PE BLOOD ALCOHOL ANALYSIS PROCEDURES

ABOUT THIS DOCUMENT

The Forensic Lab Director / Quality Manager reviews this document at least annually. If changes are made, analysts acknowledge the updated procedures. Obsolete procedures are archived and retained in the laboratory for at least two years.

TABLE OF CONTENTS

I. INTRODUCTION

Forensic alcohol analysis is defined as the practical application of specialized devices, instruments and methods by trained laboratory personnel to measure the concentration of ethyl alcohol in samples of blood from persons involved in traffic accidents or traffic violations.

This activity is carried out in laboratories certified by the Maine Department of Health and Human Services (D.H.H.S.). In order to be so licensed, personnel must meet the requirement set forth by DHHS rule 10-44 Chapter 267 - CERTIFICATION STANDARDS FOR PERSONS CONDUCTING CHEMICAL ANALYSES OF BLOOD AND BREATH FOR THE PURPOSE OF DETERMINING THE BLOOD ALCOHOL LEVEL.

The Maine Health & Environmental Testing Laboratory meets the above criteria and is a certified forensic alcohol laboratory. The method selected for the determination of alcohol content of blood samples utilizes a headspace gas chromatograph to perform a test that is both qualitative and quantitative. The procedure calls for addition of a small aliquot of sample to an internal standard solution. A portion from the headspace of this mixture is injected onto gas chromatographic columns that are capable of separating ethyl alcohol from acetone and the common aliphatic alcohols (i.e., methanol, isopropanol, etc.). Quantitation is accomplished through comparison to calibration curves. Data is captured and calculations are performed by device(s) designed to do so (i.e., integrator, workstation, laboratory automation computer). The method is available to all via SharePoint, where the lab stores controlled copies of documents, forms, etc.

A. PRINCIPLE

Aliquots of biological fluids or liquids are mixed with an internal standard solution. The samples are then analyzed by headspace gas chromatography and quantitated using the internal standard technique.

B. SPECIMEN REQUIREMENTS

1. Whole blood-serum-plasma

2. Liquids and/or beverages (not on the Lab Scope of Analysis)

C. CHEMICALS

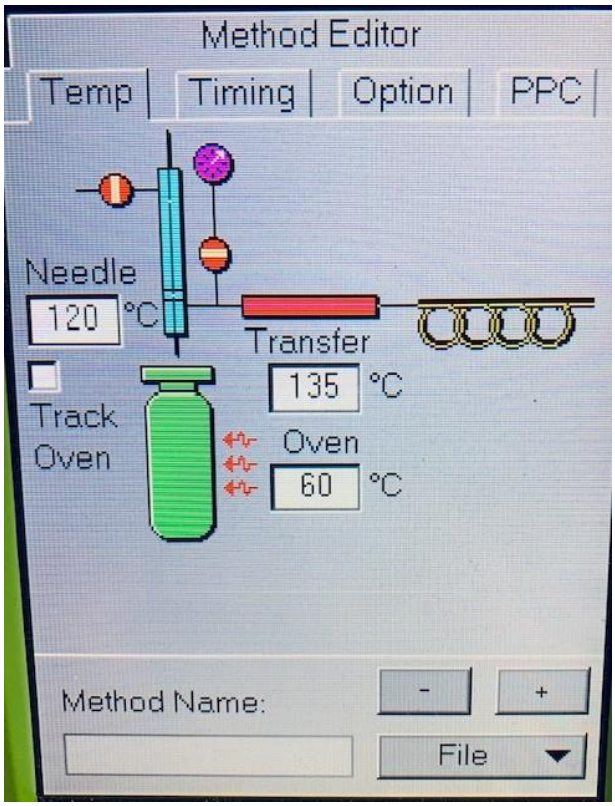
Deionized water, ethanol standards, volatiles standards, whole blood controls, and serum controls.

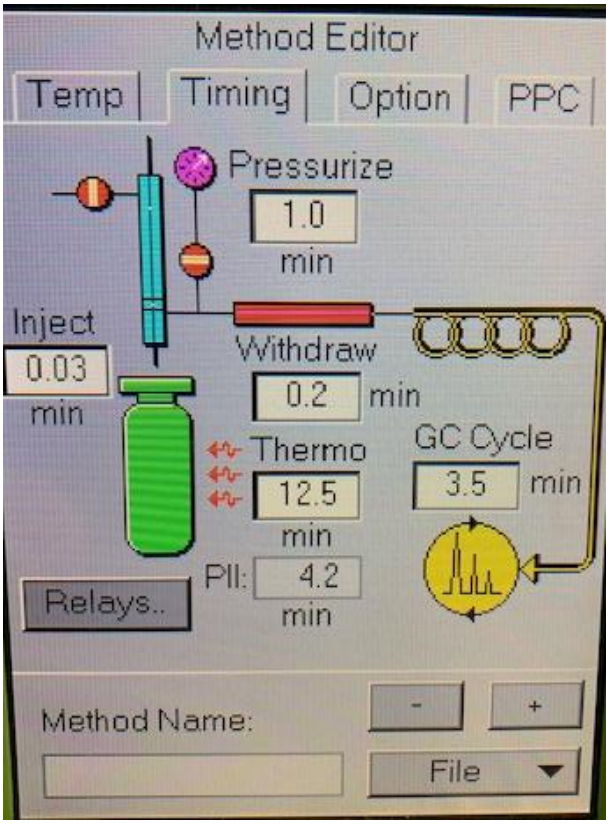
D. SAFETY PRECAUTIONS AND PPE

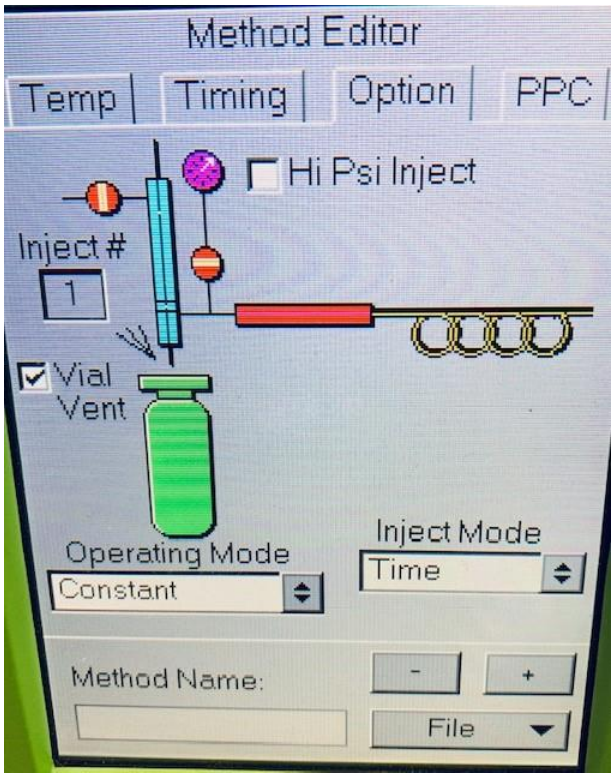
Lab coats, gloves and eye protection will be worn when handling chemicals.
Full-face shield will be worn when handling blood samples.

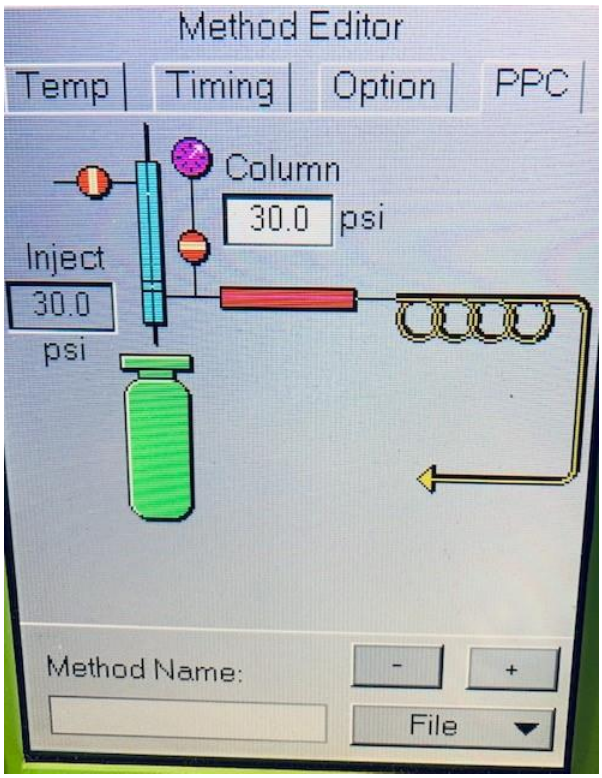
E. APPARATUS

PerkinElmer HeadSspace TurboMatrix 110 Sampler
PerkinElmer Clarius 590 gas chromatograph
Instrument Name: HS110
Detector –Flame Ionization (FID)
Columns – Elite BAC-1 Advantage 30 meter, 0.32 mmID, 1.8um df; Elite-BAC-2-Advantage 30meter, 0.32mmID, 0.6um df
Carrier Gas – Helium UHP
Detector Gas – Hydrogen
Headspace Parameters – See screen shots below









GC Parameters - See HETLBAC Method Print Out on File

F. Preparation of Solutions

Internal Standard: 0.02% by volume n-propanol
Dilute 200.0 uL n-propanol to 1.0L with deionized water (scalable as needed).

Ethanol Calibrators and Standards:

Purchased from a approved vendor (such as Cerilliant / Lipomed, etc)
Whole Blood Controls from an approved vendor (such as CliniQA Laboratories, etc)
Serum Controls from an approved vendor (such as UTAK), only run when a serum casework sample is present in the batch

Volatiles other than Ethanol: Volatile calibrators shall be purchased from an approved vendor (such as Cerilliant, Lipomed, etc). Controls containing volatiles are part of the whole blood controls from CliniQA Laboratories, and serum controls (when needed) from UTAK. Volatile calibrations will be updated twice a year, preferably prior to proficiency tests containg volatile targets.

G. QC and Sample Run Scheme:

Case samples are run in duplicate. Minimum QC batch should contain a blank (internal standard in 250ul of water), a minium reporting level standard, a mid-range standard, high-range standard, a whole blood control, a whole blood voliatile control and a serum control (if needed). The following is an example of how casework samples and QC checks/standards may appear on the batch sheet:

Blank
ETOH Standard
3 samples in duplicate
ETOH Standard
3 samples in duplicate
ETOH Standard
3 samples in duplicate
ETOH Standard
3 samples in duplicate
ETOH Standard
3 samples in duplicate
ETOH Standard
2 samples in duplicate
ETOH Standard
Whole Blood Control – Low BAC
Whole Blood Control – High BAC with Volatiles
Serum Control – 0.080 g/dL EtOH (when required)

The form used to record such information can be found on SharePoint.

II. EVIDENCE HANDLING AND PRESERVATION

All laboratory personnel will handle submitted materials in a manner that assures the integrity of the evidence. Prior to initiating and during the processing of evidence, the analyst will employ the following practices:

- The work area will be clean and free of any excess debris
- Countertops will have adequate space for working with samples
- All glassware and tools to be used will be clean
- Test tubes, capillary pipettes, Pasteur pipettes, etc are used only once, then discarded
- To prevent cross contamination of samples, **only one case will be opened by the analyst at a time**
- Reagents and solvents will be kept in closed containers when not being used in the analysis

During analysis the evidence will be under constant control by the analyst.

Evidence to be analyzed will be removed from evidence refrigerator and the reverse side of the pink Receipt/Request for Examination Form will be filled out (i.e., internal chain of custody)

The analyst will initial stickers bearing the Lab Identification Number.

If the subject's name is not available at the time of log-in, the analyst will write the subjects name on the label at the time of analysis (if known).

The analyst will verify all identification numbers and names agree with the Chain of Custody receipt.

The collection kit and all specimens will be labeled with the lab identification number, name of the subject (if known) and the analyst's initials.

All paperwork contained in the kit will be labeled with the laboratory identification number and initialed by the analyst.

The analyst will verify the case information provided with the kit matches the HETL folder, sample information from the Blood Alcohol Analysis form submitted with the sample and all Starlims labels. Any discrepancy shall be noted by the analyst within the case notes, and/or batch sheet(s).

The analyst will fill out the Blood Sample Worksheet and perform a kit inventory, making any necessary notations. The analyst will document the HETL case number on the worksheet.

The analyst will record the lot numbers of the standards, control and calibrators on the worksheet

At the time of analysis a worksheet with the specimen identification number will be created.

After analysis the remaining blood tubes will be sealed in a plastic tube container, the seal initialed by the analyst, and stored in a tube storage rack in the locked evidence refrigerator. The tube rack and position will be recorded on the Blood Sample Worksheet. All blood tubes will be held in locked storage for a period of at least six months upon completion of analysis, until being returned to the submitter or destroyed.

The sample kit (everything other than blood tubes) will be stored in an appropriately labeled box which is given a unique identification number. The box identification number will be recorded on the Blood Sample Worksheet. This box will be retained until filled in the laboratory. All filled boxes will be placed in long term storage, for a period of at least six months, until being returned to the submitter or destroyed.

III. QUALITY ASSURANCE

A. Equipment Maintenance and Calibration

Daily (when in use), before running calibration or sample sequence

- Instrument check sequence consisting of one blank and two standards (aqeuos or whole blood)

Monthly (coordinate with generator test):

- Restart Computer
- Back-up data to external hard drive
- Clean Needle

Every 6 months:

- Replace o-ring seals

Annually:

- Replace Needle Seal Assembly
- PM by vendor (when possible)

As Needed:

- Change Column
- Trim Column
- Bake Out Column

B. Reagents, Standards, and Quality Control Materials

Refer to Quality Manual

C. QUALITY CONTROL

Functional checks will be performed to check the performance of equipment and reagents used (either at regular intervals or while testing samples). Daily instrument checks will be performed, when the instrument is in use, prior to a calibration or sample sequence to check for column leaks. These checks will be reviewed by the analyst for acceptability, initialed for approval and stored in the corresponding batch folder.

Control checks will be performed during the analysis or testing process.

These checks are used to:

- Determine the performance of the analytical or testing system.

- Quantitate the variability of results from the analysis or test in terms of precision and accuracy.

The frequency of checks will be determined by:

- Currently accepted practices/standards in the discipline.
- The number of samples being run in a particular sequence.

Wherever possible, Control Charts will be set up and used to record results from selected function and control checks. Determination will be made whether the testing or analytical process is out of control and corrective actions taken will be recorded.

Control checks will be performed during the analytical or testing process. These checks are performed either with each analysis or intermittently after a specified number of analyses. These control checks include but are not limited to:

- Blanks
- Standard with known or established specifications
- Running samples in duplicate

Calibrators and standards shall be from different sources. Lot numbers and expiration dates will be recorded on worksheets.

Ethanol Calibrators and Standards will be traceable to Guide 34 compliant suppliers whenever possible. The Forensic Lab Director / Quality Manager will maintain appropriate records of approved vendors, and how vendors are approved.

D. FREQUENCY OF UPDATING THE CALIBRATION CURVE

A calibration will be updated at least once every 7 days, when samples are analyzed. This data will be stored with the ethanol control documents. Calibrators used to create the calibration curve will be of the following concentrations (g/100ml): Blank (DI water), 0.010, 0.050, 0.100, 0.200, 0.300, 0.400, 0.500. Three second source aqueous standards will be run with the curve to check accuracy of calibration curve.

Criteria for acceptance will be:

1) An r^2 value of .998 or greater for both Channels
2) All calibrators and second source standards must pass using acceptance critera defined in section VI

E. EVALUATION AND REPORTING RESULTS OUTSIDE THE RANGE OF THE CALIBRATION CURVE

Any sample determined to be greater than the largest calibrator will be reported as ‘greater than 0.xxx’ (Where xxx denotes the concentration of the largest / highest calibrator).

F. QC CHECK OF BLOOD KITS

1. Record the kit lot, blood tube lot, and PI pad lot numbers on the worksheet.
2. Add 2 mL of DI water and the liquid contents of the PI pad to one tube and vortex.
3. Transfer the resulting mixture to a second tube and vortex.
4. Label two 20mL headspace vials with identification number and suffix (A or B).
5. Using the auto dispenser re-pipetter - pipette 2500 uL of internal standard solution into each vial, and 250ul of above mixture into each vial.
6. Seal the vial by crimping the vial cap.
7. Vortex the vial.
8. Analyze by HS/GC/FID.
9. Send an email to the Quality Manager stating the kit lot number, tube lot number, PI pad lot number and the results of testing. This email will be retained with the Quality Manager.

IV. SAMPLE PREPARATION

1. Mix the sample thoroughly, allow samples to shake on rocker for at least 10 mintues
2. Label two 20mL headspace vials with identification number and suffix (A or B).
3. Prime diluter at least 3 times with DI water
4. Prepare all sample A vials at the same time, then repeate for sample B vials. Using the auto dispenser re-pipetter - pipette 2500 uL of internal standard solution into each vial, and 250ul of sample into each vial.
5. Between cases, blanks, and controls rinse the diluter with DI water.
6. Seal the vial by crimping the vial cap.
7. Vortex the vial.

Standards, controls and samples can be prepared and stored in the refrigerator for 6 days before being run on the instrument. If samples are prepared, but not able to be run within 6 days, they must be discarded and prepared again, and a note must be added to the sample worksheet. Sample preparation date will be recorded as the start date on the PE Alcohol Analysis Worksheet and control preparation date will be recorded in the column on the PE Alcohol Analysis Worksheet.

V. GAS CHROMATOGRAPHIC ANALYSIS

1. Check helium and air tanks are on and replace if necessary. Check water level on hydrogen generator. Check flames are lit.
    a. If tanks need to be changed:
        i. In TC Navigator Software
            1. Left click on Run Icon
            2. Click Release Control
        ii. On GC Touch Screen
            1. Under Tools Tab click Configure
            2. Click on B-FIDW and check disable
            3. Click on A-FIDW and check disable

iii. Change Tanks
iv. On GC Touch Screen
    1. Under Tools Tab click Configure
    2. Click on B-FIDW and uncheck disable
    3. Click on A-FIDW and uncheck disable
v. In TC Navigator Software
    1. Left click on Run Icon
    2. Click Take Control

2. Create data file path
    a. Open file explorer
    b. Navigate to C/TC DATA/Results
    c. Open most recent casework folder by navigating to the current year folder/casework/month/date
    d. Make a new folder with date and initials (mmddyyXX), subsequent runs on the same date appended with letter after date (mmddyya, mmddyyb, etc…)

3. Check carrier pressure should be ~30 psi.
4. Launch TC Navigator and log in
5. Click Build Sequence Icon
6. Edit sequence log table:
    a. Under **Type** field, fill down with 'Sample'
    b. Under **Name** field, enter all sample names and QC and standard lot numbers and exp dates
    c. Under **Number** field, right click on number column, select smart fill, sample number pattern = ##, uncheck the Synchronize with vial numbers box, enter start & end row numbers
    d. Under **Inst Method** field, right click and click folder icon to navigate to correct method, select instrument method "hetlbac" (file path: c:/tc data/methods/hetlbac)
    e. Under **Proc Method** for **Channel A** field, right click and select Change Values, click the folder icon to navigate to most recent method/date "hetlproc**a**_mmddyy ", repeat for Channel B and select most recent method/date "hetlproc**b**_mmddyy"
    f. Under **Calib Method** for **Channel A** field, right click and select Change Values, click the folder icon to navigate to most recent method/date "hetlproca_mmddyy", repeat for Channel B and select most recent method/date "hetlprocb_mmddyy"
        i. Note – Proc method and Cal method should be the same date
    g. Rpt FMT leave empty
    h. Under **Raw Data File**, right click for Channel A, select Path, check the "Apply to other channel" box, select the folder icon next to "New Path", browse to the correct results (C:\TCDATA\Resutls\Year Casework\Month\Date
        i. If data file number is not autopopulated – right click for each channel – smart fill
    i. Save As and name the same as result folder
7. Print the sequence.
8. Load vials on headspace autosampler.
9. Sequence Verification Step – As samples and controls after the vials are loaded into the sample tray read off vial labels to a second person while the second person verifies the order listed on the printed sequence. Once the correct order has been verified the second person will signoff on the sequence.
10. Enter the vial range on Headspace touch screen
11. Click Start on Headspace touch screen (method: bac)

12. Click Actions in menu bar in TC Navigator software
    a. Click set up
    b. Set up instrument window opens
        i. Check Sequence Name
        ii. Check vial numbers
        iii. Click OK

UPDATING METHOD CALIBRATION

1. Run a calibration sequence according to the above section "CREATING A SEQUENCE"
    a. Name the sequence and results folder as mmddyyCAL
2. Update old calibration with new data.
    a. Select Build Method
    b. Open the last calibration method date for A
    c. In menu bar, select Components, click Calibrate
    d. Click the folder button and navigate to the results folder of the new calibration
    e. Select files A2-A8
    f. Assign the appropriate concentration levels – make sure replace box is checked
        i. Error message will pop up because only the Ethanol calibrators are being updated.
    g. In menu bar, select Other, click Fit Analysis, save method as hetlproca_mmddyy
        i. A graph will pop up in the window of the methanol calibration.
    h. In menu bar, select Data, click Next Component
        i. Ethanol graph should now be on the screen.
    i. Print the graph
    j. Close the window
    k. Open the latest B method date
    l. Repeat for files B2-B8
3. Process the new calibration against the old calibration.
    a. Select Build Sequence, select Load sequence stored on disk
    b. Navigate to Sequences folder
    c. Under file type, select .idx with date of sequence
    d. Open the .idx file of the newest calibration sequence
    e. Under Inst Method for Channel A, right click, select Change Values, click the folder icon and navigate to the new calibration date that reads hetlproca
        i. Repeat for Channel B
    f. Under Proc Method for Channel A, right click, select Change Values, click the folder icon and navigate to the new calibration date that reads hetlproca
        i. Repeat for Channel B
    g. In menu bar, select Actions, click Batch, change the results folder file path under Change File Path to the new calibration date results folder

NOTE: Make notation on the worksheet of any instrument repair or any issue that prevents the run from going to completion. A QC check shall be performed and pass after any instrument failure, before

processing further casework samples. Minimum QC batch should contain a blank (internal standard in 250ul of water), a minium reporting level standard, a mid-range standard, high-range standard, a whole blood control, and a whole blood voliatile control. Recalibration may be necessary depending upon the scope of the repair.

ADDING NEW USER IN TOTAL CHROME SOFTWARE

1. Open TC Navigator
2. Click Admin in the file menu – click show admin tool bar
3. Click system configuration
4. Click users in menu bar – click Add
5. Enter all information (don't check box)
6. Click ok
7. Don't change anything in next window – click ok
8. Click instrument access – check all instrumnets – click ok
9. Click password – enter new password – click never expires
10. Close Software and reopen – type in user name for first time to add to drop down
11. Enter password

VI. CALCULATION and REPORTING OF RESULTS

The acceptable limits of accuracy for the standards, sample replicates during the run are as follows: ± 0.005 g/dL or ± 5% relative percent difference (RPD), whichever is greater. RPD is calculated using the following forluma: |(x2 - x1)|/((x2 + x1)/2). (Samples less than 0.010 g/dL will be reported as 'less than 0.010 g/dL' even if difference in duplicates is greater than .005 g/dL)

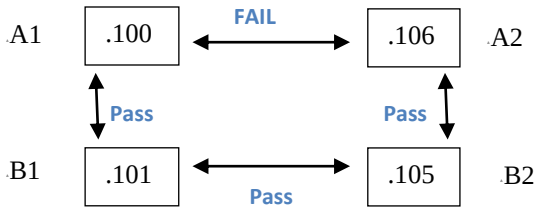
To better define and clarify exactly what is acceptable and what is not:
There are 4 results from each case (duplicate samples examined by dual column FID). Samples will be compared as such:

Vial A: Result 1 to Result 2:

Vial B: Result 1 to Result 2:

Vial A: Result 1 to Vial B: Result 1:

Vial A: Result 2 to Vial B: Result 2:

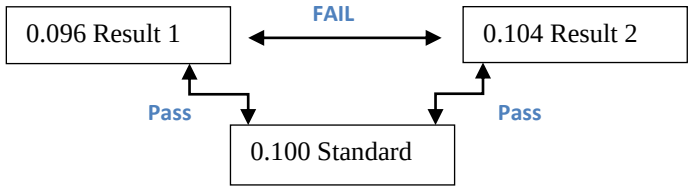


CLARIFICATION FOR STANDARDS: There are 2 results from each QC Check-Standard, one from each channel. (Result 1 – Channel A and Result 2-Channel B). Results will be compared as follows:

Result 1 to Result 2

Result 1 to nominal value of Standard

Result 2 to nominal value of Standard



For both Casework and Standards: Acceptable results are less than or equal to 5.0% RPD, or 0.005, whichever is greater. Meaning, 5.01% = Fail. 0.005 = PASS

If any sample from casework fails, at least 1 (2 if adequate sample exists) new vial(s) will be prepared and tested along with at least 1 blank, 2 aqueous controls, and 1 whole blood control.If any QC Check-Standard fails, then all casework samples that bracket the failure will be re-aliquoted and tested, with at least 1 blank, 2 aqueous standards, and 1 whole blood control.

WHOLE BLOOD CONTROLS AND SERUM CONTROLS: Whole blood controls (low and high) and the serum control each have 2 results, one from each Channel (Channel A and Channel B) and are compared to each other and to the range provided by the manufacturer. Acceptable results between Channel A and Channel B are 0.005 or 5% RPD, whichever is greater. For **whole blood controls**, when compared to the range provided by the manufacturer, both results (Channel A and B) must be within the published range. If the two values are greater than 0.005 or 5% RPD, and/or either result from Channel A or B is outside the published range, then all whole blood samples within the batch will be rejected and re-tested. For **serum controls**, both results (Channel A and B) must be within 5% of the expected concentration. If the two values are greater than 0.005 or 5% RPD, and/or or either result from Channel A or B is outside 5% of the expected contration, then all serum samples within the batch will be rejected and re-tested. The mean values of the controls are being continuously tracked by the Quality Manager.

BLANKS: Blanks results must be below 0.010 g/dL to be considered passing. If a blank fails all samples in the batch must be rerun.

REPORTING

1. All test results are recorded to three decimal places on worksheet(s).

2. Individual results from each case are averaged and always rounded down to three decimal places (example: 0.10375 is rounded down to 0.103).

An uncertainty of measurement for each result is calculated based upon the most current expanded uncertainty value. The resulting value is always rounded up to three decimal places, regardless of what the 4th significant figure is (example: 0.01424 is rounded up to 0.015). Any results less than 0.100 g/dL will be reported out with an uncertainty of 0.009 g/dL.

3. Results are reported in grams of alcohol per 100 mL of blood.

4. Serum/Plasma results will be converted to whole blood with the conversion factor 1.22:1*. A comment will be made on the report stating a serum sample was run, and indicate which color top tube was analyzed.

5. A comment will be made on the report for any non-gray top tube analyzed, noting which tube was analyzed.

6. Concentrations of ethanol below 0.010 g/100ml will be reported as “Below minimum reporting limit”

7. Worksheets and chromatograms for each sample are placed into the appropriate case folder along with a copy of batch sheet used in the calibration, and any other paperwork submitted with the case. Each page will have the case number, and initials of the analyst.

8. Final reports, worksheets, chromatograms of both samples and standards are Technically and Adminstratively Reviewed.

9. The original worksheets, sequence list and chromatograms for calibrators and standards are placed in the Ethanol Controls folder labeled with the run date. File this folder in an archives box.

10. After the administrative review is completed, the analyst will complete the notarization of the report, and it will be sent to the customer.

* Measuring Blood Alcohol Concentration for Clinical and Forensic Purposes, AW Jones and Derrick Pounder, Handbook of Drug Abuse, S Karch, MD, 1998

VII. NOTES

Blood alcohol concentrations ≥ 0.08 g/100 ml blood are prima facie evidence in operating under the influence violations.

Many volatile substances can be detected by this procedure. The most common volatiles in body fluids are ethanol, methanol, isopropanol, and acetone. All of these substances can be separated from ethanol on the gas chromatograph. HETL does not quantitate volatiles other than ethanol.

VIII. CASE DOCUMENTATION

A. CASE NOTES

The minimum information, which must be contained in the case notes are:

- Laboratory Identification Number
- Collection kit's suspect/police information paperwork
- Blood Kit Inventory Worksheet
- Run Data
- QC Data
- Comments/Results

All case notes, chromatograms and other data generated during analysis will bear the initials of the analysts and case number. Addition notes may indicate the stopper color of submitted tubes, and if a collection kit was past the published expiration date at the time of collection.

B. CASE FILE

The minimal information, which must be contained in the individual case file consists of:

- The final report
- Any preliminary, supplementary or corrected reports
- Collection kit's suspect/police information paperwork (if available)
- Worksheet(s)
- Evidence receipt
- Original chromatograms
- Technical and Administrative Review

IX. Uncertainty of Measurement

When estimating the uncertainty of measurement, all uncertainty components which are of importance shall be taken into account using appropriate testing procedures. Bias of Calibrators is also acknowledged and examined as part of the UofM procedure. Documentation, when applicable, will be retained by the Quality Manager.

A. What is being measured: Ethanol concentration in blood samples

B. Traceability of is established by using NIST / Guide 34 / ISO 17034 traceable controls, obtained by an approved vendor, and utilizing equipment calibrated to ISO 17025 standards by an accredited and approved vendor.

C. The equipment used for determining ethanol concentration:
   1) PerkinElmer Headspace Gas Chromatograph- dual-column
      i. Headspace- TurboMatrix 110 S/N HS110S1805221
      ii. Gas Chromatograph- Clarius 590 S/N 590S18053006
   2) Hamilton Diluter – 600 Series: Microlab 600 - S/N:ML600FF8801

D. The following components are recognized as potentially contributing to UofM:
   1) Whole Blood control (Reproducibility)
   2) Aqueous Controls (evaluation of bias)
   3) Temperature (liquids and ambient)
   4) Variation of time at room temperature
   5) Uncertainty stated on COA's
   6) Matrix differences
   7) Diluter: samples, controls, calibrators, and internal standard
   8) Stability of controls and calibrators
   9) Staff variability
   10) Headspace variability
   11) Concentration
   12) Internal Standard (n-propanol) stability and concentration
   13) Stability
   14) Instrument parameters
   15) Instrument precision

16) Calibration model
17) Integration parameters / processing of data

E. The following components are considered of significance:
   1) Whole Blood Matrix Control - Type A: (reproducibility of assay)
   2) Matrix - Type B: (5% administrative rule)
   3) Calibrators- Type B from COA
   4) Diluter for Calibrators - Type B from calibration certificate
   5) Diluter for Samples - Type B from calibration certificate
   6) Aqueous Controls - Type B from COA (evaluation of bias)

F. Data from controls and duplicates are tracked in a Microsoft Excel Spreadsheet. Calibration certificate(s) of the Hamilton diluter, and COA's of respective calibrators, QC standards, and whole blood controls are retained by the Quality Manager. From these spreadsheets, and in particular the WBC's, it can be determined that the Data is/is not of a normal distribution, skewed, and the mean and standard deviation calculated. Additional graphs can also be created as warranted. All values of uncertainty from individual components deemed significant (See E above) are converted to % uncertainty (See ASCLD/LAB Annex D AL-PD-3065 Ver 1.0).

G. The following calculations are performed: the standard uncertainties of the six sources identified in section E, are individually squared, and totaled. The square root is determined of the resulting sum, and this value is equal to the combined uncertainty or k. The expanded uncertainty (K2) is calculated by doubling the uncertainty (Kx2). (The method is identical to ASCLD/LAB Annex D AL-PD-3065 Ver 1.0).

$$\sqrt{(u_1)^2 + (u_2)^2 + (u_3)^2 + (u_4)^2 + (u_5)^2 + (u_6)^2} = K$$

K(2) = (Kx2) = reported UofM at 95.45% confidence interval

For reporting purposes, the lab will round up the K2 value for ease of use and understanding of case reports by our customers.

H. The schedule to review the measurement uncertainty will be conducted annually or upon the addition or replacement of laboratory equipment, staff or other factors considered of significance. The Quality Manager will retain calculations, verifications of spreadsheets, graphs, etc.

References:

ASCLD/LAB Guidance on the Estimation of Measurement Uncertainty – Annex D. (ASCLD/LAB document: AL-PD-3065 Ver 1.0 22 May 2013)

Moffat, A. C., Osselton, M. D., Widdop, B., & Watts, J. (2011). Clarke's analysis of drugs and poisons: In pharmaceuticals, body fluids and postmortem material. London: Pharmaceutical Press.