The Tromsø Heart Study: Serum selenium and risk of myocardial infarction a nested case-control study

JETMUND RINGSTAD,¹ BJARNE K JACOBSEN,¹ YNGVAR THOMASSEN,² AND DAG S THELLE¹

From the Institute of Community Medicine,¹ University of Tromsø, Norway and the Institute of Occupational Health,² Oslo, Norway

SUMMARY The association between serum selenium concentration and the risk of myocardial infarction was studied in a nested case-control study. Altogether 59 men, initially free of disease, aged 28–54 at the time of blood sampling, died suddenly or experienced a fatal or non-fatal myocardial infarction during a six year follow-up period. Case-control pairs came from a population of 9364 persons examined in 1979–80 in the second Tromsø Heart Study. No significant difference was observed between serum selenium in cases and controls ($p=0.34$). The major determinants of myocardial infarction and sudden death were raised levels of serum cholesterol and triglycerides ($p\leq 0.001$) and high systolic blood pressure ($p<0.05$). Thus, in this population with intermediate selenium intake, low serum selenium is not associated with an excess risk of myocardial infarction.

A number of reports suggest that the pathogenesis of cardiac ischaemic injury may be partly ascribed to the production of activated oxygen species.^{1–3} Therefore, much interest has been focused on the trace element selenium, which is an essential prosthetic group for the enzyme glutathione peroxidase.⁴ This enzyme reduces lipid hydroperoxides (or hydrogen peroxide) to the corresponding lipid alcohols (or water), thereby possibly protecting the epithelium from oxidative damage.⁵ Glutathione peroxidase may also influence the synthesis of prostacyclin, an agent protective against platelet aggregation.⁶

The relationship between serum selenium concentration and the risk of ischaemic heart disease has not been convincingly proved. Between-area comparisons,^{7,8} case-control studies,^{9,10} and clinical observations¹¹ have produced some support for the role of selenium in coronary heart disease. Among the published prospective studies on selenium and the risk of death from ischaemic heart disease, one study found an inverse association,¹² two studies were equivocal but suggested an inverse association among persons free of previous ischaemic heart disease,^{13,14} and in two studies no association was observed.^{15,16} These different findings have recently been discussed by Salonen.¹⁷

The present study was carried out to test the hypothesis that serum selenium is linked to the risk of myocardial infarction.

Materials and methods

For the present analysis we used data from the second Tromsø Heart Study in 1979–80, a population survey of cardiovascular risk factors. All men aged 20–54 and women 20–49, residents of Tromsø municipality (21 329 subjects), were invited; 16 621 (78%) participated. The examination comprised administration of a questionnaire identical with that used in the first Tromsø Heart Study¹⁸ and the cardiovascular studies in the Norwegian counties,¹⁹ measurement of blood pressure, height, and weight, and a collection of non-fasting blood samples for measurement of total serum cholesterol, HDL-cholesterol, triglycerides, and glucose. At the examination an extra blood sample was taken from the first 9364 subjects who attended the screening. These serum samples were frozen in closed plastic tubes at -20°C for later analysis.

Persons who in the 1979–80 survey reported a history of myocardial infarction or chest pain previous to the examination were excluded from this study. During the subsequent six years (until 31 December 1985), 65 men, free of myocardial infarction or self-reported angina pectoris at screening, died suddenly of unknown cause ($n=8$) or experienced a fatal or non-fatal myocardial infarction ($n=57$). Nearly all sudden deaths of middle-aged men in Norway are due to coronary heart disease.²⁰ No cases were found among the women.

Data about acute myocardial infarction were obtained from the only hospital in the municipality, and data on deaths from myocardial infarction were derived from the Central Bureau of Statistics, Oslo, which registers all deaths in the country. There may, therefore, be non-fatal cases of myocardial infarction not registered in this study. Furthermore, only serum from 59 persons could be obtained, as serum samples from six myocardial infarction cases had been used for other non-selenium-related research purposes. One healthy control was matched pairwise to each of these cases according to sex, age (usually within the same year), daily consumption of cigarettes (non-smoker, 1-10, 11-20, 21-30, > 30 cigarettes per day), month of blood sampling, and place of residence (one of four parts of the municipality). One case was a pipe-smoker. As no appropriate pipe-smoking control was available, he was matched with a man who smoked 1-10 cigarettes per day.

Both the cases and controls were free of present and previous cancer as assessed by crosslinking the information from the Cancer Registry of Norway.

The presented values for total cholesterol, HDL-cholesterol, and blood pressure are the results from the 1979-80 survey of the Tromsø Heart Study. Total cholesterol was measured directly by the enzymatic oxidase method, using a commercial kit (Boehringer 148393). HDL-cholesterol was assayed by the same procedure after precipitation of lower density lipoproteins with heparin and manganese chloride. All of these laboratory assessments were performed shortly after the screening by the Division of Clinical Chemistry, University Teaching Hospital of Tromsø.

The selenium analyses were performed at the Institute of Occupational Health in Oslo in October 1986, when the serum samples were thawed for the first time. Selenium concentrations were determined by electrothermal atomic absorption spectroscopy after dilution with a nickel matrix modifier as proposed by Saeed *et al.*²¹ All analyses were done in duplicate and

in a random order, and calibration was done against standards based on human serum with a known content of selenium.²¹ Quality assurance of the selenium determinations was ensured using a certified reference serum as a quality control material.²² The coefficient of variation for the reference serum during one day was 2.8%, and day-to-day variation based on the same serum sample was 5.9%. The mean concentration (and standard deviation) of selenium in the reference serum for all days was 1.17 (0.07) $\mu\text{mol/l}$, while the proposed concentration is 1.15 (0.08) $\mu\text{mol/l}$.²³

Statistical analyses were performed with the Statistical Package for the Social Sciences (SPSS).²⁴ The mean differences between cases and controls were tested for statistical significance with a T-test for paired samples.

Results

Table 1 shows the distribution of serum selenium concentration in cases and controls. The mean concentration of serum selenium (and standard deviation) was 1.57 (0.21) $\mu\text{mol/l}$ for all cases and 1.61 (0.27) $\mu\text{mol/l}$ for all controls. This difference (0.04 $\mu\text{mol/l}$) is not statistically different from zero ($p = 0.34$) (table 2). There were two particularly high values for serum selenium (2.61 and 2.68 $\mu\text{mol/l}$), both in controls. If the two pairs with these controls were

Table 1 Distribution of serum selenium in cases and controls. The Tromsø Heart Study, 1979-80

Serum selenium ($\mu\text{mol/l}$)	Cases (N)	Controls (N)
≤ 1.29	5	6
1.30-1.44	9	7
1.45-1.59	21	12
1.60-1.74	13	22
1.75-1.90	7	8
≥ 1.90	4	4

Table 2 Mean difference (and standard deviation) in serum selenium, number of cigarettes per day, age at examination, total cholesterol, HDL-cholesterol, triglycerides, blood pressure, and body mass index in 59 matched pairs of myocardial infarction cases and controls. The Tromsø Heart Study, 1979-80

	Cases	Controls	Difference	p-value
Serum selenium ($\mu\text{mol/l}$)	1.57	1.61	0.04 (0.35)	0.34
No. of cigarettes/day*	10.7	11.4	0.7 (3.4)	0.12
Age at examination*	46.3	46.3	0.0 (0.4)	0.74
Cholesterol (mmol/l)	7.55	6.50	1.05 (1.78)	<0.001
HDL-cholesterol (mmol/l)	1.41	1.51	0.10 (0.78)	0.33
Triglycerides (mmol/l)	2.31	1.70	0.61 (1.38)	0.001
Blood pressure (mmHg)				
Systolic	141.2	134.7	6.5 (24.2)	0.04
Diastolic	89.5	86.3	3.2 (16.5)	0.14
Body mass index	25.6	25.2	0.4 (4.1)	0.43

* Matching variables

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excluded from the analyses, the mean serum selenium was 1.57 and 1.58 $\mu\text{mol/l}$ in cases and controls, respectively. The selenium levels of the 21 fatal cases (1.62 $\mu\text{mol/l}$) were not significantly different from their controls (1.56 $\mu\text{mol/l}$).

We also tested whether low (in this study) serum selenium levels were associated with an increased risk of myocardial infarction. However, serum selenium levels below 1.45 $\mu\text{mol/l}$ were not associated with a risk of myocardial infarction (RR=1.0, 95% CI=0.4–2.3).

As cases and controls were matched on age and smoking, they had comparable means for age and number of cigarettes smoked daily (table 2). Of the 59 men 48 were daily smokers.

Table 2 shows the major cardiovascular risk factors in cases and controls. The differences between cases and controls were highly significant for cholesterol (1.05 mmol/l, $p<0.001$) and triglycerides (0.61 mmol/l, $p=0.001$). Also systolic blood pressure was significantly higher in cases (6.5 mmHg, $p=0.04$). HDL-cholesterol was lower in cases (0.10 mmol/l) but did not reach statistical significance.

Table 3 shows the distribution of serum selenium in cases and controls according to when the myocardial infarction occurred. The difference in serum selenium between cases and controls was not statistically significant in any period of follow-up. If the two above mentioned pairs are excluded from the analyses, the difference in serum selenium between cases and controls in all the three periods of follow-up was $\leq 0.01 \mu\text{mol/l}$.

Discussion

In this matched-pair, six-year follow-up study of men initially free of coronary heart disease, there were no differences in serum selenium concentration between cases and controls. The mean serum selenium values observed in this study are comparable with those in other Norwegian studies^{25,26} but are well above selenium levels in Finland (range 0.24–1.72 $\mu\text{mol/l}$)¹³ where the other prospective studies have been conducted.^{12–15} Salonen *et al* have proposed that a

threshold value may exist in the relation of serum selenium to ischaemic heart disease.¹² This may explain why no association was observed in this study with higher mean serum selenium levels.

We found higher serum cholesterol, triglycerides, and blood pressure in cases than in controls. One should note that these associations are univariate, and most studies have shown that the association between serum triglycerides and the risk of ischaemic heart disease disappears when adjusted for serum cholesterol.²⁷ The difference in HDL-cholesterol between cases and controls in this study is significantly less ($p<0.05$) than that found in the first Tromsø Heart Study.²⁸ One possible reason for this is the matching for smoking habits.

A bias could be introduced as we do not have information about all cases of myocardial infarction in the six year follow-up of the population screened in the second Tromsø Heart Study. However, we do not find it likely that the serum selenium concentrations of cases not available to us (due either to no available serum sample or to non-fatal myocardial infarction after migration to other parts of the country) differ from the serum selenium concentrations of cases included in this study. The follow-up of men screened in the first Tromsø Heart Study in 1974 suggests that relatively few non-fatal events took place outside Tromsø, and only one out of 60 cancer cases was registered outside Tromsø in a similar follow-up study (unpublished observations).

It may be argued that serum selenium measured in one blood sample does not reflect the selenium status over years, and, even if there were an association between serum selenium and the risk of myocardial infarction, one should not expect to find any. However, the mean serum selenium in Finland and New Zealand with a recognised low food intake of selenium is lower than the serum selenium in, for example, Norway where a higher intake has been recorded.²⁹ Furthermore, the repeated findings of a relation between low serum selenium and risk of myocardial infarction in Finland with a lower intake, suggests that serum selenium to some extent gives a valid picture of the intake.

Miettinen *et al*¹⁵ suggest that the relation between serum selenium below 0.57 $\mu\text{mol/l}$ and risk of myocardial infarction in Finland is due to a coexisting low intake of selenium and eicosapentaenoic acid. In our population, with a rather high fish consumption, it is reasonable to believe that the intake of this nutrient is also adequate.

In summary, this study suggests that in populations with an intermediate selenium intake like the Norwegians, a low serum selenium level is not associated with an excess risk of myocardial infarction. Thus, in Norway, efforts should still be

Table 3 Serum selenium (and standard deviation) ($\mu\text{mol/l}$) related to when the myocardial infarction occurred. Months after blood sampling. The Tromsø Heart Study, 1979–80

	Months after blood sampling		
	< 24	25–48	> 48
Cases	1.56 (0.21)	1.60 (0.16)	1.56 (0.24)
Controls	1.61 (0.29)	1.70 (0.37)	1.56 (0.17)
n of pairs	19	16	24
p-value for difference	0.49	0.40	0.95

directed towards reducing blood lipids and blood pressure and discouraging smoking.

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Correspondence and reprint requests to: Dr J Ringstad, Institute of Community Medicine, University of Tromsø, Box 417, N-9001, Tromsø, Norway.

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