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Original Article

Relationship between serum bilirubin concentration and nondipper hypertension

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Abstract: Background: Although nondipper hypertension has been associated with increased cardiovascular morbidity and mortality, the relationship between bilirubin levels and nondipper hypertension remains unclear. Several studies have demonstrated that higher serum bilirubin levels inhibit inflammation and the proliferation of vascular smooth muscle cells, which may suggest a relationship between serum bilirubin levels and cardiovascular disease. The aim of this study was to compare serum bilirubin levels between dipper and nondipper hypertensive patients. Methods: The present study included 80 hypertensive patients who were stratified into two groups: 50 dipper patients (mean [± SD] age 51.5 ± 8 years; 29 male) and 30 nondipper patients (mean age 50.6 ± 5.4 years; 17 male). All patients underwent 24 h ambulatory blood pressure monitoring. Results: No statistically significant differences were found between the two groups in terms of basic characteristics. Total, direct and indirect serum bilirubin levels were significantly lower among individuals with nondipper hypertension compared with patients with dipper hypertension (0.78 ± 0.6 mg/dL versus 0.42 ± 0.32 mg/dL; and 0.29 ± 0.1 mg/dL versus 0.18 ± 0.05 mg/dL; and 0.48 ± 0.52 mg/dL versus 0.25 ± 0.22 mg/dL, respectively; all $P<0.001$). Additionally, leukocyte counts were higher in patients with nondipper hypertension. Conclusion: The present study revealed a potential relationship between lower serum bilirubin levels and a nondipping pattern in hypertensive patients.

Keywords: Nondipper hypertension, serum bilirubin

Introduction

Elevated levels of systemic inflammatory markers may be associated with cardiovascular diseases such as coronary artery disease, hypertension (HT) and atrial fibrillation [1-3]. Several previously published studies have demonstrated the relationship between serum bilirubin levels and cardiovascular diseases such as coronary artery disease. Bilirubin is an important and potent endogen antioxidant and anti-inflammatory agent [4-6].

Systolic and diastolic blood pressure decreases of >10% during sleep (compared with awake hours) is considered to be a normal diurnal pattern. The term 'nondipper' refers to patients whose blood pressure does not demonstrate this diurnal pattern. Nondipper patients are at higher risk for cardiovascular events and target organ damage than dippers [7, 8].

Dipper and nondipper blood pressure patterns have been studied extensively among hypertensive patients. Nondipper HT is associated with increased cardiovascular morbidity, such as left ventricular hypertrophia, and mortality [9, 10]. To our knowledge, there have been no studies investigating the relationship between bilirubin levels, and dipper and nondipper HT. Accordingly, we compared total serum bilirubin levels between dipper and nondipper HT patients.

Methods

Patient selection

A total of 118 consecutive patients were screened: 38 were excluded from the study for various reasons (12 for diabetes mellitus, 10 for chronic renal failure, eight for moderate or severe valvular disease, five for left ventricular systolic dysfunction and three for hyperthyroid-

Table 1. Comparison of basal demographic and ambulatory blood pressure monitoring features of dipper and nondipper hypertensive patients

	Dipper (n = 50)	Non-dipper (n = 30)	*P* value
Age (years)	51.5 ± 8	50.6 ± 5.4	0.24
Sex (n, %) males	29 (58%)	17 (56.6%)	0.62
BMI (kg/m^2)	29.7 ± 3.70	28.4 ± 3.09	0.24
Smoking	15 (30%)	6 (20%)	0.20
Beta blockers	15.9%	18.7%	0.40
CCB	25.6%	28.7%	0.25
ARB	18.6%	21.4%	0.14
ACE inhibitors	35.5%	33.2 %	0.12
Diuretics (%)	70.2 %	71.5 %	0.62
Combined regimen	61.8%	63.2%	0.46
Mean SBP (mmHg) (awake)	139.1 ± 12	133.3 ± 8.7	0.07
Mean DBP (mmHg) (awake)	89.3 ± 11	85.2 ± 8	0.08
Mean SBP (mmHg) (sleep)	118.5 ± 5.8	130.2 ± 9.6	<0.001
Mean DBP (mmHg) (sleep)	69.3 ± 4.8	78.1 ± 7.2	<0.001

BMI: Body mass index, CCB: Calcium channel blockers, ARB: Angiotensin reseptor blockers, SBP: systolic blood pressure, DBP: diastolic blood pressure.

ism). The remaining 80 patients with a history of chronic HT and receiving appropriate antihypertensive medications for at least three months before study commencement were prospectively enrolled. After a 5 min rest in a seated position, blood pressure was measured in the nondominant upper limb of each patient. Patients with a systolic blood pressure ≥140 mmHg and/or a diastolic blood pressure ≥90 mmHg, and those taking antihypertensive drugs were defined as hypertensive. Patients taking oral anti-diabetic drugs or insulin, or those with two fasting blood glucose measurements ≥126 mg/dL, were considered to be diabetic and excluded from the study.

Exclusion criteria included the presence of one of the following factors: known coronary artery disease; chronic renal failure; chronic liver disorders; moderate or severe valvular disease; diabetes mellitus; congenital heart disease; left ventricular systolic dysfunction on echocardiography (ejection fraction <50%); recent acute coronary syndrome; anemia; hyperthyroidism; pregnancy; obstructive sleep apnea; secondary HT; hematological disorders; known malignancy; drug history including anti-gout medications, anti-inflammatory agents such as corticosteroids; hormone treatment such as estrogens; a white blood cell count >12,000 cells/μL or <4000 cells/μL; and body temperature >38°C. Patients with a recent history of acute infection or an inflammatory disease were also excluded. All patients provided written informed consent, and the local ethics committee approved the study.

24 h ambulatory blood pressure monitoring

24 h ambulatory blood pressure monitoring (Suntech Medical Inc, Morrisville, NC, USA) was performed. Automatic blood pressure recordings were obtained regularly every 30 min during the 24 h period. The cuff was placed around the patients' nondominant arm. Sleep and awake periods were assessed based on information provided by the patients. Nocturnal blood pressure dipping was calculated using the following formula: (%) 100 x [1-(sleep systolic blood pressure/awake systolic blood pressure)]. Nocturnal blood pressure dipping was defined as >10% decrease in both nocturnal systolic and diastolic blood pressures compared with the average daytime blood pressures. A decrease of <10% in either systolic or diastolic blood pressure was defined as nondipper HT [11].

Laboratory analyses

Blood samples were drawn by venipuncture to perform routine blood chemistry in the post-ambulatory blood pressure monitoring period. Fasting blood glucose, serum bilirubin, creatinine, total cholesterol, electrolyte and thyroid-stimulating hormone levels were measured and recorded.

Statistical analysis

SPSS version 16.0 (IBM Corporation, USA) was used for statistical analysis. All values are presented as mean ± standard deviation. Mean values of continuous variables were compared between groups using the Student's t test or Mann-Whitney U test, according to whether the data were normally distributed, as tested by the Kolmogorov-Smirnov test. $P<0.05$ was considered to be statistically significant.

Table 2. Comparison of biochemical features of dipper and nondipper hypertensive patients

	Dipper (n = 50)	Nondipper (n = 30)	*P* value
Fasting glucose (mg/dl)	95.3 ± 12.1	93.2 ± 10	0.62
Creatinin (mg/dl)	0.92 ± 0.12	1.1 ± 0.18	0.24
AST (U/L)	23.12 ± 4.79	23.80 ± 6.68	0.52
ALT (U/L)	19.39 ± 6.17	21,03 ± 8.64	0.23
ALP (U/L)	67.7 ± 11	66.9 ± 9.5	0.35
Total cholesterol (mg/dl)	198.4 ± 31.1	203.6 ± 22.1	0.23
Na (mmol/L)	141.1 ± 12.6	138 ± 10.3	0.27
K (mmol/L)	4.41 ± 0.37	4.33 ± 0.38	0.24
Ca (mg/dl)	9.7 ± 1.41	9.3 ± 1.22	0.21
Total bilirubin	0.78 ± 0.6	0.42 ± 0.32	<0.001
Direct bilirubin	0.29 ± 0.1	0.18 ± 0.05	<0.001
İndirect bilirubin	0.48 ± 0.52	0.25 ± 0.22	<0.001
TSH (μIU/mL)	1.45 ± 0.2	1.56 ± 0.4	0.68
Hemoglobin (g/dL)	13.2 ± 1.27	14.13 ± 1.4	0.51
Platelets, $10^3/mm^{-3}$	243 ± 72	247 ± 77	0.62
Leukocyte, $10^3/mm^{-3}$	6917 ± 1936	8829 ± 2263	<0.001

Results

According to 24 h ambulatory blood pressure monitoring, dipper and nondipper HT was found in 50 (62.5%) patients and 30 (37.5%) patients, respectively. Evaluation of basic clinical and demographic characteristics showed no statistically significant difference between the two groups in terms of antihypertensive medications, age, sex distribution, body mass index or smoking status. In the dipper group, awake mean (± SD) systolic and diastolic blood pressures were higher than in the nondipper group but statistically insignificant, whereas mean sleep systolic and diastolic blood pressures were lower than in nondipper patients (118.5 ± 5.8 mmHg versus 130.2 ± 9.6 mmHg, and 69.3 ± 4.8 mmHg versus 78.1 ± 7.2 mmHg, respectively; *P*<0.001) (**Table 1**).

Serum total, direct and indirect bilirubin levels were lower in patients with nondipper HT than in those with dipper HT (0.78 ± 0.6 mg/dL versus 0.42 ± 0.32 mg/dL; 0.29 ± 0.1 mg/dL versus 0.18 ± 0.05 mg/dL; and 0.48 ± 0.52 mg/dL versus 0.25 ± 0.22 mg/dL, respectively; all *P*<0.001). Leukocyte counts were higher in the nondipper group. Other hematological and biochemical parameters were not statistically significant between the two groups (**Table 2**).

Discussion

The relationship between low bilirubin levels and increased cardiovascular risk is well known. In the present study, we found that total serum bilirubin levels were significantly lower in patients in the nondipper HT group compared with those in the dipper HT group.

Recently, Tian et al [12] investigated the association between specific circulating leukocyte types and blood pressure, and found that blood pressure was specifically related to neutrophil and lymphocyte levels. Kawada et al [13] showed an independent relationship between neutrophil levels and HT. However, there are no studies that have investigated specific relationships between inflammatory cell levels and nondipper HT.

Recently, low serum bilirubin levels have been proposed as a useful biomarker to predict cardiovascular risk. Recent evidence suggests that bilirubin acts as a potent physiological antioxidant and anti-inflammatory agent. Studies have shown that elevated serum bilirubin concentrations provide important protection against atherosclerotic diseases [14-16], with several authors suggesting that bilirubin plays a role in the inhibition of lipid oxidation [17, 18]. Previous studies have shown that plasma bilirubin concentrations are inversely correlated with several risk factors for coronary artery disease, such as smoking, diabetes and obesity, and directly correlated with high-density lipoprotein cholesterol levels [17, 19]. The inverse correlation between the presence of coronary artery disease, peripheral arterial disease, carotid intima-media thickness and bilirubin levels has been reported in several studies. Subnormal levels of plasma bilirubin are associated with premature coronary artery disease and cardiovascular morbidity [20, 21]. In a previous study, the three-year incidence of coronary artery disease was significantly lower in patients with Gilbert syndrome [22].

Elevated circulating concentrations of plasma bilirubin have been suggested to be able to prevent atherogenesis. A strong ability to scavenge peroxyl radicals and the antioxidant capacity of

bilirubin, even in slightly increased concentrations, have led to the concept that bilirubin may have a physiological function in protecting against disease processes involving oxygen and peroxyl radicals [23, 24]. In a previous study, Gullu et al [25] showed that elevated concentrations of bilirubin may serve as a protective factor against the development of coronary flow reserve impairment, coronary microvascular dysfunction and, possibly, the development of coronary atherosclerosis. They concluded that bilirubin demonstrated these beneficial effects independent of known coronary risk factors.

Induced hyperbilirubinemia was associated with a significant improvement in endothelial function in patients with type 2 diabetes mellitus [26]. Furthermore, bilirubin inhibited vascular cell adhesion molecule 1 and blocked vascular smooth muscle cell proliferation [27].

To our knowledge, there is no study available in the literature that has investigated the association between nondipper HT and serum bilirubin levels. Our study is unique in this respect, and we ascertained whether there is, in fact, an association between bilirubin levels and nondipper HT.

When the two groups in our study were compared, serum levels of bilirubin in patients with nondipper HT were significantly lower than in patients in the dipper HT group, and leukocyte levels in nondipper HT patients were significantly higher than those of dipper HT patients. Additional studies are required to determine the relationship between bilirubin and nondipper HT.

In conclusion, for the first time, we have shown that patients with nondipper HT have lower bilirubin levels compared with hypertensive dipper patients. Our study found a possible association between nondipper HT and serum bilirubin levels. The measurement of bilirubin may also be used to predict an increased risk for adverse, HT-related cardiovascular events. The most important limitation of our study was the small number of patients and the absence of a control group; in addition, we studied an incompletely characterized population. There is a need for large-scale investigation of this phenomenon.

Disclosure of conflict of interest

None.

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