Introduction

Vascular diseases are commonly associated with traditional risk factors, but in the last decade other risk markers have been identified, one of them being homocysteine. The increasing interest in mild hyperhomocysteinaemia as a possible coronary and cerebrovascular risk factor has suggested that elevated plasma levels of homocysteine are associated with an increased risk of atherosclerosis and cardiovascular ischaemic events. Cardio- and cerebrovascular diseases are multifactorial, as their aetopathogenesis is determined by genetic and environmental factors and by gene-gene and gene-environment interactions. Experimental studies have shown that many possible mechanisms are implicated in the pro-atherogenic effect of homocysteine. Hyperhomocysteinaemia may confer a mild risk alone, but it increases the risk of disease in association with other factors promoting vascular lesions. Variants in genes encoding enzymes involved in homocysteine metabolism, or depletion of important cofactors or substrates for those enzymes, including folate, vitamin B12 and vitamin B6, may result in elevated plasma homocysteine levels. Several studies have been performed to elucidate the genetic determinant of hyperhomocysteinaemia in patients with vascular disease, and the MTHFR 677C>T polymorphism is the one most extensively investigated. However, the lack of homogeneity in the data and the high number of factors influencing plasma homocysteine concentrations remain conflicting. Moreover, studies on the evaluation of therapeutic interventions in improving the atherogenic profile, lowering plasma homocysteine levels, and preventing vascular events, have shown inconsistent results, which are reviewed in this paper. More prospective, double-blind, randomized studies, including folate and vitamin B interventions, and genotyping for polymorphisms in genes involved in homocysteine metabolism, might better define the relationship between mild hyperhomocysteinaemia and vascular damage.

Keywords: cardiovascular disease, cerebrovascular disease, gene, hyperhomocysteinaemia, MTHFR, polymorphisms, vascular risk factors.
farction, peripheral arterial disease, and venous thrombosis (Brattström et al. 1984; Boers et al. 1985; Stamper et al. 1992; Selhub et al. 1995; Arnesen et al. 1995; Perry et al. 1995; den Heijer et al. 1996). Moreover, results from case-control, cross-sectional, retrospective and prospective studies have shown a graded correlation between homocysteine levels and the risk or severity of cardiovascular disease, suggesting that elevated homocysteinaemia is a causal risk factor (Boushey et al. 1995; Mudd et al. 1995; Wilcken and Wilcken 1976; Brattström et al. 1984; Boers et al. 1985; Stamper et al. 1992; Selhub et al. 1995; Perry et al. 1995; den Heijer et al. 1996; Graham et al. 1997).

A further input of our understanding of hyperhomocysteinaemia has been given by biochemical investigations, which showed an association with a thermolabile variant in methylenetetrahydrofolate reductase (MTHFR). This variant exhibits a reduced catalytic activity, and is associated with an elevation of tHcy, and with atherosclerotic disease (Kang et al. 1988, 1991). Next, the identification of the 677C>T mutation in the MTHFR gene, and the observation that homozygotes for the variant showed hyperhomocysteinaemia, suggested that the 677C>T mutation could be a genetic risk factor for cardiovascular disease (Frosst et al. 1995).

The objective of this paper is to review the available literature data on relationships between plasma homocysteine levels, MTHFR gene polymorphisms, and cardio-cerebrovascular risk.

Homocysteine biochemistry

Homocysteine is a sulphur amino acid that is formed by demethylation of methionine. Methionine, an essential amino acid (present in meat, milk, eggs, legumes, etc.), is activated to form S-adenosylmethionine (SAM), the universal methyl group donor. S-adenosylhomocysteine is formed when SAM donates the methyl group; S-adenosylhomocysteine is hydrolysed to generate homocysteine and adenosine. As shown in Figure 1, homocysteine can be remethylated to methionine by methionine synthase (MTR), which requires 5-methyl-tetrahydrofolate (CH3-THF) as a methyl donor, and methylcobalamin (a biologically active form of vitamin B12) as a cofactor for its enzymatic activity. CH3-THF is formed upon the reduction of 5,10-methylenetetrahydrofolate, a reaction that is catalysed by the enzyme MTHFR. The cycle preserves methionine, which in its activated form (SAM) is the principal methyl donor in numerous reactions, e.g. in methylation of DNA, RNA, hormones, lipids and neurotransmitters (Rezvani et al. 2002; Mudd et al. 2001). In the liver, where methionine metabolism is very active, another enzyme, the betaine-homocysteine-methyltransferase (BHMT), participates in the synthesis of methionine with betaine or trimethylglycine as a methyl donor.

If an excess of dietary methionine occurs, MTR is inhibited and the transsulphuration pathway becomes active, homocysteine interacting with serine to give cystathionine. Cystathionine β-synthase (CBS) catalyses this initial step, requiring pyridoxal 5’ phosphate (the active form of vitamin B6) for its activity. Cystathionine is subsequently hydrolysed by another vitamin-B6-requiring enzyme, γ-cystathionase, to form cysteine and α-ketobutyrate. Excess cysteine is oxidized to taurine or organic sulphates or is excreted in the urine (De Vecchi et al. 1999).

In physiological conditions, a balance between homocysteine formation and degradation is present, and about 50% is remethylated to methionine. Excess homocysteine is exported into circulation, causing elevated plasma or urine levels. In the circulation, <1% homocysteine is present in the free reduced form, while 10–20% of tHcy is present as homocysteine-cysteine mixed disulphide and homocystine (the dimer of homocysteine), while the remaining 80–90% of homocysteine is protein-bound. Homocysteinaemia is the plasma total amount of all the homocysteine forms (tHcy) (Hankey and Eikelboom 1999). Plasma tHcy levels can vary within a wide range, while intracellular levels are confined to a narrow range (Moat et al. 2004). Plasma concentrations depend on different dietary habits within populations and among groups belonging to the same population. The normal plasma concentration ranges from 5 to 15 μmol L–1. Moderate hyperhomocysteinaemia occurs from 15 to 30 μmol L–1 tHcy levels, intermediate hyperhomocysteinaemia from 30 to 100 μmol L–1, and severe hyperhomocysteinaemia at levels higher than 100 μmol L–1 (Kang et al. 1992).

Hyperhomocysteinaemia

Hyperhomocysteinaemia is a multifactorial disease; smoking, coffee consumption, and lack of exercise can raise tHcy (Refsum et al. 1998; Nygard et al. 1997a; Nygard et al. 1998). Increasing age and male gender have been found to be associated with increased homocysteine concentrations (Ueland et al. 2001). In women, increasing homocysteinaemia is observed after