GREAT DEBATE

Homocysteine and the vascular endothelium

Peter Jacobs, Lucille Wood

Obstruction to the circulation has catastrophic consequences. On the high-flow arterial side acknowledged risk factors include dyslipidaemia, smoking, poorly controlled hypertension and renal disease. Additionally, venous thrombosis correlates with obesity, hormonal imbalance and reduced mobility especially when this is combined with dehydration, for example in long-distance air travel. Superimposed upon such environmental predictors of hypercoagulability is an impressively expanding list of genetic determinants that impair functional integrity of intimal cells, and this happens irrespective of anatomical site. Among these are reduced levels of antithrombin III, protein C or S and mutations in factor II or V, with the latter giving rise to resistance in protective effects of activated protein C. Furthermore there is an increasing awareness of the adverse effects that follow sustained elevation in plasma homocysteine levels. This may be hereditary and so explain the familial occurrence of premature vascular disease whether in the cerebral, coronary or peripheral circulations, but an abundance of data document the comparable hazard that is acquired with renal or hepatic failure or with use of many drugs.[1] Neither should it be overlooked that such predisposing factors can occur together with compelling data that appropriate doses of folic acid and vitamins B_{12} and B_6 can return the hyperhomocysteinaemia to normal.[2] The relevant issue is what role needs to be assigned to such intervention in primary as opposed to secondary prevention.[3]

Physiology

Physiologically the huge intimal area presents a non-thrombogenic and antiadhesive surface to the blood.[4] Protective molecules include prostacyclin and natriuretic peptide that are opposed by the endothelins and thromboxane A_2.[4] The secretion of these active mediators plays a key role in coagulation as well as modulating immune response and contributing to vascular tone.

Pathophysiology

Pathophysiologically it is seen that homocysteine inflicts its damage by directly disrupting this barrier in a dose and time-dependent manner, doing so incrementally even within the normal range. The injury results from generation of active radicals, reduction in nitric oxide and increase in adhesion proteins that enhance platelet aggregation. Such a changing phenotype shifts the balance to coagulation through release of tissue plasminogen activator, impairment of thrombomodulin and diminished generation of the naturally occurring anticoagulant protein C.[4]

Molecular level

At molecular level raised plasma concentration reflects, most typically, genetically determined decreases in cystathionine β-synthase, mutation in thermolabile variant of methylenetetrahydrofolate reductase or methionine synthase deficiency. Acquired causes range from increasing age and menopause, through lifestyle where tobacco and coffee have been incriminated to deficiencies of cobalamin, pyridoxine and folic acid. Additionally, similar elevations occur in hepatic impairment, renal dysfunction, systemic lupus erythematosus and a number of malignancies as well as solid organ transplantation.[1] Of particular note is a wide range of drugs in regular use exemplified by methotrexate, phenytoin, azathioprine, theophylline, metformin, thiazide diuretics, colestipol, nicotinic acid and oral contraceptives.[1]

Treatment

Treatment rests on appreciating that this amino acid is constantly generated from methionine. It is then disposed of by remethylation dependent upon an adequate supply of folate and maintenance of normal activity for the specific synthase requiring vitamin B_{12} as the co-factor. There is an alternative pathway in the liver where the methyl donor is betaine and this requires the presence of a specific transferase. Clearance of the offending molecule from the plasma also takes place by transsulphuration requiring vitamin B_6.[3]

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Therapeutic interventions

Therapeutic interventions are based on this biochemical background. Thus, there is a graded hazard for developing atherosclerosis as plasma levels increase. This is an independent factor equivalent to smoking and hyperlipidaemia, in which a 5 µM increment, even in the normal range, increases the risk between 60% and 80% simultaneously targeting cerebral, peripheral and coronary vasculature.[3]

Although pathophysiology is clear, controversy inexplicably persists regarding treatment. Perhaps this is because, for primary prevention, proof of benefit is not yet sufficient to justify worldwide supplementation although preliminary results are expected to reach statistical significance within 3 years.[5] Quite different is the specific use of these vitamins to avoid secondary complications. Thus, in hyperhomocysteinaemic individuals, who have had an occlusive episode, doses of cobalamin, folate and pyridoxine to reduce the levels to the low-normal range is rational. Studies in first-degree relatives are also quite appropriate and protection with replacement therapy should be offered to such people with raised values.[1]

Conclusions

Conclusions are threefold and quite clear. Rationally it is recognised that local authority or government need to add at least folate to some widely consumed food staple. Secondly, a documented or arterial venous event, in the patient with elevations in plasma level of this amino acid — especially when other risk factors are also present — should receive this quite specific form of treatment that differs in no way conceptually from pharmacological lowering of blood pressure or management of hyperglycaemia. Thirdly, close family members who may not yet have had arterial or venous thromboembolism but are similarly affected and particularly when defects in naturally occurring anticoagulant mechanisms are present, justify equivalent consideration. This is a compelling argument that needs to be widely acknowledged and responded to by doctors and nurses as well as third party payers in South Africa — as it is elsewhere in the world.

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Plasma homocysteine and arterial thromboembolic disease

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Population studies of coronary vascular disease such as the Framingham study have identified cigarette smoking, hypertension, diabetes mellitus, age, raised total cholesterol, and low high-density lipoprotein (HDL) cholesterol as major and independent risk factors for coronary heart disease (CHD).[1] These traditional risk factors may, however, be insufficient to account for all cases of CHD and they do not adequately explain the large differences in CHD rates between populations.[2] Novel risk factors such as homocysteine have therefore gained popularity as potential independent risk factors for CHD.[3]

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Background

The hypothesis that homocysteine may be causally related to thromboembolic disease stems from the observation that young adults with the rare autosomal recessive disease of congenital homocysteinuria die prematurely of atherosclerosis and thrombosis. These individuals have extremely high plasma levels of homocysteine and are generally treated with methionine/cysteine-restricted diets and some sort of vitamin B supplementation, depending on the specific underlying enzymatic defect. Homocysteine is an amino acid generated in the metabolism of dietary methionine. The latter is derived exclusively from animal protein. Homocysteine undergoes metabolism either by remethylation or transsulphuration. Genetically determined enzyme dysfunctions (e.g.

heterozygous cystathionine synthase deficiency), as well as substrate and co-factor abnormalities, including folate, vitamin B_6 and vitamin B_{12} deficiency, may lead to elevated homocysteine levels. Furthermore, methionine-rich diets, excessive coffee consumption, high alcohol intake, smoking, and lack of physical activity have been associated with high serum concentrations.[3] The prevalence of hyperhomocysteinaemia in the general population has been estimated to be between 5% and 10%, and as high as 30 - 40% in the elderly population.[4] Considering the multiple genetic and environmental factors that can influence homocysteine levels, it is not surprising that such figures vary geographically.[5] The prevalence in various population groups in South Africa is unknown.

The evidence

Over the past 30 years there has been an exponential increase in publications relating to the association between raised homocysteine levels and occlusive vascular disease. Various reviewers of the subject have concluded that elevated homocysteine is an independent risk factor for vascular disease in coronary, cerebral and/or peripheral blood vessels.[3,4,6] The initial evidence supporting such a theory came from cross-sectional and retrospective case-control studies such as the one by Clarke *et al.*[7] Subsequent prospective population-based cohort and nested case-control studies supported such findings. Nevertheless, results have not been consistent. Some studies have indicated a continuous dose-response relationship between increasing homocysteine concentrations and CHD events,[8,9] whereas others have suggested a threshold effect.[5,10] The US Physicians Study[10] demonstrated a positive association in the top 5% of the homocysteine distribution when compared with the bottom 90%. It is interesting that these findings could not be reproduced when the same cohort was followed beyond 5 years.[11]

To add to the confusion, other large population-based prospective cohort studies have failed to show an independent association.[12-14] The most recent is a 10-year follow-up of the Caerphilly cohort in the UK.[15] This was a nested case-control study comparing homocysteine levels in 312 men (of an original cohort of 2 290) who developed acute myocardial infarction or death, with 1 248 randomly selected, age frequency matched controls. Although geometric mean homocysteine concentrations were slightly higher in cases than controls, this did not reach statistical significance (12.2 µmol/l, 95% CI: 11.8 - 12.6 versus 11.8 µmol/l, 95% CI: 11.3 - 12.5, $p = 0.09$). Also, comparing the top 5% of the homocysteine concentration with the remaining 95%, the adjusted odds ratio of CHD was 1.05 (95% CI: 0.56 - 1.95, $p = 0.9$). A linear association of homocysteine levels and cardiovascular endpoints was observed, but this disappeared when controlled for confounding variables such as smoking, obesity and physical activity.

Hence, although homocysteine appears to be positively associated with vascular events, it remains to be seen whether it is an independent, and furthermore, causal risk factor for thromboembolic disease. Elevated levels of plasma homocysteine have been associated with major components of the cardiovascular risk profile, including male gender, old age, smoking, hypertension, hyperlipidaemia and a sedentary lifestyle.[16] They are also increased with renal and hepatic impairment. It may be that high levels of homocysteine are a result of, rather than the cause of, active underlying vascular disease and positive results in short-term follow-up studies may reflect prevalent subclinical vascular disease. Such reverse causation has been suggested to occur in studies of another putative vascular risk factor, lipoprotein(a).[17] It may also be that hyperhomocysteinaemia is only associated with an increased risk at very high levels and/or in young people, which may be indicative of genetic enzyme defects. A stronger association between serum homocysteine level and ischaemic heart disease at younger rather than older ages has been proposed.[9]

Regarding specific therapy for hyperhomocysteinaemia, various studies have demonstrated a lowering in homocysteine levels with folate and vitamin B therapy. According to a meta-analysis, 0.5 - 5 mg of folate reduces homocysteine levels by ~25%, whereas 0.5 mg of vitamin B_{12} produces an additional reduction of 7%. Vitamin B_6 (mean 16.5 mg) did not have any significant effect.[18] Nevertheless, the latter has been proposed as offering independent cardiovascular protection, that appears to be separate to any effect on homocysteine levels.[12] However, no results of prospective randomised controlled trials are available as yet to establish the effects of lowering homocysteine on cardiovascular outcomes, or to ascertain the effect of folate and vitamin B_6 supplementation on such events.

Recommendations

Until such time as there is a better understanding of the relationship between serum homocysteine levels, vascular events and folate/vitamin B_6/vitamin B_{12} levels and supplementation, it is premature to advocate screening and intervention programmes for elevated homocysteine levels. Such practice should be confined to a research setting. This is in agreement with the conclusions by the Canadian task force on preventive health care which conducted an extensive literature review of English-language publications between 1996 and 1999.[4] Adherence to recommended daily allowance of dietary sources of folate and vitamins B_{12} and B_6 should, however, be encouraged and coffee consumption should be kept low.[3] The usual recommendations for a cardioprotective lifestyle must be strongly advocated. Smoking cessation, ample physical activity, moderate intake of animal protein, high consumption of fresh plant produce and avoidance of excessive

alcohol should not only have a beneficial effect on homocysteine levels, but most have long been accepted as offering a benefit to patients with regard to atherosclerotic disease prophylaxis. Innovative ways of promoting such a lifestyle need to be found. Accepting the difficulty regarding the latter, the hopeful enthusiasm for yet another 'pill' and widespread vitamin food fortification programmes in an effort to reduce vascular occlusive disease can be easily understood.

Doctors in the desperate situation of treating patients with accelerated premature atherosclerotic disease in the absence of accepted risk factors or other treatable causes cannot be criticised for prescribing vitamin B complexes. This intervention is relatively cheap, in most likelihood it is safe and it may give the patient the benefit of the doubt of any potential therapeutic benefit. From a health policy and health funding perspective, however, more data are required before resources should be allocated towards the screening and intervention of hyperhomocysteinaemia.

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AIDS FOCUS

Ethical issues in voluntary HIV testing in a high-prevalence area — the case of Malawi

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The first adult case of HIV / AIDS in Malawi was identified in April 1985,[1,2] with the first paediatric case in January 1986.[3] From that time to 1997, at least 10% of the general population and 15% of the 15 - 49-year age group were infected.[4,5] Up to 30% of women attending prenatal care at the Queen Elizabeth Central Hospital, Blantyre, are HIV-infected.[6,7] HIV / AIDS has been associated with a rise in the number of orphans, now estimated at between 400 000 and 1 000 000 as no reliable data are currently available. The maternal mortality ratio, which had been estimated at about 620 deaths per 100 000 live births in the 1992 Demographic and Health Survey (MDHS),[8] has now risen to 1 120 / 100 000,[9] due *inter alia* to the HIV pandemic. Up to 70% of admissions in the medical wards of Blantyre and Lilongwe are HIV / AIDS-related and tuberculosis (TB) has

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