A Reference Value for Delta-Aminolevulinic Acid in Plasma in the Population Occupationally Unexposed to Lead

Abstracts: To obtain a reference value for delta-aminolevulinic acid in plasma (ALA-P), we determined ALA-P levels in 141 subjects (105 males and 36 females) without occupational exposure to lead. The distribution of ALA-P levels in males can be regarded to follow log-normal distribution and the geometric mean was 8.7 $\mu\text{g/l}$ with 95% confidence interval of 6.0-12.5 $\mu\text{g/l}$. These data would be useful as a baseline for evaluating the relationship between ALA and its biological effects.

Key words: Delta-aminolevulinic acid — Plasma — Reference value

Delta-aminolevulinic acid (ALA) is an intermediate in the heme-biosynthetic pathway and accumulates in tissues by lead exposure and in acute intermittent porphyria. ALA has been well known as a neurotoxic substance^{1, 2)} and its carcinogenicity has been recently reported in the relation to ALA-generated reactive oxygen species^{3, 4)}. The oxygen species hydroxylate deoxyguanosine residues in DNA and produce hydroxy-2'-deoxyguanosine, which induces G-T transversion. Thus, the knowledge of the ALA concentration in the tissue such as plasma would be very important not only in industrial health but also in the mechanism of chemical carcinogenesis. For several years, the sensitive methods of determining ALA in plasma or blood by fluorometric method using high-performance liquid chromatography (HPLC) have been developed⁵⁻⁸⁾; however, there is few study which collected the sufficient number of normal subjects and determined not only ALA in plasma (ALA-P) level but also blood lead concentration (Pb-B). In this study, we determined the concentration of ALA-P and Pb-B in large numbers of occupationally unexposed population and reported the reference value for ALA-P.

Venous blood treated with EDTA-2K was obtained from 152 subjects who did not deal with lead in their work. The workers who have had a special medical examination for lead poisoning were excluded from this study. Blood samples were collected in the physical examinations. Two workers were found to take medicine by a medical checkup. Four workers showed abnormal levels in hematocrit. Five workers whose blood were hemolyzed during sampling were also excluded. Therefore, those 11 subjects were excluded from this study. A total number of analysis was 141 for 105 males and 36 females, consisting of 42 workers of electrical appliance manufacture, 63 drivers, 33 bus conducteresses and 3 clerks.

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Plasma was separated from whole blood immediately after sampling, and stored at -80°C until analysis. ALA-P was determined as described previously⁸⁾. Pb-B was determined by flameless atomic absorption spectrometry.

Figure 1 shows the histograms of ALA-P levels in 105 male and 36 female subjects. The distribution pattern of male ALA-P levels was tested by skewness (b1) and kurtosis (b2). The original ALA-P data did not fit for normal distribution (b1 = 1.06, b2 = 5.44; $p < 0.01$, respectively). When the ALA-P values were converted to logarithm, the distribution of ALA-P in males can be regarded to

Fig. 1. Distribution of delta-aminolevulinic acid in plasma (ALA-P) in 105 males (a) and in 36 females (b).

follow log-normal distribution ($b_1 = 0.30$, $b_2 = 3.60$; $p > 0.05$, respectively). The geometric mean of ALA-P was $8.7 \mu\text{g/l}$ and the 95% confidence interval was 6.0–12.5 $\mu\text{g/l}$. The distribution of ALA in urine (ALA-U) in healthy urban males has been reported to follow log-normal distribution⁹. For the ALA-P levels in females, comments could not be given in detail, because the sample number was only 36 and their ages were distributed to younger population in this study.

Table 1 shows the ages and concentrations of ALA-P and Pb-B in the subjects in this study. Mean Pb-B levels of both genders were less than 5 $\mu\text{g/dl}$, which was coincident with the value reported by Watanabe *et al.*¹⁰ The mean $\pm$ SD of ALA-P levels in males were reported to be $8.2 \pm 4.7 \mu\text{g/l}$ ($n = 32$)⁷ $8.6 \pm 1.3 \mu\text{g/l}$ ($n = 26$)⁸. Although the Pb-B levels were not determined in these two studies, both the ALA-P levels^{7,8} were nearly equal to the value in the present study. There was no study which determined ALA-P levels in females occupationally unexposed to lead. In the present study, both ALA-P and Pb-B levels in females were statistically lower than those in males ($p < 0.01$, by Welch's test). Taguchi *et al.*¹¹ reported that ALA-U levels in females were lower than those in males in subjects occupationally unexposed to lead but when the urine was corrected for creatinine, ALA-U levels in females were higher than those in males. In this study, no significant correlation was found between ALA-P and Pb-B or age in each gender.

We previously determined both levels of ALA-P and Pb-B in 191 male lead workers and reported that the no effect level of Pb-B for increasing ALA-P might be around 5 $\mu\text{g/dl}$ ¹². This previous data are basically consistent with the present results because the number of subjects whose Pb-B levels were more than 5 $\mu\text{g/dl}$ was only 18 and the highest level of Pb-B was as low as 6.9 $\mu\text{g/dl}$ in this study. It is suggested that ALA-P levels in male adults are not associated with Pb-B levels in the normal population whose mean Pb-B levels were less than 5 $\mu\text{g/dl}$ as seen in Japan.

This is the first report of a reference value for ALA-P levels in large numbers of male adults whose mean Pb-B levels were less than 5 $\mu\text{g/dl}$. This result would be useful as a baseline for evaluating the relationship between ALA and its biological effects. This study have to be followed by extended surveys for female.

Table 1. Ages and concentrations of blood lead (Pb-B) and delta-aminolevulinic acid in plasma (ALA-P) in males and females.

		Males (n = 105)	Females (n = 36)
Age	AM	40.6 $\pm$ 8.7 (20–60)	22.5 $\pm$ 5.3 (18–40)
ALA-P ($\mu\text{g/l}$)	AM	8.8 $\pm$ 1.7 (5.7–15.6)	7.6 $\pm$ 1.3 (5.2–10.8)
	GM	8.7 $\pm$ 1.2	7.5 $\pm$ 1.2
Pb-B ($\mu\text{g/dl}$)	AM	3.9 $\pm$ 1.2 (1.8–6.9)	2.4 $\pm$ 0.7 (1.1–4.3)
	GM	3.7 $\pm$ 1.4	2.3 $\pm$ 1.3

AM: Arithmetic mean $\pm$ SD (Range).

GM: Geometric mean $\pm$ SD.

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