Received: 30 November 2017 | Revised: 19 January 2018 | Accepted: 13 February 2018

DOI: 10.1002/ajmg.a.38661

ORIGINAL ARTICLE

WILEY AMERICAN JOURNAL OF medical genetics PART A

Genetic variation in biotransformation enzymes, air pollution exposures, and risk of spina bifida

Amy M. Padula[1] | Wei Yang[2] | Kathleen Schultz[3] | Fred Lurmann[4] | S. Katharine Hammond[5] | Gary M. Shaw[2]

[1]Department of Obstetrics, Gynecology and Reproductive Sciences, University of California, San Francisco, San Francisco, California

[2]Department of Pediatrics, Stanford University School of Medicine, Stanford, California

[3]UCSF Benioff Children's Hospital Oakland, Oakland, California

[4]Sonoma Technology Inc, Petaluma, California

[5]Department of Environmental Health Sciences, University of California, Berkeley, Berkeley, California

Correspondence
Amy M. Padula, 550 16th Street, San Francisco, CA 94143.
Email: amy.padula@ucsf.edu

Funding information
Centers for Disease Control and Prevention, Grant/Award Number: U50/CCU913241; California Tobacco-Related Disease Research Program, Grant/Award Number: 16RT-0086; Environmental Protection Agency, Grant/Award Number: RD835435; National Institutes of Health, Grant/Award Number: P01ES022849, R00ES021470

Spina bifida is a birth defect characterized by incomplete closure of the embryonic neural tube. Genetic factors as well as environmental factors have been observed to influence risks for spina bifida. Few studies have investigated possible gene-environment interactions that could contribute to spina bifida risk. The aim of this study is to examine the interaction between gene variants in biotransformation enzyme pathways and ambient air pollution exposures and risk of spina bifida. We evaluated the role of air pollution exposure during pregnancy and gene variants of biotransformation enzymes from bloodspots and buccal cells in a California population-based case-control (86 cases of spina bifida and 208 non-malformed controls) study. We considered race/ethnicity and folic acid vitamin use as potential effect modifiers and adjusted for those factors and smoking. We observed gene-environment interactions between each of the five pollutants and several gene variants: NO (*ABCC2*), NO_2 (*ABCC2*, *SLC01B1*), PM_{10} (*ABCC2*, *CYP1A1*, *CYP2B6*, *CYP2C19*, *CYP2D6*, *NAT2*, *SLC01B1*, *SLC01B3*), $PM_{2.5}$ (*CYP1A1* and *CYP1A2*). These analyses show positive interactions between air pollution exposure during early pregnancy and gene variants associated with metabolizing enzymes. These exploratory results suggest that some individuals based on their genetic background may be more susceptible to the adverse effects of pollution.

KEYWORDS
air pollution, congenital anomalies, gene, gene-environment, spina bifida

1 | INTRODUCTION

Spina bifida is a human structural birth defect characterized by incomplete closure of the embryonic neural tube and is the more frequently observed phenotype among its broader group known as neural tube defects. Folic acid fortification of the food supply has been associated with reductions of neural tube defects by approximately 20% in the United States (Honein, Paulozzi, Mathews, Erickson, & Wong, 2001).

Additionally, other environmental risk factors have been hypothesized to contribute to neural tube defect risk; several studies have examined the role of air pollution (Girguis et al., 2016; Lupo et al., 2011; Padula et al., 2013). The results across studies are not consistent, though several studies do find increased risk associations between early prenatal exposure to air pollution and neural tube defects (Lupo et al., 2011), including our previous study, which found an association between carbon monoxide (CO) and nitrogen dioxide (NO_2) and increased risk of spina bifida (Padula et al., 2013).

It is thought that most structural birth defects are caused by a complex combination of genetic and environmental factors that interact to interfere with morphogenetic processes; however, few studies have examined the interaction of genetic and environmental factors. The few studies that do exist examined the interaction with smoking (Jenkins et al., 2014; Torfs, Christianson, Iovannisci, Shaw, & Lammer, 2006; Wu et al., 2012) or nutrition (Enaw et al., 2006; Wu et al., 2010)

Abbreviations: CI, confidence interval; NC, not calculated; OR, odds ratio.

and one with occupational chemical exposures (Shaw, Nelson, Iovannisci, Finnell, & Lammer, 2003). Fewer such gene-environment interaction studies have been conducted specifically on spina bifida with most focused on folate transport or metabolism.

The current study examines the interaction between gene variants in biotransformation enzyme pathways, enzyme pathways known to mediate detoxification of xenobiotic exposures, and ambient air pollution exposures and risk of spina bifida risk in a population-based case-control study the San Joaquin Valley of California.

2 | METHODS

2.1 | Study population

The California Center of the National Birth Defects Prevention Study (Reefhuis et al., 2015; Yoon et al., 2001) is a collaborative partnership between Stanford University and the California Birth Defects Monitoring Program in the Department of Public Health. Since 1997, the Center has collected data from women residing in eight counties (San Joaquin, Stanislaus, Merced, Madera, Fresno, Kings, Tulare, and Kern) in the San Joaquin Valley. The California Birth Defects Monitoring Program is a surveillance program that is population-based (Croen, Shaw, Jensvold, & Harris, 1991). To identify cases with birth defects, data collection staff visit all hospitals with obstetric or pediatric services, cytogenetic laboratories, and all clinical genetics prenatal and postnatal outpatient services.

Cases in the current analysis included infants or fetuses with spina bifida as confirmed by clinical, surgical, or autopsy reports. Cases recognized or strongly suspected to have single-gene conditions or chromosomal abnormalities or with identifiable syndromes were ineligible (Rasmussen et al., 2003), given their presumed distinct underlying etiology. Controls included non-malformed live-born infants randomly selected from birth hospitals to represent the population from which the cases arose. Maternal interviews were conducted by using a standardized, computer-based questionnaire, primarily by telephone, in English or Spanish, between 6 weeks and 24 months after the infant's estimated date of delivery. Estimated date of conception was derived by subtracting 266 days from the expected date of delivery. The expected date of delivery was based on self-report; if unknown, it was estimated from information in the medical records (<2% of participants).

Interviews were conducted with mothers of 74% of eligible cases and 69% of controls. The present analysis includes 86 cases of spina bifida and 208 controls with estimated delivery dates between October 1, 1997, and December 31, 2006. Mothers with diabetes (type 1 or type 2) prior to gestation were excluded. Mothers reported a full residential history from 3 months before conception through delivery, including start and stop dates for each residence. The Centers for Disease Control and Prevention geocoded the addresses by using Centrus Desktop (Pitney Bowes, Inc., Stamford, CT), which combines reference street networks from Tele Atlas B. V. ('s-Hertogenbosch, the Netherlands) and U.S. Postal Service data. Geocodes were available for the addresses of 95% of cases ($n = 151$) and 93% of controls ($n = 900$). Out of 138 cases and 849 controls exposed to any pollutants during the first 2 months of pregnancy, we identified 101 and 508 with blood spots or buccal samples available at lab. Out of 508 controls, we randomly selected 250 samples for further genotyping. We finally were able to genotype 96 cases and 230 controls due to missing samples. The present analysis includes 86 cases of spina bifida and 208 controls with call rate $\geq$ 89%.

2.2 | Genotyping analyses

For genetic experiments, DNA was derived from newborn bloodspots (infants only) or buccal samples (infant and mother of infants). A specific method to extract DNA was developed in the Lammer lab and has been used for numerous genotyping preparations in our molecular epidemiology work [e.g., (Shaw et al., 2003)]. We used this method to extract genomic (not amplified) DNA (gDNA) of sufficient quality and quantity from these precious bloodspots to provide excellent performance with Illumina GWAS platforms (2.5m). Genomic DNA was extracted from buccal brushes using an established protocol (NaOH extraction (Richards et al., 1993) along with the QIAquickR Purification kit (Qiagen, Valencia, CA)). Genotyping of DNA from buccal brush samples was performed on purified, unamplified genomic DNA. Further, genotyping calls from high-density polymorphism arrays (Human660W-Quad BeadChip) are highly concordant (99.9%) between DNA derived from buccals versus blood (Dr. Charlotte Hobbs, personal communication).

The TaqMan® OpenArray® PGx Panel (derived from the PharmaADME Core Marker Set) is an efficient, easy-to-use OpenArray® plate for pharmacogenomics applications. Assays were developed to detect polymorphisms in genes encoding metabolism enzymes and associated transport proteins. The panel contained 158 assays.

Although mainly known as "drug metabolizing enzymes," such biotransformation enzymes are involved in metabolizing both endogenous compounds and myriad xenobiotic chemicals (Nebert, 1997). For xenobiotics, these enzymes are important for detoxifying both parent compounds and reactive intermediate chemicals that may be teratogenic. Genetic variants have been described for a number of these metabolizing enzymes. For this project, we chose candidate genes whose variants are known to have altered enzyme activity or inducibility by xenobiotic compounds likely to be encountered in a pregnant woman's environment. These genes include, for example, the acetyl-*N*-transferases (NATs, *NAT1* 1088, *NAT11095*, and *NAT2*), cytochrome P450 (*CYP1A1*, *CYP1A2*, *CYP2A6*, *YPC2B6*, *CYP2C19*, *CYP2C8*, *CYP2C9*, *CYP2D6*, *CYP3A4*, *CYP3A5)*, and the glutathione *S*-transferases (*GSTM1* and *GSTT1*). The full list of gene variants is shown in Table 2. We also included other relevant genes like nitric oxide synthase (*NOS3*), which regulates nitric oxide production and has been associated with orofacial clefts and maternal smoking (Shaw et al., 2005).

For each gene variant, the Haploview Program (version 4.2, http://www.broadinstitute.org/scientific-community/science/programs/medical-and-population-genetics/haploview/haploview) (Barrett, Fry, Maller, & Daly, 2005) was used to calculate minor allele frequency (MAF) and to evaluate deviations from Hardy–Weinberg equilibrium (HWE) among controls. These analyses were conducted for all participants

together and separately for native-born Hispanic, foreign-born Hispanic, and non-Hispanic white mothers.

Out of 158 gene loci, there were 27 loci without variation (i.e., all were wildtype). An additional 27 SNPs failed the HWE among controls. Therefore, results include 104 SNPs. Odds ratios (ORs) were not calculated (NC) for and case/control counts < 3.

2.3 | Air pollution exposure assessment

As part of the Children's Health and Air Pollution Study, ambient air pollution measurements and traffic metrics were assigned to each of the geocoded residences reported by study subjects corresponding to their first and second months of pregnancy (this period approximates the closure of the embryonic neural tube). If there was more than 1 address during the period, exposure assignments were calculated for the number of days at each residence. Exposure assignments were made if the geocodes were within the San Joaquin Valley and were available for at least 75% of each month. Daily 24-hr averages of NO_2, nitrogen oxide, CO, particulate matter <10 μm (PM_{10}), and particulate matter <2.5 μm ($PM_{2.5}$) were then averaged over the first 2 months of pregnancy.

Ambient air quality data have been collected routinely at more than 20 locations in the San Joaquin Valley since the 1970s, and these data were acquired from the US Environmental Protection Agency's Air Quality System database (www.epa.gov/ttNCirs/airsaqs). The station-specific daily air quality data were spatially interpolated by using inverse distance-squared weighting. Data from up to 4 air quality measurement stations were included in each interpolation. Owing to the regional nature of NO_2, PM_{10}, and $PM_{2.5}$ concentrations, we used a maximum interpolation radius of 50 km. Nitrogen oxide and CO were interpolated by using a smaller maximum interpolation radius of 25 km because they are directly emitted pollutants with larger spatial gradients. When a residence was located within 5 km of 1 or more monitoring stations, the interpolation was based solely on the nearby values.

Gaseous pollutants were measured by using Federal Reference Method (US Environmental Protection Agency, Office of Air Quality Planning and Standards, Research Triangle Park, NC) continuous monitors. Particulate matter data were primarily limited to those collected with Federal Reference Method samplers and Federal Equivalent Method monitors. The national air monitoring networks began measuring $PM_{2.5}$ in 1999; therefore, births with dates of conception prior to 1999 were not part of the analyses of $PM_{2.5}$.

2.4 | Statistical analyses

Risk for spina bifida associated with each infant gene variant was calculated for both the homozygotes and the heterozygotes, with homozygous wildtypes as the referent. For each gene variant, the wildtype/reference genotype was defined as the homozygous genotype with the most frequent allele among controls. Risks were estimated as ORs with 95% confidence intervals (CIs) by logistic regression using SAS software (version 9.4, SAS Institute, Cary, NC). Regression analyses were stratified by maternal race/ethnicity (Hispanic and non-Hispanic white). Wald chi-square tests were calculated for the interaction terms to determine if the subgroups were statistically different.

Regression analyses were stratified by air pollution exposure (highest tertile versus lower two tertiles). For these analyses, homozygous variants and heterozygotes were combined and compared to homozygous wildtypes as the referent. Wald chi-square tests were calculated for the interaction terms to determine if the subgroups were statistically different. ORs were calculated for 104 genotypes and 5 pollutants for a total of 520 comparisons for the gene-environment interaction analyses. These models were adjusted for maternal race/ethnicity, vitamin use (folic acid containing in 1 month before conception and first 2 months of pregnancy), BMI (continuous), and smoking (active and/or passive versus none). These analyses were additionally stratified by maternal use of vitamins containing folic acid.

We conducted a sensitivity analysis on the maternal genotypes to determine if the results were consistent ($N = 37$).

3 | RESULTS

The study population included 86 spina bifida cases and 208 controls from the San Joaquin Valley of California. The demographic characteristics of cases and controls are presented in Table 1. Case mothers were slightly less likely to have competed >12 years of education or to have used multivitamin supplements and slightly more likely to be foreign-born Hispanic and to be exposed to passive cigarette smoke. Case mothers were more likely to be overweight or obese and control mothers had a more even distribution across age categories.

Table 2 lists the position and reference allele of the gene variants ($N = 104$) and summarizes call rates, MAFs, and HWE evaluation using the HaploView Program.

Results of analyses exploring potential gene-only effects and spina bifida are presented in Supporting Information Table S1. Of the 104 gene variants that were analyzed, 4 had ORs with accompanying 95% CIs that excluded 1 and showed associations with spina bifida—two variants of the *ABCC2* gene (rs717620, rs3740066), one CYP2C9 variant (rs9332131), and one *NAT2* variant (rs1799931). The variant genotype (TT) of *ABCC2* (rs717620) was associated with spina bifida (OR = 5.4, 95%CI: 1.3–22.4). The heterozygous genotypes of *ABCC2* (rs3740066), CYP2C9 (rs9332131), and *NAT2* (rs1799931) were associated with two-fold or more increased odds of spina bifida (OR_{ABCC2} =1.9, 95% CI: 1.1–3.4; OR_{CYP2C9} =5.1, 95% CI: 1.2–20.7, OR_{NAT2} =2.1, 95% CI: 1.1–3.9).

When stratified by race/ethnicity (White non-Hispanic vs. Hispanic), homozygous and heterozygous variants of *ABCC2* (rs3740066) had stronger associations with spina bifida among Whites, but not Hispanics (Supporting Information Table S2). Additionally, the heterozygous genotype of *CYP2D6* (rs3892097) was associated with spina bifida only among Whites (OR = 3.1, 95% CI: 1.0–9.3).

Tables 3–7 present results of the gene variant-pollutant analyses, adjusted for maternal race, vitamin use, BMI, education, and smoking. Below we note the ORs with 95% CIs excluding 1 and *p* values of the

TABLE 1 Demographic characteristics of spina bifida cases and non-malformed controls, California 1997–2006

	Cases[a] (*N* = 86) *n* (%)	Controls[a] (*N* = 208) *n* (%)
Maternal race/ethnicity		
White non-Hispanic	19 (22)	76 (37)
US-born Hispanic	26 (30)	57 (27)
Foreign-born Hispanic	33 (38)	54 (26)
Other	8 (9)	21 (10)
Maternal age at delivery (years)		
<20	8 (9)	29 (14)
20–24	27 (31)	61 (29)
25–29	32 (37)	53 (25)
30–34	13 (15)	39 (19)
35+	6 (7)	26 (13)
Maternal education (years)		
<12	26 (30)	62 (30)
12	28 (33)	52 (25)
>12	32 (37)	93 (45)
Missing	0	1 (<1)
Parity		
0	26 (30)	69 (33)
1	27 (31)	68 (33)
2+	33 (38)	71 (34)
Maternal body mass index (kg/m^2)		
Underweight (<18.5)	2 (2)	4 (2)
Normal (18.5–<25)	29 (34)	96 (46)
Overweight (25–<30)	27 (31)	49 (24)
Obese (≥30)	20 (23)	41 (20)
Missing	8 (9)	18 (9)
Plurality		
Singletons	83 (97)	207 (99.5)
Multiples	3 (3)	1 (0.5)
Infant sex		
Male	42 (49)	94 (45)
Female	43 (50)	114 (55)
Missing	1 (1)	0
Multivitamin use[b]		
No	28 (33)	56 (27)
Yes	56 (65)	148 (71)
Missing	2 (2)	4 (2)
Smoking[b]		
None	64 (74)	160 (77)
Active only	8 (9)	15 (7)
Passive only	13 (15)	18 (9)
Active and passive	1 (1)	15 (7)
Air pollution exposure—mean (*SD*)		
CO (ppm)	0.6 (0.3)	0.6 (0.3)
NO (ppb)	16.0 (12.6)	15.1 (14.7)
NO_2 (ppb)	18.8 (5.1)	18.2 (5.6)
PM_{10} ($\mu g/m^3$)	38.9 (15.4)	35.5 (14.3)
$PM_{2.5}$ ($\mu g/m^3$)	23.7 (14.0)	19.3 (11.9)

[a]Percentages may not equal 100 owing to rounding and missing.
[b]During the month before or the first 2 months of pregnancy

TABLE 2 Characteristics of gene variants of controls, Hardy–Weinberg Equilibrium (HWE) evaluated using Haploview Program (*N* = 104)

Gene Symbol	dbSNP ID	Position	Reference Allele[a]	Call Rate %	MAF[b]	HWE *p* Value
ABCB1	rs1045642	7586657	G	100	0.46	.63
ABCB1	rs1128503	7586662	G	100	0.44	.96
ABCB1	rs2032582	11711719	C	99.1	0.42	.09
ABCB1	rs2032582	11711720	C	82	0.06	<.01
ABCC2	rs56199535	2864	C	98.2	<0.01	1.00
ABCC2	rs717620	2814642	C	98.2	0.14	.86
ABCC2	rs3740066	11214910	C	98.2	0.34	.23
ABCC2	rs2273697	22272980	G	98.2	0.17	.39
ABCG2	rs2231142	15854163	G	100	0.15	.92
CYP1A1	rs1048943	25624888	T	99.1	0.22	<.01
CYP1A1	rs41279188	30634152	G	99.1	0.00	1.00
CYP1A2	rs762551	8881221	A	100	0.29	.02
CYP1A2	rs2069514	15859191	G	94.6	0.22	<.01
CYP2A6	rs1801272	27861808	A	95.5	0.02	1.00
CYP2A6	rs28399433	30634332	A	98.2	0.08	.29
CYP2A6	hCV33845966	33845966	T	99.1	<0.01	1.00
CYP2A6	rs72547591	33845974	T	100	0.00	1.00
CYP2A6	rs28399454	34816076	C	95.5	0.01	1.00
CYP2B6	rs3745274	7817765	G	87.4	0.26	.71
CYP2B6	rs12721655	30634236	A	100	<0.01	1.00
CYP2B6	rs28399499	60732328	T	98.2	0.01	1.00
CYP2C19	rs12248560	469857	C	84.7	0.17	1.00
CYP2C19	rs3758580	1329163	C	97.3	0.11	.13
CYP2C19	rs17878459	11383554	G	100	0.02	1.00
CYP2C19	rs4244285	25986767	G	100	0.12	.62
CYP2C19	rs41291556	30634130	T	95.5	0.02	1.00
CYP2C19	rs17885098	30634133	T	97.3	0.03	1.00
CYP2C19	rs28399504	30634136	A	98.2	0.01	1.00
CYP2C19	rs17886522	34816135	A	99.1	0.01	1.00
CYP2C8	rs10509681	25625782	T	99.1	0.10	.05
CYP2C8	rs11572080	25625794	C	98.2	0.08	.59
CYP2C8	rs1058930	25761568	G	100	0.04	1.00
CYP2C8	rs11572103	30634034	T	99.1	0.01	1.00
CYP2C9	rs7900194	25625804	G	100	<0.01	1.00
CYP2C9	rs1799853	25625805	C	99.1	0.07	.52
CYP2C9	rs1057910	27104892	A	100	0.04	.52

(Continues)

TABLE 2 (Continued)

Gene Symbol	dbSNP ID	Position	Reference Allele[a]	Call Rate %	MAF[b]	HWE *p* Value
CYP2C9	rs28371686	27859817	C	97.3	0.01	.01
CYP2C9	rs9332239	30634125	C	99.1	0.02	.07
CYP2C9	rs28371685	30634132	C	99.1	0.01	1.00
CYP2C9	rs9332130	30634137	A	100	<0.01	1.00
CYP2C9	rs9332131	32287221	A	97.3	0.02	<.01
CYP2C9	hCV72649992	72649992	I/D	93.7	0.01	.02
CYP2C9	rs72558190	72649995	C	98.2	0.01	.01
CYP2D6	rs28371706	2222771	G	93.7	0.02	.08
CYP2D6	rs3892097	27102431	C	99.1	0.13	1.00
CYP2D6	rs5030862	27531552	C	96.4	0.02	.07
CYP2D6	rs72549349	32407228	C	98.2	0.01	<.01
CYP2D6	rs72549350	32407229	TCT	98.2	0.01	1.00
CYP2D6	rs35742686	32407232	T	96.4	0.03	<.01
CYP2D6	rs72549353	32407233	AGTT	98.2	0.01	1.00
CYP2D6	hCV32407240	32407240	–	100	0.02	<.01
CYP2D6	rs5030655	32407243	A	88.3	0.01	1.00
CYP2D6	rs72549346	72649935	–	94.6	0.04	<.01
CYP2D6	rs5030865	30634117	C	93.7	0.02	1.00
CYP2D6	rs5030865	30634118	C	93.7	0.05	1.00
CYP3A4	rs55785340	30634204	A	98.2	0.02	<.01
CYP3A5	rs10264272	30203950	C	99.1	0.01	1.00
CYP3A5	rs55965422	30633867	A	99.1	0.01	1.00
CYP3A5	rs41303343	32287188	–	99.1	0.01	1.00
DPYD	rs1801267	8393589	C	97.3	0.01	.01
DPYD	rs1801265	9491497	A	99.1	0.24	.20
DPYD	rs3918290	30633851	C	98.2	0.01	1.00
DPYD	hCV32287186	32287186	ATGA	100	0.01	1.00
GSTP1	rs1695	3237198	A	96.4	0.42	.69
NAT1	rs4986782	1204334	G	99.1	0.01	1.00
NAT1	rs5030839	27529440	C	100	0.00	1.00
NAT1	rs4986988	27530743	C	99.1	0.03	1.00
NAT1	rs55793712	30633845	A	100	0.01	1.00
NAT2	rs1208	572769	A	100	0.37	.63
NAT2	rs1799931	572770	G	98.2	0.08	.62
NAT2	rs1799930	1204091	G	95.5	0.26	.29
NAT2	rs1799929	1204092	C	99.1	0.34	.85
NAT2	rs1801280	1204093	T	94.6	0.37	.37
NAT2	rs1041983	8684085	C	100	0.31	1.00
SLC15A2	rs2293616	385910	G	100	0.39	.89
SLC15A2	rs2257212	385920	C	100	0.38	1.00
SLC15A2	rs1143671	385930	C	99.1	0.39	.78
SLC15A2	rs1143672	7504282	G	92.8	0.39	1.00
SLC22A1	rs628031	8709275	G	98.2	0.28	1.00
SLC22A1	rs2282143	15877554	C	100	0.04	1.00
SLC22A1	rs34059508	30634080	G	100	0.01	1.00
SLC22A1	rs55918055	30634094	T	100	0.01	1.00
SLC22A1	rs72552763	34211613	GAT	99.1	0.22	.01
SLC22A2	rs316019	3111809	C	97.3	0.08	1.00
SLC22A2	rs8177507	25621236	C	100	<0.01	1.00
SLC22A2	rs8177517	25621260	T	100	<0.01	1.00
SLC22A2	rs8177516	30633923	G	100	0.01	.07
SLCO1B1	rs2306283	1901697	A	95.5	0.42	.60
SLCO1B1	rs56061388	30633903	T	100	0.01	1.00
SLCO1B1	rs72559745	30633905	A	95.5	<0.01	1.00
SLCO1B1	rs4149056	30633906	T	99.1	0.11	.50
SLCO1B1	rs55737008	30633912	A	99.1	<0.01	1.00
SLCO1B3	rs4149117	25639181	G	96.4	0.19	.26
SLCO1B3	rs7311358	25765587	A	96.4	0.19	.30
SLCO2B1	rs2306168	16193013	C	98.2	0.09	.93
TPMT	rs1142345	19567	T	93.7	0.05	1.00
TPMT	rs56161402	19569	C	100	<0.01	1.00
TPMT	rs1800460	30634116	C	98.2	0.05	1.00
UGT1A1	rs4148323	559715	G	98.2	0.02	1.00
UGT1A1	rs4124874	1432134	T	100	0.48	.96
UGT2B15	rs1902023	27028164	C	97.3	0.42	.40
UGT2B7	rs7668258	27827970	C	91	0.36	.43
UGT2B7	rs7662029	30720663	G	92.8	0.36	.16
VKORC1	rs8050894	2847860	C	98.2	0.48	.77

[a]I/D = insertion/deletion variant.
[b]MAF = minor allele frequency

Wald chi-squared test of interaction less than 0.05. The gene variant *ABCC2* (rs3740066) was associated with an increased odds of spina bifida for 4 of the 5 pollutants: $OR_{NO} = 3.0$ (1.2, 7.7); $OR_{NO2} = 2.8$ (1.2, 6.7); $OR_{PM10} = 3.9$ (1.7, 8.9); $OR_{PM2.5} = 3.9$ (1.3, 11.7). Those with high exposure to NO_2 and variants of *SLC01B1* (rs4149056) were associated with increased odds of spina bifida (OR = 3.7; 1.2, 11.7). In addition, some results showed decreased risk among those with gene variants and low exposure (e.g., PM_{10} and *ABCB1* (rs203582); NO_2 and

TABLE 3 Gene-environment interactions for carbon monoxide (CO) associations with risk of spina bifida

			High carbon monoxide[a]			Low carbon monoxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
ABCB1	rs1045642	Variant	16	33	1.1 (0.4, 2.8)	21	62	0.7 (0.3, 1.7)
ABCB1	rs1045642	Wildtype	4	15	0.4 (0.1, 1.6)	15	28	Reference
ABCB1	rs1128503	Variant	15	33	1.1 (0.4, 2.9)	24	65	0.9 (0.4, 2.2)
ABCB1	rs1128503	Wildtype	5	15	0.7 (0.2, 2.4)	12	26	Reference
ABCB1	rs2032582	Variant	11	32	0.6 (0.2, 1.6)	17	58	0.6 (0.2, 1.3)
ABCB1	rs2032582	Wildtype	8	16	0.8 (0.3, 2.3)	19	32	Reference
ABCB1	rs2032582	Variant	0	4	NC	1	6	NC
ABCB1	rs2032582	Wildtype	14	34	0.9 (0.4, 2.1)	30	67	Reference
ABCC2	rs56199535	Variant	1	0	NC	2	1	NC
ABCC2	rs56199535	Wildtype	19	48	1.1 (0.5, 2.2)	32	89	Reference
ABCC2	rs717620	Variant	6	12	2.0 (0.6, 6.5)	16	26	2.2 (0.9, 5.2)
ABCC2	rs717620	Wildtype	14	35	1.2 (0.5, 2.7)	20	65	Reference
ABCC2	rs3740066	Variant	12	24	2.5 (0.9, 7.1)	26	43	**2.8 (1.2, 6.9)**
ABCC2	rs3740066	Wildtype	7	24	1.1 (0.4, 3.5)	10	47	Reference
ABCC2	rs2273697	Variant	3	16	0.4 (0.1, 1.6)	10	31	0.7 (0.3, 1.7)
ABCC2	rs2273697	Wildtype	17	31	1.2 (0.5, 2.7)	26	57	Reference
ABCG2	rs2231142	Variant	7	13	1.3 (0.4, 3.9)	7	22	0.7 (0.3, 1.9)
ABCG2	rs2231142	Wildtype	13	35	0.8 (0.4, 1.9)	29	69	Reference
CYP1A1	rs1799814	Variant	1	4	NC	4	4	1.9 (0.4, 9.0)
CYP1A1	rs1799814	Wildtype	19	44	1.1 (0.5, 2.3)	32	86	Reference
CYP1A1	rs1048943	Variant	8	16	0.9 (0.3, 2.7)	12	30	0.7 (0.3, 1.8)
CYP1A1	rs1048943	Wildtype	12	32	0.9 (0.4, 2.2)	24	61	Reference
CYP1A1	rs41279188	Variant	0	0	NC	1	1	NC
CYP1A1	rs41279188	Wildtype	20	48	1.0 (0.5, 2.1)	35	90	Reference
CYP1A2	rs762551	Variant	8	17	1.6 (0.5, 4.6)	21	43	1.5 (0.6, 3.4)
CYP1A2	rs762551	Wildtype	12	31	1.1 (0.4, 2.9)	15	48	Reference
CYP1A2	rs2069514	Variant	7	14	0.9 (0.3, 2.8)	13	32	0.8 (0.3, 2.0)
CYP1A2	rs2069514	Wildtype	12	30	1.3 (0.5, 3.1)	20	57	Reference
CYP2A6	rs1801272	Variant	1	1	NC	1	4	NC
CYP2A6	rs1801272	Wildtype	18	45	1.0 (0.5, 2.1)	33	86	Reference
CYP2A6	rs4986891	Variant	3	5	2.0 (0.4, 9.8)	3	7	1.2 (0.3, 5.5)
CYP2A6	rs4986891	Wildtype	16	38	1.2 (0.6, 2.6)	30	82	Reference
CYP2A6	rs28399433	Variant	3	8	0.7 (0.2, 2.9)	9	12	1.9 (0.7, 5.2)
CYP2A6	rs28399433	Wildtype	17	40	1.2 (0.6, 2.6)	27	77	Reference
CYP2A6	hCV33845966	Variant	0	0	NC	0	1	NC
CYP2A6	hCV33845966	Wildtype	19	48	1.0 (0.5, 2.0)	35	90	Reference
CYP2A6	rs28399454	Variant	0	1	NC	1	3	NC
CYP2A6	rs28399454	Wildtype	20	45	1.1 (0.6, 2.2)	34	87	Reference

(Continues)

TABLE 3 (Continued)

			High carbon monoxide[a]			Low carbon monoxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP2B6	rs3745274	Variant	9	17	1.2 (0.4, 3.5)	15	41	0.7 (0.3, 1.7)
CYP2B6	rs3745274	Wildtype	7	27	0.5 (0.2, 1.4)	19	41	Reference
CYP2B6	rs12721655	Variant	0	0	NC	0	1	NC
CYP2B6	rs12721655	Wildtype	20	47	1.1 (0.5, 2.1)	35	90	Reference
CYP2B6	rs28399499	Variant	0	1	NC	0	2	NC
CYP2B6	rs28399499	Wildtype	20	45	1.1 (0.5, 2.1)	36	87	Reference
CYP2C19	rs12248560	Variant	9	11	1.6 (0.5, 4.7)	6	25	0.5 (0.2, 1.4)
CYP2C19	rs12248560	Wildtype	6	29	0.4 (0.1, 1.0)	28	51	Reference
CYP2C19	rs3758580	Variant	2	7	NC	10	18	1.6 (0.6, 4.2)
CYP2C19	rs3758580	Wildtype	18	39	1.3 (0.6, 2.7)	25	72	Reference
CYP2C19	rs17878459	Variant	1	3	NC	1	3	NC
CYP2C19	rs17878459	Wildtype	19	45	1.0 (0.5, 2.1)	35	88	Reference
CYP2C19	rs4244285	Variant	2	7	NC	10	22	1.2 (0.5, 2.9)
CYP2C19	rs4244285	Wildtype	18	41	1.1 (0.5, 2.4)	26	69	Reference
CYP2C19	rs41291556	Variant	1	0	NC	2	4	NC
CYP2C19	rs41291556	Wildtype	15	42	0.9 (0.4, 1.9)	32	84	Reference
CYP2C19	rs17885098	Variant	2	2	NC	6	11	1.4 (0.5, 4.4)
CYP2C19	rs17885098	Wildtype	18	43	1.1 (0.5, 2.2)	30	78	Reference
CYP2C19	rs28399504	Variant	0	3	NC	1	1	NC
CYP2C19	rs28399504	Wildtype	18	42	1.3 (0.6, 2.6)	30	88	Reference
CYP2C19	rs17886522	Variant	1	1	NC	2	1	NC
CYP2C19	rs17886522	Wildtype	19	47	1.0 (0.5, 2.0)	34	89	Reference
CYP2C8	rs10509681	Variant	1	9	NC	7	15	1.1 (0.4, 3.2)
CYP2C8	rs10509681	Wildtype	19	38	1.2 (0.6, 2.5)	29	74	Reference
CYP2C8	rs11572080	Variant	1	9	NC	4	13	0.7 (0.2, 2.6)
CYP2C8	rs11572080	Wildtype	19	39	1.1 (0.5, 2.3)	32	75	Reference
CYP2C8	rs1058930	Variant	1	3	NC	1	7	NC
CYP2C8	rs1058930	Wildtype	19	45	1.0 (0.5, 2.0)	35	83	Reference
CYP2C8	rs11572103	Variant	0	0	NC	1	2	NC
CYP2C8	rs11572103	Wildtype	19	48	1.0 (0.5, 1.9)	35	88	Reference
CYP2C9	rs7900194	Variant	0	1	NC	0	0	NC
CYP2C9	rs7900194	Wildtype	20	47	1.1 (0.6, 2.2)	35	91	Reference
CYP2C9	rs1799853	Variant	1	8	NC	6	12	1.4 (0.4, 4.4)
CYP2C9	rs1799853	Wildtype	19	39	1.3 (0.6, 2.6)	30	79	Reference
CYP2C9	rs1057910	Variant	1	3	NC	3	10	0.9 (0.2, 3.7)
CYP2C9	rs1057910	Wildtype	19	45	1.1 (0.5, 2.2)	32	81	Reference
CYP2C9	rs28371686	Variant	0	0	NC	2	2	NC
CYP2C9	rs28371686	Wildtype	20	46	1.1 (0.6, 2.2)	34	88	Reference

(Continues)

TABLE 3 (Continued)

			High carbon monoxide[a]			Low carbon monoxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP2C9	rs9332239	Variant	0	3	NC	1	1	NC
CYP2C9	rs9332239	Wildtype	20	45	1.2 (0.6, 2.3)	34	89	Reference
CYP2C9	rs28371685	Variant	0	0	NC	1	0	NC
CYP2C9	rs28371685	Wildtype	20	45	1.2 (0.6, 2.4)	34	91	Reference
CYP2C9	rs9332130	Variant	0	0	NC	3	1	NC
CYP2C9	rs9332130	Wildtype	20	48	1.1 (0.5, 2.2)	33	90	Reference
CYP2C9	rs9332131	Variant	0	3	NC	5	1	NC
CYP2C9	rs9332131	Wildtype	20	45	1.3 (0.6, 2.5)	30	86	Reference
CYP2C9	hCV72649992	Variant	0	1	NC	2	0	NC
CYP2C9	hCV72649992	Wildtype	19	45	1.2 (0.6, 2.4)	31	86	Reference
CYP2C9	rs72558190	Variant	0	1	NC	1	1	NC
CYP2C9	rs72558190	Wildtype	20	47	1.1 (0.6, 2.2)	34	89	Reference
CYP2D6	rs28371706	Variant	0	2	NC	0	1	NC
CYP2D6	rs28371706	Wildtype	19	44	1.0 (0.5, 2.0)	36	86	Reference
CYP2D6	rs3892097	Variant	4	13	1.5 (0.4, 5.4)	12	19	2.4 (1.0, 6.1)
CYP2D6	rs3892097	Wildtype	16	32	1.5 (0.7, 3.4)	23	71	Reference
CYP2D6	rs5030862	Variant	0	3	NC	1	2	NC
CYP2D6	rs5030862	Wildtype	20	44	1.1 (0.6, 2.3)	35	87	Reference
CYP2D6	rs72549349	Variant	0	1	NC	1	0	NC
CYP2D6	rs72549349	Wildtype	20	47	1.0 (0.5, 2.1)	35	88	Reference
CYP2D6	rs72549350	Variant	1	2	NC	0	1	NC
CYP2D6	rs72549350	Wildtype	19	46	1.0 (0.5, 2.0)	34	87	Reference
CYP2D6	rs35742686	Variant	0	3	NC	2	4	NC
CYP2D6	rs35742686	Wildtype	19	43	1.2 (0.6, 2.4)	32	84	Reference
CYP2D6	rs72549353	Variant	0	3	NC	1	1	NC
CYP2D6	rs72549353	Wildtype	20	44	1.1 (0.6, 2.3)	35	88	Reference
CYP2D6	hCV32407240	Variant	0	2	NC	1	2	NC
CYP2D6	hCV32407240	Wildtype	20	43	1.2 (0.6, 2.4)	34	89	Reference
CYP2D6	rs5030655	Variant	0	1	NC	1	2	NC
CYP2D6	rs5030655	Wildtype	18	43	1.1 (0.5, 2.2)	32	81	Reference
CYP2D6	rs72549346	Variant	1	2	NC	2	4	NC
CYP2D6	rs72549346	Wildtype	18	44	1.0 (0.5, 2.0)	34	82	Reference
CYP2D6	rs5030865	Variant	1	0	NC	1	3	NC
CYP2D6	rs5030865	Wildtype	18	47	0.9 (0.4, 1.8)	35	81	Reference
CYP2D6	rs5030865	Variant	1	6	NC	5	8	1.4 (0.4, 4.9)
CYP2D6	rs5030865	Wildtype	19	39	1.3 (0.6, 2.6)	30	78	Reference
CYP3A4	rs55785340	Variant	0	1	NC	3	4	2.0 (0.4, 10.3)
CYP3A4	rs55785340	Wildtype	20	46	1.2 (0.6, 2.4)	31	87	Reference

(Continues)

TABLE 3 (Continued)

Gene symbol	dbSNP ID	Genotype	High carbon monoxide[a]			Low carbon monoxide[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP3A5	rs10264272	Variant	0	0	NC	1	3	NC
CYP3A5	rs10264272	Wildtype	20	47	1.0 (0.5, 2.0)	35	88	Reference
CYP3A5	rs55965422	Variant	1	1	NC	1	2	NC
CYP3A5	rs55965422	Wildtype	17	47	0.9 (0.5, 1.9)	33	88	Reference
CYP3A5	rs41303343	Variant	0	1	NC	1	2	NC
CYP3A5	rs41303343	Wildtype	19	47	1.0 (0.5, 2.0)	34	87	Reference
DPYD	rs1801267	Variant	0	1	NC	0	1	NC
DPYD	rs1801267	Wildtype	18	45	1.1 (0.5, 2.2)	34	88	Reference
DPYD	rs1801265	Variant	8	22	0.7 (0.2, 1.8)	11	39	0.6 (0.2, 1.3)
DPYD	rs1801265	Wildtype	12	26	1.0 (0.4, 2.3)	25	50	Reference
DPYD	rs3918290	Variant	0	0	NC	0	3	NC
DPYD	rs3918290	Wildtype	20	48	1.0 (0.5, 2.0)	36	88	Reference
DPYD	hCV32287186	Variant	1	0	NC	0	1	NC
DPYD	hCV32287186	Wildtype	19	47	1.0 (0.5, 1.9)	36	90	Reference
GSTP1	rs1695	Variant	14	29	0.7 (0.3, 1.9)	22	61	0.5 (0.2, 1.3)
GSTP1	rs1695	Wildtype	6	19	0.5 (0.2, 1.7)	14	28	Reference
NAT1	rs4986782	Variant	1	2	NC	1	1	NC
NAT1	rs4986782	Wildtype	19	46	1.0 (0.5, 2.0)	35	89	Reference
NAT1	rs4986988	Variant	2	2	NC	3	7	1.2 (0.3, 5.0)
NAT1	rs4986988	Wildtype	18	45	1.0 (0.5, 2.0)	33	84	Reference
NAT1	rs55793712	Variant	0	0	NC	1	2	NC
NAT1	rs55793712	Wildtype	20	48	1.0 (0.5, 2.1)	34	89	Reference
NAT2	rs1208	Variant	13	26	1.1 (0.4, 2.8)	18	57	0.6 (0.3, 1.4)
NAT2	rs1208	Wildtype	6	22	0.4 (0.1, 1.3)	18	34	Reference
NAT2	rs1799931	Variant	6	3	4.1 (0.9, 18.9)	9	16	1.5 (0.5, 4.1)
NAT2	rs1799931	Wildtype	14	42	0.9 (0.4, 1.9)	27	72	Reference
NAT2	rs1799930	Variant	11	17	1.7 (0.6, 4.5)	17	42	1.2 (0.5, 2.7)
NAT2	rs1799930	Wildtype	7	28	0.6 (0.2, 1.8)	17	45	Reference
NAT2	rs1799929	Variant	12	25	1.2 (0.5, 3.1)	18	54	0.7 (0.3, 1.7)
NAT2	rs1799929	Wildtype	7	23	0.5 (0.2, 1.6)	17	37	Reference
NAT2	rs1801280	Variant	13	24	1.2 (0.5, 3.3)	17	53	0.6 (0.3, 1.5)
NAT2	rs1801280	Wildtype	6	22	0.5 (0.1, 1.4)	16	33	Reference
NAT2	rs1041983	Variant	10	21	1.3 (0.5, 3.5)	22	51	1.3 (0.6, 3.1)
NAT2	rs1041983	Wildtype	9	27	1.0 (0.4, 2.9)	14	39	Reference
SLC15A2	rs2293616	Variant	16	29	1.3 (0.5, 3.4)	20	60	0.8 (0.4, 2.0)
SLC15A2	rs2293616	Wildtype	4	19	0.4 (0.1, 1.5)	16	31	Reference
SLC15A2	rs2257212	Variant	16	28	1.4 (0.5, 3.4)	19	59	0.8 (0.3, 1.8)
SLC15A2	rs2257212	Wildtype	4	20	0.4 (0.1, 1.3)	17	32	Reference

(Continues)

TABLE 3 (Continued)

			High carbon monoxide[a]			Low carbon monoxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
SLC15A2	rs1143671	Variant	16	29	1.3 (0.5, 3.4)	20	60	0.8 (0.4, 2.0)
SLC15A2	rs1143671	Wildtype	4	19	0.4 (0.1, 1.5)	16	31	Reference
SLC15A2	rs1143672	Variant	16	27	1.8 (0.7, 4.9)	19	59	0.9 (0.4, 2.2)
SLC15A2	rs1143672	Wildtype	4	17	0.5 (0.1, 1.7)	15	30	Reference
SLC22A1	rs628031	Variant	8	25	0.9 (0.3, 2.6)	15	42	1.0 (0.4, 2.3)
SLC22A1	rs628031	Wildtype	12	21	1.3 (0.5, 3.4)	19	46	Reference
SLC22A1	rs2282143	Variant	1	5	NC	2	8	NC
SLC22A1	rs2282143	Wildtype	19	43	1.0 (0.5, 2.1)	34	83	Reference
SLC22A1	rs34059508	Variant	1	2	NC	1	2	NC
SLC22A1	rs34059508	Wildtype	19	46	1.0 (0.5, 2.0)	35	89	Reference
SLC22A1	rs55918055	Variant	0	0	NC	2	2	NC
SLC22A1	rs55918055	Wildtype	20	48	1.0 (0.5, 2.1)	34	88	Reference
SLC22A1	rs72552763	Variant	8	15	1.4 (0.5, 3.9)	13	31	1.1 (0.5, 2.6)
SLC22A1	rs72552763	Wildtype	12	33	0.9 (0.4, 2.2)	23	60	Reference
SLC22A2	rs316019	Variant	5	10	1.7 (0.5, 5.5)	3	10	0.8 (0.2, 3.3)
SLC22A2	rs316019	Wildtype	15	35	1.0 (0.5, 2.1)	32	80	Reference
SLC22A2	rs8177507	Variant	0	0	NC	0	1	NC
SLC22A2	rs8177507	Wildtype	20	48	1.0 (0.5, 2.0)	36	90	Reference
SLC22A2	rs8177517	Variant	1	1	NC	0	0	NC
SLC22A2	rs8177517	Wildtype	19	47	1.0 (0.5, 2.0)	36	91	Reference
SLC22A2	rs8177516	Variant	0	1	NC	2	3	NC
SLC22A2	rs8177516	Wildtype	20	47	1.1 (0.5, 2.2)	33	88	Reference
SLCO1B1	rs2306283	Variant	16	36	1.6 (0.6, 4.3)	26	61	1.6 (0.6, 4.0)
SLCO1B1	rs2306283	Wildtype	4	10	1.2 (0.3, 5.1)	9	28	Reference
SLCO1B1	rs56061388	Variant	0	0	NC	0	1	NC
SLCO1B1	rs56061388	Wildtype	18	48	0.9 (0.5, 1.8)	36	90	Reference
SLCO1B1	rs72559745	Variant	0	0	NC	1	1	NC
SLCO1B1	rs72559745	Wildtype	19	48	1.0 (0.5, 2.0)	34	87	Reference
SLCO1B1	rs4149056	Variant	5	8	2.2 (0.6, 8.0)	7	21	0.8 (0.3, 2.3)
SLCO1B1	rs4149056	Wildtype	15	39	0.9 (0.4, 1.9)	28	69	Reference
SLCO1B1	rs55737008	Variant	0	0	NC	2	1	NC
SLCO1B1	rs55737008	Wildtype	20	48	1.1 (0.5, 2.1)	34	89	Reference
SLCO1B3	rs4149117	Variant	8	16	1.3 (0.5, 3.8)	16	32	1.3 (0.6, 2.9)
SLCO1B3	rs4149117	Wildtype	12	29	1.1 (0.5, 2.7)	20	57	Reference
SLCO1B3	rs7311358	Variant	8	16	1.3 (0.5, 3.7)	15	31	1.2 (0.5, 2.7)
SLCO1B3	rs7311358	Wildtype	12	32	0.9 (0.4, 2.2)	21	57	Reference
SLCO2B1	rs2306168	Variant	3	7	0.9 (0.2, 4.0)	8	17	1.1 (0.4, 2.9)
SLCO2B1	rs2306168	Wildtype	17	41	1.0 (0.5, 2.2)	28	73	Reference

(Continues)

TABLE 3 (Continued)

			High carbon monoxide[a]			Low carbon monoxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
TPMT	rs1142345	Variant	1	5	NC	4	12	0.8 (0.2, 2.9)
TPMT	rs1142345	Wildtype	17	42	0.9 (0.5, 2.0)	31	75	Reference
TPMT	rs56161402	Variant	0	0	NC	2	1	NC
TPMT	rs56161402	Wildtype	20	48	1.0 (0.5, 2.1)	34	89	Reference
TPMT	rs1800460	Variant	1	6	NC	6	9	2.0 (0.6, 6.4)
TPMT	rs1800460	Wildtype	19	42	1.2 (0.6, 2.6)	29	81	Reference
UGT1A1	rs4148323	Variant	1	2	NC	0	3	NC
UGT1A1	rs4148323	Wildtype	19	46	1.0 (0.5, 2.0)	36	87	Reference
UGT1A1	rs4124874	Variant	16	36	0.7 (0.3, 1.8)	23	68	0.5 (0.2, 1.2)
UGT1A1	rs4124874	Wildtype	3	12	0.3 (0.1, 1.5)	13	23	Reference
UGT2B15	rs1902023	Variant	15	29	1.0 (0.4, 2.4)	20	64	0.6 (0.2, 1.3)
UGT2B15	rs1902023	Wildtype	5	17	0.4 (0.1, 1.5)	15	26	Reference
UGT2B7	rs7668258	Variant	10	29	0.7 (0.3, 1.9)	13	47	0.6 (0.3, 1.5)
UGT2B7	rs7668258	Wildtype	7	14	1.0 (0.3, 3.0)	18	34	Reference
UGT2B7	rs7662029	Variant	11	29	0.9 (0.3, 2.3)	17	50	0.8 (0.4, 1.9)
UGT2B7	rs7662029	Wildtype	9	16	1.2 (0.4, 3.4)	18	37	Reference
VKORC1	rs8050894	Variant	14	32	0.7 (0.3, 1.8)	19	64	0.5 (0.2, 1.2)
VKORC1	rs8050894	Wildtype	5	16	0.5 (0.2, 1.8)	16	26	Reference

[a]Highest tertile cut-off = 0.730 ppm.
[b]Adjusted for maternal race, vitamin use, BMI, education, and smoking

TABLE 4 Gene-environment interactions for nitrogen oxide (NO) associations with risk of spina bifida

			High nitrogen oxide[a]			Low nitrogen oxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aORb (95% CI)
ABCB1	rs1045642	Variant	18	34	1.4 (0.6, 3.7)	25	66	1.0 (0.4, 2.4)
ABCB1	rs1045642	Wildtype	9	17	1.1 (0.4, 3.2)	12	28	Reference
ABCB1	rs1128503	Variant	19	37	1.8 (0.7, 4.7)	28	65	1.6 (0.6, 3.8)
ABCB1	rs1128503	Wildtype	8	15	1.7 (0.5, 5.5)	9	29	Reference
ABCB1	rs2032582	Variant	13	34	1.0 (0.4, 2.4)	22	59	0.9 (0.4, 2.2)
ABCB1	rs2032582	Wildtype	13	18	1.6 (0.6, 4.1)	15	34	Reference
ABCB1	rs2032582	Variant	0	2	NC	1	8	NC
ABCB1	rs2032582	Wildtype	21	39	1.2 (0.6, 2.5)	29	66	Reference
ABCC2	rs56199535	Variant	3	0	NC	0	1	NC
ABCC2	rs56199535	Wildtype	24	51	1.3 (0.7, 2.4)	35	93	Reference
ABCC2	rs717620	Variant	9	15	2.1 (0.8, 5.7)	15	26	2.0 (0.9, 4.7)
ABCC2	rs717620	Wildtype	18	36	1.5 (0.7, 3.2)	22	68	Reference
ABCC2	rs3740066	Variant	17	26	3.0 (1.2, 7.7)	26	45	**2.5 (1.1, 5.9)**
ABCC2	rs3740066	Wildtype	9	26	1.3 (0.4, 3.6)	11	48	Reference

(Continues)

TABLE 4 (Continued)

			High nitrogen oxide[a]			Low nitrogen oxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aORb (95% CI)
ABCC2	rs2273697	Variant	6	19	0.7 (0.2, 1.9)	11	30	0.9 (0.4, 2.1)
ABCC2	rs2273697	Wildtype	21	33	1.5 (0.7, 3.2)	26	60	Reference
ABCG2	rs2231142	Variant	8	15	1.2 (0.4, 3.4)	8	24	0.7 (0.3, 1.8)
ABCG2	rs2231142	Wildtype	19	37	1.2 (0.6, 2.4)	29	70	Reference
CYP1A1	rs1799814	Variant	2	4	NC	3	5	1.3 (0.3, 6.2)
CYP1A1	rs1799814	Wildtype	25	48	1.3 (0.7, 2.5)	34	88	Reference
CYP1A1	rs1048943	Variant	7	15	0.9 (0.3, 2.7)	14	32	0.9 (0.4, 2.2)
CYP1A1	rs1048943	Wildtype	20	37	1.4 (0.7, 3.0)	23	61	Reference
CYP1A1	rs41279188	Variant	1	0	NC	0	1	NC
CYP1A1	rs41279188	Wildtype	26	52	1.2 (0.6, 2.3)	37	93	Reference
CYP1A2	rs762551	Variant	14	22	2.2 (0.9, 5.5)	20	42	1.7 (0.7, 3.9)
CYP1A2	rs762551	Wildtype	13	30	1.5 (0.6, 3.6)	16	51	Reference
CYP1A2	rs2069514	Variant	7	15	0.9 (0.3, 2.8)	14	32	0.8 (0.3, 2.1)
CYP1A2	rs2069514	Wildtype	18	33	1.6 (0.7, 3.5)	20	58	Reference
CYP2A6	rs1801272	Variant	1	1	NC	1	4	NC
CYP2A6	rs1801272	Wildtype	24	49	1.2 (0.7, 2.4)	35	90	Reference
CYP2A6	rs4986891	Variant	3	5	2.0 (0.4, 9.4)	3	10	0.8 (0.2, 3.2)
CYP2A6	rs4986891	Wildtype	22	42	1.4 (0.7, 2.7)	32	82	Reference
CYP2A6	rs28399433	Variant	4	8	1.0 (0.3, 3.9)	9	14	1.5 (0.6, 4.1)
CYP2A6	rs28399433	Wildtype	23	44	1.4 (0.7, 2.8)	28	78	Reference
CYP2A6	hCV33845966	Variant	0	0	NC	0	1	NC
CYP2A6	hCV33845966	Wildtype	26	52	1.3 (0.7, 2.4)	36	93	Reference
CYP2A6	rs28399454	Variant	0	1	NC	1	3	NC
CYP2A6	rs28399454	Wildtype	26	49	1.3 (0.7, 2.5)	35	90	Reference
CYP2B6	rs3745274	Variant	13	20	1.7 (0.7, 4.2)	16	38	1.0 (0.4, 2.2)
CYP2B6	rs3745274	Wildtype	10	27	0.8 (0.3, 2.1)	19	47	Reference
CYP2B6	rs12721655	Variant	0	0	NC	0	1	NC
CYP2B6	rs12721655	Wildtype	26	51	1.3 (0.7, 2.4)	37	93	Reference
CYP2B6	rs28399499	Variant	0	2	NC	0	1	NC
CYP2B6	rs28399499	Wildtype	27	47	1.4 (0.7, 2.6)	37	92	Reference
CYP2C19	rs12248560	Variant	13	12	2.1 (0.8, 5.6)	4	28	0.3 (0.1, 0.8)
CYP2C19	rs12248560	Wildtype	10	31	0.5 (0.2, 1.2)	29	52	Reference
CYP2C19	rs3758580	Variant	4	8	1.5 (0.4, 6.0)	10	15	2.2 (0.8, 5.6)
CYP2C19	rs3758580	Wildtype	23	42	1.6 (0.8, 3.3)	26	78	Reference
CYP2C19	rs17878459	Variant	1	3	NC	1	2	NC
CYP2C19	rs17878459	Wildtype	26	49	1.3 (0.7, 2.5)	36	92	Reference
CYP2C19	rs4244285	Variant	4	8	1.3 (0.3, 5.0)	10	19	1.5 (0.6, 3.7)
CYP2C19	rs4244285	Wildtype	23	44	1.4 (0.7, 2.9)	27	75	Reference

(Continues)

TABLE 4 (Continued)

			High nitrogen oxide[a]			Low nitrogen oxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aORb (95% CI)
CYP2C19	rs41291556	Variant	3	0	NC	1	4	NC
CYP2C19	rs41291556	Wildtype	20	46	1.1 (0.5, 2.1)	33	85	Reference
CYP2C19	rs17885098	Variant	3	2	NC	5	10	1.3 (0.4, 4.3)
CYP2C19	rs17885098	Wildtype	24	47	1.3 (0.7, 2.5)	32	82	Reference
CYP2C19	rs28399504	Variant	0	3	NC	1	1	NC
CYP2C19	rs28399504	Wildtype	24	47	1.4 (0.7, 2.8)	32	89	Reference
CYP2C19	rs17886522	Variant	1	1	NC	2	1	NC
CYP2C19	rs17886522	Wildtype	26	51	1.3 (0.7, 2.4)	35	92	Reference
CYP2C8	rs10509681	Variant	6	11	1.1 (0.4, 3.5)	2	15	NC
CYP2C8	rs10509681	Wildtype	21	38	1.3 (0.6, 2.5)	35	79	Reference
CYP2C8	rs11572080	Variant	3	11	0.6 (0.1, 2.3)	2	13	NC
CYP2C8	rs11572080	Wildtype	24	39	1.4 (0.7, 2.8)	35	80	Reference
CYP2C8	rs1058930	Variant	1	3	NC	2	7	NC
CYP2C8	rs1058930	Wildtype	26	48	1.3 (0.7, 2.5)	35	87	Reference
CYP2C8	rs11572103	Variant	0	0	NC	1	2	NC
CYP2C8	rs11572103	Wildtype	26	52	1.2 (0.7, 2.3)	36	91	Reference
CYP2C9	rs7900194	Variant	0	1	NC	0	0	NC
CYP2C9	rs7900194	Wildtype	26	51	1.3 (0.7, 2.4)	37	94	Reference
CYP2C9	rs1799853	Variant	4	10	0.9 (0.2, 3.2)	3	12	0.7 (0.2, 2.8)
CYP2C9	rs1799853	Wildtype	23	42	1.3 (0.7, 2.6)	34	82	Reference
CYP2C9	rs1057910	Variant	1	3	NC	3	9	1.0 (0.2, 4.1)
CYP2C9	rs1057910	Wildtype	26	49	1.4 (0.7, 2.6)	33	85	Reference
CYP2C9	rs28371686	Variant	0	0	NC	2	2	NC
CYP2C9	rs28371686	Wildtype	27	50	1.4 (0.7, 2.6)	35	91	Reference
CYP2C9	rs9332239	Variant	0	3	NC	1	1	NC
CYP2C9	rs9332239	Wildtype	26	49	1.3 (0.7, 2.5)	36	91	Reference
CYP2C9	rs28371685	Variant	0	0	NC	1	0	NC
CYP2C9	rs28371685	Wildtype	27	49	1.5 (0.8, 2.8)	35	94	Reference
CYP2C9	rs9332130	Variant	3	0	NC	0	1	NC
CYP2C9	rs9332130	Wildtype	24	52	1.1 (0.6, 2.2)	37	93	Reference
CYP2C9	rs9332131	Variant	2	3	NC	3	1	NC
CYP2C9	rs9332131	Wildtype	24	49	1.3 (0.7, 2.5)	34	88	Reference
CYP2C9	hCV72649992	Variant	2	1	NC	1	0	NC
CYP2C9	hCV72649992	Wildtype	23	49	1.2 (0.6, 2.4)	34	88	Reference
CYP2C9	rs72558190	Variant	0	1	NC	1	1	NC
CYP2C9	rs72558190	Wildtype	27	51	1.4 (0.7, 2.6)	35	92	Reference
CYP2D6	rs28371706	Variant	0	2	NC	1	1	NC
CYP2D6	rs28371706	Wildtype	26	47	1.3 (0.7, 2.5)	36	90	Reference

(Continues)

TABLE 4 (Continued)

			High nitrogen oxide[a]			Low nitrogen oxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aORb (95% CI)
CYP2D6	rs3892097	Variant	4	13	1.5 (0.4, 5.3)	15	21	**3.4 (1.4, 8.4)**
CYP2D6	rs3892097	Wildtype	23	36	**2.4 (1.1, 5.1)**	21	72	Reference
CYP2D6	rs5030862	Variant	0	3	NC	1	2	NC
CYP2D6	rs5030862	Wildtype	27	47	1.5 (0.8, 2.7)	36	92	Reference
CYP2D6	rs72549349	Variant	0	1	NC	1	0	NC
CYP2D6	rs72549349	Wildtype	27	51	1.3 (0.7, 2.5)	36	91	Reference
CYP2D6	rs72549350	Variant	2	1	NC	1	2	NC
CYP2D6	rs72549350	Wildtype	23	51	1.1 (0.6, 2.1)	36	90	Reference
CYP2D6	rs35742686	Variant	3	3	2.6 (0.5, 14.6)	0	4	NC
CYP2D6	rs35742686	Wildtype	23	47	1.2 (0.6, 2.4)	35	87	Reference
CYP2D6	rs72549353	Variant	0	3	NC	1	1	NC
CYP2D6	rs72549353	Wildtype	27	47	1.4 (0.8, 2.7)	36	91	Reference
CYP2D6	hCV32407240	Variant	0	2	NC	1	2	NC
CYP2D6	hCV32407240	Wildtype	27	47	1.5 (0.8, 2.8)	35	92	Reference
CYP2D6	rs5030655	Variant	0	1	NC	1	2	NC
CYP2D6	rs5030655	Wildtype	22	46	1.2 (0.6, 2.3)	35	86	Reference
CYP2D6	rs72549346	Variant	4	2	NC	0	4	NC
CYP2D6	rs72549346	Wildtype	22	47	1.1 (0.6, 2.1)	37	86	Reference
CYP2D6	rs5030865	Variant	3	0	NC	0	3	NC
CYP2D6	rs5030865	Wildtype	23	50	1.0 (0.5, 2.0)	37	84	Reference
CYP2D6	rs5030865	Variant	2	7	NC	5	7	2.1 (0.6, 7.6)
CYP2D6	rs5030865	Wildtype	23	41	1.6 (0.8, 3.2)	31	82	Reference
CYP3A4	rs55785340	Variant	1	1	NC	2	4	NC
CYP3A4	rs55785340	Wildtype	24	50	1.2 (0.6, 2.3)	34	89	Reference
CYP3A5	rs10264272	Variant	1	1	NC	0	3	NC
CYP3A5	rs10264272	Wildtype	26	50	1.2 (0.7, 2.3)	37	91	Reference
CYP3A5	rs55965422	Variant	1	1	NC	1	2	NC
CYP3A5	rs55965422	Wildtype	22	51	1.0 (0.5, 2.0)	36	90	Reference
CYP3A5	rs41303343	Variant	0	1	NC	1	1	NC
CYP3A5	rs41303343	Wildtype	26	51	1.3 (0.7, 2.4)	35	91	Reference
DPYD	rs1801267	Variant	0	1	NC	0	1	NC
DPYD	rs1801267	Wildtype	25	49	1.4 (0.7, 2.6)	35	91	Reference
DPYD	rs1801265	Variant	12	25	1.0 (0.4, 2.3)	11	39	0.6 (0.2, 1.3)
DPYD	rs1801265	Wildtype	15	26	1.2 (0.5, 2.6)	26	54	Reference
DPYD	rs3918290	Variant	0	0	NC	0	3	NC
DPYD	rs3918290	Wildtype	27	52	1.3 (0.7, 2.4)	37	91	Reference
DPYD	hCV32287186	Variant	1	0	NC	0	1	NC
DPYD	hCV32287186	Wildtype	26	51	1.3 (0.7, 2.4)	37	93	Reference

(Continues)

TABLE 4 (Continued)

			High nitrogen oxide[a]			Low nitrogen oxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aORb (95% CI)
GSTP1	rs1695	Variant	19	31	1.0 (0.4, 2.5)	23	66	0.6 (0.2, 1.3)
GSTP1	rs1695	Wildtype	8	20	0.7 (0.2, 2.1)	14	28	Reference
NAT1	rs4986782	Variant	0	1	NC	2	2	NC
NAT1	rs4986782	Wildtype	27	51	1.4 (0.7, 2.6)	35	91	Reference
NAT1	rs4986988	Variant	4	2	NC	1	8	NC
NAT1	rs4986988	Wildtype	23	49	1.1 (0.6, 2.1)	36	86	Reference
NAT1	rs55793712	Variant	1	0	NC	0	2	NC
NAT1	rs55793712	Wildtype	25	52	1.2 (0.6, 2.2)	37	92	Reference
NAT2	rs1208	Variant	17	28	1.5 (0.6, 3.6)	19	59	0.8 (0.3, 1.7)
NAT2	rs1208	Wildtype	9	24	0.7 (0.2, 1.8)	17	34	Reference
NAT2	rs1799931	Variant	8	6	2.6 (0.8, 9.1)	7	17	0.9 (0.3, 2.4)
NAT2	rs1799931	Wildtype	19	44	1.0 (0.5, 2.0)	30	72	Reference
NAT2	rs1799930	Variant	14	21	1.7 (0.7, 4.0)	17	42	1.0 (0.5, 2.3)
NAT2	rs1799930	Wildtype	10	28	0.8 (0.3, 2.1)	19	47	Reference
NAT2	rs1799929	Variant	16	26	1.8 (0.7, 4.4)	19	56	0.9 (0.4, 2.1)
NAT2	rs1799929	Wildtype	10	26	0.8 (0.3, 2.1)	16	37	Reference
NAT2	rs1801280	Variant	17	26	1.6 (0.6, 3.9)	18	55	0.7 (0.3, 1.7)
NAT2	rs1801280	Wildtype	8	25	0.6 (0.2, 1.6)	16	32	Reference
NAT2	rs1041983	Variant	14	28	1.2 (0.5, 2.9)	20	51	1.0 (0.5, 2.2)
NAT2	rs1041983	Wildtype	12	24	1.3 (0.5, 3.2)	17	42	Reference
SLC15A2	rs2293616	Variant	17	33	1.8 (0.7, 4.8)	25	61	1.5 (0.6, 3.5)
SLC15A2	rs2293616	Wildtype	10	19	1.4 (0.5, 4.1)	12	33	Reference
SLC15A2	rs2257212	Variant	17	32	1.8 (0.7, 4.5)	24	60	1.3 (0.6, 3.1)
SLC15A2	rs2257212	Wildtype	10	20	1.2 (0.4, 3.5)	13	34	Reference
SLC15A2	rs1143671	Variant	17	33	1.8 (0.7, 4.8)	25	61	1.5 (0.6, 3.5)
SLC15A2	rs1143671	Wildtype	10	19	1.4 (0.5, 4.1)	12	33	Reference
SLC15A2	rs1143672	Variant	17	31	2.5 (0.9, 6.7)	24	59	1.7 (0.7, 4.2)
SLC15A2	rs1143672	Wildtype	10	17	1.7 (0.6, 5.1)	11	32	Reference
SLC22A1	rs628031	Variant	11	28	1.3 (0.5, 3.2)	17	40	1.4 (0.6, 3.1)
SLC22A1	rs628031	Wildtype	14	20	1.9 (0.8, 4.6)	19	53	Reference
SLC22A1	rs2282143	Variant	1	4	NC	2	9	NC
SLC22A1	rs2282143	Wildtype	26	48	1.3 (0.7, 2.4)	35	85	Reference
SLC22A1	rs34059508	Variant	0	3	NC	2	1	NC
SLC22A1	rs34059508	Wildtype	27	49	1.4 (0.8, 2.7)	35	93	Reference
SLC22A1	rs55918055	Variant	2	0	NC	0	2	NC
SLC22A1	rs55918055	Wildtype	25	51	1.2 (0.6, 2.2)	37	92	Reference
SLC22A1	rs72552763	Variant	10	16	1.6 (0.6, 4.4)	15	34	1.2 (0.5, 2.7)
SLC22A1	rs72552763	Wildtype	17	36	1.3 (0.6, 2.8)	22	60	Reference

(Continues)

TABLE 4 (Continued)

			High nitrogen oxide[a]			Low nitrogen oxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aORb (95% CI)
SLC22A2	rs316019	Variant	4	9	1.4 (0.4, 5.1)	4	10	1.3 (0.4, 5.0)
SLC22A2	rs316019	Wildtype	23	40	1.5 (0.7, 3.0)	32	83	Reference
SLC22A2	rs8177507	Variant	0	0	NC	0	1	NC
SLC22A2	rs8177507	Wildtype	27	52	1.3 (0.7, 2.4)	37	93	Reference
SLC22A2	rs8177517	Variant	1	1	NC	0	0	NC
SLC22A2	rs8177517	Wildtype	26	51	1.3 (0.7, 2.4)	37	94	Reference
SLC22A2	rs8177516	Variant	2	1	NC	0	3	NC
SLC22A2	rs8177516	Wildtype	24	51	1.1 (0.6, 2.1)	37	91	Reference
SLCO1B1	rs2306283	Variant	19	37	2.1 (0.8, 5.5)	28	61	1.9 (0.8, 4.6)
SLCO1B1	rs2306283	Wildtype	7	12	2.0 (0.5, 7.0)	9	32	Reference
SLCO1B1	rs56061388	Variant	0	0	NC	0	1	NC
SLCO1B1	rs56061388	Wildtype	25	52	1.2 (0.6, 2.2)	37	93	Reference
SLCO1B1	rs72559745	Variant	1	0	NC	0	1	NC
SLCO1B1	rs72559745	Wildtype	23	52	1.0 (0.5, 2.0)	37	90	Reference
SLCO1B1	rs4149056	Variant	7	7	2.7 (0.8, 8.9)	8	26	0.9 (0.3, 2.2)
SLCO1B1	rs4149056	Wildtype	20	44	1.1 (0.5, 2.2)	28	67	Reference
SLCO1B1	rs55737008	Variant	2	0	NC	0	1	NC
SLCO1B1	rs55737008	Wildtype	25	52	1.2 (0.6, 2.2)	37	92	Reference
SLCO1B3	rs4149117	Variant	11	17	1.8 (0.7, 4.6)	16	31	1.4 (0.6, 3.2)
SLCO1B3	rs4149117	Wildtype	16	31	1.5 (0.6, 3.3)	21	62	Reference
SLCO1B3	rs7311358	Variant	11	17	1.8 (0.7, 4.6)	15	30	1.3 (0.6, 2.9)
SLCO1B3	rs7311358	Wildtype	16	33	1.3 (0.6, 3.0)	22	63	Reference
SLCO2B1	rs2306168	Variant	7	9	1.5 (0.5, 4.7)	5	17	0.6 (0.2, 1.9)
SLCO2B1	rs2306168	Wildtype	20	43	1.1 (0.5, 2.2)	32	75	Reference
TPMT	rs1142345	Variant	1	4	NC	6	14	1.0 (0.3, 3.0)
TPMT	rs1142345	Wildtype	22	46	1.1 (0.6, 2.2)	31	77	Reference
TPMT	rs56161402	Variant	1	0	NC	1	1	NC
TPMT	rs56161402	Wildtype	26	51	1.3 (0.7, 2.4)	36	93	Reference
TPMT	rs1800460	Variant	3	5	1.5 (0.3, 7.1)	6	11	1.5 (0.5, 4.5)
TPMT	rs1800460	Wildtype	23	47	1.3 (0.7, 2.5)	31	82	Reference
UGT1A1	rs4148323	Variant	1	2	NC	0	3	NC
UGT1A1	rs4148323	Wildtype	26	50	1.2 (0.7, 2.3)	37	90	Reference
UGT1A1	rs4124874	Variant	21	39	1.1 (0.5, 2.7)	25	67	0.8 (0.3, 1.8)
UGT1A1	rs4124874	Wildtype	5	13	0.8 (0.2, 2.8)	12	27	Reference
UGT2B15	rs1902023	Variant	19	29	1.4 (0.6, 3.5)	21	65	0.7 (0.3, 1.5)
UGT2B15	rs1902023	Wildtype	8	21	0.6 (0.2, 1.9)	14	27	Reference
UGT2B7	rs7668258	Variant	11	31	0.9 (0.3, 2.3)	17	47	0.9 (0.4, 2.0)
UGT2B7	rs7668258	Wildtype	12	17	1.6 (0.6, 4.2)	16	36	Reference

(Continues)

TABLE 4 (Continued)

			High nitrogen oxide[a]			Low nitrogen oxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aORb (95% CI)
UGT2B7	rs7662029	Variant	13	31	1.2 (0.5, 3.0)	20	51	1.2 (0.5, 2.7)
UGT2B7	rs7662029	Wildtype	14	17	2.3 (0.9, 6.0)	16	41	Reference
VKORC1	rs8050894	Variant	15	36	0.8 (0.3, 2.0)	22	67	0.7 (0.3, 1.5)
VKORC1	rs8050894	Wildtype	11	15	1.5 (0.5, 4.1)	14	27	Reference

[a]Highest tertile cut-off = 15.145 ppb.
[b]Adjusted for maternal race, vitamin use, BMI, education, and smoking.

TABLE 5 Gene-environment interactions for nitrogen dioxide (NO_2) associations with risk of spina bifida

			High nitrogen dioxide[a]			Low nitrogen dioxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
ABCB1	rs1045642	Variant	21	45	1.5 (0.7, 3.6)	30	77	1.2 (0.6, 2.7)
ABCB1	rs1045642	Wildtype	9	16	1.4 (0.5, 4.1)	14	38	Reference
ABCB1	rs1128503	Variant	21	44	1.7 (0.7, 4.2)	33	81	1.5 (0.6, 3.4)
ABCB1	rs1128503	Wildtype	9	18	1.7 (0.6, 5.2)	11	35	Reference
ABCB1	rs2032582	Variant	15	41	1.1 (0.5, 2.7)	25	72	1.1 (0.5, 2.4)
ABCB1	rs2032582	Wildtype	15	20	2.1 (0.8, 5.2)	16	44	Reference
ABCB1	rs2032582	Variant	0	5	NC	1	8	NC
ABCB1	rs2032582	Wildtype	23	45	1.3 (0.6, 2.4)	35	85	Reference
ABCC2	rs56199535	Variant	3	0	NC	1	1	NC
ABCC2	rs56199535	Wildtype	26	62	1.2 (0.7, 2.2)	42	113	Reference
ABCC2	rs717620	Variant	10	16	2.2 (0.9, 5.7)	15	33	1.3 (0.6, 2.9)
ABCC2	rs717620	Wildtype	20	45	1.2 (0.6, 2.4)	29	83	Reference
ABCC2	rs3740066	Variant	20	30	**2.8 (1.2, 6.7)**	31	62	1.9 (0.9, 4.1)
ABCC2	rs3740066	Wildtype	9	31	1.0 (0.4, 2.8)	13	53	Reference
ABCC2	rs2273697	Variant	7	21	0.8 (0.3, 2.2)	14	34	1.1 (0.5, 2.4)
ABCC2	rs2273697	Wildtype	23	40	1.6 (0.8, 3.2)	30	79	Reference
ABCG2	rs2231142	Variant	8	16	1.2 (0.4, 3.2)	10	31	0.7 (0.3, 1.7)
ABCG2	rs2231142	Wildtype	22	46	1.2 (0.6, 2.4)	34	85	Reference
CYP1A1	rs1799814	Variant	2	3	NC	3	6	1.2 (0.3, 5.4)
CYP1A1	rs1799814	Wildtype	28	59	1.3 (0.7, 2.5)	39	109	Reference
CYP1A1	rs1048943	Variant	14	19	1.3 (0.6, 3.2)	12	36	0.6 (0.3, 1.4)
CYP1A1	rs1048943	Wildtype	16	43	1.0 (0.5, 2.0)	32	78	Reference
CYP1A1	rs41279188	Variant	0	1	NC	1	0	NC
CYP1A1	rs41279188	Wildtype	30	61	1.3 (0.7, 2.4)	42	114	Reference
CYP1A2	rs762551	Variant	13	23	2.3 (0.9, 5.9)	27	59	1.9 (0.9, 4.0)
CYP1A2	rs762551	Wildtype	17	39	1.6 (0.7, 3.8)	16	55	Reference
CYP1A2	rs2069514	Variant	12	17	1.3 (0.5, 3.2)	13	38	0.7 (0.3, 1.6)
CYP1A2	rs2069514	Wildtype	16	41	1.2 (0.6, 2.7)	26	72	Reference

(Continues)

TABLE 5 (Continued)

			High nitrogen dioxide[a]			Low nitrogen dioxide[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP2A6	rs1801272	Variant	1	3	NC	1	3	NC
CYP2A6	rs1801272	Wildtype	27	57	1.3 (0.7, 2.4)	41	110	Reference
CYP2A6	rs4986891	Variant	3	4	2.9 (0.6, 14.7)	4	16	0.7 (0.2, 2.3)
CYP2A6	rs4986891	Wildtype	25	52	1.3 (0.7, 2.6)	36	98	Reference
CYP2A6	rs28399433	Variant	8	9	2.0 (0.7, 5.9)	9	17	1.2 (0.5, 3.0)
CYP2A6	rs28399433	Wildtype	22	52	1.2 (0.6, 2.3)	35	98	Reference
CYP2A6	hCV33845966	Variant	0	1	NC	0	0	NC
CYP2A6	hCV33845966	Wildtype	28	61	1.2 (0.7, 2.2)	44	115	Reference
CYP2A6	rs28399454	Variant	0	2	NC	1	2	NC
CYP2A6	rs28399454	Wildtype	29	58	1.4 (0.8, 2.5)	41	112	Reference
CYP2B6	rs3745274	Variant	14	22	1.9 (0.8, 4.7)	22	52	1.1 (0.5, 2.3)
CYP2B6	rs3745274	Wildtype	13	33	1.0 (0.4, 2.3)	18	52	Reference
CYP2B6	rs12721655	Variant	0	0	NC	1	1	NC
CYP2B6	rs12721655	Wildtype	29	61	1.3 (0.7, 2.3)	43	114	Reference
CYP2B6	rs28399499	Variant	0	1	NC	0	2	NC
CYP2B6	rs28399499	Wildtype	30	58	1.3 (0.7, 2.4)	44	112	Reference
CYP2C19	rs12248560	Variant	11	15	1.5 (0.6, 3.9)	8	33	0.5 (0.2, 1.3)
CYP2C19	rs12248560	Wildtype	16	39	0.8 (0.4, 1.7)	31	65	Reference
CYP2C19	rs3758580	Variant	6	8	2.4 (0.7, 8.2)	11	23	1.4 (0.6, 3.3)
CYP2C19	rs3758580	Wildtype	24	52	1.4 (0.7, 2.7)	31	91	Reference
CYP2C19	rs17878459	Variant	2	2	NC	1	6	NC
CYP2C19	rs17878459	Wildtype	28	60	1.2 (0.7, 2.2)	43	110	Reference
CYP2C19	rs4244285	Variant	6	9	1.7 (0.5, 5.7)	11	27	1.1 (0.5, 2.5)
CYP2C19	rs4244285	Wildtype	24	53	1.2 (0.7, 2.4)	33	89	Reference
CYP2C19	rs41291556	Variant	2	1	NC	3	5	1.6 (0.3, 7.3)
CYP2C19	rs41291556	Wildtype	26	54	1.4 (0.8, 2.7)	35	104	Reference
CYP2C19	rs17885098	Variant	5	3	4.0 (0.9, 18.2)	4	10	1.0 (0.3, 3.7)
CYP2C19	rs17885098	Wildtype	25	56	1.1 (0.6, 2.2)	40	102	Reference
CYP2C19	rs28399504	Variant	0	3	NC	1	1	NC
CYP2C19	rs28399504	Wildtype	26	57	1.3 (0.7, 2.5)	39	110	Reference
CYP2C19	rs17886522	Variant	2	0	NC	1	2	NC
CYP2C19	rs17886522	Wildtype	28	61	1.2 (0.7, 2.2)	43	114	Reference
CYP2C8	rs10509681	Variant	4	11	0.9 (0.3, 3.3)	7	18	1.2 (0.4, 3.2)
CYP2C8	rs10509681	Wildtype	26	49	1.4 (0.8, 2.7)	37	97	Reference
CYP2C8	rs11572080	Variant	2	11	NC	5	16	1.0 (0.3, 3.1)
CYP2C8	rs11572080	Wildtype	28	50	1.5 (0.8, 2.7)	39	98	Reference
CYP2C8	rs1058930	Variant	2	3	NC	2	11	NC
CYP2C8	rs1058930	Wildtype	28	58	1.3 (0.7, 2.3)	42	105	Reference

(Continues)

TABLE 5 (Continued)

Gene symbol	dbSNP ID	Genotype	High nitrogen dioxide[a]			Low nitrogen dioxide[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP2C8	rs11572103	Variant	0	2	NC	1	1	NC
CYP2C8	rs11572103	Wildtype	29	60	1.3 (0.7, 2.4)	43	113	Reference
CYP2C9	rs7900194	Variant	0	1	NC	0	0	NC
CYP2C9	rs7900194	Wildtype	29	61	1.3 (0.7, 2.3)	44	116	Reference
CYP2C9	rs1799853	Variant	3	10	0.8 (0.2, 3.4)	6	14	1.6 (0.5, 4.6)
CYP2C9	rs1799853	Wildtype	27	51	1.5 (0.8, 2.8)	38	102	Reference
CYP2C9	rs1057910	Variant	2	4	NC	2	11	NC
CYP2C9	rs1057910	Wildtype	28	58	1.3 (0.7, 2.4)	41	105	Reference
CYP2C9	rs28371686	Variant	0	0	NC	2	2	NC
CYP2C9	rs28371686	Wildtype	30	60	1.4 (0.8, 2.5)	42	113	Reference
CYP2C9	rs9332239	Variant	0	3	NC	1	1	NC
CYP2C9	rs9332239	Wildtype	29	59	1.3 (0.7, 2.4)	42	112	Reference
CYP2C9	rs28371685	Variant	0	0	NC	1	2	NC
CYP2C9	rs28371685	Wildtype	30	60	1.4 (0.8, 2.5)	42	113	Reference
CYP2C9	rs9332130	Variant	2	0	NC	4	1	NC
CYP2C9	rs9332130	Wildtype	28	62	1.3 (0.7, 2.4)	40	115	Reference
CYP2C9	rs9332131	Variant	1	3	NC	5	1	NC
CYP2C9	rs9332131	Wildtype	29	59	1.5 (0.8, 2.8)	37	109	Reference
CYP2C9	hCV72649992	Variant	1	1	NC	3	1	NC
CYP2C9	hCV72649992	Wildtype	28	59	1.5 (0.8, 2.7)	36	109	Reference
CYP2C9	rs72558190	Variant	0	1	NC	1	1	NC
CYP2C9	rs72558190	Wildtype	30	61	1.4 (0.8, 2.5)	42	114	Reference
CYP2D6	rs28371706	Variant	0	3	NC	1	1	NC
CYP2D6	rs28371706	Wildtype	29	56	1.3 (0.7, 2.5)	43	110	Reference
CYP2D6	rs3892097	Variant	7	15	2.2 (0.8, 6.5)	14	27	2.0 (0.9, 4.6)
CYP2D6	rs3892097	Wildtype	23	44	1.6 (0.8, 3.3)	29	88	Reference
CYP2D6	rs5030862	Variant	0	3	NC	1	2	NC
CYP2D6	rs5030862	Wildtype	30	58	1.4 (0.8, 2.5)	43	112	Reference
CYP2D6	rs72549349	Variant	0	1	NC	1	0	NC
CYP2D6	rs72549349	Wildtype	30	60	1.4 (0.8, 2.5)	42	114	Reference
CYP2D6	rs72549350	Variant	2	0	NC	1	5	NC
CYP2D6	rs72549350	Wildtype	26	61	1.1 (0.6, 2.0)	43	109	Reference
CYP2D6	rs35742686	Variant	2	3	NC	2	6	NC
CYP2D6	rs35742686	Wildtype	27	56	1.4 (0.7, 2.5)	40	108	Reference
CYP2D6	rs72549353	Variant	0	3	NC	1	1	NC
CYP2D6	rs72549353	Wildtype	30	58	1.4 (0.8, 2.5)	43	112	Reference
CYP2D6	hCV32407240	Variant	0	2	NC	1	2	NC
CYP2D6	hCV32407240	Wildtype	30	57	1.5 (0.8, 2.7)	42	113	Reference

(Continues)

TABLE 5 (Continued)

Gene symbol	dbSNP ID	Genotype	High nitrogen dioxide[a]			Low nitrogen dioxide[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP2D6	rs5030655	Variant	0	1	NC	1	2	NC
CYP2D6	rs5030655	Wildtype	26	56	1.3 (0.7, 2.3)	41	106	Reference
CYP2D6	rs72549346	Variant	3	3	2.6 (0.5, 14.1)	3	5	1.4 (0.3, 6.6)
CYP2D6	rs72549346	Wildtype	27	56	1.3 (0.7, 2.5)	39	106	Reference
CYP2D6	rs5030865	Variant	2	0	NC	2	5	NC
CYP2D6	rs5030865	Wildtype	28	59	1.2 (0.7, 2.2)	41	103	Reference
CYP2D6	rs5030865	Variant	2	6	NC	7	12	1.5 (0.5, 4.1)
CYP2D6	rs5030865	Wildtype	26	53	1.3 (0.7, 2.5)	36	96	Reference
CYP3A4	rs55785340	Variant	0	1	NC	3	4	2.0 (0.4, 10.0)
CYP3A4	rs55785340	Wildtype	28	60	1.3 (0.7, 2.4)	40	111	Reference
CYP3A5	rs10264272	Variant	1	1	NC	1	3	NC
CYP3A5	rs10264272	Wildtype	29	60	1.3 (0.7, 2.3)	43	113	Reference
CYP3A5	rs55965422	Variant	1	2	NC	2	2	NC
CYP3A5	rs55965422	Wildtype	25	60	1.1 (0.6, 2.1)	41	112	Reference
CYP3A5	rs41303343	Variant	1	2	NC	0	1	NC
CYP3A5	rs41303343	Wildtype	29	60	1.4 (0.7, 2.5)	42	113	Reference
DPYD	rs1801267	Variant	0	2	NC	0	0	NC
DPYD	rs1801267	Wildtype	28	58	1.4 (0.8, 2.6)	42	113	Reference
DPYD	rs1801265	Variant	12	29	0.9 (0.4, 2.1)	14	46	0.7 (0.3, 1.5)
DPYD	rs1801265	Wildtype	18	33	1.3 (0.6, 2.8)	30	68	Reference
DPYD	rs3918290	Variant	0	0	NC	0	3	NC
DPYD	rs3918290	Wildtype	30	62	1.3 (0.7, 2.2)	44	111	Reference
DPYD	hCV32287186	Variant	1	1	NC	0	1	NC
DPYD	hCV32287186	Wildtype	29	60	1.3 (0.7, 2.3)	44	115	Reference
GSTP1	rs1695	Variant	22	37	1.0 (0.4, 2.3)	28	82	0.6 (0.3, 1.3)
GSTP1	rs1695	Wildtype	8	25	0.7 (0.2, 1.9)	16	32	Reference
NAT1	rs4986782	Variant	2	1	NC	0	3	NC
NAT1	rs4986782	Wildtype	28	61	1.2 (0.6, 2.1)	44	112	Reference
NAT1	rs4986988	Variant	3	3	3.0 (0.5, 16.7)	3	8	1.3 (0.3, 5.2)
NAT1	rs4986988	Wildtype	27	58	1.3 (0.7, 2.3)	41	108	Reference
NAT1	rs55793712	Variant	0	0	NC	3	2	NC
NAT1	rs55793712	Wildtype	29	62	1.3 (0.7, 2.3)	41	114	Reference
NAT2	rs1208	Variant	17	32	1.7 (0.7, 4.1)	26	70	1.1 (0.5, 2.3)
NAT2	rs1208	Wildtype	12	30	0.9 (0.4, 2.4)	17	44	Reference
NAT2	rs1799931	Variant	7	5	2.9 (0.8, 10.5)	11	21	1.3 (0.6, 3.2)
NAT2	rs1799931	Wildtype	23	55	1.2 (0.6, 2.2)	33	89	Reference
NAT2	rs1799930	Variant	17	27	1.8 (0.8, 4.1)	19	49	1.1 (0.5, 2.3)
NAT2	rs1799930	Wildtype	13	32	1.1 (0.5, 2.5)	21	60	Reference

(Continues)

TABLE 5 (Continued)

Gene symbol	dbSNP ID	Genotype	High nitrogen dioxide[a]			Low nitrogen dioxide[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
NAT2	rs1799929	Variant	16	31	2.0 (0.8, 4.7)	26	65	1.3 (0.6, 2.7)
NAT2	rs1799929	Wildtype	12	31	1.0 (0.4, 2.4)	17	49	Reference
NAT2	rs1801280	Variant	17	29	2.1 (0.9, 5.0)	22	65	1.0 (0.5, 2.2)
NAT2	rs1801280	Wildtype	11	30	0.9 (0.3, 2.2)	16	43	Reference
NAT2	rs1041983	Variant	16	30	1.4 (0.6, 3.1)	23	65	0.9 (0.4, 1.9)
NAT2	rs1041983	Wildtype	13	32	1.1 (0.5, 2.6)	20	50	Reference
SLC15A2	rs2293616	Variant	22	37	1.8 (0.8, 4.1)	25	74	1.0 (0.5, 2.1)
SLC15A2	rs2293616	Wildtype	8	25	0.7 (0.3, 2.0)	18	42	Reference
SLC15A2	rs2257212	Variant	22	36	1.7 (0.8, 3.9)	24	73	0.9 (0.4, 1.8)
SLC15A2	rs2257212	Wildtype	8	26	0.6 (0.2, 1.7)	20	43	Reference
SLC15A2	rs1143671	Variant	22	37	1.7 (0.7, 3.9)	25	72	1.0 (0.5, 2.0)
SLC15A2	rs1143671	Wildtype	8	25	0.7 (0.3, 1.9)	19	42	Reference
SLC15A2	rs1143672	Variant	22	37	1.9 (0.8, 4.5)	24	70	1.0 (0.5, 2.2)
SLC15A2	rs1143672	Wildtype	8	22	0.8 (0.3, 2.3)	18	41	Reference
SLC22A1	rs628031	Variant	17	33	1.5 (0.7, 3.2)	16	51	1.0 (0.5, 2.1)
SLC22A1	rs628031	Wildtype	11	26	1.0 (0.4, 2.5)	27	63	Reference
SLC22A1	rs2282143	Variant	1	4	NC	2	10	NC
SLC22A1	rs2282143	Wildtype	29	58	1.2 (0.7, 2.3)	42	106	Reference
SLC22A1	rs34059508	Variant	1	3	NC	1	1	NC
SLC22A1	rs34059508	Wildtype	29	59	1.3 (0.7, 2.4)	42	115	Reference
SLC22A1	rs55918055	Variant	1	0	NC	3	2	NC
SLC22A1	rs55918055	Wildtype	29	61	1.3 (0.7, 2.4)	41	113	Reference
SLC22A1	rs72552763	Variant	13	24	1.7 (0.7, 4.0)	18	40	1.2 (0.6, 2.5)
SLC22A1	rs72552763	Wildtype	17	38	1.3 (0.6, 2.7)	25	75	Reference
SLC22A2	rs316019	Variant	4	11	1.2 (0.3, 4.4)	5	17	0.9 (0.3, 2.5)
SLC22A2	rs316019	Wildtype	26	48	1.4 (0.7, 2.7)	38	98	Reference
SLC22A2	rs8177507	Variant	0	0	NC	0	1	NC
SLC22A2	rs8177507	Wildtype	30	62	1.3 (0.7, 2.3)	44	115	Reference
SLC22A2	rs8177517	Variant	1	1	NC	0	0	NC
SLC22A2	rs8177517	Wildtype	29	61	1.3 (0.7, 2.3)	44	116	Reference
SLC22A2	rs8177516	Variant	1	1	NC	3	3	2.4 (0.4, 13.1)
SLC22A2	rs8177516	Wildtype	28	61	1.3 (0.7, 2.3)	41	113	Reference
SLCO1B1	rs2306283	Variant	21	41	2.3 (0.9, 5.7)	32	77	1.7 (0.7, 3.9)
SLCO1B1	rs2306283	Wildtype	9	19	1.4 (0.5, 4.4)	10	35	Reference
SLCO1B1	rs56061388	Variant	0	1	NC	0	1	NC
SLCO1B1	rs56061388	Wildtype	28	61	1.2 (0.7, 2.2)	44	115	Reference
SLCO1B1	rs72559745	Variant	1	0	NC	1	1	NC
SLCO1B1	rs72559745	Wildtype	27	61	1.2 (0.6, 2.1)	42	111	Reference

(Continues)

TABLE 5 (Continued)

Gene symbol	dbSNP ID	Genotype	High nitrogen dioxide[a]			Low nitrogen dioxide[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
SLCO1B1	rs4149056	Variant	8	8	**3.7 (1.2, 11.7)**	10	33	0.8 (0.4, 1.9)
SLCO1B1	rs4149056	Wildtype	22	53	1.0 (0.5, 2.0)	33	82	Reference
SLCO1B1	rs55737008	Variant	1	1	NC	3	0	NC
SLCO1B1	rs55737008	Wildtype	29	61	1.4 (0.8, 2.5)	40	115	Reference
SLCO1B3	rs4149117	Variant	14	25	1.6 (0.7, 3.7)	16	34	1.2 (0.5, 2.6)
SLCO1B3	rs4149117	Wildtype	16	34	1.3 (0.6, 2.9)	27	79	Reference
SLCO1B3	rs7311358	Variant	14	24	1.5 (0.7, 3.5)	15	34	1.0 (0.5, 2.2)
SLCO1B3	rs7311358	Wildtype	16	37	1.1 (0.5, 2.4)	29	78	Reference
SLCO2B1	rs2306168	Variant	5	7	1.6 (0.5, 5.8)	9	21	1.0 (0.4, 2.5)
SLCO2B1	rs2306168	Wildtype	25	55	1.2 (0.6, 2.3)	35	92	Reference
TPMT	rs1142345	Variant	2	7	NC	5	13	1.0 (0.3, 3.2)
TPMT	rs1142345	Wildtype	26	53	1.3 (0.7, 2.5)	36	98	Reference
TPMT	rs56161402	Variant	1	0	NC	2	1	NC
TPMT	rs56161402	Wildtype	29	61	1.3 (0.7, 2.4)	42	115	Reference
TPMT	rs1800460	Variant	3	7	1.3 (0.3, 5.7)	8	11	2.1 (0.8, 5.9)
TPMT	rs1800460	Wildtype	26	55	1.4 (0.7, 2.5)	36	103	Reference
UGT1A1	rs4148323	Variant	1	1	NC	0	5	NC
UGT1A1	rs4148323	Wildtype	29	61	1.2 (0.7, 2.2)	44	109	Reference
UGT1A1	rs4124874	Variant	21	45	1.3 (0.5, 3.2)	33	84	1.1 (0.5, 2.5)
UGT1A1	rs4124874	Wildtype	8	17	1.4 (0.4, 4.2)	11	31	Reference
UGT2B15	rs1902023	Variant	20	35	1.1 (0.5, 2.6)	24	84	0.5 (0.2, 1.1)
UGT2B15	rs1902023	Wildtype	10	26	0.6 (0.2, 1.6)	18	28	Reference
UGT2B7	rs7668258	Variant	15	39	0.9 (0.4, 2.1)	15	53	0.6 (0.3, 1.4)
UGT2B7	rs7668258	Wildtype	12	18	1.4 (0.5, 3.4)	23	46	Reference
UGT2B7	rs7662029	Variant	17	39	1.1 (0.5, 2.4)	18	60	0.7 (0.3, 1.6)
UGT2B7	rs7662029	Wildtype	13	20	1.4 (0.6, 3.4)	25	51	Reference
VKORC1	rs8050894	Variant	15	42	1.0 (0.4, 2.5)	31	82	1.0 (0.5, 2.3)
VKORC1	rs8050894	Wildtype	14	20	2.0 (0.7, 5.3)	12	32	Reference

[a]Highest tertile cut-off = 20.145 ppb.
[b]Adjusted for maternal race, vitamin use, BMI, education, and smoking.

$PM_{2.5}$ and *UGT2B15* (rs1902023); NO and *CYP2C19* (rs12248560)). No statistically significant interactions were observed between variants and CO and risk of spina bifida.

The interaction of PM_{10} and several variants of CYP, NAT, and SLC genes resulted in increased odds of spina bifida (Table 6). The variants included *CYP1A2* (rs762551), *CYP2B6* (rs3745274), *CYP2C19* (rs3758580), *CYP2D6* (rs3892097), *NAT2* (rs1208, rs1799931, rs1799930, rs1799929, rs1801280), *SLC01B1* (rs2306283), *SLC01B3* (rs4149117), and ORs ranged from 2.4 to 4.0. Similar to PM_{10}, high exposure to $PM_{2.5}$ and several variants of CYP and SLC genes were associated with increased odds of spina bifida (Table 7). The statistically significant ORs ranged from 3.2 to 7.4 for the following gene variants, *CYP1A2* (rs762551), *CYP2B6* (rs3745274), *CYP2C19* (rs3758580, rs4244285), *SLC01B1* (rs2306283, rs4149056), *SLC01B3* (rs4149117), *SLC15A2* (rs1143672), and *SLC22A1* (rs628031).

When stratified by maternal folic acid containing vitamin use during the 1 month prior to conception through the first 2 months of pregnancy, one statistically significant result was revealed among the non-vitamin users. Exposure to NO and a variant of *CYP1A2* (rs762551) was associated with a fivefold increased risk of spina bifida (OR = 5.2, 95%CI: 1.2–23.5). Conversely, among vitamin users with high exposure to NO and a variant of *CYP1A2*

TABLE 6 Gene-environment interactions for particulate matter <10 microns (PM_{10}) associations with risk of spina bifida

			High particulate matter <10 microns[a]			Low particulate matter <10 microns[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
ABCB1	rs1045642	Variant	29	43	2.0 (0.8, 4.6)	24	78	0.9 (0.4, 2.0)
ABCB1	rs1045642	Wildtype	8	19	1.0 (0.4, 3.0)	14	33	Reference
ABCB1	rs1128503	Variant	27	38	2.2 (0.9, 5.5)	28	85	1.0 (0.4, 2.5)
ABCB1	rs1128503	Wildtype	10	24	1.2 (0.4, 3.6)	10	28	Reference
ABCB1	rs2032582	Variant	23	36	1.4 (0.6, 3.1)	18	76	0.5 (0.2, 1.2)
ABCB1	rs2032582	Wildtype	13	26	1.0 (0.4, 2.4)	19	36	Reference
ABCB1	rs2032582	Variant	1	6	NC	1	7	NC
ABCB1	rs2032582	Wildtype	27	42	1.7 (0.9, 3.3)	32	85	Reference
ABCC2	rs56199535	Variant	0	1	NC	4	0	NC
ABCC2	rs56199535	Wildtype	35	61	1.9 (1.0, 3.3)	34	111	Reference
ABCC2	rs717620	Variant	13	21	2.3 (1.0, 5.5)	12	28	1.5 (0.6, 3.5)
ABCC2	rs717620	Wildtype	24	41	1.8 (0.9, 3.7)	26	84	Reference
ABCC2	rs3740066	Variant	27	33	**3.9 (1.7, 8.9)**	26	55	**2.5 (1.1, 5.7)**
ABCC2	rs3740066	Wildtype	9	28	1.8 (0.6, 4.9)	12	57	Reference
ABCC2	rs2273697	Variant	9	18	1.5 (0.6, 3.8)	11	36	0.8 (0.3, 1.8)
ABCC2	rs2273697	Wildtype	28	42	1.8 (0.9, 3.5)	27	75	Reference
ABCG2	rs2231142	Variant	10	15	1.6 (0.6, 4.2)	7	31	0.5 (0.2, 1.3)
ABCG2	rs2231142	Wildtype	27	47	1.5 (0.8, 2.9)	31	82	Reference
CYP1A1	rs1799814	Variant	2	3	NC	3	7	1.0 (0.2, 4.4)
CYP1A1	rs1799814	Wildtype	35	59	1.9 (1.0, 3.4)	33	105	Reference
CYP1A1	rs1048943	Variant	15	15	2.0 (0.8, 5.0)	13	41	0.7 (0.3, 1.5)
CYP1A1	rs1048943	Wildtype	22	46	1.4 (0.7, 2.7)	25	71	Reference
CYP1A1	rs41279188	Variant	0	0	NC	1	1	NC
CYP1A1	rs41279188	Wildtype	36	60	1.9 (1.0, 3.3)	37	112	Reference
CYP1A2	rs762551	Variant	18	28	**3.1 (1.3, 7.2)**	22	50	2.2 (1.0, 4.8)
CYP1A2	rs762551	Wildtype	19	33	**2.5 (1.1, 5.9)**	15	62	Reference
CYP1A2	rs2069514	Variant	15	14	2.0 (0.8, 5.3)	12	42	0.6 (0.2, 1.4)
CYP1A2	rs2069514	Wildtype	20	45	1.3 (0.6, 2.8)	21	64	Reference
CYP2A6	rs1801272	Variant	0	3	NC	2	3	NC
CYP2A6	rs1801272	Wildtype	37	57	**2.2 (1.2, 4.0)**	32	107	Reference
CYP2A6	rs4986891	Variant	3	11	0.9 (0.2, 3.8)	4	9	1.8 (0.5, 6.6)
CYP2A6	rs4986891	Wildtype	33	49	**2.4 (1.2, 4.4)**	29	97	Reference
CYP2A6	rs28399433	Variant	9	9	2.4 (0.8, 6.9)	8	18	1.1 (0.4, 2.9)
CYP2A6	rs28399433	Wildtype	28	53	1.7 (0.9, 3.2)	30	93	Reference
CYP2A6	hCV33845966	Variant	0	0	NC	0	1	NC
CYP2A6	hCV33845966	Wildtype	36	62	1.8 (1.0, 3.1)	37	111	Reference
CYP2A6	rs28399454	Variant	0	1	NC	1	3	NC
CYP2A6	rs28399454	Wildtype	36	60	1.9 (1.0, 3.3)	35	107	Reference

(Continues)

TABLE 6 (Continued)

Gene symbol	dbSNP ID	Genotype	High particulate matter <10 microns[a]			Low particulate matter <10 microns[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP2B6	rs3745274	Variant	19	24	**2.8 (1.2, 6.7)**	18	49	1.3 (0.6, 2.9)
CYP2B6	rs3745274	Wildtype	17	32	1.9 (0.8, 4.5)	14	51	Reference
CYP2B6	rs12721655	Variant	0	0	NC	1	1	NC
CYP2B6	rs12721655	Wildtype	37	61	**1.9 (1.1, 3.3)**	36	111	Reference
CYP2B6	rs28399499	Variant	0	0	NC	0	3	NC
CYP2B6	rs28399499	Wildtype	37	59	1.8 (1.0, 3.1)	38	108	Reference
CYP2C19	rs12248560	Variant	7	14	1.4 (0.5, 4.1)	12	34	0.8 (0.3, 1.9)
CYP2C19	rs12248560	Wildtype	22	40	1.2 (0.6, 2.5)	25	61	Reference
CYP2C19	rs3758580	Variant	7	6	**4.0 (1.1, 13.7)**	10	24	1.5 (0.6, 3.7)
CYP2C19	rs3758580	Wildtype	30	55	1.9 (1.0, 3.6)	26	86	Reference
CYP2C19	rs17878459	Variant	2	3	NC	1	5	NC
CYP2C19	rs17878459	Wildtype	35	59	1.8 (1.0, 3.2)	37	108	Reference
CYP2C19	rs4244285	Variant	7	8	2.3 (0.7, 7.4)	10	27	1.2 (0.5, 3.0)
CYP2C19	rs4244285	Wildtype	30	54	1.8 (1.0, 3.5)	28	86	Reference
CYP2C19	rs41291556	Variant	2	0	NC	3	6	1.3 (0.3, 5.8)
CYP2C19	rs41291556	Wildtype	32	57	1.9 (1.0, 3.4)	29	98	Reference
CYP2C19	rs17885098	Variant	3	2	NC	6	11	1.8 (0.6, 5.5)
CYP2C19	rs17885098	Wildtype	34	57	1.9 (1.0, 3.5)	32	98	Reference
CYP2C19	rs28399504	Variant	1	1	NC	0	3	NC
CYP2C19	rs28399504	Wildtype	33	55	**1.9 (1.1, 3.5)**	33	109	Reference
CYP2C19	rs17886522	Variant	2	0	NC	1	2	NC
CYP2C19	rs17886522	Wildtype	35	62	1.7 (0.9, 3.0)	37	110	Reference
CYP2C8	rs10509681	Variant	2	14	NC	9	15	2.1 (0.8, 5.7)
CYP2C8	rs10509681	Wildtype	35	47	**2.5 (1.3, 4.6)**	29	96	Reference
CYP2C8	rs11572080	Variant	2	13	NC	5	14	1.2 (0.4, 3.8)
CYP2C8	rs11572080	Wildtype	35	47	**2.2 (1.2, 4.0)**	33	98	Reference
CYP2C8	rs1058930	Variant	2	6	NC	2	8	NC
CYP2C8	rs1058930	Wildtype	35	55	1.9 (1.0, 3.4)	36	105	Reference
CYP2C8	rs11572103	Variant	0	1	NC	1	2	NC
CYP2C8	rs11572103	Wildtype	37	61	1.9 (1.0, 3.3)	36	109	Reference
CYP2C9	rs7900194	Variant	0	1	NC	0	0	NC
CYP2C9	rs7900194	Wildtype	37	61	**1.9 (1.1, 3.4)**	37	113	Reference
CYP2C9	rs1799853	Variant	3	12	0.9 (0.2, 3.8)	6	12	1.8 (0.6, 5.6)
CYP2C9	rs1799853	Wildtype	34	49	**2.2 (1.2, 4.0)**	32	101	Reference
CYP2C9	rs1057910	Variant	1	3	NC	3	12	0.9 (0.2, 3.6)
CYP2C9	rs1057910	Wildtype	36	59	1.9 (1.0, 3.4)	34	101	Reference
CYP2C9	rs28371686	Variant	1	0	NC	1	2	NC
CYP2C9	rs28371686	Wildtype	36	61	1.8 (1.0, 3.1)	37	109	Reference

(Continues)

TABLE 6 (Continued)

			High particulate matter <10 microns[a]			Low particulate matter <10 microns[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP2C9	rs9332239	Variant	1	0	NC	0	4	NC
CYP2C9	rs9332239	Wildtype	36	61	1.8 (1.0, 3.1)	36	107	Reference
CYP2C9	rs28371685	Variant	0	1	NC	1	1	NC
CYP2C9	rs28371685	Wildtype	37	61	**1.9 (1.1, 3.4)**	36	109	Reference
CYP2C9	rs9332130	Variant	0	0	NC	6	1	NC
CYP2C9	rs9332130	Wildtype	37	62	**2.1 (1.2, 3.8)**	32	112	Reference
CYP2C9	rs9332131	Variant	2	0	NC	4	4	3.0 (0.7, 12.9)
CYP2C9	rs9332131	Wildtype	35	61	1.9 (1.0, 3.4)	32	104	Reference
CYP2C9	hCV72649992	Variant	2	0	NC	2	2	NC
CYP2C9	hCV72649992	Wildtype	35	61	**2.0 (1.1, 3.6)**	29	104	Reference
CYP2C9	rs72558190	Variant	0	0	NC	1	2	NC
CYP2C9	rs72558190	Wildtype	37	62	1.8 (1.0, 3.3)	36	110	Reference
CYP2D6	rs28371706	Variant	1	1	NC	0	3	NC
CYP2D6	rs28371706	Wildtype	35	59	1.6 (0.9, 2.9)	38	104	Reference
CYP2D6	rs3892097	Variant	9	11	**3.8 (1.3, 11.2)**	11	31	1.5 (0.6, 3.5)
CYP2D6	rs3892097	Wildtype	28	49	1.9 (0.9, 3.6)	26	80	Reference
CYP2D6	rs5030862	Variant	1	0	NC	0	5	NC
CYP2D6	rs5030862	Wildtype	36	61	1.7 (0.9, 2.9)	38	105	Reference
CYP2D6	rs72549349	Variant	1	0	NC	0	1	NC
CYP2D6	rs72549349	Wildtype	36	62	1.7 (1.0, 3.1)	37	109	Reference
CYP2D6	rs72549350	Variant	3	1	NC	0	4	NC
CYP2D6	rs72549350	Wildtype	34	61	1.6 (0.9, 2.9)	36	106	Reference
CYP2D6	rs35742686	Variant	1	0	NC	3	9	1.0 (0.2, 3.9)
CYP2D6	rs35742686	Wildtype	35	61	1.7 (1.0, 3.1)	33	100	Reference
CYP2D6	rs72549353	Variant	1	0	NC	0	4	NC
CYP2D6	rs72549353	Wildtype	36	60	1.7 (0.9, 3.0)	38	107	Reference
CYP2D6	hCV32407240	Variant	1	0	NC	0	4	NC
CYP2D6	hCV32407240	Wildtype	36	61	1.7 (1.0, 3.1)	37	106	Reference
CYP2D6	rs5030655	Variant	1	0	NC	0	3	NC
CYP2D6	rs5030655	Wildtype	34	60	1.7 (0.9, 3.1)	34	99	Reference
CYP2D6	rs72549346	Variant	1	0	NC	5	8	1.8 (0.5, 6.1)
CYP2D6	rs72549346	Wildtype	35	60	1.9 (1.0, 3.4)	31	99	Reference
CYP2D6	rs5030865	Variant	1	0	NC	3	5	1.3 (0.3, 6.1)
CYP2D6	rs5030865	Wildtype	35	58	1.7 (0.9, 3.1)	35	101	Reference
CYP2D6	rs5030865	Variant	3	4	2.6 (0.5, 13.4)	6	14	1.1 (0.4, 3.1)
CYP2D6	rs5030865	Wildtype	33	55	1.8 (0.9, 3.3)	30	91	Reference
CYP3A4	rs55785340	Variant	1	0	NC	2	5	NC
CYP3A4	rs55785340	Wildtype	35	61	1.8 (1.0, 3.2)	34	107	Reference

(Continues)

TABLE 6 (Continued)

Gene symbol	dbSNP ID	Genotype	High particulate matter <10 microns[a]			Low particulate matter <10 microns[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP3A5	rs10264272	Variant	0	1	NC	2	3	NC
CYP3A5	rs10264272	Wildtype	37	60	**1.9 (1.1, 3.4)**	36	110	Reference
CYP3A5	rs55965422	Variant	1	0	NC	2	4	NC
CYP3A5	rs55965422	Wildtype	34	61	1.8 (1.0, 3.2)	33	108	Reference
CYP3A5	rs41303343	Variant	1	1	NC	0	2	NC
CYP3A5	rs41303343	Wildtype	35	61	1.7 (1.0, 3.1)	37	109	Reference
DPYD	rs1801267	Variant	0	0	NC	0	2	NC
DPYD	rs1801267	Wildtype	35	62	1.7 (0.9, 3.0)	36	106	Reference
DPYD	rs1801265	Variant	14	28	1.4 (0.6, 3.1)	14	45	0.8 (0.4, 1.8)
DPYD	rs1801265	Wildtype	23	34	1.8 (0.9, 3.8)	24	66	Reference
DPYD	rs3918290	Variant	0	1	NC	0	2	NC
DPYD	rs3918290	Wildtype	37	60	1.8 (1.0, 3.3)	38	110	Reference
DPYD	hCV32287186	Variant	0	0	NC	1	2	NC
DPYD	hCV32287186	Wildtype	37	62	1.8 (1.0, 3.2)	37	110	Reference
GSTP1	rs1695	Variant	24	39	1.6 (0.6, 3.8)	27	77	0.9 (0.4, 2.1)
GSTP1	rs1695	Wildtype	13	23	1.7 (0.6, 4.7)	11	34	Reference
NAT1	rs4986782	Variant	2	2	NC	0	2	NC
NAT1	rs4986782	Wildtype	35	60	1.7 (0.9, 3.0)	38	110	Reference
NAT1	rs4986988	Variant	1	5	NC	5	6	3.3 (0.9, 12.3)
NAT1	rs4986988	Wildtype	36	57	**2.0 (1.1, 3.7)**	33	106	Reference
NAT1	rs55793712	Variant	0	1	NC	3	1	NC
NAT1	rs55793712	Wildtype	37	61	**2.1 (1.2, 3.7)**	34	112	Reference
NAT2	rs1208	Variant	22	32	**2.6 (1.1, 6.2)**	22	69	1.3 (0.6, 2.9)
NAT2	rs1208	Wildtype	15	29	1.8 (0.7, 4.5)	14	43	Reference
NAT2	rs1799931	Variant	10	9	2.9 (1.0, 8.2)	9	17	1.5 (0.6, 3.8)
NAT2	rs1799931	Wildtype	27	52	1.6 (0.9, 3.1)	29	89	Reference
NAT2	rs1799930	Variant	19	25	2.4 (1.0, 5.5)	17	50	1.0 (0.5, 2.1)
NAT2	rs1799930	Wildtype	16	34	1.3 (0.6, 2.9)	19	56	Reference
NAT2	rs1799929	Variant	21	30	**3.0 (1.3, 7.2)**	22	65	1.5 (0.7, 3.4)
NAT2	rs1799929	Wildtype	15	31	1.8 (0.7, 4.5)	14	47	Reference
NAT2	rs1801280	Variant	21	28	**3.3 (1.4, 8.0)**	19	65	1.4 (0.6, 3.2)
NAT2	rs1801280	Wildtype	15	27	2.2 (0.9, 5.6)	12	44	Reference
NAT2	rs1041983	Variant	22	32	1.9 (0.8, 4.1)	17	62	0.7 (0.3, 1.6)
NAT2	rs1041983	Wildtype	15	30	1.3 (0.6, 2.9)	19	50	Reference
SLC15A2	rs2293616	Variant	27	36	2.1 (0.9, 4.6)	19	74	0.7 (0.3, 1.4)
SLC15A2	rs2293616	Wildtype	10	26	0.8 (0.3, 2.0)	18	39	Reference
SLC15A2	rs2257212	Variant	26	36	2.0 (0.9, 4.3)	19	72	0.7 (0.3, 1.5)
SLC15A2	rs2257212	Wildtype	11	26	0.9 (0.3, 2.2)	19	41	Reference

(Continues)

TABLE 6 (Continued)

Gene symbol	dbSNP ID	Genotype	High particulate matter <10 microns[a]			Low particulate matter <10 microns[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
SLC15A2	rs1143671	Variant	27	36	2.0 (0.9, 4.4)	19	72	0.6 (0.3, 1.4)
SLC15A2	rs1143671	Wildtype	10	26	0.7 (0.3, 1.9)	19	39	Reference
SLC15A2	rs1143672	Variant	26	34	2.1 (0.9, 4.8)	19	72	0.7 (0.3, 1.5)
SLC15A2	rs1143672	Wildtype	10	25	0.7 (0.3, 1.9)	18	36	Reference
SLC22A1	rs628031	Variant	17	27	2.0 (0.9, 4.7)	16	56	0.9 (0.4, 2.1)
SLC22A1	rs628031	Wildtype	18	34	1.4 (0.6, 3.1)	21	53	Reference
SLC22A1	rs2282143	Variant	2	5	NC	1	9	NC
SLC22A1	rs2282143	Wildtype	35	57	1.7 (1.0, 3.1)	37	104	Reference
SLC22A1	rs34059508	Variant	1	3	NC	1	1	NC
SLC22A1	rs34059508	Wildtype	36	59	1.9 (1.0, 3.3)	36	112	Reference
SLC22A1	rs55918055	Variant	0	1	NC	4	1	NC
SLC22A1	rs55918055	Wildtype	37	61	**2.0 (1.1, 3.7)**	34	110	Reference
SLC22A1	rs72552763	Variant	18	27	2.0 (0.9, 4.5)	13	37	1.0 (0.4, 2.3)
SLC22A1	rs72552763	Wildtype	19	35	1.7 (0.8, 3.5)	24	75	Reference
SLC22A2	rs316019	Variant	6	8	2.3 (0.7, 7.6)	3	17	0.6 (0.2, 2.3)
SLC22A2	rs316019	Wildtype	30	53	1.5 (0.8, 2.8)	35	93	Reference
SLC22A2	rs8177507	Variant	0	1	NC	0	0	NC
SLC22A2	rs8177507	Wildtype	37	61	1.8 (1.0, 3.2)	38	113	Reference
SLC22A2	rs8177517	Variant	1	0	NC	0	1	NC
SLC22A2	rs8177517	Wildtype	36	62	1.7 (1.0, 3.0)	38	112	Reference
SLC22A2	rs8177516	Variant	0	2	NC	4	2	NC
SLC22A2	rs8177516	Wildtype	37	60	**2.2 (1.2, 3.9)**	33	111	Reference
SLCO1B1	rs2306283	Variant	29	38	**3.2 (1.3, 8.0)**	26	78	1.3 (0.5, 3.1)
SLCO1B1	rs2306283	Wildtype	8	22	1.0 (0.3, 3.3)	10	31	Reference
SLCO1B1	rs56061388	Variant	0	0	NC	0	2	NC
SLCO1B1	rs56061388	Wildtype	36	62	1.7 (1.0, 3.1)	37	111	Reference
SLCO1B1	rs72559745	Variant	0	0	NC	2	1	NC
SLCO1B1	rs72559745	Wildtype	36	62	1.8 (1.0, 3.2)	34	107	Reference
SLCO1B1	rs4149056	Variant	8	12	2.8 (1.0, 8.1)	11	28	1.3 (0.5, 3.0)
SLCO1B1	rs4149056	Wildtype	28	50	1.6 (0.8, 3.2)	27	83	Reference
SLCO1B1	rs55737008	Variant	0	0	NC	4	1	NC
SLCO1B1	rs55737008	Wildtype	37	62	**2.0 (1.1, 3.6)**	33	111	Reference
SLCO1B3	rs4149117	Variant	17	21	2.4 (1.0, 5.5)	14	38	1.0 (0.4, 2.3)
SLCO1B3	rs4149117	Wildtype	20	40	1.5 (0.7, 3.2)	22	70	Reference
SLCO1B3	rs7311358	Variant	17	22	2.2 (1.0, 4.8)	14	36	1.0 (0.4, 2.3)
SLCO1B3	rs7311358	Wildtype	20	40	1.4 (0.7, 3.0)	24	72	Reference
SLCO2B1	rs2306168	Variant	3	4	2.3 (0.5, 11.7)	11	24	1.3 (0.5, 3.0)
SLCO2B1	rs2306168	Wildtype	34	58	1.8 (1.0, 3.4)	27	86	Reference

(Continues)

TABLE 6 (Continued)

Gene symbol	dbSNP ID	Genotype	High particulate matter <10 microns[a]			Low particulate matter <10 microns[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
TPMT	rs1142345	Variant	4	13	0.9 (0.3, 3.0)	3	7	1.3 (0.3, 5.7)
TPMT	rs1142345	Wildtype	30	48	1.9 (1.0, 3.5)	33	100	Reference
TPMT	rs56161402	Variant	0	0	NC	3	1	NC
TPMT	rs56161402	Wildtype	37	61	**1.9 (1.1, 3.4)**	35	112	Reference
TPMT	rs1800460	Variant	4	13	1.1 (0.3, 3.8)	7	5	**5.5 (1.5, 19.4)**
TPMT	rs1800460	Wildtype	33	49	**2.5 (1.3, 4.6)**	30	106	Reference
UGT1A1	rs4148323	Variant	0	2	NC	1	4	NC
UGT1A1	rs4148323	Wildtype	37	59	**1.9 (1.1, 3.3)**	37	108	Reference
UGT1A1	rs4124874	Variant	27	43	1.8 (0.7, 4.6)	28	86	0.9 (0.4, 2.2)
UGT1A1	rs4124874	Wildtype	10	18	1.4 (0.5, 4.5)	9	27	Reference
UGT2B15	rs1902023	Variant	22	34	1.8 (0.8, 4.4)	23	81	0.7 (0.3, 1.7)
UGT2B15	rs1902023	Wildtype	15	25	1.3 (0.5, 3.4)	13	30	Reference
UGT2B7	rs7668258	Variant	17	35	1.4 (0.6, 3.2)	12	53	0.7 (0.3, 1.6)
UGT2B7	rs7668258	Wildtype	18	16	**3.0 (1.3, 7.3)**	19	49	Reference
UGT2B7	rs7662029	Variant	17	40	1.2 (0.5, 2.7)	17	55	0.9 (0.4, 2.1)
UGT2B7	rs7662029	Wildtype	19	21	2.3 (1.0, 5.2)	21	51	Reference
VKORC1	rs8050894	Variant	22	43	1.4 (0.6, 3.3)	25	79	0.8 (0.3, 1.8)
VKORC1	rs8050894	Wildtype	14	19	1.8 (0.7, 4.9)	12	32	Reference

[a]Highest tertile cut-off = 38.795 μg/m^3.
[b]Adjusted for maternal race, vitamin use, BMI, education, and smoking

TABLE 7 Gene-environment interactions for particulate matter <2.5 microns ($PM_{2.5}$) associations with risk of spina bifida

Gene symbol	dbSNP ID	Genotype	High particulate matter <2.5 microns[a]			Low particulate matter <2.5 microns[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
ABCB1	rs1045642	Variant	21	28	2.5 (0.8, 7.2)	18	57	1.0 (0.4, 2.6)
ABCB1	rs1045642	Wildtype	6	13	1.2 (0.3, 4.3)	10	24	Reference
ABCB1	rs1128503	Variant	20	24	2.5 (0.8, 7.7)	19	60	1.0 (0.4, 2.6)
ABCB1	rs1128503	Wildtype	7	17	1.2 (0.3, 4.5)	9	23	Reference
ABCB1	rs2032582	Variant	17	24	1.7 (0.6, 4.6)	13	52	0.7 (0.3, 1.7)
ABCB1	rs2032582	Wildtype	8	17	1.2 (0.4, 3.8)	15	30	Reference
ABCB1	rs2032582	Variant	0	2	NC	1	7	NC
ABCB1	rs2032582	Wildtype	20	34	1.4 (0.6, 3.2)	24	60	Reference
ABCC2	rs56199535	Variant	1	0	NC	0	1	NC
ABCC2	rs56199535	Wildtype	26	40	**2.2 (1.1, 4.7)**	26	82	Reference
ABCC2	rs717620	Variant	8	14	2.4 (0.8, 7.3)	10	22	1.3 (0.5, 3.4)
ABCC2	rs717620	Wildtype	19	26	2.3 (0.9, 5.5)	18	61	Reference
ABCC2	rs3740066	Variant	18	23	**3.9 (1.3, 11.7)**	20	40	2.1 (0.8, 5.8)
ABCC2	rs3740066	Wildtype	8	17	2.3 (0.7, 7.7)	8	43	Reference

(Continues)

TABLE 7 (Continued)

Gene symbol	dbSNP ID	Genotype	High particulate matter <2.5 microns[a]			Low particulate matter <2.5 microns[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
ABCC2	rs2273697	Variant	8	11	2.0 (0.6, 6.2)	8	23	1.1 (0.4, 3.0)
ABCC2	rs2273697	Wildtype	19	29	2.2 (0.9, 5.1)	20	58	Reference
ABCG2	rs2231142	Variant	6	11	1.6 (0.5, 5.4)	7	19	1.0 (0.3, 3.0)
ABCG2	rs2231142	Wildtype	21	30	2.2 (1.0, 5.1)	21	64	Reference
CYP1A1	rs1799814	Variant	1	3	NC	2	2	NC
CYP1A1	rs1799814	Wildtype	25	37	**2.2 (1.1, 4.7)**	26	81	Reference
CYP1A1	rs1048943	Variant	6	13	1.3 (0.4, 4.7)	14	26	1.5 (0.5, 4.0)
CYP1A1	rs1048943	Wildtype	21	27	**3.1 (1.3, 7.8)**	14	56	Reference
CYP1A1	rs41279188	Variant	1	0	NC	0	1	NC
CYP1A1	rs41279188	Wildtype	26	41	1.9 (0.9, 4.0)	27	80	Reference
CYP1A2	rs762551	Variant	12	12	**3.9 (1.2, 12.7)**	17	43	1.6 (0.6, 4.2)
CYP1A2	rs762551	Wildtype	15	29	2.2 (0.8, 6.2)	11	38	Reference
CYP1A2	rs2069514	Variant	7	14	1.6 (0.5, 5.7)	14	29	1.3 (0.5, 3.6)
CYP1A2	rs2069514	Wildtype	18	25	**3.5 (1.3, 9.4)**	11	50	Reference
CYP2A6	rs1801272	Variant	0	2	NC	0	4	NC
CYP2A6	rs1801272	Wildtype	27	38	**2.2 (1.1, 4.7)**	25	77	Reference
CYP2A6	rs4986891	Variant	2	6	NC	2	12	NC
CYP2A6	rs4986891	Wildtype	23	33	2.3 (1.0, 5.1)	24	68	Reference
CYP2A6	rs28399433	Variant	7	5	3.2 (0.8, 12.6)	7	10	1.6 (0.5, 5.0)
CYP2A6	rs28399433	Wildtype	20	36	2.0 (0.9, 4.4)	21	71	Reference
CYP2A6	hCV33845966	Variant	0	0	NC	0	1	NC
CYP2A6	hCV33845966	Wildtype	27	41	2.1 (1.0, 4.4)	27	82	Reference
CYP2A6	rs28399454	Variant	0	0	NC	1	2	NC
CYP2A6	rs28399454	Wildtype	26	39	2.2 (1.0, 4.5)	26	81	Reference
CYP2B6	rs3745274	Variant	12	15	**3.6 (1.2, 11.1)**	17	31	2.1 (0.8, 5.6)
CYP2B6	rs3745274	Wildtype	11	20	2.1 (0.7, 6.4)	10	42	Reference
CYP2B6	rs12721655	Variant	0	0	NC	0	0	NC
CYP2B6	rs12721655	Wildtype	26	40	2.0 (1.0, 4.1)	28	82	Reference
CYP2B6	rs28399499	Variant	0	0	NC	0	2	NC
CYP2B6	rs28399499	Wildtype	27	39	2.0 (1.0, 4.2)	28	79	Reference
CYP2C19	rs12248560	Variant	7	9	2.6 (0.8, 9.2)	5	24	0.5 (0.2, 1.6)
CYP2C19	rs12248560	Wildtype	15	25	1.4 (0.6, 3.7)	20	47	Reference
CYP2C19	rs3758580	Variant	6	3	**7.4 (1.3, 40.4)**	7	18	1.3 (0.4, 3.7)
CYP2C19	rs3758580	Wildtype	21	38	2.1 (0.9, 4.6)	19	63	Reference
CYP2C19	rs17878459	Variant	1	2	NC	2	5	NC
CYP2C19	rs17878459	Wildtype	26	39	2.0 (1.0, 4.2)	26	78	Reference
CYP2C19	rs4244285	Variant	6	3	5.6 (1.0, 29.9)	8	23	0.9 (0.3, 2.6)
CYP2C19	rs4244285	Wildtype	21	38	1.8 (0.8, 4.0)	20	60	Reference

(Continues)

TABLE 7 (Continued)

Gene symbol	dbSNP ID	Genotype	High particulate matter <2.5 microns[a]			Low particulate matter <2.5 microns[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP2C19	rs41291556	Variant	1	0	NC	3	5	1.6 (0.3, 8.1)
CYP2C19	rs41291556	Wildtype	20	37	1.8 (0.8, 4.1)	23	71	Reference
CYP2C19	rs17885098	Variant	3	1	NC	2	8	NC
CYP2C19	rs17885098	Wildtype	24	39	1.8 (0.8, 3.8)	26	71	Reference
CYP2C19	rs28399504	Variant	0	2	NC	1	0	NC
CYP2C19	rs28399504	Wildtype	25	36	**2.3 (1.1, 5.0)**	25	79	Reference
CYP2C19	rs17886522	Variant	1	1	NC	1	1	NC
CYP2C19	rs17886522	Wildtype	26	40	2.0 (0.9, 4.1)	27	81	Reference
CYP2C8	rs10509681	Variant	4	7	1.6 (0.4, 7.1)	5	16	1.1 (0.3, 3.9)
CYP2C8	rs10509681	Wildtype	23	32	2.3 (1.0, 5.1)	23	66	Reference
CYP2C8	rs11572080	Variant	2	7	NC	3	14	1.0 (0.2, 4.2)
CYP2C8	rs11572080	Wildtype	25	31	**2.5 (1.2, 5.6)**	25	69	Reference
CYP2C8	rs1058930	Variant	1	3	NC	3	8	1.2 (0.3, 5.4)
CYP2C8	rs1058930	Wildtype	26	37	2.2 (1.0, 4.6)	25	75	Reference
CYP2C8	rs11572103	Variant	0	0	NC	1	2	NC
CYP2C8	rs11572103	Wildtype	27	41	2.1 (1.0, 4.3)	27	80	Reference
CYP2C9	rs7900194	Variant	0	1	NC	0	0	NC
CYP2C9	rs7900194	Wildtype	26	40	2.1 (1.0, 4.3)	28	83	Reference
CYP2C9	rs1799853	Variant	4	7	2.0 (0.5, 8.4)	3	12	1.2 (0.3, 5.5)
CYP2C9	rs1799853	Wildtype	23	34	2.1 (1.0, 4.6)	25	71	Reference
CYP2C9	rs1057910	Variant	1	0	NC	1	9	NC
CYP2C9	rs1057910	Wildtype	26	41	2.0 (1.0, 4.2)	26	74	Reference
CYP2C9	rs28371686	Variant	0	0	NC	2	2	NC
CYP2C9	rs28371686	Wildtype	27	39	**2.3 (1.1, 4.8)**	26	80	Reference
CYP2C9	rs9332239	Variant	0	1	NC	1	1	NC
CYP2C9	rs9332239	Wildtype	25	40	2.0 (1.0, 4.3)	27	79	Reference
CYP2C9	rs28371685	Variant	0	0	NC	1	2	NC
CYP2C9	rs28371685	Wildtype	27	39	**2.3 (1.1, 4.9)**	26	81	Reference
CYP2C9	rs9332130	Variant	2	0	NC	3	1	NC
CYP2C9	rs9332130	Wildtype	25	41	2.1 (1.0, 4.5)	25	82	Reference
CYP2C9	rs9332131	Variant	0	1	NC	6	1	NC
CYP2C9	rs9332131	Wildtype	26	40	**2.6 (1.2, 5.6)**	21	76	Reference
CYP2C9	hCV72649992	Variant	1	0	NC	3	1	NC
CYP2C9	hCV72649992	Wildtype	23	40	2.2 (1.0, 4.7)	23	76	Reference
CYP2C9	rs72558190	Variant	0	1	NC	1	1	NC
CYP2C9	rs72558190	Wildtype	27	40	**2.2 (1.1, 4.6)**	27	81	Reference
CYP2D6	rs28371706	Variant	0	1	NC	1	0	NC
CYP2D6	rs28371706	Wildtype	26	38	2.1 (1.0, 4.4)	27	78	Reference

(Continues)

TABLE 7 (Continued)

			High particulate matter <2.5 microns[a]			Low particulate matter <2.5 microns[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
CYP2D6	rs3892097	Variant	6	10	3.0 (0.8, 10.9)	10	23	1.9 (0.7, 5.3)
CYP2D6	rs3892097	Wildtype	21	29	**2.5 (1.1, 5.9)**	18	59	Reference
CYP2D6	rs5030862	Variant	0	1	NC	1	2	NC
CYP2D6	rs5030862	Wildtype	27	39	**2.2 (1.1, 4.7)**	27	78	Reference
CYP2D6	rs72549349	Variant	0	0	NC	1	0	NC
CYP2D6	rs72549349	Wildtype	27	41	**2.3 (1.1, 4.8)**	26	80	Reference
CYP2D6	rs72549350	Variant	3	3	2.6 (0.4, 17.0)	0	2	NC
CYP2D6	rs72549350	Wildtype	24	38	1.9 (0.9, 4.0)	27	78	Reference
CYP2D6	rs35742686	Variant	2	1	NC	1	5	NC
CYP2D6	rs35742686	Wildtype	25	38	2.2 (1.0, 4.7)	25	75	Reference
CYP2D6	rs72549353	Variant	0	1	NC	1	1	NC
CYP2D6	rs72549353	Wildtype	27	38	**2.3 (1.1, 4.8)**	27	80	Reference
CYP2D6	hCV32407240	Variant	0	1	NC	1	2	NC
CYP2D6	hCV32407240	Wildtype	27	38	**2.4 (1.1, 5.0)**	26	80	Reference
CYP2D6	rs5030655	Variant	0	0	NC	1	2	NC
CYP2D6	rs5030655	Wildtype	24	38	2.2 (1.0, 4.7)	25	73	Reference
CYP2D6	rs72549346	Variant	3	0	NC	2	5	NC
CYP2D6	rs72549346	Wildtype	23	37	2.0 (0.9, 4.4)	24	74	Reference
CYP2D6	rs5030865	Variant	1	0	NC	2	4	NC
CYP2D6	rs5030865	Wildtype	25	39	1.8 (0.9, 3.9)	26	70	Reference
CYP2D6	rs5030865	Variant	2	4	NC	6	9	2.2 (0.6, 7.7)
CYP2D6	rs5030865	Wildtype	23	35	2.3 (1.0, 5.2)	22	67	Reference
CYP3A4	rs55785340	Variant	0	1	NC	3	4	2.2 (0.4, 12.2)
CYP3A4	rs55785340	Wildtype	25	40	2.1 (1.0, 4.4)	25	78	Reference
CYP3A5	rs10264272	Variant	1	1	NC	0	2	NC
CYP3A5	rs10264272	Wildtype	26	39	2.0 (1.0, 4.2)	28	81	Reference
CYP3A5	rs55965422	Variant	0	0	NC	1	2	NC
CYP3A5	rs55965422	Wildtype	24	41	1.9 (0.9, 4.2)	25	79	Reference
CYP3A5	rs41303343	Variant	0	0	NC	1	2	NC
CYP3A5	rs41303343	Wildtype	26	41	2.0 (1.0, 4.3)	26	79	Reference
DPYD	rs1801267	Variant	0	0	NC	0	1	NC
DPYD	rs1801267	Wildtype	26	40	**2.3 (1.1, 4.9)**	26	80	Reference
DPYD	rs1801265	Variant	10	18	1.5 (0.5, 4.4)	9	33	0.6 (0.2, 1.7)
DPYD	rs1801265	Wildtype	17	22	1.9 (0.8, 4.7)	19	49	Reference
DPYD	rs3918290	Variant	0	1	NC	0	1	NC
DPYD	rs3918290	Wildtype	27	40	2.1 (1.0, 4.2)	28	80	Reference
DPYD	hCV32287186	Variant	0	0	NC	0	1	NC
DPYD	hCV32287186	Wildtype	27	41	2.0 (1.0, 4.1)	28	82	Reference

(Continues)

TABLE 7 (Continued)

			High particulate matter <2.5 microns[a]			Low particulate matter <2.5 microns[a]		
Gene symbol	dbSNP ID	Genotype	Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
GSTP1	rs1695	Variant	20	28	1.5 (0.5, 4.2)	18	56	0.5 (0.2, 1.5)
GSTP1	rs1695	Wildtype	7	12	1.1 (0.3, 4.4)	10	26	Reference
NAT1	rs4986782	Variant	0	1	NC	1	1	NC
NAT1	rs4986782	Wildtype	27	40	2.2 (1.0, 4.5)	27	81	Reference
NAT1	rs4986988	Variant	3	1	NC	0	7	NC
NAT1	rs4986988	Wildtype	24	39	1.7 (0.8, 3.6)	28	76	Reference
NAT1	rs55793712	Variant	1	0	NC	2	2	NC
NAT1	rs55793712	Wildtype	26	41	2.1 (1.0, 4.5)	25	81	Reference
NAT2	rs1208	Variant	18	23	1.9 (0.7, 5.2)	16	54	0.7 (0.3, 1.7)
NAT2	rs1208	Wildtype	8	18	1.0 (0.3, 3.2)	12	27	Reference
NAT2	rs1799931	Variant	5	5	3.4 (0.8, 14.7)	10	16	1.8 (0.7, 5.0)
NAT2	rs1799931	Wildtype	22	34	2.2 (1.0, 5.0)	18	62	Reference
NAT2	rs1799930	Variant	13	19	2.4 (0.9, 6.9)	13	33	1.4 (0.5, 3.5)
NAT2	rs1799930	Wildtype	11	19	1.7 (0.6, 4.8)	14	44	Reference
NAT2	rs1799929	Variant	17	22	2.0 (0.7, 5.7)	16	53	0.8 (0.3, 2.0)
NAT2	rs1799929	Wildtype	9	19	1.2 (0.4, 3.7)	12	28	Reference
NAT2	rs1801280	Variant	18	21	1.9 (0.7, 5.5)	15	52	0.6 (0.2, 1.5)
NAT2	rs1801280	Wildtype	8	18	0.8 (0.2, 2.7)	12	23	Reference
NAT2	rs1041983	Variant	12	25	1.6 (0.6, 4.7)	15	44	1.2 (0.5, 3.1)
NAT2	rs1041983	Wildtype	14	16	2.8 (1.0, 7.9)	13	38	Reference
SLC15A2	rs2293616	Variant	16	25	2.2 (0.8, 6.6)	18	57	0.9 (0.3, 2.4)
SLC15A2	rs2293616	Wildtype	10	16	1.5 (0.5, 4.7)	10	26	Reference
SLC15A2	rs2257212	Variant	16	25	2.0 (0.7, 5.8)	17	56	0.8 (0.3, 2.0)
SLC15A2	rs2257212	Wildtype	11	16	1.5 (0.5, 4.5)	11	27	Reference
SLC15A2	rs1143671	Variant	16	24	2.3 (0.8, 6.7)	18	56	0.9 (0.4, 2.5)
SLC15A2	rs1143671	Wildtype	11	16	1.7 (0.5, 5.1)	10	26	Reference
SLC15A2	rs1143672	Variant	16	22	**3.6 (1.1, 11.6)**	17	56	1.0 (0.4, 2.8)
SLC15A2	rs1143672	Wildtype	11	15	1.9 (0.6, 6.0)	9	25	Reference
SLC22A1	rs628031	Variant	12	17	**3.3 (1.1, 9.9)**	12	38	1.2 (0.5, 3.2)
SLC22A1	rs628031	Wildtype	13	22	1.8 (0.7, 4.8)	15	43	Reference
SLC22A1	rs2282143	Variant	1	3	NC	1	4	NC
SLC22A1	rs2282143	Wildtype	26	38	2.1 (1.0, 4.3)	27	79	Reference
SLC22A1	rs34059508	Variant	0	1	NC	0	1	NC
SLC22A1	rs34059508	Wildtype	26	40	2.0 (1.0, 4.1)	28	82	Reference
SLC22A1	rs55918055	Variant	1	0	NC	3	2	NC
SLC22A1	rs55918055	Wildtype	26	40	2.1 (1.0, 4.5)	25	80	Reference
SLC22A1	rs72552763	Variant	10	12	2.4 (0.8, 7.3)	11	33	1.0 (0.4, 2.6)
SLC22A1	rs72552763	Wildtype	17	29	1.9 (0.8, 4.7)	16	49	Reference

(Continues)

TABLE 7 (Continued)

Gene symbol	dbSNP ID	Genotype	High particulate matter <2.5 microns[a]			Low particulate matter <2.5 microns[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
SLC22A2	rs316019	Variant	3	6	1.8 (0.4, 8.7)	2	11	NC
SLC22A2	rs316019	Wildtype	24	34	2.1 (1.0, 4.6)	25	72	Reference
SLC22A2	rs8177507	Variant	0	1	NC	0	0	NC
SLC22A2	rs8177507	Wildtype	27	40	2.1 (1.0, 4.4)	28	83	Reference
SLC22A2	rs8177517	Variant	1	0	NC	0	0	NC
SLC22A2	rs8177517	Wildtype	26	41	1.9 (0.9, 4.0)	28	83	Reference
SLC22A2	rs8177516	Variant	1	0	NC	3	2	NC
SLC22A2	rs8177516	Wildtype	25	41	2.1 (1.0, 4.3)	25	81	Reference
SLCO1B1	rs2306283	Variant	19	26	**3.4 (1.1, 11.2)**	20	58	1.5 (0.5, 4.4)
SLCO1B1	rs2306283	Wildtype	7	11	2.2 (0.6, 8.6)	7	23	Reference
SLCO1B1	rs56061388	Variant	0	0	NC	0	1	NC
SLCO1B1	rs56061388	Wildtype	26	41	1.9 (0.9, 4.0)	28	82	Reference
SLCO1B1	rs72559745	Variant	1	0	NC	0	1	NC
SLCO1B1	rs72559745	Wildtype	25	41	1.9 (0.9, 3.9)	27	78	Reference
SLCO1B1	rs4149056	Variant	6	5	**6.1 (1.4, 25.9)**	9	24	1.4 (0.5, 3.8)
SLCO1B1	rs4149056	Wildtype	21	35	2.0 (0.9, 4.7)	18	58	Reference
SLCO1B1	rs55737008	Variant	2	0	NC	2	1	NC
SLCO1B1	rs55737008	Wildtype	25	41	2.1 (1.0, 4.4)	26	81	Reference
SLCO1B3	rs4149117	Variant	12	13	**3.2 (1.1, 9.1)**	12	26	1.4 (0.5, 3.6)
SLCO1B3	rs4149117	Wildtype	15	25	2.2 (0.9, 5.6)	15	55	Reference
SLCO1B3	rs7311358	Variant	12	15	2.7 (1.0, 7.4)	12	25	1.3 (0.5, 3.5)
SLCO1B3	rs7311358	Wildtype	15	25	2.0 (0.8, 5.1)	16	55	Reference
SLCO2B1	rs2306168	Variant	4	5	2.2 (0.5, 9.6)	7	17	1.1 (0.4, 3.1)
SLCO2B1	rs2306168	Wildtype	23	36	2.0 (0.9, 4.5)	21	65	Reference
TPMT	rs1142345	Variant	4	4	2.3 (0.5, 10.7)	2	12	NC
TPMT	rs1142345	Wildtype	19	36	1.5 (0.7, 3.4)	25	67	Reference
TPMT	rs56161402	Variant	1	0	NC	0	0	NC
TPMT	rs56161402	Wildtype	26	40	2.0 (1.0, 4.2)	28	83	Reference
TPMT	rs1800460	Variant	6	4	3.9 (0.9, 16.7)	4	9	1.9 (0.5, 7.5)
TPMT	rs1800460	Wildtype	21	37	2.0 (0.9, 4.4)	23	72	Reference
UGT1A1	rs4148323	Variant	0	1	NC	0	3	NC
UGT1A1	rs4148323	Wildtype	27	39	2.0 (1.0, 4.2)	28	79	Reference
UGT1A1	rs4124874	Variant	19	32	2.0 (0.6, 6.3)	22	61	1.2 (0.4, 3.6)
UGT1A1	rs4124874	Wildtype	7	9	2.7 (0.6, 11.9)	6	21	Reference
UGT2B15	rs1902023	Variant	19	22	1.6 (0.6, 4.5)	12	61	0.3 (0.1, 0.8)
UGT2B15	rs1902023	Wildtype	8	16	0.5 (0.2, 1.7)	16	20	Reference
UGT2B7	rs7668258	Variant	12	25	1.0 (0.3, 2.8)	8	36	0.4 (0.2, 1.2)
UGT2B7	rs7668258	Wildtype	12	11	1.8 (0.6, 5.3)	18	33	Reference
UGT2B7	rs7662029	Variant	14	25	1.5 (0.5, 4.2)	9	42	0.5 (0.2, 1.5)

(Continues)

TABLE 7 (Continued)

Gene symbol	dbSNP ID	Genotype	High particulate matter <2.5 microns[a]			Low particulate matter <2.5 microns[a]		
			Case *N*	Control *N*	aOR[b] (95% CI)	Case *N*	Control *N*	aOR[b] (95% CI)
UGT2B7	rs7662029	Wildtype	13	13	2.2 (0.8, 6.2)	18	37	Reference
VKORC1	rs8050894	Variant	15	29	1.4 (0.5, 3.8)	17	59	0.7 (0.3, 1.9)
VKORC1	rs8050894	Wildtype	11	11	2.9 (0.8, 10.0)	10	23	Reference

[a]Highest tertile cut-off = 19.860 μg/m^3.
[b]Adjusted for maternal race, vitamin use, BMI, education, and smoking.

(rs762551), there was no increased risk of spina bifida (OR = 1.5, 95%CI: 0.5–4.5).

Results of the maternal genotypes among a subset of samples were similar to those of the infant genotypes, though there was substantially less power to detect statistical significance. Interaction among the CYP gene variants and high PM_{10} and $PM_{2.5}$ showed similar associations (*CYP1A1* (rs1048943) $OR_{PM2.5} = 3.3$ (0.9, 11.7); $OR_{PM10} = 1.5$ (0.4, 6.2) and *CYP1A2* (rs2069514) $OR_{PM2.5} = 4.0$ (1.9, 16.0); $OR_{PM10} = 1.9$ (0.5, 7.5), additional data not shown).

4 | DISCUSSION

In our previous study of air pollution exposures during the first 2 months of pregnancy, we found associations between elevated levels of CO and NO_2 and risk of spina bifida ($OR_{CO} = 2.00$, 95% CI: 1.06, 3.75; $OR_{NO2} = 1.73$, 95% CI: 1.01, 2.97) (Padula et al., 2013). Our current study extends those findings and demonstrates a gene-environment interaction between each of the five pollutants and several gene variants: NO (*ABCC2*), NO_2 (*ABCC2*, *SLC01B1*), PM_{10} (*ABCC2*, *CYP1A1*, *CYP2B6*, *CYP2C19*, *CYP2D6*, *NAT2*, *SLC01B1*, *SLC01B3)*, $PM_{2.5}$ (*CYP1A1* and *CYP1A2*). These gene pathways are involved in metabolizing both endogenous compounds and myriad xenobiotic chemicals (Nebert, 1997). Their role in detoxifying air pollutant exposures has been investigated as potential modifiers in environmental health studies (Kelada, Eaton, Wang, Rothman, & Khoury, 2003). For example, airborne polycyclic aromatic hydrocarbons (a component of particulate matter) have been associated with measures of genotoxicity of *CYP1A1* and *NAT2* genes (Kelada et al., 2003). Futhermore, several studies have reported the role of specific variants in detoxification genes in association with congenital heart malformations including *CYP1A1* and *ABCB1* (Vecoli, Pulignani, & Andreassi, 2016).

We view this investigation as exploratory even though some results showed sizable ORs (>4) and 95% CIs excluding 1. Such caution seems prudent owing to sample sizes being relatively small, numerous comparisons being made, and a paucity of previous studies to corroborate these findings. With regard to the latter, we are aware of only one previous study that has investigated spina bifida risk, gene variants, and air pollutants, with the pollutants being from indoor air pollution (exposure index score based on exposure active or passive smoking and coal combustion) or placental polycyclic aromatic hydrocarbons and 12 variants of AHR and CYP genes found that *CYP1B1* modifies the effect of indoor air pollution and NTD risk. For mothers with the *CYP1B1* (rs2855658) GG variant, exposure to indoor air pollution led to a dose response relationship for NTD risk, with ORs of 3.0 (95% CI: 1.6–5.7) and 8.1 (95% CI: 3.8–17) for medium and high levels of exposure, respectively. Although our study did not examine the *CYP1B1* gene variants, we did find gene-environment interactions with several other CYP gene variants (*CYP1A1*, *CYP1A2*, *CYP2B6*, *CYP2C8*, *CYP2C19*, *CYP2D6*) (Wang et al., 2014).

A previous study on smoking, which has similar constituents to air pollution, and NAT1 C1095A variant found an increased risk of spina bifida ($p = .003$) (Jensen, Hoess, Whitehead, & Mitchell, 2005). Additional studies have examined the interaction between smoking and gene variants for their combined risk of gastroschisis and cleft palate (Jenkins et al., 2014; Torfs et al., 2006; Wu et al., 2012).

Although we know that maternal folic acid intake is associated with reduced risk of spina bifida, in general, the underlying biological mechanism has not been elucidated. Increased folic acid in the population through vitamin use and fortification of foods may have reduced spinia bifida, but the continued prevalence suggests that factors other than folic acid are involved with the etiology of spina bifida (Au, Ashley-Koch, & Northrup, 2010), Several factors including low socioeconomic status and both advanced and young maternal age have been observed as risk factors for spina bifida in epidemiologic studies; however, several observations support genetic risk factors as well. Many studies have evaluated associations of neural tube defects with candidate genes known to code for proteins/enzymes in folate transport (e.g., *SLC19A1*), methylation (e.g., *NAT1*, *NAT2*), and oxidative stress (e.g., *CYP26A1*) (Au et al., 2010). Given the mixed results, further research is warranted to examine further gene-environment interactions.

The results should be considered in context to some limitations of our study. The relatively small sample size for these types of analyses restricted the inference of our results. We performed numerous analyses and explored many potential effect modifiers without multiple comparisons made. These analyses are not meant to test a specific hypothesis, but rather serve as an initial investigation to generate hypotheses and begin the large amount of work needed to understand more complex pathways than previously examined.

5 | CONCLUSIONS

Our study is the first examination of the interaction between these gene variants and air pollutant exposures with regard to spina bifida

risk in a well-characterized population in California. Despite its limitations, this study exhibits detailed exposure assessment and targeted gene variant analyses. The results warrant further investigation of gene-environment interactions and risk of birth defects including additional exposures and gene variants.

ACKNOWLEDGMENTS

This work would not have been possible without the tremendous leadership and scholarship of our dear colleague, Dr. Ed Lammer, who sadly passed away during this research. This research was supported by funds from the Centers for Disease Control and Prevention, Center of Excellence Award U50/CCU913241, California Tobacco-Related Disease Research Program (16RT-0086), the Environmental Protection Agency (RD835435) and the National Institutes of Health (P01ES022849, R00ES021470). We thank the California Department of Public Health Maternal Child and Adolescent Health Division for providing data for these analyses. The findings and conclusions in this report are those of the authors and do not necessarily represent the views of the California Department of Public Health.

CONFLICT OF INTEREST

The authors declare no competing financial interests.

ORCID

Amy M. Padula http://orcid.org/0000-0003-1435-4814

REFERENCES

Au, K. S., Ashley-Koch, A., & Northrup, H. (2010). Epidemiologic and genetic aspects of spina bifida and other neural tube defects. *Developmental Disabilities Research Reviews*, *16*(1), 6–15.

Barrett, J. C., Fry, B., Maller, J., & Daly, M. J. (2005). Haploview: Analysis and visualization of LD and haplotype maps. *Bioinformatics (Oxford, England)*, *21*(2), 263–265.

Croen, L. A., Shaw, G. M., Jensvold, N. G., & Harris, J. A. (1991). Birth defects monitoring in California: A resource for epidemiological research. *Paediatric and Perinatal Epidemiology*, *5*(4), 423–427.

Enaw, J. O., Zhu, H., Yang, W., Lu, W., Shaw, G. M., Lammer, E. J., & Finnell, R. H. (2006). CHKA and PCYT1A gene polymorphisms, choline intake and spina bifida risk in a California population. *BMC Medicine*, *4*, 36.

Girguis, M. S., Strickland, M. J., Hu, X., Liu, Y., Bartell, S. M., & Vieira, V. M. (2016). Maternal exposure to traffic-related air pollution and birth defects in Massachusetts. *Environmental Research*, *146*, 1–9.

Honein, M. A., Paulozzi, L. J., Mathews, T. J., Erickson, J. D., & Wong, L. Y. (2001). Impact of folic acid fortification of the US food supply on the occurrence of neural tube defects. *JAMA*, *285*(23), 2981–2986.

Jenkins, M. M., Reefhuis, J., Gallagher, M. L., Mulle, J. G., Hoffmann, T. J., Koontz, D. A., … National Birth Defects Prevention Study. (2014). Maternal smoking, xenobiotic metabolizing enzyme gene variants, and gastroschisis risk. *American Journal of Medical Genetics: Part A*, *164A*(6), 1454–1463.

Jensen, L. E., Hoess, K., Whitehead, A. S., & Mitchell, L. E. (2005). The NAT1 C1095A polymorphism, maternal multivitamin use and smoking, and the risk of spina bifida. *Birth Defects Research: Part A, Clinical and Molecular Teratology*, *73*(7), 512–516.

Kelada, S. N., Eaton, D. L., Wang, S. S., Rothman, N. R., & Khoury, M. J. (2003). The role of genetic polymorphisms in environmental health. *Environmental Health Perspectives*, *111*(8), 1055–1064.

Lupo, P. J., Symanski, E., Waller, D. K., Chan, W., Langlois, P. H., Canfield, M. A., & Mitchell, L. E. (2011). Maternal exposure to ambient levels of benzene and neural tube defects among offspring: Texas, 1999–2004. *Environmental Health Perspectives*, *119*(3), 397–402.

Nebert, D. W. (1997). Polymorphisms in drug-metabolizing enzymes: What is their clinical relevance and why do they exist? *American Journal of Human Genetics*, *60*(2), 265–271.

Padula, A. M., Tager, I. B., Carmichael, S. L., Hammond, S. K., Lurmann, F., & Shaw, G. M. (2013). The association of ambient air pollution and traffic exposures with selected congenital anomalies in the San Joaquin Valley of California. *American Journal of Epidemiology*, *177*(10), 1074–1085.

Rasmussen, S. A., Olney, R. S., Holmes, L. B., Lin, A. E., Keppler-Noreuil, K. M., & Moore, C. A. (2003). Guidelines for case classification for the National Birth Defects Prevention Study. *Birth Defects Research: Part A, Clinical and Molecular Teratology*, *67*(3), 193–201.

Reefhuis, J., Gilboa, S. M., Anderka, M., Browne, M. L., Feldkamp, M. L., Hobbs, C. A., … National, B. D. P. S. (2015). The National Birth Defects Prevention Study: A review of the methods. *Birth Defects Research. Part A, Clinical and Molecular Teratology*, *103*(8), 656–669.

Richards, B., Skoletsky, J., Shuber, A. P., Balfour, R., Stern, R. C., Dorkin, H. L., … Klinger, K. W. (1993). Multiplex PCR amplification from the CFTR gene using DNA prepared from buccal brushes/swabs. *Human Molecular Genetics*, *2*(2), 159–163.

Shaw, G. M., Iovannisci, D. M., Yang, W., Finnell, R. H., Carmichael, S. L., Cheng, S., & Lammer, E. J. (2005). Endothelial nitric oxide synthase (NOS3) genetic variants, maternal smoking, vitamin use, and risk of human orofacial clefts. *American Journal of Epidemiology*, *162*(12), 1207–1214.

Shaw, G. M., Nelson, V., Iovannisci, D. M., Finnell, R. H., & Lammer, E. J. (2003). Maternal occupational chemical exposures and biotransformation genotypes as risk factors for selected congenital anomalies. *American Journal of Epidemiology*, *157*(6), 475–484.

Torfs, C. P., Christianson, R. E., Iovannisci, D. M., Shaw, G. M., & Lammer, E. J. (2006). Selected gene polymorphisms and their interaction with maternal smoking, as risk factors for gastroschisis. *Birth Defects Research: Part A, Clinical and Molecular Teratology*, *76*(10), 723–730.

Vecoli, C., Pulignani, S., & Andreassi, M. G. (2016). Genetic and epigenetic mechanisms linking air pollution and congenital heart disease. *Journal of Cardiovascular Development and Disease*, *3*(4), 32.

Wang, L., Li, Z., Jin, L., Li, K., Yuan, Y., Fu, Y., … Ren, A. (2014). Indoor air pollution and neural tube defects: Effect modification by maternal genes. *Epidemiology*, *25*(5), 658–665.

Wu, T., Fallin, M. D., Shi, M., Ruczinski, I., Liang, K. Y., Hetmanski, J. B., … Beaty, T. H. (2012). Evidence of gene-environment interaction for the RUNX2 gene and environmental tobacco smoke in controlling the risk of cleft lip with/without cleft palate. *Birth Defects Research: Part A, Clinical and Molecular Teratology*, *94*(2), 76–83.

Wu, T., Liang, K. Y., Hetmanski, J. B., Ruczinski, I., Fallin, M. D., Ingersoll, R. G., … Beaty, T. H. (2010). Evidence of gene-environment interaction for the IRF6 gene and maternal multivitamin supplementation in controlling the risk of cleft lip with/without cleft palate. *Human Genetics*, *128*(4), 401–410.

Yoon, P. W., Rasmussen, S. A., Lynberg, M. C., Moore, C. A., Anderka, M., Carmichael, S. L., ... Edmonds, L. D. (2001). The National Birth Defects Prevention Study. *Public Health Reports*, *116*(1_Suppl), 32–40.

SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

How to cite this article: Padula AM, Yang W, Schultz K, Lurmann F, Hammond SK, Shaw GM. Genetic variation in biotransformation enzymes, air pollution exposures, and risk of spina bifida. *Am J Med Genet Part A*. 2018;176A:1055–1090. https://doi.org/10.1002/ajmg.a.38661