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RESEARCH

Low carbohydrate-high protein diet and incidence of cardiovascular diseases in Swedish women: prospective cohort study


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Abstract

Objective To study the long term consequences of low carbohydrate diets, generally characterised by concomitant increases in protein intake, on cardiovascular health.

Design Prospective cohort study.

Setting Uppsala, Sweden.

Participants From a random population sample, 43 396 Swedish women, aged 30-49 years at baseline, completed an extensive dietary questionnaire and were followed-up for an average of 15.7 years.

Main outcome measures Association of incident cardiovascular diseases (ascertained by linkage with nationwide registries), overall and by diagnostic category, with decreasing carbohydrate intake (in tenths), increasing protein intake (in tenths), and an additive combination of these variables (low carbohydrate-high protein score, from 2 to 20), adjusted for intake of energy, intake of saturated and unsaturated fat, and several non-dietary variables.

Results A one tenth decrease in carbohydrate intake or increase in protein intake or a 2 unit increase in the low carbohydrate-high protein score were all statistically significantly associated with increasing incidence of cardiovascular disease overall (n=1270)—incidence rate ratio estimates 1.04 (95% confidence interval 1.00 to 1.08), 1.04 (1.02 to 1.06), and 1.05 (1.02 to 1.08). No heterogeneity existed in the association of any of these scores with the five studied cardiovascular outcomes: ischaemic heart disease (n=703), ischaemic stroke (n=294), haemorrhagic stroke (n=70), subarachnoid haemorrhage (n=121), and peripheral arterial disease (n=82).

Conclusions Low carbohydrate-high protein diets, used on a regular basis and without consideration of the nature of carbohydrates or the source of proteins, are associated with increased risk of cardiovascular disease.

Introduction

Overweight and obesity are risk factors for several common chronic diseases,[1] and they have become endemic in most economically developed countries and beyond.[2 3] Increased physical activity is one way of counteracting excessive energy intake, but reducing this intake is also important.[4] Many dietary regimens have been proposed as conducive to weight control, invoking various mechanisms including increased satiety.[5 6] The most popular among these diets emphasise reduction of carbohydrate intake, thereby encouraging high protein intake,[7] as high fat diets are generally avoided in most Western societies. Low carbohydrate-high protein diets may have short term effects on weight control,[8 9] but concerns have also been expressed, notably with respect to cardiovascular outcomes.[10 11] Although low carbohydrate-high protein diets may be nutritionally acceptable if the protein is mainly of plant origin and the reduction of carbohydrates applies mainly to simple and refined ones, the general public do not always recognise and act on these qualifications.

During the past few years, several cohort studies have evaluated the long term health effects of low carbohydrate-high protein diets, with emphasis on cardiovascular diseases. In the Nurses' Health Study in the United States, diets lower in carbohydrate and higher in protein were not associated with increased incidence of ischaemic heart disease.[12] Three smaller studies in Europe, however, indicated statistically significant increases in cardiovascular mortality in relation to low carbohydrate-high

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protein diets. One of these studies was from the same cohort of relatively young Swedish women as the investigation reported here (but presented data only on 75 cardiovascular deaths from shorter follow-up),[13] the second study was based on middle aged Greek men and women,[14] and the third study investigated elderly men in Sweden.[15] Willett has considered possible reasons for the discrepant results of the European studies and the American one, including differences in the main dietary sources of protein (animal versus plant) and the prevalence of obesity in the studied populations.[16]

Given the importance of the topic and the widespread use of low carbohydrate diets for weight control, particularly among women, we followed up the women in the Swedish Women's Lifestyle and Health Cohort,[13] focusing on incidence of cardiovascular diseases and using the valuable registry resources available in Sweden.

Methods

Participants

Women aged 30-49 years, residing in the Uppsala healthcare region in Sweden in 1991-92, were the source population for the Swedish Women's Lifestyle and Health Cohort. For this cohort, 96 000 women were randomly selected from four age strata (30-34, 35-39, 40-44, and 45-49), invited by mail to participate, and asked to fill in a questionnaire and return it in a pre-paid envelope.[17] A total of 49 261 questionnaires were returned.

Questionnaire and dietary assessment

The questionnaire used in the study was self administered and recorded information on several lifestyle variables (including detailed smoking and alcoholic drinking habits), anthropometry, and history of diagnoses of major diseases and conditions, including medical diagnosis of hypertension. For the assessment of physical activity, women rated their overall level of activity (that is, activities in the house and occupational and recreational physical activity) on a five point scale with examples attached to levels 1 (low), 3, and 5 (high). Dietary intakes were assessed with a validated food frequency questionnaire,[18 19] in which women recorded their frequency and quantities of consumption of about 80 food items and beverages, focusing on the six month period before their enrolment in the study. We formed 11 food groups from the food items: vegetables, legumes, fruits and nuts, dairy products, cereals, meat and meat products, fish and seafood, potatoes, eggs, sugars and sweets (all measured in g/day), and non-alcoholic beverages (measured in mL/day). Alcoholic beverages were listed in a separate category, and we calculated the amount of ethanol. On the basis of the Swedish national food administration database,[20] we translated food consumption into macronutrient and energy intakes.

We estimated the energy adjusted intakes of protein and carbohydrates for each woman, using the "residual method."[21] This method allows evaluation of the "effect" of an energy generating nutrient, controlling for the energy generated by this nutrient, by using a simple regression of that nutrient on energy intake to calculate the residual. For each woman, we assigned a score from 1 (very low protein intake) to 10 (very high protein intake), according to her tenth of energy adjusted total protein intake, and an inverse score from 1 (very high carbohydrate intake) to 10 (very low carbohydrate intake), according to her tenth of energy adjusted total carbohydrate intake. We studied the scores for high protein and low carbohydrate intake both separately and after adding them together to create a composite low carbohydrate-high protein score (ranging from 2 to 20), which simultaneously assessed the position of each woman in terms of protein and carbohydrate intake. Thus, a woman with a score of 2 was one with very high consumption of carbohydrates and very low consumption of proteins, whereas a woman with a score of 20 was one with very low consumption of carbohydrates and very high consumption of proteins. The use of energy adjusted residuals was necessary, because all energy generating nutrients are generally strongly positively associated with total energy intake. After controlling for energy intake, however, distinguishing the effects of a specific energy generating nutrient is all but impossible, as a decrease in the intake of one is unavoidably linked to an increase in the intake of one or several of the others.[22] Nevertheless, in this context, a low carbohydrate-high protein score allows the assessment of most low carbohydrate diets, which are generally high protein diets,[7] because it integrates opposite changes of two nutrients with equivalent energy values.

Follow-up

The primary outcome under investigation in this study was a first diagnosis of cardiovascular disease, as defined by ICD-9 (international classification of diseases, 9th revision) codes 410-414, 430-438, and 440 and ICD-10 codes I20-I25, I60-I67, I69, I70, and I74. We also studied ischaemic heart disease (ICD-9 410-414; ICD-10 I20-I25), ischaemic stroke (ICD-9 433-438; ICD-10 I63-I67, I69.3, I69.4, and I69.8.), intracerebral haemorrhage (ICD-9 431-432.9; ICD-10 I61-I62 and I69.1-I69.2), subarachnoid haemorrhage (ICD-9 430; ICD-10 I60 and I69.0), and peripheral arterial disease (ICD-9 440; ICD-10 I70 and I74) separately. We used linkages with nationwide Swedish registers, by means of the unique national registration numbers, for the follow-up of the cohort with respect to hospital discharges, death, and emigration. Information on dates of death by cause, as well as on dates of emigration to 31 December 2007, came from the Swedish Bureau of Statistics. Information on discharge diagnoses by date to 31 December 2007 came from the Swedish in-patient registry. We defined the date of return of the questionnaire during 1991-92 as the start of follow-up. We calculated observation time from the date of entry into the cohort until the occurrence of a first diagnosis of the cardiovascular event under study, death from the cardiovascular disease under study without a previous diagnosis of this cardiovascular disease, or censoring. For overall cardiovascular diseases, censoring was on account of emigration, the end of the observation period, or death from any cause other than cardiovascular disease. For the specific cardiovascular outcomes, censoring was on account of emigration, the end of the observation period, death from any cause other than the specific cardiovascular disease studied, or occurrence of cardiovascular disease other than the one studied.

Statistical analysis

Of the 49 261 Swedish women who returned the questionnaire, we excluded 1350 women from the analyses because of events before or at the cohort enrolment (10 emigrated, 136 had previous cardiovascular events, 621 had energy intake outside the extreme 1% centiles of the population energy intake (<1846.6 or >12 473.9 kJ/day), and 583 women had not filled in the food frequency questionnaire), leaving 47 911 women for the statistical analyses. After exclusion of women with missing data on any of the model covariates, 43 396 women remained for use in the statistical analyses. No major differences existed between the women excluded on account of missing data and the women who contributed to the study. For each

cardiovascular diagnostic category, we had about 680 000 person years of follow-up.

The participating women and the number of incident cases of cardiovascular disease (overall and by specific diagnostic categories) that occurred among them were distributed by non-nutritional covariates, and we calculated age adjusted incidence rate ratios by using Poisson regression models. We also calculated summary statistics of daily dietary intakes of energy generating nutrients. Subsequently, we tabulated the distribution by low carbohydrate-high protein score of the person time, the number of incident cases of cardiovascular disease (overall and by diagnostic categories), and the corresponding crude incidence rates per 10 000 woman years.

We calculated incidence rate ratios, for all cardiovascular diseases combined and for each category of cardiovascular disease, by using survival analysis with attained age as the primary underlying timescale.[23] Specifically, we fitted Poisson regression models by splitting the follow-up-time (that is, attained age between cohort entry and exit) into two year intervals and then allowing for one parameter for each such interval and an offset parameter equal to the logarithm of the observed risk time in each interval. For each participant, we considered only the first cardiovascular event. Poisson regression is commonly used in survival analysis and gives approximately the same parameter estimates and likelihood ratios as Cox proportional hazards regression when the length of follow-up is split into finer intervals (here we used two year intervals).[24 25] We used, alternatively, the high protein score, the low carbohydrate score, and the additive low carbohydrate-high protein score as the primary exposure variable, introducing the scores, alternatively, as ordered or categorical variables. We ensured that the parameter for each of high protein, low carbohydrate, and low carbohydrate-high protein score was approximately constant across time by visual inspection of the estimated log rates and by calculating likelihood ratio tests for parameters capturing the high protein, low carbohydrate, and low carbohydrate-high protein exposure by time (attained age) interactions.

We adjusted the models for the following variables as reported at enrolment: height (cm, continuously), body mass index (<25, 25-29.99 and ≥30, categorically), smoking status (never smokers, former smokers of <10 cigarettes, former smokers of 10-14 cigarettes, former smokers of 15-19 cigarettes, former smokers of ≥20 cigarettes, current smokers of <10 cigarettes, current smokers of 10-14 cigarettes, current smokers of 15-19 cigarettes, and current smokers of ≥20 cigarettes, categorically), physical activity (from 1 (low) to 5 (high), categorically), education (≤10, 11-13, and ≥14 years in school, categorically), report of ever being diagnosed as having hypertension (yes versus no), energy intake (per 1000 kJ/day, continuously), unsaturated lipid intake (per 10 g/day, continuously), saturated lipid intake (per 10 g/day, continuously), and alcohol intake (<5 g/day, 5-25 g/day, and >25 g/day, categorically). We used categories to accommodate trends that may not be log linear and also to take into account biological considerations or guidelines, as for body mass index and alcohol intake. In models evaluating the associations of the outcome variables with the low carbohydrate and the high protein scores, we repeated the analysis by also introducing the complementary score (that is, high protein in the models evaluating low carbohydrate and vice versa) without energy intake in the model.

To examine whether the dietary exposure variables were differentially associated with the five distinct cardiovascular outcomes, we also fitted Poisson regression models including all five cardiovascular conditions simultaneously.[26] The joint model, stratifying on cardiovascular outcome and under the proportional hazards assumption, allows the evaluation of separate associations between exposure and each outcome. This approach enabled us to test directly for heterogeneity in incidence rate ratios across the cardiovascular conditions by using likelihood ratio tests. We did this for the high protein, low carbohydrate, and low carbohydrate-high protein scores in separate models.

Because the origin of proteins (animal versus plant sources) may have differential effects on the occurrence of cardiovascular events,[27 28] we also evaluated the association of the cardiovascular outcomes with the low carbohydrate-high protein score, as well as the low carbohydrate and high protein scores, separately among women with intake of animal protein equal to or higher than the cohort median and among women with intake of animal protein lower than the cohort median. We calculated P values for interaction by likelihood ratio tests after the introduction of product terms of the respective scores (as continuous variables) with protein intake (as a two level categorical variable: above or equal to versus below the median).

We used SAS versions 9.22 and 9.3 for all data management and statistical analyses (PROC GENMOD). We made all tests of statistical hypotheses on the two sided 5% level of significance corresponding to using two sided 95% confidence intervals. In addition, using R software 2.12.1, we calculated the variance inflation factor, to ensure that no problems were generated by co-linearity among the model covariates.[29]

Results

Overall, the 43 396 women were followed up for an average of about 15.7 years and generated a total of 680 818 person years, with 1270 incident cardiovascular events (703 ischaemic heart disease, 294 ischaemic stroke, 70 haemorrhagic stroke, 121 subarachnoid haemorrhage, and 82 peripheral arterial disease). Table 1⇓ shows the distribution of the women in the cohort and the incident cardiovascular cases (overall and by diagnostic category) by non-nutritional variables. The table also shows the corresponding age adjusted incidence rate ratios for descriptive purposes only, as confounding influences by factors other than age are not accounted for. Nevertheless, several well known patterns are evident, including the reduced risk of cardiovascular diseases with increasing level of education and physical activity and the increased risk with tobacco smoking and history of hypertension.

Table 2⇓ shows representative values of the daily dietary intakes and gives an overview of the nutritional background of the women in the cohort. In this cohort of women, median intake of energy was 6390 kJ/day with 10th and 90th centiles of 4241 and 9053 kJ/day. The additive low carbohydrate-high protein score was positively correlated with protein intake (Spearman r=0.37), inversely correlated with carbohydrate intake (Spearman r=−0.30), and positively correlated with lipid intake (Spearman r=0.28 for saturated lipids; Spearman r=0.17 for unsaturated lipids), but, importantly, it was not correlated with energy intake (Spearman r=−0.004).

Table 3⇓ shows the women in the cohort, the person time generated, and the number of incident cases overall and for each diagnostic category as distributed by categories of low carbohydrate-high protein score. The corresponding crude incidence rates per 10 000 woman years are also shown. Although potential confounding influences have not been accounted for, the data in this table suggest that the incidence of cardiovascular outcomes tends to increase with increasing low carbohydrate-high protein score; the patterns are more

regular in the more numerous categories of overall cardiovascular diseases and ischaemic heart disease.

Table 4⇓ shows the fully adjusted (see table footnote) incidence rate ratios for overall cardiovascular diseases and the main diagnostic subcategories, per decreasing tenth of carbohydrate intake, increasing tenth of protein intake, and their addition (that is, per 1 unit increase in low carbohydrate and high protein scores and 2 unit increase in low carbohydrate-high protein score). In this table, the influence of potential confounding variables ascertained in the study has been accounted for, and a comparison with the results in table 3⇓ indicates the modest amount of confounding by these variables. A pattern of positive associations can be seen, which are statistically significant for all three scores with respect to the overall cardiovascular outcomes and for high protein and low carbohydrate-high protein scores with respect to the two more common subcategories—that is, ischaemic heart disease and ischaemic stroke. For each of the three scores, a formal statistical comparison of their associations with the distinct cardiovascular outcomes did not show statistically significant evidence of heterogeneity (P values for heterogeneity with the scores as continuous variables were 0.90 for the low carbohydrate score, 0.41 for the high protein score, and 0.79 for the low carbohydrate-high protein score). Thus, a one tenth decrease in carbohydrate intake or increase in protein intake or an increase in the low carbohydrate-high protein score by 2 units were all statistically significantly associated with increasing incidence of cardiovascular diseases overall, with point estimates of 1.04 (95% confidence interval 1.00 to 1.08), 1.04 (1.02 to 1.06), and 1.05 (1.02 to 1.08). When we evaluated the associations of various cardiovascular outcomes with low carbohydrate and high protein scores, mutually controlling for each other, but excluding total energy intake, the results were essentially unchanged.

Using the incidence rates in table 3⇓, we evaluated whether the trends shown in table 4⇓ were driven by the extreme score categories or whether they reflected an essentially smooth exposure-response pattern. With a low carbohydrate-high protein score of 6 or less as the baseline, the incidence rate ratio for overall cardiovascular diseases increased to 1.13 for women with a low carbohydrate-high protein score from 7 to 9, to 1.23 for those with a score from 10 to 12, to 1.54 for those with a score from 13 to 15, and to 1.60 for those with a score of 16 or higher. An underlying, fairly regular trend is thus apparent.

Taking into account the fact that diet was assessed at recruitment only and could have changed over time, we also examined whether a tendency exists for the associations to weaken with longer follow-up. We examined the incidence rate ratios in the first 10 years and in the remaining 5.7 years of follow-up. We found a tendency towards attenuation in the later follow-up period. Thus, contrasting the earlier with the later follow-up period, for overall cardiovascular diseases the association per one tenth increase in the scores changed from 1.05 (1.01 to 1.09) to 1.03 (0.99 to 1.07) for the low carbohydrate score, from 1.05 (1.03 to 1.07) to 1.02 (1.00 to 1.05) for the high protein score, and from 1.06 (1.03 to 1.09) to 1.04 (1.00 to 1.07) for the low carbohydrate-high protein score.

We further evaluated whether the associations evident in table 4⇓ were more pronounced among women whose protein intake was mainly of animal origin than among those whose protein intake was mainly of plant origin, by using the median intake of animal protein as cut-off value (40.9 g/day). We found a suggestion that the incidence rate ratios tended to be higher among women whose protein intake was mainly of animal rather than plant origin, although the formal tests for interaction were generally non-significant, except with respect to subarachnoid haemorrhage for the low carbohydrate-high protein score. For overall cardiovascular diseases, the incidence rate ratio per one unit increase in the low carbohydrate score was 1.06 (1.00 to 1.12) for women with animal protein intake above or equal to the median and 0.99 (0.93 to 1.06) for those with animal protein intake below the median (P for interaction=0.86); the results per 1 unit increase in the high protein score were 1.04 (1.00 to 1.08) and 1.03 (1.00 to 1.07) (P for interaction=0.92); and the results per 2 unit increase in the low carbohydrate-high protein score were 1.06 (1.01 to 1.12) and 1.03 (0.98 to 1.08) (P for interaction=0.93). For subarachnoid haemorrhage, the incidence rate ratio per 2 unit increase in the low carbohydrate-high protein score was 1.17 (1.00 to 1.38) for women with animal protein intake above or equal to the median and 0.90 (0.76 to 1.07) for those with animal protein intake below the median (P for interaction=0.04, without accounting for multiple comparisons).

Discussion

In a population based cohort study of 43 396 women followed up for an average of about 15.7 years and generating about 680 000 person years, 1270 incident cardiovascular events were recorded. After fine controlling for all assessed cardiovascular risk factors that could act as confounding variables, as well as for total energy and saturated and unsaturated fat intake, we found that women had a statistically significant 5% increase in the incidence of cardiovascular events per 2 unit increase in the 20 unit low carbohydrate-high protein score. In practical terms, and taking into account the rough correspondence in the ranking of energy adjusted and crude tenths of intake, a 20 g decrease in daily carbohydrate intake and a 5 g increase in daily protein intake would correspond to a 5% increase in the overall risk of cardiovascular disease.

We found no heterogeneity in the association of the low carbohydrate-high protein score with specific cardiovascular outcomes; the incidence rate ratio associated with a 2 unit increase in the low carbohydrate-high protein score varied from a statistically significant 1.04 for ischaemic heart disease to a statistically non-significant 1.07 for subarachnoid haemorrhage and a statistically significant 1.07 for ischaemic stroke. We consider this to be evidence of consistency, pointing to a common component in the dietary causes of the various cardiovascular outcomes.

The results followed a fairly regular exposure-response pattern. Both high protein and low carbohydrate scores were statistically significantly associated with increased incidence of cardiovascular diseases overall. The results with respect to high protein score were also statistically significant for the two more numerous cardiovascular disease categories—namely, ischaemic heart disease and ischaemic stroke. The associations of low carbohydrate, high protein, and low carbohydrate-high protein scores with cardiovascular outcomes were not, in general, statistically significantly different between women whose protein intake was mainly of animal origin and those whose protein intake was mainly of plant origin.

Low carbohydrate diets, recommended for weight control, often contain less than 15% of energy intake from carbohydrates and about 30% of energy intake from proteins.[30] Among the women studied, carbohydrate intake at the low extreme of the distribution was higher and protein intake at the high extreme of the distribution was lower than the respective intakes prescribed by many weight control diets. However, the underlying trend between low carbohydrate-high protein score and incidence of cardiovascular disease was essentially monotonic, indicating that our findings are applicable across

the spectrum of carbohydrate and protein intakes of the participating women.

Comparison with other studies

At least four cohort studies have examined the association of low carbohydrate-high protein diets and mortality from or incidence of cardiovascular diseases overall or ischaemic heart disease as the most common among categories of cardiovascular disease. Halton and colleagues,[12] reporting from the Nurses' Health Study in the United States, found that diets lower in carbohydrate and higher in protein and fat were not associated with increased incidence of ischaemic heart disease in women. In three other population based cohort studies, however, including one based on the same cohort as the study reported here,[13] low carbohydrate-high protein diets were statistically significantly positively associated with cardiovascular mortality among young women,[13] in the general population in Greece,[14] or among elderly men in Sweden.[15] The results of this investigation, which focuses on incidence of cardiovascular outcomes, are compatible with this last set of studies.

The results from the different cohorts, however, are not as divergent as they appear. Fung and colleagues,[31] reporting from the Nurses' Health Study and the Health Professionals Follow-up Study in the United States, found that a low carbohydrate score was associated with a modest, statistically non-significant increase in cardiovascular mortality. When the results were examined with the animal or plant origin of the diet taken into account, a statistically significant positive association was seen with low carbohydrate diets of mainly animal origin, but a statistically significant inverse association was seen with low carbohydrate diets of mainly vegetable origin. In studies evaluating the incidence of stroke and ischaemic heart disease among men in relation to protein intake in the Health Professionals Follow-up Study,[27 28] statistically non-significant positive associations of protein intake were reported for both outcomes; when the results were examined by animal or plant origin of protein, the associations were positive for both outcomes for animal protein, whereas they were statistically non-significantly inverse for vegetable protein with respect to both outcomes. Finally, a positive association between intake of red meat and risk of ischaemic heart disease among women was reported from the Nurses' Health Study.[32] Thus, the results of the major US cohorts do indicate an increase, though generally not statistically significant, in risk of cardiovascular disease with higher protein intake, low carbohydrate intake, or both, but they point to a possible harmful effect driven by diets primarily of animal origin. The results of our study and those of the other European cohorts point to a statistically significant harmful effect of low carbohydrate and high protein diets in general.[14 15] Differential effects by animal or plant origin of the diet were not examined in the earlier European studies, but in our study we were not able to document any effects, possibly on account of power limitations. Thus, the results of our study, although indicating that low carbohydrate-high protein diets tend in general to be detrimental to cardiovascular health, do not preclude that dietary regimens low in refined carbohydrates and high in plant proteins or unsaturated lipids could have no adverse effects on cardiovascular health. As also pointed out by Willett,[16] one has to take into account differences in the sources of proteins in various populations, as well as the nature of carbohydrates consumed. Also, differences in the way protein sources, and perhaps also carbohydrate sources, are prepared and processed might explain, to a certain extent, diverging results from studies in different populations.[33]

With respect to the biomedical plausibility of our findings, vegetables, fruits, cereals, and legumes, which have been found in several studies to be core components of healthy dietary patterns,[34 35] are important sources of carbohydrates, so that reduced intake of these food groups is likely to have adverse effects on cardiovascular health. Moreover, several studies have reported that meat consumption or high intake of protein from animal sources may increase the risk of cardiovascular disease.[31 36] Intervention studies have reported beneficial effects of regimens high in protein, low in carbohydrates, or both on blood lipids that could favourably affect cardiovascular risk, but the follow-up in these studies was short (one year or less).[37 38] Of note, a study in mice has indicated that low carbohydrate-high protein diets may have substantial adverse vascular effects not reflected in serum markers but mediated through reduction of bone marrow and peripheral blood endothelial progenitor cells,[11] which are considered a marker of vascular regenerative capacity.[39]

Strengths and limitations of study

Our study has several strengths: the cohort was population based, the dietary questionnaire was validated, several potential confounding variables were ascertained in detail, and nationwide data linkage in Sweden allowed virtually complete follow-up and objective ascertainment of cardiovascular outcomes. The healthcare system in Sweden is publicly financed, and referral to hospital does not depend on insurance status or the financial background of the patient. Concerning the analytical strategy, we followed the substitution approach,[21 22] taking into account the fact that energy generated by carbohydrates and proteins is equivalent and that a simple additive score reflecting the balance of intakes of these two energy generating nutrients was uncorrelated with total energy intake. Controlling for saturated and unsaturated fat intake and as finely as possible for other potential confounders, particularly smoking, minimised the potential for confounding bias.

Among the weaknesses of the study are concerns about misclassification of dietary exposures, particularly as diet was assessed at enrolment only and was self reported through a food frequency questionnaire, which, however, is a standard practice in large cohort studies. With respect to capturing seasonality of consumption, participants were enrolled in a time frame longer than one year, and we expect that seasonal variation was balanced and therefore largely accounted for. In any case, errors in the ascertainment of diet are generally not correlated with errors in the ascertainment of incidence of cardiovascular disease and, hence, are unlikely to generate substantial confounding. The long interval between exposure and outcome is a source of concern, because certain participants may change their dietary habits during the intervening period. However, this is more likely to generate non-differential misclassification and, thus, attenuate the evaluated association. In fact, we saw a tendency for the incidence rate ratios to decline with longer follow-up. Information on waist:hip ratio was missing for a large fraction of the cohort, and data on drugs for cardiovascular diseases were not available (the relevant registry was not operational during most of the follow-up period). Finally, we did not have data on blood cholesterol, an important risk factor for cardiovascular diseases, but even if such values were available, they would probably be, at least partly, intermediates in the association between diet and incidence of cardiovascular disease. As in all observational studies, residual confounding cannot be confidently excluded, but our control of potential confounders is as effective as that in other cohort studies with similar objectives.

Conclusions and policy implications

Having been generated from a cohort of relatively young women, the results of our study are directly relevant to a group that often resorts to weight control regimens that encourage restriction of carbohydrate with unavoidable increases in protein intake. Our results do not answer questions concerning possible beneficial short term effects of low carbohydrate or high protein diets in the control of body weight or insulin resistance. Instead, they draw attention to the potential for considerable adverse effects on cardiovascular health of these diets when they are used on a regular basis, without consideration of the nature of carbohydrates (complex versus refined) or the source of proteins (plant versus animal).

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Contributors: HOA and EW were involved in the conception and design of the initial cohort. PL, HOA, and DT were involved in the conception and design of the study. SS, PL, and DT did the data analysis. ML and PL were responsible for nutritional epidemiology. All authors were involved in interpretation of data. PL, SS, and DT drafted the manuscript, and all authors critically revised it for important intellectual content. All authors approved the final version to be published. EW is the guarantor.

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Ethical approval: The study was approved by the Swedish Data Inspection Board and the regional ethical committee.

Data sharing: No additional data available.

1 Pi-Sunyer X. The medical risks of obesity. *Postgrad Med* 2009;121:21-33.
2 Li Z, Bowerman S, Heber D. Health ramifications of the obesity epidemic. *Surg Clin North Am* 2005;85:681-701.
3 Ogden CL, Yanovski SZ, Carroll MD, Flegal KM. The epidemiology of obesity. *Gastroenterology* 2007;132:2087-102.
4 Bessesen DH. Update on obesity. *J Clin Endocrinol Metab* 2008;93:2027-34.
5 Paddon-Jones D, Westman E, Mattes RD, Wolfe RR, Astrup A, Westerterp-Plantenga M. Protein, weight management, and satiety. *Am J Clin Nutr* 2008;87:1558-61S.
6 Astrup A, Kristensen M, Gregersen NT, Belza A, Lorenzen JK, Due A, et al. Can bioactive foods affect obesity? *Ann N Y Acad Sci* 2010;1190:25-41.
7 Astrup A, Meinert Larsen T, Harper A. Atkins and other low carbohydrate diets: hoax or an effective tool for weight loss? *Lancet* 2004;364:897-9.
8 Bravata DM, Sanders L, Huang J, Krumholz HM, Olkin I, Gardner CD, et al. Efficacy and safety of low-carbohydrate diets: a systematic review. *JAMA* 2003;289:1837-50.
9 Samaha FF, Iqbal N, Seshadri P, Chicano KL, Daily DA, McGrory J, et al. A low-carbohydrate as compared with a low-fat diet in severe obesity. *N Engl J Med* 2003;348:2074-81.
10 Steffen LM, Nettleton JA. Carbohydrates: how low can you go? *Lancet* 2006;367:880-1.
11 Foo SY, Heller ER, Wykrzykowska J, Sullivan CJ, Manning-Tobin JJ, Moore KJ, et al. Vascular effects of a low-carbohydrate high-protein diet. *Proc Natl Acad Sci U S A* 2009;106:15418-23.
12 Halton TL, Willett WC, Liu S, Manson JE, Albert CM, Rexrode K, et al. Low-carbohydrate-diet score and the risk of coronary heart disease in women. *N Engl J Med* 2006;355:1991-2002.
13 Lagiou P, Sandin S, Weiderpass E, Lagiou A, Mucci L, Trichopoulos D, et al. Low carbohydrate-high protein diet and mortality in a cohort of Swedish women. *J Intern Med* 2007;261:366-74.
14 Trichopoulou A, Psaltopoulou T, Orfanos P, Hsieh CC, Trichopoulos D. Low-carbohydrate-high-protein diet and long-term survival in a general population cohort. *Eur J Clin Nutr* 2007;61:575-81.
15 Sjögren P, Becker W, Warensjö E, Olsson E, Byberg L, Gustafsson IB, et al. Mediterranean and carbohydrate-restricted diets and mortality among elderly men: a cohort study in Sweden. *Am J Clin Nutr* 2010;92:967-74.
16 Willett WC. Low-carbohydrate diets: a place in health promotion? *J Intern Med* 2007;261:363-5.
17 Veierod MB, Weiderpass E, Thorn M, Hansson J, Lund E, Armstrong B, et al. A prospective study of pigmentation, sun exposure, and risk of cutaneous malignant melanoma in women. *J Natl Cancer Inst* 2003;95:1530-8.
18 Wolk A, Bergström R, Hunter D, Willett W, Ljung H, Holmberg L, et al. A prospective study of association of monounsaturated fat and other types of fat with risk of breast cancer. *Arch Intern Med* 1998;158:41-5.
19 Khani BR, Ye W, Terry P, Wolk A. Reproducibility and validity of major dietary patterns among Swedish women assessed with a food-frequency questionnaire. *J Nutr* 2004;134:1541-5.
20 Bergstrom L, Kylberg E, Hagman U, Eriksson HB, Bruce A. The food composition database KOST: the National Food Administration's information system for nutritive values of food. *Var Foda* 1991;43:439-47.
21 Willett W, Stampfer M. Implications of total energy intake for epidemiological analyses. In: Willett W, ed. Nutritional epidemiology. 2nd ed. Oxford University Press, 1998:273-301.
22 Wacholder S, Schatzkin A, Freedman LS, Kipnis V, Hartman A, Brown CC. Can energy adjustment separate the effects of energy from those of specific macronutrients? *Am J Epidemiol* 1994;140:848-55.
23 Korn EL, Graubard BI, Midthune D. Time-to-event analysis of longitudinal follow-up of a survey: choice of the time-scale. *Am J Epidemiol* 1997;145:72-80.
24 Pawitan Y. In all likelihood: statistical modelling and inference using likelihood. Oxford University Press, 2001:326-30.
25 Whitehead J. Fitting Cox's regression model to survival data using GLIM. *J Royal Stat Soc Series C (Appl Stat)* 1980;29:268-75.
26 Lunn M, McNeil D. Applying Cox regression to competing risks. *Biometrics* 1995;51:524-32.
27 Preis SR, Stampfer MJ, Spiegelman D, Willett WC, Rimm EB. Lack of association between dietary protein intake and risk of stroke among middle-aged men. *Am J Clin Nutr* 2010;91:39-45.
28 Preis SR, Stampfer MJ, Spiegelman D, Willett WC, Rimm EB. Dietary protein and risk of ischemic heart disease in middle-aged men. *Am J Clin Nutr* 2010;92:1265-72.
29 Kleinbaum DG, Kupper LL, Muller KE. Applied regression analysis and other multivariate methods. 2nd ed. PWS-Kent, 1988.
30 Volek J, Westman E. Very-low-carbohydrate weight-loss diets revisited. *Cleve Cl J Med* 2002;69:849-58.
31 Fung TT, van Dam RM, Hankinson SE, Stampfer M, Willett WC, Hu FB. Low-carbohydrate diets and all-cause and cause-specific mortality: two cohort studies. *Ann Intern Med* 2010;153:289-98.
32 Bernstein AM, Sun Q, Hu FB, Stampfer MJ, Manson JE, Willett WC. Major dietary protein sources and risk of coronary heart disease in women. *Circulation* 2010;122:876-83.
33 Larsson SC, Virtamo J, Wolk A. Red meat consumption and risk of stroke in Swedish men. *Am J Clin Nutr* 2011;94:417-21.
34 McCullough ML, Feskanich D, Stampfer MJ, Giovannucci EL, Rimm EB, Hu FB, et al. Diet quality and major chronic disease risk in men and women: moving toward improved dietary guidance. *Am J Clin Nutr* 2002;76:1261-71.
35 Trichopoulou A, Costacou T, Bamia C, Trichopoulos D. Adherence to a Mediterranean diet and survival in a Greek population. *N Engl J Med* 2003;348:2599-608.
36 Zyriax BC, Boeing H, Windler E. Nutrition is a powerful independent risk factor for coronary heart disease in women—the CORA study: a population-based case-control study. *Eur J Clin Nutr* 2005;59:1201-7.
37 Foster GD, Wyatt HR, Hill JO, McGuckin BG, Brill C, Mohammed BS, et al. A randomized trial of a low-carbohydrate diet for obesity. *N Engl J Med* 2003;348:2082-90.
38 Noakes M, Keogh JB, Foster PR, Clifton PM. Effect of an energy-restricted, high-protein, low-fat diet relative to a conventional high-carbohydrate, low-fat diet on weight loss, body composition, nutritional status, and markers of cardiovascular health in obese women. *Am J Clin Nutr* 2005;81:1298-306.
39 Hill JM, Zalos G, Halcox JP, Schenke WH, Waclawiw MA, Quyyumi AA, et al. Circulating endothelial progenitor cells, vascular function, and cardiovascular risk. *N Engl J Med* 2003;348:593-600.

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What is already known on this topic

Sporadic reports have suggested that low carbohydrate-high protein diets can increase the risk of cardiovascular disease

Three European cohort studies relying on mortality have all provided supportive evidence, but a US cohort study based on incidence indicated no association

What this study adds

In a large cohort of women in Sweden, compelling evidence shows that low carbohydrate-high protein diets may increase cardiovascular risk

Tables

Table 1| Distributions of 43 396 women by non-nutritional variables: number of incident cases of cardiovascular diseases overall, ischaemic heart disease, ischaemic and haemorrhagic stroke, subarachnoid haemorrhage, and peripheral arterial disease with corresponding incidence rate ratios* in Swedish Women's Lifestyle and Health Cohort

		No of incident cases; incidence rate ratio (95% CI)					
Variables	No	Cardiovascular disease	Ischaemic heart disease	Ischaemic stroke	Haemorrhagic stroke	Subarachnoid haemorrhage	Peripheral arterial disease
Age at enrolment (years):							
30-34	10 414	84; 1	36; 1	21; 1	7; 1	15; 1	5; 1
35-39	11 145	178; 0.91 (0.68 to 1.20)	88; 0.83 (0.55 to 1.24)	46; 1.04 (0.59 to 1.83)	7; 0.49 (0.16 to 1.56)	23; 0.93 (0.47 to 1.86)	14; 1.78 (0.50 to 6.34)
40-44	11 449	350; 1.09 (0.82 to 1.45)	196; 1.05 (0.70 to 1.57)	72; 1.03 (0.58 to 1.85)	22; 0.96 (0.33 to 2.81)	39; 1.31 (0.63 to 2.71)	21; 1.51 (0.40 to 5.68)
45-49	10 388	656; 1.24 (0.92 to 1.66)	383; 1.27 (0.84 to 1.92)	155; 1.15 (0.63 to 2.10)	34; 0.90 (0.29 to 2.81)	44; 1.10 (0.50 to 2.45)	42; 1.79 (0.47 to 6.86)
Education (years):							
≤10	12 927	630; 1	370; 1	135; 1	32; 1	48; 1	45; 1
11-13	16 848	395; 0.65 (0.57 to 0.74)	213; 0.61 (0.52 to 0.73)	91; 0.71 (0.54 to 0.93)	22; 0.69 (0.40 to 1.21)	45; 0.82 (0.54 to 1.24)	24; 0.54 (0.33 to 0.90)
>13	13 621	245; 0.45 (0.39 to 0.52)	120; 0.38 (0.31 to 0.47)	68; 0.59 (0.44 to 0.79)	16; 0.57 (0.31 to 1.04)	28; 0.60 (0.38 to 0.97)	13; 0.33 (0.18 to 0.61)
Height (cm):							
≤160	5 394	276; 1	162; 1	63; 1	18; 1	18; 1	15; 1
161-165	12 260	392; 0.87 (0.74 to 1.01)	224; 0.85 (0.69 to 1.04)	88; 0.85 (0.62 to 1.18)	17; 0.58 (0.30 to 1.12)	41; 1.39 (0.80 to 2.42)	22; 0.90 (0.47 to 1.73)
166-170	13 911	383; 0.82 (0.70 to 0.96)	216; 0.79 (0.65 to 0.97)	81; 0.76 (0.55 to 1.06)	21; 0.69 (0.37 to 1.29)	35; 1.13 (0.64 to 2.00)	30; 1.18 (0.64 to 2.20)
>170	11 831	219; 0.77 (0.64 to 0.92)	101; 0.61 (0.48 to 0.78)	62; 0.96 (0.67 to 1.36)	14; 0.75 (0.37 to 1.51)	27; 1.39 (0.77 to 2.53)	15; 0.97 (0.47 to 1.98)
Body mass index:							
<18.5	752	34; 2.17 (1.54 to 3.07)	20; 2.53 (1.61 to 3.96)	5; 1.31 (0.54 to 3.17)	1; 1.04 (0.14 to 7.54)	5; 2.60 (1.05 to 6.40)	3; 2.93 (0.91 to 9.41)
18.5-24.9	30 628	721; 1	369; 1	177; 1	44; 1	84; 1	47; 1
25-29.9	9 532	357; 1.44 (1.27 to 1.64)	210; 1.64 (1.39 to 1.95)	77; 1.26 (0.96 to 1.64)	20; 1.33 (0.79 to 2.27)	29; 1.06 (0.70 to 1.62)	21; 1.31 (0.78 to 2.19)
≥30	2 484	158; 2.48 (2.09 to 2.94)	104; 3.16 (2.54 to 3.93)	35; 2.21 (1.54 to 3.18)	5; 1.30 (0.51 to 3.27)	3; 0.43 (0.14 to 1.35)	11; 2.66 (1.38 to 5.14)
Physical activity:							
1 (low)	1 797	90; 1	48; 1	22; 1	4; 1	7; 1	9; 1
2	4 643	170; 0.73 (0.57 to 0.94)	98; 0.79 (0.56 to 1.12)	35; 0.62 (0.36 to 1.05)	8; 0.77 (0.23 to 2.56)	15; 0.82 (0.34 to 2.02)	14; 0.60 (0.26 to 1.39)
3	25 878	796; 0.60 (0.48 to 0.74)	437; 0.61 (0.46 to 0.83)	189; 0.58 (0.37 to 0.90)	46; 0.78 (0.28 to 2.15)	79; 0.76 (0.35 to 1.65)	45; 0.34 (0.16 to 0.69)

Table 1 (continued)

Variables	No	No of incident cases; incidence rate ratio (95% CI)					
		Cardiovascular disease	Ischaemic heart disease	Ischaemic stroke	Haemorrhagic stroke	Subarachnoid haemorrhage	Peripheral arterial disease
4	7 405	148; 0.40 (0.31 to 0.52)	80; 0.40 (0.28 to 0.58)	36; 0.40 (0.23 to 0.69)	8; 0.48 (0.15 to 1.61)	14; 0.48 (0.19 to 1.18)	10; 0.27 (0.11 to 0.66)
5 (high)	3 673	66; 0.37 (0.27 to 0.51)	40; 0.43 (0.28 to 0.65)	12; 0.28 (0.14 to 0.57)	4; 0.51 (0.13 to 2.03)	6; 0.42 (0.14 to 1.25)	4; 0.23 (0.07 to 0.73)
Smoking at enrolment:							
Never smoker	17 901	330; 1	188; 1	89; 1	22; 1	22; 1	9; 1
Ex-smoker	12 826	313; 1.33 (1.14 to 1.56)	166; 1.24 (1.00 to 1.52)	71; 1.13 (0.83 to 1.55)	20; 1.28 (0.70 to 2.35)	37; 2.35 (1.39 to 3.99)	19; 2.96 (1.34 to 6.55)
Current smoker	12 669	627; 2.78 (2.43 to 3.18)	349; 2.70 (2.28 to 3.25)	134; 2.21 (1.69 to 2.89)	28; 1.86 (1.07 to 3.26)	62; 4.07 (2.50 to 6.63)	54; 8.81 (4.35 to 17.80)
Alcohol intake (g/day):							
<5	32 148	977; 1.26 (1.10 to 1.44)	548; 1.35 (1.13 to 1.62)	232; 1.39 (1.05 to 1.83)	47; 0.79 (0.47 to 1.30)	89; 0.99 (0.66 to 1.48)	61; 1.12 (0.68 to 1.86)
5-25	11 057	287; 1	151; 1	62; 1	22; 1	32; 1	20; 1
>25	191	6; 1.20 (0.53 to 2.68)	4; 1.51 (0.56 to 4.07)	0	1; 2.60 (0.35 to 19.30)	0	1; 2.85 (0.38 to 21.20)
Hypertension:							
No	39 388	1001; 1	547; 1	237; 1	56; 1	103; 1	58; 1
Yes	4 008	269; 2.50 (2.19 to 2.86)	156; 2.64 (2.21 to 3.15)	57; 2.23 (1.67 to 2.98)	14; 2.34 (1.30 to 4.21)	18; 1.69 (1.03 to 2.80)	24; 3.86 (2.40 to 6.22)
Total	43 396	1 270	703	294	70	121	82

*As derived from Poisson regression models adjusted for attained age.

Table 2| **Daily dietary intakes of 43 396 women in Swedish Women's Lifestyle and Health Cohort**

Dietary component	Mean	SD	10th centile	1st quartile	Median	3rd quartile	90th centile
Energy (kJ)	6534	1868	4241	5208	6390	7705	9053
Carbohydrates (g)	196.6	60.0	123.7	154.7	192.1	233.3	275.2
Protein (g):	62.9	19.0	39.9	49.7	61.4	74.2	87.9
Animal protein (g)	42.3	15.5	24.0	31.6	40.9	51.1	62.2
Vegetable protein (g)	20.6	6.5	12.8	16.0	20.0	24.6	29.2
Total lipids (g):	54.7	19.7	31.2	40.7	52.6	66.4	80.9
Saturated lipids (g)	24.7	10.0	13.1	17.6	23.4	30.5	38.0
Unsaturated lipids (g)	25.8	8.9	15.1	19.6	25.0	31.2	37.5

Table 3| Distributions of 43 396 women, person time, and number of incident cases per diagnostic category by value in low carbohydrate-high protein score and corresponding crude incidence rates per 10 000 woman years in Swedish Women's Lifestyle and Health Cohort

	Low carbohydrate-high protein score*					
Diagnosis	**≤6**	**7-9**	**10-12**	**13-15**	**≥16**	**All women**
No of women	8 343	8 228	10 028	8 402	8 395	43 396
Woman years†	130 965	129 553	157 388	131 577	131 262	680 745
All cardiovascular disease:						
No of incident cases	188	211	279	290	302	1 270
Incidence rate	14.4	16.3	17.7	22.0	23.0	18.7
Ischaemic heart disease:						
No of incident cases	110	99	168	161	165	703
Incidence rate	8.4	7.6	10.7	12.2	12.6	10.3
Ischaemic stroke:						
No of incident cases	41	63	57	65	68	294
Incidence rate	3.1	4.9	3.6	4.9	5.2	4.3
Haemorrhagic stroke:						
No of incident cases	11	14	13	20	12	70
Incidence rate	0.8	1.1	0.8	1.5	0.9	1.0
Subarachnoid haemorrhage:						
No of incident cases	19	20	25	24	33	121
Incidence rate	1.5	1.5	1.6	1.8	2.5	1.8
Peripheral arterial disease:						
No of incident cases	7	15	16	20	24	82
Incidence rate	0.5	1.2	1.0	1.5	1.8	1.2

*Derived by summing position of woman in tenths of descending carbohydrate intake and tenths of ascending protein intake; score ranges from 2 (very low protein and very high carbohydrate intake) to 20.

†Calculated for all cardiovascular disease outcomes combined; essentially identical for each of indicated cardiovascular disease outcomes, because of their rarity.

Table 4| Incidence rate ratios for overall cardiovascular diseases and main diagnostic subcategories, per decreasing tenth of carbohydrate intake, increasing tenth of protein intake, and their addition in Swedish Women's Lifestyle and Health Cohort

	Incidence rate ratios* (95% CI)		
Condition (No of cases)	**Low carbohydrate score (per tenth)**	**High protein score (per tenth)**	**LCHP score (per 2 units)**
All cardiovascular diseases (1268)	1.04 (1.00 to 1.08)	1.04 (1.02 to 1.06)	1.05 (1.02 to 1.08)
Ischaemic heart disease (701)	1.04 (0.99 to 1.09)	1.03 (1.00 to 1.06)	1.04 (1.00 to 1.08)
Ischaemic stroke (294)	1.05 (0.98 to 1.14)	1.05 (1.01 to 1.10)	1.07 (1.00 to 1.13)
Haemorrhagic stroke (70)	1.00 (0.86 to 1.17)	1.05 (0.96 to 1.14)	1.05 (0.93 to 1.18)
Subarachnoid haemorrhage (121)	1.07 (0.95 to 1.21)	1.05 (0.98 to 1.12)	1.07 (0.97 to 1.17)
Peripheral arterial disease (82)	1.04 (0.90 to 1.21)	1.04 (0.95 to 1.13)	1.04 (0.93 to 1.17)

LCHP=low carbohydrate-high protein.

*Incidence rate ratios per indicated increase in corresponding score. Poisson models, using attained age as timescale in 2 year intervals, and adjusting for height (cm, continuously), body mass index (<25, 25-29.99, and ≥30, categorically), smoking status (never smokers, former smokers of <10 cigarettes, former smokers of 10-14 cigarettes, former smokers of 15-19 cigarettes, former smokers of ≥20 cigarettes, current smokers of <10 cigarettes, current smokers of 10-14 cigarettes, current smokers of 15-19 cigarettes, and current smokers of ≥20 cigarettes, categorically), physical activity (from 1 (low) to 5 (high), categorically), education (≤10, 11-13, and ≥14 years in school, categorically), diagnosis of hypertension (ever versus never), energy intake (per 1000 kJ/day, continuously), unsaturated lipid intake (per 10 g/day, continuously), saturated lipid intake (per 10 g/day, continuously), and alcohol intake (<5 g/day, 5-25 g/day, and >25 g/day, categorically).