Triebner 2016

Menopause is associated with accelerated lung function decline

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Triebner 2016

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Triebner 2016

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Triebner 2016

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At a Glance Commentary

The present study is the first to investigate the effect of menopause on lung function in a large, longitudinal and population-based survey. We analyzed the decline of forced vital capacity and forced expiratory volume in one second as related to menopausal status. The study population is a hormonally well-defined group of women, aggregated by latent class analysis on measurements of follicle stimulating hormone and luteinizing hormone as well as questionnaire data. Linear mixed effects models showed that postmenopausal women and women in the menopausal transition are associated with a steeper decline in lung function, beyond the age related decline, compared to nonmenopausal women. Our findings are relevant for clinical practice and health care professionals should be aware that women undergoing the menopausal transition are at risk of accelerated lung function decline.

Online data supplement

This article has an online data supplement, which is accessible from this issue's table of content online at www.atsjournals.org

Triebner et al   1

Abstract

Rationale: Menopause is associated with changes in sex hormones, which affect immunity, inflammation, and osteoporosis and may impair lung function. Lung function decline has not previously been investigated in relation to menopause.

Objectives: To study whether lung function decline, assessed by forced vital capacity and forced expiratory volume in one second, is accelerated in women who undergo menopause.

Methods: The population-based longitudinal European Community Respiratory Health Survey provided serum samples, spirometry and questionnaire data about respiratory and reproductive health from three study waves (N=1438). We measured follicle stimulating hormone and luteinizing hormone and added information on menstrual patterns, to determine menopausal status using latent class analysis. Associations with lung function decline were investigated using linear mixed effects models, adjusting for age, height, weight, packyears, current smoking, age at completed full-time education, spirometer and including study center as random effect.

Measurements and Main Results: Menopausal status was associated with accelerated lung function decline. The adjusted mean forced vital capacity decline was increased by -10.2 ml/yr (95% Confidence interval -13.1 to -7.2) in transitional women and -12.5 ml/yr (-16.2 to -8.9) in postmenopausal women, compared to women menstruating regularly. The adjusted mean forced expiratory volume in one second decline increased by -3.8 ml/yr (-6.3 to -2.9) in transitional women and -5.2 ml/yr (-8.3 to -2.0) in postmenopausal women.

Triebner et al   2

Conclusions: Lung function declined more rapidly among transitional and postmenopausal

women, in particular for forced vital capacity, beyond the expected age change. Clinicians

should be aware that respiratory health often deteriorates during reproductive aging.

Key words

Latent class analysis; lung function decline; menopause; reproductive aging; sex hormones;

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Abbreviations used

BLUP: Best Linear Unbiased Prediction

BMI: Body Mass Index

CI: Confidence Interval

FEV_1: Forced Expiratory Volume In One Second

FSH: Follicle Stimulating Hormone

FVC: Forced Vital Capacity

LH: Luteinizing Hormone

OBS: Observations

SD: Standard Deviation

INTRODUCTION

The non-reproductive phase of a woman's life begins with menopause (1). Cessation of menstruations may be accompanied by lack of energy, hot flushes, vaginal discharge, insomnia, osteoporosis and an increased risk of developing chronic conditions like diabetes and cardiovascular disease (2-6). Menopause is associated with profound changes in the activity of the hypothalamo-pituitary-gonadal axis: as 17β-estradiol production in the ovaries ceases, follicle stimulating hormone (FSH) and luteinizing hormone (LH) concentrations rise (7, 8). Low levels of 17β-estradiol are associated with increased systemic inflammation and inflammation in the lungs (9-16). The inflammation markers C-reactive protein and Interleukin-6 are inversely associated with forced vital capacity (FVC) and forced expiratory volume in one second (FEV_1) (17, 18). These findings point towards a possible association between menopause and increased lung function decline. Given today's life expectancy far beyond the age of menopause, understanding how to maintain good health and quality of life in later years is important (1, 19-22). The role of sex hormones in respiratory health is increasingly acknowledged (23-32). However, potential changes in respiratory health as related to reproductive aging in women are still poorly understood (23, 33, 34). A recent longitudinal analysis of a large population based survey showed higher incidence of new-onset asthma and increase in respiratory symptoms as related to menopause (28). Potential changes in lung function with menopause have only been addressed in cross-sectional analyses: One population-based study of women aged 45 to 55 years, not using hormone replacement therapy, found lower lung function in postmenopausal women compared to women who still had regular menstruations (27). A recent study from the UK Biobank found an association between cessation of menstruation and lower lung function, whereas another study saw no independent effect of menopause on FVC and FEV_1 levels (35, 36). It has been reported that lifestyle factors such as smoking may be more detrimental after menopause,

Triebner et al   4

while some hormone replacement therapy regimens appear to be associated with higher lung

function in postmenopausal women (37-39). Whether obesity impacts on respiratory health in

menopause is not known, although sex hormones and body fat mass are interrelated (40, 41)

and effect modification of BMI on the role of hormone replacement therapy is suggested in

cross-sectional studies (42). The aim of the present study was to investigate the association of

menopausal status with decline in lung function over a 20-year period, using data from a

large European, population-based cohort. Some of the results of this study have been

previously reported in the form of an abstract to the European Respiratory Society annual

congress (43).

American Journal of Respiratory and Critical Care Medicine

METHODS

Study population

The European Respiratory Health Survey (ECRHS) is an international multicenter study including three waves, ECRHS 1 in 1991-1994, ECRHS 2 in 1998-2002 and ECRHS 3 in 2010-2012, and is described in detail at http://www.ecrhs.org. The age range at baseline was 25 to 48 years and the median observation time for women participating in all three waves was 19.7 (17.9 - 22.4) years. The present study includes 19 centers in Europe (see Table E1 in the online data supplement). The examinations included an interviewer-led questionnaire, spirometry and serum sampling. At ECRHS 2 and 3 women additionally answered standardized questionnaires on women's health. For 2484 women, serum samples from ECRHS 2 and/or 3 were analyzed for FSH and LH (see online data supplement). Inclusion criteria are presented in Figure 1. Ethical approval was obtained from the appropriate ethics committee and each participant provided informed written consent.

Figure 1. Study population with inclusion criteria (*height and packyears)

Classification according to menopausal status

Every woman contributed with up to three observations (ECRHS 1, 2 and/or 3, N_{obs}=3295). We attributed each observation separately to a menopausal status, at the same time as the pulmonary function tests were conducted. At baseline we included only nonmenopausal women, excluding women who retrospectively reported their last period more than one year before baseline. To determine menopausal status at ECRHS 2 and 3 we performed latent class analysis in order to identify subgroups within our population, based on menopause related factors such as hormone levels and presence and/or regularity of menstruations (44, 45). Latent class analysis is commonly used in medical research to classify subjects by selected characteristics (>2000 publications listed on www.PubMed.gov). The number of

Triebner et al 6

classes was restricted to three, retrospectively labeled: nonmenopausal, transitional and postmenopausal. Observations were assigned class-memberships based on response probabilities (see Table E2 in the online data supplement). The algorithm, grouping the most similar observations, was repeated 30 times with a maximum of 10.000 iterations. The included categorical variables were FSH in quintiles, LH in quintiles and menstrual status as reported by participants (see Table E2 in the online data supplement). Each participant was present either in one, two or all three classes, representing the reproductive stage at different waves.

Outcomes

FVC and FEV_1 were recorded following standardized methods (46, 47). The spirometers that

were used are presented in Table E1 in the online data supplement.

Statistical analysis

Data of all three waves was modeled by linear mixed effects models, allowing for random intercepts (48). By including spirometric measurements and an interaction term of the menopausal status with age, we determined the lung function decline defined as the slope of change in lung capacity, which can be interpreted as the mean annual decline [ml/yr] associated with each menopausal status. We adjusted for fixed effects of age, weight, packyears, current smoking and type of spirometer at each wave, as well as height at baseline and age at completed full time education as a socioeconomic proxy, which if missing (6.0%) was imputed as the population mean. Missing age (3.8%) and weight (9.4%) were included as age and weight at a different wave plus the mean increase during follow-up. The participant's id number, nested in the respective study center, was accounted for as random effect. Stratified analyses were performed among never-smokers, ever-smokers and BMI categories

according to the World Health Organization. To investigate whether results were driven by a history of asthma, or gynecological disorders such as polycystic ovary syndrome, endometriosis, surgical menopause (reported at ECRHS 2 or 3) or premature menopause (<40 years), we performed sensitivity analysis within the respective subgroups without these conditions. We further tested potential heterogeneity between study centers and an alternative model, accounting for change in height over the study period. Analyses were performed using R (Version 3.1.0, The R Foundation for Statistical Computing).

Triebner et al   8

RESULTS

Characteristics of the subjects at baseline are presented in Table 1. The class conditional

probabilities and standard errors for the class membership, determined by latent class

analysis, can be found in Table E2 in the online data supplement.

Table 1. Characteristics of the study population at baseline (n=1369[1])

	Median (5th, 95th percentile)
Age [yr]	36.2 (26.8, 45.0)
Height [m]	1.64 (1.52, 1.75)
Weight [kg]	61 (50, 85)
BMI [kg/m^2]	22.6 (18.8, 31.2)
Packyears	1.0 (0.0, 24.0)
Current smoking, n (%)	438 (32)
Year of PFT[1]	1992 (1991, 1993)
Observation time [yr]	19.7 (18.5, 21.0)
FVC [L]	3.85 (2.94, 4.80)
FEV$_1$ [L]	3.19 (2.37, 3.96)
Delta FVC[2] [ml/yr]	25.8 (2.4, 53.6)
Delta FEV$_1$[2] [ml/yr]	33.4 (10.7, 56.9)

[1]N$_{total}$=1438, baseline observations for 69 women were incomplete and therefore excluded (follow-up observations of these women were included)

[2] Pulmonary function test

[3]Annualized change over follow-up

The included 1438 women provided a total of 3295 complete observations (pulmonary

function tests) including data on menopausal status and covariates (up to three observations

per woman). The nonmenopausal class included 60% (n$_{obs}$=1992), the transitional class 18%

(n$_{obs}$=583) and the postmenopausal class 22% (n$_{obs}$=720) of the observations. The mean age

was 39 years in the nonmenopausal, 53 years in the transitional and 56 years in the

postmenopausal class (Table 2). Weight and BMI were highest in the transitional class (Table

2).

Triebner et al   9

Table 2. Characteristics of the observations by menopausal class, mean (SD)

	Nonmenopausal (n_{obs} =1992)	Transitional (n_{obs} =583)	Postmenopausal (n_{obs} =720)
Age [yr]	38.8 (6.8)	52.8 (8.2)	55.7 (5.4)
Height [m]	1.64 (0.07)	1.63 (0.07)	1.63 (0.07)
Weight [kg]	65.2 (12.6)	71.7 (15.2)	69.3 (12.5)
BMI [kg/m^2]	24.2 (4.5)	26.8 (5.4)	26.0 (4.4)
Packyears	6.2 (10.3)	9.2 (16.5)	9.5 (17.9)
Current smoking, n (%)	581 (29)	120 (21)	115 (16)
FVC [L]	3.81 (0.58)	3.41 (0.60)	3.35 (0.54)
FEV$_1$ [L]	3.11 (0.50)	2.64 (0.53)	2.55 (0.45)
FSH [IU/L]	9.3 (9.9)	56.8 (40.9)	146.9 (80.0)
LH [IU/L]	5.8 (5.7)	20.1 (10.0)	35.5 (11.0)

Figures 2 and 3 show the best linear unbiased predictions (BLUP´s) for FVC and FEV$_1$ of the

whole population, obtained by adding together the population predictions (based on the fixed

effects estimates) and the estimated contributions of the random effects (48). Both curves

show a negative increase in slope when a large proportion of women enter the transitional or

menopausal stage.

Figure 2. Best linear unbiased predictions for FVC of the study population (n=1438) according to the specified linear mixed effects model, including 95%CI and population distribution by menopausal class (The dotted line, as visual aid continues the initial slope)**.**

Figure 3. Best linear unbiased predictions for FEV$_1$ of the study population (n=1438) according to the specified linear mixed effects model, including 95%CI and population distribution by menopausal class (The dotted line, as visual aid continues the initial slope)**.**

The mean age dependent lung function decline for all groups was -15.9 ml/yr (95% CI -18.2

to -13.6) for FVC and -24.0 ml/yr (-25.9 to -22.0) for FEV$_1$. The interaction term of

menopausal status with age showed that the mean FVC decline was additionally increased by

-10.2 ml/yr (-13.1 to -7.2) for women classified as transitional and -12.5 ml/yr (-16.2 to -8.9)

for women classified as postmenopausal, compared to women menstruating regularly. The

mean FEV$_1$ decline additionally increased by -3.8 ml/yr (-6.3 to -2.9) for women classified as

transitional and by -5.2 ml/yr (-8.3 to -2.0) for women classified as postmenopausal.

Triebner et al 10

Current smoking was significantly associated with FEV_1 decline [-29.3 ml (-54.0 to -4.7)] compared to former smoking and lifelong non-smoking, whereas packyears were significantly associated with both FEV_1 decline [-2.3 ml/packyear (-3.0 to -1.5)] and FVC decline [-1.0 ml/packyear (-1.9 to -0.1)]. The results for other covariates of the primary analysis are presented in Tables E3 and E4 in the online data supplement.

Analysis of FVC decline as related to menopause and stratified according to BMI at ECRHS 3 showed a higher age related decline (Figure 4) and indicated a smaller menopause related decline with increasing BMI ($p_{interaction}$<0.01 for transitional women and $p_{interaction}$<0.001 for postmenopausal women) (Figure 5). Ever smokers indicated a higher age related decline and higher menopause related decline than never smokers. For FEV_1 we observed a similar but less distinct pattern (Figure 4 and Figure 5).

Figure 4. Age related lung function decline by smoking and BMI for the study population, ml/yr

Figure 5. Menopause related lung function decline by smoking and BMI for the menopausal class, ml/yr

The sensitivity analysis showed that the results were not driven by a history of asthma, by

gynecological disorders, by hypertension or cardiovascular disorders, or by surgical or premature menopause (data not given). Results were consistent for individual centers with sufficient participants (Galdakao, Albacete, Grenoble, Paris, Reykjavik, Bergen, Umeå and Uppsala).

Height changed over the study period. The age related decline in height was -0.23 mm/yr (95% CI -0.38 to -0.08). Women in the menopausal transition had an additional decline of -0.62 mm/yr (-0.79 to -0.05) and the postmenopausal class an additional decline of -0.77 mm/yr (-0.98 to -0.06). The alternative model accounting for change in height showed a 25% reduced, yet still significant, decline in FVC for both the transitional [-7.7 ml/yr (-10.6 to -4.7)] and the postmenopausal class [-9.0 ml/yr (-12.7 to -5.5)], compared to the

Triebner et al   11

nonmenopausal class. The decline for FEV_1 was reduced by circa 50% and not significant

after such adjustment (transitional class: -2.1 ml/yr (-4.7 to 0.3), postmenopausal class: -2.6

ml/yr (-5.8 to 0.5).

Triebner et al   12

DISCUSSION

This population-based study of 1438 women aged 25-48 years from nine European countries over 20 years with repeated spirometry, hormone measurements and questionnaire data, found that lung function declined more rapidly in women who were transitional or postmenopausal, as compared to women who were nonmenopausal. To our knowledge, this is the first longitudinal population-based study that investigates lung function decline in relation to menopause.

The menopause related decline was more pronounced for FVC than for FEV_1, which points towards a restrictive rather than an obstructive pattern. Stratified analyses and sensitivity analyses revealed that these findings were very robust. Effect modification by BMI seems plausible, as circulating androgens are converted to estrogens in adipose tissue. Ever smokers show a suggested steeper decline in both age and menopause related impairment of lung function, possibly because of a combined effect of smoking on the lungs and its anti-estrogenic effects. Sensitivity analyses in women without a history of asthma, surgical menopause or gynecological disorders, showed similar and statistically significant results.

We used latent class analysis to define the menopausal status rather than fixed cut-offs, as this type of analysis accounts better for the underlying biology and the information gathered from the participants. Clustering of the most similar observations forms the latent classes, and the multivariate and repetitive nature of the classification process allows for a dynamic grouping based on maximum likelihood criteria. We restricted classification to three classes as the data favored more classes by a marginally better fit, yet it supported a structure of two clearly defined classes (nonmenopausal and postmenopausal), as well as a more heterogeneous class representing different stages within the menopausal transition. The transitional class might include a minority of postmenopausal and nonmenopausal women.

Hormone levels of women above 60 years who were still categorized as transitional are on average lower than in women categorized as postmenopausal. The trajectories of FSH over the menopausal transition are not uniform across the population; e.g. women with a higher BMI or expressed sexuality are more likely to show a slower increase and lower postmenopausal level of FSH (49-51). These women are most likely postmenopausal, however hormonally they are more similar to the transitional class, as determined by the latent class analysis (7, 8). The heterogeneity of the transitional class might dilute associations slightly.

Hormonal changes in menopause, linked to complex biological events might contribute to the demonstrated acceleration in lung function decline with menopause. One possible mechanism is systemic inflammation, which is associated with menopausal hypoestrogenism and impaired lung function. The way 17β-estradiol affects inflammation depends strongly on the type of inflammatory or immunogenic stimulus, the involved cell types, the target organ's specific microenvironment and metabolism, leading to different anti- and proinflammatory effects (10, 52). Animal models suggest that low levels of 17β-estradiol might amplify inflammation and higher levels might attenuate it (11, 12). In rats and mice, surgically induced menopause enhances pleural exudation, leukocyte migration and lung myeloperoxidase activity amongst others (13, 14). Ovariectomy is further associated with increased levels of inflammatory markers, which decreased after 17β-estradiol substitution (13, 14). A comprehensive review concluded that in all available inflammatory models of the lung, 17β-estradiol was demonstrated to have an anti-inflammatory effect (10).

Further, hypoestrogenism plays a critical role in osteoporosis, which results in reduced height of the thoracic vertebrae which may mechanically reduce the expansion of the thoracic cage during inspiration and place the diaphragm in a suboptimal position (53, 54). Our findings

Triebner et al   14

suggested that the accelerated decline related to menopause was in part explained by osteoporosis, as accounting for change in height attenuated the associations of menopause with decline in FVC as well as FEV_1.

Strengths of the present study are the availability of repeatedly measured lung function and hormone measurements as well as interview/questionnaire data for large population samples. A unique window of opportunity was taken advantage of, as the study subjects were aged 25-48 years at baseline and most women went through the age of the menopausal transition during the 20-year follow-up. Hormone measurements at two time points are a major strength of this analysis, as well as the use of latent class analyses to define menopause based on both hormone measurement and questionnaire information. The crude definition of menopause at baseline, only based on questionnaire data, is a weakness of the study that may have introduced minor non-differential error and thus attenuated true results. The outcomes of the study, objectively measured lung function parameters are a strength and spirometric measurement error is unlikely to be related to menopausal status. The change in spirometers between surveys is accounted for in the analyses, however, it is possible that a residual non-differential measurements error may have attenuated the true results. The multinational and multi-centric design of the study allows for high external validity and the consistency across geographic and cultural borders suggests biologic explanatory mechanism rather than confounding by sociocultural factors.

Conclusions

The present study shows that menopause was associated with an accelerated decline in lung function, beyond the expected age related decline. This was most pronounced for FVC. The results were consistent across subgroups and independent of smoking history. The effect size for FEV_1 was comparable to smoking 20 cigarettes per day for two years, and for FVC to

smoking 20 cigarettes per day for ten years. The mechanisms underlying the associations

between reproductive aging and respiratory health need to be further investigated. Future

studies should address potential beneficial effects of hormone replacement therapy. The

results from the present study should be taken into account, as they are highly relevant for the

health and quality of life of a large and steadily increasing number of women.

American Journal of Respiratory and Critical Care Medicine

Triebner et al   16

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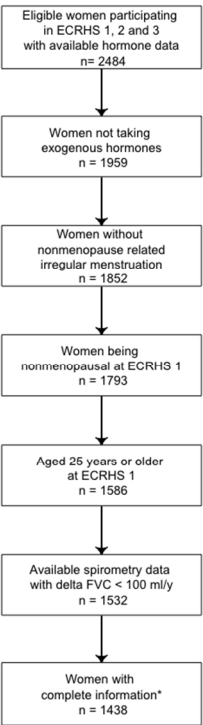
Figure 1. Study population with inclusion criteria (*height and packyears)
Figure 1
407x313mm (72 x 72 DPI)
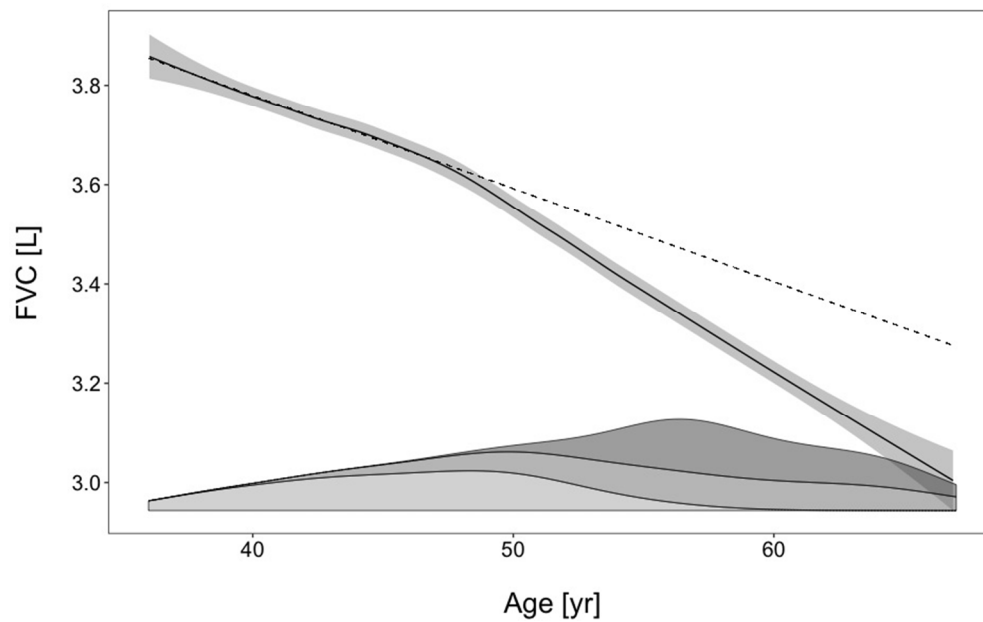
Figure 2. Best linear unbiased predictions for FVC of the study population (n=1438) according to the specified linear mixed effects model, including 95%CI and population distribution by menopausal class (The dotted line, as visual aid continues the initial slope).
Figure 2
333x205mm (72 x 72 DPI)
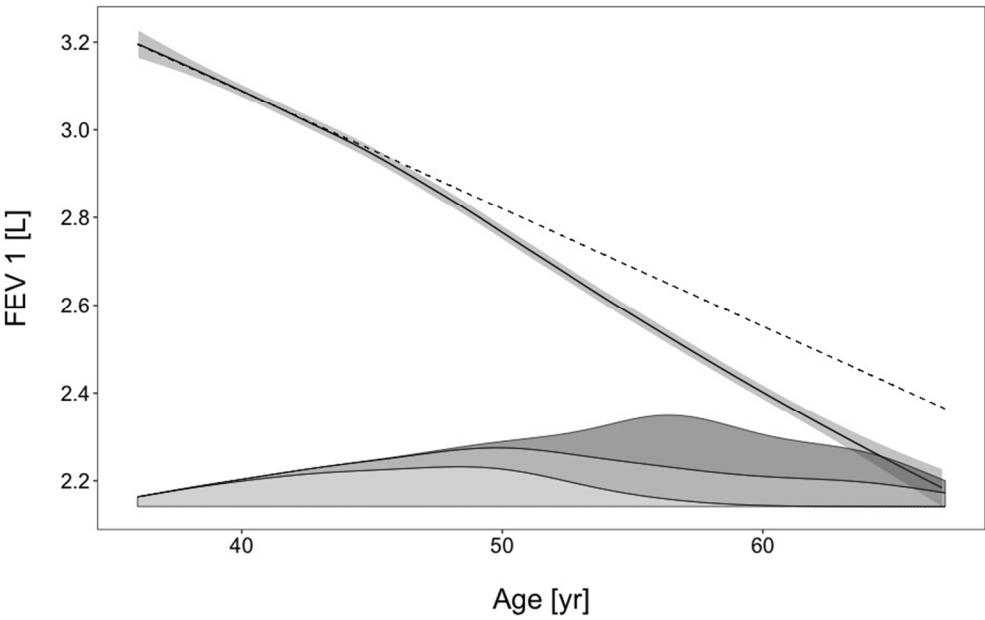
Figure 3. Best linear unbiased predictions for FEV1 of the study population (n=1438) according to the specified linear mixed effects model, including 95%CI and population distribution by menopausal class (The dotted line, as visual aid continues the initial slope).
Figure 3
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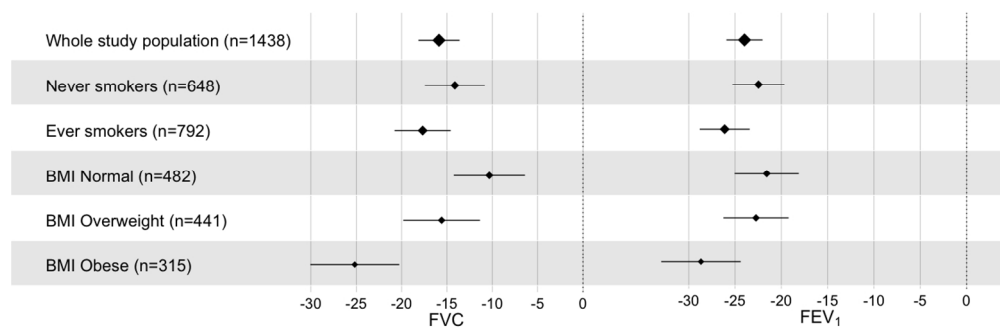
Figure 4. Age related lung function decline by smoking and BMI for the study population, ml/yr
Figure 4
479x164mm (72 x 72 DPI)
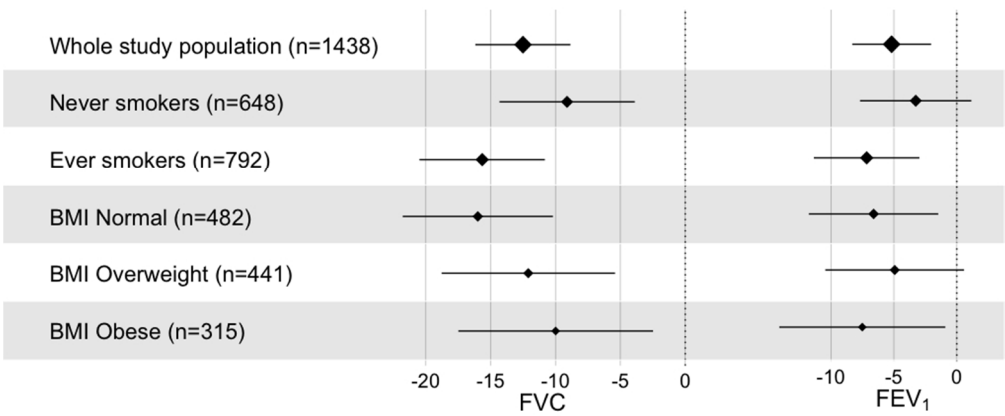
Figure 5. Menopause related lung function decline by smoking and BMI for the menopausal class, ml/yr
Figure 5
358x158mm (72 x 72 DPI)

ONLINE DATA SUPPLEMENT

Hormone measurements

At ECRHS 2 the levels of FSH and LH in the serum samples were determined by chemiluminescence in an Elecsys 2010 analyzer from Roche Diagnostics (Germany) at the Hôpital Xavier Bichat in Paris, France. At ECRHS 3 we determined FSH and LH levels using enzyme-linked immunosorbent assays provided by Demeditec Diagnostics, (Germany). All pipetting steps were executed fully automated on a liquid-handling robot from Hamilton Robotics using the protocol 4Elisa at the Core Facility for Metabolomics of the University of Bergen, Norway (www.uib.no/metabolomics). For FSH, the between day coefficient of variation was 7.0% and accuracy was 106%, for LH respectively 8.6% and 85%.

Menopausal status

Methods: At ECRHS 2 the menstrual status was obtained from the answers to the following questions: "Are your periods regular?" (Alternatives: "Yes", "No, they have never been regular", "No they have been irregular for a few months") and "Why did your periods stop?" (Alternatives: "Naturally", "Because of surgery" and "Other"; and at ECRHS 3: "Do you have regular periods?" (Alternatives: "Yes", "No, they have never been regular", "No they have been irregular for a few months" and "No, my periods have stopped").

Results: At ECRHS 2, 52% (n=586) of the participants were categorized as nonmenopausal, 28% (n=311) as transitional and 20% (n=223) as menopausal. At ECRHS 3, 26% (n=367) of the participants were categorized as nonmenopausal, 29% (n=409) as transitional and 46% (n=651) as menopausal. A reversal of menopausal status was indicated in the latent class analysis in a few cases (4%). Excluding these women from the analysis did not alter the

results. We kept the observations in the primary analysis with the corresponding class originally attributed by the latent class analysis.

Pulmonary function data

Pulmonary function data, including hormone measurements and a complete set of covariates was available at baseline for 1369 women (95%), at ECRHS 2 for 866 women (60%) and ECRHS 3 for 1060 women (74%). Of the included women 71% provided measurements at all three waves, 28% at two waves and 2% at one wave.

Table E1. Spirometers used by center and wave

Centre	ECRHS 1	ECRHS 2	ECRHS 3
Aarhus (DK)	Vitalograph spiro	-	NDD Easyone
Hamburg (DE)	Jaeger pneum	Jaeger pneum	NDD Easyone
Erfurt (DE)	Jaeger pneum	Jaeger pneum	NDD Easyone
Galdakao (ES)	Biomedin spiro	Biomedin spiro	NDD Easyone
Albacete (ES)	Biomedin spiro	Biomedin spiro	NDD Easyone
Huelva (ES)	Biomedin spiro	Biomedin spiro	NDD Easyone
Bordeaux (FR)	Vitalograph spir	Vitalograph spir	NDD Easyone
Grenoble (FR)	Biomedin spiro	Biomedin spiro	NDD Easyone
Montpellier (FR)	Biomedin spiro	Biomedin spiro	NDD Easyone
Paris (FR)	Biomedin spiro	Biomedin spiro	NDD Easyone
Reykjavik (IS)	SensorMedics spiro	SensorMedics spiro	NDD Easyone
Bergen (NO)	SensorMedics spiro	SensorMedics spiro	NDD Easyone
Gothenburg (SE)	SensorMedics spiro	SensorMedics spiro	NDD Easyone
Umea (SE)	SensorMedics spiro	SensorMedics spiro	NDD Easyone
Uppsala (SE)	SensorMedics spiro	SensorMedics spiro	NDD Easyone
Tartu (EE)	Jaeger pneum	Jaeger pneum	NDD Easyone

Triebner et al   3

Table E2. Class conditional probabilities and standard errors for the latent class analysis

	Group	First follow-up (ECRHS 2)			Second follow-up (ECRHS 3)		
		Nonmenopausal	Transitional	Menopausal	Nonmenopausal	Transitional	Menopausal
FSH quintiles	1st	41.6 (2.8)	0.0 (0.0)	0.0 (0.0)	77.1 (2.5)	0.0 (0.0)	0.5 (0.4)
	2nd	33.0 (2.3)	15.3 (2.7)	0.0 (0.0)	22.9 (2.5)	48.9 (4.2)	0.0 (0.0)
	3rd	25.0 (2.2)	22.3 (3.0)	0.0 (0.0)	0.0 (0.0)	36.3 (3.7)	20.9 (2.7)
	4th	0.4 (2.3)	62.4 (3.7)	1.6 (3.2)	0.0 (0.0)	14.8 (3.7)	34.6 (2.5)
	5th	0.0 (0.0)	0.0 (0.0)	98.4 (3.2)	0.0 (0.0)	0.0 (0.0)	44.0 (2.7)
LH quintiles	1st	39.8 (2.4)	2.0 (2.4)	0.0 (0.0)	77.2 (2.5)	0.6 (0.6)	0.0 (0.0)
	2nd	33.8 (2.3)	11.5 (2.7)	0.0 (0.0)	18.0 (2.2)	43.2 (3.3)	6.4 (1.5)
	3rd	25.2 (2.2)	24.7 (3.0)	0.0 (0.0)	2.3 (0.9)	32.6 (2.9)	21.9 (2.0)
	4th	1.3 (0.9)	47.8 (4.1)	22.7 (2.9)	0.0 (0.0)	14.9 (2.5)	34.6 (2.2)
	5th	0.0 (0.0)	14.1 (2.5)	77.4 (2.9)	2.5 (1.1)	8.7 (2.1)	37.2 (2.2)
Menses	Regular	85.3 (1.7)	65.1 (3.4)	9.2 (2.0)	70.5 (2.8)	6.5 (1.8)	1.4 (0.6)
	Irregular	6.4 (1.2)	13.6 (2.1)	17.5 (2.6)	17.8 (2.3)	15.4 (2.3)	4.8 (1.1)
	Stopped	8.3 (1.3)	21.3 (2.8)	73.3 (3.0)	11.8 (2.0)	78.1 (2.8)	93.8 (1.3)

Table E3. Determinants of decline of forced vital capacity

	Mean (95% CI)[4]	p-Value
Age[1] [ml/yr]	-15.9 (-18.2 to -13.7)	< 0.001
Height at baseline [ml/cm]	52.3 (48.6 to 55.9)	< 0.001
Weight [ml/kg]	-6.5 (-7.8 to -5.3)	< 0.001
Packyears [ml/packyear]	-1.0 (-1.9 to -0.1)	0.04
Current smoking[2] [ml]	-9.4 (-38.1 to 19.3)	0.52
Education[3] [ml/yr]	1.1 (-2.9 to 5.0)	0.60

[1]Centered at median

[2]Current smoking compared to former and never smokers

[3]Age at completed full-time education

[4]Linear mixed effect model, intercept -4.6 L (-5.2 to -4.0)

Table E4. Determinants of decline of forced expiratory volume in one second

	Mean (95% CI)[4]	p-Value
Age[1] [ml/yr]	-24.2 (-26.2 to -22.3)	< 0.001
Height at baseline [ml/cm]	37.2 (34.1 to 40.3)	< 0.001
Weight [ml/kg]	-4.3 (-5.4 to -3.2)	< 0.001
Packyears [ml/packyear]	-2.2 (-3.0 to -1.4)	0.04
Current smoking[2] [ml]	-29.3 (-54.0 to -4.7)	0.02
Education[3] [ml/yr]	1.2 (-2.2 to 4.5)	0.50

[1]Centered at median

[2]Current smoking compared to former and never smokers

[3]Age at completed full-time education

[4]Linear mixed effect model, intercept -3.0 L (-3.5 to -2.5)

Triebner et al   4

Triebner et al   5

Triebner et al   6

Triebner et al   7

American Journal of Respiratory and Critical Care Medicine