Department of Forensic Biology

Training Manual

Version 1.0
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I. Introduction

The purpose of the training program is to prepare new analysts with the theoretical and practical means necessary to perform reliable testing. For interpreting job titles, this includes training to learn how to present this information competently in court. By having a multi-phase program of practical exercises, written assignments, and oral examinations, an analyst's weak points should become obvious, and the staff can work with the analyst to bolster this aspect of his/her knowledge and competency.

Newly hired staff are trained to perform a variety of different procedures, each relating to analyzing physical evidence for DNA typing. Each trainee progresses through a series of training modules in sequence; the modules correspond to duty rotations in the laboratory: evidence examination, sexual assault kit processing, exemplar processing, Chelex extraction, QuantiBlot, and PCR amplification and typing. The modules selected depend on the job title of the trainee. Completion of the complete set of required modules is necessary for a trainee to become an interpreting analyst.

During training periods, staff will be required to spend all of their time in training. This means that flexible or compressed time schedules, attendance at professional meetings, and participation in special projects (such as MESATT) will not generally be allowed.

In total, the training will cover the theoretical and practical aspects of forensic biology. In particular it covers aspects of molecular biology, separation technology, interpretation of complex DNA results, and statistical concepts as they relate to forensic DNA analysis.

A. Training – evidence examination and serological methods

The goal of training and competency testing in the classical forensic biology methods is to establish consistency of performance between individual analysts and to maintain the highest possible level of performance over time. These analytical procedures for identifying physiological fluids are the foundation on which further individualization (DNA testing) is based, and their behavior and limitations must be understood.

The classical forensic biology training program is monitored by the Director, Deputy Director, Assistant Directors, and/or Criminalist III/IV supervisors. The training may be performed by any Criminalist II or higher who is competent and has the appropriate level of experience (generally, at least six months completed past the training period).

A Criminalist may interpret and write reports for negative blood cases, negative semen cases, semen-only cases, and species-determination cases as soon as they have successfully completed the appropriate competency tests.

B. Training - DNA analysis

The goal of training and competency testing in the DNA laboratory is to establish consistency of performance throughout the laboratory and to maintain the highest possible level of performance over time.
The DNA training program is monitored by the Director, Deputy Director, Assistant Directors, and/or Criminalist III/IV supervisors. The training may be performed by any Criminalist II or higher who is competent and has the appropriate level of experience (generally, at least six months completed past the training period).

The trainee may not interpret DNA results (STR gel/CE processing and signing DNA reports) until they become a DNA interpreting analyst: completed all training modules (except DNA mock court), all three classes, population statistics, six months DNA lab experience, and successful completion of a DNA competency test. They will be expected to manage their DNA cases and write DNA reports for their supervisor’s signature in the interim.

*If any new or additional federal and/or state requirements are imposed, they must be met prior to interpreting DNA results.*

C. Training folder

The training is documented in a training folder. This contains records (notes, worksheets, photographs, etc.) generated during training. In addition, for each topic the date and initials of the trainer should be noted. The supervisor should regularly review the contents of the training folder for accuracy and completeness.

D. Training schedule

A training schedule must be provided to each trainee and all scientific staff responsible for any aspect of the training. Because the training schedule affects many aspects of department operations, it should be adhered to as carefully as possible. Each module has adequate time allotted for the training. If necessary, for example if equipment is unavailable, a trainee may be asked to substitute a weekend day for a week day.

For Lab Associates and Criminalist I’s, the training is limited. As competency is obtained in each module, the trainee normally then is assigned to a two week assignment in that rotation performing analysis on casework samples.

For Criminalist II’s and above, the training continues non-stop. Once all required training modules are complete, the trainee joins the rotation schedule.
E. Roles and responsibilities

Training Coordinator

The training coordinator is responsible for periodic review and/or revision of the Training Manual and reference binder.

The training coordinator is responsible for preparation of training schedules, training assignments, and training folders. This includes scheduling of training given by OCME staff other than those from the Department of Forensic Biology.

The training coordinator is responsible for ensuring that practice samples and competency test samples are prepared.

The training coordinator is responsible for ensuring that reference binders and manuals are available.

Trainee

The trainee is expected to be ready to go by 9 am each day there is directly supervised training (observation or demonstration of a technique). On days where the trainee is working on practical exercises, practice samples, or competency tests there may be more flexibility.

The trainee is expected to do the required readings and be prepared to answer questions from the trainer or their supervisor on the topics as they are covered.

The trainee is expected to work on and complete the written questions during the time period of the training module and/or lecture. They should not be postponed until the end of hands-on training.

Trainer

The trainer is expected to be ready to go by 9 am each day there is directly supervised training (observation or demonstration of a technique). The trainer must realize that training has the priority; meetings or other tasks may have to be postponed. If the assigned trainer finds he/she is unavoidably unable to perform the training they must make arrangements for the training to be re-assigned.

The trainer is responsible for reinforcing the information from the required readings and lectures by discussing each technique in detail during the training, including theoretical and practical aspects.

The trainer must be available for questions on other days allocated for the module.

The trainer must review any paperwork generated during the demonstration of a technique by a trainee; the review should include checking for completeness and accuracy.
Supervisor

The direct supervisor of the trainee has the primary responsibility for the monitoring of the training process. The supervisor must plan on regularly spending time with the trainee, including:

- Discussing the topics covered by the required reading.
- Reviewing the answers to the written questions.
- Reviewing the training folder for completeness and accuracy.
- Determining the successful completion of competency tests.

Regular weekly or biweekly meeting at the end of each training module is expected.

The direct supervisor is responsible for helping the trainees choose cases for serology and DNA mock court, acting as prosecutor, and preparing them for testimony.

Technical Leader

The technical leader is responsible for final determination of the readiness of the trainee to enter the rotation. This includes:

- Final review of the training folder, including review of competency tests as needed.
- Final review of the answers to the written questions.
- Evaluation of the oral examination, including any needed remediation.
- Determination of satisfaction of state and/or federal requirements, including review of college transcripts, course syllabi, and/or textbooks as needed.

The technical leader is responsible for issuing the written notification of completion of training and the written notification of achievement of interpreting analyst status.
II. Training – general guidelines

A. Theoretical background

In addition to requiring a minimum educational background for the job title(s), the Department provides additional theoretical background necessary for trainees to understand the scientific basis behind each analytical test. This training takes place over a number of weeks through the required lectures and reading assignments. Most lectures are also available as computer presentations maintained in the departmental directory.

Each member of the scientific staff has access to literature references and reference books that the department maintains; methods manuals used in the laboratory contain bibliographies listing references in the scientific literature. Copies of publications pertaining to in-house methods are given to each trainee in the form of a Reference Binder. Additionally, OCME professional staff has library and Internet privileges at the New York University Medical School library located next door to the OCME.

B. Practical experience

Each analyst will be trained to perform the analytical procedures that are appropriate to that job title. All practical training has three phases: the trainee observes the procedure being done; the trainee demonstrates the procedure to the trainer; the trainee uses the procedure independently on practice specimens.

C. Competency testing

At the conclusion of training in any particular analytical procedure, the trainee will be asked to successfully complete a competency test using that procedure. In general, a competency test is prepared in-house with the key to the results being supplied to the supervisor, Assistant Directors, Technical Leader, and Director.

D. Written assignments and oral examination

New scientific staff (Lab Associates and above) must take and pass the written assignment (if applicable) for each module taken. New scientific staff (Criminalist II and above) must take an oral examination covering several areas of DNA theory and analysis before using DNA procedures in casework.

The written assignment is reviewed by the supervisor and Technical Leader.

The oral examination is attended by the supervisor, appropriate Assistant Director, and the Technical Leader.

E. Court preparation

An important part of training is for learning to present scientific information in court. There are several ways to prepare trainees for court and public speaking: accompanying laboratory personnel to court and observing testimony, as well as attending pre-trial
conferences at the laboratory.

Before testifying in court or grand jury, new staff must participate in a moot/mock court conducted by supervisory scientific staff; non-scientific people may also be present. The purpose of the moot/mock court is to give the analyst an introduction to the adversary process and to practice for actual testimony in a trial or grand jury. It is also a mechanism for the supervisory staff to identify and correct obvious problems the analyst may have in his/her knowledge or ability to communicate effectively. After the moot court, the constructive criticism of the trainee’s mock testimony is given, and, if needed, specific suggestions for improvement is provided.

Minimally, two moot/mock courts are required. The first, early in training, is a serology mock court on an actual small case; this covers the initial forensic biology training topics. The second, after the analyst has joined the rotation, is a DNA mock court on an actual DNA case; this covers all forensic biology training topics. If necessary, additional mock courts can be scheduled.

F. Supplemental Training

Scientists will be introduced to new procedures as they are added. Usually, this will include a lecture covering the theoretical and practical aspects of the procedure; a reading list selected from the scientific literature will be provided. The three-phase training method (trainee observes the analytical procedure, trainee demonstrates the analytical procedure, and independent practice) will also be used for most supplemental training.

For the practical aspects of the training, practice samples may be processed at the same time as other samples. Once the analysts are comfortable with the new procedure, they will be given competency test samples, which must be successfully completed for each new procedure before the analyst can use the procedure in casework.
III. Training - specific guidelines

A. Training modules

The training is divided into modules. The number of modules taken depends on the job title of the trainee; fewer or additional modules may be given depending on the particular job assignment of the trainee.

<table>
<thead>
<tr>
<th></th>
<th>Lab Associate</th>
<th>Criminalist I</th>
<th>Criminalist II</th>
</tr>
</thead>
<tbody>
<tr>
<td>introduction (personnel)</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>right to know (hygiene officer)</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>digital photography</td>
<td></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>evidence exam</td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>serology- blood presumptive</td>
<td></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>serology- species determination</td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>serology- AP and sperm</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>serology- amylase</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>small cases</td>
<td></td>
<td></td>
<td>X</td>
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<tr>
<td>35 mm photography</td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>microscopy</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>serology mock court</td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>sexual assault kits</td>
<td></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>exemplar processing</td>
<td></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>P30 ELISA</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>hemoglobin</td>
<td>selected staff</td>
<td>selected staff</td>
<td>selected staff</td>
</tr>
<tr>
<td>Chelex Extraction</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>QuantiBlot</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>PCR amp &amp; gels</td>
<td>gel prep 377 take-down</td>
<td>gel prep 377 take-down</td>
<td>X</td>
</tr>
<tr>
<td>PCR amp &amp; CE</td>
<td>PCR amp</td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>dilutions &amp; mixtures</td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>STR interpretation</td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>DNA written exam</td>
<td>limited</td>
<td>X</td>
<td></td>
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<tr>
<td>-----------------------</td>
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<td></td>
</tr>
<tr>
<td>DNA oral exam</td>
<td></td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>DNA mock court</td>
<td></td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>blood spatter</td>
<td>selected staff</td>
<td>selected staff</td>
<td>X</td>
</tr>
</tbody>
</table>

B. **Required lectures**

Most of the training modules have required lectures. The lectures are given by staff members, generally prior to beginning each training module. Many of the lectures are also available as computer presentations found in the departmental directories, and can be reviewed as necessary.

C. **Required reading**

All of the training modules have required reading. Much of the information is found in the reference binder supplied to trainees. However, the analysts are also required to read the appropriate sections of manuals, chapters in books, etc. The required reading should be completed during the time allotted to the training module.

D. **Practice samples**

For serology training (blood presumptive test, species identification, semen presumptive tests, semen confirmatory tests, and amylase) practice samples can come from a variety of sources: the trainee, stains from previous external proficiency tests, or casework extracts previously tested for P30 and/or amylase.

The number of serology training samples is variable, depending on the training module. The number of tests performed is much greater, as specified in the practical exercises of each module.

For DNA training practice samples of known DNA types are required. Because these are practice samples, the DNA types may be supplied to the trainee along with the samples. These can be stains representing laboratory personnel, exemplar stains from casework, or stains from previous external proficiency tests. *A sample from the trainee is mandatory, for addition to the LABTYPES database holding the DNA profiles of staff members.*

The number of DNA samples must include at least one of each of the following: blood stains, mixed semen stains, saliva stains, and hair samples. They should be supplied in sufficient quantity for the trainee to be able to do more than one analysis if necessary. The number of tests performed is much greater, as specified in the practical exercises of each module.

The trainee will generally use these same practice samples for all DNA procedures - Chelex extraction, QuantiBlot, amplification and DNA typing.
E. Competency samples

For the DNA modules, it is preferable that these be from previous external proficiency tests. They must be coded to render them anonymous to the trainee.

<table>
<thead>
<tr>
<th>module</th>
<th>sample type</th>
<th>minimum number of competency samples</th>
</tr>
</thead>
<tbody>
<tr>
<td>serology - blood presumptive test</td>
<td>blood/not blood</td>
<td>4</td>
</tr>
<tr>
<td>serology - species identification</td>
<td>human/not human</td>
<td>4</td>
</tr>
<tr>
<td>serology - sperm identification</td>
<td>sperm/no sperm</td>
<td>4</td>
</tr>
<tr>
<td>serology - amylase identification</td>
<td>amylase/no amylase</td>
<td>4</td>
</tr>
<tr>
<td>P30 ELISA</td>
<td>semen/no semen</td>
<td>4</td>
</tr>
<tr>
<td>Chelex extraction</td>
<td>blood and/or saliva stains</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>mixed semen stains</td>
<td>2</td>
</tr>
<tr>
<td>QuantiBlot</td>
<td>previously quantitated samples - the Chelex samples from above or others supplied by trainer</td>
<td>10 (5 sets of neat and 1/10 dilutions)</td>
</tr>
<tr>
<td>PCR amp/gels</td>
<td>blood and/or saliva stains, mixed semen stains - the Chelex samples from above</td>
<td>5</td>
</tr>
<tr>
<td>PCR amp/CE</td>
<td>adult, fetal, and/or other variant blood stains</td>
<td>5</td>
</tr>
</tbody>
</table>

The trainee will use these same competency test samples for all DNA procedures - Chelex extraction, QuantiBlot, amplification and DNA typing. Since these are competency test samples, the DNA types are not to be supplied to the trainee.

F. Review procedures

The results from the trainee’s practice samples and competency tests will be evaluated by his/her direct supervisor in terms of sensitivity, consistency, and contamination at each of the steps in the training. In addition, the supervisor must ensure that the trainee is analyzing the proper control samples, is correctly and completely filling out worksheets and logbooks used to document sample analyses, and is familiar with the operation of the equipment necessary to perform the tests. It may be helpful to include the trainer in this review process.

Problems will be addressed at each rotation and additional practice instituted, if necessary. For example, the supervisor must check the trainee’s work for contamination.
Low-level contamination (the presence of alleles that do not meet laboratory reporting criteria, such as small peaks in STR analysis) may not affect the typing results. Such contamination may often be eliminated by simply changing a reagent. However, if the analyst consistently demonstrates low-level contamination, he/she must be observed more closely during subsequent practice runs to identify the reason for the problem.

*The direct supervisor must sign off on each module, indicating completion of all practical exercises and successful completion of the competency test, if applicable.*

G  Completion of training

At the completion of each analytical training module, a written notification must be made to the trainee by the direct supervisor that he/she has successfully passed the competency test and that the analyst may now perform that technique on casework samples. The notification will generally take the form of initialing the Forensic Biology Training Checklist.

Once an analyst has completed all the requirements to become a DNA interpreting analyst, the Technical Leader must issue a written notification which acknowledges the successful completion of the requirements: completes all training modules (except DNA mock court), all three classes, population statistics, six months DNA lab experience, and successful completion of a DNA competency test. This notification is filed in the training folder.

As of that date, the analyst may interpret DNA results (STR gel/CE processing) and sign DNA reports.
MODULE 1  Digital photography

Required lecture

Basic forensic photography

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the Basic Forensic Photography handout.

Practical exercises

After observing fundamental digital camera handling techniques, do the following:

1. Practice handling and using the camera, transferring digital images to the computer, manipulating the images, and printing photographs in various formats (one to a page, two or more to a page).

Competency test

None.

KSA’s to be mastered

1. Be able to use any of the digital cameras in the laboratory.
2. Be able to control illumination.
3. Be able to produce decent quality photographs of physical evidence.
4. Be able to explain the theory to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor sign off on successful completion of module. The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
MODULE 2  Evidence examination

Required lecture

Safety and right to know

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the note taking section of the Case Management Manual.
3. Retrieve current case files from the file cabinet and study the note-taking styles of various analysts. Think about what you like and dislike about the different styles.

Practical exercises

1. Observe several Criminalists examining evidence.

Competency test

None.

KSA’s required to be mastered

1. Understand target dates, how cases are assigned, and paperwork to be filled out for case tracking purposes.
2. Understand the importance of chain of custody worksheets for evidence sign-out, return to the Evidence Unit, and the documentation of retained items.
3. Understand the need to thoroughly examine and analyze evidence items based on the scheduled analysis, including the use of evidence packaging form, clothing description form, hand-written notes and diagrams, and photography as needed.
4. Understand policies regarding substrate controls, retention of samples, and consumption of samples.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor sign off on successful completion of module. The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
MODULE 3A  Serology - blood presumptive tests

Required lecture

Body fluid identification

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the tests for blood in the Biochemistry Manual.

Practical exercises

When you run analytical procedures during training or receive samples for competency testing, take notes and fill out worksheets as if you were working on a real case.

After observing each procedure and having demonstrated each procedure to the trainer, do the following experiments.

1. Sensitivity. Check this for the KM and LMG presumptive tests by testing serial dilutions of blood up to 1/1,000,000.
2. Specificity. Check this for both presumptive reagents by testing various substances such as sweat, urine, rust, plant extracts (onions), etc.

Competency test

Obtain a bloodstain identification competency test. You must correctly determine the presence or absence of blood.

KSA’s to be mastered

1. Be able to perform the blood presumptive tests.
2. Understand the composition of blood, both its cellular components and protein makeup (including hemoglobin).
3. Understand the mechanisms of the presumptive tests for blood employed in the laboratory: Kastle-Meyer (KM), leucomalachite green (LMG), and luminol.
4. Understand which substances cross-react with which presumptive test and why.
5. Understand the sensitivity and limitations of the KM test.
6. To understand the use of controls for this procedure.
7. Be able to explain the theory and how these tests are run to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor evaluate the results of the competency test.
3. Have supervisor sign off on successful completion of module. The initials of the
supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).

MODULE 3B  

Serology - species determination

Required lecture

Body fluid identification

Required reading

1. Study the articles in the reference binder on this topic.

Practical exercises

When you run analytical procedures during training or receive samples for competency testing, take notes and fill out worksheets as if you were working on a real case.

After observing each procedure and having demonstrated each procedure to the trainer, do the following experiments.

1. Run both procedures using anti-human sera against dilutions of human blood.
2. Run both procedures using dilutions of anti-human sera against human blood.
3. Run both procedures using anti-human sera against the blood of various common animals that you might encounter in New York City.
4. Run both procedures using anti-sera of various animals against their corresponding animal blood, other animal blood, and human blood.

Competency test

Obtain a bloodstain species identification competency test. You must correctly determine the species of each sample.

KSA's to be mastered

1. Be able to perform the Ouchterlony and cross-over electrophoresis tests.
2. Understand the sensitivity and limitations of the species determination tests.
3. Understand the use of controls for the species tests.
4. Understand the immunology of antibodies and antigens, including sensitivity and specificity of the methods used.
5. Understand how antisera are made, both polyclonal and monoclonal.
6. Understand the agarose gel diffusion method and cross-over method used to identify antigens and antibodies.
7. Understand why the precipitin bands form and how this relates to the identity of a specific antigen.
8. Understand the function of the Takayama test and its limitations.
9. Be able to explain the theory and how these tests are run to someone who does not have a scientific background.
Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor evaluate the results of the competency test.
3. Have supervisor sign off on successful completion of module. *The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).*
MODULE 3C  Serology - AP and sperm

Required lecture

Body fluid identification

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the tests for semen in the Biochemistry Manual.

Practical exercises

When you run analytical procedures during training or receive samples for competency testing, take notes and fill out worksheets as if you were working on a real case.

After observing each procedure and having demonstrated each procedure to the trainer, do the following experiments.

1. Sensitivity. Using the acid phosphatase test, test various dilutions of semen extracts up to 1/1,000,000.
2. Specificity. Check for specificity of the acid phosphatase test against other substances: vaginal fluid, urine, saliva, etc.
3. Prepare slides of semen stains, semen-stained vaginal swabs, and semen-free vaginal swabs. Use two methods of preparing the slides: extracting the stains and pelleting cellular debris by centrifugation and "mashing" the stains/swabs onto a slide. Stain these slides using the Christmas Tree stain procedure.

Competency test

Obtain a semen identification competency test. You must correctly determine the presence or absence of sperm in each sample.

KSA's to be mastered

1. Be able to perform the acid phosphatase presumptive test and the Christmas Tree stain.
2. Understand the sensitivity and limitations of the AP test.
3. Understand the use of controls for the AP tests.
4. Understand the components of seminal fluid, including human sperm morphology. Get a general feeling about how sperm morphology differs in various animals.
5. Understand the mechanism of the acid phosphatase presumptive test.
6. Understand what substances interfere with the test or might give a false positive test.
8. Understand the persistence of the components of semen in the oral, anal, and vaginal tracts and why the length of time differs.
9. Be able to explain the theory and how these tests are run to someone who does not have a scientific background.
Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor evaluate the results of the competency test.
3. Have supervisor sign off on successful completion of module. *The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).*
MODULE 3D  Serology - amylase

Required lecture

Body fluid identification

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the tests for saliva sections in the Biochemistry Manual.

Practical exercises

When you run analytical procedures during training or receive samples for competency testing, take notes and fill out worksheets as if you were working on a real case.

After observing each procedure and having demonstrated each procedure to the trainer, do the following experiments.

1. Run the amylase diffusion procedure on dilutions -- up to 0/000 -- of saliva, semen, semen-stained vaginal swabs, semen-free vaginal swabs, and urine.

Competency tests

Obtain an amylase identification competency test. You must correctly determine the presence or absence of amylase in each sample.

KSA's to be mastered

1. Be able to perform the amylase test.
2. Be able to properly interpret the test for different sample types.
3. Understand the sensitivity and limitations of the amylase test.
4. Understand the use of controls for the amylase test.
5. Understand the difference between AMY1 and AMY2 and in which body fluids each is found
6. Understand the theory of how to differentiate AMY1 and AMY2 using lectins.
7. Be able to explain the theory and how these tests are run to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor evaluate the results of the competency test.
3. Have supervisor sign off on successful completion of module. The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
MODULE 4  Small cases

Required lecture

Case management

Required reading

1. Study the articles in the reference binder on this topic.

Practical exercises

1. Obtain some simple cases to work on, in consultation with the evidence exam rotation supervisor, evidence sign-in supervisor, or your supervisor. You will be the interpreting analyst on each case for the purpose of determining the presence of body fluids.
2. Review the paperwork in the casefile.
3. Examine the items in each case, documenting as needed with available forms, handwritten notes, photography, and/or diagrams.
4. Perform any presumptive and/or confirmatory tests as needed.
5. Write reports, return evidence as needed (after consultation with your supervisor), and submit case to your supervisor for review.

Competency test

None.

KSA’s to be mastered

1. Understand target dates, how cases are assigned, and paperwork to be filled out for case tracking purposes.
2. Be able to fill out chain of custody worksheets for evidence sign-out, return to the Evidence Unit, and for documentation of retained items.
3. Be able to thoroughly examine and analyze evidence items based on the scheduled analysis including use of evidence packaging forms, clothing description forms, handwritten notes and diagrams, and photography as needed.
4. Be able to write an accurate and complete report on a simple case.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor evaluate the case notes, body fluid identification, and the report.
3. Have supervisor sign off on successful completion of module. The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
MODULE 5  35 mm photography

Required lecture

Basic forensic photography

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the Basic Forensic Photography handout.

Practical exercises

After observing fundamental camera handling techniques, do the following:

1. Practice loading, rewinding, and unloading film into and from the camera.
2. Practice adding and removing photographic accessories to and from the camera (lenses, electronic flashes, etc.).
3. Practice taking basic photographs covering establishing and closeup photography, depth of field, 1:1 photography, and illumination techniques.

Competency test

Obtain a Basic Photographic Competency Test sheet. You must take all the required photographs (must be good quality).  

KSA’s to be mastered

1. Be able to use any of the Nikon 35mm SLR cameras in the laboratory.
2. Be able to produce decent quality photographs of physical evidence.
3. Know the basic photographic terms, principles, and techniques.
4. Understand the relationship, if any, between the shutter speed and physical aperture when producing an exposure.
5. Be able to explain the theory to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor sign off on successful completion of module. The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
MODULE 6  Microscopy

Required lecture

Basic microscopy

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the Procedure for Proper Microscopic Illumination handout.

Practical exercises

After observing fundamental microscopic technique, do the following:

1. Practice setting up critical illumination with different slides and magnifications.
2. Practice using the stereoscope.

Competency test

None.

KSA’s to be mastered

1. Be able to use and set up critical illumination on any of the compound microscopes in the laboratory.
2. Be able to use the stereoscope.
3. Know the basic microscopic terms, principles, and techniques.
4. Be able to explain the theory to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module. This may necessitate the direct supervisor observing the trainee demonstrate proper use of the microscope.
2. Have supervisor sign off on successful completion of module. The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
MODULE 7  Serology mock court

Required lecture

Basics of the legal system
QA/QC
Accreditation and certification

Required reading

1. Study the articles in the reference binder on this topic.

Practical exercises

To prepare for mock court, the trainee might review court transcripts, suggested questions, reading material concerning expert testimony, and observing laboratory personnel testify in court.

1. As available, attend court with Criminalists and observe testimony.
2. In consultation with your supervisor, select one of your small cases for use in a mock court. Your supervisor will be the prosecutor, and other staff members will take the roles of the defense attorney and judge.
3. Review the theoretical and practical aspects of the testing performed in the small case.
4. With your supervisor, go over the questions to be asked in the direct examination and the potential topics to be covered in cross examination.
5. Practice your answers with your supervisor and on your own, paying particular attention to making your responses loud, clear, and easily understandable to a lay person. Learn to speak slowly and enunciate carefully, directing your answers towards the jury. Learn to listen carefully to the questions, making sure the question is complete before answering; think before replying.

Competency test

Successfully complete your serology mock court. Your performance will be critiqued by the attending staff members; a written Court Testimony Evaluation form will be provided by the “judge”.

KSA’s to be mastered

1. Demonstrate poise, technical knowledge, ability to convey scientific concepts, and correct interpretation of laboratory results.

Final actions

1. Have supervisor sign off on successful completion of module.
MODULE 8 Sexual assault kits

Required lecture

Standardized sexual assault evidence collection kits

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the sexual assault kit processing section of the Case Management Manual.
3. Study the Christmas Tree staining and amylase presumptive test sections of the Biochemistry Manual.

Practical exercises

1. Observe a Criminalist processing a sexual assault kit, including at least one containing underwear or other small clothing item.
2. Demonstrate the processing of at least one sexual assault kit for the trainer, including preparation of stained slides and examination of underwear or other small clothing item.

*During training, and for a period of six months after the completion of training, new Criminalists must have sperm searches double-read by a competent Criminalist.*

Competency test

None.

KSA’s to be mastered

1. Understand target dates new cases are assigned, and paperwork to be filled out for case tracking purposes.
2. Be able to thoroughly examine and analyze a sexual assault kit based on the scheduled analysis.
3. Be able to fill out chain of custody worksheets for evidence sign-out, return to the Evidence Unit, and for documentation of retained items.
4. Understand the tasks to be done prior to transferring a semen-positive case to another Criminalist.
5. Be able to write an accurate and complete report on a simple case.
6. Understand the purpose of each sexual kit component.
7. Understand the mechanism of the Christmas Tree stain.
8. Be able to write an accurate and complete report on a simple case.
9. Be able to explain the theory and how these tests are run to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor evaluate the case notes and the report.
3. Have supervisor sign off on successful completion of module. *The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).*
MODULE 9  Exemplar processing

Required lecture

Safety and right to know

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the blood processing section of the Biochemistry Manual.

Practical exercises

Observation and demonstration only.

Competency test

None.

KSA's to be mastered

1. Understand target dates, how cases are assigned, and paperwork to be filled out for case tracking purposes.
2. Be able to thoroughly examine and analyze exemplar items (blood and oral swabs) based on the scheduled analysis, including submission for DNA extraction.
3. Be able to fill out chain of custody worksheets for evidence sign-out, return to the Evidence Unit, and for documentation of retained items.

Final actions

1. Discuss the module with a direct supervisor, including review of paperwork and discussion of theory and practical aspects of module.
2. Have supervisor sign off on successful completion of module. *The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).*
MODULE 10  P30 ELISA

Required lecture

Identification of P30 using ELISA

Required reading

1. Study the articles in the reference binder on this topic.
3. Review the P30 ELISA validation records.

Practical exercises

When you run analytical procedures during training or receive samples for competency testing, take notes and fill out worksheets as if you were working on a real case.

After observing the procedure and having demonstrated the procedure to the trainer, do the following experiments.

1. Sensitivity. Using the P30 ELISA test, test various dilutions of semen extracts up to 1/10,000,000.
2. Specificity. Check the specificity of anti-P30 (polyclonal antibody) by using other substances such as vaginal fluid, urine, and saliva using ELISA.

Competency test

Obtain a semen identification competency test. You must correctly determine the presence or absence of P30 in each sample.

KSA’s to be mastered

1. Be able to perform the P30 ELISA test.
2. Be able to correctly interpret P30 ELISA results.
3. Understand the sensitivity and limitations of the AP test.
4. Understand the use of controls for the AP tests.
5. Know about seminal plasma specific proteins. Concentrate on the prostate specific antigen, P30 (also called Prostate Specific Antigen or PSA), and how it is identified and quantified.
6. Study ELISA techniques used to quantify P30.
7. Understand the relationship, if any, between the amount of acid phosphatase, P30, spermatozoa, and the amount of semen present.
8. Be able to explain the theory and how these tests are run to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of
theory and practical aspects of module.

2. Have supervisor evaluate the results of the competency test.
3. Have supervisor sign off on successful completion of module. The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
 MODULE 11  

Chelex extraction

Required lecture

DNA extraction

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the DNA extraction methods in the PCR manual.

Practical exercises

When you run analytical procedures during training or receive samples for competency testing, take notes and fill out worksheets as if you were working on a real case.

After observing the procedure and having demonstrated the procedure to the trainer, do the following experiments.

1. Chelex extraction of bloodstains/saliva samples, including your own sample.
2. Chelex differential extraction of semen positive samples.
3. Chelex extraction of hairs.

As each extraction is finished, submit aliquots for DNA quantitation. Review the results with the supervisor; once satisfactory results are obtained on the practice samples, perform extractions on the competency test samples. Submit aliquots of competency test samples for DNA quantitation.

Competency test

Each sample must yield a typable amount of DNA, as determined by QuantiBlot. Each extraction set must have a clean extraction negative, as determined by QuantiBlot and PCR analysis. Each sample must give the correct type in the PCR systems tested.

*Lab Associates and Criminalist I's must aliquot their competency test samples for PCR analysis in one system (generally Quad) so that the typing results can be evaluated by a supervisor.*

*Criminalist II's and above will perform their own PCR analysis in other modules.*

KSA's to be mastered

1. Be able to perform Chelex extraction on all sample types.
2. Understand the preparation, handling, and function of reagents used for DNA extraction.
3. Be able to properly aliquot samples for QuantiBlot.
4. Understand the use of controls introduced at this stage of DNA typing.
5. Be able to explain the theory and how these tests are run to someone who does not have a scientific background.

Final actions
1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor evaluate the results of the competency test.
3. Have supervisor sign off on successful completion of module. *The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).*
MODULE 12  QuantiBlot

Required lecture

DNA quantitation

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the DNA quantitation methods in the PCR manual.

Practical exercises

When you run analytical procedures during training or receive samples for competency testing, take notes and fill out worksheets as if you were working on a real case.

After observing the procedure and having demonstrated the procedure to the trainer, do the following experiments.

1. Repeat DNA quantitation of the Chelex extracts of the practice samples
2. If desired, practice QuantiBlot further

Review the results with the supervisor; once satisfactory results are obtained on the practice samples, perform DNA quantitation on the competency test samples. Review the results with the supervisor before continuing.

Competency test

The competency test samples provided for Chelex extraction are used for all subsequent DNA competency tests.

The QuantiBlots must have callible standards, calibration controls, and negative. The QuantiBlot results obtained for neat samples and the 1/10 dilution should correlate with one another; if not, the samples must be repeat.

KSA’s to be mastered

1. Be able to perform QuantiBlot.
2. Understand the preparation, handling, and function of reagents used for DNA quantitation.
3. Understand the use of controls for the QuantiBlot test.
4. Understand the sensitivity and limitations of the QuantiBlot test.
5. Be able to explain the theory and how this test is run to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor evaluate the results of the competency test.
3. Have supervisor sign off on successful completion of module. *The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).*
MODULE 13  PCR amplification and gels

Required lectures

- PCR theory
- STR typing
- Basics of gel electrophoresis on the ABI 377

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the gel-based DNA methods in the PCR manual.

Practical exercises

When you run analytical procedures during training or receive samples for competency testing, take notes and fill out worksheets as if you were working on a real case.

After observing the procedure and having demonstrated the procedure to the trainer, do the following experiments.

PCR amplification and gel-based typing

1. Amplify all practice samples using one gel-based PCR system (generally Quad).
2. Prepare a gel, set up a run, and analyze the data.

Review the results with the supervisor; once correct PCR typing results are obtained on the practice samples, perform PCR amplification and typing on the competency test samples in all gel-based casework PCR systems.

Submit the PCR typing results for review by the supervisor. If the supervisor feels that additional work is necessary for the trainee, it should be completed before continuing. Once the trainee is passed, continue to the last part of practical analytical procedure training.

Competency test

The competency test samples provided for Chelex extraction are used for all subsequent DNA competency tests.

The DNA typing results must be correct. Extraction negatives must give clean results. The editing of samples must be correct.

KSA’s to be mastered

1. Be able to correctly interpret QuantiBlot results, make any necessary calculations, and submit proper amounts for amplification.
2. Understand the preparation, handling, and function of reagents used for PCR amplification and DNA typing.
3. Understand the use of controls introduced at this stage of DNA typing.
4. Be able to amplify and type samples in all DNA gel systems used in casework.
5. Be able to correctly edit electropherograms, including the correct identification of artifacts.
6. Be able to properly use the instrument and associated computers, and archive data correctly.
7. Understand the theory of PCR, the basics of STR typing, and the basics of gel electrophoresis.
8. Be able to explain the theory and how these tests are run to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module. This may necessitate the direct supervisor observing the trainee demonstrate proper archiving of data.
2. Have supervisor evaluate the results of the competency test.
3. Have supervisor sign off on successful completion of module. The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
MODULE 14  PCR amplification and capillary electrophoresis

Required lectures

PCR theory
STR typing
Basics of capillary electrophoresis on the ABI 310

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the capillary-based DNA methods in the PCR manual.

Practical exercises

When you run analytical procedures during training or receive samples for competency testing, take notes and fill out worksheets as if you were working on a real case.

After observing the procedure and having demonstrated the procedure to the trainer, do the following experiments.

PCR amplification and capillary-based typing

1. Amplify all practice samples using one capillary-based PCR system (generally Cofiler).
2. Set up a run, and analyze the data.

Review the results with the supervisor once correct PCR typing results are obtained on the practice samples, perform PCR amplification and typing on the competency test samples in all gel-based casework PCR systems.

Submit the PCR typing results for review by the supervisor. If the supervisor feels that additional work is necessary for the trainee, it should be completed before continuing. Once the trainee is passed, continue to the next part of practical analytical procedure training.

Competency test

The competency test samples provided for Chelex extraction are used for all subsequent DNA competency tests.

The DNA typing results must be correct. Extraction negatives must give clean results. The editing of samples must be correct.

KSA’s to be mastered

1. Be able to correctly interpret QuantiBlot results, make any necessary calculations, and submit proper amounts for amplification.
2. Understand the preparation, handling, and function of reagents used for PCR amplification and DNA typing.
3. Understand the use of controls introduced at this stage of DNA typing.
4. Be able to amplify and type samples in all DNA gel systems used in casework.
5. Be able to correctly edit electropherograms, including the correct identification of artifacts.
6. Be able to properly use the instrument and associated computers, and archive data correctly.
7. Understand the theory of PCR, the basics of STR typing, and the basics of capillary electrophoresis.
8. Be able to explain the theory and how these tests are run to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module. This may necessitate the direct supervisor observing the trainee demonstrate proper archiving of data.
2. Have supervisor evaluate the results of the competency test.
3. Have supervisor sign off on successful completion of module. The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
MODULE 15  PCR dilution and mixture studies

Required lecture

OCME PCR validation studies
Basics of STR mixture interpretation

Required reading

1. Study the articles in the reference binder on this topic.
2. Study the interpretation of complex STR results section in the PCR manual.

Practical exercises

At this point, the trainee will be working independently, performing dilution and mixture studies which will aid him/her to interpret complex PCR typing results.

1. Using either practice or competency test Chelex extracts, prepare a dilution series of DNA (50, 10, 5, 2, 1, 0.1 and 0.01 ng) in the final amplification volume in at least two of the autosomal PCR systems (one gel and one CE) used in casework and amplify; evaluate results.
2. Pick two practice or competency test samples which have different DNA types and prepare mixtures of the samples (20:1, 10:1, 5:1, 1:1, 1:2, 1:10, and 1:20); amplify and type in at least two of the autosomal PCR systems (one gel and one CE) used in casework.
3. Prepare a mixture containing two males and a mixture containing a male and a female (40:1, 20:1, 10:1, 5:1, 1:1, 1:5, 1:10, 1:20, and 1:40); amplify and type in Y STR’s.
4. Prepare a written interpretation of the results, including in your interpretation a statement concerning limitations of the method to detect and resolve mixtures.

Competency test

None

KSA’s to be mastered

1. Be able to identify mixtures and determine the relative proportion of the components.
2. Understand the limitations of each system to resolve mixtures of different proportions.
3. Understand the sensitivity of each system.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor evaluate the conclusions developed in the written interpretation.
3. Have supervisor sign off on successful completion of module. *The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).*
MODULE 16  PCR data interpretation

Required lecture

OCME PCR validation studies
Basics of STR mixture interpretation
Basics of population genetics and statistics

Required reading

1. Study the articles in the reference binder on this topic.

Practical exercises

The trainer will provide the trainee with a series of data tables and/or gel runs representing the range of results that are typically observed in PCR DNA typing cases. The trainee must evaluate the data tables and write a Forensic Biology report discussing the data and his/her interpretation of it. These interpretations will be discussed in a meeting with Criminalist IV’s and/or an Assistant Director.

Competency test

None

KSA’s to be mastered

1. Be able to write DNA reports, including appropriate statistics, using the standard report format and template statements of the Department of Forensic Biology.
2. Be able to evaluate initial DNA results and draw correct conclusions.
3. Be able to evaluate initial DNA results and determine what further testing might be needed.
4. Be able to determine the proper statistical information for each DNA scenario.

Final actions

1. After the meeting discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module. Review the reports that were written and the changes and suggestions made during the meeting.
2. Have supervisor sign off on successful completion of module.
MODULE 17 DNA mock court

Required lecture

Basics of the legal system

Required reading

1. Study the articles in the reference binder on this topic.

Practical exercises

To prepare for mock court, the trainee might review court transcripts, suggested questions, reading material concerning expert testimony, and observing laboratory personnel testify in court.

1. As available, attend court with Criminalists and observe testimony.
2. In consultation with your supervisor, select one of your DNA cases for use in a mock court. Your supervisor will be the prosecutor, and other staff members will take the roles of the defense attorney and judge.
3. Review the theoretical and practical aspects of the testing performed in the small case.
4. With your supervisor, go over the questions to be asked in the direct examination and the potential topics to be covered in cross examination.
5. Practice your answers with your supervisor and on your own, paying particular attention to making your responses loud, clear, and easily understandable to a lay person. Learn to speak slowly and enunciate carefully, directing your answers towards the jury. Learn to listen carefully to the questions, making sure the question is complete before answering; think before replying.

Competency test

Successfully complete your DNA mock court. Your performance will be critiqued by the attending staff members; a written Court Testimony Evaluation form will be provided by the “judge”.

KSA’s to be mastered

1. Demonstrate poise, technical knowledge, ability to convey scientific concepts, and correct interpretation of laboratory results.

Final actions

1. Have supervisor sign off on successful completion of module.
MODULE 18  Oral examination

Required lecture

All technical lectures plus QA/QC and Accreditation and Certification.

Required reading

All.

Practical exercise

None.

Competency test

New scientific staff must take and pass an oral examination covering several areas of DNA theory and analysis before using DNA procedures in casework; the scope of the questions is similar to those in the written examination. The oral examination is attended by the supervisor, appropriate Assistant Director, and the Technical Leader. In order to pass, each question must be answered to the satisfaction of the Technical Leader. If remediation is needed, it may take the form of immediate follow-up questions, answering of the question(s) at a later date with the Technical Leader, or a repeat of the entire oral examination.

KSA's to master

1. Be able to answer a wide variety of technical DNA questions.
2. Be able to answer a wide variety of questions related to QA/QC.

Final actions

1. Have supervisor and technical leader sign off on successful completion of module.
MODULE 19  Hemoglobin

Required lecture

Isoelectric focusing and hemoglobin

Required reading

1. Study the articles in the reference binder on this topic.

Practical exercises

When you run analytical procedures during training or receive samples for competency testing, take notes and fill out worksheets as if you were working on a real case.

After observing the procedure and having demonstrated the procedure to the trainer, do the following experiments.

1. Obtain and/or prepare several samples and practice hemoglobin phenotyping using isoelectric focusing until you feel proficient with the procedure.

Competency test

Obtain a hemoglobin competency test. The correct hemoglobin type must be obtained for each sample.

KSA's to be mastered

1. Be able to perform hemoglobin typing.
2. Understand the structure-function relationship of the protein and know its structure (monomer, dimer, etc.), genetics (phenotypes, alleles), and discrimination potential. Also read about its persistence in dried stains.
3. Review the different separation/detection methods of different phenotypes (cellulose acetate electrophoresis, starch gel electrophoresis, polyacrylamide gel electrophoresis, and isoelectric focusing methods).
4. Read about isoelectric focusing in ultrathin gels. Understand the concepts and why isoelectric focusing is the method of choice for hemoglobin phenotype determination.
5. Read about methemoglobin. Know what it is, why it forms, and how to identify it using isoelectric focusing gels.
6. Be able to explain the theory and how these tests are run to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor evaluate the results of the competency test.
3. Have supervisor sign off on successful completion of module. The initials of the supervisor...
indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
MODULE 20 Bloodstain Pattern Analysis

Required lectures

History of bloodstain pattern analysis
Basics of bloodstain pattern analysis and blood droplet dynamics
Basic forensic photography

Required reading

1. Study the available literature in the laboratory.
2. Study the Crime Scene Investigation and Reconstruction Manual with respect to the examination of bloodstain patterns.

Practical exercises

1. Participate in a week-long bloodstain pattern workshop (internal or external).

Competency test

None

KSA’s to be mastered

1. Be able to examine bloodstain patterns and offer an opinion as to their mode of deposition (commensurate with experience).
2. Know about the history, terminology, and basic principles of bloodstain pattern analysis and blood droplet dynamics.
3. Be able to explain the theory to someone who does not have a scientific background.

Final actions

1. Discuss the module with direct supervisor, including review of results and discussion of theory and practical aspects of module.
2. Have supervisor sign off on successful completion of module. The initials of the supervisor indicate that all practical exercises have been completed and the correct results have been obtained on the competency test (if applicable).
List of Department of Forensic Biology lectures

Technical lectures

Basics of forensic photography
Basics of microscopy
Body fluid identification
Identification of P30 using ELISA
Standardized sexual assault evidence collection kits
DNA extraction
DNA quantitation
PCR theory
Basics of STR typing
Basics of gel electrophoresis on the ABI 377
Basics of capillary electrophoresis on the ABI 310
Basics of mtDNA typing
Department of Forensic Biology PCR validation studies
Basics of STR mixture interpretation
Basics of population genetics and statistics
Isoelectric focusing and hemoglobin typing
History of bloodstain pattern analysis
Basics of bloodstain pattern analysis and blood droplet dynamics

Non-technical lectures

Safety and Right to Know (OCME Director of Health and Safety)
Case management
Quality assurance and quality control
Accreditation and certification
Basics of the legal system
CODIS

Additional lectures may be added as needed.
## FORENSIC BIOLOGY TRAINING CHECKLIST
### lectures

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## Forensic Biology Training Checklist

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## FORENSIC BIOLOGY TRAINING CHECKLIST

### modules

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Written examination

This set of questions is found in g:\users\biology\training\question. Use “save as” to save your version to your own directory (h:\users\yourname) - not the public Forensic Biology directories. Type in the answers, review with your direct supervisor, and supply to the Technical Leader prior to the oral examination.

DNA extraction:

1. Describe the DNA that is obtained by using a Chelex extraction method.

2. Describe the DNA that is obtained by using an organic extraction method.

3. What is Chelex 100 and what is its role in the extraction procedure?

4. When a bloodstain is extracted, why is it important to remove as much of the supernatant from the initial soak as possible?

5. What properties of sperm cells allow a differential extraction procedure to be effective?

6. During a differential extraction, what is the purpose of:
   - PBS
   - proteinase K
   - DTT

7. During a differential extraction, sperm are seen at steps 10 and 19 for all six samples being extracted. When the QuantiBlot results come back, none of the sperm cell fractions have DNA in them; all the epithelial cell fractions and swab remains fractions do. How can this be explained?

8. The only evidence remaining on a 1985 sexual assault case is a vaginal slide. You examine the slide and see only four sperm heads and a low amount of epithelial cells. What do you do next?

9. A decomposed body is found that remains unidentified. The Medical Examiner asks what sorts of samples should be collected for possible DNA testing. What do you tell her? List sample types in order of preference.

10. In a differential extraction, what is the purpose of the swab/substrate remains fraction?

DNA quantitation:

3. Name three methods of DNA quantitation that require the use of controls or reference standards.

4. Name one method of DNA quantitation that does not require the use of controls or reference standards.

5. Explain the mechanism of DNA quantitation using U.V. absorption.

7. What is the purpose of quantitating DNA samples using U.V. absorption for RFLP analysis?

8. Name one advantage of using fluorescence to measure DNA quantity as opposed to U.V. or yield gels.

9. What causes donut bands on a Q-blot membrane?

10. Why is there a notch in the upper left hand corner of the Q-blot membrane?

11. What is the purpose of the citrate buffer wash prior to color development of a Q-blot membrane?

12. Explain the color development reaction of the Q-blot assay.

13. What type of DNA probe is used in a Q-blot assay?

14. You get a Q-blot worksheet back and the results for your sample are inconclusive for the neat sample and 0.31 ng/20 ul for the 1/10 dilution. What volume do you submit for Quad amplification and why?

**PCR theory:**

1. What are the 3 steps in the PCR reaction?

2. Who invented PCR? Where was it invented and for what reason?

3. What is meant by “primer dimer”?

4. What is meant by “stutter”? Give an explanation for the cause of stutter one repeat unit longer and one repeat shorter than the desired product?

5. Excluding the inactivation of polymerase and the exhaustion of primers or dNTPs, give 3 other reasons why the amplification plateau occurs in PCR?

6. Explain allelic dropout and preferential amplification?

7. State 5 factors that influence primer specificity and stringency.

8. What direction does primer extension occur?

9. What is the relationship between the time of the extension phase of PCR and the size of DNA fragment being amplified?

10. What is the relationship between the concentration of Mg+2 and the concentration of dNTPS in the optimization PCR?

11. What is meant by a non-specific amplification product?

12. What is meant by non-template nucleotide addition?

13. If you were designing primers for PCR amplification how would you design the reverse
primer to reduce the amount of non-template nucleotide additions? Why?

14. *Tag* DNA polymerase offers the obvious advantage of being thermostable. How does the property of thermostability help the PCR reaction? Compare PCR using *Tag* polymerase with PCR using the Klenow fragment of *E. coli* DNA polymerase I.

15. How can the addition of NaOH be beneficial in the PCR reaction? When would you add it?

16. What are reasons for having a minimum amount of DNA to add to an amplification and what are possible outcomes of using less?

17. Nucleotide misincorporation occurs approximately once in every 1 million base pairs in PCR. Under what conditions would this adversely affect the fidelity of the PCR process?

STR’s:

1. Describe the advantages and disadvantages of forensic STR testing in comparison to RFLP testing and HLA/DQA1 polymarker.

2. Why are most of the STR’s used for forensic DNA typing tetranucleotides as opposed to dimeric?

3. Are the STR loci currently used in our lab human specific?

4. What is a non-consensus or microvariant allele? Give three examples for different loci and explain the allele nomenclature.

5. List the different classifications used to describe the complexity of the STR core repeat sequences. Give an example for each type.

6. Using fluorescent STR allele detection technology, how many reaction primers are labeled and which labeling colors do you know?

7. Explain the function of the matrix file. What are indications for a problem with the matrix?

8. What is the function of the APS and the TEMED in the Long Ranger gel mix?

9. You are troubleshooting a gel run. What are possible explanations for the following observations:
   - no PCR product present in all lanes, but red size standards visible
   - size standards and positive control show more than the expected bands
   - red streaks running vertically
   - any color streaks running horizontally
   - signals are faint on one side of the gel only
   - lanes on the left and right hand side are cut off

10. You are troubleshooting a CE run. What are possible explanations for the following observations:
    - no PCR product present in all lanes, but red size standards visible
    - most samples look OK, but one sample has neither red standard nor alleles
    - the initial samples pass, but later peaks get progressively lower and wider
- all peaks are present in all colors
- there are lots of spikes in almost every lane

11. How are the DNA molecules injected into the capillary and why is the quality of the formamide critical?

12. How does the Genotyper software assign allele names to Genescan sizing data? For the Quad and YM1 systems? For the Profiler Plus and Cofiler systems?

13. List the different scenarios where STR results might be mistaken for a DNA mixture:
   possible PCR artefacts
   possible electrophoresis and detection artefacts
   possible genetic abnormalities

14. You have obtained a VWA result in Quad of 16, 17. Upon amping and typing in ProfilerPlus, the VWA result is a 17. You have eliminated sample mix-up as a cause. What is the explanation?

mtDNA:

1. Human mtDNA consists of approximately how many base pairs?

2. How does mtDNA exist in all animal cells types?

3. Do the distribution of bases differ between the heavy and light strands of mtDNA? If so, explain?

4. The control region of mtDNA is also known as the displacement loop (D-loop). Why is this area referred to as the displacement loop? How large is this region?

5. What two regions of the mtDNA D-loop are used in forensic analysis? Why and how large are they in length?

6. What is the Anderson Sequence? How was it constructed and what is its use?

2. What is the difference between manual sequencing to automated sequencing. Include major advantages of automated sequencing?

3. There are two major types of automated sequencing kits. One utilizes dRhodamine fluorescently labeled ddNTP’s and the other utilizes BigDye chemistry. What are the advantages to each system?

4. What is the structural difference of a 3’ dNTP as compared to a 2’ 3’ddNTP. Which NTP is utilized in sequencing and why?

5. Signal dropout is common after the C stretch of HV1 when the polymorphic T is absent from within it. What is the most likely cause?

6. How can the remaining sequence be deciphered past this C stretch?

12. Circle the correct strand.
Primers are referred to as forward and reverse primers. Forward primers synthesize the F/R strand and the complement of the F/R strand. Reverse primers synthesize the F/R strand and the complement of the F/R strand.

QA/QC:

1. (a) Define accreditation.
   (b) How often is the Department of Forensic Biology required to undergo accreditation?
   (c) Who performs the accreditation?
   (d) What is the significance of an accredited lab?

2. List all of the QC tests that are used to monitor:
   (a) evidence examination
   (b) P30 ELISA
   (c) serology
   (d) chelex extraction
   (e) QuantiBlot
   (f) PCR
   (g) STR analysis

3. What is the significance of the inventory sheets that are posted at each station?

4. Give the definitions for the following terms and/or acronyms:
   (a) ABC
   (b) ASCLD and ASCLD/LAB
   (c) CODIS, NDIS, GDIS, SDIS, and LDIS
   (d) criminalist and criminologist
   (e) DAB
   (f) examining and interpreting analysts
   (g) forensic scientist
   (h) NIJ
   (i) NIST
   (j) NRC
   (k) NYCLAC and NYCLAC TWG
   (l) proficiency testing
   (m) QC and QA
   (n) TWGDAM and SWGDAM

Population genetics and statistics:

1. Assuming no laboratory error, is DNA testing always accurate? always precise? always conclusive?

2. How is population frequency calculated for autosomal STRs, Y chromosome STRs, mitochondrial DNA? How is each inherited and why are they calculated differently?

3. What is the second NRC report, who wrote it, and have you read chapters 4 and 5?

4. What is the ceiling principle and is it still used?

5. How are homozygote and heterozygote frequencies calculated under Hardy Weinberg
equilibrium?

6. What is population substructure?

7. What is theta and how is it used in calculations?

8. What degree of relatedness is assumed and what value is used

9. Is the homozygote or heterozygote frequency correct for substructure and why are homozygotes corrected or not corrected?

10. Given a best estimate of frequency, in what range does the true frequency lie? How was this determined? Why is a best estimate given and not a true value?

11. If only one allele of a locus can be reported, how can the locus frequency be calculated?

12. Do the corrected Hardy Weinberg formulas apply if the evidence and the subject is from the same subgroup (isolated “New England fishing village”) What is used?

13. If a man whose DNA type matches the sperm in a rape, says, I did not commit the rape but my younger brother did it, what is the best way to determine the probability that they share the same genotype (approaching 100% probability)? What is the best thing to do if they are identical twins?

14. If the brother in the above non-twin scenario cannot be found, what can be done?

15. Is the same formula with minor variation used to calculate relatedness for all relatives? Why or why not?

16. How do subtypes of alleles, trinucleotide core repeats (i.e. 8.3), or sequence polymorphisms affect the calculation of DNA profile frequency? How are they handled in the calculations and what happens when the population database is constructed? What is the frequency that one of these events occurs and does this invalidate non-sequencing STR DNA typing?

17. How many different Quad DNA profiles are there (show your calculations)?

18. Assume the following mixture has two contributors. How many different combinations of contributors are possible (show your calculations)?

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<td>9, 10</td>
<td>6, 7, 9, 10</td>
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19. Assume the VWA allele 14 is found in 10% of the population and allele 15 is found in 4% of the population. According to Hardy-Weinberg, how frequently are the 14, 14 and 14, 15 genotypes found? Repeat the calculations using the method recommended by the NRC.

Miscellaneous:

1. What is the relationship between the amount of DNA in a sperm cell compared to an epithelial cell?

2. How much DNA (weight) is in a single epithelial cell (show your calculations)?
3. How many sperm are necessary to yield 1 ng of DNA (show your calculations)?
STR DNA interpretation exercises

The DNA tables can be found in the departmental directory as:

g:\users\biology\training\STR-inex

These are interpretation exercises to be worked through as part of training. By practicing interpretation now, and discussing among ourselves our reasoning, you will have an easier time when a tough interpretation comes up in a case.

For each scenario, write a report. Use the standard DNA template report and import the tables by copying and pasting. Include:

- a summary section (first page of report) with statistics if appropriate
- a table of results
- an explanation to follow the table (usually more info than the summary)

Write this up on the computer as if you were reporting the results; do this independently, without consulting anyone. Some are complicated and require thought! This is not something you can do at the last minute. Use "save as" to save your version to your own directory (h:\users\yourname) - not the public Forensic Biology directories.

In each case, sperm was found on the vaginal swab.
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<tr>
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<td>6, 8</td>
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