Determinants of plasma lipoproteins and coagulation factors in men from Caerphilly, South Wales

J W G YARNELL,¹ ANN M FEHILY,¹ JANET MILBANK,^{1*} A J KUBICKI,^{1†} R EASTHAM,⁴ AND T M HAYES³

From the MRC Epidemiology Unit,¹ Cardiff, Frenchay Hospital,² Bristol, and the Department of Medicine,³ University Hospital of Wales, Cardiff, UK

SUMMARY Dietary, social, and constitutional determinants of plasma concentrations of some major risk factors for ischaemic heart disease were investigated in a cross sectional study among 711 men from the general population aged 30–69. For high density lipoprotein cholesterol (HDL-C) six variables were significantly associated with plasma concentrations, and these variables explained 12·6% of the variance. For low density lipoprotein cholesterol (LDL-C), total cholesterol (T-C), and triglyceride 8·4%, 7·5%, and 18·5% of the variance was explained by significantly associated variables. Fibrinogen concentrations determined chemically were significantly associated with age, smoking habit, body mass index, alcohol consumption, and intake of cereal fibre (24·2% variance). These data provide some encouragement for the possibility of dietary intervention to influence plasma concentrations of major risk factors for ischaemic heart disease in men.

High density lipoprotein cholesterol (HDL-C) has been rediscovered recently to be an important risk factor for ischaemic heart disease (IHD).^{1–3} Interest has focused also on coagulation factors⁴ in the aetiology of IHD. Little is known about the major determinants of these factors in plasma; the purpose of the present paper is to examine these in a prevalence study among a random sample of 711 men aged 30–69. Similar data for over 2000 men aged 45–59 will be available shortly.

Methods

STUDY POPULATION

A random sample of men was drawn from the electoral registers for the town of Caerphilly and two outlying villages (population 38 000) in South Wales. All men were sent an initial letter but were only considered eligible for inclusion in the study if aged 30–69; a total of 797 men came into this category and were invited to attend a local clinic.

SURVEY METHODS

Details of the medical history, physical measurements, and nutritional questionnaire⁵ have

been described in detail previously⁶ in a similar study among women.

LABORATORY MEASUREMENTS

A venous blood sample was taken with minimum stasis after an overnight fast (minimum eight hours, average 12 hours). HDL-C was estimated in supernatant by enzymatic assay⁷ after precipitation of the remaining lipoprotein fractions, usually within six hours by the use of sodium heparin/Mn Cl₂ (46mM).⁸ Total cholesterol (T-C) was estimated on EDTA plasma by the same enzymatic assay⁷ (Boehringer). Low density lipoprotein cholesterol (LDL-C) was calculated from the formula of Friedewald *et al*⁹

$$(\text{LDL-C} = \text{T-C} - \left[\text{HDL-C} - \frac{(\text{TG} \times 18)}{38.7} \right])$$

(mmols). Triglycerides were estimated by enzymatic assay.¹⁰ Fibrinogen was estimated by two methods; a chemical method¹¹ on EDTA plasma and a clotting assay¹² on citrated plasma. Viscosity was estimated on EDTA plasma by the method of Thorp *et al*¹³ and haematocrit by a standard capillary tube

* Present address: John Radcliffe Hospital, Oxford.

† Present address: Department of Clinical Investigation, Diabetic Department, Dudley Road Hospital, Birmingham.

method. Factor VII assay¹⁴ was carried out on citrated plasma using plasma derived from beagles. For convenience, clotting fibrinogen (fibrinogen clotting time) has been converted to similar units to those obtained biochemically; but in the case of factor VII assays a shorter clotting time indicates a higher level of factor VII activity.

Quality control was maintained during the course of the study by the use of pooled plasma standards where appropriate, duplicate samples presented "blindly" to the laboratory and second samples taken from a small group of men some four to 12 weeks after the first sample. In the case of the "split" samples the coefficients of variation for T-C, HDL-C, triglycerides, fibrinogen (chemical), fibrinogen (clotting), viscosity, and factor VII were 4.0%, 13.3%, 4.8%, 5.3%, 10.0%, 2.3%, and 3.1% respectively. For the second samples the coefficients of variation were 6.9%, 18.1%, 22.6%, 10.3%, 17.9%, 2.4%, and 4.1%.

STATISTICAL METHODS

Univariate analyses were done initially to determine which variables were entered into a multiple linear regression by a stepwise (forward inclusion) procedure. The dependent variable that explained the greatest proportion of the variance in the independent variable was entered initially; the variable that explained the greatest percentage of the remaining variance was entered next; and so on, so that each additional variable yielded the maximum increase in the proportion of the total variance explained.

Results

Of 797 men eligible for inclusion in the study, 711 (89%) were examined and 700 (88%) provided a blood sample. Dietary questionnaires were returned by 697 men (87%). Because of missing information it was not possible to obtain complete regression analyses on all members of the study sample; however, non-respondents had a similar level of morbidity to those subjects with complete information.

Table 1 lists the variables that remained significantly associated with the lipid variables after completion of the regression analyses on 670 men with complete information. LDL-C and triglyceride values were transformed because their distributions were skewed; similar variables were associated with the untransformed data.

In the case of HDL-C a total of 12.6% of the variance was explained by the significantly associated variables. An additional 1.0% of the variance was

Table 1 *Accumulative percentage of variance in plasma concentrations of lipids in 670 men, which is explained by age, smoking habit, etc*

	Accumulative percentage of variance explained	Direction of association
HDL-Cholesterol:		
	per cent	
Body mass index (Quetelet's)	5.8	-
Smoking habit	8.7	-
Alcohol	10.1	+
Age	11.2	+
Longevity	12.0	+
Vitamin C	12.6	+
LDL-Cholesterol:*		
Age	2.1	+
Milk	3.7	+
Animal protein	5.7	-
Longevity	7.2	-
Smoking habit	8.4	+
Total cholesterol:		
Age	3.3	+
Longevity	6.4	-
Milk	7.5	+
Triglyceride:*		
Body mass index	10.7	+
Alcohol	14.3	+
Smoking habit	16.0	+
Starch (%calories)	16.8	+
Age	17.7	+
Cereal fibre	18.5	-

*Values transformed to logarithms.

explained by including four more non-significantly associated variables. For LDL-C 8.4% of the variance was explained by significantly associated variables—age, milk consumption, animal protein, longevity, and smoking habit. T-C had only 7.5% of its variance explained by significantly associated variables; an additional 0.5% of the variance was explained by including an additional non-significantly associated variable (Quetelet's index). A total of 18.5% of the variance in plasma concentrations of triglyceride was explained by significantly associated variables.

Table 2 shows the accumulative percentage of the variance explained by variables significantly associated with plasma concentrations of certain coagulation factors in 658 men with complete results. Chemical fibrinogen was associated principally with age and smoking habit but body mass index, alcohol consumption, and cereal fibre contributed a further 2.9% of the variance. Clotting fibrinogen was significantly associated only with age and smoking habit. Viscosity was similarly associated principally with age, body mass index, and smoking habit. Factor VII levels were significantly, and only weakly, associated with age; but an additional 2.3% of the variance was explained by including four non-significantly associated variables.

*Some second samples were non-fasting.

Table 2 Accumulative percentage of the variance in plasma concentrations of coagulation factors in 658 men, which is explained by age, smoking habit, etc

	Accumulative percentage of variance explained	Direction of association
Fibrinogen, chemical:*		
	per cent	
Age	12.8	+
Smoking habit	21.3	+
Body mass index	22.8	+
Alcohol	23.5	-
Cereal fibre	24.2	-
Fibrinogen, clotting:*		
Age	10.2	+
Smoking habit	15.4	+
Viscosity:*		
Age	5.7	+
Body mass index (Quetelet's)	7.5	+
Smoking habit	9.3	+
Vegetable oil	10.4	-
Factor VII:*		
Age	0.9	-

*Values transformed to logarithms.

Discussion

This study shows that dietary factors can be associated with major plasma lipid components within the same population. In the case of HDL-C, however, some caution should be exercised in the interpretation of the results since different dietary factors have been reported by members of our group to be associated with plasma HDL-C in women⁶ and in a further sample of 117 men¹⁵ aged 44–60. In the present study a further plasma sample was assayed for lipoproteins in a separate laboratory (Dr C Bolton, Bristol) and slightly different results were obtained for HDL-C in which 5.3% of the variance is explained by four variables; alcohol (+ve), smoking habit (-ve), vegetable fibre (+ve), and age (+ve). A common finding, however, in this population is a slight increase in HDL-C with age.

LDL-C, T-C, and triglycerides show a larger degree of consistency in results obtained from the two laboratories. LDL-C (which was calculated by means of a direct result from VLDL-C and HDL-C in the case of the Bristol laboratory) is consistently associated with age, milk consumption, longevity, and smoking habit. T-C is consistently associated with age, longevity, and milk consumption. Triglycerides had a high percentage of the variance explained by similar factors for results from both laboratories (18.5 and 20.3% respectively). Consistent associations, all of which were positive, were shown for body mass index, alcohol consumption, smoking habit, and age. Although internal consistency within the data provides a certain measure of reassurance, these data are based on multiple comparisons of relatively small numbers

of subjects. Analysis of larger samples should provide more secure data.

Chemical and clotting fibrinogen levels have been reported to be associated with prevalent IHD¹⁶ and clotting fibrinogen with incident IHD.⁴ In the present study chemical and clotting methods were done independently in separate laboratories. Chemical fibrinogen was associated with age and cereal fibre intake in a further sample of 117 men¹⁵ aged 44–60 for whom detailed seven day weighted dietary records were available. In these present data consistent associations were noted between fibrinogen concentrations and age, smoking habit, body mass index, alcohol consumption, and intake of cereal fibre although in the case of clotting fibrinogen only age and smoking habit were statistically significant. Similar findings have been noted in another cross sectional study,¹⁷ but in this study dietary factors other than alcohol consumption were not apparently investigated. The mechanism by which an acute phase reactant such as fibrinogen¹⁸ can be influenced by intake of cereal fibre currently remains obscure, but the association would seem to warrant testing by means of an intervention study.

In these data chemical fibrinogen concentrations and plasma viscosity are highly correlated ($r = 0.60$). In a multiple logistic regression analysis¹⁹ viscosity was not independently associated with prevalent IHD, its apparent univariate association being entirely explained by fibrinogen.

Although certain of these data such as the dietary determinants of HDL-C must be regarded as provisional because of inconsistent results, they provide some encouragement for the possibility of dietary interventions to reduce levels of the major plasma risk factors.

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