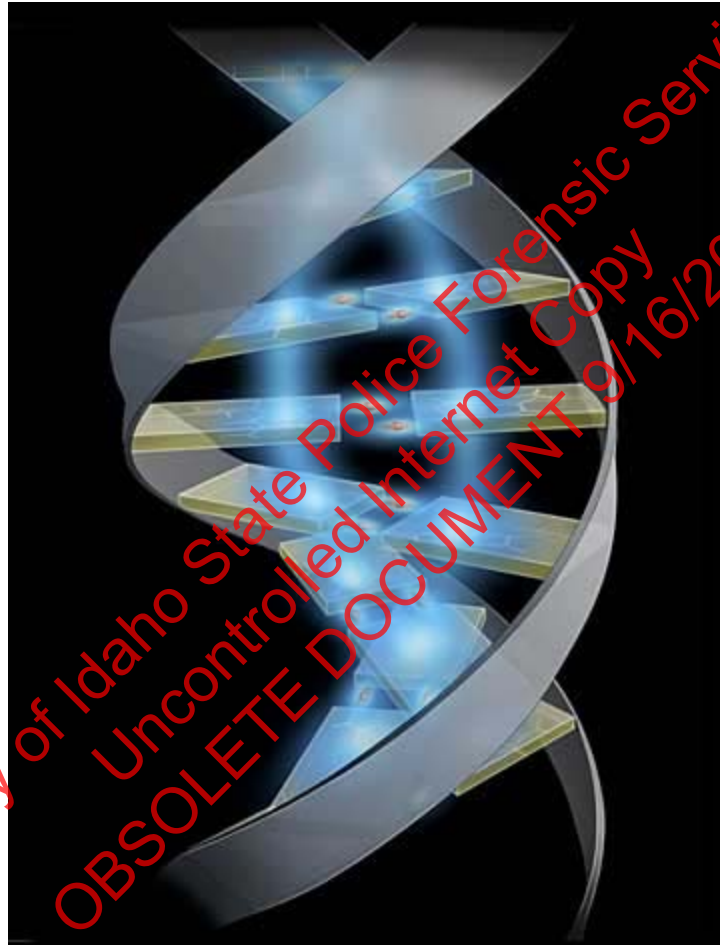


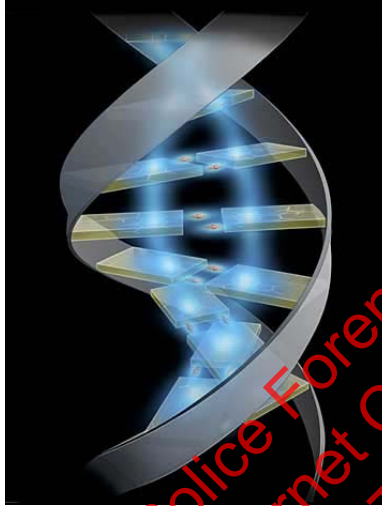
ISP FORENSIC BIOLOGY QUALITY MANUAL



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Forensic Biology Quality Manual

Revision #17



APPROVED
October 16, 2015

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Biology/DNA Technical Lead

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Forensic Biology Quality Manual

REVISION RECORD

The following table must be filled out when revisions to the Biology Quality/Procedure Manual are made.

- Date:** The date the revision(s) was completed/effective date.
Revision #: The manual revision number.
Description: A brief description of the changes made to the manual.
Addition: This column is checked if the revision reflects an addition (e.g. new SOP or form) to the manual.
Deletion: This column is checked if the revision reflects a deletion (e.g. SOP or form no longer in use) from the manual.
Initials: Initials of the Technical Leader making the revisions.

Date	Revision #	Description	Addition	Deletion	Initials
8/10/09	9	Updated Quality Policies and forms methods lists, added contingency plan and FBI quality assurance documents as appendices, separated quality/casework methods/database methods into three separate manuals, added database forms and renumbered methods/forms, updated QC functions, fixed clerical errors throughout	X		CRH
11/29/10	10	Updated 3130 references to include 3130xl, separated biology and database labs, updated CODIS security and review to reflect current NDIS procedures, renamed vault fridges/freezers, changed eye wash check to monthly			CRC
8/29/11	11	Added forms 316-BI, 404C-QC, and 406C-QC (separated casework and database QC's), defined thermal cycler performance check, allowed for FBI on-site visit for outsourcing, clarified evidence numbering scheme and need to refer to return of empty packaging in reports, report wording so blood not required reference, clerical errors	X		CRC
12/7/11	12	Removed 310 references, renumbered CODIS methods, change to casework packet order/binding.		X	CRC
5/23/12	13	Updated organization chart, replaced Mideo with Digital Imaging, changed Biomek 3000 performance check, update forms to remove Taq Polymerase/replace PP16 with PP16HS/replace 9947A with 2800M, 406C-QC not controlled, adjust mini fridge temperature, clerical errors	X	X	CRC
7/3/12	14	Vendor accreditation/calibration certificate review, insufficient male DNA report statements, updated forms			CRC
10/15/13	15	DHR website update, replace Dispatch with bleach substitute, DNA packets not required, removed PCIAA, remove water bath from weekly QC, notes/ reports and review updated to reflect ILIMS, updated forms list, revised forms, removed 101-BI; 214-BI; 218-BI, added 502-BI and 406D-BI, clerical errors	X	X	CRC
9/8/14	16	Updated database note packets and equipment section to reflect FSS i ³ expert system, updated org. chart, added temperature monitoring system, removed Amylase diffusion and Ouchterlony reagents, revised forms 404A-QC(now 404-	X	X	CRH

**Forensic Biology Quality Manual
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INTRODUCTION

1.0 STATEMENT OF PURPOSE AND OBJECTIVES

1.1 Statement of Purpose: 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 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:

- 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

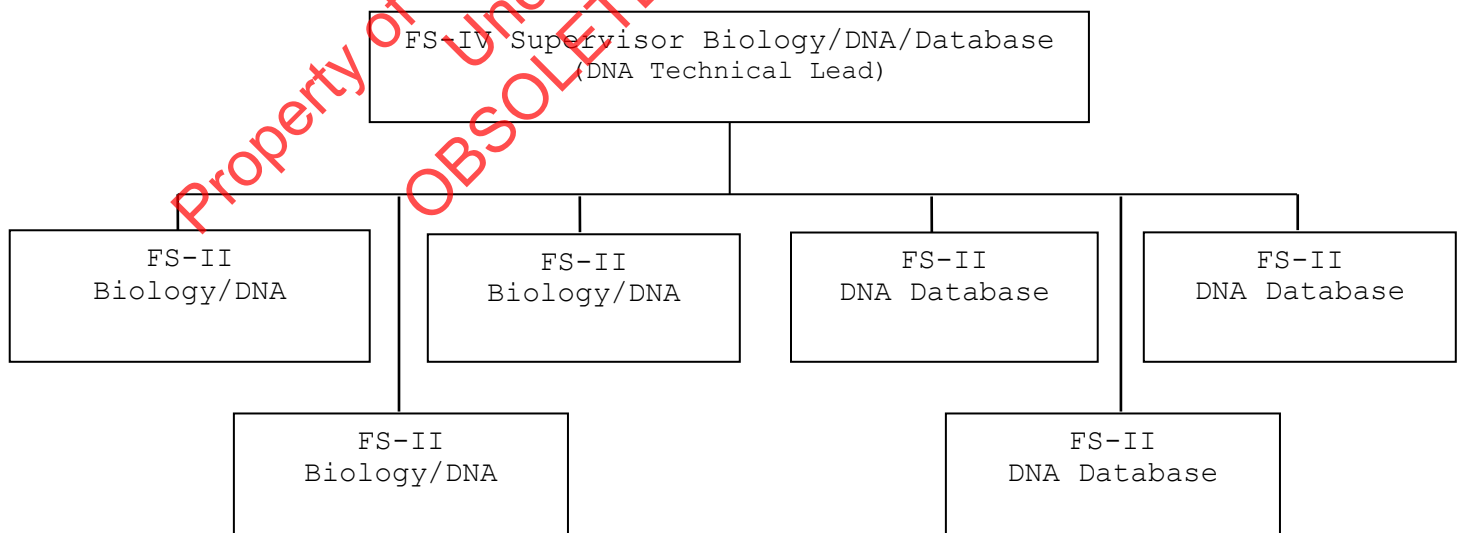
2.1 Organizational Chart and Functional Structure

2.1.1 An organizational chart for ISP Forensic Services appears in the ISP Forensic Services Quality/Procedure Manual. The Forensic Biology organization is delineated below.

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 Convicted Offender 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 positions will generally be filled by qualified DNA analysts. The ISPFBS Quality Manager and Meridian Laboratory Manager will serve those functions, respectively, in absence of DNA analysts being appointed.



3.0 PERSONNEL QUALIFICATIONS AND TRAINING

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:

(<https://labor.idaho.gov/DHR/ATS/StateJobs/JobDescriptions.aspx>).

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.

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 an FS training budget. The training will also be supplemented through the routine reading of current scientific literature. The DNA Technical Lead, or designee, will distribute a DNA-related article to each member of the biology section on a monthly basis. Each staff member will read the article and date/initial the attached sign-off sheet 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) at the bi-annual CODIS State Administrators' meetings and annual CODIS conference.

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. Periodic review of continuing education and overall performance is accomplished during the annual employee evaluation.

3.4.1 Forensic Biology and DNA Supervisor/Technical Lead

It is assumed for the purposes of this document (and is currently the case), that in a laboratory system of the size of Idaho's, these functions will be served by a single individual.

3.4.1.1 Education

Must have, at minimum, a Master of Science degree in a physical or biological science. Successful completion of a minimum of 12 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 and experience in molecular biology and DNA-based analyses from academic, governmental, private forensic and/or research laboratory(ies). 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 physical or biological science and successfully completed college coursework in genetics, biochemistry, and molecular biology. Must also have completed coursework and/or training 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 and the DNA Quality Assurance auditor training within six months of appointment if 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 mixture interpretation. Must be or have been a qualified DNA analyst.

3.4.3 DNA Analyst

The following delineate requirements for a DNA casework or database analyst whose responsibilities include performing genetic analyses on the capillary electrophoresis instruments and data interpretation. DNA extraction, quantification, 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.

3.4.3.1 Education

Must have at minimum, a Bachelor of Science degree in a physical or biological science and successfully completed college coursework in genetics, biochemistry, and molecular biology. Must also have completed coursework and/or training 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 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.

3.4.4.1 Education

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

3.4.4.2 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.

3.4.4.3 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.

3.4.5 Biology Laboratory Technician

3.4.5.1 Education

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

3.4.5.2 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.

3.4.5.3 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

4.1 Laboratory Security

Security of the Forensic Services Laboratory is covered in the ISP Forensic Services Quality/Procedure Manual.

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. Persons outside the Forensic Biology unit 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 for the duration of the individual(s) being present in the laboratory.

4.1.2 CODIS Security

The CODIS workstation is located in the locked CODIS office and the CODIS Server is located in the secured server room in the CJIS Section. The following security measures have been implemented:

4.1.2.1 Only Forensic Biology personnel will have access to the CODIS office. When a biology staff member is not present, the office will be secured by closing and locking the door.

4.1.2.2 Only the CODIS State Administrator, designated Forensic Biology staff and CJIS personnel will have access to the CODIS Server.

4.1.2.3 A differential backup of the CODIS server will be performed each weekday. A full backup will be performed once weekly with the backup tape being stored off-site. At any given time, one month of data will be stored offsite.

- 4.1.2.4 Only Forensic Biology Personnel that have gone through the NDIS application and approval process will have user-names and passwords for CODIS.
- 4.1.2.5 CODIS users must log in each time they use CODIS and log out prior to leaving the CODIS Workstation.
- 4.1.2.6 DNA Tracker, the convicted offender sample-tracking database, resides on the ISP intranet and is accessible only to personnel designated by the Biology/DNA Supervisor.
- 4.1.2.7 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.
- 4.1.2.8 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 E 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-210 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 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 tools/instruments** (i.e. forceps, scissors, etc.) will be cleaned/rinsed with ethanol or germicidal instrument cleaner prior to use and between samples. 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 **Biomek 3000** work surface trays and holders are to be removed and cleaned with 10% bleach or bleach substitute as part of the monthly QC procedure or in the event of a spill. Additionally, each of the tools is to be wiped down with ethanol, being careful not to touch the electronic end.
- 4.3.6 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.
- 4.3.7 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.
- 4.3.8 All work surfaces in the **amplification/post-amp rooms** are to be cleaned with 10% bleach or bleach substitute before and after analysis and as part of the monthly QC procedure. Clean white paper and/or a Wypall is to be placed on the

bench top prior to use. Additionally, 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.

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5.0 EVIDENCE CONTROL

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 2°C and 8°C. Frozen evidence should be stored at ≤-10°C. Standard Reference Materials will be handled, stored, and used according to the guidelines outlined on the corresponding certificate of analysis. Bloodstains certified against a NIST SRM will be used as a known standard, stored frozen (≤-10°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 Packet

It has become increasingly important to retain evidence for possible future analyses and to secure samples for non-probative casework analyses that are necessary for the validation of any new technology. Therefore, a DNA packet may be created for cases submitted for analysis to Forensic Biology, in which positive Biological screening results are obtained (See BI-102). 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. The extracts may be placed in the DNA packet if one has been made.

5.2.2 Limited Sample

In every case, care should be taken to save ~1/2 of a sample for independent testing. If testing would consume all or nearly all of a sample and there is an identified suspect charged in the case, the accused must receive appropriate notification. Written and/or verbal notification will be given to the prosecuting attorney informing him/her of possible consumption and

requesting defense counsel be notified of the situation. Before testing will commence, an allowance will be made for testing by another accredited laboratory agreed upon by both parties. Additionally, a letter from the prosecuting attorney must be received by the laboratory indicating whether or not the sample may be consumed.

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. In cases where both the evidence and associated DNA extract have been consumed, the amplified product will be assigned an item number in ILIMS and retained in a sealed container within the product room freezer.

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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 CHEMICALS/REAGENTS

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

7.1 COMMERCIALLY PURCHASED CHEMICALS

7.1.1 Biology Personnel should consult the section's electronic Chemical Inventory Log (Form 400-QC) prior to ordering. Chemical grade requirements should be checked and ordered as appropriate. The date ordered should be reflected in the log to avoid duplicate orders. An entry for chemicals not currently on the inventory will be made at this time to reflect the chemical, source, and order date. This inventory will be audited annually, at a minimum, and a printout placed in the Forensic Biology Reagent Binder.

Note: An order form/document must be filled out and approved by the section supervisor (indicated by date and initials) prior to placing the order. Reference the forensic services approved chemical list prior to ordering new chemicals.

7.1.2 Upon receipt of a chemical or reagent, the Chemical Inventory Log will be updated to reflect the new lot number, received date, quantity received, and quantity in stock. The order date will be removed at this time. The chemical(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 when removed for use. The following commercially purchased reagents do not have manufacturer expiration dates: HiDi Formamide, and 10X Genetic Analyzer buffer. These 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.hazard.com/msds>

<http://www.msds.com>

<http://www.ilpi.com/msds/>

Note: Critical Reagents listed in 7.3 will be tracked on the individual QC forms, rather than the chemical inventory log.

7.1.3 Expired chemicals will be disposed of in an appropriate manner.

7.2 REAGENTS PREPARED IN-HOUSE

7.2.1 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 7.4 and 7.5 below for individual reagent recipes.

7.2.2 Each reagent has a corresponding form to document the making of the reagent and components used. This form 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.

7.2.3 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.

7.3 CRITICAL REAGENTS

CRITICAL REAGENTS are those reagents that, if improperly functioning, could result in significant loss or destruction of DNA and are not amenable (or it's not practical) to testing immediately before (e.g., use on forensic samples) each use. 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. 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.

ABACARD® HEMATRACE® TEST KIT (Form 410-QC)

OneStep ABACARD® p30 TEST KIT (Form 412-QC)

Plexor® HY System Kit (Form 419-QC)

PowerPlex® 16 HS System Kit (Form 420-QC)

7.4 BIOLOGICAL SCREENING REAGENTS

Phenolphthalein (Kastle-Meyer) Reagent

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

May be a commercial purchase.

Phenolphthalein	1.0g
KOH	20.0g
Zinc (granular)	20.0g

Phenolphthalein, KOH, and 100ml of dH₂O 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.

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%) 10ml/90ml nanopure dH₂O

Mix the H₂O₂ with 90ml of nanopure dH₂O and store at 4°C.

Ortho-Tolidine Reagent

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

O-Tolidine 0.6g
Glacial Acetic Acid 100ml
Ethanol 100ml

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.

Ammonium Hydroxide (~3%)

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

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

Add the NH₄OH to 90ml of nanopure dH₂O, mix well and store at RT.

10X Brentamine (Sodium Acetate) Buffer

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

Sodium Acetate (Anhydrous) 1.2g
Acetic Acid (to adjust to pH 5) ≈400μl

Dissolve Sodium Acetate in 10ml of nanopure dH₂O. Add Acetic Acid to pH 5. Store refrigerated.

Brentamine Solution A

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

O-Dianisidine Tetrazotized (Fast Blue B) 50 mg
10X buffer pH 5 5 ml

Dissolve Fast Blue B Salt in 5 ml of 10X Brentamine Buffer.
Store refrigerated in a dark container.

Brentamine Solution B

(NFPA: health 2, flammability 0, reactivity 0)

α -Naphthyl Phosphate (Disodium Salt) 50 mg

Dissolve in 5 ml of nanopure dH₂O. Store Refrigerated.

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.

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 dH₂O. 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 KH₂PO₄, and 2.2g Na₂HPO₄·7H₂O (or 1.1g Na₂HPO₄ anhydrous) in 800ml nanopure dH₂O. Adjust pH to 7.4 if necessary. Q.S. to 1l with nanopure dH₂O, autoclave and store at RT.

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 dH₂O. Immediately add the Nuclear Fast Red, mix, cool and filter (paper or ≥45µm). May be stored at RT.

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 ≥45µm). May be stored at RT.

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.

Zinc Chloride 10% (w/v)

(NFPA: health 2, flammability 0, reactivity 2)

Zinc Chloride 10g/100ml 95% EtOH

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

7.5 DNA REAGENTS

1M Tris-HCl Buffer pH 7.5

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

Tris Base (tris[Hydroxymethyl]amino methane) 121.1 g

Dissolve Tris in ~800 ml nanopure dH₂O. Adjust to pH7.5 at RT by adding concentrated HCl (approximately 65ml). Q.S. to 1l with nanopure dH₂O, autoclave and store at RT.

1M Tris-HCl Buffer pH 8

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

Tris Base (tris[Hydroxymethyl]amino methane) 121.1 g

Dissolve Tris in ~800 ml nanopure dH₂O. Adjust to pH8 at RT by adding concentrated HCl (approximately 45ml). Q.S. to 1l with nanopure dH₂O, autoclave and store at RT.

0.5M Ethylenediamine Tetraacetic Acid (EDTA)

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

Na₂EDTA·2H₂O 186.1g/l

Slowly add EDTA to 800ml nanopure H₂O while stirring vigorously. Add ~20g of NaOH pellets to bring the pH to near 8.0. When fully dissolved adjust pH to 8.0 and bring final volume to 1l. Autoclave and store at RT.

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

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 dH₂O. Store at RT.

Note: Reagent contains SDS, do not autoclave.

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 dH₂O.

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

1M Sodium Acetate pH 5.2

(NFPA: health 3, flammability 2, reactivity 0)

$\text{CH}_3\text{COONa}\cdot 3\text{H}_2\text{O}$ 13.6g

Dissolve the $\text{CH}_3\text{COONa}\cdot 3\text{H}_2\text{O}$ in 80ml nanopure dH_2O . Adjust to pH5.2 by adding glacial acetic acid (approximately 2 ml). Q.S. to 100ml with nanopure dH_2O , autoclave and store at RT.

DTT Solution

(NFPA: health 2, flammability 1, reactivity 0)

Dithiothreitol (DTT) 0.77g

Dissolve the DTT in 5ml nanopure dH_2O . Add 50µl 1M Sodium Acetate, pH5.2. Dispense ~500µl each into sterile microcentrifuge tubes and store at $\approx 20^{\circ}\text{C}$.

Note: Do not autoclave.

PCR-TE (TE⁻⁴) Buffer (10mM Tris-HCl/0.1mM EDTA)

(NFPA: health 2, flammability 1, reactivity 0)

1M Tris-HCl, pH8 10ml
0.5M EDTA, pH8 0.2ml

Mix Tris-HCl and EDTA with 990ml nanopure dH_2O . Autoclave and store at RT.

5N Sodium Hydroxide

(NFPA: health 3, flammability 0, reactivity 2)

NaOH 50g

Slowly dissolve the Sodium Hydroxide in 250ml sterile nanopure dH_2O . Allow to cool and store at RT.

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

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) 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. The system will escalate to an automated text message alert when a unit has been outside of its established critical range for an extended period of time, and which could result in the deterioration of evidence or reagents. 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.). A temperature report for each unit will be generated and saved on a monthly basis. The first page of the report will be printed and placed in the applicable section QC binder. Additional reports may be generated as necessary and applicable for monitoring unit performance.

8.1.2 Analytical equipment significant to the results of examination and requiring routine calibration and/or performance verification will be listed on the BIOLOGY CRITICAL EQUIPMENT INVENTORY Spreadsheet (Form 401-QC). Information on the spreadsheet includes (as known or appropriate): equipment identity and its software, manufacturer's name, model, property number, serial number and/or unique identifier, and location. The inventory spreadsheet will be maintained in the instrument QC binder or section QC binder as appropriate.

8.1.3 OPERATING MANUALS for most equipment/instrumentation will be maintained in the product information file (Manuals for the ABI PRISM™ 3130/3130xl 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. The Biomek 3000 manual is built into the Biomek software.

8.1.4 MAINTENANCE/REPAIR/CALIBRATION LOGS will be maintained as follows:

The records for the ABI PRISM™ 3130/3130xl Genetic Analyzers, ABI 7500 Real-Time PCR System, and Thermal Cyclers will be maintained in the instrument QC binder.

Any equipment/instrumentation function (not documented on weekly, monthly, quarterly, or annual QC Check forms) will be recorded on the Equipment Maintenance/Repair form (Form 402-QC). Equipment Failure will also be reported on this form. This form and the QC check forms will be maintained in the section QC Binder, 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 Form 402-QC.

8.1.6 The SCHEDULE of QC/Performance Checks for both critical and non-critical equipment is as follows:

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
- Heating Block(s) Temperature Check
- Oven Temperature Check
- BSD600 Cleaned

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

- Biomek 3000 Cleaned
- Lab Cleaned
- Eye Wash Station Check
- Autoclave Clean and Check Sterilization
- ABI 7500 Background Assay/Contamination Test, and Function Test/Bulb Check
- BioRobot EZ1 grease D-rings
- QIAcube Cleaned
- 3130/3130xl Water Wash
- 3130/3130xl Water Trap Flush
- 3130/3130xl (C and E drives) and 7500 computer defragmentation
- Biomek 3000 computer defragmentation and restart

QUARTERLY

(once per quarter with an interval between dates not less than 30 days and not exceeding 120 days) Note: * denotes critical equipment

- Thermal Cycler* Temperature Verification
- ABI 7500* Temperature Verification
- Biomek 3000 Robotic System* Framing/Calibration Check (Form 408-QC)
- QIAcube O-ring Replacement, Centrifuge Rotor Cleaned/Oiled (Form 408-QC)
- Chemical Shower Check (Form 408-QC)
- FSS i³ Expert System Calibration Check (Form 430-QC)

ANNUALLY (Form 402-QC)

(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)
- NIST Traceable Thermometers* (outside vendor)
- Driftcon FFC Temperature Verification System* Calibration Check (outside vendor)
- Biological and Chemical Hoods Test (outside vendor)
- ABI PRISM™ 3130/3130xl* 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* Preventative Maintenance (outside vendor)
- Qiagen QIAcube* Preventative Maintenance (outside vendor)

- Biomek 3000* preventative Maintenance (outside vendor)
- Microscope Cleaning/Preventative Maintenance (outside vendor)
- Centrifuge Calibration Check (outside vendor)
- Balance* Calibration Check (outside vendor)

In addition to the above schedule, personnel should check appropriate parameter function on all instrumentation with each use [including calibration of the pH meter (documented on Form 403-QC) and water bath temperature at the time of use], and run a spatial and spectral calibration for the ABI PRISM™ 3130/3130xl Genetic Analyzers as needed or following CCD camera and/or laser replacement/adjustment.

Following the annual preventative maintenance, a sensitivity panel (previously characterized DNA) should be run on the 3130/3130xl and included in the QC binder as a verification of performance. A framing/calibration check are to be run on the Biomek 3000 and included in the Database QC binder as a performance check following the annual preventative maintenance. The Driftcon FFC will be run on each thermal cyclers (including 7500's) following repair and prior to being placed back in to service as a verification of performance. If no repairs were necessary, the pure dye calibration and ROT's will serve as the performance verification for the 7500 s following the annual preventative maintenance. Documentation will be maintained in the section QC binder.

Equipment calibrations performed by an outside vendor will be verified through a review of the calibration certificate provided by the vendor. Additionally, 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.

Any problems noted with laboratory equipment, during normal usage or as part of a QC check should be brought to the attention of the necessary supervisory personnel and documented on Form 402-QC and/or the respective QC form.

A certified NIST standard will also be run annually or if substantial procedural changes have been made. The QC run will be documented on Form 426-QC and filed in the section QC binder.

9.0 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.

9.1 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.

9.2 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.

10.0 CORRECTIVE ACTION

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

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11.0 FILE DOCUMENTATION AND REPORTS

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 case record and retention are described in the ISP Forensic Services Quality/Procedure Manual.

11.1 CASE NOTES

- 11.1.1 Notes or worksheets prepared external to ILIMS (e.g. printed photos, sketches, DNA worksheets, electropherograms, etc.) will bear the date and scientist's initials. These will be scanned to ILIMS. Following approval of the assignment, the scanned version will be considered the 'original' documentation and the hard copies will be destroyed.
- 11.1.2 Each page of 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.
- 11.1.3 All evidence submitted for biological screening should be transferred to the scientist, dated and initialed. This is the case regardless of whether or not they analyze the item of evidence (exceptions include cases where communication with investigator/attorney indicate analysis is no longer needed). Items taken into custody but not examined will be listed in the report.
- 11.1.4 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.

- 11.1.5** 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.
- 11.1.6** Evidence descriptions should be "unique" inasmuch as possible (i.e., one pair blue jeans is **NOT** adequate). They should 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 and condition (e.g., worn, clean, torn, mud-caked, blood-soaked, etc.).
- 11.1.7** 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 is removed for DNA testing ≡ Item 57.1

2 or more areas/stains removed for DNA testing)≡ Item 57.1, Item 57.2, Item 57.3 etc.

An item with multiple sub-items (e.g., a SAECK; Item 1) ≡ Item 1.1, Item 1.2, Item 1.3, etc.

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

≡ Item 25.1 (shirt) and Item 25.2 (pants)

Areas/stains removed for DNA testing ≡ Item 25.1.1, Item 25.1.2, Item 25.2.1 etc.

- 11.1.8** Photography, digital or otherwise, is often useful in documenting the appearance of evidence items. However, it is not meant to completely replace drawing, but instead as a supplement or in cases when drawing may be too difficult to accurately depict the item. Careful drawing and description result in careful and detailed examinations and, in many instances, may be a better choice than photography. 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 a jpeg for case notes, as necessary.

11.1.9 The casework note packet is considered complete when the analyst submits the assignment to be reviewed. Electronic documentation external to ILIMS (e.g. Electropherograms, tables of results, and statistics) is considered stored at this time. Any changes to the electronic data required after this point will be documented on the original hard copy, initialed, dated, and scanned to ILIMS. If the change requires reprinting, a notation on the new hard 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 scanned to ILIMS. The original scanned document will remain in ILIMS but will be unchecked for inclusion in the note packet.

11.2 DATABASE PACKETS

11.2.1 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).

11.2.2 In general, database packets will be arranged from front to back with chronological worksheets then reinjection summary. Review forms may be placed at the front of the packet for ease of plate identification. Upon completion of review, the analyst should bind (e.g. staple) the documentation together and file it appropriately.

11.2.3 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 i³ electronic data are required after this point (on or after the review date documented in the notes), the analyst will generate an output file so the dates in the audit files are updated to reflect when the changes were made. Changes to the Genemapper ID data will be documented in the note packet with analyst initials and the date.

11.3 CASEWORK REPORTS

- 11.3.1 The report will contain the title Forensic Biology Report, Forensic DNA Report, or CODIS Hit Report as appropriate.
- 11.3.2 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.
- 11.3.3 The case submission information will include: laboratory case#, agency, agency case#, offense date(s), investigating officer, report #, evidence submission date(s), analyst, report date, and principals (victim, suspect, etc.)
- 11.3.4 The body of the report will be separated from the case submission information by the following headings: Evidence Description, Conclusions and Interpretations, and Disposition of Evidence
- 11.3.5 Conclusions and interpretations statements should be, inasmuch as possible, in the following order: 1) positive results or inclusions, 2) inconclusive results, 3) negative results or exclusions and 4) items not examined.
- 11.3.6 The following results/conclusions statements are to be used in a **biology screening report**, as dictated by the analysis findings. 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:

Semen Results/Conclusions Statements:

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

Chemical and microscopic analyses for the detection of semen were conducted on (items). Semen was confirmed on (items) by the presence of a single spermatozoon (or limited 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 (items). Semen was detected on (items) by the presence of the semen specific protein, p30; however, no spermatozoa were observed, which may be insufficient for further testing at this time.

Results from presumptive chemical tests for the presence of semen were negative on (items).

Blood Results/Conclusion Statements:

Results from chemical and serological tests performed on (items) indicated the presence of human (or non-human) blood.

Results from presumptive chemical tests performed on (items) indicated the presence of blood; however, serological tests to determine the species of origin were not performed (or were inconclusive).

Results from presumptive chemical tests for the presence of blood were negative on (items).

Saliva Results/Conclusions Statements:

Results from chemical tests performed on (items) indicated the presence of an elevated level of amylase, an enzymatic component of saliva.

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

Urine Results/Conclusions Statements:

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

Feces Results/Conclusions Statements:

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

Further Testing Statements (to be included at the end of the Results of Examination Section):

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

11.3.7 The following results/conclusions statements are to be used in an **STR DNA Report**:

Note: Associations deemed to be probative based on sample type and case circumstances will be qualified with a statistic and/or source attribution statement. Probative associations for multiple items or profiles may be qualified with a single statement if the statistics are identical or exceed the source attribution threshold. 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., 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 sperm cell fraction (e.g. minor component consistent with semen donor). In these instances, a statement regarding the DNA source of this fraction is not required.

Introductory Statement:

Deoxyribonucleic Acid (DNA) Analysis, employing the Polymerase Chain Reaction (PCR), was used to generate a Short Tandem Repeat (STR) profile from the following items: "List of items".¹

Note: The following footnote will appear in all reports in which DNA testing was attempted.

¹Loci Examined: D3S1358, TH01, D21S11, D18S51, Penta E, D5S818, D13S317, D7S820, D16S539, CSF1PO, Penta D, vWA, D8S1179, TPOX, and FGA.

Profile Match Statement [meeting the 'source attribution' criterion (estimated frequency in population of ≤ 1 in 2.0×10^{10})] for single source and identifiable major contributors of a mixture:

The DNA profile obtained from Item # matches that obtained from the blood stain/sample (or reference oral swab/sample, etc.) of/from "name".

Therefore, "name" is the source of the "(DNA, blood, semen, saliva etc.) " on this item².

Note: The following footnote will appear in any report containing the above match statement.

²This conclusion is based upon the following: 1) a genetic match at the gender identity locus, Amelogenin, in addition to the "number" polymorphic STR loci listed above that have an expected population frequency of at least less than 1 in "actual (most conservative of the population groups calculated) frequency estimate", 2) a statistical frequency exceeding the source attribution criterion of 2.0×10^{10} (for $N=2.0 \times 10^7$, $\alpha=0.01$; *Forensic Science Communications* 2(3)July 2000), and 3) that "name" does not have a genetically identical twin.

Profile match Statement [not meeting the 'source attribution' criterion (estimated frequency in population of greater than 1 in 2.0×10^{10})] for single source and identifiable major contributors of a mixture:

The DNA profile obtained from Item # matches that obtained from the blood/oral sample of "name". The probability of selecting an unrelated individual at random from the general population having a DNA profile that would match the DNA profile obtained from Item # is at least less than one in "actual (most conservative of the population groups calculated) frequency estimate".

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

The DNA profile obtained from Item # also matches that obtained from the blood/oral sample of "name", however less genetic information was obtained. The probability of selecting an unrelated individual at random from the general population having a DNA profile that would match the partial DNA profile obtained from Item # is at least less than one in "actual (most conservative of the population groups calculated) frequency estimate".

The partial DNA profile obtained from Item # is consistent with that obtained from the blood sample of "name". The probability of selecting an unrelated individual at random from the general population having a DNA profile that would match the partial DNA profile obtained from Item # is at least less than one in "actual (most conservative of the population groups calculated) frequency estimate".

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.

Mixture Statements:

The DNA profile from Item# indicates a mixture of DNA from at least "X" persons. "Name(s)" is a potential contributor(s) to this mixture. "X%" of unrelated individuals randomly selected from the general population would be expected to be eliminated as potential contributors to this mixture.

The DNA profile from Item# indicates a mixture of DNA from at least two persons. "Name(s)" is a potential contributor(s) to this mixture. The DNA profile obtained from Item# is at least "X" 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.

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

The DNA profile from Item# indicates a mixture of DNA with a discernable major contributor/profile. (include match, consistent with, or exclusionary statement regarding major profile). "name" is included/excluded/cannot be excluded as a possible contributor to the minor DNA component of this mixture.

The DNA profile from Item# indicates a mixture of DNA from at least "X" persons. "Name(s)" is a potential contributor(s) to this mixture. At least one in "actual (most conservative of the population groups calculated) frequency estimate" unrelated individuals randomly selected

from the general population would be expected to be included as potential contributors to this mixture.

Exclusionary Statements:

The DNA profile obtained from the Item # does not match that obtained from the blood sample of "name". Therefore, "name" is not the source (or "a contributor" in a mixed profile situation) of the "(DNA, blood, semen, saliva etc.)" on this item.

The DNA profile obtained from the Item # was determined to be from an unknown male/female. "name" is not the source of the "(DNA, blood, semen, saliva etc.)" on this item.

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

No DNA Profile Obtained/Insufficient Male DNA Statements:

Due to insufficient quantity or degradation, no DNA profile was obtained from Item #.

Male DNA was detected on Item #; however, the quantity obtained is insufficient for STR analysis. No further testing was conducted on this item.

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

CODIS Entry Statement:

The unknown male/female (included if source is not identified) DNA profile obtained from the Item # was entered into the Combined DNA Index System (CODIS) 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.

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

Evidence Disposition Section Statements:

All items, including any remaining DNA extracts (Item #), 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 number).

The following items have 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 for return to the submitting agency.

Note: Nonsuspect cases (those with no known/identified suspect) in which biological evidence has been detected, will be forwarded for DNA testing and CODIS entry.

Evidence Description Section Examples:

A Sexual Assault Evidence Collection Kit from "name".

Two reference oral swabs from "name".

A red/brown stained swab from "location".

A red 'Hanes' t-shirt size large, labeled as "description on package if different than actual item".

11.3.9 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 (Offender ID#). 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.

11.3.10 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.

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12.0 REVIEW

Technical/administrative, document, 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 in this manual for database review criteria.

12.1 BIOLOGY/DNA CASEWORK REVIEW

- 12.1.1 100% 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 CODIS.
- 12.1.2 "Peer-review" in Forensic Biology/DNA will encompass both technical and administrative reviews of case notes, worksheets, electronic data/electropherograms, allele tables, photographs, reports, etc. The review is performed in and the completion documented in ILIMS.
- 12.1.3 The individual performing the "peer-review" will be a second scientist who is qualified in the area of the review (i.e., Biological Screening and/or STR Analysis).
- 12.1.4 Technical and administrative review criteria are outlined in the 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.
- 12.1.5 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.
- 12.1.6 Additionally, the second scientist will review the CODIS entry information and verify that all eligible profiles have been identified for CODIS entry and the correct specimen categories have been assigned. Eligible specimens will not be entered into CODIS until

review/verification is complete. The specimen details report will be reviewed and initialed by the CODIS Administrator (or alternate) following manual data entry and prior to searching at SDIS and uploading to NDIS to verify correct allele entry and specimen category.

12.1.7 Outsourced casework (when applicable) will undergo the same review as listed above, as well as for compliance with contract technical specifications.

12.2 CONVICTED OFFENDER/DATABASE SAMPLE REVIEW

- 12.2.1** 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.
- 12.2.2** The individual performing the technical review will be a second scientist who is qualified in the area of STR Analysis.
- 12.2.3** The second scientist performing the review will initial each page of the data package (and date the first and last page at a minimum).
- 12.2.4** 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 (note: the review includes the use of an expert system).
- 12.2.5** The scientist performing the review 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.
- 12.2.6** 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.

12.3 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 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).

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13.0 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 and shower.

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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 appropriate accompanying documentation will be submitted to NDIS according to NDIS Operational Procedures.

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15.0 OUTSOURCING

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

- 15.1 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.
- 15.2 Technical specifications will be outlined in the outsourcing agreement/contract and approved (approval will be documented) by the Biology/DNA Technical Manager prior to award.
- 15.3 An on-site visit of the vendor laboratory will be performed, by the technical leader or a qualified DNA analyst, and documented prior to the submission of any samples to that laboratory. Alternatively, the technical leader 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.
- 15.4 An annual on-site visit will be performed and documented for any contract extending beyond one year.
- 15.5 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.

16.0 Practices, 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).

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

BI≡Analytical Procedures or Individual Processes

- BI-100** PROCESSING LIQUID BLOOD
- BI-102** DNA PACKETS
- BI-104** PHENOLPHTHALEIN TEST FOR BLOOD
- BI-105** O-TOLIDINE TEST FOR BLOOD
- BI-106** HUMAN BLOOD IDENTIFICATION USING ABACARD® HEMATRACE® TEST
- 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** IDENTIFICATION OF SEMEN BY P30 DETECTION (ABAcad®)
- BI-122** AMYLASE TEST: PHADEBAS
- BI-126** DETECTION OF URINE (UREASE)
- BI-128** DETECTION OF URINE (CREATININE)
- BI-130** DETECTION OF FECAL MATERIAL (UROBILINOGEN)
- BI-200** EXTRACTION PROTOCOLS FOR PCR DNA TYPING TESTS
- BI-207** DNA QUANTIFICATION: REAL-TIME PCR
- BI-208** STR AMPLIFICATION: PP16HS
- BI-210** STR TYPING: CAPILLARY ELECTROPHORESIS AND DATA ANALYSIS
- BI-301** OFFENDER SAMPLE RECEIPT AND DNA TRACKER ENTRY
- BI-312** EXTRACTION PROTOCOLS FOR PCR DNA TYPING TESTS
- BI-314** DNA QUANTIFICATION : REAL-TIME PCR
- BI-316** STR AMPLIFICATION : PP16HS
- 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

Form BI≡Various forms used in each discipline

* indicates a controlled form

100-BI PHENOLPHTHALEIN REAGENT (KASTLE-MEYER)
102-BI HYDROGEN PEROXIDE 3% (v/v)
103-BI O-TOLIDINE REAGENT
104-BI AMMONIUM HYDROXIDE (~3%)
114-BI 10X BRENTAMINE (SODIUM ACETATE) BUFFER
116-BI BRENTAMINE SOLUTION A
118-BI BRENTAMINE SOLUTION B
120-BI SALINE (0.85% NaCl)
124 BI 1X PHOSPHATE BUFFERED SALINE (PBS)
126-BI XMAS TREE STAIN SOLUTION A (KERNECHTROT SOLUTION)
128-BI XMAS TREE STAIN SOLUTION B (PICROINDIGOCARMINE SOLUTION)
138-BI MERCURIC CHLORIDE 10% (w/v)
140-BI ZINC CHLORIDE 10% (w/v)
201-BI 1M TRIS-HCl BUFFER pH 7.5
203-BI 1M TRIS-HCl BUFFER pH 8
205-BI ETHYLENEDIAMINE TETRAACETIC ACID (EDTA) 0.5M
207-BI STAIN EXTRACTION BUFFER pH 8
211-BI PROTEINASE K (20 mg/ml)
222-BI 1M SODIUM ACETATE pH 5.2
223-BI DTT (1M)
229-BI PCR-TE (TE⁻⁴) BUFFER (10mM TRIS-HCl, 0.1M EDTA)
231-BI NaOH 5N
233-BI SODIUM CHLORIDE (NaCl) 5M
249-BI BOVINE SERUM ALBUMIN (BSA) 4%
200-BI DNA EXTRACTION WORKSHEET
202-BI DIFFERENTIAL DNA EXTRACTION WORKSHEET
206-BI* CASEWORK WORKSHEETS (A-D)
306-BI STR OFFENDER DATABASE REVIEW CHECKLIST
312-BI* DATABASE WORKSHEETS (A-E)
314-BI OUTSOURCED OFFENDER DATA REVIEW
316-BI DATABASE REINJECTION SUMMARY
400-QC FORENSIC BIOLOGY CHEMICAL INVENTORY
401-QC FORENSIC BIOLOGY CRITICAL EQUIPMENT INVENTORY
402-QC FORENSIC BIOLOGY EQUIPMENT MAINTENANCE/REPAIR RECORD
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
- 408-QC FORENSIC BIOLOGY QUARTERLY QC
- 410-QC* QC ABACARD® HEMATRACE® KIT
- 412-QC* QC ONESTEP ABACARD® P30 KIT
- 419-QC* QC PLEXOR® HY QUANTIFICATION KIT
- 420-QC* QC PP16HS KITS
- 422-QC 3130/3130x1 INJECTION LOG
- 426-QC* ANNUAL NIST QC RUN
- 430-QC* QUARTERLY EXPERT SYSTEM CALIBRATION CHECK
- 500-BI CODIS SAMPLE REMOVAL CHECKLIST
- 502-BI HIT CONFIRMATION CHECKLIST

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