



Best Practice Manual for the Microscopic Examination and Comparison of Human and Animal Hair

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Background

This Best Practice Manual (BPM) belongs to a series of 10 BPMs issued by the European Network of Forensic Science Institutes (ENFSI) in November 2015. The series covers the following forensic disciplines:

1. Forensic Examination of Digital Technology
2. Forensic Examination of Handwriting
3. Chemographic Methods in Gunshot Residue Analysis
4. Road Accident Reconstruction
5. Microscopic Examination and Comparison of Human and Animal Hair
6. Fingerprint Examination
7. DNA Pattern Recognition and Comparison
8. Application of Molecular Methods for the Forensic Examination of Non-Human Biological Traces
9. Forensic Recovery, Identification and Analysis of Explosives Traces
10. Forensic Investigation of Fire Scenes which have resulted in Fatalities*
11. Forensic Investigation of Fire Scenes which involve the Clandestine Manufacture of Improvised or Homemade Explosive Devices*
12. Forensic Investigation of Fire Scenes which Involve the Clandestine Manufacture of Illicit Synthetic Drugs*

* *The three specific areas on Forensic Investigation of Fire Scenes (numbers 10 -12) were combined into one BPM 'Investigation of Fire Scenes'.*

In the years 2014 and 2015, so-called Activity Teams have - in parallel - developed the 10 BPMs. The activities were performed within the project 'Towards European Forensic Standardisation through Best Practice Manuals (TEFSBPM)' and co-ordinated by the ENFSI Quality and Competence Committee. The realisation of the BPMs was supported by the Prevention of and Fight against Crime Programme of the European Commission – Directorate General Home Affairs (code: PROJECT HOME/2012/ISEC/MO/4000004278). The core project concept was that the BPMs will enhance the quality of the forensic services available to law enforcement and justice across Europe and thereby encourage forensic standardisation and cross-border cooperation between countries.

ENFSI expects that the issuing of this series will stimulate the improvement of already existing BPMs as well as the creation of new BPMs on disciplines that are not covered yet.

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The text may be translated into other languages as required. The English language version remains the definitive version.

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1 AIMS

This Best Practice Manual (BPM) aims to provide a framework for procedures, quality principles, training processes and approaches to the forensic microscopic examination of human and animal hairs. This BPM can be used by Member Laboratories of ENFSI and other Forensic Science Laboratories to establish and maintain working practices in the field of microscopic examination and comparison of human and animal hair that will deliver reliable results, maximize the quality of the information obtained and produce robust evidence. The use of consistent methodology and the production of more comparable results will facilitate interchange of data between laboratories.

The term BPM is used to reflect the scientifically accepted practices at the time of creating this document.

The term BPM does not imply that the practices laid out in this manual are the only good practices used in the forensic examination of hairs. In this series of ENFSI Practice Manuals the term BPM has been maintained for reasons of continuity and recognition.

2 SCOPE

This document provides guidelines for the entire forensic process of human and animal hair examination, including recovery at scenes of crime or in the laboratory, laboratory examination (comprising identification, comparison and analysis), evidential evaluation and interpretation and presentation of evidence.

This BPM is intended for experts in the field, and assumes prior knowledge in this discipline. It is not a standard operating procedure, and addresses the requirements of the judicial systems in general terms only.

This document also encompasses the requirements for systems, procedures, personnel, equipment and accommodation for the forensic process of hair examination.

3 DEFINITIONS AND TERMS

For the purposes of this BPM the relevant terms and definitions given these documents apply: ILAC G19 “*Modules in a Forensic Science Process*” [1] and standards such as ISO 9000 [2], ISO 17000 [3], ISO 17020 [4], ISO 17025 [5], and the relevant ENFSI documents.

The following terms are defined here to aid use of this BPM and are defined solely by how they are used in forensic hair examinations.

Anagen: The active growth phase of a hair follicle.

Ancestry (sometimes referred to as ethnic/racial origin): A term used to describe the appearance of hairs from people of differing ethnic or racial descent, generally grouped into three broad categories: European, African and Asian/Native American. The terms ‘Caucasian’, ‘Negroid’ and ‘Mongoloid’ may still be seen in older texts but are no longer in general use.

Bleaching: A chemical or a natural process used to make a hair colourless or lighter than its usual colour.

Buckling: An abrupt change in the shape and orientation of a hair shaft; often seen in pubic hairs.

Catagen: The transitional growth phase of the hair follicle.-

Characteristic: A feature or attribute of a hair.

Cortex: The anatomical region of a hair between the cuticle and the medulla.

Cortical Fusi: Small spaces in the hair cortex; they can be filled with air or liquid.

Cortical structure: The appearance of the cortical cells.

Cuticle: the outermost region of a hair composed of layers of overlapping scales.

Distal: The part of a hair remote from the point of attachment.

Dye: A chemical used to artificially colour hair.

Eumelanin: The dark brown to black pigment, naturally occurring in hair.

Follicle: The cavity in the skin from which hair grows.

Follicular Tag: Residual tissue from a hair follicle attached to the root.

Fusiform: Spindle-shaped (eg. cortical cells).

Hair Growth Cycle: The repeated growth (anagen), transition (catagen), and resting (telogen) phases of the hair follicle.

Keratin: The key structural protein of hair.

Macroscopic: Characteristics large enough to be perceived without magnification.

Medial: The area between the proximal and distal.

Medulla: The core of the hair shaft that is composed of air vacuoles and cells.

Melanin: A natural pigment of which two forms, eumelanin and phaeomelanin, determine the colour of hair.

Microscopic: Characteristics requiring magnification to be perceived (refer to section 4.2 for types of microscopes used in the routine examination of hairs).

Ocular Micrometer: Also referred to as an eyepiece graticule. This usually takes the form of a small clear disc on which is etched a linear scale marked in divisions of at least 0.1mm, inserted into the ocular lens.

Ovoid Bodies: Oval-shaped, heavily pigmented bodies that may be found in the hair cortex.

Peripheral Region: This is the portion of the hair that includes the cuticle, and the outer areas of the cortex most distant from the medullary or central region.

Phaeomelanin: The reddish-yellow pigment naturally occurring in hair.

Pigment Density: The relative concentration of pigment granules in the hair cortex when viewed microscopically.

Pigment Distribution: The spatial arrangement of the pigment granules in the hair shaft (such as uniform, peripheral, one-sided, variable or central).

Pigment Granules: Small particles that impart colour within the cortex of the hair.

Post-mortem Banding: An opaque microscopic band that can be observed near the root area of hairs from a decomposing body.

Practitioner: For the purposes of this BPM this term is used for any trained and competent individual undertaking any stage of human and animal hair examination, including recovery and examination, evidential evaluation and interpretation and presentation of evidence.

Proximal: The part of a hair close to the point of attachment.

Questioned Sample: Hairs of an unknown origin.

Reference Sample: A sample of hair representative of the range of colour and structure of a specific donor (for more information refer to section 8.1.3).

Root: The area of attachment of the hair to the follicle.

Root Sheath: The follicular tissue surrounding a root structure.

Shaft: The portion of the hair distal to the hair root (note that in some literature the shaft is defined as that portion of the hair external to the hair follicle).

Somatic Region / Origin: The area of the body from which a hair originates.

Telogen: The final phase of the hair growth cycle.

4 RESOURCES

4.1 Personnel

For the purposes of this BPM the term 'practitioner' is used for any trained and competent individual undertaking any stage of human and animal hair examination, including recovery and examination, evidential evaluation and interpretation and presentation of evidence.

Practitioner competence should be regularly assessed by proficiency tests and/or collaborative exercises. Organisations should maintain a record of the education and job-specific training, assessment and ongoing competency of their practitioners.

4.1.1 Recovery and Reference Sampling

For those practitioners recovering hairs at crime scenes and from items in the laboratory, and practitioners taking reference samples from individuals/animals, the following skills are necessary:

- Familiarity with relevant health and safety issues
- Awareness of contamination risks
- Awareness of other trace materials
- Familiarity with the requirements of the examining laboratory (eg. method of recovery, packaging, documentation)

4.1.2 Sample Preparation

For those practitioners preparing samples in the laboratory for further testing (eg. mounting, DNA-preparation, DAPI-dyeing) the following skills are necessary:

- Familiarity with relevant health and safety issues
- Awareness of contamination risks
- Awareness of other trace materials
- Familiarity with the relevant laboratory protocols

4.1.3 Examination and Microscopy

For those practitioners examining hair samples in the laboratory the following skills are necessary:

- Familiarity with relevant health and safety issues
- Awareness of contamination risks
- Awareness of other trace materials
- Competence in operating relevant instruments and equipment
- Knowledge of hair morphology
- Knowledge of hair examination protocols
- Competence in hair examination and comparison
- Familiarity with the relevant literature

4.1.4 Evaluation, Interpretation and Reporting

For those practitioners evaluating, interpreting and reporting the results of hair examinations the following skills are necessary:

- Competence in assessing case requirements, devising and directing examination strategies and evaluating examination outcomes.
- Competence in hair examination and comparison
- Understanding of the relevance of other trace materials
- Knowledge of contamination risks
- Knowledge of methodology and relevant instruments and equipment
- Knowledge of hair morphology
- Knowledge of hair examination protocols
- Knowledge of the relevant literature
- Competence in preparing and presenting evidence (written or verbal) for the relevant jurisdiction

4.1.5 Undertaking peer review (Refer to Section 5.5, Peer Review)

Practitioners who perform peer reviews need the skills relevant to the area under review, together with a level of knowledge or a period of experience, specified by the organisation.

Note: This BPM has been written primarily from the viewpoint of forensic laboratory personnel, but it is accepted that in some countries other individuals, including Police Officers, Crime Scene Technicians, Medico-legal Experts, and Medical Examiners, may play a role in recovery and examination of hairs.

4.2 Equipment

The key pieces of equipment used in hair examinations are microscopes. Several types of light microscopes are used including low power stereomicroscopes (incident / reflected light and transmitted light), and high magnification transmitted light, polarised light and comparison microscopes. In some situations, the fluorescence microscope and the scanning electron microscope (SEM) may also provide additional information. The nature and extent of the hair examinations required will dictate which type of microscope is used.

Stereomicroscope: A low power stereomicroscope (incident and/or transmitted light) with a range of magnifications is useful for the examination of macroscopic (gross) characteristics of both mounted and unmounted hairs. The objectives and eyepieces should permit observations in the range of approximately 10X to 100X magnification.

Transmitted Light Microscope: A transmitted light microscope is required to examine and identify the microscopical characteristics of hairs. The objectives and eyepieces should permit observations in the range of approximately 40X to 400X magnification.

Polarising Light Microscope: A polarising light microscope is required if optical characteristics need to be determined or measured, particularly in the case of artificial hairs. This type of microscope may also enhance the hair examiner's ability to visualize certain morphological characteristics and chemical treatments and to determine the cross-sectional shapes of the hairs.

Fluorescence Microscope: A fluorescence microscope is required for DAPI examination and may be useful when examining dyed and artificial hairs.

Comparison Microscope: The use of a high quality transmitted light comparison microscope is useful when comparing the microscopical characteristics of hairs. Examinations using this type of microscope have the advantage that both the questioned and reference samples are viewed under similar optical conditions, and the questioned samples can be easily compared with a wide range of hairs from the reference material. The objectives and eyepieces selected should permit observations in the range of approximately 40X to 400X magnification.

Scanning Electron Microscope: Scanning electron microscopy can be useful to assess surface detail such as scale pattern and for damage to hairs.

4.2.1 Microscope Maintenance and Performance Check

4.2.1.1 *Maintenance*

An equipment inventory should be maintained which records the manufacturer, model, serial number, the date of acquisition, the date placed in service and the location for each piece of equipment used in the examination and comparison of hairs.

The practitioner should be familiar with and follow the manufacturer's operating manual and maintenance recommendations for each piece of equipment used for the examination. These should be readily available, together with any repair and/or general maintenance documents.

4.2.1.2 Performance and Calibration Checks

Microscopes must be correctly set up as detailed in the manufacturer's instructions, and all users must be fully trained in their operation.

Light microscopes should be set for Köhler illumination before beginning work.

It is recommended that the performance of the microscope be checked against appropriate working standards every time it is used.

Only equipment that is operating properly should be employed in casework, and then only within the limits of its performance check.

Calibration of the Ocular Micrometer / Eyepiece Graticule: Any microscope used to precisely measure dimensions requires a calibrated ocular micrometer / eyepiece graticule for the ocular lens.

This 'working standard' ocular micrometer must be calibrated at regular intervals against a *certified* standard, generally comprising a stage micrometer (refer to section 4.3.2). It is recommended that this calibration is carried out at least annually.

Optical balance check (eg. colour and magnification): If a comparison microscope is used for microscopical hair comparisons, a paired set of reference slides must be used to ensure that the colour is uniformly balanced between the left and right fields of view. If the colour is balanced, the sample images and the background colour on both sides of the microscope should appear to be the same. The magnification should be checked to ensure that the left and right images are magnified to the same degree.

4.3 Reference Materials

4.3.1 Human and Animal Reference Hair Collections

Laboratories may hold reference collections of exemplar hairs from known human sources (for example, an assortment of hairs of varied ancestry and somatic origin, growth phases, etc.) and of known animal species and breeds to assist in hair identifications.

Where possible, hair collections should be authenticated and traceable. The provenance of the hair samples in such authenticated collections should be readily demonstrable. It is advisable that the laboratory puts into place appropriate control measures to ensure that the integrity of any authenticated samples is maintained.

It is recognised that many laboratory hair collections are not authenticated or traceable. Such collections should be used with caution and referred to for guidance and indication purposes only.

4.3.2 Calibration and Working Standards (Refer to 4.2.1.2)

Paired sets of reference slides: paired slides typically consist of a uniformly coloured sample of hairs (or fibres) cut in half and mounted on slides.

Stage micrometer: A slide with a linear scale of known dimensional divisions, against which the 'working standard' ocular micrometer / eyepiece graticule can be compared.

4.4 Accommodation and Environmental Conditions

Laboratories for the examination of items for hairs and for carrying out detailed hair examinations should be designed for efficient and effective working. Particular consideration should be given to the need for avoidance of contamination.

To minimise the chance of contamination, some consideration should be given to the following:

- Laboratories, equipment and sampling materials used for the examination and comparison of hairs should be cleaned thoroughly before and after use.
- Laboratories should provide adequate space for searching items. They should also allow for the physical separation of search areas to permit the processing of items from different suspects, victims and the scene.
- In situations where evidence types other than hair may be of potential significance the laboratories should provide accommodation to allow for the effective recovery of the different evidence types.

4.5 Materials and Reagents

All materials and chemicals used for the examination and comparison of human and animal hair should be of a suitable quality and demonstrated to be fit for purpose.

All chemicals and reagents, whether manufactured internally or obtained from external suppliers, should be labelled with their identity, concentration (if appropriate), date of preparation or receipt, date of opening, date of expiry and any special storage or safety requirements, if applicable, to comply with laboratory policy and/or appropriate regulations. The identity of the individual preparing reagents produced in the laboratory should also be recorded together with records of the preparation procedures.

Materials used in recovery and/or examination of human and animal hairs can include the following:

- forceps (preferable smooth, non-serrated tips)
- adhesive tape and backing
- packaging for questioned hairs
- glass microscope slides and cover slips
- mounting media and solvents
- ruler/measure marked in mm increments (this need not be calibrated as measurements are rarely critical).

Many suitable mounting media are available for preparing temporary and permanent hair mounts, and the selection of an appropriate mounting medium will be influenced by the particular microscope technique used. To examine the hairs in transmitted light, a colourless, non-yellowing mounting medium with a refractive index in the range of 1.50 to 1.60 should be used [6].

The analysis of surface particulates and biological material, compatibility with DNA analysis, and ease of artefact isolation should also be considered when selecting a mounting medium.

Additional equipment, materials and reagents will be required for specialist examination techniques, for example DAPI, alkaline hydrolysis, sectioning or scale casts (refer to section 5.1.4).

5 METHODS

Before any examinations are carried out, a case examination strategy should be defined as described in Section 9, Initial Assessment.

5.1 Examination Techniques

A hair examination is generally required to determine if an item of interest is:

- A hair
- From a human or another animal of a given species
- From certain body areas
- Characteristic of a certain ancestry
- Characteristic of a particular growth phase
- Damaged
- Diseased
- Associated with other trace evidence
- Chemically altered, such as dyed or bleached
- Suitable for microscopical comparison
- Suitable for nuclear DNA analysis
- Similar to a reference hair sample from a particular person or individual animal

5.1.1 Macroscopic and Low Power Stereomicroscopic Examination

Macroscopic and low power stereomicroscopic examinations are useful for determining whether a questioned item is a hair or other fibrous material and for observing hair characteristics, such as colour, length, shape, and texture.

This is an important step in identifying hairs, assessing which are suitable for comparison, determining the presence of other trace materials, and evaluating which hairs have roots suitable for nuclear DNA analysis.

Macroscopic and low power microscopic examination can also be used to identify or assist in the recovery of any adhering material on the surface of the hair, eg. biological material that may require recovery before further hair examination.

5.1.2 Transmitted Light Microscopy

The internal microscopic characteristics of hair can be observed easily in transmitted light when the hairs are appropriately mounted. Such an examination will determine whether the hair is human or animal, the somatic origin, ancestry, damage, and suitability for comparison.

One hair or multiple hairs from the same source may be mounted on a glass microscope slide with an appropriate cover slip. Each mounted hair must be clearly visible and distinguishable. Each slide must be uniquely labelled. Questioned and reference hairs should be mounted in the same type of mounting medium.

5.1.3 Hair Comparison

Hair comparisons should be made between questioned and reference hairs of the same somatic origin. This is usually carried out by high power microscopy, typically on a comparison microscope, however, the use of the unaided eye or a stereomicroscope may be sufficient for elimination purposes in some cases when the differences are obvious.

An adequate number of hairs that represent the range of characteristics present in the sample are selected for comparison with the questioned hair. The selection should be primarily based on macroscopic and low power microscopic characteristics, such as length, shape, and colour. The examination should comprise high power comparison of characteristics seen in the reference and questioned hairs (see Table 1, page 14). Documentation should record the characteristics seen in the questioned hair and establish whether these characteristics fall within the range observed in the reference sample.

The appearance of particular hair characteristics is usually not constant along successive portions from root to tip of a single hair, and therefore hair characteristics should be compared in corresponding areas along the length of the hairs.

Based on the outcome of the above examinations, the practitioner will evaluate the findings (refer to Section 12, Evaluation and Interpretation).

5.1.4 Other Techniques and Methods

In addition to the above methods, the following techniques can be used in the examination of hairs:

- DAPI-staining (4',6-diamidino-2-phenylindol): A fluorochrome staining treatment used for the microscopical visualisation and quantification of DNA eg. in hair roots
- Scale casts: A technique used to microscopically visualise the cuticle scale pattern
- Longitudinal and transverse sectioning: A technique used to microscopically visualise the structure of the medulla
- Alkaline hydrolysis: A destructive technique involving chemical dissolution of a hair to microscopically visualise the medullary structure of animal hairs
- Medullary fraction analysis: A microscopical technique used to distinguish between cat and dog hairs (often referred to as the medullary ratio)
- Scanning electron microscopy: A microscope / technique used to visualise surface detail such as scale pattern, and damage to hairs

5.2 Hair Characteristics and Identification

Human hair can be distinguished from other animal hair by examining characteristics such as scale pattern, medulla, root, colour, hair length, and shaft profile/form.

5.2.1 Human Hair

5.2.1.1 *Somatic Origin*

Hairs of different somatic origin may include scalp, pubic, facial, limb and body, and eyebrow and eyelash hairs, although there may be considerable overlap in appearance between these areas. Somatic origin of human hair may be established by considering characteristics such as length, cross-sectional shape, shaft profile and texture.

5.2.1.2 *Ancestry*

Characteristics such as colour, shaft profile, cross-sectional shape, pigment distribution, hair diameter and cuticle may be used to classify a hair as having characteristics typical of particular ancestry, such as European, African and Asian/Native American.

The practitioner should be alert to the possibility of mixed ancestral characteristics and atypical characteristics. Opinions about the ancestral origin of a hair should be formulated with caution.

5.2.1.3 Human Hair - Characteristics

For guidance purposes, Table 1 (see page 14) provides a list of characteristics that may be used for identification and comparison of human hairs. *The characteristics listed in this table are examples only; they are neither exhaustive nor prescriptive.*

It is recommended that all observations are documented on a standard hair examination form with the characteristics selected from a range of discrete options to ensure consistency within the laboratory.

5.2.2 Animal Hair

5.2.2.1 Species Identification

Characteristics, such as colour, shaft profile, cross-sectional shape, medulla characteristics, root form and cuticle may be used to classify the species of origin.

5.2.2.2 Types of Animal Hair

The coat of an animal consists of different types of hairs; guard hairs (coarse) and fur hairs (fine). These can be further classified as follows:

- Vibrissae: Large and stiff hairs, mostly with sensory function.
- Bristle hairs: Stout and rigid hairs of uniform diameter along their length
- Over hairs: Longer than the bulk of the fur coat, mostly with circular cross-section.
- Guard Hairs: Coarser hairs of the main fur coat, often with a shield in the distal part. Different types of guard hair can be differentiated, for example, primary or secondary guard hairs.
- Under hairs (fur hairs): Shorter and finer hairs of the pelage, usually not forming a shield.

Fur hairs do not exhibit sufficient characteristics to reliably identify the species of origin, and therefore the guard hairs are most useful when identifying the species of origin.

5.2.2.3 Animal Hair - Characteristics

For guidance purposes, Table 2 (see page 15) provides a list of characteristics that may be used for identification and comparison of animal hairs. *The characteristics listed in this table are examples only; they are neither exhaustive nor prescriptive.*

It is recommended that all observations are documented on a standard hair examination form with the characteristics selected from a range of discrete options to ensure consistency within the laboratory.

Table 1: Examples of Characteristics Used For Identification & Comparison of Human Hairs

FEATURE		Stereomicroscopy (Incident Light)	Transmitted Light Microscopy
General Characteristics	Colour - Described using colour chart, text, etc.	White, Blonde, Red, Brown, Black, etc.	Colourless (white), Blonde, Red, Brown, Black, etc.
	Cosmetic Treatment	Dye lines	Dyes, Styling products, Bleaches, Lighteners
	Shaft Form		Straight, Arced, Wavy, Curly, Twisted, Tightly coiled, Crimped
	Shaft Length (in mm / cm)		
	Shaft Thickness / Shaft Diameter	Fine, Medium, Coarse	Measured in μm
	Shaft Cross-section	Round, Oval, Triangular, Flattened	Round, Oval, Triangular, Flattened
	Shaft Form		Buckling, Convoluting, Shouldering, Undulating, Splitting, Regular
	Proximal End	Root presence / absence and Appearance	Root presence / absence and Appearance
	Distal End	Tip Appearance - Natural, Cut, Abraded	Tip appearance - Natural, Cut, Abraded, etc.
	Cuticle		Presence, Thickness, Margin features, Pigment
Cortex	Cortex Texture		Coarse, Medium, Fine
	Other Cortical Features		Ovoid bodies, Cortical fusi
	Pigmentation (in discrete granules)		Presence / Absence
	Pigment Size		Coarse, Medium, Fine
	Pigment Aggregation		Streaked, Clumped, Patchy
	Pigment Aggregate Size		Large, Medium, Small
	Pigment density		Absent, Light, Medium, Heavy, Opaque
	Pigment distribution		Uniform, Peripheral, One-sided, Random or variable, Central or medial, Pigment in cuticle, Banded
Medulla	Medulla Type	Presence & Appearance; Continuous, Discontinuous, Fragmented, etc.	Presence & Appearance; Continuous, Discontinuous, Fragmented, etc.

Other	Damage	Singeing, Crushing, Cutting, etc.	Singeing, Crushing, Cutting, etc.
	Special Characteristics	Post Mortem Banding, Lice, Mould, Fungal tunnels, Insect bite marks	Post Mortem Banding, Lice, Mould, Fungal tunnels, Insect bite marks
	Diseases		Pili annulati, Trichoschisis

Table 2: Examples of Characteristics Used For Identification & Comparison of Animal Hairs

FEATURE		Stereomicroscopy (Incident Light)	Transmitted Light Microscopy
General Characteristics	Colour - Described using colour chart, text, etc.	White, Yellow, Orange, Red, Brown, Black, Colour Banding/Shading, etc.	Colourless, Red, Brown, Black
	Hair Profile	With/without Shield, Straight, Curved, Undulating, Curly, Twisted, Zigzag	
	Hair Length (in mm)		
	Hair Diameter	Fine, Medium, Coarse	Measured in μm
	Cross-sectional Shape	Circular, Oval, Oblong, Biconcave, H-shaped, Dumb-bell shaped, Concave- Convex	Circular, Oval, Oblong, Biconcave, H-shaped, Dumb-bell shaped, Concave- Convex
	Proximal End	Root presence/absence, appearance and shape	Root presence/absence, Root shape, Appearance
	Distal End	Tip appearance - Natural, Cut, Abraded	Tip appearance - Natural, Cut, Abraded
Cuticle	Scale Orientation /Arrangement		Straight, Fringed, Transverse, Longitudinal, Intermediate
	Scale Margins		Smooth, Rippled, Scalloped, Dentate
	Scale Patterns		Broad Petal, Diamond Petal, Regular Wave, Irregular Wave
	Distance Between Scale Margins		Near, Distant, Close
Cortex	Cortex Thickness (in μm)		
	Pigmentation (in discrete granules)		Diffuse, Amorphous, Aggregate
	Pigment Distribution	Colour banding	In Cortex, In Medulla, Colour Banding
Medulla	Width (in μm)		Broad, Narrow
	Width Composition		Multicellular, Unicellular
	Structure		Ladder, Lattice, Isolated, Cloisonné, Filled, Crescent
	Margins		Scalloped, Straight, Fringed

Other	Medullary ratio: Width of Cortex + Cuticle / Total Hair (in μm)		
	Damage	Singeing, Crushing, Cutting	Singeing, Crushing, Cutting

5.3 Microscopy and DNA Analysis of Human and Animal Hairs

DNA analyses are destructive techniques and consume portions of the hair, and therefore it is important that detailed microscopic comparisons are carried out, where possible, prior to DNA analysis.

Microscopy and DNA analysis are often complementary comparison techniques. In some instances, the microscopical hair comparison may be inconclusive because the hair is a fragment, featureless, or the reference hair sample was collected a significant period of time after the questioned hair. These hairs can still be analysed for DNA. Hairs that are excluded as having come from a person by a microscopical examination may not require DNA analysis.

There will also be instances when mitochondrial DNA may not provide adequate discrimination among people; individuals of the same maternal line of descent will have the same mitochondrial DNA sequence. In these cases, a microscopical examination might provide further discrimination of the hair to associate a questioned hair to a particular person in that family group.

A combination of mitochondrial DNA and comparison microscopy will often help to exclude or provide a stronger association than the use of either technique alone.

5.3.1 Preparing Hair Evidence for DNA Analysis

A hair practitioner may need to isolate and prepare the hair for DNA analysis. The hair should be prepared and transferred in such a way as to minimize contamination and degradation. The relevant local procedures and protocols for preparing samples for DNA should be followed.

5.4 Other Analytical Techniques

Other analyses may be performed on hairs that have been chemically altered or have trace materials on the surface, such as dyed hairs or hair care products. These techniques are beyond the scope of these guidelines because they are not used routinely.

5.5 Peer Review

The outcome of a hair examination should be reviewed prior to the issuing of any report detailing the findings and/or any associated interpretation.

This process takes the form of a review of technical findings and critical findings during which a second competent practitioner:

- carries out an independent 'blind' examination of the hair, and is unaware of the results of the first practitioner
- reviews the documented results
- reviews any interpretation and opinion-based results

The laboratory should have a procedure in place to address and resolve any differences in opinion between two practitioners.

Note: Some legal jurisdictions may also have other specific witnessing requirements during the recovery or examination process.

5.5.1 Review of Technical Findings

In hair examination cases, 'technical findings' are considered those findings not related to opinion, such as, the observation and documentation of any macroscopic and microscopic characteristics of the questioned and/or reference hairs. These may also include the results of other analytical tests or findings which are incapable of being confirmed at a later date (eg. where destructive tests are required).

5.5.2 Review of Critical Findings

In hair examination cases, 'critical findings' are considered those findings which are based on interpretation or opinion. For example:

- The location of questioned hairs before recovery, where significant evidential weight will be attached to where the hairs were recovered, for example, hairs adhering to a weapon
- The identification of damage to hair and interpretation of the cause of the damage
- The results of microscopical hair comparisons.

6 VALIDATION AND ESTIMATION OF UNCERTAINTY OF MEASUREMENT

6.1 Validation

For the general requirements of validation, the reader is referred to the ENFSI document "*Guidelines for the Single Laboratory Validation of Instrumental and Human Based Methods in Forensic Science*", and in particular, *Section 4, Human Based Methods* [7, 8].

Validation should focus on the process of identification and comparison of hairs and does not extend to evaluating the significance of hair comparison results.

Hair **identification** is based upon well-established scientific principles supported by scientific literature extending back over a hundred years. Hair identification is soundly based on scientific peer-reviewed methodology in anatomy, physiology and taxonomy.

Forensic **comparisons** of hairs have been used and scientifically accepted worldwide for many decades. The literature dealing with hair characteristics and the reliability of forensic hair comparison is extensive. The quality of hair comparisons depends on the judgment and experience of the practitioner, which may be achieved by scientific education, training and continued professional development.

Hair identification and comparison are human-based methods of examination. Therefore, validation and verification should comprise practitioners demonstrating competence by showing that they can provide consistent, reproducible and valid results that are compatible with the results of other competent practitioners.

6.2 Estimation of Uncertainty of Measurement

Uncertainties of measurement in hair examinations can firstly relate to those arising from the use of instruments, equipment and reagents. Secondly, as a human-based method, practitioner competence forms the basis of reliable and reproducible results. In addition, hair examinations are often reliant on adequate sampling and the nature and variation of hair samples.

6.2.1 Instrumentation/equipment/reagents (Refer to Section 4.2 and 4.5)

Uncertainties of measurement attributable to microscopy can be minimised by servicing and maintenance of the equipment according to the manufacturer's specifications. In addition, regular performance and calibration checks should be performed.

Uncertainties of measurements attributable to reagents (eg. mounting media, fluorescent dyes) can be minimised by the selection of reagents of appropriate quality and/or refractive index. Failure to carry out these steps can have an impact on the practitioner's ability to visualise and recognise key characteristics / features.

Macroscopic measurements, such as hair length, are rarely critical, and therefore any uncertainty of measurement is unlikely to be of significance.

6.2.2 Practitioner competence

As a human-based method, the reliability and reproducibility of results of hair examinations are highly reliant upon practitioner competence. Well-trained and competent practitioners participating in regular proficiency testing pose minimal risk of error and uncertainty.

Practitioner competence extends to reporting, where it has been observed by some that the use of imprecise reporting of hair comparisons has, in the past, caused significant issues [9]. This is discussed further in Section 12, Evaluation and Interpretation.

6.2.3 Hair samples

Each of the following has an impact on the ability of the practitioner to attribute a hair to a source with any degree of certainty:

- Non representative reference sampling
- Highly variable hair samples
- Hair samples with limited or non-distinguishable characteristics / features

7 PROFICIENCY TESTING

Proficiency tests (PTs) supported by collaborative exercises (CEs) should be used to test and assure the quality of microscopical identification and comparison of human and animal hair.

Proficiency tests and collaborative exercises could form a part of the maintenance of a practitioner's competence. A list of currently available PT/CE schemes as composed by the QCC is available at the ENFSI Secretariat.

"Guidance on the Conduct of Proficiency Tests and Collaborative Exercises within ENFSI" [10] provides information for the ENFSI Expert Working Groups (EWGs) on how to organise effective proficiency tests and collaborative exercises for their members.

Proficiency tests in hair examination are regularly available from commercial organisations (eg. FTS in the USA) and are set biennially by the ENFSI Expert Working Group (ETHG).

Such tests typically involve:

- Distinguishing human from animal hairs
- Identifying somatic origin of human hairs
- Identifying the suitability for nuclear DNA analysis
- Comparison of human and animal hairs
- Identification of animal species

In addition, these may be supplemented by tests provided by collaborations of laboratories or within an organisation.

The performance in proficiency testing and collaborative exercises must be reviewed by the organisations in a timely manner and any anomalous results investigated and addressed with appropriate corrective actions.

8 HANDLING ITEMS

Hair examinations will be driven by the requirement of the client and the overall forensic examination strategy. Personnel should be aware that various types of evidence will be present during the processing of a crime scene or the examination of items submitted to the laboratory. Some types of evidence may be more significant to a particular case and therefore should be given higher priority. The situation has to be considered carefully before any examinations take place. All potential forensic opportunities should be considered and the appropriate experts should be consulted.

8.1 At the Crime Scene

The following recommendations apply to the examination of hairs relating to a crime scene, as well as hairs relating to victim(s) and suspect(s).

8.1.1 Avoidance of Contamination and Prevention of Loss

The accessibility to crime scenes should be restricted and any access should be documented.

Items from different sources must be physically separated from one another after seizure to reduce the potential for contamination, eg. clothing and other articles from the scene, suspect(s) and of the victim(s) must be kept separate from one another.

Any situation that could cause contamination or otherwise compromise the trace evidence examinations should be documented and communicated.

In order to protect hair traces or their substrates from loss and/or biological, chemical, or physical contaminants, personal protective equipment (PPE) should be worn by the practitioner at the scene. The choice of PPE will be determined by the aims of the examiner.

The area for examination (crime scene) may require protection against environmental conditions, eg. tents to protect against adverse weather.

Items at a crime scene which bear visible but easily lost traces, or items that are impractical to transport, should be appropriately documented, and the traces collected by an appropriate technique.

To prevent the loss of evidence, consideration should be given to the recovery of hairs at the scene by an appropriate method (refer to section 8.1.2). Alternatively, items can be removed from the scene and hairs recovered later in the laboratory. If this is the case, these items should be packaged immediately in an appropriate manner (refer to section 8.1.4).

Steps should be taken to avoid post-incident contamination between the scene, victim(s) and suspect(s), eg. by the use of different personnel or PPE.

8.1.2 Search and Recovery

8.1.2.1 *Searching*

Some hairs are visible to the naked eye under adequate illumination and can therefore be located, recovered and secured for further examination. Supplementary lighting or a magnification glass might be advantageous.

8.1.2.2 *Manual Recovery*

Generally, manual recovery of hairs is the preferred method for recovering hairs and should take place before any other examinations of that item.

Loose hairs visible to the unaided eye may be collected from an object by picking them off individually by gloved hand or with clean forceps. Hairs that are embedded in or adhering to a substrate must be carefully inspected before removal. The location of these hairs should be documented. Care must be taken not to contaminate, alter or damage the hairs.

Manual recovery of hairs does not generally affect other evidence types but may be time consuming.

8.1.2.3 *Adhesive Tape Lifting*

Hairs not visible to the naked eye may also be collected from items using adhesive tape lifts which are applied to a surface. Loosely adhering trace evidence will stick to the tape and can easily be preserved by being placed on a backing such as an acetate or polyethylene sheet, or by folding the tape against itself. Care should be taken not to overload tapes. Consider the use of additional tape to seal the edges of insecure tapes.

Be aware that the adhesive from the lifting material could interfere with the analysis of surface treatments that might be present on the hairs.

Sequential taping is the standard tape lifting method used. If additional information about the distribution of hairs on an item is required, then zonal or 1:1 taping may be more appropriate.

Although manual recovery of hairs is preferable, tape lifting is acceptable but may alter or destroy other evidence types and can also be problematic in adverse conditions, such as wet or extremely dusty surfaces.

8.1.2.4 *Combing*

When recovering transferred hairs (and other trace materials) from a person's head or pubic region, the combing technique or taping can be used. When combing, always use a new comb or brush. Lacing the teeth of a comb with clean cotton or gauze may help to retain hairs and debris on the comb. Place a piece of clean paper under the area that is combed to catch loose hairs and debris; this paper should be included in the evidence package with the comb.

The use of fine-toothed combs (eg. lice combs) is not recommended.

Transferred hairs and other trace material should always be recovered before taking reference samples from a given individual.

8.1.2.5 *Other Recovery Methods*

Hairs can also be collected from an item by brushing or vacuuming. Brushes must be new and the appropriate vacuum filter, trap and other relevant parts must be changed and/or rigorously

cleaned between each vacuuming to avoid contamination. These methods recover all types of loose debris which still requires separation and the original position of any evidence will be unknown.

8.1.3 Reference Sampling

Reference samples should be requested from all persons or animals who might reasonably be considered a source of a questioned hair.

Reference hairs should be sampled from specific somatic regions for comparisons to questioned hairs. Every effort should be made to ensure that these hairs are sampled as soon as possible after the incident.

Sampling should be carried out in the following sequence:

1. Combing to recover telogen hairs; recovered material may include both transferred hairs and other trace material.
2. Pulling and cutting of the reference samples.

Different hairs from the same body region of a person exhibit variation in microscopical characteristics and features. Therefore, it is important to obtain a sufficient number of hairs in order to adequately represent the range of characteristics present. If the range is large, it becomes necessary to obtain a larger number of hairs.

- A reference human head hair sample should consist of hairs from the five different areas of the scalp (top, front, back including nape, and both sides). Ideally, a minimum of 20 hairs *per region* should be taken from the scalp (i.e. a minimum of 100 hairs in total).
- A reference human pubic hair sample or a sample from any other somatic region should ideally consist of a minimum of 20 hairs.
- A reference animal hair sample should ideally represent the range of colour and hair types (refer to Table 2, page 15) present at the different body regions (head, back, tail, belly, limbs) of the animal. If possible, a photograph of the animal of interest should be provided.

Notes:

- A comparison can still be performed with fewer than the recommended number of hairs, but this may increase the likelihood of a false exclusion.
- Whilst pulled hair samples are preferable for microscopical comparison, it is recognised that such samples may be impractical to obtain.
- Combed hair samples must be packaged and labelled separately. Additionally, it is desirable for hairs from each sampled region (eg. from the head regions) to be packaged and labelled individually.

8.1.4 Preservation and Packaging

The method of preservation and packaging of hairs and items to be examined for hairs will vary. Packaging must prevent loss, deterioration, or contamination of the hairs.

All evidence packages must be properly sealed in a manner to prevent tampering and eliminate loss or contamination of the hairs through open edges.

Loose hairs and tape lifts must be secured in clean, unused containers or packaging. Large items, such as whole garments, should preferably be sealed individually in clean, unused packaging.

Wet recovered items bearing hairs or wet hair samples should be air dried as soon as possible, without exposure to heat or sunlight, in a secured area in a manner that will prevent loss or contamination of the hairs. Alternatively, wet hair samples may be frozen. An arrangement to collect hairs and any other trace evidence that may fall from the item during drying should be used.

Small or manageable items at a crime scene that bear visible, firmly attached hairs should be documented, packaged intact, and transported to the laboratory for examination. Items at a crime scene that bear visible but easily lost hairs, or items that are impractical to transport, should be documented and the hairs collected using an appropriate technique.

8.1.5 Labelling and Documentation

The techniques used for detection, collection, and preservation of the items should be recorded together with the location from which they are removed.

The labelling of any items recovered should follow the local jurisdiction guidelines.

Documentation of the handling and examination of items and traces at the scene should include:

- The location of the scene, the name of the victim/suspect
- The date (and time, when appropriate) the item or trace was recovered
- The name of the person recovering the item or trace
- A short description of the recovered item or trace
- A unique identifying mark for each item or trace, e.g. case number and item number
- The position of relevant items (e.g. documented by notes, sketches, measurements and/or photographs)

8.1.6 Transport

There are no particular problems or risks associated with the transportation of hair samples provided that they have been packaged appropriately so as to avoid any risk of contamination or tampering.

8.1.7 Storage

Any items recovered from the scene, a victim(s) or a suspect(s) should be stored in a manner which prevents loss, deterioration, or contamination, eg. loose hairs and tape lifts must be secured in clean, unused containers or packaging. Ideally hairs should be stored in a dry and dark environment.

8.2 In the Laboratory

8.2.1 Anti-contamination Procedures

The accessibility to examination areas should be controlled.

The recovery of hairs from different sources (scenes, victims, suspects) must be undertaken separately from one another to reduce the potential for contamination. Once the hairs are recovered, secured and labelled the risk of inter-sample contamination is minimised. To demonstrate the integrity of the examination, notes should include details of item packaging and the examination undertaken, together with the examination date, time and location.

Any situation that could cause contamination or otherwise compromise the trace evidence examination should be documented and communicated.

In order to protect hair traces or their substrates from loss and/or biological, chemical, or physical contaminants, PPE should be worn by the practitioner. The choice of PPE will be determined by the examination strategy.

It is desirable to clean any packaging of exhibits immediately prior to opening to prevent any contamination.

Laboratories, examination areas and equipment should be cleaned according to laboratory anti-contamination protocols before and after examinations are carried out.

8.2.2 Search and Recovery

8.2.2.1 *Searching*

Some hairs are visible to the naked eye under adequate illumination and can therefore be located, recovered and secured for further examination. Supplementary lighting and the use of microscopes might be advantageous.

8.2.2.2 *Manual recovery*

Generally, manual recovery of hairs is the preferred method for recovering hairs and should take place before any other examinations of that item.

Loose hairs visible to the unaided eye may be collected from an object by picking them off individually by gloved hand or with clean forceps. Hairs that are embedded in or adhering to a substrate must be carefully inspected before removal. The location of these hairs should be documented. Care must be taken not to contaminate, alter or damage the hairs.

Manual recovery of hairs does not generally affect other evidence types but may be time consuming.

8.2.2.3 *Adhesive Tape Lifting*

Hairs not visible to the naked eye may also be collected from items using adhesive tape lifts which are applied to a surface. Loosely adhering trace evidence will stick to the tape and can easily be preserved by being placed on a backing such as an acetate or polyethylene sheet, or by folding the tape against itself. Care should be taken not to overload tapes. Consider the use of additional tape to seal the edges of insecure tapes.

Be aware that the adhesive from the lifting material could interfere with the analysis of surface treatments that might be present on the hairs.

Sequential tape lifting is the standard tape lifting method used. If additional information about the distribution of hairs on an item is required, then zonal or 1:1 taping may be more appropriate.

Although manual recovery of hairs is preferable, tape lifting is acceptable but may alter or destroy other evidence types and can also be problematic in adverse conditions, such as wet or extremely dusty surfaces.

8.2.2.4 *Other Recovery Methods*

Hairs can also be collected from an item by brushing or vacuuming. Brushes must be new and the appropriate vacuum filter, trap and other relevant parts must be changed and/or rigorously cleaned between each vacuuming to avoid contamination. These methods recover all types

of loose debris which still requires separation and the original position of any evidence will be unknown.

8.2.3 General Considerations

If the hair samples require washing or cleaning, then the potential significance of adhering material should be assessed and documented before this is undertaken.

The presence of a small amount of blood or debris on a hair may not interfere with the microscopical examination. A washed hair should be allowed to air dry prior to mounting.

Hair exhibiting thermal or mechanical damage may be more brittle and should be handled minimally and with more care.

8.2.4 Storage Conditions

Hairs must be stored in a manner which prevents loss, deterioration, or contamination, eg. loose hairs and tape lifts must be secured in clean, unused containers or packaging. Ideally hairs should be stored in a dry and dark environment.

A record of movement including storage should be held.

9 INITIAL ASSESSMENT

The significance and evidential value of hair evidence relies upon the correct detection, collection and preservation of this evidence and the appropriate sequence of examinations. Furthermore, the success of hair examinations and interpreting the evidential value of results is strongly dependent on the initial case assessment and forensic examination strategy, devised by the relevant personnel involved.

Prior to starting any examinations, the requirements of the client must be clearly defined and documented. Detailed information about the circumstances of the case is usually necessary to assess any potential limitations in the examinations proposed and their effect on the perceived outcome.

Factors to be considered when establishing the client's requirement include:

- What area(s) and level of expertise is required
- Whether the laboratory has the capacity, suitable resources, facilities and equipment available to perform the necessary examinations
- Whether there is any time or other constraints that might affect the overall examination strategy, for example, where other evidence types are involved in the examination sequence

In relation to hair evidence specifically, this may require consideration of the potential for the transfer and retention of hair based on the information available. This includes:

- What is suspected or known to have occurred before, during and after the incident
- The persons (or animals) involved
- The sequence of events and the time frames involved, including those associated with the recovery of items submitted for examination

Before examination in the laboratory commences, the potential risk of contamination prior to laboratory submission should be assessed by establishing whether:

- There was any opportunity for the transfer of hairs between the individuals, items and/or surfaces involved prior to the incident
- There was an opportunity for hair transfer between the individual(s), items and/or surfaces involved following the incident
- The items relating to the individual(s), and/or items involved were properly handled during recovery and packaged appropriately
- There was any opportunity for secondary transfer to individuals, items and/or surfaces involved, such as contact with other individuals and/or seating.

In instances where more than one scientific discipline is involved and/or different evidence types need to be considered, a coordinated examination strategy will determine the actions to be taken and the most appropriate methods to be used for the search and recovery of hairs.

10 PRIORITISATION AND SEQUENCE OF EXAMINATIONS

The prioritisation and sequence of examinations is determined by the overall forensic examination strategy (refer to Section 9, Initial Assessment).

Consideration should be given to the following before commencing any recovery and examinations of hair:

- The urgency and priority of the client's need for information
- Other types of forensic evidence, and the associated examinations which may have to be carried out on the same items
- Which items have the potential to provide the most evidential information
- To minimise the possibilities of contamination it is preferable to examine all items relating to one individual or scene before commencing with items relating to others.

In general the examination protocol might be as follows:

- Recovery of visible material, such as hairs, glass, etc., adhering loosely to the item
- Recovery of any potential material not visible to the naked eye. In some instances a low power microscopical search of the item may be relevant. This method decreases any potential contamination and loss of unforeseen evidence
- Any dry testing (eg. testing for the presence of blood)
- Recovery of material for further testing (eg. DNA sampling)
- Any wet testing (eg. tests for the presence of semen and recovery of such material)
- Any comparison testing (eg. hair or fibre comparison)
- Any testing of recovered material (eg. body fluids, cosmetic products)
- Destructive testing (eg. DNA analysis)

During the examination there may be a conflict of interest, and there may be occasions where one test could prevent others being attempted or where the ability to obtain a result is compromised. In such instances the practitioner should consider:

- What is the chance of obtaining informative results from the different tests?
- What would be the probative values of the results?
- Which test is most likely to answer the client's questions?

Such considerations should be documented and reviewed by another competent practitioner.

11 RECONSTRUCTION OF EVENTS

In some cases it might be necessary to carry out experimentation to evaluate the findings. This

might take the form of a simulation or reconstruction exercise designed to mimic the proposed causative actions. For example, tests to replicate heat-damaged or crush-damaged hairs.

Such tests should be designed so as to answer the issues raised during the examination process, or by the client. Any experiments or tests carried out should be well documented, including any assumptions made and limitations of the tests. Before carrying out destructive testing in reconstruction experiments it is advisable to consider the use of replicate samples. Where replicates are used (eg. weapons) they should closely reflect the form and condition of the original item.

When carrying out any experimentation, it is preferable not to alter the original evidence (depending on the legal jurisdiction). Where it is altered by experimentation, this has to be clearly documented and disclosed to the Court.

12 EVALUATION AND INTERPRETATION

Evaluation and interpretation of the findings should reflect the issues identified in the initial assessment and should answer the specific requirements of the client (refer to Section 9, Initial Assessment).

Knowledge and understanding of the dynamics of the transfer and persistence of hair, and the techniques available for the detection, collection, examination and analysis of hair evidence play an important role in the evaluation and interpretation of hair evidence.

12.1 Hair Identification (species; ancestry and somatic origin):

The identification of both human and animal hair depends on the presence of key characteristics (eg. scale pattern, medullary structure, root appearance). Where those characteristics are absent, the confidence in identification is lowered (eg. small hair fragments). Comparison against laboratory reference collections can assist and give greater confidence in the identification of an unknown sample.

There is a reduced degree of certainty in the identification where there is an overlap in characteristics between different hair types, for example ascribing a hair to a specific human ancestral origin or somatic type (eg. mixed ancestry and transition areas on the body) and ascribing an animal hair to a species (eg. hare and rabbit hair).

12.2 Hair Comparison (hair colour and characteristics):

Because no two hairs are exactly the same in every detail (identical), it must be determined what range of characteristics is represented within the reference hair sample. If the characteristics exhibited by the questioned hair(s) fit within the range of characteristics present within the reference hair sample then it is possible to conclude that the hairs could share a common origin.

Microscopical examination of hair does not lead to the identification of the donor to the exclusion of all others. Therefore, having established that the questioned hair possesses characteristics that fall within the range of characteristics seen in the reference hair sample, to evaluate the significance of this finding the practitioner must attempt to assess how common those shared characteristics might be. In Bayesian terms, this is assessing the likelihood of finding a hair with these characteristics if it did *not* come from the same source as the reference sample. The presence of some types of hair characteristics may increase the confidence that a questioned

and a reference hair sample have a common source, for example the presence of similar dyes or hair abnormalities. The confidence that the two hair samples have a common source may be reduced, for example when the number of characteristics that can be used for comparison is limited.

Over the years there has been much conflicting debate as to how the evidential value of hair comparisons should be evaluated [for example, 11, 12, 13, 14, 15]. There is no association between the number of characteristics shared by the questioned and reference hairs and the probability that the samples have a common origin. In addition, no scientifically accepted statistics exist regarding the frequency with which particular morphological characteristics occur in the population [9].

Although some laboratories have built up internal reference collections to help assess the frequency of some characteristics, the assessment of the strength of the findings still primarily relies on the scientist's experience.

Some US studies have highlighted that on occasion hairs apparently 'associated' by microscopic comparisons have later been shown by mitochondrial DNA analysis to have originated from different sources [9]. This is not unexpected given the relative discriminating powers of the two techniques. The problem here lies in imprecise reporting terminology, where an unqualified 'associated with' or 'could be from' can suggest individualisation. This can then cause concern when the results of subsequent DNA analysis of the hair apparently contradict the microscopy findings.

The current view is that microscopy and mitochondrial DNA analysis are complementary, with the two methods combined providing an additional level of information that provides greater accuracy than either method alone [16], although to date there have been no studies to quantify the reliability of their joint use [9].

Where the characteristics exhibited by the questioned hair(s) do not fall within the range of those in the reference hair sample, the questioned hairs may be excluded as having come from the same source (assuming that the reference hair sample is truly representative).

On occasion, the results of a hair comparison can be termed inconclusive where the examination cannot establish whether the hairs share a common origin or not. For example, where hairs do not exhibit sufficient distinguishing characteristics or the number of hairs in the reference sample is inadequate.

12.3 Cognitive Bias

Any hair comparison inevitably involves an element of subjectivity and is therefore prone to cognitive bias; this can take various forms. 'Confirmation bias' occurs when the practitioner sees what they expect to see, and this can have a particular effect where the findings are relatively ambiguous. Outside information, such as case notes, police opinions, awareness of any particularly emotive aspects of a case ('contextual bias'), or knowledge of the opinion of any earlier examination by a colleague, can all influence the interpretation. The effects of such cognitive bias are now widely recognised [for example, 17, 18, 19], and although procedures have been suggested to minimise cognitive influences in hair comparison [20], these have not been widely adopted. The effects of cognitive influences can be minimised by assessing specific characteristics from a limited set of options available on a hair examination form. In addition, the use of independent peer review of hair comparisons can, to some extent, minimise the effects of cognitive influences.

12.4 The Significance of Transfer and Persistence

The opportunity for hair transfer will depend primarily on the nature and extent of the contact between the individual (or animal) and the items or surfaces involved.

Transferred hairs can be lost or redistributed after the initial transfer. The rate of loss will be influenced by factors such as:

- the nature of the recipient item
- the environmental conditions to which that item is subjected
- the physical attributes of the hair
- post-transfer activity

Where the method of transfer (eg. primary or secondary transfer) and the time of occurrence is an issue, published literature can provide some guidance. In such instances, assessment at the initial examination stage should formulate likely outcomes and expectations against which the results of the examination can be evaluated.

For general information relating to evaluative reporting, the reader is referred to the ENFSI document "*Guideline for Evaluative Reporting in Forensic Science*" [21].

13 PRESENTATION OF EVIDENCE

The overriding duty of those providing expert testimony is to the Court and to the administration of justice. As such, evidence should be provided with honesty, integrity, objectivity and impartiality.

Evidence can be presented to the court either orally or in writing. Only information which is supported by the examinations carried out should be presented. The presentation of evidence should clearly state the results of any evaluation and interpretation of the examination.

After issuing their report, the practitioner may review and alter their opinion based on new information given to them.

13.1 Presentation of Written Evidence

The practitioner's findings and opinion are normally provided in written form, as a statement of evidence or a report. These can be used by the investigator, the prosecutor, the defence or the court. Depending on the client's requirements, either a shortened or a complete report can be issued.

Written reports should include all the relevant information in a clear, concise, structured and unambiguous manner, as required by the relevant legal process for the country of jurisdiction, as well as the guidelines of the institute. Reports should clearly state the results of any evaluation and interpretation of the examination, and written reports must be peer reviewed.

13.2 Presentation of Oral Evidence

At Court, the practitioner should only respond to matters arising from their report, or those matters raised in Court, which fall within their area of expertise. Expert witnesses should resist responding to questions that will take them outside their field of expertise, unless specifically directed by the court, and even then a declaration as to the limitations of their expertise should be made.

14 HEALTH AND SAFETY

The relevant national health and safety regulations must be complied with.

14.1 At the Crime Scene

There are no specific hazards associated with the recovery of hairs at the crime scene.

Generic hazards that apply to any crime scene, for example risks associated with exposure to body fluids, trip hazards, etc., will always need to be considered and may be documented as part of the formal risk assessment process prior to scene entry. Control measures should be put in place to either remove the risk or minimise it to an acceptable safe working level.

No specific protective clothing is necessary for hair recovery at crime scenes, and the usual anti-contamination clothing (including head hair protective covering) is adequate.

14.2 In the Laboratory

Generic hazards that apply to any laboratory examination, for example risks associated with exposure to body fluids, etc., will always need to be considered. Control measures should be put in place to either remove the risk or minimise it to an acceptable safe working level.

No specific protective clothing is necessary for hair recovery or examination at the laboratory, and the usual laboratory anti-contamination clothing (as dictated by local laboratory procedure) is adequate.

Risks in the examination process at the laboratory include:

- Microscopic examination and comparison of hairs can be a lengthy process, during which time the operator may be seated for prolonged periods at the microscope. There is a risk of eye strain and postural discomfort, which should be alleviated by frequent breaks from the examination process.

The laboratory may consider the need for a documented risk assessment of the workplace environment, to ensure that laboratory lighting is adequate and seating position is optimised, with seating providing the appropriate lumbar support and set at the appropriate height relative to the laboratory bench and microscope etc.

- Exposure to chemicals can also be an issue. Solvents may be used by some laboratories to remove hairs from tape lifts, whilst some laboratories may utilise solvent-based mountants for mounting hairs on microscope slides. Other chemicals, for example DAPI, or those used for scale casting, may on occasion be used during the examination process. The risks associated with the various chemicals, solvents and mountants used will vary.

The laboratory should undertake and document a risk assessment of the hazards associated with the use of any chemicals, solvents or mountants, in a specific activity and identify the precautions to be taken during their use in that activity to mitigate the risks. Some chemicals, solvents and mountants may have to be used in a fume hood or while wearing a suitable protective mask and safety spectacles. In some instances certain personnel should avoid any exposure to the particular chemical. For example, xylene-based mountants may pose a risk to unborn children and exposure of pregnant female staff to these chemicals must be avoided.

14.3 At Court

There are no specific safety risks associated with hair evidence where the materials may be brought into the public domain, such as Courts.

However, if the hair is known to present a biohazard, for example, it is contaminated with lice or body fluids, it should be appropriately packaged and clearly labelled to indicate the biohazard risk.

14.4 Other

Sampling reference samples may have associated risks, particularly as the sampling will usually require the use of sharp scissors or similar. For example, sampling hair from an uncooperative human suspect or from an animal poses the risk of sharps injury to the sampler and subsequent risk associated with potential exposure to body fluids. Sampling animal reference hair samples may also have health risks specific to the animal concerned, for example sampling hair from dangerous undomesticated animals or animals posing a biohazard risk to humans.

In such instances a risk assessment specific to the risks associated with the activity should be documented, identifying the risks and the precautions to be taken to mitigate the risks.

15 REFERENCES

- 1 ILAC/G19: 08/2014, Modules in a Forensic Science Process.
- 2 EN ISO/IEC 9000:2005 Quality Management Systems - Fundamentals and Vocabulary.
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16 AMENDMENTS AGAINST PREVIOUS VERSION

Not applicable.

APPENDIX - BIBLIOGRAPHY FOR HAIR PRACTITIONERS

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