



TRAINING PROGRAM FOR BODY FLUID IDENTIFICATION
NORTH CAROLINA STATE BUREAU OF INVESTIGATION FOR
TECHNICIANS

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1. PURPOSE AND SCOPE

The purpose of this manual is to provide a consistent training program for the analysis of forensic evidence by Technicians at the Molecular Genetics Section at the North Carolina State Bureau of Investigation. This program provides individuals with the theoretical background and the working knowledge to assist Qualified Analysts conducting scientific tests on forensic evidence using validated procedures under the direction/supervision of a Qualified Body Fluid Analyst. Heavy emphasis shall be placed on quality assurance of all tests performed, data integrity via thorough documentation, and excellence in obtaining consistent and congruous results.

The training program detailed in this document provides the following:

- Theoretical knowledge of the principles of body fluid identification testing.
- Working knowledge of the principles and practices of serological theories such as antigen antibody reactions, immunodiffusion, etc. as they relate to the forensic analysis of body fluids.
- The ability to perform accurate and consistent forensic analysis on forensic case material.

2. REQUIREMENTS FOR QUALIFICATION

2.1 Prerequisites

Individuals must possess a strong scientific background and have completed a Bachelor's of Science in a Science and possess course work in biology, chemistry, biochemistry and genetics.

2.2 Competency Tests

A Qualified Technician must pass a competency test in the specific area that he/she will be assisting the Qualified



Analyst. These tests are to determine the trainee's ability to consistently analyze body fluids from a variety of sources. Competency tests may include blood, mixed fluids, and simulated cases when appropriate.

2.3 Written Examination

A written examination shall examine the trainee's understanding of the theoretical and working knowledge of body fluid identification tests and how they are applied in the laboratory.

2.4 Proficiency Testing

The Qualified Technician will participate in a Proficiency Testing Program in the specific area in which they were trained as soon as possible after training has been completed.

3. **INSTRUCTIONS FOR THE TRAINING OFFICER**

3.1 This program is designed to provide each trainee with the theoretical background and working knowledge to reliably assist in the analysis of forensic evidence utilizing body fluid identification tests. The training officer must pay very close attention to detail and ensure that all quality assurance guidelines are being followed for every sample processed in the training program. By ensuring each trainee maintains a high degree of concentration and awareness during the performance of his/her training, the proper techniques will be learned and later successfully applied to actual casework.

3.2 The order of topics listed in this manual are not necessarily in the chronological order that the tests will be performed. It may be necessary to learn and perform some techniques out of order. In this case, it is the responsibility of the training officer to provide the trainee with a clear explanation of any missing points or steps and later logically tie everything together.

3.3 It is the responsibility of the Training Officer to point out pertinent scientific literature and technical manuals included in the bibliography to the trainee so that they may become familiar with these readings.

3.4 It is the responsibility of the Training Officer to explain potential safety hazards to each Trainee BEFORE performing a task that may involve said potential safety hazard.



4. INSTRUCTIONS FOR THE TRAINEE

4.1 The trainee is required to keep files on all work completed. These files should include but are not limited to the Training Manual Log Sheet, worksheets, and copies of reports issued in cases they participate in. These files will be checked periodically by the Training Officer and/or SAC.

4.2 The readings assigned are very important. While it is not necessary to memorize reagent recipes, it is necessary to become familiar with the principles of each test and the protocol and be able to perform all duties independently. The trainee is expected to become familiar with the literature that pertains to the forensic analysis of body fluid identification testing that is included in the bibliography.

5. SAFETY ISSUES, INTRODUCTION TO THE LAB, AND ORGANIZATION

5.1 There are many potential hazards that exist in the laboratory. While the exposure to all hazards can be minimized or avoided, it is the responsibility of the Training Officer to ensure the Trainee is aware of all potential hazards. These potential hazards include but are not limited to the following:

Infectious Agents

- A. Viral agents, including HIV and Hepatitis
- B. Bacteria, including sexually transmitted diseases
- C. Fungi
- D. Parasites

Hazardous Materials

- A. Caustic Agents (Acids and Bases)
- B. Carcinogens/Mutagens
- C. Teratogens
- D. Organic Chemicals

Electrical Hazards

Shock from any piece of electrical equipment

Burn Hazards

Autoclaves Bunsen burners
Sterilizers Hot Plates



Eye Damage

Alternate light sources

5.2 Laboratory Safety Procedures

5.2.1 Individuals must be trained in laboratory safety by the Section Safety Officer prior to the commencement of training. Various manuals are provided that must be followed to ensure safety of all laboratory personnel. The following manuals are to be used for reference and guidance for laboratory safety: MSDS Notebook, Molecular Genetics Section Manuals, Chemical Hygiene Program, Bloodborne Pathogen Program, and the DOJ Safety Manual. The trainee will also be briefed on the fire evacuation plan for the laboratory.

5.2.2 It is the responsibility of the training officer to alert the trainee to safety hazards specific to this laboratory, including all items mentioned in 5.1.

5.3 Laboratory Orientation

The trainee may be taken throughout the laboratory and shown areas of interest to their work. They will also be provided with a written job description, an organizational chart, and various manuals including the Crime Laboratory Procedures Manual, SBI Policy Manual and the Section Quality Assurance Manual.

6. **ASEPTIC TECHNIQUE AND CONTAMINATION CONTROL**

6.1 This Section uses the Polymerase Chain Reaction (PCR) technology which allows very small amounts of DNA to be amplified over a billion times. Because of the sensitivity of this technique, contamination control is a very serious issue that must be emphasized and practiced with every sample, starting with the identification of the body fluids present on the evidence. The Evidence Handling Procedures must be strictly followed.

6.1.1 All items used in the identification, transfer and isolation of forensic samples must be sterile and/or free of contaminate DNA.

6.1.2 Gloves must be worn at all times while handling samples. This is to protect both the analyst and sample.



6.1.3 A fresh, sterile pipette tip must be used for each transfer of fluid or chemical to be used for DNA analysis.

6.1.4 Special precautions must be taken to ensure that the bench and surrounding areas have been properly decontaminated.

6.1.5 Scissors, tweezers, and other instruments used for cutting or extractions shall be sterilized in between each sample.

6.1.6 The preparation of the known and unknown samples shall be separated by time.

7. DOCUMENTATION

7.1 Goals

7.1 To provide protocols for the preparation of reagents and performance of tests to ensure consistent, reliable results.

7.2 To provide a thorough record of events for each case analysis.

7.2 Protocol Notebook

The Body Fluid Identification Procedures Manual shall be made available to each Technician. The analyst shall not deviate from any procedure without permission from the SAC. Any deviation (purposely or by mistake) from the protocol shall be thoroughly documented on the worksheet at the time of occurrence.

7.3 Buffers and Solutions Recipe Book



The working copy of the appropriate QC forms, which includes procedures for preparing solutions, will be maintained in the QC notebook.

7.4 Worksheets

The purpose of the worksheets are to provide a means to thoroughly document each step of the analytical process. Each worksheet is to be completed either during or as soon as possible following the step.

8. RECEIVING AND HANDLING OF EVIDENCE

8.1 Goals

- 8.1.1 To obtain a working knowledge of factors and conditions that influence the deterioration of evidence as it relates to packaging, handling, and storage conditions and time.
- 8.1.2 To develop a thorough understanding of evidence handling procedures.
- 8.1.3 To develop a thorough understanding of the necessity for detailed comprehensive notes and adequate labeling of evidential materials.

8.2 Tasks

- 8.2.1 Read and become thoroughly familiar with the SBI Evidence Handling Procedures regarding receiving, identifying, and handling of evidence; as well as specific guidelines for handling biological evidence within the Section.

9. ANALYSIS OF BODY FLUID SAMPLES

9.1 Goals

- 9.1.1 To develop a basic understanding of the methodology and theory of chemical, microscopic, and immunological testing procedures used to identify blood and the species from which the blood may have originated.



- 9.1.2 To develop a basic understanding of the methodology and theory of chemical, microscopic, and immunological testing procedures used to identify semen and sperm.
- 9.1.3 To develop a basic understanding of the methodology and theory of chemical testing procedures used to identify amylase.
- 9.1.4 To develop skills that will allow the trainee to independently and successfully analyze forensic samples.
- 9.1.5 To become familiar with the sensitivity and limitations of the procedures used.
- 9.1.6 To develop a cognizant understanding of contamination issues and the steps necessary to avoid contamination.
- 9.1.7 To understand the use of controls during each procedure.
- 9.1.8 To become familiar with and understand the function of any buffers, solutions, or reagents used.
- 9.1.9 To become familiar with all documentation required.

- 9.2 Tasks
 - 9.2.1 Prepare any buffers, solutions, or reagents necessary for testing.
 - 9.2.1 Understand and perform quality control checks necessary on buffers, solutions, reagents and test kits used.
 - 9.2.3 Perform testing on various sample types which allow for ample testing for each analytical procedure used (**see Appendix III for specific training tasks for each procedure**). The training samples will represent materials commonly encountered in forensic casework. The Training Officer will initially present the trainee with a short lecture on the analytical procedure in question. The trainee is then given samples on which to practice. Once a block of instruction is completed, the trainee will receive a written test and a set of competency samples to analyze. The exact number of samples will be determined by the training officer in accordance with experience and ability of the trainee. Upon successful completion of the block of instruction, the trainee will start on the next block of instruction.



9.2.4 Blocks of instruction the trainee will complete to identify blood include:

9.2.4.1 Phenolphthalein testing (presumptive chemical tests)

9.2.4.2 Takayama testing (microcrystalline identification)

9.2.4.3 Species origin testing (immunological methods)

9.2.4.3 ABACard HemaTrace testing

Note: See Appendix III for specific training tasks for each procedure.

9.2.5 Blocks of instruction the trainee will complete to identify semen include:

9.2.5.1 Acid Phosphatase testing (presumptive chemical tests)

9.2.5.2 Spermatozoa identification (microscopic identification)

9.2.5.3 ABACard P30 testing

Note: See Appendix III for specific training tasks for each procedure.

9.2.6 Blocks of instruction the trainee will complete to identify amylase include Phaedebas testing (presumptive chemical tests).

Note: See Appendix III for specific training tasks for this procedure.

9.2.7 Complete documentation for all procedures.

10. REPORT WRITING

10.1 Goals

10.1.1 To develop the skills necessary to effectively report body fluid identification results.

10.1.2 To develop a working knowledge of the terminology for analysis and results.

10.1.3 To become familiar with pertinent scientific literature.



10.2 Tasks

10.2.1 Read and understand pertinent scientific literature provided in the Bibliography (Appendix III).

10.2.2 Thoroughly understand and be able to accurately and concisely answer the questions in Appendix II.

10.2.3 Draft Body Fluid Identification Reports. May include but not be limited to proficiency tests, draft reports from cases previously analyzed by the section, mock cases, draft reports from current cases (NOTE: These MUST be reviewed and approved by a Trained Analyst before Technical Review and Administrative Review.)

11. COMPETENCY TESTING

11.1. Goals

Upon successful completion of all blocks of instruction, the trainee will be given a series of competency tests that closely mimic forensic evidence. The trainee must score 100% accuracy in these tests. This test is the final one that the trainee must complete before being allowed to work on forensic cases.

11.2 Tasks

Successful completion of the competency tests.

12. Qualified Technician

12.1 Goals

Once the trainee completes their competency tests, they are placed with a qualified Body Fluid Analyst. Qualified technicians will work cases under the direct supervision of a Qualified Body Fluid Examiner.

12.2 Tasks

12.2.1 The qualified technician will assist the Analyst in conducting the analysis of cases, the preparation of the notes, and may write the report.

NCSBI MOLECULAR GENETICS SECTION

Training Manual

Body Fluid Identification

Body Fluid Identification Technician Training Manual: Rev. 00



12.2.2 The Analyst will observe all results, initial the notes, and evidence, and review all work conducted. The case will be assigned to the experienced examiner, and the experienced examiner will sign the laboratory report.

Revision History		
Effective Date	Revision Number	Reason
3		al Document

OVAL SIGNATURES	
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**APPENDIX I. NORTH CAROLINA STATE BUREAU OF INVESTIGATION:
Body Fluid Identification Training Manual Log Sheet**

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Training Manual

Body Fluid Identification

Body Fluid Identification Technician Training Manual: Rev. 00



Training Area	Date Completed/Initials	Trainers Initials
Education		
Safety		
1. Chemical Hazards/MSDS Sheets		
2. Electrical Safety		
3. Blood borne Pathogen Training		
4. Readings		
Laboratory Orientation and Organization		
1. Lecture		
2. Readings		
Blood Identification		
1. Lecture		
2. Readings		
Semen Identification		
1. Lecture		
2. Readings		
Saliva Identification		
1. Lecture		
2. Readings		
Quality Control and Documentation		
1. Use of QC Reagent Book		
Training Area (continued)	Date Completed/Initials	Trainers Initials
2. Use of QC forms		

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3. Documentation using Worksheets		
Receiving and Handling Evidence		
Read SBI Evidence Procedures		
Laboratory Training		
Aseptic Technique and Contamination Control		
Cleaning of Equipment		
Use of Bio-safety Hood		
Handling of Samples		
Blood Identification		
Phenolphthalein Testing		
1.1 Read and Understand Procedure		
1.2 Demonstration of Test		
1.3 Testing washed and heated samples		
1.4 Sensitivity Testing		
1.5 Specificity Testing		
1.6 Supervised Testing of Known Samples		
1.7 Competency testing		
Okayama Testing		
2.1 Read and Understand Procedure		
2.2 Demonstration of Test		
2.3 Supervised Testing of Known Samples		
Training Area (continued)	Date Completed/Initials	Trainers Initials
2.4 Testing of materials prepared for Phenolphthalein Testing		

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Body Fluid Identification

Body Fluid Identification Technician Training Manual: Rev. 00



2.5	Testing of catalase, peroxidase, and old bloodstains		
2.6	Absorbing and testing blood placed on various matrices		
2.7	Competency Testing		
Species Origin Tests			
3.1.	Read and Understand Procedures		
3.2.	Demonstration of Tests		
3.3	Supervised Testing of Known Samples		
3.4	Testing of human and animal blood samples		
3.5	Competency Testing		
Human Identification			
Acid Phosphatase Testing			
1.1	Read and Understand Procedures		
1.2	Demonstration of Tests		
1.3	Supervised Testing of Known Samples		
1.4	Testing of a variety of stains		
1.4	Competency Testing		
Confirmatory Testing			
2.1	Read and Understand Procedures		
Training Area (continued)		Date Completed/Initials	Trainers Initials
2.2	Demonstration of Tests		
2.3	Supervised Testing of Known Samples		
2.4	Testing of a variety of stains		

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Body Fluid Identification

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2.5	Competency Testing		
Christmas Tree Stain			
3.1	Read and Understand Procedures		
3.2	Demonstration of Tests		
3.3	Supervised Testing of Known Samples		
3.4	Testing of a variety of stains		
3.5	Competency Testing		
30 Analysis			
4.1	Read and Understand Procedures		
4.2	Demonstration of Tests		
4.3	Supervised Testing of Known Samples		
4.4	Testing of a variety of stains		
4.5	Competency Testing		
Abacard Analysis			
5.1	Read and Understand Procedures		
5.2	Demonstration of Tests		
5.3	Supervised Testing of Known Samples		
5.4	Testing of a variety of stains		
5.5	Competency Testing		
Training Area (continued)		Date Completed/Initials	Trainers Initials
Aliva Identification			
Aldehyde test Testing			
1.1	Read and Understand Procedure		
1.2	Demonstration of Test		

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Training Manual

Body Fluid Identification

Body Fluid Identification Technician Training Manual: Rev. 00



1.3	Supervised Testing of Known Samples		
1.4	Testing of a variety of stains		
1.5	Competency Testing		
Training Area (continued)		Date Completed/Initials	Trainers Initials
Report Writing			
	Ability to Draft a Satisfactory Report		
Competency Testing			
	Successful Completion of Practical Competency Tests		
	Successful Completion of Final Written Test		

**APPENDIX II. NORTH CAROLINA STATE BUREAU OF INVESTIGATION: General Questions For The Trainee**

1. Describe the basis for presumptive tests for blood in general.
2. Name two classes of substances that react with the presumptive tests other than blood.
3. Why is a three step procedure better than a one step blood testing procedure?
4. List the advantages and disadvantages to ABA Card testing.
5. List and explain the appropriate controls used in Ouchterlony species identification.
6. What environmental factors affect the stability of a bloodstain?
7. How does heat, such as autoclaving, affect a bloodstain?
8. Which is more sensitive, a blood presumptive test or a human origin test?
9. What is semen and list as many components that you can think of that are found in semen? Specify which are semen specific and which are not.
10. List three sources of Acid Phosphatase.
11. What components identify semen conclusively?
12. Briefly describe and draw a spermatozoa labeling the major parts.
13. Name the staining procedure used for sperm identification and identify the names of the stains that we use.
14. Are sperm haploid or diploid?
15. What does azoospermic mean? How does a person become azoospermic (list three)?
16. What is p30?
17. How do we detect p30? Describe the controls used in each test.
18. Describe the principle of the Phadebas testing procedure.
19. Describe the procedure for testing saliva with Phadebas tablets.
20. Is Phadebas testing a confirmatory or presumptive test for saliva?