Section: General Medicine

Original Research Article

CORRELATION BETWEEN ELEVATED SERUM HOMOCYSTEINE LEVELS AND CAROTID INTIMAL THICKNESS IN PATIENTS WITH THROMBOTIC STROKES

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Abstract

Background: Elevated plasma levels of homocysteine, a sulphur-containing amino acid produced during methionine metabolism, had been associated with an increased risk of atherosclerotic and ischemic events in a dose-dependent manner. Ischemic stroke has been a leading cause of mortality and morbidity across the globe, causing a significant burden to patients and families. There are several known factors elevating the risk of ischemic stroke, which include transient ischemic attack (TIA), arterial diseases, atrial fibrillation, improper diet and/or obesity and physical inactivity. Notably, less than half of the different types of strokes can be linked to recognized causal risk factors. **Materials and Methods:** This Prospective observational case control study was carried on 50 patients over a period of 1 year, with following criteria's. Clinical information including age, sex, history of current evidence of hypertension (HTN): systolic blood pressure (SBP) - 140mmHg and diastolic BP - 90mmHg, Diabetes Mellitus (DM): fasting blood glucose 7 mmol/L/126 mg, cardiac disease, life style, diet pattern, family history of vascular diseases was recorded for all subjects. Serum total cholesterol (CH), HDL cholesterol, LDL cholesterol VLDL cholesterol and triglycerides were measured by using standard enzymatic procedures. Serum homocysteine was estimated by Chemilunescent Immuno assay method, variant of standard ELISA. **Result:** In our study distribution of 50 patients according to serum homocysteine. Majority of the patients (72%) had high homocysteine. The majority of non-diabetic patients (64%) had high homocysteine. There is no significant correlation between hypertension and homocysteine. 48% of hypertension patients of study population, had high homocysteine. There is no significant correlation between them. Among patients with high homocysteine, 44% had carotid artery involvement and 28% had vertebral artery involvement. There was a statistically significant positive correlation between serum homocysteine with CIMT (p=0.001). Majority of patients with high homocysteine (68%) had high CIMT levels. There was a statistically significant positive correlation between serum homocysteine with total cholesterol (p=0.001), triglycerides (p=0.001) and LDL(p=0.001). Majority of patients with high total cholesterol (62%), high triglycerides (66%), low HDL (42%) and high LDL (66%). **Conclusion:** People at risk for cerebrovascular diseases having risk factors such as, smoking, dyslipidemia should be screened for hyperhomocysteinemia. Our study revealed that hyperhomocysteinemia appears to be an important risk factor for cerebrovascular accidents. It is therefore important to use serum homocysteine level as an important tool to investigate all cases of young cerebrovascular accidents and also in those who are at risk of developing it.

INTRODUCTION

Elevated plasma levels of homocysteine, a sulphur-containing amino acid produced during methionine metabolism, had been associated with an increased risk of atherosclerotic and ischemic events in a dose-dependent manner.[1]

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As a demethylated by-product of methionine, homocysteine (Hct) is considered a toxic amino acid. It is usually removed by methionine synthase in a remethylation process that uses methylcobalamin as a co-factor and 5-methyl tetrahydrofolate as a methyl donor.[2] In the brain, Hct catabolism heavily relies on its remethylation to methionine using methylcobalamin and folate.[3]

Ischemic stroke has been a leading cause of mortality and morbidity across the globe, causing a significant burden to patients and families.[4] There are several known factors elevating the risk of ischemic stroke, which include transient ischemic attack (TIA), arterial diseases, atrial fibrillation, improper diet and/or obesity and physical inactivity. Notably, less than half of the different types of strokes can be linked to recognized causal risk factors.[5]

Lately, attention has been given to Hct and its role in stroke incidences or events. Hct is a type of amino acid that contains sulfur that is considered a causal risk factor for vascular diseases such as stroke. Furthermore, available empirical evidence shows that there is a positive correlation between iHct and ischemic stroke, which has helped develop practical procedures for screening and treating iHct as a modifiable risk factor.[6] Nevertheless, there are still some controversies regarding the causal role of iHct in stroke, especially due to failure of vitamin B-supplementing trials in preventing strokes.[7]

There is growing evidence that high homocysteine levels contribute to the pathogenesis of ischemic stroke.[8] Various studies are currently assessing the role of homocysteine as a independent risk factor in stroke in ischemic stroke patients and its possible implication in prevention. This study will correlate the serum homocysteine level in patients with ischemic stroke.

MATERIALS AND METHODS

This Prospective observational case control study was carried on 50 patients over a period of 1 year, with following criteria's.

Inclusion Criteria

- First ever episodes of ischemic stroke
- Cases presenting within two weeks of the event
- Age group 15 years to 45 years
- Willing to give informed consent.

Exclusion Criteria

- Non-hemorrhagic stroke
- Renal, hepatic, thyroid dysfunction
- Collagen vascular diseases
- Chronic diseases like HIV, Syphilis, TB, RHD, and cancer
- Patient on steroids
- Pregnancy state and postpartum period.

Clinical information including age, sex, history of current evidence of hypertension (HTN): systolic blood pressure (SBP) - 140mmHg and diastolic BP - 90mmHg, Diabetes Mellitus (DM): fasting blood glucose 7 mmol/L/126 mg, cardiac disease, life style, diet pattern, family history of vascular diseases was recorded for all subjects. Serum total cholesterol (CH), HDL cholesterol, LDL cholesterol VLDL cholesterol and triglycerides were measured by using standard enzymatic procedures.[5]

Borderline for normal values were: total cholesterol 200- 239 mg, HDL-C < 60 mg, LDL-C <130-159 mg, VLDL< 1.1 mmol/L and triglyceride < 150 mg.[5]

Serum homocysteine was estimated by Chemilunescent Immuno assay method, variant of standard ELISA.

The upper limit of the manufacturer and the laboratory was 15 µmol/L. Values above 15 µmol/L were accepted as high.[6]

Statistical Analysis

In order to compare these parameters between patients and controls, student's t-test was applied and the results were presented in tabular form.

RESULTS

[Table 1] shows the distribution of 50 patients according to serum homocysteine. Majority of the patients (72%) had high homocysteine.

[Table 2] shows distribution of cases according to age with serum homocysteine. There was a statistically significant positive correlation between age and serum homocysteine(P=0.016). The majority of cases (24%) with high serum homocysteine were in the age group of 41-50.

[Table 3] shows the distribution of cases according to gender with serum homocysteine. There was no correlation between gender and homocysteine. According to the above table, majority of male patients (24%) and female patients (48%) had high homocysteine.

[Table 4] shows statistically significant positive correlation between diabetes and homocystiene (p=0.001). The majority of non-diabetic patients (64%) had high homocysteine.

[Table 5] shows no significant correlation between hypertension and homocysteine.48% of hypertension patients of study population, had high homocysteine.

[Table 6] shows distribution of MRA with serum homocysteine. There is no significant correlation between them. Among patients with high homocysteine, 44% had carotid artery involvement and 28% had vertebral artery involvement.

[Table 7] shows the distribution of serum homocysteine with CIMT levels. There was a statistically significant positive correlation between serum homocysteine with CIMT (p=0.001). Majority of patients with high homocysteine (68%) had high CIMT levels.

[Table 8] shows there was a statistically significant positive correlation between serum homocysteine with total cholesterol (p=0.001), triglycerides (p=0.001) and LDL(p=0.001). Majority of patients with high total cholesterol (62%), high triglycerides (66%), low HDL (42%) and high LDL (66%).

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Table 1: Distribution of Cases Based On Homocysteine Level

Homocysteine	No. of Patients	Percentage
<40 mg/dl	14	28
>40 mg/dl	36	72

Table 2: Age Distribution of Cases With Serum Homocysteine

Age	Low Homocysteine N(%)	High Homocysteine N(%)
31-40	05(10)	11(22)
41-50	05(10)	12(24)
51-60	0	11(22)
61-70	03(06)	0
>71	01(02)	02(04)

P=0.016*

Table 3: Gender Distribution of Cases According To Serum Homocysteine

Sex	Low Homocysteine N(%)	High Homocysteine N(%)
Male	05(10)	12(24)
Female	09(18)	24(48)

P=0.873

Table 4: Distribution of Diabetes and Serum Homocysteine

Homocysteine	Diabetes	
	Yes (N%)	No (N%)
<15	11(22)	03(06)
>15	04(08)	32(64)

P=0.001*

Table 5: Hypertension And Serum Homocysteine

Homocysteine	Hypertension	
	Yes n (%)	No n (%)
<15	10(20)	07(14)
>15	24(48)	09(18)

(P=0.501)

Table 6: MRA and Serum Homocysteine Distribution

MRA	Low Homocysteine	High Homocysteine
Carotid	11(22)	22(44)
Vertebral	03(06)	14(28)

Table 7: Correlation Between Homocysteine and Cimt

Homocysteine	CIMT (<0.75)	CIMT (>0.75)
<15	12(24.0)	02(4.0)
>15	02(4.0)	34(68.0)

P=0.001*

Table 8: Correlation between Serum Homocysteine and Lipid Profile

	Low/High	Homocysteine		
		Low N(%)	High N(%)	
TC	<200mg/dl	12(24.0)	05(10.0)	0.001*
	>200 mg/dl	02(4.0)	31(62.0)	
TG	<150 mg/dl	11(22.0)	03(6.0)	0.001*
	>150 mg/dl	03(6.0)	33(66.0)	
HDL	<40 mg/dl	12(24.0)	21(42.0)	0.099
	>40 mg/dl	02(4.0)	15(30.0)	
LDL	<130 mg/dl	11(22.0)	03(6.0)	0.001*
	>130 mg/dl	03(6.0)	33(66.0)	

DISCUSSION

Stroke is emerging as an increasing public health problem, especially due to long-term disability among patients. Early detection and intervention represent the best way to protect against stroke complications. Nevertheless, there is no biomarker thus far that may correlate with stroke. High level of Hct has been found in atherosclerosis patients. Previous in vivo and in vitro studies suggest that Hct changes the endothelium to a thrombogenic phenotype. Therefore, Hct thus has adverse effects on endothelial function, stimulates smooth muscle proliferation, and is a procoagulant, thus increasing the risk of major vascular events.[10]

Patients with ischemic stroke disease have higher Hct levels than controls. Three studies identified large differences but the vast majority of them

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demonstrated a tendency of higher levels of Hct in patients with stroke compared to controls, which was confirmed in the sensitivity analysis that excluded those three outliers.[11]

Dai D et al. establish that inverse association between Glasgow Coma Scale and Hct concentrations among patients with hemorrhagic stroke, especially in ever smokers, or in participants with higher systolic blood pressure or total cholesterol levels.[12] Hct level may be an aggravating factor in atherosclerosis, which is positively associated with a high risk of stroke.[12]

It is highly likely that iHct can be regarded as an aggravating factor that elevates the risk of ischemic stroke, since it triggers the necrosis of vessel walls and pathophysiology of endothelial degeneration. It can interfere with the stability of the blood-brain barrier through an excitotoxicity pathway related to NMDA channels, and it can decrease the activity of numerous essential cellular components such as Na/K ATPase, superoxide dismutase, and glutathione peroxidase.[13]

Hct activates the transcription of the factor in nervous tissue, which increases inflammation by increasing the levels of inflammatory cytokines. The role of this molecule in epilepsy, anxiety, cardiovascular diseases and stroke leaves no doubt that it could be a key aspect of ischemic stroke pathogenesis.[14]

Hyperhomocysteinemia is currently recognized as a risk factor for ischemic stroke and human vascular disorders. Nonetheless, it is very imprecise to determine if iHct triggers the pathogenesis of the diseases or it signifies a biomarker of metabolic aberrations. Hyperhomocysteinemia changes how vascular endothelial cells function as well as activating homocysteinylation and thiolation of enzymes and plasma proteins which negatively affect the brain parenchyma and cerebrovascular. Under iHct conditions, ischemic reperfusion injury usually triggers degeneration processes as well as engage in deregulation of intracellular signaling.[15]

Moreover, Acampa et al. showed that iHct can favor atrial cardiopathy and silent episodes of atrial fibrillation occurrence with multiple mechanisms: direct effects on atrial ionic channels (electrical remodeling); biochemical damage on atrial extracellular matrix (structural remodeling); prothrombotic state, favoring atrial thrombosis and possible subsequent ischemic embolic stroke.[16]

Another study by Acampa et al. also shows that iHct is associated with an alteration in electrical atrial conduction, possibly contributing, at least in part, to the increased risk of cardiac arrhythmias and, therefore, a risk factor for stroke.[17]

Shi et al. observed that high plasma Hct levels in the acute ischemic stroke are strong mortality predictors in patients with severe atherosclerosis.[18] There is a relevant, independent association with high Hct levels and the risk of mortality from patients affected with ischemic stroke. This effect may be mediated at least partly through elevations of homocysteine levels. Kwon et al. proposed that high Hct increase the risk of early neurological deterioration.[18]

It has been proven that the elevation of plasma Hct is often correlated with the development of atherosclerosis as well as the impairment of the vascular endothelium. Additionally, it triggers serine elastase synthesis in the vascular smooth muscle cells leading to elastolysis by degrading the extracellular matrix and releasing reactive oxygen species.

van Beynum et al. reported an association between occlusive vascular and stroke disease and hypehomocysteinemia.[19] In a prospective study was observed 20% higher risk of ischemic stroke in men associated with homocysteine values. High plasma Hct levels are associated with intracranial strong plaque and carotid plaque and intima-media thickness.[20]

CONCLUSION

People at risk for cerebrovascular diseases having risk factors such as, smoking, dyslipidemia should be screened for hyperhomocysteinemia. Our study revealed that hyperhomocysteinemia appears to be an important risk factor for cerebrovascular accidents. It is therefore important to use serum homocysteine level as an important tool to investigate all cases of young cerebrovascular accidents and also in those who are at risk of developing it. Significant correlation has been found between homocysteine concentration and ischemic stroke.

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