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ADDENDUM

Following this symposium and the writing of this article, a second monogenic cause of hypertension has been elucidated with the description of the molecular basis of Liddle's syndrome. Liddle's syndrome is a further 'rare' cause of endocrine hypertension characterised by sodium retention, but with low renin and low aldosterone levels.[1] Hypokalaemia may occur but is by no means an invariable finding. The phenotype results because of a mutation in the carboxy-terminal domain of the $\beta$-subunit of the human epithelial sodium channel, which, for reasons which are still obscure, results in constitutive activation of the apical sodium channel, sodium reabsorption and subsequent hypertension.[2]

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# LESSONS FROM A SYMPOSIUM ON FIBRINOGEN AND CARDIOVASCULAR DISEASE HELD IN THE COLLEGE ON 1–2 NOVEMBER 1994

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At the First International Symposium on Fibrinogen and Cardiovascular Disease held in Vienna, 1992, the importance of plasma fibrinogen as a cardiovascular risk factor was established. The multi-disciplinary and international audience at this Second International Symposium consisted of many pioneers in this area of research, together with others who had gained an interest and were keen to learn of recent developments. During the two days, there were sessions on: progress on the measurement of plasma fibrinogen; aspects of the pathogenic role of high fibrinogen levels; the epidemiology of fibrinogen in relation to risk factors and genotype; fibrinogen and ischaemic heart disease, peripheral arterial disease and cerebrovascular disease; and the therapeutic lowering of raised plasma fibrinogen levels.

*Biochemistry and measurement*

The term fibrinogen covers a large family of closely related molecules, which are heterogeneous in each of their three chains: A$\alpha$, B$\beta$ and $\gamma$. Methods for the determination of fibrinogen can be divided into two groups; those that measure clottable fibrinogen (for example, the widely used Clauss method) and those that quantify the fibrinogen molecule in other ways (for example, by heat precipitation or immunologically). Due to the biological variability of fibrinogen (both within and between individuals) and the different assays used by different studies, there was much confusion in recent years as to what the population reference range of fibrinogen should be. In 1991, work was initiated on the formulation of a standard by a scientific sub-committee of the International Society on Thrombosis and Haemostasis. The international collaborative study involving 22 laboratories has allowed the establishment of the 1st International Standard for plasma fibrinogen (89/644). This standard was reported to be a valuable and reliable calibrator for the measurement of plasma fibrinogen over a wide concentration level in clotting assays.

*Fibrinogen and pathogenesis of arterial disease*

There are a number of mechanisms whereby high fibrinogen levels are likely to predispose to atherosclerosis and thrombosis, although most are not fully understood. After vascular injury, the coagulation system acts to secure haemostasis by the production of thrombin and thus the conversion of soluble fibrinogen to insoluble fibrin forming a stable clot. In addition, fibrinogen is an important cofactor in platelet aggregation and also a major determinant of blood and plasma viscosity. Fibrinogen is present in the normal arterial intima and in atherosclerotic lesions. In early proliferative (gelatinous) lesions, fibrin is also present and provides the scaffolding along which smooth muscle cells migrate and proliferate, binds thrombin and lipoproteins; and is a source of fibrin

degradation products which have atherogenic effects. Recent studies have suggested that free α-thrombin may be present in abnormal intima, perhaps promoting fibrin formation within the lesion. In addition, as an acute-phase protein reactant, synthesis of fibrinogen is increased following injury, surgery and acute infections. This is probably due to hepatocyte stimulation by fibrin degradation products and/or by cytokines released from activated monocytes in damaged tissues.

*Genetic influences on fibrinogen*

Several reports have suggested that individuals with otherwise similar cardiovascular risk profiles may have substantially different plasma fibrinogen levels. There is considerable evidence to suggest that individual responses to the environmental factors that influence plasma fibrinogen levels (for example, smoking) are genetically determined, although the extent of this influence has yet to be established. Estimates of the degree of genetic regulation vary from around 30% in studies of twins, up to about 50% in family studies.

Because synthesis of the Bβ chain is the rate-limiting step in the formation of fibrinogen, mutations affecting the production of this chain would be most likely to influence fibrinogen levels and hence most genetic studies have focused on this area. Several restriction fragment length polymorphisms linked to the β-fibrinogen gene have been identified and their allelic frequencies have been related to fibrinogen levels. Results have, however, been inconsistent. Other genetic mutations affecting the γ and α-genotypes may also be of clinical significance. While it is difficult to identify the size of the contribution that the fibrinogen genotype contributes to the risk profile for cardiovascular disease (due to the interactions between genetic and environmental risk factors), this was highlighted as an area of growing importance.

*Epidemiology of fibrinogen*

Since there is increasing evidence to indicate fibrinogen as a risk factor for cardiovascular disease, it is important to define which factors determine plasma fibrinogen levels. In published epidemiological studies, there is a consensus that plasma fibrinogen levels increase with age and are higher in women than in men (men however appear more susceptible to the cardiovascular risk associated with increasing fibrinogen levels). Smoking has the strongest influence showing a positive dose-response effect with the number of cigarettes smoked and also a consistent decline on cessation of smoking (although fibrinogen levels take 5–10 years to return to those of a life-long non-smoker). It is not clear what constituents of tobacco smoke increase plasma fibrinogen level, but the results of one presented paper suggested that the detrimental effect of smoking on fibrinogen was *not* mediated by circulating nicotine.

Among women, fibrinogen levels are higher after the menopause. Users of hormone replacement therapy have lower levels than non-users, whereas oral contraceptive use raises fibrinogen levels. Obesity is related to higher fibrinogen levels, whilst physical activity and exercise training tend to be associated with lower levels. Alcohol consumption and increased fish oil intake are associated with lower levels, whilst macronutrients such as dietary fat appear to have no effect. Other factors that have been reported to be positively associated with fibrinogen include low density lipoprotein cholesterol, triglycerides, leucocyte



count, fasting insulin and low birth weight. In contrast, high density lipoprotein cholesterol and social class have both shown negative correlations. Several studies have suggested that fibrinogen levels vary by season, and some have speculated that this may reflect a response to the seasonal variation in respiratory infections. However, results from a Dutch population study suggested that this seemed an unlikely explanation and that physical activity and dietary habits may have a role. Fibrinogen levels are higher in patients with essential hypertension than in normotensive controls. Diabetics also show raised levels and correlations between blood sugar level and fibrinogen have been reported. In conclusion, plasma fibrinogen seems to correlate with all of the common cardiovascular risk factors in a non-specific way.

*Fibrinogen and ischaemic heart disease*

An increasing number of prospective studies have consistently shown a direct, independent and statistically significant association between fibrinogen level and the subsequent incidence of ischaemic heart disease. In some studies, this association was at least as strong as that between cholesterol and ischaemic heart disease. For example, in the sixteen year follow-up of the Northwick Park Heart Study, a one standard deviation increase in fibrinogen (about 0·6 g/l) was associated with an increase in incidence of ischaemic heart disease of about 40% (similar to that for a one standard deviation difference in cholesterol). A recent meta-analysis of six epidemiolgical studies covering a total of 92,147 person years showed a continuous increase in risk of cardiovascular events from the lowest to the highest tertile of plasma fibrinogen, with the summary odds ratio of all studies being 2·3 (95% confidence interval 1·9 to 2·8). Considering the diversity of study designs, samples selected, fibrinogen assays, follow-up periods and end-point criteria, the results were very uniform. At this meeting, similar results were reported from the 8-year follow-up of 8,800 men and women in the Scottish Heart Health Study. A report from the Glasgow MONICA Study also defined the cut-off between middle and upper tertiles in middle-aged men and women as 3·0 g/l, using the Clauss assay and International Fibrinogen Standard.

These prospective studies have been supplemented by similar findings in prevalence and cross-sectional studies, although these have the limitation of being unable to distinguish between cause and consequence. Several angiographic studies have shown a correlation between fibrinogen level and the extent of coronary artery disease, with the relationship perhaps due to luminal occlusion as well as vessel wall involvement. In addition, fibrinogen levels remain elevated many years after a myocardial infarction, and are also predictive of recurrent disease in those who have previously survived an infarction.

*Fibrinogen and peripheral arterial disease*

The relationship between plasma fibrinogen and peripheral arterial disease has not been studied as extensively as that between fibrinogen and ischaemic heart disease. Several small case-control studies have examined hospital patients with peripheral arterial disease and have shown higher levels in these patients compared to controls. In cross-sectional studies, higher fibrinogen levels were noted in claudicants (the Scottish Heart Health Study and the Edinburgh Artery Study) and in patients with major asymptomatic disease (the Edinburgh Artery Study) compared to normals. However, there is little data relating plasma fibrinogen to

incident cases of peripheral vascular disease, although both the Speedwell and the Framingham studies have noted an effect.

Using the ankle brachial pressure index (ABPI) as an indicator of the degree of peripheral atherosclerosis, the association of plasma fibrinogen with the ABPI seems to be stronger in men than in women. This may provide clues about the increased susceptibility of men to atherosclerotic disease. Cigarette smoking appears to be a more important risk factor for peripheral arterial disease than for ischaemic heart disease, although the relationship between plasma fibrinogen and peripheral arterial disease was reported to be largely independent of smoking. Finally, plasma fibrinogen has been found to be an independent predictor of coronary deaths among claudicants and may also predict treatment outcome (for example, the patency of infra-inguinal grafts).

*Fibrinogen and cerebrovascular disease*

Reports from two large prospective studies (the Gothenburg Study and the Framingham Study) have identified the independence of plasma fibrinogen as a risk factor for cerebrovascular disease in men. Although several case-control studies report elevated levels among stroke patients, their interpretation is limited by the well-documented reactive elevation of fibrinogen within hours of the stroke, after which the level gradually normalises. Consequently, this association has been attributed to an acute-phase reaction due to brain tissue necrosis. However, fibrinogen levels have been shown to be significantly elevated in patients who experience transient ischaemic attacks and minor ischaemic strokes, suggesting that fibrinogen may have some other releationship with cerebrovascular events. A further point to note is that 85% of strokes arise from cerebral infarction (occlusive stroke) and 15% from haemorrhage. Since it is difficult to differentiate the two groups based on clinical signs and symptoms, many studies have failed to accurately define the distribution of stroke types in their population. If the relationship of fibrinogen varies between the two types of stroke, then this would have obvious consequences for intervention. As with coronary and peripheral arterial disease, plausible mechanisms by which high fibrinogen levels increase the risk of cerebrovascular events include: thrombosis through a hypercoagulable state; the acceleration of atherosclerosis; and the reduction of blood flow via rheological effects.

*Therapeutic reduction of fibrinogen*

The ultimate test of whether fibrinogen is a cause, rather than a consequence, of cardiovascular disease would be a randomised controlled trial to therapeutically lower fibrinogen levels in patients at risk, and to determine the subsequent cardiovascular outcome. Such a trial would need a drug that reduces fibrinogen levels safely and selectively. Unfortunately, no such drug is available at present. However, since the overviews and presentations during the symposium strongly suggest that the association between fibrinogen and cardiovascular disease is, in fact, causal, then the therapeutic lowering of fibrinogen becomes an important issue. Of all the life-style changes that would affect fibrinogen level, smoking cessation is by far the most effective modification, whilst weight reduction or an increase in exercise may have less pronounced effects.

The range of medications that have been shown to reduce plasma fibrinogen levels is wide, and such drugs are generally marketed for other cardiovascular



actions (for example, to lower lipids). Among the fibrate group of lipid lowering drugs, clofibrate, bezafibrate, fenofibrate and ciprofibrate have all been shown to lower fibrinogen levels. Bezafibrate has been demonstrated to lower fibrinogen by 25% on average and has the most consistent effect across several different trials. Possible mechanisms that may explain this effect of fibrates are an interaction with apolipoprotein A, and a direct inhibition of the hepatic fibrinogen synthesis (possibly via effects on cytokines). Platelet inhibitors (such as ticopidine), $\beta$-adrenergic-receptor blockers, pentoxifylline and other vasodilators, calcium dobesilate and various other drugs have all, in varying degrees, been claimed to lower fibrinogen levels. Interpretation of such trials is difficult, because the drugs were given to treat a defined disease or symptom, rather than to lower fibrinogen levels, and the observed reduction in fibrinogen may thus be confounded by the clinical effect of the therapy. Further research is needed concerning the physiological control of fibrinogen levels and the mechanisms by which therapeutic intervention might decrease them.

In conclusion, at the present time, there is no sound evidence on which to base the prescription of drugs for the *sole* purpose of reducing fibrinogen. However, two large, ongoing randomised controlled trials of bezafibrate should soon establish whether or not fibrinogen reduction is effective in secondary prevention of cardiovascular events in survivors of myocardial infarction and in patients with claudication.

REFERENCE

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