I, Michael N Fries, hereby submit this original work as part of the requirements for the degree of Master of Science in Industrial Hygiene (Environmental Health).

It is entitled:
The Validation of LeadCare © II Portable Blood Lead Analyzer through Graphite Furnace Absorption Spectroscopy (GFAAS)

Student’s name: Michael N Fries

This work and its defense approved by:

Committee chair: Glenn Talaska, Ph.D.

Committee member: Nicholas Newman, D.O., M.S.

Committee member: Michael Maier, Ph.D.
The Validation of LeadCare © II Portable Blood Lead Analyzer through Graphite Furnace Absorption Spectroscopy (GFAAS)

A thesis submitted to the Graduate School of the University of Cincinnati in partial fulfillment of the requirements for the degree of Master of Science

in the Department of Environmental Health of the College of Medicine by

Michael Fries

July 2015

B.S. Purdue University

2013

Committee Chair: Glenn Talaska, Ph.D., CIH
Abstract
This study correlates a LeadCare © II blood analyzer with Graphite Furnace Absorption Spectroscopy (GFAAS) and performs a comparison of the lead level results. Subjects who currently occupied a lead containing building that was under construction were recruited. The study consisted of 12 total participants. One sample was collected from each subject via venipuncture. The samples were then analyzed with both a LeadCare © II analyzer and also via Graphite Furnace Absorption Spectroscopy (GFAAS). The sample mean for the GFAAS analysis was 1.56 µg/dL with a SD ± 1.93. Many of the samples were reported at or below the limit of detection of 1.0 µg/dL. The reportable range of the Lead © II device is 3.3 to 65 µg/dL with most of the results being reported <3.3 µg/dL. Due to the analysis methods having different reportable ranges and most being below these measurable spectrum, it was considered illogical to make a comparison of measurements between these two devices.
Acknowledgements

I would like to begin by thanking my Advisor and Primary Investigator, Glenn Talaska, PhD, CIH, for his tremendous support, instruction and guidance throughout this study. I would like to thank Susan Pinney, PhD for taking the time to analyze the study protocol and allowing us to work with her outstanding phlebotomist. I would especially like to show my sincere thanks and gratitude to Victoria Straughn, for all of her dedication to this study and taking time to draw and collect the individual samples throughout the process. Victoria was phenomenal with all of the subjects. A big thanks to Peter Russell at Cincinnati Children’s for his coordination with the sample pick up and analysis. Thank you to Andrew Maier, PhD, CIH, DABT and Nicholas Newman, DO, MS, FAAP for taking the time to review all documents and being available to address all of my questions. Finally, I would like to thank the Hamilton County Department of Health for donating the Leadcare© II to the Department of Environmental Health at the University of Cincinnati.
Table of Contents

Abstract .......................................................................................................................... ii
Acknowledgements ..................................................................................................... iv
Table of Contents ........................................................................................................ v
List of Tables and Figures ............................................................................................ vi
Background .................................................................................................................. 1
Results ........................................................................................................................... 16
Discussion ....................................................................................................................... 18
Study Limitations .......................................................................................................... 20
Bibliography .................................................................................................................. 22
List of Tables and Figures

Tables:

Table 1: Blood Lead Levels and Health Effects

Table 2: QC Level 1 and 2 Mean and Standard Deviation

Table 3: Study Demographics

Figures:

Figure 1: Chart of average blood level measured by the CCHMC/GFAAS method

Figure 2: Distribution of blood lead level values from GFAAS analysis
Background

Lead is found all throughout our environment—the air, the soil, the water, and even inside our own homes. Most exposures to lead come from human activities including past uses of leaded gasoline, types of industrial facilities, and lead-based paint in homes. Other household products containing lead include ceramics, pipe and plumbing materials, solders, gasoline, batteries, ammunition, and cosmetics\(^1\). Lead is beneficial for many processes due to its density, durability, malleability, low melting point, long life-span, and ability to resist corrosion\(^2\).

Some of the more common occupations that result in lead exposure include construction workers, steel welders, plumbers and pipe fitters, auto repairers, firing range instructors, and shipbuilders\(^3\). Lead is most commonly found as a dust or fume in the workplace. Routes of exposure for lead include breathing in lead dust or fumes and ingesting lead dust. Lead is readily absorbed through inhalation, particularly lead fumes which are created by the condensation of gaseous lead. Lead dust and fumes do not have an odor resulting in most employees not realizing that they are being exposed.\(^4\) Ingestion can occur by lead dust settling on food, water, clothes, and other objects. It is particularly absorbed through the eyes, nose, and mouth.\(^4\)

The major sources of lead exposure in homes include paint, pipes, solders, water, batteries, and cosmetics.\(^1\) Lead exposure in homes remains a concern due to children being able to absorb more lead than adults and their brains and nervous systems being more sensitive to the damaging effects.\(^1\) Lead exposure in children has been known to lead to behavior and learning problems, lower IQ and hyperactivity, slowed growth, hearing problems, and anemia. Children also tend to touch more objects around their homes and then place their hands in their mouths creating a route of exposure. This is a primary reason for ingestion being a significant route of exposure in young children. This is especially dangerous in older homes that may contain lead based paint.
In fact, it was estimated that nearly 38 million households in the United States have lead based paint. In 2013, it was also discovered that approximately 75,760 children under the age of 6 had lead levels between (5-9 µg/dL). Lead exposure to children has become a major concern due to its ability to mimic one of the most important nutrients for children, Calcium. Children have a desire for calcium in their younger years due to rapid bone growth. With lead mimicking the absorption method of calcium, kids who are exposed could be effected because nearly 50% of the lead ingested is able to be absorbed by the body. This is a significantly higher value in comparison to adults with only 10% of what they ingest being absorbed.

Another major concern for lead is that usually no symptoms are associated with exposure and it is also capable of damaging nearly all systems and organs of the body. The symptoms of lead exposure have been separated into both short and prolonged exposure. The short term exposure health effects consist of abdominal pain, fatigue, headaches, loss of appetite, pain or tingling in the hands and feet, and weakness. Many of these symptoms can be caused by other factors which usually results in people overlooking lead exposure. The health effects from prolonged exposure involve abdominal pain, constipation, depression, being easily distracted, forgetfulness, irritability, and nausea. Further symptoms could include high blood pressure, heart disease, kidney disease, reduced fertility, and complications with the systems of the human body particularly the nervous system. As mentioned, lead is particularly dangerous to younger children causing neurological effects and mental retardation. With the long half-life of lead, even a single exposure can lead to an internal dose that lasts for years. The bottom line is that there is no safe lead level in the body. The following table provides a representation of the health effects that could arise from different blood lead levels.
<table>
<thead>
<tr>
<th>Lead Level</th>
<th>Health Effect</th>
</tr>
</thead>
<tbody>
<tr>
<td>&gt;80 µg/dL</td>
<td>serious, permanent health damage may occur, (extremely dangerous)</td>
</tr>
<tr>
<td>Between 40 and 80 µg/dL</td>
<td>serious health damage may be occurring, even if there are no symptoms (seriously elevated)</td>
</tr>
<tr>
<td>Between 25 and 40 µg/dL</td>
<td>regular exposure is occurring. There is some evidence of potential physiologic problems (elevated)</td>
</tr>
<tr>
<td>Between 10 and 25 µg/dL</td>
<td>lead is building up in the body and some exposure is occurring</td>
</tr>
</tbody>
</table>

*Lead levels and effects were provided by the New York State Department of Health*

**Table 1: Blood Lead Levels and Health Effects**

To protect the safety of employees in the workplace against substances such as lead that could be harmful to one’s health, agencies have established Occupational Exposure Limits (OELs) which enforce and recommend maximum exposure levels for a standard work day. However, not all people will have the same health effects when exposed to similar levels. There have been cases of employees becoming affected by a substance when the levels were below the OEL.

The Occupational Safety and Health Administration has been established to assure safe and healthy working conditions for all working men and women by setting and enforcing standards through training, outreach, education, and assistance. The OSHA regulations regarding lead include no worker being exposed to over 50 µg/m$^3$ over a standard 8-hour workday. Also, the action limit for lead exposure is set at 30 µg/m$^3$ meaning if levels surpass this limit, continued monitoring and medical surveillance must be performed. Employees must also undergo blood lead tests every six months. The National Institute for Occupational Safety and Health (NIOSH) provided a recommended exposure limit (REL) of 50 µg/m$^3$ over an 8 hour workday. The American Conference of Governmental Hygienists (ACGIH) provides a threshold limit
value (TLV) and biological exposure index (BEI) of 50 µg/m³ and 30 µg/100 mL lead in the blood.¹³

The appropriate blood lead level in adults has continued to remain a controversial topic. Fortunately, NIOSH has set a reference blood level of 10 µg/dL for adults. NIOSH has obtained this reference level through continued research primarily by creating the Adult Blood Lead and Epidemiology Surveillance (ABLES). ABLES began in 1987 and has continued to work to monitor laboratory-reported adult blood levels. Due to current research associating decreased renal function at 5 µg/dL and an increased risk of hypertension at levels below 10 µg/dL, the Department of Health and Human Services has recommended that blood lead levels among all adults should be reduced to <10 µg/dL.¹⁴

Trends overtime have indicated that one of the major contributors for elevated population lead levels has been leaded gasoline. Studies have supported this statement through monitoring lead levels in different populations from the time it was beginning to be phased out in 1976 to present day. A study by Pirkle et al. (1994) conducted a study from 1976 to 1991 monitored the blood lead levels of Mexican Americans who resided in the southwest part of the United States. The findings of the study included the population aged from 1 to 74 having their mean blood levels decline by 78% (12.8 µg/dL to 2.8 µg/dL). The study concludes that the major cause of this observed decline could be attributed to the removal of 99.8% of lead from gasoline.²¹

The current study was developed in 2013 when the Kettering Laboratory on the East Campus of the University of Cincinnati proposed to demolish the North wing. The North wing has traditionally been home to the Department of Environmental Health (DEH) and had been since the late 1930’s when Dr. Robert A. Kehoe was able to receive financial support to construct the
laboratories. The labs were initially used by Dr. Kehoe to work with toxicologists, industrial hygienists, physicians, chemical analysts, and engineers to evaluate and conduct experiments on occupational and environmental health problems.\textsuperscript{15}

Demolition of the North wing raises major concerns primarily due to the age of the building itself. Many of the older materials during initial construction are still present inside the building. Specifically lead based paint, chemical residue, and lead containing pipes. Also, experiments that involved mercury and beryllium were also conducted in the North wing of the building. The DEH faculty traditionally have offices in other parts of the building. However, many laboratories, offices, and classrooms that are used by the department reside in the North wing.

As previously stated, many construction materials contain lead due to its versatility. With the North wing being completely demolished, the employees of Kettering and residents in the surrounding areas could be exposed by the lead dust particles that will be generated. Exposure can occur when lead dusts and fumes are inhaled, or when lead is ingested via contaminated hands, food, water, cigarettes, or clothing. Lead that enters the body is then released into the blood and distributed throughout the body.\textsuperscript{16}

Monitoring lead levels can become difficult due to the laboratory equipment required for analysis. Monitoring levels via venipuncture and lab analysis are effective but create obstacles such as transportation and time to receive results. Field portable blood lead analyzers have been created to provide a faster analysis of blood lead levels. Instead of having to wait multiple days for the lab results, one can obtain blood lead levels of the individual within minutes. Also, one does not have to store or transport samples which reduces both costs and the possibility of samples becoming contaminated. The portable analyzers were initially developed for
measurements in children, however, using them in an occupational setting for adults has been evaluated as well. The initiative to develop an effective portable blood lead analyzer began in 1991 after the CDC provided a recommended blood level of 10 µg/dL for all children. Research emphasized the analyzer using anodic stripping voltammetry (ASV). The first instrument was created in 1997 and was called the “LeadCare I”. A second generation system with “lower regulatory requirements” was created in 2006 and called the “LeadCare II Blood Lead Test System”. The benefit of the LeadCare © II is that anyone can perform analysis on it by following the manual instructions. It has also been approved by the FDA for utilization in a non-laboratory setting.

The Department of Environmental Health at the University of Cincinnati obtained a LeadCare © II device that was donated by the Hamilton County Department of Health. In order to determine if the instrument would prove to be accurate in the field, multiple studies have been conducted that compared the LeadCare © II device to other analytical methods. The first study conducted by Kuyat (2014) involved the comparison of blood lead results between the University of Cincinnati’s LeadCare © II analyzer to the Cincinnati Children’s Hospital Medical Center (CCHMC). The objective of the study was to determine if LeadCare © II device would be applicable to monitoring employee blood lead levels during the demolition/renovation of the Kettering Laboratory North wing. The study found that between the blood lead level results, the values were slightly different but not enough to be significant. This suggests that one could consider using the LeadCare © II device to monitor the blood levels of employees.

The current study has been taken a step further with through comparison of a different method that CCHMC is using other than the LeadCare © I. To further support the theory of being able
to use the LeadCare© II analyzer for employee blood lead analysis, the device was compared to
the Graphite Furnace Atomic Absorption Spectroscopy (GFAAS) method. All information
provided here in the background emphasizes the health concerns associated with lead and the
continuing problems that it has caused in the workplace. Building demolition and renovation
proves to be particularly dangerous when handling lead due to the possibility of dust exposure.
If demolition or renovation must take place, one should ensure that proper measures are taken to
enclose the area to prevent the particles from spreading. Both the construction workers and all
occupants in the area are at risk whenever these processes are performed. According to a case
study by the CDC in 1988, five workers developed lead poisoning after a bridge in
Massachusetts was demolished. The blood lead levels of the workers ranged from 67 to 160
µg/dL. Four out of the five workers developed symptoms that were consistent with lead
poisoning and had to receive intravenous chelation therapy. 22

Even if proper control methods are taken, it is still important measure blood lead levels to
monitor exposure levels. The LeadCare © II device would serve as an efficient and quick
method to monitor these levels for both the construction workers and occupants during the
demolition/renovation process at the Kettering Laboratory.

Methods

Recruitment

All material and methodology of this study were annually reviewed and approved by the
University of Cincinnati’s Institutional Review Board. This study undergoes annual review
because it has been ongoing (IRB approval number 2014-0763) in an effort to analyze blood
levels during the demolition of the north wing of Kettering Laboratories. The participants
contacted were involved in the previous study “Validation of Lead Care © II Portable Blood
Lead Analyzer” (Kuyat, 2014). Participants in the previous study were required to be adult (18+) employees in the Department of Environmental Health (DEH). The study did not allow any pregnant women. All contact was made with the participants via email and office visits where the study was able to be reviewed and consents were collected. All participants also filled out a preliminary questionnaire used to determine possible lead exposures other than the building demolition. Each participant was assigned an identification number to assist in organizing all samples and results. Once employees had agreed to participate, emails were exchanged to determine times of availability to collect the samples.

**Venous Blood Sampling**

All participant blood samples were taken by a certified phlebotomist who was an employee of the University of Cincinnati. Approximately 3 milliliters (mL) were collected from each subject in a K2 ethylenediaminetetraacetic acid (EDTA) lead free tube with a tan cap. One sample was collected from each participant in February and March of 2015. All samples were collected in an unoccupied office on 2nd floor of the Kettering Building in the Environmental Health Department. Standard procedures were followed for taking venous blood samples. The area of the arm (cubital fossa) was wiped down with an alcohol swipe before a 21 gauge butterfly needle was used to puncture the vein. Each subject was asked to verify their date of birth for identification purposes. Each sample was given a specific I.D. and placed into a styrofoam cooler with ice packs. Dates and times of the samples were also documented onto a research processing log sheet provided by CCHMC. One copy was sent with the samples to CCHMC and other copies were made for the phlebotomist and study coordinator. Once all samples were taken for the specific date, CCHMC was then contacted where a pick-up was coordinated. Once
analyzed by CCHMC, the samples were then returned to the Biological Monitoring laboratory in the Kettering Building.

*Storage for Specimen*

The protocol stated that the blood samples were to be stored at approximately 50-90 degrees Fahrenheit and should be analyzed on the LeadCare © II device within 24 hours of being collected. If refrigerated, the samples can be analyzed within 7 days.7

*LeadCare © II ASV Analysis*

The way that the LeadCare © II conducts analysis is through a process known as Anodic Stripping Voltammetry (ASV). The process begins with the blood sample being mixed with the LeadCare Treatment Reagant which lyses the red blood cells (RBCs) causing the lead in the RBC wall to be released. A sensor within the device then has a negative potential applied to it which allows for accumulation of the lead atoms on the test electrode. This potential during this is rapidly reversed to release the lead ions. The current that produced from the previous step is directly proportional to the concentration of lead in the sample. A curve is created from measuring the current (microAmps) on the y-axis and the potential (mV) on the x-axis. The area underneath this curve is then utilized to calculate the qualitative blood lead result.

The LeadCare © II analyzer kit consisted of: 48 blood lead sensors, 48 LeadCare treatment reagent tubes, 50 LeadCare II heparinized capillary tubes and plungers, 50 LeadCare II droppers, a level 1 and level 2 control, calibration button, package insert, labels, and worksheets to record the data. To ensure valid results, all equipment must be used before the provided expiration date. The specific kit used would not expire until April of 2016. The LeadCare © II had a recordable range of 3.3-65.0 µg/dl with any value under 3.3 being reported as “Low” and any above 65.0 as “High”. The device allowed the blood samples to be collected either via a finger prick and
drawing the blood through a capillary tube or drawing a venous sample. The venous drawing method was used throughout the duration of this study.

Prior to analysis, lab attire such as safety goggles, nitrile gloves, and a lab coat had to be worn at all times. All supplies used during the analysis were sterilized with all cleaning equipment being kept away from the lead analysis device. During the analysis, all worksheets were immediately filled out to ensure accurate results. All reagent and test tubes were placed in a holder to prevent any spills or contamination.

**LeadCare © II Device Setup/Calibration**

Before the device was placed onto the Biomonitoring lab bench, the entire area was wiped down to prevent the machine from becoming polluted. The analyzer needed to be plugged into an outlet and the lever on the back needed to be turned to the “ON” position. As soon as the light on the main screen came on, a message reading “Please calibrate analyzer with button” was displayed. The yellow calibration was found in the provided LeadCare © II kit. On the front of the button, a specific QC Lot number was provided. When the button was placed onto the reader, it was held down until the machine beeped. A lot number was then provided on the device screen where it was verified that it matched the lot number on the yellow button. The specific message read “Prepare sample use sensor lot XXXXX or recalibrate then insert sensor”. If the lot numbers matched up, then the LeadCare © II was ready for analysis.
**Quality Control Tests**

To ensure the LeadCare © II results are precise; two levels of quality control (QC) tests were performed both before and after a set of blood samples were analyzed. On the blood testing system data sheet they were labeled as “Level 1” and “Level 2” with a section where the lead level results were recorded. Each QC was designated a target lead value range that the result was required to meet when analyzed to ensure accuracy. The QC tests were treated and prepared as if it were an actual blood sample. The sample preparation method can be found below. According to the Clinical Laboratory Improvement Amendments (CLIA) guidelines, controls should be run for each new lot, each new shipment of materials, any new operator, monthly to check on storage conditions, and when any other problems are identified. Both the level 1 and 2 solutions were provided in the LeadCare © II test kit. During each QC test, the level 1 control was performed first followed by the level 2. Both of the controls were required to be mixed before being used with the cap being placed on an uncontaminated surface. Once the results were obtained, the level was compared to the provided target range. The target range for level 1 was 7.5 +/−3.0 µg/dl and 24.8+/-4.0 µg/dl for level 2. The target ranges were found on the labels of the appropriate level tubes. Blood samples were not to be analyzed if any of the controls did not meet the acceptable range. If the range was not met, operators were to call the LeadCare © II support line to troubleshoot the present issue. The means and standard deviations of both QC levels that were obtained during the analysis are provided in table 2 below.
Table 2. QC Levels 1 and 2 Mean and Standard Deviation

<table>
<thead>
<tr>
<th></th>
<th>QC Level 1 (µg/dL)</th>
<th>QC Level 2 (µg/dL)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Target Range</strong></td>
<td>7.5 ±3.0</td>
<td>24.8±4.0</td>
</tr>
<tr>
<td><strong>Mean</strong></td>
<td>6.6</td>
<td>23.6</td>
</tr>
<tr>
<td><strong>Standard Deviation</strong></td>
<td>0.82</td>
<td>0.93</td>
</tr>
</tbody>
</table>

Preparing the Samples

Sample preparation began with a clean treatment reagent tube being labeled with the same sample ID that was assigned during the blood drawing. The tubes were labeled by writing the number on stickers that were provided in the LeadCare © II test kit. Once the cap of the reagent tube was removed, it was placed onto a clean surface and the tube was placed into a holder. It was crucial that the inside of the cap did not touch anything in order to avoid contamination. Before being mixed with the reagent, the blood sample in the EDTA tube was introverted approximately ten times to mix the specimen. Once mixed, the cap from the EDTA tube was removed and placed onto a clean surface. A capillary tube from the test kit was then inserted horizontally into the EDTA tube to draw up approximately 50 µL of blood. This was measured with a black line on the capillary tube. The blood sample tube was then placed back into the tube holder with the cap placed back on. Any excess blood in the area was cleaned up with the provided Kimwipes. The capillary tube was then analyzed to ensure that it was filled all the way and did not have any bubbles or contaminants. The full capillary tube was then placed into the treatment reagent where a plunger from the test kit was the placed at the top of the capillary tube and pressed down. The operator was required to push the plunger all the way down to ensure that the entire 50 µL was dispensed into the reagent tube. The reagent tube cap was then
replaced and the tube was introverted approximately ten times. Once the contents in the reagent
tube turned brown, the test sample was ready for analysis. The capillary tube, Kimwipe, plunger,
and any other material contaminated with blood were soaked in bleach water and then discarded
into a biohazard waste container.

*Analyzing the Sample*

Once the sample was prepared, a sensor from the sensor container was removed with the lid to
the container being immediately closed. The sensor was grabbed at the end without the black
bars. The sensor was then inserted completely into the analyzer on the front of the device with
the black bars facing up. The operator had to check that the sensor was under the sensor guides
and sat correctly on the analyzer deck. This was done by hearing a “beep” and seeing the
following message on the display screen:

```
“ADD SAMPLE
TO X ON SENSOR
SENSOR LOT XXXXX”
```

Both the sensor lot number on the display and the sensor lot number on the sensor container
should have matched. The sample mixture was then made sure that it as being used at room
temperature and was also uniformly mixed. The cap from the reagent treatment tube was then
removed and placed onto a clean surface. A transfer dropper was then taken from the test kit and
placed into the reagent tube. In order to draw up the sample with the dropper, the walls were
squeezed and then released when the tip was placed into the sample. The dropper was able to
draw up approximately ½ of the sample. The dropper was then removed from the reagent tube with the dropper tip being placed on the “X” of the sensor. The dropper walls were then squeezed to release the sample onto sensor. The analyzer would then beep when enough of the sample was dropped. After the beep, the 180 second (3 minute) countdown would begin on the display screen. The message on the screen read: “TESTING, XXX SECONDS TO GO”. The analyzer would beep again once the time was up and the results were presented on the display screen in µg/dL. The official message of the displayed results read:

“RECORD TEST RESULT

XX µG/dL Pb

THEN REMOVE SENSOR

SENSOR LOT XXXXX”

The results were immediately recorded onto the “Blood Lead Testing System Data Sheet”. The sensor was then removed from the analyzer which cleared the display screen making the machine ready for the next sample. All contaminated materials such as the sensor, reagent tube, and dropper were placed into bleach water and then discarded into a biohazard waste container. This procedure was repeated for every blood sample that was taken. When the analyzer produced a level result as “Low”, this was interpreted as the blood lead level being lower than 3.3 µg/dL and the result was recorded as “<3.3 µg/dL”. Any “High” display meant that the blood lead level was “>65 µg/dL” and further laboratory tests needed to be conducted. Once all samples were analyzed and the necessary data was recorded, the analyzer was switched to “OFF”. The EDTA
tubes containing the blood samples were placed into a biohazard packet and properly stored in
the biological monitoring refrigerator.

_CCHMC Analysis, GFAAS_

CCHMC used a GFAAS device to conduct the blood lead analysis. The specific machine that
conducted the analysis was the PinAAcle 900T atomic absorption spectrometer with an AS 900
furnace autosampler. This method has proved to be both cost effective and also allows for
detection limits that are under the blood-lead standards. The blood analyzed had to be collected
within 24 hours and stored in EDTA lead free tubes. The venous blood samples were sent to
CCHMC the day of collection to meet these criteria. All samples were prepared by following the
procedures provided in the PinAAcle 900T © User’s Guide. The GFAAS method had an
Analytical Measurement Range (AMR) of 1.0 to 60 µg/dL. Samples below this range were
reported as <1.0 µg/dL and samples above were reported as >60.0 µg/dL. There are many ways
to deal with non-detects, for the purpose of this study, any sample that was reported below the
range were reported as 1.0 µg/dL for the statistical analysis.

_Providing Results to Subjects_

Results were provided on a CCHMC portal (CareEvolve) that was username and password
protected. Results were presented as a pdf which simply stated the date the samples were
received along with the blood levels that were measured in mcg/dL. The pdf copy with the
results was then emailed to participants individually along with a letter stating the NIOSH
reference blood levels for adults.
Results

Demographics

The study consisted of a total of 12 participants who had previously participated in a study done Kuyat (2014) validating the Lead Care © II Portable Blood Analyzer. The mean age of the study population was 51.25 years (SD ± 16.72). The preliminary questionnaire provided to this population resulted in approximately 33.3% living in a home that contained lead pipes or brass and copper pipes that were soldered with lead. Also, 8.3% of the population currently resided in homes that contained lead based paint. Finally, 8.3% of the participated in lead containing hobbies. (Table 2).

<table>
<thead>
<tr>
<th>Characteristic/Demographic</th>
<th>n=12</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (mean years ± SD)</td>
<td>51.25 ±16.72</td>
</tr>
<tr>
<td>Visited countries still using lead gasoline in past 2 years (% yes)</td>
<td>0</td>
</tr>
<tr>
<td>Involved in any research using lead (% yes)</td>
<td>0</td>
</tr>
<tr>
<td>Home contains lead based paint (% yes)</td>
<td>8.3</td>
</tr>
<tr>
<td>Home contains lead pipes or brass and copper pipes soldered with lead (% yes)</td>
<td>33.3</td>
</tr>
<tr>
<td>Spouse has occupational exposure to lead (% yes)</td>
<td>0</td>
</tr>
<tr>
<td>Involved with hobbies that may contain lead (pottery, jewelry, shooting, etc.) (% yes)</td>
<td>8.3</td>
</tr>
<tr>
<td>Decant wine into leaded (crystal) glass (% yes)</td>
<td>0</td>
</tr>
<tr>
<td>Current smoker (% yes)</td>
<td>0</td>
</tr>
</tbody>
</table>

Table 3: Study Demographics/Questionnaire Results

LeadCare © II Analysis Results

The venous blood samples were analyzed with the LeadCare © II Analyzer on March 3, March 11, and April 16 of 2015. Samples were analyzed as soon as they were returned by CCHMC. The reportable range of the device is 3.3 to 65 µg/dL. From the LeadCare © II, all values were recorded as being under 3.3 µg/dL or “Low” with the exception of ID number 004 producing a value of 7.7 µg/dL.
**CCHMC Results, GFAAS**

Venous blood samples were collected on 2/20/15, 3/4/15, and 3/31/15. Multiple collection dates had to be used due to scheduling conflicts with the subjects. No complications with the blood collections were experienced which resulted in all being included in the final data set. CCHMC samples from the GFAAS analyzer ranged from 1.0 to 7.4 µg/dL. This resulted in a sample mean of 1.56 µg/dL ± 1.93. All samples were sent to the CCHMC lab the day of collection to ensure accurate results.

![CCHMC/GFAAS Analysis Blood Lead Results](chart)

**Figure 1. Chart of average blood level measured by CCHMC/GFAAS method**
Results indicate that the overall levels had a low sample mean in comparison to the range. This argument is strengthened by the distribution chart portrayed in figure 2 as a majority of the blood lead results were at or below 1.0 µg/dL.

With I.D. #004 having a lead level above the “Low” range for both measuring devices, a comparison was able to be made between the two. For the LeadCare © II analyzer, the recorded value was 7.7 µg/dL and 7.4 µg/dL for the GFAAS method. As shown, these values are very similar but not exact.

**Discussion**

The overall method involved comparing a LeadCare © II analyzer to the Graphite Furnace Atomic Absorption method. The reportable range at the lower end of the devices is 3.3 µg/dL for the LeadCare © II and 1.0 µg/dL for GFAAS. The blood samples analyzed by the LeadCare
© II were mostly reported as <3.3 µg/dL which made it difficult to compare to the GFAAS method. Most of the samples were also recorded as 1.0 µg/dL or even lower for GFAAS which did not allow for any comparison. The exception came with sample ID #004 which had a 7.7 µg/dL level from the LeadCare © II and a 7.4 µg/dL level for GFAAS. Even here with a comparable value one can see that the recordable levels are not exact. This small difference does not raise a great concern due to it being within the error of the device. Both values were also close enough together that if medical attention was required, the treatment methods would be similar. However, the accuracy of the measurements could decrease as the lead levels increase raising concern for true values and clinical applications. Some of the samples were analyzed by the LeadCare © II device up to two weeks after collection which could have contributed to variation in the results.

Storage of the samples could have also led to differences within the results. According to the protocol, samples were required to be stored at precisely 50-90 degrees Fahrenheit and should have been analyzed on the LeadCare © II within 24 hours of collection. If this was not practiced, the blood could have also been mixed with the reagent and analyzed within 48 hours or could have also been refrigerated and analyzed within 1 week. A number of the samples were not able to be analyzed by the LeadCare © device within the 48 hours or one week time frame due to the lack of available reagent during the day of collection. All samples were immediately stored into coolers with ice packs and transported to the CCHMC Laboratories immediately after collection. CCHMC then stored the samples in their labs based on GFAAS requirements so these results were not affected. The samples were then returned to Kettering Laboratory within 5 business days. All samples were refrigerated until they were able to be analyzed with the LeadCare © II device.
Due to only one set of samples being collected, it was difficult to determine if levels between the LeadCare II device and the GFAAS method differed. However, it was known that no blood level was recorded above the NIOSH REL of 10 µg/dL for adults. A majority of the participants were right at or even below the lower level detection limit of the GFAAS at 1.0 µg/dL. For the subject with a recorded level of 7.4 µg/dL, the direct source for this level was not determined. However, when the results were reviewed, the subject did explain that they currently reside in an older home and have frequently been making home improvements to the basement area. If the basement area had lead containing material, this could be a possible source of exposure. The subject was informed on how to take the proper precautionary steps and to stay updated on their blood lead levels.

**Study Limitations**

The most noticeable limitation of the study deals with the small sample size of only 12 participants. Only a total of twelve subjects were able to participate. Having a larger population would have strengthened the data by providing more results and leading to a better correlation. Also, only taking one sample made it difficult to notice any trends in the data. At least two samples should have been taken from subjects at different times to notice any increases or decreases in the data. Many of the subjects had very low lead exposure levels which made the exposure analysis difficult. Our lead analyzer is only capable of detecting levels at 3.3 µg/dL or above. Most subjects had exposure levels below this value. It would have improved the overall comparison by comparing subjects who had higher lead exposure levels preferably above this range. Even most of the GFAAS results were measured at 1.4 µg/dL or lower. Studies in the past have reported the reporting accuracy decreasing as the levels have increased. The sample storage methods used did not follow the exact LeadCare II storage protocol which could have
affected the final results. Storing the samples according to the LeadCare © II requirements could have increased the overall accuracy of the readings.
Bibliography


