Influence of maternal BMI and macrophage activation on asthma exacerbations in pregnancy

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Word Count: 3083

Key Words

Asthma; pregnancy; exacerbation; macrophage; CD163; inflammation; eosinophil; exhaled nitric oxide

Abbreviations

ACQ	Asthma control questionnaire
ANOVA	Analysis of variance
BDP	Beclomethasone dipropionate
BMI	Body Mass Index
CRP	C-reactive protein
ECO	Exhaled carbon monoxide
ED	Emergency Department
FENO	Fractional exhaled nitric oxide
FEV_1	Forced expiratory volume in 1 second
FVC	Forced vital capacity
GWG	Gestational weight gain
ICS	Inhaled corticosteroids
IL	Interleukin
IOM	Institute of Medicine
IQR	Interquartile range
OCS	Oral corticosteroids
OR	Odds Ratio
RCT	Randomised controlled trial
sCD-163	Soluble CD-163
SD	Standard deviation
TNF	Tumour necrosis factor

Financial Support Statement

Project grant funding was provided by National Health and Medical Research Council (NHMRC, project grant ID 455593). Dr Vanessa Murphy was the recipient of an Australian Research Training Fellowship (Part-time, grant ID 455626) and Career Development Fellowship from NHMRC (grant ID 1084816). Prof Peter Gibson was the recipient of a Practitioner Fellowship from NHMRC.

Highlights Box:

What is already known about this topic?

- Obesity is a risk factor for exacerbations of asthma

What does this article add to our knowledge?

- Maternal overweight/obesity and systemic macrophage activation increase exacerbation risk for asthma during pregnancy

How does this study impact current management guidelines?

- This study highlights the potential importance of pre-pregnancy weight reduction for improving asthma outcomes, as well as perinatal outcomes, during pregnancy.

Abstract

Background: Obesity is a risk factor for exacerbations of asthma, but the mechanisms of this effect in pregnancy are unknown.

Objective: This study determined the influence of maternal body mass index (BMI), gestational weight gain, eosinophilic inflammation and systemic macrophage activation on the risk of exacerbations during pregnancy.

Methods:Women with asthma (n=164) participated in the study. BMI recorded at baseline (17 weeks gestation) was categorised as healthy weight (18.5-24.9 kg/m^2), overweight (25-29.9 kg/m^2) or obese (>30 kg/m^2). Exacerbations requiring medical intervention were recorded prospectively. Asthma control, medication use and fractional exhaled nitric oxide (FENO) were assessed monthly; additional visits occurred during exacerbations. Peripheral blood was collected at baseline for the measurement of eosinophils, soluble CD-163, C-reactive protein (CRP) and interleukin (IL)-6.

Results:

Exacerbations occurred in a higher proportion of overweight (51.1%) and obese (48.4%) women compared to healthy weight women (25%, P=0.026). Excess weight gain during pregnancy was not associated with exacerbation risk. Macrophage activation (elevated serum sCD-163) was associated with exacerbations requiring oral corticosteroids (OCS, P=0.043), while high peripheral blood eosinophils or FENO were not associated with exacerbation or OCS use.

Conclusions:

Being overweight or obese confers a greater risk of asthma exacerbation during pregnancy, and may be due to systemic macrophage activation.

Introduction

Asthma affects 8-12% of pregnant women worldwide [1,2], and up to 45% of these women have exacerbations requiring medical intervention during pregnancy [3]. Asthma contributes to an increased risk of poor perinatal outcomes including preterm birth [4], and neonatal hospitalisations [5], with some outcomes, such as low birth weight, linked to exacerbations [6]. Obesity is now known to adversely impact asthma [7]. In pregnancy, obesity can occur as a pre-existing condition (elevated BMI) or due to excessive gestational weight gain (GWG).

The prevalence of overweight and obesity among pregnant women has greatly increased in recent years [8]. Only one previous study has examined the relationship between maternal obesity and asthma exacerbations in pregnancy [9]. Hendler et al studied a cohort of American women with asthma from 1994 to 1999 and found that obese women with a pre-pregnancy BMI ≥30 kg/m^2 were 30% more likely to have an exacerbation requiring medical intervention during pregnancy than women who were non-obese (adjusted odds ratio [OR] 1.3, 95% confidence interval 1.1, 1.7) [9]. However, the mechanisms involved in this association have not been explored.

Inflammation has been proposed as a key mediator of adverse pregnancy outcomes associated with obesity [10]. Inflammation in asthma is often associated with allergen-driven eosinophilic airway inflammation. However, obesity itself is a pro-inflammatory state where macrophages in adipose tissue are increased and exhibit an activated pro-inflammatory (M1) phenotype which results in secretion of cytokines such as interleukin (IL)-6 and tumour necrosis factor (TNF)-α [10]. IL-6 leads to increased liver production of C-reactive protein (CRP), which is a typical feature of systemic inflammation in obesity. Toll-like receptor activation by fatty acids induces cleavage of the macrophage surface marker CD-163, which results in increased circulating soluble CD-163 (sCD-163), a marker of macrophage activation [11]. In addition, macrophages have an important role in the placenta, with significantly more placental macrophages in obese compared to non-obese women, accompanied by increased expression of pro-inflammatory cytokines including IL-6, and higher gene expression of macrophage markers including CD-68 [12].

This study tested the hypothesis that the risk of asthma exacerbation would be greater among overweight or obese pregnant women compared to healthy weight women, and that exacerbations would be driven by systemic inflammation rather than eosinophilic airway inflammation.

Materials and Methods

This is a secondary analysis of pregnant women with physician-diagnosed asthma, recruited from the John Hunter Hospital antenatal clinics, between April 2007 and November 2009, to a prospective study of exacerbations of asthma during pregnancy [13]. Concurrently, some women also participated in a randomised controlled trial (RCT, the Managing Asthma in Pregnancy Study, Australian and New Zealand Clinical Trials Registry: 12607000561482) in which women in the control group had their asthma treatment adjusted monthly according to symptoms and lung function, while women in the intervention group had their asthma treatment adjusted monthly according to fractional exhaled nitric oxide (FENO) as well as symptoms and lung function [14]. Inclusion and exclusion criteria were given in detail in previous publications [13,14]; women with pre-existing diabetes or hypertension were not excluded. Ethics approval was provided by the Hunter New England Area Health Service and the University of Newcastle Research Ethics Committees and women provided written informed consent for participation.

Data were included from all women who had height and weight measurements made at the baseline study visit, with a BMI ≥ 18.5 kg/m^2. Women with an early pregnancy BMI from 18.5 to 24.9 kg/m^2 were considered healthy weight, those with a BMI 25 to 29.9 kg/m^2 were considered overweight, and those with a BMI ≥30 kg/m^2 were considered obese, according to the Institute of Medicine (IOM) guidelines [15].

GWG was calculated as the average weight gain (kg/week) over the second and third trimester (from study recruitment to the final visit) and compared to IOM guidelines for rate of weight gain in pregnancy. These guidelines recommend that

women with a BMI in the healthy weight range gain 0.45 kg body weight / week in the second and third trimester (total weight gain 11.3 – 15.9 kg), while those who are overweight and obese have a recommended weight gain of 0.27 kg / week (total weight gain 6.8 – 11.3 kg) and 0.23 kg / week (total weight gain 5.0 – 9.7 kg), respectively [15].

This study involved monthly clinical visits, phone calls between visits (every 14 days), and additional visits during asthma exacerbations. Exacerbations requiring medical intervention were recorded prospectively and defined as hospitalisation, emergency department (ED) presentation, unscheduled doctor visit, and/ or a prescribed course of oral corticosteroids (OCS). Exacerbations which occurred at least 14 days apart were considered separate events. Each month, and during asthma exacerbations, asthma control was assessed using the validated Asthma Control Questionnaire (ACQ-7) [16]; lung function was measured by spirometry (EasyOne Spirometer, NicheMedical, North Sydney, Australia); and inhaled corticosteroid (ICS) use was assessed by direct questioning of prescribed dose and adherence. Smoking was assessed by self-report, and confirmed by urinary cotinine at visit 1 or 2 (≥ level 5 or 2840 nmol/L, Nicalert, NYMOX, St-Laurent, QC, Canada), and exhaled carbon monoxide (ECO) measurements (≥ 10ppm, piCO Smokerlyzer Breath CO Monitor, Bedfont, UK) at monthly visits. Perinatal outcomes were extracted from medical records after delivery.

The inflammatory profile was assessed using FENO; assessed using the Ecomedics chemiluminescence analyser (Ecomedics, Duernten, Switzerland), at a controlled flow rate of 50 ml/s, and serum measurements. Blood samples were collected via venepuncture at baseline (early second trimester) and tested for soluble CD163 (sCD-163, Trillium Diagnostics, IQ Products, The Netherlands), IL-6 (high sensitivity ELISA, R&D Systems, Minneapolis, USA) and CRP (high sensitivity ELISA, MP Biomedicals, Solon, Ohio) using ELISA. Peripheral blood eosinophil counts were measured by Hunter Area Pathology Service (Newcastle, NSW, Australia) using an automated analyser (Beckman Coulter LH780, Miami FL, USA). The baseline sample was used to assess atopy as previously described [14]. Subjects were considered atopic if the specific serum IgE to aeroallergen was ≥0·35 kUA/L.

Statistical analysis was performed using Stata 11 (StataCorp, College Station, TX) and GraphPad Prism 6 (GraphPad Software, Inc, La Jolla, CA). Results are presented as mean ± standard deviation (SD) or median (interquartile range, IQR) with Student's t-test or Mann Whitney test, and analysis of variance (ANOVA) or Kruskal Wallis test applied as appropriate. The STATA kwallis2 test was used to test for post-hoc significance [17]. The Chi square test was used to compare proportions. Two-sided tests with $P<0{\cdot}05$ were considered significant.

Results

One-hundred and sixty-eight pregnant women with asthma were recruited to the primary study. Three women were excluded from the secondary analysis because they were underweight (BMI<18.5kg/m^2), and one woman was excluded due to a missing baseline weight measurement. Of the 164 women remaining, 45 were healthy weight (27.4%), 53 were overweight (32.3%) and 66 were obese (40.2%). The majority of subjects were also enrolled in the RCT of asthma management in pregnancy (n=138, 84.1%), however there was no difference in allocation to the intervention within each BMI category (P=0.965). Three women had pre-existing diabetes (1 in overweight group, 2 in obese group), while one woman had pre-existing hypertension (obese group).

Table 1 shows subject characteristics across BMI categories. Maternal age, gestational age at baseline, atopy, smoking status and lung function were not significantly different between groups (P>0.05), while gravidity was significantly higher in the obese group (P=0.008). The obese women used a significantly higher ICS dose at baseline compared to the healthy weight women (P=0.022). In a subgroup of women with late pregnancy weight measurements (n=115, 70%), GWG was above guideline recommendations in 70% of the healthy BMI group, 79% of the overweight group, and 71% of the obese group. Median GWG in each group was 8.0 (7.2, 10.2) kg, 7.3 (5.7, 28.0) kg and 6.4 (4.1, 10.4) kg, respectively (P=0.088), over the course of the study.

Data on exacerbations were available for 91% of participants (40 healthy weight, 45 overweight and 64 obese). Exacerbations requiring medical intervention occurred in

a higher proportion of women in the overweight (51.1%) and obese groups (48.4%), compared to the healthy weight group (25%, Figure 1a, P=0.026, post-hoc significance for healthy weight vs overweight, P=0.014). Ten women of healthy BMI had exacerbations requiring medical intervention (12 events, consisting of one hospital admission and 11 unscheduled doctor visits, with two women prescribed OCS); 23 overweight women had exacerbations (36 events, all unscheduled doctor visits, with nine women prescribed OCS for 15 exacerbations); and 31 obese women had exacerbations (37 events, consisting of two hospital admissions, six ED presentations and 28 unscheduled doctor visits, with eight women prescribed OCS). The proportion of women requiring OCS for exacerbations in pregnancy was not significantly different between the groups (20% in healthy group, 39% in overweight group, 26% in obese group, P=0.437).

Data on exacerbations and GWG were available for 70% of participants (Figure 1b). There was no significant difference in the proportion of women with exacerbations based on GWG below/within guideline recommendations (45%) versus above guideline recommendations (46%, Figure 1b, P=1.0), nor was the proportion using OCS different between groups (19.4% vs. 9.5%, P=0.198).

Biomarkers

Serum sCD-163, CRP and IL-6 were significantly higher in overweight and obese subjects compared to healthy weight subjects at baseline (Table 1); the fold increase with BMI category is shown in Figure 2. Biomarkers were not significantly different between the GWG categories (data not shown).

Women were characterised according to baseline serum sCD-163 levels, as being below or above the median level (1113 mg/ml). The difference in proportion of women with exacerbations (sCD-163 below median [32.8%] vs. sCD-163 above median [49.3%]) did not reach statistical significance (P=0.053, Figure 3). However, a significantly higher proportion of women with sCD-163 levels above the median experienced at least one exacerbation requiring OCS during pregnancy, compared to women with sCD-163 levels below the median (19.4% vs. 7.5%, P=0.043, Figure 3).

The proportion of women with exacerbations (38% vs 44%, P=0.502), or OCS use (12% vs 12%, P=0.949), did not differ between women with baseline serum CRP levels at or below, versus above, the median level (7.0 mg/L). Likewise, when using a CRP cut-point of 3.0 mg/L, exacerbations were not significantly different between women with low CRP or high CRP (26.9% vs 44.4%, P=0.103) and OCS use was not significantly different between women with low CRP or high CRP (7.7% vs 13.9%, P=0.394).

There were no significant differences in the proportion of women with exacerbations (39% vs 52%, P=0.188), or OCS use (14% vs 10%, P=0.563), when comparing women with baseline serum IL-6 levels at or below, versus above, the median (0.97 pg/ml).

Women were characterised as having peripheral blood eosinophils which were low ($<0.3 \times 10^9$/L) or high ($\geq 0.3 \times 10^9$/L)[18]. There was no significant difference in the proportion of women with exacerbations between these groups (44% vs 47.7%,

P=0.691, Figure 4), or in OCS use between these groups (10.7% vs 22.7%, P=0.069).

Women were characterised as having FENO that was low (≤29 ppb) or high (>29 ppb) [14]; there was no significant group difference in the proportion of women with at least one exacerbation (41.5% vs 48.6%, P=0.451, Figure 5), or in OCS use (11.3% vs 16.2%, P=0.440).

Perinatal outcomes were extracted from the medical records after delivery. There were 47 infants born to the healthy weight mothers (2 sets of twins), 53 infants to the overweight mothers and 68 infants to the obese mothers (2 sets of twins). There was a significant difference between the groups in the proportion of mothers who had pregnancy-induced hypertension or gestational diabetes, which was higher in the obese group (Table 2, $P<0.05$).

There were 32 infants born (2 sets of twins) to women with GWG below or within guideline recommendations, and 86 born (1 set of twins) to women with GWG above the guideline recommendations. Infants whose mothers had GWG greater than recommendations were significantly less likely to be low birth weight or to be admitted to neonatal intensive care ($P<0.05$). There were trends towards higher rates of pregnancy-induced hypertension and gestational diabetes with excess GWG, but results did not reach significance (data not shown).

Discussion

Maternal overweight and obesity in early pregnancy, but not excess gestational weight gain, were associated with a greater proportion of women experiencing asthma exacerbations requiring medical intervention during pregnancy. sCD-163 was a stronger biomarker for exacerbation risk than peripheral blood eosinophils, in this population with actively treated asthma, and may have potential as a clinical tool in the future. Well described risk factors for asthma exacerbation during pregnancy include respiratory viral infection [13], maternal smoking [3] and non-adherence to ICS therapy [19]. Previously, obesity, but not overweight, has been associated with asthma exacerbations [9]. Our results showed that twice as many overweight or obese women had exacerbations compared to healthy weight women, compared to a 30% increase in obese vs non-obese women reported previously [9]. With pre-existing overweight and obesity, as opposed to excess gestational weight gain, there is likely to be established adipose tissue inflammation and the sCD-163 results support obesity-related macrophage activation leading to asthma exacerbation.

The rate of obesity in our population (40%) was high, compared to previous publications on asthmatic populations [9], and previous Australian population studies [8]. The increased gravidity of the obese women may have contributed to the higher obesity rates and BMIs observed in our study. Women who are obese or overweight are at risk of excess gestational weight gain, and when this is not lost post-partum, weight gain compounds with increasing gravidity. A recent study from South Australia of pregnant women with asthma, reported their average BMI to be 28.1 (6.8) kg/m^2 [20]. Hendler et al (1994-1999) reported that 30.7% of U.S. women with asthma were obese, which was significantly higher than the rate of obesity in women without asthma (25.5%) [9]. A study of asthmatic women from Quebec (1990-2002) reported that 20% of women were overweight and 17% were obese before pregnancy, and gestational weight gain was higher than recommendations in the obese women [21]. Over 70% of all women in our study gained more weight than recommended during pregnancy, however this was not associated with asthma exacerbations, or macrophage activation or systemic inflammation. Despite the lack of effect observed for asthma outcomes, our findings indicate that interventions for women planning pregnancy and to manage GWG are needed. Indeed, the metabolic

effects were observed, with a higher proportion of overweight and obese women experiencing poor pregnancy outcomes, (hypertension and gestational diabetes), which increase the short-term and long-term risk for adverse maternal and fetal outcomes.

One of the limitations of our study was that BMI was determined from measurements made early in pregnancy, rather than prior to pregnancy. However, this may be more accurate than self-reported pre-pregnancy weight, which is likely to be underestimated. In Callaway's study, the pre-pregnancy BMI was also estimated based on a measured early pregnancy weight, and an estimate of pregnancy associated weight gain [8]. Another limitation was that multivariate analysis was not conducted due to the sample size. However, the potential demographic confounders that were captured including maternal age, smoking status, ethnicity and education level were balanced between groups (Table 1, E Table 1).

One of the novel findings of the present study is that systemic macrophage activation was associated with asthma exacerbations in pregnancy. Women with "high" levels of sCD-163 (above the median) had significantly more exacerbations requiring OCS. sCD-163 was found to be correlated with the proportion of android fat (measured by dual energy X-ray absorptiometry) in obese non-pregnant women [22], and in obese girls with asthma, increasing sCD-163 was correlated with worse asthma control [22]. A study in pregnancies with gestational diabetes found elevated serum sCD-163 levels in mothers, as well as increased CD-163 positive cells and increased release of CD-163 from the placenta and adipose tissue, compared to pregnancies without gestational diabetes [23]. Our results were consistent with previous studies which have found that obese pregnant women have higher serum levels of CRP [12] and IL-6 [24], than women of healthy BMI [25]. However, these biomarkers were not associated with a greater risk of exacerbation in pregnancy. In addition, our study did not show a significant association between measures of eosinophilic inflammation and exacerbation risk in pregnancy, suggesting that obesity-related markers of macrophage activation may be more important in this context.

This population had regular monitoring of asthma control and active adjustment of ICS, including a concurrent study in which a subgroup had active management by

FENO, which would reduce exacerbation risk [14] and is the likely reason we failed to identify an effect of blood eosinophils. This is important since our results show that the effect of an elevated BMI on asthma exacerbations is still significant in a treated asthma population.

Our data on perinatal outcomes by BMI category was consistent with previous data in non-asthmatic populations [8], and to an extent with data from asthmatic women, reported by Hendler et al [9]. Callaway et al reported that overweight and obese women were at increased risk of hypertensive disorders of pregnancy, gestational diabetes, caesarean section and premature delivery [8]. Hendler et al also found obesity to be associated with hypertensive disorders, gestational diabetes and caesarean section; however, there was no influence on preterm birth [9]. Data from Quebec demonstrated no significant difference in adverse perinatal outcomes in obese asthmatic women, including preterm delivery and small for gestational age infants [21]. Our study was not adequately powered to detect a difference in all adverse outcomes based on maternal BMI; however, we did observe a significantly higher rate of pre-eclampsia and gestational diabetes with obesity.

Maternal obesity and the inflammation associated with obesity, have also been linked to alterations in immune responses, lung development and increased wheeze or asthma in the offspring [26,27]. Data from a large Dutch cohort study, demonstrated that early pregnancy maternal CRP in the highest quartile was associated with eczema at age 4 years, while high cord blood CRP was associated with an increase in wheezing [28]. A New Zealand study described a relationship between more subcutaneous fat (and greater gain in subcutaneous fat) during pregnancy and more wheeze in 18 month old offspring [29]. The Generation R study showed that amongst mothers with a history of asthma or allergy, there was an increased risk of preschool wheeze with maternal obesity [30]. These observations are important because women with asthma have an increased risk of having children with wheeze or asthma themselves [31], and obesity may increase this risk even further.

In conclusion, we showed that early pregnancy overweight and obesity was associated with an increase in asthma exacerbations during pregnancy, while

excess gestational weight gain was not. Markers of macrophage activation and systemic inflammation were elevated in the obese group, suggesting systemic activation of macrophages may be a contributing mechanism to exacerbation risk in pregnancy. This study highlights the potential importance of pre-pregnancy weight reduction for improving asthma outcomes, as well as perinatal outcomes, during pregnancy.

Acknowledgements

The authors thank Kelly Steel and Karen McLaughlin (University of Newcastle) for assistance with clinical assessments, and Michelle Gleeson, Dr Katie Baines and A/Prof Jodie Simpson (University of Newcastle) for assistance with ELISA measurements.

Figure Legends

*Figure 1: The proportion (with 95% confidence interval) of women with exacerbations requiring medical intervention during pregnancy according to (a) body mass index (BMI) category; and (b) gestational weight gain (GWG) category. * Indicates statistical significance (Healthy vs Overweight, P=0.0137 post-hoc analysis, healthy vs obese, P=0.0173 post-hoc analysis)*

*Figure 2: Fold increase in serum biomarkers (sCD163, CRP, IL-6) in the overweight and obese groups compared to the healthy BMI group. *indicates P<0.05 vs healthy, ^ indicates P<0.05 vs overweight.*

*Figure 3: The proportion (with 95% confidence interval) of women with exacerbations requiring medical intervention, or exacerbations requiring OCS use, according to sCD163 level (below or above median). * Indicates P<0.05*

Figure 4: The proportion (with 95% confidence interval) of women with exacerbations requiring medical intervention or exacerbations requiring OCS use according to peripheral blood eosinophils

Figure 5: The proportion (with 95% confidence interval) of women with exacerbations requiring medical intervention or exacerbations requiring OCS use according to FENO

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Table 1: Baseline subject characteristics of pregnant women with asthma

	Healthy Weight n = 45	Overweight n = 53	Obese n = 66	P value
Demographics				
Age (years)	27.9 (4.9)	29.7 (6.2)	28.3 (5.4)	0.215
Weight (kg)	60.0 (54.1, 66.3)	72.0 (68.0, 78.0) #	92.2 (85.0, 104.8) #^	**<0.0001**
BMI (kg/m^2)	22.3 (21.1, 24.0)	26.4 (25.6, 28.0)#	33.6 (31.5, 39.7)#^	**0.0001**
Gestational age (weeks)	16.9 (15.1, 19.1)	17.4 (14.9, 18.7)	16.7 (14.9, 18.7)	0.813
Parity	0 (0, 1)	1 (0, 1)	1 (0, 2)	0.076
Gravidity	2 (1, 3)	2 (1, 3)	2.5 (2, 4) #	**0.008**
Pre-bronchodilator lung function	n=42	n=46	n=50	
FEV_1 (% predicted)	93.3 (13.9)	93.9 (15.0)	95.1 (15.0)	0.837
FVC (% predicted)	105.9 (18.3)	104.3 (14.3)	102.8 (13.4)	0.641
FEV_1/FVC (%)	77.5 (9.0)	78.4 (8.3)	80.4 (6.8)	0.194
SABA use (times/week)	1.5 (0, 11) n=42	1 (0, 14) n=51	2 (0, 7) n=61	0.892
Maintenance ICS use	14 (31.1%)	14 (26.4%)	20 (30.3%)	0.854
ICS used in past week (adjusted for non-adherence), BDP equivalent (mcg/day)	343 (157, 554)	571 (400, 929)	800 (600, 1500) #	**0.022**
ACQ7 score	0.94 (0.73)	1.23 (1.16)	1.08 (0.88)	0.309
Uncontrolled asthma (ACQ7 > 1.5), n (%)	10 (22.2%)	20 (37.7%)	19 (28.8%)	0.240
Atopy	34 (77.3%)	35 (71.4%)	41 (66.1%)	0.459
Smoker	8 (17.8%)	15 (28.3%)	12 (18.2%)	0.323
Asthma History				
Age of diagnosis (years)	7.0 (4.5, 13.5)	6.0 (3.0, 14.0)	9.5 (4.0, 15.8)	0.280
Hospital admissions past 2 years, n (%)	0	1 (1.9%)	2 (3.0%)	0.504
ED Visits in past 2 years, n (%)	3 (6.7%)	8 (15.1%)	10 (15.2%)	0.351
OCS in past 2 years, n (%)	6 (13.3%)	10 (18.9%)	10 (15.2%)	0.741
Pregnancy in past 2 years, n (%)	15 (33.3%)	19 (35.8%)	30 (45.5%)	0.371
Biomarkers				
FENO (ppb)	15.7 (6.4, 31.0)	16.3 (6.7, 35.1)	12.3 (6.6, 22.6)	0.544
Peripheral blood eosinophils (10^9/L)	0.2 (0.1, 0.3) n=40	0.2 (0.1, 0.3) n=42	0.2 (0.1, 0.3) n=58	0.781

Serum sCD163 (mg/ml)	954.8 (782.8, 1189.3)	1064.3 (899.1, 1459.9)	1287.4 (982.4, 1877.6) [#]	**P=0.0016**
Serum CRP (mg/l)	3.7 (1.9, 5.8)	6.6 (4.2, 10.7) [#]	12.5 (6.0, 20.0) [#^]	**P<0.0001**
Serum IL-6 (pg/ml)	0.64 (0.48, 0.80) n=22	0.95 (0.78, 1.31) [#] n=31	1.28 (0.95, 1.75) [#] n=44	**P<0.0001**

Values are mean (SD) or median (interquartile range) or n (%).

ACQ, asthma control questionnaire; BMI, body mass index; BDP, beclomethasone diproprionate; CRP, C-reactive protein; ED, emergency department; FENO, fractional exhaled nitric oxide; FEV_1, forced expiratory volume in 1 second; FVC, forced vital capacity; ICS, inhaled corticosteroids; IL-6, interleukin-6; OCS, oral corticosteroids

[#] *post hoc significant vs Healthy;* ^ *post hoc significant vs Overweight*

Table 2: Perinatal outcomes according to maternal BMI

	Healthy Weight	Overweight	Obese	P Value
Number of mothers	45	53	66	
Number of infants	47	53	68	
Gestational age at delivery (weeks)	39.6 (38.3, 40.7) n=45	40.0 (39.1, 40.3) n=52	39.4 (38.4, 40.3) n=66	0.534
Preterm Delivery <37 completed weeks, n (%)	4/45 (8.9%)	4/52 (7.7%)	9/66 (13.6%)	0.533
Birth weight (g)	3340 (3010, 3719) n=46	3250 (2975, 3590) n=51	3500 (3060, 3880) n=65	0.201
Low Birth Weight (<2500g), n (%)	7/46 (15.2%)	6/51 (11.8%)	4/65 (6.2%)	0.289
High Birth Weight (>4000g), n (%)	5/46 (10.9%)	8/51 (15.7%)	12/65 (18.5%)	0.551
Birth length (cm)	50.8 (49.4, 53.0) n=36	51.0 (49.0, 53.0) n=46	51.5 (49.5, 53.0) n=55	0.754
Birth head circumference (cm)	34.5 (33.0, 35.0) n=45	34.0 (33.3, 35.0) n=51	34.7 (33.5, 36.0) n=65	0.069
Spontaneous Labour, n (%)	24/45 (53.3%)	38/52 (73.1%)	35/65 (53.8%)	0.129
Vaginal Delivery, n (%)	37/46 (80.4%)	41/52 (78.8%)	42/65 (64.6%)	0.103
Caesarean Delivery, n (%)	9/46 (19.6%)	11/52 (21.2%)	23/65 (35.4%)	0.103
Pre-eclampsia, n (%)	2/44 (4.5%)	4/52 (7.7%)	3/65 (4.6%)	0.232
Pregnancy induced hypertension, n (%)	1/44 (2.3%)	2/52 (3.8%)	9/65 (13.8%)	**0.038**
Gestational Diabetes, n (%)	0	3/52 (5.8%)	8/65 (12.3%)[#]	**0.041**
Postpartum haemorrhage, n (%)	3/44 (6.8%)	1/52 (1.9%)	4/65 (6.2%)	0.465
Stillbirth, n (%)	0	0	1/66 (1.5%)	0.479
Neonatal intensive care admission, n (%)	10/46 (21.7%)	7/52 (13.5%)	8/66 (12.1%)	0.345

Values are mean (SD) or median (interquartile range) or n (%).

[#] *post hoc significant vs Healthy*

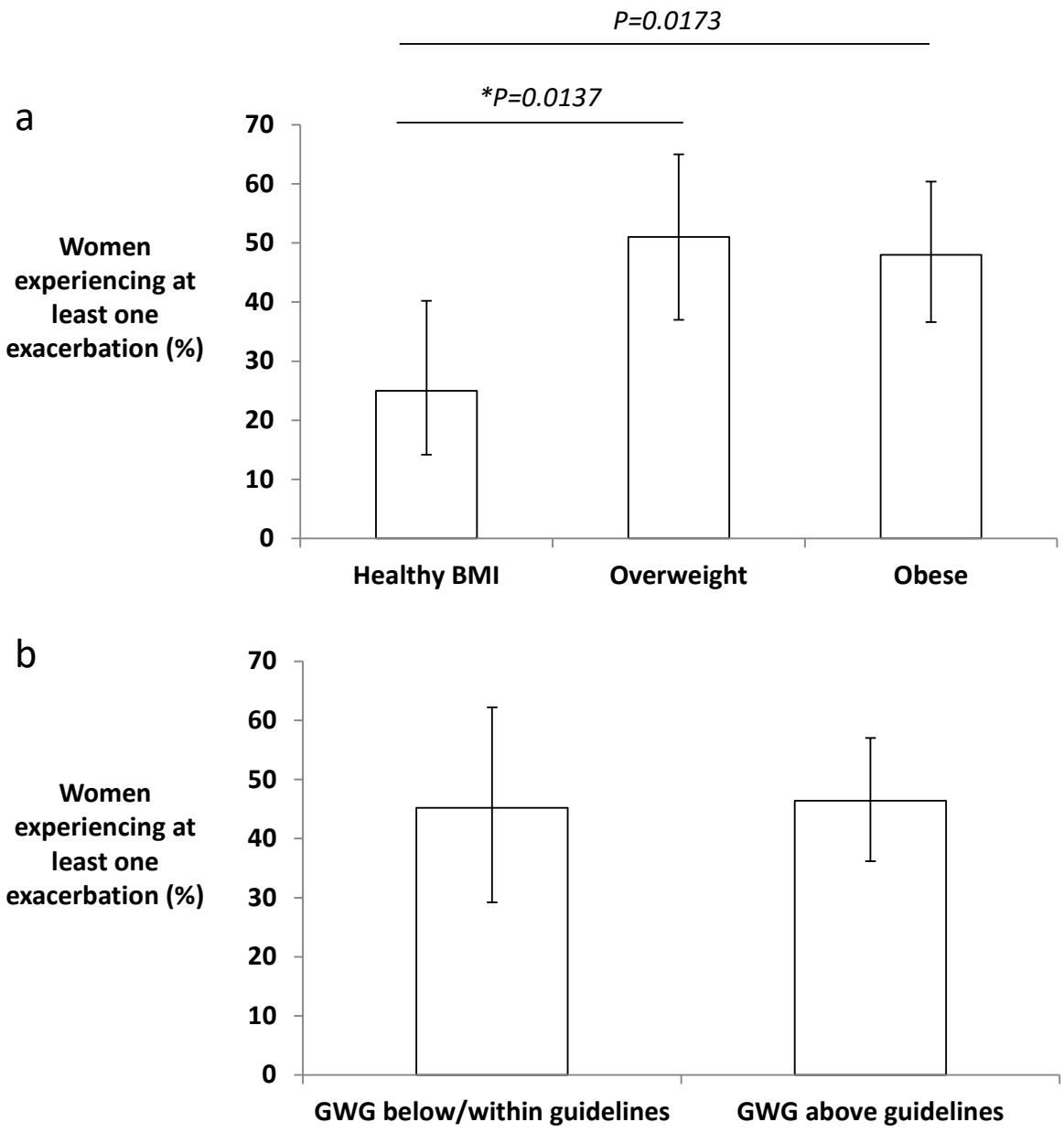
a
P=0.0173
*P=0.0137
Women experiencing at least one exacerbation (%)
70
60
50
40
30
20
10
0
Healthy BMI
Overweight
Obese
b
Women experiencing at least one exacerbation (%)
70
60
50
40
30
20
10
0
GWG below/within guidelines
GWG above guidelines

Fig1

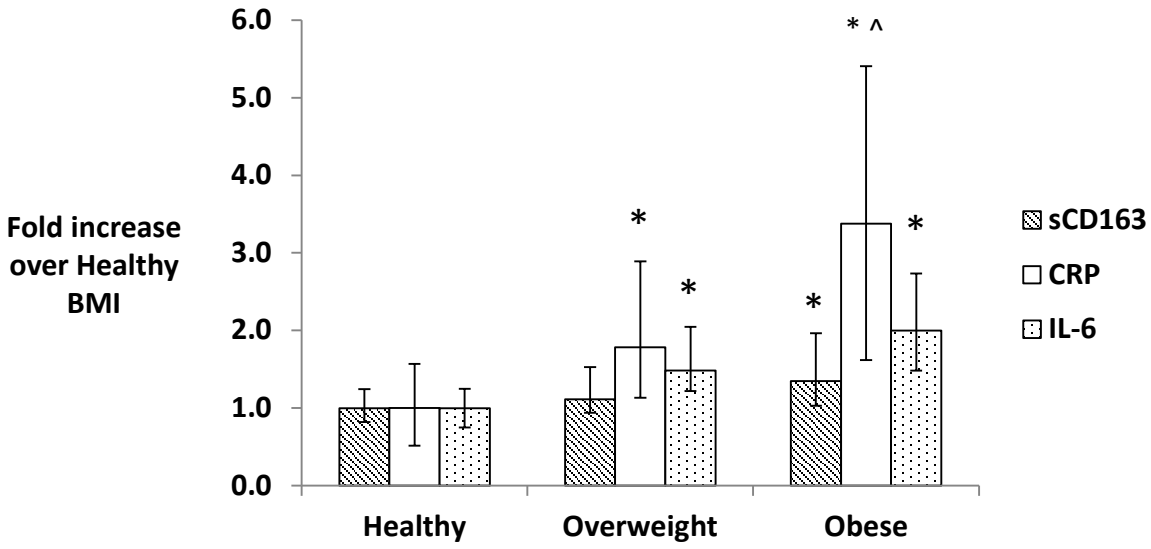
Fold increase over Healthy BMI
6.0
5.0
4.0
3.0
2.0
1.0
0.0
* ^
*
*
*
*
Healthy
Overweight
Obese
sCD163
CRP
IL-6

Fig2

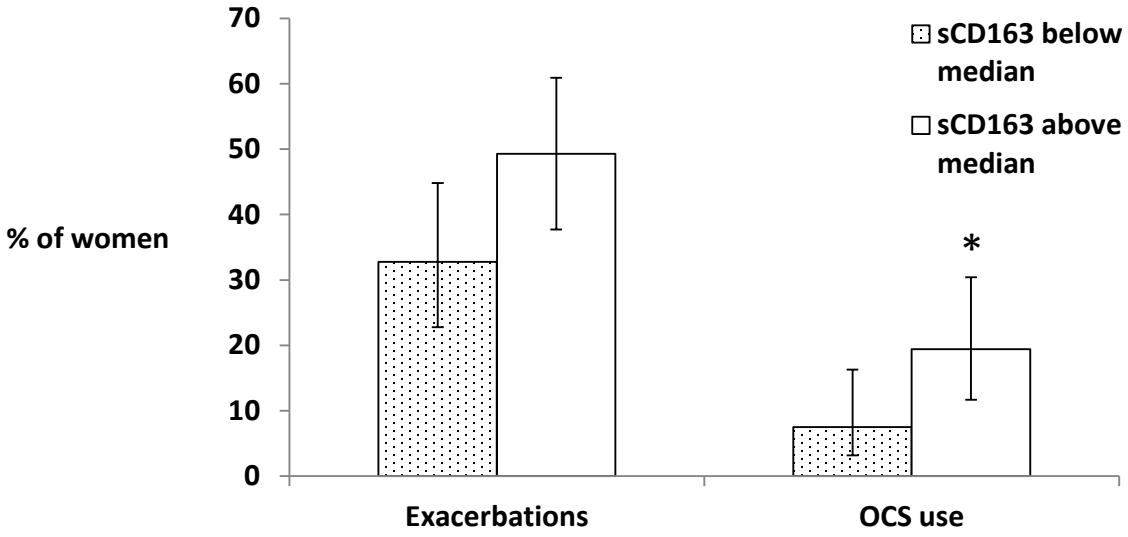
70
60
50
40
30
20
10
0
% of women
sCD163 below median
sCD163 above median
*
Exacerbations
OCS use

Fig3

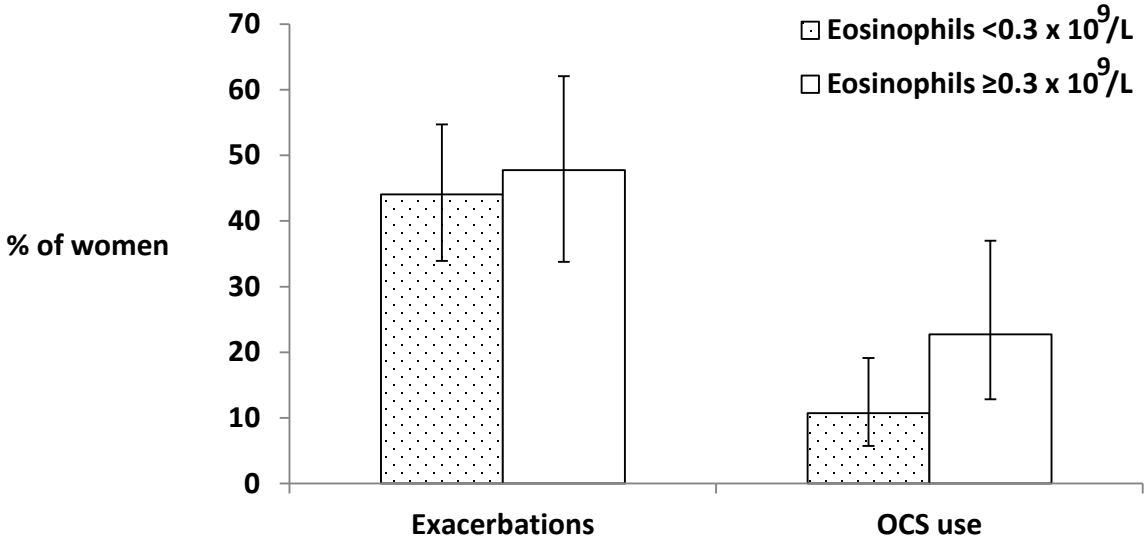
% of women
70
60
50
40
30
20
10
0
Eosinophils <0.3 x 10^9/L
Eosinophils ≥0.3 x 10^9/L
Exacerbations
OCS use

Fig4

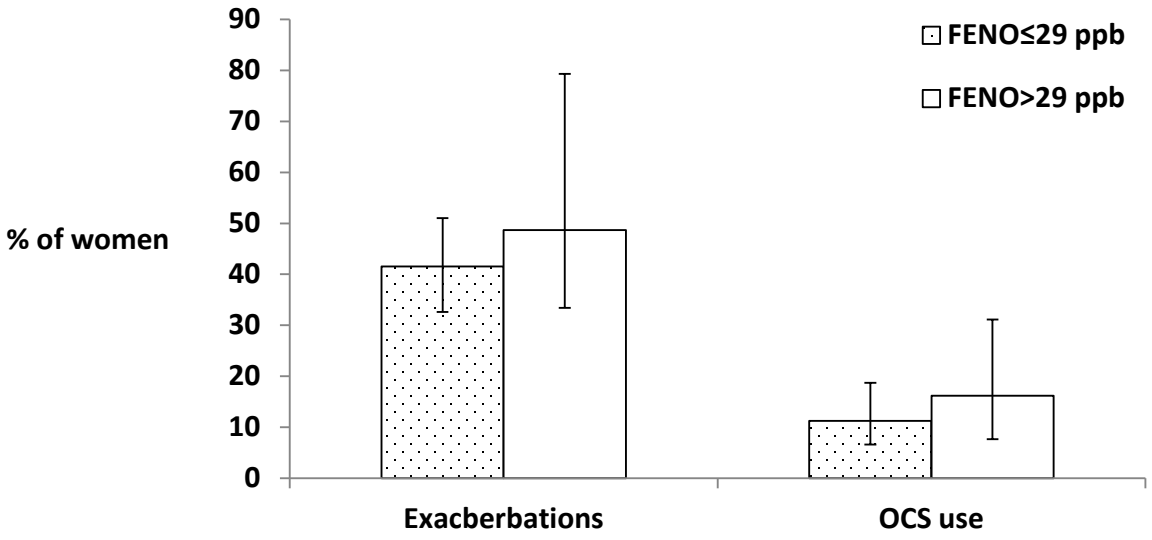
90
80
70
60
50
40
30
20
10
0
% of women
FENO≤29 ppb
FENO>29 ppb
Exacberbations
OCS use

Fig5