Interaction between Maternal Passive Smoking during Pregnancy and CYP1A1 and GSTs Polymorphisms on Spontaneous Preterm Delivery

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Abstract

Objective: The present study aimed to examine the association between maternal passive smoking during pregnancy and the risk of spontaneous PTD and to explore the potential interaction of the single or joint gene polymorphism of CYP1A1 and GSTs with maternal passive smoking on the risk of spontaneous PTD.

Method: We investigated whether the association between maternal passive smoking and PTD can be modified by 2 metabolic genes, i.e. cytochrome P4501A1 (CYP1A1) and glutathione S-transferases (GSTs), in a case-control study with 198 spontaneous preterm and 524 term deliveries in Shenzhen and Foshan, China. We used logistic regression to test gene-passive smoking interaction, adjusting for maternal socio-demographics and prepregnancy body mass index.

Results: Overall, maternal passive smoking during pregnancy was associated with higher risk of PTD (adjusted odds ratio = 2.20 [95% confidence interval: 1.56–3.12]). This association was modified by CYP1A1 and GSTs together, but not by any single genotype. For cross-categories of CYP1A1 Msp I and GSTs, maternal passive smoking was associated with higher risk of PTD among those women with CYP1A1 “TC/CC" + GSTs “null”, but not among women with other genotypes; and this interaction was significant (OR = 2.66 [95% Cl: 1.19–5.97]; P-value: 0.017). For cross-categories of CYP1A1 BsrD I and GSTs, maternal passive smoking was associated with higher risk of PTD only among those women with CYP1A1 “AG/GG” + GSTs “null”, but not among women with other genotypes; and this interaction was significant (OR = 3.00 [95% Cl: 1.17–7.74]; P-value: 0.023).

Conclusions: Our findings suggest that the combined genotypes of CYP1A1 and GSTs can help to identify vulnerable pregnant women who are subject to high risk of spontaneous PTD due to passive smoking.

Introduction

Preterm delivery (PTD, <37 completed weeks of gestation) is a big clinical and public health challenge globally. Despite increased awareness and improved prenatal care, the percentage of PTD remains unacceptably high in both developed (e.g. 7% in U.S. [1]) and developing societies (e.g. 5–15% in China [2]). About 15% preterm babies die within one month after birth [3]. PTD also leads to many other short- and long-term health problems and poses enormous burden to both health care system and the child’s family [4]. Preterm delivery is often classified into spontaneous and medically indicated subgroups, and the majorities (75–83%) of PTDs are spontaneous [5]. The causes and underlying biological mechanisms of spontaneous PTD are still unclear [6], although previous studies have identified a long list of risk factors, including low socio-economic status [7], parity, maternal age [8], drug abuse, life events [9], racial origin [10], maternal active or passive smoking [11], air pollution [12,13] intraterine infection [14], and genetic heterogeneity [9,15].

Maternal active smoking is a well-established risk factor for PTD, and it accounts for about 14% of all PTDs [16]. But only a relatively small proportion of smoking pregnant women end with PTD, which may be explained by the substantial variability in genetic susceptibility across individuals. Some evidences show that gene polymorphism in cytochrome P4501A1 (CYP1A1) and glutathione S-transferases (GSTs) may modify the link between maternal active smoking and PTD [17]. CYP1A1 is one of cytochrome P450 (CYP450) family genes that are responsible for phase I detoxication by converting exogenous exposures, e.g. tobacco compounds, into intermediate metabolites. CYP1A1 mutation can lead to higher enzyme activity. GSTs are one of
phase II detoxication enzymes that protect cells from toxicants by conjugation with glutathione. Inherited homozygous deletion of GSTs genes can result in lack of phase II detoxification activity [18,19,20], which thus increases the accumulation of intermediate metabolites of exogenous in human body. Limited researches focused on the gene (CYP1A1, GSTs)-maternal active smoking on the risk PTD yielded inconsistent findings [17,21,22].

Passive smoking is one of the most important public health problems in many developing countries, such as China, where many non-smoking pregnant women are exposed to environmental tobacco smoke at home, workplace, and public places [23]. Perera et al. found the levels of serum cotinine and DNA adducts in passive smoking mothers were significantly higher than those in non-passive smoking mothers [24]. Some previous studies, all done in developed countries, suggest that maternal passive smoking may shorten gestational age and thus increase the risk of PTD [23]. It is unclear whether this association holds in pregnant women in developing countries. Moreover, little is known about the potential interaction between maternal passive smoking and genes (CYP1A1, GSTs) polymorphisms in the risk of PTD.

Therefore, the present study aimed to fill these important research gaps in this filed. Specially, we had 2 objectives in this study: 1) to examine the association between maternal passive smoking during pregnancy and the risk of spontaneous PTD among Chinese pregnant women; 2) to explore the potential interaction between the single or joint gene polymorphism of CYP1A1 and GSTs with maternal passive smoking on the risk of spontaneous PTD.

Materials and Methods

Study Population

We conducted a case-control study from September 2009 to March 2011 at two Women and Children’s Hospitals at Shenzhen and Foshan, Guangdong Province, China. Figure 1 shows the flow chart of our study participants flowchart was listed in figure 1. A total of 379 preterm (gestational age <37 full weeks) babies were born in these two hospitals during the study period. Among them, 223 spontaneous singleton babies were eligible for this study (see below). Finally, 198 (88.8%) mothers of 223 eligible babies agreed to participate in the study and were included as cases. We randomly selected controls from those mothers who delivered term singleton babies (gestational age 37–42 full weeks) with normal birth weight (2500–4000 g) in the same hospital. To increase the comparability, we matched 563 eligible controls with eligible cases by delivery date (+/-3 days). Among them, 524 (93.1%) mothers of the controls agreed to participate in this study. We approached participating mothers within 12–36 hours after delivery.

For the purpose of this study, we excluded the preterm babies whose mothers: 1) actively smoked cigarettes during pregnancy (n = 2); 2) had one or more pre-existing chronic conditions, including heart failure, chronic renal diseases, lung diseases, diabetes, hypertension, hyperthyroidism, Mediterranean anemia etc. (n = 24); 3) had induced or accidental abortion (n = 23); 4) had multiple births or newborns with birth defects (n = 32); or 5) had medically indicated PTD due to obstetric complications, such as severe preeclampsia, antepartum hemorrhage, placenta previa etc (n = 75).

All the participants understood and signed a written consent form. This study was approved by the Ethics Committees of Sun Yat-sen University in Guangzhou, China.

Data Collection

We collected data through interview, medical records review, and blood lab tests. At the postnatal face-to-face interview, each mother completed a structural questionnaire and reported her passive smoking status during pregnancy, socio-demographics, reproductive history, medical history, psychosocial stress, health behavior and lifestyles. Pregnant women were expected to complete up to 12 routine prenatal care visits for routine obstetric examinations starting from the 20th week of gestation: 2 visits during 20–27 weeks of gestation, bi-weekly during 28–35 weeks of gestation, and then weekly after 36 weeks of gestation. We

![Figure 1. Flow chart of study participants.](doi:10.1371/journal.pone.0049155.g001)
obtained maternal and fetal health information from obstetrical medical records, including last menstrual period, ultrasound assessment, maternal chronic diseases, and obstetric complications. We collected maternal blood samples within 12 hours after admission to the hospital in tubes with anticoagulants (EDTA K2), and then stored blood samples in a refrigerator at temperature of −80°C.

**Measures**

**PTD outcome.** Pregnant women self-reported their last menstrual period (LMP) at the 1st prenatal care visit (usually at 0–10th week of gestation). The LMP was confirmed by early ultrasound assessment (gestational age<20 full weeks). If self-reported LMP was unavailable, ultrasound estimated LMP based on the crown-rump length in early pregnancy was used instead [25]. We calculated gestational age as the interval between LMP and delivery date. We defined PTD as gestational age less than 37 complete weeks.

**Passive smoking exposures.** In this study, we measured maternal passive smoking during pregnancy by combining self-report and serum cotinine test. At the interview, participants retrospectively self-reported their passive smoking status during pregnancy.

We also measured the cotinine level in maternal peripheral blood using enzyme immunoassay technique (Immunalysis Corp., Pomona, California, US; Manufacturer-reported detection limit, 1 ng/ml). Briefly, we first added 10 μl of serum, calibrator, or control to 3 separate assay wells, and mixed them with 100 μl of cotinine enzyme. Then we incubated these samples for 30 minutes at room temperature, and washed the micro-plate 6 times with 350 ul buffer. Then we added 100 μl of substrate solution and measured the absorbance spectrum of samples at 450 nm using a micro-plate reader within 60 minutes.

We defined passive smoking as: 1) self-reported exposure to cigarette smoke by others (at home, work or public places) during pregnancy (n = 199), or 2) serum cotinine level >3 ng/ml (n = 230). This combined use of self-report and serum cotinine could reduce misclassification of passive smoking [26,27]. Overall, there was relatively high concordance between self-reported passive smoking when serum cotinine cut-off level was set as 3 ng/ml in our sample (Kappa-value: 0.752). However, 53 women (7.3%) who did not report passive smoking but had serum cotinine level ≥3 ng/ml and 22 (3.0%) women who reported passive smoking but had serum cotinine level <3 ng/ml (Table S1).

**Genotypes.** We purified DNA from venous whole blood samples using DNS purification kits (Takara, Biot. Ltd, China).We used polymerase chain reaction (PCR) and restriction fragment

| Table 1. Comparison of characteristics between spontaneous PTD cases and controls. |
|-----------------------------------------------|---------------------------|--------------------------|
| Cases (n = 198)                                | Controls(n = 524)         | P-value                  |
| Mean ± SD or N. (%)                            | Mean ± SD or N. (%)       |                          |
| Maternal age (years, mean ± SD)                | 28.5±4.9                 | 28.8±4.3                 | 0.474                     |
| Marital status (%)                             |                           |                          |
| Married                                        | 193(97.5)                | 502(95.8)                | 0.290                     |
| Unmarried                                      | 5(2.5)                   | 22(4.2)                  |                          |
| Race/ethnicity (%)                             |                           |                          |
| Han                                            | 191(97.0)                | 507(96.9)                | 0.992                     |
| Minority                                       | 6(3.0)                   | 16(3.1)                  |                          |
| Education level (%)                            |                           |                          |
| Junior high school or lower                    | 70(35.4)                 | 136(26.0)                | 0.020                     |
| High school                                    | 55(27.8)                 | 142(27.1)                |                          |
| College or higher                              | 73(36.9)                 | 246(46.9)                |                          |
| Family income (%)                              |                           |                          |
| Low                                            | 44(23.0)                 | 141(27.2)                | 0.248                     |
| Middle                                         | 32(16.8)                 | 102(19.7)                |                          |
| High                                           | 115(60.2)                | 276(53.2)                |                          |
| Parity (%)                                     |                           |                          |
| Nullparous                                     | 134(67.7)                | 398(76.0)                | 0.024                     |
| Parous                                         | 64(32.3)                 | 126(24.0)                |                          |
| Alcohol use during pregnancy, %                | 4(2.0)                   | 17(3.2)                  | 0.383                     |
| Prepregnancy BMI (kg/m²) (%)                   |                           |                          |
| Underweight (<18.5)                            | 45(22.7)                 | 124(23.7)                | 0.606                     |
| Normal (18.5–23.9)                             | 137(69.2)                | 346(66.0)                |                          |
| Overweight or obesity(>24.0)                   | 16(8.1)                  | 54(10.3)                 |                          |
| Child gender (%)                               |                           |                          |
| Male                                           | 109(35.6)                | 257(51.3)                | 0.105                     |
| Female                                         | 89(64.4)                 | 271(48.7)                |                          |

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length polymorphism (RFLP) methods to genotype CYP1A1 polymorphisms. We digested PCR products by Msp I (to identify CYP1A1 m1 mutation) and BsrD I (to identify CYP1A1 m2 mutation) and then detected the genotypes of our interest, including the homozygous wild type "TT" "AA", heterozygous variant "TC" "AG", and homozygous variant "CC" "GG". The PCR primers (Sangon biotech, shanghai, co., Ltd.) for CYP1A1 polymorphisms included Msp I forward 5′-CAG TGA AGA GGT GTA GCC GCT-3′ and reverse 5′-TAG GAG TCT TGT CTC ATG CCT-3′; BsrD I forward 5′-CTG TCT CCC TCT GGT TAC AGG AAG C-3′ and reverse 5′-TTC CAC CCG TTG CAG CAG CAT AGC C-3′. The PCR primers for GSTs polymorphisms included GSTM1 forward 5′-GAA CTC CCT GAA AAG CTA AG-3′ and reverse 5′-GTG GAG GGA GCC AGAC ACC TAT ATG GTG G-3′; and GSTT1 forward 5′-TTC CTT CCT GGT GCT CAC ATC TC-3′ and reverse 5′-TCA CCG GAT CAT GGC CAG CA-3′. As the internal control, a 268-bp fragment of the human β-globin gene was coamplified with a second set of primers (5′-CAA CTT CAT CCA GGT TCA CC-3′) and (5′-GAA GAG CCA GAG AAG AAT GGT AGC-3′). Due to small numbers of participants with homozygous variant genotypes, we combined TC (n = 310) and CC (n = 127) as TC/CC, AG (n = 266) and GG (n = 71) as AG/GG. For the same reason, we combined GSTM1 and GSTT1 genotypes as a binary variable GSTs that was coded as “null” if both GSTM1 or GSTT1 were null and as “present” otherwise.

**Table 2.** Associations of maternal passive smoking during pregnancy, CYP1A1, and GSTs genotypes with risks of spontaneous PTD.

|                          | Case (n = 198) | Control (n = 524) | Crude OR (95% CI) | OR* (95%CI) | P-value |
|--------------------------|---------------|------------------|------------------|-------------|---------|
| Passive smoking during pregnancy |               |                   |                  |             |         |
| No                       | 101(51.0)     | 369(70.4)        | 1                | 1           |         |
| Yes                      | 97(49.0)      | 155(29.6)        | 2.29 (1.63–3.20) | 2.20 (1.56–3.12) | 0.000 |
| CYP1A1 Msp I             |               |                   |                  |             |         |
| TT                       | 73(36.9)      | 212(40.5)        | 1                | 1           |         |
| TC/CC                    | 125(63.1)     | 312(59.5)        | 1.16 (0.83–1.63) | 1.15 (0.82–1.63) | 0.418 |
| CYP1A1 BsrD I            |               |                   |                  |             |         |
| AA                       | 95(48.0)      | 290(55.3)        | 1                | 1           |         |
| AG/GG                    | 103(52.0)     | 234(44.7)        | 1.34 (0.97–1.87) | 1.30 (0.93–1.81) | 0.133 |
| GSTM1                    |               |                   |                  |             |         |
| Present                  | 97(49.0)      | 275(52.5)        | 1                | 1           |         |
| Null                     | 101(51.0)     | 249(47.5)        | 1.15 (0.83–1.60) | 1.12 (0.80–1.57) | 0.515 |
| GSTT1                    |               |                   |                  |             |         |
| Present                  | 95(48.0)      | 241(46.0)        | 1                | 1           |         |
| Null                     | 103(52.0)     | 283(54.0)        | 0.92 (0.67–1.28) | 0.97 (0.69–1.37) | 0.877 |
| GSTs                     |               |                   |                  |             |         |
| Present                  | 137(69.2)     | 386(73.7)        | 1                | 1           |         |
| Null                     | 61(30.8)      | 138(26.3)        | 1.25 (0.87–1.78) | 1.29 (0.89–1.87) | 0.182 |

*OR* adjusted for family income, maternal age, education level and prepregnancy BMI.

**Statistical Analyses**

We first conducted Chi-square/t-test to examine the overall characteristic balance between PTD case and control groups. Chi-square test was adopted to test the associations between maternal passive smoking during pregnancy, single gene polymorphism with the risk of PTD, we then fitted multivariable logistic regression models to estimate the odds ratios (OR) and their 95% confidence intervals (CI), adjusting for potential confounders.

To examine whether the association between maternal passive smoking and PTD could be modified by maternal CYP1A1 and GSTs polymorphisms, we tested their interactions on multiplicative scale. Specifically, we added the interaction terms "passive smoking × gene polymorphism" (products) to the multivariable logistic regression models which already included the main effect terms for passive smoking and CYP1A1 and GSTs genotypes, as well as the potential confounders. A significant departure of the OR value for an interaction term from 1 indicated the existence of interaction on a multiplicative scale. To better demonstrate joint gene-passive smoking interaction, we also classified participants into 8 exclusive groups by passive smoking status (yes vs. no), CYP1A1 ("wild" vs. "variant"), and GSTs ("present" vs. "null"). We set non-passive smoking mothers with low risk genotypes (CYP1A1 "TT" or “AA”+GSTs “present”) as the reference group
and then compared it with the other 7 groups. All analyses were completed in SPSS 16.0 software (SPSS Inc, Chicago, Illinois, USA). The statistical significance level was set to 0.05 (two-sided).

## Results

Table 1 shows the characteristics of our study sample. Overall, the PTD case and control groups were comparable in terms of maternal age, marital status, socio-economic status, alcohol use during pregnancy, prepregnancy BMI, and the child gender. However, mothers in the case group had lower education level (36.9% vs. 46.9% with college or higher degree), and were also more likely to be parous (32.3% vs. 24.0%), when compared with mothers in the control group.

Table 2 shows the overall associations (main effect) of maternal passive smoking during pregnancy and single gene polymorphism with risk of PTD. The proportion of passive smoking mothers in the PTD group (49.0%) was much higher than those in the control group (29.6%), and the adjusted OR was 2.20 (95% CI: 1.56–3.12). However, the distribution of CYP1A1 Msp I, CYP1A1 BsrD I, GSTM and GSTT1 genotypes were similar between the two groups.

Table 3 shows the interactions between maternal passive smoking during pregnancy and single gene loci polymorphism on risk of PTD, after adjusting for potential confounders. Overall, there was no significant interaction between maternal smoking during pregnancy with any of the 4 selected gene loci (CYP1A1 Msp I, CYP1A1 BsrD I, GSTM1 and GSTT1) on multiplicative scale.

### Table 3. Interaction between maternal passive smoking during pregnancy and single genotype of CYP1A1 and GSTs on risk of spontaneous PTD.

| Passive smoking | Genotype | Case(n = 198) | Control(n = 524) | OR*(95%CI) | P-value |
|-----------------|----------|---------------|-----------------|------------|---------|
|                 |          | N. (%) | N. (%) |             |         |
| CYP1A1 Msp I    |          | TT      | 41 (20.7) | 151 (28.8) | 1       |
|                 |          | TC/CC   | 60 (30.3) | 218 (41.6) | 0.96 (0.61–1.52) | 0.876 |
|                 | Yes      | TT      | 32 (16.2) | 61 (11.6)  | 1.75 (1.00–3.07)* | 0.052 |
|                 | Yes      | TC/CC   | 65 (32.8) | 94 (17.9)  | 2.42 (1.50–3.89)* | 0.000 |
|                 | Interaction |       |          |           | 1.44 (0.71–2.92) | 0.319 |
| CYP1A1 BsrD I   |          | AA      | 50 (25.3) | 203 (38.7) | 1       |
|                 |          | AG/GG   | 51 (23.5) | 166 (31.7) | 1.18 (0.75–1.85) | 0.483 |
|                 | Yes      | AA      | 45 (22.7) | 87 (16.6)  | 1.98 (1.22–3.22)* | 0.007 |
|                 | Yes      | AG/GG   | 52 (26.3) | 68 (13.0)  | 2.90 (1.78–4.72)* | 0.000 |
|                 | Interaction |       |          |           | 1.24 (0.62–2.47) | 0.541 |
| GSTM           | No       | Present | 49 (24.7) | 195 (37.2) | 1       |
|                 | No       | Null    | 52 (26.2) | 174 (35.7) | 1.15 (0.73–1.81) | 0.519 |
|                 | Yes      | Present | 48 (24.2) | 80 (15.3)  | 2.30 (1.41–3.74) | 0.001 |
|                 | Yes      | Null    | 49 (24.7) | 75 (29.5)  | 2.43 (1.49–3.97) | 0.000 |
|                 | Interaction |       |          |           | 0.92 (0.46–1.83) | 0.806 |
| GSTT1          | No       | Present | 49 (24.7) | 163 (31.1) | 1       |
|                 | No       | Null    | 52 (26.2) | 206 (39.3) | 0.84 (0.53–1.33) | 0.454 |
|                 | Yes      | Present | 46 (23.2) | 78 (14.9)  | 1.61 (1.10–2.98) | 0.020 |
|                 | Yes      | Null    | 51 (25.8) | 77 (14.7)  | 2.22 (1.37–3.62) | 0.002 |
|                 | Interaction |       |          |           | 1.46 (0.73–2.91) | 0.286 |
| GSTs           | No       | Present | 73 (36.9) | 267 (51.0) | 1       |
|                 | No       | Null    | 28 (14.1) | 102 (19.5) | 1.00 (0.60–1.66) | 0.999 |
|                 | Yes      | Present | 64 (32.3) | 119 (22.7) | 1.86 (1.24–2.81) | 0.003 |
|                 | Yes      | Null    | 33 (16.7) | 36 (6.9)   | 3.39 (1.95–5.91) | 0.000 |
|                 | Interaction |       |          |           | 1.83 (0.84–3.96) | 0.126 |

*OR* adjusted for family income, maternal age, education level and prepregnancy BMI. GSTs “null” if both GSTM1 and GSTT1 “null”, GSTs “present” if either GSTM1 or GSTT1 “present”. % for distribution within the case and control groups respectively. doi:10.1371/journal.pone.0049155.t003
Table 4. Interaction between maternal passive smoking during pregnancy and joint genotype of CYP1A1 Msp I and GSTs on risk of spontaneous PTD.

| Passive smoking | Genotype | Case (n = 198) | Control (n = 524) | OR*(95%CI) | P-value |
|-----------------|----------|---------------|------------------|------------|---------|
|                 | CYP1A1 Msp I | GSTs | N. (%) | N. (%) |          |
| No              | TT       | Present | 30 (15.2%) | 110 (21.0%) | 1       |
| No              | TT       | Null    | 11 (5.6%)  | 41 (7.8%)  | 1.04 (0.47–2.29) | 0.918 |
| No              | TC/CC    | Present | 43 (21.7%) | 157 (30.0%) | 0.98 (0.57–1.67) | 0.947 |
| No              | TC/CC    | Null    | 17 (8.6%)  | 61 (11.6%) | 0.95 (0.47–1.93) | 0.902 |
| Yes             | TT       | Present | 25 (12.6%) | 45 (8.6%)  | 1.82 (0.95–3.48) | 0.072 |
| Yes             | TT       | Null    | 7 (3.5%)   | 16 (3.1%)  | 1.64 (0.61–4.41) | 0.326 |
| Yes             | TC/CC    | Present | 39 (19.7%) | 74 (14.1%) | 1.85 (1.04–3.26) | 0.035 |
| Yes             | TC/CC    | Null    | 26 (13.1%) | 20 (3.8%)  | 4.72 (2.28–9.77) | 0.000 |
| Interaction     |          |          | 2.66 (1.19–5.97) |          | 0.017 |

*a.OR* adjusted for family income, maternal age, education level, prepregnancy BMI and CYP1A1 BsrD I genotype.
*b.GSTs “null” if both GSTM1 and GSTT1 “null”, GSTs “present” if either GSTM1 or GSTT1 “present”. |
*c.% for distribution within the case and control groups respectively.
*d.The variable of passive smoking+CYP1A1 Msp I+ GSTs with eight levels was set as dummy independent variable, and the first level as reference category.

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Table 5. Interaction between maternal passive smoking during pregnancy and joint genotype of CYP1A1 BsrD I and GSTs on risk of spontaneous PTD.

| Passive smoking | Genotype | Case (n = 198) | Control (n = 524) | OR*(95%CI) | P-value |
|-----------------|----------|---------------|------------------|------------|---------|
|                 | CYP1A1 BsrD I | GSTs | N. (%) | N. (%) |          |
| No              | AA       | Present | 38 (19.2%) | 149 (28.4%) | 1       |
| No              | AA       | Null    | 12 (6.1%)  | 54 (10.3%) | 0.94 (0.45–1.95) | 0.863 |
| No              | AG/GG    | Present | 35 (17.7%) | 118 (22.5%) | 1.14 (0.67–1.95) | 0.636 |
| No              | AG/GG    | Null    | 16 (8.1%)  | 48 (9.2%)  | 1.19 (0.59–2.42) | 0.613 |
| Yes             | AA       | Present | 30 (15.2%) | 61 (11.6%) | 1.80 (1.01–3.22) | 0.050 |
| Yes             | AA       | Null    | 15 (7.6%)  | 26 (5.0%)  | 2.33 (1.11–4.89) | 0.030 |
| Yes             | AG/GG    | Present | 34 (17.2%) | 58 (11.1%) | 2.17 (1.23–3.83) | 0.008 |
| Yes             | AG/GG    | Null    | 18 (9.1%)  | 10 (1.9%)  | 7.01 (2.91–16.86) | 0.000 |
| Interaction     |          |          | 3.00 (1.17–7.74) |          | 0.023 |

*a.OR* adjusted for family income, maternal age, education level, prepregnancy BMI and CYP1A1 Msp I genotype.
*b.GSTs “null” if both GSTM1 and GSTT1 “null”, GSTs “present” if either GSTM1 or GSTT1 “present”. |
*c.% for distribution within the case and control groups respectively.
*d.The variable of passive smoking+CYP1A1 BsrD I+ GSTs with eight levels was set as dummy independent variable, and the first level as reference category.

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PTD accompanied by histologic chorioamnionitis. Genotypes (CYP1A1''AG/GG'' or 'TC/CC') seemed to amplify the risk of spontaneous PTD associated with passive smoking during pregnancy. This novel finding has important clinical and public health implications. It not only contributes to better understanding the pathogenic pathways through which maternal passive smoking increases risk of spontaneous PTD, but also helps to identify vulnerable pregnant women who are subject to high risk of spontaneous PTD due to maternal passive smoking.

Conclusion

In summary, we found maternal passive smoking during pregnancy was associated with higher risk of spontaneous PTD. The genotypes of CYP1A1 “AG/GG” or “TC/CC” + GSTs “null” seemed to amplify the risk of spontaneous PTD associated with passive smoking during pregnancy. This novel finding has important clinical and public health implications. It not only contributes to better understanding the pathogenic pathways through which maternal passive smoking increases risk of spontaneous PTD, but also helps to identify vulnerable pregnant women who are subject to high risk of spontaneous PTD due to maternal passive smoking.

Supporting Information

Table S1 Concordance of maternal passive smoking measured by self-report and by maternal serum cotinine level.

(DOC)

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Author Contributions

Analyzed the data: YJL. Contributed reagents/materials/analysis tools: YJL. Wrote the paper: YJL. Generated hypothesis and designed the experiments: WQC YJL XZW TL. Conducted the survey and experiments: YJL PD YHH CBX JmL SXY XLG DQJ LHC BZH. Edited manuscript: XZW WQC.

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