# Comparison of capillary earlobe and venous blood monitoring for occupational lead surveillance

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Biological monitoring for occupational lead exposure involves routine venous blood draws from exposed employees. This uncomfortable procedure normally yields more blood than what is needed for analysis. Capillary blood sampling is less invasive but introduces the possibility of surface contamination. The objective of this study was to compare venous and capillary (earlobe) blood lead samples obtained from occupationally exposed individuals. Phlebotomists trained specifically in the collection of blood samples for lead determination collected 2 venous blood samples and 2 capillary earlobe samples from each participating employee. Before the capillary draw, the employee's earlobe was cleansed with an alcohol wipe in an effort to remove potential lead contamination. A second alcohol wipe was then used to sanitize the lancing area and was retained for lead analysis. Both the venous and capillary samples were subsequently analyzed with the use of graphite furnace atomic absorption spectrometry (GFAAS). GFAAS of venous blood specimens was considered the reference method of sampling and analysis. We collected and analyzed 126 paired earlobe and venous samples. Earlobe sampling was preferred to venous sampling by 54% of the employees surveyed. The mean difference between the capillary and venous results was 38.8  $\pm$  48.1  $\mu$ g/dL. Lead concentrations in earlobe blood were more than twice those found in venous samples in more than half of the samples (64 of 126). Despite simple cleansing with an alcohol wipe and no visible skin contamination, 94% of the wipe samples from earlobes contained more than 1  $\mu$ g of lead. Even low concentrations of contamination can significantly alter the concentration of lead in the blood; for example, sample contamination of 0.3  $\mu g$  lead in a 200- $\mu L$  blood sample would yield an increase of 150 µg/dL in the measured lead concentration. The findings of this study suggest that until satisfactory skin cleansing and decontamination techniques are identified and evaluated, earlobe sampling should be avoided in the surveillance of occupational blood lead levels. (J Lab Clin Med 2004;143:217-24)

**Abbreviations:** ARC = American Red Cross; CDC = Centers for Disease Control and Prevention; CLIA = Clinical Laboratory Improvements Amendment; EDTA = ethylenediaminetetraacetic acid; GFAAS = graphite furnace atomic absorption spectrometry; NCEH = National Center for Environmental Health; NIOSH = National Institute for Occupational Safety and Health; OSHA = Occupational Safety and Health Administration; PEL = permissible exposure limit; TWA = time-weighted average

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In the United States, OSHA regulates employers with workers exposed to lead in general industry and in the construction industry. The OSHA PEL for airborne lead exposure is a TWA of 50  $\mu$ g/m³ during a typical workday.¹ Employers are required to have workers participate in a medical surveillance program if they are exposed to airborne lead environments of 30  $\mu$ g/m³ as a TWA for at least 30 days a year. Because air samples are not a surrogate for biologic monitoring, lead concentrations in employees' blood are tested regularly. The medical surveillance program consists of an initial medical examination and biologic monitoring at least every 6 months. Given the high potential for surface contamination with capillary samples, venous samples are typically collected.

Because capillary sampling is less invasive, individuals are typically more favorably inclined toward it than they are toward venous sampling. Because both the earlobe and the finger tend to bleed quite freely, the American Red Cross has used both capillary earlobe and finger-stick sampling to screen individuals for blood donation purposes.<sup>2,3</sup>

Traditional finger-stick blood sampling for the monitoring of lead concentrations in worker populations is not used because of problems with contamination even after the use of cleansing procedures. Uncontaminated capillary samples from finger-stick specimens are difficult to obtain from workers whose hands are callused or contaminated with lead. For example, in a recent NIOSH Health Hazard Evaluation, lead contamination averaging 530  $\mu$ g was evident on employees' hands even after the workers had thoroughly washed them. Lead contamination may persist on the human hand because the intricate pattern of crevices and grooves on the skin retains lead particles. Additionally, lead is excreted by way of sweat onto the skin. 5.6

Earlobe sampling has been used previously for occupational monitoring of lead and cholinesterase. <sup>7-10</sup> In the earliest use of earlobe blood sampling to monitor lead concentrations, the blood sample was immediately transferred to filter paper, which was subsequently analyzed with the use of disc atomic absorption spectroscopy. <sup>7</sup> In previous studies the workers preferred earlobe sampling. <sup>7,10</sup>

Hemoglobin is the oxygen-carrying pigment of the erythrocyte. Once lead is in the bloodstream, it binds to both erythrocytes and free proteins in the blood. 11,12 Because a difference in hemoglobin concentration may be associated with a difference in lead concentration per unit volume in the blood, comparison of hemoglobin concentrations between venous and capillary earlobe blood may be important. Early research has suggested that the hemoglobin concentrations in venous blood correlate well with hemoglobin concentrations

from capillary earlobe blood. 13,14 Because the ARC uses hematocrit and hemoglobin measurements as a screening tool for blood donors, most research comparing venous and capillary hemoglobin concentrations has been completed in the blood donation arena. More recently, researchers have indicated that differences may exist between the hemoglobin concentrations in capillary earlobe blood and those in capillary samples from the fingertip or arm. 2,3,15-18 Overall, the percentage of hemoglobin in capillary samples from the earlobe has been reported to be 7% to 10% higher than those in finger-stick and venous samples.

In this study we sought to compare lead concentrations in venous and capillary earlobe blood samples obtained from occupationally exposed individuals. Both types of samples were analyzed with the use of GFAAS in a CLIA- and OSHA-certified CDC reference laboratory. (GFAAS is also referred to as electrothermal atomic-absorption spectrometry by the International Union of Pure and Applied Chemistry.) To investigate whether our results were influenced by different hemoglobin concentrations in capillary and venous blood, we also conducted hemoglobin measurements.

#### **METHODS**

**Recruitment.** Representatives of industries historically associated with lead exposure (eg, battery manufacturing, abrasive blasting in construction, lead smelting) were contacted to request their facilities' participation in this study. Facilities whose management expressed interest were initially screened on the basis of the diversity of exposures and the number of employees, and 2 sites were selected on the basis of information obtained during walkthrough surveys. The study design and protocol were reviewed and approved by the CDC Institutional Review Board.

All employees enrolled in each facility's medical surveillance program were eligible to participate in the CDC study. The study was described to small groups of employees (n < 30) in meetings, during which informed consent was obtained from each volunteer. Each participant was then assigned a 20-minute appointment during which blood samples were collected and a brief interview was conducted to obtain demographic and exposure information. After the blood draw was complete, the participant was asked which sampling method (ie, venous or capillary) he or she preferred.

**Lead in air and on surfaces.** At each site, airborne particulate samples were collected on 37-mm filter cassettes containing mixed cellulose ester filters (SKC, Inc, Eighty Four, Pa) for 8 hours inside the room where the blood samples were drawn. These samples were collected and analyzed for lead in accordance with NIOSH method 7082<sup>19</sup> (GFAAS model 4100, Perkin-Elmer, Norwalk, Ct). The limit of detection for this method was 2.6 µg lead/filter sample.

The surfaces of tabletops where blood samples were obtained from workers were thoroughly cleansed with Wash N

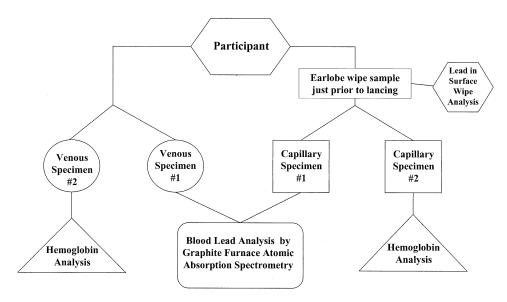


Fig 1. Blood collection and analysis.

Dri wipes (Softsoap Enterprises, Chaska, Minn). At the start of each sampling period, we tested tabletops for lead contamination using the surface-wipe spot-test method of Esswein et al. <sup>20</sup> This spot test involves wipe sampling followed by treatment of the wipe with vinegar and then an aqueous rhodizonate solution (prepared from 99% + sodium rhodizonate; Aldrich Corporation, Milwaukee, Wis). The reaction of rhodizonate with lead (collected on the wipe) to yield a characteristic color change from yellow to red is indicative of the presence of lead. If spot test wipe results were positive for lead, we performed additional surface cleaning until subsequent spot test results from surface wipe samples were negative. Between workers, tabletops were covered with clean, fresh absorbent pads.

**Blood draws.** Blood samples were collected in an occupational health clinic or in a training room inside the facility. All supplies used for blood sampling, including needles, evacuated tubes, and alcohol wipes were prescreened for lead contamination. Using universal precautions, 22,23 phlebotomists trained specifically in the collection of blood samples for the determination of lead concentrations sought to collect 2 venous blood samples and 2 capillary earlobe samples from each participant. The collection and analysis protocol is depicted in Figure 1.

Before the venous blood draw, the puncture site, in the antecubital area of the employee's arm, was cleansed with a wetted wipe (Wash N Dri) to remove any lead contamination. A second wipe was then used to sanitize the antecubital area. Each participating employee donated 2 venous samples of 2 mL each (Venipuncture Needle-Pro, SIMS Portex, Inc, Keene, NH). Each venous blood sample was drawn into a Vacutainer tube (K<sub>3</sub> EDTA, BD 36-9651; Becton Dickinson, Rutherford, NJ) labeled with a bar code for identification. In an effort to prevent clotting, we immediately placed all sam-

ples in a rocker (model U-51401; Cole-Parmer, Vernon Hills, Ill) to quickly induce mixing of the anticoagulant with the sampled blood.

Any employee wearing a pierced earring was asked to remove it from his or her earlobe before the capillary blood draw. The phlebotomist gently massaged the employee's earlobe for approximately 10 seconds to improve blood circulation. The earlobe was then cleansed with an alcohol wipe to remove any lead contamination. A second wipe was used to sanitize the skin on the earlobe. The second alcohol wipe was deposited in a 30-mL high-density-polyethylene screw cap container (Nalgene Corporation, Rochester, NY). The back of the earlobe was held in a triangular shape and a spring-loaded lancet (Mictrotainer, 2.2-mm blade depth; Becton Dickinson, Franklin Lakes, NJ) was used to lance the earlobe. The first drop of blood was not collected. One sample of capillary blood was drawn through a capillary tube, with the use of a gentle nonsqueezing motion, into a 200-µL EDTA-treated microvial (Becton Dickinson; Fig 2). If the earlobe continued to bleed, a second sample was collected in another microvial. Each capillary-blood sample was marked with a bar-code identification label. The microvials were manually shaken immediately after sampling in an effort to ensure that the anticoagulant was mixed thoroughly with the blood. Capillary samples were then placed in the rocker to minimize clotting. The specific dates and times of blood draws were documented in a master log.

GFAAS blood lead analysis. One venous sample (Vacutainer tube) and 1 earlobe sample (EDTA-treated vial) from each sampling pair were refrigerated and sent by way of overnight courier to the CDC reference laboratory. These samples were analyzed for blood lead in accordance with CDC whole blood method 1080C by means of GFAAS (model 5100-ZL with Zeeman background correction; Per-



Fig 2. Capillary earlobe sampling.

kin-Elmer), which was based on the method described by Miller et al.<sup>24</sup> We regarded GFAAS analysis of venous samples as the reference method for sampling and analysis.

Hemoglobin analysis. To determine any differences between venous and earlobe hemoglobin, we analyzed all blood samples on site for hemoglobin, using a HemoCue photometer (HemoCue AB, Angelholm, Sweden). To conduct hemoglobin analysis, we pipetted a small bead of blood from the second blood specimen (venous or capillary) onto clean paraffin paper. From each sample, we drew a small quantity of blood into a disposable microcuvette (HemoCue) by means of capillary action. The microcuvette was then inserted into the HemoCue instrument and the hemoglobin concentration (grams per deciliter) measured (by means of colorimetric detection of hemoglobinazide adduct).

As part of the quality assurance process, we used a control cuvette with an optical interference filter to verify the instrument's basic electronic/optical status. Liquid quality control materials (HemoCue) at 3 concentration levels were used to ensure that the performance of both the instrument and the cuvette were within acceptable limits.

**Lead in earlobe surface wipes.** After sample collection, we sent the 30-mL containers containing the alcohol wipe samples obtained from earlobes to a commercial laboratory for analysis. The laboratory was accredited by the American Industrial Hygiene Association for lead analysis under the National Lead Laboratory Accreditation Program. The wipes were analyzed for lead with the use of GFAAS in accordance with NIOSH method 9100 (model 5100ZL; Perkin-Elmer). The limit of detection for this method was 0.1 μg lead/wipe sample. A minimum frequency of 5% of field blanks was submitted for analysis along with the earlobe wipe samples.

**Statistical analysis.** We summarized the demographic characteristics of the study population using frequency distributions. Descriptive statistics were determined for the lead content of venous and capillary earlobe samples and earlobe wipe samples, as well as venous and capillary hemoglobin concentrations. Normally, blood lead results from laboratory

analysis are reported to patients in integer figures. To avoid biasing the statistical analysis by rounding, we left blood lead results at 1 decimal place during statistical analysis. We conducted nonparametric (signed-rank) tests to compare venous and capillary blood lead results, as well as venous and capillary hemoglobin results. Linear regression analysis was conducted in an attempt to evaluate the difference between venous and capillary hemoglobin concentrations.

#### **RESULTS**

**Study demographics.** Site 1 was a lead battery manufacturing facility; site 2 was a lead smelting facility. Both study sites had medical surveillance programs and diverse workplace exposures. Because the participating employers could use the venous analysis results to satisfy the OSHA medical surveillance requirements at no cost, employer cooperation was high during facility recruitment.

We recruited a total of 206 eligible study participants from the 2 sites, 66 from the battery manufacturing plant and 140 from the lead smelter. The study population had a mean age of  $37.3 \pm 10.7$  years and was mostly male (83.2%) and white (96.2%); 40.4% were smokers. The site 1 population was older (40.4 vs 35.9 years), included fewer men (54.6% vs 96.5%), had fewer smokers (36.4% vs 42.3%), and reported less use of respirators (74.2% vs 98.6%).

Venous and capillary lead analysis. Although 206 venous samples were collected from study participants and then analyzed, just 182 capillary earlobe samples were collected. Twenty participants' earlobes did not yield a capillary sample of 200  $\mu$ L, and 6 participants' schedules did not allow sufficient time for capillary samples. Of the 182 capillary samples donated, 33 were not analyzed because the sample volume was insufficient for the analytical method. Another 23 capillary samples were not successfully analyzed because the initial analysis result was above the calibration curve and additional sample volume was insufficient for dilution and reanalysis. Therefore only 126 pairs of venous and capillary earlobe samples were available for statistical analysis.

Capillary blood lead concentrations are plotted against venous results in Fig 3. The results of lead measurements—including venous and capillary earlobe concentrations of lead detected on GFAAS, alcoholwipe samples, and venous and capillary earlobe hemoglobin concentrations—are summarized in Table I.

The mean venous lead concentration was  $25.2 \pm 8.2 \, \mu \mathrm{g/dL}$ , whereas the mean capillary earlobe concentration was  $64.0 \pm 50.7 \, \mu \mathrm{g/dL}$ . The mean difference between the capillary and venous results was  $38.8 \pm 48.1 \, \mu \mathrm{g/dL}$ . The difference between capillary earlobe lead on GFAAS minus the venous GFAAS result plot-

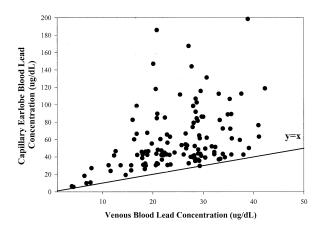


Fig 3. Lead concentrations in capillary and venous blood.

ted against the venous GFAAS result is presented in Fig 4. In more than 50% of the blood-lead results, capillary earlobe lead concentrations were at least twice the venous lead results. The signed-rank test revealed a significant difference between venous and capillary results (P = .0001).

Hemoglobin differences. We analyzed both venous and capillary blood samples for hemoglobin (n = 113). The mean venous and capillary hemoglobin concentrations (and ranges) were 15.4 g/dL (12.3–22.0 g/dL) and 15.7 (12.4-19.8) g/dL. The mean difference between capillary and venous results was -0.26 g/dL. The mean levels were within the normal hemoglobin range of 13 to 18 g/dL for American males at sea level.<sup>26</sup>

The differences between venous and capillary hemoglobin concentrations are plotted against venous hemoglobin results in Fig 5. Although the differences between the paired venous and capillary hemoglobin concentrations were statistically significant on the signed-rank test (P < .0001), these small differences were not clinically significant. The difference between venous and capillary hemoglobin measurements was within 2 g/dL for 90% of the samples (102 of 113).

Regression analysis detected no relationship between the difference in venous and capillary hemoglobin concentrations and the difference between venous and capillary lead concentrations (P < .0001). The variance of the difference between the venous and capillary hemoglobin measurements was 2.2 (g/dL)<sup>2</sup>, significantly higher than the variance (0.013–0.80 [g/dL]<sup>2</sup>) reported for the HemoCue hemoglobin measurement instrument. 27,28

Instrument variation, although a minor contributor, probably does not completely explain the differences between venous and capillary hemoglobin concentrations. These small observed differences in measurements of hemoglobin concentrations therefore do not account for the large differences observed between the measured venous and capillary lead concentrations.

Earlobe wipe samples. We used alcohol wipes on the participants' earlobes as a means of sanitizing them and also to obtain dermal wipe samples to investigate the presence of lead contamination on the skin before earlobe blood sampling. Despite the use of the cleansing procedure and observation of a visibly "clean" earlobe, 94% of the earlobe-wipe samples (n = 105) contained lead in amounts greater than 1  $\mu$ g/wipe sample. The amount of lead in the wipes varied from the detection limit of 1.0  $\mu$ g to 300  $\mu$ g/sample. The mean lead concentration of the alcohol wipe was  $38.6 \pm 54.3$  $\mu$ g/wipe sample. The difference between the capillary and venous lead concentrations was correlated with the earlobe wipe (Pearson's correlation coefficient .49, P < .0001). Results from field blanks (n = 6) were all below the limit of detection.

Lead in air samples. All airborne lead samples were analyzed and reported to be below the limit of detection, 2.6 µg/filter sample. Blood sample contamination resulting from airborne lead is therefore likely not a source of increased lead in capillary samples.

Employee impression of capillary earlode sampling. After blood draws were complete, each employee was asked whether he or she preferred one of the techniques —earlobe or venous sampling—or whether he or she had no preference. As shown in Table II, more than 50% percent of the employees preferred earlobe sampling. Some workers mentioned that their preference in blood sampling site would be directly related to the expertise of the phebotomist.

### DISCUSSION

Sample collection. Capillary earlobe blood sampling was more time consuming than we originally anticipated. Although some individuals' earlobes bled easily, other individuals' blood clotted rapidly while the sample was being drawn. If a sample was difficult to obtain, the sample collection took several minutes to complete. In 20 cases, no earlobe blood sample could be obtained from the participant. The tendency for the earlobe to bleed may be a limiting factor in capillary earlobe blood lead monitoring in occupational biological monitoring. Only 69% of the earlobe blood samples (126 of 182) yielded a sample volume sufficient for successful blood lead analysis.

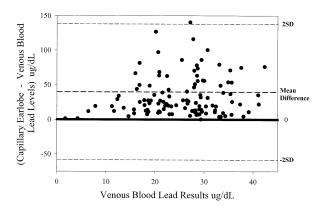
Hemoglobin analysis. A difference in hemoglobin concentrations between venous and capillary blood might be associated with a difference in blood lead concentration per unit volume. Other studies involving various techniques of hemoglobin determination have revealed that hemoglobin concentrations vary as much

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Table I. Descriptive statistics from lead and hemoglobin analysis

Sample Type	Units	N	Mean ± SD	Range
Venous blood lead (overall)	μg/dL	206	23.8 ± 8.62	1.7–42.3
Venous blood lead (paired)*	μg/dL	126	$25.2 \pm 8.15$	3.9-42.3
Capillary earlobe blood lead	μg/dL	126	$64.0 \pm 50.7$	5.6-361.7
Earlobe-wipe samples	μg	123	$38.6 \pm 54.3$	<1.0-300
Venous hemoglobin	g/dL	113	$15.4 \pm 1.52$	12.3-22.0
Capillary hemoglobin	g/dL	113	$15.7 \pm 1.60$	12.4-19.8

<sup>\*</sup>Paired samples are subsets of venous samples for which there are corresponding capillary samples from the same individuals. Not all subjects donated capillary samples that could be successfully analyzed.



 ${\bf Fig}~{\bf 4.}$  Difference plot of lead concentrations in capillary and venous blood.

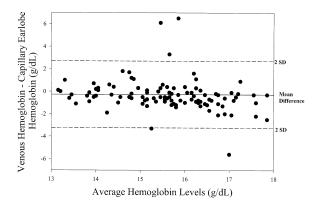


Fig 5. Difference plot of hemoglobin concentrations in capillary and venous blood.

as 10% from different sampling sites.  $^{2,15-18}$  However, one study has shown that the HemoCue performs acceptably on ear-stick samples compared with other hemoglobin measurement techniques.  $^{29}$  The results of this study indicate that the percent mean difference between hemoglobin concentrations in venous and capillary earlobe samples was just -2.1% (0.3 g/dL). The large disparity between blood lead results cannot be

**Table II.** Blood-sampling-technique preferences

Sampling technique	% preferred by participating workers
Capillary	54
Venous	21
No preference	25

solely attributed to hemoglobin differences between the 2 sampling sites.

Skin cleansing techniques. Although we cleansed the skin with an alcohol wipe and evaluated the area visually for the absence of dermal contamination, the earlobe wipe results (Table I) indicate that the earlobes remained contaminated with lead. Even low concentrations of surface contamination can significantly alter the blood lead determination. For example, a sample contamination of 0.3  $\mu$ g of lead in a 200- $\mu$ L blood sample would cause a 150  $\mu$ g/dL increase in the assessment of blood lead concentration. Surface contamination appears to explain the poor correlation between lead concentrations in capillary and venous blood in this study. Without a more vigorous cleansing technique, capillary earlobe sampling will not yield accurate results.

Some cleansing methods have been evaluated during finger-stick and venous blood sample comparisons in pediatric populations.<sup>30,31</sup> One study illustrated that washing of the hands with soap and water before the fingers were cleaned with alcohol wipes, just before lancing, was a marginally better technique for removing skin contamination.<sup>30</sup> Battery workers who washed with soap and water still left 30% of the lead contamination on the skin surface.<sup>32</sup>

More vigorous cleansing techniques for the earlobe should be evaluated before capillary earlobe sampling can be seriously considered in the occupational setting. Removal techniques for other analytes, such as pesticides on the skin, have been studied and may provide a foundation on which to base the evaluation of dermal decontamination of other substances such as lead. 33,34

**Portable blood lead analyzers.** In conjunction with this study, we have published a companion evaluation of a portable blood lead analyzer. <sup>35</sup> Although this analyzer requires just 50  $\mu$ L of sample, which can be provided through capillary earlobe sampling, problems with sample collection remain. Earlobe sample collection is not recommended for use with the portable blood lead analyzer among worker populations at this time.

Concluding remarks. In this study we evaluated the potential of capillary earlobe blood sampling as a monitoring technique among lead exposed workers. To our knowledge, it is the largest effort to date to evaluate the efficacy of capillary earlobe sampling for blood lead screening. Capillary earlobe sampling among lead-exposed workers falls victim to the same obstacle as finger-stick sampling: surface lead contamination. Until a satisfactory dermal decontamination technique is identified and evaluated, capillary earlobe sampling should be avoided as a method of occupational blood lead surveillance.

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