(4)

Blood Alcohol Procedure

Effective Date: 2018/09/19

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Pitt County Sheriff's Office Forensics Services Unit Issued by the Technical Leader

Technical Procedure for Blood Alcohol Analysis

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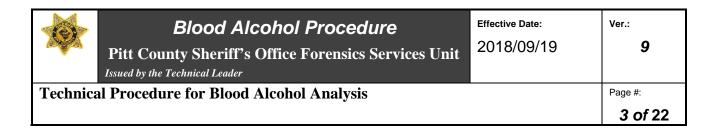
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1 PURPOSE

The purpose of this procedure is to provide detailed instruction on how to quantitatively determine the amount of alcohol in forensic blood specimens utilizing Headspace Gas Chromatography (HSGC).

2 SCOPE

The scope of the blood alcohol testing process is to allow the Forensic Chemist to:

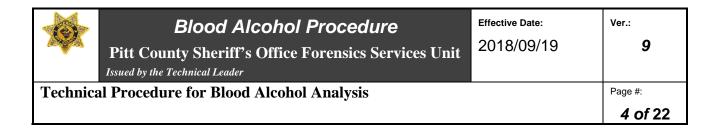
- Provide timely forensic alcohol results.
- > Provide scientifically sound expert testimony on the accuracy and reliability of forensic testing.
- ➤ Provide expert testimony when interpreting alcohol levels and impairments as it relates to driving.
- ➤ Function as a resource on alcohol analysis and impairment to the Sheriff's Office and community.

In order to meet the goals and guidelines, there are qualifications that the Forensic Chemist must meet:

- A current permit from the NC Department of Health and Human Services.
- Must be qualified as a Forensic Chemist with a minimum of a Bachelor's Degree in Chemistry, Physics, Biology or Forensic Science. Experience in a forensic or evidence laboratory setting or a combination of education and experience sufficient to successfully perform the essential duties of the job such as those listed above.
- > Training and developmental courses.
- ➤ Complete the PCSO Forensic Services Blood Alcohol Training Program.

3 DEFINITIONS

- ➤ <u>Alcohol</u> Any substance containing any form of alcohol, including ethanol, methanol, propanol, and isopropanol
- ➤ Aliquot a small representation of the sample
- ➤ <u>Analyte</u> is a substance or chemical constituent that is of interest in an analytical procedure
- ➤ <u>Calibration</u> Adjustment or standardization of the accuracy of a measuring instrument, usually by comparison with a certified reference or standard.



- ➤ <u>Calibration Standards</u> analytes used to check the calibration of the forensic alcohol method. They are analyzed at the beginning of every run as part of the daily check. Prepare the solutions using absolute ethanol diluted with Type I water.
- ➤ Commercial Reagent A purchased solvent or chemical.
- ➤ <u>Individual Volatile Standard (IVS)</u> Utilizes the individual volatiles, methanol, ethanol, *n*-propanol, isopropanol, and acetone, to establish retention times (RT's) and elution order. In some cases, these volatiles can be found in blood. IVS will be combined with Type 1 water when preparing a HSGC sample vial.
- ➤ <u>Internal Standard (ITSD)</u> an analyte used for the quantitative determination of ethanol. Presence of consistent peak areas and RTs of the internal standard in each sample ensure reliability of the accuracy and precision of the determined ethanol value.
- Linearity Check Standards (LCS) analytes used to check the linearity of the calibration curve before and after each blood sample. This is to confirm that the linearity of the curve has remained intact throughout the analysis.
- ➤ <u>Negative Control</u> used to show the absence of analytes in the Type I water and on the column. Type I water is used for this control.
- ➤ <u>NIST Calibration Standards</u> -certified ethanol standards utilized to generate a 6 point calibration curve. They are prepared in triplicate on 5 separate days when a new batch of internal standard is prepared.
- ➤ Performance Check Standard (PCS) analytes used for the initial confirmation of the reliability of equipment, instrumentation and/ or reagents; also, used to check complete separation of known analytes. A solution of reference materials containing *n*-propanol, isopropanol, ethanol, methanol, and acetone combined with Type 1 water when preparing HSGC sample vial.
- ➤ <u>Positive Control</u> Any sample with known alcohol concentrations, such as: NIST standards, PCS, 5 Calibration Standards, and Linearity Checks.
- ➤ Prepared Reagent Mixture of two or more reagents or a dilution.
- ➤ <u>Primary Reference Material</u> Any reference material obtained from a commercial source which has documentation issued by the manufacturer certifying its chemical composition or has documentation stating the manufacturer's specifications for the material. This material may be certified reference material if available and practicable.
- ➤ <u>Sampling</u> The testing of a representative portion of a substance (*i.e.*, blood) and reporting on the whole substance.
- > Type I Water Water that has been purified by ionic exchange and used for preparation of laboratory applications. It is utilized for reagent grade and advanced analytical techniques.

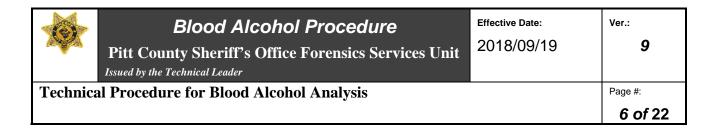
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<u>Uncertainty of Measurement-</u> A parameter associated with the result of a measurement that characterizes the distribution of values that could reasonably be attributed to that being measured. Sources contributing to the uncertainty include, but are not limited to: the operator; reference materials used; methods and equipment used; environmental conditions; properties and condition of the item being tested or calibrated.

4 EQUIPMENT AND REAGENTS

4.1 Equipment

- 1. Agilent Gas Chromatograph with Flame Ionization Detector and Software
- 2. Helium, Ultra High Purity
- 3. Hydrogen, Ultra High Purity
- 4. Nitrogen, Ultra High Purity
- 5. Quality Air, Ultra Zero
- 6. Vortex Mixer
- 7. VWR Nutating Mixer
- 8. Automatic pipette
- 9. Disposable pipette tips
- 10. Analytical Balance
- 11. Class A volumetric flasks and stoppers
- 12. Headspace vials and caps
- 13. Dispenser System
- 14. Various glassware
- 15. Centrifuge



4.2 REAGENTS

- 1. Type 1 water
- 2. Ethanol (ACS grade or equivalent)
- 3. Acetone (ACS grade or equivalent)
- 4. Isopropanol (ACS grade or equivalent)
- 5. Methanol (ACS grade or equivalent)
- 6. *n*-Propanol (ACS grade or equivalent)
- 7. Cerilliant Ethanol Standards or equivalent (NIST-traceable)

When received in lab and prior to use of reagents, they shall be inspected, initialed, and dated.

5 INTERFERENCES

There are no known interferences for this method.
Use proper sampling techniques to avoid cross contamination and carryover.

6 REAGENT PREPARATION

6.1 Preparation of Solutions

Containers of prepared reagents shall be identified with the concentration/name, date prepared & preparer's initials, and the expiration date. Lot numbers of the commercial reagents used in the preparation will be recorded in the reagent log.

6.1.1 Individual Volatile Standards

Prepare a 0.10 gram/ 100 mL solution of primary reference material of each verification solution for ethanol, methanol, acetone, *n*-propanol and isopropanol. Record the weight of each analyte. Run during the initial validation of the method and anytime the type of column is changed.

Record date prepared, expiration date and initials in the BAC reagent log and on the flask.

Expiration date = One year.

Storage = Room temperature.

6.1.2 Performance Check Standard

Prepare a mixture of 0.10 gram/ 100 mL solution of the following: ethanol, methanol, acetone, *n*-propanol and isopropanol. Run with each batch.

Record date prepared, expiration date and initials in the BAC reagent log and on the flask

Expiration date=One year.

Storage = Room temperature.

6.1.3 NIST Calibration Standards

Certified ethanol standards utilized to generate a 6 point calibration curve. They are prepared in triplicate on 5 separate days after a new batch of internal standard is prepared. The concentrations are as follows: 0.01g/100mL, 0.08g/100mL, 0.10g/100mL, 0.20 g/100 mL, 0.30g/100mL, and 0.50g/100mL.

Expiration date = As indicated by manufacturer.

Storage = Refrigerate.

6.1.4 Calibration Standards

Prepare 0.03, 0.15, 0.25, 0.35, and 0.45 g/100 mL calibration solutions from ethanol.

Record date prepared, expiration date and initials in the BAC reagent log and on the flask.

Expiration date = Six months.

Storage = Room temperature.

6.1.5 Linearity Check Standards

Prepare a 0.06 gram/ 100 mL and a 0.22 gram/ 100 mL Linearity Check Standard from ethanol. This QC control standard is used to check the entire system from sampling to analysis. Run prior to and after each blood sample.

Record date prepared, expiration date and initials in the BAC reagent log and on the flask

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Expiration date = Six months.

Storage = Room temperature.

6.1.6 BAC Internal Standard (*n*-propanol)

Prepare 0.015 g/ 100mL of primary reference material *n*-propanol.

Record date prepared, expiration date and initials in the BAC reagent log and on the flask.

Expiration date = Six months.

Storage = Room temperature.

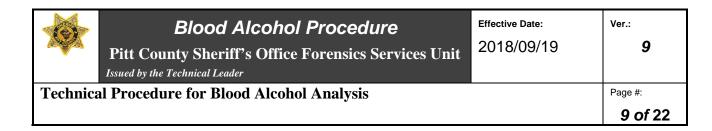
7 CALIBRATION/ VERIFICATION

7.1 NIST Calibration

- 1. Prepare 3 headspace vials for each of the 6 NIST standard concentrations. Run a sequence beginning with 2 blanks followed by each of the 6 concentrations and then repeat that order 2 more times for the remaining 2 sets of concentrations.
- 2. If the peak area for either analyte in an individual NIST calibrator is outside of the expected range, a new calibration vial will be prepared and used for that data point in the calibration curve.
- 3. The NIST calibration curve shall be fitted to a linear model.
- 4. The NIST calibration curve for ethanol shall show a correlation coefficient (R²) of 0.995 or greater. If the calibration has a correlation coefficient of less than that, appropriate action (*e.g.*, maintenance or new solution preparations) shall be made and the calibration repeated.
- 5. After each successful calibration, save as a new master method (mmddyy BAC Method).

7.2 Calibration Verification

- 1. Verify the Calibration Standards against the NIST calibration curve.
- 2. Quantitate the samples with the updated calibration table.
- 3. Each analyte shall be identified by the instrument software on both columns and the baseline visually resolved.



- 4. All analyte results shall be within $\pm 2\%$ of the target RT. If not, appropriate action shall be taken and calibration repeated.
- 5. For ethanol concentrations:

Standards whose values are \leq 0.20 (W/V) must have results < 0.005 (W/V) of the known value.

Standards whose values are > 0.20 (W/V) and ≤ 0.45 (W/V) must have results within < 0.01 (W/V) of the known value.

Standards whose values are > 0.45 (W/V) must have results < 0.03 (W/V) of the known value.

- 6. Calibration standards shall meet all requirements for ethanol to be quantitated in casework. If ethanol concentrations do not meet all requirements it shall be noted on the laboratory notes sheet.
- 7. If the calibration is found to be unacceptable for ethanol, appropriate action (*e.g.*, maintenance or new solution preparation) shall be taken and the calibration repeated.
- 8. Calibration data shall be reviewed by a Forensic Chemist qualified to perform headspace chromatography.

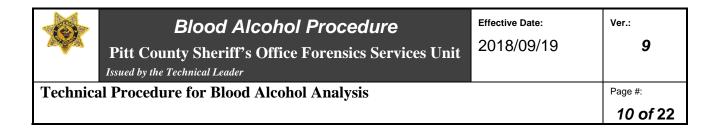
7.3 Standards and Controls

7.3.1 Daily System Check

- 1. The Daily System Check shall be performed the day of analysis and preceding any sample analysis. The Daily System Check is valid for twenty-four (24) hours after the first injection.
- 2. Prepare a PCS sample with Type 1 water, 5 calibration standard samples with ITSD, and a negative control sample using Type 1 water.
- 3. Run the 5 calibration standards, as quality control samples, using the current data analysis method corresponding to the internal standard used. Ensure verification sample analytes are identified by the instrument software on both columns and are visually baseline resolved. All analyte results shall be within the acceptable RT window. The negative control shall not contain any identifiable methanol, ethanol, isopropanol, acetone or n-propanol.
- 4. If the daily system check is found to be unacceptable for any analyte, appropriate action shall be taken and the daily system check repeated.
- 5. Record any discrepancies.

7.3.2 Quality Control

1. The first and last samples of a batch sequence shall be a negative control sample with the remaining control samples distributed throughout the batch. A Negative



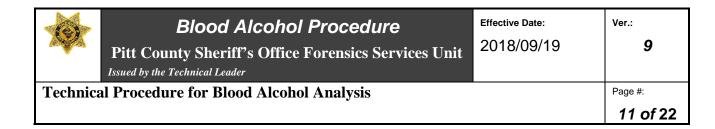
Control shall also be run after the PCS sample, after the highest calibration standard, and after the highest LCSs.

- 2. Linearity controls and calibration standards shall be selected to avoid duplicate concentrations within a sequence when possible.
- 3. Quality control samples in a sequence shall meet all requirements for each analyte to be quantitated in casework. If quality control samples do not meet all requirements it shall be noted.
- 4. If a quality control sample in a sequence is found to be unacceptable, the sequence will be aborted, if still running, and the sequence shall be re-run starting with the unacceptable quality control sample, through the end of the sequence. If the sequence has completed prior to the identification of the unacceptable quality control sample, the sequence may be re-run starting with the unacceptable quality control sample; if the sequence re-run cannot be completed within the 24 hour time frame of the Daily Check, a new daily check must be completed prior to re-running the sequence. This shall be documented.
 - Record the unacceptable QC sample on the laboratory notes sheet.
 - Correct any apparent problem and document the action on the laboratory notes sheet

7.4 Procedure

7.4.1 Sample Selection

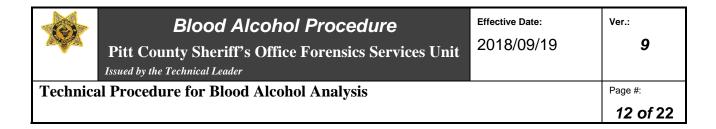
- 1. Two (2) blood tubes are submitted in one evidence DWI kit and identified as one item. If possible, only use one (1) tube to perform analysis. Identify the sample HS vial tested first as the PCSO case number followed by "BAC1" and the subsequent sample as "BAC2," etc.
- 2. Allow the blood samples to equilibrate to room temperature and utilize the VWR Nutating Mixer, and the vortex mixer if necessary, to ensure the samples are homogeneous.
- 3. Ensure that blood is homogeneous by visually inspecting them.
 - ➤ If an aliquot cannot be obtained due to the consistency of the blood sample, the blood tube will be centrifuged, a notation will be made on the laboratory notes sheet with a copy placed in the case file, and the alcohol concentration will be calculated with a 1.2 ratio.
 - ➤ If a homogeneous sample cannot be obtained because the blood cells have been separated from the liquid, a notation will be made on the laboratory notes sheet with a copy placed in the case file and the alcohol concentration shall be calculated with a 1.2 ratio.



- ➤ If a homogeneous sample cannot be obtained for any other reason, a notation shall be made on the laboratory notes sheet with a copy placed in the case file detailing the condition of the sample and its handling.
- 4. When using the Hamilton Dispenser System, the supply tubing should be inserted in the internal standard. Prime the instrument with the internal standard multiple times until you see no bubbles in the tubes. Dispense approximately 15-30 mL to prime the Dispenser.
- 5. While using the Dispenser, go to Wizards \rightarrow aliquot. Enter 1800 μ L and refill "on." Push the hand pedal and fill the sample vials with internal standard using the hand held apparatus.
- 6. Using the manual pipette, dispense $200 \,\mu\text{L}$ of standard or blood to be analyzed into the vial labeled with the appropriate identifying information and screw cap securely.
- 7. Each evidence blood sample is to be prepared in 2 separate sample vials to be analyzed.
- 8. After setting up sample sequence, save the sequence according to the date or case #, using lowercase letters in alphabetical order to indicate any supplemental runs (*e.g.*, 081115, 081115a, or 081115b).

7.5 Identification of Volatiles

- 1. Ethanol, methanol, isopropanol, *n*-propanol, and acetone shall be run in the appropriate RT window on both columns in both sample preparations to be identified.
- 2. Include the sample chromatograms, calibration data, daily system check data and quality control data in the case file. Each page should be initialed and dated by the Forensic Chemist.
- 3. Volatiles, other than those listed, may be identified using a reference material relative retention time (RRT) comparison relative to the internal standard.
 - The gas chromatographic RRT of the sample and reference material shall not differ by more than $\pm 2\%$ on each column in both sample preparations.
 - The reference material shall be analyzed under the same chromatographic conditions as the sample.
 - Include the sample chromatograms, the sample RRT, the reference material RRT, calibration data, daily system check data and QC data in the case file.



7.6 Determination of Alcohol Concentration in Blood

- 1. The concentration of ethanol shall be measured and calculated to 4 decimal places by the instrument software utilizing the most current calibration table that corresponds to the instrument used and the lot of BAC internal standard solution used to prepare the samples. The mean of the 4 measured blood sample values obtained for each analyte is the concentration of that analyte.
- 2. If any of the 4 measured blood sample ethanol values are outside of $\pm 5\%$ of the mean, the sequence shall be re-run from the first unknown sample (BAC1).
- 3. The BAC is the average of the identified ethanol in the two blood sample HS vials. Each average shall be truncated to the hundredths place for the final BAC recorded on the laboratory report. And the reported average, with the uncertainty, shall be truncated to the third decimal place.
- 4. Clotted blood samples that cannot be rendered homogenous, and samples in which the blood cells have been separated from the liquid (including serum and plasma) shall be converted to an equivalent whole blood alcohol concentration by dividing the alcohol concentration by 1.2 to compensate for the water distribution ratio of serum to whole blood. Do not convert the uncertainty of measurement.

7.7 Uncertainty of Measurement

The uncertainty of measurement (UOM) shall be calculated by the following formulas:

- 1. The Forensic Chemist shall determine an estimation of the UOM for calculating the blood alcohol concentration.
- 2. The contributions of UOM shall be evaluated using Type A methods (by a statistical analysis of measured values obtained under defined measurement conditions such as repeatability and/ or reproducibility, including measurement assurance data) and Type B methods (by other means of analysis of analytes from such things as instrument readability, calibration certificate reported uncertainty, etc.).
- 3. The equation for uncertainty of measurement is as follows:

 $C_{Corr} = (C_0 R / X_{bar}) * f_{calib}$

where: C_{Corr} = the corrected mean BAC results

 C_0 = the mean of the original measurement results

R=the traceable reference control value

X_{bar}=the mean results from measuring the controls f_{calib}=the correction factor for the linear calibration

4. The uncertainty on C₀ will be calculated from a plot of standard deviation versus blood alcohol concentration

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- 5. The uncertainty on R will be obtained from the certificate of analysis on the NIST standard.
- 6. The uncertainty on X_{bar} will be obtained from the NIST standard calibration points generated from the calibration curve for the standard with the closest concentration to the BAC.
- 7. The uncertainty on f_{calib} will be obtained from the linear equation generated from the NIST calibration plot.
- 8. Once all the above uncertainties are obtained, the combined uncertainty will be calculated from the following equation.

$$\mu_c = C_{Corr} * \sqrt{(CV_{Co}^2 + CV_R^2 + CV_{Xbar}^2 + CV_{fcalib}^2)}$$

where μ_c = combined uncertainty and CV = individual identified sources of uncertainty.

- 9. The expanded uncertainty (EU) shall be calculated to provide a minimum 99.73% coverage probability by multiplying the μ_c by the appropriate coverage factor, K.
- 10. Round the EU to three decimal places. Do not perform rounding prior to this step and, report the average BAC concentration to the same amount of decimal places.

8 OPERATING PARAMETERS

An Agilent 7890B model GC equipped with two different gas chromatographic columns terminating in two flame ionization detectors is utilized. Column A is an Agilent DB-ALC1 or equivalent and Column B is an Agilent DB-ALC2 or equivalent.

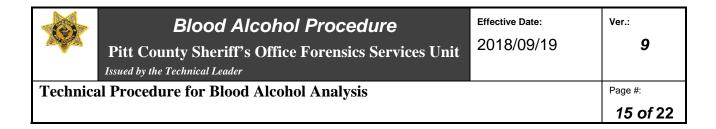
8.1 GC Parameter Table

The nominal operating parameters are:

GC Method Parameters	Agilent 7890B	
Isothermal Column Temperature:	30°C	
Injector Temperature:	150 °C	
Detector Temperature	250 °C	
Column Flow:	9 mL/min, nominal, constant flow	
Split Ratio:	3:1	
Run Time:	300 seconds	
Detector Make up Flow:	18 mL/ min.	
Detector H ₂ Flow:	30 mL/ min.	
Detector Air Flow:	300 mL/ min.	
Headspace Autosampler Method	Agilent G6509B	
Parameters		
Incubation Temperature:	70° C	
Incubation Time:	600 seconds	
Agitation Speed:	500 rpm	
Agitator On/ Off:	5 seconds on/ 2 seconds off	
Runtime:	360 seconds	
Syringe Temperature:	75 °C	
Sample Volume:	1000 μL	
Syringe fill speed:	100 μL/ second	
Flush time:	150 seconds	

8.2 Establishing Sequence Table

Setting up the sequence table involves using the software available on the GC/ FID along with the current method that is being used.



8.3 Nomenclature of Sequence

1. There is a systematic way to loading a sequence table in order to make it easier to locate files for future dates.

2. The following nomenclature will be used with the subsequent abbreviation and example. If the sample is case specific, it will include a case number (e.g., 1500035) and if it does not it will include the date (e.g., 061615):

Volatile – AcetoneIVA061615Volatile – MethanolIVM061615Volatile – IsopropylIVI061615Volatile – n-propanolIVN061615Volatile – EthanolIVE061615

Linearity Control 061615LC0600 (concentration)
Calibration Standards 061615C1500 (concentration)

NIST Calibration Standards DRNIST0500 (day-run-NIST-concentration)
Calibration Negative Control DRBLANKA/B (day-run-BLANK-A or B)

Performance Check Standard PCS061615 Negative Control NC061615 Blood Sample 1500035BAC1

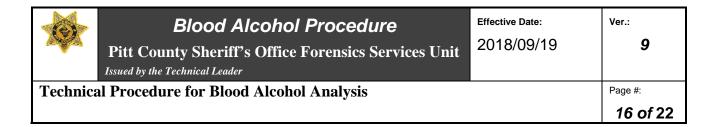
9 MAINTENANCE

- ➤ The Forensic Chemists in the BAC unit have authority to determine maintenance needs.
- > Record all maintenance on the electronic maintenance log after it is performed.
- When maintenance (excluding minor routine maintenance, such as: changing the septa, changing the inlet liner, changing the syringe, replacing the ignitor, etc.) is performed, the instrument shall be out of service until the daily check and/or calibration is successfully completed and recorded in the maintenance log.
 - After a helium or nitrogen tank is exchanged, a QC check is to be performed using at least one control solution. This shall be recorded in the electronic maintenance log.

Gas Chromatogram:

An outside vendor will perform the PM yearly. Follow the suggested vendor maintenance schedule for consumables. Frequency of use of the instrument may alter the need for maintenance. Record all maintenance on the electronic maintenance log.

Septum



Replace as per vendor specification.

Syringe

Replace approximately yearly or as needed.

<u>Liner</u>

Replace approximately yearly or as needed.

Column

Replace or cut as needed based upon quality of resolution obtained in the daily system check.

10 QUALITY ASSURANCE/ QUALITY CONTROL

Internal Standard: n-propanol

The internal standard (ITSD) is used in the calculation for the determination of ethanol. The internal standard is used to dilute all standards (except PCS and IVSs) and samples to be analyzed in a batch. A peak for the internal standard must be present in the chromatograms of each item analyzed in a run with the exception of the negative control and IVSs. Each internal standard peak must have a RT within the acceptable range.

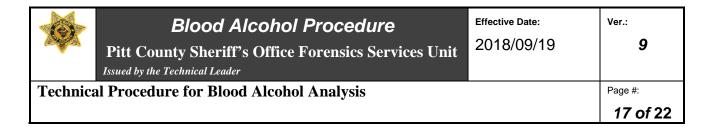
Calibration and Linearity Check Standards

The beginning of the sequence will proceed as follows: a negative control, PCS, negative control, 5 calibration standards (run from lowest to highest concentration), and another negative control. Then two linearity check standards (run from lowest to highest concentration), followed by a negative control are placed at the beginning and end of each blood sample. Values for the calibration standards and LCSs must meet the same criteria described in Section 7.2 in order for a run to be accepted as valid and the sample results to be used for reporting.

Performance Check Standard

The performance check standard (PCS) is analyzed after the first negative control leading a sequence and preceding the calibration standards. The RT for each volatile in the performance check standard must fall within the acceptable RT range for that volatile. All peaks must be present in the chromatogram of the performance check standard and there must be complete baseline separation between each peak.

Negative Control



The negative control is used to show the absence of analyte carryover in the column. This standard is always run after the highest concentration calibrator. Type 1 water is used for this control.

Proficiency Testing

Proficiency testing is required at least once per year to demonstrate that each Forensic Chemist is proficient in quantitating ethanol. Proficiency samples must be received from an outside agency or private company who provides blood alcohol samples for proficiency testing. The samples should come from an approved facility. Records of all proficiency testing must be maintained as long as the lab is performing blood alcohol analysis.

11 REPORTING GUIDELINES

11.1 Report Writing

1. When alcohol is identified and quantitated, state the concentration in the following format:

The final reported alcohol concentration is **0.XX** grams of alcohol per 100 milliliters of whole blood.

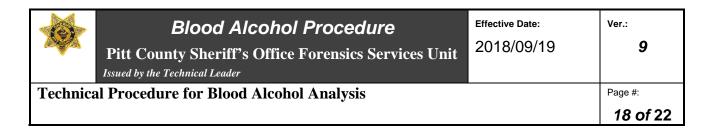
The final result is derived from two (or more) analyses with a mean of **0.XXX** ± **0.XXX** grams of alcohol per 100 milliliters of whole blood at a coverage probability of 99.73%.

2. If a serum conversion factor is applied to the measured alcohol concentration, use the statements below.

The blood alcohol concentration is **0.XX** grams of alcohol per 100 milliliters, as defined by NCGS 20-4.01 (1b).

The reported blood alcohol concentration is a calculated value resulting from a converted serum alcohol concentration. The measured serum alcohol concentration is $0.XXX \pm 0.XXX$ grams of alcohol per 100 milliliters.

3. If no alcohol is identified or if any of the quantitative results of analysis before averaging are below the lowest calibrator of 0.01 grams of alcohol per 100 milliliters of whole blood, use the statement below.



The blood alcohol concentration is 0.00 grams of alcohol per 100 milliliters, as defined by NCGS 20-4.01 (1b).

Report Reviews

After the report has been generated and reviewed by the performing Forensic Chemist, it must be technically and administratively reviewed by someone trained and authorized to perform the review other than the chemist who performed the run. Once the report has been finalized, it must be signed and notarized.

Include the lab report, chain of custody, FS1, and data including: sample chromatograms and any additional documents that are created. The analyst must initial and date the Case Review Sheet after complete technical and administrative review of the data and report generated.

Prior to reporting results of an analysis, the analysts will ensure the following meet acceptance criteria:

- 1. The RTs for each standard and sample on the chromatograms.
- 2. The R^2 value of the calibration curve.
- 3. The quantitative values of the Calibration Standard and LCSs.
- 4. The negative controls do not contain analytes.
- 5. The %RSD of the internal standard areas must be less than or equal to 10%.
- 6. The two quantitative results for each sample.

11.2 Technical Review of the Results

Technical review of a run is performed by someone trained and authorized to perform the review other than the chemist who performed the run. The technical reviewer will ensure the following meet acceptance criteria:

- 1. Check the RTs for each standard and sample.
- 2. Check the appropriate boxes on the case review checklist.
- 3. Check the quantitative values of the Calibration Standards and LCSs.
- 4. Confirm the absence of analytes in the negative controls.
- 5. Confirm the %RSD of the internal standard areas.
- 6. Confirm that the two results for each sample.
- 7. If additional analyses were performed, confirm proper inclusion and/or exclusion of results for mean calculation.
- 8. Confirm the final reported result (to the second decimal) is the truncated mean.

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- 9. Confirm the truncated mean (to the third decimal) and measurement uncertainty, rounded to three decimal places, are reflected accurately on the report.
- 10. Initial and date the Administrative/Technical Case Review Form to confirm to all requirements are met.

11.3 Reporting the Results

Each sample is analyzed from 2 separate headspace sample vials that are run individually and must meet the criteria defined in Section 7.6. The analyst will:

- 1. Record the results in the appropriate BAC UM Template worksheet for the ITSD used.
- 2. Confirm that the results meet the acceptance criteria.
- 3. Record the mean truncated to the third decimal.
- 4. Calculate the Uncertainty of Measurement (UOM) to the appropriate number of decimal places (using the BAC UM Template worksheet).
- 5. Report the UOM to the third decimal (same significance as the mean).
- 6. Record the final result to the truncated second decimal.

11.4 Administrative Review of Report

Administrative review of a report is performed by someone trained and authorized to perform the review other than the analyst who prepared the report. The administrative reviewer can also be the technical reviewer. The administrative reviewer shall:

- 1. Confirm the technical review of each run is completed.
- 2. Confirm the results from the BAC UM worksheet match the results listed on the Report.
- 3. Check the report information against the information submitted on the FS1 form:
 - a. Confirm submitting agency and case number
 - b. Subject's Name
 - c. Type of Offense
 - d. Date of Offense
 - e. Requesting Officer

11.5 Finalizing the Report

Once the technical and administrative review is completed, the final report is signed by the analyst in the presence of the notary, and then notarized.

11.6 Release and Distribution of the Report

The final report is released according to the Quality System Procedure for reporting results and the BAC SOP. Final report is distributed to the appropriate personnel. The original report with the raised notary public seal is maintained in the BAC Master File for that case. Copies of the report are distributed, via email/mail, to the Clerk of Court's Office, District Attorney's Office, Department of Motor Vehicles, Department of Health and Human Services (DHHS), and the submitting officer.

12 RECORDS

- 1. Calibration Table
- 2. Case File
- 3. Report
- 4. Analytical Data

13 REFERENCES

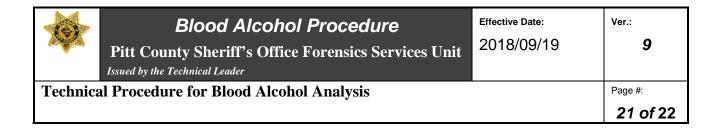
Garriott's Medicolegal Aspects of Alcohol, 6th Ed. (2015)

Operation Manual(s) for the gas chromatograph.

Operation Manual(s) for the headspace autosampler.

14 ADDITIONAL DOCUMENTS

- 1. Laboratory Notes Sheet
- 2. Case Review Sheet
- 3. UM Calculation Worksheet



15 Revision History

REVISION HISTORY			
CURRENT VERSION	EFFECTIVE DATE	SUMMARY OF CHANGES	
0	2016/07/01	Original	
1	2016/07/01	Moved Document to DM	
2	2016/10/17	Changed laboratory notebook to laboratory notes sheet (sections: 7.2 #6, 7.3.2 #5, 7.4.1 #3, & 14 #1) and changed laboratory notebook to laboratory balance binder (sections: 6.1.1, 6.1.2, 6.1.4, 6.1.5, & 6.1.6).	
3	2017/01/10	Changed HS Autosampler runtime from 300s to 360s in section 8.1. Added VWR Nutating Mixer to Section 4.1 and changed Section 7.4.1 from using the Vortex Mixer to using the Nutating Mixer.	
4	2017/04/20	Retention time criteria changed from ±10% to ±2% in Sections 7.2, 7.3.1, 7.5, 10, 11.1, & 11.2. Technical reviewer description changed under Section 11.1 to match Section 11.2. Changed NIST calibration standards from "run" to "prepared" in triplicate in Sections 3 and 6.1.3.	
5	2017/07/12	Removed Analytical Balance paragraph in Section 9: Maintenance. Added centrifuge to Sections 4.1 & 7.4.1. Changed Section 11.2 #7 to "rounded." Reworded Section 7.4.1 #2.	
6	2017/10/23	Changed Header from Rev. to Ver. & changed document title to "Technical Procedure for Blood Alcohol Analysis. Changed Revision History Table to new format. Added to procedure to Section 7.1 for addressing calibration point data that is not consistent with expected analyte peak areas. Changed "significant figures to "decimal places" in Sections 7.6 #1, 7.8 #10, 11.2 #7, and 11.3 #4.	
7	2018/02/21	Updated Purpose section. Updated Qualifications and punctuation under Scope Section. Clarified three definitions. Clarified statement in 4.2 Reagents. Changed "laboratory balance binder" to	



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REVISION	REVISION HISTORY		
CURRENT EFFECTIVE VERSION DATE		SUMMARY OF CHANGES	
		"BAC QC Check binder" throughout document. Removed "technically" from Section 7.2 #8. Removed Section 7.7 and changed Section 7.8 to 7.7. Adjusted spacing and page breaks throughout document. Updated Table of Contents. Corrected Effective date for Version 6 in Revision History table.	
8	2018/04/01	Changed Effective Date. Changed header from Issued by "Forensic Chemistry Technical Leader" to "Technical Leader".	
9	2018/09/19	Removed redundant definitions and those not used in procedure, added "positive control" and removed from body of procedure, clarified other definitions. Made all references to Type I water consistent in naming. Removed redundant mentions of the acceptable criteria. Removed quantification criteria for the Negative control throughout the document. Defined routine maintenance exceptions in Section 9, and added a statement about QC checking after changing helium or nitrogen tanks. Removed references to "other volatiles" and replaced with "n-propanol". Adjusted minimum and maximum NIST and calibration standard concentrations to allow the NIST calibration curve to encompass concentrations 0.01 to 0.50 g/100mL. Added nomenclature for the NIST calibration blanks to Section 8.3. Added storage conditions for control solutions in Section 6. Clarified actual procedures in Sections 6, 7, 11.4, & 11.5. Added coverage probability to result statement #2 in Section 11.1. Changed "case record" to "case file" and "Diluter" to "Dispenser System" throughout the document. Changed terms previously defined with an abbreviation, to the abbreviation in the rest of the document. Misc. grammar/punctuation/formatting corrections throughout	