MATERNAL OBESITY AND SMOKING DURING PREGNANCY PERIOD ASSOCIATED WITH DEVELOPMENT OF POLYCYSTIC OVARY SYNDROME IN THE OFFSPRING IN LATER LIFE: A NATIONAL REGISTRY-BASED COHORT STUDY

H Valgeirsdottir[a*], E Vanky[b,c], I Sundstrom-Poromaa[a], N Roos[d], T S Lovvik[b], O Stephansson[d,e], A-K Wikstrom[a,d]

[a]Department of Women's and Children's Health, Uppsala University, Uppsala, Sweden.

[b]Department of Obstetrics and Gynecology, St. Olav's Hospital, Trondheim University Hospital, Trondheim, Norway.

[c]Department of Laboratory Medicine, Children's and Women's Health, Norwegian University of Science and Technology, Trondheim, Norway.

[d]Department of Medicine, Solna, Clinical Epidemiology Unit, Karolinska Institute, Stockholm, Sweden.

[e]Department of Women's and Children's Health, Karolinska Institute, Stockholm, Sweden.

Precis: Maternal obesity and smoking are associated with increased risk of PCOS in the offspring. The data give further support to continue the ongoing work in antenatal care to reduce these risk factors

H Valgeirsdottir et al

Correspondance: Heiddis Valgeirsdottir MD

Department of Women's and Children's Health, Uppsala University, 751 85 Uppsala, Sweden.

Tel +46 736 537 476

Email heiddis.valgeirsdottir@kbh.uu.se

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Abstract

Objective: To study the associations between prenatal exposures and risk of developing PCOS.

Design: National registry-based cohort study.

Setting: Sweden.

Study population: All girls born in Sweden during the years 1982 to 1995 (N=681 123).

Methods: The girls were followed until the year 2010 for a diagnosis of PCOS. We estimated the associations between maternal BMI, smoking, preeclampsia and diabetes in the index pregnancy, size at birth and the risk of developing a PCOS diagnosis. Risks were calculated by adjusted hazard ratio (cHR and aHR) and 95% confidence intervals (CI). Adjustments were made for the other exposures, as well as for maternal age, parity, years of involuntary childlessness, country of birth, education and heritability for PCOS on the maternal side.

Main outcome measures: PCOS diagnosis at 15 years or later.

Results: During the follow-up 3738 girls were diagnosed with PCOS (0.54%). Compared to girls who had a mother with normal BMI during pregnancy, girls with overweight or obese mothers had 1.5-2.0 times higher risk of PCOS (aHRs; 95% CIs 1.52; 1.36-1.70 and 1.97; 1.61-2.41, respectively). The risk of PCOS was also increased if the mother smoked during pregnancy (1-9 cigarettes/day: aHR 1.31; 95% CI: 1.18-1.47 and $\geq$ 10 cigarettes/day: aHR 1.44; 95% CI: 1.27-1.64). Being born small and having a small head circumference for gestational age were both associated with later diagnosis of PCOS in crude estimates, but the associations were not significant after adjustments.

Conclusions: This study provides new information on epidemiology of prenatal exposure of smoking and BMI and development of PCOS in the offspring.

Keywords: birthweight / body mass index / prenatal care / polycystic ovary syndrome / smoking

Introduction

Polycystic ovary syndrome (PCOS) is an endocrine condition affecting 6-10% of women (1) and besides polycystic ovarian morphology, the syndrome is associated with menstrual irregularity and

hyperandrogenism. (2) Women with PCOS frequently present with infertility, central adiposity, insulin resistance and have an increased risk of developing type 2 diabetes mellitus. (3)

The etiology of PCOS is multifactorial and not fully understood. Evidence suggests that both heritable and environmental factors play a role in the pathogenesis of PCOS. (4) Several genes associated with PCOS have been identified (5-7) and diverse genes seem responsible for different phenotypes of PCOS, (8) implying a complex genetic disorder. Ethnicity, (9) family history, (10) and lifestyle (11) also influence the risk of PCOS. Obesity and greater weight gain are associated with PCOS status (12) and smoking increases the free androgen index in women with PCOS. (13)

It is possible that various factors affecting the female fetus in the intrauterine life contribute to PCOS development later in life. For instance, increased levels of testosterone during fetal life have been suggested to be involved in the PCOS pathogenesis. A number of animal models have been used, demonstrating that sheep (14), monkeys (15), rats (16, 17) and mice (18, 19) treated with testosterone or dihydro-testosterone prenatally exhibit ovarian and endocrine traits similar to women with PCOS, such as luteinizing hormone hypersecretion, enlarged polyfollicular ovaries, and functional hyperandrogenism. Some human studies have suggested that high and low birthweight are associated with increased risk of developing PCOS, (4, 20, 21) while other studies have not shown such association. (22-24) One study indicated that women whose mothers had diabetes had higher risk of being diagnosed with PCOS. (4) Apart from these exposures, human data is essentially lacking.

We thus designed this study to investigate if prenatal exposures were associated with the risk of developing PCOS in women.

Methods

The Swedish National Board of Health and Welfare gave access to information from the Swedish Medical Birth Registry, the Patient Registry and the Cause of Death Registry. Statistics Sweden provided data from the Education Registry and the Total Population Registry. Individual record linkage between the registries was possible through each individual's unique personal registration number, which is assigned to Swedish residents at birth or immigration. (25)

The Birth Registry contains data on 98% of all births in Sweden since 1973 and includes prospectively collected demographic data, information on reproductive history, and complications that occur during pregnancy, delivery and the neonatal period. In Sweden, antenatal care is standardized

and free of charge. (25) During the first antenatal visit, usually taking place at the end of the first trimester, the mother is interviewed about her medical and obstetric history, including her height and current smoking habits. Data on some chronic diseases (such as pre-gestational diabetes) and smoking habits are recorded by check boxes. Maternal weight is measured wearing light indoor clothing. Complications during pregnancy and delivery are classified according to the International Classification of Diseases (ICD), as noted by the responsible obstetrician at discharge from the delivery hospital. Information on each pregnancy and delivery is forwarded to the Birth Registry through copies of standardized antenatal, obstetric, and pediatric records.

The Patient Registry includes information on dates of hospital admissions and diagnoses since the 1960s, with full national coverage since 1987. (26) Since 2001 the Registry also covers specialized outpatient visits, including visits to a gynaecologist or reproductive specialist. Diagnoses are classified according to ICD-codes.

Study population

All singleton, live-born girls between the 1st of January 1982 and the 31st of December 1995 who reached at least 15 years of age were included in the study population (Figure 1). The mothers' height and weight were registered in the Birth Registry 1982-1989 and from 1992 and onwards, enabling the calculation of early pregnancy body mass index (BMI). Data on smoking habits in early pregnancy and years of involuntary childlessness before the index pregnancy have been recorded in the Birth Registry since 1982. Information on gestational diabetes was only available from 1989 and forward. We started to follow the offspring in the Patient Registry when they turned 15 years of age and we were able to follow the population to a maximum of 28 years of age at the end of 2010.

Variables

We collected data from the Swedish Birth Registry on maternal height, weight, smoking habits, age at delivery, parity and involuntary childlessness before the index pregnancy, presence of preeclampsia or diabetes during the index pregnancy, and the offspring's size and gestational age at birth. Preeclampsia and pre-gestational diabetes were identified by ICD codes and pre-gestational diabetes was further identified by the check box from the first antenatal visit. Diagnoses are coded according to

the Swedish versions of the International Classification of Diseases (ICD). ICD version 7 was applicable between 1964 and 1968, version 8 between 1969 and 1986, version 9 between 1987 and 1996 and version 10 from 1997. Preeclampsia was defined as ICD codes: ICD-8: 637.03-99 and ICD-9: 642E-G. Pre-gestational diabetes was defined as a mark in a corresponding check-box at the first antenatal visit and/or ICD-8 codes 250 and 761.1 and ICD-9 codes 250 and 648A. Gestational diabetes was only available in ICD-9 and refers to 648W. We had different definitions to estimate offspring size at birth. First we categorized by birthweight (<2500, 2500-4499, ≥4500 g). Secondly, we categorized standardized birthweight according to gestational age and sex-specific Swedish birthweight curves. (27) Appropriate for gestational age (AGA) describes infants whose weight is within the normal range for the gestational age. Small for gestational age (SGA) were infants with birthweight below the 10th percentile for the gestational age and large for gestational age (LGA) were infants with birthweight above the 90th percentile. During the mid-1980's ultrasound estimation of gestational age was introduced in Sweden. If no ultrasound scan was available, the first day of the last menstrual period was used to calculate gestational age at delivery. Thirdly, we calculated the ponderal index, defined as birthweight/height^3×100. Low and high ponderal index were defined as a ponderal index score among the 10% lowest or highest in the study population. Finally, we calculated the head circumference for gestational age. Small and large head circumference for gestational age were defined as more than 10% below the mean head circumference for gestational age, respectively, according to the Swedish reference curve for newborn infants. (28) Variables were categorized as shown in Tables 1 and 2.

Data on maternal education in 2010, country of birth and maternal PCOS diagnosis were collected from the Education Registry, the Registry of the Total population and the Patient Registry, respectively.. Variables were categorized as maternal education ≤ 11 years or ≥ 12 years, maternal country of birth as Nordic or non-Nordic, and if the mother had PCOS diagnosis or not (during the years 1964 to 2010). Until year 1990 the criteria for Stein-Levendahl syndrome was used for the diagnosis of PCOS syndrome in Sweden. Year 1990 the NIH criteria was induced for PCOS diagnosis. According to NIH criteria the woman needs to fulfil both of these criteria for diagnosis of PCOS: Clinical or biochemical hyperandrogenism and chronic anovulation. (29) From 2003, Rotterdam criteria were swiftly introduced in clinical practice in Sweden. Even so, the NIH criteria are still in practice in adolescent girls, when transvaginal ultrasound is considered unfeasible. According to

the Rotterdam criteria the woman needs to fulfil two of three criteria for PCOS diagnosis: Oligo- or anovulation, clinical or biochemical hyperandrogenism and polycystic morphology of the ovaries. (2) PCOS diagnosis was defined as ICD codes: ICD-7, 275.20, ICD-8, 256.90, ICD-9, 256E and ICD-10, E28.2. In the Swedish version of ICD-7 and ICD-8 the codes correspond to the formerly known Stein-Leventhals syndrome. For versions ICD-9 and ICD-10 the code is specified as polycystic ovary syndrome and Stein-Leventhals syndrome.

The primary outcome variable was a PCOS diagnosis in the offspring at ≥15 years of age. The PCOS diagnosis was retrieved from the Patient Registry.

Statistical analyses

Cox regression analysis was used to estimate hazard ratios with 95% confidence intervals (CI) for the association between maternal and newborn characteristics and risk of a PCOS diagnosis in the offspring. Test of Schoenfeld residuals was performed and showed fitness for the variables in the model. Adjustments were made for all maternal characteristics (Table 1) when testing each variable in turn.

Since information on BMI was missing in many cases before 1992, we re-ran our analyses without inclusion of BMI in our model. We thereafter estimated the association between maternal smoking and birthweight for gestational age and risk of PCOS diagnosis in those with and those without maternal BMI data.

The statistical software package SAS 9.4 (version 6.1; SAS, Cary, NC, USA) was used for analysis. The study was approved by the regional ethical committee board in Stockholm, Sweden (2008/1182-31/4 and 2011/1856-32).

Funding

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Results

In the final cohort of 681 123 girls, 3738 (0.54%) had been diagnosed with PCOS. In total, the follow-up included 4 516 270 person years.

Table 1 illustrates diagnosis of PCOS in the offspring and maternal characteristics. Girls who developed PCOS had more often mothers with BMI in the higher categories, who smoked during pregnancy, were younger, primiparous, and had more years of involuntary childlessness before the index pregnancy than girls without PCOS. Mothers of the PCOS girls had more often also a lower educational level, were more often born in non-Nordic countries and had more often been diagnosed with PCOS themselves (1.2% compared with 0.2% of the mothers to the girls without PCOS diagnosis). However, pre-gestational diabetes, gestational diabetes or preeclampsia were not more common in the mothers of the PCOS offspring.

Table 2 illustrates newborn characteristics and later diagnosis of PCOS in the offspring. Girls who later developed PCOS were more often born small for gestational age, were more often thinner at birth and had smaller head circumferences than those without PCOS. Birthweight <2500 g and preterm birth (<37 gestational weeks) were not associated with a later PCOS diagnosis.

Table 3 illustrates the adjusted associations between maternal characteristics and later diagnosis of PCOS. A dose-response pattern was noted between maternal BMI and maternal smoking in the index pregnancy and risk of PCOS diagnosis in the offspring. The adjusted risk for PCOS was increased 2-fold if the mother was obese (BMI $\geq$30 kg/m^2) compared with mothers with normal BMI, and the risk was increased by 1.44-fold if the mother was a heavy smoker compared with a non-smoker. Mothers born in a non-Nordic country, and mothers with PCOS, more commonly gave birth to a girl who developed PCOS. There were no associations between maternal age, parity, involuntary childlessness, diabetes or preeclampsia in the index pregnancy and development of PCOS in the offspring.

Table 4 illustrates the associations between birth size and later diagnosis of PCOS. According to crude hazard ratio being born SGA as well as having small head circumference for gestational age increased the risk of PCOS later in life.

Supporting information, Table S1, illustrates the stratified analyses on women with and without information about BMI. The effect of maternal smoking and birthweight for gestational age on PCOS diagnosis in the girl offspring was similar independent on whether information about BMI was available or not.

Discussion

Main findings

In this large population-based cohort we found associations between maternal obesity and smoking during pregnancy and a later PCOS diagnosis in the offspring. Being born SGA and having a small head circumference for gestational age were both associated with later diagnosis of PCOS in crude estimates, but the associations were not significant after adjustments.

Interpretations

Our results indicate that a high maternal BMI is an independent risk factor for later development of PCOS in the offspring. This may be a direct or indirect effect of maternal fat tissue synthesis and secretion of adipocytokines, or of raised or altered inflammatory factor profiles, disturbed glucose metabolism or hormonal effects on the fetus, but it could also be a consequence of genetic factors. An abundance of fat tissue may also have direct effects on the placenta. In overweight and obese women placental mitochondrial dysfunction, reduced mitochondrial biogenesis and a decrease in placental ATP levels have been noted. (30) Further, it was recently demonstrated that obesity interferes with the initial steps of placental cholesterol metabolism, leading to lower estradiol and progesterone synthesis. (31) In addition, BMI is positively correlated with maternal testosterone levels during pregnancy, (32) and maternal weight gain is associated with amniotic fluid testosterone levels in female fetuses. (33) Such effects of maternal obesity may affect the fetal hormonal profile and, in turn, influence PCOS development in the offspring. Presumably, daughters to women with high BMI, have similar nutritional habits as their mothers, which by itself could increase their risk of overweight and obesity, and thereby their risk of developing PCOS. In this study we did not have information on the BMI of the grown-up offspring girls to substantiate such a relation.

In the present study we also noted a dose-dependent association between maternal smoking and offspring PCOS diagnosis, even after adjustment for maternal BMI, educational level and maternal PCOS diagnosis. Maternal smoking has not previously been associated with a risk for PCOS, (34) but it has been associated with decreased levels of reproductive hormones, (35) and reduced fertility in the adult female offspring. (36) Smoke-exposed fetal ovaries exhibit a higher density of primordial follicles, (37) reduced germ cell and somatic cell proliferation, (38-40) and genes

important for ovarian development are dysregulated, (37) suggesting that maternal smoking may have direct effects on the fetal ovaries.

Association with development of PCOS and relative birthweight was noted. Prior studies in this area have been inconsistent, demonstrating that both high and low birthweight are associated with PCOS development. (4, 20, 21) Melo et al. reported a higher prevalence of PCOS in women born SGA than AGA. (21) Davies et al. did not find a higher prevalence of PCOS in SGA or LGA infants but reported that a low ponderal index was associated with all three diagnostic criteria for PCOS. (20) In a large cohort study, Mumm et al. observed an association between PCOS and high birthweight, but not with birthweight when adjusted for gestational age. (4) We did not observe association between later PCOS and actual birthweight, which is consistent with several other studies. (22, 24, 41, 42) However, we demonstrate association between later PCOS and low birthweight and small head circumference for gestational age in crude analyses. This replication of the relative size at birth and development of PCOS clearly points to the importance of antenatal factors for the fully expressed syndrome. After adjustment for maternal factors like ethnicity, country of birth, maternal PCOS diagnosis or preeclampsia in the index pregnancy the association was attenuated and no longer significant in the present study. This finding indicates that maternal factors might be the underlying cause of the association between relative size at birth and development of PCOS.

Strength and limitations

An important strength of this study is the large size of the cohort including a whole country, which enabled us to assess more prenatal exposures than in former studies. (4, 20-22, 24) Information on the exposure variables was prospectively collected through the birth registration, which limits recall bias. Free and standardized antenatal and delivery care minimized the possibility of residual confounding and increased the generalisation of the results.

The major limitation of the study is the low prevalence of PCOS, 0.54%, compared to 5-10% in studies that have aimed to report the prevalence of PCOS. (1) This low prevalence depends on the register-based study design and is in line with the prevalence of PCOS in studies with similar design. (4, 34) Thus, only women who sought medical care for their symptoms and were diagnosed with PCOS in hospitals (inpatient or outpatient) were included, which could lead to misclassification of

PCOS women to the non-PCOS groups and driving our estimates towards no real difference. Further, our cases of PCOS received their diagnosis before the age of 28 years. Irregular menstruation and polycystic morphology of the ovaries are more common in adolescents and therefore it is important for clinicians to only set the PCOS diagnosis in juvenile population when PCOS symptoms are severe and clinically important. Earlier studies have shown that young patients with PCOS diagnosis are likely to reflect the more extreme end of the PCOS-associated clinical presentations, including severe ovulatory dysfunction and hirsutism. (44). Our results are therefore presumably more likely to be generalizable to the most severe PCOS phenotypes. Information on BMI was missing in one third of the women, largely because BMI was not collected in the maternal report for a certain period of years. However, when we performed a stratified analysis on women with and without information about BMI, our results were similar in both groups indicating that this did not influence the findings.

Conclusion

Factors during intrauterine life such as maternal smoking and obesity may influence the risk for later PCOS in the offspring. Hopefully it will be possible to restrict maternal smoking or obesity with education and interventions in the primary maternal care or even at family planning.

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Figure 1. Flow chart showing the identification of the study population

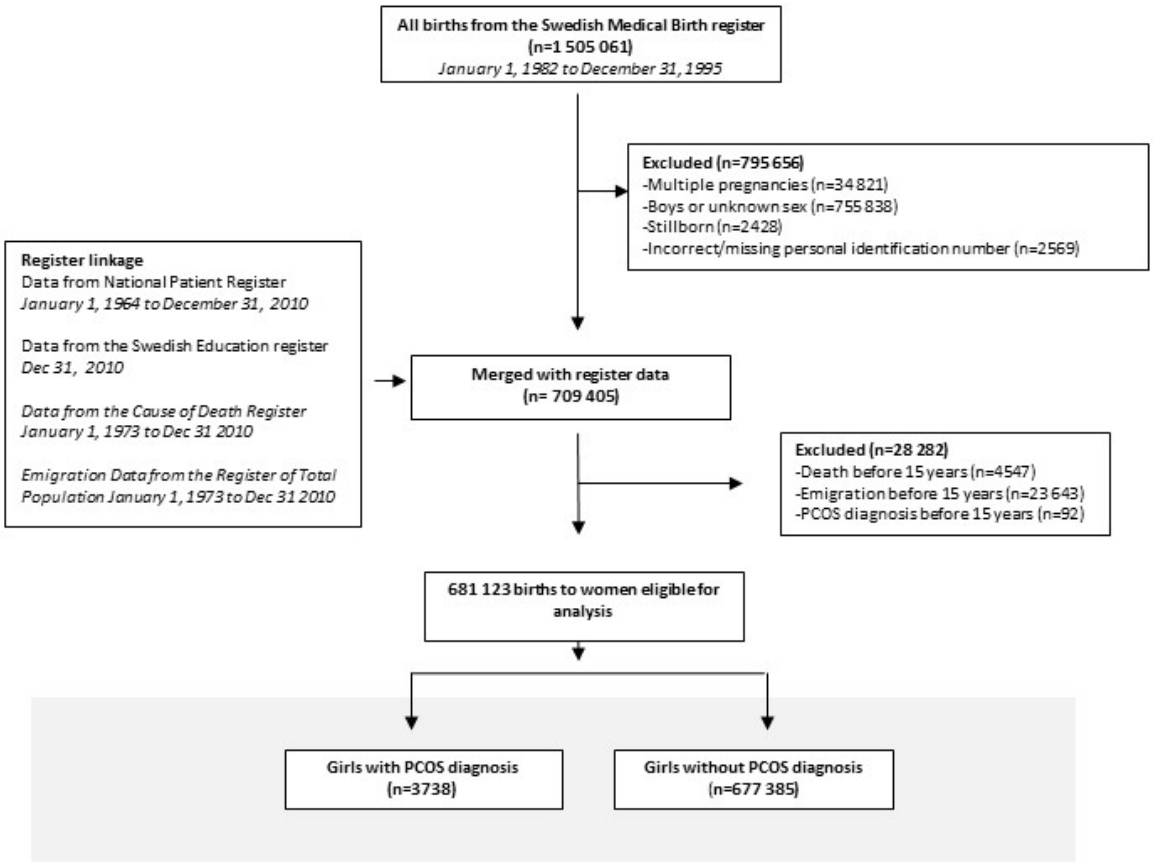

Table 1. Maternal characteristics and later diagnosis of PCOS in the offspring, among singleton girls born between 1982 until 1995 in Sweden

	PCOS diagnosis in the offspring girls			
	Yes		No	
Characteristic	No. of births (N=3738)	Rate (%)	No. of births (N=677 385)	Rate (%)
Maternal Body-mass index in early pregnancy[a]				
≤ 18.4	151	6.4	25 473	6.1
18.5-24.9	1637	69.4	310 280	74.0
25.0-29.9	441	18.7	66 290	15.8
≥ 30.0	130	5.5	17 304	4.1
Data missing	1379		258 038	
Maternal daily smoking in early pregnancy				
No	2125	65.2	461 338	74.6
1-9 cigarettes/day	653	20.0	97 147	15.7
≥ 10 cigarettes/day	482	14.8	59 753	9.7
Data missing	478		59 147	
Maternal age at birth (years)				
13-19	153	4.1	19 608	2.9
20-24	1008	27.0	154 288	22.8
25-29	1320	35.3	252 556	37.3
30-34	838	22.4	171 715	25.4
≥ 35	419	11.2	79 212	11.7
Data missing	0		6	
Parity at index pregnancy				
0	1602	42.0	278 691	41.1
≥ 1	2136	57.1	398 694	58.9
Involuntary childlessness before index pregnancy (years)				
0- <1	3469	92.8	639 099	94.4
1-2	153	4.1	22 276	3.3
> 2	116	3.1	16 010	2.4
Maternal education (years)				

≤ 11	2544	70.0	417 802	63.1
≥ 12	1092	30.0	244 644	36.9
Data missing	102		14 939	
Maternal country of birth				
Nordic	3270	88.4	623 391	92.6
Non-Nordic	430	11.6	49 904	7.4
Data missing	38		4 090	
Maternal diabetes during index pregnancy				
No	3702	99.0	670 575	99.0
Pre-gestational	22	0.6	3769	0.6
Gestational	14	0.4	3041	0.5
Maternal preeclampsia during index pregnancy				
No	3636	97.3	661 388	97.6
Yes	102	2.7	3636	2.4
Maternal PCOS diagnosis[c]				
No	3693	98.8	676 005	99.8
Yes	45	1.2	1,380	0.2

[a]BMI was available between 1982 and 1989 and 1992-1995

[c]Gestational diabetes was available in 1989 and later

[c]During the years 1964 to 2010

Table 2. Characteristics at birth and later diagnosis of PCOS, among singleton girls born between 1982 until 1995 in Sweden

	PCOS diagnosis in the offspring girl			
	Yes		No	
Characteristic	No. of births (N=3,738)	Rate (%)	No. of births (N=677,385)	Rate (%)
Birthweight for gestational age[a]				
Small (SGA)	401	10.8	56 166	8.3
Average (AGA)	2914	78.7	543 609	80.8
Large (LGA)	389	10.5	73 418	10.9
Data missing	34		4192	
Ponderal Index[b]				
Low (“thin”)	348	9.5	54 780	8.2
Average	2943	79.9	537 922	80.4
High (“plump”)	392	10.6	76 689	11.5
Data missing	55		7994	
Head circumference for gestational age[c]				
Small	438	12.0	63 576	9.7
Average	2883	79.0	532 633	81.0
Large	326	8.9	61 405	9.3
Data missing	91		19 771	
Birthweight (g)				
< 2500	144	3.9	24 065	3.6
2500-4499	3482	93.8	635 721	94.2
≥ 4500	88	2.4	15 113	2.2
Data missing	24		2486	
Gestational age at birth (weeks)				

Very preterm (< 32)	17	0.5	3686	0.6
Moderately preterm (32-36)	170	4.6	29 013	4.3
Term or postterm (≥ 37)	3540	95.0	642 883	95.2
Data missing	11		1803	

[a]Small and large birthweight for gestational age were defined as a birthweight of more than 10% below or above the mean weight for gestational age, respectively, according to the Swedish sex-specific fetal growth curve (Marsal)

[b]Low and high ponderal index were defined as a ponderal index score among the approximately 10% lower or higher ponderal index in the study population

[c]Small and large head circumference for gestational age were defined as more than 10% below the mean weight for gestational age, respectively, according to the Swedish reference curve for new born infants (Niklasson)

Table 3. Risks of PCOS diagnosis by maternal characteristics, among singleton girls born between 1982 until 1995 in Sweden

	PCOS diagnosis in offspring girl		
	Rate (%)	Hazard Ratio (95% CI)	
		Crude	Adjusted[a]
Maternal Body-mass index in early pregnancy[b]			
≤ 18.4	0.6	0.93 (0.78-1.10)	0.89 (0.74-1.06)
18.5-24.9	0.5		Reference
25.0-29.9	0.7	1.60 (1.44-1.78)	1.52 (1.36-1.70)
≥ 30.0	0.8	2.31 (1.93-2.76)	1.97 (1.61-2.41)
Maternal daily smoking in early pregnancy			
No	0.5		Reference
1-9 cigarettes/day	0.7	1.27 (1.16-1.38)	1.31 (1.18-1.47)
≥ 10 cigarettes/day	0.8	1.48 (1.34-1.63)	1.44 (1.27-1.64)
Maternal age at birth (years)			
13-19	0.8	1.33 (1.12-1.57)	1.01 (0.79-1.29)
20-24	0.6	1.17 (1.08-1.27)	1.04 (0.93-1.16)
25-29	0.5		Reference
30-34	0.5	0.95 (0.87-1.04)	0.90 (0.80-1.01)
≥ 35	0.5	1.05 (0.94-1.17)	0.99 (0.85-1.16)
Parity at index pregnancy			
0	0.6		Reference
≥ 1	0.5	1.07 (1.00-1.14)	1.03 (0.94-1.14)
Involuntary childlessness before index pregnancy (years)			
0- <1			Reference
1-2		1.12 (0.95-1.31)	1.05 (0.86-1.29)

> 2		1.23 (1.02-1.48)	1.14 (0.90-1.45)
Maternal education (years)			
≤ 12	0.4	1.31 (1.22-1.40)	1.10 (1.00-1.22)
> 12	0.6		Reference
Maternal country of birth			
Nordic	0.5		Reference
Non-Nordic	0.8	2.07 (1.87-2.29)	2.00 (1.73-2.32)
Maternal diabetes during index pregnancy			
No	0.6		Reference
Pre-gestational	0.6	0.99 (0.64-1.47)	0.61 (0.29-1.29)
Gestational	0.5	1.64 (0.97-2.77)	0.87 (0.32-2.33)
Maternal preeclampsia during index pregnancy			
No	0.6		Reference
Yes	0.6	1.17 (0.96-1.43)	1.05 (0.80-1.38)
Maternal PCOS diagnosis[d]			
No	0.2		Reference
Yes	1.2	8.02 5.97-10.76	6.59 (4.40-9.89)

[a]Adjusted for all the other variables in the Table (maternal age, parity, years of involuntary childlessness before index pregnancy, presence of diabetes or preeclampsia at index pregnancy, as well as for maternal education level, country of birth and PCOS diagnosis) and for infant birthweight for gestational age.

[b]Information on maternal BMI was available between 1982-1989 and 1992-1995.

[c]Gestational diabetes was available in 1989 and later

[d]During the years 1964 to 2010

Table 4. Risks of PCOS diagnosis by size at birth, among singleton girls born between 1982 until 1995 in Sweden

	PCOS diagnosis in offspring girl		
	Rate (%)	Hazard Ratio (95% CI)	
		Crude	Adjusted[a]
Birthweight for gestational age[b]			
Small (SGA)	0.7	1.23 (1.11-1.37)	1.09 (0.94-1.26)
Average (AGA)	0.5		Reference
Large (LGA)	0.5	1.04 (0.93-1.15)	1.01 (0.88-1.17)
Ponderal Index[c]			
Low (“thin”)	0.6	1.11 (0.99-1.25)	1.12 (0.96-1.30)
Average	0.5		Reference
High (“plump”)	0.5	1.02 (0.92-1.14)	1.06 (0.92-1.21)
Head circumference for gestational age[d]			
Small	0.7	1.16 (1.05-1.28)	1.04 (0.90-1.19)
Average	0.5		Reference
Large	0.5	0.96 (0.85-1.07)	1.12 (0.97-1.29)
Birthweight (g)			
< 2500	0.6	1.06 (0.90-1.25)	0.94 (0.73-1.21)
2500-4499	0.5		Reference
≥ 4500	0.6	1.17 (0.94-1.44)	1.26 (0.96-1.66)

[a]Adjusted for all the maternal characteristics (maternal age, parity, years of involuntary childlessness before index pregnancy, presence of diabetes or preeclampsia at index pregnancy, as well as for maternal education level, country of birth and PCOS diagnosis).

[b]Small and large birthweight for gestational age were defined as a birthweight of more than 10% below or above the mean weight for gestational age, respectively, according to the Swedish sex-specific fetal growth curve (Marsal)

[c]Low and high ponderal index were defined as a ponderal index score among the approximately 10% lower or higher ponderal index in the study population

[d]Small and large head circumference for gestational age were defined as more than 10% below the mean weight for gestational age, respectively, according to the Swedish reference curve for new born infants (Niklasson)

Table S1. Association between maternal smoking during pregnancy and birthweight for gestational age and later diagnosis of PCOS in the offspring, stratified by those with and without maternal BMI data from index pregnancy

	PCOS diagnosis in offspring girl					
	Without maternal BMI data			With maternal BMI data		
Characteristic	No. of Births (N=3738)	Rate (%)	Adjusted[a] Hazard Ratio (95% CI)	No. of Births (N=677 385)	Rate (%)	Adjusted Hazard Ratio (95% CI)
Daily smoking in early pregnancy						
No	723	0.4	Reference	1402	0.5	Reference
1-9 cigarettes/day	194	0.6	1.09 (0.92-1.29)	459	0.7	1.31 (1.17-1.46)
≥ 10 cigarettes/day	164	0.7	1.47 (1.22-1.76)	318	0.8	1.46 (1.28-1.66)
Birthweight for gestational age[b]						
Small (SGA)	165	0.7	1.06 (0.86-1.31)	236	0.7	1.06 (0.91-1.22)
Average (AGA)	1057	0.5	Reference	1857	0.6	Reference
Large (LGA)	144	0.5	1.14 (0.94-1.39)	245	0.5	1.07 (0.93-1.24)

[a]Adjusted for the other variable in the Table, and maternal age, parity, years of involuntary childlessness before index pregnancy, presence of diabetes or preeclampsia at index pregnancy, as well as for maternal education level, country of birth and PCOS diagnosis

[b]Small and large birthweight for gestational age were defined as a birthweight of more than 10% below or above the mean weight for gestational age, respectively, according to the Swedish sex-specific fetal growth curve (Marsal)