Lack of a relationship between plasma pyridoxal phosphate levels and ischaemic heart disease

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Summary

The 'vitamin B₆-homocysteine theory' has been proposed as an alternative to the widely accepted lipid hypothesis in the aetiology of ischaemic heart disease (IHD). In a cross-sectional study of 71 white men with evidence of IHD and 110 male controls (all aged 45 - 54 years) we have been unable to demonstrate any differences in plasma pyridoxal phosphate (PLP) levels between the groups. It is therefore unlikely that deficiency of vitamin B₆ has a primary causal role in development of IHD. However, 31% of the overall study population had low plasma PLP levels, and the possibility that underlying vitamin B₆ deficiency may facilitate the actions of the primary risk factors for IHD therefore cannot be excluded.

Subjects and methods

Subjects and controls were drawn from among men aged 45 - 54 years who participated in the 1983 Coronary Risk Factor (CORIS) Resurvey carried out in the districts of Robertson, Swellendam and Riversdale in the south-western Cape Province (unpublished data). 'Probable IHD' was regarded as present if there was a history of chest pain (angina or previous infarct on the Rose questionnaire) suggestive of IHD and/or a resting ECG showing Q waves (Minnesota codes) ST-segment depression (4.1, 4.2), T-wave inversion (5.1, 5.2) or complete bundle-branch block (7.1, 7.2). These codes have previously been shown to correlate with the presence of IHD in the CORIS male populations. If there was no history of suggestive chest pain but the presence of other codable ECG changes, i.e. lesser Q-wave (1.3), ST-segment (4.3, 4.4) and T-wave (5.3) changes and conduction defects (7.3, 7.4), together with left axis deviation (2.1), left ventricular hypertrophy (3.1), first-degree atrioventricular block (6.3), extrasystoles (8.1) and atrial fibrillation (8.3), 'possible IHD' was regarded as present. Controls had no history of chest pain or codable ECG items. Subjects who were taking vitamin supplements were excluded from the study. All 100 eligible subjects with evidence of IHD and 151 randomly selected controls were invited to participate. In the probable IHD and possible IHD groups response rates of 70% (33 subjects) and 72% (38 subjects) respectively were obtained, and in the control group the response rate was 75% (113 subjects). Three of the control subjects were subsequently excluded since they were taking vitamin supplements, leaving 110.

Blood samples (subjects did not fast) were taken into a 10 ml Vacutainer containing ethylenediamine tetra-acetic acid and separated within 2 hours, whereafter the plasma was kept frozen at -4°C. The samples were protected from light throughout, and within 2 weeks were analysed on a blind basis at the Metabolic Unit of Tygerberg Hospital using the enzymatic-isotopic method of Chabner and Livingstone. This measures the ¹⁴C-labelled carbon dioxide released from tyrosine by tyrosine decarboxylase obtained from bacteria grown in a vitamin B₆-deficient medium. The activity of the enzyme therefore depends on the concentration of the biologically active vitamin B₆ (pyridoxal phosphate co-enzyme) present in the deproteinized plasma test sample. The normal range of
plasma pyridoxal phosphate (PLP) levels in this laboratory is 6 - 20 ng/ml.

**Results**

The individual results are plotted in Fig. 1, which shows that the distribution of plasma PLP values appears similar in the control group and the two IHD groups. This is confirmed in Table I, from which it can be seen that the means, medians and ranges were very similar. The percentage of subjects with PLP values below 6 ng/ml appears somewhat higher in the two IHD groups (33.3% and 39.5% v. 27.3% in the controls), but the differences in proportions were not significant on chi-square testing, irrespective of whether the IHD groups were considered separately or together.

Further analysis of the PLP levels in different categories of subjects with probable IHD, e.g. comparing those with Q waves on ECG (which can be regarded as the firmest evidence of IHD) with those with a history of chest pain only, showed no significant differences.

**Discussion**

This study did not yield any evidence of a direct relationship between plasma PLP levels and the presence of IHD in men aged 45 - 54 years. (In contrast, the CORIS study has yielded highly significant associations between the primary risk factors and IHD.13) This conclusion is in direct contrast to that of Serfontein et al.,1 which was based on the study of a small group of myocardial infarct patients. Their observation of lower plasma PLP levels in the infarct group may, however, be confounded by uncertainties about the effect of a recent infarct on PLP levels, the validity of their statistical analysis and, more importantly, by the 10-year average age difference between the controls and the patients,14 since the plasma vitamin B6 level is known to decrease markedly with age.15 The only other published study of circulating vitamin B6 levels in relation to IHD comes from behind the Iron Curtain.16 In that study lower plasma vitamin B6 levels, which could be corrected by diet or supplementation, were found in subjects with IHD than in controls. However, the report is difficult to evaluate because the experimental methods, selection criteria, and age range of the controls are not adequately described.17

The present study did, however, indicate that biochemical vitamin B6 deficiency is very common in the age range studied. In all, 31% of the study population had levels below 6 ng/ml. The latter cut-off point is close to the 5 ng/ml level obtained from metabolic depletion-repletion studies using methodology for vitamin B6 measurement similar to ours (H. E. Sauberlich — personal communication). The overall mean plasma PLP level of 8 ng/ml in our study is fairly close to the 7 ng/ml found by Hamfeld18 for subjects of this age. It would seem that vitamin B6 deficiency is an inevitable consequence of westernized eating habits. This conclusion is strengthened by the finding that 47% of males and 82% of females aged 15 - 64 years in the 1979 CORIS study had dietary intakes of vitamin B6 below the recommended dietary allowance (RDA) (unpublished data). Similarly, 62 - 80% of coloured people in the Cape Peninsula and 96 - 100% of institutionalized white elderly subjects have been found to have vitamin B6 intakes below the RDA in various studies performed by the National Research Institute for Nutritional Diseases (NRIND). Even among nutritionally aware subjects, such as the staff of the NRIND, there was a high prevalence of suboptimal intakes (31%), and this was also the case among 28 - 66% of vegetarians. High prevalences of apparently inadequate dietary vitamin B6 intakes have also been found elsewhere.19

It has been suggested that vitamin B6 status should be judged in relationship to protein intake20 or, more specifically, to methionine intake,4 since an excess of the latter may, according to the vitamin B6-homocysteine theory, have an adverse effect even in the presence of marginally adequate vitamin B6 status. It can be shown that lacto-ovo vegetarians can attain the suggested dietary ratio of vitamin B6/protein of 0.02 mg/g, but omnivores generally do not (M. Faber and E. Gouws — unpublished data). The vegetarians, however, often have a vitamin B6 intake below 1.25 mg/d, which may in itself be undesirable. Sauberlich (personal communication) has accumulated data showing that vitamin B6 requirements are not affected by whether dietary proteins are obtained from plant or animal sources. Nevertheless, it remains possible that a mainly meat-eating population has an underlying adverse vitamin B6/methionine ratio, which may predispose it to IHD.
However, vitamin B6 levels themselves, as judged on the basis of the present evidence, do not appear to play a primary role in the genesis of IHD.

It should be borne in mind that the present study population has been subject to lipid-lowering dietary intervention for 3 years before the vitamin B6 estimations were carried out, and a proportion of the IHD subjects had been informed of their status after the 1979 survey. Thus, dietary adaptations may have obscured any pre-existing differences in plasma PLP levels that were originally present. This, however, is unlikely in view of the generally low PLP levels in all three study groups.

Clearly, a prospective study would be required to confirm or deny the existence of a true association between IHD and vitamin B6 status. It would also be necessary to demonstrate that individuals with IHD have a lower functional vitamin B6 status, e.g. by measurement of vitamin B6-dependent enzyme systems or urinary excretion of tryptophan metabolites. Especially important would be the measurement of homocysteine levels themselves. In this regard heterozygotic relatives of homocysteinuric patients generally do not have elevated fasting plasma homocysteine levels in spite of having markedly depressed cystathionine synthetase activity. Therefore, the demonstration of low plasma PLP levels may not necessarily imply impairment of homocysteine metabolism. Finally, it must be shown that treatment with vitamin B6 improves the outcome in IHD before a causal relationship can be entertained.

What facilitatory role, if any, vitamin B6 plays in conjunction with individual susceptibility must be investigated further before the vitamin can be considered a worthwhile additional modality for the prevention or treatment of IHD. Even if the vitamin B6-homocysteine theory should be shown to have causal significance, the logical approach would be to moderate methionine (animal protein) intake and increase dietary vitamin B6 intake in the form of unrefined cereals, pulses, nuts, vegetables and fruits. Except for not emphasizing the importance of a low fat intake, this dietary advice does not differ substantially from that conventionally given for the prevention of IHD.

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16. Canham JE, Baker EM, Sauberlich HE, Plough le. Dietary protein intake in the form of unrefined cereals, pulses, nuts, vegetables and fruits. Except for not emphasizing the importance of a low fat intake, this dietary advice does not differ substantially from that conventionally given for the prevention of IHD.

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**Nuus en Kommentaar/News and Comment**

**Sensitive to your own hormones**

A vast number of external stimuli can cause anaphylaxis but it is extremely uncommon to pinpoint an internal stimulus triggering off the attack. Where no external cause has been discovered on investigation, the patient is usually labelled as suffering from recurrent idiopathic anaphylaxis. The possibility that there is a specific cause in these cases is raised by a report of a 36-year-old woman with a lifelong history of seasonal allergic rhinitis and asthma, previous anaphylactic reactions to penicillin and streptomycin, and a strong family history of allergy (Meggs et al., N Engl J Med 1984; 311: 1236). At the age of 33 years she began to experience acute anaphylactic attacks every 5-10 days, frequently requiring emergency treatment for hypotension, laryngeal oedema and asthma. The attacks were reduced in severity but not prevented by long-term management with antihistamine drugs. In the end, a permanent tracheal fenestration was created because of her attacks of laryngeal oedema.

The first clue to the cause of her attacks occurred when she became pregnant and her attack rate increased during the second and third trimesters, but the episodes ceased altogether during the period of lactation. Two weeks after she had stopped lactating, the attacks resumed. It then became clear that a hormonal factor was involved.

When the patient was challenged with luteinizing hormone-releasing hormone (LH-RH) the frequency of attacks increased and she was therefore started on therapy with the LH-RH analogue which suppresses pituitary response to LH-RH. This effectively reduced gonadotrophin and progesterone levels and the attacks ceased. A provocation skin test with progesterone was followed by profound anaphylaxis.

Some months later the patient developed urinary incontinence and she was operated upon for a uterine prolapse. After hysterectomy and oophorectomy had been performed, the patient became entirely free of anaphylactic attacks without any treatment at all. Incidentally, it was noted that the patient had previously received parenteral progesterone during pregnancy, and this may well have been a sensitizing factor.