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Environmental Pollutants in Relation to Complications of Pregnancy

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Certain complications of pregnancy, e.g., threatened spontaneous abortion, toxemia, emesis, and anemia, were studied in pregnant women living in industrial areas contaminated by smelters and the petrochemical industry. Exposure to lead or aromatic hydrocarbons was assessed in parallel by the determination of these agents or their metabolites in blood and urine. Comparison of respective exposure levels was made between women with normal pregnancies and those with complications. Significantly higher levels of lead in blood and increased excretion of the metabolic products of organic solvents were found in women with complicated pregnancies compared to those with normal pregnancies. Threatened spontaneous abortion, toxemia, and anemia were associated with higher lead exposure in the vicinity of smelters. In these patients, evidence of disturbances of blood glutathione equilibrium and increased lipid peroxidation were found indicating a decreased ability to compensate for the effects of exposure. Styrene exposure in a petrochemical industrial area was associated mainly with late toxemia and nephropathy. Patients with these complications also had a tendency to elevated exposure to other aromatic hydrocarbons. It is suggested that complications of pregnancy may be induced by environmental agents at levels lower than those that result in pregnancy loss or preterm birth.

Introduction

A number of environmental agents have been associated with unfavorable pregnancy outcomes such as spontaneous abortion, prematurity, and stillbirth. Although these end points have frequently been studied in connection with exposure to environmental factors, less attention has been given to complications occurring during pregnancy such as anemia, toxemia, emesis, and threatened spontaneous abortion, which may not end in fetal death or premature birth but nevertheless are a frequent threat to maternal health and prenatal development. Eclampsia from toxemia of pregnancy is one of the principal causes of antepartum maternal death (*1*).

It is possible that disorders occurring in the course of pregnancy could be influenced by environmental agents at exposure levels lower than those provoking loss of pregnancy. Our recent studies (*2*) have shown a significantly higher prevalence of toxemia, spontaneous abortion, and prematurity among the general population living in areas polluted by the heavy metal and chemical industries. Environmental lead contamination has had a highly significant correlation with the rate of maternal toxemia.

The present study was designed to assess the relationship between complications of pregnancy and biological exposure to agents known to be developmental and reproductive hazards, *viz.*, lead and organic solvents, in pregnant women residing in areas of industrial pollution.

Materials and Methods

A total of 305 pregnant women were studied during their last trimester of pregnancy, of whom 175 were residents of metal-smelting areas polluted by lead to differing degrees, and 130 lived in a region contaminated with hydrocarbons and organic solvents from a petrochemical plant. The extent of the exposure to these pollutants was assessed by analysis of the substances or their metabolites in blood and urine.

For assessment of lead exposure, blood lead levels (AAS determination) and urinary excretion of δ-aminolevulinic acid (*3*) were assayed. In addition, blood glutathione (*4*) and lipid peroxides in blood (*5*) were determined as indicators of lead binding to thiol groups and lead-induced peroxidation in body tissues.

For assessment of exposure to aromatic hydrocarbons in the petrochemical plant area, urinary excretion of hippuric acid, a metabolic product of toluene, xylene, benzene, and its derivatives (*6*), and urinary excretion of mandelic acid, a metabolite of styrene (*7*), were determined.

In each individual case, data were collected documenting the presence of complications in the course of pregnancy such as anemia, hyperemesis, toxemia and threat-

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Table 1. Parameters of lead exposure, blood glutathione, and lipid peroxides in normal and pathological pregnancy in a metal-smelting region.[a]

	Blood lead concentration, μg/dL (range)	δ-ALA in urine, mg/g creatinine (% over 5 mg/g)	Blood glutathione, mg %			Lipid peroxides in blood, μEq/mL
			Total	Oxidized	Reduced/total, %	
Normal pregnancy ($n=22$)	5.2 ± 0.2 (2.9–7.4)	4.08 ± 0.45 (27)	18.7 ± 0.9	8.0 ± 0.9	59	0.66 ± 0.10
Anemia ($n=15$)	6.4 ± 0.5* (3.3–11.2)	4.46 ± 0.67 (33)	21.1 ± 1.9	7.2 ± 1.2	66	0.69 ± 0.13
Toxemia, all cases ($n=19$)	6.5 ± 0.5* (3.3–11.2)	4.22 ± 0.54 (32)	19.7 ± 1.5	9.2 ± 1.1	54	0.70 ± 0.11
Proteinuria and arterial hypertension ($n=7$)	6.8 ± 1.1* (4.1–11.2)	4.54 ± 0.92 (43)	19.0 ± 2.2	9.9 ± 1.2	51	0.72 ± 0.23
Hyperemesis ($n=5$)	6.7 ± 1.0* (5.8–10.4)	4.17 ± 0.91 (20)	22.6 ± 3.8	8.9 ± 1.1	57	0.81 ± 0.26
Threatened spontaneous abortion ($n=24$)	7.1 ± 0.7* (3.9–22.2)	4.22 ± 0.51 (30)	21.8 ± 1.4	10.3 ± 1.1	55	0.44 ± 0.08
Hospitalized cases ($n=7$)	9.9 ± 2.2† (5.8–22.2)	3.44 ± 0.11 (17)	24.2 ± 3.1	10.8 ± 1.9	54	0.58 ± 0.22

Abbreviations: ALA, aminolevulinic acid.
[a]Values are means ± SE.
*$p < 0.05$.
†$p < 0.01$.

ened spontaneous abortion and/or premature birth. Toxemia was diagnosed when patients developed edema, proteinuria, or arterial hypertension; the combination of all three was diagnosed as nephropathy. To reduce the effect of confounding factors, patients with a past or present history of renal, hepatic, endocrine, blood, or cardiovascular disease, habitual abortions, previous sterility, or abnormal gynecological status were excluded from the study. Other relevant factors such as age, parity, order of pregnancy, previous spontaneous or induced abortion, medication and drug use during pregnancy, maternal and paternal occupation, smoking and drinking habits, and socioeconomic status were taken into account. Personal interviews and medical records in local clinics that documented the regular monthly clinical and paraclinical examination of all registered pregnancies were used as sources of information.

Each of the groups studied were further subdivided into groups of either normal pregnancy or pregnancy with complications. For each pollutant studied, comparisons were made between the levels of exposure in the cases of normal and complicated pregnancies. Statistical evaluations were performed using Student's t tests and chi-square test.

Results

Lead Exposure and Complications of Pregnancy

Table 1 summarizes the results of the study performed in the area with the higher level of lead pollution. The average monthly concentration of lead in the ambient air of this region reached as high as 3.5 μg/m^3. Among the study cases, 22 had a normal pregnancy without any complications. The blood lead levels in these 22 normal cases ranged from 2.9 to 7.4 μg/dL, with a mean of 5.3 μg/dL. Threatened spontaneous abortion was the most common complication, followed by toxemia and anemia. The highest lead levels were detected in the threatened spontaneous abortion subgroup (mean 7.1 μg/dL; range 3.9–22.2 μg/dL). The mean blood lead levels of the subgroups of toxemia and anemia were similar to each other ranging from 3.3 to 11.2 μg/dL (6.4 μg/dL). However, higher mean values were detected in certain categories of toxemia, notably those with associated hyperemesis (6.7 μg/dL) or those with proteinuria and arterial hypertension (6.8 μg/dL). The majority of cases did not require hospitalization, however, in the more severe cases who were hospitalized, the blood lead levels averaged 9.9 μg/dL (range: 5.8–22.2 μg/dL), and this was the highest level of all the subgroups.

The proportion of patients with blood lead levels greater than 7 μg/dL within each of the subgroups is shown in Table 2. Only 4.5% of the normal cases had levels above 7

Table 2. Incidence of blood lead concentrations above 7 μg/dL in normal and complicated pregnancies.[a]

Diagnosis	Blood lead over 7 μg/dL, % of cases
Normal pregnancy	4.5 (1/22)
Anemia	40.0 (6/15)*
Threatened spontaneous abortion	41.7 (10/24)*
Toxemia (total)	47.3 (9/19)*
Toxemia (late)	50.0 (7/14)†
Hospitalized patients (all causes)	57.1 (4/7)‡

[a]Same patients as in Table 1.
*$p < 0.05$.
†$p < 0.01$.
‡$p < 0.001$.

μg/dL, whereas 40–50% of patients with complicated pregnancies had levels above this value, most frequently in the toxemia group, and especially among those requiring hospitalization.

δ-Aminolevulinic acid (δ-ALA) excretion in urine was moderately elevated in association with complicated pregnancies. Higher levels were detected in the cases of anemia and toxemia diagnosed in late gestation, and particularly among those with proteinuria and arterial hypertension. It should be noted that all cases of anemia had been treated with folates, vitamins, and iron-containing drugs at the time of analysis, which suggests that δ-ALA excretion might have been higher before treatment.

Urinary excretion of δ-ALA was lower in patients with threatened spontaneous abortion or hyperemesis than in the above conditions, but blood lead levels were higher, suggesting that recent lead exposure was of greater importance for threatened abortion and emesis than for toxemia and anemia. An explanation for this may be that spontaneous abortion and emesis both occur most often in early pregnancy, whereas lead mobilization from the maternal body pool takes place later, after the 24th gestational week (*8*). This conclusion is further supported by our finding in the group of hospitalized patients that had only threatened spontaneous abortion and hyperemesis, who had a higher mean blood lead level than any other group. δ-Aminolevulinic acid excretion, however, was lower than that found in the other groups (Table 1). A δ-ALA level of over 5 mg/g creatinine was recorded in only one of the hospitalized patients in whom signs of toxemia (edema gravidarum) were observed.

Evidence of disturbed glutathione equilibrium, e.g., increased oxidized form and a decrease in the active reduced form, was found in association with increased lead exposure (Table 1), suggesting an impairment of the compensatory mechanisms of the patients against the well known damaging oxidative effect of lead on tissues (*9*). Products resulting from this damage such as lipid hydroperoxides can amplify the tissue-damaging process. This effect was more pronounced in patients with late toxemia (proteinuria and arterial hypertension), who had the lowest ratios of reduced/total glutathione combined with elevated levels of lipid peroxides in the blood. A slight elevation in the levels of total glutathione was detected in all categories of complications, suggesting a compensatory increase in glutathione synthesis. This compensation, however, appears to be insufficient to counteract the increased formation of lipid peroxides, as these were detected in elevated concentrations in cases of hyperemesis, late toxemia and anemia. That lead exposure accounted for the increased lipid peroxidation and changes in glutathione equilibrium was confirmed by comparison with an area that had a lower level of lead pollution (Table 3). This comparison only involved normal pregnancy cases in order to avoid any possible interference caused by the process of the pregnancy complication itself. Blood lead levels in cases from the less polluted area were half as high and were parallelled by markedly lower levels of lipid peroxides in blood, lower oxidized glutathione, and greater reserves of its reduced form.

Table 3. Blood lead, glutathione, and lipid peroxides in areas of high and low lead exposure (cases of normal pregnancy).

Parameter	High-exposure area	Low-exposure area
Lead in blood, μg/dL		
Mean ± SE	5.17 ± 0.24*	2.23 ± 0.18
Range	2.9–7.4	1–3.3
Glutathione in blood, mg %		
Total (mean ± SE)	18.2 ± 0.9	18.1 ± 1.2
Oxidized (mean ± SE)	8.0 ± 0.9	6.7 ± 0.9
Total/reduced (%)	58.7	63.9
Lipid peroxides in blood, μEq/mL (mean ± SE)	0.66 ± 0.10*	0.06 ± 0.02

*$p < 0.01$.

Analysis of confounding factors (Table 4) revealed a higher percentage of maternal smoking before and during gestation among complicated pregnancies compared to normal, and an increased proportion of women with supplementary occupational exposure in the toxemia group, but the differences were not statistically significant. A significantly higher incidence of previous spontaneous abortion was found among the patients with complicated pregnancies, especially in the cases of threatened spontaneous abortion in the present pregnancy.

Exposure to Aromatic Hydrocarbons and Complications of Pregnancy

Higher exposures to aromatic hydrocarbons, as shown by the excretion of their metabolic products in urine, were found in women in the petrochemical plant area who had complications of pregnancy (Table 5). The mean value of the styrene metabolite mandelic acid was 60.6 mg/L for the total group of complicated pregnancies compared to 42.4 mg/L for the cases of normal pregnancy, and this difference was statistically significant. Excretion of hippuric acid was elevated but to a proportionally lesser degree (69.8 mg/L compared to 60.6 mg/L), and the difference did not reach statistical significance due to considerable variations in the individual values.

The highest mean levels of aromatic hydrocarbon metabolites, according to the type of complication, were found in late toxemia (nephropathy, arterial hypertension, proteinuria) and hyperemesis (Table 5). The mean values in cases of anemia and threatened spontaneous abortion did not differ significantly from normal pregnancy, although there was a tendency to elevation of mandelic acid levels. However, when patients hospitalized for threatened abortion were analyzed separately, a significant difference in mandelic acid excretion was found, indicating higher exposure levels to styrene in these more severe cases.

The elevation of mandelic acid level was more pronounced in all groups than that of hippuric acid, suggesting a prevailing exposure to styrene. Hippuric acid level was increased only in cases of early and late toxemia, indicating a possible parallel exposure to other organic solvents, but the differences were not statistically significant due to large individual variations. All individual

Table 4. Pregnancy complications and confounding factors in women exposed to the metal smelter industry.

Parameter	Normal pregnancy	Pregnancy with complications			
		Anemia	Threatened spontaneous abortion	Toxemia (total)	Toxemia (late)
Number of women (total = 94)	22	15	24	19	14
Age, years (mean ± SE)	22.7 ± 1.1	23.7 ± 1.3	24.3 ± 0.9	24.6 ± 1.4	24.7 ± 0.9
Order of pregnancy (mean ± SE)	1.7 ± 0.2	1.9 ± 0.4	2.0 ± 0.2	1.9 ± 0.2	1.9 ± 0.2
Number of births (mean ± SE)	0.5 ± 0.2	0.4 ± 0.2	0.5 ± 0.1	0.5 ± 0.1	0.6 ± 0.1
Previous abortions, %					
Spontaneous	0	20*	25*	15.8*	14.3
Induced	13.6	20	20.8	26.5	21.4
Smoking, %					
Before pregnancy	31.8	40	62.5	52.6	64.3
During pregnancy	4.5	26.7	20.8	15.8	21.4
Occupational exposure, %					
Maternal	4.5	6.6	4.2	10.5	7.2
Paternal	9.0	0	8.4	5.3	7.4

*$p < 0.05$.

values were within the physiological range of 34–440 mg/L, which is wide because other factors such as smoking and consumption of food with a natural content of benzoic acid influence the amount of hippuric acid excretion (*6*). In contrast, mandelic acid is not normally present in urine, so its excretion is considered to be a specific indicator of exposure to styrene (*7*). In this study the mean concentration of mandelic acid was 42 mg/L in patients with normal pregnancy, while the mean concentration in cases of complicated pregnancy was 60.6 mg/L. Table 6 compares the incidence of mandelic levels above 60 mg/L in normal and pathological pregnancies according to the type of complication. A gradation in the level of exposure is evident, the highest prevalence of concentrations greater than 60 mg/L being found among cases of early and late toxemia, reaching up to 100% for nephropathy, whereas such levels were present in only 16% of women with a normal pregnancy.

The analysis of other interfering factors showed no significant differences among the groups studied (Table 7). Patients with anemia during pregnancy had a higher incidence of maternal occupational exposure to solvents and a lower proportion of smokers, but these differences were not statistically significant.

Table 5. Urine excretion of metabolic products of aromatic hydrocarbons in normal and pathological pregnancy in a region of petrochemical industry.

Parameter	Mandelic acid, mg/L (mean ± SE)	Hippuric acid, mg/L (mean ± SE)
Normal pregnancy (n = 25)	42.0 ± 4.1	60.6 ± 9.7
Anemia (n = 19)	53.2 ± 4.9	70.0 ± 22.7
Threatened spontaneous abortion		
Total (n = 45)	57.7 ± 3.2	62.2 ± 9.6
Hospitalized (n = 14)	68.6 ± 4.4*	63.2 ± 11.3
Proteinuria (n = 24)	61.0 ± 5.5*	63.3 ± 13.8
Hyperemesis (n = 7)	67.9 ± 7.2*	102.1 ± 36.8
Arterial hypertension (n = 6)	70.0 ± 2.2†	87.6 ± 12.5
Nephropathy (n = 6)	72.5 ± 1.7†	96.7 ± 34.8
Pathological pregnancy, total	60.6 ± 2.9*	69.8 ± 10.5

*$p < 0.01$.
†$p < 0.001$.

Discussion

In summary, significantly higher levels of exposure to environmental toxic substances, such as lead and organic solvents, were found among women residents of areas polluted by metallurgical and petrochemical industries who had complications of their pregnancies. A previous study (*2*) has indicated an increased incidence of complications of pregnancy in those areas compared to the average complication rate over the whole country.

In the lead-polluted area, threatened spontaneous abortion, toxemia and anemia were associated with higher levels of lead exposure compared to noncomplicated pregnancies. Blood lead levels in excess of 7 μg/dL were found 10 times more commonly in patients with complicated pregnancies than those with normal pregnancies. In the Port Pirie study (*10*), maternal blood levels of over 8 μg/dL were associated with an increased risk of preterm delivery. The lower lead values that we have found in cases of threatened spontaneous abortion or other complications of pregnancy may be explained by the fact that in our study all the pregnancies were preserved at the time of examination. It is possible that levels of lead exposure lower than those leading to fetal death or premature birth may be involved in transient complications of pregnancy.

Table 6. Incidence of mandelic acid concentration above 60 mg/L urine in normal and complicated pregnancies in the region of a petrochemical plant.

Parameter	% of cases with mandelic acid over 60 mg/L urine (number/total)
Normal pregnancy	16 (4/25)
Anemia	31.6 (6/19)
Threatened spontaneous abortion	
Total	46.7 (21/45)*
Hospitalized	71.4 (10/14)†
Proteinuria	58.3 (14/24)*
Hyperemesis	71.4 (5/7)*
Arterial hypertension	100 (5/5)*
Nephropathy	100 (6/6)†

*$p < 0.05$.
†$p < 0.01$.

Table 7. Pregnancy complications and confounding factors in women living in the petrochemical plant area.

Parameter	Normal pregnancy	Pregnancy with complications		
		Anemia	Threatened spontaneous abortion	Toxemia
Number of women (total = 130)	25	19	45	41
Age, years (mean ± SE)	22.9 ± 0.9	24.5 ± 1.0	24.0 ± 0.7	24.2 ± 0.7
Order of pregnancy (mean ± SE)	1.8 ± 0.2	1.7 ± 0.2	1.7 ± 0.1	1.9 ± 0.1
Number of births (mean ± SE)	0.7 ± 0.1	0.6 ± 0.2	0.6 ± 0.1	0.7 ± 0.1
Previous spontaneous abortions, %	16	10.5	11.1	7.3
Smoking before and during pregnancy, %	24	5.3	28.9	26.8
Occupational exposure, %				
Maternal	16	36.8	13.3	9.8
Paternal	8	5.3	11.1	9.8

The correlation between environmental lead pollution and toxemia which was found in a previous study (*2*) was substantiated by the present investigation. This correlation could be explained by the known toxic effect of lead on the renal, cardiovascular, and nervous systems, which are involved in the pathogenesis of toxemia. In addition, the present study has demonstrated that lead exposure can lead to a decreased ability of the individual to cope with free radicals and other oxidizing substances, and an increased level of lipid peroxidation, which can cause tissue damage. Bearing in mind that environmental exposure is always complex and involves a variety of agents, the reduced compensatory reserves of the individual may render her more vulnerable to the impact of the various environmental factors. Our studies have shown that the correlation between lead exposure and toxemia is enhanced by simultaneous exposure to other agents such as chromium and hydrogen sulfide (*11*), which are typical pollutants in addition to lead in industrial metal-smelting areas.

Organic solvents have also been found to be involved in pregnancy complications. In the particular locality we studied, exposure to styrene was primarily associated with late toxemia, and especially with nephropathy. Styrene exposure appeared to influence threatened spontaneous abortion and anemia to a lesser extent, although a significantly higher level of exposure was detected in patients requiring hospitalization for threatened spontaneous abortion. Our results are supported by the observation of other authors (*12*) that there is a higher incidence of toxemia and anemia among women occupationally exposed to styrene.

The results of this investigation suggest that complications of pregnancy are important end points in the assessment of the impact of environmental agents on reproduction.

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