Original Contribution

Associations Between Ozone and Preterm Birth in Women Who Develop Gestational Diabetes

Yu-Ting Lin, Chau-Ren Jung, Yungling Leo Lee, and Bing-Fang Hwang*

* Correspondence to Dr. Bing-Fang Hwang, Department of Occupational Safety and Health, College of Public Health, China Medical University, No. 91 Hsueh-Shih Road, Taichung, Taiwan 40402, R.O.C. (e-mail: bfhwang@mail.cmu.edu.tw).

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Prenatal exposure to ambient air pollutants might cause adverse birth outcomes; however, there have been few studies in which the association between air pollution and preterm birth was examined after stratifying by pregnancy complications. We conducted a population-based case-control study of 1,510,064 singleton births from the Taiwanese birth registry during 2001–2007. Of the total of 1,510,064 births, we designated all 86,224 preterm births as the case group and then randomly selected an additional 344,896 from the remaining births (equivalent to 4 full-term births for every 1 preterm birth) as the control sample. We used an inverse distance weighting approach to calculate an average exposure parameter for air pollutants. The adjusted odds ratio for preterm birth per 10-ppb increase in ozone was 1.12 (95% confidence interval: 1.01, 1.23) for women with gestational diabetes mellitus who were exposed in the third trimester and 1.02 (95% confidence interval: 1.01, 1.03) for women without gestational diabetes (P for interaction <0.001). These findings suggest that exposure to ozone in pregnancy is associated with an increased risk of preterm birth, particularly for women who have gestational diabetes mellitus.

Abbreviations: CI, confidence interval; OR, odds ratio; PM10, particulate matter with an aerodynamic diameter of 10 µm or less.

Preterm birth is a significant perinatal health problem across the globe, and it is associated with an increased risk of neonatal morbidity and mortality compared with full-term birth (1–4). In Taiwan, this is especially important because of the declining crude birth rate (from 15.52% to 10.08% from 1992 to 2012). The prevalence of preterm births worldwide is 9.6% (1), and the average in Taiwan is 9.2%. There is growing evidence that ambient air pollution might contribute to health problems, such as premature death, adverse reproductive outcomes, and respiratory and cardiovascular diseases, particularly in pregnant women and newborn children (5–9). In the past decade, the ambient air concentrations of most pollutants have decreased in Taiwan. The exception is ozone, the concentration of which has gradually increased (10). Most previous studies of the links between air pollution and preterm births have explored the association of traffic-related pollutants or particulate matter with preterm birth rather than ozone (11–14). In an animal experiment, Kavlock et al. (15) reported that exposure to ozone during late gestation in rats led to abnormal reproductive function. However, it is still unclear which biological mechanisms operate during ozone exposure in humans. Numerous epidemiologic studies have provided evidence that ambient air pollution plays an important role in preterm birth. The evidence of an association between ozone and preterm birth, however, has been inconsistent (16–21). These inconsistent results may be due to differences in the prevalence of conditions that modify the association or in the average concentrations of ozone in the areas studied.

In addition to adverse birth outcomes, the association between pregnancy complications and air pollution has been examined in previous studies (19, 22–25). However, to our knowledge, no study has yet explored the association between air pollution and pregnancy complications and the potential effect of that association on the risk of preterm birth. We conducted a population-based case-control study to assess the association between air pollution and preterm birth in single-pollutant and multipollutant models. We also investigated the link between exposure to air pollution and the risk...
of preterm birth in the subgroups of pregnant women who experienced pregnancy complications.

METHODS

Study population

We conducted a population-based case-control study to assess the association of air pollution with preterm birth and to see whether it varied by whether or not the woman experienced pregnancy complications. By law, all births in Taiwan must be reported to the Taiwan Birth Registry within 15 days of delivery. Almost 99% of pregnant Taiwanese women have free prenatal care, which includes at least 10 prenatal care visits during pregnancy that are covered by national health insurance. The Taiwanese birth registry is a valid source of data on preterm births, with a low rate of missing information (1.6%), high sensitivity and specificity (92.8% and 99.6%, respectively), and high reliability (Cohen’s k statistic, 0.92) (26).

In the birth registry, preterm birth is defined as a gestational age of less than 37 weeks based on routine ultrasound examination. The source population comprised 1,510,064 singleton births registered in the Taiwanese birth registry from 2001 to 2007. We excluded infants born with any birth defect (International Classification of Diseases, Ninth Revision, Clinical Modification codes 740–758; n = 9,073) and infants born to mothers with a chronic disease (chronic diabetes, chronic hypertension, or heart disease; n = 5,090). Birth defects and other health conditions were primarily diagnosed by physicians. We excluded births to women who smoked (n = 394), infants with a birth weight less than 500 g or greater than 5,000 g (n = 469), infants born at less than 20 gestational weeks or greater than 42 gestational weeks (n = 329), and those with a birthplace in an area in which air pollution monitoring data were missing (n = 15,074). The final study population used in the statistical analysis included 86,224 preterm birth cases and 344,896 randomly selected control subjects. The study was approved by the Institutional Review Board of the China Medical University Hospital, and it complied with the principles outlined in the Helsinki Declaration. Because the data were anonymized, the institutional review board specifically waived the need to obtain consent from each subject.

Exposure assessment

Data on 5 of the major air pollutants (ozone, carbon monoxide, nitrogen dioxide, particulate matter with an aerodynamic diameter of 10 μm or less (PM10), and sulfur dioxide) were obtained from 72 Taiwan Environmental Protection Agency air quality monitoring stations on Taiwan’s main island. Data on air pollution concentrations are continually measured by fully automated monitoring stations on an hourly basis. Carbon monoxide is measured using nondispersive infrared absorption, nitrogen dioxide is measured using chemiluminescence, ozone is measured using ultraviolet absorption, PM10 is measured using β-gauge, and sulfur dioxide is measured using ultraviolet fluorescence.

The map coordinates of air monitoring stations and the data on air pollution were identified and managed using a geographic information system (ArcGIS, version 10; ESRI, Redlands, California). The air pollutant measurements were integrated into monthly data points and interpolated to surface-level data using an inverse distance weighting approach with suitable spatial resolution (100 m) (27). For the inverse distance weighting approach, we used the inverse square distance method by using the 3 closest monitoring stations within 25 km of each grid cell to calculate the monthly mean concentration for each air pollutant. To obtain postcode-level pollutant concentrations, we integrated the monthly air pollution data with the postcode area for each grid cell and then assigned it to individual women using their own postcode number. The postal code was typically representative of 1 block in urban areas but was larger in rural areas.

The average concentrations were set as the daily maximum of the 8-hour period from 10:00 AM to 6:00 PM for ozone and the 24-hour average concentrations for carbon monoxide, nitrogen dioxide, PM10, and sulfur dioxide for the duration of pregnancies during 2000–2007. We averaged the air pollution data over the first trimester (1–3 months), second trimester (4–6 months), and third trimester (7 months to birth) of gestation based on the birth date and gestational age reported on the birth registry.

Covariates

The covariates constructed from the routinely available birth registration data included maternal age (<20, 20–24, or ≥24 years), sex of the infant (male or female), season of conception (spring, summer, fall, or winter), and year of conception. We obtained salary data from the Bureau of National Health Insurance to calculate the average annual household income in each postcode area and assigned this to subjects by postcode. We stratified the socioeconomic status of each postcode area into quartiles: ≥75th percentile, 75th–50th percentile, 50th–25th percentile, and <25th percentile.

Statistical methods

For the dichotomous outcome variable, we applied a logistic regression model with and without adjustment for the covariates to explore the association among air pollution, pregnancy complications, and preterm births in the 3 trimesters of pregnancy. We identified adjustment factors, including the covariates listed above, that were associated with preterm birth or air pollution based on χ2 tests and t tests. Further, we also selected adjustment factors a priori and included them in the final model.

Single-pollutant models were fitted to estimate the association of an individual pollutant with preterm birth. We also fitted multipollutant models to examine the association of ozone exposure with preterm birth while controlling for other pollutants. However, we did not include carbon monoxide and nitrogen dioxide data in the same model because of their high collinearity (r = 0.83).

The association of each pollutant with the risk of preterm birth was estimated as the odds ratio and 95% confidence interval per each 10-ppb change for ozone and nitrogen dioxide, per each 1-ppb change for sulfur dioxide, per each 100-ppb change for carbon monoxide, and per each 10-μg/m3 change for PM10. We used SAS, version 9.3 (SAS Institute, Inc., Cary, North Carolina) to perform all statistical analyses. Statistical significance was set at P < 0.05 based on a 2-sided test.
As a first step, we selected air pollutants that had significantly positive associations with preterm birth. We hypothesized that common pregnancy complications, such as gestational diabetes mellitus, gestational hypertension, and preeclampsia, could be worsened by exposure to air pollutants. We then performed a stratified analysis to explore the association between the pollutant and preterm birth by 2 levels of pregnancy complications (no disease and disease). Secondly, we created an interaction term (exposure × pregnancy complication) to add into the model used Wald’s method to assess the multiplicative interaction. We only assessed the interaction during the second and third trimesters because the general diagnostic periods for gestational hypertension and preeclampsia are after 20 weeks of gestation, and routine screening for gestational diabetes mellitus begins after 24–28 weeks of gestation.

RESULTS
Characteristics of cases and control subjects

The distribution of maternal and infant characteristics in the study population is presented in Table 1. More cases than controls were male (57%), had mothers less than 20 years of age or 35 years of age or older, and were born into a lower quintile of household income.
socioeconomic group (<25th percentile). The odds of preterm delivery were higher for those mothers who experienced pregnancy complications, including gestational diabetes mellitus, gestational hypertension, and preeclampsia (Table 1).

### Air pollution

The distributions of monthly mean air pollutant concentrations and correlations for the 3 trimesters of the pregnancy are presented in Tables 2 and 3. The average concentrations of nitrogen dioxide and carbon monoxide were highly correlated in all 3 trimesters (correlation coefficients ranged from 0.83 to 0.84), reflecting their common emission source (motor vehicles). The concentrations of ozone were moderately associated with PM$_{10}$ concentration ($r = 0.52–0.53$) and positively but weakly correlated with sulfur dioxide concentration ($r = 0.16–0.17$). Ozone was negatively correlated with the mainly traffic-related pollutants (Table 3).

### Air pollution and preterm births

Figure 1 shows the association estimates for preterm birth during the 3 trimesters in the single-pollutant model. A 10-ppb increase in ozone level was significantly positively associated with preterm birth during the second trimester ($r = 0.23$).

#### Table 2. Distribution of Average Air Pollution Concentrations During Each Trimester of Pregnancy in the Study Population ($n = 431,120$), Taiwan, 2001–2007

<table>
<thead>
<tr>
<th>Pollutant and Trimester</th>
<th>Mean (SD)</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Interquartile Range$^a$</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>8-Hour maximum ozone</strong>, ppb</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>First</td>
<td>42.74 (9.33)</td>
<td>17.40</td>
<td>77.68</td>
<td>38.45–48.22</td>
</tr>
<tr>
<td>Second</td>
<td>42.94 (9.27)</td>
<td>17.40</td>
<td>77.68</td>
<td>36.72–48.43</td>
</tr>
<tr>
<td>Third</td>
<td>43.30 (9.08)</td>
<td>14.11</td>
<td>86.55</td>
<td>37.14–48.98</td>
</tr>
<tr>
<td><strong>24-Hour average nitrogen dioxide</strong>, ppb</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>First</td>
<td>21.60 (6.58)</td>
<td>1.11</td>
<td>47.78</td>
<td>16.84–25.92</td>
</tr>
<tr>
<td>Second</td>
<td>21.18 (6.60)</td>
<td>1.11</td>
<td>47.78</td>
<td>16.27–25.60</td>
</tr>
<tr>
<td>Third</td>
<td>21.09 (6.42)</td>
<td>1.02</td>
<td>47.78</td>
<td>16.37–25.41</td>
</tr>
<tr>
<td><strong>24-Hour average carbon monoxide</strong>, ppm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>First</td>
<td>0.68 (0.23)</td>
<td>0.08</td>
<td>2.56</td>
<td>0.52–0.79</td>
</tr>
<tr>
<td>Second</td>
<td>0.66 (0.23)</td>
<td>0.07</td>
<td>2.35</td>
<td>0.51–0.78</td>
</tr>
<tr>
<td>Third</td>
<td>0.66 (0.22)</td>
<td>0.07</td>
<td>2.35</td>
<td>0.50–0.77</td>
</tr>
<tr>
<td><strong>24-Hour average PM$_{10}$, µg/m$^3$</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>First</td>
<td>59.81 (19.36)</td>
<td>20.06</td>
<td>135.80</td>
<td>45.36–70.40</td>
</tr>
<tr>
<td>Second</td>
<td>58.60 (18.97)</td>
<td>19.13</td>
<td>135.80</td>
<td>44.81–68.22</td>
</tr>
<tr>
<td>Third</td>
<td>59.10 (18.29)</td>
<td>17.51</td>
<td>144.27</td>
<td>45.98–68.32</td>
</tr>
<tr>
<td><strong>24-Hour average sulfur dioxide</strong>, ppb</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>First</td>
<td>4.56 (2.22)</td>
<td>0.18</td>
<td>18.91</td>
<td>3.19–5.36</td>
</tr>
<tr>
<td>Second</td>
<td>4.52 (2.18)</td>
<td>0.18</td>
<td>18.91</td>
<td>3.16–5.33</td>
</tr>
<tr>
<td>Third</td>
<td>4.52 (2.14)</td>
<td>0.15</td>
<td>19.55</td>
<td>3.21–5.30</td>
</tr>
</tbody>
</table>

$^a$ First quartile to third quartile.

#### Table 3. Correlations for Average Air Pollutants Concentrations During Each Trimester of Pregnancy in the Study Population ($n = 431,120$), Taiwan, 2001–2007

<table>
<thead>
<tr>
<th>Pollutant</th>
<th>First Trimester</th>
<th>Second Trimester</th>
<th>Third Trimester</th>
</tr>
</thead>
<tbody>
<tr>
<td>CO, ppm</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>NO$_x$, ppm</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>O$_3$, ppb</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>PM$_{10}$, µg/m$^3$</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
</tr>
<tr>
<td>SO$_2$, ppb</td>
<td>1.00</td>
<td>1.00</td>
<td>1.00</td>
</tr>
</tbody>
</table>

Abbreviations: CO, carbon monoxide; NO$_x$, nitrogen dioxide; O$_3$, ozone; PM$_{10}$, particulate matter with an aerodynamic diameter of 10 µm or less; SO$_2$, sulfur dioxide.
with preterm birth; the adjusted odds ratio was 1.03 (95% confidence interval (CI): 1.02, 1.04) for the first trimester, 1.02 (95% CI: 1.01, 1.02) for the second trimester, and 1.02 (95% CI: 1.01, 1.03) for the third trimester. Each 10-µg/m³ increase in PM10 exposure was slightly associated with a 1% increase in the risk of preterm birth in the first trimester but was negatively associated during the second and the third trimesters. Negative associations were found for traffic-related pollutants (carbon monoxide and nitrogen dioxide), but no association was found with sulfur dioxide in any of the trimesters (Figure 1).

Figure 2 presents the relationship between ozone concentration and preterm birth in 9 multipollutant models. In the 2-pollutant models (models 1, 2, and 3), the association estimates for ozone while controlling for other pollutants ranged from a 1% to 4% increase in the risk of preterm birth, and the relationship remained significant during each trimester. We found a similar association during all 3 trimesters; the association estimates of ozone with combinations of the other 2 pollutants in the 3-pollutant models (model 5 and model 6). However, the associations between ozone concentration and preterm birth were diluted in models 7, 8, and 9 (Figure 2).

Table 4 shows the association of ozone exposure with preterm birth for 2 different levels of pregnancy complications. In a stratified analysis that was adjusted for maternal age, infant sex, season of conception, and year of conception, the estimates for the association of ozone exposure with preterm birth among women who developed gestational diabetes mellitus (in the second trimester, adjusted odds ratio (OR) = 1.15, 95% CI: 1.05, 1.27; in the third trimester, adjusted OR = 1.12, 95% CI: 1.01, 1.23) were higher than for women who did not develop gestational diabetes mellitus. We found an apparent effect modification (P for interaction <0.01) between ozone exposure and gestational diabetes mellitus on the risk of preterm birth during the second and third trimesters. However, we did not find any interaction with gestational hypertension and preeclampsia (Table 4).

DISCUSSION

In the present study, we used a large population base to investigate the association between air pollution and preterm birth and examined whether the association varied in the presence of pregnancy complications. We found that each 10-ppb increase in ozone exposure during the 3 trimesters was associated with a 2%–3% increase in the risk of preterm birth. Our results showed a risk increase of 1%–4% in the odds of preterm birth per each 10-ppb increase in ozone level after adjustment for copollutant exposure in the multipollutant models. We also found that exposure to elevated concentrations of ozone during the second and third trimesters of pregnancy was associated with a higher risk of preterm birth among those women with gestational diabetes mellitus. More than 99% of pregnant Taiwanese women have free prenatal care comprising at least 10 prenatal care visits during pregnancy. Pregnancy complications are mostly diagnosed by obstetricians and gynecologists. We excluded infants with birth defects and those whose mothers had chronic diseases.
from the case and control subjects to reduce the possible influence of those factors on the risk of preterm birth. There are other potential confounding factors for which we did not adjust in the present study, such as maternal nutrition, body mass index, stress level, and occupational, behavioral, and environmental factors (28). This was due to the absence of such information in the Taiwanese birth registry database. Although some of the potential factors above might have regional variation, we adjusted for a postcode-level measure of socioeconomic status to control for unknown local factors. However, we cannot rule out confounding from other factors, such as other environmental toxicants and indoor air quality.

Women in Taiwan have a lower prevalence of smoking (<4%) during pregnancy than women in many other countries (29, 30). In the present study, we found that the prevalence of smoking during pregnancy was probably underreported in the records (0.1% among mothers of cases and <0.03% among mothers of controls), which could have affected the results.

An important limitation of this study was uncertainty in the classification of personal exposures when relying on data from ambient air pollution monitoring stations. We used the inverse distance weighting approach to estimate air pollution concentration based on residential postcode rather than address during pregnancy, which may have increased the possibility of random exposure misclassification. This method is also best for secondary pollutants that vary on a larger geographical scale and are more homogeneously distributed (e.g., ozone) than for those that are more localized and dependent on combustible sources (e.g., carbon monoxide, nitrogen dioxide) (17, 31). In a previous study, Navidi and Lurmann (32) reported that using municipal-level air pollution monitoring data to estimate personal exposure could underestimate the associations when compared with results using data obtained at individual level.

As in most previous studies, we did not have data about maternal residential mobility during pregnancy. However, 2 previous studies in New York (33) and Canada (34) found that most of the mothers who did move only moved short distances, and the majority remained in the same exposure region, so the exposure misclassification due to mobility was minimal. Whether this was also the case in Taiwan is unclear. One previous study reported a slightly strengthened association between exposure air pollution and preterm birth in women who had not moved during pregnancy (21). The present study shows a positive association between ozone concentration and preterm birth throughout pregnancy. These results are inconsistent with the findings of 6 previous studies from Australia (16, 17), California (20, 21), Canada (18), and Sweden (19). In 3 previous studies (18, 20, 21), no association between ozone exposure and preterm birth during the pregnancy was reported. Other studies reported that the risk of
preterm birth during the first trimester was significantly associated with ozone exposure (per 7.1 ppb, OR = 1.26, 95% CI: 1.10, 1.45; per 1 ppb, OR = 1.01, 95% CI: 1.01, 1.02; per 10 µg/m³, OR = 1.04, 95% CI: 1.01, 1.08) (16, 17, 19), but only 2 of the studies above reported no association in the third trimester (16, 17). Because the concentration of ozone has gradually increased in the past decade in Taiwan, a possible reason might be the higher mean concentration of ozone level (an 8-hour average ozone level of approximately 43 ppb) in Taiwan compared with the concentrations those countries mentioned above (a 1-hour maximum ozone level of approximately 31 ppb).

Our results show a negative association between traffic-related pollutants (carbon monoxide and nitrogen dioxide) and the risk of preterm birth. This was possibly due to an inverse correlation with ozone. In the multipollutant models, the association with ozone remained significant except when carbon monoxide or nitrogen dioxide were added to the model.

Atmospheric pollution is a well-known risk factor in complications in human reproduction, particularly during pregnancy (8, 35). In an animal experiment, Kavlock et al. (15) reported that exposure to ozone late in gestation in rats led to abnormal reproduction. However, it is still unclear whether exposure to ozone in all 3 trimesters at pregnancy might influence preterm birth. A possible explanation is that exposure to ozone might increase the release of proinflammatory mediators from the airway epithelial cells, macrophages, and monocytes, which is associated with preterm birth (36–38). For example, asthma is an inflammatory disease, and ozone exposure might lead to inflammation and asthma exacerbation. In 1 previous study, Shiloh et al. (39) reported that a history of gestational diabetes mellitus in women might be associated with upper respiratory infection, which could exacerbate the risk due to exposure to air pollution.

Oxidative stress has been identified as the toxic effect of air pollution that is most relevant for adverse health effects and reproductive outcomes (35, 40, 41). Two toxicology studies reported that ozone exposure might increase levels of lipid peroxidation products and inflammatory cytokines (42, 43).

Ozone is a known strong oxidizing agent that can generate hydrogen peroxide, hydroxyl radicals, and superoxides (9), which might influence the rate of preterm birth.

The present study suggests that exposure to ozone in outdoor air may increase the risk of preterm births over any of the trimesters of pregnancy. Our finding also indicates that the association of ozone with preterm birth is modified by whether the mother develops gestational diabetes mellitus.

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Author affiliations: Department of Public Health, College of Public Health, China Medical University, Taichung, Taiwan (Yu-Ting Lin, Chau-Ren Jung); Institute of Epidemiology and Preventive Medicine, College of Public Health, National Taiwan University, Taipei, Taiwan (Yungling Leo Lee); and Department of Occupational Safety and Health, College of Public Health, China Medical University, Taichung, Taiwan (Yu-Ting Lin, Chau-Ren Jung, Bing-Fang Hwang).

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Conflict of interest: none declared.

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