Birth defects in Tarnaveni area, Romania – preliminary study results

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PUBLIC HEALTH

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Abstract

Background and aims. In the development of any human body, defects may occur, resulting in the occurrence of congenital malformations, also referred to as birth defects. The aim of this preliminary study was to assess the prevalence of birth defects registered during a period of 5 years in Tarnaveni area. As Tarnaveni is located in close proximity to a former chemical plant, a recognized hazardous waste site, we conducted this pilot study to assess the prevalence of birth defects, in order to evaluate the need for a more comprehensive investigation of a potential relation between the exposure to toxic metals contaminating the environmental media as a result of the past industrial activities, and the prevalence of the birth defects in this area.

Methods. We abstracted birth information (gestational age at delivery (GA), birth weight (BW), birth length (BL), head circumference (HC), and major structural birth defects), from medical records at "Dr. Gheorghe Marinescu" Tarnaveni Municipal Hospital, of the 2010-2014 period. We expressed BW as Z-scores relative to expected mean values at each gestational age for a reference population, calculated the ponderal index, and determined the 5 years birth defects prevalence among live births during the study interval.

Results. The 5 years (2010-2014) prevalence of birth defects, was 3.3% (95% confidence interval (CI): 2.47, 4.09). There were n = 163 (8.7%) preterm deliveries (less than 37 weeks of gestation at delivery), mean birth weight was 3108.3 g (standard deviation (SD) = 517.1), ranging from 450–4600 g, and n = 187 (10%) were low birth weight (LBW) (less than 2500 g). The ponderal index was 2.2 g/cm^3 on average (SD = 0.5), with range 1.2–20.7 g/cm^3.

Conclusions. While preliminary, our data show a 5 years (2010-2014) prevalence of major structural birth defects among newborns from Tarnaveni area of 3.3%. These pilot results indicate the need for a more comprehensive investigation of a potential relation between the exposure to toxic metals contaminating the environmental media as a result of the past industrial activities and the prevalence of the birth defects in Tarnaveni area.

Keywords: hazardous waste site, heavy metals, birth defects, low birth weight, Tarnaveni

Introduction

Birth defects, also known as congenital anomalies, congenital disorders or congenital defects have been defined as anomalies of the structure and/or function of an organ and/or a system in the body, detectable prenatally (by ultrasound examination), at birth or during infancy [1]. Their prevalence at a European level, reported for 2003-2007 period, by European Surveillance of Congenital Anomalies (EUROCAT) was 2.39 per 100 births [2]. However, EUROCAT reported 2010-2014 birth defects prevalence of 2.5 per 100 births for Ukraine, 1.35-2.25 per 100 births for Poland, and 2.25-2.75 per 100 births for Austria [3]. Yet, for Romania, Zavate et al. [4] reported a birth defects prevalence of 1.3 per 100 live births during the 2003-2007 period, lower than the reported prevalence at the European level. The most frequent severe birth defects worldwide, are congenital heart defects, neural tube defects and Down syndrome.

While the etiology of most birth defects is unknown, genetic factors (e.g. single gene defects, chromosomal disorders, multifactorial inheritance) and environmental factors (e.g. environmental chemicals with teratogenic effects, high doses of radiations, iodine and/or folic acid diet deficiency, and biological environmental agents such as rubella virus, among others) are likely to cause birth

defects [5]. Also, advanced maternal age is a risk factor for non-disjunction related abnormalities in fetal development (chromosomal abnormalities, such as Down syndrome) [6].

Previous reports linked higher risks for birth defects with proximity to hazardous waste sites [7-9] and to heavy metals exposure [10,11]. Thus, arsenic (As), lead (Pb) and manganese (Mn) exposure via contaminated environmental media has been linked to higher frequencies of birth defects, such as congenital heart defects and neural tube defects in human populations [12-14].

Birth defects are an important cause of infant death, worldwide. In the European Region, the World Health Organization (WHO) estimated that up to 25% of neonatal deaths are attributable to congenital anomalies [6]. Also, congenital disorders are an important issue for the health services, requesting multidisciplinary medical care and significant financial resources [1].

The aim of this preliminary study was to assess the prevalence of birth defects registered during the 5 years period from 2010-2014, in Tarnaveni area. Tarnaveni is located in close proximity to a former chemical plant and is recognized as a hazardous waste site with high levels of heavy metal contamination [15]. Thus, we conducted this pilot study to assess the prevalence of birth defects, in order to evaluate the need for a future comprehensive study of a potential relation between the toxic metals exposure via contaminated environmental media and the prevalence of the birth defects in this area.

Methods
Study area
Tarnaveni is a town in Transylvania (Mures County), located on the Tarnava Mica River, with 22,075 inhabitants [16]. A chemical plant located close to the town operated until 2007, and produced chemical products containing hexavalent chromium (Cr^{6+}). Millions of tons of chemical waste, including Cr^{6+} and other heavy metals remained on the former industrial platform, in close proximity to the town and river [17].

Birth outcomes/defects data collection and analysis
We abstracted de-identified birth information from the birth registries between 2010-2014, including GA, BW, BL, and HC. We collected information on structural birth defects at delivery from clinical observation sheets corresponding to each delivery and ultrasound examination results. All records were kept at the Gynecology and Obstetrics Department of the "Dr. Gheorghe Marinescu" Municipal Hospital in Tarnaveni. Birth defects were defined as anomalies of structure and/or organ/system function diagnosed by a physician before delivery using ultrasound, or after delivery, but prior to hospital discharge.

We calculated a 5 years prevalence of birth defects among the total number of live births during that period of time (as number of birth defects x 100/number of live births). We did not include n=14 cases of systolic murmur in the calculation of the 5 years prevalence, as these cases did not need further therapeutic intervention and so, were more likely to reflect a delayed fetal growth trajectory rather than a true defect. However, for comparison, we also calculated the 5 years prevalence including the 14 cases of systolic murmur. We used Excel 2016 (Microsoft Co., Redmond, Washington, USA) to calculate the 5 years prevalence of birth defects and 95% CI, overall and according to maternal age, and also, for plotting the percent of diagnosed birth defect among the live births in Tarnaveni, between 2010-2014 (categorized at organ and system level).

We considered GA as a continuous variable and dichotomized at less than 37 weeks, to indicate preterm delivery [18]. We calculated the Z-score for BW (BW-Z), using reference values published by Kramer et al. [19], as (BWs – BWr)/SDr, where BWs = BW of our population sample; BWr = the mean values of BW at each gestational age for the reference population; SDr = standard deviation for the BW of the reference population). We defined small for gestational age (SGA) infants, an indicator of intrauterine growth restriction, as birth weights less than the 10th percentile of the expected value from the reference population [18]. We also dichotomized BW, to indicate LBW at less than 2500 g [18], and calculated the ponderal index (PI), an indicator of growth proportionality, as BW (g) x $100/BL^3$(cm).

We used Stata v12 (StataCorp LP, College Station, TX, USA) to generate descriptive statistics, including mean, SD, median, 25th and 75th percentiles, and the range of values for continuous variables (GA, BW, BL, HC, PI, BW-Z) and to generate frequency tables for categorical variables (n, %). We defined statistical significance as p <0.05 for a 2-tailed hypothesis test.

Results
Table I describes the distribution of birth outcomes during 2010-2014 among Tarnaveni area newborns.

Table II shows the type of birth defects diagnosed in newborns from Tarnaveni area during 2010-2014, and the prevalence of birth defects among live births in this area. The 5 year birth defects prevalence was 3.3% overall excluding n=14 cases of systolic murmur, and 4.03% overall, when including the 14 cases of systolic murmur. Most birth defects were isolated anomalies affecting an organ (e.g. anomalies of the genital organs - cryptorchidism) or system (e.g. nervous system – congenital hydrocephalus), but there were also several (n=3) multiple organ and system abnormalities.

In Table III, we show the frequency and prevalence of birth defects by mother's age in our study area, with 95% CI. The prevalence of total birth defects increased with mother's age, with the highest prevalence among mothers aged 35-40 years (7%; 95% CI 2.58, 11.37).

The birth defects diagnosed in newborns from Tarnaveni area, between 2010-2014, included congenital defects of the cardiac chambers and connections, chromosomal

abnormalities, defects of the digestive system, nervous system, urinary and genital organs, and endocrine glands, ear structural defects, defects of the musculoskeletal system, and multiple birth defects (Figure 1). The birth defects of the musculoskeletal system comprised the largest proportion (36.1%) of total diagnosed birth defects, closely followed by the of the cardiac chambers and connections defects (22.9%), and then, by defects of the urinary and genital organs (18.0%).

Table I. Distribution of birth outcomes in Tarnaveni area, in 2010-2014 period.

Outcome	N	Mean (%)	SD	Min.	25th percentile	Median	75th percentile	Max.
Gestational age (weeks)	1871	38.8	1.9	23	38	39	40	43
Caesarean birth	322	17.2%	-	-	-	-	-	-
Preterm delivery (less than 37 weeks)	163	8.7%	-	-	-	-	-	-
BW (g)	1871	3108.6	517.2	450	2800	3100	3450	4600
LBW (less than 2500 g)	187	10%						
BW-Z	1863a	-0.5	0.8	-3.4	-1.1	-0.6	0.02	3.2
SGA	189a	10%						
BL (cm)	1871	51.9	2.6	21	51	52	53	60
PI (g/cm³)	1871	2.2	0.5	1.2	2.1	2.2	2.3	20.7
HC (cm)	1871	33.1	1.7	20	32	33	34	52
Male baby	982	52.5%						

BL, birth length; BW, birth weight; LBW, low birth weight; BW-Z, birth weight Z score; HC, head circumference; SGA, small for gestational age; PI, ponderal index; N, frequency; mean, arithmetic average; SD, standard deviation; Max., maximum reported value; Min., minimum reported value; median, 50th percentile.
[a] calculated only for singleton live births

Table II. Prevalence (%) of birth defects relative to the total number of live births (N), in Tarnaveni area, in 2010-2014 period.

	N	5 years birth defects prevalence (%) and 95% CI	Diagnosed birth defects	Frequency of the reported birth defects (n cases)
Live births	1861[a]			
Birth defects	61[b]	3.3% (95% CI: 2.47, 4.09)	**Cardiac chambers and connections defects, unspecified**	**14**
			Musculoskeletal system defects	**22 (total)**
			congenital clubfoot	*11*
			congenital dislocation of the hip, unilateral	*1*
			diastasis rectus abdominis	*1*
			polydactyly, unspecified	*3*
			syndactyly of toes (fused toes)	*1*
			unspecified congenital defect of the left hand	*1*
			unspecified congenital bone and joint defect	*2*
			omphalocele	*1*
			bilateral inguinal hernia	*1*
			Endocrine glands defects	**1**
			congenital hypothyroidism	1
			Nervous system defects	**5 (total)**
			congenital hydrocephalus	*1*
			congenital generalized hypertonia	*2*
			congenital generalized hypotonia	*1*
			unspecified congenital neurological disease	*1*
			Congenital ear defect, unspecified	**1**
			Urinary and genital organs defects	**11 (total)**
			bilateral undescended testis	6
			cryptorchidism	1
			hypospadias	2
			congenital ovarian cyst	1
			unspecified congenital defect of the kidney	1
			Digestive system defects	**1 (total)**
			anorectal atresia	*1*
			Chromosomal abnormalities	**3 (total)**
			Down syndrome	*3*
			Multiple birth defects, unspecified	**3**

N, total reported number of live births
[a] newborns with Apgar score = 0 were excluded (only live births were included in the analysis;
[b] excluded n=14 infants with minor cardiac defects clinically expressed as systolic murmur
Note: the 5 year prevalence of birth defects increases to 4.03% (95% CI: 3.14, 4.92) when including those n=14 infants.

Table III. Frequency (n cases) and prevalence (%) of birth defects by mothers' age (years), in Tarnaveni area, in 2010-2014 period

Diagnosed birth defects	13-20	20-30	30-35	35-40	above 40
Defects of cardiac chambers and connections	6	4	3	1	0
Chromosomal abnormalities	1	0	1	1	0
Defects of digestive system	0	1	0	0	0
Ear malformation	0	1	0	0	0
Endocrine glands malformation	1	0	0	0	0
Multiple defects	2	0	0	1	0
Musculoskeletal system defects	3	12	4	3	0
Nervous system defects	0	2	1	1	1
Urinary and genital organs defects	1	6	2	2	0
Total defects	**14**	**26**	**11**	**9**	**1**
Total live births	**367**	**1002**	**348**	**129**	**25**
Prevalence (%) of total birth defects among total live birth, by mother's age and (95% CI)	*3.8* *(1.85, 5.77)*	*2.6* *(1.61, 3.58)*	*3.2* *(1.32, 5.00)*	*7.0* *(2.58, 11.37)*	*4.0* *(-3.68, 11.68)*
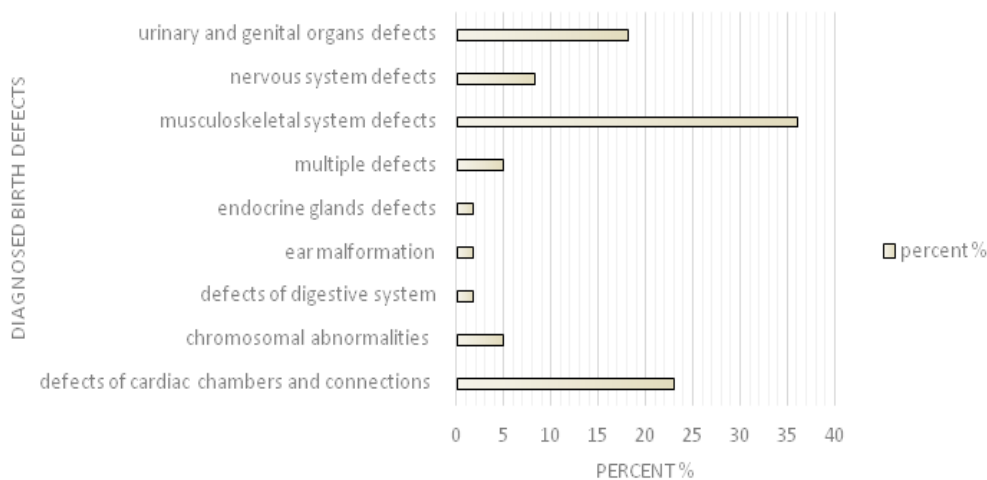
Figure 1. Proportion of total congenital defects by organs and system, in Tarnaveni area, in the 2010-2014 period

Discussion

In this preliminary study we report a prevalence of 3.3% birth defects, among Tarnaveni area newborns, for the 5 years period from 2010-2014. This value is higher than the 1.3% prevalence reported for Romania overall, during 2003-2007 [4]. But yet, we should not rule out the possibility that this difference to be only apparent, due to the differences in detection/survival, and the small population sample size in our pilot study may also, account, in part, for this difference. We also showed that, 9% of deliveries in Tarnaveni area between 2010 and 2014, were preterm deliveries, 10% were LBW, and 10% were SGA.

The birth defects prevalence rate in Tarnaveni area was higher than that reported for all of Cluj County during 2003-2007, 1.33% [4], Tarnaveni's birth defect prevalence also eclipsed the 0.69% prevalence reported for the entire 6 counties North West region of Romania (i.e., Bihor, Bistrita-Nasaud, Cluj, Maramures, Satu Mare, and Salaj counties) during the same time period (2003-2007) [4]. We can speculate that the differences might be due in part to time trends, as the Tarnaveni data are from 2010-2014, and due to differences in the ascertainment, diagnosis, and reporting of birth defects. Preconception care and socio-economic differences between Cluj County and the

Romanian North West region as compared to Tarnaveni area, are also likely to be important. However, the higher birth defects prevalence in Tarnaveni area might also be attributed in part to reproductive toxicity associated with the presence of the hazardous waste site in close proximity to the town, which has contaminated the environmental media in the surroundings with heavy metals.

We conducted this investigation to assess the prevalence of birth defects in this area of Romania, as birth defects are a leading cause of infant mortality worldwide. In Romania, infant mortality due to birth defects was 21.2 per 10,000 live births in 2012, a decrease from 40.2 per 10,000 live births in 1990 [20]. The etiology remains unknown in approximately half of birth defects [21], while the other half is attributed to genetic factors and to complex gene-environment interactions [22]. Previous studies that looked into the etiology of birth outcomes and morphological abnormalities, have indicated that environmental contaminants may be associated with a significant increase in the prevalence of birth defects [23,24]. The exposure to a toxic chemical compound might cause a birth defect (congenital malformation) via a mechanism of preconceptional mutation (including chromosomal abnormalities) or via a mechanism of teratogenic action after conception [23]. Brent and Beckman [25,26] estimated that 10% of major birth defects are caused by teratogenic chemicals, while 37% result from the interaction between genetic and environmental factors. More recent estimates suggest that as much as 80% of developmental diseases in children are due to gene-environment interactions [27]. Still, many birth defects caused by chromosomal abnormalities are incompatible with survival and affected fetuses are spontaneously aborted early in gestation [28,29]. Thus, identifying modifiable risk factors for the risk of congenital defects is an urgent public health priority [30].

Previous studies link environmental heavy metal exposures to birth defects. Higher risks for congenital cardiovascular defects (odds ratio (OR) = 2.2; 95% CI: 1.3, 3.9) were reported for children born from mothers living in close proximity to contaminated sites, who were exposed to heavy metals and cyanides during pregnancy [31]. A study conducted in North Carolina [14], reported an association between higher levels of exposure to Mn (prevalence ratio (PR): 1.6; 95% CI: 1.1, 2.5) and Pb (PR: 1.7; 95% CI: 0.9, 3.3) via drinking water and an increased prevalence of heart defects. In another study, higher levels of Mn in the placenta were linked to a 4-fold (95% CI: 1.23, 14.79) higher risk of neural tube defects and a 7-fold (95% CI: 1.52, 39.64) higher risk of spina bifida [32]. Elevated maternal blood levels of Pb, As, cadmium (Cd) and copper (Cu) were risk factors for neural tube defects in a study conducted in Turkey [33]. A study by Vinceti et al., indicated a higher risk for cardiovascular defects, oral clefts, and musculoskeletal defects associated with exposure to Pb in northern Italy [34]. A study conducted in California, showed that women living in the surroundings of a factory using Cr^{6+}, had an increased risk for pregnancy loss, and the risk for birth defects, such as genitourinary, eye, ear, face, neck, cleft defects and chromosomal anomalies in offspring, was also elevated [35]. A recent study by Ou et al. [36], evaluating the relationship between heavy metal levels measured in maternal blood (Pb, Cd, Cr, Cu, mercury (Hg), and selenium (Se)) and congenital heart defects in offspring, found that high maternal blood Pb (OR=12.09, 95% CI: 2.81, 51.97) was associated with heart defects in offspring, suggesting that exposure to levels at the current biological limit for protecting human health (10μg/dL), may still harm an unborn child.

In the light of scientific literature on this topic, we conducted our pilot study to assess the prevalence of birth defects in Tarnaveni, an area highly contaminated with heavy metals, for comparison with the reported birth defects prevalence values for other areas of Romania and at the national level. The higher prevalence of the birth defects in Tarnaveni area that we show in this pilot study, indicates the need for a more comprehensive investigation of risk factors for adverse birth outcomes and birth defects, in particular from maternal exposure to the toxic metals contaminating the local environment. To investigate the role of exposure to hazardous substances in the environmental media as risk factors for birth defects in the population living in this area, we plan to collect soil, vegetable, and biological samples as part of a future analysis.

This pilot study is a preliminary component of a larger ongoing research project, in which we will analyze heavy metal concentrations in soil, vegetables, and maternal blood specimens in a population sample from Tarnaveni, to evaluate the relationships with the prevalence of birth defects and adverse birth outcomes. The successful completion of our extended study will provide scientific data that are currently missing in this area, concerning the exposure to heavy metals in the environmental media and the potential related effects on fetal in utero development. Also, these data might be used to design interventions to reduce the prevalence of birth defects in Tarnaveni, by offering regulators and legislators a valid scientific foundation upon which to make critical public health decisions and to initiate management and risk communication programs for protecting the health of susceptible populations.

To the best of our knowledge, this is the first study that has analyzed data on birth defects in Tarnaveni area historically polluted with Cr^{6+}, a highly toxic contaminant for which there is a body of scientific evidences suggesting that gestational exposure may be associated with low birth weight and internal and skeletal malformations, and is reporting a higher prevalence of birth defects in this area than in other regions and at national level, raising serious concerns on a possible association of this higher prevalence with the environmental exposure to Cr^{6+}.

Conclusions

While preliminary, our data showed a 5 years (2010-2014) prevalence rate of birth defects higher in Tarnaveni area than for other North West Romanian Counties and for Romania overall. We found the highest prevalence for the birth defects of the musculoskeletal system, and also, as expected, the prevalence of birth defects increased with mother's age. We have an ongoing investigation in this area (soil, vegetables and biological samples collection and analysis) to evaluate potential associations between exposure to toxic metals in the residential areas surrounding the site of the former chemical plant in Tarnaveni, and birth defects prevalence.

Acknowledgements

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