Reproductive and dietary determinants of the age at menopause in EPIC-Heidelberg

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1. Introduction

The menopausal status is an important determinant for the development of some tumours as well as of other chronic diseases [1]. There is evidence that the age at natural menopause is inversely related to all-cause and ischemic heart disease mortality [2,3]. In addition, early menopause promotes bone loss. On the other hand, early menopause appears to have a strong protective effect on breast cancer, which is attributed to changes in the circulating oestrogen levels [4]. Thus, age at menopause

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has implications for the health of postmenopausal women.

The estimated average age at natural menopause in Western countries is between 50 and 51 years [5]. The cessation of menses is a consequence of the loss of ovarian function. At birth the number of follicles is in large part determined, and thereafter the number of follicles declines bi-exponentially [6]. At the age of 45–46 years a critical number of a few thousands follicles is reached [7]. The association between the family history of early menopause and age at menopause suggests involvement of a genetic component on the age at menopause [8]. The period preceding the natural menopause may generally be linked to reproductive aging, and thus serves as retrospective indicator for reproductive events, such as subfertility and the end of fertility [9].

Due to the association of menopause with the risk of chronic diseases, determinants of the onset of natural menopause are of interest. Smoking and extreme malnutrition [10] were consistently found to be associated with an earlier age at menopause [11]. In addition, low social economic class [12,13] was associated with younger age at menopause (reviewed in 1), however, the data were inconclusive. The results regarding oral contraceptives (OC) use are conflicting, since an association with an older age at menopause [14], younger age at menopause [31] or no effect on menopause was reported [13,42]. Studies on diet-related factors and onset of menopause are scarce, though an influence of dietary factors on the onset of menopause is biological plausible. A cross-sectional study in Japanese women found a higher intake of fat, cholesterol and coffee to delay menopause [15]. A prospective study revealed an inverse association between green and yellow vegetable intake and age at menopause [16]. Torgerson et al. found in a cross-sectional study that higher alcohol and meat intake was related to older age at menopause [17]. Compared to meat-eaters for vegetarians lower oestrogen levels were measured which may lead to an earlier onset of menopause [18].

Given the importance of age at menopause determinants in women's health, and the limited data on the role of dietary factors in this relationship, we investigated the influence of reproductive and dietary factors on the age at menopause in a large-scale prospective study.

2. Material and methods

2.1. Study population

EPIC-Heidelberg is a prospective cohort aiming at the investigation of the association between diet, lifestyle and chronic diseases with emphasis on cancer. The study was conducted in Heidelberg and comprises about 25,500 subjects, including 13,612 women aged 35–65 years at recruitment (June 1994 until October 1998) [19,20]. EPIC-Heidelberg contributes to the European prospective investigation into cancer and nutrition (EPIC). The baseline assessment of the cohort included a self-administered food frequency questionnaire (FFQ) [21,22] as a dietary assessment tool, a computerized lifestyle interview and standardized anthropometric measurements [23]. Subjects were recruited in the general population of Heidelberg and surrounding communities. In women aged 35–65 years the overall participation rate was 40% [19]. The response rate of the first follow-up, which took place between 1998 and 2000, was 93.5%. The second follow-up with a response rate of 91.6% was completed in 2004. Selection factors of the cohort recruitment are described in detail elsewhere [19]. Briefly, comparison with the reference population showed that more women with higher educational level were included in the study. Additionally, the proportion of smokers and ex-smokers was higher and the frequency of obesity was lower than in the total population [19].

2.2. Follow-up and outcome

At baseline, 6098 women without self-report of cancer, hysterectomy or ovarectomy (uni- or bilateral) were menstruating. Women with extreme values of total energy intake (below 1000 or above 7500 kcal/day) were excluded. At the time of the analysis 5568 women had been consequently followed-up, since the time of enrolment into the study and 5511 women provided data on menopausal state. Women who experienced menopause were identified through comparison of baseline and follow-up data. From the analysis we excluded 324 women who underwent hysterectomy, ovarectomy (uni- and bilateral) or the combination of both, and 77 women with missing values, leaving 5110 women in the study. During a median follow-up time of 5.8 years, 1009 women (21%) expe-

rienced natural menopause, which was defined as at least 12 consecutive months of amenorrhea not due to surgery [24]. These were compared with 3798 women who remained premenopausal. The remaining 303 women who experienced amenorrhoe for less then 12 menses during the past year were considered to be in perimenopausal state. Age at menopause was the main outcome variable.

2.3. Covariates

Education was categorized as low (up to 9 years of schooling without a specific vocational training), middle (high school degree (10 years) and vocational or technical training) and high (13 years of high school or a university degree). Participants were categorized as non-smokers, ex-smokers, or current smokers at baseline. Body mass index (BMI) was calculated as weight (kg)/height (m^2) and classified in <25, 25–29.9 and ≥30 kg/m^2. Leisure time physical activity was classified as none, 0.1–2, 2–5 and >5 h/week.

The number of full term pregnancies was classified as nullipara, 1–2 and ≥3 births at recruitment and the age at first full term pregnancy as <20, 20–26, 27–31 and ≥32 years. The time of breast-feeding was summarized across pregnancies and classified as none, 1–5, 6–11 and ≥ 12 months. Age at menarche (<11, 12, 13, 14 and ≥15 years), and time between menarche and regular menses (<1, 1–2, 3–4 and ≥5 years) were also considered in the analysis. Exposure to exogenous hormones was classified as ever-use of oral contraceptives and ever-use of hormone replacement therapy (HRT).

Dietary intake at food group level and alcohol consumption was calculated based on the data collected with a self-administered and validated food frequency questionnaire. Nutrient intake data were calculated from the food intake data by means of the German Food Composition Table BLS, Version II.3 [25]. The quartile cut-points were based on consumption levels among the subjects in the analyses. Alcohol consumption was expressed as ethanol intake in grams per day.

2.4. Statistical methods

Medians and percentiles were calculated using the SAS procedure UNIVARIATE. Associations between age at menopause and subjects' characteristics were evaluated by either Chi Square Test or Wilcoxon test (SAS procedure NPAR1WAY).

Using the product limit life table method, Kaplan Meier survival curves were plotted separately for each variable and Log Rank tests were applied. Cox proportional hazard regression models were fitted for each nutrient to identify predictors of the age at natural menopause [26]. Regression models included age, education, age at menarche, OC-use, HRT-use, parity, BMI, time of breast feeding, age at first full term pregnancy, smoking habit, ethanol intake and leisure time physical activity. In addition, the results were adjusted for total energy intake in order to control partially for the error of the estimated nutrients because of the high correlation between dietary components. The adjusted hazard ratios (HR) and the corresponding 95% confidence intervals (95% CI) were calculated using the PROC PHREG procedure of the statistical software package SAS release 8.2 (SAS Institute, Cary, NC, USA). Tests for linear trend with two-sided p-values were calculated by using the median values for each category as continuous exposure score.

3. Results

During the follow-up of the cohort, 1009 women (mean age at menopause 51.3 years S.D. 4.7 years) experienced natural menopause and were compared with 3798 women who remained premenopausal. Another 303 women remained in perimenopausal state and were excluded from the analysis. The median follow-up time for both groups was 5.8 (5.1–9.2) years. Table 1 shows the HR for reproductive and lifestyle factors. Increasing age at first full term pregnancy was associated with delayed onset of menopause, although the test of trend was not statistically significant. A longer interval between menarche and the onset of regular menses was associated with later menopause (p for trend 0.012). Compared to nullipara, women with 1–2 children experienced menopause at older age (HR: 0.74, 95% CI: 0.62–0.87). Breast-feeding for 1–5 months was associated with older age at menopause (HR: 0.81; 95% CI: 0.69–0.94). Smoking at enrolment in the study was related to younger age at menopause (HR: 1.40, 95% CI: 1.13–1.58), whereas for ex-smokers no effect on the age at menopause was found (HR: 1.01, 95%CI: 0.86–1.17). Educational

Table 1
Subjects' characteristics of women who became postmenopausal during follow-up ($N = 1009$) and who remained in premenopausal state ($N = 3789$) and age-adjusted hazard ratios (HR) and 95% confidence intervals (95% CI) for younger age at menopause

	Postmenopausal state ($N = 1009$)		Premenopausal state ($N = 3798$)		Age at menopause[a] (post vs. premenopausal state)		
	N	%	N	%	HR	95%CI	P for trend
Educational level[*]							0.90
High (university)	70	6.94	194	4.74	1		
Medium (professional training)	639	63.39	2271	59.79	1.05	0.81–1.37	
Low (no professional training)	299	29.66	1333	35.10	0.99	0.75–1.30	
Parity[*]							0.56
Nulliparous	207	20.56	971	25.57	1		
1, 2 children	662	65.74	2345	61.76	0.74	0.62–0.87	
≥3 children	138	13.70	481	12.67	0.83	0.66–1.04	
Age at first full term pregnancy/years[**]							0.11
<20	86	10.72	154	5.45	1		
20–26	371	46.26	1095	38.73	0.76	0.59–0.98	
27–31	233	29.05	1046	37.00	0.72	0.55–0.94	
≥32	112	13.97	532	18.82	0.69	0.51–0.94	
Breast feeding/months[**]							0.68
None	360	35.68	1295	34.10	1		
1–5	403	40.24	1088	28.65	0.81	0.69–0.94	
6–11	115	11.40	519	13.67	0.83	0.66–1.03	
≥12	128	12.69	869	23.59	0.92	0.74–1.14	
OC-use (ever)							
No	106	10.51	344	9.06	1		
Yes	903	89.49	3453	90.94	0.91	0.74–1.13	
HRT-use (ever)[**]							
No	698	69.11	2677	92.60	1		
Yes	312	30.89	214	7.40	0.96	0.81–1.14	
Age at menarche/years							0.80
≤11	138	13.69	530	14.04	1		
12	266	26.39	989	26.67	1.09	0.88–1.36	
13	297	30.39	1189	30.68	0.98	0.80–1.22	
14	190	29.46	682	18.23	0.90	0.71–1.13	
≥15	117	18.85	407	10.38	0.88	0.68–1.14	
Time till regular menses/years[**]							0.01
<1	682	67.66	2154	56.74	1		
1–2	221	21.92	1094	28.82	0.88	0.75–1.03	
3–4	48	4.76	225	5.93	0.82	0.60–1.13	
≥5	57	5.65	323	8.51	0.67	0.50–0.89	
Leisure time physical activity h/week							0.10
None	380	37.77	1417	37.34	1		
0, 1–<2	276	27.44	1032	27.19	1.08	0.84–1.39	
2–<5	269	26.74	1064	28.04	1.20	0.92–1.55	
>5	71	8.05	282	7.43	1.17	0.90–1.51	
Smoking status							0.32
Never	441	43.75	1552	40.87	1		
Ex	321	31.85	1214	31.99	1.01	0.86–1.17	
Current	246	24.40	1030	27.14	1.40	1.13–1.58	

Table 1 (*Continued*)

	Postmenopausal state (*N* = 1009)		Premenopausal state (*N* = 3798)		Age at menopause[a] (post vs. premenopausal state)		
	N	%	*N*	%	HR	95%CI	*P* for trend
Body mass index (BMI)**							0.33
<25	607	60.16	2656	60.93	1		
25–29.9	276	27.35	809	21.30	1.00	0.86–1.16	
≥30	126	12.49	333	8.77	1.18	0.96–1.45	

Chi Square test: $p < 0.05$.

[a] The models included as adjustment variables: age (continuous).

* $p < 0.01$.

** $p < 0.001$.

level, BMI, leisure time physical activity, OC-use, HRT-use and age at menarche had no effect on age at menopause.

Table 2 shows HRs for selected dietary factors. Increasing carbohydrate consumption was associated with younger age at natural menopause, but the test for trend failed statistical significance (*p* for trend 0.119). The third quartile of vegetable intake compared to the lowest intake group (first quartile) was associated with earlier menopause (HR: 1.32, 95% CI: 1.09–1.60). High intake levels of cereal products, fibre and soy products were associated with earlier menopause and the test of trend nearly reached statistical significance (*p* for trend 0.06, 0.16 and 0.06, respectively). High total fat consumption was associated with delayed menopause (third quartile: HR: 0.78, 95% CI: 0.59–0.79). Additionally, increasing meat intake was associated with later menopause (*p* for trend = 0.046), as was in tendency protein intake (*p* for trend = 0.086). The consumption of dairy products, sweets, added vegetal fat, added animal fat and intake of fish and alcohol was not associated with the age at menopause.

4. Discussion

In our study sample, later natural menopause was associated with increasing age of first full term pregnancy and increasing time between menarche and the onset of regular cycles. Compared to never smokers, current smokers experienced menopause at younger age. Among the dietary variables, the consumption of carbohydrates, vegetables, fibre, soy products and cereal products was associated with younger age at

menopause, whereas increasing fat, meat and protein consumption was associated with later menopause.

Among the reproductive variables older age at first full term pregnancy was associated with later menopause. Age at menarche was not significantly associated with the age at natural menopause. Previous data on the association between age of menarche and menopause have been inconsistent. Whereas some authors reported early menarche to be associated with early menopause [27], others failed to find an association [13,14]. Later menopause was associated with the time till occurrence of regular menses after menarche. Our data suggest that the interval until regular occurrence of menstrual cycles had a stronger impact on age at menopause than menarche itself. It can be speculated that anovulatory cycles increase the remaining follicle pool. Between parity, time of breast feeding and age at menopause we found no association in contrast to most previous work reporting that nulliparous women experience menopause at older age [12,17]. The inclusiveness of the data on reproductive factors and age at menopause may be caused by different adjustment approaches between the studies regarding the selection and classification of covariates.

For the ever-use of oral contraceptives we found no association with age at menopause. As the decline of follicle stimulation hormone (FSH) results in delayed age at menopause, the use of OC, which suppress FSH levels, may lead to a postponed onset of menopause [28,29]. However, the literature on prior OC-use and the age at natural menopause is conflicting, since some authors found a delaying effect [28], whereas others observed no effect on menopause [12,30] or an earlier onset of menopause [11]. de Vries et al. found for the use of high dose OCs over at least 3 years an increased

Table 2
Distribution of nutrients and among pre- and postmenopausal women and adjusted hazard ratios (HR) for younger age at menopause and corresponding 95% confidence intervals (95% CI)

Nutrient in g/day	Postmenopausal		Premenopausal		Age at menopause, adjusted				P for linear trend across quartiles
	Median	75 percentile	Median	75 percentile	1. Quartile lowest	2. Quartile	3. Quartile	4. Quartile highest	
Total fat	66.4	82.8	68.8	85.9					0.153
Cases					313	263	224	209	
HR					1	0.87	0.78	0.79	
(CI)						0.72–1.04	0.65–0.97	0.59–1.07	
Protein	60.9	71.8	60.5	73.1					0.086
Cases					272	252	267	218	
HR					1	0.99	0.97	0.78	
(CI)						0.82–1.19	0.79–1.19	0.59–1.04	
Carbohydrates	186.3	230.5	188.6	233					0.119
Cases					270	261	261	217	
HR					1	1.30	1.44	1.47	
(CI)						1.07–1.58	1.15–1.80	1.07–2.02	
Alcohol	6.01	15.7	8.06	17.3					0.289
Cases					225	226	271	287	
HR					1	0.91	1.02	0.83	
(CI)						0.75–1.11	0.84–1.24	0.69–1.01	
Meat	67.4	94.8	65.9	98.2					0.046
Cases					244	261	288	216	
HR					1	0.99	0.91	0.75	
(CI)						0.82–1.20	0.75–1.10	0.61–0.93	
Dairy products	209.7	310	203.5	309.2					0.998
Cases					254	249	248	258	
HR					1	1.15	1.12	1.04	
(CI)						0.95–1.39	0.92–1.35	0.86–1.27	
Fish	15.1	23.0	14.8	22.5					0.163
Cases					243	218	280	268	
HR					1	1.09	1.14	1.14	
(CI)						0.90–1.33	0.94–1.37	0.95–1.38	
Vegetables	112.2	145.9	107.7	143.4					0.427
Cases					242	240	269	258	
HR						1.11	1.32	1.18	
(CI)						0.91–1.34	1.09–1.60	0.97–1.43	

	Q1	Q2	Q3	Q4	p
Fruit					0.477
Cases	212	260	265	272	
HR	1	1.08	1.14	1.08	
(CI)		0.89–1.32	0.94–1.14	0.88–1.32	
Median	129.4	214.1	126.3	207.3	
Cereal products					0.059
Cases	285	240	253	231	
HR	1	1.09	1.23	1.25	
(CI)		0.90–1.31	1.02–1.48	1.02–1.53	
Median	166.7	211.5	169	219.8	
Fibre					0.158
Cases	261	252	228	268	
HR	1	1.24	1.22	1.30	
(CI)		1.03–1.50	0.99–1.50	1.03–1.65	
Median	18.7	23.6	18.6	23.1	
Soy products					0.060
Cases	313	261	233	202	
HR	1	0.92	0.96	1.08	
(CI)		0.77–1.10	0.79–1.15	0.89–1.31	
Median	0.1	0.3	0.2	0.4	
Sweets					0.697
Cases	283	263	236	227	
HR	1	1.21	1.10	1.12	
(CI)		1.00–1.45	0.91–1.34	0.88–1.41	
Median	79.4	127.3	83.8	131.9	
Added animal fat					0.466
Cases	284	246	247	232	
HR	1	0.92	0.98	1.03	
(CI)		0.77–1.11	0.81–1.18	0.85–1.24	
Median	8.3	15.4	10.2	17.6	
Added vegetable fat					0.498
Cases	258	254	238	259	
HR	1	1.05	0.96	0.87	
(CI)		0.87–1.27	0.79–1.16	0.72–1.05	
Median	8.9	13.7	9.0	13.9	

The model included as adjustment variables: age (years, continuous), total energy intake (kcal/day, continuous), educational level (low, medium, high), body mass index (<25, 25–29.9 and ≥30 kg/m^2), leisure time physical activity (0, 1–<2, 2–<5 and ≥5 h/week), alcohol intake (none, 0.1–<2, 2–<5 and ≥5, >5–18 and >18 ethanol g/day), smoking (never, ex and current), number full term pregnancies (0, 1–2 and >3), age at menarche (>11, 11, 12, 13, 14 and ≥15 years), time till regular menses occurred after menarche (<1, 1–2, 3–4 and ≤5 years), age at first full term pregnancy (<10, 20–26, 17–31 and ≥32 years) ever HRT-use (yes and no).

risk of earlier age at menopause [31], implicating that a dose-depended relationship exists.

Our results that current smoking is associated with earlier age at menopause are in agreement with former findings [12,17,32]. In several studies a dose–response trend for earlier menopause was observed with increasing number of cigarettes [33,34]. In animal model systems, it was shown that polycyclic aromatic hydrocarbons, as a toxic component of cigarette smoke, destroy primordial oocytes [35]. This mechanism, mediated by an up-regulation of the Bax pro-apoptosis gene, results in follicle damage and premature ovarian failure [36]. Other smoke related components might decrease oestrogen production or shift the oestrogen pathway towards 2-hydroxylation leading to less active oestrogens [37,38].

Former smokers have only a slightly earlier natural menopause, indicating that the toxic effect of smoking may not be permanent, but related to exposure that increases the intrafollicular oxidative stress resulting in impaired folliculogenesis and decreased follicular density in smokers [32,39,40].

In our data we found no clear association between the consumption of alcohol and the age at menopause. Torgerson et al. observed among women aged 45–49 years a delay of menopause with increasing alcohol consumption [17]. This effect may be caused be the estrogenic effect of alcohol at moderate level of consumption. However, a study dealing with hormonally determined menopause on the basis of FSH measurements did not reveal an association with alcohol intake [41]. The overall effect of alcohol intake on menopause may be due to different potential mechanisms possibly also depending on the types alcoholic beverages consumed.

In the present study, population no effect of the BMI on the age at menopause was observed. In general, the results on this association are not univocal [16,42,43]. Our observation is consistent with former data [12,28], but also an association with younger age at menopause was reported [44]. Obese women are characterized by a more estrogenic environment due to the conversion of androstenedione in oestrone and an increased level of free oestradial due to decreased sex hormone binding globulin. The resulting higher levels of free oestradiol may lead to a delayed menopause. Recent results on pre- and perimenopausal women enrolled in the SWAN study showed in that BMI is strongly related to menstrual cycle characteristics, since whole cycle hormone levels are lower in obese women [45].

In our study leisure time physical activity was not associated with age at menopause. Vigorous exercise is known to suppress ovarian function by lowering serum estrogens and increasing sex hormone binding globulin [46]. A recent intervention study among postmenopausal women found in the exercise group a decline in the serum oestrone, oestradiol and free oestradiol levels [47], indicating that prior intensive sport activities may lead to an earlier age at menopause.

Diet influenced the onset of natural menopause. Among vegetarians, menopause occurred earlier. This may be linked to increased fibre intake or decreased animal fat intake, which may alter the levels of the luteinizing hormone (LH) and follicle stimulating hormone and the length of the menstrual cycle [48]. In consistency with a cross-sectional study among Japanese women [15], we found high total fat intake associated with delayed menopause. An intervention study on the reduction of dietary fat intake has shown a reduction of serum oestradiol levels in pre- and postmenopausal women [49]. Since dietary fat intake increases the β-glucoronidase activity in the intestinal microflora, the reabsorption of oestrogens may be increased [50], which may postpone the onset of menopause. In our study, women with higher meat consumption tended to experience menopause at older age, which is consistent with a former result of Torgerson et al. [17]. The association between increasing protein intake and delayed onset of natural menopause in our analysis is not consistent with findings from other studies [15,16].

In the present study, increasing carbohydrate consumption was associated with earlier age at menopause, whereas other authors did not find an association [16]. Carbohydrates may lead to dyslipidemia and insulin resistance via glucose levels [51]. In case of insulin resistance an increase of serum oestrogen levels and decrease of sex hormone binding globulin levels can be anticipated, which may lead to an older age at menopause. However, the effect on the postprandial glucose and insulin response may depend on the type of carbohydrate and the physical properties of specific food items [52]. Since in the EPIC-Heidelberg cohort, main sources of carbohydrates in women are bread, potatoes, fruit and vegetables [53], the observed effects for carbohydrates may be explained by the food compo-

nents, which is supported by our findings on vegetable, cereal products and fibre intake.

High vegetable intake was associated with younger age at menopause in our data. This is in line with results presented by Nagata et al. who observed in Japanese women an association between the consumption of green and yellow vegetables and earlier age at menopause [16]. Antioxidants provided by vegetables may reduce oxidative stress, which inhibits steroidogenesis in granulosa cells and induce atresia of ovarian follicles [54]. However, also other nutrients, such as minerals or phytochemicals, may contribute to the observed effect. For increasing intake of soy products we observed an inverse relationship with the age at menopause, but the association failed to reach significance. In Germany, the consumption of soy products may be too low to reveal a potential effect. Nagata et al. observed in a cross-sectional study in Japanese women an inverse association between soy intake and later menopause [15], which was not found in a later prospective study in Asia [16]. However, dietary intervention with soy products resulted in decreased serum oestradiol levels [55].

The observation that high meat and fat intake were associated with delayed menopause, whereas high vegetable, fibre and cereal products consumption correlated with earlier menopause seems to contradict the inverse association between these dietary factors and mortality. The hormonal changes at menopause are mainly attributed to oestrogen depletion suggesting a stronger impact of hormone related dietary factors in the premenopause. The cessation of menstruation is considered to lead to an accretion of iron body stores and the occurrence of a more atherogenic blood fat profile, which may increase cardiovascular disease (CVD) risk. Due to the physiological changes following menopause, the effects of diet may be mediated differently in pre- and postmenopausal women. Consequently, dietary determinants impacting on the age at natural menopause and the age at natural all-cause or CVD mortality may differ.

Dietary intake data may be prone to measurement error. The food frequency questionnaire used in this prospective study was validated by comparison with results of 24 h dietary recalls obtained from the same individual [56]. There is evidence that habitual dietary and lifestyle habits appear to be rather stable in this age range [57], since the recall of the past diet may

be seen as a function of consistency in diet over time [58]. This is strongly supported by the preliminary results comparing the results of the first FFQ at baseline (1994–1998) with the second FFQ (2001–2004). We obtained only marginal differences at the food group or nutrient level within subjects.

However, the strength of the present study is its prospective design so that misclassification is likely to be non-differential with respect to future menopausal status. In the present study, educational level was not associated with age at natural menopause, however, socio-economic circumstances beyond the scope of this investigation may have influenced the age at menopause [59]. Subjects were recruited from the general population. There was concern about the participation of more educated (high school: 27% versus 4%) subjects, more current smokers (23% versus 14%) and less obese women (BMI > 35: 5% versus 7%) than in the underlying population [19]. This might result in overestimation of current smoking and underestimation of the educational level and obesity on the age at menopause. In cohort studies bias resulting from differential selection at the start of follow-up can be viewed as confounding [60]. In the analyses of dietary factors potential confounding factors were considered. Further adjustment of the reproductive variables did not change the risk estimates substantially, therefore age-adjusted estimated are presented. Caution is needed in generalizing the results. However, the prospective study design with almost completeness of follow-up and the control of a wide range of potential confounders strengthen our results. Though the age at menopause was self-reported, the follow-up time intervals of 2–3 years may facilitate a timely recall of the onset of menopause. Due to the number of statistical tests carried out, the results have to be interpreted with caution, since positive associations might have been generated by chance only.

In conclusion, the results of this prospective study corroborate the well-established association between current smoking and younger age at menopause. Older age at the first full term pregnancy and the time until regular menses occurred after menarche were associated with older age at menopause. Moreover, associations between dietary factors and the age at natural menopause were observed. In particular, high intake of carbohydrates, vegetables, fibre and cereal products was associated with younger age at natural menopause, whereas women with higher total fat, protein and

meat intake experienced a delayed onset of natural menopause. Further studies including biological markers are needed to clarify these associations.

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