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Evaluation of a Portable Blood Lead Analyzer With Occupationally Exposed Populations

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Background *This project evaluated a portable electroanalytical instrument that is used to rapidly analyze blood lead levels in individuals, using a fresh whole blood sample (venous).*

Methods *Samples were obtained from 208 lead-exposed employees who donated two 2 ml venous blood samples into "lead-free" evacuated tubes. One blood sample was analyzed onsite using the portable field instrument while the second sample was analyzed using graphite furnace atomic absorption spectrometry (GFAAS).*

Results *According to GFAAS results, employee venous blood lead levels ranged from 1 μg/dl to 42 μg/dl. The mean difference between the results from the field instrument and GFAAS was less than 1 μg/dl. Analysis indicates that the results from the field instrument yielded a slight positive bias overall (P value = 0.0213), with less bias for blood lead levels above 10 μg/dl (P value = 0.0738).*

Conclusions *Within the blood range evaluated (1–42 μg/dl), the instrument performed adequately according to Clinical Laboratory Improvements Amendments (CLIA) proficiency requirements. The ability of the instrument to perform rapid analysis makes it potentially valuable to occupational health professionals for medical monitoring or on-site investigations.* Am. J. Ind. Med. 40:354–362, 2001. *Published 2001 Wiley-Liss, Inc.*†

KEY WORDS: electroanalytical instrument; blood lead analysis; blood lead monitoring; instrument evaluation; anodic stripping voltammetry

INTRODUCTION

The health effects of lead exposure are well documented. Lead can be inhaled into the lungs, absorbed through the skin, or ingested from contaminated hands, food, or cigarettes. Lead entering the body via inhalation is absorbed based upon particle size and solubility. In adults, approximately 10% of the ingested lead is absorbed into the gastrointestinal tract [Anderson et al., 1995]. In the bloodstream, the majority of the lead (>95%) is bound to erythrocytes. Long-term deposition of lead results in the distribution of this metal to the brain, kidney, and skeletal system [LaDou, 1990]. Lead is a known animal carcinogen, and the International Agency for Research on Cancer

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classifies it as a possible human carcinogen [International Agency for Research in Cancer (IARC), 1987; Agency for Toxic Substances and Disease Registry, 1997].

Traditionally, fingerstick capillary samples from occupationally exposed employees are difficult to obtain from hands that are often callous and contaminated with lead. Unfortunately, even slight lead contamination can seriously alter the analysis results of a blood lead sample. In a recent National Institute for Occupational Safety and Health (NIOSH) Health Hazard Evaluation, employee skin contamination averaged 530 μg lead/wipe per two hands on a wipe even after employees had thoroughly washed their hands [Esswein et al., 1996]. Additionally, lead is excreted in sweat onto the skin, thereby causing additional skin contamination [Omokhodion et al., 1991]. Given this high potential for surface contamination, venous samples are considered the primary method for blood lead testing in occupationally exposed individuals.

Data from the National Health and Nutrition Examination Survey predicted approximately 700,000 adults with blood lead levels greater than 25 μg/dl in the US [Brody et al., 1994]. In 1998, the NIOSH Adult Blood Lead Epidemiology and Surveillance (ABLES) program documented that approximately 18% of reported blood lead levels were greater than 25 μg/dl [Centers for Disease Control and Prevention, 1999]. Industries with the highest lead exposures include battery manufacturing, shipbuilding and repair, lead smelting, and abrasive blasting in the construction industry [National Institute for Occupational Safety and Health, 1997; Centers for Disease Control and Prevention, 1999].

The Occupational Safety and Health Administration (OSHA) regulates employers with lead-exposed workers. These regulations are in place not only to protect the individual employee, but also to reduce the potential contamination of employees' homes. The OSHA permissible exposure limit (PEL) for airborne lead exposure is 50 $\mu g/m^3$ during a typical workday [Code of Federal Regulations, 1978]. Employers are required to have workers participate in a medical surveillance program if workers are exposed to airborne lead environments of 30 $\mu g/m^3$ for at least 30 days in a year. Since air samples are not a surrogate for biological monitoring, employees must have their blood lead levels tested regularly. The medical surveillance program consists of an initial medical examination and biological monitoring at least every 6 months.

If a worker's blood lead level is greater than or equal to 40 μg/dl, the worker must have his/her blood lead monitored every 2 months and a medical examination on an annual basis. If the worker's average blood lead level throughout three subsequent blood lead tests is above 50 μg/dl, and his/her last test blood lead level was not below 40 μg/dl, the worker must be removed from work involving lead exposure. An employee may return to lead work after two successive test results at a blood level at or below 40 μg/dl. The frequency of blood lead monitoring suggests the need for a convenient, low-cost analytical method, particularly to help employer compliance with OSHA regulations.

Currently, OSHA regulations do not mandate a specific analytical method for blood lead analysis. Rather, they require that a laboratory be "OSHA approved." Since all laboratories conducting clinical diagnostic tests must obtain Clinical Laboratory Improvement Amendments (CLIA) certification, "OSHA approved" laboratories must have dual approval and certification. Successful participation in an ongoing proficiency-testing program is necessary to remain CLIA certified and OSHA approved in blood lead analytical techniques. The CLIA of 1988 was created to establish the regulations that all laboratories must meet to be certified to perform testing on human specimens [Federal Register, 1997]. CLIA mandates that a blood lead analysis laboratory must report proficiency testing results within ±4 μg/dl or within 10%, whichever is greater, of the target (true) value. If these criteria are met, then the analysis method is considered acceptable under the terms of CLIA.

LeadCare® Instrument

In 1991, the Centers for Disease Control and Prevention (CDC) lowered the blood lead level of concern for children to 10 μg/dl. Since this recommendation, CDC has been supporting research for a quick, cost-effective technique to monitor pediatric blood lead levels. Early instruments (based on Anodic Stripping Voltammetry [ASV]) achieved success, but some needed constant adjustment due to a copper interference [Roda et al., 1988]. These instruments are more suited for use in a laboratory and have become a popular means of determining lead in blood [Ashley, 1994]. In recent years, ESA, Inc. (Chelmsford, MA) and AndCare, Inc. (Durham, NC) developed a small, rapid, hand-held portable LeadCare® instrument for on-site monitoring applications.

A field portable, blood lead monitoring instrument could provide a valuable service to occupational professionals. Instead of waiting days or weeks to obtain results, an employee's blood lead level would be known quickly (in minutes or hours). As the investment cost of this instrument is rather low, the portable blood lead analyzer could be a cost effective alternative to the traditional fixed-site laboratory technology. Although the LeadCare® instrument had received FDA market clearance, the supporting sampling and analysis was conducted in hospital environments. Given the potential challenges of an occupational environment, it was deemed necessary to evaluate the instrument in a workplace setting.

METHODS

Recruitment

Industries historically associated with lead exposures, such as battery manufacturing, abrasive blasting in construction, and lead smelting were contacted to participate in this study. The objective of the recruiting process was not to select typical exposures for each industry, but rather to include workers who were expected to have blood lead levels throughout a wide range of concentrations. Interested facilities were initially screened based upon the diversity of exposures and the number of employees and were finally selected based upon information obtained during walk-through surveys. The study design and protocol was reviewed and approved by the CDC Institutional Review Board.

All employees enrolled in the facility's medical surveillance program were eligible to participate in the CDC study. Prior to the sampling program, we posted flyers at the facilities describing the study to employees. Upon our arrival, we briefly described the study in meetings to small groups ($n < 30$) of volunteering employees. During these meetings, we also obtained the informed consent and enrolled participants in the study. Each participant was then assigned a 20-min appointment where blood samples were collected and a brief interview was conducted to obtain demographic information, including age, race, sex, smoking history as well as work history and practices. We devised a target population of at least 200 participants based upon an α of 0.05, a β of 0.90, and the detection of $\pm 12.5\%$ difference between the two analytical methods.

Venous Blood Sampling

Following ''universal precautions,'' phlebotomists with specific training for the collection of venous blood lead samples collected two blood samples from the antecubidal area of each participant's arm (inside arm opposite the elbow) [Centers for Disease Control and Prevention, 1988,1991]. The blood samples were collected in an occupational health clinic or in a training room inside the facility. All supplies used for the blood sampling including needles, evacuated tubes, and alcohol wipes were pre-screened for lead contamination. The screening was conducted using a dilute nitric acid solution as described elsewhere [National Committee for Clinical Laboratory Standards (NCCLS), 2001].

At both study sites, the sampling area utilized a separate ventilation system from the manufacturing process. Air samples were collected to determine whether there was any airborne lead contamination in the analysis room. Prior to the venous blood draw, the employee's arm was cleaned with an alcohol wipe to remove potential lead contamination. A second alcohol wipe was then used to sterilize the sampling area. Each participating employee donated two 2 ml venous samples for the study. Each blood sample was drawn into a Vacutainer^{TM} tube (K_3EDTA, BD# 36-9651) identified with a bar-coded identification label. The samples were immediately placed in a rocker to prevent any clotting of the blood. The specific date and time of the blood draw was documented in a master log.

Laboratory Blood Lead Analysis

One Vacutainer tube from each sampling pair was refrigerated and sent via overnight courier to the to the CDC laboratory. This sample was analyzed for blood lead according to the CDC Whole Blood Method 1080C using graphite furnace atomic absorption spectrometry (GFAAS) (Perkin-Elmer Model 4100-ZL GFAAS with Zeeman-effect background correction) based on the method described by Miller et al. [1987]. The GFAAS results were considered the (true) reference method.

ASV Blood Lead Analysis

The other paired sample was analyzed for blood lead onsite using one of three portable ASV instruments. All venous blood samples were kept at room temperature and were analyzed on the ASV instrument within 2 h of collection. At site one, the ASV analysis was conducted in the same large room as the blood collection. At site two, the blood analysis was conducted in an adjoining room.

Calibrations were uploaded onto each instrument every time the instrument was turned on and were repeated if a new reagent test kit was opened. Each reagent test kit includes a calibration ''button'' that is placed on the calibration reader of the instrument. In order for a calibration to be successful, the three-letter code on the calibration button must match the reagent kit as well as the code digitally listed on the LeadCare® instrument. ESA Inc. LeadCare® quality control samples were analyzed on each instrument at the beginning and end of each analytical session. Quality Control samples consisted of two concentrations: low-normal and elevated lead concentrations (7.0 and 25.8 μg/dl, respectively).

At each site, airborne lead samples were collected inside the room where the portable ASV analysis was conducted. These samples were analyzed for lead according to NIOSH Method 7082 and were reported as lower than the detection limit (LDL) [NIOSH, 1994].

The LeadCare® instrument uses electrochemistry with a small, screen-printed colloidal gold electrode to measure the amount of lead in whole blood [Wang and Tian, 1992]. The LeadCare® instrument has received 510(k) market clearance from the Food and Drug Administration (FDA) and has been classified under the CLIA of 1988 as a

TABLE I. Characteristics of Study Participants, Stratified by Site

		By site	
Characteristic	**Both sites (n = 208)**	**Site 1 (n = 66)**	**Site 2 (n = 142)**
Age (mean years ± SD)	37.3 ± 10.7	40.4 ± 11.4	35.9 ± 10.2
Gender (% male)	83.2	54.6	96.5
Race (% white)	96.2	97.0	95.8
Current smoker (%)	40.4	36.4	42.3
Years of employment (mean years ± S.D.)	5.8 ± 6.1	6.5 ± 5.2	5.5 ± 6.4
Current use of respirator (%)	90.9	74.2	98.6

moderately complex medical diagnostic device. It has been used with success during pediatric screening programs [Shannon and Rifai, 1997]. However, blood lead levels in children are significantly lower than the blood lead levels normally present in occupationally exposed adults. The operational range of the LeadCare® instrument is 0–65 μg/dl, which should encompass the vast majority of employee blood-lead levels encountered in the US. The FDA market clearance of the portable ASV instrument excludes all blood lead samples below the limit of detection. If a blood sample contains a lead concentration greater than 65 μg/dl, the instrument responds with the message "HI" in the digital display.

To begin the analysis with the LeadCare® instrument, 50 μl of fresh, whole blood was mixed with a proprietary treatment reagent [ESA, Inc, 1997]. This reagent disrupts the red blood cells that contained most of the lead and chemically disassociates the lead from the red blood cell components. "Free" lead is released in the treatment reagent in the form of divalent lead. This lead species is then available for detection on the sensor electrodes. After the blood-treatment reagent mixture is transferred to the sensor and the test is started, an electrical potential is applied by the analyzer which causes the lead to collect (plate) onto the test electrode. The analyzer then causes the removal (stripping) via a potential (voltage) sweep to more positive potentials. The resultant current associated with stripping is then measured and automatically converted into a blood lead concentration value (μg/dl) which is displayed on the analyzer [ESA, Inc, 1997]. This technique is known as ASV [Wang and Tian, 1992; Wang, 1996].

Statistical Analysis

Demographic characteristics of the study population were collected and then summarized using frequency distributions. Normally, blood lead values from laboratory analysis are reported to patients in integer figures. The ASV instrument reports blood lead values to one decimal place. In order to not bias the statistical analysis by rounding, blood lead values resulting from both the GFAAS and the ASV instrument were left to one decimal place during statistical analysis. Both parametric (*t*-tests and univariate regression analysis) and non-parametric (sign rank test) were conducted to compare the ASV portable instrument values to the GFAAS analysis values. Additionally, a regression analysis was performed to detect any differences between genders, smoking status, study sites, and the three ASV instruments.

RESULTS

Study Demographics

Site 1 was a lead battery manufacturing facility while site 2 was a lead smelting facility. Both facilities had a medical surveillance program and diverse workplace exposures. Since the participating employers could use the analytical results to satisfy the OSHA medical surveillance requirements at no cost, employer cooperation was high during facility recruitment.

A total of 208 eligible study participants were recruited from the two sites, 66 from Site 1 and 142 from Site 2 (Table I). The total study population had a mean age of 37.3 years (SD = 10.7), was mostly male (83.2%) and white (96.2%), and 40.4% were current smokers. The mean period of employment was 5.8 years (SD = 6.1), and 90.9% reported current use of a respirator. When comparing Site 1 to Site 2, Site 1 was older (40.4 vs. 35.9 years), had fewer males (54.6 vs. 96.5%), had fewer smokers (36.4 vs. 42.3%), and reported less current use of a respirator (74.2 vs. 98.6%).

The FDA market clearance of the portable ASV instrument excludes all blood lead samples below the limit

TABLE II. Quality Control Standards and Precision

Quality control standard (limits)	**N**	**Mean ± SD**	**Precision**
Level 1 (low)	30	7.4 ± 1.4	0.19
Level 2 (Normal)	30	25.8 ± 2.1	0.08

TABLE III. LeadCare Bias

Blood lead range	N	(Bias) mean percent difference ± SD	*P*-values	Mean difference ± SD	*P*-values
BLL ≤ 10.0 μg/dl	16	0.45 ± 1.1	0.1252	3.41 ± 7.61	0.0928
BLL > 10.0 μg/dl	190	0.03 ± 0.2	0.0738	0.57 ± 5.35	0.1441
Overall	206	0.06 ± 0.4	0.0213	0.79 ± 5.59	0.0436

SD, Standard Deviation.

of detection. Accordingly, two participants were excluded from the analysis because they had blood lead levels below the limit of detection (1.4 μg/dl) of the portable ASV instrument.

Quality control results were analyzed to estimate the instrument's precision. Table II presents the mean precision and standard deviation for the two quality control standards. The bias or mean percent difference between the portable ASV results and the GFAAS results is presented in Table III. Analysis indicates that the results from the field instrument yielded a slight positive bias overall (*P*-value = 0.0213), with less bias for blood lead levels above 10 μg/dl (*P*-value = 0.0738).

Method Comparison

Figure 1 presents a histogram of the differences between the portable ASV instrument results and the GFAAS results. In this data set, the statistical analysis was conducted upon the difference between the two analytical methods.

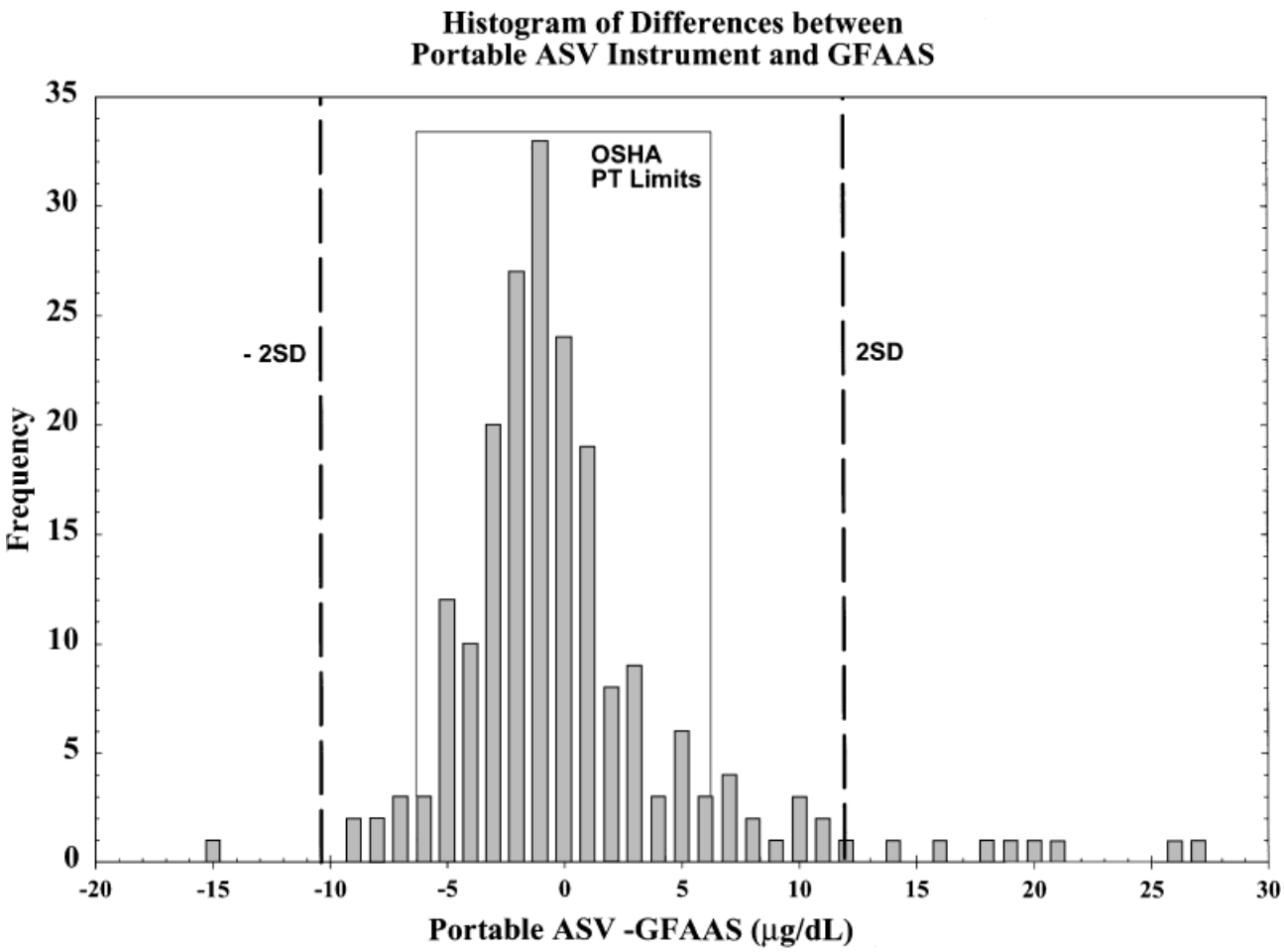

FIGURE 1. Histogram of differences between portable ASV instrument and GFAAS.

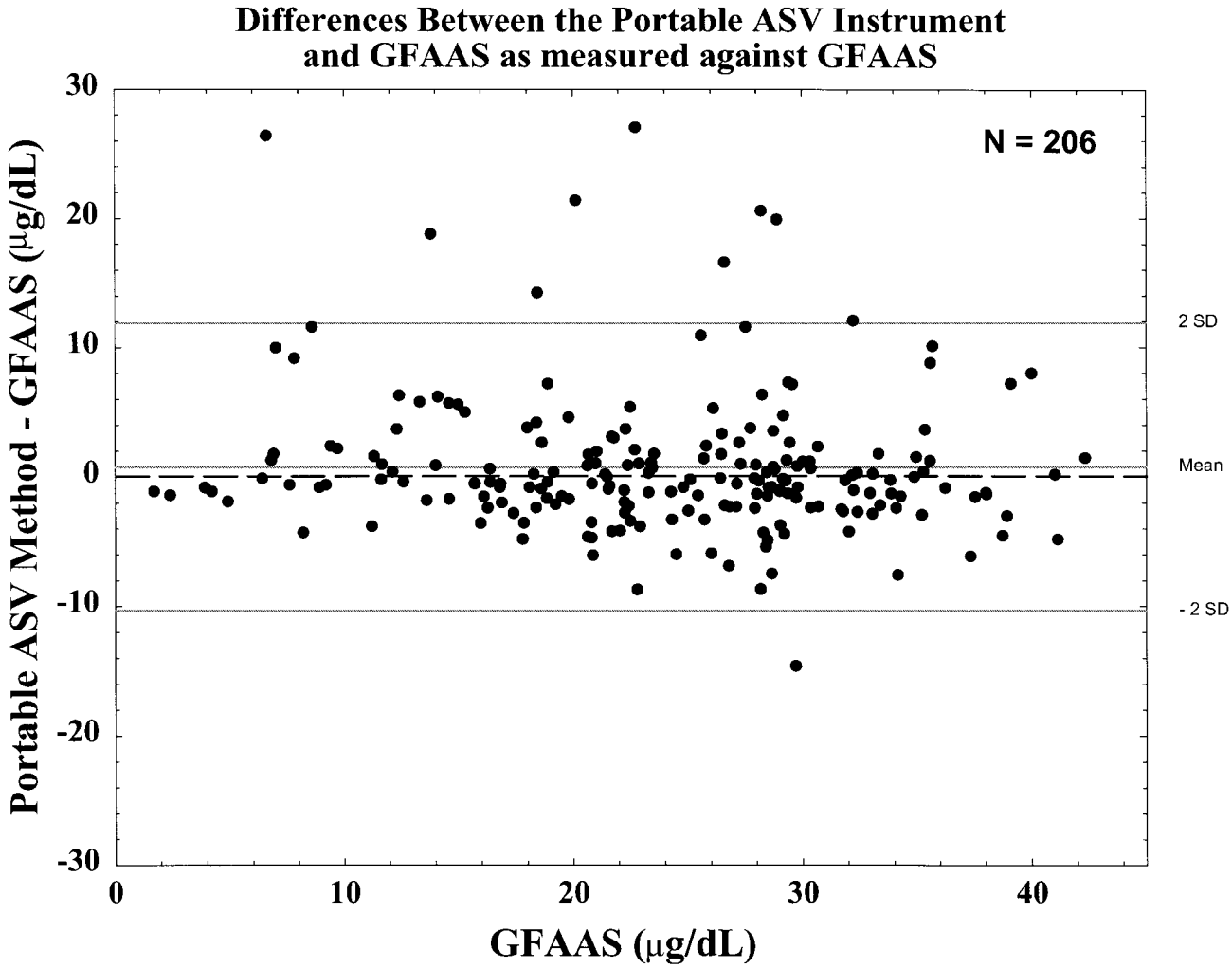

FIGURE 2. Differences between the portable ASV instrument and GFAAS as measured against GFAAS.

Since the differences between the two methods are nearly normally distributed, a logarithmic transformation was not conducted on the differences. The mean difference between the blood leads obtained from the portable ASV instrument and GFAAS was 0.79 µg/dl (SD = 5.59; P = 0.0436) indicating that on average the portable ASV instrument overestimated the true blood lead value by less than 1 µg/dl.

The differences between the portable ASV instrument results and the GFAAS blood lead analyses are plotted against the results of the laboratory blood lead results (which is considered the ''true'' blood lead concentration) in Figure 2. Approximately 95% (196/206) of the differences fell within ±11µg/dl, and the differences between the two techniques remained relatively consistent over the range of the blood lead concentrations. However, it is important to note that although the mean difference was ± 1 µg/dl, nine data points overestimated the true blood value by greater than 11 µg/dl. A simple linear regression detected a statistically significant association between the portable ASV instrument and the laboratory results (Fig. 3) [slope (standard error) = 0.91 (0.046); intercept (standard error) = 3.0 (1.17)].

Portable ASV Instrument Differences

To evaluate variability in analytical method differences and gender, age, smoking status, study site, operator team, and instrument categories, a multivariable ANOVA test was conducted. No significant differences by gender, age, smoking status, study site, or operator team were observed. However, there was a statistically significant difference between the instruments; a Tukey test indicated that results from instrument 2 were statistically different from instrument 3 (Table IV). In absolute units, the mean difference between instrument 2 and instrument 3 was 2.4 µg/dl.

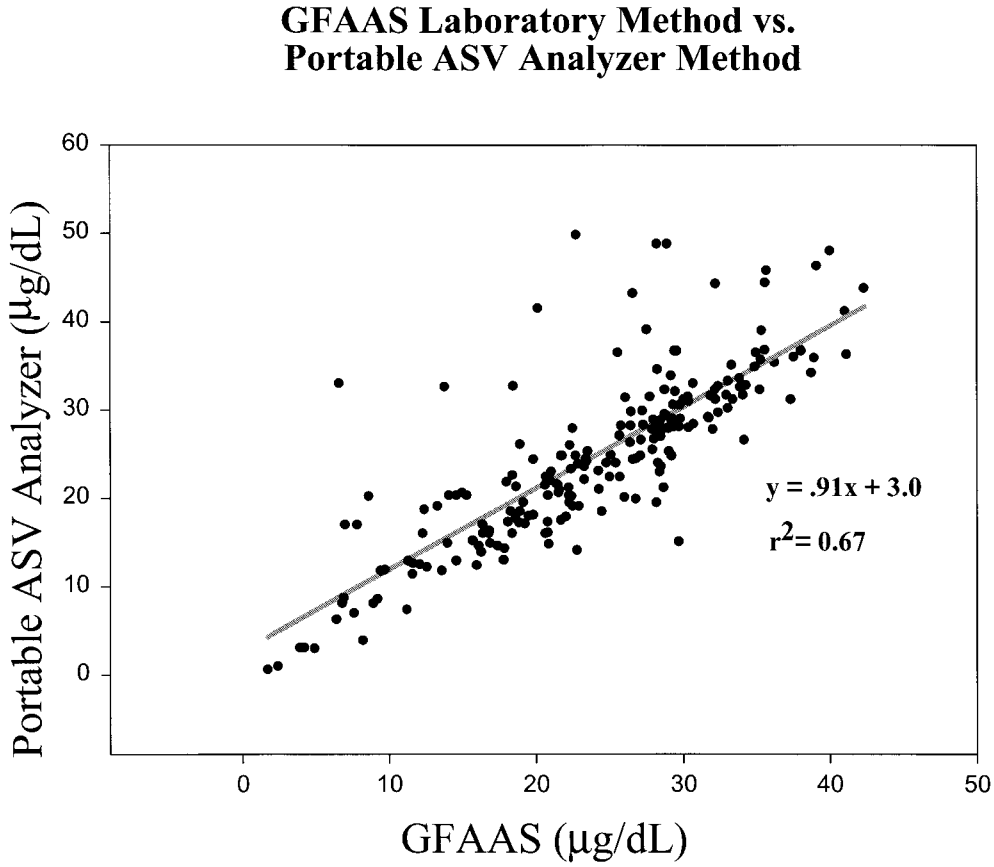

FIGURE 3. GFAAS laboratory method vs. portable ASV analyzer method.

DISCUSSION

The portable ASV instrument was evaluated within the blood lead range of 1.4–42.0 µg/dl (Figs. 2 and 3). The mean difference between the field instrument and the laboratory results was less than 1 µg/dl. Since treatment options would not be dramatically altered by a change in blood lead of ±1 µg/dl, the mean difference between the two analytical methods holds very little clinical significance.

It is unclear why the portable ASV instrument occasionally overestimates the blood lead levels by greater than ±11 µg/dl. In this data set, nine out of ten data points outside two SDs from the mean resulted in an overestimation of the true blood lead level. Occupational health professionals should evaluate the occurrence of these values when determining if the ASV instrument is appropriate for their application.

The portable ASV instrument has working range of 0–65 µg/dl whereas the GFAAS has a normal calibration

TABLE IV. Comparing the Mean Differences (µg/dl) and SD of Venous Values (Leadcare® Instrument Minus CDC Laboratory) Between Three Leadcare® Instruments

			Tukey test for significant differences (*P*-value)		
Instrument number	**N**	**Mean and standard error**	**Instrument 1**	**Instrument 2**	**Instrument 3**
Instrument 1	25	−0.99 ± 0.97	—	0.17	0.93
Instrument 2	97	0.95 ± 0.49	0.17	—	0.01
Instrument 3	53	−1.43 ± 0.67	0.93	0.01	—

Since the instrument number was not recorded on 31 samples, the total data set (n = 206) was reduced to (n = 175) for this instrument analysis.

range of 0–50 μg/dl and an effective "working range" of 0–300 μg/dl (with appropriate dilutions). The level of detection (LOD) for the portable ASV instrument is 1.4 μg/dl while the GFAAS has a LOD of 0.3 μg/dl. Overall, the GFAAS has approximately twice the precision of the LeadCare® instrument. Although the precision is less, the portable ASV instrument still has significant value as an analytical tool in blood lead analysis. Since the implementation of the OSHA lead standard and the risk of medical removal at 50 μg/dl, few employee blood lead levels in the US are greater than 65 μg/dl. Therefore, the instrument's upper operational limit will encompass the majority of employee blood lead levels. Secondly, the portability of the instrument gives it a unique advantage compared to the traditional laboratory techniques with higher precision. This instrument could be useful for occupational health professionals who work with a dynamic workforce such as the construction industry, where the workforce is transient and exposures are highly variable. Additionally, this instrument could also be used in some international locations where a high quality blood lead laboratory infrastructure does not exist.

The investment cost of this instrument is relatively low in comparison to the traditional GFAAS equipment. Depending upon the volume of samples analyzed, the cost per sample is equivalent or less than GFAAS analysis.

CLIA Certification Requirements

The current LeadCare® instrument requires the user to be CLIA certified for moderately complex testing. Obtaining CLIA certification is a resource intensive process, and should be fully considered when evaluating the potential use of this instrument in an occupational setting.

Compliance Uses

Under current interpretations, if a facility successfully participates in the proficiency program using the LeadCare® instrument, then the LeadCare® results could be used for OSHA compliance [Personal Communication, 2000].

Despite less rigorous requirements mandated by OSHA, many employees participate in monthly or bimonthly blood lead monitoring programs. If a facility is CLIA certified, the portable ASV instrument could also be utilized during non-compliance, monitoring periods.

State Reporting Requirements

Currently, many states operate registry programs that require approved blood lead laboratories to submit adult blood lead results to the state health department when the levels are above a mandated level. There is a concern among state surveillance officials that widespread utilization of this instrument could cause a bypass of the current registry systems. Although this is a possibility, there is little reason to believe that a laboratory utilizing the portable ASV instrument would be less likely to abide by state reporting requirements than a laboratory utilizing alternate analytical methods.

Study Limitations

In an effort to enroll employees with a wide range of blood lead levels, employees from all departments at each site were included in the study. Although this goal was achieved, employee blood lead levels only ranged from 1–42 μg/dl. Manufacturer specifications indicate the LeadCare® instrument has a working range from 1.4–65 μg/dl. Therefore, this study cannot comment on the instrument's performance above a blood lead concentration of 42 μg/dl. However, an evaluation at higher blood lead levels could be conducted and is an opportunity for additional research.

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