Risk of Asthma Onset After Natural and Surgical Menopause: Results From the French E3N Cohort

Bobette Matulonga-Diakiese, Dominique Courbon, Agnès Fournier, Margaux Sanchez, Annabelle Bédard, Sylvie Mesrine, Camille Taillé, Gianluca Severi, Gabriel Thabut, Raphaëlle Varraso, et al.

ORIGINAL RESEARCH

Title: Risk of asthma onset after natural and surgical menopause: Results from the French E3N Cohort

Bobette Matulonga-Diakiese[1,2], Dominique Courbon[1,3], Agnès Fournier[4,5], Margaux Sanchez[6,7], Annabelle Bédard[6,7], Sylvie Mesrine[4,5,8], Camille Taillé[1,9], Gianluca Severi[4,5], Gabriel Thabut[1,3], Raphaëlle Varraso[6,7] and Bénédicte Leynaert[1,3].

1. UMR 1152, Pathophysiology and Epidemiology of Respiratory Diseases, Epidemiology Team, Inserm, Paris, France;
2. Faculty of Medicine, Univ Paris-Sud, Le Kremlin-Bicêtre, France;
3. UMR 1152, Univ Paris Diderot Paris7, Paris, France ;
4. Univ Paris-Saclay, Univ Paris-Sud, UVSQ, CESP Health across Generations, INSERM, Villejuif, France;
5. Gustave Roussy, Villejuif, France;
6. U1168, VIMA (Aging and chronic diseases. Epidemiological and public health approaches), Inserm, Villejuif, France;
7. UMR-S 1168, UVSQ, Univ Versailles St-Quentin-en-Yvelines, St-Quentin-en-Yvelines, France
8. Department of Gynecology, Hôpital Bretonneau, CHRU de Tours, Tours, France;
9. Service de Pneumologie A et Centre de Compétence des Maladies Pulmonaires Rares, Hôpital Bichat, Assistance Publique-Hôpitaux de Paris, Paris, France;

Corresponding author:

Bénédicte Leynaert

Inserm UMR 1152, Physiopathologie et épidémiologie des maladies respiratoires - Equipe Epidémiologie

Faculté de Médecine site Bichat

16 Rue Henri Huchard

75890 PARIS cedex 18, France

Telephone number: +33. 1.57.27.75.68

Email : benedicte.leynaert@inserm.fr

HIGHTLIGHTS

- The only two longitudinal studies on menopause and asthma had conflicting findings
- We showed that surgical menopause is associated with an increased risk of asthma
- We found no change in asthma onset with natural menopause in normal weight women
- The incidence of asthma increased after menopause, only in overweight/obese women
- Metabolic factors, rather than female sex hormones alone, are likely to be involved

Abstract

Background: The gender switch in asthma incidence around puberty has been put forward to suggest a role of sex hormones in asthma. However, there are limited and inconsistent findings on change in asthma incidence with menopause. We aimed to investigate the associations between menopause and asthma incidence, and interactions with overweight/obesity.

Methods: Asthma incidence was assessed in 67,872 women free of asthma at baseline (aged 41-68 years) and regularly followed-up as a part of the French E3N cohort. Adjusted hazard-ratios (aHR) were derived from Cox models considering age as the time-scale, menopausal status as a time-varying covariate and taking into account menopausal treatment.

Results: During 843,243 person-years of follow-up, 1,205 new-onset asthma cases were identified. Compared to pre-menopause, surgical menopause was associated with an increased risk of asthma onset (aHR=1.33 [95%CI 1.01-1.75]) but no association was observed for natural menopause (aHR=1.05 [0.84-1.32]). In women with natural menopause, a further analysis separating the transition through menopause and the later post-menopausal period did not show any change in asthma incidence with menopause in the total sample and in normal-weight women. However, in overweight/obese women, peri-menopausal and post-menopausal women had an increased risk of developing asthma as compared to pre-menopausal women of the same age (aHR=1.91 [1.00-3.66] and aHR=2.08 [1.07-4.06] respectively).

Conclusion: Surgical menopause was associated with an increased risk of asthma onset. For natural menopause, no change in asthma incidence was observed in normal-weight women. However, overweight/obese women had an increased risk of developing asthma after natural menopause.

Keywords

Asthma, menopause, Body mass index, epidemiology, reproductive aging, sex hormones, E3N.

Data statement.

The authors have applied for E3N data access for this specific analysis and do not have permission to share the data collected in E3N surveys. The codes for the models used in the analysis will be shared on request.

1. Introduction

The most compelling argument for a role of sex hormones in asthma is that asthma is more frequent in boys in childhood and that there is a gender switch around puberty [1, 2]. Features of the women's reproductive life, such as menstrual cycles, age of menarche, pregnancy, parity or use of oral contraception have been found also to be associated with asthma [3, 4]. Studies have shown that the incidence of asthma, in particular non-allergic asthma, was higher in women as compared to men over all the reproductive years [2, 5]. More recently, menopause has been found to be associated with an accelerated lung function decline[6, 7]. Altogether, these findings suggest that sex hormones might influence women respiratory health [2, 5]. However, whether changes in sex hormones related to the transition through menopause are related to changes in asthma is still unknown. Most studies have focused on the role of menopausal hormone therapy (MHT) on asthma [8, 9] but the effect of menopause itself has been little studied and mostly through cross-sectional analyses [10]. Only two longitudinal studies, the Nurses' Health Study (NHS) in 1995 [11] and the Respiratory Health In Northern Europe (RHINE) survey in 2016 [12] have investigated the associations between menopausal status and asthma onset. These studies show discrepant results, with a lower [11] or higher [12] risk of asthma-onset in menopausal women as compared to pre-menopausal women of the same age. Inconsistencies between studies might relate to differences in the definition of the menopausal status, study design, statistical modeling or in the selection of the study population. Potential interaction with body mass index (BMI) also appears to be particularly relevant because of the change in adiposity with menopause, and the associations between obesity and asthma onset in women [3, 13-15].

The aim of the present study was to investigate the associations between menopause and asthma incidence, considering separately natural and surgical menopause, and considering possible interaction with obesity. The E3N cohort (**E**tude **E**pidémiologique auprès de femmes de la mutuelle

générale de l'**E**ducation nationale) represents a unique opportunity to investigate these associations as it comprises regular updating of the reproductive history, BMI and asthma, and includes a large sample of ageing women followed for more than 12 years.

2. Methods

2.1 Study design and population

Briefly, E3N is a prospective cohort of 98,995 French women born between 1925 and 1950, insured by a health insurance plan covering mostly teachers [16]. Participants, who gave written informed consent, completed self-administered questionnaires sent biennially since 1990[16]. The study protocol was approved by the French Institutional Ethics Committee and the French National Commission for Data Protection and Privacy. A question on ever asthma and date of diagnosis was included from the 1992 questionnaire onwards and asthma status was updated every two years. A question inquiring whether a doctor confirmed the asthma diagnosis was added in the 2002 and 2005 questionnaires. Women without asthma at baseline (1992 questionnaire) and who completed at least one of the follow-up questionnaires up to 2005 were followed-up until the age of asthma-onset, age at last answered questionnaire, or December 2005 (end of study), whichever occurred first.

2.2 Definitions

Incident cases of asthma were women without asthma at baseline who reported to have had asthma at any biennial questionnaire. The age at asthma onset reported in the questionnaire in which women reported asthma attack for the first time was used to define the “age at asthma onset”. Women who reported an age at asthma onset before baseline were excluded (figure 1).
Menopausal status and age at menopause were determined from biennially updated questionnaires as previously in E3N [17, 18]. Women were considered post-menopausal if they had had 12 consecutive months without menstrual periods (unless due to hysterectomy), had undergone bilateral oophorectomy, had used MHT, or reported that they were post-menopausal [18]. Surgical menopause was defined as bilateral oophorectomy, with or without hysterectomy, with age at oophorectomy used as age at menopause. Age at natural menopause was defined as the age at

the last menstrual period (if occurred before any MHT use), or, in decreasing order of priority, self-reported age at menopause, age at start of MHT or age at start of menopausal symptoms. We excluded women with hysterectomy without oophorectomy (as their menopausal status could not be defined).

2.3 Statistical analyses

In a first analysis, hazard-ratios (HR) for asthma onset were estimated using Cox proportional-hazard models, with age as the time-scale [19] and menopausal status considered as a three-level time-varying covariate (pre-menopause *vs*. natural post-menopause /surgical post-menopause) (Figure E1, in supplementary data). The proportional hazards assumption was assessed by introducing an interaction term with age. However, no interaction was observed. Analyses were adjusted for BMI, smoking status, physical activity and current use of MHT (taking into account the type of MHT), treated as time-varying covariates, updated from biennial questionnaires, and hay fever (as a proxy of allergic status), educational level and cohort of birth included as fixed covariates. Details on variables used for adjustment are provided in the supplementary data. Sensitivity analyses were carried-out in participants who completed all surveys (N=50 026), and considering the American Thoracic Society (ATS) criteria for self-reported “doctor-diagnosed asthma” [9] (positive answer to both questions, “have you ever had an asthma attack?” and “was this confirmed by a doctor?”). Because earlier studies showed interaction between obesity and hormonal factors on asthma [3, 12], the analysis was stratified according to BMI at baseline. The interaction of BMI (“normal-weight” *vs* “overweight/obese”) was further tested by introducing an interaction term in the model. Because the incidence of asthma has been shown to increase over cohort of birth [5], a stratified analysis was carried by birth cohort to appraise potential residual bias related to the overrepresentation of women from more recent cohort among pre-menopausal women. We also stratified analyses by allergic status because non-allergic and allergic asthma

phenotypes may be differently associated with risk factors and because women's hormonal status might be more strongly associated with non-allergic asthma [2].

The Cox model described above considered age at menopause as a cut-point between pre and post menopause. However, because natural menopause is not an abrupt event, we considered an alternative model in women who had reached menopause naturally. This second model considered a "transition period" between "pre" and "post" menopause, as derived from the Stages of Reproductive Aging Workshop classification [20] (figure E1). All analyses were performed with SAS software version 9.3.

3. Results

Our study population consisted of 67,872 women (mean age 51.7; standard deviation (SD) 6.7) without asthma at baseline and with complete data on asthma and menopausal status at follow-up (figure 1). Comparison of women included and excluded from the analysis is provided in the supplementary data. As compared to naturally post-menopausal women, surgically post-menopausal women were more frequently overweight/obese and used more frequently estrogen alone as MHT (table 1). At the end of follow-up (median length of follow-up 13.2 years), 2,088 (3.1%) women were still pre-menopausal; 59,519 (87.7%) were naturally post-menopausal and 6,265 (9.2%) had undergone surgical menopause. Overall 1,205 women developed asthma over the follow-up.

3.1 Asthma onset after natural or surgical menopause as compared to before menopause (model 1)

The incidence of asthma (per 1000 Person-Years) was 1.20[95% confidence interval (CI): 1.04-1.36] before menopause, 1.43[1.33-1.53] after natural menopause and 1.94[1.61-2.27] after surgical menopause. A higher risk of asthma incidence was found in women with hay fever, in overweight and obese women, in smokers and in women from more recent birth cohorts. The use of MHT, and particularly estrogen-MHT, was also independently associated with a higher incidence of asthma (HR=1.21 [1.02-1.43]). After adjustment for all these factors, the Cox model did not show any difference in asthma incidence in naturally post-menopausal women as compared to pre-menopausal women (adjusted Hazard-Ratio (aHR)=1.05[0.84-1.32], table 2), but there was an increased risk of new-onset asthma in surgically post-menopausal women (aHR=1.33[1.01-1.75]). Sensitivity analyses conducted among never-smokers, among MHT never-users, and using self-

reported doctor-diagnosed asthma showed also a higher risk of new-onset asthma associated with surgical menopause, but no association was found for natural menopause (table 2).

The incidence of asthma was much higher in women with hay fever as compared to women without hay fever and in women born after 1945 as compared to women born before 1945. The analysis stratified by allergic status or by birth cohort consistently did not show any association between natural menopause and asthma whatever the strata. Surgical menopause remained associated with an increased risk of asthma in women born before 1945 (aHR=1.47[0.96-2.24]) and in allergic women (aHR=1.95[1.22-3.13]).

The incidence of asthma was higher in overweight women as compared to normal-weight women and in obese women as compared to overweight women, whatever the menopausal status (figure E2 in the supplementary material). For the stratified analysis, overweight and obese women were grouped together (Table 2). Surgical menopause remained associated with a higher risk of asthma in overweight/obese women (aHR=1.73[1.04-2.87]), but this association was reduced in normal-weight women (aHR=1.16[0.83-1.62]) (p for interaction=0.26) (table 2). The hazard-ratio for asthma onset in naturally post-menopausal women as compared to pre-menopausal women, was very close to 1.00 (0.99[0.76-1.29]) and showed no association in normal-weight women. It was somewhat higher (1.25[0.79-1.97]) but still not significant in overweight/obese women.

3.2 Asthma-onset before menopause, during the transition to natural menopause and in the later post-menopausal period (model 2)

In the analysis carried-out only in women who had reached the menopause naturally, and considering a transition period between “pre” and “post” menopause (figure E1), no difference in asthma incidence was observed during the transitional and in the later period after menopause as

compared to pre-menopause, in the total sample (aHR=1.06[0.79-1.41] and 1.04[0.77-1.28] respectively). The analysis stratified by BMI did not show any change in asthma incidence with menopause in normal-weight women (figure 2a). In overweight/obese women (figure 2b), an increased risk of asthma-onset was observed both during the transition period (aHR=1.91[1.00-3.66]) and in the later post-menopausal period (aHR=2.08[1.07-4.06]) as compared to pre-menopause. Similarly to the main analysis, the sensitivity analyses carried-out in never-smokers, in women who never used MHT, in women who completed all the questionnaires and when considering self-reported doctor-diagnosed asthma, showed no association between natural menopause and asthma, in normal weight women. The higher risk of asthma associated with natural menopause in overweight/obese women, was confirmed in all the sensitivity analyses, except in women who had never used MHT, in whom the association was reduced and no longer significant (Table 3).

3.3 Findings from supplementary analyses

Finally, to appraise the effect of surgical menopause as compared to natural menopause, a subsidiary analysis was performed using “natural post-menopause” as the reference. In this analysis, surgical menopause was also associated with an increased risk of asthma onset (aHR=1.26[1.05-1.52]) as compared to natural menopause.

4. Discussion

4.1 Main findings

This study is the first prospective study to investigate change in asthma incidence with menopause taking into account the type of menopause and potential modification effect by BMI. In this large prospective study of women, including 1,205 incident cases of asthma, we did not find any difference in the risk of asthma onset in natural post-menopausal women, as compared to pre-menopausal women of the same age in the total sample, and in normal-weight women. In overweight/obese women, an increased risk of asthma onset was observed during the transition to natural menopause and in the later post-menopausal period, although this increased risk was not found in the limited sample of women who never used MHT. Surgical menopause was associated with an increased risk of asthma both as compared to pre-menopause and to natural post-menopause.

4.2 Comparison with other studies

Our finding of an increased risk of asthma after surgical menopause is consistent with the cross-sectional analysis of Health Survey for England in which surgical menopause was associated with an increased risk of doctor-diagnosed asthma [8]. In normal-weight women, we found no evidence for any change in the incidence of asthma around menopause, which is consistent with a recent meta-analysis by Zemp et al [10]. Noteworthy, in the most recent longitudinal analysis on asthma and menopause (RHINE), the increased risk of asthma in women who were post-menopausal at the end of follow-up as compared to premenopausal women was observed only in women with higher BMI [12] which is consistent with our findings. However, in the RHINE analysis, surgical and natural menopause were not considered separately, and data from the present study shows that up to 9% of post-menopausal women had undergone surgical menopause. Our results contrast with those from the first longitudinal study in the Nurse Health Study showing a decreased risk of asthma in

naturally post-menopausal women who never used MHT as compared to pre-menopausal women [11]. Unfortunately, this analysis of the NHS was not stratified by BMI. In our analysis, in normal weight women, the hazard-ratio for natural menopause and asthma was lower than 1.00 in some sensitivity analyses (e.g. HR=0.78 (0.51-1.19) for diagnosed asthma). However, these associations were not at all significant. Differences in population and work-related exposure (such as exposure to cleaning disinfectants, which have been found to be associated with asthma in hospital workers) might partly explain the different findings [21].

Several studies, including the E3N cohort, showed an increased risk of asthma associated with MHT use, and more particularly with postmenopausal use of estrogens [9]. In our sample, 68% of the post-menopausal women had ever used MHT. In our main analysis, the hazard-ratio for the risk of asthma associated with menopause was adjusted for the use of MHT, which was defined as a time-dependent variable (for each type of MHT, post-menopausal women were classified as "users"/then re-classified as "non-users" when they started/stopped using the specific treatment). We also run a sensitivity analysis after censoring women when they started using MHT. Noteworthy, the hazard-ratios showing an increased risk of asthma associated with peri- and post- menopause in the analysis adjusted for MHT among overweight/obese women (HR=1.91 and HR=2.08; table 3) were reduced and not significant in this sensitivity analysis (HR=1.47(0.74-2.92) for perimenopause and HR=1.17(0.52-2.64) for post-menopause). Therefore, we cannot exclude that the excess risk of asthma incidence associated with menopause in overweight/obese women might be due to residual confounding by MHT use. However, these findings have to be interpreted with caution, as this group of women who had never used MHT might be highly selected (including women with specific health-seeking behaviors, or women with little or no trouble with menopause-related symptoms), and not representative of post-menopausal women.

4.3 Strengths and limitations

The strengths of our study include the prospective design with biennial updating on asthma, menopausal status and MHT use, the sample size and the large number of incident cases of asthma, which ensured adequate power for sub-group analyses. We used age as the time-scale because it has been shown that bias can occur in Cox's regression analysis of epidemiologic cohort when time-on-study is used as the time-scale instead of age, especially for time-dependent covariates associated with age such as menopause [19]. To overcome residual confounding with smoking, which has anti-estrogen effects and is associated with early menopause, and to limit possible misclassification with chronic obstructive pulmonary disease (COPD), a sensitivity analysis was performed in never smokers. To avoid potential confounding by indication or selection bias [3], we adjusted for MHT instead of excluding MHT-users. However, to highlight the effect of natural changes in sex hormone levels around menopause, we also conducted a sensitivity analysis in women who had not used MHT. Altogether, the stratified and sensitivity analyses support the robustness of our findings and do not suggest that asthma incidence might change after natural menopause in normal-weight women. Although we do not have objective data to confirm asthma diagnosis, asthma defined by questionnaire has been found to have a high specificity when compared to a clinical diagnosis of asthma [22]. In addition, we found consistent findings in the sensitivity analysis considering the ATS definition for self-reported doctor-diagnosed asthma that has been used in several E3N analyses [9, 23]. The prospective design and the biennial updating of information on asthma and menopausal status in the E3N study reduce possible recall bias and risk of misclassification when assessing whether asthma onset occurred before or after menopause. Analyses comparing ages of asthma onset declared in different questionnaires in E3N showed good agreement [24]. Menopausal status and age at menopause were also self-reported. To date, there is no adequate independent biological marker to define menopausal status and it is advised to base

the diagnosis of menopause on menstrual cycle criteria [20]. Furthermore, a validation study on self-reported menopausal status and age at menopause in E3N has shown sufficient accuracy [17]. In our sample, 72% of women had completed all the six follow-up questionnaires, and findings were unchanged in this subgroup. The use of the model that includes a “transition period” should further reduce possible classification bias due to inaccurate reporting of age at menopause or age at asthma. In addition, the use of this model better reflects the continuum of the reproductive aging, and allows comparing women with more contrasted hormonal profiles. The indication of oophorectomy was not available, which limits speculation on the association observed between surgical menopause and asthma-onset. Because most of E3N participants have been recruited among teachers, our results might not be generalizable to other populations with different exposure.

4.4 Possible underlying mechanisms

In the present analysis, the increased risk of asthma onset associated with natural menopause, found in the model considering a transition period between pre and post menopause, was restricted to overweight/obese women. After menopause, adipose tissue becomes the main source of estrogen production due to androgen aromatization [25]. One study reported higher estradiol levels in women who developed asthma after their menopause as compared to healthy menopausal women [26]. Estrogen treatment after the menopause has been found to be associated with asthma-onset [9]. Altogether, these observations suggest that estrogen levels above the normal range for a given stage of the reproductive aging, or unbalanced estrogen/progesterone ratio might increase the risk of asthma [27]. Although debated, higher plasma estrogen levels have been shown in overweight/obese women around menopause [28], which might explain the higher asthma incidence we observed after natural menopausal in these overweight/obese women. Menopause is associated with changes in body fat distribution.[29] Weight gain and change in body silhouette have been associated with an increased risk of asthma onset in the E3N and other prospective

studies. Noteworthy, obese women gained more weight after natural menopause than women with BMI<25kg/m² (+0.49 kg/m^2 (standard deviation; SD 1.49) vs. 0.16 kg/m^2 (SD 0.93)). Although our analyses were adjusted for BMI as a time-dependent variable, we could not adjust for change in adiposity. Finally, we could not adjust for potential confounding by obesity-related comorbidities such as gastroesophageal reflux or obstructive sleep apnea.

A plausible explanation for the increased incidence of asthma among surgically post-menopausal women as compared to both pre-menopausal and naturally post-menopausal women might relate to the abrupt decrease of sex hormones and the different hormonal profile of surgically post-menopausal women [8, 30]. Women with bilateral oophorectomy have been found to have lower levels of testosterone and lower sex hormone-binding globulin and similar levels of estradiol as compared to natural menopause [30]. It has been suggested that androgens might have a protective immunomodulating effect against asthma development [31]. Studies *in vivo* suggest that testosterone might be partly responsible for the decreased Th2 cell responses in males, and murine models have shown that castrated mice developed more severe airways inflammation [32]. Noteworthy, the few studies that have investigated the associations between the changes in asthma around puberty and sexual maturation were not able to demonstrate any relationship between estrogens and asthma, but pointed toward a possible effect of androgens [1, 31]. The underlying pathology leading to surgical menopause in obese women might also be associated with an increased risk of asthma [8]. Altogether, the increased risk of asthma among surgically post-menopausal women, the association between natural menopause and asthma observed only in overweight women, put together with the reported change in the sex-ratio for asthma around puberty, raise the question of a protective role of androgens in asthma or the unbalance between androgens/estrogens rather than a negative effect estrogens taken alone.

4.5. Conclusion

To summarize, in this large prospective study, we did not find any difference in the incidence of asthma before and after natural menopause in the total sample, and in normal-weight women. Only overweight/obese women were at increased risk of developing asthma after natural menopause, which strengthens the hypothesis of a close interplay between metabolic factors and sex hormones in asthma. Surgically menopausal women were at increased risk of asthma onset. Our findings suggests that factors other than change in estrogen levels have to be explored to elucidate the gender switch in asthma incidence around puberty and higher incidence of asthma in women as compared to men throughout the reproductive life.

Acknowledgments

The authors thank all those who participated to the setting of the E3N study (Etude Epidémiologique auprès de femmes de la MGEN (Mutuelle Générale de l'Education Nationale)) on the various aspects. The authors are grateful to R Chait, M Fangon, M Niravong, C Kernaleguen and L Hoang for managing the data. They are indebted to all the individuals who participated without whom that study would not have been possible.

Fundings

This work was supported by the French League against Cancer, the Mutuelle Générale de l'Education Nationale, the Gustave Roussy Institute of Cancérologie, the National Institute of Health and Medical Research, the Programme Hospitalier de Recherche Clinique (PHRC) PHRC-National 2010 and PHRC-National 2012.

Conflict of Interest: none declared

Author's contributions :

B Matulonga, B Leynaert, R Varraso were involved in the conception, hypotheses delineation, and design of the study analysis strategy.

Gianluca Severi, A Fournier, S Mesrine, A Bédard, M Sanchez, and R Varraso participated in the acquisition of the data.

B Matulonga, D Courbon and B Leynaert, conducted the statistical analyses and interpreted the findings.

B Matulonga and B Leynaert wrote the manuscript.

All authors participated in the interpretation of the findings, reviewed the manuscript and revised it critically before submission.

All authors have seen and approved the final version of the manuscript.

References

1. Almqvist, C., M. Worm, and B. Leynaert, *Impact of gender on asthma in childhood and adolescence: a GA2LEN review.* Allergy, 2008. **63**(1): p. 47-57.
2. Leynaert, B., et al., *Gender differences in prevalence, diagnosis and incidence of allergic and non-allergic asthma: a population-based cohort.* Thorax, 2012. **67**(7): p. 625-31.
3. Macsali, F., et al., *Respiratory health in women: from menarche to menopause.* Expert Rev Respir Med, 2012. **6**(2): p. 187-200.
4. Hansen, S., et al., *Early menarche and new onset of asthma: Results from the SAPALDIA cohort study.* Maturitas, 2017. **101**: p. 57-63.
5. Hansen, S., et al., *Gender differences in adult-onset asthma: results from the Swiss SAPALDIA cohort study.* Eur Respir J, 2015. **46**(4): p. 1011-20.
6. Campbell, B., et al., *Menopause, lung function and obstructive lung disease outcomes: a systematic review.* Climacteric, 2018. **21**(1): p. 3-12.
7. Triebner, K., et al., *Menopause is Associated with Accelerated Lung Function Decline.* Am J Respir Crit Care Med, 2016.
8. Jarvis, D. and B. Leynaert, *The association of asthma, atopy and lung function with hormone replacement therapy and surgical cessation of menstruation in a population-based sample of English women.* Allergy, 2008. **63**(1): p. 95-102.
9. Romieu, I., et al., *Postmenopausal hormone therapy and asthma onset in the E3N cohort.* Thorax, 2010. **65**(4): p. 292-7.
10. Zemp, E., et al., *Asthma and the menopause: a systematic review and meta-analysis.* Maturitas, 2012. **73**(3): p. 212-7.
11. Troisi, R.J., et al., *Menopause, postmenopausal estrogen preparations, and the risk of adult-onset asthma. A prospective cohort study.* Am J Respir Crit Care Med, 1995. **152**(4 Pt 1): p. 1183-8.
12. Triebner, K., et al., *Menopause as a predictor of new-onset asthma: A longitudinal Northern European population study.* J Allergy Clin Immunol, 2016. **137**(1): p. 50-7 e6.
13. Brumpton, B., et al., *General and abdominal obesity and incident asthma in adults: the HUNT study.* Eur Respir J, 2013. **41**(2): p. 323-9.
14. Real, F.G., et al., *Lung function, respiratory symptoms, and the menopausal transition.* J Allergy Clin Immunol, 2008. **121**(1): p. 72-80 e3.
15. Lobo, R.A., *Metabolic syndrome after menopause and the role of hormones.* Maturitas, 2008. **60**(1): p. 10-8.
16. Clavel-Chapelon, F., et al., *[E3N, a cohort study on cancer risk factors in MGEN women. Description of protocol, main characteristics and population].* Bull Cancer, 1996. **83**(12): p. 1008-13.
17. Clavel-Chapelon, F. and N. Dormoy-Mortier, *A validation study on status and age of natural menopause reported in the E3N cohort.* Maturitas, 1998. **29**(2): p. 99-103.
18. Morois, S., et al., *Menopausal hormone therapy and risks of colorectal adenomas and cancers in the French E3N prospective cohort: true associations or bias?* Eur J Epidemiol, 2012. **27**(6): p. 439-52.
19. Thiebaut, A.C. and J. Benichou, *Choice of time-scale in Cox's model analysis of epidemiologic cohort data: a simulation study.* Stat Med, 2004. **23**(24): p. 3803-20.
20. Harlow, S.D., et al., *Executive summary of the Stages of Reproductive Aging Workshop + 10: addressing the unfinished agenda of staging reproductive aging.* Menopause, 2012. **19**(4): p. 387-95.
21. Dumas, O., et al., *Occupational exposure to disinfectants and asthma control in US nurses.* Eur Respir J, 2017. **50**(4).
22. Toren, K., J. Brisman, and B. Jarvholm, *Asthma and asthma-like symptoms in adults assessed by questionnaires. A literature review.* Chest, 1993. **104**(2): p. 600-8.

23. Bedard, A., et al., *Time-dependent associations between body composition, physical activity and current asthma in women using marginal structural models.* Am J Epidemiol, 2017.
24. Sanchez, M., et al., *Temporal asthma patterns using repeated questionnaires over 13 years in a large French cohort of women.* PLoS One, 2013. **8**(5): p. e65090.
25. Simpson, E.R., *Sources of estrogen and their importance.* J Steroid Biochem Mol Biol, 2003. **86**(3-5): p. 225-30.
26. Foschino Barbaro, M.P., et al., *Menopausal asthma: a new biological phenotype?* Allergy, 2010. **65**(10): p. 1306-12.
27. de Oliveira, A.P., et al., *Cellular recruitment and cytokine generation in a rat model of allergic lung inflammation are differentially modulated by progesterone and estradiol.* Am J Physiol Cell Physiol, 2007. **293**(3): p. C1120-8.
28. Freeman, E.W., et al., *Obesity and reproductive hormone levels in the transition to menopause.* Menopause, 2010. **17**(4): p. 718-26.
29. Davis, S.R., et al., *Understanding weight gain at menopause.* Climacteric, 2012. **15**(5): p. 419-29.
30. Kotsopoulos, J., et al., *The relationship between bilateral oophorectomy and plasma hormone levels in postmenopausal women.* Horm Cancer, 2015. **6**(1): p. 54-63.
31. Vink, N.M., et al., *Gender differences in asthma development and remission during transition through puberty: the TRacking Adolescents' Individual Lives Survey (TRAILS) study.* J Allergy Clin Immunol, 2010. **126**(3): p. 498-504 e1-6.
32. Hayashi, T., et al., *Less sensitivity for late airway inflammation in males than females in BALB/c mice.* Scand J Immunol, 2003. **57**(6): p. 562-7.

Table1. **Characteristics of the study population according to menopausal status at inclusion**

Baseline characteristics	**Pre-menopause N=32,489**	**Natural menopause N=30,710**	**Surgical Menopause N=4,673**
Age at baseline, mean (SD)	46.4 (3.1)	56.6 (5.1)	56.2 (6.0)
Age at menopause, mean (SD)	-	50.1 (3.04)	44.8 (5.8)
Birth cohort, %			
Born ≤ 1945	43.0	98.2	93.4
Born after 1945	57.0	1.8	6.6
BMI*, %			
<25 kg/m^2	84.6	76.5	69.4
≥25 kg/m^2	15.4	23.5	30.6
Smoking, %			
Never Smokers	63.8	71.9	73.4
Ex-smokers	26.1	20.6	20.7
Current smokers with <15 cig/day	5.9	4.4	3.7
Current smokers with ≥15 cig/day	4.2	3.1	2.2
Educational level, %			
< High school diploma	9.9	15.8	21.8
High school to 2-level university	48.0	53.7	53.5
≥ 3-level university	42.1	30.5	24.7
Physical activity†, %			
Low	40.7	35.1	36.2
Moderate	31.8	30.6	29.3
High	27.5	34.4	34.5
Allergic rhinitis	8.1	7.2	7.7
Current use of MHT‡, %			
No MHT	-	48.1	41.7
Estrogen alone	-	10.5	19.0
Estrogen + micro progest/dydrogest	-	18.2	17.7
Estrogen with other progestagen	-	20.5	16.4
Other	-	2.7	5.2

Note: * Body mass index was calculated as weight in kilogram divided by the square of height in meters.

† Physical activity was assessed using questions on several activities converted into the metabolic equivalent of task (MET) and added to yield the energy expended per week. Sum of METs was thereafter categorized into tertiles ("low", "moderate" and "high").

Abbreviations: BMI (Body mass index)

‡ MHT: menopausal hormone therapy

Table 2. **Associations between menopausal status and the risk of asthma-onset**

	New-onset asthma	Person-years	Adjusted HR* (95%CI)
			Total N=67,872
Pre-menopause	203	169,797	1.00 (ref)
Natural menopause	865	602,959	1.05 (0.84-1.32)
Surgical menopause	137	70 487	1.33 (1.01-1.75)
Sensitivity analyses			
Among never smoker†			Total N=44,742
Pre-menopause	110	102,745	1.00 (ref)
Natural menopause	511	405,320	1.12 (0.82-1.53)
Surgical menopause	96	49,540	1.58 (1.10-2.26)
In women who had never used MHT ‡			Total N=54,180
Pre-menopause	203	169,797	10.00 (ref)
Natural menopause	264	204,668	1.11 (0.88-1.41)
Surgical menopause	45	23,788	1.44 (0.97-2.13)§
Among women who answered to all questionnaires			*Total N=50,026*
Pre-menopause	163	125,651	1.00 (ref)
Natural menopause	668	477,131	0.95 (0.73-1.23)
Surgical menopause	108	54,215	1.26 (0.92-1.72)
Doctor-diagnosed asthma-onset †††			Total N=50,026
Pre-menopause	143	123,598	1.00 (ref)
Natural menopause	545	470,232	1.12 (0.82-1.53)
Surgical menopause	85	53,429	1.30 (1.00-1.82)
Stratified analysis according to BMI at inclusion ǂ			
In women with BMI<25			Total N=54,308
Pre-menopause	158	145,311	1.00 (ref)
Natural menopause	610	483,134	0.99 (0.76-1.29)
Surgical menopause	76	50,944	1.16 (0.83-1.62)**
In women with BMI≥25			Total N=13,551
Pre-menopause	45	24,482	1.00 (ref)
Natural menopause	255	119,789	1.25 (0.79-1.97)
Surgical menopause	61	19,543	1.73 (1.04-2.87)**

Note: * Hazard-ratios (HR) were derived from Cox model using women age as the time-scale and menopausal status as a time-dependent variable. Naturally and surgically post-menopausal women were compared to pre-menopausal women for their risk of developing asthma at the same age. HR are adjusted for BMI, smoking, current use of MHT, physical activity included as time-varying covariates and hay fever, birth cohort and educational level included as fixed covariates.

† In women who had never smoked in their life.

§p=0.07

‡Women who have reported use of MHT at inclusion were excluded. Women, who began using MHT during follow-up, accounted for person-years until the age of first use of MHT (or age of asthma onset) and were censored afterwards.

†† in women who participated to all surveys.

ǂHR are adjusted for all other confounders and for BMI as a continuous variable (included as time-varying covariate.

**p for interaction=0.26

Abbreviations: HR (Hazard-ratios)
MHT (menopausal hormone therapy)
BMI (body mass index)

Table 3. Change in asthma incidence during transition through natural menopause according to BMI at inclusion

	N incident	BMI< 25	BMI≥25	p for interaction
		Hazard-ratio* (95% CI)	Hazard-ratio* (95% CI)	
Among all women (n=61,607)				
Pre-menopause	104	1.00 (ref)	1.00 (ref)	
Transition to menopause	119	0.91 (0.66-1.25)	1.91 (1.00-3.66)	0.11
Post-menopause	832	0.86 (0.62-1.21)	2.08 (1.07-4.06)	0.09
Among never smokers† (n=40,395)				
Pre-menopause	55	1.00 (ref)	1.00 (ref)	
Transition to menopause	75	1.26 (0.82-1.94)	2.43 (1.02-5.79)	0.14
Post-menopause	485	0.98 (0.62-1.57)	2.13 (0.85-5.32) ‡	0.09
In women who had never used MHT (n=46,948)				
Pre-menopause	104	1.00 (ref)	1.00 (ref)	
Transition to menopause	96	0.87 (0.62-1.23)	1.47 (0.74-2.92)	0.20
Post-menopause	253	1.06 (0.68-1.64)	1.17 (0.52-2.64)	0.23
Among women who answered to all questionnaires (n=45,480)				
Pre-menopause	83	1.00 (ref)	1.00 (ref)	
Transition to menopause	93	0.97 (0.68-1.39)	2.07 (0.99-4.31)	0.12
Post-menopause	645	0.75 (0.51-1.11)	2.81 (1.32-5.97)	0.07
Doctor-diagnosed asthma onset (n=44,779) ǂ				
Pre-menopause	74	1.00 (ref)	1.00 (ref)	
Transition to menopause	83	1.06 (0.72-1.56)	2.30 (1.05-5.01)	0.10
Post-menopause	521	0.78 (0.51-1.19)	3.09 (1.38-6.93)	0.06

Note: Pre-menopause is defined as the period up to the 2 years before the age at menopause; transition to menopause is the period from 2 years before to 1 year after the age at menopause and post-menopause starts after 1 year of amenorrhea.

*Hazard ratios were adjusted for BMI (continuous variable), MHT, physical activity, smoking defined as time-varying covariates and hay fever, birth cohort and educational level defined as fixed covariates.

†In women who have never smoked in their life.

‡p=0.10

ǂin women who participated to all surveys

Abbreviations: BMI (body mass index)
CI (confident interval)
MHT (menopausal hormone treatment)