CRIMESCENE

Newsletter of the Northwest Association of Forensic Scientists

FALL 2003 VOLUME 29 ISSUE 4

INSIDE:

PRESIDENT'S MESSAGE BILL **MARSHALL**

My first job was selling peanuts at a baseball stadium in Boise when I was 14. I cleared about a penny and a half per bag. Over the years, and especially going to school, I have had several other jobs; washing dishes, waxing floors, driving trucks, making pizza and pouring beer. I was even an encyclopedia salesman for a couple of weeks. Hey, it was a job.

I just compared a recent membership list of the Northwest Association with one from 1974. Out of fifty-nine members on the earlier list, fourteen are still members and nine of these are still active in the field, another half dozen are still active but no longer members, at least 25 are retired, and a few have unfortunately passed on. There are little over a dozen names that I don't know what they have done. The one point I want to make is that the majority of these people have made a career of forensic science.

. . . we are still involved in answering the basic questions - who, what, when, where. (*Why* is one I'm not sure anyone ever knows) . . .

The newer list contains over three times the number of names as the first one and many of these are people who have also spent many years in the field of forensic science. It also has some newer names of those just starting out. I suppose this message is for you.

The field has changed. FTIR replaced grating IRs, capillary GC has replaced packed columns, mass spectroscopy is now standard. Obviously, DNA has revolutionized what can be done with biological evidence and databases (spectral, AFIS, CODIS, etc.) have changed how we can use the information once it is generated by the newer instruments/ techniques. But we are still involved in answering the basic questions – who, what, when, where. (*Why* is one I'm not sure anyone ever knows) Sometimes, we get a chance to solve puzzles.

For the new people coming in, I hope that if someone compares the membership list in the future that, like those on the 1974 membership list, forensic science has been a career and not just a job.

- BILL MARSHALL

CONTENTS

CRIMESCENE
FALL 2003

On The Cover...

A criminalist uses an alternate light source to enhance the imaging of any potential biological stains. *For demonstration purposes only*. **Do Not Try This At Home!**

CRIMESCENE is the official publication of the Northwest Association of Forensic Scientists. It is published four times a year in the months of February, May, August, and November. The Newsletter welcomes submissions from its membership, such as: technical tips, case studies, literature compilations, workshop or training notifications, reference citations, commentary, historical accounts, and other topics of interest to the membership. While not currently required, please submit material for publication in Microsoft Word for Windows format as an e-mail attachment or on a 3.5" floppy disk. For more information regarding the Newsletter or to make a submission please contact editorial secretary, Joshua S. Spatola at the California Department of Justice, Central Valley Crime Laboratory, 1306 Hughes Lane, Ripon, CA, 95366 (phone: 209.599.1418 / fax: 209.599.1240 / email: josh.spatola@doj.ca.gov).

Sample Handling Considerations for Biological Evidence and DNA Extracts

Theresa F. Spear
California Department of Justice
California Criminalistics Institute

Introduction

This article will make recommendations for handling biological evidence from sample collection to sample storage. Depending upon the type of biological evidence, it may be very difficult to detect certain types of samples. Saliva and semen stains are often not visible. Even bloodstains may be difficult to find if they are on a dark substrate. Although specialized, forensic light sources are sometimes helpful in visualizing these stains, they cannot always be relied upon to find all biological stains. It is especially difficult to detect saliva or semen on some dark colored fabric with any of the widely used forensic light sources. Thus, a stereomicroscope or chemical mapping techniques may be required to improve the chances of finding all biological evidence. After a sample has been detected, steps must be taken to insure that the sample integrity is maintained and the chances of contamination are minimized.

Sample Collection in the Field

Once a sample has been located and a decision has been made to take the sample, issues arise as to how to collect, dry and package it for transportation to the laboratory. Considerations relevant at this stage include the ability to obtain as much sample as possible, to minimize degradation and finally to insure that samples are not inadvertently contaminated with other biological samples.

The standard recommendation for collecting biological evidence is *not* to remove the stain from an object but rather to collect the object with the stain. The advantages of this strategy are that the entire stain is obtained, it is not necessary to collect an "unstained control" sample and there are no further manipulations required that might negatively impact the sample. If the stain is on a smooth, non-porous surface (i.e. it can be easily "flaked" off), it will be necessary to protect the stain from contact with other objects. Depending upon the nature of the evidence, a stain can be protected by immobilizing the evidence item in a cardboard box (e.g. with pieces of wire) or by taping a piece of paper over the stain (if this will not destroy other evidence, such as fingerprints). *Provided that the stain can be adequately protected, this is the optimum collection procedure.*

Given that some stains are found on immovable objects, it is not always possible to collect the object with the stain. Some samples will need to be collected in the field. If the entire object cannot be collected then *the next best way to collect biological evidence is to remove the stain by cutting it out (e.g. from a piece of carpet).* Remember to use clean scissors and to cut out an "unstained" control. Scissors or tweezers can be cleaned by rinsing with clean water and then drying with tissue. Repeat this cleaning process twice prior to each sampling.
It is not necessary to clean tools with bleach. Improperly used, bleach could destroy biological evidence.

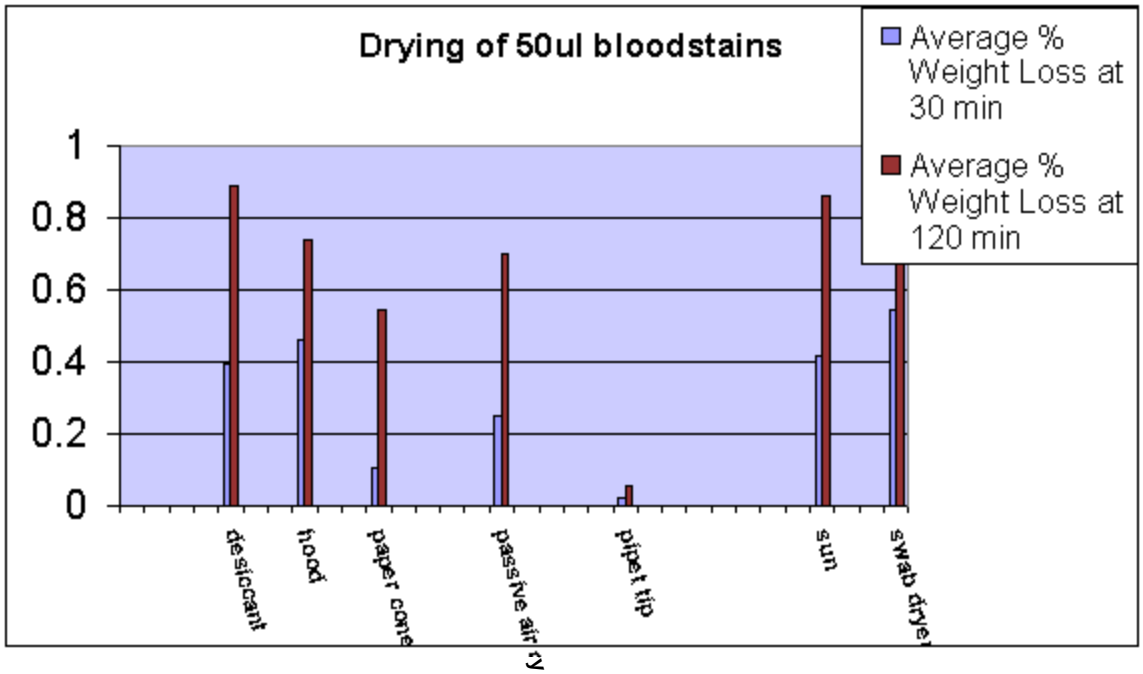
There will be occasions when it is not possible to collect a stain by cutting it from an object (e.g. stain is on a concrete floor). The two methods traditionally used to collect these stains are: (1) to use a dampened cotton swab, thread or piece of gauze to collect the stain or (2) to use a clean implement (e.g. razor blade) to scrape the stain into a clean paper bindle. *With these two methods, it will typically be necessary to take an "unstained" control sample.* Scraping is practical only when the *stain is in the form of a crust* and it can be lifted from a smooth, non-porous surface. The most significant problem encountered while scraping stains is that samples tend to "powder" when scraped and it may be difficult to control the retrieval of the entire sample. The "powdered" stain, which is not retrieved, may contaminate adjacent stains. Given these considerations, most samples will not lend themselves to the scraping technique and will need to be collected by swabbing.

The best method for swabbing a stain is to use a minimum amount of distilled water to dampen an appropriate, clean substrate (e.g. cotton swab or cotton gauze) and then absorb the stain onto the slightly dampened substrate. An unstained control is taken in the same manner as the stain and is taken from an unstained area as close as possible to the biological evidence sample. It may be useful to test the "unstained" area with an appropriate presumptive test to see if it contains a biological sample. In an effort to keep the stain as concentrated as possible, the size of substrate used to absorb the stain is important. Ideally, the smaller the stain, the smaller the substrate used to collect that stain. Thus one might choose to use a small piece of cotton gauze rather than a swab to collect a small blood spatter (1 or 2 mm in

diameter). In order to protect the integrity of a small sample, the dampened gauze should only be handled with clean tweezers. If a stain is not very small, *swabs are probably easiest to use* since they do not require the use of tweezers or any other intermediate object.

Cotton swabs have traditionally been used to collect biological samples. They are readily available in sterile packages and they are very absorbent. Do *not* use "calcium alginate" swabs for sample collection since these swabs interfere with many of the DNA extraction procedures. Cotton gauze, which does not have any additive (e.g. fabric sizing), is also an acceptable substrate.

Once a stain is collected, the next consideration is drying the swabbed sample. Drying biological samples is critical to preserving these samples because water is necessary in most of the biochemical reactions that result in degradation. Try to dry this sample as fast as possible to minimize degradation of the sample. Packaging a wet swab into a plastic tube with holes or a pipet tip will considerably slow the drying process. Samples placed into devices such as these frequently take more than 24 hours to dry. A drying process that extends over many hours may jeopardize a marginal or small sample. Saliva samples placed onto swabs and then into swab containers with small holes show more degradation and yield less DNA than saliva samples placed on swabs and then into paper envelopes. Ideally, samples should be left open to the atmosphere and allowed to air dry before they are packaged in paper envelopes. In all but very humid climates, they should be dry enough to package into paper envelopes in two hours. The use of desiccants should be considered in humid environments. The following graph illustrates drying rates of 50ul bloodstains using a variety of different techniques.



One last consideration is collecting a sample with a minimum amount of surrounding material. This should be done to minimize the contact of the biological sample with potentially deleterious material. For example, it has been found that cigarette ashes tend to inhibit the PCR reaction resulting in a failed DNA test. Therefore, when collecting cigarette butt(s) from ashtrays, it is a good idea to only collect the cigarette butt and not the accompanying ashes. This also applies to collecting biological samples from soil. To the extent that it is possible to separate the sample from the soil matrix, there will be a greater chance of a successful DNA test.

Packaging Materials

After the sample has been collected, it should be placed into paper packaging that will protect the item against loss and contamination. This packaging can also be used to record the chain of custody and to describe the evidence item. The most important feature about this aspect of evidence packaging is that paper is a porous material and will allow air exchange with the atmosphere. This property will allow samples the chance to completely dry, even when they are not entirely dry at the time of initial packaging. *Paper envelopes or bags are the easiest way to accomplish this goal*. If

the evidence is a heavily bloodstained garment, it should be allowed to dry. A piece of clean paper can then be placed over the top of the item and the item can be folded so that the paper helps to prevent direct contact between separate stains. Depending upon the stained item, more than one sheet of paper may be required. Ideally, each evidence item should be packaged in a separate, paper container. Choose a paper container large enough to easily accommodate the evidence item so that it is not necessary to "stuff" the object in the package. As long as paper bags and envelopes are not reused, they will not be a source of contamination. Once the item is ready for storage, it needs to be sealed according to agency policy.

Sample Handling Considerations in the Laboratory

Before Analysis

If it was not possible to completely air-dry samples in the field, they can be dried upon return to the laboratory. Make sure evidence is clearly labeled before it is placed into a laboratory hood or swab dryer to finish the drying process. If the item is large, try to dry one evidence item at a time in the hood. The dried evidence can then be returned to paper envelopes or bags. The next decision that will likely need to be made is: How should the evidence be stored until it is analyzed? Some types of evidence may require that the biological sample be removed from the surrounding material before storage. This would be done to minimize contact of the biological sample with potentially harmful material. For example, leather substrates may contain compounds that tend to destroy biological samples and inhibit PCR reactions. For this reason, it may be wise to remove a bloodstain from leather by swabbing the stain as soon as possible rather than cutting out the stain for analysis. Certain plant materials and soil are examples of other materials that might need to be separated from biological samples as soon as possible.

The next issue that needs to be addressed is: Where will the evidence be placed until it can be analyzed? This type of storage can usually be regarded as "short-term" storage. Three possibilities exist: (1) room temperature, (2) refrigeration or (3) frozen. If the evidence is on a smooth, non-porous material (e.g. plastic/glass/metal) or requires fingerprint processing, it should be stored at room temperature. Room temperature storage avoids the problem of condensation that is typically seen with large temperature changes that occur when the evidence item is removed from the freezer.

In general, DNA in biological samples is very stable and can tolerate room temperature storage conditions very well. However, over an extended period of time, frozen storage is generally considered to be the optimum storage condition for biological samples. This means that evidence, such as bloodstained clothing, should be held frozen. Liquid blood samples, stored in glass or plastic containers, should be held in the refrigerator since these samples can break in the freezer. Liquid blood samples containing preservatives (e.g. EDTA) are stable for a very long period of time in the refrigerator.

During Analysis

Once analysis begins, an analyst needs to decide where to sample and how much sample to take. Evidence stains are not always uniform and it may be necessary to sample in more than one area to obtain all relevant information. Since each stain is unique, it is always a judgment call on how much sample to consume in order to optimize the chance of getting a clear result. Another simultaneous consideration is attempting to leave a sufficient amount of sample so that a second test can be conducted by the defense. If there is not enough sample to achieve both goals, it may be necessary to consult with the local district attorney's office on how to proceed.

After a sample is selected and placed into a tube for analysis, it is important that consideration be given to maintaining the quality and quantity of the evidence. Once water is added back to a sample, bacteria can begin to degrade DNA. Thus, if it is necessary to soak a sample in phosphate buffer saline to remove the cellular material, this step should be done in the refrigerator to minimize degradation by any bacteria. Certain buffers (e.g. those containing EDTA) are designed to inhibit the activity of nucleases that can breakdown DNA and these can preserve DNA in liquid form better than other liquid extractants. Another variable to keep in mind is that the efficiency of DNA extraction techniques, or how much DNA is obtained, vary with the method and the analyst. It is important to spend time to optimize each extraction method so that reasonable yields of DNA can be routinely obtained.

With respect to issues of contamination, the most at-risk samples are small stains. These samples may consist of old and degraded samples or very small volume stains. With these stains, it may be advisable to take special precautions to insure the integrity of these samples. Contamination can be minimized by preventing contact of evidence samples with

any other form of biological sample. Sample tubes and reagents should be autoclaved before use. Any implement or glove that contacts the sample should be clean. It may also be important not to talk over evidence samples. Finally, when working with marginal samples it may be advisable to work in a biosafety hood.

After Analysis

Frequently, DNA extracts or amplified products are generated during the analysis of evidence. The labeling information on these samples should be traceable to the original evidence item. This can be accomplished by assigning "sub" item numbers that correspond to the original evidence item but also reflect the exact stain.

It may not always be possible to preserve all of the extracted DNA remaining after a sample has been typed. Extracted DNA can be lost from tubes held frozen. Some of this DNA may bind to the sides of the storage tubes or be destroyed by nucleases. Recommendations that help preserve extracted DNA include using an appropriate TE buffer to store extracted DNA and keeping the extracted DNA as concentrated as possible. Dilute DNA solutions (e.g. 0 .1ng/ul), even made in the appropriate TE buffer, do not appear to be very stable over time. Finally, consider using storage tubes that seal well so that samples do not dry to a residue in the freezer.

If at all possible, keep biological evidence in the freezer for long-term storage. Although it has been possible to successfully analyze biological evidence that has been stored at room temperature for more than 25 years, the resulting STR profiles show degradation effects. In addition, relatively small amounts of DNA are usually obtained from samples held for long periods of time at room temperature indicating that DNA loss has occurred over time. Although freezing biological samples does not prevent all DNA loss or degradation, it slows DNA degradation down much more than room temperature storage. There is some anecdotal information to indicate that dry biological samples might benefit from being stored in sealed plastic bags in the freezer. The final reason to consider frozen storage is that small or marginal samples that are maintained at room temperature could degrade so that little or no typing information can be obtained.

SUMMARY OF RECOMMENDATIONS FOR THE COLLECTION OF BIOLOGICAL EVIDENCE FOR DNA ANALYSIS

Goals of Biological Evidence Collection

1. Collect as much sample as possible from a single source.
   Keep biological evidence stain concentrated/do not spread a small stain over a large substrate.
2. Insure that the sample is not inadvertently mixed with other biological samples.
   Wear gloves. Make sure that nothing contacts the evidence samples which contains another biological sample. Do not talk over biological evidence.
3. Handle the sample in a manner, which minimizes deterioration of the sample.
   Air-dry the sample as fast as possible.

Recommended Methods for Collection of Biological Evidence

Listed in Order of Desirability

1. When feasible, take object with stain. Do *not* remove stain. If stain can be easily dislodged (e.g. stain is on non-porous surface), protect it from contact with another object. One way this can be done is by immobilizing the evidence item in a cardboard container.

2. If evidence stains are found on objects that can be cut (e.g. a rug), the evidence stain can be removed by cutting it out with a pair of clean scissors. Remember to also take an "unstained" control cutting (e.g. the substrate without the stain) from the object.

3. If object cannot be moved, use a **slightly dampened** substrate (cotton swab or piece of plain, cotton cloth or gauze) to collect stain. Remember to collect an unstained control by swabbing an unstained area of the evidence object as very few stains can be safely collected by scraping them.

- The size of the stain should influence the size of a substrate used to collect the stain. Thus, use a small part of a swab or a small piece of cotton cloth or piece of gauze to collect a small stain. Do not smear a small stain over a large surface

- Use a minimum amount of distilled water to dampen the swab or cloth/gauze substrate.
- To keep the stain concentrated, collect the stain on the smallest area of the swab or cotton cloth.
- Because of the attached wooden applicator stick, cotton swabs are easiest to use (tweezers are not needed).
- Use a pair of clean tweezers to manipulate cotton cloth/gauze; do not handle stain with bare hands.
- Allow all samples to air-dry as quickly as possible. The longer an evidence stain is kept wet, the more it can deteriorate.

Other Considerations

1. To avoid contamination, do not allow the biological evidence stain to come into contact with any other biological sample.

- Each individual stain should be collected separately. Do not collect or package two separate stains together in the same container.
- Do not allow evidence samples to come into contact with any surface, which contains residue from another biological sample (e.g. dirty tweezers, bloodstained glove, contaminated work surface).
- Use tweezers with smooth, easy-to-clean working surfaces.
- Tools (e.g. tweezers, scissors) can be cleaned by thoroughly rinsing with a stream of distilled water and drying thoroughly with paper tissue. Repeat this process *twice* before using tool to manipulate another sample.
- Do not talk or cough over biological evidence.
- Working surfaces (that could contact other samples) should be cleaned with a freshly made 10% solution of bleach. Insure that any bleach residue is thoroughly removed by wiping it down with another clean paper towel.

2. Small biological evidence (e.g. 2-mm size bloodstain or hair) is most susceptible to contamination.

- Put on a new pair of gloves before handling these small stains. Consider wearing a mask.
- Work on clean surfaces. If possible, work in a biosafety hood.

3. Package all biological evidence in **paper** bags or envelopes. Do **not** use plastic.

- Allow stains to air-dry before placing in paper bag or envelope.
- Work on one evidence item at a time.
- Insure that paper containers used to package biological evidence are sufficiently large to allow some air circulation around evidence item.
- "Unstained controls" must be packaged separately from the evidence stain.
- Package different evidence items in separate paper containers.
- A piece of clean paper can be placed over a garment with a number of bloodstains. The garment can be folded so that the paper helps to prevent contact between different stains

Storage of Biological Evidence

1. If evidence sample is at risk, it might be desirable to remove it from the evidence item before storage
    a. A crust of blood on a smooth, non-porous item may be easily dislodged and lost
    b. A biological sample on a potentially harmful substrate like leather should be removed
2. In general, the DNA in biological samples is relatively stable at room temperature. Short-term, some evidence should be held at room temperature:
    a. Any evidence item that requires both fingerprint processing and DNA typing should be held at room temperature conditions until analysis is completed.
    b. Biological evidence on smooth, non-porous surfaces where condensation from large temperature changes might be a problem
3. Liquid blood samples (with preservatives) are stable in the refrigerator for extended time periods. For long-term storage, frequently bloodstains are made from these liquid samples and the stains are held frozen.
4. Over any extended period of time, biological evidence is best preserved in the freezers. Freezers that do not routinely go through a defrost cycle are preferable to freezers that are "frost-free". Clothing with biological stains should be held frozen.
5. DNA extracts should be kept as concentrated as possible, stored in TE buffer and held frozen in airtight tubes.

References

Kobilinsky, L. Recovery and Stability of DNA in Samples of Forensic Science Significance. Forensic Sci. Rev 4:67, 1992.

Gialamas, D. and Stockwell, D., Forensic Biology Sample Collection and Handling Techniques: A Look at Methods Utilized by California Crime Labs, CAC News, Summer 1995.

Fall 2003 Conference MEETING **MINUTES**

NWAFS Business Meeting * October 17, 2003 * Portland, Oregon

Meeting was called to order by President Tom Barnes at approximately 9am.
A quorum of the membership was established.
A list of members in attendance was kept by membership secretary Dan Alessio.

The minutes from the Reno Spring meeting were presented as posted in the newsletter. Motion for approval (W. Marshall/S. Jacobson) Passed

Rhonda Banks, Secretary/Treasurer's Report

- Reno Spring 2003 meeting gain: $2529.27
- $2,680.24 was returned to NWAFS main account from the Spokane Spring 2002 meeting
- Taxes are up to date.
- Financial report:

NWAFS Net Worth Report
for the period of 04/07/03 through 10/07/03

	04/07/03	10/07/03
Cash and Bank Accounts		
Checking	2,739.65	2,270.36
Savings	2,546.64	7,973.44
Investment accounts		
Dreyfus Liquid Assets*	22,047.30	22,126.47
Total Assets	27,333.59	32,370.27
Change in net worth 04/07/03 through 10/07/03		5,036.68

NWAFS Income & Expense Report
for the period of 04/07/03 through 10/07/03

Expenses	
Newsletter expenses	(1,548.56)
Accountant	(950.00)
Reno spring 2003 meeting expenses	(210.73)
Missoula spring 2004 meeting expenses	(2,000.00)
Total Expenses	(4,709.29)
Income	
Dividend income (Dreyfus Liquid Assets)*	79.17
Interest income (savings)	16.56
Membership dues	4,230.00
Reno spring 2003 proceeds	2,740.00
Return seed $ Spokane spring 2002	2,680.24
Total income	9,745.97
Total income & expenses 04/07/03 through 10/07/03	5,036.68

*Reflects account balance from most recent statement period ending 6/30/03.

- Motion for approval of treasurer's report (S. Jacobson/W. Marshall) Passed

Dan Alessio, Membership Secretary's Report

- The following names are submitted for membership and advancement:

New Memberships

Associate:

Sam Blittman, Arson Laboratory Consultants, Antioch, CA
Chad Day, Intern, Montana State Crime Lab, Missoula, MT
Eric McCollum, BATF, Cheyenne WY
Robinette Struckel, King County Medical Examiners Office, Seattle, WA
Kristin Walti, Criminalist, South Dakota State Forensic Lab, Pierre, SD

Regular:

Dylan Argyle, DPS-BFS, Ogden, UT
Terry Coons, OSP, Portland, OR
Samantha Evans, CA DOJ, Santa Rosa, CA
Matthew Gamette, WSP, Spokane, WA
Kris Gates, OSP, Portland, OR
Jennifer Gauthier, WSP, Seattle WA
Rebecca Gaxiola, CA DOJ, Redding, CA
Christopher Hamburg, WSP, Tacoma, WA
Brianne Huseby, WSP, Seattle, WA
Lori Hutchinson, Montana Forensic Lab, Missoula, MT
Lori Knops, WSP, Marysville, WA
Shannon Larson, ISP, Pocatello, ID
Dr. John Lundy, OSP Medical Examiners Office, Portland, OR
Rocklan McDowell, ISP, Pocatello, ID
Barry Miller, CA DOJ, Redding, CA
Corinna Owsley, ISP, Meridian, ID
Dr. Eric Person, WSP, Marysville, WA
Dale Purcell, OSP, Portland, OR
Glen Spencer, OSP, Springfield, OR
Katherine Woodard, WSP, Seattle, WA

Members Elevated

Prov. Associate to Associate:

Michael Frost, Weber State University, Layton, UT
Bruce Thomas, American River College, McClellan, CA

Prov. Regular to Regular:

Jeff Borngasser, OSP Springfield
Sara Day, CA DOJ, Redding, CA
Adam Dutra, San Diego PD Crime Lab, San Diego, CA
Elizabeth Selya, CA DOJ, Santa Rosa, CA

- No objections were made to any of the proposed advancements/membership applications.
- Motion to accept the list as a whole (B. Carpenter/K.D. Sommer) Passed
- There have been no nominations for the two positions up for election, member-at-large and technical secretary
- Nomination from the floor: Annalivia Bishop nominates Bill Schneck for member-at large , Bill accepts this nomination
- Brad Putnam gave a brief description of the technical secretary position
- Nomination from the floor: Beth Carpenter nominates Nici Wanek for technical secretary, Nici accepts
- Bill Marshall and Stuart Jacobson are the nominees for President and Vice-president respectively
- President Tom Barnes proposes a vote for the positions of President, Vice-President, Member-at-large, and Technical Secretary as a block.
- A motion is made to elect the above named to office (F. Boshears/T. McAdam) Passed

Josh Spatola, Editorial Secretary's Report (presented by Tom Barnes)

- Josh is looking for articles/information for the newsletter
- Tom Barnes presented information regarding the award of a free registration for outstanding contribution to the newsletter

Nothing to report:

Bill Marshall, Vice President
Brad Putnam, Editorial Secretary
Stuart Jacobson, Member-at-Large

Upcoming Meetings:

Spring 2004 in beautiful Missoula, MT
Hosted by the Montana State Crime Lab Annalivia Bishop as chairperson
Hotel and workshop information

Fall 2004 in Ashland, OR
Hosted by the National Fish and Wildlife Laboratory with Ed Espinoza as chairperson

There are no proposed sites for meetings beyond Fall 2004. Analivia has approached the IAI about a joint meeting. They are not interested in 2005, but possibly 2006. She will act as a contact for them. Suggestions were made to approach SWAFS about a joint meeting and the RCMP about hosting a meeting.

Tom Barnes, President's Report

- Proposed changes to the ethics committee policy were discussed. A motion was made to accept the changes as published in the newsletter (M. Jorgenson/ S. Jacobson) Passed
- Proposed changes to the quorum rule discussed. By show of hands, the membership present does not support this change. No motion to vote was put forth. It was noted that there is a constitutional provision for a mail ballot if a quorum is not established.
- Proposed new wording regarding the status of life members was discussed. A motion was made (S. Jacobson/V. Wanek) and passed to change the wording to the following:

LIFE MEMBER

(1) Life membership may be bestowed upon a Regular member in good standing upon nomination by three (3) Regular members in writing to the Membership Secretary, the recommendation of the Membership committee, and a simple majority vote of the Regular and Life members present at the Fall Business meeting. The criteria for evaluating the candidate's qualifications to be bestowed Life membership may include, but are not limited to:
   a) Charter membership; or
   b) service as a past officer of the Association; or
   c) on-going, active participation on the working committees of the Association; and
   d) continued service to the Association.

(2) A Life Member shall maintain the privileges of a Regular Member, including:
   a) voting rights on issues placed before the Membership for action;
   b) the right to hold an elected office in the Association; and
   c) shall receive the Association's newsletter at no cost.

(3) A Life Member shall be exempt from paying annual Association dues.

(4) A Life Member shall be entitled to one meeting registration per annum.

- Discussion was presented regarding the nomination of Arnold Melnikoff as life member and the Board of Directors decision to not move this nomination forward to a vote at this time. Arnie gave an explanation of his current status with Washington State Patrol. A motion was made to override to BOD decision and move the nomination forward to a vote. (M. Noedel/J. Jagmin) This motion failed. The nomination was not put to vote.

Meeting adjourned approximately 10:05am

Fall 2003 Conference OPEN**LETTER**

October 13-17, 2003 * Portland, Oregon

Dear NWAFS,

I'd like to thank and express my appreciation to Rhonda Banks, Devon Sommer, Tom Barnes, Deborah Newville, and all the others that helped in putting on the Fall 2003 NWAFS meeting. It was a wonderful week in Portland; the workshop I attended (Bloodstain Pattern Analysis) was top-notch and I heard similar comments from those who attended other workshops. The after-hours activities were a lot of fun and offered opportunities to exchange ideas in an informal setting with other scientists. The technical session was also enjoyable with good quality presentations on a range of topics, though there is one thing that I think we can work on as an organization with regards to our technical sessions.

On both Thursday and especially Friday, I noticed some of the presentations ran a little long. Typically it was up to 5 minutes long for the 20-minute presentations, however, one presentation ran 20 minutes long (doubling the amount of time allocated to it). This frustrated me for a number of reasons.

First, following speakers may feel obligated to "make up time" by speeding up their own presentations. In fact, a later speaker actually made comments to this effect during his talk. Second, it may create problems for the meeting coordinator who relies on the schedule for coordinating lunch breaks and catering. And third, it is unfair to those presenters who rehearsed their presentations and pared them down to be sure they stayed within the 20-minute time allotment.

I'd like to suggest that as an organization, we purchase a light or some other device that could be placed in the back of the room and light up when a certain amount of time is left. This would be a silent but noticeable indicator to the presenter that time is running out and to wrap things up. It might also be less awkward than the session mediator having to interrupt a speaker to tell him/her that time is short. Also, maybe a reminder to presenters when putting out the call for papers would be appropriate.

Sincerely,

Jennifer Riedel
Forensic Scientist
Oregon State Police

Fall 2003 Conference ABSTRACTS

Technical Session * October 13-17, 2003 * Portland, Oregon

An Interesting Officer Involved Shooting Case

Dan Alessio, Oregon State Police Crime Lab – Portland, OR

On August 13, 2003 the Oregon State Police Portland Crime Lab was requested to respond to the scene of an officer involved shooting at a busy rest area along the I-5 freeway south of Portland. What seemed to be the routine documentation of a scene soon turned into a bit of a puzzle. Some lessons can be learned about information exchange at a scene and an officer under duress during a shooting situation.

Detection of Chemical Residues Using Near-Infrared Dyes and Latent Blood Using Leuco Dyes

Gabor Patonay, Department of Chemistry, Georgia State University - Atlanta, GA

Near-Infrared (NIR) chromophores have been used successfully in several applications. The advantage of NIR fluorescence spectroscopy is especially obvious in applications where the inherently low background fluorescence of the long wavelength spectral region reduces the need for sample preparation, but other areas of analytical chemistry also benefit.

The basic operation of NIR dye probes, light sources, and detectors for forensic applications will be discussed. A new NIR dye has been developed that can be used to detect pepper spray residues using NIR fluorescence detection. An area of interest such as clothing is sprayed with the dye solution followed by illumination with a NIR light source, e.g. a NIR filtered alternative light source (Crime Scope). Pepper spray residues will exhibit NIR fluorescence spots that can be observed using an appropriately filtered pocket scope, digital camera, CCD camera, or other NIR detection systems. A similar application will be discussed for gun shot residue detection as well. The main advantage of NIR detection is that the color of the background has very little influence on detection and that there are very few materials that would interfere by exhibiting NIR fluorescence. The use of pocket scopes permits sensitive and convenient detection. Once the residues are located, samples can be extracted for further analyses. This NIR detection method does not interfere with LC-MS, GC-MS, or similar methods. In addition, several other promising new NIR chromophores designed to detect latent prints, blood, metal ions, and other trace chemicals will be discussed.

Leuco dyes typically exhibit zero or very little fluorescence when applied to surfaces. If latent blood residues are present leuco dyes are oxidized and become fluorescent dyes. Fluorescin has been used frequently for this purpose. The chemistry used in the past has several disadvantages for example, rapidly diminishing contrast after application

This presentation will discuss a new chemistry for fluorescin latent blood detection. This new chemistry results in excellent contrast for several hours and in a significant number of cases the contrast remains for days or weeks. The FBI's Evidence Response Team and DNA I unit worked together with Georgia State University to validate this new fluorescin chemistry for use in the field. This new chemistry requires no preparation at the crime scene. Several examples will be presented to illustrate how time and conditions affect contrast and false positive results. The performance of the new fluorescin chemistry on different substrates will be discussed. While this new chemistry is fully compatible with presently used light sources, new truly portable blue LED light source can be used as well.

In addition to the visualization of latent bloodstains, this kit is also suitable to observe latent fingerprints on certain surfaces. This feature that is due to the delayed self-oxidation of the leuco dye, does not interfere with latent bloodstain detection, because it appears at a later time. In addition to fluorescein, rhodamines can be prepared in their leuco form and are oxidized by latent bloodstains. This new rhodamine chemistry requires no preparation at the crime scene similar to fluorescein. The longer wavelength absorption and fluorescence properties of rhodamine can be very useful on certain substrates of high interference.

Correlation of Growth Phase with DNA Results Obtained from Hair Roots

Rhonda Banks, Oregon State Police Crime Lab – Portland, OR

With the advent of more sensitive DNA analysis capabilities, forensic hair examiners are more and more frequently asked to perform hair comparisons as a screening tool for DNA analysis rather than for the value of the comparison itself. Some forensic laboratories no longer perform traditional microscopic hair comparisons. Frequently, however, the number of hairs in a given case is large and many of the hair roots do not yield a DNA result. This study correlates the growth phase of the hair root and the DNA results obtained in an effort to determine if it is possible at the screening level to target hairs that will yield useable DNA results.

The Death of Barbara Hardegger: A Homicide Case Study

Jennifer Riedel, Oregon State Police Crime Lab – Springfield, OR

In September 2001, the body of Barbara Hardegger was found buried in a rural area south of Eugene, Oregon. Sheriff's deputies were lead to the location by her husband, Gary Hardegger, who placed a 911 call stating that he "had done something terrible to his wife." As the investigation continued, one of the Hardegger's sons was also implicated in the crime and it was determined she had been buried alive.

The laboratory assisted with the crime scene investigation including excavation of the body and crime scene reconstruction. In-lab analysis of the physical evidence was also performed and a number of significant physical evidence findings were used to assess the accuracy of statements made by both Gary and his nephew. Ultimately, the case was negotiated without a trial.

The case history will be discussed along with some of the significant physical evidence results. A brief overview of OC (pepper) spray will also be presented, along with the analysis of OC spray from a cloth substrate using ethyl acetate to extract the stain and then analyzing the extract using a GC/MS. The extraction scheme was successful with an evidence sample that was over 3 ½ months old.

Species identification of ginseng roots by HPLC and discriminant analysis

Edgard Espinoza, DrPH, National Fish and Wildlife Forensic Laboratory - Ashland, OR

American ginseng (*Panax quinquefolius)*, which is listed as a Threatened and Endangered species, is illegally harvested from Fish &Wildlife Refuges and National Parks. Often the apprehended suspects claim that the roots in their possession are Korean ginseng (*Panax ginseng)* and not the protected species. Botanists rely on morphological characteristics of the plant for making species inferences, and find they are unable to determine species of root without the stems and leaves.

These two ginseng species contain a wide variety of saponins called ginsenosides. An HPLC study of the ginsenosides show that their presence and abundance differs and can be the basis for determining species of origin. Discriminant function analysis of the HPLC data confirms that the principal components of the evidentiary roots are in agreement with the analysis of ginseng root standards.

This analytical approach was applied to commercial ginseng root preparations purchased at health food stores. The results indicate that, by and large, the manufacturers are truthful in their claims of species origin.

A Visual Technique for Sex Determination in Humans Utilizing the Distal Humerus

Nici Wanek, Oregon State Police Crime Lab – Portland, OR

Sex determination of unidentified skeletal remains has historically been performed on bony elements such as the pelvis and the cranium. When these elements are absent or fragmented, however, the sex of the unknown individual must be determined through other means. The distal humerus, a largely overlooked skeletal element, exhibits marked differences between males and females. 649 individuals from the Smithsonian Institute and the Cleveland Museum of Natural History were used to test a visual technique of sex determination from the distal humerus. Results of this research strongly indicate that characteristics of this element are accurate in sex determination, regardless of the biological affiliation of the individual. This technique can be used with great success in contemporary forensic cases, as well as multiple grave recoveries, mass disasters, and plane crashes.

3D Spectrofluorometric Fingerprinting of Automobile Chassis Petroleum Products

Dale Purcell, Oregon State Police Crime Lab – Portland, OR

Petroleum product residues are often present on chassis of automobiles. These residues are complex mixtures of oils, greases, coolants and hydraulic fluids. This work investigates the use of three dimensional (3D) fluorescence data, visualized as contour plots, to characterize these residues with the goal of associating the transferred residue with its source. A method was developed to separate different classes of petroleum compounds: oils, asphaltenes, and pre-asphaltenes. Absolute identification of individual compounds was not attempted. This study shows that sufficient individual characterization can be obtained when all three sample fractions are considered together to associate an unknown sample to a known sample.

Identification of Some Components of Pyrodex® by Hot-Stage Microscopy

Chris Hamburg, Washington State Patrol Crime Lab – Tacoma, WA

Experiments were conducted to create an easier method to identify the components of some black powder substitutes. Traditional methods of component identification of black powder substitutes have consisted of particle picking and

extractions that can potentially produce mixtures. The differences in melting points and sublimation characteristics of some components, along with extractions and particle picking, help in obtaining component separation. The use of a hot-stage greatly assists in the identification by giving melting point data and sublimation temperature ranges. This approach can also be used with traditional (and inexpensive) hot plates and alcohol lamp.

On-line SFE GC/MS of Human Hairs for Age Discrimination

Jeremiah Bishop, George Washington University

Human head hairs from 13 female Caucasians of varying ages were analyzed for surface lipids by supercritical fluid extraction (SFE) gas chromatography – mass spectrometry (GC/MS) to determine if the age of an individual could be discriminated using the extracted surface lipids. Approximately 70 ?g of a hair sample was extracted using on-line carbon dioxide SFE and analyzed by GC/MS to determine what compounds were present. The extraction and analysis was completed a second time on each hair sample to ensure that as much extractable material as possible was removed. The reproducibility of these extractions and analyses were determined to be adequate for age discrimination by analyzing several samples multiple times. Using squalene, cholesterol, odd chain length free fatty acids, and wax esters age discrimination was accomplished. All but one of the known samples were correctly discriminated with respect to the originating individuals age. Two of the three blind study samples analyzed were correctly discriminated with respect to age. The third blind sample contained a contamination that prevented this sample from being discriminated correctly. Age discrimination of evidential hairs using on-line carbon dioxide SFE extraction coupled with GC/MS analysis may be a beneficial tool in the analysis of human head hairs.

A Look at Explosive Disrupters

Jeff Jagmin, Washington State Patrol Crime Lab - Tacoma, WA

Disrupters have become the primary tool used by bomb squads to disable explosive devices. A study was conducted to examine the various types of disrupters and what to expect when "rendered safe" evidence is submitted to the laboratory. Also discussed will be recommendations on what items should be submitted along with the "rendered safe" evidence, and questions to ask your bomb technician.

The role of the prosecution and defense scientists in defense of murder cases

James Pex and Peter Faye – former Chief Prosecutor, Lincoln County District Attorney Office

Overview of Services Provided to Law Enforcement Agencies

Wilma Jolly – National Law Enforcement and Corrections Technology Center, Western Region, California

Clandestine Ammonia Generation

Eric Person, Washington State Patrol Crime Laboratory - Marysville, WA

Northwest Washington has seen rapid growth of the use of ammonium sulfate fertilizer in generating ammonia for use in the manufacture of methamphetamine. It is unlikely that ammonia generated in this fashion would be 'anhydrous,' raising the concern of whether water would interfere in the chemical reduction. The tolerance of the lithium-ammonia method to water, methyl alcohol, and isopropyl alcohol was investigated.

DNA Special Topics Session * October 13-17, 2003 * Portland, Oregon

DNA Profiles in Bone Marrow Recipients

Elaine Kitano, Oregon State Police Crime Lab – Portland, OR

Mixtures of DNA in evidence samples can be a normal occurrence in forensics, however, it is possible to see a mixture in the reference standard if the individual giving the standard has had a bone marrow transplant. The oral and blood DNA profiles were compared from two bone marrow transplant recipients and their respective donors. The STR analyses of the oral samples from the individuals who had received a bone marrow transplant revealed a mosaic of DNA within their body.

Post-amplification yield gels

Dona Scarpone, Oregon State Police Crime Lab – Portland, OR

Are your CEs back-logged? Analysts spending a lot of time doing re-injections? Running a post-amplification yield gel may be your answer to a quick and easy way to alleviate some of your CE overcrowding.

Behind the Scenes of BodePlex: Implementation of miniSTR multiplexes on samples from World Trade Center disaster victims

Courteney W. McCoppin, The Bode Technology Group – Springfield, VA

The terrorist attacks on the World Trade Center (WTC) two years ago launched the largest DNA identification project ever. After the analysis of over 20,000 bones and tissue extracts with traditional STR multiplexes, it became clear that further analysis would be needed. Of the nearly 13,000 bones tested, less than half yielded useful profiles. There was an even lower rate of success from the tissue extracts. In order to obtain results from samples that previously proved unsuccessful, two novel miniSTR multiplexes were developed, BodePlex 1 and BodePlex 2. The manufacture and quality control took place at The Bode Technology Group. Individual primers ordered from outside vendors had to pass through rigorous quality control checks before they could be combined to form the multiplexes. A final stringent quality control check had to be performed on each multiplex batch. In all, 8771 samples were amplified in duplicate with both multiplexes (35,048 total amplifications). The eventual outcome was the development of several hundred more profiles that were used to identify WTC victims.

An Effective Design for Starting a Convicted Offender Program

Chrystal Bell, Oregon State Police Crime Lab – Portland, OR

When Oregon went to an all-felons database in January 2002, there were nearly 40,000 offenders that were required to submit samples for CODIS. The challenge for us was to find a low cost alternative to buying expensive commercial DNA collection kits. Using local businesses and modern software technology, we found innovative ways to both design our own kit and manage the large volume of samples coming into the lab.

Troubleshooting the Genetic Analyzer 310

Christina Stivers, Oregon State Police Crime Lab – Portland, OR

Troubleshooting the Genetic Analyzer 310 can be a hair splitting activity – getting to the root of the problem takes patience, time and skill. This talk will cover basic troubleshooting techniques to use when your CE has a loss in sensitivity, raised baseline, resolution issues, matrix issues, an increase or loss in current and error messages.

Correlation of Growth Phase with DNA Results Obtained from Hair Roots

Susan Hormann/Rhonda Banks, Oregon State Police Crime Lab – Portland, OR

With the advent of more sensitive DNA analysis capabilities, forensic hair examiners are more and more frequently asked to perform hair comparisons as a screening tool for DNA analysis rather than for the value of the comparison itself. Some forensic laboratories no longer perform traditional microscopic hair comparisons. Frequently, however, the number of hairs in a given case is large and many of the hair roots do not yield a DNA result. This study correlates the growth phase of the hair root and the DNA results obtained in an effort to determine if it is possible at the screening level to target hairs that will yield useable DNA results.

SWGDAM Update

Phil Kinsey, Oregon State Police Crime Lab – Portland, OR

Validation of the Qiagen Biorobot EZ1 for the DNA Extraction and Purification of Reference and Evidence Samples for Forensic Casework

Shawn Montpetit – San Diego Police Department – San Diego, CA

CAPTION**THIS!** CURRENT

Ladies and gentlemen, welcome to the always popular "CAPTION THIS!," the captioning game that is sweeping the nation. The power to deliver a witty slogan, subtitle, or description is finally in your hands.

The best caption submitted for the photo presented above will win a STARBUCK'S Coffee Gift Card.

Decision of the editor is final. Bribery may not be tolerated.

EMAIL: JOSH.SPATOLA@DOJ.CA.GOV

WINNER CAPTION**THIS!**

WE HAVE A WINNER!!!

"Why should I choose Y-STRs when I've got all these other chromosomes to choose from?**"**

-Daniel Petersen, Ph.D.
Oregon State Police Crime Lab

Congrats! Your STARBUCK'S Coffee Gift Card is on its way!

Original Caption...

Electron Tubes *March 11, 1959* - Inner assemblies and completed units of RCA's new high-performance, thimble-size electron tubes appear to be dancing in mid-air as Mary Tomed of the company's Harrison, N.J., plant inspects them. The photographer made this unusual shot by focusing his camera into a mirror beneath a glass plate. RCA's new tube, called a "Nuvistor," opens the way to more compact, more efficient electronic instrument for the home, the military, and industry. Original Caption by Science Service

2002 - 2003 NWAFS OFFICERS

EXECUTIVE COMMITTEE

PRESIDENT:	**WILLIAM MARSHALL** Washington State Patrol Toxicology Laboratory 2203 Airport Way South Seattle, WA 98134-2027	206.464.5435 (phone) wmarsha@wsp.wa.gov
VICE-PRESIDENT:	**STUART JACOBSON** Idaho State Police Forensic Services 1000 Hubbard, Suite # 240 Coeur d' Alene, ID 83814	208.769.1410 (phone) 208.769.1410 (fax) stuart.jacobson@isp.state.id.us
MEMBER-AT-LARGE:	**BILL SCHNECK** Washington State Patrol Crime Laboratory - Spokane W. 1100 Mallon Ave. Rm 100 Spokane, WA 99201-2043	509.456.4144(phone) bill.schneck@wsp.wa.gov
SECRETARY-TRESURER:	**RHONDA BANKS** Oregon State Police Crime Laboratory 1111 SW 2nd Ave. Rm 1201 Portland, OR 97204-3258	503.229.5017 (phone) 503.229.6638 (fax) rhonda.banks@state.or.us

SECRETARIES

MEMBERSHIP:	**DAN ALESSIO** Oregon State Police Crime Laboratory 1111 SW 2nd Ave. Rm 1201 Portland, OR 97204-3258	503.229.5017 (phone) 503.229.6638 (fax) dan.alessio@state.or.us
TECHNICAL:	**NICI WANEK** Oregon State Police Crime Laboratory 1111 SW 2nd Ave. Rm 1201 Portland, OR 97204-3258	503.229.5017 (phone) 503.229.6638 (fax) nici.wanek@state.or.us
EDITORIAL:	**JOSH SPATOLA** California Department of Justice Central Valley Crime Lab 1306 Hughes Lane Ripon, CA 95366	209.599.1418 (phone) 209.599.1240 (fax) josh.spatola@doj.ca.gov

NWAFS Spring Meeting
April 19-23, 2004
Holiday Inn Parkside
Missoula, MT

Come enjoy beautiful Montana! Our conference will feature the traditional meeting format with three days of workshops followed by a day and a half technical session.

Dates: April 19 – 23, 2004 **Location:** Holiday Inn Parkside
Missoula, Montana
Reservations 406-721-8550

The Holiday Inn Parkside is located along the Clark Fork River between beautiful downtown Missoula and University of Montana, and while it is a nice hotel, it's certainly not too classy for our group!

Scheduled Workshops:

Hair Examination for DNA Analysts	*	Bone Identification
Trends in Ammunition	*	Bloodspatter
Shooting Scene Reconstruction	*	Fast GC
Forensic Applications of LC/MS	*	GC/MSD Maintenance
Sexual Assault Evidence Collection	*	SEM Calibration
Conflict Resolution in the Workplace	*	Clan Lab Synthesis
Trace Evidence from Ammunition	*	Forensic Epidemiology

Calculating and Presenting DNA Evidence in Court

Social Activities:

Tuesday Evening Fly Fishing Demonstration
Wed Evening Vendor Reception Featuring Vendor Sumo Wrestling
Thursday Evening Banquet Featuring Sperm Races and Dancing
Friday Morning Breakfast Buffet Featuring Surprise Keynote Speaker!

Contact Information: Annalivia Bishop
Montana State Crime Lab
406-728-4970
abishop@state.mt.us