Association between Serum Homocysteine, Folate and B12 Concentration with Coronary Artery Disease in Thai Patients

Worachat Moleerergpoom MD*, Thunyachai Sura MD**, Piyamitr Sritara MD***

* Division of Cardiology, Police General Hospital  
** Division of medical genetics and molecular medicine, Faculty of Medicine Ramathibodi Hospital  
*** Division of Cardiology, Faculty of Medicine, Ramathibodi Hospital

Background: Hyperhomocysteinemia, associated with low folate and low B12 levels, is known to be an independent risk factor for atherosclerosis. Only a few available data has been demonstrated in Thai patients.

Objective: To evaluate serum fasting homocysteine, folate and B12 levels whether to see they are associated with coronary artery disease (CAD).

Method and Result: Three hundred and one consecutive suspected CAD patients who underwent coronary angiography at the Police General Hospital were studied. The mean age of the patients, 195 males and 106 females, was 63.0 ± 10.0 year (range 39-85). A total of 218 patients were angiographically demonstrated as having CAD. The mean serum homocysteine level of CAD patients had a non significant higher level than those of 83 non CAD patients: 11.4 ± 6.2 vs 10.2 ± 4.2 umol/L, p = 0.06. Means of folate and B12 level in the CAD patients and non CAD patients were 6.6 ± 4.6 vs 7.0 ± 4.3 nmol/L, p = 0.49 and 650.9 ± 415.4 vs 613.3 ± 443.2 pmol/L, p = 0.56 respectively. No significant correlations were found between homocysteine with folate and B12 levels. Logistic regression analysis showed a significant association between homocysteine and CAD with OR = 1.08 (95%CI, 1.01-1.16), p = 0.03 after being adjusted for age, sex, DM, HT, history of hyperlipidemia, smoking, BMI, folate and B12 levels. No significant association between homocysteine level with the number of coronary vessel stenosis, age, BMI, DM, HT, smoking and history of hyperlipidemia was observed in the present study.

Conclusion: Hyperhomocysteinemia, but not folate and B12 levels, may be an independent risk factor for coronary artery disease in Thai patients

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Material and Method

Study population

A total of 301 patients suspected of having CAD underwent coronary angiography in the Police General Hospital with the following indications: 55.5% with chronic stable angina, 20.7% with unstable angina, 12.5% with preoperative evaluation, 4.7% with cardiomyopathy and 6.6% of others. The study was performed from December 1999 to February 2001. Consent was obtained from all the patients at their first visit which included history taking and complete physical examination. Twelve hours fasting blood samples of all eligible patients were collected at the time the coronary angiography was performed. Patients taking either regular oral vitamins or serum creatinine > 2 mg/dl or malnutrition status were excluded from this study. Conditions defined in patients in the present study followed the standard criterias of WHO; hypertension those who had a history of taking antihypertensive drugs or blood pressure > 140 mmHg systolic and/or > 90 mmHg diastolic, hyperlipidemia those who had a total cholesterol > 200 mg/dl or had a history of taking any lipid lowering drugs, smoking those who were a current smokers or stop smoking for less than 2 years, and coronary heart disease defined by angiographic stenosis at least 50% of lumen diameter in any of the main vessels, left main stenosis without stenosis of the right coronary was classified as two-vessel disease. The control group were patients who had normal coronary angiography or lumen stenosis less than 50% in all vessels involved.

Biochemical Measurements

Fasting plasma of eligible patients was separated from blood cells by immediate centrifugation. Serum was obtained and kept at a temperature of -27 C for analysis serum homocysteine (Hcy), folate and B12 levels by high-performance liquid chromatography (HPLC) method (Ubbink 20, 21, 22)

Statistical Analysis

Student’s t-test and ANOVA were employed for comparing means between groups. Pearson Correlation was used for determining correlation between homocysteine, folate, B12, age and BMI. The association between groups categorized by variables was determined by Chi square and logistic regression analysis was used for determining association between homocysteine and CAD by adjusting confounding risk factors such as age, sex, BMI, smoking, HT, hyperlipidemia. All analysis were two tailed and p value less than 0.05 was considered significant.

Results

301 patients were eligible for determining serum concentration of homocysteine which had a mean 11.0 ± 5.7 umol/L and a median = 10.05 umol/L (range 1.5-48.2).

Fig. 1 shows normal distribution of CAD and control group with median = 10.1 umol/L (range 1.5-48.2) and 9.9 mol/L (range 2.75-22.5), respectively.

Table 1 shows the baseline characteristics of patients with CAD and the control group. Mean age of the patients was 63.0 ± 10.0 years, 195 males and 106 females. The control patients had a higher mean age than those with CAD. Most of CAD patients were male 67.9% vs 57.1% in the control group. There was significantly more diabetes and hyperlipidemia in the CAD group. No significant difference in BMI, HT and smoking was found between the two groups. Mean of homocysteine level in the CAD was slightly higher than that in the control but was not significant, 11.4 ± 6.2 vs 10.2 ± 1.2 umol/L respectively, p 0.06. There was also not significantly different means of folate and B12 level in the CAD and Control groups, 6.6 ± 4.6 vs 7.0 ± 4.3, p = 0.49 and 650.9 ± 415.4 vs 613.3 ± 443.2, p = 0.56.

Means of homocysteine in the CAD were not significantly higher than those in the control group in both the male and female group (Fig. 2). Means of homocysteine in males higher than those of females but also not significant 11.4 ± 5.9 vs 10.2 ± 5.1 umol/L,
Correlation between homocysteine vs folate and B12 levels (Fig. 3)

No significant correlations were found between homocysteine vs folate and vs B12 levels with r = -0.001, p = NS and r = 0.072, p = NS respectively.

Association between homocysteine with coronary risk factors and the numbers of vessels stenosis

There was no association between homocysteine and various coronary risk factors such as age, BMI, smoking, DM, hyperlipidemia and HT. Means of homocysteine were not different in the CAD patients according to the number of vessel stenosis; 10.8 ± 4.8, 11.9 ± 6.9 and 11.1 ± 6.1 umol/L in a single vessel, double vessel and triple vessel disease respectively.

Multivariate Analysis

Fig. 4 shows the logistic regression model for homocysteine, folate, B12 and various factors which could be confounders such as age, BMI, sex, DM, hyperlipidemia, HT and smoking were analyzed as to whether there was independent association with CAD. Homocysteine was weakly significantly associated with CAD with OR = 1.08 (1.01-1.16) p = 0.03. There was also significant association between CAD and age, DM and hyperlipidemia in the present study.

Discussion

The average of homocysteine level from population-based survey in various ethnic groups ranged from 7.0-9.6 umol/L. Age, sex and ethnicity were major factors of variation. A review of data from a cross-sectional and retrospective case control study published by Hankey GJ et al demonstrated that the homocysteine level in CAD ranged from 11.3-16.7 umol/L while the control group was slightly lower 8.9-14.7 umol/L. The present data is concordant...
with other investigators; however, the present study could not prove a significant difference in means of the homocysteine level between these two groups as in many previous reports. The explanation may be the advanced age of the control group. Following the multivariate analysis for adjusting many confounding factors, homocysteine was found to be an independent risk for CAD with OR 1.08 (1.01-1.16) \( p = 0.03 \). The interpretation was every 1 umol/L of homocysteine increment increased risk of CAD by 1.08 folds (or 8%). According to this weak association, can we accept homocysteine to be a new risk factor in Thai? Larger number of patients need to be explored later but we should concern this new risk factor when atherosclerosis develop in any young or in whom have no other risk factors. Regarding Vitamin B12 and folate, both study groups had comparable Vitamin B12 and folate levels. Use of fish sauce in Thai cooking may be the reason for these similarities as it contains high folate and fresh vegetables are a natural source of Vitamin B12. Due to a small size of the study population, the relationship of homocysteine with age, sex and other coronary risk factors could not be identified. Selhub J, et al proposed to use the level above 95th percentiles of population-based survey to classify hyperhomocysteinemia\(^{(17)}\). Homocysteine data in a Thai population is not available; therefore, the population in the present study could not be classified. Pathogenesis of homocysteine involved in atherosclerosis is still unclear. Numerous studies explored the sites of adverse effect of homocysteine induced vascular injury interaction with plasma lipoproteins clotting factors and platelets. Altered vascular endothelium function including proliferation of vascular smooth muscle cells and collagen deposit in plaque was also observed. Folic acid supplementation is the major therapeutic intervention for the treatment of elevated homocysteine levels\(^{(19)}\). Although numerous studies of folic acid supplementation in hyperhomocysteinemia have been performed, it remains to be confirmed that homocysteine-lowering therapy will prevent important atherosclerosis vascular events in patients with hyperhomocysteineemia. Several large randomized clinical trials are needs to address this issue.

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References

ความสัมพันธ์ระหว่าง โฮมิคิสทีน โฟเลต และ วิตามิน บี 12 กับโรคหลอดเลือดหัวใจตีบในคนไทย

วรชาติ โมฬีฤกษ์ภูมิ, ธันยชัย สุระ, ปิยะมิตร ศรีธารา

ภาวะ สารโฮมิคิสทีน สูงในเลือด เป็นปัจจัยเสี่ยง ต่อการเกิดโรคหลอดเลือดหัวใจตีบ ซึ่งมีความสัมพันธ์กับ ระดับสาร โฟเลต และ วิตามินบี 12 ในเลือดที่ดี ข้อมูลส่วนใหญ่เป็นข้อมูลที่ได้จากต่างประเทศ จึงทำการศึกษาภาวะนี้ในคนไทย โดยศึกษาผู้ป่วยที่มีอาการหลอดหัวใจ จำนวน 301 คนที่โรงพยาบาลตำรวจ เป็นผู้ป่วย 106 คน อายุเฉลี่ย 63±10 ปี (39-85 ปี) พบว่า มีโรคหลอดเลือดหัวใจตีบ 218 คน และ กลุ่มควบคุม คือ ไม่เป็นโรค มีหลอดเลือดหัวใจปกติ 83 คน ค่าเฉลี่ยของสารโฮมิคิสทีน สารโฟเลต และ วิตามินบี 12 ในกลุ่มที่เป็นโรคหลอดเลือดหัวใจเท่ากับ 11.4±6.2 vs. 10.2±4.2 เอนโดมิคิสทีน ไม่แตกต่างกัน แต่สารโฟเลต สารวิตามินบี 12 ในกลุ่มหลอดเลือดหัวใจตีบเท่ากับ 6.6±4.6 vs. 7.0±4.3 ไม่แตกต่างกัน แต่ค่า Odds ratio ของสารวิตามินบี 12 การวิเคราะห์ Logistic regression แสดง ความสัมพันธ์ ระหว่าง ระดับ โฮมิคิสทีน กับ โรคหลอดเลือดหัวใจตีบ ค่า Odds ratio 1.08 (95%CI, 1.01-1.16) ระดับ สารโฟเลต และ วิตามินบี 12 ไม่แตกต่างกัน แต่ค่า Odds ratio ได้เป็นปัจจัยเสี่ยงต่อการเกิดโรคหลอดเลือดหัวใจ ดีป่าในคนไทย