Air pollution is an environmental risk factor of concern in urban centers all over the world because of widely known adverse effects on human health. Many studies reported the strong association between air pollution and cardiovascular and respiratory diseases (Schwartz, 2006; Pope, Ezzati, & Dockery, 2009; Dockery, 2009), adverse perinatal outcome such as low birth weight, miscarriages, preterm birth (Gouveia, Bremner, & Novaes, 2004; Mohorovic, Petrovic, Haller, & Micovic, 2010; Calderon-Garciduenas et al., 2011), impaired neurodevelopment in children (Yorifuji et al., 2016), poorer semen quality (Lafuente, Garcia-Blaquez, Jacquemin, & Checa, 2016), or change in secondary sex ratio (Maraglia et al., 2013; Lichtenfels et al., 2007).

Worldwide, the human sex ratio at birth is fairly consistent—the male proportion of all births being 51.4% (James, 1996). Studies indicate that in many countries the proportion of male births has been declining during the past five decades (Allan, Brant, Seidel, & Jarrell, 1997; van der Pal-de Bruin, Verloove-Vanhorick, & Roeleveld, 1997; Marcus et al., 1998; Møller, 1998; Parazzini, La Vecchia, Levi, & Franceschi, 1998). It has been suggested that the declining proportion of male births may be attributed by human exposure to air pollution; however due to limited data on this issue, the obtained results should be confirmed in longitudinal studies.
were liquefied at 37ºC for 20 min before analysis. The semen samples after 2–3 days of abstinence. Samples were collected into sterile containers. Participants were instructed to collect semen samples on the same day, full details of the parent study have been described elsewhere (Radwan et al., 2015). Study group comprised of 195 subjects. Men with normal sperm concentration (> 15 mln/ml) (WHO, 1999) were enrolled into the study. A sampling was calculated in order to represent the average and maximum average temperature were analyzed. Each participant was assigned a grid location according to their ZIP code of residence. For each pollutant, the average value for the 90 days preceding semen sampling was calculated in order to represent the average exposure to which those sperm had been exposed.

Materials and Methods

Study Population and Semen Analysis

The study was performed according to the Declaration of Helsinki, and the procedures were approved by the local ethics committee. Men with normal sperm concentration (> 15 mln/ml) (WHO, 1999) were enrolled into the study from infertility clinic between 2008 and 2011 (Jurewicz et al., 2014). Study group comprised of 195 subjects. Questionnaires were performed by trained interviewers who recorded general participant characteristics, including demographic and lifestyle factors. Study participants provided urine, saliva, and semen samples on the same day. Full details of the parent study have been described elsewhere (Radwan et al., 2015).

Semen samples were collected by masturbation into sterile containers. Participants were instructed to collect the semen samples after 2–3 days of abstinence. Samples were liquefied at 37ºC for 20 min before analysis. The spermia, pH, color, and viscosity were determined for each sample. The rationale and methods of semen analysis have been previously described (Jurewicz et al., 2014).

Sperm cells were assayed for Y- or X-bearing cells using fluorescent in situ hybridization as described previously by Jurewicz et al. (2016). Briefly, on the day of FISH procedure the sperm suspensions were spread onto clean glass slides and air-dried. The slides were then washed in 2× standard saline citrate (SSC) solution and were incubated at room temperature for 30 min in 0.1mol/l Tris–HCl buffer containing 10 mmol/l dithiothreitol in order to allow DNA decondensation. Slides were then washed twice in 2× SSC, dehydrated in an ethanol series (70%, 85%, 100%) and air-dried. The slides were viewed by fluorescence microscopy Nikon Eclypsy 80i equipped with LUCIA Cytogenetics-Karyo/FISH software. About 1,000 spermatozoa for each man were analyzed by FISH.

Air Quality Data and Exposure Assessment

Indices of air pollution were obtained from the AirBase database (http://www.eea.europa.eu/data-and-maps/data/airbase-the-european-air-quality-database-6#tab-data-by-country). AirBase is the air quality information system maintained by the European Environmental Agency through the European topic center on air pollution and climate change mitigation. For the purpose of this study, daily levels of particulate matter >10 μm in aerodynamic diameter (PM₁₀), particulate matter <10 μm in aerodynamic diameter (PM₂.₅), and sulfur dioxide (SO₂) (reported as μg/m³), the data represent the 24-hr average, carbon monoxide CO (reported as μg/m³) and ozone (reported as μg/m³), the data represent the maximum 8-hr average, nitrogen dioxide NO₂ (reported as μg/m³) the data represent the maximum 1-hr average and minimum average and maximum average temperature were analyzed. Each participant was assigned a grid location according to their ZIP code of residence. For each pollutant, the average value for the 90 days preceding semen sampling was calculated in order to represent the average exposure to which those sperm had been exposed.

Statistical Analysis

In linear regression models, the effects of exposure to examined air pollutants on the fraction of Y chromosomes were evaluated using R program (R Development Core Team 2013, R Foundation for Statistical Computing, Vienna, Austria. ISBN 3-900051-07-0; URL: http://www.R-project.org) (R Core Team, 2012). A p-value of .05 was defined as the level of statistical significance.

Age (years), current smoking (yes/no), sexual abstinence (days), past diseases (yes/no), and alcohol...
consumption (none or less than 1 drink per week, 1 to 3 drinks per week, 4 to 7 drinks per week) were suggested to affect sex ratio (Hilsenrath, Swarup, Bischoff, Buster, & Carson, 1997; Fukuda, Fukuda, Shimizu, Andersen, & Byskov, 2002), and were therefore considered as potential confounders. If these variables were associated with the Y chromosome fraction (p < .20) they were included in the models, one at a time, together with the exposure variable.

Results

Table 1 presents the characteristics of the patients enrolled in this study. Most of the participants had secondary (39.5%; n = 77) and higher (36.9%; n = 72) education, and were nonsmokers (71%; n = 138) (verification was based on the level of cotinine in saliva). The mean age of men participating in this study was 32 years (Table 1).

Table 2 reports the semen quality, sex ratio, and the level of air pollution among study participants. The semen quality among study participants was in normal range of WHO reference value for semen analysis. The percentage of sperm with abnormal morphology was 51.78%. The fraction of Y was 51.5% ± 1.85.

The average PM<sub>2.5</sub>, PM<sub>10</sub>, SO<sub>2</sub>, NO<sub>x</sub>, and CO concentrations during the study periods were 30.15 μg/m<sup>3</sup> (range: 8.98–89.45 μg/m<sup>3</sup>), 37.07 μg/m<sup>3</sup> (range: 7.56–115.60 μg/m<sup>3</sup>), 40.53 μg/m<sup>3</sup> (range: 10.37–160.47 μg/m<sup>3</sup>), 36.70 μg/m<sup>3</sup> (range: 2.80–170.10 μg/m<sup>3</sup>), and 0.60 μg/m<sup>3</sup> (range: 0.15–1.83 μg/m<sup>3</sup>), respectively. The average O<sub>3</sub> concentration was 43.60 μg/m<sup>3</sup> (range: 11.96–87.54 μg/m<sup>3</sup>). Additionally individual values occasionally exceeded air quality standards. The statistically significant negative associations were observed between exposure to PM<sub>10</sub> and CO and the proportion of Y/X chromosome bearing sperm (p = .039 and p = .045) (Table 3). A p-value of .05 was defined as the level of statistical significance.

After adjusting for smoking, abstinence, past diseases, age, and alcohol consumption the significant reduction was observed in the proportion of Y/X chromosome bearing sperm and exposure to PM<sub>10</sub> (p = .009) and PM<sub>2.5</sub> (p = .023) (Table 3).

Discussion

The main results of the present study were a negative association between exposure to PM<sub>10</sub> and PM<sub>2.5</sub> and the proportion of Y/X chromosome bearing sperm. As this is the first such study, the comparisons of the findings may be difficult. In the study performed in Sao Paulo, Brazil, a significant negative association between particular matter PM<sub>10</sub> and secondary sex ratio (fewer male births) was observed. In the same study an animal model with male mice identified a significant reduction in the secondary sex ratio suggesting that ambient air pollution may interfere with sex distribution by altering the X:Y sperm proportion in pollution-exposed males (Lichtenfels et al., 2007). The negative significant association between PM<sub>10</sub> and male to female ratio was also observed in the next study performed in Sao Paulo (Miraglia et al., 2013).

Among studies conducted in Scotland and Taiwan two studies reported a lower sex ratio at birth in residential areas at risk from air pollution (Williams et al., 1992; Yang et al., 2000a) whereas the other two studies (Williams et al., 1995; Yang et al., 2000b) reported no convincing evidence that exposure to airborne pollution was associated with sex ratio.

Potential toxicological mechanisms that might explain and give strength to the environmental contamination

---

### Table 1. Characteristics of the Study Population.

| Characteristic                                | N = 195 |
|-----------------------------------------------|---------|
| **Education, n (%)**                          |         |
| Vocational                                    | 46 (23.6) |
| Secondary                                     | 77 (39.5) |
| Higher                                        | 72 (36.9) |
| **Smoking determined by cotinine level, n (%)**|         |
| No                                            | 138 (70.8) |
| Yes                                           | 54 (27.7) |
| Missing data                                  | 3 (1.5) |
| **Alcohol use, n (%)**                        |         |
| None or <1 drink/week                         | 88 (45.13) |
| 1–3 drinks/week                               | 100 (51.28) |
| Everyday                                      | 7 (3.59) |
| **Past diseases, which may have impact on semen quality, n (%)** |         |
| No                                            | 168 (86.2) |
| Yes                                           | 27 (13.8) |
| **Duration of couple’s infertility [years], n (%)** |         |
| 1–2                                          | 74 (37.9) |
| 2–3                                          | 61 (31.3) |
| 3–5                                          | 31 (15.9) |
| >5                                           | 29 (14.9) |
| **Abstinence [days], n (%)**                   |         |
| <3                                           | 24 (12.3) |
| 3–7                                          | 139 (71.3) |
| >7                                           | 32 (16.4) |
| mean (SD)                                     | 5.2 ± 2.5 |
| median (min–max)                              | 5.0 (0.0–20.0) |
| **Age (years)**                               |         |
| mean (SD)                                     | 32.2 (4.7) |
| median (min–max)                              | 31.9 (22.7–44.8) |

Note. N = number of participants; SD = standard deviation; min = minimum; max = maximum; mean = arithmetic mean; median = the value separating the higher half of a population, or a distribution from the lower half.
causes in the determination of the sex ratio are still inconclusive.

The biological plausibility for these epidemiological observations may be associated with the fact that air pollution may interfere in sex distribution by altering the testicular functioning as a whole leading to excess of X sperm production in exposed males. The experimental study with male mice raised in nonfiltered open-top chambers identified a significant reduction in secondary sex ratio suggesting that ambient air pollution may interfere with sex distribution by altering the X:Y sperm production in pollution-exposed males (Lichtenfels et al., 2007). The testicular histological analysis also reported a significant reduction in sperm count, total number of sperm cells, and elongated spermatids. In addition, a decrease in sperm concentration in the caudal proportion of the epididymus was identified in the exposed mice (Lichtenfels et al., 2007). But there are many paths to pursue to clarify the mechanisms responsible for the findings in the current study.

When evaluating the results of the current study, several potential biases need to be considered. The men in this study were from a fertility clinic as opposed to the general population, but they have normal sperm concentration [>15 mln/ml (WHO, 1999)]. Although participants may differ from men in the general population, there is no evidence that they would differ in their response to air pollution. The results may apply to general population samples as well. Participants were heterogeneous in their semen profiles and had normal semen parameters, so the selection bias is of minor concern. Additionally the residual confounding is probably not an issue of great concern, as all potential confounders known to us in the analysis have been considered. Imperfect measurements of the confounders that have caused some residual confounding cannot be excluded. But the measurements of some confounders were confirmed by assessment of biomarker of exposure, like in case of smoking that was verified by measuring the cotinine level in saliva. Other limitations of the study need to be addressed.

The lack of hybridization was only in case of 2% of spermatozoa. Overall hybridization efficiency was high from 98.91% to 99.36% which is higher with the hybridization efficiency (97%) reported by other groups (Johannisson et al., 2002; Martin, 2006).

This is the first study to evaluate the impact of air pollution exposure on sex chromosome ratio in sperm and the results indicate that sperm Y:X ratio is influenced by exposure to air pollution. The design of the study does not allow for the clarification of mechanisms behind the observed association.

### Table 2. Sex Ratio, Main Semen Parameters, and the Level of Air Pollution.

| Variables                      | Median | Min  | Max  | Mean | SD  | N  |
|-------------------------------|--------|------|------|------|-----|----|
| Fraction of Y (%)             | 51.50  | 44.80| 57.9 | 51.5 | 1.85| 195|
| Concentration (10^6/ml)       | 48.30  | 15.00| 360.0| 64.81| 55.73| 195|
| Motility (%)                  | 55     | 20   | 99   | 57.88| 19.8 | 195|
| Abnormal morphology (%)       | 50     | 15   | 97.00| 51.78| 20.1 | 195|
| Ozone (µg/m³)                 | 41.01  | 11.96| 87.54| 43.60| 19.33| 195|
| PM₁₀ (µg/m³)                  | 34.00  | 7.56 | 115.60| 37.07| 30.60| 195|
| PM₂·₅ (µg/m³)                 | 26.34  | 8.98 | 89.45| 30.15| 20.16| 195|
| SO₂ (µg/m³)                   | 40.12  | 10.37| 160.47| 40.53| 16.22| 195|
| NO₂ (µg/m³)                   | 29.54  | 2.80 | 170.10| 37.22| 37.22| 195|
| CO (µg/m³)                    | 0.55   | 0.15 | 1.83 | 0.60 | 0.45 | 195|

*Note.* NOx = nitrogen dioxide; SO₂ = sulfur dioxide; CO = carbon monoxide; PM₁₀ = particulate matter >10 mg/m³ in aerodynamic diameter; PM₂·₅ = particulate matter <10 mg/m³ in aerodynamic diameter.

### Table 3. The Association Between Air Pollutants and the Proportion of Y/X Chromosome Bearing Sperm.

| Selected chemicals | Coef.  | 95% CI            | p-Value |
|--------------------|--------|-------------------|---------|
| Ozone crude        | 0.189  | [−0.164, 0.542]   | .296    |
| Ozone adjusted     | 0.176  | [−0.240, 0.592]   | .408    |
| PM₁₀ crude         | −0.394 | [−0.766, −0.022]  | .039    |
| PM₁₀ adjusted      | −0.552 | [−0.961, −0.142]  | .009    |
| PM₂·₅ crude        | −0.343 | [−0.769, 0.083]   | .116    |
| PM₂·₅ adjusted     | −0.559 | [−1.036, −0.081]  | .023    |
| SO₂ crude          | 0.029  | [−0.091, 0.149]   | .635    |
| SO₂ adjusted       | −0.016 | [−0.149, 0.117]   | .812    |
| NO₉ crude          | −0.211 | [−0.502, 0.081]   | .158    |
| NO₉ adjusted       | −0.159 | [−0.482, 0.164]   | .335    |
| CO crude           | −2.571 | [−5.070, −0.071]  | .045    |
| CO adjusted        | −2.759 | [−5.621, 0.103]   | .061    |

*Note.* NOx = nitrogen dioxide; SO₂ = sulfur dioxide; CO = carbon monoxide; PM₁₀ = particulate matter >10 mg/m³ in aerodynamic diameter; PM₂·₅ = particulate matter <10 mg/m³ in aerodynamic diameter; Coef. = β coefficient; CI = confidence interval.

*Adjusted for:* smoking, abstinence, past diseases, age, alcohol consumption.
In conclusion, the observed effects of a lower Y:X sperm chromosome ratio among men exposed to air pollution support the evidence that the trend of declining sex ratio in several societies over past decades has been due to exposure to air pollution. These findings are evidence that air pollution exposure can result in selective genetic pressure in human males (X- vs. Y-bearing sperm) and that this could be expressed as changed sex ratios in offspring. Elucidation of the intratesticular mechanisms affecting the distribution of Y and X sperm may not only add to understanding of the biological mechanisms regulating the offspring sex ratio but also contribute to better understanding of the process of spermatogenesis. Patterns of reduced sex ratio need to be carefully assessed to determine whether they occur more generally or whether temporal or spatial variations are evident.

Declaration of Conflicting Interests
The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding
The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This study was performed under the project “The association between environmental exposure to widespread man made endocrine disrupting chemicals and level of hormones associated with the activity of hypothalamic pituitary testicular axis among young men” supported by National Science Centre in Poland from grant no. UMO-2014/13/B/NZ7/02223 and the project financed with a grant for statutory activity IMP 10.23/2015.

References
Allan, B. B., Brant, R., Seidel, J. E., & Jarrell, J. F. (1997). Declining sex ratios in Canada. Canadian Medical Association Journal, 156(1), 37–41.
Calderon-Garciduenas, L., Engle, R., Mora-Tiscareno, A., Stynner, M., Gómez-Garza, G., Zhu, H., … D’Angiulli, A. (2011). Exposure to severe urban air pollution influences cognitive outcomes, brain volume and systemic inflammation in clinically healthy children. Brain and Cognition, 77(3), 345–355.
Davis, D. L., Gottlieb, M. B., & Stampnitzky, J. R. (1998). Reduced ratio of male to female births in several industrial countries- a sentinel health indicators? Journal of the American Medical Association, 279(13), 1018–1023.
Dockery, D. W. (2009). Health effects of particulate air pollution. Annals of Epidemiology, 19(4), 257–263.
Fukuda, M., Fukuda, K., Shimizu, T., Andersen, C. Y., & Byskov, A. G. (2002). Parental periconceptional smoking and male: Female ratio of newborn infants. The Lancet, 359(9315), 1407–1408.
Gouveia, N., Bremner, S. A., & Novaes, H. M. D. (2004). Association between ambient air pollution and birth weight in São Paulo, Brazil. Journal of Epidemiology and Community Health, 58(1), 11–17.
Hilsenrath, R. E., Swarup, M., Bischoff, F. Z., Buster, J. E., & Carson, S. A. (1997). Effect of sexual abstinence on the proportion of X-bearing sperm as assessed by multicolor fluorescent in situ hybridization. Fertility and Sterility, 68(3), 510–513.
James, W. H. (1996). Evidence that mammalian sex ratios at birth are partially controlled by parental hormone levels at the time of conception. The Journal of Theoretical Biology, 180(4), 271–286.
Johannisson, R., Leuschner, E., Huppe, M., Hinrichs, F., Al-Hasani, S., Diedrich, K., … Mennicke, K. (2002). Increased frequency of X-bearing sperm in males from an infertility clinic: Analysis by two-color fluorescence in situ hybridization. Cytogenetic and Genome Research, 98(4), 240–244.
Jurewicz, J., Radwan, M., Sobala, W., Bochenek, M., & Hanke, W. (2014). Lifestyle and semen quality- role of modifiable risk factors. Systems Biology in Reproductive Medicine, 60(1), 43–51.
Jurewicz, J., Radwan, M., Sobala, W., Radwan, P., Jakubowski, L., Wielgomas, B., … Hanke, W. (2016). Exposure to widespread environmental endocrine disrupting chemicals and human sperm sex ratio. Environmental Pollution, 213, 732–740.
Lafuente, R., Garcia-Blazquez, N., Jacquemin, B., & Checa, M. A. (2016). Outdoor air pollution and sperm quality. Fertility and Sterility, 106(4), 880–896.
Lichtenfels, A. J. F. C., Gomes, J. B., Pieri, P. C., Miraglia, S. G. E. K., Hallak, J., & Saldiva, P. H. N. (2007). Increased levels of air pollution and a decrease in the human and mouse male-to-female ratio in Sao Paulo, Brazil. Fertility and Sterility, 87(1), 230–232.
Marcus, M., Kiely, J., Xu, F., McGeehin, M., Jackson, R., & Sinks, T. (1998). Changing sex ratio in the United States, 1969–1995. Fertility and Sterility, 70(2), 270–273.
Martin, R. H. (2006). Meiotic chromosome abnormalities in human spermatogenesis. Reproductive Toxicology, 22(2), 142–147.
Miraglia, S. G. E. K., Veras, M. M., Amato-Lourenço, L. F., Rodrigues-Silva, F., & Saldiva, P. H. N. (2013). Follow-up of the air pollution and the human male-to-female ratio analysis in Sao Paulo, Brazil: A times series study. British Medical Journal Open, 3(7), e002552. doi:10.1136/bmjopen-2013-002552.
Mohorovic, L., Petrovic, O., Haller, H., & Micovic, V. (2010). Pregnancy loss and maternal methemoglobin levels: An indirect explanation of the association of environmental toxics and their adverse effects on the mother and the fetus. International Journal of Environmental Research and Public Health, 7(12), 4203–4212.
Møller, H. (1998). Trends in sex-ratio, testicular cancer and male reproductive hazards: Are they connected? Acta Pathologica, Microbiologica et Immunologica Scandinavica, 106(1–6), 232–238.
Parazzini, F., La Vecchia, C., Levi, F., & Franceschi, S. (1998). Trends in male: Female ratio among newborn infants in
29 countries from five continents. *Human Reproduction, 13*(5), 1394–1396.

Pope, C. A., III, Ezzati, M., & Dockery, D. W. (2009). Fine-particulate air pollution and life expectancy in the United States. *The New England Journal of Medicine, 360*(4), 376–386.

R Core Team. (2012). *R: A language and environment for statistical computing*. Vienna: R Foundation for Statistical Computing. ISBN 3-900051-07-0, Retrieved from http://www.R-project.org/

Radwan, M., Jurewicz, J., Wielgomas, B., Piskunowicz, M., Sobala, W., Radwan, P., ... Hanke, W. (2015). The association between environmental exposure to pyrethroids and sperm aneuploidy. *Chemosphere, 128*, 42–48.

Robbins, W. A., Rubes, J., Selevan, S. G., & Perreault, S. D. (1999). Air pollution and sperm aneuploidy in healthy young men. *International Journal of Hygiene and Environmental Health, 1*, 125–131.

Rubes, J., Selevan, S. G., Evenson, D. P., Zudova, D., Vozdova, M., Zudova, Z., ... Perreault, S. D. (2005). Episodic air pollution is associated with increased DNA fragmentation in human sperm without other changes in semen quality. *Human Reproduction, 20*(10), 2776–2783.

Schwartz, J. (2006). Long-term effects of exposure to particulate air pollution. *Clinics in Occupational and Environmental Medicine, 5*(4), 837–848.

Selevan, S. G., Borkovec, L., Slott, V. L., Zudová, Z., Rubes, J., Evenson, D. P., & Perreault, S. D. (2000). Semen quality and reproductive health of young Czech men exposed to seasonal air pollution. *Environmental Health Perspectives, 108*(9), 887–894.

Šrám, R. J., Beneš, I., Binková, B., Dejmek, J., Horstman, D., Kotešovec, F., ... Lewtas, J. (1996). Teplice program—the impact of air pollution on human health. *Environmental Health Perspectives, 104*(suppl 4), 699–714.

van der Pal-de Bruin, K. M., Verloove-Vanhorick, S. P., & Roeleveld, N. (1997). Change in male: Female ratio among newborn babies in Netherlands. *The Lancet, 349*(9044), 62.

Van Larebeke, N. A., Sasco, A. J., Brophy, T. J., Keith, M. M., Gilbertson, M., & Watterson, A. (2008). Sex ratio changes as sentinel health events of endocrine disruption. *International Journal of Occupational and Environmental Health, 14*(2), 138–143.

Watanabe, N., & Kurita, M. (2001). The masculinization of the fetus during pregnancy due to inhalation of diesel exhaust. *Environmental Health Perspectives, 109*(2), 111–119.

Williams, F. L., Lawson, A. B., & Lloyd, O. L. (1992). Low sex-ratios of births in areas at risk from air-pollution from incinerators, as shown by geographical analysis and 3-dimensional mapping. *International Journal of Epidemiology*, 21(2), 311–319.

Williams, F. L., Ogston, S. A., & Lloyd, O. L. (1995). Sex-ratios of births, mortality, and air-pollution: Can measuring the sex-ratios of births help to identify health-hazards from air-pollution in industrial environments. *Occupational and Environmental Medicine, 52*(3), 164–169.

WHO (World Health Organization). (1999). *WHO laboratory manual for the examination of human semen and sperm-cervical mucus interaction* (4th ed.). Cambridge: Cambridge University Press.

WHO (World Health Organization). (2010). WHO Laboratory Manual for the Examination of Human Semen and Sperm-Cervical Mucus Interaction. 5th ed. Cambridge, UK: World Health Organization, Cambridge University Press.

Yang, C. Y., Tsai, S. S., Cheng, B. H., Hsu, T. Y., & Wu, T. N. (2000a). Sex ratio at birth associated with petrochemical air pollution in Taiwan. *The Bulletin of Environmental Contamination and Toxicology, 65*(1), 126–131.

Yang, C. Y., Cheng, B. H., Hsu, T. Y., Tsai, S. S., Hung, C. F., & Wu, T. N. (2000b). Female lung cancer mortality and sex ratios at birth near a petroleum refinery plant. *Environmental Research, 83*(1), 33–40.

Yorifuji, T., Kashima, S., Diez, M. H., Kado, Y., Sanada, S., & Doi, H. (2016). Prenatal exposure to traffic-related air pollution and child behavioral development milestone delays in Japan. *Epidemiology, 27*(1), 57–65.