. Some clinical results strongly suggest that enhanced IL-6 concentration is associated with diabetic kidney disease- enhanced stimulation of mesangial cell proliferation which interferes with the extracellular dynamics of matrix formation at the podocyte level. Christian Herder et al, in their study had revealed associations between biomarkers of subclinical inflammation and depressive symptoms in patients with diabetes mellitus[18]. Meta analytic studies performed, provide a better understanding that IL-6 produced by a variety of cells involved in inflammation and act as signals in pathological processes associated with chronic inflammation[19]. Studies reveal circulatory levels of IL-6 and TNF- 〈 are elevated in diabetic patients. Hyperglycemia increases circulating cytokine concentration by oxidative mechanism [20]. Also numerous studies have confirmed that both schizophrenia and diabetes underlie chronic low-grade inflammation with elevated IL-6 indicating that disturbances in immune response might be involved in concurrent onset of both conditions. Tulshi Chakraborty et al, in their study has mentioned that biomarkers like Autoantibodies, C-reactive protein (CRP), Fibrinogen, Interleukin-1 receptor antagonist (IL-1RA), Plasminogen activator inhibitor (PAI-1), etc. are used for diagnosis; Adiponectin, microRNA, Acylcarnitine, Hemoglobin A1c, etc. are used for treatment, and 1,5-anhydroglucitol are used for both diagnosis and treatment of diabetes mellitus[21].

6. CONCLUSION

Raised levels of Interleukin-6 shows progression of inflammatory mechanisms in diabetes patients. High pro-inflammatory marker IL-6 results in development of microvascular and macrovascular complications. In addition to periodic checking of HbA1c, it is paramount important to check the inflammatory status in diabetics to prevent diabetic complications. Anti-IL-6 drugs have been developed and already used for treatment of various diseases. This study provides valuable information to clinicians for developing clinical management protocols for patients with type 2 diabetes mellitus.

7. ACKNOWLEDGMENTS

We express our sincere gratitude to all teaching and technical staff members of Department of Biochemistry, Department of Medicine Sree Balaji medical college and Hospital,

8. CONFLICT OF INTEREST

No conflicts of interest are declared by the authors.

9. AUTHORS CONTRIBUTION STATEMENT

Sample collection, processing was done by Dr. Mary Chandrika, discussion and write up done by Dr. V. S. KalaiSelvi.

10. REFERENCES

1. Marc Y. Donath &Steven E. Shoelson Type 2 diabetes as an inflammatory disease Nature Reviews Immunology 14 January 2011 11:98–107
2. Al-ShookriG.L.Khor, Y.M.Chan, S.C.Loke and M. Al-Maskari, Type 2 diabetes in the sultanate of Oman ,Malayasian Journal of nutrition. 2011;17(1):129-141.
3. Papanicolaou DA, Vgontzas AN. Interleukin-6: the endocrine cytokine. J ClinEndocrinolMetab 2000; 85:1331–1333.
4. Barnes TC, Anderson ME, and Moots RJ. The Many Faces of Interleukin-6: The Role of IL-6 in Inflammation, Vasculopathy, and Fibrosis in Systemic Sclerosis. Hindawi Publishing Corporation International Journal of Rheumatology 2011; 721608:1-6.
5. Pradhan AD, Manson JE, Rifai N, Buring JE and Ridker PM. C-reactive protein, interleukin 6, and risk of developing type 2 diabetes mellitus. JAMA 2001; 18:286(3):327-34.
6. Narayanan KM, Boyle JP, Thompson TJ et al. Lifetime risk for diabetes mellitus in the US. JAMA 290 .18841890, 2003
7. Francesco Andreozzi, EmanuelaLaratta,CristinaProcopio, Marta LetiziaHribal, Angela Sciacqua, Maria Perticone, Claudia Miele, Francesco Perticone, Giorgio Sesti. Interleukin-6 Impairs the Insulin Signaling Pathway, Promoting Production of Nitric Oxide in Human Umbilical Vein Endothelial Cells. December 2020. DOI: https://doi.org/10.1128/MCB.01340-06.
8. Rasool, Roohi et al. "Study of serum interleukin (IL) 18 and IL-6 levels in relation with the clinical disease severity in chronic idiopathic urticaria patients of Kashmir (North India)." Asia Pacific allergy vol. 4,4 (2014): 206-11. doi:10.5415/apallergy.2014.4.4.206
9. Metus P, Ruzzante N, Bonvicini P, Meneghetti M, Zaninotto M, Plebani M. Immunoturbidimetric assay of glycated hemoglobin. J Clin Lab Anal. 1999;13(1):5-8.
10. He Q, Dong M, Pan Q, Wang X, Guo L. Correlation between changes in inflammatory cytokines and the combination with hypertension in patients with type 2 diabetes mellitus. Minerva Endocrinol 2019; 44:252-8. DOI: 10.23736/S0391-1977.18.02822-5
11. Nicholas Bowker, Rupal L. Shah, Stephen J. Sharp, Jian'an Luan, Isobel D. Stewart, Eleanor Wheeler,

Manuel A.R. Ferreira, ArisBaras, Nicholas J. Wareham, Claudia Langenberg, Luca A. Lotta, Meta-analysis investigating the role of interleukin-6 mediated inflammation in type 2 diabetes, E-BioMedicine, Volume 61, 2020, 103062, ISSN 2352-3964

12. Singh, VarunParkash et al. "Advanced glycation end products and diabetic complications." The Korean journal of physiology &pharmacology : official journal of the Korean Physiological Society and the Korean Society of Pharmacology vol. 18,1 (2014): 1-14. doi:10.4196/kjpp.2014.18.1.1
13. Zahran A, Enas S. EssaWaleedF.andAbdElazeem Study of Serum Tumor Necrosis Factor Alpha and Interleukin 6 in Type 2 Diabetic Patients with Albuminuria. Life Science Journal 2012; 9(1): 877-882.
14. Spranger J, Kroke A, Mohlig M, Hoffmann K, Bergmann MM, Ristow M, Boeing H, Pfeiffer AF. Inflammatory cytokines and the risk to develop type 2 diabetes: results of the prospective population-based European Prospective Investigation into Cancer and Nutrition (EPIC)-Potsdam Study. Diabetes 2003;52 (3):812-7
15. Pradhan AD, Manson JE, Rifai N, Buring JE, Ridker PM. C-Reactive Protein, Interleukin 6, and Risk of Developing Type 2 Diabetes Mellitus. JAMA. 2001;286(3):327–334.
16. Nalysnyk L, Hernandez-Medina M, Krishnarajah G. Glycaemic variability and complications in patients with diabetes mellitus: evidence from a systematic review of the literature. Diabetes, Obesity & Metabolism. 2010; 12:288–298
17. Gabay, Cem. "Interleukin-6 and chronic inflammation." Arthritis research & therapy vol. 8 Suppl 2,Suppl 2 (2006): S3. doi:10.1186/ar1917
18. Herder C, Fürstos JF, Nowotny B, Begun A, Strassburger K, Müssig K, Szendroedi J, Icks A, Roden M; GDS Group. Associations between inflammation-related biomarkers and depressive symptoms in individuals with recently diagnosed type 1 and type 2 diabetes. Brain Behav Immun. 2017 Mar; 61:137-145.
19. Tanaka T, Narazaki M, Kishimoto T. IL-6 in inflammation, immunity, and disease. Cold Spring HarbPerspect Biol. 2014;6(10):a016295. Published 2014 Sep 4. doi:10.1101/cshperspect.a016295
20. Esposito K, Nappo F, Marfella R, Giugliano G, Giugliano F, Ciotola M, Quagliaro L, Ceriello A, Giugliano D. Inflammatory cytokine concentrations are acutely increased by hyperglycemia in humans: role of oxidative stress. Circulation. 2002 Oct 15;106(16):2067-72.
21. TulshiChakraborty, Dr.Sumeet Gupta, VipinSaini, ArindamTalukdar , Biomarkers: An Important Tool for Diagnosing and Treating Diabetes Mellitus.(2021).Int. J. Life Sci. Pharma Res.11(2), P123-129 http://dx.doi.org/10.22376/ijpbs/lpr.2021.11.2.P123-129