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Homocysteine and Cognitive Impairment in Thai Elderly

Duangkamol Viroonudomphol[1], Saowanee Kajanachumpol[2], Chaiwat Prawettongsopon[3]

[1]Faculty of Nursing, Siam University, Bangkok, Thailand
[2]Faculty of Medicine Ramathibodi Hospital, Mahidol University, Bangkok, Thailand
[3]Faculty of Science, Mahidol University, Bangkok, Thailand
Email: v_duangkamol@yahoo.com, saowanee.kan@mahidol.ac.th, yo_scbi@hotmail.com

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Abstract

Background: The prevalence and incidence of dementia increase dramatically with age. Cognitive impairment is one major symptom of dementia. Older persons increase in our society, which means a big number of people with decreased cognitive function. So it is important to find out risk factors. The amino acid homocysteine may be a risk factor. **Objective:** The aim was to determine the independent association of homocysteine and cognitive performance in Thai elderly. **Design:** Concentrations of homocysteine were measured in fasting blood samples of 100 Thais aged 60 - 80 years. Global cognitive function was assessed by using with mini-Mental State Examination score (MMSE), and cognitive functions were assessed by a neuropsychological test battery. The relationship between homocysteine levels and neuropsychological test scores was assessed by multiple linear regression. **Results:** In the crude model, homocysteine was inversely associated with scores for learning slope test (B = −0.048, $p = 0.042$) and verbal pair total test (B = −0.124, $p = 0.032$). After adjusting for confounders, no association was found between homocysteine and cognitive impairment. Age (B = −0.129, $p = 0.007$) was found to be a significant determinant of decreased learning slope score. Similarly, age (B = −0.298, $p = 0.009$) and education (B = 0.267, $p = 0.029$) were found to be significant determinants of decreased verbal pair total score. **Conclusions:** In this study, it was found that no association between homocysteine and cognitive impairment in a population of institutionalized subjects. Age and education were more significantly associated with cognitive impairment scores than homocysteine.

Keywords

Homocysteine, Cognitive Impairment, Thai

1. Introduction

The prevalence and incidence of dementia increase dramatically with age. It affects 8% of people age over 65 and more than 60,000 new cases each year in Canada. Alzheimer's disease accounts more than 50% of dementia cases in Canada [1]. Since 2002, Thailand has been an ageing society with older persons constituting more than 10% of population. In Thailand, studies showed that the prevalence of dementia is 1.8% - 10.2% in the age group of 55 years and above [2] [3]. Health Systems Research Institute of Thailand surveyed 21,960 people aged over 60 years in 2008-2009. The findings showed that 12.4% of people aged over 60 years had dementia and 9.8% being male while 15.1% were female. According to the latest population census surveyed in 2010 by the Office of National Statistics, it reports that the ageing population accounted for 12% of the whole population, and it is expected that the number will increase to 17% by the year 2020. It is estimated that there are as huge number as at least 300,000 people who have been diagnosed as having dementia across the nation. Cognitive impairment is one major symptom of dementia. Older persons increase in our society, which means a big number of people with decreased cognitive function. Therefore, it is important to search for modifiable risk factors. The amino acid homocysteine may be such a risk factor [4]. Most longitudinal studies have found associations between cognitive function scores and homocysteine levels. As can be seen in **Table A**, studies involving healthy elderly people yielded conflicting results. Some have shown significant associations between homocysteine levels and cognitive function [5]-[11], whereas others have not [12] [13].

Studies investigate the relation between homocysteine and cognitive function scores, as shown in **Table A** [1].

Therefore, it is important to search for modifiable risk factors. The amino acid homocysteine may be such a risk factor [4].

Homocysteine is metabolized through 2 different pathways (**Figure 1**) [1] [14]. The effects of homocysteine in the brain are multiple but can be broadly divided into nurotoxic and vascular effects (**Box 1**) [1] [14].

2. Subjects and Methods

2.1. Subjects

The 100 subjects in this study were recruited from the Baan Bangkae Social Welfare Development Center for Older Persons, Bangkok and the vicinity. The study was approved by the Medical Ethics Committee of Ramathibodi Hospital, Mahidol University, Bangkok, Thailand, and written informed consent was obtained from all participants. All resident who were aged 60 - 80 years were invited to participate. During a visit, trained interviewers administered a questionnaire covering, among other areas, sociodemographic background, medical history, and medication use. This was followed by 2 visits to the Baan Bangkae Social Welfare Development Center, where subjects underwent clinical examinations, including neuropsychologic testing.

Table A. Studies investigating relation between homocysteine and cognitive function scores [1].

Study	Study population	Study design	Cognitive assessment	Results	Comment
Budge *et al*., 2002 [5]	158 community dwelling people age 60 - 91 yr	Cross section of prospective cohort	CMCOG, MMSE, GDS	Higher tHcy levels associated with lower memory scores per umol/L (OR 1.15, 95% CI 1.10 - 1.27)	OR adjusted for age, sex, serum cystatin C level and systolic blood pressure
Duthie *et al*., 2002 [6]	334 community dwelling people who had participated in Scottish Mental Survey of 1932 and 1947	Cross section	MMSE,NART, RPM, AVLT, WAIS	tHcy levels negatively associated with scores on RPM, WAIS in older cohort with higher tHcy levels (mean 10.9 umol/l, 95% CI 10.1 - 11.5)	Results adjusted for childhood intelligence quotient
Pins *et al*., 2002 [7]	1077 people aged 60 - 90 yr in Rotterdam Scan Study	Cross section of prospective cohort	Abbreviated Stroop tesatl Letter-Digit Substitution Task, Verbal fluency test, PPMST, Modified Rey's test	Patients with tHcy > 14 umol/l had lower scores for global cognitive function (diference-0.20, 95%CI-0.30 - 0.11)	Results adjusted for age, sex, education level, depression, serum creatinine level
Miller *et al*., 2003 [8]	1789 community dwelling people aged $\geq$ 60 yr in Sacramento Area Latino Study on Aging	Cross section of prospective cohort	3MSE, verbal and visual memory tests, object naming conceptualization and attention span tests	Inverse relation between tHcy levels and scores on 3MSE ($p = 0.02$), picture association ($p = 0.05$), verbal attention span ($p = 0.04$), and recognition tests ($p = 0.001$),	Multiple linear regression model included folate, cobalamin, age creatinine, sex, education and acculturation
Ravaglia *et al*., 2003 [9]	650 community dwelling people aged 65 - 91 yr (mean 73 yr) with normal cognitive function in Conselice Study	Population based study	MMSE	Inverse relation between odds of tHcy level > 15 umol/l and MMSE scores	Results adjusted for age, income, education level, serum creatinine level, serum vitamin B index, active lifestyle, coffee and meat consumption
Garcia *et al*., 2004 [10]	281 community dwelling people aged >65 yr	Cross section	Stroop, Mattis DRS, CVLT	Subjects with elevated tHcy levels (>13.9 umol/l) had lower stroop scores than those with normal tHcy levels in univariate analysis ($p < 0.05$)	Strongest association found between methylcitric acid and cognitive scores
Dufouil *et al*., 2003 [11]	1241 people aged >60 yr in Epidemiology of Vascular Aging Study	prospective cohort; 4-yr follow-up	MMSE, Trail Making Test Part B, Digit Symbol Substitution Test from the WAIS, Finger Tapping Test	Odds of cognitive decline 2.8 (95% CI 1.2 - 6.2) in patients with tHcy level $\geq$ 15 umol/l	OR adjusted for age, sex, education level, baseline cognition, BMI, alcohol consumption, smoking, hypertension, hypercholesterolemia, Glycemix status, history of vascular disease, and folate and B_{12} levels
Kalmijn *et al*., 1999 [12]	702 community dwelling people aged > 55 yr in Rotterdam Study	prospective cohort; mean follow-up 2.7 yr	MMSE	No association between tHcy and cognitive impairment (highest v. lowest tertile, OR 0.91, 95% CI 0.52 - 1.58)	OR adjusted for age, education level, and baseline MMSE score
Ravaglia *et al*., 2000 [13]	54 people aged > 65 yr in Conselice Study	Cross section of prospective cohort	MMSE, clock drawing test, prose memory test, Corsi block tapping task, Mental Deterioration Battery	No association between tHcy and cognitive test scores	Results adjusted for age, sex, education level, smoking status, alcohol or coffee consumption, and previous cardiovascular disease

Neurotoxic
-Hyperactivation of N-methyl-D-aspartate receptors
-Apoptosis
Vascular
-Increased proliferation of smooth muscle cells
-Increased platelet aggregation
-Increased number of strokes and white matter lesions

Box 1. Effects of elevated homocysteine levels in the brain

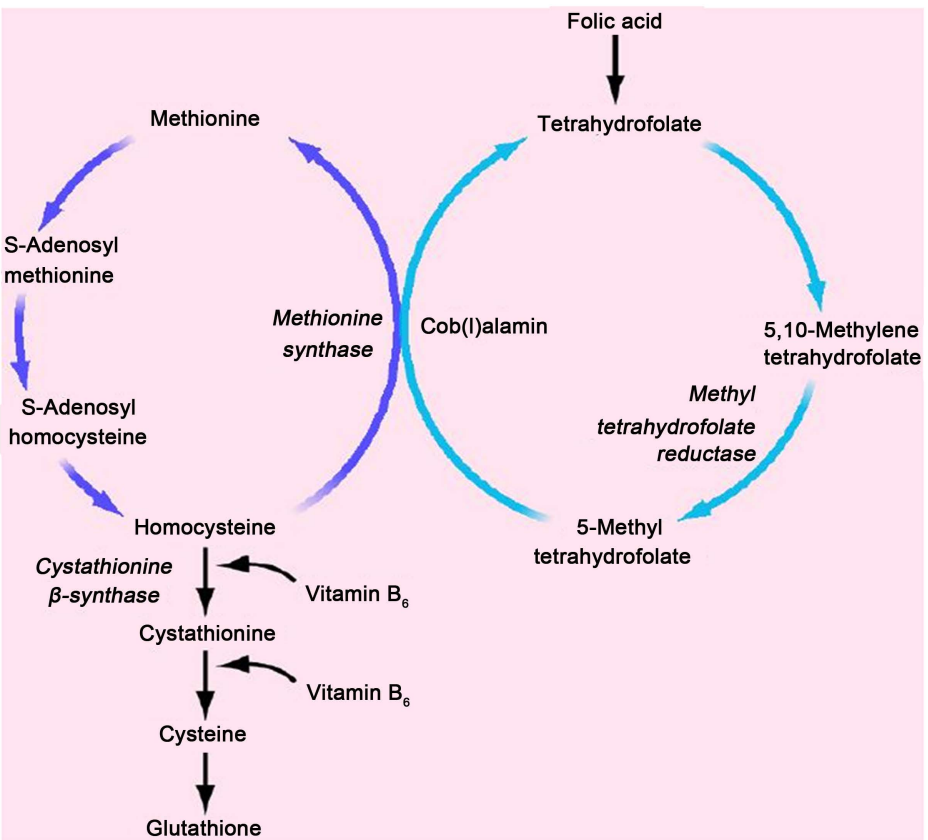

Figure 1. Homocysteine is metabolized through 2 different pathways: the methionine synthase pathway and the cystathionine pathway. It has been proposed that impaired remethylation of homocysteine produces an increase of intracellular homocysteine that is toxic to neurons, and a decrease in S-adenosyl methionine. Impairment of this reaction would occur in states of cerebral oxidative stress, which would augment oxidation of an intermediate form of vitamin B_{12} (cobalamin) produced in the methionine synthase reaction and compromise the conversion of the vitamin to its metabolically active form. By Canadian Medical Association, Angeles Garcia, and Katherine Zanibbi, CMAJ, 2004, 171, 897-904.

2.2. Laboratory Measurement

Venous blood sample after overnight fasting were drawn according to standard procedure. Plasma or serum was isolated and stored at −80˚C before analysis. Serum total homocysteine was measured at the clinical chemistry laboratory of the Ramathibodi Hospital, Mahidol University, Bangkok, using automated chemiluminescent enzyme

immunoassay method (Diagnostic Products Corporation, Los Angeles, CA); the CV ranged from 4.1% to 10.2%.

Global cognitive function was assessed with the Thai language by using mini-Mental State Examination score (MMSE), and cognitive functions were assessed by a neuropsychological battery test for memory, executive function, attention, visual-spatial organization, information processing and motor speed.

Other measurements: The following variables were considered as possible confounders: age; sex; cigarette smoking (current, former, never); alcohol consumption, assessed with a semi quantitative food frequency questionnaire [15]; level of education, group into 4 levels (complete primary education, lower vocational or general education, intermediate vocational or general education, and higher vocational training, college, or university) [16]; and hypertension, which was defined in accordance with the WHO [17] as a systolic blood pressure of 160 mmHg or more, a diastolic blood pressure of 95 mmHg or more, or use of antihypertensive medication.

2.3. Statistical Analysis

Multiple linear regression analysis was used to examine the relations between homocysteine levels and neuropsychological test scores with control for potential confounding variables to evaluate whether the relations were altered by these other variables. All tests were two-sided, and a p value of less than 0.05 was considered to be statistically significant. All data analyses were done with SPSS version 17.0 (SPSS Inc., Chicago, IL).

3. Results

The characteristics of the study population are summarized in **Table 1** and **Table 2**.

In **Table 1**, the mean ± SD age of participants was 72.8 ± 4.6 y. About one-half of the population had a history of hypertension (54.6%), 24.1% had diabetes mellitus and 15.1% had cardiovascular disease, respectively.

Table 2 for neuropsychological test found that the highest % abnormality test of C-W stimulustest, follow by digit symboltest, C stimulus, trial marking test D-KEFS condition 5, verbal pair total (VP total), retention, verbal paired associates 2(VP2), and block design (73.1, 38.3, 34.4, 26.6, 22.3, 15.6, 13.8 and 13.8), respectively.

Table 3 in the model 1, homocysteine was inversely associated with scores for learning slope test (B = −0.048, p = 0.042) and verbal pair total test (B = −0.124, p = 0.032). After adjusting for confounders, no association was found between homocysteine and cognitive impairment. Age (B = −0.129, p = 0.007) was found to be a significant determinant of decreased learning slope score (**Table 4**, model 2). Similarly, age (B = −0.298, p = 0.009) and education (B = 0.267, p = 0.029) were found to be significant determinants of decreased verbal pair total score (**Table 5**, model 3).

4. Discussion

High homocysteine levels have been associated with an increased risk of stroke and other cardiovascular events [18], which, in turn have been related to decreased cognitive

Table 1. Demographic and laboratory characteristics of the study subjects.

Variables	Value	% abnormal laboratory (only)
Age (y)	72.8 ± 4.6 (61 - 80)	
Education (y)	6.3 (0 - 18)	
Hypertension (%)	54.6	
Diabetes mellitus (%)	24.1	
Cardiovascular disease (%)	15.1	
Homocysteine (μmol/L)	14.2	34.0
Vitamin B 12 (pg/mL)	612.1	4.26
Serum Folic (ng/ml)	11.6	0.0
RBC folate (ng/ml)	498.5	0.0
Cholesterol (mmol/L)	5.54	60.0
LDL (mmol/L)	3.49	50.0
HDL (mmol/L)	1.36	23.40
Creatinine (μmol/L)	70.8	4.3 (female) 12.0 (male)

Table 2. Cognitive performance of the study subjects.

Test score	Median (95% CI)	% abnormal (<1.5 SD)
MMSE	27.0 (25.6 - 28.3)	0.0
VP1	0.0	0.0
Learning slope	2.09 (1.9 - 2.1)	3.2
VP2	1.5 (1.43 - 1.57)	13.8
VP total	5.0 (4.75 - 5.25)	22.3
Recognition	23.0 (21.85 - 24.15)	0.0
Retrieval	21.0 (19.95 - 22.05)	0.0
Retention	66.7 (63.36 - 70.03)	15.6
Digit forward	8.0 (7.6 - 8.4)	0.0
Digit backward	4.0 (3.8 - 4.2)	0.0
Block design	9.0 (8.5 - 9.5)	13.8
Trial Marking Test D-KEFS condition 5	52.6 (50.0 - 54.2)	26.6
Digit symbol	16.5 (15.6 - 17.4)	38.3
C stimulus	112.0 (106.4 - 117.6)	34.4
C-W stimulus	36.0 (34.2 - 37.8)	73.1

Table 3. Association between homocysteine and cognitive test score (model 1).

Cognitive test	Homocysteine (β)	p-value
MMSE	−0.010	0.752
VP1	−0.010	0.384
Learning slope	−0.048 (*)	0.042 (*)
VP2	−0.021	0.273
VP total	−0.124 (*)	0.032 (*)
Recognition	0.007	0.869
Retrieval	0.032	0.373
Retention	0.528	0.415
Digit forward	−0.007	0.798
Digit backward	0.004	0.870
Block design	−0.063	0.436
Trial Marking Test D-KEFS condition 5	0.584	0.081
Digit symbol	−0.255	0.196
C stimulus	−0.069	0.586
C-W stimulus	−0.498	0.087

Verbal pair total: VP total, *significant $p < 0.05$.

Table 4. Multiple linear regression between homocysteine, B12, folic, age, education, Cr and learning slope (model 2).

Cognitive test	B	p-value
Homocysteine	−0.015	0.520
Vitamin B 12	0.000	0.824
RBC folate	−1.126	0.971
Serum folic	0.005	0.876
Age	−0.129	0.007 (**)
Education	0.067	0.129

Table 5. Multiple linear regression between homocysteine, B12, folic, age, Cr and verbal pair total (model 3).

Cognitive test	B	p-value
Homocysteine	−0.080	0.252
Vitamin B 12	0.010	0.894
RBC folate	−0.006	0.716
Serum folic	0.013	0.786
Age	−0.298	0.009 (**)
Education	0.267	0.029 (*)

function and dementia [19] [20] [21]. Thus, we hypothesized that a high level of homocysteine was associated with cognitive function. However, in the present study, there was no significant association between high levels of homocysteine and cognitive impairment in a population of institutionalized subjects (Baan Bangkae). Several methodological

Considerations arise when we try to explain our negative findings. We used only one global measure of cognitive function, the MMSE. However, the MMSE is a valid and reliable test [22]. However, the MMSE was not developed to estimate change in cognitive impairment. In addition, with our definition of cognitive decline, we were able to find an association with known risk factors, such as age and education. Similar results from Seshadri *et al*. [23] which found that cognitive decline associated with age education, and stroke. Still, random misclassification may have diluted our results for cognitive decline [12]. Demographic variables, particularly age and education, were significantly associated with cognitive function scores than was homocysteine. The reasons may be the small number of sample size (100), normal level of homocysteine in blood samples (14 umol/l, 34.04% hyperhomocysteinemia) used cut-off point >15 umol/l compared with other study which had a higher homocysteine levels (cut-off point >13 umol/l) [1] [24] [25] [26] than our study 1, age of elderly Baan Bangkae, higher than other communities and had many diseases (HT, hyperlipidemia, DM, CVD) that effect homocysteine level and cognitive impairment.

5. Conclusion

In summary, although an association between homocysteine and cognitive impairment was biologically plausible, homocysteine did not seem to be a risk factor for cognitive impairment in this general population of the elderly. However, the possibility that homocysteine is truly not related to cognitive impairment cannot be discarded. Further researches such as a large number of participants and subjects in community are needed to find out for better quality of life in elderly population.

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