Evaluation of Amino acid Homocysteine in Hypertensive Patients

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ABSTRACT :

BACKGROUND :

Hypertension is a world wide disease and in 90% of cases the cause is unknown . Its early detection and treatment can prevent serious complications such as ischemic heart disease (IHD) . Furthermore the association of lipids with IHD is a well-known fact . However abnormally high levels of homocysteine were found to be strongly linked to an increase risk of coronary artery disease .

OBJECTIVES:

To evaluate the total plasma homocysteine concentration in hypertensive patients.

METHODS :

Total plasma homocystein concentrations were measured using High Performance Liquid Chromatography(HPLC) with Ultraviolet(UV) detector in 60 hypertensive patients (27 male and 33 female) aged 35 years and more . Cholesterol , triglyceride , HDL-cholesterol , LDL-cholesterol , VLDL-cholesterol were determined .The prevalence of high total homocysteine values were determined by comparison with normal reference population.

RESULTS :

Total plasma homocysteine levels were significantly higher in patients than in normal population. Total serum cholesterol and triglyceride concentrations were also significantly higher in patients than in normal population with no association to the level of homocysteine which is regarded as a special independent vascular risk factor.

CONCLUSION :

The study involves the evaluation of homocysteine in hypertensive patients plasma homocysteine levels were significantly higher in patients than in control groups. There were no significant differences between male and female patients.

KEY WORDS : Atherosclerosis , Homocysteine , Hypertension , Ischemic heart disease .

INTRODUCTION :

The diagnosis of hypertension in adult is made when the average of two or more diastolic and systolic blood pressure measurements on at least two subsequent visits are more than 90 and 140 mmHg respectively (1). Its early detection and treatment can prevent a lot of serious complications such as heart attack, stroke , kidney diseases and heart failure(2). Among the most important available risk factors for hypertention is serum lipids particularly serum cholesterol which is a solid alcohol of high molecular weight present in diet and is mainly synthesized in the liver and small intestine and excreted unchanged in bile or converted to bile acids to be excreted (3). It was also found that abnormally high blood level of homocysteine is strongly linked to an increased risk of coronary heart diseases as it may harm the lining of the arteries and contribute to blood clotting (4) by causing endothelial injury followed by platelet activation and thrombus formation (5).

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Homocysteine is a sulfur-containing aminoacid (M.Wt.268) formed during the metabolism of methionine (6)which can be found in meats and dairy products ,therefore high dietary consumption of such products can result in the overproduction of homocysteine. Elevation in plasma homocycteine is either caused by genetic defects in the enzyme involved in its metabolism or due to nutritional deficiencies of vit.B6 , B12 or folic acid because these vitamins are essential co-factors for enzymes involved in the metabolism of homocysteine like methionine synthase and 5,10 methylene tetrahydrofolic acid reductase(7).

Hyperhomocysteinemia may be associated with several disease states and medications. It may increase in chronic renal failure often approaching concentrations that are up to four times the normal value which may explain the observed acceleration of atherosclerosis in end stage renal disease(8).

There is also some association between hyperhomocysteinemia and patients with hypothyroidism and rheumatoid arthritis which suggest a potential mechanism for the high incidence of vascular disease observed in those

patients Several drugs like methotrexate and phenytoin interferes with folate metabolism and may cause mild hyperhomocysteinemia(9,10).

METHODS :

The study was conducted on 60 hypertensive patients (27 male , 33 female) attending Kadimya Teaching Hospital as an out patient over 6 months period with an average age of 35- 85 years. Another 20 healthy subjects were studied as a control group . A full personal and family history was taken from each patient. Seven ml of blood was drown into a sterile disposable syringe.

Five ml from the sample were allowed to clot after 30 minutes in a plain tube and serum was separated by centrifugation at 3000 rpm under room temperature for 5 minutes. The serum was immediately analyzed for lipid profile .

The remaining 2-ml were put in a K3 EDTA tube and immediately put in a crashed ice . The plasma was separated from RBC by cooling centrifugation at 3000 rpm under 4C for 5 minutes then stored under – 20C until analyzed. Total cholesterol was determined by Cholesterol Enzymatique PAP Kits (BIOMERIUX). Triglyceride was determined using enzymatic method supplied by BIOMERIUX Lab. HDL-Cholesterol serum level was determined by precipitation of VLDL-Cholesterol and LDL-Cholesterol by the addition of phosphotungastic acid in the presence of magnesium ions.

The supernatant obtained after centrifugation contains only HDL-Cholesterol on which the cholesterol fraction was done by the same enzymatic method mentioned in cholesterol. Serum level of LDL cholesterol was calculated by Friedwald formula. Determination of the biologically active homocysteine (THCY)was done by using HPLC with Schimodzu SPD-6AV UV-visible detector within a wavelength of 195 – 700nm(11).

RESULTS :

The study showed a significant higher total plasma homocysteine concentration in our hypertensive patients (56 ±40.15 µmol/l) than in control group (14±3.9 µmol/l) with P< 0.05but the study showed no significant differences in homocysteine levels between male and female as shown in table-1. serum triglyceride concentrations in patient group were also higher than in control group as shown in table– 4.

The table also shows that there were significant differences between patient The severity of homocysteinemia is shown in table –2 where intermediate hyperhomocysteinemia was representing 55% of all cases compared to moderate and sever degrees which were representing 30% and 15% of all cases respectively(11). Table –3 shows that controlled hypertensive patients representing 73.33% of all cases with a mean homocysteine level of 50.7±29.2 µmol/l , while 26.66% were uncontrolled hypertensive patients with a mean homocysteine level of 62.6 ±27.4 µmol/l (12).

The study also demonstrated that total serum cholesterol concentrations in patient group were significantly higher than the concentration in control group. Mean and control groups in serum concentrations of HDL-Cholesterol , LDL-Cholesterol and VLDL-Cholesterol.

It is also clear from the study that 32 patients who had hypercholesterolemia more than 220 mg/dl and 21 patients who had hypertriglyceridemia more than 160 mg/dl had no relation to the level of homocysteine which is regarded as a specific risk factor independent from other risk factors as it is demonstrated in table-5.

Table 1 :Total plasma homocysteine concentration in patients and control groups .P< 0.05 .

	[Homocysteine µmol/l		
	N	Mean	SD
Patients	Male 27	58.15	3.66
	Female 33	55.45	4.36
	Total 60	56.8	4.014
Control	20	14	3.9

Table 2 :Classification of plasma homocysteine according to concentrated *level in male and female patients.*

		Male			Female		
		N	Mean±SD	%	N	Mean±SD	%
Moderate	15-30 µmol/l	8	22.9±7.8	29.6	10	20.3±5.2	30.3
Intermediate	30-100 µmol/l	15	55±20.6	55.6	18	45.7±15.8	54.5
Sever	>100µmol/l	4	102.8±40.1	14.8	5	117.4±50.5	15.2

Table 3 : Plasma homocysteine in controlled and uncontrolled hypertension

	Homocysteine µmol/l			
	N	Mean	SD	%
Controlled hypertension	44	50.7	29.2	73.33
Uncontrolled hypertension	16	62.6	27.4	26.66

Table 4 :Lipid profile in patients and control groups

	Patient		Control		P<0.05
	Mean	SD	Mean	SD	
Cholesterol (mg\dl)	221.94	45.82	173.20	18.57	Significant
Triglyceride (mg\dl)	182.09	111.04	117.89	43.22	Significant
HDL-chol. (mg\dl)	41.95	14.31	52.72	12.41	Significant
VLDL-chol. (mg\dl)	36.13	22.41	19.11	8.81	Significant
-LDL	138.19	49.70	83.55	18.31	Significant

Table 5 :Plasma homocysteine with hypercholestrolemia and hypertriglyceridemia

	Homocysteine µmol/l			
	N	Mean	SD	%
Hypercholestrolemia >220 mg/dl	32	53.99	33.74	53.3
Hypertriglyceridemia >160 mg/dl	21	56.47	31.9	35

DISCUSSION :

The study had demonstrated a significant increase in plasma homocystein concentration in hypertensive patients and an essential linear relationship between homocystein level and vascular risk which was also found by Graeme et al.(13). There was no significant difference in plasma total homocystein concentration between male and female patients with moderate and intermediate hyperhomocystein level.

This difference increased in patients with sever hyperhomocystinemia which is similar to the finding of Stamler ,et al (14) . Hyperhomocystinemia was found in 73.33% of all patients which may be due to along interval (without control of hypertension)of disease which was more than 10 years . The high concentrations of homocystein in uncontrolled hypertensive patients might be due to sever hypertension as a result of neglection of treatment by antihypertensive drugs(15).

Total serum cholesterol and triglyceride concentrations was significantly higher than the concentration in control group and this result agree with Neaton and coworkers who have reported increases in cardiovascular outcome events with increasing cholesterol levels (16).This also agree with a previous finding in another study which found a significant triglyceride – coronary disease association (17) . HDL-Cholesterol in patients and control groups were significantly different and a more recent study showed a basic association of high HDL and low risk of coronary diseases .

A similar finding for both LDL and VLDL were obtained . The results showed that increase in cholesterol and triglyceride levels have no relation to the levels of homocystein which is regarded as a special independent risk factor(18) .

These findings agree with Konecky et al. who showed that hyperhomocysteinemia is an independent risk factor for aortic diseases (19).

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