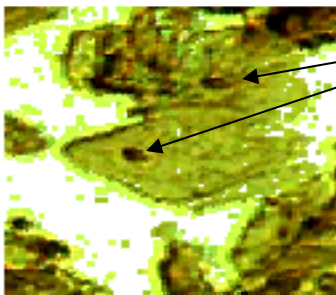


Collecting a Buccal Swab – An Art or a Cinch?

By

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An increasing number of cases tried in the courtroom involve DNA evidence. If a DNA profile is recovered from crime scene evidence, it is typically necessary to collect reference samples from suspects and victims, for comparison purposes. The preferred form of a reference sample is a buccal (buccal is Latin for cheek) or oral swab. Buccal cells are found inside the mouth, lining the cheeks. Buccal cell samples are easy to collect, collection devices are inexpensive, and buccal cells are a reliable source of DNA. In order to obtain court admissible DNA results, a buccal swab must be collected, handled, and stored properly. Below are some pointers on how to maintain the integrity of buccal swab evidence.



Buccal cells magnified 400x
Arrows show nuclei which contain DNA

Obtaining a buccal swab sounds easy enough. Simply find a volunteer to stick a swab in someone's mouth, right? Wrong! Although taking a buccal swab may not require specialized training or certification, taking the time to learn the proper technique of collecting and storing a swab and understanding what can go wrong if the swab is not collected properly is essential. Swab collection and the way it is handled post-collection can determine if a sample will maintain its integrity and be useful in a courtroom or be deemed useless due to complications such as contamination, degradation, and insufficient yield.

FOUR STEPS OF PROPER BUCCAL SWAB COLLECTION

For the purposes of this paper, the buccal swab collection has been broken down into four basic steps: Verification, Preparation, Collection, and Packaging/Storage. It is important that all four steps are performed properly to ensure full integrity of the buccal swab. It is equally important to have all of the necessary supplies at the time of swabbing. An ideal strategy is to have a pre-made buccal swab collection kit. A generic collection kit usually contains a set of directions, two swabs, two pairs of gloves, a mask, collection envelopes, and evidence tape (for sealing of envelopes). Pictured below are the contents of a typical buccal swab collection kit.



Verification

To verify that you are swabbing the subject of interest, ask for a photo identification, such as a driver's license or a passport and photograph it. If the subject is incarcerated, compare the subject's name and booking number from their jail or prison identification to the name of the subject of interest. Take a photograph of the subject's jail or prison identification. Additionally, take a headshot of the subject that is being swabbed. If you bypass this step, you could end up with a story similar to that played out in the first case in which forensic DNA analysis was utilized. Colin Pitchfork paid a friend to donate a DNA sample in his name during the investigation of the death of two girls in the 1980's. Pitchfork would have gotten away with the crimes if his imposter friend had not been overheard telling a friend about the sample switch.

Preparation

Some jurisdictions require a consent form to be signed by the subject being swabbed. In preparation for collecting the swab, be sure the consent form is signed so that the data obtained from the sample can be admissible in court. Before physically collecting the swab, confirm that the subject's mouth is empty. If there is food, gum, or tobacco in the subject's mouth, not only will you get a disgusting swab, but the swab will be introduced to substances that could cause complications with the DNA typing procedures. Put on gloves and a mask and inform the subject that you will be placing a swab on the side of the cheek and gently rubbing the cheek for about 5-10 seconds. Emphasize to the subject that they will not experience any pain.

Collection

Immediately before collecting the sample, open the outer packaging of the swab, remove the swab from the packaging without handling the actual swab, and immediately swab the subject's mouth. The swab should not touch anything before entering the subject's mouth. After swabbing, place the swab directly into the receptacle or the supplied packaging. Do not set the swab down or wave it in the air, simply place it into the proper packaging to dry. Most swab kits come with a receptacle or some sort of packaging that contains a desiccant, which is a chemical agent that absorbs moisture to promote timely and proper drying of the swab.

Packaging/Storage

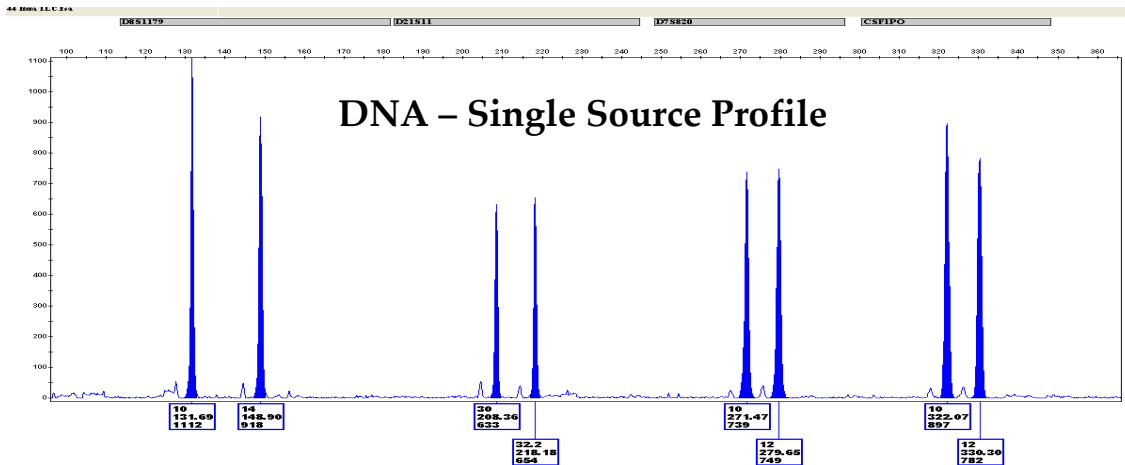
After it is safely in the receptacle, label the receptacle and place into an envelope. Label the packaging with the required information, such as the subject's name, the time, date, and place of collection, person collecting the sample, along with the identification information provided. Tape seal, initial, and date the outer packaging for chain of custody purposes, which is essential for cases that go to court. Keep the swab at room temperature until it can be stored in a freezer.

POTENTIAL COMPLICATIONS

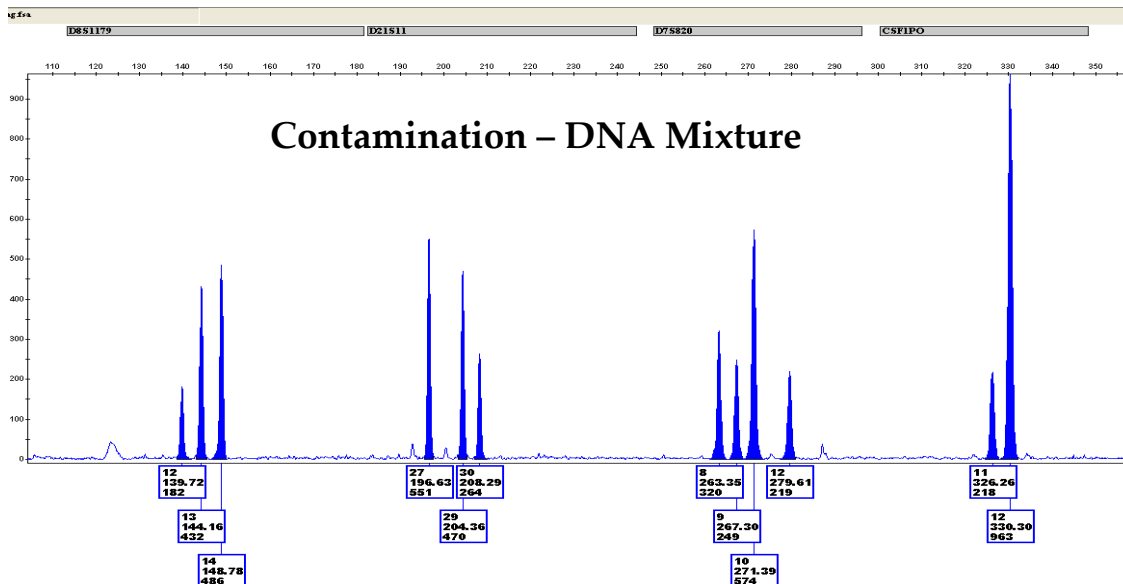
After reading the four steps, it is obvious that taking a buccal swab is far from complicated. But in reality, if the swab is not collected and/or handled properly, there are potential problems that can occur that will cause complications when interpreting the DNA profile. These potential problems include contamination, degradation, and insufficient yield. These are complications that should **NOT** be encountered when typing a reference sample. When they appear in a sample that should be pristine, the information obtained from the sample is useless.

Contamination

When analyzing a buccal swab, contamination is considered present when there are DNA profiles from more than one individual. The profiles are obtained by analysts in the form of an electropherogram. Electropherograms are a visual and graphical representation of the data collected from typically 14 to 16 different locations of the DNA. The electropherograms pictured below show only 4 of the 16 common DNA locations analyzed by forensic laboratories. In the single source profile at each location, there are two peaks. The two peaks represent the phenomenon that half of the DNA is inherited from the mother and the other half is from the father. The parents contribute equally to the genetic make up of their offspring by each contributing a peak. Also notice that the heights of the peaks are fairly even or equal.



Looking at the contaminated DNA mixture result, notice at three of the same four DNA locations, there are more than 2 peaks. The peaks at every location vary in height. No evenness between the peaks is present. These are indicators that more than one contributor is present. It is not known which peaks belong to the individual swabbed. A sample that contains a DNA mixture can not be used as a reference sample. When contamination occurs with a reference swab, the swab has to be collected again and that costs more money, plus the time of the individual collecting it and the analyst processing the sample.

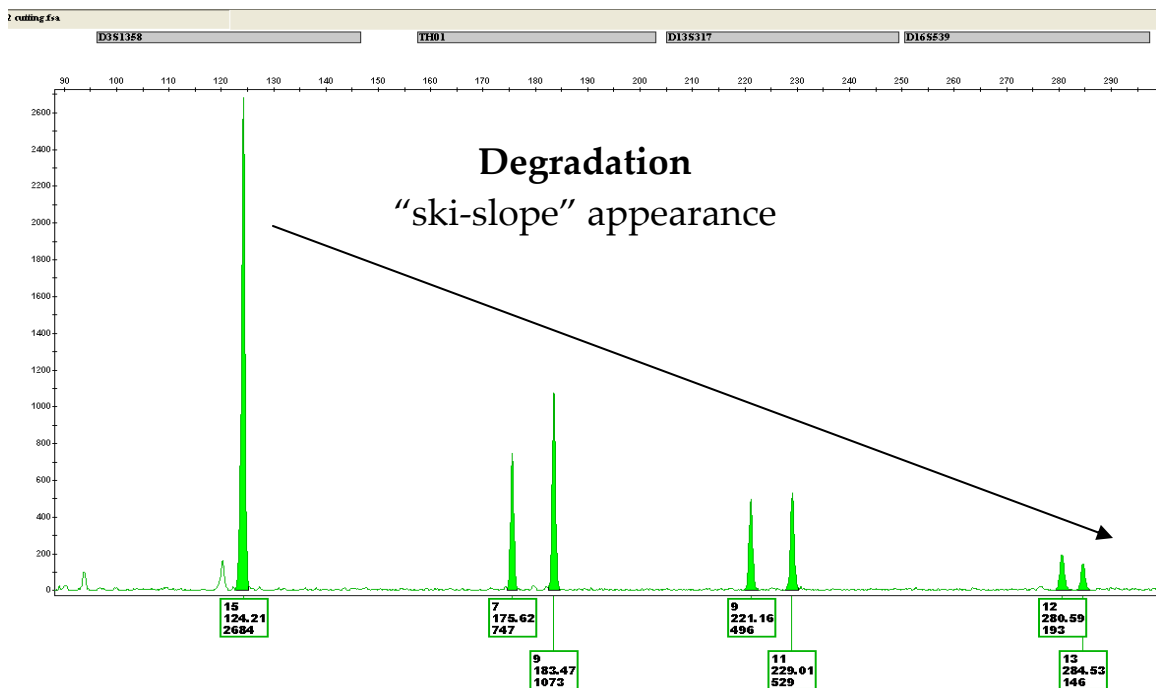


During buccal swab collection, there are steps that can be taken to avoid contamination. While taking the swab do not talk and make sure a mask is worn. No one, including myself, likes to wear a mask, but keep in mind that saliva contains buccal cells and buccal cells contain DNA. When talking, sneezing, and laughing, saliva is expectorated. If any of this expectorated saliva lands on a buccal swab, the swab is contaminated. With this in mind, it is preferable to collect the sample in a low traffic area. Do not open the swab packaging until it is time for the collection of the swab. It is good to be prepared, but opening the package too early introduces additional opportunities for the swab to be contaminated. After the swab is collected, place the swab in the receptacle packaging immediately versus placing it on any kind of surface. The goal is to protect the swab from contaminants and also to promote drying of the swab to prevent bacterial growth.

Degradation

Another potential concern when collecting a reference sample is the possibility of degradation. Degradation occurs when the swab is exposed to different factors such as sunlight, heat, humidity, water, and chemicals. When DNA is degraded it means that bacteria and/or chemicals have caused breaks in the DNA strand. These breaks disrupt the DNA analysis process which ultimately results in a DNA profile, like the one pictured below, that has what is referred to as a “ski slope” appearance. There are no longer nice, even peaks throughout the profile. Similar to meeting the height requirements for a ride at an amusement park, there are height requirements for peaks, meaning they have to be a certain height, or exceed what analysts call “threshold”, to be considered reliable. A peak height “threshold” is determined in each laboratory for each piece of equipment. For example, if the threshold was determined to be ‘200’, in the profile below, the peak heights at three of the four locations can be used. But the smallest locations cannot be used because the peaks are not tall enough and fall below the interpretation thresholds. Losing information reduces the discriminatory power and complicates the interpretation.

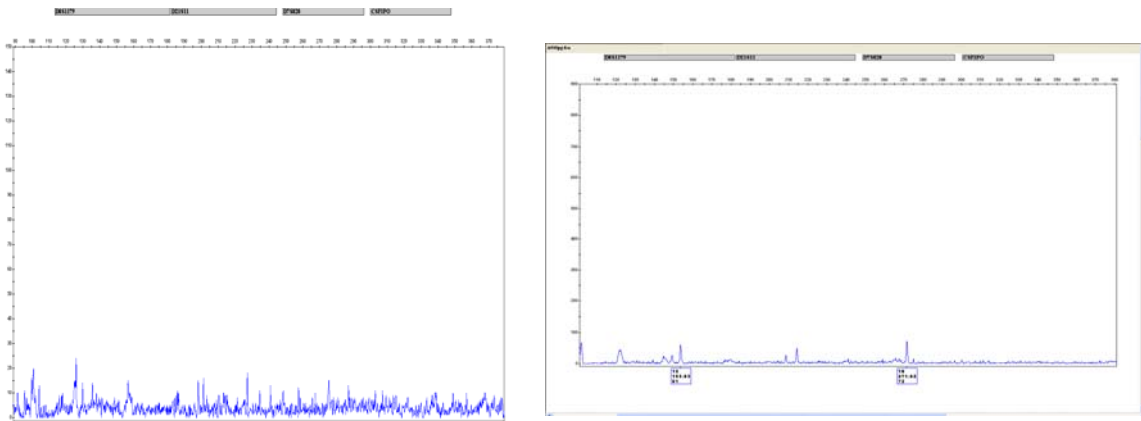
Ways to avoid degradation are to limit the swabs exposure to the factors already mentioned, sunlight, heat, etc. Keeping the swab in a cool dry environment slows down the degradation process of bacteria substantially. The best practice is to protect the swab by immediately placing it in its receptacle and by keeping it at room temperature until it is properly stored in a freezer.



Insufficient Yield

One last potential problem faced is obtaining a DNA result from a reference swab that displays insufficient yield. When forensic scientists use the phrase “insufficient yield”, it means there is either no DNA present or only a partial profile was obtained. As discussed earlier, the forensic community analyzes 14-16 DNA locations. A partial profile means there are DNA results (peaks) at some, but not all of the DNA locations. This results in less information to work with. Pictured below are two examples of what a DNA result will look like if there is not enough DNA. On the left, the electropherogram pictured is a profile with absolutely no DNA present at all. On the right the electropherogram pictured is considered a partial profile. Two out of the four locations pictured have small peaks that may or may not reach the determined threshold. The data cannot be reliably interpreted because there is not enough DNA present. The idea of insufficient data should never be a concern with reference samples if collected and stored properly.

Insufficient DNA



To avoid obtaining an insufficient amount of DNA from a reference swab, make sure the swab actually touches the cheek of the subject. After the swab is touching the cheek, it needs to be rubbed **and** rotated against the cheek for approximately 5-10 seconds, making sure that the whole surface of the swab has made contact with the cheek. Sometimes the subject likes to take their own swab, which is appealing because it is much more comfortable for both the swabber and the subject. But that leaves potential for an insufficient sample. If the subject does not actually touch his/her cheek with the swab or they fail to properly rub and rotate the swab, the amount of DNA collected may not be enough to obtain a full DNA profile. It is advised that a properly trained individual who understands the complications of a swab collection gone wrong actually collect the sample. After the swab is collected, make sure the swab is packaged, labeled, and stored properly, which has already been discussed earlier in the article.

CONCLUSIONS

At first blush, the idea of collecting a buccal swab appears to be simple task. But in reality, there are factors that have to be considered, such as contamination, degradation, and insufficient yield while taking the sample. Because DNA testing techniques are so sensitive, it is surprisingly easy to compromise a sample. It is important to collect, package, and store reference swabs properly to obtain the best DNA results.

As already mentioned, collecting a buccal swab for a reference sample may not require specialized training or certification, but regardless, obtaining some basic training or attending a short hands-on introductory seminar is recommended.