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Original Research

Total bilirubin and fasting plasma glucose levels are associated with coronary collateral development in elderly patients

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Abstract

Background and objective: We aimed to investigate biochemical factors affecting coronary collateral circulation development in an elderly population aged 75 years and over.

Material and methods: The study group consisted of patients with a prior coronary angiography for stable coronary artery disease (CAD). Patients with total occlusion of at least one vessel were included in the study. Enrolled patients were divided into two groups, good collateral (GC; n = 73) and bad collateral (BC; n = 55), in accordance with the Cohen-Rentop's classification system.

Results: In comparison to the GC group, bilirubin levels were significantly lower ($p < 0.001$), and fasting plasma glucose (FPG) levels were significantly higher in the BC group ($p = 0.026$). Low-density lipoprotein cholesterol (LDL-C) and high-density lipoprotein cholesterol (HDL-C) levels were significantly lower in the BC group when compared to the GC group ($p = 0.002$ and $p < 0.001$, respectively). Backward elimination stepwise logistic regression analysis identified bilirubin and FPG as variables that strongly predicted the presence of a well-developed coronary collateral circulation and a poorly developed coronary collateral circulation, respectively.

Conclusion: Bilirubin and FPG were seemed as the most important factors affecting coronary collateral circulation development in patients with stable CAD who were older than 75 years.

Keywords

Collateral; Bilirubin; Coronary artery disease; Older patients

1. Introduction

Coronary artery disease (CAD) remains a major cause of all-cause mortality [1]. The coronary collateral circulation can be considered as the body's defense mechanism against coronary artery disease. Coronary collaterals are believed to be present from birth, but their number, size, and the extent of their coverage increases in the presence of an epicardial coronary artery occlusion [2, 3]. A previously published study found that when intermittent ischemia was induced, collateral vessels that were non-functional at rest became functional with exercise [4]. Well-developed coronary collateral arteries have been associated with smaller necrotic areas and ventricular aneurysm formation in patients with acute myocardial infarction, resulting in improved heart remodeling, as well as reduced ischemia and chest pain in chronically ill patients [5–7]. The development of coronary collateral vessels is thought to occur via both angiogenesis, the formation of new vessels through sprouting of new capillaries from existing blood vessels, and via arteriogenesis, the growth and maturation of existing anastomotic channels between coronary arteries. These two mechanisms can act concomitantly, and also independently, following initial blood vessel formation [8].

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TABLE 1. Demographic and clinical characteristics of the patients in the good collateral (GC) and bad collateral (BC) groups.

	GC group (n = 73)	BC group (n = 55)	*p*-value
Age (± SD, year)	77.8 ± 2.6	77.4 ± 2.2	0.345
Sex			0.840
Male	40 (54.8%)	32 (58.2%)	
Female	33 (45.2%)	23 (41.8%)	
HT	37 (50.7%)	35 (63.6%)	0.200
DM	36 (49.3%)	26 (47.3%)	0.819
HL	51 (70.8%)	27 (55.1%)	0.114
History of smoking	24 (32.9%)	14 (25.5%)	0.475
History of myocardial infarction	29 (39.8%)	21 (38.0%)	0.886
Ejection fraction	50 ± 10	48 ± 9	0.696
Number of chronically occluded coronary vessels			0.083
Single vessel	47 (64.4%)	44 (80.0%)	
Two vessels	26 (35.6%)	11 (20.0%)	
Vessel involvement			>0.999
Single vessel	9 (12.3%)	7 (12.7%)	
Multiple vessels	64 (87.7%)	48 (87.3%)	
LAD	31 (42.5%)	21 (38.2%)	0.759
RCA	30 (41.1%)	24 (43.6%)	0.915
CX	12 (16.4%)	10 (18.2%)	0.982
Rentrop grade			N/A
0	N/A	10 (18.2%)	
1	N/A	45 (81.8%)	
2	46 (63.0%)	N/A	
3	27 (37.0%)	N/A	

HT, hypertension; DM, diabetes mellitus; HL, hyperlipidaemia; LAD, Left anterior descending artery; Cx, Circumflex artery; RCA, right coronary artery, N/A, Not applicable.

Age is one of the most important risk factors for CAD. More than half of the deaths associated with CAD occur in individuals older than 70 years [9]. The diagnosis of CAD in the elderly population can be more challenging. In elderly patients, CAD is more diffuse and severe anatomically, and presents with greater calcification [10]. As elderly patients frequently present with multiple comorbidities, conservative approaches rather than invasive options are commonly used to treat CAD, despite studies showing the beneficial effects invasive strategies among patients older than 75 years with stable angina pectoris [11]. The presence of well-developed coronary collaterals in older patients may also play an effective role in preventing ischemia. In the current study, the aim was to determine clinical and biochemical factors affecting the development of coronary collaterals in an elderly population.

2. Materials and methods

This retrospective study included patients older than 75 years who underwent coronary angiography for stable CAD in the Department of Cardiology, Gaziantep University between 2014 and 2017. All of the patients had class 3 or 4 angina according to The Canadian Cardiovascular Society (CCS) Angina Score. Coronary angiograms of the patients were reviewed in a double-blinded fashion by two experienced invasive cardiologists. Patients with at least one completely occluded coronary vessel, as demonstrated by coronary angiography, were enrolled in the study. The angiographic characteristics of the patients were also assessed (e.g., single-vessel versus multiple-vessel disease). The development of coronary collaterals was graded according to the Cohen-Rentrop's classification method (grade 0 = no visible filling of any collaterals with contrast agent; grade 1 = filling of the side branches of the infarct-related artery, without visualization of the epicardial segment; grade 2 = partial filling of the epicardial segment with collateral vessels; and grade 3 = complete filling of the epicardial segment by collateral vessels) [12]. For patients with multiple collaterals, the vessel with the highest collateral grade was recorded.

The patients were divided into two groups based on their Cohen–Rentrop grade. Patients scoring 0–1, poor coronary collateralization, were assigned to the bad collateral group (BC group; n = 55), whereas patients with a Cohen–Rentrop score of 2–3, well-developed coronary collateralization, were assigned to the good collateral group (GC group; n = 73). Clinical and demographic characteristics for each patient were retrieved from the hospital records. Venous blood samples were drawn from all patients at least 12 h prior to coronary angiography in order to undertake a fasting biochemistry panel and a complete blood count.

The study protocol was reviewed and approved by the institutional ethics committee in accordance with the Declaration of Helsinki.

Patients with known peripheral artery disease, a history of cerebrovascular events or acute coronary syndrome, autoimmune diseases or cancer, renal failure or hepatic dysfunction

were excluded, as were patients who had undergone a coronary intervention within the last month.

The Kolmogorov–Smirnov test was used to analyse whether continuous numerical variables were normally distributed. The homogeneity of variance was assessed using the Levene's test. Descriptive statistics are presented as mean ± standard deviation for continuous numerical variables or median (the width of inter-quartile ranges). Categorical variables are expressed as the number of cases (N) and percentage (%).

The significance of mean between-group differences was assessed using the Student's *t* test. The Mann–Whitney *U* test was used to analyse significant differences in continuous numerical variables when assumptions for parametric test statistics were not met. Categorical variables were analysed using a chi-square test corrected for continuity or Pearson's chi-square test.

Backward elimination stepwise logistic regression analysis was used to determine the variables that strongly predicted "good" or "bad" collateral development. Subsequently, variables with a p value of <0.10 from the univariate statistical analyses were included in a multivariate logistic regression model as candidate risk factors. Odds ratios, with their 95% confidence intervals (95% CI) and Wald statistics were estimated for each variable.

Data analyses were performed using an IBM SPSS software package, version 17.0 (IBM Corporation, Armonk, NY, USA). A p value of <0.05 was considered as statistically significant.

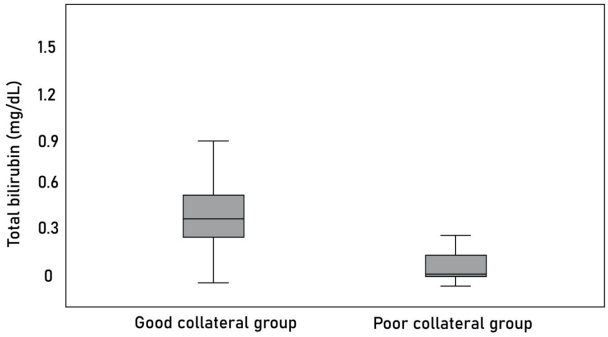

FIG. 1. Comparison of serum bilirubin levels in good and poor collateral groups.

3. Results

There were no statistically significant differences between the GC and BC groups with respect to the mean age and sex distribution of the patients (p = 0.345 and p = 0.840, respectively). There were also no significant between-group differences relating to the patients histories of hypertension, diabetes mellitus or hyperlipidaemia ($p > 0.05$). The number of chronically occluded coronary arteries, vessels involved, and distribution of the vessels were not statistically significantly different between the two groups ($p > 0.05$; Table 1). The ejection fraction was not statistically significantly different between the two groups ($p > 0.05$; Table 1). In total, 21 patients within the poor collateral group and 28 patients from the good collateral group had a history of myocardial infarction (MI). There were no statistical differences between the two groups which related to their history of MI.

Comparisons between the results from the biochemical panel for the two groups are presented in Table 2. The low-density lipoprotein (LDL) and high-density lipoprotein (HDL) levels were significantly lower in the BC group than observed in the GC group (p = 0.002 and $p < 0.001$, respectively). In comparison to the GC group, the BC group levels of bilirubin were significantly lower ($p < 0.001$; Fig. 1), and fasting plasma glucose (FPG) were significantly higher (p = 0.026). There were no significant between-group differences in any other biochemical parameters ($p > 0.05$). In addition, the distribution of medications used was not statistically significantly different between the GC and BC groups ($p > 0.05$; Table 3).

TABLE 2. Results of biochemical measurements in the good collateral (GC) and bad collateral (BC) groups.

	GC group (n = 73)	BC group (n = 55)	*p*-value
HGB (g/dL)	12.40 ± 2.27	12.39 ± 1.80	0.972
WBC (×10^3/mm^3)	8.99 (3.84)	9.15 (3.85)	0.922
PLT (×10^3/mm^3)	242.00 (99.00)	255.50 (87.25)	0.738
MPV (fl)	10.45 (1.40)	10.15 (1.08)	0.336
RDW (%)	14.35 (1.88)	14.15 (1.82)	0.475
PCT (%)	0.26 (0.11)	0.25 (0.11)	0.809
PDW (%)	12.10 (3.00)	11.95 (2.73)	0.747
NEU (×10^3/mm^3)	5.36 (3.34)	5.73 (3.30)	0.715
LYM (×10^3/mm^3)	1.93 (1.29)	1.72 (0.83)	0.585
NLR	2.63 (3.06)	3.35 (2.45)	0.230
LDL (mg/dL)	129.00 (22.50)	119.00 (28.50)	0.002
HDL (mg/dL)	42.00 (6.75)	36.50 (7.00)	<0.001
TRIG (mg/dL)	167.50 (140.75)	145.00 (81.00)	0.509
Urea (mg/dL)	48.55 (33.75)	44.60 (23.80)	0.089
Creatinine (mg/dL)	0.97 (0.50)	0.97 (0.38)	0.904
AST (IU/L)	29.00 (27.50)	22.50 (16.50)	0.077
ALT (IU/L)	17.50 (14.50)	16.00 (12.00)	0.135
Direct Bilirubin (mg/dL)	0.40 (0.24)	0.10 (0.11)	<0.001
Total Bilirubin (mg/dL)	0.82 (0.29)	0.44(0.14)	<0.001
FPG (mg/dL)	126.00 (56.00)	148.00 (101.00)	0.026
Uric acid (mg/dL)	5.65 (2.85)	5.70 (2.10)	0.544

HGB, haemoglobin; WBC, white blood cell; PLT, platelet; MPV, mean platelet volume; RDW, red cell distribution width; PCT, platecrit; PDW, platelet distribution width; NEU, neutrophil; LYM, lymphocyte; NLR, neutrophil/lymphocyte ratio; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; TRIG, triglyceride; AST, aspartate aminotransferase; ALT, alanine aminotransferase FPG, Fasting plasma glucose.

The backward elimination stepwise logistic regression analysis showed that the variables relating to bilirubin and FPG strongly predicted good and bad collateral development, respectively. The analysis revealed a 2.072-fold statistically significant increase ($p < 0.001$) in the odds of developing poor coronary collateral circulation (95% CI 1.512–2.840) for every 0.1 unit decrease in bilirubin levels. In addition for every 50-unit increase in FPG was associated with a

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TABLE 3. Distribution of medications used by patients in the good collateral (GC) and bad collateral (BC) groups.

	GC group (n = 73)	BC group (n = 55)	*p*-value
ASA	46 (63.0%)	35 (63.6%)	>0.999
Clopidogrel	24 (32.9%)	18 (32.7%)	>0.999
ACE-I	21 (28.8%)	18 (32.7%)	0.773
ARB	24 (32.9%)	20 (36.4%)	0.823
Statins	40 (54.8%)	26 (47.3%)	0.399
Beta-blocker	50 (68.5%)	34 (61.8%)	0.549
Calcium channel blockers	26 (35.6%)	22 (40.0%)	0.747
Diuretic	13 (17.8%)	11 (20.0%)	0.932
OAD	26 (35.6%)	18 (32.7%)	0.879
Insulin	10 (13.7%)	8 (14.5%)	>0.999

ASA, acetylsalicylic acid; ACE-I, angiotensin-converting enzyme inhibitor, ARB, angiotensin receptor blocker; OAD, oral antidiabetic drug.

statistically significant (p = 0.044) 1.438-fold increase in the odds of developing poor coronary collateral circulation (95% CI 1.010–2.048; Table 4).

TABLE 4. Distinguishing factors in the good collateral (GC) and bad collateral (BC) groups.

	Odds ratio	95% confidence interval		Wald	*p*-value
		Lower limit	Upper limit		
Bilirubin	2.072	1.512	2.840	20.534	<0.001
FPG	1.438	1.010	2.048	4.053	0.044

FPG, fasting plasma glucose.

4. Discussion

Our study provides additional novel data on the factors that affect coronary collateral development in elderly patients. We found that higher serum bilirubin levels were associated with good coronary collateral development, whereas higher FPG levels were associated with bad coronary development in this special patient population.

Due to their advanced age, older patients are less likely to receive invasive therapeutic interventions than younger patients. Another complicating factor is that comorbidity incidence increases with age. The coronary collateral circulation may play important coronary perfusion restorative roles in this patient group. Coronary collateral circulation development is an active process driven mainly by tissue ischemia and shear forces along the pressure gradient that develops in response to coronary stenosis, leading to the formation of new functional vessels. The process involves not only proliferation of endothelial and smooth muscle cells, and the release of several cytokines from the endothelium, it also requires growth factor expression and monocyte and macrophage recruitment [13]. Thus, it seems likely that factors affecting this whole cascade of events (e.g., oxidative stress, diabetes mellitus, hypertension, hyperlipidaemia and smoking) may impact coronary collateral growth.

Bilirubin is an end-product of heme catabolism, and is known to have antioxidant and anti-inflammatory properties, it has been associated with a number of disorders [14–17]. Elevated serum bilirubin levels are inversely associated with coronary atherosclerosis [18, 19]. Increased coronary artery calcium scores have also been associated with lower bilirubin levels in patients with coronary artery calcification [20, 21]. Lower levels of bilirubin have also been inversely related to increased carotid intima-media thickness and impaired flow-mediated dilation, an indicator of endothelial dysfunction [22]. One mechanisms of interest is that bilirubin prevents LDL oxidation, thereby inhibiting a major step in atherosclerosis [23]. In previous research, low bilirubin levels were identified as an independent risk factor for coronary artery disease and have also been associated with risk factors for CAD. Serum bilirubin levels are inversely associated with the prevalence of metabolic syndrome [24]. Furthermore, individuals with increased serum bilirubin levels had a reduced likelihood of developing type 2 diabetes over the next 4 years, irrespective of other factors [25]. Higher HbA1c values were reported in diabetic patients with low bilirubin levels [26]. It has also been suggested that hyperbilirubinaemia may play a role in the prevention of thrombotic events by inhibiting platelet activation [27]. In a study exploring the association of bilirubin levels with the development of the coronary collateral circulation, increased bilirubin levels correlated with well-developed coronary collaterals [28]. This finding is in line with our results. Previous studies have suggested that bilirubin prevents endothelial dysfunction through its antioxidant properties, resulting in improved coronary collateral development. Contradictory findings were obtained in studies relating to the association of oxidative stress and coronary collateral development. Demirbag e*t al.* [29] reported a positive correlation with the oxidative stress index in patients with a well-developed coronary collateral circulation. As oxidative stress occurs secondary to initial tissue hypoxia, it may act as a positive stimulus for the development of the coronary collateral circulation. However, it seems more plausible that at a certain level, oxidative stress inhibits coronary collateral formation by disrupting the actions of molecules involved in endothelial dysfunction and collateral development. Age is also of interest when investigating the role of bilirubin. Previously published work showed that coronary collateral development was impaired in elderly individuals when compared to younger individuals [30]. Animal studies have also demonstrated age-related impairment of collateral remodeling after peripheral vascular occlusion [31]. The mechanisms implicated in poor development of coronary collaterals, as seen in the elderly, include reduced expression of vascular endothelial growth factor, reduced nitric oxide release, increased formation of apoptotic proteins and increased oxidative stress. Reduced telomerase activity may also play a key role in the aforementioned mechanisms [32]. Asymmetric dimethylarginine, which inhibits the synthesis of nitric oxide, has been associated with poor coronary collateral development [33]. Oxidative stress restrains nitric oxide production by nitric oxide synthase and directly affects its function [34]. In light of these data, bilirubin likely improves coronary collateral development in the elderly population via

its antioxidant and anti-inflammatory properties.

In the present study, different bilirubin values were observed between the two Cohen–Rentrop grade based groups, however it must be highlighted that laboratory values for bilirubin can vary even within the same laboratory [35, 36]. We calculated the measurement uncertainty of serum Total Bilirubin and direct bilirubin levels based on the "top-bottom" approach, as defined in the Nordtest guidelines. Levels were estimated using internal and external quality control data, these values are compared with the total permissible error (TEa%) as directed within the 2019 The Clinical laboratory Improvement Amendments (CLIA). Serum total bilirubin and direct bilirubin analysis measurement uncertainty (U) were 12.18%, and 15.58% respectively in the 95% confidence interval, and these values did not exceed the 2019 CLIA% total permissible error value which is determined as 20%. Therefore, our results were within a safe range.

The risk factors for coronary artery disease have differing effects on coronary collateral development. Metabolic syndrome and diabetes mellitus have been reported to affect coronary collateral development adversely [37, 38]. In the present study, although the rate of diabetes mellitus was similar in both the BC and GC study groups, every 50 g/dL increase in FPG was associated with a 1.438-fold increase in the odds of developing poor coronary collateral circulation. In addition, although there was no between-group differences in the presence of hyperlipidaemia, levels of both LDL and HDL were significantly higher in the GC group. Elevated HDL and hypercholesterolemia were previously shown to be associated with good collateral development [39, 40]. Our findings are consistent with these data. In the current study, medications used by the patients were not associated with coronary collateral development. Whereas some published studies indicate the impacts of statin use on coronary collateral formation, the present study showed no statin effects in the cohorts [41]. This lack of effects from statin use on coronary collateral development in our sample may be explained by the age of the population and/or by differences in the duration of statin administration.

The retrospective design of the study, and the small sample size, are limitations of the present study. An additional limitation was the absence of data relating to the duration of chronic total occlusion of coronary vessels. Larger prospective studies are needed to establish a cause-effect relationship between bilirubin levels and coronary collateral development.

5. Conclusions

In conclusion, serum bilirubin and FPG seem to affect coronary collateral development in an elderly population with stable CAD.

Author contributions

Conception and design: EV, FYC; Writing the article: EV, IVD; Critical revision of the article: MS, IVD; Data collection: EA, GA; Statistical analysis: EV, MK; Overall responsibility: EV, IVD.

Ethics approval and consent to participate

All subjects gave their informed consent for inclusion before they participated in the study. The study was conducted in accordance with the Declaration of Helsinki, and the protocol was approved by the Ethics Committee of Gaziantep University with the approval number 237 in 2019.

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Conflict of interest

The authors declare no conflict of interest.

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