American Journal of Epidemiology
Copyright © 2001 by The Johns Hopkins University School of Hygiene and Public Health
All rights reserved
Vol. 153, No. 6
Printed in U.S.A.

Association of Transposition of the Great Arteries in Infants with Maternal Exposures to Herbicides and Rodenticides

Christopher A. Loffredo,[1,2] Ellen K. Silbergeld,[1,2] Charlotte Ferencz,[1] and Jianyi Zhang[1]

The Baltimore-Washington Infant Study, a case-control study of congenital heart defects in liveborn infants conducted in 1981–1989, interviewed parents about a wide range of environmental exposures that occurred during and before the pregnancy. In the period 1987–1989, the questionnaire was expanded to include a detailed inquiry about exposures to pesticides. An analysis of these latter data revealed an association of maternal exposure to any pesticides during the first trimester with transposition of the great arteries in their infants (TGA; $n = 66$ infants), relative to 771 control infants, with an odds ratio of 2.0 (95% confidence interval (CI): 1.2, 3.3). No other heart defects were associated with pesticides. When analyzed by type of pesticide and adjusted for covariates, there were associations of TGA with maternal exposures to herbicides (odds ratio (OR) = 2.8; 95% CI: 1.3, 7.2) and to rodenticidal chemicals (OR = 4.7; 95% CI: 1.4, 12.1) but not to insecticides (OR = 1.5; 95% CI: 0.9, 2.6). No data were collected on specific chemicals or brand names. These results raise new questions about the possible epidemiologic association of TGA with some classes of pesticides and warrant new, carefully targeted investigations. *Am J Epidemiol* 2001;153:529–36.

heart defects, congenital; herbicides; pesticides; rodenticides; transposition of great vessels

During the past 20 years, environmental risk factors for human birth defects have drawn attention from the public and scientific communities. Major birth defects are diagnosed in approximately 4 percent of livebirths in this country, are the leading cause of infant mortality, account for nearly one third of pediatric hospitalizations, and carry major economic costs in the first years of life (1–3). Only a fraction of affected infants have heritable genetic defects. For example, in the Baltimore-Washington Infant Study, only 20 percent of 4,390 infants with congenital heart disease had chromosomal or syndromic anomalies (4). Despite the public health importance of birth defects, little is known about potential environmental risk factors.

Pesticide exposures are of possible concern in this context for a number of reasons. Many of these compounds are ubiquitous in the environment because of commercial and household applications. Some compounds, such as dichlorodiphenyltrichloroethane (DDT), persist in the environment and in the body for many years because of high lipid solubility and resistance to breakdown. Dichlorodiphenyltrichloroethane and other stable organochlorine pesticides have been detected in breast milk and in the adipose tissue of neonates (5). Much of the US population now has detectable levels of certain pesticide compounds in adipose tissue (6). Some pesticides, such as 2,4-dichlorophenoxy acetic acid (2,4-D), once widely sold as a yard and garden weed killer, have been shown to induce birth defects in laboratory animals (7), raising concern for possible teratogenic effects among exposed human populations.

Numerous studies have examined the possible association between parental exposures to pesticides and the occurrence of congenital anomalies (7–19). Some of these studies detected increased rates of birth defects as a whole compared with the rate in the general population, while others looked at specific anomalies such as orofacial clefts and hypospadias. However, many of these studies, particularly those involving case reports and occupational cohorts, have been limited by small numbers of affected infants, lack of appropriate control subjects, and inadequate power to detect increased risks for specific malformations. Ecologic studies may have suffered from misclassification of exposures. Few studies have examined potential associations between pesticide exposures and specific cardiovascular malformations.

Congenital heart defects represent a major segment of congenital anomalies (prevalence = 4/1,000 livebirths in the Baltimore-Washington Infant Study) and carry a disproportionate burden of public and private costs associated with diagnosis, treatment, counseling, and social services (20). Malformations of the cardiac outflow tract are among the most severe congenital heart defects. Such cases with normal visceral situs may be classified anatomically into two main groups: those with and without transposed great arteries (21–23). In infants with transposition of the great arteries

Received for publication July 14, 1999, and accepted for publication July 18, 2000.

Abbreviations: CI, confidence interval; OR, odds ratio; TGA, transposition of the great arteries.

[1] Department of Epidemiology and Preventive Medicine, University of Maryland School of Medicine, Baltimore, MD.

[2] Program in Human Health and the Environment, University of Maryland School of Medicine, Baltimore, MD.

Reprint requests to Dr. Christopher A. Loffredo, Department of Epidemiology and Preventive Medicine, University of Maryland, 660 West Redwood St., Baltimore, MD 21201 (e-mail: cal9@gunet.georgetown.edu).

Downloaded from https://academic.oup.com/aje/article-abstract/153/6/529/136848 by guest on 11 April 2019

(TGA), the aorta arises from the right ventricle (instead of the left) and the pulmonary artery arises from the left ventricle (instead of the right). TGA is associated at birth with hypoxia, cyanosis, and high neonatal mortality, but survival is now possible with surgical intervention (24). Infants with TGA are predominantly male, free from additional birth defects, and tend to be born at full term with near-normal birth weight. The group of outflow tract defects with normally related great arteries ("non-TGA") is heterogeneous, including tetralogy of Fallot, double outlet right ventricle, truncus arteriosus, and several rare phenotypes. Like TGA, this group is characterized by hypoxia at birth, high neonatal mortality, and the need for early surgical interventions. Unlike TGA, infants with non-TGA outflow tract anomalies show little variation by sex, and many are born growth retarded and with multiple birth defects. Despite these clinical differences between infants with TGA and non-TGA outflow tract anomalies, most epidemiologic studies have tended to combine the malformations into one category when examining associations with potential risk factors (25, 26).

The Baltimore-Washington Infant Study, a hypothesis-generating case-control study of the etiology of cardiovascular malformations, was undertaken between 1981 and 1989, with the aim of generating hypotheses of genetic and environmental risk factors for specific congenital heart defects (4). Because of mounting public and scientific interest in the health effects of pesticides, the original Baltimore-Washington Infant Study questionnaire was expanded in 1987 to include a detailed, self-reported parental assessment of pesticide exposures at home and on the job. An analysis of those data has not been previously reported. Therefore, the aim of the present study is to report the maternal pesticide exposure data of this later subset of the Baltimore-Washington Infant Study (1987–1989) for possible associations with TGA and non-TGA outflow tract anomalies. This approach addresses the possible heterogeneity of cardiac outflow tract anomalies and provides an opportunity to examine the associations of these birth defects with specific classes of pesticides.

MATERIALS AND METHODS

The Baltimore-Washington Infant Study collected data from 1981 to 1989 from parents of case infants with congenital heart defects and parents of randomly selected control infants who did not have heart defects. The details of the study design and the questionnaire have been published (4). The vast majority of interviews (90 percent among cases, 91 percent among controls) took place at home, and 90 percent of all interviews took place before the infant's first birthday. Response rates were 90 percent among the case families and 95 percent among the controls. Reported family histories of birth defects in first degree relatives were confirmed by medical records.

Cases were defined as infants born alive in 1981–1989 to parents who were residents of the study area (Maryland, the District of Columbia, and adjacent counties of northern Virginia). All six pediatric cardiology centers in the region participated in the study. The diagnosis of specific structural heart disease was confirmed before 1 year of age by pediatric cardiologists, and a search of all hospital pathology logbooks and death certificates in the region identified additional cases who died prior to referral. Transposition of the great arteries was defined as TGA with or without ventricular septal defect, pulmonary atresia, tricuspid atresia, and double-outlet right ventricle. The non-TGA group of cardiac outflow tract anomalies consisted of an aortic-pulmonary window, a supracristal ventricular septal defect, a double-outlet right ventricle, a common arterial trunk, and tetralogy of Fallot.

Controls were liveborn infants who had no congenital heart defects and were born in the region during 1981–1989. They were selected randomly from the liveborn birth cohort, stratified by year and hospital of birth. For the present study, we analyzed the subset of Baltimore-Washington Infant Study subjects born in 1987–1989 to investigate the association of specific heart defects with specific maternal pesticide exposures.

We considered a mother exposed to pesticide chemicals if she reported such an exposure during the "critical period" for cardiovascular development, that is, the first trimester of pregnancy and the preceding 3 months. This exposure window allows for uncertainty concerning the date of conception. Exposure information obtained from the questionnaire included: *type of exposure*, which classified pesticide products by type of pest, including those used to kill fleas, flying insects, crawling insects, rodents, and weeds; *mode of exposure*, which included area-wide sprays from aircraft, professional exterminator services, hand-held spray, "bomb" applicator, and "pellet, powder, or food imitators"; *places where the exposure occurred*, which included home, job, and other site; *frequency of exposure*, which included no exposure, one exposure in the critical period, once a month, once a week, a few times a week, and daily; and *time of exposure*, by trimester. Nonchemical controls (e.g., rodent traps) were excluded from analysis.

From the answers to these questions, we grouped mothers into the following mutually exclusive groups: 1) nonexposure, that is, no exposure to any pesticides at any time 6 months before or during pregnancy; 2) critical period exposures; 3) exposures to any pesticides 4–6 months prior to the pregnancy; and 4) late gestational exposures (4–9 months of pregnancy).

Variables under consideration as potential confounders or effect modifiers of pesticide exposures included the following: sex and race of the child (White/non-White); family socioeconomic status score (defined below); maternal age; family history of heart defect (yes/no) and other family birth defects (yes/no); maternal diabetes (yes/no); maternal use of marijuana (yes/no), cigarettes (nonsmoker, <1 pack/day, 1 pack/day, >1–<2 packs/day, ≥2 packs/day), and alcohol (never, <1 time/week, 1 time/week, almost 1 time/day); maternal exposures (yes/no) to painting, paint stripping/sanding, varnishing, dry cleaning solvents, degreasing solvents, and other solvents; and history of paternal exposure to pesticides during the 1–6 months before pregnancy (yes/no). The socioeconomic status score and the maternal cigarette and alcohol amounts were used as continuous variables in data

Downloaded from https://academic.oup.com/aje/article-abstract/153/6/529/136848 by guest on 11 April 2019

analysis. The socioeconomic status score was defined as the sum of the following variables: maternal education (0 for none; 1 for 1–6 years; 2 for 7–9 years; 3 for 10–11 years; 4 for 12 years; 5 for 13–15 years; 6 for 16 or more years) plus household income (1 for $10,000; 2 for $10,000–$19,000; 3 for $20,000–$29,000; 4 for $30,000 or more) plus head-of-household occupation (1 for professional; 0 for other). For analysis the following categories were studied: low (0–4), medium (5–8), and high (9–12).

Associations between maternal pesticide exposures and congenital heart defects were studied using logistic regression in SAS computer software (27) to produce crude and adjusted odds ratios and 95 percent confidence intervals. Potential effect modifications were tested by including interaction terms in the regression models; confounding effects were tested with the method of Kleinbaum et al. (28).

RESULTS

Number of cases and controls

A total of 4,390 cases of congenital heart disease were identified in the Baltimore-Washington Infant Study during 1981–1989. The subset of Baltimore-Washington Infant Study cases born in 1987–1989 totaled 1,832 cases of congenital heart defects (analyzed in the present study), among whom there were 66 infants with TGA and 114 with non-TGA outflow tract anomalies. In this time period we enrolled 771 control infants.

Associations of pesticides with specific types of heart defects

We examined specific subsets of congenital heart defects, each having at least 30 cases, for association with maternal exposure to any pesticides during the critical period (table 1). These subsets included defects of laterality and looping, TGA, non-TGA outflow tract anomalies, endocardial cushion defects, left-sided heart lesions, pulmonic valve stenosis, ventricular septal defect, and atrial septal defect. Only TGA was significantly associated with pesticides; all other associations were not statistically significant, and their odds ratios ranged from 0.8 to 1.5. For all cardiovascular malformations as a whole, the association was not significant (odds ratio (OR) = 1.1). Other than TGA, there were no significant associations between congenital heart defects and specific categories of pesticides, modes of application, places of exposure, or frequencies of exposure. The subsequent data analyses focused on TGA and non-TGA outflow tract anomalies.

Characteristics of the infants and their parents

Of the infants with TGA, 10 percent had extracardiac anomalies, compared with 33 percent with non-TGA outflow tract anomalies. Compared with controls, the infants with TGA were more likely to be male, but this was not true of the infants with non-TGA outflow tract anomalies (table 2). The three groups of infants had a similar race distribution. Infants in the non-TGA case group were more likely than TGA infants and controls to be born prematurely and to have a low birth weight. Characteristics of the parents, including age, socioeconomic factors, and most environmental exposures, were distributed similarly among the three groups. Maternal exposures to solvents and paternal pesticide exposures were more frequently reported by parents of infants with TGA than by parents of controls.

Pesticide exposures among infants with TGA and other outflow tract anomalies

Analysis of maternal responses to the questionnaire revealed that pesticide exposures, especially to chemicals used against crawling insects (e.g., cockroaches), were fairly common during the critical period (table 3). Mothers of infants with TGA were more likely than controls to report taking measures against infestations of rodents and weeds. For all three groups of mothers, the most common mode of application was a hand-held spray, but the greatest case-control difference was in the use of pellet, powder, and food imitator among mothers of TGA infants. For all groups, the home was by far the most common site of exposure. Frequent exposures, that is, once a week or more often, were

TABLE 1. Association of congenital cardiovascular malformations with maternal exposure to any pesticides during the critical period of pregnancy, Baltimore-Washington Infant Study, 1987–1989

Type of heart defect	Total	Exposed		OR*	95% CI*
		No.	%		
Laterality and looping defects	31	7	22.6	0.8	0.3, 1.8
Transposition of the great arteries	66	29	43.3	2.0	1.2, 3.3
Non-TGA* cardiac outflow tract anomalies	114	30	26.3	1.0	0.6, 1.5
Endocardial cushion defect	87	31	35.6	1.5	0.9, 2.4
Left-sided obstructive lesions	147	38	25.9	0.9	0.6, 1.4
Pulmonic stenosis	80	25	31.3	1.2	0.7, 2.0
Ventricular septal defect	373	104	27.9	1.0	0.9, 1.5
Atrial septal defect	103	34	33.0	1.3	0.8, 2.1
All cardiovascular malformations	1,001	298	29.8	1.1	0.9, 1.3
Controls	771	209	27.1	1.0	Reference

* OR, odds ratio; CI, confidence interval; TGA, transposition of the great arteries.

Downloaded from https://academic.oup.com/aje/article-abstract/153/6/529/136848 by guest on 11 April 2019

TABLE 2. Parental and infant characteristics of cases and controls, Baltimore-Washington Infant Study, 1987–1989

Characteristic	Infants with outflow tract anomalies		Control infants (*n* = 771)
	TGA* group (*n* = 66)	Non-TGA group (*n* = 114)	
Infant			
Sex (% male)	62.1	53.5	50.8
Race (% White)	68.2	58.8	69.4
Gestational age (%)			
<38 weeks	6.0	25.3	10.4
≥38 weeks	94.0	73.7	89.6
Birth weight, g (mean (SD*))	3,421 (541)	2,960 (747)	3,362 (603)
Parents			
SES* score† (%)			
Low (1–4)	74.2	51.8	60.1
Middle (5–8)	24.4	42.9	35.9
High (9–12)	1.5	5.3	4.0
Maternal age at conception (%)			
<20 years old	6.1	13.2	11.9
20–35 years old	83.3	75.4	81.2
>35 years old	10.6	11.4	6.9
Maternal cigarette smoking (%)			
None	74.2	65.8	69.4
<1/2 pack/day	16.7	14.9	13.4
≥1/2–1 pack/day	6.1	15.8	12.8
>1–<2 packs/day	3.0	1.8	3.2
≥2 packs/day	0	2.6	1.2
Maternal alcohol use (%)			
None	38.5	37.7	44.4
<Once/week	38.5	37.7	33.3
Once/week	15.4	18.4	16.1
Daily	7.4	6.1	6.2
Family history of heart defects (%)	1.5	2.6	2.1
Family history of other birth defects (%)	6.1	9.6	5.8
Maternal diabetes (%)	1.5	3.5	1.2
Maternal marijuana use (%)	3.0	9.6	4.7
Maternal cocaine use (%)	1.5	2.6	1.7
Maternal house painting (%)	12.1	8.8	11.3
Maternal exposure to solvents‡ (%)	6.1	0	2.0
Father exposed to pesticides (%)	62.1	46.5	45.3

* TGA, transposition of the great arteries; SD, standard deviation; SES, socioeconomic status.
† Summary score based on maternal education, family income, and head-of-household occupation.
‡ Paint thinners, brush cleaners, and art and photography solvents.

rarely reported. Only one mother of an infant with TGA reported both herbicide and rodenticide exposures, compared with none in the non-TGA group and none among the controls.

We examined these variables with unadjusted odds ratios and 95 percent confidence intervals to identify associations in need of further evaluation. There were no significant associations between TGA and infestations of fleas, flying insects, or crawling insects. However, there were significant associations between TGA and exposures to rodent killers (OR = 3.5; 95 percent CI: 1.5, 8.5) and weed killers (OR = 3.7; 95 percent CI: 1.6, 8.4). In terms of mode of exposure, only the use of pellet, powder, or food imitator was significantly associated with TGA (OR = 4.0; 95 percent CI: 1.7, 9.8). Specific places of exposure were not significantly associated with TGA. The trend in odds ratios with increasing frequency of total exposures was also examined; relative to unexposed pregnancies, the odds ratio for only one exposure during the critical period was 1.6 (95 percent CI: 0.7, 3.4) and was 3.4 (95 percent CI: 1.8, 6.4) if the frequency of exposure was once a month or greater. This apparent dose-response relation did not continue when we examined separately the small number of mothers who reported being exposed once a week (OR = 1.2), a few times a week (OR = 0.8), or daily (no exposures reported).

We also evaluated the association of non-TGA cardiac outflow tract anomalies with pesticide exposure variables. The crude odds ratios indicated significant associations only with job exposures (OR = 3.8; 95 percent CI: 1.4, 10.6), but no single type of exposure mode or chemical subgroup predominated among these reports. None of the parents were professional exterminators. There were no significant asso-

Downloaded from https://academic.oup.com/aje/article-abstract/153/6/529/136848 by guest on 11 April 2019

TABLE 3. Characteristics of maternal pesticide exposures among TGA,* non-TGA, and control infants, Baltimore-Washington Infant Study, 1987–1989

Exposure characteristic during the critical period	TGA group (n = 66)		Non-TGA group (n = 114)		Control infants (n = 771)	
	No.†	%	No.†	%	No.†	%
Any pesticide exposure	29	43.9	30	26.3	209	27.1
Type of pest						
Fleas	2	3.0	2	1.8	28	3.6
Flying insects	2	3.0	2	1.8	22	2.9
Crawling insects	14	21.2	22	19.3	130	16.9
Rodents	5	7.5	4	3.5	12	1.6
Weeds	8	12.1	0		28	3.6
Multiple pests	4	6.0	6	5.3	9	3.0
Mode of pesticide application						
Area spray	3	4.5	2	1.8	13	1.7
Exterminator	6	9.1	8	7.0	34	4.4
Bomb	2	3.0	1	0.9	10	1.3
Hand-held spray	14	21.2	16	14.0	116	15.0
Pellet, powder, or food imitator	7	10.6	5	4.4	22	2.9
Other modes	4	6.1	3	2.6	19	2.5
Multiple modes	1	1.5	1	0.9	13	2.0
Place of exposure						
Home	23	34.8	24	21.1	195	25.3
Job	3	4.5	6	5.3	11	1.4
Other sites	2	3.0	1	0.9	3	0.4
Multiple sites	0		1	0.9	1	0.1
Maximum frequency of exposure						
One time only	9	13.6	8	7.0	84	10.9
Once a month	15	22.7	9	7.9	66	8.6
Once a week	3	4.5	8	7.0	37	4.8
A few times a week	1	1.5	3	2.6	18	2.3
Daily	0		2	1.8	4	0.5

* TGA, transposition of the great arteries.
† Number of mothers reporting exposure.

ciations between these heart defects and different types of pests, modes of application, or frequencies of the exposure.

Multivariate analysis

We adjusted the crude associations reported above for potential effect modifiers and confounders (table 4). We focused the multivariate analyses on two specific exposures, rodenticides and herbicides, as these were most strongly associated with TGA in the unadjusted analyses. The crude odds ratio for rodenticidal chemicals was significantly elevated at 4.7 (95 percent CI: 1.6, 13.8), which remained stable after taking into account race, socioeconomic status, maternal age, maternal smoking and alcohol use, family history of heart defects, and paternal pesticide exposures (adjusted OR = 4.8). There were no significant interactions. The adjusted odds ratios for herbicide exposures during the critical period were 2.8 (95 percent CI: 1.2, 6.9), indicating a possible increase in the risk of having an infant with TGA. Restricting this exposure window to only the first trimester did not change the risk estimate (OR = 2.76).

For comparison with TGA associations, we examined the crude and adjusted associations of non-TGA heart defects with maternal exposures to rodenticides. The results (data not shown) indicated that this type of exposure did not increase the risk of having an infant with non-TGA types of cardiac outflow tract anomalies (OR = 1.6; 95 percent CI: 0.6, 4.2). Unlike mothers in the TGA group, no mothers in this non-TGA group reported using herbicides during the critical period.

Effect of exposures during different time periods of pregnancy

The association of "any pesticides" with TGA at different time periods before and during the pregnancy is shown in table 5. The odds ratio for any type of pesticide exposure at the critical period was 2.1 (95 percent CI: 1.2, 3.4), but it was slightly lower (OR = 1.8) in the period of 4 months before the inception of pregnancy, in contrast to the odds ratio of 1.2 for late gestational exposure. Insecticides were not associated with TGA in any time period. For both roden-

Downloaded from https://academic.oup.com/aje/article-abstract/153/6/529/136848 by guest on 11 April 2019

TABLE 4. Association of maternal herbicide and rodenticide exposures with transposition of the great arteries, Baltimore-Washington Infant Study, 1987–1989

Exposure	Cases		Controls		Model*	OR†	95% CI†
	No.	%	No.	%			
Herbicides	8	12.1	28	3.6	Unadjusted	3.7	1.6, 8.4
					Adjusted	2.8	1.2, 6.9
Rodenticides	5	7.6	13	1.7	Unadjusted	4.8	1.6, 13.8
					Adjusted	4.7	1.5, 14.2

* Adjusted model includes race of infant, socioeconomic status score, maternal age, maternal smoking and alcohol use categories, family history of heart defects, maternal diabetes, maternal solvent exposures, and paternal pesticide exposures.
† OR, odds ratio; CI, confidence interval.

ticides and herbicides, the associations with TGA were significant if they occurred in the critical period (i.e., ORs = 5.1 and 3.6 for rodenticides and herbicides, respectively), or if they occurred earlier, but not if the exposures took place after the first trimester.

DISCUSSION

This study suggests an association between maternal exposures to herbicides and rodenticides early in pregnancy and the occurrence of transposition of the great arteries in their infants. The exposures occurred mainly in the home and less than weekly for nearly all of the subjects. These results raise new questions about the possible risks associated with the use of pesticides in the home.

Exposures to pesticides

There are over 600 pesticide chemicals actively used and marketed in 45,000–50,000 different formulations (29). There are many differences among these compounds in their physical properties, toxicity, metabolism, and other characteristics. Our study found significant associations involving rodenticides and herbicides. We do not have data on the specific products used by the parents in our study. However, during this time period (1987–1989), the predominant chemicals in rodenticides were anticoagulants such as brodifacoum and other derivatives of coumarin (warfarin) (30–32). A wider range of chemicals was sold as commercial herbicides (32), especially chlorophenoxy compounds and glyphosphate. Some types, such as 4-chloro-2-methylphenoxyacetic ethylester and 2-methyl-4-chlorophenoxyacetic acid, induced teratogenic effects at high doses among laboratory animals (33, 34), including ventricular septal defects, while other compounds have not proven to be teratogenic or have not been tested adequately (7, 35). If the associations in our study are real, that is, not due to chance or to confounding with another, unknown teratogen or other factor, it may be that there are underlying similarities in the biologic activities of these chemicals

TABLE 5. Infants with transposition of the great arteries and timing of pesticide exposure in relation to the time of pregnancy, Baltimore-Washington Infant Study, 1987–1989

Type of exposure	Exposure time period	No. of mothers exposed		OR*	95% CI*
		Cases (n = 66)	Controls (n = 771)		
Any pesticides	4–6 months before pregnancy	22	168	1.8	1.0, 3.0
	Critical period	29	209	2.1	1.2, 3.4
	4–9 months during pregnancy	19	195	1.2	0.7, 2.0
Insecticides	4–6 months before pregnancy	18	141	1.6	0.9, 2.9
	Critical period	21	179	1.5	0.9, 2.6
	4–9 months during pregnancy	16	163	1.2	0.7, 2.1
Rodenticides	4–6 months before pregnancy	4	8	6.1	1.8, 20.7
	Critical period	5	12	5.1	1.7, 14.9
	4–9 months during pregnancy	3	11	3.2	0.9, 14.9
Herbicides	4–6 months before pregnancy	5	13	4.7	1.6, 13.6
	Critical period	8	28	3.6	1.6, 8.2
	4–9 months during pregnancy	4	26	1.8	0.6, 5.4

* OR, odds ratio; CI, confidence interval.

Downloaded from https://academic.oup.com/aje/article-abstract/153/6/529/136848 by guest on 11 April 2019

that affect the development of the cardiovascular system. It is unclear, however, why such exposures would increase the risk of TGA but not of other types of cardiovascular malformations.

Some pesticides, including the coumarin derivatives used in rodenticide baits, have relatively long biologic half-lives (100–200 days) (31). Numerous pesticides pass readily from the maternal to the fetal circulation and have also been detected in breast milk. In a study of adipose tissue from 183 healthy neonates, high concentrations of chlorinated hydrocarbon pesticide residues were detected even before the infants took their first food (5). It is therefore plausible that maternal pesticide exposures early in the pregnancy, or even just before the pregnancy, could result in the exposure of the fetus to biologically effective concentrations of pesticides. Our data showing that the strength of the association with TGA depends on the time period of exposure lend additional support to a potential teratogenic role of these exposures.

Regarding the dose-response relation, we found that the risk of TGA was sequentially increased when mothers who reported one-time exposures and mothers reporting monthly exposures were each compared with unexposed mothers. Lack of a dose-response relation at more frequent exposure levels (e.g., weekly) might be explained by underreporting, low power to detect effects at extremely rare exposure levels, or fetal demise at high exposure levels.

Cardiac outflow tract anomalies

The development of the human cardiovascular system begins around the third intrauterine week and is essentially complete by the end of the 12th week. This window of vulnerability to toxic disturbances coincides with the period of exposure we found to be most strongly associated with an increased risk of TGA. Maldevelopment of the outflow tract of the heart, which includes the aorta and pulmonary artery and their septa and valves, results in a wide spectrum of cyanotic and acyanotic conditions detectable in neonates. The most common such malformations are TGA and tetralogy of Fallot (combined prevalence = 5.5/10,000 livebirths), but even these two malformations exhibit anatomic variations that correlate with their clinical severity (36). Previous epidemiologic studies have tended to group all outflow tract anomalies as a single entity, a classification system based on anatomy, not on etiology (23). Recent experimental studies of cardiovascular morphogenesis suggest that outflow tract anomalies are heterogeneous (37, 38) and tend to support the idea of TGA as distinct from other malformations of the cardiac outflow tract. As mentioned above, clinical evidence based on birth weight, sex ratio, and associated malformation syndromes is also consistent with this classification. In other reports from the Baltimore-Washington Infant Study, in which TGA and non-TGA outflow tract anomalies were separately analyzed for potential risk factor associations, TGA was unique in its association with maternal exposures to influenza, ibuprofen, and organic solvents (36), as well as high maternal intake of retinol supplements (39).

The scarcity of epidemiologic studies of potential environmental risk factors for TGA makes it difficult to compare our results with those from other researchers. Tikkanen and Heinonen (25) reported associations between "conal heart malformations" and maternal exposures to chemicals, dyes, lacquers, and paints at work, but they did not look at TGA separately. Adams et al. (26) examined potential environmental risk factors for the combined group of "conotruncal" heart defects and found elevated odds ratios associated with maternal work in agricultural trades, a possible marker of pesticide exposures. Separate analyses of TGA and non-TGA heart defects were not performed in that study.

Methodological considerations

Maternal recollection of periconceptional events may be influenced by the birth of a child with a congenital malformation, calling to mind events and exposures suspected as being teratogenic. If mothers of cases with TGA in this study had overreported their exposures to weed killers and rodenticides relative to control mothers, this bias should have been discernible in other severe heart defects. However, we found that pesticide associations with heart defects other than TGA were weak and statistically nonsignificant. It seems unlikely that recall bias affects only the mothers of infants with TGA. Furthermore, there were no case-control differences in maternal reports of exposures to insecticides, and it is unclear why recall bias would affect some classes of pesticides but not others.

Another methodological concern, nondifferential inaccuracy in reporting distant events, was minimized in our study by interviewing mothers of cases and controls within a 1-year period after their infants were born, so distortion of exposure reporting by poor memory is unlikely to be materially different in the two groups. In any event, such nondifferential errors in recall would most likely bias the odds ratios toward the null value. Selection bias due to differences in participation rates was probably minimal since these rates were similar across the groups. Finally, uncontrolled or poorly controlled confounding due to factors such as social class and poor housing stock may help to explain the observed association with pesticides, especially in view of the trend of lower socioeconomic status markers in TGA families relative to those in controls.

A major strength of the study is that it is population based. The cases were actively identified through multiple sources within a defined geographic area, and controls were selected to represent a random sample of the birth cohort. All cardiac diagnoses were confirmed by pediatric cardiologists. These methods substantially reduced potential selection biases and diagnostic misclassification. Finally, we were able to examine subsets of pesticide exposures by type of pesticides, mode of application, place of exposure, and frequency of exposure and to adjust for potential covariates and effect modifiers in the statistical analysis.

In conclusion, this study suggests that TGA may be a unique cardiovascular malformation, whose occurrence is associated with environmental pesticide exposures during the periconceptional period of pregnancy. Our data suggest that the risk of TGA is increased with maternal exposures to

Downloaded from https://academic.oup.com/aje/article-abstract/153/6/529/136848 by guest on 11 April 2019

herbicides and rodenticides. Multicenter studies should be carried out to achieve a larger sample size, with a carefully focused research design to further investigate these associations.

ACKNOWLEDGMENTS

The study was supported by a grant from the National Heart, Lung, and Blood Institute (R37 HL25269) and by a MERIT award to Dr. Charlotte Ferencz, principal investigator.

REFERENCES

1. Trends in infant mortality attributable to birth defects—United States, 1980–1995. MMWR Morb Mortal Wkly Rep 1998;47:773–8.
2. Economic costs of birth defects and cerebral palsy—United States, 1992. MMWR Morb Mortal Wkly Rep 1995;44:694–-9.
3. Garson A Jr, Allen HD, Gersony WM, et al. The cost of congenital heart disease in children and adults. A model for multicenter assessment of price and practice variation. Arch Pediatr Adolesc Med 1994;148:1039–45.
4. Ferencz C, Rubin JD, Loffredo CA, et al. Epidemiology of congenital heart disease: the Baltimore-Washington Infant Study 1981–1989. Mt. Kisco, NY: Futura Publishing Company, 1993.
5. Teufel M, Niessen KH, Sartoris J, et al. Chlorinated hydrocarbons in fat tissue: analysis of residues in healthy children, tumor patients, and malformed children. Arch Environ Contam Toxicol 1990;19:646–52.
6. Robinson PE, Mack GA, Remmers J, et al. Trends of PCB, hexachlorobenzene, and β-benzene hexachloride levels in the adipose tissue of the U.S. population. Environ Res 1990;53:175–92.
7. Schardein JL. Chemically induced birth defects. 2nd ed. New York, NY: Marcel Dekker, Inc, 1993.
8. Nelson CJ, Holson JF, Green HG, et al. Retrospective study of the relationship between agricultural use of 2,4,5-T and cleft palate occurrence in Arkansas. Teratology 1979;19:377–83.
9. Gordon JE, Shy CM. Agricultural chemical use and congenital cleft lip and/or palate. Arch Environ Health 1981;36:213–21.
10. Hanify JA, Metcalf P, Nobbs CL, et al. Aerial spraying of 2,4,5-T and human birth malformations: an epidemiological investigation. Science 1981;212:349–51.
11. McDonald AD, McDonald JC, Armstrong B, et al. Congenital defects and work in pregnancy. Br J Ind Med 1988;45:581–8.
12. Romero P, Barnett PG, Midtling JE. Congenital anomalies associated with maternal exposure to oxydemeton-methyl. Environ Res 1989;50:256–61.
13. Lin S, Marshall EG, Davidson GK. Potential parental exposure to pesticides and limb reduction defects. Scand J Work Environ Health 1994;20:166–79.
14. Garry VF, Schreinemachers D, Harkins ME, et al. Pesticide appliers, biocides, and birth defects in rural Minnesota. Environ Health Perspect 1996;104:394–9.
15. Kristensen P, Irgens LM, Andersen A, et al. Birth defects among offspring of Norwegian farmers, 1967–1991. Epidemiology 1997;8:537–44.
16. Garcia AM, Benavides FG, Fletcher T, et al. Paternal exposure to pesticides and congenital malformations. Scand J Work Environ Health 1998;24:473–80.
17. Kogevinas M, Sala M. Pesticides and congenital malformations—how many studies will it take to reach a conclusion? Scand J Work Environ Health 1998;24:445–7.
18. Shaw GM, Wasserman CR, O'Malley CD, et al. Maternal pesticide exposure from multiple sources and selected congenital anomalies. Epidemiology 1999;10:60–6.
19. Garcia AM, Fletcher T, Benavides FG, et al. Parental agricultural work and selected congenital malformations. Am J Epidemiol 1999;149:64–74.
20. Yoon PW, Olney RS, Khoury MJ, et al. Contribution of birth defects and genetic diseases to pediatric hospitalizations. A population-based study. Arch Pediatr Adolesc Med 1997;151:1096–103.
21. Clark EB. Cardiac embryology. Its relevance to congenital heart disease. Am J Dis Child 1986;140:41–4.
22. Clark EB. Growth, morphogenesis, and function: the dynamics of cardiac development. In: Moller JH, Neal WA, eds. Fetal, neonatal, and infant cardiac disease. Norwalk, CT: Appleton and Lange, 1990:3–23.
23. Pexieder T. Conotruncus and its septation at the advent of the molecular biology era. In: Clark EB, Markwald RR, Takao A, eds. Developmental mechanisms of heart disease. Mt. Kisco, NY: Futura Publishing Co, 1995:227–47.
24. Lupinetti FM, Bove EL, Minich LL, et al. Intermediate-term survival and functional results after arterial repair for transposition of the great arteries. J Thorac Cardiovasc Surg 1992;103:421–7.
25. Tikkanen J, Heinonen OP. Risk factors for conal malformations of the heart. Eur J Epidemiol 1992;8:48–57.
26. Adams MM, Mulinare J, Dooley K. Risk factors for conotruncal cardiac defects in Atlanta. J Am Coll Cardiol 1989;14:432–42.
27. SAS Institute, Inc. SAS/STAT user's guide, version 6. 4th ed. Cary, NC: SAS Institute, Inc, 1989.
28. Kleinbaum DG, Kupper LL, Muller KE, et al. Applied regression analysis and other multivariable methods. Pacific Grove, CA: Duxbury Press, 1997.
29. Baker SR, Wilkinson CF, eds. The effects of pesticide on human health. In: Advances in modern environmental toxicology. Vol XVIII. Princeton, NJ: Princeton Scientific Publishing Co, 1990.
30. Dorman DC. Anticoagulant, cholecalciferol, and bromethalin-based rodenticides. Vet Clin North Am Small Anim Pract 1990;20:339–52.
31. van Sittert NJ, Tuinman CP. Coumarin derivatives (rodenticides). Toxicology 1994;91:71–6.
32. Lewis GR. 1,001 chemicals in everyday products. New York, NY: Van Nostrand Reinhold, 1994.
33. Yasuda M, Maeda H. Teratogenic effects of 4-chloro-2-methylphenoxyacetic acid ethylester (MCPEE) in rats. Toxicol Appl Pharmacol 1972;23:326–33.
34. Buslovich SIu, Aleksashina ZA, Kolosovskaia VM. Effect of phenobarbital on the embryotoxic action of 2-methyl-4-chlorophenoxyacetic acid. (In Russian). Farmakol Toksikol 1979;42:167–70.
35. Shepard TH. Catalog of teratogenic agents. 8th ed. Baltimore, MD: Johns Hopkins University Press, 1995.
36. Ferencz C, Loffredo CA, Correa-Villasenor A, et al. Genetic and environmental risk factors of major cardiovascular malformations: the Baltimore-Washington Infant Study 1981–1989. Armonk, NY: Futura Publishing Company, 1997.
37. Manner J, Seidl W, Steding G. Correlation between the embryonic head flexures and cardiac development. Anat Embryol (Berl) 1993;188:269–85.
38. Manner J, Seidl W, Steding G. Embryologic observations on the morphogenesis of double-outlet right ventricle with subaortic ventricular septal defect and normal arrangement of the great arteries. Thorac Cardiovasc Surg 1995;43:307–12.
39. Botto LD, Loffredo CA, Scanlon KS, et al. Cardiac outflow tract defects in the offspring of mothers who took retinol supplements. Proc Greenwood Genet Center 1997;16:146–7.

Downloaded from https://academic.oup.com/aje/article-abstract/153/6/529/136848 by guest on 11 April 2019