



NSP CRIME LABORATORY CRIME LABORATORY MANUAL BIOLOGY



I. Introduction

The admission of biological evidence in courts of law has become commonplace. Indeed, such evidence can have an influence on the outcome of litigation. Because genetic typing results can be statistically expressed, the significance of the evidence can often be clearly articulated.

The crime laboratory is using DNA testing procedures that utilize the Polymerase Chain Reaction (PCR) to analyze Short Tandem Repeats (STRs). The crime laboratory tests for the 13 core STR loci established by the Federal Bureau of Investigation (FBI). These same 13 loci are used by the vast majority of crime laboratories throughout the United States, allowing law enforcement agencies to share information for criminal investigations. This sharing of information is accomplished by the use of the Combined DNA Index System (CODIS).

CODIS utilizes a national DNA database maintained by the FBI that contains DNA profiles of convicted felons and evidence from unsolved crimes from all fifty states. The Nebraska State Patrol Crime Laboratory is electronically linked to CODIS. CODIS can be searched to identify suspects for crimes committed in Nebraska by matching DNA from crime scenes to convicted offenders. CODIS searches are useful for linking serial violent crimes which can assist in criminal investigations.

Various guidelines provided in this manual are designed to assist the forensic DNA /serology laboratory in establishing and maintaining standards by which the law enforcement agencies (the user of the services) may assess the quality of the biological evidence analysis.

The present state-of-the-art technology in forensic DNA/serology relies on two basic principles:

- A. The exchange principle: Contact between suspect and victim which results in cross-transfer of hair or other evidence.
- B. Genetic marker polymorphism: Biological evidence such as blood, semen, saliva, urine, bones, hair and many other biological samples can provide individuality through serological analysis and DNA typing which can be associated with donor(s) and exclude non-donors.

Conventional serology and forensic DNA testing utilize certain types of biological evidence which bear some measure of individuality which can exclude or include potential non-donors or donors, respectively.



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It is recognized that the field of forensic DNA/serology is not a static one.

Amendments to the following guidelines will be inevitably required in the future to encompass new scientific developments.

In the following sections, various types of evidence typically received in the DNA/serology section of the Crime Laboratory will be reviewed. For optimal success, various DNA/serology evidence should be collected and preserved according to the guidelines provided. Biological evidence deteriorates rapidly. Therefore, DNA/serological evidence must be preserved appropriately and transported from the crime scene to the Laboratory as soon as possible. The "chain of custody" of such evidence must be carefully maintained.

II. General Guidelines for Collection and Preservation of DNA/Serology Evidence

A. Biological evidence that can be analyzed by serology and DNA (Deoxyribonucleic acid) analysis is generally of the following types.

1. Blood and bloodstains.
2. Semen, seminal stains, vaginal and seminal fluid mixtures.
3. Saliva and salivary stains.
4. Tissues and cells.
5. Bones, organs.
6. Urine.
7. Hairs.

B. It should be noted that while research results have indicated successful polymorphic genetic marker typing from the above listed biological specimens, not all biological evidence submitted to the Crime Laboratory in casework can be successfully typed, either due to degradation, environmental insults, or simply by the very nature of its being biological.

C. Evidence such as perspiration, saliva, serum, etc. now may yield significant information due to Polymerase Chain Reaction (PCR) DNA technology.

D. Biological evidence should be handled with "Universal Precautions." Gloves must be worn while the evidence is being handled and protective garments should be worn for protection from HIV, hepatitis or other infectious agents. All biological samples MUST BE ASSUMED to be infectious at all times regardless of their history.

E. Biological evidence can be transferred from one person to another directly or indirectly. This can be used to link various individuals with one another

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(suspect's semen on victim, victim's blood on suspect's clothing, suspect's hair in victim's hand) or to an object (knife, gun, etc.) or a place (blood of the victim in suspect's house or car).

F. Direct Transfer of Biological Specimens

Liquid specimens such as blood, saliva, semen, urine, etc. are often deposited directly and usually identified in the crime scene as dried stains. Seminal fluid or other samples such as bone, tissue or hair can be directly or indirectly transferred to a location.

G. Secondary (Indirect) Transfer

This is a common type of transfer in hair evidence and sometimes occurs in case of biological specimens also. An uninvolved person can pick up a victim's hair from a suspect's car seat and transfer that to another object or location unknowingly. Blood from a victim's clothing can be transferred to a witness' clothing or an ambulance.

A secondary indirect transfer does not provide a direct connection between a crime and a person or between a victim and a suspect.

H. Collection and Preservation of Biological Evidence

Successful analysis of biological evidence depends a great deal upon the type of evidence and the manner in which it is collected and preserved. Collected evidence should be properly documented, collected, packaged and preserved. The chain of custody should be maintained to meet the legal and scientific admissibility standards. It is most important to avoid cross contamination, particularly due to the highly sensitive nature of PCR DNA analysis.

I. Documentation

The original condition and the position and location of each item of evidence should be recorded prior to collecting the evidence. Each item should be photographed and/or videotaped before being handled or collected.

1. The condition of each item should be noted. For example, a yellow stain (suspected urine) on snow that is melting should be noted as such.



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2. The packaging should be paper, such as a paper bag. Each item of evidence should be labeled, sealed and noted with an exhibit or other identifying number.
3. In order to perform serological and DNA analysis on biological evidence, it is of utmost importance to collect the correct types of evidence and use the correct preservation procedure.
 - a. Some of the issues that might be raised for admissibility of evidence in court regarding such procedures are:
 - 1) Techniques used to collect and document evidence.
 - 2) Type of evidence.
 - 3) Location from where collected.
 - 4) Packaging method used.
 - 5) Preservation method used.
4. Please remember that the biological activity of an item depends a great deal upon proper collection and preservation, which will minimize degradation and decomposition.
5. **AVOID CROSS-CONTAMINATION** of evidence. This is a legal issue of utmost importance in DNA analysis.

III DNA Typing

Characterization, or "typing," of deoxyribonucleic acid (DNA) for purposes of criminal investigation can be thought of as an extension of the forensic typing of blood that has been common for more than 50 years; it is actually an extension from the typing of proteins that are coded for by DNA to the typing of DNA itself. Genetically determined variation in proteins is the basis of blood groups, tissue types, and serum protein types. Developments in molecular genetics have made it possible to study the person-to-person differences in parts of DNA that are not involved in coding for proteins, and it is primarily these differences that are used in forensic applications of DNA typing to personal identification. DNA typing can be a powerful adjunct to forensic science.

- A. DNA, the active substance of the genes, carries the coded messages of heredity in every living thing: animals, plants, bacteria, and other microorganisms. In humans, the code-carrying DNA occurs in all cells that have a nucleus, including white blood cells, sperm, cells surrounding hair roots, and cells in saliva. These would be the cells of greatest interest in forensic studies.



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- B. Human genes are carried in 23 pairs of chromosomes, long thread-like or rod-like structures that are a person's archive of heredity. Those 23 pairs, the total genetic makeup of a person, are referred to as the human "diploid genome." The chemistry of DNA embodies the universal code in which the messages of heredity are transmitted. The genetic code itself is spelled out in strings of nucleotides of four types, commonly represented by the letters A, C, G and T (standing for the bases adenine, cytosine, guanine and thymine), which in various combinations of three nucleotides spell out the codes for the amino acids that constitute the building blocks of proteins. A gene, the basic unit of heredity, is a sequence of about 1,000 to over 2 million nucleotides. The human genome, the total genetic makeup of a person, is estimated to contain 50,000-100,000 genes.
- C. Much of the DNA, the part that separates genes from one another, is noncoding. Variation in the genes, the coding parts, are usually reflected in variations in the proteins that they encode, which can be recognized as "normal variation" in blood type or in the presence of such diseases as cystic fibrosis and phenylketonuria, but variations in the noncoding parts of DNA have been most useful for DNA typing.
- D. Except for identical twins, the DNA of a person is for practical purposes unique. That is because one chromosome of each pair comes from the father and one from the mother; which chromosome of a given pair of a parent's chromosomes that parent contributes to the child is independent of which chromosome of another pair that parent gives to that child. Thus, the different combinations of chromosomes that one parent can give to one child is 2^{23} , and the number of different combinations of paired chromosomes a child can receive from both parents is 2^{46} .
- E. Forensic DNA typing often involves samples that are degraded, contaminated, or from multiple unknown sources. The procedures sometimes cannot be repeated, because there is too little sample. It often involves matching of samples from a wide range of alternatives in the population and thus lacks built-in consistency checks. Except in cases where DNA evidence excludes a suspect, assessing the significance of a result requires statistical analysis of population frequencies.
- F. When only very minute quantities of DNA are available, amplification of the samples using the Polymerase Chain Reaction (PCR) is necessary for obtaining sufficient material for analyses. This technique has revolutionized forensic DNA typing dramatically. DNA samples from sources such as paraffin embedded tissue, saliva, and hair roots can be successfully amplified and typed.

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- G. Techniques for analyzing DNA are changing rapidly. PCR method, for example, allows detection even from a few hundred nucleated cells. New ways to demonstrate variations in the genome are being developed. The current techniques are likely to be superseded by others that provide unambiguous individual identification.

IV CODIS

CODIS is short for the Combined DNA Index System. CODIS is a law enforcement tool that blends forensic science with computer technology for solving violent crimes. It enables federal, state and local laboratories to exchange and share DNA profiles electronically. CODIS can be used to identify suspects for unsolved crimes, such as linking an unsolved crime in Nebraska to a known offender from California. CODIS can also be used for linking serial crimes to one another. So if a serial criminal is committing crimes in Nebraska, Colorado, Iowa and Kansas, and leaving DNA evidence behind, CODIS can help established the connection.

- A. CODIS has three hierarchal levels: Local, State and National. The National DNA Index System is the highest level and is what enables all local and state laboratories to exchange and compare DNA information. The state DNA index is the repository for all the DNA profiles in the state. The national DNA index is the repository for all the DNA profiles in the country. The Nebraska State Patrol Crime Laboratory is the only CODIS laboratory in Nebraska.
- B. CODIS uses two indexes for DNA searches to gather investigative information. These are the forensic index and the convicted offender index. The forensic index contains DNA profiles from evidence collected from crime scenes, such as semen stains from a sexual assault or blood from a homicide. The convicted offender index contains DNA profiles from people convicted of qualifying crimes. In Nebraska those people who are required to submit a DNA sample for CODIS are those convicted of sex crimes and violent crimes.
- C. The state DNA index can be searched to compare DNA profiles from within the state. The national DNA index can also be searched to look for matches. National searches are performed on a weekly basis. Searches using the national DNA index compare DNA profiles from the entire United States. Of course matches can only be identified with DNA profiles in the database. If the perpetrator is not in the database, no offender match will be made.



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V. Examinations

A. Sexual Assaults

Sexual assaults often are secluded crimes in which the victim and the assailant are the only witnesses. Frequently, even the victim cannot provide eye witness testimony as to the attacker's identity. In such circumstances, the biological and physical evidence factors that are interchanged between the victim and assailant assume a leading role in the investigative and prosecutive aspects of the cases. Rape differs from other violent assaultive crimes in that very often the perpetrator leaves personal biological evidence such as semen at the scene.

Semen can provide proof of sexual contacts and, in being analyzed for polymorphic genetic markers and DNA, can often provide the identity of the semen donor. Therefore, it is essential for the investigator to follow the guidelines provided below in using the sexual assault kit and collection of specimens from victims of sexual assault.

Laboratory tests can identify and confirm the presence of semen. DNA testing can be used to determine the semen donor. DNA testing may also be successful for identifying the victim's vaginal fluid on items such as the suspect's underwear and used condoms.

B. Blood

Dried bloodstains comprise the bulk of the evidence submitted to the Laboratory from homicide crime scenes. A few examples of liquid and dry blood stains of value to investigators include blood left behind by wounded suspects on victim's body or clothing, sinks, window sills, doorways and even in the snow. Polymorphic genetic markers and DNA analysis can be performed on such evidence. Please see the guidelines below for collection of such bloodstains.

Laboratory tests are necessary to positively identify blood. The appearance of blood can vary greatly, depending on age and other factors. Laboratory tests can determine the species of the source. If human, DNA testing can be used to determine the identity of the donor.

C. Other Significant Body Fluids

Many other types of tissues make their way to the DNA/serology section. Such samples can be used for DNA testing.



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Many types of physiological secretions and excretions may be transferred or deposited at crime scenes. They include saliva (i.e., cigarette butts, cups, bite marks) perspiration (clothing), vaginal secretions (clothing, condoms), and urine to name the majority of such fluids. Many of the same genetic markers found in blood are also present in dried stains of many physiological fluids including semen, saliva and vaginal secretions. Please follow the guidelines provided in collection and preservation of such evidence.

D. Hair Evidence

Forensic hair analysis is performed to identify the species of origin and if human, determine race and body part from which it originated. Microscopic hair comparisons (to determine the donor of question hairs) are not performed at the crime lab. Although not performed at the crime lab, human hairs can be compared microscopically with standard hairs from potential donors and a conclusion drawn whether the suspected donor is eliminated or is a potential source. Human hair cannot be positively associated with any one individual by microscopic examination. The purpose of hair evidence is to establish a link between a possible suspect and the victim. Finding common hair would be of little significance if it is well established that the victim and the suspect have had frequent prior contact. Hair of a suspect found on victim's clothing etc. with whom the latter had shared a common residence, for example, would be expected and therefore has little evidential value.

Hair examinations will be very limited and will only be done in extreme case situations when no other biological evidence is available for DNA analysis and with prior consultation.

DNA testing can be performed on hair root sheaths even when the hairs have been processed for microscopic examination. Mitochondrial DNA testing can be performed on the hair shafts by specialized laboratories. The crime lab does not perform mitochondrial DNA testing.

VI. Guidelines for Collection and Preservation of DNA/Serology Evidence

A. Reference DNA Samples

Suitable reference samples for DNA analysis are generally liquid reference blood samples collected in purple top vials or buccal swabs. Please note the crime laboratory may require liquid reference blood samples depending on specific case circumstances. In cases such as homicide, it may not be possible to collect reference liquid blood

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depending on case circumstances. Other tissues such as pulled head hairs or bodily tissues (i.e., liver, kidney) collected at autopsy can be used. If there are any questions about which type of reference samples to submit, please contact the crime laboratory for discussion and consultation.

1. Liquid Blood Samples. For the collection of reference blood samples, please refer to section VI. C. 1. below.
2. Buccal swabs. Buccal swabs can be collected as reference DNA samples using the following procedures:
 - a. The subject should not smoke, eat, drink or chew gum or tobacco for at least 30 minutes prior to swab collection.
 - b. Put on a pair of biohazard barrier gloves (i.e. latex gloves).
 - c. Open a sterile swab package being sure not to touch the cotton portion of the swab.
 - d. Remove a swab from the package and place swab in subject's mouth. Vigorously brush the swab between the gum line and cheek for 5 to 10 seconds being sure to cover the entire surface of the swab. Remove the swab from the subject's mouth and place it on a sterile surface (such as the inside of the swab package) or on a swab drying rack or similar device.
 - e. Repeat using a new swab on the other side of the mouth.
 - f. Let the swabs air dry for approximately 30 minutes.
 - g. Label a clean, unused, paper envelope or swab box and placed both swabs inside.
 - h. Seal the package with tape and initial the seal.
 - i. Transport to the crime laboratory as soon as possible.
 - j. Do not leave evidence in a warm, humid environment. Dry swabs packaged in paper envelopes can be temporarily stored at room temperature. They should be place in the freezer for long term storage.
3. Pulled hairs. Pulled hairs can be used if necessary as reference DNA samples. Please refer to sections VI. B. 2. f. and g. below for collection.
4. Other bodily tissues. Bodily tissues such as liver, heart, muscle, bone marrow, etc. may be collected at autopsy and can be used if necessary as reference DNA samples.

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B. Sexual Assault Kit

An ideal sexual assault evidence collection kit for collecting evidence from the victim should contain all of the following items:

1. One 5 ml purple top vial (EDTA) for blood
2. Swab or filter paper for saliva
3. One paper box each for
 - a. vaginal swabs, 4 swabs should be collected
 - b. oral swabs, 4 swabs should be collected
 - c. rectal swabs, 4 swabs should be collected.
4. One cardboard slide holder each for
 - a. vaginal slides (one or two)
 - b. anal slides (one or two)
 - c. oral slides (one or two)
5. Envelopes for
 - a. pulled pubic hair sample
 - b. combed pubic hair sample
 - c. pulled head hair sample
6. One paper bag for panties
7. Patient Information Form

Each of the above items should be contained in individual paper envelope with proper labels. All envelopes with items should then be placed inside a paper box.

The Crime Laboratory does not endorse a certain product. However, several companies have sexual assault evidence collection kits available. They usually have standard kits with the ability to make custom kits. Please choose a kit that contains at least the items listed above. Here is a list of some companies from whom the Crime Laboratory has received kits that are considered appropriate. Please note that this is not intended to be a complete list of companies who have sexual assault evidence collection kits available.

| | |
|--|--|
| Tri-Tech Inc. 4019 Executive Park Boulevard SE Southport, NC 28461 1-800-438-7884 www.tritechusa.com | Sirchie Finger Print Laboratories, Inc 100 Hunter Place Youngsville, NC 27596 1-800-536-7311 www.sirchie.com |
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| Lynn Peavy Company PO Box 14100 Lenexa, KS 66285-4100 1-800-255-6499 www.lynnpeavey.com | Serological Research Institute 3053 Research Drive Richmond , CA 94806 1-510- 223-7374 www.serological.com |
|---|--|

Please contact the Crime Laboratory if you have any questions.

2. Collection of Specimens from Sexual Assault Victims:

DO NOT STAPLE ANY BAGS - USE TAPE TO SEAL BAGS

In order to determine if there is any material from an assailant, it is necessary to determine what materials came from the victim by comparing the questioned evidentiary material with known specimens from the victim. A sexual assault kit is designed to assist you in doing the most thorough job possible in collecting evidentiary specimens which will be examined by the crime laboratory serving the agency investigating the case of the alleged sexual assault. Your cooperation is requested in collecting the following items:

NOTE: Sexual Assault kits have expiration dates. These dates usually refer to the EDTA in the blood collection tubes. If a sexual assault kit is expired, the blood collection tube can be replaced with a hospital provided purple top tube. Make note of the change on the sexual assault kit.

a. Blood Sample: To be examined for standard serology and DNA.

The known blood sample should be collected by the hospital personnel in a 5ml purple top tube (EDTA). The preservative contained in these tubes has been found to be very satisfactory for DNA typing.

In homicide cases where sexual assault is also suspected, approximately 2ml of blood from the body (without any preservative) should be placed on a piece of cotton cloth, air dried and placed in an envelope.

b. Saliva Sample: (Used as additional DNA reference sample)

Have victim place the circular or a small piece of filter paper in her/his mouth, saturate it with saliva, then have victim place the filter paper on envelope labeled "Saliva Sample" and let AIR DRY.



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Then place filter paper in the envelope, seal and complete the required information on the face of the envelope.

- c. Swabs: To be used for semen, possible blood group, enzymes, DNA, etc.

Do not aspirate any body fluid. Collect AT LEAST 4 EACH vaginal, anal and oral swabs. ONE OF EACH WILL BE USED BY THE CRIME LABORATORY AND THE OTHERS WILL BE SAVED FOR INDEPENDENT TESTING.

Swab the vaginal vault. These swabs can be used to make smears on the two slides while still moist. Then allow swabs to AIR DRY.

Do not dilute secretions on swabs by irrigation or aspiration. Do not collect vaginal fluid in plastic or glass vial.

NOTE: In the event of anal and oral assault where anal and oral swabs and smears are needed, but are not included in the kit, use additional swabs (hospital provided) or use another assault kit and label accordingly. **PUT ALL SWABS IN PAPER ENVELOPES. LABEL ACCORDINGLY. DO NOT PUT IN PLASTIC OR GLASS AIR-TIGHT CONTAINERS.**

- d. Smears: To be examined for spermatozoa.

Smears from previously mentioned swabs should be AIR DRIED. Do not stain or chemically fix these smears. Slides should be labeled with pencil on frosted end with source of smear (vaginal (v), oral (o), rectal (r), etc.), victim's name and medical personnel's initials. Place slides in slide mailer, seal and label.

NOTE: If the medical facility does its own testing, such as motile sperm and quantitative acid phosphatase, it should take the samples needed in addition to the sexual assault kit. Please do not use the kit materials for the hospital Laboratory.

- e. Pubic Hair Combing: May contain hairs from assailant.

Place a clean paper towel under pubic area of victim. Comb pubic area with enclosed comb. Transfer the comb and hairs into the envelope labeled "Pubic Combing" and seal. Complete the required



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information on the face of the envelope.

- f. Pulled Pubic Hairs: To be compared with hairs in combed samples.

Pull (DO NOT CUT) at least 25 pubic hairs from various areas (regions) of victim's pubic area by using thumb and forefinger. Place in envelope labeled "Pulled Pubic Hairs". If forceps are used for pulling hair, place the forceps in the envelope with the hairs. Seal and complete the required information on the face of the envelope.

- g. Pulled Head Hairs: To be compared with questioned hair samples.

Pull (DO NOT CUT) at least 20 of the victim's head hairs from each of the five regions of the head using thumb and forefinger for a total of 100 hairs. Place in envelope labeled "Pulled Head Hairs". If forceps are used for pulling hair, place the forceps in the envelope with the hairs. Seal and complete the required information on the face of the envelope.

- h. Custody Receipt: Give to law enforcement agency investigating this case.

- i. Victim's (patient's) Clothing: Victim's clothing should be collected and packaged in separate paper (hospital provided) bags. Properly label and seal each bag. Do not use plastic bags!

- j. Bite marks: Sexual assault cases can involve preserving and analyzing bite marks on the victim's body. This area can also yield valuable forensic DNA/serology evidence. The area should be swabbed gently without alteration of the bite marks with a distilled-water-soaked swab. The latter should be preserved in an envelope and included in the sexual assault kit.

NOTE: For the collection and submission of relevant evidence when date rape drugs are suspected, refer to the Toxicology section of this manual.

Seal sexual assault kit after evidence collection using enclosed evidence tape. Also use enclosed chain of possession label.

Evidence should be submitted to Laboratory AS SOON AS POSSIBLE!! or keep in the refrigerator until submitted. A completed Evidence Submittal form must accompany the evidence.



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Please submit known samples of the suspect's blood, saliva and hair. An evidence collection kit for this purpose is available from Tri-Tech Inc. and from other sources.

The following guidelines should be used for processing various types of serology and DNA evidence. The guidelines are according to the reference: Journal of Forensic Identification. 344/41(5), 1991. Lee et al.

C. Liquid Blood

1. Reference Liquid Blood Samples from a Person

- a. Liquid blood from a person should be collected by qualified medical personnel.
- b. One tube of blood, at least 5 ml, should be collected in vacuum collection tube with EDTA as an anticoagulant. If conventional serological analyses and/or drug or alcohol analyses are to be conducted, additional tubes of blood should be collected. Generally, the blood samples for conventional serological analyses should contain no preservative (red-top tube) and the tube for drug or alcohol testing should contain NaF (sodium fluoride) (grey-top tube).
- c. Each tube should be labeled with the date, time, subject's name, location, collector's name, case number, and exhibit number.
- d. Blood samples must be refrigerated (NOT FROZEN) and submitted to the forensic laboratory as soon as possible. If mailed, it should be sent by certified or registered mail or overnight delivery service. No ice is required and dry ice should never be used to cool the tube of blood.

2. Liquid Blood Specimens at the Crime Scene

- a. Liquid blood should be collected with a clean syringe or disposable pipette and transferred to a clean (preferable sterile) test tube. (DO NOT MOUTH PIPETTE ANY BODY FLUID REGARDLESS OF THE SOURCE!)
- b. A blood clot can be transferred to a clean test tube with a clean spatula.
- c. A clean cotton cloth, gauze, or cotton swabs can be used to soak up liquid blood or a blood clot if other equipment is not

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available. If this method is used and a significant delay occurs in getting the specimen to the laboratory, the blood should be air dried out of direct sunlight prior to submission to the laboratory.

- d. The specimens should be labeled with case number, item number, date, time, location, and evidence collector's name.
- e. Wet blood samples, if they are collected, must be preserved in a suitable anticoagulant (EDTA for DNA analysis) and kept in a refrigerator. These specimens must be submitted to the laboratory as soon as possible.

3. Liquid Blood Specimens in Snow or Water

- a. Blood samples found on snow or in water should be collected immediately to avoid further dilution.
- b. The largest possible quantity of these samples should be collected in a clean, suitable container, avoiding any contaminant as much as possible.
- c. The specimens should be labeled as previously indicated.
- d. These specimens should be frozen, if possible.
- e. The specimens should be submitted to the laboratory as soon as possible.

D. Wet Bloodstains

1. Garments with Wet Bloodstains

- a. Garments bearing wet bloodstains should be placed on a clean surface or hung up and allowed to air dry out of direct sunlight without a heat source, such as a hair dryer.
- b. A wet garment or garment with a wet bloodstain should never be collected in a sealed, airtight container or plastic bag. This practice causes the specimens to retain moisture, and promotes bacterial growth as well as sample deterioration.
- c. Once the garments and stains are dry, they should be packaged in a PAPER container with proper labeling.

2. Objects with Wet Bloodstains

- a. Small objects bearing wet bloodstains should be allowed to air dry, then collected as is.
- b. An effort should be made to preserve the integrity of any bloodstain pattern during packaging and transportation.

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- c. Large objects that cannot be removed from a crime scene may have wet bloodstains on them. The wet blood should be transferred onto a clean cloth.
- d. Bloodstained cotton cloth must be allowed to air dry before packaging in a paper container.
- e. Each object and container must be properly labeled.

E. Dried Bloodstains

1. Dried Bloodstains on Removable Items

- a. Dried bloodstains on weapons, garments, and other movable objects should be collected separately and by collecting the entire item.
- b. Each item should be placed in its own paper container, and each should be sealed and labeled properly.

2. Dried Bloodstains on Solid, Nonabsorbent Surfaces of Immovable Objects

- a. The bloodstain pattern should be photographed, documented, and sketched to the extent necessary.
- b. The stains can be scraped off the object onto a clean piece of paper.
- c. The paper is then folded into a "druggist fold" and placed in an envelope which is properly sealed.
- d. Each item must be labeled properly.

3. Dried Bloodstains on Large or Immovable Objects where Stains cannot be Cut, such as Walls, Concrete, etc.

- a. The bloodstain pattern should be documented, photographed, and sketched as necessary.
- b. The bloodstain may be scraped off the object directly, or eluted onto clean, cotton threads moistened with sterilized saline (or distilled water) by rubbing the threads on the stained area. The threads are then allowed to dry and are placed in a paper fold packet.
- c. Blood crust can be scraped directly onto a clean paper fold packet.
- d. The packet is then placed in an envelope which is sealed and properly labeled.
- e. A control must always be obtained by repeating the procedure on an adjacent but unstained area of the surface



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containing the bloodstain.

4. Dried Bloodstains on Carpet, Upholstery, Wood, or Other Objects that can be Cut
 - a. The stained area should be documented as previously described.
 - b. A portion of the item containing the bloodstain can be removed by cutting with a sharp instrument.
 - c. Each cutting should be packaged separately and labeled accordingly.
 - d. An unstained portion of the item should be collected and packaged as a control.

5. Dried Small Blood Spatters
 - a. It is best to collect the entire object without any attempt to remove the blood spatters. If that is not feasible, then try to remove the blood spatters.
 - b. Small blood spatters are often difficult to remove from their surfaces. It may be possible to collect them using the tape lift method.
 - c. Each piece of tape should be packaged and labeled properly.
 - d. Each piece of tape is placed into a larger plastic container. The ends of the tape lift should be taped in place to secure the lift.
 - e. The tape with the blood spatter portion should be suspended in the middle of the container.
 - f. The container should be sealed and labeled appropriately.

6. Bloodstains on Vehicles Involved in Hit and Run Cases
 - a. The exterior surface of the vehicle should be examined thoroughly for the presence of fabric impressions, imprint evidence, paint transfer, hairs, tissue, fibers, blood, and other trace evidence. All findings should be properly documented.
 - b. The under carriage of the vehicle should also be examined for the presence of tissue, hair, blood, fibers, and other evidentiary material.
 - c. All imprint and impression evidence should be photographed.
 - d. All trace evidence should be removed and properly

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- packaged.
- e. Blood stains are preferably removed from painted surfaces by chipping with a sharp object such as a chisel or screwdriver while at the same time removing paint samples which can be analyzed and compared to any found on the victim. Stains may also be scraped and placed in a clean paper fold if no paint sample is required.
- f. The envelopes should be labeled and sealed accordingly.
- g. This type of evidence should be recovered prior to any method of chemical latent fingerprint detection with the exception of lasers or alternate light sources.

F. Semen and Seminal Stains

1. Liquid Semen Evidence Found at a Scene

- a. The semen evidence should be documented by use of notes, photography, videotape, and sketching.
- b. A clean syringe or disposable pipette should be used to transfer liquid semen to a clean, sterile test tube. (DO NOT MOUTH PIPETTE ANY BODY FLUID REGARDLESS OF THE SOURCE!)
- c. The tube should be labeled with the case and item number, date, time, location, and name of the collector.
- d. The specimen should be kept refrigerated and submitted to the laboratory as soon as possible.
- e. Alternatively, liquid semen can be transferred onto clean cotton cloth or gauze by absorption. The cloth is then air dried, packaged, sealed, and labeled properly.

2. Seminal Stains on Movable Objects

- a. Seminal stains on panties, clothing, bed sheets, pillow, and other movable objects should be collected as is.
- b. If an article has a wet stain on it, the stain must be allowed to air dry thoroughly prior to collection of the article.
- c. Each item should be packaged separately in a clean paper container.
- d. Each item's packaging must be properly sealed and labeled.
- e. Packaged items should be refrigerated, if possible, and submitted to the laboratory as soon as possible.

3. Seminal Stains on Large Objects that can be Cut

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- a. Examples of large objects that can be cut, and that could have seminal stains on them are carpeting, bedding, and upholstery.
 - b. The evidence should be documented as previously described.
 - c. A clean scalpel or scissors should be used to cut the stained area from the rest of its surface.
 - d. Each cutting should be placed in a separate piece of clean paper.
 - e. A "druggist fold" of an appropriate paper container is made for each cutting, and the container should be sealed and labeled properly.
4. Seminal Stains of Immovable, Nonabsorbent Surfaces
- a. Examples of these surfaces are floors, counters and metal surfaces.
 - b. The seminal stain evidence should be documented as previously described.
 - c. A clean scalpel should be used to scrape the semen stain onto a clean paper, and the paper should be folded into a "druggist fold" container.
 - d. The scalpel must be cleaned or changed in between each use to avoid any contamination.
 - e. Each "druggist fold" should be placed into its own appropriate paper container.
 - f. Each container should be sealed and labeled properly.
5. Seminal Evidence from Sexual Assault Victims
- a. Sexual assault victims are always medically examined in hospitals or a physician's office.
 - b. Physical evidence should be collected using established procedures. Please see Section VI A above.
 - c. A standard rape kit must be used to collect vaginal, oral, and anal evidence as necessary.
 - d. Each item should be packaged, sealed, and labeled properly.
 - e. Evidence should be delivered to the laboratory as soon as possible. If there is a delay in this, the rape kit should be kept in the refrigerator (NOT FROZEN).
- G. Tissue, Organ, or Bone Specimen



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1. Fresh Tissue, Organ or Bone
 - a. Each item of evidence should be described in notes, and documented by photography, sketches, and videotape.
 - b. This type of evidence item can be picked up with a clean pair of forceps.
 - c. Each item should be placed in a clean container without any added fixatives.
 - d. Each container should be properly sealed and labeled, and stored in a freezer.
 - e. Evidence should be submitted to the laboratory as soon as possible.

2. Old Tissue, Organ, or Bone
 - a. Each item of evidence should be photographed and sketched before collection. The size, shape, and pattern of each item, as well as the spatial relationships to the rest of the scene should be properly documented.
 - b. Each item can be picked up with a clean-gloved hand. Evidence still connected should be collected together.
 - c. Care must be exercised not to contaminate any item with material from another item. Gloves should be changed for every different item.
 - d. Each item should be placed in a clean container; the container then should be sealed and properly labeled as previously described.
 - e. Evidence can be kept cool or stored at room temperature and submitted to the laboratory as soon as possible.

H. Urine, Saliva, and Other Body Fluids

1. Liquid Samples
 - a. Liquid urine or saliva should be transferred to a clean, sterilized container (plastic or glass bottle) as soon as possible.
 - b. Each container should be sealed and labeled properly.
 - c. Evidence should be stored frozen and submitted to the laboratory as soon as possible.

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2. Stains

- a. Urine stains, saliva stains, and other body fluid stains, can be collected as is, or removed from their substrata by scraping or cutting.
- b. Each stain sample should be placed in a clean, paper container. Scrapings or cuttings should be collected in a "druggist fold" made from clean paper. The "druggist fold" is then placed into a secondary paper container.
- c. Containers should be sealed and labeled properly.
- d. Samples should be transported to the laboratory as soon as possible.

I. Hair Evidence

1. Hair evidence can be picked up using a clean pair of forceps.
2. Each group of hair evidence items should be packaged separately, the package then sealed and labeled properly.
3. Care should be exercised during the collection to prevent damage to any hair root tissue that may be present.
4. Hairs mixed with blood, tissue, or other body fluids should be treated with care. Each item should be placed in a clean container that is then sealed and labeled properly.
5. Evidence should be stored in a refrigerator and submitted to the laboratory as soon as possible.
6. If hairs are mixed with wet body fluids and a delay of submission is unavoidable, the mixture must be dried thoroughly.

VIII Storage of DNA/Serology Samples

Samples intended for DNA/serology analysis should be stored in a manner that ensures the integrity of the biological evidence.

- A. Liquid reference blood samples should be stored in the refrigerator. This includes sexual assault kits.
- B. Liquid blood and body fluid evidence
 1. Liquid blood and body fluid specimens collected at the crime scene should be stored frozen a suitable container (i.e. 15

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- mL conical tube).
2. Liquid blood and body fluid specimens in snow or water should be stored frozen in a suitable container (i.e. 15 mL conical tube).
- C. Items with wet bloodstains or biological stains (i.e. semen)
1. Garments bearing wet bloodstains or biological stains should be placed on a clean surface or hung up and allowed to air dry out of direct sunlight without a heat source.
 2. A wet garment or garment with a wet bloodstain or biological stains should never be stored in a sealed, airtight container or plastic bag. This practice causes the specimens to retain moisture, and promotes bacterial growth as well as sample deterioration.
 3. Once the garments and stains are dry, they should be packaged in a paper container.
 4. An effort should be made to preserve the integrity of any stain pattern during packaging and storage
 5. Once items are dry and packaged in paper containers, they may be stored at room temperature
- D. Items with dried bloodstains or biological stains can be stored at room temperature in a paper container. Small items (i.e. swabs) can be placed in the refrigerator or freezer.
- E. Tissue, organ or bone
1. Fresh tissue, organ or bone specimens should be stored frozen a suitable container (i.e. 15 mL conical tube).
 2. Old tissue, organ, or bone specimens can be stored at room temperature in paper packaging. They should be placed in the freezer for long term storage.
- F. Hairs can be stored at room temperature in paper envelopes. They may be placed into a piece of weigh paper which is folded into a bindle before placing into the coin envelope. They should be placed in the freezer for long term storage.
- G. DNA samples should be stored in the refrigerator or freezer. DNA samples should be placed in the freezer for long term storage.
- H. Cuttings, scrapings, swabbings, etc. collected from evidence at the crime laboratory are preserved in paper containers (i.e., coin



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envelopes). They and can be stored at room temperature, but should be placed in the freezer for long term storage.

NOTE: Refrigerator storage is 2 to 8 C and freezer storage is -15 to -25 C.

IX. DNA/Serology Limitations

In recent years, there has been a steady increase in the DNA/serology caseload. The results of DNA analyses are now expected and given great weight in our judicial system in helping to determine the guilt or innocence of a defendant.

Because of our ever-increasing workload in our DNA/serology section we have become more selective in the items and examinations on which we spend time. We will strive to limit our examinations to only those that might have a high evidential value in any possible court proceedings. Our goal is to best use our limited time and staff in making the most meaningful examinations for the law enforcement community.

The following are some of the areas in which we will be more selective in our examinations:

- A. Cases for which DNA testing is requested will be prioritized and examined based on individual case circumstances. The specific number and types of items tested for each case may be limited and will be at the discretion of the crime laboratory.
- B. Sometimes the locations where some sexual assaults take place might contain possible semen, vaginal secretions, saliva, hairs, etc. from previous sexual encounters. In these cases only the rape kits and the victim's immediate clothing will be examined.
- C. No examinations, other than the presence of semen, will be conducted in cases of statutory rape or consensual sex, where the identity of the semen donor is not in question.
- D. No examinations will be performed when the suspect has pled guilty.
- E. Many law enforcement agencies use the Crime Laboratory for screening purposes and then submit the samples to a private laboratory for DNA testing, particularly in homicide cases. Unfortunately this takes up much time. This practice will have to be curtailed or extremely limited.

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- F. Additional evidentiary samples are often sent to the Crime Laboratory in homicide cases immediately prior to the trial date. Quite often the original evidence examination in these cases occurred a year or more prior to the subsequent request.. Because of other ongoing case commitments, most of the time we will be unable to analyze these samples in time for the trial.
- G. Microscopic hair comparisons are not performed. Other microscopic hair examinations will only be done in extreme case situations when no other biological evidence is available for DNA analysis and with prior consultation.
- H. References (known) samples are necessary for all cases where body fluid or tissue of a donor is suspected and the suspect is known. Without reference samples, only the presence or absence of body fluids will be reported, and time consuming DNA testing will not be performed on these items.
- I. Convenience items (a discarded cigarette butts, cups, etc.) collected from a suspect are not considered legally obtained reference samples and cannot be used for courtroom purposes. A legally collected reference DNA sample must be obtained and submitted for comparison and statistical analysis.
- J. Known samples (blood and/or saliva) must be submitted for any person(s) who have had sexual contact with the victim up to 72 hours prior to the assault.
- K. Known DNA samples may be required from any individual that may have had access to the crime scene or items being DNA tested for elimination purposes. This may include, but is not limited to victims, household members, law enforcement officer and first responders to the scene.
- L. Although an individual may have their DNA profile in the Convicted Offender database, it is still necessary to submit a legally obtained reference DNA sample from the individual for comparison to the specific case evidence.
- M. Only probative evidence profiles can be submitted to the Combined DNA Index System (CODIS) to search for matches. The Crime Laboratory cannot search an individual against the CODIS database to search for matches to unsolved crimes unless the individual is required by state statutes to provide a convicted offender sample.

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- N. The Crime Laboratory will not perform DNA testing on any item that has already resulted in a positive identification of an individual using some other method such as latent fingerprints.
- O. Items being tested for trace levels of DNA, such as gun swabs or other “touch” evidence will be consumed in testing in order to obtain optimal results. Please inform the Crime Laboratory upon submittal of the item if a portion of the evidence must be retained for independent testing.
- P. Analysis of blood from the victim's clothing in a homicide case, where the suspect has not been injured, provides no useful information and thus will not be performed.
- Q. The crime laboratory does not perform paternity testing (even in cases of rape when a child was conceived) or DNA testing on bone, teeth, or paraffin embedded tissue. If victim's known samples are not available for DNA analysis, the biological parents can be used to infer the victim's DNA profile (as in paternity). The Human DNA Identity Laboratory at the University of Nebraska Medical Center performs this type of testing for a fee. You can call (402) 559-7220 for information and instructions.

The Laboratory realizes that each case is accompanied by a unique set of circumstances and evidence. The previously-stated selective areas are general and will never totally define the Laboratory's analysis role in any one case. Therefore should any law enforcement agency feel that the analysis of a particular piece(s) of evidence is extremely critical for the ultimate success of the case it is strongly suggested that they contact either the DNA/serology section supervisor or the Laboratory Director for discussion and consultation. Only through communication can important and useful evidence be identified so that the full potential of the Laboratory can be focused on meaningful examinations of these items.

X. DNA Databasing

State law requires persons convicted of felony sexual and violent offenses, burglary and robbery to submit a DNA sample for the purpose of the DNA database. Please refer to the DNA Identification Information Act (Article 41) for completed information.

Nebraska administrative code, Title 272 - Nebraska State Patrol, Chapter 20 sets forth procedures for the collection of DNA Database samples.

- A. Only DNA Database Collection Kits provided by the crime laboratory are acceptable for the collection of these samples.

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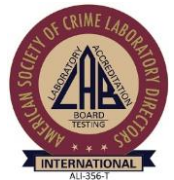
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- B. To ensure the viability of the blood or buccal sample, it must be delivered to the Crime Laboratory within five working days of its collection.
- C. The blood samples should be refrigerated until they are transported to the laboratory.
- D. Because the laboratory is not staffed on week-ends or on state holidays, careful planning relative to the dates blood samples are collected in order that they can be delivered to the laboratory within five working days of collection will be necessary.
- E. If the blood samples are not hand delivered to the laboratory, it is recommended that an overnight mail service be used.

The crime laboratory tests the DNA database samples for 16 STR loci, including the 13 core CODIS loci and the results are entered in CODIS. The crime laboratory is also responsible for storage of the DNA database samples for an indefinite period of time.