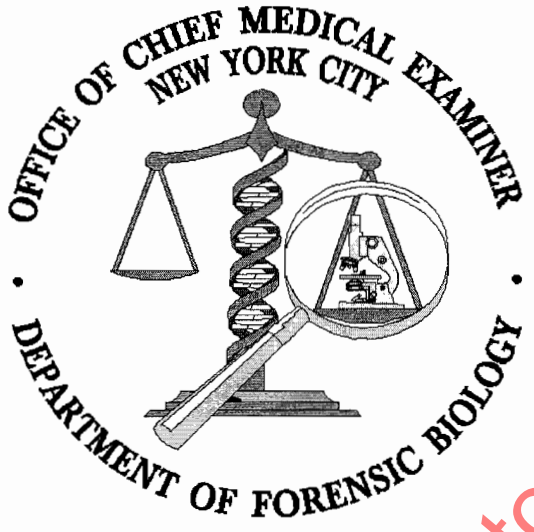
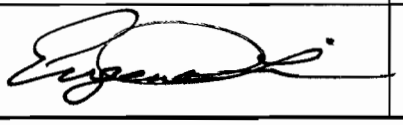


QUALITY ASSURANCE MANUAL
VERSION 5.0

Effective date: July 14, 2008

REVIEWED/APPROVED BY			
Title	**Print Name**	**Signature**	**Date**
Technical Leader, Nuclear DNA Operations	**Eugene Y. Lien**		**July 11, 2008**

1. INTRODUCTION

As of this date, the Quality Assurance Manual, Version 4.0 supersedes all previous Quality Assurance (QA) and/or Quality Control (QC) Manuals in the Department of Forensic Biology at the New York City Office Of Chief Medical Examiner (OCME). Where appropriate, references have been made to the Forensic Biology Administrative Manual, Case Management Manual, Forensic Biochemistry Methods Manual, and Protocols for Forensic STR Analysis.

The Quality Assurance Manual consists of various sections that address the current FBI Quality Assurance Standards and the ASCLD/LAB Manual. Its appendices contain reagent sheets (Appendix A), Quality Control procedures (Appendix B), and a list of usage and maintenance logs (Appendix C) that are currently being used in the laboratory.

A. Section 1 through Section 7

These sections address the current FBI Quality Assurance and ASCLD/LAB Standards and specify the policies and procedures followed by the Department of Forensic Biology. These sections are controlled and must be approved by the Director or his/her designee prior to being implemented and/or changed.

B. Reagent sheets (Appendix A)

The Department of Forensic Biology documents the preparation of all internal critical reagents. This documentation is in the form of a reagent sheet that lists the chemical makeup and procedures necessary for the preparation of a given reagent. All current reagent sheets are filed in a series of **Reagent Sheet Binders**. A copy of each reagent sheet has also been included in this manual as Appendix A. Reagent sheets are worksheets, and do not require the Director or his/her designee's approval prior to being implemented and/or changed, but must be reviewed by the Quality Assurance Manager.

C. Quality Control Testing Procedures (Appendix B)

The purpose of a Quality Assurance Program is to ensure that the laboratory meets a specified standard of quality. The Quality Assurance Program does this through the monitoring, verifying, and documenting of the performance of the laboratory. To accomplish these tasks, the Forensic Biology Quality Assurance Program has established a series of Quality Control Testing Procedures that are designed to monitor critical aspects of forensic sample analysis in order to ensure that the resulting product conforms to the current standards set forth by the ASCLD/LAB Manual, FBI Quality Assurance Standards, and Scientific Working Group for DNA Analysis Methods (SWGDAM). These Quality Control Testing Procedures are contained in Appendix B and are identified by specific QC numbers. As an appendix, Quality Control Testing Procedures do not require the Director or his/her designee's approval prior to being implemented and/or changed, but must be reviewed by the Quality Assurance Manager.

D. Miscellaneous (Appendix C)

Appendix C lists miscellaneous items associated with the Quality Assurance Program of the laboratory. As an appendix, they do not require the Director or his/her designee's approval prior to being implemented and/or changed, but must be reviewed by the Quality Assurance Manager.

Appendix C-1 lists the usage and maintenance logs used by the laboratory to provide documentation of equipment use, calibration, and maintenance. This documentation aids the QA program in identifying trends in equipment operation and analyst performance. This information can also assist the QA program in identifying potential or existing problems of quality.

Appendix C-2 shows a list of quality control testing "procedures" used in the Department of Forensic Biology. Each procedure may be a combination of several quality control tests listed in Appendix B. If a reagent sheet lists a "procedure" for its quality control, then the reagent must pass all the quality control tests listed. If it lists a specific "QC" number, then the reagent must pass that quality control tests only.

2. ASCLD/LAB MANUAL		
DATE EFFECTIVE 07-14-2008	VERSION 5.0	PAGE 1 OF 3

In accordance to the Quality Assurance Manual guidelines (See Discussion of Standard 1.4.2.1) set forth by the American Society of Crime Laboratory Directors/Laboratory Accreditation Board (ASCLD/LAB), this manual contains or references the documents or policies/procedures pertaining to the following:

- **A quality policy statement including objectives and commitments by management.**
  This is listed in the Forensic Biology Administrative Manual.

- **The organization and management structure of the laboratory, its place in any parent organization, and relevant organizational charts.**
  This is diagrammed and discussed in the Forensic Biology Administrative Manual.

- **The relationships and responsibilities of management, technical operations, and support services in implementing the quality system.**
  This is presented in the Forensic Biology Administrative Manual.

- **Job descriptions, education, and up-to-date training records of laboratory staff.**
  Job descriptions for all laboratory personnel are described in the Forensic Biology Administrative Manual. In addition, Civil Service job specifications for each job title are located in a filing cabinet containing ASCLD/LAB and FBI QAS criterion files. Training records of laboratory staff are kept in a filing cabinet located near the departmental administrative office.

- **Control and maintenance of documentation of case records and procedure manuals.**
  The control and maintenance of documentation of case records is discussed in the Forensic Biology Administrative Manual.

The Laboratory Director, or his/her designee, has the ultimate responsibility for all procedural manuals and assigns the writing and editing of manuals to the Deputy Director, Assistant Directors, Quality Assurance Manager and/or Criminalist IVs on a regular basis. Minor revisions to each manual are made when necessary. The finalization of each revision occurs when (i) the Director or his/her designee, and if necessary, the Technical Manager, Deputy/Assistant Directors, Quality Assurance Manager or other laboratory members have reviewed the change(s), and (ii) the Director, or his/her designee, signs an approval to indicate that a newly revised manual will be in effect. The Quality Assurance Manager shall maintain the original signed approval for each procedural manual and keeps track of all changes that have been made. The original controlled version of each procedural manual shall remain on the Departmental network drive. Every effort will be made to inform the laboratory of changes to the procedural manuals, however, it is the responsibility of each analyst to ensure that if they have a personal (uncontrolled) copy of a manual that it corresponds to the most up-to-date version.

- **The laboratory's procedures for ensuring that measurements are traceable to appropriate standards, where available.**
  These are listed in the "NIST Standards" and "Equipment Calibration and Maintenance" sections of this manual.

- **The type and extent of examinations conducted by the laboratory.**
  These are listed and described in detail in the Forensic Biology Biochemistry Manual and the Forensic Biology Protocols for Forensic STR Analysis.

- **Validation of test procedures used.**
  This is described in the Forensic Biology Administrative Manual.

- **Handling evidence.**
  This is described in the Forensic Biology Administrative Manual and the Forensic Biology Case Management Manual.

- **The use of standards and controls in laboratory procedures.**
  These are discussed in the "Reference Standards" and "Equipment Calibration and Maintenance" sections of this manual. These are also discussed in the Forensic Biology Biochemistry Manual and the Forensic Biology Protocols for Forensic STR Analysis under each analytical procedure.

- **Calibration and maintenance of equipment.**
  This is presented in the Forensic Biology Administrative Manual and in the "Equipment Calibration and Maintenance" section of this manual.

- P**ractices for ensuring continuing competence of examiners including interlaboratory comparisons, proficiency testing programs, and internal quality control schemes (e.g., technical peer review).**
  Proficiency testing, sample re-analysis, and technical review procedures are discussed in the Forensic Biology Administrative Manual. External proficiency testing for DNA analysis is conducted in the laboratory according to the FBI Quality Assurance Standards and the National DNA Index System (NDIS) standards for the operation of the Combined DNA Index System (CODIS).

- T**aking corrective action whenever analytical discrepancies are detected.**
  This is discussed in the Forensic Biology Administrative Manual.

- **Monitoring court testimony to ensure the reporting of scientific findings in an unbiased and effective manner.**
  This is discussed in the Forensic Biology Administrative Manual. All documents monitoring the court testimony of Criminalists, Assistant Directors, and Director are filed in a binder located in a designated area of the Forensic Biology Laboratory.

- **Laboratory protocol permitting departures from documented policies and procedures.**
  The specific procedures for analytical techniques done in this laboratory are thoroughly presented in the Forensic Biology Biochemistry Manual and the Forensic Biology Protocols for Forensic STR Analysis. Any deviations from the procedures must be clearly documented on the data sheets (eg. worksheets, electropherograms, etc.) that are generated.

- **Dealing with complaints.**
  This is discussed in the Forensic Biology Administrative Manual.

- **Disclosure of information.**
  This is discussed in the Forensic Biology Administrative Manual.

- **Audits and quality system review.**
  The Department of Forensic Biology Laboratory conducts audits annually in accordance to the standards dictated by ASCLD/LAB, the FBI Quality Assurance Standards, and NDIS; this is further discussed in the Forensic Biology Administrative Manual.

3. FBI QUALITY ASSURANCE STANDARDS

DATE EFFECTIVE	VERSION	PAGE
07-14-2008	5.0	1 OF 2

In accordance to the FBI Quality Assurance Standards (See Standard 3.1.1), this manual contains or references the documents or policies/procedures pertaining to the following:

- **Goals and Objectives**
  The goals and objectives of the Department of Forensic Biology are listed in the Forensic Biology Administrative Manual

- O**rganization and management structure**
  The organization and management structure of the laboratory are diagramed and discussed in the Forensic Biology Administrative Manual.

- **Personnel qualifications and training**
  Job descriptions for all laboratory personnel are described in the Forensic Biology Administrative Manual. In addition, the Civil Service specifications for each job title are kept in the laboratory along with personnel transcripts, resumes, and documentation of continuing education and training.

- **Facilities**
  This is presented in the subsequent sections of this manual.

- **Evidence Control**
  Evidence control, handling, and documentation procedures are discussed in the Forensic Biology Administrative Manual and the Forensic Biology Case Management Manual. These procedures have been designed to ensure the integrity of all physical evidence that enters the laboratory.

- **Validation**
  Validation is conducted according to the FBI Quality Assurance Standards and is described in the Forensic Biology Administrative Manual.

- **Analytical Procedures**
  This is presented in the subsequent sections of this manual and in various procedural manuals of the laboratory.

- **Calibration and Maintenance**
  This is presented in the subsequent sections of this manual and in the Forensic Biology Administrative Manual.

- **Proficiency Testing**
  Proficiency testing is discussed in the Forensic Biology Administrative Manual. External proficiency testing for DNA analysis is conducted in the laboratory according to the FBI Quality Assurance Standards and the National DNA Index System (NDIS) standards for the operation of the Combined DNA Index System (CODIS).

- **Corrective Action**
  This is discussed in the Forensic Biology Administrative Manual.

- **Reports**
  Written procedures for writing and issuing reports are presented in the Forensic Biology Case Management Manual, the Forensic Biology Administrative Manual, and the Forensic Biology Protocols for Forensic STR Analysis.

- **Review**
  Case review and related issues are discussed in the Forensic Biology Administrative Manual and the Forensic Biology Case Management Manual.

- **Safety**
  The Department of Forensic Biology has a documented environmental health and safety program as listed in the Forensic Biology Administrative Manual. This documentation is kept in the **Safety Binder**. The OCME building safety officer conducts at least three inspections each year of the laboratory.

- **Audits**
  The Department of Forensic Biology Laboratory conducts audits annually in accordance to the standards dictated by ASCLD/LAB, the FBI Quality Assurance Standards, and NDIS; this is further discussed in the Forensic Biology Administrative Manual.

4. FACILITIES

DATE EFFECTIVE	VERSION	PAGE
07-14-2008	5.0	1 OF 5

A. Security

Laboratory and building security are discussed in the Forensic Biology Administrative Manual.

B. Contamination

1. Prevention

Several measures have been taken to prevent contamination within the Department of Forensic Biology. The laboratory is divided into physically isolated areas for evidence examination, DNA extraction, pre-amplification (amplification setup) and post-amplification (amplification and DNA typing). Each area has its own dedicated equipment. Once samples are accepted into the laboratory, they move through these areas in one direction only. Samples are first processed in the evidence examination area. They are then moved to the DNA extraction area. Following DNA extraction, aliquots of each sample are quantitated in the DNA quantitation area. Following DNA quantitation, aliquots of each sample are moved into the pre-amplification area. Here fresh kit reagents are stored and samples are prepared for amplification. Finally, the samples are amplified and typed in the post-amplification area. This laboratory setup helps eliminate cross contamination from amplified DNA areas back into non-amplified DNA areas.

To avoid cross contamination between specimens, exemplar samples are processed separately from evidence samples. Also, only one sample is processed at a time using single-use disposable supplies whenever possible (eg. pipet tips), and scissors/tweezers are thoroughly cleaned between each sample (see Protocols for Forensic STR Analysis and Case Management Manuals for additional procedures to avoid cross contamination).

By far, the best defense against contamination is training for the analysts. The analysts must understand what is happening to the DNA at every step of the procedure. They must understand the rationale behind the laboratory setup and the methods of sample handling, so they are able to prevent problems before they arise. In this way, they are equipped to assess and to modify their individual habits as they practice each test of the training program.

2. Identification

Contamination can be identified as 1) the presence of signal in Rotorgene, P30, and Amylase negatives (reagent blanks), 2) presence of ***alleles*** in extraction negatives (reagent blanks) or amplification negatives (reagent blanks), 3) presence of extraneous alleles in positive controls, or 4) presence of extraneous alleles in case samples. Contamination problems reflect a system failure or contamination of the samples by an outside source. The source may be equipment, reagents, the working environment, laboratory personnel, or an analytical error. Contamination can either be a single isolated event such as cross contamination between two samples or it can be persistent, such as contamination of a reagent or equipment. To remedy contamination caused by a single isolated event, the appropriate extraction, quantitation, amplification and/or STR analysis is repeated (also see the STR Results Interpretation section in the Forensic Biology Protocols for Forensic STR Analysis).

The Quality Assurance Manager must be notified if contamination occurs. The source of contamination should be identified, if possible, and eliminated. To demonstrate the elimination of the persistent contamination, a clean run (see QC155) may be performed. During a clean run, control samples are processed along with a series of negative controls. Negative controls are run at the extraction, amplification, and typing steps. The results from these samples will indicate the area in which contamination appears. By focusing attention on one area at a time, the source or sources of contamination can be systematically eliminated. In addition, recent casework may be reviewed and selected samples may be repeated later to verify the results. The analysts will be informed of any corrective action adopted to prevent the recurrence of the problem.

3. Troubleshooting

Often, the source of a contamination problem can be identified on the basis of experience. For example, in a Rotorgene run, a) a value greater than accepted in the no template control, or b) a value greater than accepted in an extraction negative indicates (i) contamination of the reagents used during the extraction procedure, (ii) contamination of the solutions used during the Rotorgene run, (iii) consistent contamination by the analyst during extraction, or (iv) equipment contamination by improper cleaning. In the former case, this contamination may represent a build up of DNA in the reagents over the course of many extractions. Generally, fresh reagents will eliminate this problem. In the latter case, if necessary, corrective action in the form of counseling and/or retraining will be given to the identified analyst(s).

Electrophoresis runs, which appear to have the same mixture of DNA types across all the samples, indicate a more serious contamination problem at the level of the instrument or amplification step. If tubes or reagents are contaminated during the pre-amplification set up, the contaminant DNA will be amplified along with the sample. The sample signals may even be overwhelmed by the contaminant. To solve this problem, the pre-amplification room must be cleaned out and the bench washed with a 10% bleach solution. All of the kit reagents must be changed and new reaction tubes must be aliquoted.

Documentation resulting from troubleshooting experiments is kept in the **QA/QC Incident Report** binder**.**

4. Quality Control Testing Procedures

In addition to proper technique on the part of the analyst, care must also be taken in the preparation of all in-house reagents and in keeping all apparatus that come in contact with forensic samples free of contamination. To this end, various QC procedures have been developed and are part of routine laboratory operation (see Appendix B).

a. Reagent Preparation

Clean laboratory glassware is an essential in reagent preparation (see QC175). Furthermore, all aliquots of deionized water are first sterilized using an autoclave (see QC115) or irradiated prior to distribution throughout the laboratory. This procedure protects these reagents from possible bacterial contamination that could later result in the degradation of sample DNA. In addition, autoclaving conditions help to keep these solutions DNA-free. Other working reagents that are kept in the laboratory for long periods of time (e.g. 0.5M EDTA) may also be autoclaved to increase their shelf life.

b. Equipment Decontamination

Various QC procedures have also been developed to help maintain a DNA-free environment at the points of sample contact with the various apparatus used in DNA analysis. A 10% bleach solution is extremely effective in degrading DNA and is thus used for general cleanup procedures of equipment and the laboratory environment (e.g. laboratory desks and benches). Regular decontamination procedures with 10% bleach are used for the disinfection of the P30 ELISA Plate Washer (QC235), micropipetman (QC215), microcentrifuges (QC140), thermocyclers (QC290), and biosafety/fume hoods (QC125).

Documentation of these various decontamination procedures is kept in the Plate Washer Maintenance Log Binder, Thermocycler Calibration and Maintenance Log Binder and/or Laboratory Cleanup Binder.

5. ANALYTICAL PROCEDURES

A. Introduction

Analytical procedures that are used by the Forensic Biology Laboratory are described in the Biochemistry Methods Manual and Protocols for Forensic STR Analysis Manual. These manuals also include general guidelines for the interpretation of data. References to scientific literature on which these procedures are based are also included in these manuals.

B. Reagents

Reagents used to perform various analytical procedures in the laboratory are purchased from commercial vendors or prepared in the laboratory. Reagents that are purchased from commercial vendors (e.g. calibrator standards and SYBR Green I for quantitation of human DNA, sodium dodecyl sulfate, sodium hydroxide, etc.) are used either directly in a given analytical procedure (eg. calibrator standards and SYBR Green I for quantitation of human DNA) or in the preparation of in-house reagents (e.g. sodium dodecyl sulfate, sodium hydroxide).

Each reagent has a corresponding **reagent sheet** which may include the identity and application of the reagent, date of preparation, identity of individual preparing the reagent, reagent lot number (if critical reagent), standard batch size, ingredients of the reagent, procedure to follow when preparing the reagent, data log section, and the quality control procedures to be performed before the reagent is released for use into the laboratory (see Appendices A and B). Working copies of the reagent sheets are kept in the **Reagent Binders**.

At a minimum, every reagent (or its container) that is prepared by the Department of Forensic Biology is labeled with the identity of the reagent, the date of preparation or expiration, and the identity of the individual preparing the reagent. The reagent sheets may further dictate what, in addition, must be indicated on the label.

1. Lot Numbers

All critical reagents are assigned a lot number. Subsequent lots increase in numerical order (e.g. 51, 52, 53, etc.). Some reagents that are usually made fresh for a given procedure and/or are not critical reagents are not assigned lot numbers. Where applicable, the reagent sheet indicates the lot number of that reagent and the lot numbers of the ingredients that were used for making the reagent. The reagent sheets for each lot are also filed in the Reagent Binders along with any supporting quality control documentation.

2. Standard Batch Size

Each reagent sheet indicates the standard batch size routinely prepared for each lot. The quantities listed in the ingredients section have been calculated for this standard batch. Occasionally, it may be convenient to prepare a batch larger or smaller than the standard batch size. In such cases, the preparer must note the adjusted amount of each ingredient added for preparation of the reagent. If changes in demand persist over time, the reagent sheet may be modified to reflect the new batch size.

3. Ingredients

An ingredient may be either purchased from an outside vendor or prepared in-house. The ingredients required for the preparation of the reagent and the amounts of each ingredient required for the standard batch size are listed at the top of the reagent sheet. When suitable, final concentrations, and/or a tolerance of measurement are also listed next to the amount of a given ingredient. The tolerance of measurement is calculated to define an acceptable range of variation that will not significantly change the final concentration of a given reagent. Also, certain ranges have been adopted based upon recommendations for optimum performance. Volume measurements, which are made in the appropriate, size graduated cylinders and which appear to the eye to be exact, fall well within the range of tolerance listed in the ingredients section.

4. Procedure

The procedure describes how to prepare the solution step by step and includes important notes regarding the safe handling of hazardous chemicals. The completed sheets must document exactly how the solution was prepared. Any deviation from the printed procedure must be clearly documented on the reagent sheet.

5. Data Log

The **Data Log** records information regarding the ingredients used in the preparation of reagents. This information includes the source of the ingredient, lot number of the ingredient, amount of ingredient used, date of preparation, and the identity of the individual preparing the reagent. Reagents prepared in the laboratory may also be listed as ingredients (eg. BSA Solution 5 mg which is used in the preparation of YM1 PCR Reaction Mixture). In those cases, the source is listed as FB (Forensic Biology) and the laboratory lot number is recorded.

6. Quality Control

The quality control section lists the appropriate QC tests to be performed, if any, before the solution is released for use in the laboratory. These QC test procedures have been assigned QC numbers and names (eg. QC145 Chelex Extraction).

The type and number of quality procedures required to be done on a given reagent is dictated by the nature of that reagent. For example, QC620 Real Time Quantitative PCR, is listed in the quality control section for 10,000X SYBR Green I (see SYBR Green I (Raw Material) reagent sheet in Appendix A). To evaluate the performance of this component, it is not necessary to amplify and type test samples. Only the Rotorgene procedure is necessary to establish quality of the Quantiblot 10,000X SYBR Green I. On the other hand, the QC procedure for 5% Chelex (QC145) requires an extraction, human DNA quantitation, amplification, and STR analysis of the appropriate controls. The newly prepared 5% Chelex solution is released into the laboratory when all the tests have been passed.

More than one solution may be tested with a given QC procedure. In this case, the quality test must be sufficient for all of the components. For example, if a single run is to be performed for 5% Chelex and 10,000X SYBR Green I, the quality test must begin with the extraction. QC145 Chelex Extraction is the appropriate test for the Chelex, and the procedure encompasses the Rotorgene run required for the 10,000X SYBR Green I.

7. Documentation

After a quality test has been performed, the supporting documentation is attached to the original solution sheet and submitted for review. If the reagent performance is satisfactory, it will be released for general use in the laboratory. If the reagent fails to meet the standards set forth in the QC procedure, it may be submitted for further testing or discarded.

After a reagent has passed quality control and been released, the reagent sheet and quality control documentation are filed in the appropriate QC reagent binder. If more than one reagent has been tested for quality control in a single test run, the original quality control documents will be filed with one solution sheet and cross referenced on the reagent sheet of the other.

C. Critical Reagents

By definition, "critical reagents are determined by empirical studies or routine practice to require testing on established samples before use on evidentiary samples in order to prevent unnecessary loss of sample." (FBI QAS, 7/2004). Thus, all critical reagents in the Forensic Biology Laboratory have a QC test procedure listed on each respective reagent sheet. This QC test procedure must be performed in order for the reagent to be released for use in routine casework analysis.

D. Reference Standards

The laboratory must check its DNA procedures annually or whenever substantial changes are made to the protocol(s) against an appropriate and available National Institute of Standards and Technology (NIST) standard reference material (SRM) or controls that are traceable to a NIST standard.

Standard reference materials (SRM) for STR analysis may be purchased from the National Institute of Standards and Technology (NIST) and tested annually as a quality check on the equipment and procedures that are used by the lab for STR typing. The laboratory determines the DNA profiles of the given SRM samples. The results of these experiments are compared to the allele identification results that are also provided by NIST. Secondary standards may be created by identify controls and running them against NIST SRM's, which in turn makes these controls NIST traceable. The laboratory determines the DNA profiles of the controls. The results of these experiments are compared to the allele identification results of the original run. This information is filed in the **PCR NIST Standards Binder**.

Positive and negative controls are run for every analytical procedure that is done in the laboratory. A discussion of the purpose for various types of negative controls used in the laboratory is presented in the Forensic Biology Protocols for Forensic STR Analysis. A list of the correct DNA profiles for various positive controls used in STR typing is presented in the same section of the Protocols for Forensic STR Analysis Manual (see subsection Amplification Positive Control)

Equipment calibration and maintenance is essential for establishing confidence in the results that are generated during routine testing of forensic DNA samples. Equipment calibration and maintenance procedures can be subdivided into three (3) separate categories:

A. Weights and Measures

1. Temperature

The Department of Forensic Biology monitors the temperatures of all freezers, refrigerators, heat blocks and incubators that are used for storage of evidence and all types of casework samples on a daily basis, when the laboratory is open. Room temperature readings are also recorded in each laboratory. Acceptable temperature readings for each specific apparatus are noted below.

Equipment	Set Temperature	Acceptable
Freezers	-20°C	-1 to -25°C
	-80°C	-60 to -85°C
Refrigerators	4°C	1 to 13°C
56°C heat block	56°C	56 ± 3°C
65°C heat block	65°C	65 ± 3°C
95°C heat block	95°C	95 ± 3°C
100°C heat block	100°C	100 ± 3°C
37°C incubator	37°C	37 ± 3°C

The laboratory may choose to use more stringent values. However, the above minimum acceptable values must be observed.

Digital thermometers that are used to monitor heat blocks and water baths are calibrated or are replaced by new units according to the vendor specifications (e.g., recalibration date; see QC270). Digital thermometers and dedicated RTD probes used in calibrating thermocyclers are calibrated annually to National Institute of Standards and Technology (NIST) traceable standards. Each of these measuring instruments or probes are calibrated yearly to National Institute of Standards and Technology (NIST) traceable standards (see QC270 and QC280). The date of calibration is documented on the appropriate log sheet and filed in the **Temperature Equipment Maintenance Log Binder**. All new temperature measuring instruments/probes must have proof of calibration (e.g. documentation of traceability to NIST standards) prior to being used in the laboratory.

The **Amega Temperature Monitoring System** is currently in place to monitor the temperature of laboratory refrigerators, freezers, cold rooms, heat blocks and room temperatures. The Amega system is checked twice daily on weekdays by an assigned member of the Quality Assurance group. Weekends are checked on the following Monday. Results are recorded in the Amega Temperature Binder. This system is calibrated according to the manufacturer's recommendations.

Any additional maintenance performed on refrigerators and freezers is documented in the **Temperature Equipment Maintenance Log Binder**.

2. Balances

Analytical balances are used to weigh chemicals for the preparation of all laboratory reagents. At a minimum, balances must be calibrated annually to NIST traceable standards (see QC120). Documentation of each calibration is kept in the **General Equipment Maintenance Binder**.

3. pH Meter

The pH meter is used to measure the pH of reagents. A two-point calibration and verification of the pH meter is performed at least weekly (see QC245) and is documented in the **pH Log & Water System Binder**.

4. **Micropipettes**

Micropipettes are used routinely in the laboratory to measure and dispense accurate volumes of reagents used for a given protocol. All micropipettes are calibrated once each year by an outside vendor (see QC215). In addition, if at any time, there is reason to suspect that a micropipette may not be performing to its specifications, a quick gravimetric check may be done by weighing specific volumes of water on an analytical balance. If the micropipette differs significantly from specifications, the Quality Assurance Manager must be notified and the micropipette under question will be removed from laboratory operations and will be sent for calibration with the next outgoing shipment. When possible, spare calibrated micropipettes will be used as temporary replacements for any micropipettes that have been removed by this manner from regular operation. Micropipette calibration is documented in the **Pipettes PM and Calibration Database, located in the Quality Assurance network folder.**

B. Analytical Methods

Equipment that is used for specific analytical methods in the laboratory is also calibrated on a regular basis according to the specific QC procedure indicated below.

Documentation of calibration and maintenance procedures performed for equipment is done on specific equipment log sheets that are filed in each specific equipment logbook. Each logbook is located near the equipment under consideration.

Equipment	Analytical Procedure	Calibration/ Maintenance Protocol
ABI 3130xl Genetic Analyzer	STR Capillary Electrophoresis (3130xl)	QC360
BioRad Benchmark and 680 XR Microplate Readers	P30 ELISA	QC230
GeneAmp PCR System 9700	STR PCR	QC302

C. Lab Personnel Safety

The laboratory has chemical fume hoods and biological containment hoods that are inspected annually by an outside vendor (see QC125). Documentation of inspections is kept in a binder with the Quality Assurance Unit.

7. REAGENTS

This section shows a list of reagents used in the Department of Forensic Biology. They are further classified as "Critical" or "Non-Critical" reagents. As per the FBI Quality Assurance Standards, a "Critical reagent" requires testing on established samples before use in order to prevent unnecessary loss of sample. In addition, the Department of Forensic Biology may quality control test "Non-Critical" reagents to reduce the chances that analyses are rerun.

REAGENT	CRITICAL
Acid Phosphatase Test Reagent	Y
Agarose	N
Alkaline Substrate Buffer	Y
AmpFℓSTR Cofiler PCR Amplification Kit	Y*
AmpFℓSTR Identifiler PCR Amplification Kit	Y*
AmpFℓSTR Profiler Plus PCR Amplification Kit	Y*
AmpliTaq Gold DNA Polymerase Kit (all components)	Y*
Amylase Gel Buffer	Y
BigDye Terminator Cycle Sequencing Kit	Y
BSA Solution, 5 mg/mL	Y
Calibrator for Real Time Quantitative PCR	Y
Casein Stock Solution	Y
Cell Lysis Buffer (CLB)	Y
Cells	Y
Centrisep columns, strips, and plates	N
Chelex, 20%	Y
Chelex, 5%	Y
Chloroform-Isoamyl Alcohol	N
Chromogen	Y
Citrate Buffer	N
Coomassie Blue Stain	N

REAGENT	CRITICAL
Deoxynucleotide Triphosphates, 2.5 mM (dNTPs)	Y
Destain Solution	N
Digest Buffer	Y
Dithiothreitol (DTT), 1M	Y
DMSO	N
EB1	Y
EB2	Y
EDTA, 0.5 M	N
Enzyme Conjugate	Y
Ethidium Bromide (mtDNA)	Y
ExoSAP-IT	Y
Genetic Analyzer Buffer (ABI)	N
HiDi Formamide	N
Human Leukemia 60 (HL60)	Y
Hydrogen Peroxide, 3%	N
Iodine Solution, 0.01 N	N
Kastle-Meyer (KM) Reagent	Y
Leucomalachite Green (LMG) Reagent	Y
Linear Array Denaturation Solution	N
Linear Array Wash Buffer	Y
Magnesium Chloride (MgCl2)	N
Negative female control DNA for Y STR analysis	Y
Nuclear Fast Red	Y
Orange G Loading Dye	Y
Organic Extraction Buffer	Y

REAGENT	CRITICAL
PBS for Chelex Extraction	Y
PBS Solution for P30 ELISA (PBS tablets)	Y*
PBS Solution, Irradiated (LCN DNA)	Y
PBS-BSA Solution	N
Phase lock gel tubes	N
Phenol Chloroform Isoamyl Alcohol (PCIA)	Y
Picric Indigo Carmine (PIC)	Y
Pop 4	N
Pop 6	N
Poly A RNA	Y
Positive Male Control DNA for Y STR Analysis	Y
Potassium Cyanide Solution (KCN), 0.05%	N
PowerPlex 16 System	Y*
Primer, DYS19/1	Y
Primer, DYS19/2	Y
Primer, DYS389/1	Y
Primer, DYS389/2	Y
Primer, DYS390/1	Y
Primer, DYS390/2	Y
Primer, FBI – A1, B1, C1, D1, C2, D2, A4, B4, HVIF, HVIR, HVIIF, HVIIR (mtDNA)	Y
Proteinase K solution	Y
Roche Primer and Reaction Mix	Y
Saline (0.85% NaCl)	N
Sarkosyl, 20%	N
SDS, 20%	N

REAGENT	CRITICAL
SDS, 0.01% (LCN DNA)	Y
Sequencing Loading Buffer	Y
Sodium Acetate, 0.1 M	N
Species Agarose Gel	N
Species Tank Buffer	N
SSPE, 20X	N
Stain Extraction Buffer	Y
Standard DNA for Real Time Quantitative PCR	Y
Sterile Deionized Water	Y
SYBR Green I	Y
Terg-a-zyme	N
TBE buffer	N
TNE, 10X	N
TNE, 1X	N
Tris-EDTA, 1X	Y
Tris-HCl, 1M (pH 8.0)	N
Water, Irradiated (LCN DNA)	Y
XIV molecular weight ladder	N
Xylene	N
YM1 STR/PCR Reaction Mixture	Y*

***Tested for each new vendor lot/shipment**.

This section shows a list of "DNA Critical Equipment" used in the Department of Forensic Biology. As per the FBI Quality Assurance Standards, "critical equipment or instruments" are those requiring calibration or a performance check prior to use and periodically thereafter. Preventative maintenance and/or calibration protocols for each are further described elsewhere in the Forensic Biology Quality Assurance Manual.

Equipment
3130XL Genetic Analyzers
9700 Thermal Cyclers
Rotorgenes
Qiagen M48 Robots
Biotek Plate Readers
Pipetters
Thermometers
Balances
Thermal Cycler Verification Systems