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Section 1 Responsibility:

DNA Section Personnel.

DNA Training Program: new employees or additional training, DNA Unit

This program is designed to train personnel in laboratory-specific forensic DNA protocols. Here within is an outline of formal procedures for the training and assessment of new examiners/technicians in the DNA unit. The goal of this program is to develop a DNA examiner/technician capable of performing independent forensic DNA analysis. Successful completion of this program will take approximately six months. However, the training program may be abbreviated as warranted for examiners and technicians that have previous experience in forensic DNA methods with approval of the TL.

This training program is designed to supplement successful completion of college coursework in biochemistry, molecular biology, genetics, and population genetics/statistics as required by the FBI QAS. The training program will expose the DNA trainee to the scientific principles underlying each DNA test used by the State of Connecticut Division of Scientific Services. Assessment will be made at the completion of each module. Each module will be approved by the Training Coordinator, Supervisor, or Training Coordinator designee once the trainee has been deemed competent at the specified tasks.

The trainee will maintain a training folder containing the results of each module as they are being completed. The final paperwork will be retained by the Laboratory. DNA Unit personnel must pursue continuing education in the form of workshops, seminars, professional conferences or collegiate coursework as well as further on-the-job training as specified by FBI QAS.

For any necessary re-training of DNA Unit personnel, please refer to GL-14 General Training.

T. Introduction

This section must be completed by Forensic Science Examiners 1, 2, and 3, and Laboratory Assistants.

Goal:

Upon completion, the examiner will be familiar with the general forensic laboratory operations and his/her individual responsibilities.

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Tasks:

- 1. Orientation to the laboratory facility, personnel, table of organization, and the chain of command.
- 2. Instruction on the laboratory code of ethics: (GL-5) (Additional Reading: ASCLD/LAB Guiding Principles of Professional Responsibility for Crime Laboratories)
- 3. Instruction on the security and confidentiality requirements of the laboratory. (GL-3 Security)
- 4. Overview of the Quality Control/Quality Assurance policies of the laboratory (including Quality Manual).
- 5. Orientation to the Laboratory Safety Procedures and DNA specific safety procedures. (chemical, biohazard, incident reports, fire and emergency, blood-borne pathogens). (GL-2 Safety)

| TASK | Trainee Initials Date | Supervisor/Designee Approval | Date |
|---------------|-----------------------|---------------------------------|------|
| 1.Orientation | | | |
| 2.Ethics | | | |
| 3.Security | | | |
| 4.QC/QA | | | |
| 5.Safety | | | |

Required Reading:

- 1. Laboratory Quality Manual
- 2. DESPP A & O Manual.
- 3. DNA SOP, Work Instructions and Quality Records.
- 4. SWGDAM Guidelines (01-14-10).
- 5. Quality Assurance Standards for Forensic DNA Testing Laboratories (09/01/11).

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6. Quality Assurance Standards for Convicted Offender DNA Databasing Laboratories (09/01/11) DAB Audit Document.

| Required Reading | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|-------------------|------------------|------|------------------------------|------|
| 1.Lab QM /GL | | | | |
| 2.DESPP A&O | | | | |
| 3.DNA SOP, WI, QR | | | | |
| 4.SWGDAM | | | | |
| 5.QAS/Forensic | | | | |
| 6.QAS/Database | | | | |

Assessment of Section: I Introduction:

1. Oral and/or written evaluation by the supervisor or designee.

| Assessment | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|--------------------|------------------|------|------------------------------|------|
| 1.Eval./supervisor | | | | |

II. Evidence Handling / Examination

This section must be completed by Forensic Science Examiners 1, 2, and 3, and Laboratory Assistants.

Goals:

- 1. To handle evidentiary samples in an appropriate manner.
- 2. To preserve evidence that may need to be analyzed by other sections within the Laboratory.
- 3. To learn the operation of the LIMS computer system.
- 4. To demonstrate competency in the basic tasks necessary to complete evidence documentation and handling for DNA (QR-1 and QR-1A).

Tasks:

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- 1. To learn the laboratory procedures for receipt, transfer, storage, and return of evidence on the LIMS computer system for DNA casework.
- 2. To learn the written procedures and LIMS computer system for receipt, transfer, and storage of convicted offender samples for DNA database analysis.
- 3. To learn how and when to create sub-items for evidence on the LIMS computer system
- 4. To demonstrate knowledge of appropriate storage conditions for different types of evidence submitted for DNA analysis.
- 5. To demonstrate knowledge of safe handling procedures of evidence (to avoid contamination of evidence or exposure of examiner/co-workers to potential biohazards).
- 6. To demonstrate knowledge of maintaining the chain of custody and integrity of evidence.
- 7. To learn the laboratory case acceptance policies.

| Task | Trainee Initials | Ďate | Supervisor/Designee | Date |
|-------------------|------------------|------|---------------------|------|
| | | | Approval | |
| 1.LIMS/casework | | | | |
| 2.LIMS/database | | | | |
| 3.LIMS/subitems | | | | |
| 4.Storage | | | | |
| 5.Safe handling | | | | |
| 6.Chain/integrity | | | | |
| 7.Case acceptance | | | | |

Required Reading:

- 1. Quality Manual for LIMS (GL-4).
- 2. Forensic Applications of DNA Typing. Part 2: Collection and Preservation of DNA Evidence (1998).

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| Reading # | Trainee Initials | Date | Supervisor/Designee | Date |
|-------------------|------------------|------|---------------------|------|
| | | | Approval | |
| 1. GL-4 | | | | |
| | | | | |
| 2. Forensic Appl. | | | | |

Assessment of Section: II Evidence Handling:

 Demonstrate/document proficiency/understanding regarding tasks #1 − 7 to the supervisor of DNA or designee.

| Assessment | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|--------------------|------------------|------|------------------------------|------|
| 1.Eval./supervisor | | | | |

III. Foundational Scientific Knowledge

This section must be completed by Forensic Science Examiners 1, 2, and 3. Laboratory assistants may complete tasks deemed appropriate by the supervisor or designee.

1. <u>Goal</u>: To ensure that examiners have both the formal education and a working knowledge of the fundamental scientific concepts underlying DNA analysis in a forensic laboratory setting.

Tasks:

- 1. Document coursework and/or training in the following general subject areas and a working knowledge of the principles listed:
 - **A. Genetics**: alleles; Mendelian inheritance; genotype vs. phenotype; coding vs. non-coding; DNA vs. protein markers.

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- **B. Biochemistry:** is the study of the nature of biologically important molecules in living systems, DNA replication and protein synthesis, and the quantitative and the qualitative aspects of cellular metabolism.
- **C. Molecular Biology:** is the study of the theories, methods, and techniques used in the study and analysis of gene structure, organization, and function.
- **D.** Population genetics and statistics: Hardy-Weinberg equilibrium; allele and genotypic frequencies; ideal population considerations; theta, population databases and minimum size; the use of different race/ethnicity population databases; population substructure; expected frequency (match probability) vs. likelihood ratio calculations; basic probability concepts (product rule, independence).
- 2. Document in training binder any relevant continuing education and/or training (in-house or outside agency).

| Task | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|---------------------------------------|------------------|------|------------------------------|------|
| 1A.Genetics | | | | |
| 1B.Biochemistry | | | | |
| 1C.Molecular Biology | | | | |
| 1D.Population Genetics and Statistics | | | | |
| 2.Continuing education/training | | | | |

Required reading:

- 1. NRC I (1992)
- 2. NRC II (1996)
- 3. Advanced Topics in Forensic DNA Typing: Methodology (Copyright 2012)

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| Reading | Trainee Initials | Date | Supervisor/Designee | Date |
|--------------|------------------|------|---------------------|------|
| | | | Approval | |
| 1.NRC I | | | | |
| 2.NRC II | | | | |
| 3.DNA Typing | | | | |

Assessment of Section: III Foundational Scientific Knowledge

- 1. Provide transcripts of relevant coursework.
- 2. Oral examination covering basic principles and required reading above by supervisor or designee.
- 3. Certificates and/or topic outlines from continuing education/training.

| Assessment | Trainee Initials Date | Supervisor/Designee Date |
|----------------|-----------------------|--------------------------|
| | | Approval |
| 1.Transcripts | | |
| 2.Oral exam | | |
| 3.Certificates | | |

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IV. Applied Scientific Knowledge

This section must be completed by Forensic Science Examiners 1, 2, and 3. Laboratory assistants may complete tasks deemed appropriate by the supervisor or designee.

Goals:

- 1. To train an examiner in the details of forensic DNA analyses such that he/she will be able to
 - (a) Apply the knowledge to the processing of forensic DNA evidence and
 - (b) Provide the court with an appropriate explanation of how DNA testing is performed.

Tasks:

- 1. To provide instruction on the theory and legal issues regarding the following:
 - Basic biochemical formulas: calculating molarity of a solution, performing serial dilutions, determining concentrations, etc.
 - DNA extraction methods: To provide knowledge of the protocol differences for DNA B. extractions (manual and automation) of blood, buccal cells, hair, bone, teeth, tissue, and differential DNA extractions.
 - C. DNA Quantitation Method: Quantifiler Duo or Trio using 7500 real-time PCR and software.
 - D. Southern Blot Analysis/RFLP analysis: (historical overview).
 - PCR-based methods: (historical and technical aspects) E.
 - 1. PM/DQA1 reverse dot blot technique.
 - 2. D1S80 analysis.
 - 3. Autosomal STR analyses
 - 4. Y-STRs, including statistics.
 - 5. Mitochondrial DNA, including statistics.
 - 6. Minifiler

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F. Population statistics:

- 1. Determining allele frequencies.
- 2. Calculating the combined match probability of a multi-locus DNA profile.
- 3. Mixture calculations (CPI).
- 4. Paternity calculations.
- 5. Connecticut Population DNA Databases

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|------------------------|------------------|------|---------------------|------|
| Tasks | Trainee Initials | Date | Supervisor/Designee | Date |
| | | | Approval | |
| | | | Approvar | |
| 1A.Formulas | | | | |
| | | | | |
| 1B.Extraction | | | | |
| | | | | |
| 1C.Quantitation | | | | |
| | | | | |
| 1D.RFLP(history) | | | | |
| ` " | | | | |
| 1E1: PM/DQA1 | | | | |
| | | | | |
| 1E2: D1S80 | | | | |
| | | | | |
| 1E3: Autosomal STR | | | | |
| | | | | |
| 1E4: Y-STRs | | | | |
| | | | | |
| 1E5: Mito DNA | | | | |
| | | | | |
| 1E6: Minifiler | | | | |
| | * | | | |
| 1F1: Allele freq. | | | | |
| 1 | | | | |
| 1F2: Profile freq. | | | | |
| | | | | |
| 1F3: Mixtures | | | | |
| | | | | |
| 1F4: Paternity | | | | |
| | | | | |
| 1F5: CT Pop. Databases | | | | |
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Required reading:

- 1. Validation study summaries of currently used methodologies performed by the CT State Forensic Laboratory.
- 2. Developmental validation studies. (currently used amplification kits)
- 3. Review articles for RFLP, PCR, and STRs (Autosomal & Y). (found in DNA Literature Binder)

| Required Reading | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|------------------------|------------------|------|------------------------------|------|
| 1.Internal valid. | | | | |
| 2.Developmental valid. | | | | |
| 3.Review articles | | | | |

Assessment of Section: IV. Applied Scientific Knowledge

1. Demonstrate/document theoretical understanding of tasks #1A-1F5 above and required reading 1-3 to supervisor or designee.

| Assessment | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|--------------|------------------|------|------------------------------|------|
| 1. Dem./doc. | | | | |

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V. Laboratory Analytical Procedures

V-I. Casework Analytical Procedures

This section must be completed by Forensic Science Examiners 1, 2, and 3 working with casework evidentiary and casework known samples.

Laboratory assistants must complete tasks #1, #2, #3A through #3E and others deemed appropriate by the supervisor or designee.

Definitions:

Trainer: Laboratory personnel competent in the specific workflow process being taught.

Trainee: Laboratory personnel learning the workflow process.

Observed: Trainee will observe the trainer perform the workflow process.

Supervised: Trainer will observe the trainee perform the workflow process.

Independent: Trainee will perform the workflow process without supervision.

*: Extracted samples will be quantitated, amplified, run on a genetic analyzer, and analyzed to determine if the correct DNA profile (knowns) or a DNA profile (evidentiary-like/unknowns) is detectable from the extracted genomic DNA.

Goal:

To provide practical instruction on the analytical procedures to be used by the examiner. Concluded with a competency test of each.

Note: Steps in the work flow process do not need to be done separately (unless otherwise noted). Samples can be worked in a similar fashion to the casework work flow (i.e. samples can be examined, extracted, quantified, amplified, injected, and analyzed and the process repeated till the sample goal is reached.)

Note: Completion of Training

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A. The trainer and trainee can evaluate the training and determine if additional or less training in a specific area is necessary. This alteration to the training must be documented and agreed upon by training coordinator, trainer, trainee, and Technical Leader.

B. Examination and competency test will not be taken until training coordinator, trainer, and trainee deem the trainee ready.

Tasks:

1. DNA extraction (50 + samples)

Processing the minimum # of samples in tasks 1B-1E (DNA Extraction: Knowns, Evidentiary, and Differential Samples on EZ1 does not equal 50 samples, so the trainee shall process additional samples with any of the extraction methods through 1st analysis to attain 50 samples before taking any of the extraction method competency tests in this section. Additionally, the trainee shall begin with sections 1B (DNA Extraction of Knowns on EZ1) and 2 (DNA Quantitation). After completing step 1B-3 (Independent DNA Extraction of Knowns on EZ1), the trainee will complete 2C-3 (Independent performance of DNA Quantitation) using the samples generated in section 1B-3. The trainee will then complete the DNA Quantitation Competency (step 2D) before continuing with the remaining elements of DNA Extraction.

A.

- 1. Review reagent components and QC for procedures
- 2. Training in how to use DNA worklist and workbook macros
- B. DNA extraction from blood/buccal samples for known processing procedures (Qiagen EZ1).
 - 1. Observed: 1 blood, 1 buccal, and 1 positive and 1 negative extraction control
 - 2. Supervised: 1 blood, 1 buccal, and 1 positive and 1 negative extraction control*
 - 3. Independent: 1 blood, 1 buccal, and 1 positive and 1 negative extraction control*
- C. Standard DNA extraction from a range of samples representative of evidentiary material using the Qiagen EZ1.
 - 1. Observed: Minimum 1 sample, and 1 positive and 1 negative extraction control

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2. Supervised: 1 sample, and 1 positive and 1 negative extraction control*

3. Independent: At least 10 samples (including 2 hair samples), and 1 positive and 1 negative extraction control*

Samples may be extracted in more than 1 extraction set, but each set needs the 2 controls.

- D. Differential DNA extraction from semen containing samples using Qiagen EZ1.
 - 1. Observed: Minimum 1 sample, and appropriate extraction controls
 - 2. Supervised: 1 sample, and appropriate extraction controls*
 - 3. Independent: At least 5 samples, and appropriate extraction controls*

 Samples may be extracted in more than 1 extraction set, but each set needs the 2 controls
- E. Complete Competency for Known sample extraction on Qiagen EZ1
- F. Complete Competency for evidentiary-like sample extraction using Qiagen EZ1
- G. Complete Competency for differential extraction on Qiagen EZ1

2. DNA quantitation.

- A. Review PowerPoint presentation on theory of qPCR
- B. Review reagent components, QC procedure, and protocol (SOP-3).
- C. Perform qPCR procedure and estimate of yield based on quant standards.
 - 1. Observed: One set of standard curve samples and a negative control
 - 2. Supervised: One set of standard curve samples and a negative control
 - 3. Independent: Samples produced in DNA extraction section 1B-3
- D. Complete Competency.
- E. Receive training in "Stop at Quant" (see WI-07)
- 3. DNA amplification using the STR systems.
 - A. Review reagent components and QC for procedures.
- B. Amplify samples extracted in section 1 above using the Autosomal STR kit, and some select samples with the Y-STR kit, currently used for casework.

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C. Learn routine maintenance of 3130 instrument and make and evaluate a new spectral/spatial.

- D. Review 3130 operation and prepare an injection plate for running samples.
- E. Using ABI 3130 Collection software, set up run of amplified samples from 3B.
- F. Complete "To be 2nd Analyzed" Samples
- G. Complete "Known" Analysis Samples
- H. Complete "Forensic" Analysis Samples
- I. Analysis of samples processed by trainee in Task #1.
- J. Complete "Competency" Sample analysis
- 4. Concentration of DNA Samples
 - A. Select 3 appropriate samples and controls from task 1C for concentration.
 - B. Complete concentration of samples and controls
- C. Amplify, run on genetic analyzer and analyze concentrated samples and controls to determine if a DNA profile is detectable from the concentrated genomic DNA and verify the expected result of the controls.
- 5. CODIS

Complete required CODIS elements per State CODIS Administrator DNA SOP-10 to DNA SOP-16.

- 6. Minifiler
 - A. Review Validation Summaries
 - B. Complete Analysis of Minifiler Samples
 - C. Competency Test
- 7. Use of worklist, workbook and other macros used in DNA Unit.
 - A. Receive training in how to use macros
 - B. Demonstrate ability to use macros

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| TASKS | Trainee Initials | Date | Supervisor/Designee | Date |
|---|------------------|------|---------------------|------|
| | | | Approval | |
| 1. DNA extraction | 1 | | L | |
| A1. Review reagents/QC | | | | |
| A2. Use of worklist and workbook macros | | | | |
| B.Known Extraction with Qiagen EZ1 | | | | |
| B1.Observed | | | | |
| B2.Supervised | | | | |
| B3.Independent | | | | |
| C.Standard Extraction of Evidentiary-like samples | using EZ1 | | | |
| C1.Observed | | | | |
| C2.Supervised | | | | |
| C3.Independent | | | | |
| D.Differential Extraction using EZ1 | | | | 1 |
| D1.Observed | | | | |
| D2.Supervised | | | | |
| D3.Independent | | | | |
| E. COMPETENCY - Known EZ1 | | | | |
| F. COMPETENCY - Standard EZ1 & Organic | | | | |
| G. COMPETENCY - Differential EZ1 | | | | |
| 2. DNA quantification | | | | |
| A. Review reagents/QC | | | | |
| B. PowerPoint on theory | | | | |
| C. Perform qPCR | | | | |
| C1.Observed | | | | |
| C2.Supervised | | | | |
| C3.Independent | | | | |
| D. Stop at quant training | | | | |
| E.COMPETENCY | | | | |
| 3.DNA Amplification using Autosomal & Y STR Kits | | | | |
| A. Review reagents/QC | | | | |
| B. Amplify samples (Autosomal and Y STR) | | | | |
| C. 3130 maintenance incld. spatial/spectral | | | | |
| D. 3130 Operation and inject. plate setup | | | | |

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Approval

E. 3130 Collection Software and run setup
F. Complete "To Be 2nd Analyzed"

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|--|------------------|------|-----------------------|------|
| | | | Approval | |
| E. 3130 Collection Software and run setup | | | | |
| F. Complete "To Be 2 nd Analyzed" | | | | |
| G. Complete "Known" Analysis | | | | |
| H. Complete "Forensic" Analysis | | | | |
| I. Complete Analysis of Task #1 samples | | | | |
| J. COMPETENCY TEST | | | | |
| 4. Concentration of DNA samples | | | | |
| A. Completion of concentration of 3 samples | | | | |
| B. Expected results of controls | | | | |
| 5.CODIS Elements | | | | |
| 6. Minifiler | | | | |
| A.Review Validation Summaries | | | | |
| B. Analysis Training | | | | |
| C. COMPETENCY TEST | | | | |
| 7. Macros Training | | | | |
| A. Review how to use casework workbook and | | | | |
| controlled macros | | | | |
| B. Demonstrate ability to use macros | | | | |

Required reading:

- 1. Product inserts and instruction manuals.
- 2. State database statutes (CODIS).
- 3. State database regulations (CODIS).

| Required Reading | Trainee Initials | Date | Supervisor/Designee | Date |
|---------------------|------------------|------|---------------------|------|
| | | | Approval | |
| 1.Product inserts | | | | |
| 2.State DB statutes | | | | |

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| Required Reading | Trainee Initials | Date | Supervisor/Designee | Date |
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| | | | Approval | |
| 3.State DB regs. | | | | |

Assessment of Section: V. Laboratory Analytical Procedures

- 1. Documentation of analysis of at least 50 samples for STR DNA testing.
- 2. Evaluation of performance (bench skills) by supervisor or designee.
- 3. Successful completion of all competency tests for each method relevant to the examiner.
- 4. Documentation of readiness for casework by supervisor as specified in Quality Manual.

| Assessment | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|-----------------------|------------------|------|------------------------------|------|
| 1.Document 50 samples | | | | |
| 2.Eval./supervisor | | | | |
| 3.Compentency test | | | | |
| 4.Memo/casework | | | | |

V-II. Database Laboratory Analytical Procedures

This section must be completed by Forensic Science Examiners 1, 2, and 3 working with database samples and/or casework known samples.

Laboratory assistants must complete tasks deemed appropriate by the training coordinator, supervisor, or trainer.

Definitions:

Trainer: Laboratory personnel competent in the specific workflow process being taught.

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Trainee: Laboratory personnel learning the workflow process.

Observed: Trainee will observe the trainer perform the workflow process.

Supervised: Trainer will observe the trainee perform the workflow process.

Independent: Trainee will perform the workflow process without supervision.

Goals:

To provide practical instruction on the analytical procedures to be used by the trainee. Concluded with a competency test of each task/procedure.

Tasks:

NOTE: steps in the work flow process do not need to be done separately (i.e. plates can be setup and worked to completion).

Note: SOPs will be referenced in corresponding training module.

- 1. Plate Set-up/Punching (SOP 12)
 - A. Ability to create sample list of consecutive database samples, setup plate workbook (using LIMS and EXCL), and punch plate using the BSD Duet.
 - 1. Observed: 1 full plate
 - 2. Supervised: 1 full
 - 3. Independent: 2 full
 - B. Ability to create sample list of non-consecutive database samples and/or casework known samples, setup plate workbook (using LIMS and EXCL), and punch plate using the BSD Duet.
 - 1. Observed: 1 partial plate
 - 2. Supervised: 1 partial plate
 - 3. Independent: 2 partial plates
 - C. Review BSD Duet cleaning, maintenance, and users' manual.
- 2. DNA extraction (SOPs 12 & 19)
 - A. Ability to extract single source samples on the Biomek 2000 and/or Biomek 3000 using DNA-IQ (punched in 1A & 1B).

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1. Review robotic safety, DNA IQ reagent components, QC procedures, and Biomek 2000 and/or Biomek 3000 maintenance, protocols, and users' manuals.

2. Observed: 1 full plate & 1 partial plate

3. Supervised: 1 full & 1 partial plate

4. Independent: 2 full & 2 partial plates

- B. Ability to extract single source samples on the Qiagen EZ1.
 - 1. Review robotic safety, EZ1 reagent components, QC procedures, and EZ1 maintenance, protocols, and users' manual.
 - 2. Observed: 1 run of samples
 - 3. Supervised: 1 run of samples
 - 4. Independent: 2 runs of samples
- 3. DNA Quantification (SOP 3)
 - A. Review PowerPoint presentation on theory of qPCR.
 - B. Review reagent components, QC procedures, and 7500 maintenance, protocols, and users' manual.
 - C. Use qPCR procedure and estimate [DNA] based on standards.
 - 1. Quantify samples from DNA Qiagen EZ1 step:
 - a. Observed: 1 run of samples
 - b. Supervised: 1 run of samples
 - c. Independent: 2 runs of samples
 - 2. Quantify failed samples from Biomek 2000 and/or Biomek 3000 (samples from all prior steps) (Cherry-picking).
- 4. DNA amplification using Identifiler Plus (SOP 4)

Note: Minifiler and Y-STR amplification training may be added using DNA QR-283 "Addendum Training Record" and section V-I: Casework Analytical Procedures of this training manual.

A. Review reagent components, QC procedures, and thermal cycler maintenance, protocols, and users' manual.

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B. Amplify samples extracted in Part 2 (DNA extraction):

1. Biomek 2000 and/or Biomek 3000 Amplification setup

a. Observed: 1 full plate

b. Supervised: 1 full

c. Independent: 2 full

2. Manual Amplification setup: Biomek 2000 and/or Biomek 3000 partial plate samples

a. Observed: 1 partial plate

b. Supervised: 1 partial plate

c. Independent: 2 partial plates

3. Manual Amplification setup of samples from EZ1

a. Observed: 1 run of samples

b. Supervised: 1 run of samples

c. Independent: 2 runs of samples

- C. Reamplify failed samples from all prior steps (independent)
- 5. Capillary Electrophoresis using 3130 (SOP 4)
 - A. Review reagent components, QC procedures, and 3130 maintenance, protocols, and users' manual.
 - B. Setup of Plate and Operation of 3130

1. Observed: 1 full plate & 1 partial plate

2. Supervised: 1 full & 1 partial plate

3. Independent: 2 full & 2 partial plates

C. Reinjections (Cherry-picking): setup of plate and operation of 3130 (samples from all prior steps).

1. Observed: 1 reinjection

2. Supervised: 1 reinjection

3. Independent: 2 reinjections

6. Analysis of known samples using GeneMapper (SOP 5)

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- A. Review GeneMapper program parameters and users' manual.
- B. Complete "Known" Analysis Folders (6 run folders).
- C. Complete analysis of all samples run on 3130 in part 5 and confirm profiles obtained (using CODIS and/or staff index).
- 7. CODIS & Convicted Offender Samples (SOPs 10-15)
 - A. Review CODIS SOPs (SOPs 10-15).
 - B. CODIS training on the CODIS WAN (modules taken at the discretion of the CODIS Administrator).
 - C. Import/upload of database samples: including checking on duplicates, dispositions, etc.
 - 1. Observed: 1 full plate & 1 partial plate
 - 2. Supervised: 1 full & 1 partial plate
 - 3. Independent: 2 full & 2 partial plates
 - D. Hit Confirmations
 - 1. Understanding workflow:
 - a. Checking with CODIS Administrator for new hit confirmations.
 - b. Pulling database cards.
 - c. COLLECT training (taken at the discretion of the CODIS Administrator)

/COLLECT Search.

- d. Processing of Hit Confirmation Paperwork.
- 8. Post Processing of Convicted Offender Samples (SOP 11)
 - A. Review Procedure and SOP.
 - B. Post Processing Convicted Offender Samples
 - 1. Observed: Post Processing of Samples (minimum 10 samples)
 - 2. Supervised: Post Processing of Samples (minimum 10 samples)
 - 3. Independent: Post Processing of Samples (minimum 10 Samples)
- 9. Completion of Training

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A. The trainer and trainee can evaluate the training and determine if additional or less training in a specific area is necessary. This alteration to the training must be documented and agreed upon by training coordinator, trainer, trainee, and Technical Leader.

B. Examination and competency test will not be taken until training coordinator, trainer, and trainee deem the trainee ready.

| Tasks | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|----------------------------------|---------------------|------|------------------------------|----------|
| 1.Database Plate setup/punching | | 1 | | - |
| A.Full plate consecutive samples | | | | |
| A1.Observed | | | | |
| A2.Supervised | | | | |
| A3.Independent | | | | |
| B. Partial plate non-consecutive | samples | 7 | | |
| B1.Observed | | | | |
| B2.Supervised | 101 | | | |
| B3.Independent | 14 | | | |
| C. BSD Duet cleaning/maintenance | ce | | | |
| 2. DNA extraction | | 1 | 1 | . |
| A. Extraction: Biomek 2000/Bior | nek 3000 | | | |
| A1 Review Procedures | | | | |
| A2.Observed | | | | |
| A3.Supervised | | | | |
| A4.Independent | | | | |
| B. Extraction: EZ1 | | | 1 | |

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|-----------------------------------|-----------------------|------|------------------------------|------|
| B1.Review Procedures | | | | |
| B2.Observed | | | | |
| B3.Supervised | | | | |
| B4.Independent | | | | |
| 3. Quantification | - | l | | |
| A. qPCR theory | | | | |
| B. Review System | | | | |
| C1. qPCR EZ1 | | | | |
| C2. qPCR ReAmps | | | | |
| 4. Amplification: ID+ | | | | |
| A. Review System | | | | |
| B1. Biomek 2000/Biomek 3000 A | mp setup: Full plates | | | |
| B1a.Observed | | | | |
| B1b.Supervised | | | | |
| B1c.Independent | | | | |
| B2. Manual Amp setup: Partial pla | ntes | | | |
| B2a.Observed | | | | |
| B2b.Supervised | | | | |
| B2c.Independent | | | | |
| B3. Manual Amp setup: EZ1 samp | bles | | 1 | I |
| B3a.Observed | | | | |
| B3b.Supervised | | | | |
| B3c.Independent | | | | |

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|--------------------------------|---------------------|------|------------------------------|------|
| C. ReAmp samples (independent) | | | | |
| 5. Capillary Electrophoresis | | | I. | |
| A. Review System | | | | |
| B. setup/operation 3130 | | | | |
| B1.Observed | | | | |
| B2.Supervised | | | | |
| B3.Independent | | | | |
| C. Reinjections | | | | |
| C1.Observed | | | | |
| C2.Supervised | | | | |
| C3.Independent | | | • | |
| 6. Analysis | | | | |
| A. Review System | | | | |
| B. Known analysis folder | | | | |
| C. Analysis of samples in V | | | | |
| 7. CODIS | | l | | l |
| A. Review Procedures | | | | |
| B. CODIS Training | | | | |
| C. Import/Upload | | | | |
| C1.Observed | | | | |
| C2.Supervised | | | | |
| C3.Independent | | | | |
| D. Hit Confirmations | | | | |

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| Tasks | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|------------------------|---------------------|------|------------------------------|------|
| D1c. COLLECT training | | | | |
| 8. Post Processing | | | | |
| A. Review Procedures | | | | |
| B. Post Processing | | | | |
| 9. Additional Training | | | | |

Required reading:

- 1. Validation study summaries of currently used methodologies performed by the CT State Forensic Laboratory.
- 2. Developmental validation studies. (currently used amplification kits)
- 3. Review articles for RFLP, PCR, and STRs (Autosomal & Y), contamination, automation:

Biomek/EZ1, and DNA IQ (found in DNA Literature Binder)

| Required Reading | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|-------------------------|------------------|------|------------------------------|------|
| 1. Internal valid. | | | | |
| 2. Developmental valid. | | | | |
| 3.Review articles | | | | |

Assessment of Section V: Database Laboratory Analytical Procedures

- A. Oral or written of tasks #1-8 above (which were completed by the trainee) and required reading #1-3 given by supervisor or designee.
- B. Competency Test

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| Assessment | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|--|-------------------------|------|------------------------------|------|
| A.Exam | | | | |
| 1.Plate set-up | | | | |
| 2. DNA Extraction | | | | |
| 3. Quantification | | | | |
| 4. Amplification | | | | |
| 5. Capillary Electrophoresis | | | | |
| 6. Analysis | | , | | |
| 7. CODIS | | | | |
| 8. Post Processing | | | | |
| B.Competency Test: entire workflow process | | | | |
| 1. Biomek 2000/Biomek 3000 | | | | |
| 2. EZ1 | | | | |

VI. Analytical Comparisons and Report Writing

This section must be completed by Forensic Science Examiners 1, 2, and 3. Section V-I must be completed before beginning this section.

Goals:

- 1. To provide training in interpretation of DNA results (including mixtures, single source profiles, parentage testing)
- 2. To write a comprehensive report that accurately reflects the DNA typing results.

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Tasks:

1. To provide the examiner with instruction on the following (from SOP & Quality Manual):

- A. Use of report templates and standardized wording for report consistency.
- B. Laboratory policy for discussing case results with inspectors and attorneys via fax, phone or a pre-trial meeting.
- C. The laboratory policy for sharing information with a defense attorney when evidence was submitted by the prosecution or defense.
- D. The laboratory policy for verbal information to be shared with the news media or other interested parties, family members, etc.
- E. The laboratory interpretation guidelines for DNA analyses. (DNA SOP-5)
- F. The laboratory policy for reporting population statistics for the expected frequency of a DNA profile.
- G. The laboratory policy on content of the case jacket.
- H. Documentation of reports in the LIMS computer system and in the DNA notebooks.
- 2. Mock Administrative Review (AR).
 - A. Observed: Trainer will demonstrate AR with 1 case.
 - B. Supervised: Trainee will perform a Mock AR on 1 previously completed case
 - C. Independent: Trainee will perform Mock AR on 5 previously completed cases (Document each case on a QR-4 form and discuss each Mock AR with the trainer)
- 3. Mock 1st Analysis (1A), Interpretation of STR Results (IR) (see SOP-5.8), and Report Writing (RW).

Trainee will perform 1A, IR, and RW on 5 previously completed cases and discuss each result with the Trainer.

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| TASK# | Trainee Initials | Date | Supervisor/Designee | Date |
|--------------------------------|------------------|------|---------------------|------|
| | | | Approval | |
| 1A.Report templates | | | | |
| 1B.Results disclosure | | | | |
| 1C.Information sharing | | | | |
| 1D.Information to media/family | | | | |
| 1E.Interpretation guidelines | | | | |
| 1F.Reporting stats | | | | |
| 1G.Case jacket | | | | |
| 1H.LIMS/notebook documentation | | | | |
| 2.Mock Administrative Review | | | | |
| 2A. Observed | | | | |
| 2B. Supervised | | | | |
| 2C. Independent (5) | | | | |
| 3. Mock 1A, IR, RW on 5 cases | | | | |

Required reading:

1. Review Report Templates in SOP-6 and in folder on S:\ Drive titled Current NUC Report Templates.

| Required Reading | Trainee Initials | Date | Supervisor/Designee | Date |
|-------------------------|------------------|------|---------------------|------|
| | | | Approval | |
| 1.ReportTemplate Review | | | | |

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Assessment of Section: VI. Analytical Comparisons and Report Writing

- Analyze data, interpret results, and write reports on one previously completed case file as a competency test.
- 2. After completion of the competency test, the trainee may be approved to be an Administrative Reviewer of cases. This approval is documented below as well as in a memo by the TL.

| Assessment | Trainee Initials | Date | Supervisor/Designee | Date |
|----------------------------|------------------|------|---------------------|------|
| | | | Approval | |
| 1. Competency 1A, IR, & RW | | | | |
| 2. Approved to conduct | | | | |
| Administrative Review | | | | |

VII. Legal Issues

This section must be completed by Forensic Science Examiners 1, 2, and 3.

Goals:

- 1. To give the examiner an overview of the criminal justice system regarding expert witnesses.
- 2. To become familiar with the legal requirements for testimony in Connecticut and the expectations of the CT DESPP Division of Scientific Services.

Tasks:

- 1. The examiner will receive instruction on the following:
 - A. Examiner qualifications (voir dire).
 - B. Expectations for courtroom appearance (attire and demeanor).
 - C. Review of case notes prior to testimony.
 - D. Discussion of testimony and mock court training with supervisor.

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- E. Discovery and admissibility rules.
- F. Presentation of findings.
- G. Ethical responsibility of an expert witness.
- H. Structure and pertinent rules of a courtroom.

| Tasks | Trainee Initials | Date | Supervisor/Designee | Date |
|-----------------------------|------------------|------|---------------------|------|
| | | | Approval | |
| 1A.Qualifications | | | | |
| 1B.Appearance | | | | |
| 1C.Case review | | | | |
| 1D.Mock court discussion | | | | |
| 1E.Discovery | | | | |
| 1F.Presentation of findings | | | | |
| 1G.Ethics | | | | |
| 1H.Court rules | | | | |

Required reading:

- 1. Transcripts and sample testimony for presentation of DNA test results.
- 2. State admissibility standard (State vs. Porter)
- 3. Federal admissibility standards (Frye, Daubert).

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| Required Reading | Trainee Initials | Date | Supervisor/Designee | Date |
|-------------------|------------------|------|---------------------|------|
| | | | Approval | |
| 1Transcripts | | | | |
| 2.State admiss. | | | | |
| 3.General admiss. | | | | |

Assessment of Section: VII. Legal Issues

- Demonstrate competency/understanding regarding tasks #1A-1H above to the supervisor or 1. designee
- 2. Demonstrate competency in testimony by mock court exercise.
- 3. Documentation of successful completion of mock court (written evaluation by supervisor or designee).

| | Trainee Initials | Date | Supervisor/Designee | Date |
|---------------------|------------------|------|---------------------|------|
| | | | Approval | |
| 1.Demo. /document. | | | | |
| 2.Court proficiency | | | | |
| 3.Written eval. | | | | |

VIII. Oral Examination

This section must be completed by Forensic Science Examiners 1, 2, and 3.

Topics to be covered:

- 1. General DNA Theory
- 2. Genetics, Population Genetics, Forensic DNA Typing Methods.
- 3. Legal Issues & Lab QC.

Successful completion of the exam will be documented by the supervisor or designee.

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| | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|------------------|------------------|------|------------------------------|------|
| Oral Examination | | | | |

IX. Estimated Schedule for Module Completion (Based on time for new employee with no previous experience.)

| Module # | Estimated Time to Complete |
|----------|----------------------------|
| 1 | 4-8 weeks |
| 2 | 4-6 weeks |
| 3 | 6 months |
| 4 | 6 months |
| 5 | 6 months |
| 6 | 4-8 weeks |
| 7 | 4-6 weeks |
| 8 | 1-2 weeks |
| | |

Technical Leader of the DNA Section will approve all the training. Currently qualified analysts that have completed this training manual may use DNA QR-283 "Addendum Training Record" to document successful completion of new methodologies and/or platforms. This record will be kept with all other training records.

Note: If the trainee has met all the QAS requirements for being a DNA analyst and has completed all of the training (including the oral examination), except the completion of mock court, the trainee may become an analyst with the approval of the Training Coordinator and the Technical Leader.

Mock court will be completed prior to an analyst going to court.

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Technical Review Authorization

The analyst will perform casework and after gaining experience in conducting analysis and reporting findings in a variety of evidence type/conclusions in the discipline, their experience will be evaluated for the ability to conduct technical reviews.

Once the analyst has completed the required number of cases, they may be deemed competent to conduct technical reviews.

| Required number cases | Trainee Initials | Date | Supervisor/Designee Date |
|-----------------------|------------------|------|--------------------------|
| completed | | | Approval |
| New Analyst (20) | | | |
| Analyst with prior | | | |
| experience (10) | | | |



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Training program: New employees or additional training-DNA Unit, mtDNA Section

This program is designed to assess the background knowledge of personnel for their educational and practical experience in forensic mtDNA analysis. It outlines formal procedures for the training of personnel in mtDNA analysis procedures and defines how an individual examiner will be assessed for proficiency in each section of training. The ultimate goal of this training program is to develop a mtDNA examiner/technician capable of performing independent forensic mtDNA analysis. For examiners/technicians that have previous experience in forensic mtDNA methods, the training program may be abbreviated as warranted with approval of the Technical Leader.

This training program is designed to supplement successful college coursework in biochemistry, molecular biology, genetics, and population genetics/statistics as required by the DAB. The training program will expose the mtDNA trainee to the basic scientific principles underlying each mtDNA test used by the Laboratory.

Assessments will be made at the completion of each module. Each module will be approved by the technical leader once the trainee has been deemed proficient at the specified tasks. The trainee will maintain a training folder containing the results of each module as they are being completed. The final paperwork will be retained by the laboratory. mtDNA Section personnel must pursue continuing education in the form of workshops, seminars, professional conferences or collegiate coursework as well as further on-the-job training as specified by DAB standards.

The Laboratory complies with the coursework requirements set forth in the Scientific Working Group on DNA Analysis Methods (SWGDAM) *Guidelines for a Quality Assurance Program for DNA Analysis* and the DNA Advisory Board's standards, *Quality Assurance Standards for Forensic DNA Testing Laboratories*. Examiners must have completed coursework and/or training in Molecular Biology, Genetics, Biochemistry, and Population Genetics/Statistics prior to performing casework.

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I. Introduction

Goal:

Upon completion the examiner will be familiar with the general forensic laboratory operations and his/her individual responsibilities.

Tasks:

- 1. Orientation to the laboratory facility and personnel.
- 2. Instruction on the laboratory organization structure, the code of ethics (GL-5, ASCLD/LAB Guiding Principles of Professional Responsibilities for Crime Labs), and the chain of command.
- 3. Instruction on the security and confidentiality requirements of the laboratory (GL-3 Security).
- 4. Introduction to the Quality Control/Quality Assurance policies of the laboratory (including Quality Manual).
- Orientation to the Laboratory Safety Procedures and DNA specific safety procedures to include chemical, biohazard, incident reports, fire and emergency, blood-borne pathogens (GL-2 Safety, DNA SOP-1, mtDNA SOP-1).

| Task # | <u>Completed</u> | <u>Initials</u> | <u>Date</u> | <u>Supervisor</u> |
|---------------|------------------|-----------------|-------------|-------------------|
| 1.Orientation | | | | |
| 2.Ethics | | | | |
| 3.Security | | | | |
| 4.QC/QA | | | | |
| 5.Safety | | | | |

Required Reading:

- 1. Laboratory Quality Manual, GL-1 through GL-20, Administrative Directives.
- 2. DESPP A & O Manual.
- 3. Safety Manual.
- 4. mtDNA and nuclear DNA Procedure Manuals (SOP's), Work Instructions, and Quality Records.

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- 5. SWGDAM Guidelines (2013).
- 6. Quality Assurance Standards for Forensic DNA Testing Laboratories and QAS Audit Document (effective date 09/01/11).
- 7. Quality Assurance Standards for Convicted Offender DNA Databasing Laboratories and QAS Audit Documents (effective date 09/01/11).

| Reading # | Completed | <u>Initials</u> | <u>Date</u> | Supervisor |
|--------------------|-----------|-----------------|-------------|------------|
| 1.Lab QM, GL's | | | | |
| 2.DESPP A&O | | | | |
| 3.Safety | | | | |
| 4.SOP's,WI's, QR's | | | | |
| 4-1: mtDNA | | | | |
| 4-2: nDNA | | | | |
| 5.SWGDAM | | | | |
| 6.QA/Testing | | | Y | |
| 7.QA/Database | | | | |

Assessment of Section I: Introduction

- 1. Oral and/or written evaluation by the laboratory QC manager and laboratory supervisor(s).
- 2. Oral evaluation by supervisor or designee.

| Assessment # | Completed | <u>Initials</u> | <u>Date</u> | <u>Supervisor</u> |
|--------------------|-----------|-----------------|-------------|-------------------|
| 1.Eval./QC | | | | |
| manager/lab sup. | | | | |
| 2.Eval./supervisor | | | | |

This section must be completed by Examiners.

II. Evidence Handling

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Goals:

1. To handle evidentiary samples in an appropriate manner.

- 2. To preserve evidence that may need to be analyzed by other sections within the laboratory.
- 3. To learn the operation of the LIMS computer system.
- 4. To demonstrate proficiency in the basic tasks necessary to complete evidence documentation and handling for mtDNA.

Tasks:

- 1. To learn the laboratory procedures for receipt, transfer, storage, and return of evidence on the LIMS computer system for mtDNA casework.
- 2. To learn how and when to create sub-items for evidence on the LIMS computer system.
- 3. To demonstrate knowledge of appropriate storage conditions for different types of evidence submitted for mtDNA analysis.
- 4. To demonstrate knowledge of safe handling procedures for evidence (to avoid contamination of evidence or exposure of examiner/coworkers to potential biohazards).
- 5. To demonstrate knowledge of maintenance of chain of custody and integrity of evidence.
- 6. To learn the laboratory case acceptance policies.

| Task # | Completed | <u>Initials</u> | <u>Date</u> | Supervisor |
|-------------------|-----------|-----------------|-------------|------------|
| 1.LIMS/casework | | | | |
| 2.LIMS/subitems | | | | |
| 3.Storage | | | | |
| 4.Safe handling | | | | |
| 5.Chain/integrity | | | | |
| 6.Case acceptance | | | | |

Required Reading:

- 1. Quality Manual (see Section I for completed date)
- 2. LIMS training manual and work instructions

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3. Forensic Applications of DNA Typing. Part 2: Collection and Preservation of DNA Evidence (1998).

| Reading # | <u>Completed</u> | <u>Initials</u> | Date | <u>Supervisor</u> |
|--------------------|------------------|-----------------|------|-------------------|
| 2. LIMS /Work Ins. | | | | |
| 3. Forensic appl. | | | | |

Assessment of Section II: Evidence Handling

1. Demonstrate/document proficiency/understanding regarding tasks #1 – 7 to the Technical Leader of mtDNA or designee.

| Assessment # | Completed | <u>Initials</u> | <u>Date</u> <u>Supervisor</u> |
|------------------|-----------|-----------------|-------------------------------|
| 1.Dem./document. | | | |

This section must be completed by Examiners.

III. Foundational Scientific Knowledge

Goal:

To ensure that examiners have both the formal education and a working knowledge of the fundamental scientific concepts underlying mtDNA analysis in a forensic laboratory setting.

Tasks:

- 1. Document coursework and/or training in the following general subject areas and a working knowledge of the principles listed:
- A. **Basic genetics**: alleles; Mendelian inheritance; genotype vs. phenotype; coding vs. non-coding; DNA vs. protein markers.
- B. **Population genetics and statistics**: Hardy-Weinberg equilibrium; allele and genotypic frequencies; ideal population considerations; theta, population databases and minimum size; the use of different

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race/ethnicity population databases; population substructure; expected frequency (match probability) vs. likelihood ratio calculations; basic probability concepts (product rule, independence).

- C. **Biochemistry and molecular biology**: DNA structure and function; DNA replication; DNA hybridization (complementary base-pairing); theory behind basic DNA analysis techniques; theory behind mtDNA analysis.
- D. **mtDNA specific principles:** Role of mitochondria in the cell; composition of the mtDNA genome; application of mtDNA analysis to forensic casework (i.e. heteroplasmy, maternal inheritance, disease, and population databases).
- 2. Document any continuing education and/or training (in-house or outside agency).

| <u>Task #</u> | Completed | <u>Initials</u> | <u>Date</u> | <u>Supervisor</u> |
|-------------------|-----------|-----------------|-------------|-------------------|
| 1A.Basic genetics | | | 7// | |
| 1B.Pop. genetics | | | | |
| 1C.Biochem./Mol. | | | | |
| Biology | | | | |
| 1D.mtDNA | | | | |
| 2.Continuing ed. | | | | |

Required reading:

- 1. NRC I (1992)
- 2. NRC II (1996)
- 3. Fundamentals of Forensic DNA Typing (Butler, 2010), Advanced Topics in Forensic DNA Typing: Methodology (Butler, 2012).
- 4. Readings from the mtDNA bibliography located in the appendix of this document.

It is expected that the trainee will be knowledgeable about the research, development and validation of mitochondrial DNA analysis as well as topics such as heteroplasmy, maternal inheritance, disease, and databases.

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| Reading # | Completed | <u>Initials</u> | <u>Date</u> | <u>Supervisor</u> |
|------------------|-----------|-----------------|-------------|-------------------|
| 1.NRC I | | | | |
| 2.NRC II | | | | |
| 3. DNA Typing | | | | |
| 4.mtDNA readings | | | | |

Assessment of Section III: Foundational Scientific Knowledge

- 1. Provide transcripts of relevant coursework.
- 2. Oral examination covering basic principles by supervisor or designee.
- 3. Certificates and/or topic outlines from continuing education/training.

| Assessment # | <u>Completed</u> | <u>Initials</u> | <u>Date</u> | Supervisor |
|----------------|------------------|-----------------|-------------|------------|
| 1.Transcripts | | | | |
| 2.Oral exam | | | | |
| 3.Certificates | | | | |

This section must be completed by Examiners.

IV. Applied Scientific Knowledge

Goals:

1. To train an examiner in the details of general forensic DNA analyses as well as mtDNA analysis such that he/she will be able to (a) apply the knowledge to the processing of forensic DNA evidence and (b) provide the court with an appropriate explanation of how general DNA testing and mtDNA testing is performed.

Tasks:

1. To provide instruction on the theory and legal issues regarding the following:

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- A. <u>Basic biochemical formulas</u>: calculating molarity of a solution, performing serial dilutions, determining concentrations, etc.
- B. <u>DNA extraction methods</u>: To provide knowledge of the protocol/technique differences for both nDNA and mtDNA extractions of blood, buccal cells, hair, bone, teeth and tissue (refer to SOP's).
- C. <u>DNA quantitation methods</u>: (basis for the Quantifiler method used in nDNA section as well as other historical methods).
- D. Southern Blot Analysis/RFLP analysis: (historical overview).
- E. <u>PCR-based methods</u>: (historical and technical aspects)
 - 1. PM/DQA1 reverse dot blot technique.
 - 2. D1S80 analysis.
 - 3. STR analyses (Profiler Plus, COfiler, Identifiler/Identifiler Plus, PowerPlex Y/Yfiler, Minifiler).
 - 4. Y-STRs, including statistics.
 - 5. Mitochondrial DNA, including statistics.
- 5-1. Contamination minimization during mtDNA analysis procedures.
- 5-2. Methods used to recover mtDNA from forensic biological specimens
- 5-3. Basis for controls in the extraction procedures
- 5-4. mtDNA amplification procedures
- 5-5. Basis for controls in amplification procedures.
- 5-6. Post-amplification assessment using capillary electrophoresis
- 5-7. Various DNA sequencing methods to include automated dye terminator cycle sequencing
- 5-8. Electrophoretic separation of fluorescently-labeled mtDNA fragments

F. Population statistics:

1. Technical expertise with the CODIS^t mtDNA population database search program.

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2. Ability to perform statistical analysis to determine weight of evidence with existing databases.

| Task # | Completed | <u>Initials</u> | <u>Date</u> | Supervisor | |
|--------------------|-----------|-----------------|-------------|------------|--|
| 1A.Formulas | | | | | |
| 1B.Extraction | | | | | |
| 1C.Quantitation | | | | | |
| 1D.RFLP | | | | | |
| 1E1.PM/DQA1 | | | | | |
| 1E2.D1S80 | | | | | |
| 1E3.STRs | | | | | |
| 1E4.Y-STRs | | | | | |
| 1E5.Mito. DNA | | | | | |
| 5-1.Contam. min. | | | | | |
| 5-2.Rec. methods | | | | | |
| 5-3.Extr. controls | | | | | |
| 5-4.Amp. proc. | | | | | |
| 5-5.Amp. controls | | | | | |
| 5-6.Post-amp | | | | | |
| 5-7.Seq.methods | | | | | |
| 5-8.EP sep. | | | | | |
| 1F1.CODIS | | | | | |
| 1F2.Stat.anal. | | | | | |

Required reading (employee training bibliography):

- 1. Validation studies (internal) performed by the Laboratory
- 2. External validation studies.

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3. Review articles for RFLP, PCR, STRs (autosomal & Y) and mtDNA.

| Reading # | Completed | <u>Initials</u> | <u>Date</u> | <u>Supervisor</u> |
|-------------------|-----------|-----------------|-------------|-------------------|
| 1.Internal valid. | | | | |
| 2.External valid. | | | | |
| 3.Review articles | | | | |

Assessment of Section IV: Applied Scientific Knowledge

Demonstrate/document theoretical understanding of tasks #1A-1F2 above to supervisor or designee.

| Assessment # | Completed | <u>Initials</u> | <u>Date</u> | Supervisor |
|--------------|-----------|-----------------|-------------|------------|
| Dem./doc. | | 4 | | |

This section must be completed by Examiners.

V. Laboratory Analytical Procedures

Goal:

To provide practical instruction on the analytical procedures to be used by the examiner.

Tasks:

1.mtDNA extraction (16+). The number of samples should ensure a minimum of 50 successful amplifications of mtDNA hypervariable regions in accordance with the SWGDAM training guidelines (October 2001, *Forensic Science Communications*, Volume 3, Number 4).

- A. Review reagent components and QC for procedure.
- B./C. mtDNA extraction from a range of samples to include 5 hairs, 1 bone, 3 teeth, 4 blood and 3 saliva.
- 2. DNA amplification using the mtDNA systems. (50+)

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- A. Review reagent components and QC for procedure.
- B. Amplify samples extracted from a range of samples.
- 3. Post-amplification purification using ExoSAP-IT.
 - A. Review reagent components and QC for procedure.
- 4. DNA quantification using Agilent 2100 Bioanalyzer.
 - A. Review reagent components and QC for procedure.
 - B. Agilent procedure and estimate of yield.
- 5. Cycle sequencing using Big Dye v1.1 chemistry. (Sequence 5 hairs, 1 tooth, 1 bone and 3 blood or saliva samples from the samples extracted and amplified above)
 - A. Review reagent components and QC for procedure.
- 6. Sequencing using the ABI 3130.
 - A. Review reagent components and QC for procedure.
 - B. Review ABI 3130 Collection software, including sample sheet preparation and required maintenance.
 - C. Review 3130 operation and run sample sets.
- 7. mtDNA analysis including Sequencing Analysis 5.2 and Sequencher 4.1.4Fb19 software systems.

8. CODIS

A. Review CODIS 7.0. - software and SOP's.

| Task # | | Completed | <u>Initials</u> | <u>Date</u> | Supervisor |
|------------------------------|---|-----------|-----------------|-------------|------------|
| 1. mtDNA extraction (16+) | | | | | |
| Review reagents/QC | A | | | | |
| Organic method | В | | | | |
| Qiagen method | С | | | | |
| 2. mtDNA amplification (50+) | | | | | |
| Review reagents/QC | A | | | | |
| Amplify samples | В | | | | |

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| 3. Post-amp purification | | | |
|--------------------------|---|----------|--|
| Review reagents/QC | A | | |
| 4. DNA quantification | | | |
| Review reagents/QC | A | | |
| Agilent analysis | В | | |
| 5. Cycle sequencing | | | |
| Review reagents/QC | A | | |
| 6. Sequencing | | | |
| Review reagents/QC | A | | |
| Collection software | В | | |
| Operation | С | T | |
| 7. mtDNA Analysis | | | |
| 8. CODIS | | | |
| Software | A | | |

Required reading:

- 1. mtDNA section protocols.
- 2. Product inserts and instruction manuals.
- 3. CODIS user's manual.
- 4. State database statutes.
- 5. State database regulations.
- 6. Attorney General's opinions regarding database issues.

| Reading # | Completed | <u>Initials</u> | <u>Date</u> | Supervisor |
|---------------------|-----------|-----------------|-------------|------------|
| 1.Section protocols | | | | |
| 2.Product inserts | | | | |
| 3.CODIS manual | | | | |
| 4.State DB statutes | | | | |
| 5.State DB regs. | | | | |
| 6.Attorney General | | | | |

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Assessment of Section V: Laboratory Analytical Procedures

- 1. Documentation of analysis of 10 samples for mtDNA testing.
- 2. Evaluation of performance (bench skills) by supervisor or designee.
- 3. Successful completion of competency test for each method relevant to the examiner.
- 4. Documentation of readiness for casework by supervisor in as specified in Quality Manual.

| Assessment # | Completed | <u>Initials</u> | <u>Date</u> | <u>Supervisor</u> |
|---|-----------|-----------------|-------------|-------------------|
| 1. Analyze 10 samples (5 hairs, 1 bone, | | | | |
| 1 tooth and 3 blood/buccal) | | | | |
| 2. Eval./supervisor | | | | |
| 3.Competency test | | | | |
| 4.Memo/casework | | | | |

This section is completed by Examiners.

VI. Report Writing

Goals:

- 1. To write a comprehensive report that accurately reflects the mtDNA typing results.
- 2. To provide training in interpretation of mtDNA results (including single source profiles and parentage testing)

Tasks:

- 1. To provide the examiner with instruction on the following (from SOP & Quality Manual):
 - A. Use of report templates and standardized wording for report consistency.

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- B. Laboratory policy for discussing case results with inspectors and attorneys via fax, phone or a pre-trial meeting.
- C. The laboratory policy for sharing information with a defense attorney when evidence was submitted by the prosecution or defense.
- D. The laboratory policy for verbal information to be shared with the news media or other interested parties, family members, etc.
- E. The laboratory interpretation guidelines for mtDNA analyses.
- F. The laboratory policy for reporting population statistics for the expected frequency of a mtDNA profile.
- G. The laboratory policy on content of the case jacket.
- H. Documentation of reports in the LIMS computer system.

| Task # | Completed | <u>Initials</u> | <u>Date</u> | Supervisor |
|--------------------|-----------|-----------------|-------------|------------|
| 1A.Report | | | | |
| templates | | | | |
| 1B.Results | | | * | |
| disclosure | | | | |
| 1C.Information | | | | |
| sharing | | | | |
| 1D.Information to | | | | |
| media & family | | | | |
| 1E.Interpretation | | | | |
| guidelines | | | | |
| 1F.Reporting stats | | | | |
| 1G.Case jacket | | | | |
| 1H.LIMS | | | | |
| documentation | | | | |

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Required reading:

- 1. Validation studies (internal) performed by the Laboratory.
- 2. External validation studies.
- 3. Review of sample case jackets (various scenarios) as determined by the supervisor or designee.
- 4. LIMS training manual.

| Reading # | <u>Completed</u> | <u>Initials</u> | <u>Date</u> | Supervisor |
|--------------------|------------------|-----------------|-------------|------------|
| 1.Int. validation | | | | |
| 2.Ext. validation | | | | |
| 3.Mock case review | | | | |
| 4.LIMS training | | | | |

Assessment of Section VI: Report Writing

- 1. Sample data (20) representative of their job function and/or results given to the examiner to provide a written interpretation of the data according to laboratory policy.
- 2. Completion of a written interpretation of competency test results.

| Assessment # | Completed | <u>Initials</u> | <u>Date</u> | <u>Supervisor</u> |
|--------------------|-----------|-----------------|-------------|-------------------|
| 1.Sample data sets | | | | |
| 2.Comp. report | | | | |

This section is completed by Examiners.

VII. Legal Issues

Goals:

1. To give the examiner an overview of the criminal justice system regarding expert witnesses.

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2. To become familiar with the legal requirements for testimony in Connecticut and the expectations of the Laboratory.

Tasks:

- 1. The examiner will receive instruction on the following:
 - A. Examiner qualifications (voir dire).
 - B. Expectations for courtroom appearance (attire and demeanor).
 - C. Review of case notes prior to testimony.
 - D. Practice testimony and mock court training.
 - E. Discovery and admissibility rules.
 - F. Presentation of findings.
 - G. Ethical responsibility of an expert witness.
 - H. Structure and pertinent rules of a courtroom.

| Task # | | Completed | <u>Initials</u> | <u>Date</u> | Supervisor |
|----------------------|----------|-----------|-----------------|-------------|------------|
| 1A.Qualifications | | | | | |
| 1B.Appearance | | | | | |
| 1C.Case review | | | | | |
| 1D.Mock court | | | | | |
| 1E.Discovery | | | | | |
| 1F.Presentation of e | evidence | | | | |
| 1G.Ethics | | | | | |
| 1H.Court rules | | | | | |

Required reading:

1. Transcripts and sample testimony for presentation of mtDNA test results.

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2. State admissibility standard (State vs. Porter).

3. Federal admissibility standards (Frye, Daubert)

| Reading # | Completed | <u>Initials</u> | <u>Date</u> | <u>Supervisor</u> |
|-------------------|-----------|-----------------|-------------|-------------------|
| 1.Transcripts | | | | |
| 2.State admiss. | | | | |
| 3.General admiss. | | | | |

Assessment of Section VII: Legal Issues

- 1. Demonstrate competency/understanding regarding tasks #1A-1H above to the supervisor or designee
- 2. Demonstrate competency in testimony by mock court exercise.
- 3. Documentation of successful completion of mock court (written evaluation by supervisor or designee).

| Assessment # | Completed | <u>Initials</u> | <u>Date</u> | Supervisor |
|---------------------|-----------|-----------------|-------------|------------|
| 1.Demo./document. | | | | |
| 2.Court proficiency | | | | |
| 3.Written eval. | | | | |

This section must be completed by Examiners.

VIII. Oral Examination

All analysts will successfully complete 1 oral exam.

1. DNA Theory and Principles, including mitochondrial and nuclear DNA.

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- 2. mtDNA protocol and paperwork, instrumentation and administrative (nuclear protocols where applicable.
- 3. Lab safety and Lab QC.
- 4. Genetics, Population Genetics, Forensic mtDNA Typing Methods.
- 5. Legal Issues.

Successful completion of the oral exam will be documented by the supervisor or designee.

| Assessment # | Completed | <u>Initials</u> | <u>Date</u> | <u>Supervisor</u> | |
|--------------|-----------|-----------------|-------------|-------------------|--|
| Exam | | | | | |

IX. Schedule for Module Completion (new employee, no experience)

| Module # | Estimated Time to Complete |
|----------|-----------------------------------|
| 1 | 4-8 weeks |
| 2 | 4-6 weeks |
| 3 | 6 months |
| 4 | 6 months |
| 5 | 6 months |
| 6 | 4-8 weeks |
| 7 | 4-6 weeks |
| 8 | 1-2 weeks |
| | |

^{*}Oral examination and mock court will occur at the end of the 6 month training period.

The Technical Leader of the DNA Section will approve all training. Currently qualified analysts that have completed this training manual may use DNA-QR283 "Addendum Training Record" to document successful completion of new methodologies and/or platforms. This record will be kept with all other training records.

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Technical Review Authorization

The analyst will perform casework and after gaining experience in conducting analysis and reporting findings in a variety of evidence type/conclusions in the discipline, their experience will be evaluated for the ability to conduct technical reviews.

Once the analyst has completed the required number of cases, they may be deemed authorized to conduct technical reviews.

| Required number cases | Trainee Initials | Date | Supervisor/Designee Date |
|-----------------------|------------------|------|--------------------------|
| completed | | | Approval |
| New Analyst (5) | | | |
| Analyst with prior | | | |
| experience (3) | | | |



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APPENDIX mtDNA Bibliography

The following bibliography is an extensive, but not all-inclusive list of references covering issues related to forensic mtDNA analysis. It is expected that the trainee will delve deeper into the topics enumerated below as needed to gain a comprehensive understanding of issues related to mtDNA, and will strive to keep abreast of newly published literature related to forensic mtDNA analysis.

General Information

Anderson, S. et al. Sequence and organization of the mitochondrial genome, *Nature* (1981)290:457-465.

Andrews, R. M. *et al.* Reanalysis and revision of the Cambridge reference sequence for human mitochondrial DNA, *Nature Genetics* (1999) 23:147.

Chomyn, A. and Attardi, G. Mitochondrial gene products, *Current Topics in Bioenergetics* (1987) 15:295-307.

Clayton, D. A., Structure and function of the mitochondrial genome, *Journal of Inherited Metabolic Disease* (1992) 15:439-447.

Grivell, L. A. Nucleo-mitochondrial interactions in mitochondrial gene expression, *Critical Reviews in Biochemistry and Molecular Biology* (1995) 30:121-164.

Hatefi, Y. The mitochondrial electron transport and oxidative phosphorylation system, *Ann. Rev. Biochem.* (1985) 54:1015-1069.

Manifredi, G. et al. The fate of human sperm-derived mtDNA in somatic cells, American Journal Human Genetics (1997) 61:953-960.

Evolution

Horai, S. et al. Recent African origin of modern humans revealed by complete sequences of hominoid mitochondrial DNAs, *Proceedings of the National Academy of Sciences USA* (1995) 92: 532-536.

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Merriwether, D. A. *et al.* Genetic variation in the new world: ancient teeth, bone, and tissue as sources of DNA, *Experientia* (1994) 50:592-601.

Stoneking, M. and Soodyall, H. Human evolution and the mitochondrial genome, *Curr. Op. Gen. Devel.* (1996) 6:731-736.

Wilson, A. C. and Cann, R. L. The recent African genesis of humans, *Scientific American* (1992) April: 68-73.

Forensics

Allen, M. et al. Mitochondrial DNA sequencing of shed hairs and saliva on robbery caps: sensitivity and matching probabilities, *Journal of Forensic Sciences* (1998) 43:453-464.

Bar, W. et al. DNA Commission of the International Society for Forensic Genetics: Guidelines for mitochondrial DNA typing, *International Journal of Legal Medicine* (2000) 113:193-196. Also in *Forensic Science International* (2000) 110:79-85.

Budowle, B. *et al.* Mitochondrial DNA: A possible genetic material suitable for forensic analysis. In: *Advances in Forensic Sciences*. H. C. Lee and R. E. Gaensslen, (Ed). Year Book Medical Publishers, Chicago, 1990, pp. 76-97.

Budowle, B. *et al.* Forensics and mitochondrial DNA: applications, debates, and foundations, *Annu. Rev. Genomics Hum. Genet.* (2003) 4:119-141. Carracedo, A. *et al.* Reproducibility of mtDNA analysis between laboratories of the European DNA Profiling Group (EDNAP), *Forensic Science International* (1998) 97:165-170.

Fisher, D.L. *et al.* Extraction, evaluation, and amplification of DNA from decalcified and undecalcified United States Civil War bone, *Journal of Forensic Sciences* (1993) 38:60-68.

Fourney, R. M. Mitochondrial DNA and forensic analysis: a primer for law enforcement, *Canadian Society of Forensic Science Journal* (1998) 31:45-53.

Gill, P. *et al.* Identification of the remains of the Romanov family by DNA analysis, *Nature Genetics* (1994) 6:130-135.

Ginther, C. *et al.* Identifying individuals by sequencing mitochondrial DNA from teeth, *Nature Genetics* (1992) 2:135-138.

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Higuchi, R. G. et al. DNA typing from single hairs, Nature (1988) 332:543-546.

Holland, M. M. et al. Mitochondrial DNA sequence analysis of human skeletal remains: identification of remains from the Vietnam War, *Journal of Forensic Sciences* (1993) 38: 542-553.

Holland, M. M. and. Parsons, T. J. Mitochondrial DNA sequence analysis - validation and use for forensic casework, *Forensic Science Review* (1999) 11(1):21-50. Published comments by Budowle, Wilson, and DiZinno and authors' reply appear in *Forensic Science Review* (1999) 11(2): 175.

Hopgood, R. *et al.* Strategies for automated sequencing of human mitochondrial DNA directly from PCR products, *BioTechniques* (1992) 13:82-92.

Houck, M.M. and Budowle, B. Correlation of microscopic and mitochondrial DNA hair comparisons, *Journal of Forensic Sciences* (2002) 47: 964-967.

Isenberg, A. R. and Moore, J. M. *Forensic Science Communications* (1999) 1(2), http://www.fbi.gov/fbi library/forensic science communications/back issues.

Ivanov, P. L. *et al.* Mitochondrial DNA sequence heteroplasmy in the Grand Duke of Russia Georij Romanov establishes the authenticity of the remains of Tsar Nicholas II, *Nature Genetics* (1996) 12:417-420.

Jehaes, E. *et al.* Evaluation of a decontamination protocol for hair shafts before mtDNA sequencing, *Forensic Science International* (1998) 94:65-71.

Linch, C. A. *et al.* Evaluation of the human hair root for DNA typing subsequent to microscopic comparison, *Journal of Forensic Sciences* (1998) 43:305-314.

Orrego, C. and King, M. C. Determination of familial relationships. In: *PCR Protocols: A Guide to Methods and Applications*. M. A. Innis, D. H. Gelfand, J. J. Sninsky, and T. J. White (Ed.) Academic Press, San Diego, CA, 1990, p. 416-426.

Paabo, S. et al. Mitochondrial DNA sequences from a 7000-year old brain, *Nucleic Acids Research* (1988) 16:9775-9778.

Piercy, R. *et al.* The application of mitochondrial DNA typing to the study of white Caucasian genetic identification, *International Journal of Legal Medicine* (1993) 106: 85-90.

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Prieto, L. *et al.* The 2000-2001 GEP-ISFG collaborative exercise on mtDNA: assessing the cause of unsuccessful mtDNA PCR amplification of hair shaft samples, *Forensic Science International* (2003) 134:46-53.

Reynolds, R. *et al.* Detection of sequence variation in the HVII Region of the human mitochondrial genome in 689 individuals using immobilized sequence-specific oligonucleotide probes, *Journal of Forensic Sciences* (2000) 45:1210-1231.

Stoneking, M. *et al.* Population variation of human mtDNA control region sequences detected by enzymatic amplification and sequence specific oligonucleotide probes, *American Journal of Human Genetics* (1991) 48:370-382.

Stoneking, M. *et al.* Establishing the identity of Anna Anderson Manahan, *Nature Genetics* (1995) 9:9-10. Errata published in *Nature Genetics* (1995) 9:218.

Sullivan, K. M. *et al.* Automated amplification and sequencing of human mitochondrial DNA, *Electrophoresis* (1991) 12:17-21.

Sullivan, K. M. *et al.* Identification of human remains by amplification and automated sequencing of mitochondrial DNA, *International Journal of Legal Medicine* (1992) 105: 83-86.

Tully, G. et al. Rapid detection of mitochondrial sequence polymorphisms using multiplex solid-phase fluorescent minisequencing, *Genomics* (1996) 34:107-113.

Tully, G. *et al.* Considerations by the European DNA profiling (EDNAP) group on the working practices, nomenclature and interpretation of mitochondrial DNA profiles, *Forensic Science International* (2001) 124:83-91.

Wilson, M. R. et al. Validation of mitochondrial DNA sequencing for forensic casework analysis, *International Journal of Legal Medicine* (1995) 108:68-74.

Wilson, M. R. *et al.* Guidelines for the use of mitochondrial DNA sequencing in forensic science, *Crime Laboratory Digest* (1993) 20:68-77.

Wilson, M. R. *et al.* Extraction, PCR amplification, and sequencing of mitochondrial DNA from human hair shafts, *Bio Techniques* (1995) 18(4):662-669.

Wilson, M. et al. Recommendations for consistent treatment of length variants in the human mitochondrial DNA control region, *Forensic Science International* (2002) 129:35-42.

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Wilson, M. *et al.* Further discussion of the consistent treatment of length variants in the human mitochondrial DNA control region, *Forensic Science Communications* (October 2002, Volume 4, Number 4, www.fbi.gov.

Disease

Luft, R. The development of mitochondrial medicine, *Proceedings of the National Academy of Sciences USA* (1994) 91:8731-8738.

Wallace, D. C. Mitochondrial DNA in aging and disease, Scientific American (1997) August: 40-47.

Wallace, D. et al. Mitochondrial biology, degenerative diseases and aging, BioFactors (1998)7:187-190.

Maternal Inheritance and Heteroplasmy

Alonso, A. *et al.* Results of the 1999-2000 collaborative exercise and proficiency testing program on mitochondrial DNA of the GEP-ISFG: an inter-laboratory study of the observed variability in the heteroplasmy level of hair from the same donor, *Forensic Science International* (2002) 125:1-7.

Awadalla, P. *et al.* Linkage disequilibrium and recombination in hominid mitochondrial DNA, *Science* (December 24, 1999)286:2524-2525.

Bendall, K. E. and Sykes, B. C. Length heteroplasmy in the first hypervariable segment of the human mtDNA control region, *American Journal of Human Genetics* (1995) 57:248-256.

Bendall, K. E. *et al.* Heteroplasmic point mutations in the human mtDNA control region, *American Journal of Human Genetics* (1996) 59:1276-1287.

Bendall, K. E. *et al.* Variable levels of a heteroplasmic point mutation in individual hair roots, *American Journal of Human Genetics* (1997) 61:1303-1308.

Brandstatter, A. and Parson, W. Mitochondrial DNA heteroplasmy of artifacts - a matter of the amplification strategy? *International Journal of Legal Medicine* (2003) 117:180-184.

Budowle, B. *et al.* Critique of interpretation of high levels of heteroplasmy in the human mitochondrial DNA hypervariable region I from hair, *Forensic Science International* (2002) 126:30-33.

Budowle, B. *et al.* Characterization of heteroplasmy and hypervariable sites in HVI: critique of D'Eustachio's interpretations, *Forensic Science International* (2002) 130:68-70.

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Calloway, C.D. *et al.* The frequency of heteroplasmy in the HVII Region of mtDNA differs across tissue types and increases with age, *American Journal of Human Genetics* (2000) 66:1384-1397.

Comas, D. *et al.* Heteroplasmy in the control region of human mitochondrial DNA, *Genome Research* (1995) 5:89-90.

D'Eustachio, P. High levels of mitochondrial DNA heteroplasmy in human hairs by Budowle *et al.*, *Forensic Science International* (2002) 130:63-67.

Eyre-Walker, A. *et al.* How clonal are human mitochondria? *Proceedings of the Royal Society of London B* (1999) 266, 477-483.

Grzybowski, T. Extremely high levels of human mitochondrial DNA heteroplasmy in single hair roots, *Electrophoresis* (2000) 21:548-553. See also *Electrophoresis* (2001) 22:180-182 for letter and response

Grzybowski, T. *et al.* High levels of mitochondrial DNA heteroplasmy in single hair roots: reanalysis and revision, *Electrophoresis* (2003) 24:1159-1165.

Gyllensten, U. et al. Paternal inheritance of mitochondrial DNA in mice, Nature (1991) 352:255-257.

Hagelberg, E. et al. Evidence for mitochondrial recombination in a human population of island Melanesia, *Proceedings of the Royal Society of London B* (1999) 266, 485-492. Errata published in *Proceedings of the Royal Society of London B* (2000) 267:1595-1596.

Hauswirth, W. W. and Laipis, P. J. Mitochondrial DNA polymorphism in a maternal lineage of Holstein cows, *Proceedings of the National Academy of Sciences* (1982) 79:4686-4690.

Hutchinson, C. A. et al. Maternal inheritance of mammalian mitochondrial DNA, *Nature* (1974)251:536-538.

Macaulay, V. *et al.* Mitochondrial DNA recombination - no need to panic, *Proceedings of the Royal Society of London B* (1999) 266:2037-2039. Comments published by Eyre-Walker *et al.* in *Proceedings of the Royal Society of London B* (1999) 266:2041-2042.

Paabo, S. Mutational hot spots in the mitochondrial microcosm, *American Journal of Human Genetics* (1996) 59:493-496.

Parsons, T. J. et al. A high observed substitution rate in the human mitochondrial DNA control region, *Nature Genetics* (1997) 15:363-368.

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Schwartz, M. and Vissing, J. Paternal inheritance of mitochondrial DNA, *New England Journal of Medicine* (2002) 347:576-580.

Siguroardottir, S. *et al.* The mutation rate in the human mtDNA control region, American Journal of Human Genetics (2000) 66:1599-1609.

Stewart, J. E. B. *et al.* Length variation patterns in the human mitochondrial DNA control region, *Journal of Forensic Sciences* (2001) 46:862-870.

Wakeley, J. Substitution rate variation among sites in hypervariable region 1 of human mitochondrial DNA, *Journal of Molecular Evolution* (1993) 37:613-623.

Wilson, M. R. *et al.* A family exhibiting heteroplasmy in the human mitochondrial DNA control region reveals both somatic mosaicism and pronounced segregation of mitotypes, *Human Genetics* (1997) 100:167-171.

Series of letters regarding paternal inheritance/recombination: *Science* (June 25, 1999) 284:5423 *Science* (August 6, 1999) 285: unknown *Science* (June 16, 2000) 288:1931a (5 pages) *The Lancet* (September 9, 2000) 356:941

Database Issues

Forster, P. To err is human, Annals of Human Genetics (2003) 67:2-4.

Miller, K.W.P. and Budowle, B. A compendium of human mitochondrial DNA control region: development of an international standard forensic database, *Croatian Medical Journal* (2001) 42:315-327.

Monson, K. L. et al. The mtDNA population database: an integrated software and database resource for forensic comparison, Forensic Science Communications, April 2002 Volume 4 Number 2, www.fbi.gov.

Population Studies

Allard, M.W. *et al.* Characterization of the Caucasian haplogroups present in the SWGDAM forensic mtDNA dataset for 1771 human control region sequences, *Journal of Forensic Sciences* (2002) 47:1215-1223.

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Bonatto, S. L. and Salzano, F. M. Diversity and age of the four major mtDNA haplogroups, and their implications of the peopling of the new world, *American Journal of Human Genetics* (1997) 61:1413-1423.

Budowle, B. et al. Mitochondrial DNA regions HVI and HVII population data, Forensic Science International (1999) 103:23-35.

Budowle, B. et al. HVI and HVII mitochondrial DNA data in Apaches and Navajos, *International Journal of Legal Medicine* (2002) 116:212-215.

Connor, A. and Stoneking, M. Assessing ethnicity from human mitochondrial DNA types determined by hybridization with sequence-specific oligonucleotides, *Journal of Forensic Sciences* (1994) 39(6): 1360-1371.

Melton, T. and Stoneking, M. Extent of heterogeneity in mitochondrial DNA of ethnic Asian populations, *Journal of Forensic Sciences* (1996) 41:591-602.

Melton, T. et al. Extent of heterogeneity in mitochondrial DNA of sub-Saharan African populations, *Journal of Forensic Sciences* (1997) 42:582-592.

Melton, T. et al. Extent of heterogeneity in mitochondrial DNA of European populations, *Journal of Forensic Sciences* (1997) 42:437-446.

Mountain, J. L. *et al.* Demographic history of India and mtDNA sequence diversity, *American Journal of Human Genetics* (1995) 56:979-992.

Ward, R. H. et al. Extensive mitochondrial diversity within a single Amerindian tribe, *Proceedings* of the National Academy of Sciences USA (1991) 88:8720-8724.

Methodology

Butler, J. M. *et al.* Quantitation of PCR products by capillary electrophoresis using laser fluorescence, *Journal of Chromatography A* (1994) 658:271-280.

Dugan, K. A. *et al.* An improved method for post-PCR purification for mtDNA sequence analysis, *Journal of Forensic Sciences* (2002) 47:811-818.

Hanekamp, J. S. *et al.* Screening for human mitochondrial DNA polymorphisms with denaturing gradient gel electrophoresis, *Human Genetics* (1996) 98:243-245.

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Koop, B. F. *et al.* Sequence length and error analysis of Sequenase and automated *Taq* cycle sequencing methods, *BioTechniques* (1993) 14:442-447.

McCord, B. R. *et al.* Capillary electrophoresis of PCR-amplified DNA using fluorescence detection with an intercalating dye, *Journal of Chromatography A* (1993) 652:75-82.

McCord, B. R. *et al.* High resolution capillary electrophoresis of forensic DNA using a non-gel sieving buffer, *Journal of Chromatography A* (1993) 16:1963-1981.

Parker, Li T. *et al.* (1996) Amplitaq DNA polymerase, FS dye-terminator sequencing: analysis of peak height patterns, *BioTechniques* 21:694-699.

Rosenblum, B. B. et al. New dye-labeled terminators for improved DNA sequencing patterns, *Nucleic Acids Research* (1997) 25:4500-4504.

Saiki, R. K. *et al.* Primer-directed enzymatic amplification of DNA with a thermostable DNA polymerase, *Science* (1988) 239:487-491.

Sanger, F. et al. DNA sequencing with chain-terminating inhibitors, *Proceedings of the National Academy of Sciences USA*, (1977) 74:5463-5467.

Stewart, J.E.B. *et al.* Evaluation of a multicapillary electrophoresis instrument for mitochondrial DNA typing, *Journal of Forensic Sciences* (2003) 48:571-580.

Zakeri, H. *et al.* Peak height pattern in dichloro-rhodamine and energy transfer dye terminator sequencing, *BioTechniques* (1998) 25:406-414.

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DNA Training program: Kinship Analysis

Upon successful completion of the DNA training program (nuclear and/or mitochondrial), select personnel may be trained in Kinship Analysis. This program outlines formal procedures for the training of personnel in Kinship analysis procedures and defines how an individual examiner will be assessed for competency. The goal of this training program is to develop an examiner capable of performing independent Kinship analysis for non-criminal cases. For examiners that have previous experience in Kinship analysis methods, the training program may be abbreviated as warranted with approval of the Technical Leader.

This training program is designed to supplement successful college coursework in biochemistry, molecular biology, genetics, and population genetics/statistics as required by the FBI QAS. The training program will expose the DNA trainee to the scientific principles underlying Kinship analysis used by the Laboratory.

Assessments will be made at the completion of each module. Each module will be approved by the technical leader once the DNA trainee has been deemed competent at the specified tasks. The DNA trainee will maintain a training folder containing the results of each module as they are being completed. The final paperwork will be retained by the Laboratory. DNA Section personnel must pursue continuing education in the form of workshops, seminars, professional conferences or collegiate coursework as well as further on-the-job training as specified by the FBI QAS standards.

The Laboratory complies with the coursework requirements set forth in the Scientific Working Group on DNA Analysis Methods (SWGDAM) *Guidelines for a Quality Assurance Program for DNA Analysis* and the DNA Advisory Board's standards, *Quality Assurance Standards for Forensic DNA Testing Laboratories*. Examiners must have completed coursework and/or training in Molecular Biology, Genetics, Biochemistry, and Population Genetics/Statistics prior to performing casework.

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I. Kinship Analysis

Goal:

Upon completion, the examiner will be familiar with kinship calculations utilized for non-criminal cases.

Tasks:

- 1. Read SOP-25 Kinship Analysis.
- 2. Review CT DESPP Internal Kinship Validations and Summaries.
- 3. Read SWGDAM Guidelines for Missing Persons Casework (effective 01/09/2014).
- 4. Perform practice exercises in Popstats.
- 5. Perform kinship calculations by hand.

| Task # | Completed | <u>Initials</u> | <u>Date</u> | Supervisor |
|---|-----------|-----------------|-------------|------------|
| 1.Read SOP-25. | | | | |
| 2. Review validations and summaries. | | | | |
| 3. Read SWGDAM Guidelines for Missing Persons Casework. | | | | |
| 4. Practice exercises in Popstats. | | | | |
| 5. Hand calculations. | | | | |

Assessment of Section: I Kinship Analysis:

1. Oral and/or written evaluation by the supervisor or designee.

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| Assessment | Trainee Initials | Date | Supervisor/Designee Approval | Date |
|--------------------|------------------|------|------------------------------|------|
| 1.Eval./supervisor | | | | |

This section must be completed by Examiners.

II. Schedule for Module Completion (new employee, no experience)

Module # Estimated Time to Complete 10-12 weeks