EDITORIAL COMMENT

Plasma Hyperhomocysteinemia, *MTHFR* Polymorphism and Thromboembolic Disease: An Example of Gene-nutrition Interactions in Chronic Disease

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Many studies have shown that hyperhomocysteinemia may be an independent risk factor for thromboembolic disease. However, the results among studies and populations have not been consistent. Furthermore, the possible mechanisms of hyperhomocysteinemia in vascular diseases are controversial. Mutations in the *MTHFR* (5,10-methylenetetrahydrofolate reductase) gene, resulting in rising levels of plasma homocysteine or dietary folate, and vitamin B6 and B12 intake deficiency, may play a role in this complicated process. In this editorial comment, I discuss the metabolism of homocysteine, hyperhomocysteinemia in vascular diseases, and MTHFR deficiency (*MTHFR* gene mutation) and/or nutritional deficiency and their impact on thromboembolic diseases. Finally, based on previous study results, the possible gene-nutrient interactions in the pathophysiology of hyperhomocysteinemia in thromboembolic diseases are clarified.

Homocysteine, a sulfur-containing amino acid, is an intermediate product in methionine metabolism. Most of the dietary methionine is converted to S-adenosylmethionine and then to S-adenosylhomocysteine. Hydrolysis of S-adenosylhomocysteine leads to adenosine and homocysteine. Homocysteine may then be metabolized either by transsulfuration or transmethylation, depending on the availability of methionine.[1,2] Several factors are associated with the regulation of methionine and homocysteine metabolism that depends on the presence of adequate enzyme levels and their kinetic properties. In general, plasma homocysteine levels increase with age in both genders; men have higher plasma homocysteine levels than women. However, both genetic and environmental factors play important roles in this metabolic pathway. For genetic factors, cystathionine beta-synthase (CBS) deficiency and MTHFR deficiency are usually associated with elevated plasma homocysteine levels. For environmental factors, nutritional intake such as dietary deficiencies of folate, vitamin B6 and B12 are associated with raised plasma homocysteine concentrations. Riboflavin is one of the cofactors for MTHFR that may be correlated with high plasma homocysteine when it is deficient.[3,4]

Several studies have shown that excessively high concentrations of plasma homocysteine may be associated with premature vascular diseases and thromboembolic vascular lesions. Plasma homocysteine is considered to be an independent, graded and strong risk factor for cardiovascular disease.[5–10] Wilcken and Wilcken[11] first proposed the possible association between plasma homocysteine level and vascular disease in 1976. Since then, many studies have shown that high plasma homocysteine level is an independent predictor for the subsequent development of cardiovascular disease.[6,7] However, the mechanisms behind the influence of high plasma homocysteine on cardiovascular disease remain unclear. Plasma homocysteine levels correlate negatively with plasma folate, vitamin B6 and B12 levels that indicate inadequate dietary intake in patients with cardiovascular disease. More interestingly, decreased activity of certain enzymes involved in homocysteine metabolism could also play a role in these patients. The thermolabile

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form of MTHFR is associated with impaired homocysteine metabolism resulting in higher plasma homocysteine levels and higher risk of developing cardiovascular disease.[3,12–14]

MTHFR plays an important role in homocysteine metabolism. It catalyzes the conversion of 5,10-methylenetetrahydrofolate to 5-methyltetrahydrofolate. Deficiency of MTHFR is associated with hyperhomocysteinemia and may be related to neurologic abnormalities, mental retardation, atherosclerosis and thrombosis.[3,12–15] In 1995, Frosst et al[13] reported a thermolabile variant of MTHFR that has a common 677 C-to-T mutation in *MTHFR*. The thermolabile MTHFR enzyme activity was 17% in patients with coronary heart disease and only 5% in normal subjects. In 1996, Jacques et al[3] measured MTHFR activity and folate levels in 365 subjects and found that those who were homozygous for C677T had higher homocysteine and lower folate levels when compared with those who had normal genotypes. Guttormsen et al[4] screened 18,043 subjects and found that hyperhomocysteinemia was associated with low plasma folate levels and with the thermolabile *MTHFR* mutation. Sunder-Plassmann and Fodinger[14] examined several different gene polymorphisms, such as *MTHFR*, *GCP2*, *RFC1*, and *TCN2*, in homocysteine metabolism. The most consistent effect on plasma homocysteine levels was observed for the 677 C-to-T mutation in *MTHFR*, whereas the other gene polymorphisms had no major effect on plasma homocysteine concentrations. Based on these studies, although the main regulating enzymes of homocysteine metabolism are CBS and MTHFR, the *MTHFR* 677 C-to-T mutation seems to play a more important role in affecting plasma homocysteine levels.[15]

Apart from genetic factors, the dietary intake of folate, vitamins B6 and B12 is also associated with plasma homocysteine levels. For homocysteine metabolism, vitamin B6 is a cofactor involved in CBS metabolism, which metabolizes homocysteine. Folate and vitamin B12 are cofactors involved in MTHFR metabolism, another metabolic pathway to convert homocysteine back to methionine. In the Framingham Heart Study population, Selhub et al[10] reported that intake and blood levels of both folate and vitamin B6 were inversely related to plasma homocysteine level, but for vitamin B12, only blood levels, not intake, were associated with lower homocysteine, which may be due to variation in absorption rather than to dietary intake being the main determinant of vitamin B12 levels. Deficiencies in folate, vitamins B6 and B12 usually cause elevation of homocysteine levels, and there is negative correlation with all of these vitamins, even in subjects with normal vitamin levels. More interestingly, a lower plasma vitamin level is associated with a steeper rise in homocysteine levels.[4]

Many studies have evaluated MTHFR deficiency (*MTHFR* gene mutation) and/or nutritional deficiency in thromboembolic diseases. Meleady et al[16] conducted a case-controlled study of 711 patients with vascular diseases and 747 controls to evaluate the effects of *MTHFR* genotype, and plasma homocysteine and folate levels on the risk of vascular disease. There was a strong dose-response relationship between plasma homocysteine level and vascular disease risk, and the *MTHFR* genotype plays a key role in determining plasma homocysteine concentration. However, the vascular risk of the TT genotype (homozygotes for the thermolabile mutation) was strengthened after adjusting for conventional cardiovascular disease risk factors but was attenuated after adjusting for plasma homocysteine and folate levels. Further, there is more evidence to suggest that *MTHFR* mutation is only associated with plasma homocysteine levels but not the risk of cardiovascular diseases in certain populations.[17–19] Brattstrom et al[20] had similar findings based on a meta-analysis of 23 studies, i.e. that *MTHFR* gene mutation only leads to hyperhomocysteinemia but not to vascular disease. However, in those studies, dietary folate or vitamin B intakes were not fully considered and may be potential confounders for those populations with vascular disease and with low-normal folate and/or vitamin B intakes.[15] Rimm et al[21] found that supplement and dietary intake of both folic acid and vitamin B6 were associated with reduced risk of coronary heart disease in the Nurses' Heart Study cohort.

These current data on mutation of the *MTHFR* gene do not tell us the whole story of how hyperhomocysteinemia plays a causal role in vascular disease. More complicated gene-nutrient interactions indicate that not only *MTHFR* gene mutation but also dietary intakes of folate and B vitamins play significant roles in the occurrence of thromboembolic disease.

In this issue of the *Journal of the Chinese Medical Association*, Ho et al[22] examine 106 subjects with type 2 diabetes, deep vein thrombosis or coronary artery disease to evaluate the influence of *MTHFR* C677T polymorphism, and plasma folate, vitamin B6 and B12 levels on thromboembolic disease in Chinese patients. They found that *MTHFR* C677T polymorphism, vitamin B12 and triglyceride levels were the most significant factors associated with plasma homocysteine level. Patients with coronary heart disease had higher plasma homocysteine levels than healthy subjects and those with diabetes. These results are

consistent with previous hypotheses and suggest that a *MTHFR* TT homozygous genotype and vitamin B12 level are important factors in the determination of plasma homocysteine levels, and homocysteine levels are associated with thromboembolic diseases. More interestingly, in this study, the authors found that plasma triglyceride had the greatest influence on plasma homocysteine level in healthy subjects, coronary artery disease patients, and all subjects together. Whether these characteristics are only present in Chinese or are related to special dietary intake needs further evaluation. However, perhaps due to the small sample size in their study, the authors did not examine the independent effects of plasma homocysteine, folate, vitamins B6 and B12 levels and *MTHFR* polymorphism on different disease states to determine which factor is the most significant predictor on the development of thromboembolic disease in Chinese. From their study, homozygous *MTHFR* may play a role in higher plasma homocysteine and the development of cardiovascular end-points. Furthermore, the dietary intakes and plasma levels of folate, vitamins B6 and B12 may also play synergistic roles in cardiovascular disease occurrence.

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