An ecological study of determinants of coronary heart disease rates: a comparison of Czech, Bavarian and Israeli men

Martin Bobak,a Hans-Werner Hense,b Jeremy Kark,c Bernhard Kuch,b Petr Vojtisek,d Ronit Sinnreich, Johannes Gostomzyk,e Mai Bui,f Arnold von Eckardstein,g Ralf Junker,g Manfred Fobker,g Helmut Schulte,g Gerd Assmann§ and Michael Marmota

Background The large differences in cardiovascular disease rates between Eastern and Western Europe have largely developed over the last few decades, and are only partly explained by classical risk factors. This study was set up to identify other potential determinants of these differences.

Methods This was an ecological study comparing random samples of men aged 45–64 years selected from three cities representing populations with different rates of cardiovascular mortality: Pardubice (Czech Republic), Augsburg (Bavaria, Germany), and Jerusalem (Israel). In total, 191 (response rate 70%), 153 (70%) and 162 (62%) men, respectively, participated. All centres followed the same study protocol. Lifestyle, anthropometry and biochemical risk factors were assessed by identical questionnaires, standardized medical examination, and central analyses of fasting blood samples.

Results The mortality rates in the study populations, as well as the prevalence of coronary heart disease in study samples, were highest in Czech, intermediate in Bavarian and low in Israeli men. This pattern was replicated across the three samples by mean blood pressure ($P < 0.001$), cigarette smoking (not significant), triglycerides ($P < 0.05$), fibrinogen or D-dimer levels ($P < 0.05$). On the other hand, the prevalence of diabetes and obesity were similar; total and high density lipoprotein (HDL)-cholesterol, apolipoprotein B, lipoprotein (Lp(a)) and glucose did not differ between Czech and Bavarian men; and Czechs had particularly low levels of serum insulin and factor VIIc. Israelis had low fasting glucose and total cholesterol, as well as HDL-cholesterol levels and a high Lp(a) (each $P < 0.001$) compared with the two other samples. Striking differences were found for plasma homocysteine (10.5 in Czechs versus 8.9 µmol/l in Bavarians, $P < 0.001$) and for alpha-carotene (geometric mean in Czechs 16, Bavarians 21 and Israelis 30 µg/l), beta-carotene (60, 110 and 102 µg/l), and lycopene (84, 177 and 223 µg/l, respectively; all $P$-values $< 0.001$). Adjustment for obesity or smoking did not change these estimates. There were no differences in the levels of tocopherol and retinol.

Conclusions Czech men had high levels of blood pressure, triglycerides, fibrinogen and D-dimer but many other traditional risk factors, as well as indicators of metabolic disorders and vitamins A and E, did not differ between the study samples. The low levels of carotenoids and high concentrations of homocysteine in Czech men seem to reflect their low dietary intakes of fruit and vegetables. The results

---

"International Centre for Health and Society, Department of Epidemiology and Public Health, University College London, 1–19 Torrington Place, London WC1E 6BT, UK.

b Institute of Epidemiology and Social Medicine, University of Münster, Germany.

c Department of Social Medicine, Hadassah Medical Organisation and Hebrew University, Jerusalem, Israel.

d Department of Internal Medicine, City Hospital, Pardubice, Czech Republic.

e Gesundheitsamt der Stadt Augsburg, Germany.

f Institut Suisse des Vitamines, University of Lausanne, Switzerland.

g Institute of Arteriosclerosis Research, University of Münster, Germany."
The gap in life expectancy between Central and Eastern Europe and Western Europe is more than 6 years, and more than half of this difference is due to higher mortality rates from cardiovascular diseases, and particularly coronary heart disease (CHD) in middle ages. Despite the magnitude of the mortality gap and its implication for public health and policy, there has been little systematic research into this problem. Interestingly, the few reports available to date seem to suggest that population levels of classical risk factors play only a minor role in the explanation of between-population differences in CHD.

The development of the East-West gap has been paralleled by the trends in mortality in the Czech Republic and Germany. Mortality rates in these two neighbouring countries with similar ethnicity, history, culture, lifestyle, and dietary habits were comparable until the 1960s when they started to diverge. This study was set up to identify factors, or groups of factors, which may contribute to the gap in CHD. To do so, we compared the levels of a wide range of risk factors for cardiovascular disease in men in the Czech Republic and Bavaria, the German federal state bordering with the Czech Republic. Our objective was to identify characteristics of the populations, rather than of individuals, that offer potential explanations for the differences in CHD rates, and to generate hypotheses that could be tested in more sophisticated studies.

The Czech-Bavarian comparisons were complemented by a geographically and culturally distinct sample from the East Mediterranean recruited in Israel. The rationale for including Israel was, that, as it has low rates of CHD, it could serve as a useful contrast to Central Europe. The cities of Pardubice, Augsburg and Jerusalem were selected to represent the Czech Republic, Bavaria and Israel, respectively. Pardubice and Augsburg participated in the WHO MONICA project; the recently published results of a community-based monitoring of coronary events confirmed both the difference in morbidity rates and their representativeness for national rates.

This study focused on men in the age group 45–64 years, as it is middle-aged men where the differences in morbidity and mortality are the largest. The characteristics studies in this project included classical risk factors, an extended lipid profile, fasting insulin and glucose levels, coagulation factors, homocysteine, and vitamins with presumed antioxidant properties.

Methods
Age-stratified random samples of men aged 45–64 years, assigning equal sampling probabilities to each decade, were drawn from population registers in each city. In Pardubice, where new access to the population register was denied in 1995, we invited a random subset of men (both participants and non-participants) selected for the third Czech MONICA survey in 1992 by sampling strategy identical to that in the other two cities described above.

After repeated contacts by mail and phone, 191 men in Pardubice (70% of eligible contacts), 153 men in Augsburg (70%), and 162 men in Jerusalem (62%) were examined after an overnight fast. Data were obtained by a structured questionnaire, physical examination and analyses of blood samples. All study procedures were standardized, and identical instruments and equipment were used in all centres. Compliance with the identical protocol was enforced by central training of staff. The surveys in the Czech Republic and Bavaria were carried out from April to June 1995 and in Israel between September 1995 and October 1996.

Interview questionnaires assessed smoking, alcohol consumption, use of medications, self-reported medical history, including the Rose angina questionnaire, and sociodemographic characteristics. Body weight was measured in light clothing by newly calibrated mechanical scales with precision to the nearest 0.1 kg. Body height was measured without shoes by free standing stadiometers to the nearest 0.5 cm. Body mass index (BMI) was calculated as weight (in kg) divided by height (in m) squared. Waist circumference was measured as the smallest circumference below the costal margin, hip circumference at the most lateral point over the greater trochanter, using a tape measure, with precision to the nearest 0.5 cm. The waist-to-hip ratio (WHR) was calculated from these measures.

Blood pressure was measured three times by an oscillometric automated sphygmomanometer (BOSO Oscillomat). The average of the second and third measurement were used for this analysis. The oscillometric device had previously been validated against Korotkoff and intra-arterial measurements. The comparison was further standardized by the use of a limited set of four measurement devices that were circulated between centres. In addition, each participant was asked about intake of drugs in the 7 days preceding the examination and antihypertensive drugs were identified from brand names.

Fasting blood samples were taken at the end of the physical examination. Serum and EDTA/citrate plasma samples were processed identically according to a detailed common protocol and subsequently stored at −70°C in the Czech Republic and Israel and in a liquid nitrogen cryostore (−196°C) in Bavaria. The frozen samples were later transported on dry ice. Laboratory analyses of Czech, German, and first part of Israeli samples (n = 76) were carried out simultaneously in mixed batches. As data collection in Israel started later, the second part of the Israeli samples (n = 80) was analysed separately, with quantitative assessments of between-run laboratory drifts; such differences between runs, noted the fibrinogen, D-dimer, lipoprotein (Lp(a)), apolipoprotein B (apo-B), high density lipoprotein (HDL)-cholesterol and glucose, were adjusted for by

---

**Keywords**
Risk factors, carotenoids, homocysteine, coagulation factors, cardiovascular disease rates, ecological study, Eastern Europe

**Accepted** 10 November 1998
employing additive correction factors. Homocysteine concentrations were measured only in samples from Czech and Bavarian men.

Serum lipids, insulin, glucose, and coagulation factors were determined in the Arteriosclerosis Research Laboratory at the University Meunster, Germany, and vitamin levels were analysed by the Swiss Vitamin Institute in Lausanne, Switzerland. Concentrations of total cholesterol, HDL-cholesterol, triglycerides, Lp(a), and apo-B were quantified using a Hitachi 917 autoanalyzer. Enzymatic tests for total cholesterol and triglycerides and immunoturbidimetric tests for apo-B were purchased from Boehringer Mannheim (Germany). The Lp(a) was measured with a latex-enhanced immunoturbidimetric test from Immuno (Vienna, Austria) and HDL-cholesterol was measured after precipitation of apo-B containing lipoproteins with phosphotungstic acid/MgCl₂ (Boehringer Mannheim, Germany). Determinations of cholesterol, triglycerides, and HDL-cholesterol were controlled by the NHLBI Lipid Standardization Program organized by the Centers for Disease Control (CDC, Atlanta, GA, USA). Coefficients of variation for the immunological tests were below 5%. Insulin was measured by the use of a commercially available radioimmunoassay from DPC/Biermann (Bad Nauheim, Germany). Factor-VII activity (F-VIIc) and fibrinogen were measured on a Amelung KC 10 coagulation analyser (Lemgo, Germany). The F-VIIc was determined by a one-step assay using thromboplastin (Thromborel S) and factor-VII deficient plasma (Behringwerke Marburg, Germany). Fibrinogen was determined according to Clauss using thrombin (Multifibrin) and standards from Behringwerke (Marburg, Germany). The concentrations of D-dimer were assessed using an enzyme-linked immunosorbent assay (Boehringer Mannheim, Germany).

All-trans retinol, alpha-tocopherol and carotenoids (alpha- and beta-carotene, all-trans lycopene, lutein-zeaxanthin) were assayed in EDTA plasma by liquid chromatography as previously described.¹⁰ These were available only for a part of the Israel sample (n = 76); Israeli men with and without vitamin measurements were compared in terms of their differences in age, body mass index, waist-to-hip ratio, and total cholesterol levels. There was no indication of any statistically or clinically significant differences between these two groups. Plasma homocysteine was measured by a high performance liquid chromatography kit (Chromsystems, Munich, Germany). Reverse-phase chromatography was performed on an isotropic Kontron liquid chromatograph (Neufahrn, Germany) interfaced with a model RF-535 fluorescence detector (Shimadzu, Kyoto, Japan).

Arithmetic means were calculated for variables with approximately normal distribution. Variables with skewed distribution were transformed logarithmically and their geometric means with the approximation of standard deviations are given. Although Lp(a) has a bimodal distribution with a substantial proportion of subjects having undetectable levels,⁰¹ we present its geometric means as results of more sophisticated analyses were very similar. Each pair of centres was compared separately. Prevalence of categorical risk factors were compared between centres by χ² tests, and differences in arithmetic or geometric means of continuous variables were assessed by t-test for two independent samples. Although the samples differed in age (Czechs and Israelis 54.4 years, Bavarians 55.9 years, P = 0.066), age-adjusted results were virtually identical with unadjusted results. Therefore, only the latter are presented in this report.

Results

Mortality

Age-standardized mortality from all causes and CHD among men aged 45–64 years in Pardubice, Augsburg and the national rates for Israel are shown in Figure 1; rates in Pardubice and Augsburg are consistent with the Czech and Germany national rates.

Traditional risk factors (Table 1)

In parallel to cardiovascular disease mortality, Czechs were more likely to report a history of a previous myocardial infarction or symptom of angina than Bavarians (19% versus 11%, P = 0.020) or Israelis (6.5%, P = 0.001). Likewise, the prevalence of cigarette smoking were 34%, 30% and 24%, respectively (P = 0.06 Czech versus Israelis). Mean systolic and diastolic blood pressure of Czechs exceeded those in the other two populations (P < 0.001). Consistent with this, the prevalence of antihypertensive drug use was 23.5% in Czechs, 19.6% in Bavarians and only 12.5% in Israelis. By contrast, mean BMI and WHR were similar in the three samples.

Lipid and glucose metabolism (Table 2)

Frequencies of diabetes mellitus were similar (10%, 7% and 10%, respectively). Total and HDL-cholesterol, apo-B, and Lp(a) were similar in Czech and Bavarian men while fasting triglycerides were higher (P = 0.03), glucose levels were the same, and insulin was lower (P = 0.02) in Czechs compared to Bavarians. Relative to these two samples, Israelis had both lower total as well as lower HDL-cholesterol levels (P < 0.001), similar apo-B, and higher Lp(a) concentrations (P < 0.01). Geometric means for fasting triglycerides and insulin were in between the values
of Czechs and Bavarians while glucose levels in Israelis were significantly lower ($P < 0.01$).

**Haemostatic factors (Table 3)**

Czech men showed the highest concentrations of fibrinogen (relative to Bavarians only, $P < 0.001$) and of D-dimer ($P < 0.05$). Factor-VII activity was significantly lower in Czechs than in Bavarians and Israelis ($P < 0.001$).

**Homocysteine and antioxidant vitamins (Table 4)**

Measurements of homocysteine were available only for Czech and Bavarian men. Homocysteine concentrations were considerably higher in Czech (10.5 µmol/l) than in Bavarian men (8.9 µmol/l; $P < 0.001$). Major differences were observed in plasma concentrations of antioxidant vitamins. Specifically, mean levels of alpha-carotene, beta-carotene, and lycopene were substantially and significantly lower in Czech men. The average levels of these carotenoids reached only about half or less of those in Bavaria or Israel. There were consistent shifts of the entire distribution of each carotenoid to lower values in the Czech sample (Figures 2 and 3). Adjustment for smoking and BMI did not change these estimates. This contrasted with similar retinol, alpha-tocopherol and lu-tien-zeaxanthin concentrations in Czechs and Bavarians. Lower levels of these vitamins in Israelis were largely attenuated by adjustment for cholesterol.

Discussion

The primary objective of this study was to identify factors that can contribute to explaining the high rates of cardiovascular disease in the Czech Republic compared with Bavaria and Israel. We have chosen an ecological design, as it is the bulk of the population, rather than the extremes, which shape the mortality patterns. Czech men displayed a more adverse risk profile with regard to higher levels of blood pressure, triglycerides, fibrinogen and D-dimer, and had substantially lower concentrations of carotenoids and higher levels of homocysteine in plasma. Adverse risk-factor constellations observed for Czechs relative to Bavarians were also present with the same or greater magnitude against Israeli men, with the salient exception of HDL-cholesterol, low levels of which are characteristic of the Israeli Jewish population.

The study populations were selected on the basis of their previous involvement in international collaborations but their characteristics agree well with the national level situation. In all centres, data were collected by jointly trained staff using identical methods and instruments, and blood samples were analysed in central laboratories, with samples from all centres mixed in one batch. It is therefore unlikely that measurement bias could have affected our results. By design, the number of subjects were too low for individual analyses within study centres and did not allow further detailed investigation of...
the interrelations between risk factors and potential effect modifiers. Sample sizes were chosen to allow detection of biologically meaningful differences in population means with a power of 80% or more at a 95% confidence level. Different biochemical factors are often correlated; rather than concentrating on differences in individual variables between the study populations, we put more emphasis on the pattern of factors.

The response rates in Czech and Bavarian samples were identical, and given the similarity in culture and mentality, it is unlikely that non-responders would be markedly different

<table>
<thead>
<tr>
<th>Table 3</th>
<th>Means and standard deviations (SD) of haemostatic factors in Czech (Cz), Bavarian (Bav) and Israeli (Isr) men</th>
</tr>
</thead>
<tbody>
<tr>
<td>Czech Republic</td>
<td>Bavaria</td>
</tr>
<tr>
<td>(n = 188)</td>
<td>(n = 153)</td>
</tr>
<tr>
<td>P-values</td>
<td>Cz versus Bav</td>
</tr>
<tr>
<td>Fibrinogen (g/l)</td>
<td>3.03</td>
</tr>
<tr>
<td>D-dimer (mg/dl)</td>
<td>242.7</td>
</tr>
<tr>
<td>Factor-VII activity (% of standard)</td>
<td>124.6</td>
</tr>
</tbody>
</table>

a Geometric mean.

<table>
<thead>
<tr>
<th>Table 4</th>
<th>Means and standard deviations (SD) of plasma homocysteine and antioxidant vitamins in Czech (Cz), Bavarian (Bav) and Israeli (Isr) men</th>
</tr>
</thead>
<tbody>
<tr>
<td>Czech Republic</td>
<td>Bavaria</td>
</tr>
<tr>
<td>(n = 188)</td>
<td>(n = 153)</td>
</tr>
<tr>
<td>P-values</td>
<td>Cz versus Bav</td>
</tr>
<tr>
<td>Homocysteinea (µmol/l)</td>
<td>10.5</td>
</tr>
<tr>
<td>Vitamin A (mg/l)</td>
<td>0.73</td>
</tr>
<tr>
<td>Vitamin E (mg/l)</td>
<td>14.9</td>
</tr>
<tr>
<td>Lutein-Zeaxanthin (µg/l)</td>
<td>198.1</td>
</tr>
<tr>
<td>Alpha-carotene (µg/l)a</td>
<td>15.6</td>
</tr>
<tr>
<td>Beta-carotene (µg/l)a</td>
<td>60.0</td>
</tr>
<tr>
<td>Lycopene (µg/l)</td>
<td>82.5</td>
</tr>
</tbody>
</table>

a Geometric mean.

Figure 2 Cumulative distributions of beta-carotene in Czech, Bavarian and Israeli men
between these two cities. The response rate was lower in Israel; and the reasons for non-response here may be different from Central Europe. Similarly, while the Czech and Bavarian samples were examined at the same time of the year to avoid seasonal bias, the data collection in Israel took more than a year. Although the seasonal differences in food intake in Israel are thought to be minimal, the lower response rate and different period of data collection urge more caution when interpreting the data from Israel. From this reason, we have seen the results from Israel as complementary, and the main emphasis was put on Czech-Bavarian comparisons. The only aspect in which the samples were not strictly comparable is the fact that the Czech sample was a re-examination of the 1992 MONICA sample. As a result of being examined in 1992, Czech men could be more aware of their risk factor status, have adopted a healthier lifestyle and thus appear healthier. However, a comparison with their 1992 data does not support the existence of such bias.

The geographically close Czech and Bavarian populations were remarkably similar in many aspects. Mean height, weight and obesity were practically identical, and the prevalence of diabetes, smoking, and hyperlipidaemia was also similar. As expected, the distributions of Lp(a), a largely genetically controlled factor, were also nearly identical in the two population samples. Higher average Lp(a) levels in Israel were partly due to a significantly lower proportion of men with undetectable Lp(a) concentrations (7.5%, compared to 13.7% in Czechs and 13.2% in Bavarians). Our findings suggest that the divergence in disease rates cannot be explained by population differences in those factors, and corroborate results from other studies which also failed to identify many of the classical risk factors as the major determinants of international variations in cardiovascular disease rates.

We found, however, that mean blood pressure and rate of antihypertensive treatment were higher in Czechs. It seems unlikely that higher rates of hypertension or cardiovascular disease in Czech men are attributable to factors related to insulin resistance. Mean values of fasting glucose were identical in Czechs and Bavarians, and fasting insulin was higher in Bavaria. The product of fasting glucose and insulin, suggested as a more valid index of insulin resistance, was therefore highest in Bavaria and lowest in Israel.

Levels of fibrinogen were raised in Czechs and Israelis while D-dimer, an indicator of increased fibrinogen turnover and fibrin degradation, was significantly elevated only in Czech men. Raised levels of these factors possibly reflect chronic, low-grade inflammation due to a higher prevalence of more active and progressive atherosclerotic lesions in Czech men. They have been independently associated with the occurrence of cardiovascular events and offer one possible explanation for the high disease rates in this population. Higher levels in Bavarians and Israelis of factor-VIIc, another procoagulatory predictor for cardiovascular disease, seem to be contradicting this pattern. However, the evidence for factor-VIIc being a risk predictor has been inconsistent and awaits further confirmation.

There were substantial differences in homocysteine levels between Czechs and Bavarians. Although not directly analysed in this study, previously published work has shown that Israelis have lower homocysteine levels than most European populations including Germany. As moderate hyperhomocysteinaemia has been generally accepted as a strong and independent predictor of cardiovascular disease, our results are consistent with the mortality differences among the study population.

The prominent finding of this study concerns the surprisingly low carotenoid concentrations in Czech men. The entire
distribution of carotenoids and lycopene is shifted to lower values in Czech men (Figures 2 and 3). Different intakes of dietary sources of carotenoids are the most plausible explanation of their different plasma levels, although we do not have the data to directly substantiate this contention. An intervention trial has shown that increased consumption of fruits and vegetables raised plasma levels of carotenoids and vitamin C, but no vitamin E and A. This indicates possible sources of the differences in vitamin levels in our study. Diet could also have contributed to the high homocysteine levels of Czech men. Fresh leafy vegetables are dietary sources of both folic acid and carotenoids, and dietary intake of folic acid reduces plasma levels of homocysteine. It has been estimated that adding two or three servings of these vegetables daily would decrease plasma homocysteine by some 2 µmol/l.

In our view, the low levels of antioxidants and the high cardiovascular rates in the Czech populations may be related. Several mechanisms have been proposed for the protective role of carotenoids and lycopene in observational studies that suggest cardiovascular diseases are associated with low levels of carotenoids and lycopene. On the other hand, large trials failed to demonstrate a protective effect of antioxidant supplementation, and the protective effects found in observational studies may in fact be due to other nutrients in fruits and vegetables, the common sources of antioxidants.

The reports of low consumption of fresh fruits and vegetables in Central and Eastern Europe are consistent with this hypothesis. The highly pronounced seasonality in diet, with intakes of fresh fruits and vegetables over a large part of each year, may further contribute to high mortality in Eastern Europe. Albania, the only post-communist country with low coronary mortality, has had high consumption of fresh fruits and vegetables. Two other ecological studies found low levels of antioxidants in Central and Eastern Europe. A small case-control study in the Czech Republic found increased risk of non-fatal myocardial infarction in men with low levels of betacarotene. It is interesting that dietary changes, including consumption of fruits and vegetables, have also been implicated in the changes in mortality trends in the Czech Republic and Poland, although the temporal sequence of changes in Central Europe is not entirely clear.

Ecological comparisons are suggestive rather than conclusive of the causation of a disease, but they are an indispensable tool in the investigation of the population determinants of health. Comparison of small samples has been often used as a first step to provide hypotheses for future research. In the case of the East-West gap, it is unlikely that a single factor can offer a universal explanation. The patterns of risk factors found in this study suggest that, in addition to known risk factors such as hypertension or cloting variables, factors related to low dietary intake of fruits and vegetables may contribute to the high mortality in Central and Eastern Europe.

Acknowledgements

This study was supported by grants from the German-Israeli Foundation for Scientific Research and Development, and from the Wellcome Trust. MB was recipient of the Wellcome Trust research fellowship in clinical epidemiology, and MM is supported by a professorship from the Medical Research Council.

References


