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Tracking Trends in Emissions of Developmental Toxicants and Potential Associations with Congenital Heart Disease in Alberta, Canada

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Abstract: Congenital heart disease (CHD) is a serious anomaly for which the etiology remains elusive. We explored temporal trend associations between industrial developmental toxicant (DT) air emissions and CHD in Alberta. Patients born between 2004–2011 with a diagnosis of CHD and 18 DTs from the National Pollutant Release Inventory (2003–2010) were identified. We applied principal component analysis (PCA) to DT amounts and toxicity risk scores (RS) and defined yearly crude CHD and septal defects rates for urban and rural regions. Correlations between DT groups and CHD rates were examined with Spearman test and Bonferroni correction was conducted for multiple comparisons. PCA identified three DT groups: Group 1 (volatile organic compounds (VOCs) and other gases,) Group 2 (other VOCs), and Group 3 (mainly heavy metals). Province-wide, we found associations between Group 1 DTs and CHD and septal defect rates, when using amounts (r = 0.86, CI 0.39, 0.97 and r = 0.89, CI 0.48, 0.98, respectively) and RS (r = 0.88, CI 0.47, 0.98 and r = 0.85, CI 0.36, 0.97, respectively). Rural Group 2 DTs were positively associated with septal defect rates in both amounts released and RS (r = 0.91, CI 0.55, 0.98 and r = 0.91, CI 0.55, 0.98, respectively). In this exploratory study, we found a temporal decrease in emissions and CHD rates in rural regions and a potential positive association between CHD and septal defect rates and mixtures of organic compounds with or without gases.

Keywords: congenital heart disease; planetary health; industrial emissions; air pollution; developmental toxicants

1. Introduction

Congenital heart disease (CHD) is the most common and serious congenital anomaly affecting 1% of all live births and a higher number of conceptions worldwide [1]. CHDs are the most common cause of neonatal death among birth defects and they importantly contribute to mortality and morbidity-related economic costs [2]. Genetic risk factors, such as Mendelian inheritance in some families, and non-syndromic single gene and chromosomal anomalies account for 15% of CHD [3,4]. A further 30% of CHD is thought to be multifactorial and is attributed to recognized non-inherited risk

factors, such as diabetes mellitus, infections, like rubella, and exposures to teratogenic medications [5,6]. However, for more than half of the affected children the cause is not known. It has been long suspected that CHD may, in part, relate to complex interactions between parental environmental exposures with or without a genetic predisposition [7]. The Baltimore–Washington Infant Study was the largest epidemiological study to document a potential role for chemical exposures (domestic and occupational) in the development of CHD [8]. Possible associations between ambient urban air pollutants such as sulphur dioxide, nitrogen dioxide, carbon monoxide, and particulate matter, as well as organic solvents have also been reported by other groups [9,10]. The majority of these investigations have explored associations of single pollutants with very few studies examining the relationship between multipollutant exposures and CHD [11]. This area of study has also been largely limited to the use of a few monitored ambient air pollutants as listed above. Finally, access to regional and national CHD databases has further limited research in this area.

Canada established a National Pollutant Release Inventory (NPRI) [12], a mandatory government registry that maintains annual reports of industrial chemical releases to air, water, soil, disposals and recycling for the whole country. Among the reported pollutants are developmental toxicants (DTs), chemicals believed to have some impact on fetal and childhood development and health, but have not been definitively recognized as cardiac teratogens by the Office of Environmental Health Hazard Assessment Proposition 65 [13].

Alberta is a Canadian province located in Western Canada along the Canada-US border. It spans 661,185 square kilometers and boasts a rich diverse landscape consisting of forests, prairies, the Rocky Mountains, glaciers, lakes and rivers, amongst others. Over the past five decades it has witnessed an exponential population growth from one million people to currently 4.2 million people of whom 80% reside in urban vs. 20% in rural regions [14]. This trend has been attributed to rapid industrialization and accompanying economic opportunities. The footprint from various industrial sectors varies in urban and rural Alberta (professional, scientific, and technical services vs. mining and oil and gas extraction, agricultural and forestry, respectively) [15,16]. Concerns around the exploitation of the oil sands and its impact on the planetary health of the ecosystems, biodiversity, natural landscapes, and human population have been raised [17]. In addition, the Public Health Agency of Canada has reported that the CHD prevalence in Alberta is higher than the national average [18].

Given access to the NPRI which captures toxic releases by industry and the fact that the Province of Alberta has two centralized pediatric cardiology referral centers with a captive population of CHD patients, we conducted an exploratory study to investigate the potential relationship between industrial pollutants and CHD through an ecologic study in Alberta and its urban and rural regions. The aims of this study were two-fold: (1) to track the trends of multipollutant groups of developmental toxicants (DTs) emitted by industry and the trends of CHD, and (2) to explore potential associations between trends of multipollutant groups of DTs and CHD in Alberta and its urban and rural regions.

2. Results

2.1. CHD in Alberta

A total of 2415 CHD infants were born in Alberta between 2004–2011 representing an overall incidence rate of 6.6 per 1000 live births with a slightly higher incidence in rural regions (6.9 per 1000 live births vs. 6.5 per 1000 live births in urban regions). The proportions of all of the embryological groups of CHD are shown in Table 1. Temporally, CHD rates revealed a statistically significant downward trend in the province as a whole, and in its rural regions paralleled by changes specifically in septal defects (Table 2), (Figure 1A,B, respectively). CHD rates did not change significantly in urban regions during this same period. The other embryological groups were small, and no further analysis was attempted.

Table 1. Proportions of CHD in Alberta 2004–2011.

Embryological Group	Count (n = 2415)	Percentage (%)	Prevalence (per 1000 Live Births)
Septal	1320	54.7	3.67
LHO	360	14.9	1.00
Conotruncus	263	10.9	0.73
RHO	220	9.1	0.61
AVSD	109	4.5	0.30
PDA	48	1.9	0.13
Heterotaxy	34	1.4	0.09
APVR	34	1.4	0.09
Complex/SV	21	0.9	0.06
Other	6	0.2	0.002

APVR = anomalous pulmonary venous return, AVSD = atrio-ventricular septal defect, LHO = left heart obstruction, PDA = patent ductus arteriosus, RHO = right heart obstruction, SV = single ventricle, other = abnormal valves (3), anomalous left coronary from pulmonary artery (2) and pulmonary vein stenosis (1).

Table 2. Trends of CHD and septal defects rates for exposure years 2003–2010 in Alberta and its urban and rural regions.

Variables	Region	Regression Coefficient	95% CI	* *p* Value
CHD Rates	Province	−0.4	−0.6; −0.2	0.005 *
	Rural	−0.3	−0.5; −0.2	0.003 *
	Urban	−0.2	−0.4; 0.1	0.133
Septal Defect Rates	Province	−0.2	−0.3; −0.1	0.012 *
	Rural	−0.2	−0.3; −0.02	0.025 *
	Urban	−0.1	−0.3; 0.03	0.105

* p value $\leq$ 0.05 = significant.

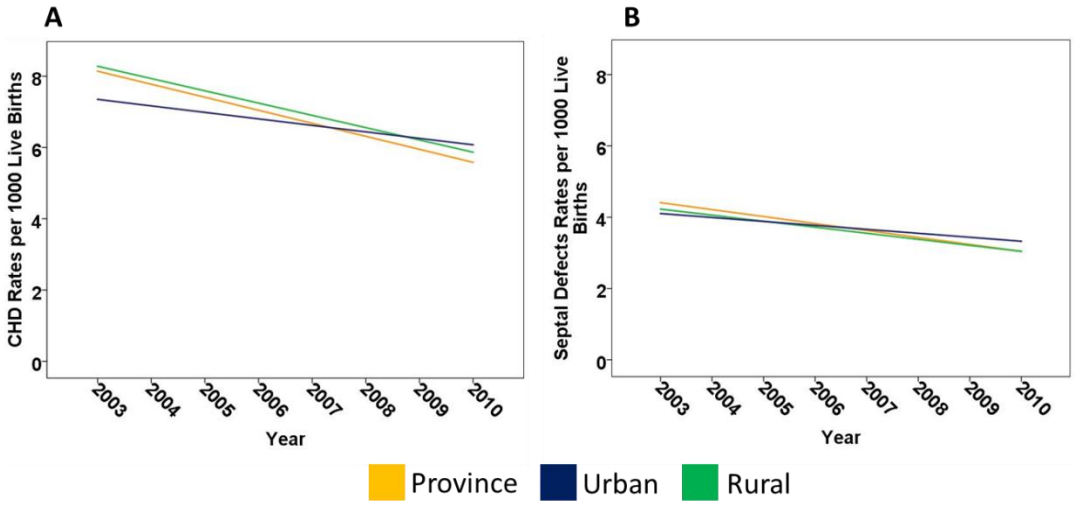

Figure 1. (**A**) Crude CHD rates in the province and rural regions show a significant decreasing trend, (* p = 0.005 and 0.003, respectively) not found in urban regions (p = 0.133); and (**B**) crude septal defects rates showed a significant downward trend in the province and rural regions (* p = 0.012 and 0.025, respectively), but not urban regions (p = 0.105).

2.2. Developmental Toxicants in Alberta

Emission proportions in absolute amount and risk score: Of the 139 reported chemicals emitted to air in Alberta, 18 were DTs, representing 51% of the provincial emissions. Of the 18 DTs, 59% of the amounts in tonnes were emitted by facilities located in rural areas and 40.3% in urban areas. There was

a total of 3537 developmental toxicant emitting facilities (DTEF) in the province, 2700 (76%) in rural and 837 (24%) in urban regions for the study period. The PCA matrix revealed three groups of DTs which were selected based on the correlation coefficient ≥0.6 (Table 3). Group 1 predominantly had four volatile organic compounds (VOCs) and two gases; Group 2 had six other VOCs; and Group 3 consisted primarily of four heavy metals. Group 1 DTs represented the largest group emitted in Alberta (urban and rural) (Table 4). DT RS were higher in rural compared to urban areas (71.5% vs. 28.5%). RS of Group 3 had the highest proportion of the emitted DTs (province > rural > urban), whilst Group 2 contributed the least.

Table 3. Principal component analysis of 18 developmental toxicants.

Developmental Toxicants	Principal Components		
	Group 1	Group 2	Group 3
Benzene	***0.98***	0.11	0.01
Carbon Disulfide	***0.95***	−0.09	−0.04
Carbon Monoxide	***0.95***	0.21	0.12
Sulphur Dioxide	***0.86***	−0.11	0.47
Toluene	***0.86***	−0.04	0.04
1,3-Butadiene	***0.64***	***0.66***	0.01
Chloroform	0.02	***0.85***	0.04
Ethylene Oxide	0.17	***0.96***	0.06
Methanol	0.03	***0.86***	0.06
Methyl-isobutyl-ketone	0.11	***0.90***	0.05
Trichloroethylene	0.11	***0.79***	0.05
Arsenic	0.16	0.11	***0.95***
Cadmium	0.36	0.06	***0.60***
Hexachlorobenzene	−0.15	−0.05	***0.91***
Lead	0.29	0.29	***0.72***
Mercury	−0.52	−0.08	***0.97***
2-Ethoxyethanol	−0.05	−0.04	−0.08
N-Methyl-2-Pyrrolidone	−0.03	0.04	0.01

DTs with a correlation coefficient ≥0.6 (bold italics) were selected and kept in the principal component they represent. DTs = developmental toxicants.

Table 4. Proportions of emissions by region using amounts and risk scores.

Region	Groups	Amount	%	Risk Score	%
Province	Group 1	4,834,586	99.6	9,773,565	15.2
	Group 2	18,220	0.4	3623	0.01
	Group 3	36	0.00	54,578,189	84.8
Total		4,852,844		64,355,377	
(AV ± SD)		95,153 ± 145,728		2,681,474 ± 2,986,681	
Urban	Group 1	1,946,446	99.4	4,274,371	23.3
	Group 2	11,637	0.6	3031	0.02
	Group 3	15	0.00	14,059,472	76.7
Total		1,958,101	40.3	18,336,874	28.5
(AV ± SD)		92,984 ± 141,235		6,112,291 ± 5,883,839	
Rural	Group 1	2,888,139	99.7	5,499,194	11.9
	Group 2	6583	0.3	592	0.00
	Group 3	21	0.00	40,518,716	88
Total		2,894,743	59.7	46,018,502	71.5
(AV ± SD)		98,669 ± 147,335		15,339,501 ± 17,945,349	

AV = Average, SD = Standard Deviation.

Emission trends of DTs in amounts and RS: DT emissions decreased in total amounts provincially and were driven by the rural emissions (Table 5). Based on total amounts, Group 1 and 2 showed a significant downward trend in rural regions whilst Group 3 showed a significant increasing trend in the whole province (Figure 2A–C). Based on RS, urban regions showed an overall significant increase in DT emissions. Comparing the three groups by RS, Group 1 showed a significant downward trend in the

province driven by rural regions, Group 2 showed a significant decrease in all regions, whilst Group 3 showed a significant increase only in urban regions (Table 5).

Table 5. Trends of developmental toxicant emissions in Alberta, 2003–2010.

Variables	Region	Regression Coefficient	95% CI	* *p* Value
DT Amounts				
Overall	Province	−14,003	−21,446; −6539	0.004 *
	Rural	−9226	−16,016; −2436	0.016
	Urban	−4634	−9337; 69	0.053
Group 1	Province	−13,962	−21,427; −6497	0.004 *
	Rural	−9188	−15,970; −2405	0.016
	Urban	−4658	−9372; 58	0.052
Group 2	Province	−42	−87; 3	0.060
	Rural	−39	−67; −10	0.016
	Urban	23	−18; 63	0.221
Group 3	Province	0.4	0.3; 0.6	<0.001 *
	Rural	0.5	0.05; 0.4	0.021
	Urban	0.2	0.04; 0.3	0.016
DT Risk Scores				
Overall	Province	128,140	−44,647; 300,928	0.120
	Rural	−68,809	−338,413; 200,794	0.555
	Urban	196,950	72,289; 321,611	0.008
Group 1	Province	−33,772	−45,486; −22,057	<0.001 *
	Rural	−29,410	−36,996; −21,824	<0.001 *
	Urban	−4361	−17,512; 8789	0.448
Group 2	Province	−65	−107; −24	0.008
	Rural	−4	−6; −1	0.016 *
	Urban	−62	−102; −22	0.009
Group 3	Province	161,977	−7609; 331,563	0.058
	Rural	−39,396	−305,239; 226,447	0.729
	Urban	201,373	71,479; 331,268	0.009

* p value $\leq$ 0.004 = significant after Bonferroni adjustment.

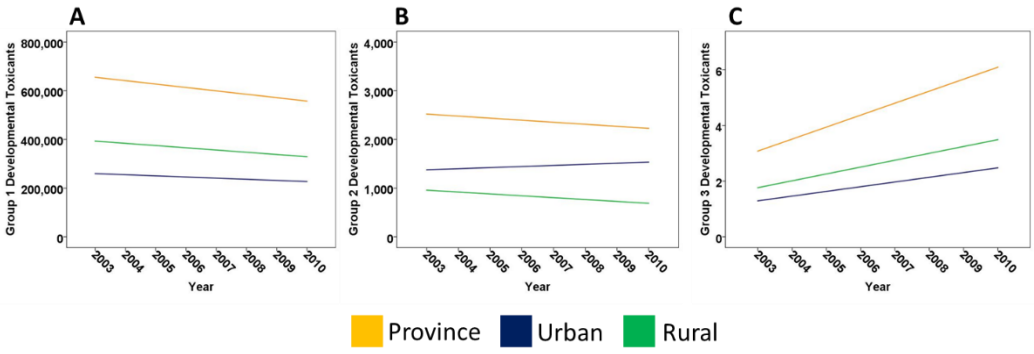

Figure 2. Developmental toxicants in amounts (tonnes). (**A**) Group 1 DTs demonstrated a decreasing trend in the province, (* p = 0.004); (**B**) There was no statistically significant decrease in Group 2 emissions in the province and its urban and rural regions; (**C**) Group 3 DTs showed a statistically significant increase in the province overall, (* p < 0.001). DT = developmental toxicant, * p value $\leq$ 0.004 = significant post Bonferroni adjustment.

2.3. Associations between Groups of DTs and CHD Rates in Alberta

Emissions Amount: There were marginal statistically significant associations between the total amount of the 18 DTs emitted in the province and with CHD rates (r = 0.86, 95% CI: 0.39, 0.97, $p < 0.007$), whilst the total of the remaining 121 chemicals showed no association (r = 0.38, 95% CI: −0.44, 0.86, $p = 0.352$). We found positive associations between Group 1 emissions in the province and septal defect rates. For Group 2 DTs, we found positive associations with septal defects in rural regions, whilst in the urban regions there were negative between Group 3 DTs and CHDs overall (Table 6).

Table 6. Associations of CHD and Septal Defect Rates with DT Groups.

Region	Variable	Spearman's Rho (95% CI)	* p Value	Spearman's Rho (95% CI)	* p Value
DT Amounts					
		CHD		**Septal**	
Province	Group 1	0.86 (0.39, 0.97)	0.007	0.89 (0.48, 0.98)	0.003 *
	Group 2	0.50 (−0.32, 0.89)	0.207	0.79 (0.16, 0.96)	0.023
	Group 3	−0.74 (−0.95, −0.07)	0.037	−0.76 (−0.95, −0.12)	0.031
Rural	Group 1	0.62 (−0.15, 0.92)	0.102	0.60 (−0.19, 0.92)	0.120
	Group 2	0.79 (0.18, 0.96)	0.021	0.91 (0.55, 0.98)	0.002 *
	Group 3	−0.64 (−0.93, 0.11)	0.086	−0.81 (−0.96, −0.24)	0.015
Urban	Group 1	0.71 (0.02, 0.94)	0.047	0.74 (0.07, 0.95)	0.037
	Group 2	−0.07 (−0.74, 0.67)	0.867	−0.02 (−0.72, 0.69)	0.955
	Group 3	−0.88 (−0.98, −0.47)	0.004 *	−0.83 (−0.97, −0.31)	0.010
DT Risk Scores					
		CHD		**Septal**	
Province	Group 1	0.88 (0.47, 0.98)	0.004 *	0.85 (0.36, 0.97)	0.007
	Group 2	0.86 (0.39, 0.97)	0.007	0.97 (0.84, 0.99)	<0.001 *
	Group 3	−0.41 (−0.86, 0.42)	0.320	−0.50 (−0.89, 0.31)	0.204
Rural	Group 1	0.88 (0.47, 0.98)	0.004 *	0.76 (0.12, 0.95)	0.028
	Group 2	0.79 (0.18, 0.96)	0.021	0.91 (0.55, 0.98)	0.002 *
	Group 3	−0.02 (−0.72, 0.69)	0.955	−0.12 (−0.76, 0.64)	0.779
Urban	Group 1	0.69 (−0.03, 0.94)	0.058	0.64 (−0.11, 0.93)	0.086
	Group 2	0.69 (−0.03, 0.94)	0.058	0.86 (0.39, 0.97)	0.007
	Group 3	−0.79 (−0.96, −0.18)	0.021	−0.81 (−0.96, −0.24)	0.015

CHD = congenital heart disease, DT = developmental toxicants, * p value ≤ 0.004 post Bonferroni adjustment.

Risk Scores: Although there were no associations with the total RS of the 18 DTs emitted in the province and CHD, we found positive associations between Group 1 and CHD rates and also with Group 2 and septal defects rates. In the rural regions, we found positive associations with Group 1 and CHD and Group 2 with septal defect rates (Table 6).

3. Discussion

Our exploratory study found important downward air emission trends in both amounts and RS (potential toxicity) which differed between rural and urban regions of Alberta, with a reduction in emissions potentially influencing CHD rates in rural regions only. They reflected province-wide positive associations between Group 1 emissions (benzene, carbon monoxide, carbon disulfide, sulphur dioxide, toluene, and 1,3 butadiene) and CHD and septal defect rates using both amounts and risk scores. Group 2 emissions (1,3 butadiene, chloroform, ethylene oxide, methanol, methyl-isobutyl-ketone, and trichloroethylene) were associated with septal defects when using both the amounts and RS in rural regions. In addition, we found positive associations between rural Group 1 emissions and CHD rates based on the RS only. In urban regions, we found negative associations between Group 3 emission amounts (arsenic, lead, cadmium, hexachorobenzene, mercury) and CHD.

To our knowledge, this is one of very few studies that utilize a national pollutant registry to explore multipollutant groups of industrial emissions and their potential relation to CHD [19].

Investigations examining associations between air pollution and CHD rates have largely relied on data from monitoring stations in urban settings which capture ambient concentrations of a few monitored pollutants (e.g., carbon monoxide, sulphur oxides, nitrogen oxides) [9]. This approach has less capacity to examine a broader range of emitted industrial pollutants and the impact of multipollutant combinations on health outcomes. Therefore, our study generates new hypotheses as to how multiple pollutants could potentially contribute to CHD, a direction recently recognized as important in understanding how environmental exposures contribute to health [20].

In this study, we examined trends of DT emitted by facilities located in rural and urban regions without considering the impact they may have in neighboring regions. That rural regions host the greatest proportion of industrial facilities compared to urban regions could have accounted for the higher proportion of DT emissions in those regions. There were greater decreases in DT emission amounts in rural compared to urban regions over the study period. Likewise, the proportion of CHD cases was higher in rural regions compared to urban regions and we found a significant temporal decrease in CHD and septal defects rates in the province and rural regions. In contrast, the urban facilities demonstrated a marginally statistically significant temporal decrease in emissions and CHD rates.

Throughout the study period, Group 1 DTs had a greater geographic footprint in both urban and rural regions compared to Group 2 and 3 DTs. Associated reductions in emissions of Group 1 and 2 DTs and CHDs could suggest that a reduction in industrial emissions in rural regions positively impacted the health in those areas. The urban located facilities continued to emit significantly more toxic Group 3 DTs into the environment. The combination of those two factors and additional factors not examined here (e.g., other pollutants, socioeconomic status) may have contributed to the lack of change in CHD rates in urban areas. The observed decreasing trend in the emission of DTs in our study can be attributed to multiple factors which may act alone or in combination, including new legislation, use of prevention and mitigation technology, cycles and variation in production, implementation of government strategy for environmentally sustainable development and industrial self-regulation [21]. Studies of environmental regulations suggest that legislation alone does not seem to explain the observed decrements in emission [22]. To this effect, economic factors may be stronger contributors to the behavior of emissions in time. For example, the 2008 economic downturn affected the manufacturing sector particularly wood manufacturing resulting in a decrease of benzene emissions [15].

To better examine the association between multiple pollutants groups with CHD rates, we explored both the trends of emission amounts as well as their RS. The use of pollutant toxicity has been found to be important, particularly in equity studies, to better quantify the risk posed by industries to nearby communities [23]. Some of the DTs may be emitted in small quantities but are highly toxic when factoring their toxic equivalent potential (e.g., heavy metals). In the current exploratory study, the most significant associations found with Groups 1 and 2 DTs remained regardless of using amounts or RS. Recently, using an inverse distance weighted (IDW) approach to understand the effect of maternal residential proximity to industrial facilities on the development of CHD [24], we identified that only the highest exposures to the three DTs Groups were associated with urban CHD while, in rural regions, associations occurred with Groups 1 and 3 DTs and not Group 2 DTs [24]. Despite the fact that rural regions had more facilities and emissions, their impact was larger in surrounding urban postal codes. In the current study it is not clear whether urban or rural regions are the main driver of the Group 1 associations at province level.

The Group 3 emissions which are dominated by heavy metals present contradictory and intriguing results. We found strong negative associations with CHD in urban regions based on amounts emitted. In fact, in our previous study, [24] using a more precise exploratory approach, we found positive associations in both urban and rural postal codes exposed to the highest levels of Group 3 emissions [24].

Our findings are consistent with published studies where the relationship between heavy metals and CHD remains inconclusive with some studies reporting positive and others negative associations [25]. This suggests that the associations between CHD and Group 3 DTs may be more complex.

Reported experimental animal models have demonstrated congenital anomalies with exposure to some of the chemicals we examined in our study, however whether there is a truly causal effect is still to be explored. A study by Holson et al. which examined arsenic (present in Group 3 chemicals in our study), demonstrated congenital anomalies only at very high metal exposures [26], suggesting that very high doses of metals may be required to produce effects in humans. In addition, there is evidence that some of the volatile organic compounds (VOCs) identified in our study, trichloroethylene (TCE) in drinking water resulted in CHD in animal models; however, exposure to inhaled TCE, as would be the case in the present study, has not clearly affected cardiac morphogenesis in these models [27]. No previous experimental models have been used to examine the role of multipollutant (e.g., VOCs and gases), and yet, based on the findings of our study, possible chemical combinations could be teratogenic. Mechanisms through which these chemicals contribute to CHD evolution could include oxidative stress-mediated dysregulation of developmental signals during cardiac morphogenesis [28], complex gene–environmental interactions during the vulnerable window of cardiac embryogenesis, and/or altered epigenetic transcription factors involved in neural crest migration or other cell processes during cardiogenesis [29–31]. Further investigations that explore the impact of exposure to pollutant mixtures on cardiac development are necessary to better define this relationship on a whole organ, cellular, and molecular level.

4. Materials and Methods

This is an exploratory ecologic study, which examined industrial DTs and CHD rate trends aggregated temporarily in the province of Alberta as a whole and its urban and rural regions. Ecologic study designs have been used to explore research ideas around rare diseases with limited knowledge in a time- and cost-effective manner, and to generate hypotheses for testing in more robust research methods [32].

4.1. Study Population

We searched for all children born in Alberta between January 2004 and August 2011 with echocardiography-confirmed CHD from the pediatric echocardiographic Xcelera (Philips, Markham, ON, Canada) regional databases. Other data for each case included birth date, study date, and postal code at the time of diagnosis. Ethics approval from the participating institutions was obtained.

Case ascertainment was performed by retrieving all echocardiographic and surgical reports to confirm a diagnosis of CHD. Cases were aggregated according to their suspected embryological derivations as previously described [33]. For patients with multiple echocardiographic examinations, the most consistent major umbrella diagnosis was accepted as the diagnosis, and when there was uncertainty regarding the primary embryological group, the echocardiogram was reviewed by a pediatric echocardiographer or the operative diagnosis was used. We considered all cases with structural heart abnormalities, including those with a patent ductus arteriosus (PDA) present at >6 months and those with an atrial septal defect (ASD) after one year, or in whom surgical or device closure was necessary. Patients with cardiomyopathies and no structural CHD, neonatal peripheral pulmonary stenosis, a PDA at less than six months, an ASD at <1 year, and all cases born outside of the province were excluded.

4.2. Pollution Data

We accessed the NPRI to identify annual reports of all chemicals released and geographic coordinates of emitting facilities in Alberta from 2003–2010. We found that overall, 99% of emissions had been released to air, and therefore we focused on air emissions. We then identified chemicals

recognized as DTs based on a list compiled by the US Environmental Protection Agency from the State of California known as Proposition 65 [13].

4.3. Spatio-Temporal Aggregation of DTs

As the study population consisted of births between January 2004 and August 2011, we used the DTs emitted to air in the year in which the first trimester occurred between 2003 and 2010, as a surrogate for exposure during the period of cardiac morphogenesis. We worked under the assumption that the cases were born at term. For the cases, whose first trimester straddled two years, the case was assigned to the preceding year as the year of exposure. Live births for the study period were obtained from Statistics Canada and assigned to the year when the first trimester occurred for the sake of consistency.

4.4. Statistical Methods

CHD rates: We calculated yearly crude rates for all CHD and for septal defects observed for the exposure years 2003–2010 and described their trends. We used the second digit of first three characters of the Alberta postal code to identify cases in urban and rural areas. The population at <1 year of age was aggregated at the postal code level using Statistics Canada population data in order to calculate CHD rates.

DT emissions: We sought to explore potential associations using the amounts of DTs reported as tonnes or taking into consideration the potential toxicity of the DTs to the neighborhood defined as a Risk Score (RS). The RS is calculated by multiplying the amount of pollutant released by its corresponding toxic equivalent potential (TEP) which is determined by international agencies and the US government and then reported in Scorecard, which is a website compiled by Environmental Defense, a US nongovernmental agency [34,35]. This solution allows for comparisons of chemical releases on a common scale that takes into account differences in their chemical toxicity.

In order to reduce the number of pollutant variables in the analyses and create multipollutant groups, we applied principal component analysis (PCA) to both provincial amounts and RS metrics. The correlation matrix of the PCA used standardized individual DTs due to large variations in emitted amounts. To fulfill the required criteria that the number of observations should be greater than the number of variables [36], we selected the DTs according to sectors using the North American Industrial Classification System at level 2 [37]. We used orthogonal varimax rotation and we retained three uncorrelated principal components (PCs) which accounted for 74% of cumulative variability in tonnes and 83% of cumulative variability for RS. We selected DTs with a correlation coefficient $\geq |0.6|$ to keep in the corresponding groups, which we named Groups 1 to 3. We then summed the yearly amounts and RS of the DTs in their respective groups and determined their annual trends. We used simple linear regression to determine coefficients for the amounts of emissions and RS and Bonferroni adjusted p-trend values. Since we did not have many comparisons for the CHD trends, we accepted a p value of $\leq$0.05 to be significant.

We tested potential associations between yearly sum of the amount and RS of each DT group and CHD rates at provincial, rural and urban levels using Spearman test. We considered associations with r values >0.7, scatter plots with a linear tendency and reported the 95% CI for the correlation coefficient. Although Bonferroni adjustments are not strictly recommended in exploratory studies [38], here we are presenting conservative p values in order to avoid inflated type 1 error due to multiple tests. Applying Bonferroni approach, we obtained the adjusted p-value threshold by dividing α (0.05) by the number of the independent hypotheses. Due to a high correlation between CHD and Septal (r = 0.90, p = 0.002), we consider CHD and Septal as one outcome in term of multiple-testing adjustment. As province data is just the sum of urban and rural, the province overall model is, therefore, not independent of rural-urban stratified models. On the other hand, due to the low correlation between amounts and risk score (r = -0.33, p = 0.42), we consider them two independent exposures. Similarly, the models with three chemical groups are independent as the three PCA groups are uncorrelated. Therefore, the total number of "independent" tests is 12 (= 2 $\times$ 3 $\times$ 2; 2 for two different metrics of the exposure (amounts

and risk score) × three chemical groups × two rural/urban strata, and considered a p value $\leq$0.004 to be significant after Bonferroni correction. All analyses were performed using STATA 12 (StataCorp LP, College Station, TX, USA) and SPSS 21 (IBM Corp, Armonk, NY, USA).

4.5. Study Limitations

Although our study sheds light on a potential association between multipollutant organic compounds and gases emitted into air by industry and CHD, certain limitations must be recognized. Being an ecological study, the observations made at aggregate level cannot be inferred to individuals. Given our source of patient data, we were unable to account for other variables associated with CHD including genetic abnormalities, maternal health and exposure to drugs, and the impact of folic acid supplementation [39]. In addition, other subtypes of CHD were not examined because they did not reveal any temporal trend and most likely due to small sample size. Therefore, we committed to report only the septal group of CHDs and this could contribute to selection bias. We were also unable to include other environmental confounders like the socio-economic status and traffic related pollutants and, therefore, the estimates observed in our study could be an over or under estimation of the associations of industrial pollutants and CHD.

We did not have data on gestational age at birth and yet some of our CHD cases could have been delivered prematurely which could have resulted in exposure misclassification errors. In addition, the emissions are reported annually making it difficult to assign them more closely to the window of cardiac morphogenesis.

We did not include terminated pregnancies with fetal CHD; however, the observed temporal decrease in CHD rates was unlikely to have occurred due to pregnancy terminations, as we had previously observed no increase in pregnancy terminations for CHD in the province during the study period, and absolute termination rates are quite low in our province [40]. Furthermore, the CHD rates we reported are consistent with previously published Alberta case ascertainment rates [41].

5. Conclusions and Recommendations

In this exploratory study, we have observed downward temporal patterns of emissions accompanied by a parallel decrease in CHD rates in rural regions which potentially implies that efforts at reducing emissions could impact positively on reducing CHD in our children and the general health of all living organisms on the planet. Furthermore, we observed consistent positive associations between VOC emissions and CHDs in Alberta, predominantly in rural areas between 2003 and 2010. The relationship between industrial DTs and CHD may not represent the effect of exposure to a single pollutant, but rather multipollutant exposures which require further investigation by the research community. We believe that the approach of using amounts and RS in this study complemented each other in attempting to quantify the risk posed by industries to nearby communities. We would like to recommend the establishment of comprehensive prospective birth defect registries which will capture maternal environmental factors, detailed perinatal variables, genetic, socio-economic, and pollutant environmental factors from various sources which will enhance more robust future epidemiological studies. Finally, our study was limited to human populations and the evidence from studies on other species [42,43] needs to be consolidated in order to begin to understand the role industrial chemical pollution and adverse health outcomes on the planet and its inhabitants.

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Conflicts of Interest: The authors declare no conflict of interest.

Ethics Approval: Ethics approval was obtained from the two participating institutions' boards: University of Alberta's Health Research Ethics Board-Health Panel approved the study and assigned it a project number, study ID: Pro00025428. University of Calgary's Conjoint Health Research Ethics Board of the Faculties of Medicine, Nursing and Kinesiology, ethics ID: E-24758.

Availability of Data Materials: The air pollution dataset generated for the current study is publicly available from the Government of Canada's National Pollutant Release Inventory, https://www.ec.gc.ca/inrp-npri/default.asp?lang=en&n=0EC58C98. (Accessed 11 July 2018. The congenital heart disease dataset is not publicly available due to privacy and confidentiality clauses of the Government of Alberta's Health Information Act Section 2).

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