Original Research article

Correlation of Homocysteine with C- reactive protein in Hypertensive patients

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Abstract

Background: Hypertension is regarded as a major health problem throughout the world because of its high prevalence and it is a risk factor of coronary artery diseases (CAD). Hyper homocysteinemia (HHcy) is known to increase the risk of many diseases and it has been regarded as a risk factor in CAD.

Aim: The present study was conducted to assay Homocysteine levels in newly diagnosed primary and secondary hypertensive patients and its association with C- reactive protein (CRP).

Materials and methods: A total 80 hypertensive patients with the age group of 30 to 50 years were selected for this study and 40 healthy age matched subjects were selected as a control group. Serum Homocysteine was assessed by ELISA, C-reactive protein was assessed by turbilatex method and routine investigations were done by ERBA EM-360 fully automated analyzer.

Results: The serum Homocysteine was significantly elevated in hypertensive patients compared with controls, and there was also significant difference observed between primary hypertension and secondary hypertension patients. Homocysteine levels were positively correlated with C- reactive protein, Cholesterol, TGL, LDL and negatively correlated with and HDL.

Conclusion: Elevated levels of homocysteine and CRP promote atherogenesis and monitoring of these levels might be useful to reduce cardiovascular mortality in hypertensive patients.

Keywords: Hypertension **,** Homocysteine (HCY), C-reactive Protein (CRP)

I. INTRODUCTION

Hypertension is an important risk factor contributing to an increased risk of coronary artery diseases (CAD) worldwide [1]. Despite the longstanding recognition of an association between hypertension and myocardial infarction (MI), the precise mechanisms that account for this relationship remain unclear. Early diagnosis and treatment strategies of hypertension, played a major role in recent dramatic declines in CAD and stroke mortality in developing countries .The prevalence of hypertension in the last decade has increased from 2% to 25% among urban residents and from 2% to 15% among the rural residents of India [2].

Homocysteine is a thiol-containing non-protein amino acid that is formed during the metabolism of the essential amino acid methionine and is recognized as an independent cardiovascular risk factor, such as arterial vascular diseases [3,4]. Increase in homocysteine level can lead to damage of endothelial cells, decreased flexibility of blood vessels leading to aortic stiffness and to reduction of the speed of blood flow, reduced production of the vasodilator nitric oxide (NO) [5-8]. C-reactive proteins (CRP) is a classical acute-phase protein present in trace amounts (1mg/L) in healthy subjects whose concentration increases in response to injury, infection or

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inflammation. CRP is named so for its ability to precipitate the somatic C-polysaccharides of Streptococcus pneumoniae and is the first acute phase protein to be described [9,10]. CRP is produced mostly by liver hepatocytes in response to cytokines such as interleukin-1 (IL-1), interleukin-6 (IL-6), and tumor necrosis factor [11,12]. So the objective of this study was to evaluate serum homocysteine levels in primary and secondary hypertension, its association with CRP a generalized inflammatory marker.

II. MATERIALS AND METHODS

A total 80 hypertensive patients of both sexes aged between 30-50 years attending Department of Medicine, Nimra Institute of Medical Sciences, Jupudi, Andhra Pradesh, India were selected for our study after approval of Institutional Human ethics committee. The included hypertensive patients were categorized into two groups according to Eighth Joint National Committee guidelines (JNC-8) .Group I- 40 Primary hypertension patients with systolic blood pressure (140-159) or diastolic blood pressure (90-99). Group-II 40 Secondary hypertension patients with systolic blood pressure (≥160) or diastolic blood pressure (≥100). The clinical characteristics of patients (age, gender, height, body weight, duration of disease, waist and hip circumferences) were collected. We excluded the patients based on the following criteria: subjects with diabetes mellitus, other cardiovascular diseases, renal dysfunction, chronic alcoholics, smokers, pregnant women and patients on medication such as antioxidant supplements and lipid lowering drugs. Forty healthy sex and age matched subjects were selected as controls.

Biochemical analysis:

Fasting blood samples were obtained from the subjects. Blood samples were centrifuged at 2000×g for 10 min. Samples were analyzed for glucose, lipid profile (Total Cholesterol, HDL, LDL, triglycerides), creatinine using by ERBA EM-360 fully automated analyzer. Serum Homocysteine was assessed by Enzyme Linked Immuno Sorbent Assay (ELISA) and C-reactive protein was assessed by turbilatex method

Statistical analysis: Statistical analysis were carried out with SPSS 20.0. Values were expressed as mean ± standard deviation, p value < 0.05 was considered statistically significant. Normally distributed data were analyzed by using one-way ANOVA. The Pearson correlation test was used for correlation analysis.

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RESULTS

Table 1: Baseline parameters in control and Hypertensive subjects.

Parameters	Controls (n=40)	Group I Primary hypertension (n=40)	Group-II Secondary hypertension (n=40)
Age	40.3±5.2	40.9±4.6	41.8±5.8
Body mass index	24.3±1.3	25.9±4.1 [a*]	26.06±2.7 [b**]
Waist/Hip ratio	0.90±0.05	0.92±0.06	0.93±0.04 [b*]
Males (%)	82.8	85.7	88.5
Females (%)	17.2	14.3	11.5
Systolic BP (mm Hg)	115.4±5.6	150.4±5.8 [a**]	187.0±11.9 [b**, c**]
Diastolic (mm Hg)	74.4±3.1	95.0±4.1 [a**]	107.8±5.6 [b**, c**]

Data are expressed as mean ±SD, **p<0.001,*p<0.05 was considered statistically significant.

a= comparison between Control and Primary hypertensive subjects

b=comparison between Control and Secondary hypertensive subjects

c=comparison between Primary and Secondary hypertensive subjects

Table 2: FPG, Lipid profile , Urea, Creatinine , Homocysteine and CRP levels in control and Hypertensive subjects

Parameters	Controls (n=40)	Group I Primary hypertension (n=40)	Group-II Secondary hypertension (n=40)
FPG(mg/dl)	85.6±7.0	88.3±9.4[a*]	91.6±8.0 [b**]
Serum cholesterol (mg/dl)	170.3±8.6	204.1±16.4 [a**]	210.7±15.7 [b**,c*]
Serum Triglycerides (mg/dl)	100.6±14.5	145.8±46.4 [a**]	147.9±44.6 [b**]
HDL cholesterol (mg/dl)	43.09±3.1	39.5±2.6 [a**]	39.3±2.4 [b**]
LDL cholesterol (mg/dl)	118.6±9.6	144.8±24.2 [a**]	156.1±22.4 [b**,c*]
Serum Urea(mg/dl)	24.7±4.3	29.2±3.2 [a**]	29.6±2.7 [b**]
Serum Creatinine(mg/dl)	0.6±0.19	0.8±0.25 [a**]	0.9±0.21 [b**]

Homocysteine (μ mol/L)	6.6±1.2	8.9±2.8 [a**]	12.2±3.5 [b**,c*]
CRP (mg/L)	1.13±0.26	3.16±0.69 [a**]	4.54±0.8 [b**,c**]

Data are expressed as mean ±SD, **p<0.001,*p<0.05 was considered statistically significant.

a= comparison between Control and Primary hypertensive subjects

b=comparison between Control and Secondary hypertensive subjects

c=comparison between Primary and Secondary hypertensive subjects

Table 3: **Correlation between Serum Homocysteine &measured parameters**

Parameters	Correlation Coefficient(r)
C-reactive protein	0.683**
Cholesterol	0.487**
TGL	0.475**
HDL	-0.278*
LDL	0.545**
Waist /Hip ratio	0.185
Systolic BP	0.636**
Diastolic BP	0.741**

**Correlation is significant at the 0.01 level (2-tailed).

Discussion

Hypertension is a major health problem and contributing factor of cardiovascular morbidity and mortality in India. The purpose of this study was to explore homocysteine levels in primary and secondary hypertensive patients compared with healthy individuals and to find out its association with inflammatory marker CRP. In the present study, we observed that serum homocysteine levels was significantly increased in both primary and secondary hypertensive patients, and it also had significant positive correlation with CRP. Elevated levels of Hcy and CRP have been associated with endothelial dysfunction and cardiovascular events in hypertension [13-14]. Sen U and Tyagi SC [15] reported that homocysteine may play a role in the pathogenesis of essential hypertension is based on the fact that homocysteine induces arteriolar constriction, renal dysfunction and increased sodium reabsorption, increasing arterial stiffness [15]. Homocysteine increases oxidative stress, which causes oxidative injury to the vascular endothelium, diminishing vasodilation by nitric oxide, stimulating proliferation of vascular smooth muscle cells and altering the elastic properties of the vascular wall, leading to an increase in hypertension [16].

The mechanisms by which Hyperhomocysteinaemia (HHcy) induces endothelial dysfunction are likely to be many, and include oxidative inactivation of nitric oxide (NO) and reactive oxygen species (ROS) generation when it is auto-oxidized to homocystine in the plasma [17-20]. The present evidence also suggests that Hcy-stimulated change in methylation of DNA and proteins may mediate its effects on endothelial cell phenotype. Cellular methyltransferases catalyze the transfer of the methyl group from S-adenosylmethionine (SAM) to substrates, among which are dC bases in CpG dinucleotides, leading to the formation of S-

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adenosylhomocysteine (SAH), which inhibits the trans-methylation reaction [21]. HHcy increases the cellular pool of SAH, decreasing SAM/SAH ratio. This mechanism has been implicated in Hcy-induced p21ras-mediated inhibition of endothelial cell growth[22]. In addition, hypomethylation of the cyclin A gene locus, and consequent decrease in cyclin A expression, has been shown to play a part in Hcy-induced inhibition of endothelial cell growth [23] .Furthermore, elevated SAH levels are associated with DNA hypomethylation in endothelial cells [24].

In addition our study also exhibits disturbances in lipid profile as in the earlier studies. Homocysteine levels shows strong positive correlation between total cholesterol, LDL, TGL and negative correlation with HDL. Dyslipidemia, one of the strong predictors of cardiovascular disease, causes endothelial damage and loss of physiological vasomotor activity [25-28]. The decrease in HDL-C could stimulate compensatory changes, as synthesis and accumulation of phospholipid-rich VLDL which binds bacterial products and other toxic substances, resulting in hypertriglyceridemia. The final consequence is an increased accumulation of cholesterol in cells. When the compensatory response (inflammation) is not able to repair injury, it turns into a harmful reaction, and the lipid changes will become chronic, either by repeated or overwhelming stimulus, enhancing the formation of atherosclerotic lesions [29]. In this regard, the current data support for the hypothesis that, all these factors are contributing elevated CRP levels. This is substantiated by several studies which have shown inflammatory markers such as CRP as an independent determinant of endothelium dependent vascular function among patient with coronary heart disease (CHD) and this situation may also exist in patients with hypertension [30]. CRP inhibits formation of nitric oxide by endothelial cells which in turn promote vasoconstriction, leukocyte adhesion, platelet activation, oxidation and thrombosis. Moreover, high levels of CRP may upregulate angiotensin receptors and enhance expression of plasminogen activator inhibitor-1 by endothelial cells [31]. In conclusion elevated levels of homocysteine and CRP promote atherogenesis and monitoring of these levels might be useful to reduce cardiovascular mortality in hypertensive patients.

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