Gene-environment interaction among *GSTT1*, *PON2* polymorphisms and organic solvents on gestational age in a Chinese women cohort

Shuai Li · Kai Fang · Wenjian Wang · Yonghua Hu · Dafang Chen

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Abstract

Purpose To investigate interactions on gestational age among two environmental risk factors and four maternal genetic polymorphisms: organic solvents, passive smoking, *CYP1A1* *rs4646903* (*MspI*), *EPHX1* *rs2234922* (*His139Arg*), *GSTT1* and *PON2* *rs12026* (*Ala148Gly*).

Methods A pregnant women cohort was conducted at Beijing Yanshan Petrochemical Corporation, and 1,097 mothers with live singleton births were included in analysis. Generalized Multifactor Dimensionality Reduction (GMDR) method was used to explore interactions among these factors with adjustment for important potential confounders. Multiple linear regression models were used to estimate the association of interaction with gestational age.

Results A three-factor model of organic solvents, *GSTT1* and *PON2* *rs12026* had the highest testing balanced accuracy (57.05 %) and best cross-validation consistency (10/10). Compared with organic solvents unexposed mothers with *GSTT1* non-null genotype and *PON2* *rs12026* CC genotype, organic solvents exposed mothers with *GSTT1* null genotype and *PON2* *rs12026* CG + GG genotype had the largest reduction in gestational age (−0.36 weeks, 95%CI: −0.70 to −0.02). The significant reductions in different groups were from 0.24 weeks to 0.36 weeks.

Conclusions Maternal genetic susceptibility *GSTT1* and *PON2* *rs12026* could significantly modify the association of organic solvents with gestational age.

Keywords Gene-environment interaction · Gestational age · Glutathione transferase · Paraoxonase

Abbreviations

CI	confidence interval
CYP1A1	cytochrome P-450 1A1
EPHX1	epoxide hydrolase 1
GMDR	generalized multifactor dimensionality reduction
GSTT1	glutathione S-transferase theta-1
hCG	human chorionic gonadotrophin
OSHA	Occupational Safety and Health Administration
PON2	paraoxonase2

Introduction

Shortened gestational age, like preterm delivery (gestational age < 37 weeks), contributes greatly to infant mortality [1]. Because of its high prevalence, its association with mortality and morbidity, as well as the cost of hospitalization and long-term disability, shortened gestational age has become a major public health concern [2].

Identifying risk factors is essential to the prevention of shortened gestational age. Organic solvents and smoking are common environmental risk factors. Many epidemiological studies have found associations between organic solvents and adverse pregnancy outcomes [3–6]. As to smoking, both active smoking and passive smoking are associated with the risk of adverse birth outcomes [7–9].

Capsule Organic solvents exposure and maternal genetic variants: *GSTT1* and *PON2* *rs12026* have an interaction on gestational age in Chinese women.

S. Li · K. Fang · Y. Hu (✉) · D. Chen (✉)
Department of Biostatistics and Epidemiology, School of Public Health, Peking University, No.38, Xueyuan Road, Beijing, China
e-mail: yihu@bjmu.edu.cn
e-mail: dafangchen@bjmu.edu.cn

W. Wang
The Bureau for Health Inspection and Supervision of Haidian District, No.12 Ganjiakou, Beijing, China

Maternal genetic susceptibility is another type of risk factor. Four functional genes: *CYP1A1*, *EPHX1*, *GSTT1* and *PON2* have received attention. These genes all encode enzymes involving in metabolizing and detoxifying xenobiotics: *CYP1A1* encodes a member of cytochrome P450 superfamily enzymes which serves as the major enzyme in phase I metabolism; *EPHX1* encodes epoxide hydrolase which can active and detoxify epoxides from the degradation of toxins; *GSTT1* encodes glutathione S-transferase (GST) theta-1, a member of superfamily enzymes playing an important role in phase II metabolism; and *PON2* encodes a member of paraoxonase family which can protect cells from oxidative stress. Molecular epidemiological studies have provided evidences that these genes were associated with adverse pregnancy outcomes: Wang et al. found that *CYP1A1* *MspI* Aa + aa genotype was associated with low-birth-weight infants (OR=3.2, 95%CI: 1.6 to 6.4) [10]; Nukui et al. found that *GSTT1* null genotype was associated with the risk of premature birth ($P=0.001$) [11]; Wu et al. found an association between *EPHX1* *Tyr113His* His/His genotype and reduced birth weight (−167.9 g, 95%CI: −329.6 to −6.1) [12]; and an association between *PON2* *S311C* CC genotype and preterm delivery was found by Chen et al. (OR=4.6, 95%CI: 1.5 to 14) [13].

There are also gene-gene interactions or gene-environment interactions among these four genes, organic solvents and smoking on adverse pregnancy outcomes. Epidemiological studies have found these interactions on gestational age: Tsai et al. reported a joint association between maternal smoking, *CYP1A1* *MspI* (Aa/aa) and *GSTT1* (null) genotypes and preterm delivery (OR=5.8, 95%CI: 2.0 to 21.1) [14]; Wang et al. found that benzene exposure, *CYP1A1* *HincII* (AA) and *GSTT1* (null) could synergistically increase the risk of shortened gestational age (−0.79 weeks, $P=0.002$) [15]. However, these studies only searched interactions among limited candidate risk factors. Interactions among all six factors on gestational age are not well studied.

Due to reasons above, in this study, we used the data from a prospective reproductive cohort study conducted in China to explore interactions among all six risk factors above. No subject in this study smoked, so we used passive smoking instead of active smoking. The genetic polymorphisms we chose were *CYP1A1* *rs4646903* (*MspI*), *EPHX1* *rs2234922* (*His139Arg*), *GSTT1* and *PON2* *rs12026* (*Ala148Gly*). We hypothesized that there were combined association between these six risk factors and shortened gestational age.

Subjects and methods

Study site and population

This prospective cohort study was conducted at Beijing Yanshan Petrochemical Corporation (BYPC), located in a suburban area of Beijing, China. In BYPC, the major occupational exposure of

organic solvents includes benzene, toluene, styrene and their derivatives. The level of organic solvents exposure in BYPC is very low. In study period, the time-weighted average for benzene during an 8-h shift for exposed workers ranged from 0.017 to 0.191 ppm, a concentration far below the limit of 1 ppm as an 8-h time weighted average recommended by the Occupational Safety and Health Administration (OSHA) [16].

The study population was female workers who newly got married and wanted pregnancy between June 1995 and June 2000. The eligibility criteria for women who enrolled in this study were as follows: full-time employed workers; live singleton birth; and aged 20–34 years. Women who had multiple gestations, birth with major congenital defects or medically diagnosed gynecological or endocrine disorders were excluded from the study.

Data collection procedures

Eligible women were enrolled by the BYPC Staff Hospital, which is the only regional hospital that provides routine physical examination, family planning counseling, prenatal care and delivery services to female employees of BYPC. After informed consent was obtained, questionnaire investigation was administered by trained interviewers to collect pre-pregnancy baseline information: demographic characteristics, medical and reproductive history and personal habits. A physical examination was performed to obtain pre-pregnancy height and weight according to a standard protocol. If a woman reported a missed or late period or had early signs or symptoms of pregnancy, she was introduced to the hospital for a check-up and to give a urine sample to confirm pregnancy. A woman who was confirmed to be pregnant received regular prenatal care and delivery services according to standard clinical guidelines and was followed-up for pregnancy outcomes. Birth outcomes such as infant sex, gestational age, and birth weight were obtained from medical records by a trained nurse. Blood samples were obtained from mothers via venipuncture by a skilled phlebotomist. The study protocol was approved by the ethics committee of Peking University Health Science Center.

Exposure assessments

The measurement of maternal organic solvents exposure in this study was based on a specialized industrial hygiene method. Briefly, a checklist of 55 potential reproductive hazards present in the industry was developed based on toxicologic literature. This checklist was used to identify potential exposures for each subject. An industrial hygienist who was familiar with the BYPC production process but did not have knowledge of birth outcomes classified those women into exposed or unexposed group according to their workshops and job activities. Pregnant workers stopped working at about 28 weeks of gestation.

Information on other exposures such as passive smoking, noise, vibration, dust, and perceived work stress were also obtained from the questionnaire interview by trained

interviewers. For passive smoking, the specific question is “Do your family members smoke at home?”; for noise, the specific question is “Do you work in a noisy environment?”; for vibration, the specific question is “Are you affected by vibration in your work?”; for dust, the specific question is “Are you affected by dust in your work?”; and for perceived work stress, the specific question is “Do you feel stressful when working?”.

Gestational age assessment

In this study, the first day of the last menstrual period recorded on the first prenatal visit was used to estimate the gestational age. This measurement was accurate in this population for several reasons. In China, married couples who plan to have a child need to apply for birth permission at the local family planning administration. Due to the one child policy, families highly concern about healthy pregnancy and healthy babies. All women received a human chorionic gonadotrophin (hCG) assay to verify pregnancy in the BYPC Staff Hospital soon after missing a menstrual period. Furthermore, the gestational age was calculated in exact days then divided by 7 instead of rounded completed weeks.

Genotyping methods

CYP1A1 rs4646903 polymorphism was detected according to the method of Kawajiri et al. [17]. This process resulted in 295-, 160-, and 135-bp products and was able to detect all three genotypes for the polymorphism.

EPHX1 rs2234922 polymorphism was detected according to the method of Hassett et al. [18]. This process resulted in 298-, 177-, and 121-bp products and was able to detect all three genotypes for the polymorphism.

GSTT1 polymorphism was detected according to the method of Nelson et al. [19]. The non-null genotype of *GSTT1* was indicated by an amplified 457-bp product, while the null genotype was indicated by the absence of a 457-bp band.

PON2 rs12026 polymorphism was detected by the method of Sanghera et al. [20]. This process resulted in 153-, 123- and 30-bp products and was able to detect all three genotypes for the polymorphism.

Statistical methods

Considering the low number of preterm infants (1.64 %) and to preserve statistical power, we analyzed our data using gestational age as a continuous variable.

At first, we used the χ^2 test for goodness-of-fit to check whether the distribution of genotypes in this population deviated from the Hardy–Weinberg Equilibrium. Then we used a univariate linear regression model to investigate individual association between gestational age and different risk factors.

In order to detect interactions among organic solvents exposure, passive smoking, *CYP1A1 rs4646903*, *EPHX1 rs2234922*, *GSTT1* and *PON2 rs12026* on gestational age, we used the Generalized Multifactor Dimensionality Reduction (GMDR) method [21]. Involving multiple factors in a traditional model means lots of multifactorial combinations, and some combinations may have few or no data. One major advantage of the GMDR method is that higher dimensions of multiple factors can be reduced to one dimension (low or high risk) within all possible combinations of factors. It permits adjustment for covariates and is applicable to both dichotomous and continuous phenotypes. Briefly, we at first calculated a score for gestational age with adjustment for covariates as following [15, 22, 23]: age (25, 25–28 and ≥ 28 years), education (elementary or middle, high school, and college or above), pregnancy history (no, yes), infant sex (male, female), pre-pregnancy BMI (<18.5, 18.5–24, 24–27 and ≥ 27), noise exposure (no, yes), vibration exposure (no, yes), and perceived work stress (no, yes). Then the data was divided into 10 sets: 9 for training and 1 for testing. *N* factors were selected from the training set and their combinations were represented in *n*-dimensional space. The GMDR classified each combination (multifactorial class) as ‘high risk’ or ‘low risk’, thus reduced the *n*-dimensional space to one dimension with two levels. For each possible model size (one-factor, two-factor, etc.), the model with the lowest misclassification error was selected. Leave-one-out cross-validation was used to calculate the prediction error and to evaluate the predictive ability of the model in the test data. The result was a set of models, a final model was chosen on the basis of minimum prediction error and maximum cross-validation consistency. After obtaining the final model, 1000 permutation tests were performed to obtain an empirical *P* value. Then we used multiple linear regression models to estimate the joint association of factors in the final model with gestational age with adjustment for covariates described above.

All *P* values were two-sided, and statistical significance was defined as *P*=0.05. The GMDR procedure was performed by using the GMDR software Beta Version 0.7 (<http://www.healthsystem.virginia.edu/internet/Addiction-Genomics/>). The other statistical analyses were performed by using the SAS software Version 9.1 (SAS Institute Inc., Cary, North Carolina).

Results

The study population contained 1,334 mothers who delivered infants. 237 mothers were excluded for having multiple gestations, birth with major congenital defects, failure of extracting DNA, failure of genotyping or missing data. Finally, a total of 1,097 mothers were included in final analysis. Since *GSTT1* had only two genotypes: non-null and null, we

Table 1 Gestational age by different groups of characteristics, exposures and polymorphisms

Variables ^a	No. (%)	Mean (SD) ^b	β ^c	95 % CI ^d	<i>P</i> value
Total sample	1,097 (100.0)	39.96 (1.30)			
Age					
<25	245 (22.3)	39.98 (1.47)	reference		
25–28	607 (55.3)	39.98 (1.27)	0.00	−0.19, 0.19	0.997
≥28	245 (22.3)	39.89 (1.20)	−0.09	−0.33, 0.14	0.424
Pre-pregnancy BMI					
<18.5	98 (8.9)	39.59 (1.31)	−0.37	−0.64, −0.10	0.008
18.5–24	661 (60.3)	39.96 (1.22)	reference		
24–27	211 (19.2)	39.95 (1.37)	−0.01	−0.22, 0.19	0.884
≥27	127 (11.6)	40.24 (1.54)	0.28	0.03, 0.53	0.026
Education					
≤middle school	276 (25.2)	40.07 (1.24)	reference		
high school	529 (48.2)	39.96 (1.34)	−0.12	−0.31, 0.07	0.229
≥college	292 (26.6)	39.86 (1.29)	−0.21	−0.43, 0.00	0.053
Infant sex					
male	548 (50.0)	39.89 (1.34)	reference		
female	549 (50.0)	40.03 (1.26)	0.15	−0.01, 0.30	0.065
Pregnancy history					
no	636 (58.0)	39.95 (1.30)	reference		
yes	461 (42.0)	39.98 (1.31)	0.03	−0.13, 0.18	0.721
Organic solvents					
no	737 (67.2)	40.03 (1.27)	reference		
yes	360 (32.8)	39.82 (1.36)	−0.21	−0.37, −0.05	0.012
Passive smoking					
no	590 (53.8)	39.95 (1.31)	reference		
yes	507 (46.2)	39.97 (1.30)	0.02	−0.13, 0.18	0.791
Stress exposure					
no	734 (66.9)	39.90 (1.34)	reference		
yes	363 (33.1)	40.09 (1.21)	0.19	0.03, 0.36	0.021
Noise exposure					
no	831 (75.8)	39.96 (1.27)	reference		
yes	266 (24.2)	39.97 (1.40)	0.01	−0.17, 0.19	0.916
Vibration exposure					
no	1,018 (92.8)	39.96 (1.29)	reference		
yes	79 (7.2)	39.97 (1.48)	0.02	−0.28, 0.31	0.920
Dust exposure					
no	1,014 (92.4)	39.98 (1.25)	reference		
yes	83 (7.6)	39.75 (1.79)	−0.22	−0.51, 0.07	0.136
<i>CYP1A1</i> rs4646903					
TT	419 (38.2)	39.97 (1.41)	reference		
TC	527 (48.0)	39.99 (1.23)	0.02	−0.14, 0.19	0.778
CC	151 (13.8)	39.83 (1.23)	−0.13	−0.38, 0.11	0.283
<i>EPHX1</i> rs2234922					
AA	871 (79.4)	39.96 (1.28)	reference		
AG + GG	226 (20.6)	39.94 (1.39)	−0.02	−0.21, 0.17	0.811
<i>GSTT1</i>					
non-null	585 (53.3)	40.00 (1.28)	reference		
null	512 (46.7)	39.91 (1.33)	−0.09	−0.25, 0.06	0.248

Table 1 (continued)

Variables ^a	No. (%)	Mean (SD) ^b	β ^c	95 % CI ^d	<i>P</i> value
<i>PON2 rs12026</i>					
CC	713 (65.0)	39.96 (1.29)	reference		
CG + GG	384 (35.0)	39.95 (1.32)	−0.02	−0.18, 0.15	0.842

^a For *CYP1A1 rs4646903*, C is the minor allele; for *EPHX1 rs2234922*, G is the minor allele; and for *PON2 rs12026*, G is the minor allele

^b SD, Standard Deviation

^c β represents the difference in mean gestational age (weeks) for each group compared reference group in characteristics, exposures and polymorphisms

^d 95 % CI, 95 % confidence interval

did not test its departure from the Hardy–Weinberg Equilibrium. The other three polymorphisms followed the Hardy–Weinberg Equilibrium (for *CYP1A1 rs4646903*, $\chi^2=0.521$, $P=0.471$; for *EPHX1 rs2234922*, $\chi^2=1.180$, $P=0.277$; and for *PON2 rs12026* $\chi^2=1.298$, $P=0.255$).

Individual associations between gestational age and different risk factors in these 1,097 mothers are shown in Table 1. On account of the low frequency of *EPHX1 rs2234922* GG genotype, we combined them with AG genotype for analysis, and so as for *PON2 rs12026*. We found no significant association between gestational age and passive smoking. As to the four polymorphisms, we did not find significant individual association, neither. However, we found that those with BMI<18.5 or with BMI $\geq$ 27 had shorter gestational age compared with subjects with normal BMI (18.5–24); organic solvents exposure was associated with shortened gestational age; mothers unexposed to stress tended to have shorter gestational age.

Then we used the GMDR method to detect interactions among organic solvents exposure, passive smoking and those four polymorphisms. Best models of different sizes are shown in Table 2. We found the most significant ($P=0.001$) interaction on gestational age among following three factors: organic solvents exposure, *GSTT1*, and *PON2 rs12026*. This three-factor model had the highest testing balanced accuracy (57.05 %) and best cross-validation consistency (10/10). It was the best and final model of interaction that we chose.

Then we performed 1,000 permutation tests, and the empirical *P* value for this three-factor model was 0.01.

Finally, we used multiple linear regression models to estimate the joint association between *GSTT1*, *PON2 rs12026* and organic solvents exposure and gestational age, and the result is shown in Table 3. We found that except the combination “no organic solvents exposure, *GSTT1* null genotype and *PON2 rs12026* CG + GG genotype”, all the other combinations could shorten gestational age when compared with the reference combination “no organic solvents exposure, *GSTT1* non-null genotype and *PON2 rs12026* CC genotype”. Among these six combinations associated with shortened gestational age, four combinations had significant reductions (−0.36 weeks, −0.33 weeks, −0.27 weeks and −0.24 weeks, respectively). The largest significant reduction was found in the combination “organic solvents exposure, *GSTT1* null genotype and *PON2 rs12026* CG + GG genotype” (−0.36 weeks, 95 % CI −0.70 to −0.02).

Discussion

In our study, we explored the interaction among maternal organic solvents exposure, passive smoking and four polymorphisms on gestational age by using the GMDR method. This study has some unique features: 1) it is a study

Table 2 Results of GMDR analysis on gestational age ^a

Model size	Factors in model	Testing balanced accuracy (%)	Cross-validation consistency	<i>P</i> value ^b
One factor	Organic solvents	53.34	10/10	0.0547
Two factors	<i>GSTT1</i> , <i>PON2</i>	55.03	9/10	0.0107
Three factors	<i>GSTT1</i> , <i>PON2</i> , Organic solvents	57.05	10/10	0.0010
Four factors	<i>CYP1A1</i> , <i>GSTT1</i> , <i>PON2</i> , Organic solvents	54.05	10/10	0.1719
Five factors	<i>CYP1A1</i> , <i>EPHX1</i> , <i>GSTT1</i> , <i>PON2</i> , Organic solvents	50.27	10/10	0.6230
Six factors	<i>CYP1A1</i> , <i>EPHX1</i> , <i>GSTT1</i> , <i>PON2</i> , Organic solvents, passive smoking	49.92	10/10	0.8281

^a Adjustment for age (25, 25–28 and $\geq$ 28 years), education ($\leq$ middle, high school, $\geq$ college), pregnancy history (no, yes), infant’s gender (male, female), pre-pregnancy BMI (<18.5, 18.5–24, 24–27 and $\geq$ 27), noise exposure (no, yes), vibration exposure (no, yes), and perceived work stress (no, yes)

^b *P* value obtained using the nonparametric sign test for testing accuracy

Table 3 Joint association of organic solvents exposure, *GSTT1* and *PON2 rs12026* with gestational age ^a

Organic solvents	<i>GSTT1</i>	<i>PON2 rs12026</i>	Adjusted mean	β ^b	95 % CI ^c	<i>P</i> value
No	Non-null	CC	40.11	reference		
No	Non-null	CG + GG	39.84	−0.27	−0.52, −0.02	0.037
No	Null	CC	39.87	−0.24	−0.46, −0.02	0.034
No	Null	CG + GG	40.25	0.15	−0.13, 0.42	0.290
Yes	Non-null	CC	39.97	−0.14	−0.41, 0.14	0.326
Yes	Non-null	CG + GG	39.97	−0.14	−0.47, 0.19	0.403
Yes	Null	CC	39.77	−0.33	−0.61, −0.06	0.017
Yes	Null	CG + GG	39.75	−0.36	−0.70, −0.02	0.037

^a Adjustment for age (25, 25–28 and ≥ 28 years), education ($\leq$ middle, high school, $\geq$ college), pregnancy history (no, yes), infant's gender (male, female), pre-pregnancy BMI (<18.5 , 18.5–24, 24–27 and ≥ 27), noise exposure (no, yes), vibration exposure (no, yes), and perceived work stress (no, yes)

^b β represents the difference in adjusted mean gestational age (weeks) for each combination compared reference combination

^c 95 % CI, 95 % confidence interval

researching on the interaction on gestational age among several candidate environmental risk factors and maternal genetic risk factors; 2) it uses the GMDR method to reduce the dimensionality of multifactor, which can solve the problem “curse of dimensionality” [24]; 3) the study population is an overall low-risk population: non-smoking, non-drinking, optimal maternal age, planned pregnancy among married couples and early prenatal care, which provides a precious opportunity to detect the interaction without potential socio-demographic and environmental confounders.

In this study, we could not provide evidence that there's individual association among passive smoking, *CYP1A1 rs4646903*, *EPHX1 rs2234922*, *GSTT1* and *PON2 rs12026*. This result might due to following reasons: 1) we used passive smoking instead of active smoking. Passive smoking may have less harm than active one that we could not find it in our population. 2) Some significant genetic variants were found in populations other than Chinese population [10, 11, 14]. The allele frequencies in different populations are different. For example, the MAF of *CYP1A1 rs4646903* in European population is 0.1; however, it is 0.417 in Asian population.

We found a three-way gene-environment interaction among *GSTT1*, *PON2 rs12026* and organic solvents exposure on the length of gestational age. Organic solvents exposure, even at a level at least five times lower than the safety level required by the Occupational Safety and Health Administration, was significantly associated with shortened gestational age. When considering the two polymorphisms in addition, the largest statistically significant reduction was observed in the combination “organic solvents, *GSTT1* null genotype and *PON2 rs12026* CG + GG genotype”, suggesting that these two polymorphisms together could modify the association between organic solvents and shortened gestational age.

Detecting risk factors of preterm birth is the primary interest of many studies. However, in our study population, the

percentage of preterm birth was 1.64 %, and the magnitude of reduction in gestational age was not of major significance. Since the population is low-risk as described above, the low percentage of preterm birth is reasonable. However, our findings have some important implications: 1) organic solvents exposure and susceptible genotypes are prevalent in general population, so the 0.2–0.4 weeks leftward shift in the gestational age distribution curve among the high-risk population could lead a significantly increased number of preterm births; 2) the result reveals that genetic susceptibility can modify the association between organic solvents exposure and reduced gestational age, which supports the importance of considering genetic susceptibility when evaluating reproductive toxicants.

To our knowledge, this study firstly provides evidence for a gene-environment interaction among *GSTT1*, *PON2 rs12026* and organic solvents on gestational age. Although with no other studies supporting our results, our finding is biologically plausible. Organic solvents have been proved to be reproductive toxicants by epidemiological studies [3, 6, 23, 25]. Since organic solvents are liquid soluble, they can pass through the placenta and can reach the fetus and induce harmful effects [26]. They can induce DNA damage, including sister chromatid exchanges [27], micronuclei formation [28] and chromosomal aberrations [29]. The reproductive toxicity of organic solvents also involves oxidative stress [30, 31]. Glutathione S-transferase serves as a major role in the detoxification of organic solvents. The *GSTT1* enzyme is important in protecting against organic solvents damage, such as sister chromatid exchange and the formation of hemoglobin adducts [32, 33]. The *GSTT1* gene encodes GSTT1 enzyme, and its complete deletion leads to the loss of enzyme activity [34, 35]. Paraoxonase-2, encoded by *PON2* gene, is not well studied for its function so far. Several studies have demonstrated its antioxidative potential [36, 37], which can reduce the toxicity of organic solvents. Little information is available for the association between *PON2 rs12026* and paraoxonase-2

activity. However, according to the linkage disequilibrium (LD) information available for Han Chinese in Beijing China in the International HapMap Database (<http://www.hapmap.org>), *PON2 rs12026* and *PON2 rs7493* (Ser311Cys, G allele is the minor allele) are in strong LD ($D=1.0$, $r^2=1.0$), and *rs12026* G allele and *rs7493* G allele are in one haplotype. Stoltz et al. found that *PON2 rs7493* G allele could impair the lactonase activity of paraoxonase-2 [38]. Thus it is plausible that *PON2 rs12026* G allele associates with the activity reduction of paraoxonase-2 and the detoxification impairment of organic solvents. Regarding the biological plausibility described above, subjects with *GSTT1* null genotype and *PON2 rs12026* G allele would be less resistant to organic solvents toxicity. Consistently, our study found that subjects with *GSTT1* null genotype and *PON2 rs12026* G allele had the shortest gestational age when exposed to organic solvents.

Several methodological limitations should be taken into consideration when interpreting our results. First, subjects of this study were low-risk, so it should take caution to generalize our results to other populations. Second, we combined benzene, toluene, etc. as one exposure, which made it hard to detect the effect of a particular organic solvent. Third, many exposures were assessed based on self-report, which might generate bias. In addition, we adjusted many covariates in the GMDR analysis and regression analysis. However, we could not rule out the possible confounding effects by other uncontrolled or inadequately controlled risk factors.

In summary, searching among six risk factors, we find that maternal genetic susceptibility *GSTT1* and *PON2 rs12026* significantly modify the association of organic solvents with gestational age, which provides a new insight of gene-environment interaction on gestational age.

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Ethical issue This study was approved by the ethics committee of Peking University Health Science Center.

All participants gave their informed content prior to participating in this study.

Conflict of interest The authors declare that they have no conflict of interest.

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