Lead Toxicity in The Pregnant Rat. II. Effects of Low-level Lead on δ-aminolevulinic Acid Dehydratase Activity in Maternal and Fetal Blood or Tissue

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Abstract: Lead was administered to pregnant rats in drinking water (5 ppm) from day 1 to day 18 or 21 of gestation, at which time the dams and fetuses were sacrificed. The red blood cell count and hemoglobin concentration were significantly lower in the lead-treated fetuses than in untreated controls on day 21 of gestation, but not in the lead-treated dams. The delta-aminolevulinic acid dehydratase (ALAD) activity of erythrocytes was significantly reduced in the lead-treated dams and their fetuses on days 18 and 21 of gestation. ALAD activity in the liver was significantly higher in the lead-treated fetuses than in the controls. There was no significant difference in placental ALAD activity between the lead-treated and control animals. Lead in tissues increased significantly in the liver of the lead-treated fetuses.

There were significant positive correlations between the maternal and fetal blood lead level, liver lead level and erythrocyte ALAD activity. There was a significant negative correlation between the maternal and fetal liver ALAD activity.

Key words: ALAD activity—Lead poisoning—Pregnant rats—Fetus—Erythrocyte—Tissue

INTRODUCTION

In a previous paper, the author reported the effects of lead on hematological parameters (red blood cell count, packed cell volume and hemoglobin concentration) and biochemical parameters (ALAD activities of the blood, liver and placenta) of the heme biosynthetic pathway in dams and fetuses following the administration of drinking water containing a high concentration (500 ppm) of lead to pregnant rats. He pointed out that (1) lead was transferred readily from the mother to the fetus, (2) lead had no apparently ill effects on the rats in the course of pregnancy, (3) the red blood cell count and hemoglobin concentration were markedly reduced in the lead-treated fetuses, but not in the lead-treated
dams, and (4) erythrocyte ALAD activity was significantly lower in both lead-treated dams and fetuses than in untreated controls. The results suggest that a strong inhibition of heme synthesis may have already occurred in the fetus.

Human beings, however, do not experience oral ingestion of a large amount of lead in normal daily life. Exposure to a low concentration of lead from the atmosphere and food is a serious problem in living organisms\(^2-3\). With the same biochemical parameters as adopted before\(^1\), this study was carried out to elucidate the effects of a low lead level on pregnant rats and their fetuses.

**MATERIALS AND METHODS**

Adult female Wistar rats, 10 weeks old, were caged overnight with a male of the same strain. The presence of a vaginal plug on the following morning was taken as indication of conception (day 1 of gestation). Five pregnant rats were given drinking water containing 5 ppm lead as lead acetate salt over a period beginning on day 1 of gestation. Five controls were maintained with distilled water. On days 18 and 21 of gestation, the dams and fetuses were sacrificed. The collection of blood, tissues and amniotic fluid has been described in detail in a previous paper\(^1\).

The following biochemical analyses were undertaken on whole blood: ALAD activity (\(\mu\text{MPBG}/100\text{ml RBC/hr}\)), packed cell volume (expressed in % PCV), hemoglobin concentration (expressed in g Hb/100 ml whole blood) and blood lead level (expressed in \(\mu\text{g Pb}/100 \text{ml whole blood}\)).

On whole homogenates of liver and placenta, the following measurements were carried out: ALAD activity (\(\mu\text{MPBG/g/hr}\)) and lead level (expressed in \(\mu\text{g Pb/g wet weight}\)). The lead level of amniotic fluid was also determined. The analytical methods used were the same as described previously\(^1\). The data were analyzed statistically by Student's t test.

**RESULTS**

1. *Reproductive data*

Reproductive parameters, including the numbers of implantation sites, resorption sites and fetuses, and fetal and placental weight are presented in Table 1. The percentage of resorption was higher in the lead-treated group than in the control group on day 18 of gestation. It was higher in the control group than in the lead-treated group on day 21 of gestation. The difference was not significant between the lead-treated group and the controls. Fetal weight was significantly lower in the lead-treated group than in the control group on days 18 and 21 of gestation. No external malformation was observed in living or dead fetuses.
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2. Hematological data and ALAD activity in dams

Hematological values and ALAD activity in the erythrocytes and liver are given in Table 2. There were no differences in the RBC count, PCV or Hb between the lead-treated group and control group. In the lead-treated group, erythrocyte ALAD activity decreased significantly to show 44.0% and 43.9% reduction on days 18 and 21 of gestation, respectively. Liver ALAD activity in the lead-treated group was reduced by 18.7 and 4.8% from that in the controls on days 18 and 21 of gestation, respectively, but there was no significant difference in the activity between the lead-treated group and the control group.

3. Hematological data and ALAD activity in fetuses

Hematological values and ALAD activity in the erythrocytes, liver and placenta are shown in Table 3. Both the RBC count and Hb concentration were markedly reduced in the lead-treated group on day 21 of gestation. Erythrocyte ALAD activity was significantly lower in the lead-treated group than in the control group.
It showed a 27.9% and 18.7% reduction on days 18 and 21 of gestation, respectively. Liver ALAD activity was significantly higher in the lead-treated group than in the control group. There was no significant difference in placental ALAD activity between the lead-treated and control groups.

4. Lead concentration

Table 4 presents the lead concentrations in the blood, liver, placenta and amniotic fluid. In the dams, there was no significant difference in the lead level of the blood or liver between the lead-treated and control groups, except for liver lead level on day 21 of gestation. In the fetuses, the lead concentrations of the blood and liver were significantly higher in the lead-treated group, except for blood lead level on day 18 of gestation. There was no significant difference in
the lead level of amniotic fluid between the lead-treated and control groups.

5. Correlation between parameters

Table 5 presents a correlation matrix that expresses the correlation between any two parameters in the dams and in the fetuses. In the dams, significant positive correlations were observed among the lead levels of the blood, liver, and placenta. On the other hand, significant negative correlations were found between blood lead and liver ALAD activity, between liver lead and erythrocyte ALAD activity, and between placenta lead and erythrocyte ALAD activity.

In the fetuses, blood lead level is correlated with liver and placenta lead, or with liver ALAD activity. Liver lead level is correlated with placenta lead, or with erythrocyte and liver ALAD activity. Furthermore, ALAD activity in the fetal erythrocytes correlated with placenta lead and liver ALAD activity.

Statistically significant positive correlations were found between dams and fetuses in blood lead level (Fig. 1), liver lead level (Fig. 2), and erythrocyte ALAD activity (Fig. 3). There was a significant negative correlation between the maternal and fetal liver ALAD activity (Fig. 4).

**DISCUSSION**

No significant difference in the blood lead level was seen between the lead-treated and control groups except for the fetuses on day 21 of gestation. However, erythrocyte ALAD activity of the lead-treated dams and fetuses significantly decreased on days 18 and 21 of gestation. On the other hand, liver lead level was significantly higher in the lead-treated than in the control groups. These findings indicate that erythrocyte ALAD activity was suppressed when lead
Fig. 1. Correlation between lead concentrations in maternal and fetal blood.

Fig. 2. Correlation between lead concentrations in maternal and fetal livers.
entered the blood stream, but, since the lead rapidly moved to the liver and other target organs, the blood lead level did not increase.

The lead level of the blood in the lead-treated fetuses was significantly higher on day 18 than on day 21 of gestation. On the other hand, the lead level of the placenta in the lead-treated groups was significantly increased. These observations suggest that lead is easily transferred from the dams to the fetuses during the early phase of gestation and is blocked by the placenta during later gestation stages. The same idea might be applied to the control fetuses, i.e., it was considered that lead stored in the dams before mating may rapidly shift to embryos when the placental formation is not complete.

The difference in the elevated liver ALAD in the fetus compared with that in the adult may be explained as follows: (1) ALAD and ALA synthetase activities are greatest in the liver; (2) the activity of ALA synthetase is 10 times higher in the fetal rat liver than in adults and (3) hemin injection depresses liver ALA synthetase activity in the adult, but not in the fetus.

The author has reported that the hepatic ALAD activity was reduced when the liver lead levels in the dams and their litters were 17 and 27 times higher than...
normal, respectively). Therefore, the liver ALAD activity is not hindered when the subject is exposed to only a small amount of lead.

The data in this study confirm the sensitivity of ALAD to intoxication by lead. The reduction of ALAD activity found in the fetuses is, however, much smaller than that seen in the erythrocytes of the dams. Kuhnert et al. pointed out that greater cell volume of the fetal erythrocytes allows a correspondingly greater amount of the cytoplasmic enzyme ALAD. As a result, the fetal erythrocyte ALAD activity may be suppressed to a lesser extent than that of the dams. In addition to a reduction of erythrocyte ALAD activity, however, a decrease of the RBC count, Hb concentration and a retardation in body growth were observed in the lead-treated fetuses. Jaquet et al. and Gerber and Maes reported that the impaired synthesis of heme in lead-intoxicated mouse embryos appears to be sufficient for the reduction of their body mass during the late phase of gestation.

From the facts described above, lead exposure during pregnancy should be assumed to influence the fetus, even though it may be such a low level that clinical signs of poisoning are not observed.

Fig. 4. Correlation between ALAD activity in maternal and fetal livers.
REFERENCES


