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Idaho State Police Forensic Services

BIOLOGY/DNA  
QUALITY MANUAL

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## Revision History

| Revision # | Description of Changes                                                                                                                                                                                                                                                                                                                                                                                          |
|------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| 1          | Original issue in new template                                                                                                                                                                                                                                                                                                                                                                                  |
| 2          | Updated organizational chart, changed to electronic journal review documentation, removed Biomek 3000, removed DNA packets, replaced PP16HS with Fusion, removed FSS i <sup>3</sup> , added EZ1 Advanced XL, added monthly fire extinguisher checks, modified critical reagents section, updated DNA reports section for STRmix™ likelihood ratios, removed source attribution, clarifications, clerical errors |
| 3          | Added semi-annual refrigerator/freezer calibration, updated Brentamine preparation, removed Bovine Serum Albumin preparation, updated case notes section, added hair results/conclusion statements, added consumption statement, added positive kinship statement, clarifications, clerical errors                                                                                                              |
| 4          | Added Y-screen, added SwabSolution and 5X AmpSolution as critical reagents, added critical reagent QC instructions and passing criteria, updated CODIS information, updated for ILIMS DNA module, clarifications, clerical errors                                                                                                                                                                               |
| 5          | Updated organizational chart and added FS-III, updated journal article review frequency, changed statistics to coursework requirement, added FES lab access, added UV cross-linker, added Y-STRs, modified temperature monitoring requirement, added reagent expiration check to weekly QC, updated reporting statements                                                                                        |
| 6          | Removed organization chart, revised literature review process, separated technical lead and supervisor descriptions, updated CODIS security and sample entry requirements, revised consumption notification, modified QC schedules, updated reporting statements and casework review                                                                                                                            |
| 7          | Updated qualifications, Kit QC procedures, performance verification frequencies, consumption policy, and proficiency testing requirements; added clarification language throughout; added annual case review; removed annual NIST.                                                                                                                                                                              |
| 8          | Updated consumption policy, forms list, equipment and chemical inventory wording, added EZ1 and Qiacube performance verification, clarified annual case review                                                                                                                                                                                                                                                  |
| 9          | Updated annual case file review, extraction worksheets updated to controlled, added Y-screen inconclusive statement, updated amplification QC process, updated                                                                                                                                                                                                                                                  |

|    |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              |
|----|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
|    | database packets for electronic process, removed requirement to retain amplified product for consumed samples                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                |
| 10 | Updating QC/maintenance procedures to reflect new electronic format and retiring of Form 402-QC, removed photography vs sketch statement (10.1.8), clarified positive and negative requirements for kit QC's (9.3.5.1 and 9.3.6.1)                                                                                                                                                                                                                                                                                                                                                                                                                                                           |
| 11 | Updated report wording throughout, removed suggestion regarding reporting order, removed references to Qiacube, updates made to sections: 5.2.2, 7.3, 8.1.1, 9.1.1, 9.1.2, 10.1.6                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                            |
| 12 | Updated throughout for Fusion 6C and 3500/3500xl implementation, moved shower checks to monthlies and removed quarterlies                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                    |
| 13 | Added "annual" to the "Case file/Database packet review" section, added extract tube minimum labeling requirements, updated external vendor certification review requirements, modified select reagent recipes for smaller quantities                                                                                                                                                                                                                                                                                                                                                                                                                                                        |
| 14 | Updated report formatting guidelines and wording, removed Form 423-QC (3500/3500XL Maintenance Log), removed 7500 and 3500 computer defragmentation requirements, removed references to classic EZ1s, added PunchSolution to Section 9, removed references to Fusion 5C, removed Form 420-QC (Fusion 5C QC)                                                                                                                                                                                                                                                                                                                                                                                  |
| 15 | Removed references to 3130 genetic analyzers, updated semen conclusions for p30, further clarified section 10.1.3 to better match the ISPFs Quality/Procedure Manual, added additional specifications to 3.3 for the CODIS conference attendance, clarified that the annual temperature verification for the 7500s may be performed by an outside vendor, updated form number for the database reinjection summary form (moved to part of the DNA Database Worksheets), removed hyperlinks that no longer function, removed statements regarding photos during QCs, updated headers for BI-106 and BI-120, added clarification to 9.1.2, modified select reagent recipes for smaller amounts |
| 16 | Added Hamilton STARlet, removed TL approval requirement for consultations with lab personnel, updated cleaning options for reusable tools, removed reference to allele summary tables, specified what constitutes a limited amount of sperm, updated blood inconclusive statements, added Brentamine working solution recipe, administrative edits throughout                                                                                                                                                                                                                                                                                                                                |

## 1.0 Introduction

1.1 Statement of Purpose/Background: ISP Forensic Biology exists to provide quality, unbiased and cost-effective analyses in the identification of biological substances and their source(s) relevant to the investigation and prosecution of criminal offenses in Idaho. The ISP Forensic Biology Quality Manual, along with the ISP Forensic Services (ISPFS) Quality/Procedure Manual, provide the framework for the evaluation of QC (Quality Control) measures utilized in Forensic Biology to achieve that purpose. A system-wide mission and objectives are enumerated in the ISP Forensic Services Quality/Procedure Manual.

### 1.2 Objectives/Scope

- 1.2.1 To develop and maintain, through annual review and revision (where necessary), a system of quality procedures, analytical methods, and controls to ensure quality up-to-date personnel training, biological screening and DNA analyses.
- 1.2.2 To evaluate (and revise where appropriate) through proficiency testing, audits, and other means of review, the thoroughness and effectiveness of biology personnel training, procedures and QC measures.
- 1.2.3 To remain scientifically neutral by basing case/evidence acceptance and analysis decisions, case reports and testimony solely on sound scientific rationale.
- 1.2.4 To develop and use practices that respect and protect the right of privacy for the genetic profiles developed in forensic casework or for database entry.
- 1.2.5 To provide high quality training, technical and informational assistance, biological analyses, written reports and testimony.
- 1.2.6 To provide all services in a cost-effective and timely manner.

## 2.0 Organization and Management

### 2.1 Organizational Chart and Functional Structure

- 2.1.1 An organizational chart for ISP Forensic Services appears in the ISP Forensic Services Quality/Procedure Manual and includes the Forensic Biology Casework and Database sections.
- 2.1.2 An organizational chart for the Idaho State Police appears in the ISP Policy Manual.

### 2.2 Authority and Accountability in Forensic Biology

- 2.2.1 The Quality Assurance Standards for Forensic DNA Testing Laboratories and DNA Databasing Laboratories (QAS), serve as a model for the ISP Forensic Biology QA Program. These standards delineate specific responsibilities and authority for the DNA Technical Lead and DNA CODIS Administrator. Additionally, the ISP Forensic Services Quality/Procedure Manual designates specific authority for the DNA Technical Lead and DNA CODIS Administrator. The CODIS Administrator and Alternate Administrator position designations are located in the ISP Forensic Services organizational chart.

## 3.0 Personnel

### 3.1 Job Descriptions

General personnel qualifications and responsibilities, as well as personnel record retention policies, are described in the ISP Forensic Services Quality/Procedure Manual. Complete job descriptions are available through the Idaho Division of Human Resources web site.

### 3.2 Training

Refer to ISP Forensic Biology Training Manual and the ISP Forensic Services Quality/Procedure Manual for specific training requirements and retention of training and continuing education records. Retraining of analysts is outlined in the ISPFs Quality/Procedure Manual. If an analyst has been removed from casework or database, the retraining will include competency testing.

### 3.3 Continuing Education

Forensic Biology personnel must stay abreast of developments relevant to forensic DNA analyses through the attendance (and participation) at DNA related presentations, seminars, courses and/or professional meetings, for a minimum of 8 hours per calendar year. Opportunities are provided by a training budget. The training will also be supplemented through the periodic reading of current scientific literature. The table of contents of relevant journals is distributed to laboratory staff on a regular basis. Each DNA analyst will provide the DNA Technical Lead with a list of DNA related articles, rulings, documents, etc. that they have read during the calendar year. This will occur by the end of each year. Additionally, the DNA Technical Lead, or designee, may distribute a DNA-related article to each member of the biology section on a periodic basis. Each staff member will read the assigned article and provide electronic documentation to indicate the completion of the reading. The article may, optionally, be presented and discussed in a journal club format as well. Additionally, the CODIS Administrator must stay abreast of developments relevant to CODIS/NDIS database management, computer and data security and computer networks through the attendance (personal or that of the Alternate CODIS Administrator or an approved NDIS user) at the bi-annual CODIS State Administrators' meetings and annual CODIS conference. If attendance by the CODIS Administrator or Alternate is not possible, approval from NDIS will be required prior to sending another representative.

### 3.4 Qualifications

Education, training and experience for Forensic Biology personnel are formally established in the following minimum requirement specifications (Minimum requirements for individual positions are listed in the ISPFs Quality/Procedure manual. These may be reviewed at the time of job announcement and may exceed

those delineated below). The minimum degree and education requirements are verified by review of transcripts as well as course descriptions, as necessary, during the application process. The DNA Technical Lead approves the degree and coursework prior to a job offer being extended to any potential hire. The date of hire/appointment/promotion will determine the applicable version of the QAS to be used for education, experience, and training review. Periodic review of continuing education and overall performance is accomplished during the annual employee evaluation.

#### 3.4.1 Biology/DNA Technical Lead

The Technical Lead is responsible for the technical operations of the laboratory; approves validations and procedure modifications, training/qualifications, retraining plans, procedures, biology quality assurance, and proficiency testing programs; as well as performs duties of a DNA analyst as outlined in section 3.4.4.

##### 3.4.1.1 Education

Must have, at minimum, a Master of Science degree in a biology, chemistry, or forensic science related area. Successful completion of a minimum of 12 semester or equivalent credit hours, including a combination of graduate and undergraduate coursework in genetics, biochemistry, molecular biology and statistics (or population genetics) as specified in the current QAS.

##### 3.4.1.2 Training

Training in each technology currently utilized in the laboratory within one year of appointment if not currently or previously qualified. Must also complete the DNA Quality Assurance auditor training within 1 year of appointment, if not already completed.

##### 3.4.1.3 Experience

Must have a minimum of three years forensic human DNA laboratory experience as a qualified analyst.

#### 3.4.2 CODIS Administrator

The functions of casework and database CODIS Administrators will typically be served by a single individual. An Alternate CODIS Administrator will also be appointed and must meet the same qualifications as the CODIS Administrator. The CODIS Administrator is responsible for administering the laboratory's CODIS network, scheduling and documenting the computer training for analysts, as well as assuring the security and quality of data and match dispositions are in accordance with state and/or federal law and NDIS operational procedures.

##### 3.4.2.1 Education

Must have at minimum, a Bachelor of Science degree in a biology, chemistry, or forensic science related area and successfully completed a minimum of 9 semester or equivalent credit hours in genetics, biochemistry, and molecular biology. Additionally, must have successfully completed a course in statistics (or population genetics).

#### 3.4.2.2 Training

A combination of training and experience in the use of computers, and database systems in a laboratory/scientific setting. Must also complete the FBI's CODIS software training within six months and the DNA Quality Assurance auditor training within one year of appointment if either was not already completed.

#### 3.4.2.3 Experience

Must possess a working knowledge of computers, computer networks, computer database management and have an understanding of DNA profile interpretation for database and casework functions, to include training in mixture interpretation. Must be or have been a qualified DNA analyst.

#### 3.4.3 Biology/DNA Supervisor

The Supervisor oversees the general operations and personnel related duties for the casework and/or database sections of the laboratory. The supervisor also performs the duties of a DNA analyst as outlined in section 3.4.4.

##### 3.4.3.1 Education

Must have at minimum, a Bachelor of Science degree in a biology, chemistry, or forensic science related area and successfully completed a minimum of 9 semester or equivalent credit hours in genetics, biochemistry, and molecular biology. Additionally, must have successfully completed a course in statistics (or population genetics).

##### 3.4.3.2 Training

Training in DNA analyses through academic, governmental, private forensic and/or research laboratory(ies). If received elsewhere, documented training must meet or exceed that outlined in the ISP Forensic Biology training manual. Must successfully complete a qualifying examination prior to performing analyses on database or forensic casework samples.

##### 3.4.3.3 Experience

Must have a minimum of six months forensic human DNA laboratory experience.

#### 3.4.4 DNA Analyst

The following delineate requirements for a DNA casework or database analyst whose responsibilities include performing genetic analyses on the capillary electrophoresis (CE) instruments and/or data interpretation. DNA extraction, quantitation, and amplification set-up may be performed by appropriately trained laboratory technicians and/or those performing the biological screening of evidence following task-specific training and successful completion of a qualifying examination.

#### 3.4.4.1 Education

Must have at minimum, a Bachelor of Science degree in a biology, chemistry, or forensic science related area and successfully completed a minimum of 9 semester or equivalent credit hours in genetics, biochemistry, and molecular biology. Additionally, must have successfully completed a course in statistics (or population genetics).

#### 3.4.4.2 Training

Training in DNA analyses through academic, governmental, private forensic and/or research laboratory(ies). If received elsewhere, documented training must meet or exceed that outlined in the ISP Forensic Biology training manual. Must successfully complete a qualifying examination prior to performing analyses on database or forensic casework samples.

#### 3.4.4.3 Experience

Must have a minimum of six months forensic human DNA laboratory experience.

### 3.4.5 Forensic Biologist

The following delineate requirements for those individuals responsible for the screening of evidence for the presence of biological substances and reporting and giving testimony regarding their findings.

#### 3.4.5.1 Education

Must have a Bachelor of Science in a physical or biological science.

#### 3.4.5.2 Training

Training specific to this job function in a governmental and/or private forensic laboratory. If received elsewhere, documented training must meet or exceed that outlined in the ISP Forensic Biology training manual. Must successfully complete a qualifying examination prior to performing forensic casework.

#### 3.4.5.3 Experience

Prior to participating in independent forensic casework, must have a minimum of six months forensic laboratory experience in the area of biological screening and/or DNA analysis.

### 3.4.6 Biology Laboratory Technician

If the technician performs interpretation of data, to include quantitation and/or CE, the minimum requirements of an analyst in 3.4.4 must be met.

3.4.6.1 Education

Must have a Bachelor of Science degree in a physical or biological science.

3.4.6.2 Training

Must receive on-the-job training specific to assigned duties and successfully complete a qualifying examination before participating in forensic DNA typing or forensic casework responsibilities.

3.4.6.3 Experience

Prior to participating in any forensic DNA typing responsibilities or forensic case processing activities, technician must have a minimum of six months forensic laboratory experience in the area of Biology/DNA.

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## 4.0 Facilities and Security

### 4.1 Laboratory Security

#### 4.1.1 Forensic Biology Security

When not under the direct control of Forensic Biology personnel, evidence and in-progress work product will be secured either by closing and locking the Forensic Biology door or by its return to secure storage (one of the locked evidence refrigerators/freezers or the analyst's personal evidence cabinet). Only Forensic Biology personnel will have access to the locked storage and laboratory areas. Designated evidence personnel may have limited access to the locked laboratory areas for evidence transfer and/or database sample accessioning. Persons outside the Forensic Biology unit (and designated evidence personnel) will not be allowed access to the Forensic Biology laboratories. Exceptions will be made in case of emergencies, for maintenance, safety, and/or equipment service needs, and for required annual quality and DNA audits. At these times, access will be limited to only required individuals, the individual(s) will be accompanied by biology program personnel, and all evidence will be placed in secured storage or a locked exam room for the duration of the individual(s) being present in the laboratory. An exception may be made for consultation with personnel from another unit of the laboratory.

#### 4.1.2 CODIS Security

The CODIS workstation is located in the secure forensic services laboratory area and the CODIS Server is located in the secured server room in the ISP-IT Section. The following security measures have been implemented:

- 4.1.2.1 Only the CODIS State Administrator, Alternate CODIS State Administrator, and designated IT personnel will have login access to the CODIS Server.
- 4.1.2.2 A full backup of the CODIS server will be performed nightly. Data will be kept onsite for 100 days and offsite for 365 days.
- 4.1.2.3 Only Forensic Biology personnel that have gone through the NDIS application and approval process will have user-names and passwords for CODIS.
- 4.1.2.4 CODIS users must log in each time they use CODIS and log out prior to leaving the CODIS Workstation.
- 4.1.2.5 DNA Tracker, the convicted offender sample-tracking database, resides on the ISP intranet and is accessible only to personnel designated by the DNA Technical Lead and/or DNA Database Supervisor.

- 4.1.2.6 Personal and identifying information on convicted offenders (hard and electronic/DNA Tracker copies) are stored separately from the DNA profile (CODIS) obtained. The DNA profiles are directly associated only with a unique Idaho Convicted Offender ID number, assigned by DNA Tracker upon sample entry.
- 4.1.2.7 CODIS samples and corresponding information are released only in accordance with 19-5514 of the Idaho DNA Database Act of 1996, the Privacy Act Notice in Appendix B of NDIS procedures, and the FBI/CODIS Memorandum of Understanding.

#### 4.2 Forensic Biology Laboratory Set-up

The Forensic Biology and Database Laboratories are designed to minimize contamination potential during the processing and analysis of forensic and convicted offender samples. Separate areas for evidence examination, DNA extraction, PCR Amplification Set-up and Amplified DNA processing and storage are delineated. Some steps of the pre-amplification processes may be conducted in the same area of the main laboratory; however, these steps are separated by time.

#### 4.3 Laboratory Cleaning and Decontamination

In order to minimize the potential for sample contamination, careful cleaning of laboratory work areas and equipment must be conducted on a routine basis. The efficacy of the procedures used is monitored through the use of controls within the analysis process (see the interpretation guidelines section in BI-212 and BI-318). It is also important that each analyst use proper 'clean technique' at all times when in the laboratory, which includes but is not limited to, using only disposable barrier pipette tips and autoclaved (or UV cross-linked) microcentrifuge tubes, using a tube de-capping tool, and wearing gloves, a lab coat, and masks as appropriate.

- 4.3.1 All working benchtop surfaces will be cleaned with 10% bleach or bleach substitute before and after use and as part of the monthly QC procedure. Clean white paper and/or a Wypall will be placed on the workbench prior to use and changed as appropriate and necessary.
- 4.3.2 All small reusable tools/instruments (i.e. forceps, scissors, etc.) will be cleaned/rinsed with germicidal instrument cleaner or 10% bleach prior to use and between samples. Metal reusable tools cleaned with bleach should then be rinsed with ethanol. Kimwipes, used to dry the instrument after cleaning/rinsing, will be single use only.

- 4.3.3 Pipettes are to be cleaned thoroughly with bleach/bleach substitute solution as part of the monthly QC procedure and anytime the barrel comes in contact with DNA or any biological fluid.
- 4.3.4 All centrifuges are to be wiped down (interior and exterior) with bleach/bleach substitute solution as part of the monthly QC procedure and in the event of a spill.
- 4.3.5 The exterior surfaces of the BSD600-Duet Puncher are to be wiped down with a damp cloth as part of the weekly QC. In addition, the chute and punch mechanism are to be cleaned by removing and separating the inner and outer chutes. The inner chute is to be cleaned with ethanol, followed by compressed air blown through both chutes, the hole in the underside of the manifold, and between the punch guide and die. Do not use ethanol on the outer chute or around any electrical components.
- 4.3.6 The thermal cyclers, to include the heating block and exterior surfaces, are to be wiped down with ethanol or bleach/bleach substitute solution as part of the monthly QC procedure. Individual wells should be cleaned as needed.
- 4.3.7 As part of the monthly QC procedure, the following are to be conducted: the exterior surfaces of the genetic analyzers and real-time instruments wiped down with ethanol or bleach/bleach substitute solution, top and handles of the refrigerator/freezers and surface underneath each genetic analyzer wiped down/dusted, and floor mopped.

## 5.0 Evidence Control and Handling

Evidence, Individual Characteristic Database (Convicted Offender) samples, in progress work product, and applicable Standard Reference Materials, that are collected, received, handled, sampled, analyzed and/or stored by ISP Forensic Services is done so in a manner to preserve its identity, integrity, condition and security.

### 5.1 Laboratory Evidence Control

Procedures detailing evidence handling are contained in the ISP Forensic Services Quality/Procedure Manual. Refrigerated evidence should be stored between 1°C and 12°C. Frozen evidence should be stored at  $\leq -10^{\circ}\text{C}$ . Standard Reference Materials will be handled, stored, transported, and used according to the guidelines outlined on the corresponding certificate of analysis. Bloodstains and/or semen samples certified against a NIST SRM used as a known standard, will be stored frozen ( $\leq -10^{\circ}\text{C}$ ), and handled as a potential biohazard. Portions of individual evidence items that are carried through the analysis process (i.e. substrate cuttings, extracts, amplified product and/or portions thereof) are considered work product while in the process of analysis and do not require sealing. Work product will be identified by labeling the individual sample tube with a unique identifier, or documenting the locations of individual samples within a plate of samples.

### 5.2 Forensic Biology Evidence Control/Sample Retention

#### 5.2.1 DNA Extracts

Any remaining DNA extracts, upon completion of analysis, will be placed into a sealed container (such as a plastic zip bag or envelope) and assigned an item number in ILIMS. Tubes should be labeled, at minimum, with the case number, item number, date and initials of the analyst who performed the extraction.

#### 5.2.2 Limited Sample

In every case, care should be taken to save  $\sim 1/2$  of a sample for independent testing whenever possible. Samples of limited size and/or quantity may be consumed if necessary, unless the submitting agency has indicated not to. When a request not to consume has been received, or if sample consumption is a result of retesting due to contamination, written permission to consume must be received from the prosecuting attorney or submitting agency prior to proceeding with the testing. DNA Database samples will not be consumed without prior authorization from the DNA Technical Lead.

#### 5.2.3 Amplified Product

Amplified DNA product will not be retained after 1) the report has been issued in the case or 2) review of the offender sample data has been

completed and certified for CODIS entry (or approximately two weeks after amplification of the offender sample).

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## 6.0 Validation

Procedures for the validation and/or performance verification of methods used in ISP Forensic Services are outlined in the ISP Forensic Services Quality/Procedure Manual. Validation/performance verification data, results and summaries for those methods employed in Forensic Biology will be maintained in that section.

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## 7.0 Analytical Methods and Forms

The following is a list of general practices/administrative procedures, analytical methods and forms utilized in Forensic Biology. Each follows the numbering scheme of: Biology Screening (1XX), DNA Casework Analysis (2XX), Database Analysis (3XX), QC Functions (4XX), and CODIS (5XX). Where applicable electronic equivalents in ILIMS may be used in place of some forms.

### 7.1 MBI≡Schemes, generally encompassing many procedures.

|         |                                                  |
|---------|--------------------------------------------------|
| MBI-100 | EXAMINATION OF BLOODSTAINED EVIDENCE             |
| MBI-102 | EXAMINATION OF EVIDENCE FOR SEMEN                |
| MBI-104 | EXAMINATION OF EVIDENCE FOR BODY FLUIDS          |
| MBI-200 | INDIVIDUALIZATION OF DNA SOURCES BY STR ANALYSIS |
| MBI-300 | INDIVIDUALIZATION OF DNA SOURCES BY STR ANALYSIS |

### 7.2 BI≡Analytical Procedures or Individual Processes

|        |                                                         |
|--------|---------------------------------------------------------|
| BI-100 | PROCESSING LIQUID BLOOD                                 |
| BI-104 | PHENOLPHTHALEIN TEST FOR BLOOD                          |
| BI-105 | O-TOLIDINE TEST FOR BLOOD                               |
| BI-106 | ABACARD® HEMATRACE® TEST FOR BLOOD                      |
| BI-110 | BIOLOGICAL SCREENING: USE OF ALTERNATE LIGHT SOURCE     |
| BI-111 | BIOLOGICAL SCREENING: USE OF INFRA RED LIGHT            |
| BI-114 | BRENTAMINE TEST FOR ACID PHOSPHATASE                    |
| BI-116 | SAMPLE EXTRACTION FOR SEMEN IDENTIFICATION              |
| BI-118 | SEMEN IDENTIFICATION: MICROSCOPIC EXAMINATION           |
| BI-119 | DIGITAL IMAGING                                         |
| BI-120 | ABACARD® P30 TEST FOR SEMEN                             |
| BI-122 | AMYLASE TEST: PHADEBAS                                  |
| BI-126 | DETECTION OF URINE (UREASE)                             |
| BI-130 | DETECTION OF FECAL MATERIAL (UROBILINOGEN)              |
| BI-132 | DETECTION OF MALE DNA ON SEXUAL ASSAULT KIT EVIDENCE    |
| BI-200 | EXTRACTION PROTOCOLS FOR PCR DNA TYPING TESTS           |
| BI-207 | DNA QUANTITATION: REAL-TIME PCR                         |
| BI-208 | STR AMPLIFICATION: POWERPLEX® FUSION 6C SYSTEM          |
| BI-210 | STR TYPING: CAPILLARY ELECTROPHORESIS AND DATA ANALYSIS |
| BI-212 | STR INTERPRETATION GUIDELINES AND STATISTICAL ANALYSES  |
| BI-214 | Y-STR ANALYSIS: POWERPLEX® Y23 SYSTEM                   |
| BI-215 | DNA QUANTITATION SETUP USING THE HAMILTON® STARLET      |

|        |                                                         |
|--------|---------------------------------------------------------|
| BI-216 | DNA AMPLIFICATION SETUP USING THE HAMILTON® STARLET     |
| BI-301 | OFFENDER SAMPLE RECEIPT AND DNA TRACKER ENTRY           |
| BI-312 | EXTRACTION PROTOCOLS FOR PCR DNA TYPING TESTS           |
| BI-314 | DNA QUANTITATION: REAL-TIME PCR                         |
| BI-316 | STR AMPLIFICATION: POWERPLEX® FUSION 6C SYSTEM          |
| BI-318 | STR TYPING: CAPILLARY ELECTROPHORESIS AND DATA ANALYSIS |
| BI-400 | DRIFTCON FFC: TEMPERATURE VERIFICATION                  |
| BI-500 | CODIS SAMPLE DATA ENTRY AND UPLOAD                      |
| BI-501 | CODIS DATABASE HIT VERIFICATION                         |
| BI-502 | CODIS SAMPLE REMOVAL                                    |

7.3 Form BI≡Various forms used in each discipline (\* indicates a controlled form)

|          |                                               |
|----------|-----------------------------------------------|
| 132-BI*  | Y-SCREEN WORKSHEETS (A-B)                     |
| 200-BI*  | DNA EXTRACTION WORKSHEET                      |
| 202B-BI* | DIFFERENTIAL DNA EXTRACTION WORKSHEET         |
| 206-BI*  | CASEWORK WORKSHEETS (A-D)                     |
| 207-BI*  | FUSION 6C CASEWORK WORKSHEETS (A-F)           |
| 208-BI*  | HAMILTON STARlet WORKSHEETS (A-E)             |
| 210-BI   | DNA BATCH REVIEW CHECKLIST                    |
| 211-BI   | Y-SCREEN BATCH REVIEW CHECKLIST               |
| 306-BI   | STR OFFENDER DATABASE REVIEW CHECKLIST        |
| 312-BI*  | DATABASE WORKSHEETS (A-G)                     |
| 314-BI   | OUTSOURCED OFFENDER DATA REVIEW               |
| 400-BI   | REAGENT PREP LOG                              |
| 403-QC   | FORENSIC BIOLOGY pH CALIBRATION RECORD        |
| 404-QC   | BIOLOGY/DNA CASEWORK WEEKLY QC                |
| 405-QC   | DNA DATABASE WEEKLY QC                        |
| 406A-QC  | BIOLOGY/DNA CASEWORK MONTHLY QC               |
| 406B-QC* | BIOLOGY/DNA CASEWORK MONTHLY QC               |
| 406C-QC  | DNA DATABASE MONTHLY QC                       |
| 406D-QC* | DNA DATABASE MONTHLY QC                       |
| 410-QC   | QC ABACARD® HEMATRACE® KIT                    |
| 412-QC   | QC ONESTEP ABACARD® P30 KIT                   |
| 418-QC   | QC SWABSOLUTION™ AND 5X AMPSOLUTION™ REAGENTS |
| 419-QC   | QC PLEXOR® HY QUANTITATION KIT                |
| 420A-QC  | QC POWERPLEX FUSION 6C SYSTEM KITS            |
| 421-QC   | QC POWERPLEX Y23 SYSTEM KITS                  |
| 427-QC   | INFREQUENT TEST LOG                           |
| 500-BI   | CODIS SAMPLE REMOVAL CHECKLIST                |
| 502-BI   | HIT CONFIRMATION CHECKLIST                    |

## 8.0 Equipment Calibration and Maintenance

General laboratory procedures for the calibration and maintenance of equipment are covered in the ISP Forensic Services Quality/Procedure Manual.

### 8.1 BIOLOGY EQUIPMENT/INSTRUMENTATION

- 8.1.1 Refrigerators and freezers in the biology section (to include the walk-in evidence units) that are used to store evidence or critical reagents will be monitored using a TEMPERATURE MONITORING SYSTEM. The system will send automated email alerts to designated personnel when a unit falls outside of its established critical temperature range. Frost-free freezers may temporarily rise above the high end of the temperature range during its routine defrost cycle. These occurrences typically appear as repetitive temperature spikes within a range of data and may be disregarded. Alerts will be routinely monitored and cleared, as appropriate, with applicable notations made regarding the out of range temperature (e.g. freezer defrost, door opened for extended time, maintenance, etc.). Temperature records are stored indefinitely by the vendor and may be pulled as needed. At least once every two years each refrigerator and freezer temperature monitoring device will either have a calibration check performed by an outside vendor or be replaced.
- 8.1.2 Equipment significant to the examination procedure will be listed on an equipment inventory. Information on the inventory includes (as known or appropriate): equipment identity and its software and firmware, manufacturer's name, model, property number, serial number and/or unique identifier, and location.
- 8.1.3 OPERATING MANUALS for most equipment/instrumentation will be maintained in the product information file (Manuals for the ABI 3500/3500xl Genetic Analyzers, ABI 7500 Real-Time PCR System, Thermal Cyclers, and Driftcon FFC will be maintained in the Amp/PostAmp Room in close proximity to the instruments). Exceptions may be made for manuals referred to for instructions. In these cases, the manual will be maintained in close proximity to the instrument.
- 8.1.4 MAINTENANCE/REPAIR/CALIBRATION LOGS will be maintained as follows:

The records for the ABI 3500/3500xl Genetic Analyzers, ABI 7500 Real-Time PCR System, and Thermal Cyclers will be maintained in the instrument QC binder or an electronic equivalent.

Any equipment/instrumentation function (not documented on weekly, monthly, quarterly, or annual QC Check forms) will be recorded on the Instrument Repair/Maintenance Log. Equipment Failure will also be reported on this form. This form and the QC check forms will be maintained in the section QC Binder or electronic equivalent, except as listed above.

- 8.1.5 EQUIPMENT FAILURE will result in that equipment being 'taken out of service'; an 'out of service' sign will be placed on the equipment and it will not be returned to service until it has passed appropriate performance testing. Actions are reported on the Instrument Repair/Maintenance Log.
- 8.1.6 The SCHEDULE of QC/Performance Checks for both critical and non-critical equipment is as follows:
- 8.1.6.1 WEEKLY (Form 404-QC and 405-QC)  
(once per week with an interval between dates not less than 3 days and not exceeding 10 days)
- Nanopure System Check
  - BSD600 Cleaned
  - Reagent Expiration Check
  - 3500/3500xl Wash Pump and Channels
  - 3500/3500xl Computer/Instrument Restart
  - Hamilton STARlet Weekly Maintenance (Note: Daily maintenance must also be completed within 24 hours of a run. The software will not perform a run unless this requirement is met. This maintenance will be documented on the associated worksheets.)
- 8.1.6.2 MONTHLY (Form 406A/B/C/D-QC)  
(once per calendar month with an interval between dates not less than 15 days and not exceeding 45 days)
- Pipettes Cleaned
  - Centrifuges Cleaned
  - Lab Cleaned
  - Eye Wash Station Check
  - Autoclave Cleaned
  - ABI 7500 Background Assay/Contamination Test, Function Test/Bulb Check, and Disk Cleanup
  - BioRobot EZ1 Advanced XL grease O-rings

- 3500/3500xl Water Trap Flush
- 3500/3500xl Cathode Buffer Septa Changed
- 3500/3500xl plate record cleanout
- Fire Extinguisher Check
- Chemical Shower Check

#### 8.1.6.3 ANNUALLY (Instrument Repair/Maintenance Log)

(once per calendar year with an interval between dates not less than 6 months and not exceeding 18 months) Note: \* denotes critical equipment

- Mechanical Pipette\* Calibration Check (outside vendor)
- Thermal Cycler\* Temperature Verification
- ABI 7500\* Temperature Verification (may be performed by an outside vendor)
- Driftcon FFC Temperature Verification System\* Calibration Check (outside vendor)
- Biological and Chemical Hoods Test (outside vendor)
- ABI 3500/3500xl\* Genetic Analyzer Preventative Maintenance (outside vendor)
- ABI 7500\* Real-Time PCR System Preventative Maintenance (outside vendor)
- ABI 7500\* Pure Dye Calibration, Optical Calibration, and Regions of Interest (ROI's) verification (see 7500 Maintenance Guide for procedures/may be part of PM by request)
- Qiagen BioRobot EZ1 Advanced XL\* Preventative Maintenance (outside vendor)
- Hamilton STARlet\* Preventative Maintenance (outside vendor)
- Microscope Cleaning/Preventative Maintenance (outside vendor)
- Centrifuge Calibration Check (outside vendor)
- Balance Calibration Check (outside vendor)

#### 8.1.6.4 BIENNIALY

(once per two calendar years with interval prior to certificate expiration)

- NIST Traceable Thermometers (outside vendor)
- Refrigerator/Freezer Calibration Check (outside vendor)

#### 8.1.6.5 In addition to the above schedule, personnel should check appropriate parameter function on all instrumentation

with each use [including 3-point calibration of the pH meter (documented on Form 403-QC; the reading must fall with  $\pm 0.50$  pH for the calibration to be confirmed by the meter) and autoclave sterilization], and run a spatial and spectral calibration for the 3500/3500xl Genetic Analyzers as needed or following CCD camera and/or laser replacement/adjustment.

After annual preventative maintenance, the following will be conducted as a verification of performance (documentation will be maintained in the QC binder or electronic equivalent):

- 3500/3500xl: a sensitivity panel (previously characterized DNA) will be run.
- Thermal cycler (including 7500's): the Driftcon FFC will be run on each following repair and prior to being placed back into service. If no repairs were necessary, the pure dye calibration and ROI's will serve as the performance verification for the 7500's following the annual preventative maintenance.
- EZ1XL: an extraction control and reagent blank will be extracted on each instrument, quantitated, and evaluated for the expected concentration per sample type.
- STARlet: A set of standards will be created by the instrument, the plate set up, and then quantitated for evaluation. Alternatively, control samples (ie: a positive and negative) will be set up for amplification by the instrument and run on a CE instrument for evaluation.

A review of the vendor's accreditation will be performed to verify their competency within the scope of the service provided.

Documentation of the review will be maintained in the appropriate QC binder or electronic equivalent.

Any problems noted with laboratory equipment, during normal usage or as part of a QC check should be brought to the attention of the Technical Lead and necessary supervisory personnel and documented on the Instrument Repair/Maintenance Log and/or the respective QC form.

Heat blocks/thermal mixers used as part of an analytical procedure (i.e. DNA lysis) are considered critical equipment and will be monitored with traceable thermometers.

POP may be left on the 3500/3500XL instruments for up to 30 days. If the POP is removed from the instrument for storage, a Pouch Cap will be placed onto the pouch, and it will be stored at 2-8°C until ready for use. Either an empty POP4 pouch or conditioning reagent will be placed on the connector to prevent desiccation of any residual polymer on the connector while the POP is in storage.

The 3500/3500XL capillary arrays may be used past their recommended injection number and expiration date. The array will be changed at minimum when the separation of the alleles on the ladder with single bp difference (i.e. TH01 9.3, 10) are no longer clearly separated.

ARCHIVED

## 9.0 Chemicals, Supplies, and Reagent Preparation

General laboratory policies and procedures regarding the purchase of chemicals and preparation of reagents are covered in the ISP Forensic Services Quality/Procedure Manual.

### 9.1 COMMERCIALY PURCHASED CHEMICALS

- 9.1.1 Biology Personnel should consult the section's reagent inventory and the forensic services supplies/services list prior to ordering. Chemical grade requirements should be checked and ordered as appropriate. Note: An order form/document must be filled out prior to placing the order.
- 9.1.2 Upon receipt of a chemical or reagent, the reagent inventory should be updated to reflect the new lot number and expiration date. The reagent(s) will be marked with the date received and the individual's initials. If it is an outer container that the chemical/kit remains in until use, the inner container will be labeled with this information either when received or when removed for use. HiDi Formamide does not have manufacturer expiration date. This reagent will additionally be labeled with a laboratory assigned expiration date of 2 years from the date of receipt. Packing slips should be checked to ensure appropriate accounting, including proper reagent grade, where applicable (this will be indicated by dating and initialing the packing slip and making notations as necessary). The packing slip and corresponding order document will be retained in the biology section. If an MSDS sheet came with the chemical, the MSDS binder and/or electronic MSDS folder should be checked for the presence of an MSDS for that chemical. If one exists, no additional copy is kept; however, if a newer version is received, the old one should be replaced. If one does not already exist, place the one received in the binder/folder. For chemicals without a hard copy MSDS, the manufacturer or one of the following websites may be consulted for information as needed:
- <http://www.msds.com>  
<http://www.ilpi.com/msds/>
- 9.1.3 Expired chemicals will be disposed of in an appropriate manner.

## 9.2 REAGENTS PREPARED IN-HOUSE

- 9.2.1 All biology reagents will be made with great care, following all quality and safety procedures. A mask will be worn by analysts during reagent preparation to help avoid the potential for contamination. See 9.4 and 9.5 below for individual reagent recipes.
- 9.2.2 Each reagent has a corresponding form to document the making of the reagent and components used. This form or an electronic equivalent in ILIMS must be filled out. A reagent label must be made that has the reagent name, the lab lot number (which consists of the first few letters of the reagent name followed by the date prepared, in the form 'MMDDYY'), and the preparer's initials. The NFPA designation will be completed on all labels. Refillable squirt-bottles of water or ethanol will be labeled but need not bear dates or initials (unless dilutions are made, i.e.: 70% ethanol).
- 9.2.3 An effort should be made to use in-house reagents within one year of preparation; however, they do not expire and may continue to be used beyond the one year timeframe.

## 9.3 CRITICAL REAGENTS

The reagents listed below have been identified as critical in Forensic Biology/DNA and will be stored according to the manufacturer's guidelines. These reagents must undergo a QC ASSAY BEFORE use on forensic casework and/or Convicted Offender samples. QC instructions and passing criteria are detailed below. Reagents received at a later date but having the same lot number as those previously tested and determined acceptable need not have a QC check performed. Critical Reagents (in addition to other DNA-related reagents with manufacturer expiration dates) may be used beyond the listed expiration date for training purposes without any further testing, so long as expected results are obtained for all associated controls. The reagent must be labeled 'for training only' if it is to be retained once the expiration date has been reached.

### 9.3.1 ABACARD® HEMATRACE® TEST KIT

- 9.3.1.1 Perform test as usual with one ~2mm<sup>2</sup> cutting and one ~2mm thread from known bloodstain. Record results (include time it took for positive reaction to be visible).
- 9.3.1.2 The ~2mm<sup>2</sup> cutting sample must have a positive reaction within 10 minutes for passing. The ~2mm thread should ideally be positive within 10 minutes but is used primarily as a sensitivity indicator of the given test lot. The kit may still be deemed as passing without a positive result for the thread.
- 9.3.1.3 The QC will be documented on Form 410-QC or electronic equivalent in ILIMS.

### 9.3.2 OneStep ABACARD® p30 TEST KIT

- 9.3.2.1 Perform test as usual with a known semen extract, as well as ~10ng/ml (10µl of a 1:500 dilution) and ~50ng/ml (10µl of a 1:100 dilution) of Seri Semen Standard. Record results (include time it took for positive reaction to be visible).
- 9.3.2.2 The semen extract must have a positive reaction within 10 minutes for passing. The Seri standards are used to estimate the range of sensitivity of the kit lot.
- 9.3.2.3 For the semen standard dilutions, if a positive reaction is not obtained at 10 minutes, continue to monitor and record result at the end of 15 minutes. In addition, run a 250ng/ml (50µl of the 1:100 dilution to 150µl of extraction buffer) or a 1:10 dilution of the semen stain extract to ensure the kit is operating within reasonable limits for forensic identification. In addition to the neat semen extract, this control sample (250ng/ml or 1:10 extract) must result in a positive reaction within 10 minutes.
- 9.3.2.4 The QC will be documented on Form 412-QC or electronic equivalent in ILIMS.

### 9.3.3 SwabSolution™ and 5X AmpSolution™ Reagents

These instructions are for the QC check of SwabSolution™ and 5X AmpSolution™ as used in the method for Detection of Male DNA on Sexual Assault Kit Evidence (BI-132). This may include the complete SwabSolution™ kit which contains both the SwabSolution™ reagent and 5X AmpSolution™ reagent and/or supplemental 5X AmpSolution™ reagent purchased separately from the kit if the lot # (as listed on the tube) is different from the 5X AmpSolution™ reagent in the kit. See 9.3.5.2 for QC of 5X AmpSolution™ used in direct amplification of offender samples and/or direct amplification of casework reference samples.

- 9.3.3.1 To check the new lot(s), perform DNA quantitation as usual using either BI-132 or BI-207. Include a NIST traceable sample of male origin and a reagent blank in which the lysates were prepared according to BI-132 using the new lot(s) of SwabSolution™ reagent and/or 5X AmpSolution™ reagent for these samples. Record the autosomal and male DNA quantitation results (ng/µl) for both the sample and reagent blank. The NIST traceable sample should yield a male DNA result comparable to the autosomal DNA result and be consistent with expectations for the type/amount of sample. The reagent blank should be absent of true

- detectable signal for both the male and autosomal targets. Obtaining the expected results will constitute a pass.
- 9.3.3.2 The QC will be documented on Form 418-QC or electronic equivalent in ILIMS. If applicable, attach the 7500 Load Sheets, Standard Curves, and Results Sheets.
- 9.3.4 Plexor® HY System Kit
- 9.3.4.1 To check the new kit lot, perform quantitation as usual with a 1:10 dilution of the PowerQuant® Male gDNA Standard as sample. A pass will be achieved when the standard curve slopes are within the acceptable range and gDNA quant values are near the expected value. The sample should yield a male DNA result comparable to the autosomal DNA result. Record the slopes obtained for the standard curves, as well as the autosomal and male quantitation results.
- 9.3.4.2 The QC will be documented on Form 419-QC or electronic equivalent in ILIMS. If applicable, attach the 7500 Load Sheets, Standard Curves, and Results Sheets.
- 9.3.5 PowerPlex® Fusion 6C System Kit
- 9.3.5.1 A positive and negative control from the kit undergoing the QC are to be processed from amplification. A pass will be achieved by obtaining the expected results for each of the samples run and data of acceptable quality (e.g. sufficient RFUs). Comments regarding quality concerns are to be noted as appropriate.
- 9.3.5.2 If the kit is to be used for the direct amplification of offender or casework reference samples the direct amplification method is to be followed for the QC including the addition of 5X AmpSolution™ (and PunchSolution™ for offender samples and SwabSolution™ for casework references).
- 9.3.5.3 The QC will be documented on Form 420A-QC, or electronic equivalent in ILIMS.
- 9.3.6 PowerPlex® Y23 System Kit
- 9.3.6.1 A male positive and negative control from the kit undergoing the QC are to be processed from amplification. A pass will be achieved by obtaining the expected results for each of the samples run and data of acceptable quality (e.g. sufficient RFUs). Comments regarding quality concerns are to be noted as appropriate.
- 9.3.6.2 If the kit is to be used for the direct amplification of casework reference samples, the direct amplification

method is to be followed for the QC including the addition of 5X AmpSolution™ and SwabSolution™.

9.3.6.3 The QC will be documented on Form 421-QC or electronic equivalent in ILIMS.

#### 9.4 BIOLOGICAL SCREENING REAGENTS

##### 9.4.1 Phenolphthalein (Kastle-Meyer) Reagent

(NFPA: health 3, flammability 1, reactivity 2)

May be a commercial purchase.

|                 |       |
|-----------------|-------|
| Phenolphthalein | 2.0g  |
| KOH             | 20.0g |
| Zinc (granular) | 20.0g |

Phenolphthalein, KOH, and 100ml of dH<sub>2</sub>O are refluxed, in a fume hood, with Zinc until solution is colorless (producing phenolphthalin in ~4 hours). Store stock solution refrigerated in dark bottle to which ~5g mossy zinc has been added to keep the solution in its reduced form. Remove for working solution as needed.

Working solution: Mix 2ml stock solution with 8ml Ethanol

Caution: Zinc is flammable. The unreacted portions and used filter paper are to be disposed of properly.

##### 9.4.2 Hydrogen Peroxide 3% (v/v)

(NFPA: health 0, flammability 0, reactivity 1)

Generally a commercial purchase, however, may be made from a 30% Solution (which is a commercial purchase) as follows:

|                         |                                         |
|-------------------------|-----------------------------------------|
| Hydrogen Peroxide (30%) | 2.5ml/22.5ml nanopure dH <sub>2</sub> O |
|-------------------------|-----------------------------------------|

Mix the H<sub>2</sub>O<sub>2</sub> with 22.5ml of nanopure dH<sub>2</sub>O and store at ~4°C.

##### 9.4.3 Ortho-Tolidine Reagent

(NFPA: health 3, flammability 1, reactivity 2)

|                     |        |
|---------------------|--------|
| O-Tolidine          | 0.075g |
| Glacial Acetic Acid | 12.5ml |
| Ethanol             | 12.5ml |

Dissolve O-tolidine in Acetic Acid/Ethanol mixture consistent with ratios above. O-tolidine is light sensitive and should be stored in dark reagent bottle and kept refrigerated when not in use.

9.4.4 Ammonium Hydroxide (~3%)  
(NFPA: health 3, flammability 1, reactivity 2)

Ammonium Hydroxide (Concentrated ~30%) 10ml/100ml

Add the  $\text{NH}_4\text{OH}$  to 90ml of nanopure  $\text{dH}_2\text{O}$ , mix well and store at RT.

9.4.5 Brentamine Solution A  
(NFPA: health 3, flammability 2, reactivity 1)

|                                        |        |
|----------------------------------------|--------|
| O-Dianisidine Diazotized (Fast Blue B) | 0.25 g |
| Sodium Acetate Trihydrate              | 5.0 g  |
| Glacial Acetic Acid                    | 2.5 ml |
| Nanopure $\text{dH}_2\text{O}$         | 25 ml  |

Mix thoroughly and store refrigerated.

9.4.6 Brentamine Solution B  
(NFPA: health 0, flammability 0, reactivity 0)

$\alpha$ -Naphthyl Phosphate (Monosodium Salt) 0.4 g

Dissolve in 2.5 ml of nanopure  $\text{dH}_2\text{O}$ . Store refrigerated.

Working Solution: 4.5 mL nanopure  $\text{dH}_2\text{O}$  + 500  $\mu\text{l}$  Brentamine Solution A + 1 drop of Brentamine Solution B

9.4.7 Saline (0.85% NaCl)  
(NFPA: health 1, flammability 0, reactivity 0)

NaCl 4.25g/500ml

Dissolve the NaCl in 500 ml nanopure water. Sterilize by autoclaving. Store refrigerated.

9.4.8 1X Phosphate Buffered Saline (PBS)  
(NFPA: health 1, flammability 0, reactivity 1)

PBS 1 commercial pre-made packet

Dissolve one packet of powdered PBS in 1l of nanopure dH<sub>2</sub>O. Check that pH $\approx$ 7.4, autoclave and store at RT.

If pre-made packets are not available, PBS may be prepared by dissolving 0.2g KCl, 8.0g NaCl, 0.2g KH<sub>2</sub>PO<sub>4</sub>, and 2.2g Na<sub>2</sub>HPO<sub>4</sub>·7H<sub>2</sub>O (or 1.1g Na<sub>2</sub>HPO<sub>4</sub> anhydrous) in 800ml nanopure dH<sub>2</sub>O. Adjust pH to 7.4 if necessary. Q.S. to 1l with nanopure dH<sub>2</sub>O, autoclave and store at RT.

9.4.9 X-mas Tree Stain Solution A (Kernechtrot Solution)  
(NFPA: health 1, flammability 0, reactivity 0)

May be a commercial purchase.

|                  |      |
|------------------|------|
| Aluminum Sulfate | 5g   |
| Nuclear Fast Red | 0.1g |

For 100ml, Dissolve the Aluminum Sulfate in 100ml HOT nanopure dH<sub>2</sub>O. Immediately add the Nuclear Fast Red, mix, cool and filter (paper or  $\geq 45\mu\text{m}$ ). May be stored at RT.

9.4.10 X-mas Tree Stain Solution B (Picroindigocarmine Solution)  
(NFPA: health 2, flammability 2, reactivity 2)

May be a commercial purchase.

|                                |       |
|--------------------------------|-------|
| Saturated Picric Acid Solution | 100ml |
| Indigo Carmine                 | 0.33g |

For 100ml, dissolve the Indigo Carmine in 100ml of the Picric Acid. Mix and filter (paper or  $\geq 45\mu\text{m}$ ). May be stored at RT.

9.4.11 Mercuric Chloride 10% (w/v)  
(NFPA: health 4, flammability 0, reactivity 1)

|                   |                    |
|-------------------|--------------------|
| Mercuric Chloride | 10g/100ml 95% EtOH |
|-------------------|--------------------|

Dissolve the Mercuric Chloride in 100ml of 95% Ethanol, mix well and store at RT.

9.4.12 Zinc Chloride 10% (w/v)  
(NFPA: health 2, flammability 0, reactivity 2)

Zinc Chloride

1g/10ml 95% EtOH

Dissolve the Zinc Chloride in 10ml of 95% Ethanol, mix well and store at RT.

## 9.5 DNA REAGENTS

### 9.5.1 1M Tris-HCl Buffer pH 7.5

(NFPA: health 2, flammability 1, reactivity 1)

Tris Base(tris[Hydroxymethyl]amino methane) 6.0 g

Dissolve Tris in ~40 mL nanopure dH<sub>2</sub>O. Adjust to pH7.5 at RT by adding concentrated HCl (approximately 3.25 mL). Q.S. to 50 mL with nanopure dH<sub>2</sub>O, autoclave and store at RT.

### 9.5.2 1M Tris-HCl Buffer pH 8

(NFPA: health 2, flammability 1, reactivity 1)

Tris Base(tris[Hydroxymethyl]amino methane) 6.0 g

Dissolve Tris in ~40 mL nanopure dH<sub>2</sub>O. Adjust to pH8 at RT by adding concentrated HCl (approximately 2.25 mL). Q.S. to 50 mL with nanopure dH<sub>2</sub>O, autoclave and store at RT.

### 9.5.3 0.5M Ethylenediamine Tetraacetic Acid (EDTA)

(NFPA: health 1, flammability 1, reactivity 0)

Na<sub>2</sub>EDTA·2H<sub>2</sub>O 9.3 g

Slowly add EDTA to 40 mL nanopure H<sub>2</sub>O while stirring vigorously. Add ~1 g of NaOH pellets to bring the pH to near 8.0. When fully dissolved adjust pH to 8.0 and bring final volume to 50 mL. Autoclave and store at RT.

Note: EDTA will not go into solution without the pH adjustment.

### 9.5.4 Stain Extraction Buffer pH8 (10mM EDTA/10mM Tris-HCl/50mM NaCl/2% SDS)

(NFPA: health 2, flammability 1, reactivity 1)

1M Tris-HCl, pH7.5 5ml

|           |       |
|-----------|-------|
| 0.5M EDTA | 10ml  |
| 5.0M NaCl | 5ml   |
| 10% SDS   | 100ml |

Mix the Tris-HCl, EDTA, NaCl and SDS with ~380ml nanopure dH<sub>2</sub>O. Store at RT.

Note: Reagent contains SDS, do not autoclave.

#### 9.5.5 Proteinase K (20mg/ml) (NFPA: health 1, flammability 1, reactivity 0)

May be a commercial purchase of 20mg/ml solution.

|              |      |
|--------------|------|
| Proteinase K | 0.2g |
|--------------|------|

Dissolve the ProK in 10ml sterile nanopure dH<sub>2</sub>O.

Dispense ~500µl (commercial purchase or in-house prep.) each into sterile microfuge tubes and store at  $\cong 20^{\circ}\text{C}$ .

#### 9.5.6 1M Sodium Acetate pH 5.2 (NFPA: health 3, flammability 2, reactivity 0)

|                                         |       |
|-----------------------------------------|-------|
| CH <sub>3</sub> COONa·3H <sub>2</sub> O | 13.6g |
|-----------------------------------------|-------|

Dissolve the CH<sub>3</sub>COONa·3H<sub>2</sub>O in 80ml nanopure dH<sub>2</sub>O. Adjust to pH 5.2 by adding glacial acetic acid (approximately 2ml). Q.S. to 100ml with nanopure dH<sub>2</sub>O, autoclave and store at RT.

#### 9.5.7 DTT Solution (NFPA: health 2, flammability 1, reactivity 0)

|                      |       |
|----------------------|-------|
| Dithiothreitol (DTT) | 0.77g |
|----------------------|-------|

Dissolve the DTT in 5ml nanopure dH<sub>2</sub>O. Add 50µl 1M Sodium Acetate, pH5.2. Dispense ~500µl each into sterile microcentrifuge tubes and store at  $\cong 20^{\circ}\text{C}$ .

Note: Do not autoclave.

#### 9.5.8 PCR-TE (TE<sup>-4</sup>) Buffer (10mM Tris-HCl/0.1mM EDTA) (NFPA: health 2, flammability 1, reactivity 1)

|                  |      |
|------------------|------|
| 1M Tris-HCl, pH8 | 10ml |
|------------------|------|

0.5M EDTA, pH8

0.2ml

Mix Tris-HCl and EDTA with 990ml nanopure dH<sub>2</sub>O. Autoclave and store at RT.

9.5.9 5N Sodium Hydroxide  
(NFPA: health 3, flammability 0, reactivity 2)

NaOH 50g

Slowly dissolve the Sodium Hydroxide in 250ml sterile nanopure dH<sub>2</sub>O. Allow to cool and store at RT.

Caution: NaOH is highly caustic. This reaction generates heat.

9.5.10 5M Sodium Chloride  
(NFPA: health 1, flammability 0, reactivity 0)

May be a commercial purchase of 5M solution.

NaCl 146.1g/500ml

Dissolve the NaCl in 500 ml nanopure water. Sterilize by autoclaving.

## 10.0 Documentation and Report Writing

Meticulous documentation is an important aspect of forensic work. In casework, the scientist's knowledge of case circumstance (and therefore their ability to discern potential significance) may be limited. It is also common to be called upon to testify months, or even years, after processing evidence for a given case. Careful observation and detailed note-taking will not only refresh the scientist's memory and provide support for the conclusion in the laboratory report, but might also provide additional information not thought to have been important at the time of evidence processing. General laboratory policies regarding release of results/confidentiality, case record, and retention are described in the ISP Forensic Services Quality/Procedure Manual.

### 10.1 CASE NOTES

- 10.1.1 Notes or worksheets prepared external to ILIMS (e.g. photos, sketches, DNA worksheets, electropherograms, etc.) will bear the date and scientist's initials or electronic user ID. These will be attached to ILIMS. Following approval of the assignment, the attached version will be considered the 'original' documentation and any hard copies will be destroyed.
- 10.1.2 Each page of the ILIMS generated note packet will have the laboratory case number, scientist's initials and page number (in a form indicating page/total pages). Additionally, dates of analyses will be reflected throughout.
- 10.1.3 In accordance with the ISP Forensic Services Quality/Procedure Manual, if an item of evidence is checked out and opened, a report will be issued describing what was done; all items checked out at that time by that analyst associated with that assignment will be listed on that report.  

Note: Items must also be taken into an analyst's custody to create a subitem.
- 10.1.4 Evidence packaging will be documented for all items and will include the type of package and seal, whether the seal is initialed, labeling, and condition of package/seal if appropriate (e.g. torn, leaking, partially open, etc.). Differences between evidence descriptions on a package, in ILIMS and/or what the evidence is once opened should be noted.
- 10.1.5 Whenever feasible, every attempt should be made to gain entry into the evidence without breaking the original seals. Any seal altered or created by a scientist will bear their initials and date across the seal.
- 10.1.6 Evidence descriptions may include, as appropriate and necessary for identification, colors, sizes (measurements where appropriate- e.g.,

- knife and blade), manufacturer, model, brand, serial numbers or other identifiers.
- 10.1.7 Evidence numbering must be unique for the purpose of possible later CODIS entry and chain of custody tracking. Items should be numbered as follows:

A single item (e.g., a baseball cap; Item 57) for which:

1 area/stain portion is removed for DNA testing  $\equiv$  Item 57.1

2 or more area/stain portions removed for DNA testing)  $\equiv$  Item 57.1, Item 57.2, Item 57.3 etc.

An item with multiple sub-items (e.g., a SAECK; Item 1)

$\equiv$  Item 1.1, Item 1.2, Item 1.3, etc.

Multiple items (Item 25) packaged together (e.g. shirt and pants)

$\equiv$  Item 25.1 (shirt) and Item 25.2 (pants)

Area/stain portions removed for DNA testing  $\equiv$  Item 25.1.1, Item 25.1.2, Item 25.2.1 etc.

- 10.1.8 Photography, digital or otherwise, is often useful in documenting the appearance of evidence items. Digital photographs used for case notes will be attached to ILIMS. Images requiring editing (e.g. contrast or brightness) will be transferred to and stored within the Digital Imaging System; refer to BI-119 for instructions. These images may be printed and scanned to ILIMS or attached as an electronic file for case notes, as necessary.
- 10.1.9 The casework note packet is considered complete when the analyst submits the assignment to be reviewed. Electronic documentation external to ILIMS (e.g. electropherograms and statistics) is considered stored at this time. Any changes to the electronic data required after this point will be documented on the original copy, initialed, dated, and attached to ILIMS. If the change requires reprinting, a notation on the new copy will be made as to the changes made. The new printed copy will bear the date the changes were made/reprinted and will be attached to ILIMS. The original attached document will remain in ILIMS but will be unchecked for inclusion in the note packet.

## 10.2 DATABASE PACKETS

- 10.2.1 Each page of the database packet should have the following: plate identifier, date, scientist's initials, and page number (in a form indicating page/total pages).
- 10.2.2 In general, database packets will be arranged from front to back with chronological worksheets then reinjection summary.
- 10.2.3 The database note packet is considered complete when the analyst submits the packet to be reviewed. Electronic documentation is considered stored at this time. If changes to the GeneMapper IDX electronic data are required after this point (on or after the review date documented in the notes), the analyst will re-export the GeneMapper IDX project so the updated project reflects when the changes were made.

### 10.3 CASEWORK REPORTS

- 10.3.1 The report will contain the title Forensic Biology Report, Forensic DNA Report, or CODIS Search Report as appropriate.
- 10.3.2 For clarity, when a statement(s) is about a particular Item (or multiple items listed individually), the "I" will be capitalized as in a name. When writing in general terms (i.e., the following items:) the "i" will remain lowercase.
- 10.3.3 The case submission information will include: laboratory case #, agency, agency case #, offense date(s), investigating officer, report #, evidence submission date(s), analyst, and principals (victim, suspect, etc.)
- 10.3.4 The body of the report will be separated from the case submission information by the following headings: Evidence Description, Conclusions and Interpretations, Notes, Further Actions, and Disposition of Evidence. Headers for each evidence item will typically be included in the Conclusions and Interpretations section, with their corresponding conclusion(s). Exceptions may be made (i.e.: paternity and CODIS reports).
- 10.3.5 The following results/conclusions statements should be used in a biology screening report, as dictated by the analysis findings. However, they may not cover all scenarios encountered during forensic DNA casework. Situations not covered by the standard method will be handled on a case by case basis upon consultation with the DNA Technical Lead. Where appropriate, descriptions, quantity, and/or locations of individual stains may be included in the corresponding statements. Portions of individual statements may be combined as needed.

#### 10.3.5.1 Semen Results/Conclusions Statements:

Chemical and microscopic analyses for the detection of semen were conducted on this item. Semen was confirmed by the presence of spermatozoa on this item. (or) Semen was not detected on this item. (or) No identifiable spermatozoa were observed on this item.

Chemical and microscopic analyses for the detection of semen were conducted on this item. Semen was confirmed on this item by the presence of a single spermatozoon (or limited [ $\leq 3$ ] number of spermatozoa), which may be insufficient for further testing at this time.

Chemical, microscopic, and serological analyses for the detection of semen were conducted on this item. Semen was indicated on this item by the presence of the semen specific protein, p30; however, no spermatozoa were observed, which may be insufficient for further testing at this time. (or) Semen was not detected on this item.

Results from presumptive chemical tests for the presence of semen were negative on this item.

#### 10.3.5.2 Blood Results/Conclusion Statements:

Results from chemical and serological tests performed on this item indicated the presence of human blood.

Results from chemical and serological tests performed on this item did not indicate the presence of human blood.

Results from presumptive chemical tests performed on this item indicated the presence of blood.

Due to possible interference from the substrate, results from presumptive chemical tests performed on this item were inconclusive for the presence of blood.

Results from presumptive chemical tests performed on this item indicated the presence of blood. However, based on the visual

appearance of the stain(s), results may be due to a non-specific reaction.

Results from presumptive chemical tests for the presence of blood were negative on this item.

Notes Section: The test for human blood, HemaTrace™, cross-reacts with upper primates and members of the genus Mustela; examples of which include the long-tailed weasel and domestic ferret.

#### 10.3.5.3 Saliva Results/Conclusions Statements:

Results from chemical tests performed on this item indicated (or did not indicate, or were inconclusive for) the presence of amylase, a component of saliva.

#### 10.3.5.4 Urine Results/Conclusions Statements:

Results from presumptive chemical tests performed on this item indicated (or did not indicate or were inconclusive for) the presence of urine.

#### 10.3.5.5 Feces Results/Conclusions Statements:

Results from presumptive chemical tests performed on this item indicated (or did not indicate or were inconclusive for) the presence of feces.

#### 10.3.5.6 Hair Results/Conclusions Statements:

This item was examined microscopically and found to contain an apparent hair with no root, which is insufficient for nuclear DNA analysis.

This item was examined microscopically and found to contain a hair with a root which is likely insufficient for nuclear DNA analysis.

This item was examined microscopically and found to contain a hair with a root which may be sufficient for nuclear DNA analysis.

#### 10.3.5.7 Male DNA Results/Conclusions Statements:

Male DNA was detected on this item.

A low level of male DNA was detected on this item, which may be insufficient for further testing at this time. (Applicable when Y quantitation result is less than 0.01ng/μL.)

Due to possible inhibition and/or non-specific amplification it is inconclusive as to whether or not male DNA is present on this item.

No male DNA was detected on this item.

Due to low level results, it is inconclusive as to whether or not male DNA is present on this item.

#### Notes Section:

Deoxyribonucleic Acid (DNA) quantitation, employing real-time Polymerase Chain Reaction (PCR), was performed on (items).

#### 10.3.5.8 Further Actions Statements:

For questions regarding the content of this report, please contact the analyst named below or the DNA section of the laboratory.

If additional testing is desired, please contact the laboratory.

If DNA testing is desired, known reference samples from (names) are required. Please contact the laboratory regarding the resubmission of evidence.

A known reference sample from (name) is required for DNA comparison. Please contact the laboratory regarding the status of this known reference sample.

Documentation authorizing consumption of (items) must be submitted prior to DNA testing. Please contact the laboratory regarding the analysis request.

If Y-STR testing is desired, a known reference sample from [NAME] is required. Please contact the laboratory prior to resubmission.

- 10.3.6 The following results/conclusions statements should be used in an STR DNA Report (refer to BI-214 of Biology/DNA Casework Analytical Methods for Y-STR reporting statements). However, they may not cover all scenarios encountered during forensic DNA casework. Situations not covered by the standard method will be handled on a case by case basis upon consultation with the DNA Technical Lead.

Note: Associations deemed to be probative based on sample type and case circumstances will be qualified with a statistic. Probative associations for multiple items or profiles may be qualified with a single statement if the statistics are nearly identical. There will be slight variation in the likelihood ratio statistic calculated by STRmix™ even when the DNA profile is the same. If more than one of the same full single source profile is obtained, it is only necessary to calculate one likelihood ratio in STRmix™ (generally for the profile with the lowest overall peak heights). A qualitative statement clearly expressing the significance of the association may be used in situations where the presence of an individual's DNA on an item is reasonably expected.

The epithelial cell fraction of intimate samples, such as vaginal/rectal swabs, etc., and the sperm cell fraction of penile swabs, are not considered probative if the testing results in a single profile matching the individual from which the sample was collected or if the results do not provide any additional probative information from that of the other cell fraction (e.g. minor component consistent with semen donor on vaginal/rectal, etc. swab, or the epithelial donor on a penile swab). In these instances, a statement regarding the DNA source of the non-probative fraction is not required.

Statistics will typically be included using charts:

| Inclusion Chart: |                       |                  |
|------------------|-----------------------|------------------|
| Name             | Likelihood Ratio (LR) | Level of Support |
|                  |                       |                  |

| Exclusion Chart: |                           |                  |
|------------------|---------------------------|------------------|
| Name             | 1/Likelihood Ratio (1/LR) | Level of Support |
|                  |                           |                  |

Exceptions may be made on a case-by-case basis (i.e.: paternity reports). LRs reported will be the most conservative of the population groups calculated.

#### 10.3.6.1 Profile match statement for single source:

The DNA profile obtained from this item matches that obtained from the known reference sample (or reference oral swab/sample, etc.) of/from "name". This DNA profile is at least (LR) times more likely to be seen if (NAME) is the source than if an unrelated individual randomly selected from the general population is the source.

The DNA profile obtained from this item is consistent with that obtained from the known reference sample (or reference oral swab/sample, etc.) of/from "name".

Note: Qualitative statements such as "consistent with" without an associated statistic will only be used for non-probative associations or associations between evidence items.

#### 10.3.6.2 Partial Profile Statement [profile consistent with item(s) in match statement above]:

The DNA profile obtained from this item also matches that obtained from the known reference sample of "name", however less genetic information was obtained. This partial DNA profile is at least (LR) times more likely to be seen if (NAME) is the source than if an unrelated individual randomly selected from the general population is the source.

The partial DNA profile obtained from this item is consistent with that obtained from the known reference sample of "name". This

partial DNA profile is at least (LR) times more likely to be seen if (NAME) is the source than if an unrelated individual randomly selected from the general population is the source.

#### 10.3.6.3 Positive Paternity Statement [profiles consistent with being a biological child]:

Based upon evaluation of the DNA profiles obtained from the above individuals, "name" cannot be excluded as being the biological father of "name". The probability of paternity (assuming a prior probability of 0.5) is "X%" relative to an unrelated man randomly selected from the general population. The combined paternity index for the loci examined is "X". At least "X%" of the male population would be expected to be excluded from the possibility of being the biological father of "name".

Note: The most conservative of the population groups calculated is reported for the statement above.

Relationship statistics are truncated to the fourth digit after the decimal (i.e. 99.9998%).

#### 10.3.6.4 Positive Kinship Statement

Based upon evaluation of the DNA profiles obtained from the above individuals, "name" cannot be excluded as being the biological father (or mother) of "name". The single parentage index for the loci examined is "X". At least "X%" of the male (or female) population would be expected to be excluded from the possibility of being the biological father (or mother) of "name".

#### 10.3.6.5 Mixture Statements:

The DNA profile from this item indicates a mixture of DNA. "Name" is a potential contributor to this mixture. Assuming a two person mixture, this DNA profile is at least (LR) times more likely to be seen if it were the result of a mixture of DNA from (NAME) and an unrelated, randomly selected individual than if it resulted from two unrelated individuals randomly selected from the general population.

The DNA profile from this item indicates a mixture of DNA. "Names" are potential contributors to this mixture. Assuming a two person mixture and that "name" is a contributor, this DNA profile is at least (LR) times more likely to be seen if it were the

result of a mixture of DNA from “name” and (NAME) than if it resulted from “name” and an unrelated individual randomly selected from the general population.

The DNA profile from this item indicates a mixture of DNA. “Names” are potential contributors to this mixture. Assuming a two person mixture, this DNA profile is at least (LR) times more likely to be seen if it were the result of a mixture of DNA from (NAME 1) and (NAME 2) than if it resulted from two unrelated individuals randomly selected from the general population.

The DNA profile obtained from this item indicates a mixture of DNA with a major profile, which matches that obtained from the known reference sample of “name”. Assuming the mixture is from two individuals, this DNA profile is at least (LR) times more likely to be seen if it were the result of a mixture of DNA from (NAME) and an unrelated, randomly selected individual than if it resulted from two unrelated individuals randomly selected from the general population.

Note: Descriptive terms such as major and/or minor may be used to qualitatively describe an individual’s relative contribution level to a mixture; however, the likelihood ratio applies to the mixture as a whole rather than a component of the mixture and will be stated as such.

The above examples contain wording for two person mixtures; however, the general format applies to mixtures of greater than two individuals.

In general, the likelihood ratio(s) reported will be the most reasonable based on the information available to the DNA analyst at the time of analysis. The use of alternate propositions and/or assumptions will result in different likelihood ratio statistics. Additional calculations may be performed as appropriate upon request. Relatedness likelihood ratios are automatically calculated in STRmix™ for each proposed hypothesis. These statements (10.3.6.7) will only be provided when

the alternate proposition is regarding an untested relative of the defendant, and only upon request.

10.3.6.6 Relatedness Statements [profile proposed to be from an untested relative]

The DNA profile obtained from this item matches that obtained from the known reference sample (or reference oral swab/sample, etc.) of/from "name". This DNA profile is at least (LR) times more likely to be seen if (NAME) is the source than if an untested (relationship) is the source.

The DNA profile obtained from this item matches that obtained from the known reference sample (or reference oral swab/sample, etc.) of/from "name". This DNA profile is at least (LR) times more likely to be seen if (NAME) is the source than if an unknown individual randomly selected from the general population is the source, to include the possibility of biological relatives of "name".

The DNA profile from this item indicates a mixture of DNA. "Names" are potential contributors to this mixture. Assuming the mixture is from two individuals and that "name" is a contributor, this DNA profile is at least (LR) times more likely to be seen if it were the result of a mixture of DNA from "name" and (NAME) than if it resulted from "name" and an untested (relationship) of "name".

The DNA profile from this item indicates a mixture of DNA. "Names" are potential contributors to this mixture. Assuming the mixture is from two individuals and that "name" is a contributor, this DNA profile is at least (LR) times more likely to be seen if it were the result of a mixture of DNA from "name" and (NAME) than if it resulted from "name" and an unknown individual randomly selected from the general population, to include the possibility of biological relatives of "name".

Additional partial profile or mixture statements may also be used following the format in 10.3.7.3 and 10.3.7.6 and substituting the relatedness portion of the wording as above.

#### 10.3.6.7 Exclusionary Statements:

The DNA profile obtained from this item does not match that obtained from the known reference sample of "name". (NAME) is not the source of DNA on this item.

The DNA profile obtained from this item was determined to be from an unknown male/female. (NAME) is not the source of DNA on this item.

(NAME) is excluded as being a contributor to the mixture of DNA obtained from this item.

Based upon evaluation of the DNA profiles obtained from the above individuals, "name" is not the biological father of "name".

#### 10.3.6.8 Inconclusive/Uninterpretable Statements

Based on the likelihood ratio result (LR = actual [smallest value of the population groups calculated] likelihood ratio), it is inconclusive whether "name" is a possible contributor to this DNA profile. (NOTE: for comparisons to previous Fusion 5C or PP16/PP16HS evidence data)

The DNA profile from this item indicates a mixture of DNA from at least five people (, at least # of which are male). Due to the number of contributors and complexity of this mixture, no additional conclusions can be made.

Due to insufficient quantity or degradation, only a partial DNA profile was obtained from this item. Due to the low level results and limited data, no conclusions can be made.

#### 10.3.6.9 No DNA Profile Obtained/Insufficient Male DNA Statements:

Due to insufficient quantity or degradation, no DNA profile was obtained from this item.

(A low level of) Male DNA was detected on this item; however, due to a high level of female DNA detected, this item is not suitable for STR analysis. No further testing was conducted.

No male DNA was detected on this item. No further testing was conducted.

#### 10.3.6.10 Notes:

Deoxyribonucleic Acid (DNA) Analysis, employing the Polymerase Chain Reaction (PCR), was used to generate a Short Tandem Repeat (STR) profile from a portion of Items (item numbers).

Loci Examined: Amelogenin, D3S1358, D1S1656, D2S441, D10S1248, D13S317, Penta E, D16S539, D18S51, D2S1338, CSF1PO, Penta D, TH01, vWA, D21S11, D7S820, D5S818, TPOX, D8S1179, D12S391, D19S433, SE33, D22S1045, DYS391, FGA, DYS576, and DYS570.

A differential extraction was performed on samples with results from Fractions 1 and/or 2, which is intended to separate sperm cells from non-sperm cells. However microscopic confirmation of spermatozoa was not performed during this procedure. Fraction 2 should contain any sperm cells present in the sample. Results may not be listed in the report for non-probative fractions.

Deoxyribonucleic Acid (DNA) extraction and quantification, employing real-time Polymerase Chain Reaction (PCR), were performed on a portion of Item #.

The level of support is based on SWGDAM recommendation 1.2 from the SWGDAM recommendations for Genotyping Results Reported as Likelihood Ratios. This recommendation states "...A qualitative statement that conveys the degree of support indicated by the likelihood ratio may be reported in addition to the numerical value for the likelihood ratio...". The scale of verbal qualifiers from the SWGDAM document are as follows:

| <b><i>LR for <math>H_p</math> Support<br/>and <math>1/LR</math> for <math>H_a</math><br/>Support</i></b> | <b>Verbal Qualifier</b> |
|----------------------------------------------------------------------------------------------------------|-------------------------|
| 1                                                                                                        | Uninformative           |
| 2 – 99                                                                                                   | Limited Support         |
| 100 – 9,999                                                                                              | Moderate Support        |
| 10,000 – 999,999                                                                                         | Strong Support          |
| $\geq 1,000,000$                                                                                         | Very Strong Support     |

Assumptions for number of contributors are based on interpretation of the data.

Exclusions are based on the comparable portion of the profile. No comparisons are being made to any portion of the profile deemed not suitable for comparisons.

Statistical analyses resulting in a likelihood ratio (LR) of 1/1000 or less, including an LR of 0, will be reported as "N/A". LR values of 1/1000 (0.001) or less support reporting a direct exclusion. The exact statistic can be found in the case file and/or provided upon request (Supported by SWGDAM recommendation 2.1 in the recommendations for Genotyping Results Reported as Likelihood Ratios). "N/A" may also be used for direct exclusions made by the reporting analyst during interpretation with no additional statistical analyses necessary.

#### 10.3.6.11 Further Actions Statements:

For questions regarding the content of this report, please contact the analyst named below or the DNA section of the laboratory.

Further interpretation may be performed upon submission of a known reference sample from SUSP (and any consensual partner(s)).

The unknown male/female (included if source is not identified) DNA profile obtained from Item # was entered into the Combined/State DNA Index System (CODIS/SDIS) to be routinely searched against the database. The case agency will be notified in the event of a profile match.

Note: This statement is included when an eligible DNA profile has been developed, regardless of whether the profile is from a known or unknown source. Eligibility of forensic profiles for entry into CODIS and upload to NDIS is according to current NDIS procedures and include both solved and unsolved cases in which the profile is associated with a crime and believed to be attributable to the putative perpetrator. Profiles matching the victim(s) and any elimination samples (e.g. consensual partner samples) may not be entered.

If YSTR testing is desired, a known reference is required from SUSP [and/or VIC's consensual partner]. Please contact the laboratory prior to resubmission.

If additional DNA analysis is required, please contact the laboratory.

10.3.7 The following statements are to be used in both biology screening and DNA STR reports:

10.3.7.1 Evidence Disposition Section Statements (these statements will reflect the status of items at the time the report is written):

All items have been returned to the main laboratory evidence vault for return to the submitting agency.

The following items have been retained in the laboratory pending latent fingerprint processing: (list all item numbers).

A portion of the following items has been forwarded for DNA analysis: (list all item numbers). Results will follow in a separate

report. All remaining items have been returned to the main laboratory evidence vault pending DNA analysis.

A portion of Item #s has been forwarded for DNA analysis; however, testing may not proceed until the required known reference sample is received. All remaining items have been returned to the main laboratory evidence vault pending DNA analysis.

Note: Non-suspect cases (those with no known/identified suspect) in which biological evidence has been detected, will be forwarded for DNA testing and applicable CODIS entry. Qualifying samples from legislatively mandated sexual assault evidence collection kits will also be forwarded; however, an attempt should be made to obtain any necessary reference samples.

#### 10.3.7.2 Evidence Description Section Examples:

Sexual Assault Evidence Collection Kit from "name"

Reference oral swabs from "name"

Submitted swab from "location"

Red Hanes t-shirt, labeled as "description on package if different than actual item"

#### 10.3.8 The following statement is to be used in CODIS search reports.

A routine search of the State/National DNA Index System identified a match between the DNA profile previously obtained from Item # (See Report dated...) and a known sample, which is said to have been collected from "Name". Convicted Offender samples are not considered to be evidentiary; therefore, submission of a known reference sample from "Name" is necessary for verification of this match.

#### 10.3.9 It should be noted that the statements (in either the Forensic Biology Screening or DNA Reports) regarding evidence examination, testing and conclusions are not all-inclusive. There may be situations for which none of these statements is optimal. Portions of statements may also be combined as needed.

## 11.0 Review

Technical/administrative, document/quality system, and testimony review (to include retention); as well as conflict resolution is addressed in the ISP Forensic Services Quality/Procedure Manual. See also, forms 306-BI and 502-BI for database review criteria.

### 11.1 BIOLOGY/DNA CASEWORK REVIEW

- 11.1.1 100% of the examinations and reports documented and/or issued from Forensic Biology/DNA will be "peer-reviewed". This review must be completed prior to issuing results (including verbal results) and/or entering eligible profiles into a searchable specimen category of CODIS.
- 11.1.2 "Peer-review" in Forensic Biology/DNA will encompass both technical and administrative reviews of case notes, worksheets, electronic data/electropherograms, photographs, reports, etc.
- 11.1.3 The individual performing the "peer-review" will be qualified and approved to perform the specific area of the review (i.e., Biological Screening and/or DNA Analysis or specified portions thereof). When performing analysis as part of a batch process, the scientist conducting the batch review may be different than the scientist conducting the case review.
- 11.1.4 Technical and administrative review criteria are outlined in the batch review and/or ILIMS review checklists and include a review of data, controls, internal size standards, ladders, DNA profiles obtained, statistics, chain of custody/disposition of evidence, and a check for clerical errors.
- 11.1.5 The report will be reviewed to ensure conclusions are supported by the data and are in accordance with laboratory policy, and that all tested items/probative fractions are addressed.
- 11.1.6 Additionally, the case review scientist will review the CODIS entry information and verify that all eligible profiles have been identified for CODIS entry, the correct specimen categories have been designated, and that alleles/profiles entered are correct. Eligible specimens will not be placed into a searchable specimen category in CODIS until review/verification is complete. The review will be documented by initialing the specimen details report and forwarding to the CODIS Administrator (or alternate) for subsequent updating of the specimen category.
- 11.1.7 Outsourced casework (when applicable) will undergo the same review as listed above, as well as for compliance with contract technical specifications.

## 11.2 CONVICTED OFFENDER/DATABASE SAMPLE REVIEW

- 11.2.1 100% of Convicted Offender sample data (including hit confirmations and outsourced data, when applicable) will be technically and administratively reviewed prior to CODIS entry and subsequent NDIS upload.
- 11.2.2 The individual performing the technical review will be qualified and approved to conduct the specific area of review.
- 11.2.3 Technical and administrative review criteria are outlined on the review checklist (Form 306-BI) and include a review of data, controls, internal size standards, ladders, DNA profiles obtained, and a check for clerical errors.
- 11.2.4 The reviewer of outsourced data (when applicable) will document in an appropriate manner, the review of data for compliance with contract technical specifications and that the .cmf file, if present, contains the correct DNA profiles.
- 11.2.5 Additionally, a documented administrative review will be performed on CODIS hit confirmation letters containing an offender's personally identifiable information, prior to release. The reviewer will date and initial the confirmation letter. The review will be documented on the Hit Confirmation Checklist (Form 502-BI).

## 11.3 TESTIMONY REVIEW

Review of courtroom testimony of Forensic Biology personnel shall be accomplished at least once in each calendar year. Preferably, this review will be performed by the Technical Lead, Biology/DNA Supervisor, or another qualified analyst and documented on the Forensic Services courtroom testimony evaluation form. Alternatively, the evaluation may be completed by criminal justice personnel (e.g., the judge, prosecutor or defense counsel).

## 11.4 CASE FILE/DATABASE PACKET REVIEW

An annual case file and database packet review will be conducted by the Technical Lead or their designee. This review shall be separate from an external audit, but may be conducted concurrently with an internal audit. It will consist of a second technical review of at least two cases/database packets per analyst completed within the previous 12 month period.

## 12.0 Proficiency Testing

General laboratory guidelines and practices for proficiency testing and retention are outlined in the ISP Forensic Services Quality/Procedure Manual. Additional Biology/DNA requirements are delineated below.

### 12.1 External DNA Proficiency Test Requirement

DNA analysts will participate in external proficiency tests, twice in every calendar year, in accordance with The FBI Quality Assurance Standards. Analysts qualified in more than one technology (i.e. STR and Y-STR) will be tested in each technology at least once per year. Multiple technologies may be reported on a single proficiency test. When using a batch approach, analysts must proficiency test in each methodology (using at least one method) for which they are qualified at least once per year.

### 12.2 Inconclusive/Uninterpretable Proficiency Test Results.

Typically, sample size/quantity in PCR DNA Proficiency Tests is sufficient for multiple analyses to be performed. Therefore, results of DNA proficiency tests are not likely to be either inconclusive, or uninterpretable (e.g., not meeting minimal rfu and/or statistical threshold for inclusion/exclusion). However, in the event data obtained in a proficiency test does not meet the standard guidelines for interpretation/conclusion, it will first be determined, by re-testing and communication with the vendor, that this is not an issue with a given sample(s). Once that determination has been made, the analyst obtaining the inconclusive data will be removed from casework/database sample analysis until satisfactory completion of a competency test and review of the analyst's casework/database analysis performed since the last successful proficiency test.

## 13.0 Corrective Action

Laboratory corrective-action and retention procedures are detailed in the ISP Forensic Services Quality/Procedure Manual.

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## 14.0 Audits

Quality audits and retention schedules are delineated in the ISP Forensic Services Quality/Procedure Manual. Specific Biology/DNA audit requirements are delineated below.

- 14.1 A DNA audit, using the current FBI DNA Quality Assurance Standards Audit Document(s), will be conducted on an annual basis.
- 14.2 The interval between annual audits will be in accordance with the current FBI Quality Assurance Standards.
- 14.3 Every other year, at a minimum, the DNA audit must be an external audit.
- 14.4 The completed audit document(s) (Quality Assurance Standards Audit for Forensic DNA Testing Laboratories and for DNA Databasing Laboratories) and applicable corrective action documentation will be reviewed by the Technical Lead and a copy provided to the CODIS Administrator. The external audit documentation will be submitted to the FBI within 30 days of receiving the final audit document.

## 15.0 Safety

Laboratory safety practices are addressed in the ISP Forensic Services Health and Safety Manual. In Forensic Biology, personnel are introduced to these practices in Module 1 of the ISP Forensic Biology Training Manual. In addition, Section 8 of this manual addresses the monitoring of the chemical eye-wash, fire extinguishers, and shower.

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## 16.0 Outsourcing

Outsourcing/Subcontracting policies and procedures are described in the ISP Forensic Services Quality/Procedure Manual.

- 16.1 Approved vendor laboratories must provide documentation of accreditation and compliance with the Quality Assurance Standards for Forensic DNA and/or Database Testing Laboratories prior to contract award and for the duration of the contract.
- 16.2 Technical specifications will be outlined in the outsourcing agreement/contract and approved (approval will be documented) by the Biology/DNA Technical Lead prior to award.
- 16.3 An on-site visit of the vendor laboratory will be performed, by the Biology/DNA Technical Lead or a qualified DNA analyst and documented prior to the submission of any samples to that laboratory. Alternatively, the Biology/DNA Technical Lead may review and accept (the review and acceptance will be documented) an on-site visit conducted by designated FBI personnel or another NDIS laboratory using the same testing platform.
- 16.4 An annual on-site visit will be performed and documented for any contract extending beyond one year.
- 16.5 When outsourcing convicted offender samples, at least one quality control sample shall be included with each batch. Additionally, at least 5% of the total outsourced samples shall be re-tested and compared for consistency and data integrity.
- 16.6 ISPF does not take ownership of vendor data for CODIS entry purposes without an outsourcing agreement.