This document sets forth background materials on the scientific research supporting examinations as conducted by the forensic laboratories at the Department of Justice. It also includes a discussion of significant policy matters. This document is provided to assist a public review and comment process of the related Proposed Uniform Language for Testimony and Reports (posted separately). It is not intended to, does not, and may not be relied upon to create any rights, substantive or procedural, enforceable by law by any party in any matter, civil or criminal, nor does it place any limitation on otherwise lawful investigative and litigative prerogatives of the Department.

SUPPORTING DOCUMENTATION FOR DEPARTMENT OF JUSTICE
PROPOSED UNIFORM LANGUAGE FOR TESTIMONY AND REPORTS
FOR THE FORENSIC HAIR EXAMINATION DISCIPLINE

Background

Hairs are threadlike outgrowths from the skin of mammals which consist primarily of dead, keratinized cells. Once shed or removed, these hairs may transfer directly or indirectly from one location to another. Questioned hairs recovered from an item can be microscopically examined to determine human or animal (non-human) origin, characteristics of ancestry, somatic origin, artificial treatment, damage, decomposition, and growth stage. Moreover, microscopic comparisons of hair characteristics may determine if a person can or cannot be included as a possible source of a questioned hair. However, microscopic comparison of hairs cannot be a basis for personal identification. Following microscopic comparison, human hairs that are associated microscopically are assessed for DNA analysis. Hairs with apparent tissue at the root end and/or hairs in the active growth phase (anagen) are designated for nuclear DNA analysis. Hairs in the resting phase (telogen) with no adhering tissue and hairs without a root are designated for mitochondrial DNA analysis.

Principles of Forensic Hair Examination

When conducting hair comparisons, the examiner assesses the microscopic characteristics of the hair evidence in order to come to a conclusion. An examiner may analyze hairs microscopically to determine somatic origin, characteristics of ancestry, animal (non-human) hair classification, growth stage, damage, artificial treatment, characteristics of decomposition, and suitability for comparison. Depending on the microscopic characteristics that are present, an examiner can come to one of three conclusions regarding the comparison of a questioned hair to a known hair sample: inclusion, exclusion, or inconclusive. Additionally, examiners need to be aware that the science of microscopic hair comparison does not support statements of individualization, statistical weight, or zero error rate.


Theory of Microscopic Hair Examination

The microscopic examination of hairs relies on differences in morphology to classify and distinguish hairs. There are two main regions of a hair, the shaft and the root. The root of the hair lies in the follicle, extending through most of the dermis layer of the skin.\(^3\) The shaft of the hair is made of three layers called the cuticle, cortex, and medulla (see Figure 1). The cuticle surrounds the exterior of the hair and is composed of overlapping scales. Underneath the cuticle is the main body of the hair known as the cortex. The cortex contains pigment granules that provide color, small air pockets called cortical fusi, and opaque structures known as ovoid bodies. The innermost layer of cells located near the core of the hair is the medulla. Documented variations in the presence, distribution, appearance, and arrangement of the characteristics described above is the basis for microscopic hair examinations and comparisons.\(^4\)

![Figure 1: Microscopic layers of the hair shaft.](image)

Microscopic examination of hairs has been used in criminal investigations since the late 1800s. The first human hair case was reported in Germany in 1861 by Rudolf Virchow.\(^5\) Since this time, numerous publications have described the microscopic characteristics of hairs in detail, including the reliability of using the characteristics to distinguish between individuals.\(^6\) In a

---


1934 publication entitled “Histological Variability of Human Hair”, Dr. Leon Hausman stated the following:

From the results of studies noted in this paper recently made by the writer, and here described, it would appear that a) certain microscopic structural elements in the hair shaft are relatively fixed in their correlations, and b) others appear to be individual enough to be usable as criteria for personal identification.\(^7\)

Studies have demonstrated how variation in hair morphology between individuals provides meaningful comparisons when utilizing comparison microscopy. A blind study by Strauss\(^8\) demonstrated that comparison microscopy correctly associated 100 questioned hairs to the 100 known hair samples. In addition, all 100 questioned hairs were correctly characterized into the appropriate racial classification. A series of studies performed by Gaudette in the 1970s\(^9\) demonstrated that both head hairs and pubic hairs could be reliably distinguished from one another. In the head hair studies, a total of 370,230 intercomparisons were conducted, with only nine pairs of hairs that could not be distinguished. In the pubic hair studies, a total of 102,831 intercomparisons were conducted, with only sixteen pairs of hairs that could not be distinguished. In these studies, Gaudette developed a probability estimate for head hair and pubic hair comparisons.

In a later study, Gaudette\(^10\) stated that “the significance of this research is not in the actual probability numbers found but in experimental proof of the proposition that macroscopic and microscopic hair comparison is a useful technique and that hair evidence is good evidence.” In this study, he provided a single known head hair sample and 100 questioned hairs to examiner trainees. Two of the trainees correctly associated the correct questioned hair to the known sample, and the third examiner trainee found three questioned hairs to be similar to the known sample. In the second part of this study, Gaudette provided a single questioned hair and 100 known hair samples to examiner trainees. Two of the examiner trainees correctly associated the questioned hair to the correct known sample, and the third examiner trainee associated the questioned hair to the correct known hair sample and to one additional known hair sample.

Bisbing et al. further demonstrated how the method can also be used to distinguish between twins.\(^11\) In one study, he obtained head hair samples from 17 pairs of twins and one set of

---


triplets. In the first part of this study, duplicate hair samples from each twin were compared. The researchers were able to pair each hair sample to its correct duplicate sample without any incorrect associations or exclusions. The second part of this study was designed to simulate forensic case work, where seven separate questioned hairs were compared to several randomly selected known samples obtained from the twins or triplets. In the seven tests, one examiner correctly excluded 47 out of 52 samples, and a second examiner correctly excluded 49 out of 52 samples.

The microscopic hair comparison process is not limited to human hair comparisons; it can also be applied to animal (non-human) hairs. In 1988, Suzanski\textsuperscript{12} conducted a blind study involving the comparison of 15 questioned hairs to 25 known hair samples taken from purebred German Shepherd dogs. Six of the fifteen questioned hairs were correctly associated to their source, and no incorrect inclusions were made in the test. In a second study,\textsuperscript{13} 25 questioned samples were compared to 100 known samples obtained from mixed-breed and purebred dogs. All 25 of the questioned samples were correctly associated to their source, with no incorrect associations.

Based on these and other published studies, microscopic hair comparison has been demonstrated to be a valid and reliable scientific methodology. These studies have also shown that microscopic hair comparisons alone cannot lead to personal identification and it is crucial that this limitation be conveyed both in the written report and in testimony.

The science of microscopic hair comparison acknowledges that the microscopic characteristics exhibited by a questioned hair may be encompassed by the range of characteristics exhibited by known hair samples of more than one person. If a questioned hair is associated with a known hair sample that is truly not the source, it does not mean that the microscopic hair association is in error. Rather, it highlights the limitation of the science in that there is an unknown pool of people who could have contributed the questioned hair. However, studies have not determined the number of individuals who share hairs with the same or similar characteristics.

**Microscopic Hair Comparison Process**

There are different methodologies and processes for conducting a microscopic hair comparison examination. The Department shares information regarding some appropriate processes below. The Department does not suggest that the processes outlined here are the only valid or appropriate processes.

The general procedure for microscopic hair analysis requires the use of comparison microscopy (higher magnification) to examine the microscopic characteristics.


Prior to a comparison, hairs are characterized and classified according to their morphology. The first step is determining whether the hair is of human or animal (non-human) origin. Human or animal (non-human) origin can be determined by examining features in both the root and shaft portions. Table 1 lists some of the differences between human hairs and animal (non-human) hairs.

<table>
<thead>
<tr>
<th>Feature</th>
<th>Human</th>
<th>Animal (non-human)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Color</td>
<td>Relatively consistent along length</td>
<td>Often showing profound color change and banding</td>
</tr>
<tr>
<td>Cortex</td>
<td>Cortex is larger than the width of the medulla</td>
<td>Cortex is often less than the width of the medulla</td>
</tr>
<tr>
<td>Distribution of pigment</td>
<td>Even or slightly more towards cuticle</td>
<td>Denser towards medulla</td>
</tr>
<tr>
<td>Medulla</td>
<td>Less than one-third width of shaft with amorphous irregular appearance</td>
<td>Often greater than one-third width with defined structure</td>
</tr>
<tr>
<td>Cuticle</td>
<td>Imbricate (no repeating pattern), flattened scales. Similar along length of shaft and relatively smooth.</td>
<td>Wide range of scale patterns. Often show variation in structure along length of shaft.</td>
</tr>
</tbody>
</table>

Animal (non-human) hairs are further examined to determine the type of hair (e.g., fur, guard), the type of animal (non-human) (e.g., dog, cat), and suitability for comparisons. Human hairs are further characterized to determine characteristics of somatic origin, ancestral group classifications, growth, presence of damage, presence of artificial treatment, presence of characteristics of decomposition, and suitability for comparisons.

Somatic origin (body area) classifications are based on the macroscopic and microscopic characteristics that are typically observed in hairs from different areas of the body. A human hair may be classified according to its region of the body using features such as root appearance, diameter, diameter variation, texture, and medulla. Ancestral group classifications are based

---


16 Guard hairs are the thicker hairs that contain the characteristics used to distinguish between mammals.

on characteristics that are typically observed in hairs from individuals of different ancestral groups and may or may not correspond with how an individual identifies his or her race. Characteristics of ancestry can often be determined using features such as pigment distribution and cross-sectional shape.\textsuperscript{18}

Hair follicles have a growth cycle where they actively grow (anagen phase) for weeks to years before they eventually enter a resting stage (telogen phase) that typically lasts one to three months. Hairs in the anagen stage have a hair bulb connected to tissue at the bottom of the follicle and require some force to be removed; however, the amount of force required to remove a specific hair is unknown. The root bulb of these hairs becomes stretched when removed from the follicle. Hairs in the telogen phase are no longer growing and are in the process of moving closer to the surface of the skin (moving upward in the follicle. These hairs are easily removed and often pushed out by a new, actively growing hair. A hair at the end of its growth cycle (telogen phase) forms a club shaped root.\textsuperscript{19} An examiner can describe a root as exhibiting characteristics of the anagen or telogen growth phase.

Damage to, artificial treatment of, and characteristics of decomposition in hair typically leave microscopic characteristics that can aid in the hair comparison process. Hairs that have been burned or singed often exhibit round air pockets within the cortex and/or medulla. Hairs that have been crushed exhibit a widened shaft that often has separation in the cortex. Broken hairs exhibit uneven separation with elongated tags while cuts with scissors or a razor have a cleaner separation leaving a smooth edge.\textsuperscript{20} Bleached and dyed hairs (artificial treatments) often exhibit a distinct color change (line of demarcation) between the untreated and treated portion of the shaft. This is due to a portion of the hair in the follicle being protected during the treatment process. An estimated time since treatment may also be provided by measuring the length of


untreated portion and comparing that against available growth rates. Postmortem banding is a characteristic of decomposition, and is displayed as an opaque band at the root end of hairs. This characteristic may be observed in hairs removed from a decomposing body. Putrid roots are another form of hair decomposition in which the root end exhibits a tapered or brush-like appearance. While characteristics of decomposition at the root end may be observed in hairs removed postmortem, the underlying process that causes these characteristics is unknown. These characteristics can be used as part of the comparison process, and may provide additional information to the investigative process.

In order to conduct hair comparisons, it must be determined whether or not the questioned hair is suitable for microscopic comparison purposes. Research has demonstrated that head hairs and pubic hairs with sufficient microscopic characteristics are suitable for meaningful comparisons. Head hairs must be compared with known head hair samples and pubic hairs must be compared to known pubic hair samples. While there is no minimum number of characteristics necessary to reach a conclusion, the quality of the characteristics present must be sufficient to conduct meaningful comparisons. Featureless hairs (e.g., white/gray hairs), hair fragments, and hairs from body areas other than the head and pubic region are not considered suitable for meaningful microscopic comparisons. Hairs from other body areas and hairs with limited characteristics may be compared; however, Department laboratory practices require that a statement will be included in the report regarding the limited conclusions that can be reached.

Comparisons are conducted using a high quality comparison microscope to allow side-by-side examination of the questioned hair and known hair sample. All of the characteristics present in the questioned and known sample are considered as part of the comparison process. In


addition, corresponding portions of the questioned and known hairs are compared as the characteristics of a hair can vary from root to tip.  

In order to account for the range of characteristics that may be present, the Department laboratory requests approximately twenty-five hairs be submitted for each known human hair sample. The examiner must determine whether or not the characteristics in the questioned hair are represented by the range of characteristics in the known hair sample. If all of the characteristics of the questioned hair are represented in the known hair sample, the source of the known sample can be included as a possible source of the questioned hair. For animal (non-human) hairs, the examiner may conclude that the questioned animal (non-human) hair is microscopically consistent with the known animal (non-human) hair sample and, accordingly, the source of the known hair sample can be included as a possible source of the questioned hair. However, animal (non-human) hairs do not typically possess sufficient differences in microscopic characteristics to distinguish between animals (non-human) of similar breed and color. For human hair, the examiner may conclude that the questioned human hair is microscopically consistent with the known hair sample and, accordingly, the source of the known hair sample can be included as a possible source of the questioned hair. 

Microscopic hair comparisons are meaningful due to the variation in macroscopic and microscopic characteristics between individuals. However, the comparison of hair characteristics does not constitute a basis for personal identification and the number of individuals who could be included as a possible source of a specific hair is unknown. While there have been published attempts to assign a probability to microscopic hair comparisons, 


there is currently no valid statistical number that can be assigned to a microscopic hair
association.

If there are significant differences between the characteristics in the questioned hair and the
known hair sample, the source of the known sample cannot be included as a possible source of
the questioned hair. A significant difference is defined as a characteristic(s) in the questioned
hair that is not represented by the known hair sample. The accurate exclusion of an individual as
the source of a questioned hair relies on an adequate known sample that is representative of the
person’s hair. There are several factors that can lead to an inadequate known sample including
poor collection methods and/or a change in microscopic characteristics between deposition of the
questioned hair and collection of the known hair sample. Microscopic characteristics of hairs
can change over time or can be intentionally altered (e.g., artificial treatment) in such a way that
the questioned hair is no longer consistent with the known sample.

If the characteristics in the questioned hair and known sample are similar in most respects,
but there are slight dissimilarities, the examiner may reach an inconclusive opinion. There are
several reasons hairs may exhibit both similarities and dissimilarities to the known sample. The
dissimilarity could be attributed to an inadequate known sample, a change in microscopic
characteristics based on a substantive time period between deposition of the questioned hair and
collection of the known hair sample, and/or because the source of the known sample has altered
his/her hair. Another explanation is that the questioned hair did not originate from the source of
the known sample, but from another person who has hairs with similar characteristics.

Hairs recovered from an item may also be of limited value for microscopic comparisons.
Examples of questioned hairs that are of limited value are head/pubic/facial hairs that are
featureless (e.g., white/gray) or short head/pubic/facial hair portions. These types of hairs may
be compared for exclusionary purposes but are not suitable to reach meaningful associations due
to their limited nature.

It also is important to note that neither the absence of questioned hair on an item/person nor
the exclusion of a questioned hair from a given source necessarily means that the given source
did not come into contact with the item/person. This is because of the many factors involved in
the transfer and persistence of hair evidence. For example, hairs may not transfer during contact
or hairs that have transferred may be lost prior to collection.

Policy Considerations

A. DNA analysis of hairs

With the implementation of DNA testing, alternative analyses became available to be used in
conjunction with microscopic hair analysis. If the Department determines an evidentiary hair
possesses sufficient tissue, nuclear DNA (nDNA) analysis may be conducted; however, crime
scene hairs typically do not contain sufficient tissue for nDNA analysis. Accordingly,
mitochondrial DNA analysis (mtDNA) is the most frequently used approach for DNA typing of
evidentiary hairs.\textsuperscript{32} nDNA or mtDNA analysis can be used in conjunction with microscopic hair comparison when appropriate. These two techniques “can be used in tandem and may add to one another’s value for classifying a common source.”\textsuperscript{33} This premise was supported by a 2002 study, which indicated that out of 80 microscopic associations, approximately 88\% were also included by additional mtDNA testing.\textsuperscript{34}

B. 2009 National Academy of Sciences Report

In 2006, Congress authorized the National Academy of Sciences (NAS) to conduct a study on forensic science, which culminated in a 2009 report.\textsuperscript{35} The NAS report discussed the basis for and benefit of the forensic discipline of forensic hair analysis stating:

Forensic hair examiners generally recognize that various physical characteristics of hairs can be identified and are sufficiently different among individuals that they can be useful in including, or excluding, certain persons from the pool of possible sources of the hair. The results of analyses from hair comparisons typically are accepted as class associations; that is, a conclusion of a “match” means only that the hair could have come from any person whose hair exhibited—within some levels of measurement uncertainties—the same microscopic characteristics, but it cannot uniquely identify one person. However, this information might be sufficiently useful to “narrow the pool” by excluding certain persons as sources of the hair.\textsuperscript{36}

The report also identified specific concerns about microscopic hair analysis, specifically:

- There are “[n]o scientifically accepted statistics about the frequency with which particular characteristics of hair are distributed in the population.”\textsuperscript{37}
- “There appear to be no uniform standards on the number of features on which hairs must agree before an examiner may declare a ‘match.’”\textsuperscript{38}


\textsuperscript{34} Houck, M. M., Budowle, B. (2002). Correlation of Microscopic and Mitochondrial DNA Hair Comparisons. Journal of Forensic Sciences, 47, 964-967. Out of the 80 hairs associated by microscopy, 9 were excluded by mtDNA. The authors warn that results of this study should not be construed as a false positive rate for microscopy or a false exclusion rate for mtDNA. In casework, there are often numerous exclusions by microscopy that are not submitted for mtDNA and therefore, were not included in this study. Furthermore, it is known that both techniques have limitations in that they cannot identify a person through hair analysis, but rather include or exclude a person as a possible source. Therefore, an inclusion by one technique and exclusion by the other does not imply error, but rather highlights the need for both analyses in order to gain the most information and reduce the number of possible contributors to a questioned hair. Over the four year time period that the authors studied, microscopy narrowed the number of known samples submitted in these cases to 80; mtDNA further narrowed the number to 71.

\textsuperscript{35} NAS Report at 161.

\textsuperscript{36} Id. at 155-156.

\textsuperscript{37} Id. at 160.

\textsuperscript{38} Id.
• “The categorization of hair features depends heavily upon examiner proficiency and experience.” 39
• Imprecise reporting terminology can lead to misunderstanding and wrongly imply individualization.40
• Although “Microscopy and mtDNA analysis can be used in tandem and may add to one another’s value for classifying a common source … no studies have been performed specifically to quantify the reliability of their joint use.”41

39 Id. at 160.
40 Id. at 161.
41 Id.