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Original Research

Association of inflammatory marker, glycosylated hemoglobin, circulating lipids with microvascular complications and glycemic control of type 2 diabetes mellitus in South Indian population

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Abstract

Background and Aims: Inflammatory processes provoke the synthesis of various acute-phase proteins like C-reactive protein (CRP) in the liver and orchestrate a predominant role in the development of insulin resistance. Against this backdrop, the present study was undertaken to evaluate the association of inflammatory markers, glycated hemoglobin, and circulating lipids with microvascular complications in type 2 DM patients. **Material and Methods:** Prospective hospital-based longitudinal study was conducted with 384 patients to complete the study. The metabolic profiles were measured at baseline, 3rd month (review I), 6th month (review II), and at 12th month (review III). Lipid profiles were measured at baseline. CRP was measured at baseline, 6th month and at 12th month. **Results:** The recruited patients were followed for three reviews. Blood sugar level, HbA_{1c}, and CRP were significantly decreased when compared to baseline. In this study baseline, CRP showed a positive correlation with baseline sugar levels, LDL, total cholesterol, renal parameters except for VLDL and a strong negative correlation with baseline HDL and ankle-brachial index but no significant correlation was observed with cardiac autonomic neuropathy and vibration perception threshold. **Conclusion:** This study concluded that due to effective pharmacotherapy the CRP level and the metabolic profiles were reduced after 12 months of follow-up.

Keywords: inflammation, CRP, HbA_{1c}, lipid profiles, microalbumniria.

Introduction

Diabetes mellitus (DM) is one of the global public health problems and the estimated range of 8.8 million adults populated are affected during the year 2015 and the number escalates in the future. Reports suggest that, in the year 2040 the global diabetic adult population is estimated to be increased by 10.4% reaching the total diabetic population of 642 million diabetic patients [1]. Type 2 diabetes mellitus (T2DM) accounts for 90–95% of DM and it imposes significant morbidity and mortality. The morbidity and mortality associated with T2DM are mainly due to micro- and macro-vascular complications [2]. The diabetic microvascular complications encompass diabetic kidney disease, retinopathy, and peripheral neuropathy. Albeit, strict glucose control decreases the development of diabetic microvascular complications, but the morbidity among these subjects are still on rising [3]. Apart from hyperglycemia, the other risk factors involved in the progression of microvascular complications are hyperlipidemia hypertension and silent inflammation [4].

Chronic inflammatory reactions orchestrate a cardinal role in the development of vascular complications in T2DM patients [5, 6]. The metabolic alteration such as insulin resistance, dyslipidemia, and central obesity induces a state of low-grade inflammation. This low-grade systemic inflammation displays an increased level of circulatory inflammatory proteins such as C-reactive protein (CRP) and various pro-inflammatory

cytokines [7]. CRP belongs to the group of acute-phase protein which is primarily synthesized in the liver and during any inflammatory reactions the plasma concentration of CRP act as a reliable marker of elevated inflammatory conditions in the vascular structures [8]. Mounting studies shows elevated CRP levels in patients affected with metabolic syndrome [9, 10]. Further, various clinical studies show the significant association between increased level of CRP and chronic diabetic vascular complications [11–13]. However, to date only little credible shreds of evidences exist regarding the increased level of CRP that can be used as a significant predictor for diabetic microvascular complications. Meanwhile, only a few literatures are available in substantiating the association between CRP level and diabetic microvascular complications and whether strict glucose control, as well as metabolic components, can decrease the inflammatory response, particularly CRP in T2DM subjects. In this scenario, the present study was conducted to find out the association of C-reactive protein, glycated hemoglobin, and circulating lipids with microvascular complications in type 2 diabetes mellitus patients with strict glycemic control.

Materials and methods

This was a prospective longitudinal study conducted among the type 2 diabetic patients from Jan 2017 to Dec 2018, at Aruna Diabetes Centre, Chennai, India. The study was approved by the Institutional Ethical Committee with reference number IEC/PHD 2015/2016/01. Initially, 450 patients were enrolled in the study and at the end of the study, the patients were lost regular follow-up (n=31), finally 384 patients completed the study.

Inclusion criteria

Inclusion criteria were based on patients who were diagnosed with type 2 diabetes either fasting blood sugar >126 mg/dl or post prandial blood sugar >200 mg/dl, a patient who consented to get screened for complications of diabetes 2 mellitus, patients with regular follow-up and treatment.

Exclusion criteria

After careful history taking and physical examination, patients with acute infections, chronic inflammatory diseases (inflammatory bowel disease, osteoarthritis, rheumatoid arthritis, chronic hepatitis, gout, and bronchial asthma), and cancer were excluded. Further, the patients who were suffering from type 1 diabetes mellitus, diabetic patients with pregnancy, who were not willing to participate, patients who were not under regular treatment were excluded.

Data collection

Demographic information and clinical factors associated with increased diabetic complications were collected using case extract forms from the patient's case book. The following information's like age, gender, weight, height, duration of diabetes, hypertension status, smoking status, history of CAD, history of CVD, sugar profile, and Glycated hemoglobin (HbA_{1c}) and lipid profile were collected and recorded. Blood samples were collected after a 12 hours overnight fast. Metabolic profiles like fasting and post-prandial plasma glucose, HbA_{1c} were measured at baseline, 3rd month, 6th month, and at 1 year. Triglyceride, total cholesterol, LDL-C, and HDL-C were measured at baseline. CRP was measured at baseline, 6th month and at 1 year. During the 1 year metabolic profiles were controlled. BMI was calculated by dividing weight in kg with height (in m^2). Both systolic and diastolic blood pressure was measured by a sphygmomanometer. Blood samples were analyzed for FBS, PPBS, TC, TGL, LDL, HDL, VLDL, and creatinine by a fully automated analyzer using standard enzymatic methods. HbA_{1c} was assayed using high-performance liquid chromatography. Serum C reactive protein was measured by nephelometer analyzer.

Diabetic vascular complications

Diabetic nephropathy was defined by urine excretion abnormalities and it was identified in the urine for microalbumin. It was

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estimated by using the immunoassay method. Diabetic peripheral neuropathy is a painful condition caused by nerve damage from diabetes. Biothesiometer was used to check the vibration perception threshold (VPT), done to identify early peripheral neuropathy of feet in volts. Ankle Brachial Index (ABI) test was used to assess the blood flow to the lower limbs. Because the lower limb arteries were more prone to atherosclerosis. ABPI is the ratio of blood pressure at the ankle to the upper arm used to check peripheral arterial disease. Cardiac autonomic neuropathy (CAN) is another complication that is common among diabetics. This leads to a silent heart attack (Painless myocardial infarction). This test was done with the help of Dyansys (ANSiscope) used to identify the involvement of Cardiac nerves (parasympathetic and sympathetic) due to diabetes.

Data analysis

Statistical analysis was performed using IBM-SPSS version 23. Descriptive statistics were presented either as mean±SD or median (min and max). The statistical significance of the difference between repeated measures was assessed by one-way ANOVA. The choice of the descriptive and inferential statistical methods were based on distribution normality (by normal probability plot). The strength of association between the two variables was assessed by correlation coefficient. A p-value of <0.05 was considered statistically significant and the level of confidence is 95%.

Results

In the present study, out of 384 recruited patients who have completed the study, the average age was found to be 55.5±13.0 years. Regarding the gender of the patients, 232 were males and 183 were females. The average duration of diabetes was found to be 10.8±7.4 years. The BMI of the recruited patients was found to be 27.6±11.6. The fasting and post-prandial blood sugar level was found to be 155.1±54.1 and 205.5±74.1 mg/dl, respectively. The baseline HbA_{1c} value was found to be 8.5±2.1%. Table 1 depicts the demographics and baseline biochemical investigations of the recruited subjects.

Further, the biochemical parameters such as FBS, PPBG, HBA_{1c} were compared at baseline, 3 months, 6 months, and 12 months after the initiation of pharmacotherapy. Meanwhile, the CRP level was compared between baseline, 6th, and 12th months after the initiation of pharmacotherapy.

Comparison of blood glucose levels from baseline and review I–III

The fasting blood sugar level for baseline, review I, II and III was found to be 155.00, 134.63, 119.99, and 108.51 mg/dl. In comparison to baseline values, the FBG levels were significantly ($p<0.05$) decreased in the review I–III.

Further, the post-prandial blood glucose was significantly ($p<0.05$) decreased in review I–III as compared to the baseline. The values were found to be 205.5, 194, 176.7, and 141.8 mg/dl, respectively.

Comparison of glycated hemoglobin levels from baseline and review I–III

The baseline and review I–III values of glycated hemoglobin were found to be 8.5, 8.1, 7.8, and 6.9%. The HbA_{1c} levels in review II and review III were significantly lower as compared to the baseline values.

Correlation between CRP and blood glucose level at baseline, review II and III

In the present study, the CRP showed a significant positive correlation with blood glucose. CRP a showed significant ($p=0.000$) strong correlation with baseline FBG and PPBG level and the values were found to be 0.752 and 0.560. However, CRP showed a significant ($p=0.000$) weak correlation with review II and III FBG and PPBG level and the values were found to be review II (0.298 and 0.175) and review III (0.272 and 0.081), respectively. The results were shown in Table 2.

Table 1: Demographics and baseline biochemical investigations of the recruited subjects.

Parameters	Values (Mean ± SD)	
Demographics		
Duration of diabetes mellitus (years)	10.8±7.4	
Age (years)	55.5±13.0	
Gender	Male	232
	Female	183
Biochemical profiles		
Fasting blood sugar – Baseline (mg/dl)	155.1±54.1	
Post-prandial blood sugar – Baseline (mg/dl)	205.5±74.1	
HbA_{1c} – Baseline (%)	8.5±2.1%	
Blood pressure – Systole (mm/hg)	137.35	
Blood pressure – Diastole (mm/hg)	80.34	
Total cholesterol - Baseline (mg/dl)	170.6±19.3	
Triglycerides (mg/dl)	142.6±60.6	
Low density lipoprotein (mg/dl)	83.5±19.9	
Very low-density lipoprotein (mg/dl)	28.2±10.9	
Serum creatinine (mg/dl)	1.0±0.3	
Estimated glomerular filtration rate (eGFR) mg/dl	87±24.6	
Urine for microalbuminuria mg/dl	28.2±9.4	
C-reactive protein – Baseline (mg/dl)	4.74±0.76	
Nerve conduction velocity (%)	32±14.17	
Ankle brachial index	1.0±0.1	
Vibration perception threshold (volts)	14.0±16.6	

Table 2: Correlations between CRP levels with FBS and HbA_{1c} at various time points.

Independent variable	Dependent variable	Correlation coefficient	p-Value
FBS-baseline	CRP-baseline	0.752	0.000
FBS-6th month	CRP-6th month	0.298	0.000
FBS-12th month	CRP-12th month	0.272	0.000
HbA_{1c}-baseline	CRP-baseline	0.739	0.000
HbA_{1c}-6th month	CRP-6th month	0.429	0.000
HbA_{1c}-12th month	CRP-12th month	0.352	0.000

$p<0.05$ were considered as statistically significant.

Regarding association between lipid profiles and CRP at baseline there was significant positive correlation with total cholesterol (r=0.604; p=0.000), LDL (r=0.115; p=0.024), triglycerides (r=0.104; p=0.04). There was a strong correlation between cholesterol and CRP and a weak correlation for LDL and triglycerides. Further, there was a significantly strong negative correlation between HDL and CRP (r= -0.692; p=0.000). However, there was no significant relationship between VLDL and CRP in the present study (r=0.093; p=0.069). The results were shown in Table 3.

In this study, there was a significant association between baseline diastolic blood pressure and CRP (r=0.119; p=0.020) and but there was no

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Table 3: Correlations between baseline CRP level and lipid profiles.

Independent variable	Dependent variable	Correlation coefficient	p-Value
Tot. cholesterol	CRP-baseline	0.604	0.000
LDL	CRP-baseline	0.115	0.024
HDL	CRP-baseline	-0.692	0.000
VLDL	CRP-baseline	0.093	0.069 NS
Triglycerides	CRP-baseline	0.104	0.042

NS Non Significant. p<0.05 were considered as statistically significant

Table 4: Correlations between baseline CRP level and end organ function.

Independent variable	Dependent variable	Correlation coefficient	p-Value
Urine for micro albuminuria	CRP-baseline	0.169	0.001
Cardiac autonomic neuropathy	CRP-baseline	0.105	0.040
Ankle brachial Index	CRP-baseline	-0.055	0.285 NS
VPT	CRP-baseline	0.056	0.273 NS

NS Non Significant. p<0.05 were considered as statistically significant

significant association between diastolic blood pressure and CRP (r=0.092; p=0.07).

Further, to find out the association between end-organ function and its association with CRP level in type 2 diabetes patients the following markers were analyzed. In this study, the baseline urea for microalbuminuria had displayed a significant association between CRP baseline (r=0.169; p=0.0001). Further, baseline cardiac autonomic neuropathy was significantly associated with baseline CRP (r=0.105; p=0.040). Further, baseline Ankle-brachial Index (r= -0.055; p=0.28) and VPT (r=0.056; p=0.273) were not significantly associated with baseline CRP. The results were shown in Table 4.

Discussion

The present study was conducted to find out the association of inflammatory marker (C-reactive protein), glycated hemoglobin, and circulating lipids with microvascular complications in type 2 diabetes mellitus patients with strict glycemic control in the South Indian population.

The average age among the study participants is found to be 55.5±13.0 years, which is in accordance with the study conducted by Elimam et al. where the average of the diabetic subjects is 50.83±8.26 years [14]. Previous studies show that microvascular complications have a significant association with diabetes duration [15]. In our study, the average duration of diabetes is 10.8±7.4 years. Similar to our report, in a study done by Bhuyan et al. [16]. The average age of diabetes patients with the presence of cardiac autonomic dysfunction (CAD) is 11.56±6.15 years [16]. According to the BMI category, the study participants are overweight with an average BMI of 27.6±11.6 kg/m^2. In a study conducted by Borgharkar et al. [17]. Diabetic subjects with strict glycemic control are overweight with an average BMI of 26.13±3.35 kg/m^2 [17].

In our study, the baseline FBG and PPBG levels are 155.1±54.1 and 205.5±74.1 mg/dl, respectively, and it is near to normal and the baseline HbA_{1c} is 8.5± 2.1%. Similar to our report in diabetic patients with HbA_{1c} level >7% the FBG and PPBG level is reported to be 129.42±18.41 and 204.81±44 mg/dl, respectively [17]. In contrast, a multi-centric ICMR-INDIAB phase I study (n=480) reported a higher (31%) proportion of patients with glycemic control, with a mean HbA_{1c} of 9.1% [18]. Thus elevated HbA_{1c} levels in diabetic patients are prone to develop the risk of micro- and macro-vascular complications.

In this study, baseline lipid profiles total cholesterol, triglycerides, LDL and VLDL are found to be normal among the diabetic subjects.

However, the baseline HDL level is markedly decreased in the range of 35.8±8.5mg/dl. Our result is in accordance with the study done by Gatti et al. [19] where the low HDL cholesterol is strongly interconnected with elevated HbA1C levels. They reported that, 68.5% of patients displayed HbA_{1c} >7 and HDL level was <40–50 mg/dl in 47.4% [19].

Previous studies show that there is a strong association between elevated blood glucose levels and inflammation [20]. During diabetic complications such as nephropathy, retinopathy, and peripheral neuropathy there has been a substantial increase in the level of pro-inflammatory cytokine (TNF-α and IL-6) and acute-phase proteins such as C-reactive protein [CRP] [21]. In this study, the average baseline CRP is 3.5±0.76 mg/dl and it is in corroboration with the study conducted by Gamit et al. where the mean CRP level is 3.08±1.62 mg/dl among the diabetic subjects [22]. Peripheral artery disease (PAD) is none of the major complications of type 2 diabetes mellitus with the amputation of legs. Ankle Brachial Index (ABI) is the reliable parameter to measure PAD among diabetics [23]. The ABI in the present study is 1.0±0.1. Previous reports show that the cut-off value of ABI for the highest sensitivity and specificity for PAD screening is between 1.0 and 1.1 and it is in line with the present study [24]. Vibration Perception Threshold (VPT) is a technique used to evaluate peripheral neuropathy in diabetic patients [25]. In our study, the VPT is 14.0±16.6 volts and it reflects as a mild peripheral neuropathy among diabetic patients.

In the present study, there is a significant correlation between CRP, fasting blood glucose, and HbA_{1c} at baseline, 6 and 12 months (p=0.000). Similar to our report, Roopakala et al. reported a positive significant association between CRP and HbA_{1c} (r=0.887; p=0.008) [26]. Further, we also found a significant correlation between baseline CRP and lipid profiles. HDL is negatively associated with CRP, while total cholesterol shows a strong positive association with CRP. Meanwhile, LDL and triglycerides show a weak positive association with CRP and VLDL is not significantly associated with CRP. Our reports are consistent with the study done by Meriga et al. where cholesterol, triglycerides showed a positive association, whilst the HDL showed a negative association with CRP [27].

In our study, baseline CRP shows a significant correlation with baseline urine for microalbuminuria, cardiac autonomic neuropathy (CAN), whilst ankle-brachial index (ABI) and VPT displayed no significant association. Similar to our study, Mojahedi et al. also reported a significant correlation between HS-CRP level and urine albumin level (r=0.43; p<0.001) [28]. In a study conducted among the Japanese diabetic patient's elevated levels of HSCRP are associated with depressed cardiovascular autonomic function [29]. Thejaswini et al. showed a significant negative correlation between ABI and CRP in diabetics which is a contrast to the present study [30]. On the contrary, Wang et al. reported an increased level of CRP in diabetic peripheral neuropathy which is measured by vibration perception [31].

Conclusion

Subclinical inflammation is a hallmark event in type 2 diabetes mellitus, even in patients with strict glucose control. In this study, CRP level was increased in diabetic subjects and it has a significant association with fasting blood glucose, HbA_{1c}, lipid profiles, urine for microalbuminuria, and CAN. Thus, during diabetes management, monitoring of CRP level was important and strategies to reduce the inflammation are also needed.

Conflict of Interest

The authors declare no conflict of interest.

References

1. Ogurtsova, K., da Rocha Fernandes, J. D., Huang, Y., et al. (2017). IDF Diabetes Atlas: Global estimates for the prevalence of diabetes for 2015 and 2040. *Diabetes Res Clin Pract*. 128:40–50.
2. Chawla, A., Chawla, R., Jaggi, S. (2016). Microvasular and macrovascular complications in diabetes mellitus: Distinct or continuum? *Indian J Endocrinol Metab*. 20(4):546–551.
3. ADVANCE Collaborative Group, Patel, A., MacMahon, S., Chalmers, J., et al. (2008). Intensive blood glucose control and vascular outcomes in patients with type 2 diabetes. *N Engl J Med*. 358:2560–2572.

 https://doi.org/10.46389/rjd-2021-1039

4. Barrett, E. J., Liu, Z., Khamaisi, M., King, G. L., Klein, R., Klein, B. E. K., Hughes, T. M., Craft, S., Freedman, B. I., Bowden, D. W., Vinik, A. I., Casellini, C. M. (2017). Diabetic microvascular disease: An endocrine society scientific statement. *J Clin Endocrinol Metab*. 102(12):4343–4410.
5. Domingueti, C. P., Dusse, L. M., Carvalho, Md., de Sousa, L. P., Gomes, K. B., Fernandes, A. P. (2016). Diabetes mellitus: The linkage between oxidative stress, inflammation, hypercoagulability and vascular complications. *J Diabetes Complic*. 30(4):738–745.
6. Donate-Correa, J., Martín-Núñez, E., Muros-de-Fuentes, M., Mora-Fernández, C., Navarro-González, J. F. (2015). Inflammatory cytokines in diabetic nephropathy. *J Diabetes Res*. 2015:948417.
7. Herder, C., Carstensen, M., Ouwens, D. M. (2013). Anti-inflammatory cytokines and risk of type 2 diabetes. Diabetes Obes Metab. 15(Suppl 3):39–50.
8. Coppola, G., Corrado, E., Muratori, I., Tantillo, R., Vitale, G., Lo Coco, L., Novo, S. (2006). Increased levels of C-reactive protein and fibrinogen influence the risk of vascular events in patients with NIDDM. *Int J Cardiol*. 106(1):16–20.
9. F. Guerrero-Romero, F., Rodriguez-Moran, M. (2003). Relation of C-reactive protein to features of the metabolic syndrome in normal glucose tolerant, impaired glucose tolerant, and newly diagnosed type 2 diabetic subjects. *Diabetes Metab*. 29:65–71.
10. Mahajan, A., Jaiswal, A., Tabassum, R., Podder, A., Ghosh, S., Madhu, S. V., et al. (2012). Elevated levels of C-reactive protein as a risk factor for metabolic syndrome in Indians. *Atherosclerosis* 220:275–281.
11. Song, J., Chen, S., Liu, X., Duan, H., Kong, J., Li, Z. (2015). Relationship between C-reactive protein level and diabetic retinopathy: A systematic review and meta-analysis. *PLoS One*. 10(12):e0144406.
12. Shaheer, A. K., Tharayil, J. K., Krishna, P. W. (2017). A comparative study of high sensitivity C-reactive protein and metabolic variables in type 2 diabetes mellitus with and without nephropathy. *J Clin Diagn Res*. 11(9):BC01–BC04.
13. Del Cañizo Gómez, F. J., Fernández Pérez, C., Moreno Ruiz, I., de Gorospe Pérez-Jáuregui, C., Silveira Rodríguez, B., González Losada, T., Segura Galindo, A. (2011). Microvascular complications and risk factors in patients with type 2 diabetes. Endocrinol Nutr. 58(4):163–168.
14. Elimam, H., Abdulla, A. M., Taha, I. M. (2019). Inflammatory markers and control of type 2 diabetes mellitus. *Diabetes Metab Syndr*. 13(1):800–804.
15. Zoungas, S., Woodward, M., Li, Q., Cooper, M. E., Hamet, P., Harrap, S., et al. (2014). Impact of age, age at diagnosis and duration of diabetes on the risk of macrovascular and microvascular complications and death in type 2 diabetes. Diabetologia. 57(12):2465–2474.
16. Bhuyan, A. K., Baro, A., Sarma, D., Choudhury, B. (2019). A study of cardiac autonomic neuropathy in patients with type 2 diabetes mellitus: A Northeast India experience. *Indian J Endocr Metab*. 23:246–250.
17. Borgharkar, S. S., Das, S. S. (2019). Real-world evidence of glycemic control among patients with type 2 diabetes mellitus in India: the TIGHT study. *BMJ Open Diabetes Res Care*. 7(1):e000654. doi: 10.1136/bmjdrc-2019-000654. PMID: 31413840; PMCID: PMC6673766.
18. Unnikrishnan, R., Anjana, R. M., Deepa, M., et al. (2014). Glycemic control among individuals with self-reported diabetes in India--the ICMR-INDIAB Study. *Diabetes Technol Ther* 16:596–603.
19. Gatti, A., Maranghi, M., Bacci, S., et al. (2009). Poor glycemic control is an independent risk factor for low HDL cholesterol in patients with type 2 diabetes. *Diabetes Care*. 32(8):1550–1552. doi: 10.2337/dc09-0256.
20. Olefsky, J. M., Glass, C. K. et al. (2010). Macrophages, inflammation, and insulinresistance. *Annu Rev Physiol*. 72:219–246.
21. Nguyen, D. V., Shaw, L. C., Grant, M. B. (2012). Inflammation in the pathogenesis of microvascular complications in diabetes. *Front Endocrinol (Lausanne)*. 3:170.
22. Gamit, N. C., Kantharia, N. D., Vaghasiya, K. B., Vataliya, A. J., Shah, A. B. (2015). Study of effects of metformin on C-reactive protein level in type-2 diabetes mellitus. *Int J Basic Clin Pharmacol*. 4:46–50.
23. Potier, L., Abi, K. C., Mohammedi, K., Roussel, R. (2011). Use and utility of ankle brachial index in patients with diabetes. *Eur J Vasc Endovasc Surg*. 41(1):110–116.
24. Nukumizu, Y., Matsushita, M., Sakurai, T., Kobayashi, M., Nishikimi, N., Komori, K. Comparison of Doppler and oscillometric ankle blood pressure measurement in patients with angiographically documented lower extremity arterial occlusive disease. *Angiology* 58:303e8.
25. Dash, S., Thakur, A. K. (2017). Perception of vibration threshold is a marker of diabetic neuropathy. *Natl J Physiol Pharm Pharmacol*. 7(9):1003–1006.
26. Roopakala, M. S., Pawan, H. R., Krishnamurthy, U., Wilma Delphine Silvia, C. R., Eshwarappa, M., Prasanna Kumar, K. M. (2012). Evaluation of high sensitivity creactive protein and glycated hemoglobin levels in diabetic nephropathy. *Saudi J Kidney Dis Transpl*. 23:286–289.
27. Meriga, R. K., Nareddy, V. A., Bhavya, S. V. S. S., Sreekeerthi, C. (2020). Correlation between glycemic control, lipid profile and C-reactive protein in adults with type 2 diabetes mellitus done in a tertiary care hospital of Nellore, Andhra Pradesh, India. *Int J Adv Med*. 7:1312–1317.
28. Mojahedi, M. J., Bonakdaran, S., Hami, M., Sheikhian, M. R., Shakeri, M. T., Aiatollahi, H. (2009). Elevated serum C-reactive protein level and microalbuminuria in patients with type 2 diabetes mellitus. *Iran J Kidney Dis*. 3(1):12–16.
29. Anan, F., Takahashi, N., Nakagawa, M., Ooie, T., Saikawa, T., Yoshimatsu, H. (2005). High-sensitivity C-reactive protein is associated with insulin resistance and cardiovascular autonomic dysfunction in type 2 diabetic patients. *Metabolism*. 54(4):552–558.
30. Thejaswini, K. O., Roopakala, M. S., Dayananda, G., Chandrakala, S. P., Prasanna Kumar, K. M. (2013). Study of association of ankle brachial index (ABI) and the highly sensitive C - reactive protein (hsCRP) in type 2 diabetic patients and in normal subjects. *J Clin Diagn Res*. 7(1):46–50.
31. Wang, D. D., Bakhotmah, B. A., Hu, F. B., Alzahrani, H. A. (2014). Prevalence and correlates of diabetic peripheral neuropathy in a Saudi Arabic population: a cross-sectional study. *PLoS One*. 9(9):e106935. doi: 10.1371/journal.pone.0106935. PMID: 25184511; PMCID: PMC4153691.