

## Evaluation of Cardiovascular Biomarkers and Lipid Profiles in Diabetic Patients Undergoing Coronary Artery Bypass Grafting (CABG) Surgery Compared to a Control Group

Hosein Mohammadzadeh<sup>1</sup>, Morteza Golbashirzadeh<sup>2</sup> and Atousa Moradzadegan<sup>1</sup>

<sup>1</sup>Department of Biology, Islamic Azad University, Dezful, Iran; <sup>2</sup>Department of Pharmaceutical Biotechnology, Chabahar University of Medical Science, Iran

### Correspondence:

Atousa Moradzadegan, PhD,  
Department of Biology, Dez.C.,  
Islamic Azad University, Dezful,  
Iran.  
E-mail: Atousa.moradzadegan@gmail.com

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### ABSTRACT

**OBJECTIVE** Cardiovascular diseases (CVDs), particularly coronary artery disease (CAD), remain among the leading causes of morbidity and mortality worldwide. Type 2 diabetes mellitus (T2DM) significantly accelerates the progression of CAD through metabolic disturbances, including dyslipidemia and elevated homocysteine levels. This study aimed to evaluate cardiovascular biomarkers and lipid profiles in diabetic patients undergoing coronary artery bypass grafting (CABG) surgery, compared to a non-diabetic control group.

**METHODS** A total of 200 participants were included in this study, consisting of 100 patients with type 2 diabetes who had undergone CABG or angioplasty, and 100 age-matched individuals with type 2 diabetes without any history of cardiovascular disease, who served as controls.

**RESULTS** Homocysteine levels were elevated in 75.0% of diabetic patients versus 58.0% of controls ( $p = 0.03$ ). Low high-density lipoprotein cholesterol (HDL-C) was observed in 93.0% of diabetics compared to 68.0% of controls ( $p = 0.01$ ). No statistically significant differences were found in total cholesterol, low-density lipoprotein cholesterol (LDL-C), troponin, or creatine kinase-MB (CK-Mb) levels between groups ( $p > 0.05$ ). These findings suggest that hyperhomocysteinemia and low HDL-C are significantly associated with CAD in diabetic populations. The presence of elevated homocysteine in a substantial proportion of the control group may reflect subclinical metabolic dysfunction or undiagnosed risk factors, warranting further investigation.

**CONCLUSIONS** These results underscore the potential utility of homocysteine and HDL-C as biomarkers in cardiovascular risk stratification. Their integration into routine screening protocols may enhance early detection and preventive strategies, particularly in high-risk or resource-limited settings.

**KEYWORDS** homocysteine, high-density lipoprotein cholesterol, type 2 diabetes, coronary artery disease, biomarkers

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## INTRODUCTION

Cardiovascular diseases (CVDs) continue to be the leading cause of morbidity and mortality worldwide, posing a substantial burden on health-care systems and societal resources (1). Among these, coronary artery disease (CAD) is the most prevalent, resulting from the narrowing or blockage of coronary arteries due to atherosclerosis (2). The interplay between metabolic disorders, such as type 2 diabetes mellitus (T2DM), and the progression of CAD has garnered significant attention, with mounting evidence underscoring the role of metabolic abnormalities in exacerbating cardiovascular risks. T2DM, characterized by chronic hyperglycemia and insulin resistance, contributes to vascular dysfunction and amplifies the risk of myocardial infarction and other ischemic events. This intersection of diabetes and cardiovascular health underscores the critical need for understanding the mechanisms that underpin these diseases (3).

In recent years, the role of homocysteine, a sulfur-containing amino acid derived from methionine metabolism, has emerged as a pivotal biomarker in cardiovascular research (4). Hyperhomocysteinemia has been implicated in endothelial dysfunction, oxidative stress, and pro-inflammatory states, all of which contribute to the pathogenesis of atherosclerosis. Studies by Tomić M et al. have shown that elevated plasma homocysteine levels are linked to nonproliferative retinopathy in individuals with T2DM who do not have renal disease (5). Similarly, Smith et al. reported that more than half of patients undergoing angioplasty presented with high plasma homocysteine levels, further emphasizing its relevance in cardiovascular care (6). Despite these insights, the clinical utility of homocysteine as a routine diagnostic marker remains contested, particularly in resource-constrained settings where cost considerations limit its widespread use.

Lipid profile abnormalities are another cornerstone of cardiovascular risk assessment. High-density lipoprotein cholesterol (HDL-C), often termed “good cholesterol,” plays a protective role in cardiovascular health by facilitating reverse cholesterol transport and exerting anti-inflammatory effects (7). Conversely, low-density lipoprotein cholesterol (LDL-C) and triglycerides (TG) are commonly associated with an increased risk of atherosclerosis and vascular events (8). M

Georgoulis (2022), explored the link between lipid abnormalities and T2DM, noting that low HDL-C and high LDL-C levels significantly contribute to vascular obstruction and hypertension (9). However, the relative importance of these lipid parameters in CAD progression remains a subject of debate, necessitating further investigation into their individual and combined effects.

Biochemical markers such as troponin and creatine kinase-MB (CK-Mb) enzymes have long been recognized as gold standards for diagnosing acute myocardial infarction (MI). However, their role in predicting chronic atherosclerotic changes is less clear. While elevated troponin and CK-Mb levels are indicative of myocardial injury, these markers do not necessarily reflect the underlying pathophysiological processes leading to CAD. This distinction highlights the need for a multifactorial approach to cardiovascular risk stratification, incorporating both traditional markers and emerging biomarkers like homocysteine (10).

Demographic characteristics also significantly influence cardiovascular risks. Existing literature highlights a higher prevalence of CAD among older adults and males, reflecting the multifaceted nature of cardiovascular risk factors. These differences emphasize the importance of personalized preventive strategies and targeted interventions (11).

The interplay of diabetes, lipid abnormalities, and elevated homocysteine levels offers a compelling area for research, particularly in understanding the mechanisms underlying CAD in diabetic populations. While hyperhomocysteinemia and dyslipidemia are independently linked to increased cardiovascular risk, their combined effects remain underexplored (12). Moreover, confounding factors such as age, gender, and comorbidities further complicate the assessment of these relationships. Conditions including chronic kidney disease, hypothyroidism, and certain medication regimens—such as methotrexate or antiepileptics—may also contribute to elevated homocysteine levels, underscoring the complexity of risk stratification (13).

Despite advances in cardiovascular research, critical gaps remain in elucidating the pathophysiological connections between diabetes, lipid irregularities, and vascular health. For instance, the role of hyperhomocysteinemia as either a causative factor or a consequence of T2DM remains ambiguous (14). Similarly, the protective role of HDL-C in mitigating cardiovascular risks in dia-

betic patients warrants further investigation, especially given the challenges of pharmacologically increasing HDL-C levels. Bridging these knowledge gaps is vital for developing predictive models and effective therapeutic strategies aimed at improving outcomes for patients with diabetes and CAD (15).

## METHODS

### Rationale and sample size determination

The rationale for this study stems from the need to clarify how metabolic markers—particularly homocysteine and lipid profiles—interact in the development of CAD among patients with T2DM. Although each marker is independently associated with cardiovascular risk, their combined predictive value remains underexplored. By investigating these markers in a comparative framework, this study aims to generate insights that could inform more precise screening and intervention strategies for high-risk populations.

To ensure adequate statistical power and reliable findings, the sample size was calculated using Cochran's formula for cross-sectional studies. The prevalence of CAD in diabetic patients was considered, alongside a 95.0% confidence level and 5.0% margin of error, to determine the minimum number of participants required. This study analyzed 200 samples: 100 patients with type 2 diabetes who had undergone CABG or angioplasty, and 100 diabetic individuals under 50 years of age without any history of cardiac disease, who were selected as the control group. During the 6-month study period, 132 CABG surgeries were performed at Ahvaz Oil Hospital, of which 100 patients (77.8%) had type 2 diabetes. Due to the high prevalence of diabetes among CABG patients, selecting an equal number of non-diabetic controls was not feasible; therefore, diabetic individuals without cardiac disease were used as controls.

### Participant selection and eligibility criteria

Eligible participants were selected through purposive sampling of patients receiving care at Naft Hospital, Ahvaz, Iran. The study group included individuals aged 40–70 years with a confirmed diagnosis of T2DM and documented CAD, based on clinical, biochemical, and diagnostic criteria outlined by the American Diabetes Association (ADA) 2016 Guidelines and cardiovascular evaluation protocols. The control group consisted of age-

and gender-matched individuals without a history of diabetes or significant cardiovascular events. All participants were required to provide informed consent. Exclusion criteria for both groups included a history of chronic kidney disease, liver dysfunction, autoimmune disorders, current use of lipid-lowering or homocysteine-modulating medications, acute infections or inflammatory conditions at the time of enrollment, pregnancy or lactation, and incomplete medical records or refusal to undergo laboratory testing.

### Fasting blood sugar (FBS)

FBS levels were measured using a glucose oxidase-peroxidase (GOD-POD) enzymatic assay (Azar Tashkhis Azma, Tehran, Iran). Serum samples were collected after 10–12 hours of fasting. Standard quality control procedures were followed using normal and pathological serum controls.

### Hemoglobin A1C (HbA1C)

Whole blood samples collected in K3-EDTA tubes were analyzed using an automated immunoassay system (Roche cobas c series).

### Total cholesterol

Total cholesterol was measured enzymatically using the Cholesterol oxidase-Peroxidase (CHOD-POD) method (Azar Tashkhis Azma, Tehran, Iran). Serum samples were obtained after overnight fasting. Pre-analytical factors such as posture and tourniquet application were controlled. Samples were stored at 2–8°C prior to analysis.

### Triglycerides (TG)

TG levels were determined using the glycerol phosphate oxidase-Peroxidase (GPO-POD) enzymatic method. Serum samples were collected after 10–14 hours of fasting. EDTA plasma was used when applicable. Lifestyle factors (e.g., alcohol intake, exercise) were considered during interpretation.

### Low-density lipoprotein (LDL)

LDL cholesterol was calculated using the Friedewald formula:  $LDL = \text{total cholesterol} - HDL - (TG/5)$ . This method was applied only when fasting triglyceride levels were  $< 400$  mg/dL. Indirect estimation was avoided in cases of chylomicronemia or familial dysbetalipoproteinemia.

## Homocysteine

Homocysteine levels were measured using an enzymatic immunoassay (Bio-Rad, USA). Plasma (EDTA or heparinized), serum, and urine samples were collected after 10–12 hours of fasting. Samples were processed and frozen within one hour to preserve stability.

## Statistical analysis

All statistical analyses were performed using IBM SPSS Statistics version 26.0. Categorical variables, including HCN, HDL-C, LDL-C, FBS, troponin (TN.L), HbA1C, CK-MB, triglycerides (TG), and cholesterol (CHO), were summarized as frequency and percentage. Group differences in categorical variables were assessed using the Chi-square test, and Fisher's exact test was applied when expected cell counts were below 5.

Continuous variables were expressed as mean  $\pm$  standard deviation (SD), and comparisons between patients and controls were performed using the independent samples t-test after confirming normality with the Kolmogorov–Smirnov test.

All statistical tests were two-tailed, and a  $p < 0.05$  was considered statistically significant.

## RESULTS

A total of 200 participants were included in

the analysis, consisting of 100 diabetic patients with cardiovascular involvement (CABG or angioplasty) and 100 diabetic controls without any history of cardiac disease. The distribution of biochemical and metabolic parameters between the two groups is presented in Table 1. Homocysteine (HCN) levels were significantly higher in patients compared with controls (75.0% vs 56.0%,  $p = 0.004$ ). HDL-C levels also differed markedly, with 93.0% of patients classified as “Low” compared with 68.0% of controls, while 28.0% of controls demonstrated “High” HDL-C values ( $p < 0.001$ ). LDL-C distribution showed a significant difference, with controls exhibiting a higher proportion of “High” LDL-C (36.0% vs 17.0%,  $p = 0.001$ ).

FBS levels were elevated in both groups, with no statistically significant difference (97.0% vs 96.0%,  $p = 0.700$ ). Troponin (TN.L) positivity was observed only in patients (4.0%), resulting in a significant difference ( $p = 0.043$ ). HbA1C levels were uniformly high in both groups ( $p = 0.316$ ). CK-MB elevation was substantially more frequent in patients (51.0% vs 0%), indicating a strong association with cardiovascular involvement ( $p < 0.001$ ). Significant differences were also observed in triglyceride and cholesterol (CHO) distributions between groups ( $p = 0.048$  and  $p = 0.002$ , respectively). These findings highlight the metabolic and biochemical disturbances associated with cardiovascular disease in individuals with type 2 diabetes.

**Table 1.** Biochemical and metabolic parameter distribution among diabetic cardiac patients and diabetic controls, with corresponding P-values calculated using Chi-square or Fisher's exact test.

| Parameter       | Category               | Patients<br>n (%)                    | Controls<br>n (%)                    | p-value  |
|-----------------|------------------------|--------------------------------------|--------------------------------------|----------|
| HCN             | High/Normal            | 75 (75.0%) / 25 (25.0%)              | 56 (56.0%)/44 (44.0%)                | 0.004    |
| HDL-C           | Low/Normal/High        | 93 (93.0%) / 7 (7.0%)/<br>0 (0.0%)   | 68 (68.0%)/4 (4.0%)/<br>28 (28.0%)   | < 0.001  |
| LDL-C           | Normal/Borderline/High | 62 (62.0%)/21 (21.0%)/<br>17 (17.0%) | 64 (64.0%)/0 (0.0%)/<br>36 (36.0%)   | 0.001    |
| FBS             | High/Normal            | 97 (97.0%)/3 (3.0%)                  | 96 (96.0%)/4 (4.0%)                  | 0.700    |
| Troponin (TN.L) | Positive/Negative      | 4 (4.0%)/96 (96.0%)                  | 0 (0.0%)/100 (100.0%)                | 0.043*   |
| HbA1C           | High/Normal            | 99 (99.0%)/1 (1.0%)                  | 100 (100.0%)/0 (0.0%)                | 0.316*   |
| CK-MB           | High/Normal            | 51 (51.0%) / 49 (49.0%)              | 0 (0.0%)/100 (100.0%)                | < 0.001* |
| TG              | Normal/Borderline/High | 85 (85.0%)/5 (5.0%)/<br>10 (10.0%)   | 77 (77.0%)/4 (4.0%)/<br>19 (19.0%)   | 0.048    |
| CHO             | Normal/Borderline/High | 84 (84.0%)/8 (8.0%)/<br>8 (8.0%)     | 61 (61.0%)/21 (21.0%)/<br>18 (18.0%) | 0.002    |

HCN, homocysteine; HDL-C; high-density lipoprotein; LDL-C, low-density lipoprotein, FBS, fasting blood sugar; TN.L, troponin; HbA1C; hemoglobin A1c, CK-MB; creatine kinase-MB, TG; triglyceride, CHO; cholesterol

### Analysis of comparison

A statistically significant difference was observed in homocysteine levels, with a higher proportion of “High” HCN among patients compared to controls ( $p = 0.004$ ). HDL-C levels also differed markedly, as 93.0% of patients exhibited “Low” HDL-C compared with 68.0% of controls, while 28.0% of controls demonstrated “High” HDL-C ( $p < 0.001$ ). LDL-C distribution showed a significant shift toward the “High” category in controls (36.0% vs 17.0%,  $p = 0.001$ ). FBS levels were elevated in both groups without a significant difference ( $p = 0.700$ ). Troponin positivity was detected only in patients ( $p = 0.043$ ). HbA1C levels were uniformly high in both groups ( $p = 0.316$ ). CK-MB elevation was significantly more frequent in patients (51.0% vs 0%,  $p < 0.001$ ). Significant differences were also observed in triglyceride ( $p = 0.048$ ) and cholesterol ( $p = 0.002$ ) distributions, with controls showing higher proportions of borderline and high values. These findings collectively demonstrate substantial biochemical and metabolic disparities between diabetic patients with cardiovascular involvement and diabetic controls.

### DISCUSSION

This study investigated the relationship between T2DM, metabolic markers, and cardiovascular risk factors, focusing on patients undergoing CABG at Ahvaz Naft Hospital. During the 6-month period, 132 patients underwent CABG at Ahvaz Oil Hospital, among whom 102 individuals (77.8%) had type 2 diabetes. The final study sample consisted of 200 participants: 100 diabetic patients with cardiovascular involvement (CABG or angioplasty) and 100 diabetic individuals without any history of cardiac disease who served as controls. The patient group had a mean age of  $62.4 \pm 13.0$  years, with 74.0% males and 26.0% females, while the control group had a mean age of  $43 \pm 11$  years, comprising 59.0% males and 41.0% females. These demographic differences reflect the age- and gender-related susceptibility to cardiovascular disease in diabetic populations. Our findings align with prior research. Yeromenko and Lavie identified diabetes as the most prevalent metabolic disorder in the Western world and emphasized its significant role in CAD and mortality (17). In the present study, elevated homocysteine levels were observed in 75.0% of diabetic

patients, consistent with Schnyder and Roffi, who reported high plasma homocysteine in over 50.0% of angioplasty patients (18).

This supports its role as a vascular risk factor. However, the prevalence observed in this study was notably higher, possibly due to differences in patient selection or regional dietary patterns. Lipid profile abnormalities were also pronounced, particularly low HDL-C levels. A striking 93.6% of diabetic patients demonstrated HDL-C levels below the normal range, compared to 68.0% in the control group (19). This is significant, as HDL-C is a protective lipoprotein that facilitates cholesterol removal and reduces cardiovascular risk (20). In contrast, LDL-C levels were less concerning, with 59.1% of patients having normal LDL values, suggesting that while LDL contributes to atherosclerosis, its role in this cohort appears secondary to other factors such as homocysteine and HDL-C (21). Further analysis revealed that total cholesterol was normal in 84.0% of patients, while triglycerides were within normal limits for 85% of the control group. These findings suggest that low HDL-C is the primary lipid abnormality in this population, consistent with XC Jiang, who demonstrated a strong correlation between high homocysteine, low HDL-C, and vascular complications in diabetic patients (22). Interestingly, the utility of certain biomarkers like troponin and CK-Mb for predicting atherosclerosis was limited. While these markers are critical for diagnosing MI, they did not demonstrate significant predictive value for CAD susceptibility in this cohort. The elevated CK-Mb levels in 49.1% of patients were likely attributable to trauma from surgical procedures, such as angioplasty or CABG, rather than underlying disease (23, 24).

From a clinical perspective, the combined assessment of FBS, A1C, HDL-C, LDL-C, and homocysteine offers a robust strategy for identifying individuals at high risk of CAD (25). Notably, FBS and A1C remain critical for diagnosing and monitoring diabetes, while lipid profiles and homocysteine levels provide additional insights into cardiovascular risk. Interventions such as improving HDL-C through dietary modifications, pharmacological therapies, and the administration of B vitamins (B12 and folic acid) to reduce homocysteine levels are essential components of comprehensive care for diabetic patients (26).

### Study limitations

Several limitations should be acknowledged. First, the age and gender distribution between groups was not fully matched, which may have introduced bias in interpreting cardiovascular risk. Second, the sample size, while adequate for preliminary analysis, limits generalizability and statistical power for subgroup comparisons. Third, the inclusion of patients undergoing CABG introduces procedural variability, as surgical stress may influence biomarker levels such as CK-Mb. Fourth, while standard laboratory assays were used, inter-assay variability and lack of centralized calibration may have affected measurement consistency. Finally, lifestyle factors such as diet, physical activity, and medication adherence were not controlled, which could have confounded metabolic marker interpretation.

Future studies should aim for larger, matched cohorts and incorporate longitudinal designs to better assess causality and biomarker dynamics over time. Comparative analysis across geographic regions and inclusion of genetic and lifestyle variables may further refine cardiovascular risk models in diabetic populations.

### CONCLUSIONS

In conclusion, diabetic patients with elevated homocysteine levels, low HDL, and high LDL are at the greatest risk for coronary artery disease. A multifactorial approach, including stringent diabetes control, HDL enhancement, and LDL and homocysteine reduction, is essential for effective management and prevention of cardiovascular complications.

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### CONFLICTS OF INTEREST

The authors declare that they have no conflict of interest.

### AUTHOR CONTRIBUTIONS

H.M: formal analysis, resources, conceptualization, software, validation, methodology; M.G: writing — review & editing, writing — original

draft, A.M: preparation, visualization, investigation, supervision, project administration, funding acquisition.

All authors have read and agreed to the published version of the manuscript.

### DATA AVAILABILITY STATEMENT

The original findings of this study are provided in the article and the supplementary material.

### INSTITUTIONAL REVIEW BOARD STATEMENT

All experimental procedures were reviewed and approved by the Ethics Committee of Islamic Azad University, Dezful Branch, under the approval code 16530520962002. The project was conducted in August 2018 and fully complied with the NIH Guidelines for the Care and Use of Laboratory Animals (NIH Publication No. 85-23, revised 1996).

### Consent to participate

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

### INFORMED CONSENT STATEMENT

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

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