Idaho State Police
Forensic Services
Toxicology Section

Section Five
Quality Assurance

## 5.1 Pipette Calibration

### 5.1.1 Option One: PCS 2™ Pipette Calibration System

#### 5.1.1.1 BACKGROUND

Colorimetry measures the intensity of a color and relates it to the concentration of the solution. The relationship between concentration and intensity is obtained through determining the degree of light absorbance by a solution at a particular wavelength. The fraction of the incident light that is absorbed by a solution depends on the thickness of the sample, the concentration of the absorbing compounds in the solution, and the chemical nature of the absorbing compound. This relationship is defined by the Beer-Lambert law:

$$A = \epsilon bc$$

A = Absorbance
$b$ = Internal path length (cm) of solution vial
$c$ = Concentration of sample solution
$\epsilon$ = Molar absorptivity of sample solution

The ARTEL PCS 2™ pipette calibration system is a colorimetric method for pipette calibration which utilizes a photometer coupled with NIST-traceable reagents to measure liquid delivery. The system is set up so that as additional solution ($V_{P1}$) is added to pre-mixed volume of blank solution ($V_B$), the absorbance change is proportional to the volume delivered by the pipette. The volume of solution pipetted ($V_{P1}$) is calculated as follows:

$$V_{P1} = V_B \left[ A_1 / \epsilon bc - A_1 \right]$$

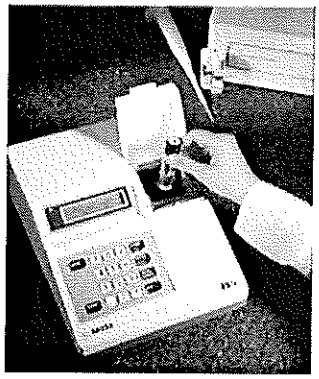
The volume of repetitions ($V_{P2}$) is determined by the following relationship:

$$V_{P2} = V_B + V_{P1} \left[ A_2 - A_1 / \epsilon bc - A_2 \right]$$

#### 5.1.1.2 SCOPE

The reliability of the volume delivered by piston or plunger operated volumetric apparatus is dependent upon verification of calibration. This method utilizes a pipette calibration system which optimizes the application of the Beer-Lambert Law to provide a reliable, time efficient, pipette calibration check that is traceable to NIST standards.

## 5.1.1.3 EQUIPMENT, SUPPLIES AND REAGENTS



5.1.1.3.1 PCS 2™ Instrument
(Artel PCS-300-00 or equivalent)

5.1.1.3.2 DPU 311 Printer
(Artel PCS-721-00 or equivalent)

5.1.1.3.3 Thermal Printer Paper
(Artel PCS-700-00 or equivalent)

5.1.1.3.4 Calibration Kit
(Artel PCS-600-00 or equivalent)

5.1.1.3.5 Appropriate Reagent Kit

Refer to PCS 2™ Calibration System Standard Operating Procedure, page 3 and PCS 2™ Pipette Calibration System Procedure Guide, page 2 for reagent kit information.

## 5.1.1.4 PROCEDURE

Refer to Standard Operating Procedure for the PCS 2™ Pipette Calibration System and PCS 2™ Pipette Calibration System Procedure Guide.

## 5.1.1.5 QUALITY ASSURANCE

5.1.1.5.1 PCS 2™ Instrument Calibration

5.1.1.5.1.1 The calibration of PCS 2™ instrument is valid for one-month providing the instrument stays in proper working order.

5.1.1.5.1.2 PCS 2™ calibration printouts, and/or a copy thereof, are to be initialed and placed in PCS 2™ logbook. A copy is permissible due to the nature of the thermal paper printout.

5.1.1.5.1.3 The calibration kit lot number, imprecision and inaccuracy results of the instrument calibration should be recorded on the PCS 2™ instrument calibration log sheet.

5.1.1.5.1.4 The results of the calibration check should be evaluated and a pass or fail indicated on the PCS 2™ instrument calibration log sheet.

5.1.1.5.2 PCS 2™ Calibration Checks

5.1.1.5.2.1 The requirement for a particular pipette to have periodic calibration checks will be indicated in the applicable SOP.

5.1.1.5.2.2 In-house calibration of pipettes by an analyst or laboratory technician will be valid for four-

months provided no maintenance was necessary during this period.

5.1.1.5.2.3 PCS 2™ instrument printouts, and/or a copy thereof, are to be initialed and placed in PCS 2™ logbook. A copy is permissible due to the nature of the thermal paper printout.

5.1.1.5.2.4 A PCS 2™ calibration check log sheet should be maintained for each pipette by serial number of other unique identifier.

5.1.1.5.2.5 The imprecision and inaccuracy results of the calibration check should be recorded on the appropriate PCS 2™ calibration check log sheet.

5.1.1.5.2.6 The results of the calibration check should be evaluated as described in section 5.1.1.5 and a pass or fail indicated on the appropriate PCS 2™ calibration check log sheet.

5.1.1.5.2.7 A minimum of 10 data points is to be collected for each check of the pipette's calibration.

5.1.1.5.2.8 A pipette not in-use need not be calibrated, however, the pipette must have its calibration prior to use.

5.1.1.5.2.9 A calibration check should be performed any time a pipette is serviced.

5.1.1.5.3 Manufacturer Data Acceptance Criteria

5.1.1.5.3.1 Artel recommendations for Piston-stroke Pipette Tolerance Limits

| *Pipette Volume* | *Inaccuracy* | *Imprecision* |
|---|---|---|
| 2µL | 5.0% | 2.0% |
| 10µL | 5.0% | 2.0% |
| 20µL | 5.0% | 2.0% |
| 100µL | 5.0% | 2.0% |
| 200µL | 5.0% | 2.0% |
| 1000µL | 5.0% | 2.0% |

5.1.1.5.3.2 Recommendations for Eppendorf Piston-stroke Fixed Volume Pipette Tolerance Limits

| *Pipette Volume* | *Inaccuracy* | *Imprecision* |
|---|---|---|
| 1µL | ±2.5% | ≤1.8% |
| 2µL | ±2.0% | ≤1.2% |
| 10µL | ±1.5% | ≤0.8% |
| 20µL | ±1.0% | ≤0.5% |
| 100µL | ±0.8% | ≤0.3% |
| 200µL | ±0.7% | ≤0.3% |
| 1000µL | ±0.6% | ≤0.2% |

5.1.1.5.3.3 Recommendations for Eppendorf Repeater Plus Pipette Tolerance Limits

| | *Inaccuracy* | *Imprecision* |
|---|---|---|
| Combitip Plus 0.1mL (beige piston) | | |
| 2µL | ±1.6% | ≤3.0% |
| 20µL | ±1.0% | ≤2.0% |
| Combitip Plus 0.2mL (blue piston) | | |
| 4µL | ±1.3% | ≤2.0% |
| 40µL | ±0.8% | ≤1.5% |
| Combitip Plus 0.5mL | | |
| 10µL | ±0.9% | ≤1.5% |
| 100µL | ±0.8% | ≤0.6% |
| Combitip Plus 1mL | | |
| 20µL | ±0.9% | ≤0.9% |
| 200µL | ±0.6% | ≤0.4% |
| Combitip Plus 2.5mL | | |
| 50µL | ±0.8% | ≤0.8% |
| 500µL | ±0.5% | ≤0.3% |
| Combitip Plus 5mL | | |
| 100µL | ±0.6% | ≤0.6% |
| 1000µL | ±0.5% | ≤0.25% |
| Combitip Plus 10mL | | |
| 200µL | ±0.5% | ≤0.6% |
| 2000µL | ±0.5% | ≤0.25% |
| Combitip Plus 25mL | | |
| 500µL | ±0.4% | ≤0.6% |
| 5000µL | ±0.3% | ≤0.25% |

| Combitip Plus 50mL | | |
|---|---|---|
| 1000µL | ±0.3% | ≤0.5% |
| 10000µL | ±0.3% | ≤0.25% |

5.1.1.5.4 Calibration Acceptance Criteria

5.1.1.5.4.1 Initially the tolerance limits recommended by ARTEL {5.1.1.5.3.1} should be applied.

5.1.1.5.4.2 When a history for an individual pipette is established, the tolerance limits should be fine-tuned and tightened accordingly.

5.1.1.5.4.3 Refer to ARTEL publication issue 5 (March 1999) for information regarding tolerance setting.

5.1.1.5.4.4 Refer to package insert for tolerance limits observed by manufacturer for each individual pipette.



## 5.1.1.6 REFERENCES

5.1.1.6.1 Standard Operating Procedure for the PCS 2™ Pipette Calibration System, Artel Document #310A2715A, April 1997,

5.1.1.6.2 PCS 2™ Pipette Calibration System Procedure Guide, Artel Document # 15A2135, Version 5.1, 03-28-1997.

5.1.1.6.3 ASTM Method E 1154-89 (reapproved 2003), **Standard Specification for Piston or Plunger Operated Volumetric Apparatus.**

5.1.1.6.4 Segel, I.H., Spectrophotometry and Other Optical Methods. pp. 324-329. *In:* "Biochemical Calculations", Second ed., John Wiley & Sons, New York, 1976.

5.1.1.6.5 Kolthoff, I.M., Sandell, E.B., Meehan, E.J. and Bruckenstein, S., Absorption Spectrophotometry. pp. 967-970, *In:* "Quantitative Chemical Analysis", Fourth ed., Macmillan, New York, 1969.

5.1.1.6.6 Setting Tolerances for Pipette Performance, Artel lab report, Issue 5, March 1999.

5.1.1.6.7 Curtis, R.H., *Performance Verification of Manual Action Pipets: Part I,* Am. Clin. Lab. 12(7):8-9; 1994.

5.1.1.6.8 Curtis, R.H., *Performance Verification of Manual Action Pipets: Part II,* Am. Clin. Lab. 12(9):16-17; 1994.

5.1.1.6.9 Eppendorf Series 2000 Reference Fixed-Volume Pipettes Instruction Manual

5.1.1.6.10 Eppendorf Series 2000 Reference Adjustable-Volume Pipettes Instruction Manual

5.1.1.6.11 Eppendorf Repeater® Plus Pipette Instruction Manual

5.1.1.6.12 Eppendorf Repeater™ Pipette Instruction Manual

5.1.1.6.13 MLA Macro and Macro Selectable Pipette Operator's Manual

**Idaho State Police**
**Forensic Services**
**Toxicology Section**

## History Page

**Toxicology Procedural Manual**
**Section Five- Quality Assurance**
**5.1 Pipette Calibration**
**5.1.1 Option One: PCS 2™ Pipette Calibration System**

| Revision # | Issue Date | History |
|---|---|---|
| 0 | 11-27-2001 | Original Issue |
| 1 | 02-02-2005 | Quality requirements detailed and updated |



## Approval

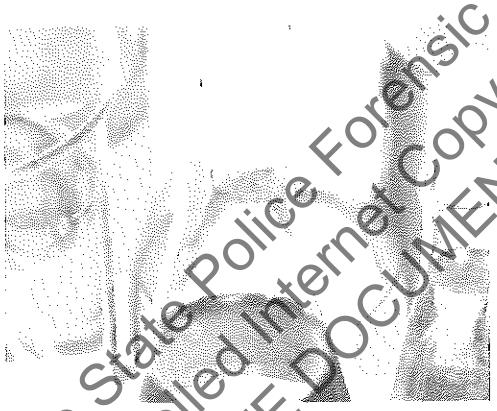
**Technical Leader:** _____ **Date:** _____
Susan C. Williamson

## Issuance

**QC Manager:** _____ **Date:** _____
Richard D. Groff

# *Idaho State Police*
## ***Forensic Services***

## *Approval for Quality System Controlled Documents*





Discipline/Name of Document: Toxicology
5.1.1 Option One: PCS 2™ Pipette Calibration System

Revision Number: 2

Issue Date: 5/07/2007

APPROVED BY: Corinna C Dudley
Quality Manager

5/7/07
Date Signed

# Section Five
# Quality Assurance

## 5.1 Intermediate Check for Verification of POVA Calibration
### 5.1.1 Option One: PCS 2™ Pipette Calibration System

#### 5.1.1.1 BACKGROUND

Colorimetry measures the intensity of a color and relates it to the concentration of the solution. The relationship between concentration and intensity is obtained through determining the degree of light absorbance by a solution at a particular wavelength. The fraction of the incident light that is absorbed by a solution depends on the thickness of the sample, the concentration of the absorbing compounds in the solution, and the chemical nature of the absorbing compound. This relationship is defined by the Beer-Lambert law:

$A = \epsilon bc$

A = Absorbance

$b$ = Internal path length (cm) of solution vial

$c$ = Concentration of sample solution

$\epsilon$ = Molar absorptivity of sample solution

The ARTEL PCS 2™ pipette calibration system is a colorimetric method for an intermediate check of pipette dispensing accuracy and precision. The system utilizes a photometer coupled with NIST-traceable reagents to measure liquid delivery. The system is set up so that as additional solution ($V_{P1}$) is added to pre-mixed volume of blank solution ($V_B$), the absorbance change is proportional to the volume delivered by the pipette. The volume of solution pipetted ($V_{P1}$) is calculated as follows:

$$V_{P1} = V_B \left[ A_1 / \epsilon bc - A_1 \right]$$

The volume of repetitions ($V_{P2}$) is determined by the following relationship:

$$V_{P2} = V_B + V_{P1} \left[ A_2 - A_1 / \epsilon bc - A_2 \right]$$



#### 5.1.1.2 SCOPE

The reliability of the volume delivered by piston or plunger operated volumetric apparatus is dependent upon verification of calibration. This method utilizes a pipette calibration verification system which optimizes the application of the Beer-Lambert Law to provide a reliable, time efficient, pipette intermediate check that is traceable to NIST standards.

#### 5.1.1.3 EQUIPMENT, SUPPLIES AND REAGENTS

5.1.1.3.1 PCS 2™ Instrument
5.1.1.3.2 Printer
5.1.1.3.3 Printer Paper
5.1.1.3.4 Calibration Kit
5.1.1.3.5 Appropriate Reagent Kit

**5.1.1.4 PROCEDURE**

Refer to manufacturer's Standard Operating Procedure for the PCS 2™ Pipette Calibration System and PCS 2™ Pipette Calibration System Procedure Guide.

**5.1.1.5 QUALITY ASSURANCE**

5.1.1.5.1 PCS 2™ Instrument Calibration Check

5.1.1.5.1.1 The calibration check of PCS 2™ instrument is valid for one-month providing the instrument stays in proper working order.

5.1.1.5.1.2 PCS 2™ calibration check printouts, and/or a copy thereof, are to be initialed and placed in PCS 2™ logbook. A copy is permissible due to the nature of the thermal paper printout.

5.1.1.5.1.3 The calibration kit lot number, imprecision and inaccuracy results of the instrument calibration check should be recorded on the PCS 2™ instrument calibration log sheet.

5.1.1.5.1.4 The results of the calibration check should be evaluated and a pass or fail indicated on the PCS 2™ instrument calibration log sheet.

5.1.1.5.2 PCS 2™ Calibration Checks

5.1.1.5.2.1 The requirement for a particular pipette to have periodic intermediate checks will be indicated in the applicable analytical method. As a rule, all methods involving quantitative analysis require periodic checks.

5.1.1.5.2.2 Intermediate check of pipette calibration is valid for four-months provided no maintenance was necessary during this period.

5.1.1.5.2.3 PCS 2™ instrument printouts, or a copy thereof, are to be initialed and placed in PCS 2™ logbook. A copy is permissible due to the nature of the

thermal paper printout.

5.1.1.5.2.4 A PCS 2™ intermediate calibration check log sheet must be maintained for each pipette by serial number or other unique identifier.

5.1.1.5.2.5 The imprecision and inaccuracy results of the intermediate check must be recorded on the appropriate PCS 2™ calibration check log sheet.

5.1.1.5.2.6 The results of the calibration check should be evaluated as described in section 5.1.1.5.4 and a pass or fail indicated on the appropriate PCS 2™ calibration check log sheet.

5.1.1.5.2.7 A minimum of 10-data points is to be collected for each check of the pipette's calibration.

5.1.1.5.2.8 A pipette not in-use need not be calibrated, however, the pipette must have its calibration verified prior to use.

5.1.1.5.2.9 An intermediate calibration check must be performed after any pipette repair/maintenance.

5.1.1.5.3 Manufacturer Data Acceptance Criteria

5.1.1.5.3.1 Artel recommendations for Piston-stroke Pipette Tolerance Limits

| *Pipette Volume* | *Inaccuracy* | *Imprecision* |
|---|---|---|
| 2µL | 5.0% | 2.0% |
| 10µL | 5.0% | 2.0% |
| 20µL | 5.0% | 2.0% |
| 100µL | 5.0% | 2.0% |
| 200µL | 5.0% | 2.0% |
| 1000µL | 5.0% | 2.0% |

5.1.1.5.3.2 Recommendations for Eppendorf Piston-stroke Fixed Volume Pipette Tolerance Limits

| *Pipette Volume* | *Inaccuracy* | *Imprecision* |
|---|---|---|
| 1µL | ±2.5% | ≤1.8% |
| 2µL | ±2.0% | ≤1.2% |
| 10µL | ±1.5% | ≤0.8% |
| 20µL | ±1.0% | ≤0.5% |
| 100µL | ±0.8% | ≤0.3% |
| 200µL | ±0.7% | ≤0.3% |
| 1000µL | ±0.6% | ≤0.2% |

5.1.1.5.3.3 Recommendations for Eppendorf Repeater Plus Pipette Tolerance Limits

| | *Inaccuracy* | *Imprecision* |
|---|---|---|
| Combitip Plus 0.1mL (beige piston) | | |
| 2µL | ±1.6% | ≤3.0% |
| 20µL | ±1.0% | ≤2.0% |
| Combitip Plus 0.2mL (blue piston) | | |
| 4µL | ±1.3% | ≤2.0% |
| 40µL | ±0.8% | ≤1.5% |
| Combitip Plus 0.5mL | | |
| 10µL | ±0.9% | ≤1.5% |
| 100µL | ±0.8% | ≤0.6% |
| Combitip Plus 1mL | | |
| 20µL | ±0.9% | ≤0.9% |
| 200µL | ±0.6% | ≤0.4% |
| Combitip Plus 2.5mL | | |
| 50µL | ±0.8% | ≤0.8% |
| 500µL | ±0.5% | ≤0.3% |
| Combitip Plus 5mL | | |
| 100µL | ±0.6% | ≤0.6% |
| 1000µL | ±0.5% | ≤0.25% |
| Combitip Plus 10mL | | |
| 200µL | ±0.5% | ≤0.6% |
| 2000µL | ±0.5% | ≤0.25% |
| Combitip Plus 25mL | | |
| 500µL | ±0.4% | ≤0.6% |
| 5000µL | ±0.3% | ≤0.25% |
| Combitip Plus 50mL | | |
| 1000µL | ±0.3% | ≤0.5% |
| 10000µL | ±0.3% | ≤0.25% |

5.1.1.5.4 Calibration Acceptance Criteria

5.1.1.5.4.1 Initially the tolerance limits recommended by ARTEL {5.1.1.5.3.1} should be applied.

5.1.1.5.4.2 When a history for an individual pipette is established, the tolerance limits should be fine-tuned and tightened accordingly.

5.1.1.5.4.3 Refer to ARTEL publication issue 5 (March 1999) for information regarding tolerance setting.

5.1.1.5.4.4 Refer to package insert for tolerance limits observed by manufacturer for each individual pipette.

## 5.1.1.6 REFERENCES


5.1.1.6.1 Standard Operating Procedure for the PCS 2™ Pipette Calibration System, Artel Document #310A2715A, April 1997,

5.1.1.6.2 PCS 2™ Pipette Calibration System Procedure Guide, Artel Document # 15A2135, Version 5.1, 03-28-1997.

5.1.1.6.3 ASTM Method E 1154-89 (reapproved 2003), **Standard Specification for Piston or Plunger Operated Volumetric Apparatus.**

5.1.1.6.4 Segel, I.H., Spectrophotometry and Other Optical Methods. pp. 324-329. *In:* "Biochemical Calculations", Second ed., John Wiley & Sons, New York, 1976.

5.1.1.6.5 Kolthoff, I.M., Sandell, E.B., Meehan, E.J. and Bruckenstein, S., Absorption Spectrophotometry. pp. 967-970, *In:* "Quantitative Chemical Analysis", Fourth ed., Macmillan, New York, 1969.

5.1.1.6.6 Setting Tolerances for Pipette Performance, Artel lab report, Issue 5, March 1999.

5.1.1.6.7 Curtis, R.H., *Performance Verification of Manual Action Pipets: Part I,* Am. Clin. Lab. 12(7):8-9; 1994.

5.1.1.6.8 Curtis, R.H., *Performance Verification of Manual Action Pipets: Part II,* Am. Clin. Lab. 12(9):16-17; 1994.

5.1.1.6.9 Eppendorf Series 2000 Reference Fixed-Volume Pipettes Instruction Manual

5.1.1.6.10 Eppendorf Series 2000 Reference Adjustable-Volume Pipettes Instruction Manual

5.1.1.6.11 Eppendorf Repeater® Plus Pipette Instruction Manual

5.1.1.6.12 Eppendorf Repeater™ Pipette Instruction Manual

5.1.1.6.13 MLA Macro and Macro Selectable Pipette Operator's Manual

Section Five

Quality Assurance

## 5.1 Intermediate Check for Verification of Pipette Calibration

### 5.1.1 Option One: PCS 2™ Pipette Calibration System

| Revision # | Issue Date | History |
|---|---|---|
| 0 | 11-27-2001 | Original Issue |
| 1 | 02-02-2005 | Quality requirements detailed and updated |
| 2 | 05-07-2007 | Updated QA measures and reformatting. |

Idaho State Police
Forensic Services
Toxicology Discipline

Section Five
Quality Assurance

## 5.1 Piston or Plunger Operated Volumetric Apparatus (POVA) Calibration Verification
## 5.1.2 Option Two: Gravimetric POVA Calibration



### 5.1.2.1 BACKGROUND

The initial calibration of piston or plunger operated volumetric apparatus (POVA) is performed by the manufacturer. Upon receipt of a newly obtained pipette or syringe and thereafter periodically, the calibration should be checked to substantiate that the volume delivered is both accurate and precise. This is accomplished by determining the mass of a volume of liquid of known density that has been delivered into a closed vessel.

### 5.1.2.2 SCOPE

The reliability of the volume delivered by POVA is dependent upon verification of calibration. This manual weighing technique is an option to evaluate the performance of each POVA. The procedure is most applicable when larger volumes (≥1mL) are employed. This SOP applies to air displacement pipettes as well as syringes attached to dilutors and dispensers.

### 5.1.2.3 EQUIPMENT

5.1.2.3.1 Analytical Balance
- Capable of accurately weighing volumes of interest.

5.1.2.3.2 Thermometer
- Long Solid-Stem.
- Traceable to NIST Standards.
- Subdivisions of ≤0.5 degree.
- Capable of reading 20° - 28°C

5.1.2.3.3 Weighing Vessel with Lid
- Nonporous material.
- Assorted sizes to accommodate volume under consideration.

5.1.2.3.4 Timer
- Capable of accurately monitoring seconds.
- Traceable to NIST Standards.

5.1.2.3.5 Appropriate disposable pipette tips.

**5.1.2.4 REAGENTS**

5.1.2.4.1 Deionized/distilled water.

**5.1.2.5 PROCEDURE**

5.1.2.5.1 General

5.1.2.5.1.1 The requirement for a particular POVA to have periodic calibration checks will be indicated in the applicable SOP.

5.1.2.5.1.2 Each POVA should be tracked by its serial number and/or other unique identifier.

5.1.2.5.1.3 In-house calibration of POVAs by an analyst or laboratory technician will be valid for four-months provided no maintenance was necessary during this period.

5.1.2.5.1.4 A POVA not in-use need not have a current calibration, however, the POVA must be checked prior to use for an application that requires a calibrated POVA.

5.1.2.5.1.5 A calibration check should be performed any time a POVA is serviced.

5.1.2.5.2 Initial set-up

5.1.2.5.2.1 The water used for the calibration process should be allowed to equilibrate at room temperature for at least two hours prior to the start of this procedure. Verify that the room and water temperature are the same prior to the start of this procedure.

5.1.2.5.2.2 Fill out identifying information on the top portion of POVA calibration check worksheet.

5.1.2.5.2.3 For adjustable volume POVA, the volume of interest should be recorded.

5.1.2.5.2.4 Determine and record the water temperature on the logsheet at the beginning and at the end of determinations.

5.1.2.5.2.5 Place a volume of water in the weighing vessel, which completely covers the bottom of the container, and cap.

5.1.2.5.2.6 Place the weighing vessel on the balance and tare.

5.1.2.5.3 First Evaporation Loss (*e*) Check

5.1.2.5.3.1 Record the initial weight of the vessel and the time of recorded on the logsheet.

5.1.2.5.3.2 Remove the lid for approximately 20 seconds. This time should be adjusted to correspond to the approximate time interval between repetitions in 5.1.2.5.4.

5.1.2.5.3.3 Record a post-weight of the vessel and the time noted.

5.1.2.5.4 POVA Determinations

5.1.2.5.4.1 Use designated POVA, to dispense appropriate volume of temperature-equilibrated water into the weighing vessel and cap.

5.1.2.5.4.2 A minimum of ten individual repetitions ($W_i$), along with their corresponding time, should be recorded.

5.1.2.5.4.3 Calculate the Mean Delivered Weight ($\overline{W}$), record on logsheet.

5.1.2.5.5 Second Evaporation Loss (*e*) Check

5.1.2.5.5.1 Record the weight of the vessel and the time of recorded on the logsheet.

5.1.2.5.5.2 Remove the lid for approximately 20 seconds.

5.1.2.5.5.3 Record a post-weight of the vessel and the time recorded.

5.1.2.5.5.4 Calculate the mean evaporation weight, record on logsheet.

5.1.2.5.6 Mean Delivered Volume

5.1.2.5.6.1 From the table below, note the conversion factor **(*Z*)** for the mean water temperature. The conversion factor is based upon an air pressure of 1013 hPa.

| *Temperature °C* | *Conversion Factor (Z) (µL/mg)* |
|---|---|
| 20.0 | 1.0029 |
| 20.5 | 1.0030 |
| 21.0 | 1.0031 |
| 21.5 | 1.0032 |
| 22.0 | 1.0033 |
| 22.5 | 1.0034 |
| 23.0 | 1.0035 |
| 23.5 | 1.0036 |
| 24.0 | 1.0038 |
| 24.5 | 1.0039 |
| 25.0 | 1.0040 |
| 25.5 | 1.0041 |
| 26.0 | 1.0043 |
| 26.5 | 1.0044 |
| 27.0 | 1.0045 |
| 27.5 | 1.0047 |
| 28.0 | 1.0048 |



5.1.2.5.6.2 Calculate the Mean Volume Delivered ($V_t$) at the mean recorded temperature.

$$V_t = (\overline{W} + e) \cdot Z$$

5.1.2.5.7 Inaccuracy Calculation

5.1.2.5.7.1 Determine inaccuracy by calculating the percent error ($E_t$) between the expected ($V_o$) and calculated mean ($V_t$) volume.

$$E_t = V_t - V_o / V_o \times 100$$

5.1.2.5.7.2 Record % error on log sheet.

5.1.2.5.8 Imprecision Calculation

5.1.2.5.8.1 Calculate the standard deviation ($s$) for the replicate weights.

$$s = \frac{\sum (W_i - \overline{W})^2}{n - 1}$$

$n$ = Total number of repetitions

5.1.2.5.8.2 Record $s$ on worksheet.

5.1.2.5.8.3 Determine the imprecision by calculating the coefficient of variation (CV%). This is also referred to as relative standard deviation (RSD).

$$\mathbf{CV\%} = s \cdot 100 / \overline{W} + e$$

5.1.2.5.8.4 Record CV % on worksheet.

5.1.2.5.9 Evaluation of Accuracy and Precision
Refer to SOP 5.1.1 for *Acceptance Criteria.*

**5.1.2.6 REFERENCES**


5.1.2.6.1 ASTM Method E-1154-89 (reapproved 2003), Standard Specification for Piston or Plunger Operated Volumetric Apparatus.

5.1.2.6.2 Curtis, R.H., *Performance Verification of Manual Action Pipets: Part I,* Am. Clin. Lab. 12(7):8-9; 1994.

5.1.2.6.3 Curtis, R.H., *Performance Verification of Manual Action Pipets: Part II,* Am. Clin. Lab. 12(9):16-17; 1994.

5.1.2.6.4 Byer, B.J., How to Use and Check Pipetting Equipment, Scientific Newsletters, Inc., 1977.

5.1.2.6.5 ISO 8655-6:2002, Piston-operated volumetric apparatus – Part 6: Gravimetric method for the determination of measurement error.

# Idaho State Police
# Forensic Services
# Toxicology Discipline

## History Page

**Toxicology Procedural Manual**
**Section Five- Quality Assurance**
**5.1 POVA Calibration**
**5.1.2 Option Two: Gravimetric POVA Calibration**

| Revision # | Issue Date | History |
|---|---|---|
| 0 | 10/1997 | Original Issue |
| 1 | 11-27-2001 | Reworked/reformatted |
| 2 | 03-22-2005 | Quality requirements detailed and updated |



## Approval

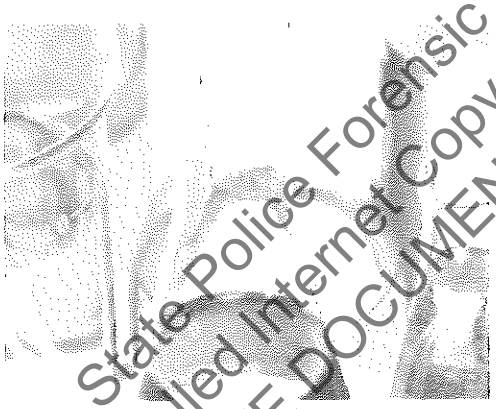
**Technical Leader:** _____ **Date:** _____
Susan C. Williamson

## Issuance

**QC Manager:** _____ **Date:** _____
Richard D. Groff

# *Idaho State Police*
## ***Forensic Services***

## *Approval for Quality System Controlled Documents*





Discipline/Name of Document: Toxicology
5.1.2 Option Two: Gravimetric Intermediate Check

Revision Number: 3

Issue Date: 5/07/2007

APPROVED BY: Corinna C Owsley
Quality Manager

5/7/07
Date Signed

# Section Five
# Quality Assurance

## 5.1 Piston or Plunger Operated Volumetric Apparatus (POVA) Intermediate Check
## 5.1.2 Option Two: Gravimetric Intermediate Check

### 5.1.2.1 BACKGROUND

The initial calibration of piston or plunger operated volumetric apparatus (POVA) is performed by the manufacturer. Upon receipt of a newly obtained pipette or syringe and thereafter periodically, the calibration must be checked to substantiate that the volume delivered is both accurate and precise. This is accomplished by determining the mass of a volume of liquid of known density that has been delivered into a closed vessel.

### 5.1.2.2 SCOPE

The reliability of the volume delivered by POVA is dependent upon verification of calibration. This manual weighing technique is an option to evaluate the performance of each POVA. The procedure is most applicable when larger volumes (≥1mL) are employed. This analytical method applies to air displacement pipettes as well as syringes attached to dilutors and dispensers.

### 5.1.2.3 EQUIPMENT

5.1.2.3.1 Analytical Balance
- Capable of accurately weighing volumes of interest.

5.1.2.3.2 Thermometer
- Long Solid-Stem
- Traceable to NIST Standards
- Subdivisions of ≤0.5 degree
- Capable of reading 20° - 28°C

5.1.2.3.3 Weighing Vessel with Lid
- Nonporous material
- Assorted sizes to accommodate volume under consideration

5.1.2.3.4 Timer
- Capable of accurately monitoring seconds
- Traceable to NIST Standards

5.1.2.3.5 Appropriate disposable pipette tips

**5.1.2.4 REAGENTS**

5.1.2.4.1 Deionized/distilled water

**5.1.2.5 PROCEDURE**

5.1.2.5.1 General

5.1.2.5.1.1 The requirement for a particular POVA to have periodic intermediate check will be indicated in the applicable analytical method.

5.1.2.5.1.2 Each POVA should be tracked by its serial number and/or other unique identifier.

5.1.2.5.1.3 Intermediate checks of POVAs by an analyst or laboratory technician will be valid for four-months provided no maintenance was necessary during this period.

5.1.2.5.1.4 A POVA not in-use need not have a current intermediate check, however, the POVA must be checked prior to use for an application that requires a calibrated POVA.

5.1.2.5.1.5 An intermediate check must be performed any time a POVA is serviced.

5.1.2.5.2 Initial set-up

5.1.2.5.2.1 The water used for the intermediate check process should be allowed to equilibrate at room temperature for at least two hours prior to the start of this procedure. Verify that the room and water temperature are the same prior to the start of this procedure.

5.1.2.5.2.2 Fill out identifying information on the top portion of POVA intermediate check worksheet.

5.1.2.5.2.3 For adjustable volume POVA, the volume of interest should be recorded.

5.1.2.5.2.4 Determine and record the water temperature on the logsheet at the beginning and at the end of determinations.

5.1.2.5.2.5 Place a volume of water in the weighing vessel, which completely covers the bottom of the container, and cap.

5.1.2.5.2.6 Place the weighing vessel on the balance and tare.

5.1.2.5.3 First Evaporation Loss (*e*) Check

5.1.2.5.3.1 Record the initial weight of the vessel and the time of recorded on the logsheet.

5.1.2.5.3.2 Remove the lid for approximately 20 seconds. This time should be adjusted to correspond to the approximate time interval between repetitions in 5.1.2.5.4.

5.1.2.5.3.3 Record a post-weight of the vessel and the time noted.

5.1.2.5.4 POVA Determinations

5.1.2.5.4.1 Use designated POVA, to dispense appropriate volume of temperature-equilibrated water into the weighing vessel and cap.

5.1.2.5.4.2 A minimum of ten individual repetitions ($W_i$), along with their corresponding time, should be recorded.

5.1.2.5.4.3 Calculate the Mean Delivered Weight ($\overline{W}$), record on logsheet.

5.1.2.5.5 Second Evaporation Loss (*e*) Check

5.1.2.5.5.1 Record the weight of the vessel and the time of recorded on the logsheet.

5.1.2.5.5.2 Remove the lid for approximately 20 seconds.

5.1.2.5.5.3 Record a post-weight of the vessel and the time recorded.

5.1.2.5.5.4 Calculate the mean evaporation weight, record on logsheet.

5.1.2.5.6 Mean Delivered Volume

5.1.2.5.6.1 From the table below, note the conversion factor (***Z***) for the mean water temperature. The conversion factor is based upon an air pressure of 1013 hPa.

| *Temperature °C* | *Conversion Factor (Z) (µL/mg)* |
|---|---|
| 20.0 | 1.0029 |
| 20.5 | 1.0030 |
| 21.0 | 1.0031 |
| 21.5 | 1.0032 |
| 22.0 | 1.0033 |
| 22.5 | 1.0034 |
| 23.0 | 1.0035 |
| 23.5 | 1.0036 |
| 24.0 | 1.0038 |
| 24.5 | 1.0039 |
| 25.0 | 1.0040 |
| 25.5 | 1.0041 |
| 26.0 | 1.0043 |
| 26.5 | 1.0044 |
| 27.0 | 1.0045 |
| 27.5 | 1.0047 |
| 28.0 | 1.0048 |



5.1.2.5.6.2 Calculate the Mean Volume Delivered ($V_t$) at the mean recorded temperature.

$$V_t = (\overline{W} + e) \cdot Z$$

5.1.2.5.7 Inaccuracy Calculation

5.1.2.5.7.1 Determine inaccuracy by calculating the percent error ($E_t$) between the expected ($V_o$) and calculated mean ($V_t$) volume.

$$E_t = V_t - V_o / V_o \times 100$$

5.1.2.5.7.2 Record % error on log sheet.

5.1.2.5.8 Imprecision Calculation

5.1.2.5.8.1 Calculate the standard deviation ($s$) for the replicate weights.

$$s = \frac{\sum (W_i - \overline{W})^2}{n - 1}$$

$n$ = Total number of repetitions

5.1.2.5.8.2 Record $s$ on worksheet.

5.1.2.5.8.3 Determine the imprecision by calculating the coefficient of variation (CV%). This is also referred to as relative standard deviation (RSD).

$$\mathbf{CV\%} = s \cdot 100 / \overline{W} + e$$

5.1.2.5.8.4 Record CV % on worksheet.

5.1.2.5.9 Evaluation of Accuracy and Precision
Refer to Analytical Method 5.1.1 for *Acceptance Criteria.*

**5.1.2.6 REFERENCES**


5.1.2.6.1 ASTM Method E-1154-89 (reapproved 2003), Standard Specification for Piston or Plunger Operated Volumetric Apparatus.

5.1.2.6.2 Curtis, R.H., *Performance Verification of Manual Action Pipets: Part I,* Am. Clin. Lab. 12(7):8-9; 1994.

5.1.2.6.3 Curtis, R.H., *Performance Verification of Manual Action Pipets: Part II,* Am. Clin. Lab. 12(9):16-17; 1994.

5.1.2.6.4 Byer, B.J., How to Use and Check Pipetting Equipment, Scientific Newsletters, Inc., 1977.

5.1.2.6.5 ISO 8655-6:2002, Piston-operated volumetric apparatus – Part 6: Gravimetric method for the determination of measurement error.

# *Revision History*

## Section Five
**Quality Assurance**

### 5.1 POVA Intermediate Check
### 5.1.2 Option Two: Gravimetric POVA Intermediate Check

| Revision # | Issue Date | History |
|---|---|---|
| 0 | 10/1997 | Original Issue |
| 1 | 11-27-2001 | Reworked/reformatted |
| 2 | 03-22-2005 | Quality requirements detailed and updated |
| 3 | 05-07-2007 | Updated QA measures and reformatting. |

Idaho State Police
Forensic Services
Toxicology Section

Section Five
Quality Assurance

## 5.2 Balance Calibration

### 5.2.1 GENERAL REQUIREMENTS

#### 5.2.1.1 SCOPE

The described requirements pertain when an analytical balance is used to prepare solutions and standards for application with toxicology procedures.

#### 5.2.1.2 BACKGROUND

5.2.1.2.1 All balances will be calibrated and serviced yearly by a qualified vendor.

5.2.1.2.2 Weights used for calibration will be calibrated yearly by a qualified vendor.

5.2.1.2.3 In-house calibration of balance by a criminalist or laboratory technician will be performed on a monthly basis. Calibration results to be recorded in logbook. Balances not in-use need not be calibrated, however, the balance must then be calibrated prior to use.

#### 5.2.1.3 EQUIPMENT

5.2.1.3.1 ANSI/ASTM Type I, Class 1 or Class 2 laboratory weights.

5.2.1.3.2 Manufacturer's manual for balance.

5.2.1.3.3 Top-Loading, Direct-Reading Laboratory Balance

#### 5.2.1.4 GENERAL PROCEDURES

5.2.1.4.1 Inspect balance pan, clean if necessary.

5.2.1.4.2 Inspect level bubble, level if necessary.

5.2.1.4.3 Tare balance with weighing paper.

5.2.1.4.4 Place ASTM weight on balance and record observed weight in balance log.

5.2.1.4.5 Place second weight on balance and record observed weight.

5.2.1.4.6 Inspect recorded weight values to determine if each value falls within the allowable range designated in

manual sections 5.2.2 and 5.2.3 for individual balances.

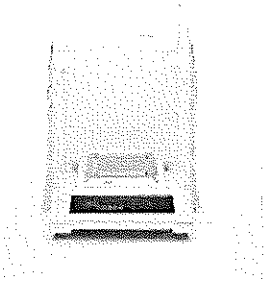
5.2.1.4.7 The calibration procedure should be repeated if the corrected value does not fall within the acceptable range. If the inaccuracy is still noted contact service vendor to set up a service call. A note should be placed on the balance to indicate that it is not in range and should not be used.

5.2.1.4.8 In the case of a significant departure in accuracy that is still within range, the calibration procedure should be repeated. If the inaccuracy is still noted, contact service vendor to set up a service call.

**5.2.1.4 REFERENCES**

5.2.1.4.1 ASTM Method E-617-97 **Standard Specification for Laboratory Weights and precision Mass Standards.**

## 5.2.2 METTLER TOLEDO AT400



### 5.2.2.1 SPECIFICATIONS

| | |
|---|---|
| ▪ Maximum capacity: | 405 g |
| ▪ Readability: | 0.1 mg |
| ▪ Taring range: | 0...405 g |
| ▪ Repeatability: | 0.05...0.15 mg |
| ▪ Linearity: | ± 0.5 mg |
| ▪ Stabilization time [typical]: | 4...6 s |
| ▪ Adjustment with internal weights: | fully automatic adjustment |
| ▪ Adjustment with external weights: | 400 g |
| ▪ Linearization: | automatic linearization with each internal adjustment |
| ▪ Sensitivity (temperature drift): | ± 1.5 ppm/°C |
| ▪ Sensitivity (long-term stability): | ± 0.00015 % |
| ▪ Size of weighing pan: | 80x80 mm |
| ▪ Free height above weighing pan: | 239 mm |
| ▪ Dimensions: | 241x433x289 mm |



### 5.2.2.2 SPECIFIC PROCEDURES for AT400

5.2.2.2.1 Daily (when-in-use) check

- Press menu button
- Display will indicate ***CAL Int.***
- To trigger adjustment hit set/zero bar
- The door will now close.
- When complete the display will indicate ***CAL end***. The balance will then beep and return to weighing mode.

5.2.2.2.2 Monthly check (when in use)

5.2.2.2.2.1 Record weights for the 100mg and 100g weights. Weights should be transferred with plastic tipped forceps.

5.2.2.2.2.2 Add or subtract correction factor for weight as determined by yearly

weight calibration certificates. Refer to top portion of balance log for values.

5.2.2.2.2.3 Record corrected weight on balance logsheet.

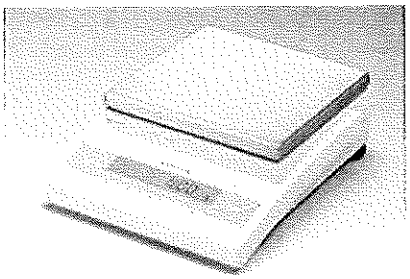
5.2.2.2.2.4 If the corrected value does not fall within the acceptable range or if there is a considerable loss in accuracy, refer to sections 5.2.1.4.6 and 5.2.1.4.7.

| *Weight* | *Tolerance* | *Acceptable Range* |
|---|---|---|
| 100mg (0.1g) | ±1.0mg (0.001g) | 99.0mg - 100.1mg |
| 100g | ±0.1g | 99.1g - 100.1g |

5.2.2.2.3 REFERENCES

5.2.2.2.3.1 Mettler Toledo AT Balance Operating Instructions

## 5.2.3 SARTORIUS Basic $^{plus}$ BP 3100S



### 5.2.3.1 SPECIFICATIONS

BasicPlus: design 0.01 g with rectangular weighing pan

- Weighing capacity 3100 g
- Readability 0.01 g
- Pan size 180x180 mm
- Response time 1.5 s
- Calibration manual - external weight
- Verification scale interval
- Verifiable for legal metrology

### 5.2.3.2 SPECIFIC PROCEDURES for BP3100S

5.2.3.2.1 Monthly calibration (when in use)

5.2.3.2.1.1 After tare is complete, press ***cal*** button. 2000 will be displayed on the screen.

5.2.3.2.1.2 Using cloth gloves, place 2kg weight on the center of the weighing pan.

5.2.3.2.1.3 When LED ***cal*** shuts off, calibration is complete.

5.2.3.2.1.4 Record weights for the 10g and 100g weights on balance logsheet. Weights should be transferred with cloth gloves.

| *Weight* | *Tolerance* | *Acceptable Range* |
|---|---|---|
| 10.00g | ±0.02g | 9.98g - 10.02g |
| 100.00g | ±0.2g | 99.8g - 100.2g |

5.2.3.2.1.5 If the corrected value does not fall within the acceptable range or if there is a considerable loss in accuracy, refer to sections 5.2.1.4.6 and 5.2.1.4.7.

**Idaho State Police**
**Forensic Services**
**Toxicology Section**

## History Page

**Toxicology Procedural Manual**
**Section Five - Quality Assurance**
**5.2 Balance Calibration**

| Revision # | Issue Date | History |
|---|---|---|
| 1 | 09-18-01 | Original Issue |



## Approval

**Technical Leader:** _____ **Date:** _____
S. C. Williamson

## Issuance

**QC Manager:** _____ **Date:** _____
Rick D. Groff

Idaho State Police
Forensic Services
Toxicology Section

## Section Five
Quality Assurance

### 5.3 Instrument Maintenance

#### 5.3.1 Maintenance Schedule for Toxicology Program Hewlett Packard 6890 Gas Chromatograph equipped with a 5973 Mass Selective Detector (GC/MSD)



**5.3.1.1 Scope**

This maintenance schedule applies to the GC/MSD used for the analysis of samples related to toxicology. Periodic and routine (as needed) maintenance requirements are described along with references to manufacturer provided manuals.

**5.3.1.2 Background**

It is considered an essential quality assurance component for a laboratory to monitor and document that all analytical instruments are maintained and operated properly. This document addresses routine and as needed maintenance for a gas chromatograph equipped with a mass selective detector (GC-MSD). Daily tuning of the instrument ensures that appropriate mass-to charge assignments and abundances of specific ions are established. The instrument tune and the system verification are valuable tools to detect performance problems. Performance is improved by performing periodic maintenance on the GC inlet and routine maintenance on the MSD.

**5.3.1.3 Equipment**

Replacement parts and cleaning supplies required for GC/MSD maintenance should be stocked to reduce the time that an instrument is off-line. Refer to the MSD Hardware Manual or the Agilent/HP Consumables and Accessories Catalog for items not listed below.

5.3.1.3.1 GC Inlet

- Gold plated seal [HP/Agilent #1874020885
- Merlin Microseal replacement septa [HP/Agilent # 5182-3444]
- Vespel-Graphite ferrules [HP/Agilent #5181-3323 or equivalent]
- Splitless Liner [HP/Agilent #5181-3316 or equivalent]
- Split Liner [HP/Agilent #5183-4647 or equivalent]

- O-rings for liner [HP/Agilent #5180-4182 or other appropriate liner]

5.3.1.3.2 MSD

- Vespel-Graphite ferrules [HP/Agilent # 5062-3508 or equivalent]
- Filaments [HP/Agilent # G1099-60053]
- Diffusion Pump Fluid [HP/Agilent # 6040-0809]
- Foreline Pump Oil [HP/Agilent # 6040-0834]
- Pellets for Edwards Rough Pump Trap [HP/Agilent # 8500-1233]
- PFTBA [HP/Agilent # 8500-0656]

5.3.1.3.3 Cleaning Supplies

- MSD cleaning kit tools [Refer to Hardware Manual]
- Sonicator
- Abrasive Cleaning Paper [HP/Agilent # 5061-5896]
- Alumina Powder [HP/Agilent # 8660-0791]
- Cotton-tipped applicators
- Assorted size beakers
- Methylene chloride
- Methanol
- Lint-free gloves [HP/Agilent # 8650-0030]
- Lint-free cloths [HP/Agilent # 05980-60051]
- Metal brush, equivalent to nominal 30 caliber handgun

5.3.1.3.4 Column test mix

- GC/MS Drug Standard Mix [Alltech 01463 or equivalent mixture which contains caffeine in addition to other commonly encountered analytes and/or drug metabolites]



**5.3.1.4 Maintenance**

5.3.1.4.1 When-in-use Maintenance

**5.3.1.4.1.1 Autotune Tune Requirements**

- Autotune should be performed before a sequence run is initiated.
- An optional *System Verification* can be run prior to a sequence run.

**5.3.1.4.1.2 Auto Tune Report**

The Tune Report is printed as a result of performing an Autotune on the MSD. The report includes the final values of the MS parameters which were set by the tuning process. The report includes the actual mass

spectral data obtained using these parameters. The analyst should inspect the Autotune report and make certain that the values are within acceptable ranges.

#### 5.3.1.4.1.3 System Verification Report

The system verification report accesses the data and indicates values that fall outside of acceptable ranges. This report will indicate "OK" for values that fall within limits. When a values falls outside the designated range, the report will indicate whether the value is low or high or above or below the range.

#### 5.3.1.4.1.4 Evaluation of Performance Indicators

The major indicators which are used to evaluate the performance of the MSD are peak width, mass peak assignments, relative abundance, isotope ratio, precursor check and leak detection. If a value on the Tune or System Verification Report falls outside of the acceptable range, troubleshooting procedures should be initiated.

| Indicator | Criteria | Hardware (HW) or Software (SW) Manual Reference | Tune Report | System Verification |
|---|---|---|---|---|
| Basepeak Determination | Basepeak should be 69 or 219 | | ☠ | Report |
| Peak Width *(Pw)* | • Peaks of the ions mass 69, 219 and 502 resulting from PFTBA.<br>• Peak widths should be 0.5±0.1amu.<br>• Range: 0.4 to 0.6amu | HW p.74<br>SW p.58 | ☠ | --- |
| Mass Peak Assignment | • *Autotune* Upper profile: Within ±0.2 amu of 69, 219 and 502.<br>• *Autotune* Lower Scan: Within ±0.1 amu of 69, 219 and 502.<br>• *System Verification* indicates the position of each mass. | HW p.74<br>SW p.58 | ☠ | ☠ |
| Relative Abundance | • Abundance of each tune mass reported as a percentage of the most abundant mass.<br>• Mass 69 at 100%<br>• Mass 219 at >40%<br>• Mass 502 at 2.4%<br>• A relative abundance value for mass 502 of ≤3% may indicate a dirty ion source | HW p.75<br>SW p.58 | ☠ | ☠ |
| Isotope Peak Position | • Isotope mass assignments should be 1 amu > the mass assignments of the respective parent peaks.<br>• System Verification evaluates: Positions of isotope mass 70, mass 220 and mass 502. | HW p.73 | --- | ☠ |

| Indicator | Criteria | Hardware (HW) or Software (SW) Manual Reference | Tune Report | System Verification |
|---|---|---|---|---|
| Isotope Ratio | ♦ Recommended ranges for isotope ratios are:<br>ϖ Mass 70 to mass 69 ☞ {0.5 to 1.6%}<br>ϖ Mass 220 to mass 219 ☞ {3.2 to 5.4%}<br>ϖ Mass 503 to mass 502 ☞ {7.9 to 12.3%}<br>ϖ Mass 219 to mass 69 ☞ {>40%}<br>ϖ Mass 502 to mass 69 ☞ {>2.4%}<br>♦ System Verification evaluates:<br>ϖ Ratio of mass 70 to mass 69, mass 220 to mass 219, mass 503 to mass 502, mass 219 to mass 69, and mass 502 to mass 69.<br>♦ Isotopes missing or having incorrect ratios may be indicative of a dirty ion source. | HW p.73<br>SW p.58 | ☠ | ☠ |
| Precursor Check | ♦ Evaluation of precursors to tune masses.<br>♦ Small precursors are not unusual<br>♦ Recommended values are:<br>ϖ Precursor to Mass 69 ☞≤3%<br>ϖ Precursor to Mass 219 ☞≤6%<br>ϖ Precursor to Mass 502 ☞≤12%<br>♦ High precursor values may be an indication of a dirty ion source. | HW p.74 | --- | ☠ |
| Leak Detection | ♦ Air leak can be detected by the presence of unusual amounts of nitrogen and water.<br>♦ Water Detection<br>ϖ Relative abundance of mass 18 to mass 69.<br>ϖ Allowable percentage is <20%.<br>♦ Nitrogen<br>ϖ Relative abundance of mass 28 to mass 69.<br>ϖ Allowable percentage is <10%. | HW p.68-72<br>SW p. 60 | ---* | ☠ |

*Perform an Air and Water Check. Refer to Software Manual p.46



### 5.3.1.5 Overview of Troubleshooting

In the advent that the MSD reports values for any of the above parameters which fall outside of the acceptable ranges, troubleshooting procedures will be initiated to determine the origin of the problem. Commonly encountered reasons for the MSD to not pass the tune criteria include:

| *Potential Source of Problem* | *Hardware Manual Reference* |
|---|---|
| Calibration vial is empty | p.120-123, 128, 212 |
| Excessive foreline or vacuum manifold pressure | p. 58, 77 |
| Dirty ion source | p. 69, 72-76 |
| Calibration valve is not working correctly | p. 74 |
| Bad signal cable connection | p. 69, 72 |
| Filament has failed or is not connected properly | p. 69,75 |
| Bad ion source wiring connection | p. 69,72 |
| Bad detector wiring connection | p. 69,72 |
| Failed electron multiplier horn | p. 69 |

For Additional trouble shooting information refer to volumes 1 through 3 of the 6890 operating manual and the hardware manual for the 5973.

### 5.3.1.6 As Needed Maintenance

| *Task* | *Indications* | *Manual Reference* |
|---|---|---|
| Replace ultra pure helium gas cylinder | At no lower than 100psi | --- |

### 5.3.1.6 As Needed Maintenance (continued)

| *Task - Record in Logbook* | *Indications* | *Manual Reference* |
|---|---|---|
| Replace inlet/injection port seal<br>Options:<br>1. Merlin Microseal™ Septum<br>2. Septa | ▪ Longer or shifting retention time<br>▪ Loss of response<br>▪ Noisy detector signal<br>▪ Autotune indicates an air leak | Operating Manual<br>Volume 2. Inlets<br>p. 18, 36-42 |
| Replace inlet/injection port liner and O-ring | ▪ Loss of response<br>▪ Visual Inspection | Operating Manual<br>Volume 2. Inlets<br>p. 19-20, 39-42, |
| Replace inlet/injection port base seal | ▪ Ghost peaks<br>▪ Visual Inspection | Operating Manual<br>Volume 2. Inlets p. 43-45 |
| Clean inlet reducing nut | ▪ When replacing inlet base seal and/or column | Operating Manual<br>Volume 2. Inlets p. 35,54-55 |
| Replace ion source filaments | ▪ Evaluation of Autotune | Hardware p. 160-163 |
| Clean ion source | ▪ Evaluation of Autotune | Hardware p. 68-76, p. 142-173 |
| Replace column | ▪ Evaluation of chromatography and Autotune | Operating Manual<br>Volume 2. Inlets p. 35 |
| Replace or clean split/splitless vent line trap | ▪ Evaluation of split ratio<br>▪ Clogged trap will not provide proper split ratio | Operating Manual<br>Volume 2. Inlets<br>General on split mode,<br>p. 123-142. |
| Lubricate seals-Side plate O-ring | ▪ Autotune indicates an air leak | Hardware p. 138-139 |

### 5.3.1.7 Weekly Maintenance

| *Task* | *Manual Reference* |
|---|---|
| Check foreline pump oil fluid level | Hardware p. 100-101 |

### 5.3.1.8 Monthly Maintenance

| *Task* | *Manual Reference* |
|---|---|
| <u>Column Performance Check</u><br>Analysis of Standard Drug/Metabolite Test Mix<br>▪ Compare chromatography for newly installed column with subsequent runs to monitor deterioration in column performance<br>▪ Evaluate detection, retention time, resolution, peak shape symmetry and signal abundance of the components of the standard mix<br>▪ Place print-out of TIC and individual scans in test-mix binder | ---- |

### 5.3.1.9 Semi-annual Maintenance

| *Task* | *Manual Reference* |
|---|---|
| <u>Pump Maintenance</u><br>▪ Drain and replace foreline pump fluid<br>▪ Remove and replace oil trap | <br>Hardware p. 102-105<br>Hardware p. 106-107 |
| Check PFTBA calibration vial. | Hardware p. 102-105 |

Idaho State Police
Forensic Services
Toxicology Section

## Section Five
Quality Assurance

### 5.3 Instrument Maintenance
#### 5.3.1 Maintenance Schedule for Toxicology Program Hewlett Packard 6890 Gas Chromatograph equipped with a 5973 Mass Selective Detector (GC/MSD)

| Revision # | Issue Date | History |
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| 1 | 11-27-01 | Original Issue in SOP format |



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Rick D. Groff

Idaho State Police
Forensic Services
Toxicology Section

## Section Five
Quality Assurance

### 5.3 Instrument Maintenance
### 5.3.2 Maintenance Schedule for Toxicology Program Perkin Elmer HS40XL Automatic Headspace Sampler and a AutoSystem XL Gas Chromatograph



**5.3.2.1 Scope**

Maintenance schedule applies to gas chromatograph/headspace analyzer GC/HSA used for the analysis of toxicology related samples. Instrument maintenance requirements are described along with references to manufacturer provided manuals.

**5.3.2.2 Background**

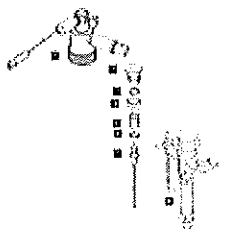
It is considered an essential quality assurance component that a forensic laboratory monitor and document that all analytical instruments involved in the analysis are maintained and operated properly.

**5.3.2.3 Equipment**

Replacement parts and cleaning supplies required for GC/HSA maintenance should be stocked to reduce the time that an instrument is off-line.

5.3.2.3.1 GC FID Detector

- Nozzle Replacement Tool for Auto-Ignite FID for PerkinElmer AutoSystem Series GC
  Part Number: N6103188
- Jet Replacement Tool for Auto-Ignite FID for PerkinElmer AutoSystem Series GC
  Part Number: N6101297
- GC Jet Assembly for Auto-Ignite Flame Ionization Detector (FID) for PerkinElmer AutoSystem XL GC
- Part Number: N6100361
- FID collector O-ring
  Part Number 09902143
- Collector Head Assembly for Auto-Ignite Flame Ionization Detector (FID) for PerkinElmer AutoSystem XL GC
  Part Number: N6100357
- Pipe cleaners
- Laboratory soap





History pg
Lists Rev 1

5.3.2.3.2 HSA

- 10 A Fuse
  Part Number 09991682
- 2mm Allen key
- Wide-Bore Stainless Steel
  Replacement Needle
  Part Number: B0131385
- Replacement Needle Seal Assembly,
  (without O-Rings)
  Part Number: B0500833
- Replacement O-Rings for PerkinElmer HS 40XL
  Needle Seal Assembly, Pkg. 10
  Part Number: B0198110
- O-ring tool
  Part Number: B0147449

**5.3.2.4 Procedure**

5.3.2.4.1 As Needed/Indicated Maintenance - Gas Chromatograph

| *Task* | *Indications* | *Manual Reference* |
|---|---|---|
| Replace Ultra Pure Helium Gas Cylinder | At no lower than 100psi | --- |
| **Gas Chromatograph** | | |
| Replace FID Jet | ▪ Failure to ignite<br>▪ No signal<br>▪ Noisy signal | AutoSystem XL GC<br>User's Guide p. 15-32 to 15-33 |
| Replace O-Ring In The FID Collector | ▪ Brittle or broken<br>▪ Noisy signal | AutoSystem XL GC<br>User's Guide p. 15-35 |
| Clean The FID Collector And Cap | ▪ Noisy signal | AutoSystem XL GC<br>User's Guide p. 15-36 |

## 5.3.2.4.2 As Needed/Indicated Maintenance - HS-40 Headspace Analyzer

| Headspace Analyzer | | |
|---|---|---|
| Replace HSA Fuse | ▪ Longer or shifting retention time<br>▪ Loss of response<br>▪ Noisy detector signal<br>Autotune indicates an air leak | HS 40 XL Headspace Sampler User's Guide p. 3-4 to 3-5 |
| Replace Sampling Needle | ▪ If damaged | HS 40 XL Headspace Sampler User's Guide p. 3-6 to 3-7 |
| Every 2500 injections a maintenance reminder is displayed. It will indicate the need to check the needle and o-ring seals. Press any key to acknowledge notice. | | |
| Replace Needle Seal | ▪ Check ~ every 2500 injections | HS 40 XL Headspace Sampler User's Guide p. 3-9 |
| Replace O-Ring Seals | ▪ Excessive carrier gas use<br>▪ May be required ~every 500 injections<br>▪ Upon inspection of needle seal, only O-ring may need to be replaced.<br>▪ Retention time shifts | HS 40 XL Headspace Sampler User's Guide p. 3-14 to 3-15 |
| Replace Upper Seal Assembly One O-ring | | HS 40 XL Headspace Sampler User's Guide p. 3-10 to 3-11 |
| Replace Lower Seal Assembly Two O-rings | | HS 40 XL Headspace Sampler User's Guide p. 3-12 to 3-13 |

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Quality Assurance

**5.3 Instrument Maintenance**

**5.3.2 Maintenance Schedule for Toxicology Program Perkin Elmer HS40XL Automatic Headspace Sampler and a AutoSystem XL Gas Chromatograph**

## Revision # Issue Date History

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Idaho State Police
Forensic Services
Toxicology Section

Section Five
Quality Assurance

## 5.4 Specimen Reanalysis Requests and Ancillary Concerns

**5.4.1 BACKGROUND**

In the event that an individual disputes the results of laboratory analysis, the sample will be forwarded to a reputable outside laboratory for reanalysis.

**5.4.2 REQUEST PROCESS**

5.4.2.1 The request for reanalysis must originate from an authorized party. Authorized parties include, but are not limited to, the judge over-seeing the case, the subject's attorney, or the subject, if they elect to represent themselves. The authorized party must work through the prosecutor's office involved with the case in question to file the request.

5.4.2.2 The party requesting reanalysis of the urine specimen should contact a reputable laboratory and establish an account. The laboratory should be advised that the specimen is forensic in origin and should have all chain-of-custody considerations applied. Many laboratories analyze both clinical and forensic specimens; forensic specimens are subjected to more rigorous tracking.

5.4.2.3 The selected laboratory should provide ISP-Forensic Services with necessary information and tracking paperwork (bar code sticker, etc) required by their laboratory.

5.4.2.4 On receipt of the necessary forwarding information, Forensic Services will forward the urine specimen in a cooler box to ensure specimen content integrity.

**5.4.3 INTERPRETATION OF REANALYSIS DATA**

5.4.3.1 When interpreting the data obtained from the reanalysis of a sample it should be kept in mind that drug compounds are not detectable in a urine specimen indefinitely.

5.4.3.2 In the case of initial drug concentration that is weak, after several months of storage, the specimen may no longer test positive.

**5.4.4 ALTERNATIVE TO REANALYSIS**

5.4.4.1 The data obtained for the specimen in question is available for an outside expert in toxicology to scrutinize.

5.4.4.2 Forensic Services files for each specimen include associated standards and quality controls.

**5.4.5 FORENSIC SERVICES ANALYSIS INFORMATION**

5.4.5.1 Specimens are analyzed by two or three independent methods with different aliquots of the specimen used for each test.

5.4.5.2 Preliminary screening tests utilized are enzyme immunoassay (EIA), gas chromatography with a nitrogen phosphorus detector (GC-NPD) and thin layer chromatography (TLC).

5.4.5.3 Confirmatory testing is accomplished with a gas chromatography with a mass selective detector (GC-MSD).

5.4.5.4 A negative finding does not indicate that no drug was present, only that it was not detected by our testing protocols.

5.4.5.5 A positive result indicates that the subject has been exposed to a detected drug compound. Due to the variable pharmacological factors which effect its detection time (drug dose, route of administration, and rates of metabolism and excretion), determination of time since use and the level of current impairment can not be established solely on the basis of the results of the toxicological testing.

5.4.5.6 For DUI, the results of toxicological testing should be considered along with the outcome of field sobriety tests or a Drug Recognition Examination (DRE) and other circumstances surrounding the incident.

5.4.5.7 The outside laboratory selected for reanalysis should employ the GC/MSD for drug compound verification procedures.

5.4.5.8 Forensic Services analysts are not advocates for either the prosecution or the defense. Our mission is to provide impartial scientific analysis of high quality to the criminal justice system.

# Idaho State Police
# Forensic Services
# Toxicology Section

## History Page

**Toxicology Procedural Manual**
**Section Five- Urine Toxicology**
**5.4 Specimen Reanalysis Requests and Ancillary Concerns**

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Forensic Services
Toxicology Section

Section Five
Quality Assurance

## 5.5 Urine Toxicology Specimen Retention

### 5.5.1 BACKGROUND

Upon completion of analysis, urine specimens are retained by Forensic Services. To delay the degradation of drug compounds in urine, specimens are stored in secure evidence freezers at approximately minus 20 degrees celsius. The stability of drug positive urine specimens stored at -20°C in a stable environment is superior to storage under refrigeration or at room temperature. Stability becomes an issue in regards to specimen reanalysis considerations (Reference section 5.4).

### 5.5.2 POSITIVE URINE SAMPLES

5.4.2.1 Retention Time

Upon completion of analysis Forensic Services will freeze the urine specimen and store it for ≥ ninety (90) days from the date of report, at which time the sample will be destroyed.

5.4.2.2 Request for Extended Storage

When the circumstances of a case make it necessary for a specimen to be retained by Forensic Services for longer than 90 days or returned to the submitting party, the request must be made in writing to Forensic Services. Forensic Services does not take responsibility for the integrity of samples returned to an agency and reanalyzed. Refer to section 5.4 for specimen reanalysis request protocol.

### 5.5.3 NEGATIVE URINE SAMPLES

5.4.2.1 Retention Time

Upon completion of analysis Forensic Services will freeze the urine specimen and store it for ≥ thirty (30) days from the date of report, at which time the sample will be destroyed.

**Idaho State Police**
**Forensic Services**
**Toxicology Section**

## History Page

**Toxicology Procedural Manual**
**Section Five- Quality Assurance**
**5.5 Urine Toxicology Sample Retention**

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Idaho State Police
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Toxicology Discipline

Section Five
Quality Assurance

## 5.7 Toxicology Competency and Proficiency Tests

### 5.7.1 BACKGROUND

In accordance with the *Toxicology Training Plan,* upon the completion of training, the trainee will complete a competency test consisting of ≥six (6) specimens, which contain representative commonly encountered analytes.

### 5.7.2 SCOPE

As set forth in the Idaho State Police Forensic Services *Procedure and Quality Manual,* proficiency testing is an integral part of a quality program. This procedure describes the criteria to be applied to the evaluation of results obtained for competency and proficiency testing for the toxicology discipline.

### 5.7.3 PROCEDURE

5.7.3.1 Ethanol and Other Volatiles Analysis

5.7.3.1.1 **Competency Tests**

5.7.3.1.1.1 The competency test can be ordered through a reliable vendor.

5.7.3.1.1.2 The acceptable alcohol concentration range is determined from the target value provided by the manufacturer of the competency test.

5.7.3.1.1.3 Reported values must fall within ±10% of the target value reported by manufacturer.

5.7.3.1.1.4 If all volatiles are not detected and/or the quantitative ethanol value(s) reported does not fall within the allowable range, analysis procedures will be reviewed and additional training may be required as deemed appropriate by the Toxicology Program Discipline Leader. The analyst will be required to perform an additional competency test.

5.7.3.1.2 **Proficiency Tests**

5.7.3.1.2.1 The blood alcohol proficiency test can be ordered through an ASCLD/LAB approved vendor and/or Department of Transportation (DOT).

5.7.3.1.2.2 To comply with ASCLD/LAB proficiency test requirements it is necessary for each laboratory to successfully complete one external test from an ASCLD/LAB approved provider.

5.7.3.1.2.3 As described in section 5.6 of this manual, in order to comply with IDAPA 11.03.01 (approval to perform alcohol determinations for legal purposes), a laboratory must take part in an Idaho State Police Forensic Services (ISP-FS) recognized proficiency testing program.

5.7.3.1.2.4 A single proficiency test can be used to comply with both IDAPA and ASCLD/LAB requirements as long as it is an ASCLD/LAB approved test.

5.7.3.1.2.5 The appropriate tests to be ordered will be evaluated yearly by the Quality Assurance Manager with input from the Toxicology Discipline Leader.

5.7.3.1.2.6 The acceptable alcohol concentration range is determined from the target value provided by the manufacturer of the competency test.

5.7.3.1.2.7 Reported values must fall within 2 standard deviations of the target value compiled by manufacturer after evaluation of all submitted results.

5.7.3.1.2.8 If the value reported does not fall within the allowable range, analysis procedures will be reviewed and additional training may be required as deemed appropriate by the Toxicology Program Discipline Leader. The analyst will be required to perform a competency test prior to resuming casework.

5.7.3.2 Urine and Blood Drug Analysis

5.7.3.2.1 **Competency Testing**

5.7.3.2.1.1 The competency test will be designed to verify that an analyst has a through working knowledge of appropriate analytical method(s).

5.7.3.2.1.2 The competency test can be prepared by the Toxicology Program Discipline Leader, their designee, or ordered through a reliable vendor.

5.7.3.2.1.3 When required, quantitative values must agree with manufacturer or discipline leader determined values ±20% of target.

5.7.3.2.1.4 If the analyst does not correctly identify all target analytes and/or quantitative values do not fall within range, the analyst's training will be reviewed and additional training may be required as deemed appropriate by the Toxicology Program Discipline Leader. The analyst will be required to complete additional competency test samples. The number of samples will be determined by the nature of the discrepancy.

5.7.3.2.2 **Proficiency Testing**

5.7.3.2.2.1 Only analytes that are tested for with current ISP-FS analytical methods and approach to analysis will be evaluated.

5.7.3.2.2.2 When reported, quantitative values must agree with manufacturer determined values within ±20%.

5.7.3.2.2.3 If the analyst does not correctly identify all target analytes and/or quantitative values do not fall within range, the analyst's training will be reviewed and additional training may be required as deemed appropriate by the Toxicology Program Discipline Leader. The analyst will be required to complete a competency test prior to resuming casework. The number of samples will be determined by the nature of the discrepancy.

# Idaho State Police
# Forensic Services
# Toxicology Discipline

## History Page

**Toxicology Analytical Method Manual**
**Section Five**
**Quality Assurance**
**5.7 Review of Toxicology Proficiency and Competency Tests**

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| 1 | 05-24-2006 | Update, additional test providers added |



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Idaho State Police
Forensic Services
Toxicology Discipline

Section Five
Quality Assurance

## 5.8 Quality Assurance Measures - Urine and Blood Toxicology

### 5.8.1 BACKGROUND

The quality assurance measures applied towards analysis promote confidence in results. This analytical method was created so that the shared requirements did not have to be included in every toxicology discipline analytical method.

### 5.8.2 SCOPE

This analytical method addresses general acceptance requirements for qualitative and quantitative analysis data obtained through analysis by gas chromatography equipped with a nitrogen phosphorus or a mass selective detective. Requirements for analysis with other instrumentation are addressed in relevant analytical methods.



### 5.8.3 EQUIPMENT AND SUPPLIES

5.8.3.1 Tube Rocker (Fisher Scientific or equivalent)

5.8.3.2 Laboratory Centrifuge (Fisher Marathon or equivalent)

5.8.3.3 Waterbath (Fisher or equivalent)

5.8.3.4 Drybath (Fisher or equivalent)

5.8.3.5 Evaporative Concentrator (Zymark TurboVap or equivalent) equipped with nitrogen tank.

5.8.3.6 Glassware

Refer to appropriate analytical method for extraction glassware.
GC/MS ALS vials (HP 5182-0865 or equivalent)
GC/MS vial microinsert (HP 5183-2088 or equivalent)

5.8.3.7 Gas Chromatograph equipped with a mass selective detector (HP 6890/5973 or comparable) and a HP-5MS Ultra low bleed (5%-Diphenyl-95%-Dimethylsiloxane co-polymer) capillary column (25M) or comparable.

### 5.8.4 REAGENTS

5.8.4.1 Refer to appropriate analytical method and manual sections 2.6 and 3.8 for solution preparation instructions.

### 5.8.5 ANALYSIS QUALITY ASSURANCE

5.8.5.1 Qualitative Analysis

5.8.5.1.1 Non-extracted Standards (NES)

5.8.5.1.1.1 Standards must be prepared and analyzed as designated in appropriate analytical method.

5.8.5.1.1.2 Acquired data should be comparable to authentication data. No significant differences in the MS data should be apparent.

5.8.5.1.2 Matrix Controls

5.8.5.1.2.1 Controls should be prepared and analyzed as designated in the appropriate analytical method.

5.8.5.1.2.2 Positive controls should exhibit proper retention time and mass spectral characteristics for compounds of interest.

5.8.5.1.2.3 Negative controls should be examined for compound(s) of interest and interfering substances.

5.8.5.1.3 Solvent Blanks

5.8.5.1.3.1 An appropriate solvent blank should be run between sample extracts.

5.8.5.1.3.2 If the solvent blank contains a reportable analyte of interest, the corrected area of the analyte peak must be a minimum of 10 times stronger than the corresponding peak in the blank preceding it. Ideally, no contamination should be apparent.

5.8.5.1.3.3 Reportable is defined as a complete fragmentation pattern at the appropriate retention time. Analytes of interest include, but are not limited to, analytes routinely reported.

5.8.5.1.3.4 If significant contamination is present, as discussed in 5.8.5.1.3.2, evaluate the analysis of a newly obtained solvent blank and the sample extract in question. If the contamination is still apparent,

troubleshoot the instrument to determine the source of contamination. In addition, the sample in question should be reextracted prior to reanalysis on rectified instrument.

5.8.5.2 Quantitative Analysis

Quality measures are optimized for the analytes in question and are addressed in each individual quantitative analytical method.

5.8.5.3 Distribution of Quality Data

5.8.5.3.1 Originals of casework standards and matrix controls will be stored in a designated central location in the laboratory where the analysis was performed.

5.8.5.3.2 Copies of all quality assurance control data need not be placed in each case file except those required under 5.8.5.3.3.

5.8.5.3.3 Copies of analytical standards used to substantiate the identification of each drug compound must be included in each case file if not otherwise indicated in the relevant analytical method.



## 5.8.6 REFERENCES

5.8.6.1 Wu Chen, N.B. Cody, J.T., Garriott, J.C., Foltz, R.L., et al., *Report of the Ad Hoc Committee on Forensic GC/MS: Recommended guidelines for forensic GC/MS procedures in toxicology laboratory associated with offices of medical examiners and/or coroners*, J. Foren. Sci, 236 (35): 236-242, 1990.

5.8.6.2 Goldberger, B.A., Huestis, M.A., Wilkins, D.G., *Commonly practiced quality control and quality assurance procedures for gas chromatograph/mass spectrometry analysis in forensic urine drug-testing laboratories*, For Sci Review, 9(2): 60-79, 1997.

5.8.6.3 SOFT/AAFS Forensic Toxicology Laboratory Guidelines, 2002

# Idaho State Police
# Forensic Services
# Toxicology Discipline

## History Page

**Toxicology Analytical Method Manual**
**Section Five**
**Quality Assurance**
**5.8 Evaluation of Quality Assurance Measures**

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| 0 | 10-18-2002 | Original Issue |
| 1 | 04-16-2003 | Clarifications, Updated. |
| 2 | 07-23-2003 | Clarification of authentication process. |
| 3 | 03-09-2005 | Reformatted, scope broadened. |
| 4 | 05-24-2006 | Clarifications, authentication process moved to Analytical Method 5.10. |



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Richard D. Groff

Idaho State Police
Forensic Services
Toxicology Discipline

Section Five
Quality Assurance

## 5.9 Testing Guidelines

### 5.9.1 BACKGROUND

To best utilize the resources available to support the ISP-FS toxicology discipline, the degree of analysis pursued should be dictated by all available information. It may not always be necessary and/or appropriate to confirm all drug compounds present. When a subject is using prescription drugs, the charge may be justified solely by drugs-of-abuse, the prescription drug may not be a consideration. With drugs of abuse, confirming the presence of additional drugs may not enhance the charge.



### 5.9.2 SCOPE

This SOP addresses the factors to consider when determining the extent of analysis a toxicology case sample requires. It is intended to provide guidance to analysts, however, the decision to pursue testing remains at the discretion of each analyst. The goal of these considerations is an efficient utilization of resources in order to provide timely analysis results to user agencies.

### 5.9.3 PROCEDURE

5.9.3.1 When available, the type of case associated with a toxicology sample should be determined.

5.9.3.2 The extent of analysis should be based on background information and the charges pending.

5.9.3.3 If no background information is provided; it is at the discretion of the analyst to perform only basic testing.

5.9.3.4 Blood and Urine samples submitted for determination of drugs of abuse and other impairing substances should be tested up to the point of justifying the charge in question. The extent of testing is at the discretion of each analyst; however, the following situations and examples should be factored into the evaluation process.

## 5.9.4 ETHANOL/VOLATILES DETERMINATION

5.9.4.1 When the ethanol concentration exceeds 0.12g/100cc, further testing for additional drugs, in either blood or urine, should not be pursued unless extenuating circumstances are involved.

5.9.4.2 Extenuating circumstances could include the following:

- Fatality or injury accident where additional drug use is suspected.
- Drug Recognition Exam (DRE) supports additional drug use. The DRE officer is reliant on a confirmation of their observations to maintain their certification.
- Drug related charges stemming from controlled substance and/or paraphernalia recovered from vehicle. Analysis of blood and/or urine would serve to support a possession charge.

5.9.4.3 The submitting officer or agency is responsible for providing justification for additional testing. Justification could take the form of a memo or letter outlining the situation and/or a case report.

5.9.4.4 If the ethanol concentration is 0.12 or lower, future testing for other impairing drugs will not be pursued if the additional testing is not requested on the *Toxicology Evidence Submittal Form.*

## 5.9.5 OTHER DRUG TESTING GUIDELINES

5.9.5.1 Enzyme Immunoassay Positive for Prescription Drugs

5.9.5.1.1 When current prescription drug therapy has the ability to trigger a positive enzyme immunoassay (EIA) response, the presence does not have to be confirmed in all situations.

5.9.5.1.2 If the drug in question is recovered in the extraction procedure for another compound, it may be confirmed provided quality assurance requirements are met.

5.9.5.1.3 **Example One**

Positive enzyme immunoassay (EIA) screen result for methamphetamine and benzodiazepines is indicated. The sample is collected as the result of a suspected DUID. The submittal form indicates symptoms consistent with stimulant use and lists diazepam as current drug therapy. When the methamphetamine confirmation data is processed, nordiazepam is present. The qualitative presence of nordiazepam may be confirmed in this sample. If no benzodiazepine had been present in the extraction to recover methamphetamine, no additional

testing has to be pursued for a benzodiazepines class drug.

5.9.5.1.4 **Example Two**

A sample indicates a positive enzyme immunoassay (EIA) benzodiazepines screen. The case is a probation violation. The submittal form lists diazepam as current drug therapy. In this situation, no additional testing should be pursued for a benzodiazepines class drug.

5.9.5.1.5 **Qualifying Statements**

In the above examples, if no analysis for the benzodiazepine is pursued, a qualifying statement should be placed on the analysis report.

**Preliminary testing indicates the presence of a _____ class compound. Confirmatory testing was not pursued because the _____**
***(state drug class)***

**_____ is said to be a part of current**
***(state specific drug compound)***
**prescription drug therapy.**

5.9.5.2 Prescription Drugs Not Covered by EIA Screen

5.9.5.2.1 When a prescription drug compound is detected in a general extraction procedure, the confirmation of its presence is not required if other drugs present serve to justify the charge. This exception applies whether or not the compound is included in current prescription drug therapy.

5.9.5.2.2 **Example One**

Positive enzyme immunoassay (EIA) screen results for methamphetamine and opiates. The sample is collected as the result of a suspected DUID. The submittal form indicates symptoms consistent with stimulant and narcotic analgesic use. Effexor (venlafaxine) is listed as current drug therapy. When the methamphetamine confirmation data is processed, venlafaxine is present. It is at the discretion of an analyst of whether or not to run a venlafaxine standard and confirm its presence.

5.9.5.3 Enzyme Immunoassay Positive for Several Drugs-of-Abuse

5.9.5.3.1 When positive EIA screen results are indicated for several drugs of abuse, all involved drug compounds need not be confirmed.

5.9.5.3.2 **Example Three**
EIA screen is positive for amphetamine, methamphetamine, opiates, and cocaine metabolite. Initial confirmatory analysis indicates the presence of amphetamine, methamphetamine, codeine, morphine and 6-monoacetylmorphine. No cocaine or ecgonine methyl ester is detected. After consideration of all available information, it is at the discretion of the analyst of whether or not to pursue the qualitative confirmation of benzoylecgonine.

5.9.5.4 Confirmation of Metabolites When Parent Drug is Detected

5.9.5.2.1 For qualitative analysis, when a parent drug compound is detected, the confirmation of the presence of associated metabolites is recommended but not required.

5.9.5.2.2 **Example One**
General basic extraction indicates the presence of propoxyphene. The confirmation of the presence of norpropoxyphene is at the discretion of the analyst.

# Idaho State Police
# Forensic Services
# Toxicology Section

## History Page

**Toxicology Procedural Manual**
**Section Five**
**Quality Assurance**
**5.9 Testing Guidelines**

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Toxicology Discipline

Section Five
Quality Assurance

## 5.10 Authentication of Reference Materials - Urine and Blood Toxicology

### 5.10.1 BACKGROUND

The quality assurance measures applied towards analysis promote confidence in results. This analytical method was created so that the shared requirements did not have to be included in every toxicology discipline analytical method.

### 5.10.2 SCOPE

This analytical method addresses qualitative and quantitative authentication of reference material used to verify analysis performance by thin layer chromatography (TLC) and gas chromatography with a nitrogen phosphorus or mass selective detective. Reference materials include both analytical reference standards and commercially obtained matrix controls.



### 5.10.3 EQUIPMENT AND SUPPLIES

5.10.3.1 Tube Rocker (Fisher Scientific or equivalent)

5.10.3.2 Laboratory Centrifuge (Fisher Marathon or equivalent)

5.10.3.3 Waterbath (Fisher or equivalent)

5.10.3.4 Drybath (Fisher or equivalent)

5.10.3.5 Evaporative Concentrator (Zymark TurboVap or equivalent) equipped with nitrogen tank.

5.10.3.6 Glassware

Refer to appropriate analytical method for extraction glassware.
GC/MS ALS vials (HP 5182-0865 or equivalent)
GC/MS vial microinsert (HP 5183-2088 or equivalent)

5.10.3.7 Gas Chromatograph equipped with a mass selective detector (HP 6890/5973 or comparable) and a HP-5MS Ultra low bleed (5%-Diphenyl-95%-Dimethylsiloxane co-polymer) capillary column (25M) or comparable.

### 5.10.4 REAGENTS

5.10.4.1 Refer to appropriate analytical method for solution preparation instructions.

## 5.10.5 REFERENCE MATERIAL AUTHENTICATION

5.10.5.1 General

5.10.5.1.1 Appropriate authentication should be documented for reference materials prior to official use.

5.10.5.1.2 Reference materials applied for qualitative purposes must have their chemical identity well established.

5.10.5.1.3 The manufacturer of reference standards used for quantitative purposes should utilize balances calibrated with weights traceable to National Institute of Standards and Technology (NIST) standards. The certificate of analysis should be consulted to verify compliance with this requirement.

5.10.5.1.4 When a standard or control contains more than one constituent, only the compound(s) of interest need be authenticated.

5.10.5.1.5 When possible, reference materials used for quantitation should be analyzed against existing calibration standards.

5.10.5.1.6 Whenever possible, source of reference standard used to prepare matrix controls should differ from that used to prepare calibration standards. If different vendors are not available, the standards and controls should be prepared separately.

5.10.5.1.7 For qualitative authentication, evaluate a single GC-MSD analysis obtained in full scan mode.

5.10.5.1.8 For quantitative authentication, a minimum of three determinations, in a single analysis run, should be evaluated.

5.10.5.1.9 For quantitative determinations, utilize analytical method and GC-MSD conditions optimized for the analyte under evaluation.

5.10.5.1.10 Whenever possible, the GC-MSD data should be compared to a previous lot of reference material.

5.10.5.1.11 The mean quantitative concentration should fall within 20% of the target value listed on *Certificate of Analysis* for standards or *Package Insert* for matrix controls. The precision between replicates should be ≤ 5%.

5.10.5.1.12 Certificate of Analysis (COA) for all standards and package inserts for commercially obtained matrix controls, will be stored centrally in the laboratory performing the authentication.

5.10.5.2 Reference Standard Authentication

5.10.5.2.1 Reference standards are used for both qualitative and quantitative purposes.

5.10.5.2.2 Whenever possible, qualitative authentication is accomplished comparing the instrumental data obtained through the instrumental analysis of the new standard with data from a peer reviewed scientific journal, reference standard compendium, instrumental data and/or library searches in conjunction with the data provided on the COA, when available.

5.10.5.2.2.1 Comparison must result in no significant differences.

5.10.5.2.3 When comparison to a journal, compendium or other document, is not an option, mass spectral interpretation may be used in conjunction with the COA. This would apply in cases where instrumental data for a drug metabolite is not yet published but a structurally similar compound is available to assist with interpretation.

5.10.5.2.4 For the quantitative authentication of the concentration of a component of a reference standard, evaluate gas chromatography-mass spectrometry (GC-MS) data in conjunction with the certificate of analysis (COA) provided by the manufacturer.

5.10.5.2.5 Deuterated internal standards may also be evaluated in SIM mode prior to use.

5.10.5.3 Matrix Control Authentication

5.10.5.3.1 Matrix controls are analyzed in parallel with casework samples to demonstrate that a procedure performed as intended.

5.10.5.3.2 Matrix controls serve to validate a calibration curve.

5.10.5.3.3 Matrix controls may be prepared with authenticated reference standards and appropriate matrix or obtained through a vendor.

5.10.5.3.4 When possible, the qualitative identity of component(s) in a commercially obtained matrix control should be based on the package insert or certificate of analysis. If the analyst is unfamiliar with the MS of the component, reference materials should be consulted as described in 5.10.5.2.2.

5.10.5.3.5 The matrix control should be extracted as described in the appropriate analytical method. For controls containing a mixture of analytes such as Toxi-Control 19, a reference mixture can be prepared from authenticated drug standards.

5.10.5.3.6 To authentication the qualitative presence of components when the manufacturer does not provide a certificate of analysis or package insert and an another lot of the control is in use, a new lot of a commercially obtained matrix control must be analyzed against the existing lot.

5.10.5.3.7 To authenticate the concentration of a component of a commercially obtained matrix control for quantitative applications, evaluate gas chromatography-mass spectrometry (GC-MS) data in conjunction with the package insert provided by the manufacturer.

5.10.5.3.8 Controls in use prior to the issue date of this analytical method revision can be used until consumed.

5.10.5.4 Authentication Documentation

5.10.5.4.1 A coversheet providing the information necessary for authentication will be prepared and placed with the MSD data. The coversheet for qualitative validation should, at a minimum, list the lot number, vendor, date of analysis, analyst, and mode of authentication. For quantitative authentication, the coversheet should include an evaluation of quantitative data.

5.10.5.4.2 Coversheet and GC-MSD data should be initialed and stored centrally in a designated location.

5.10.5.4.3 The stock container for the standard or control may be designated as "authenticated" after the authenticity of the standard has been validated.

5.10.5.4.4 Unopened stock reference material must be stored in a designated area until official use.

5.10.5.4.5 It is the responsibility of each analyst to verify that each standard or control used has been properly authenticated.



## 5.10.6 REFERENCES

5.10.6.1 Wu Chen, N.B. Cody, J.T., Garriott, J.C., Foltz, R.L., et al., *Report of the Ad Hoc Committee on Forensic GC/MS: Recommended guidelines for forensic GC/MS procedures in toxicology laboratory associated with offices of medical examiners and/or coroners*, J. Foren. Sci, 236 (35): 236-242, 1990.

5.10.6.2 Goldberger, B.A., Huestis, M.A., Wilkins, D.G., *Commonly practiced quality control and quality assurance procedures for gas chromatograph/mass spectrometry analysis in forensic urine drug-testing laboratories*, For Sci Review, 9(2): 60-79, 1997.

5.10.6.3 SOFT/AAFS Forensic Toxicology Laboratory Guidelines, 2002.

# Idaho State Police
# Forensic Services
# Toxicology Discipline

## History Page

**Toxicology Analytical Methods Manual**
**Section Five**
**Quality Assurance**
**5.10 Reference Material Quality Assurance**

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## Approval

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