

European Guidelines for Workplace Drug and Alcohol Testing in Hair

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Abstract

These guidelines for Legally Defensible Workplace Drug Testing have been prepared and updated by the European Workplace Drug Testing Society (EWDTS). They are based on the 2015 version published by Alberto Salomone, Lolita Tsanaclis, Ronald Agius, Pascal Kintz and Markus Baumgartner (European guidelines for workplace drug and alcohol testing in hair. *Drug Test Anal.* 2016; 8(10):996-1004) and in concordance with the Society of Hair Testing consensus for drugs.

(https://www.soht.org/images/pdf/Consensus_DoA_2021.pdf) and alcohol markers (https://www.soht.org/images/pdf/Revision_2019_Alcoholmarkers.pdf). The European Guidelines are designed to establish best practice procedures whilst allowing individual countries to operate within the requirements of national customs and legislation. The EWDTS recommends that all European laboratories that undertake legally defensible workplace drug testing should use these guidelines as a template for accreditation.

1. General

1.1 Objectives

- To provide a common framework for European providers of workplace drug testing services in Europe within which Workplace Drug Testing (WDT) in hair should be performed.
- To provide guidance to European laboratories involved in the determination of drugs of abuse in hair, in order to provide reliable results for the purpose of WDT.
- To promote and harmonise best practises by providing guidelines which are accepted at the European level.
- To ensure that the processes undertaken are capable of legal scrutiny.
- To provide safeguards to protect the specimen donors.
- To define common quality assurance and quality control criteria for laboratories that are capable of being accredited by an external body.

2. Definitions

For purposes of these guidelines the following definitions have been adopted:

Adulteration	See Tampering
Aliquot	A fractional part of a hair sample used for testing. It is taken as a sample representing the whole sample.
Authorising Scientist	A person who reviews all pertinent data and quality control results to attest to the validity of the laboratory's test reports.
Calibrator	A solution of known concentration used to calibrate a measurement procedure or to compare the response obtained with the response of a test sample/sample. The concentration of the analyte of interest in the calibrator is known within limits ascertained during its preparation. Calibrators may be used to establish a calibration curve over a concentration range of interest.
Chain of Custody	Procedures to account for each specimen by tracking its handling and storage from point of collection to final disposal. These procedures require that the donor identity is confirmed and that a chain of custody form is used from time of collection to receipt by the laboratory. Within

	the laboratory appropriate chain of custody records must account for the samples until disposal.
Chain of Custody Form	A form used to document the procedures from time of collection until receipt by the laboratory.
Collecting officer	A person trained to collect hair specimens from donors.
Collection Site	A place where individuals present themselves for the purpose of providing a hair specimen for analysis.
Confirmation Test	An analytical procedure, performed in addition or in replacement of a screen test (if used), which is able to identify and quantify the presence of a specific drug or metabolite present in the hair sample. Where a screen test has also been performed, the confirmation test should use a different technique or chemical principle.
Customer	The organisation requesting the drug testing service.
Cut-off	A concentration level set to determine whether the donor of the sample is a regular or occasional user.
Donor	The individual from whom a hair specimen is collected.
Laboratory	The facility providing the analytical services to detect drugs and related compounds.
Negative result	A result reported by the laboratory that indicates that either a compound is not present in the sample or that is present below the limit of detection or the cut-off concentration.
Positive result	A result reported by the laboratory showing that a compound is present in the sample tested at a level greater than or equal to the limit of detection or the cut-off concentration.
Quality control sample	A sample used to evaluate whether an analytical procedure is operating within predefined tolerance limits and under control.

Medical Review Officer (MRO)	A medical physician responsible for receiving laboratory results from the drug-testing laboratory who has knowledge of substance abuse and has appropriate training or experience to interpret and evaluate an individual's positive test result, in light of declared information.
Sample	A representative portion of a specimen submitted to a laboratory for testing.
Screening Test	A testing procedure able to eliminate negative samples from further consideration and to identify the presumptive positive samples that require confirmation testing.
Service Provider	The organisation contracted to provide the drug testing service. This may be a laboratory, or a third party providing other elements of the service, and sub-contracting the tests to another laboratory.
Specimen	The portion of hair that is collected from a donor.
Standard (1)	A reference material of known purity or a solution containing a reference material at a known concentration.
Standard (2)	An agreed protocol or procedure (e.g. EN ISO/IEC 17025 and EN ISO 15189)
Standard Operating Procedure (SOP)	A written document giving the detailed steps to be followed when undertaking a particular task (e.g. the analysis of a given drug in a hair sample).
Tampering	Any process by which an individual knowingly interferes with (or attempts to interfere with) the processes of specimen collection, transport, or analysis with the intention of avoiding a legitimate test result.
Toxicologist	A person responsible for interpreting a toxicological analytical result for the customer or the customer's designated Medical Review Officer.

3. Scope of hair testing

The five common samples widely accepted for the detection and monitoring of drug use are blood, sweat, urine, saliva and hair. They have different characteristics that mean

they have different applications and as all have inherent and differing limitations in measuring the timing, duration, frequency, and intensity of drug use. The specimen of choice depends on the context and requirements of the testing. In a workplace setting, the choice of the sample to use for drug testing depends on the purpose of the testing. Drug testing using either urine or oral fluid reflects drug use for a relatively short period before sample collection. Hair as a sample for drug testing allows a long-term historical window of the subject's drug intake, based on hair's length.

Laboratories performing hair analysis should ideally be accredited to ISO/IEC 17025 standards with consideration of the guidelines of the SoHT (Society of Hair Testing) in view of each country's legislative requirements. Samples and records need to be stored securely within the laboratory. All analytical work needs to be fully traceable, usually by a laboratory information management system (LIMS) using a unique code, and preferably all transfers and input tracked by barcode (or suitable alternative) for each stage of analysis. In this way the progress and status of any sample can be identified at any time. The reports will include the sample's unique number as stated on the chain of custody, the specific substance(s) tested and substance grouping, together with whether any substances have been detected, together with the level(s) detected.

4. Specimen Collection

4.1 Introduction

The collection of donor specimens involves some of the most difficult and sensitive areas of the WDT process. It must ensure the integrity of the entire process. The collector must be very sensitive to each individual's privacy and respect the dignity of the donor while at the same time ensuring that the sample is accurately collected and has not been tampered with in any way.

To balance between the privacy of the donor and the need to ensure the proper identification and integrity of the specimen, the following steps must be documented:

- The verification of the identity of the donor e.g. by valid and unexpired ID card. It may be lawful in certain countries to take a photograph of the donor if necessary to evidence identification.
- The proper identification of the specimen with its donor.
- Ensuring that no adulteration or tampering took place.
- Ensuring that no unauthorised access to the specimen was possible.
- The secure transfer of the specimen to each person handling it.

The documentation of the collection process is the first link in what is referred to as the chain-of-custody process. This process enables a data trail that, when reconstructed at a later date, can be used to prove that the final result properly matches the sample to the donor.

The procedures for collection of hair specimens for WDT are very specific. It is essential for each collection site to have written standard operating procedures (SOPs) and for collectors to comply with those procedures, in order to minimise the possibility of procedural or administrative errors.

A collector is a trained individual who instructs and assists the donor at a collection site, who receives and makes an initial inspection of the specimen provided by the donor, and who initiates and completes appropriate sections of the Chain of Custody Form (CCF).

The specimen collection kit is usually provided by the testing laboratory.

The collector should conduct only one collection at a time, to prevent specimen misidentification and avoid distraction that could compromise specimen security. The collector must guarantee the secure handling and storage of the specimen from the time the specimen is received from the donor until the specimen leaves the collection site for transport to the testing laboratory. Example of a hair collection technique is provided in Appendix A.

4.2 Collector Qualifications

Specimens must be collected by suitably trained personnel (Collecting Officers). Although no healthcare professional education is required, documented training, which includes a demonstration of competence, must be undertaken before collections are performed. The training can for example be organised by a laboratory or an independent organisation or company.

Collectors can be trained by various methods (video, classroom, internet, etc).

Training must include, as a minimum, the following:

- The collection process
- The chain-of-custody process
- The process involved with “problem” collections (e.g. baldness)

- The responsibility of the collector for maintaining donor privacy, confidentiality of information, and specimen integrity
- Ethical issues, especially regarding the declaration by the donor of past and present use of prescribed medications which may influence the result
- Legal, ethical and human rights of the donor

It is highly recommended that, upon completion of the training, each collector is tested on all subject matters covered in the training course to verify their understanding of the topics. It is also highly recommended that each training course include mock collections to assess collector competency. On successful completion of training, the trainee collector may begin performing collections. However, there are a few instances in which a collector should not perform a collection. These situations are:

- If the collector is the immediate supervisor of the donor (unless no other collector is available), or if the collector is a co-worker, a relative or a close friend of the donor.
- An individual working for a drug testing laboratory may not act as a collector if that individual can link the donor with the specimen drug test result unless he/she is bound by professional confidentiality.

4.3 Collection Site

A collection site is a facility (permanent or temporary) selected by the employer where donors present themselves for the purpose of providing a specimen.

Access to the facility must be restricted. Procedures for collection of specimens should allow for individual privacy. Preferably, there should be a sign outside prohibiting entry while a collection is occurring.

The collection area must be thoroughly cleaned, dedicated to sample collection and cannot be used for storage of any potential source of contamination, such as drugs.

4.4 Chain of Custody Form (CCF)

Chain-of-custody is the term used for the process of documenting the handling and storage of the hair specimen from the time the donor gives it to the collector until it is destroyed. A CCF is used to document the collection procedure and the chain-of- custody

of the specimen. In Europe, there are many different types of CCFs. Almost every laboratory that performs WDT has its own version of this form which may also include a different number of copies for each form. The CCF is numbered with a unique specimen identification number and includes a sample label that is printed with the same specimen identification number as the CCF. A tamper-evident label must be applied across the hair collection envelope. The numerical sample label can be used as a tamper-evident label and could also serve to seal the collection kit by applying it across the hair collection envelope containing the collected hair specimen. A purpose tamper-evidence seal can also be utilised.

. The information on the CCF should include:

- Unique specimen identification number, name, address, e-mail address, and phone number of the testing laboratory. Information identifying the donor details such as birth date, name, and home address. However, the identification could be an identification code purposely used so the sample remain anonymous, but it will be linked to the individual being tested, known by the employer. The testing laboratory will not know the actual identity of the individual being tested but the employer will. CCF should include date and time of the collection showing names and signatures of all individuals who had custody of the specimen during the collection process.
- The information on the required analysis including period to be tested and drugs is optional as collection may be part of a separate contract.
- Hair sample information such as colour, length of the hair lock, body site and any obvious cosmetic treatment of the hair should be recorded. The donor should be asked to declare all cosmetic treatments in addition to the observed treatments including whether heat treatment was applied to the hair. The donor should declare the use of prescribed and non-prescribed medications,
- When relevant, the Medical Review Officer (MRO) occupational physician information (i.e. name, address, telephone, e-mail and fax numbers); collection site information (i.e. collector name, telephone number).
- The number of CCF copies may vary between laboratories and countries. Typically, a minimum of three parts or copies of the CCF are distributed by the

collector as follows: one part or copy to the testing laboratory (with the specimen); a second part or copy to the donor; and a third part or copy retained by the collector

4.5 Collection Process

The following describes steps for a hair collection (an example of a hair collection technique is provided in Appendix A):

i **Verification of donor's identity.**

Annotate the CCF: The collector completes the appropriate sections of the CCF. **Hair collection.** The specimen must be cut from the posterior vertex region of the head, as close as possible to the scalp, since this is the region of least variation in growth rate. A lock of hair, with the thickness of a thin pencil – about 0.3 – 0.5 cm in diameter, is usually collected. The root end must be clearly indicated. Unless otherwise required by local laws or employer policy, the collector is permitted to submit a single specimen. **When both A and B specimens are required, two specimens should be collected side by side.** The amount of hair collected will vary according to the needs of each laboratory. Sufficient hair must be collected to allow initial testing, followed by confirmatory or re-testing of the sample if necessary. In cases where head hair is not available, body hair (i.e. chest and pubic) can be collected, if permitted by the company's WDT policy and the donor. Other source of body hair (i.e. arm and leg) may be a suitable alternative. Axillary hair should be avoided because drugs and alcohol biomarkers may be underestimated. Pubic hair is to be avoided when testing for ethylglucuronide

ii **Place the lock(s) of hair into the collection kits.** The whole length of the hair lock is placed inside the sample kit for despatch to the laboratory. When A and B samples are required, the two locks of hair are placed into two different collection kit envelopes. Each envelope will contain the minimum quantity needed for the analysis. Both specimen A and B should be stored in a dry environment, at room temperature and protected from daylight

- iii **Seal the kit.** In front of the donor, the collector places the tamper evident label/seal over both collection kit envelopes. The label/seal should be arranged in a way that any manipulation can be detected
- iv **Instruct the donor to annotate kit seals.** The collector instructs the donor to record her/his initials and date on each of the specimen kit seal.
- v **Annotate the CCF.** The collector completes the appropriate sections of the CCF with donor information (e.g. date of birth, telephone numbers), collection information (e.g. date and time of the collection), and chain-of-custody entries, and instructs the donor to sign the CCF.
- vi **Check the CCF.** The collector checks all relevant copies of the CCF for legibility and completeness. If all copies are legible and complete, the collector then provides the donor a copy of the CCF and permits the donor to leave.
- vii **Prepare specimens for shipment.** The collector places the specimen kits along with the laboratory copy (original) of the CCF in an envelope. It is important that the collector ensures each specimen collected is shipped (or picked up by the laboratory's courier) in accordance with the company policy. Specimens must be stored and shipped at room temperature, away from direct sunlight and humidity.

5. Laboratory Organisation

It is recommended that analyses are directed to designated laboratories, which have the analytical capacity to achieve the performances required by hair analysis. The complex nature of hair analysis and the concomitant analytical challenges necessitates trained scientists and sufficiently equipped laboratories.

The laboratory should use as minimal hyphenated techniques such as chromatography-mass spectrometry (GC-MS, LC-MS and tandem MS technologies), well-documented experience in method validation, data handling and expertise in reporting of results. Staff should have the ability to interact with investigating officers and with employment tribunals and the courts of law.

6. Laboratory analyses and procedures

6.1 Sample handling

Receipt of specimens in the laboratory should be indicated by a handwritten or electronic signature (or initials). Any transfer of specimens must be documented as part of the permanent laboratory record.

Once the analyses are completed, specimens must be stored for the time period agreed with the customer in a secure manner. However, a minimum 1-year storage period is recommended.

Areas of possible contamination must be considered before and during the analysis and when interpreting the results. These may include external drug exposure and laboratory contamination.

The sample and any aliquots or extracts must be handled and stored in a manner so as to minimise degradation, loss of analytes, or contamination from other sources. Dry hair should be protected from UV light sources and at room temperature whilst in storage.

6.2 Sample decontamination

When hair analysis is being used to identify drug use, potential external contamination of the hair needs to be minimised. In general, a decontamination strategy to remove gross environmental contamination must include a wash protocol before analysis of the sample. The wash protocol could be a combination of aqueous and/or organic solvents which has been validated by the laboratory. The wash residues can be stored for later analysis if necessary. After washing, the required hair segment is submitted to a validated extraction protocol that may involve drying, pulverising, cutting into small pieces or chemical disintegration of the hair sample.

6.3 Sample extraction

The minimum amount of hair used in the analysis varies between different laboratories and each laboratory must establish their requirement during method validation. Extraction procedures vary between different laboratories and each laboratory must validate their method of choice prior to use.

6.4 Screening tests

A preliminary immunoassay screening can be applied to eliminate all negative samples and identify presumptive positive samples. All presumptive positive samples need to be confirmed with a second different method according to international guidelines. The second method is a mass spectrometry (MS)-based technique, e.g., GC-MS or LC-MS.

6.5 Confirmation tests

National or European guidelines for mass spectrometric identification and quantification of drugs must be taken into account. MS criteria for identification by either scanning (e.g. Full Scan, Product Ion Scan) or non-scanning (e.g. Selected Ion Monitoring, Selected Reaction Monitoring) techniques are based on the presence and relative abundance of a number of ions which are defined by the Laboratory as diagnostic for the analyte. Any data processing (e.g. integration, subtraction, averaging, etc.) shall be performed consistently across the analytical batch.

The use of deuterated internal standards is recommended whenever possible.

7. Quality assurance and quality control

7.1 Accreditation

Laboratories performing hair analysis should ideally be accredited to ISO/IEC 17025 standards with consideration of the guidelines of the SoHT (Society of Hair Testing) in view of each country's legislation requirements. Different countries have different accreditation bodies and individual countries have their own accreditation body that follows international standards that usually certify laboratory competency.

7.2 Personnel

7.2.1 Head of Laboratory

This person is responsible for the day-to-day management of the drug testing laboratory. Some of the functions may be delegated to other appropriately qualified personnel but the overall responsibility for any delegated functions will remain with the designated Laboratory Head (typically the Laboratory Manager).

7.2.2 Authorising Scientist

A person responsible for the review and certification of pertinent data and quality control results, prior to release of analytical results.

7.2.3 Laboratory Analyst

A person responsible for undertaking the day-to-day analytical procedures.

7.2.4 Toxicologist

A person responsible for interpreting a toxicological analytical result for the customer or the customer's designated Medical Review Officer.

7.2.5 Expert Witness

A person to present evidence to administrative or disciplinary proceedings that are based on analytical results reported by the laboratory.

7.2.6 Other personnel

Other technical or non-technical staff must have the necessary training and skills for the tasks assigned.

7.2.7 Quality Manager

The person responsible for quality assurance within the laboratory organisation.

7.3 Accommodation and Environmental Conditions

The laboratory must be accessible only to authorised visitors.

Visitors should always be accompanied and are required to sign a log-book upon entry and departure from the laboratory, recording date and purpose of the visit and times of arrival and departure.

Due to their high surface area to volume ratio, hair samples are highly susceptible to external contamination. As a measure to avoid contamination, the analysis and the storage of hair samples in laboratories where seized drugs are analysed must be carried

out in separate rooms. As an additional precaution, laboratory coats must be changed if the laboratory analyst is handling seized drugs and also carrying out the analysis of hair samples.

8. Interpretation of results

8.1 Introduction

Interpretation of results must be achieved by an experienced scientist.

The following aspects need to be taken into account when interpreting a hair analysis result.

8.2 Theoretical Background

Hair is a unique matrix because no active metabolism and excretion is present to remove drugs once deposited.

Drug concentration in normally treated hair depends mainly on:

- dosage of abused drug
- metabolism
- distance from the root – significant decrease in drug concentration can be observed after several months due to washing and U.V. radiation
- cosmetic chemical or heat treatment
- position along the hair and polarity of drug
- hair colour
- percentage of hair in the anagen and telogen phase

Therefore, it is often not straightforward to correlate the concentration of drugs found in hair with consumption patterns.

The growth rate of head hair could range from 0.7 cm - 1.4 cm/month when about 80%-95% of the follicles remain in the anagen phase (active growing phase). Growth rate could be influenced by therapeutic drugs, age, sex, race and depends even on seasonal fluctuations. The calculation of the period covered by head hair uses the average of 1 cm/month to give an approximate period of the detection covered by the hair sample or segment analysed.

Body hair has a slightly slower growth rate (0.5 - 1.1 cm/month) but a different percentage in the three stages of growth cycle in comparison to scalp hair. The calculation of the period covered by body hair uses the average of 1 cm/month to give an approximate period of the detection covered by the length of body hair, but because 40-80% remains in the resting phase, the period covered may be extended by 3-4 months. Body hair is not suitable for segmental analysis.

8.3 Cut-offs - Criteria for a Positive Drug Test

The positive result of a hair analysis may be used to confirm if a person has frequently used or was exposed to a drug. The Society of Hair Testing (SoHT) has recommended specific cut-offs for hair testing in forensic cases^[1-3]. Different countries may have different WDT legislations with their specific cut-offs, which have to be taken in consideration from the testing laboratory.

8.3.1 Alcohol

The direct determination of ethanol itself in hair is not possible due to its volatility and its potential absorption from external sources. Instead, the minor ethanol metabolites ethyl glucuronide (EtG) and optionally, ethyl palmitate (EtPa) are measured in hair as a direct alcohol consumption marker.

The SoHT consensus for the analysis of alcohol markers in hair should be followed: https://www.soht.org/images/pdf/Revision_2019_Alcoholmarkers.pdf

A concentration greater than or equal to 30 pg/mg EtG in the proximal head hair segment with a length of 3 cm up to 6 cm strongly suggests chronic excessive alcohol consumption.

A concentration lower than or equal to 5 pg/mg EtG in the proximal head hair segment with a length of 3 cm up to 6 cm does not contradict self-reported abstinence.

8.3.2 Drugs and related compounds

The most common drugs and related compounds tested in hair samples in the workplace sector are: opiates/opioids; cocaine; amphetamines; cannabinoids; methadone; buprenorphine; benzodiazepines and analogues; ketamine

The SoHT consensus for the analysis of drugs and related compounds in hair should be followed: https://www.soht.org/images/pdf/Consensus_DoA_2021.pdf

When relevant, other groups may be tested for, like New Psychoactive Substances (NPS) or steroids. In this case, the results interpretation should be done by an experienced hair testing analyst, because for most of these drugs it is not possible to establish the minimal detectable dose, the frequency of use and the dosage based only on the concentration.

8.4 Reporting the Results

From the analytical point of view, the uncertainties of the final analytical measurement of hair samples can be greater than those in urine or oral fluid and variable between different laboratories. This is because the analysis of hair samples may be more cumbersome to analyse than urine or oral fluid. For example, the hair needs to be decontaminated and pulverise and or digested before analysis and losses during these processes may increase variability of the results and are likely to affect the detection of drugs and metabolites.

Awareness of the analytical pitfalls of hair testing is key to understanding its results. It is possible for an individual to provide a negative hair result if they have used or been exposed to a drug infrequently or in low doses. It is not possible to know precisely from the results of hair analysis how much or how often an individual used drugs and classify them as a heavy user or a light user.

The levels of drugs detected in hair are currently best used as a guide to changes of use in the individual when sectional analysis is performed, or two different periods are compared in the same individual. This attribute can be used to monitor drug use patterns, demonstrating increasing or decreasing doses being used by the same individual over longer time periods.

With hair analysis, when results are nil or below cut-off, results are usually reported as 'negative' or 'not detected'. However, a result below cut-off does not absolutely prove that

an individual has not used drugs. Low levels in hair are usually correlated to the small quantities of drugs used, although the minimal detectable dose that can be detected in hair is unknown.

While a positive result of a hair analysis indicates that a person has used or was exposed to a drug, a negative result does not refute use of or exposure to the drug because of the use of cut-offs by the testing laboratories. Clients may need to be offered the option of being informed when drugs are unequivocally detected at levels below cut-off in some critical industries.

If the results reports are reviewed by Medical Review Officer (MRO) or occupational physician, it must be ensured that the quality criteria for both the screening and confirmation tests are fulfilled. This must be checked by 2 qualified personnel (for instance the laboratory analyst and the authorizing scientist) and documented in the laboratory information system. The minimum quality criteria are:

- (i) the daily internal Q.C. (for both the screening and confirmation tests) must meet the acceptance criteria and
- (ii) the fulfillment of the chromatographic and mass spectral identification criteria.

The test report should contain as a minimum the following information:

- Coded identification of the donor
- Date of sample collection
- Date of receipt of the hair sample in the laboratory
- Address and/or fax of occupational physician/MRO ordering the test
- Characterisation of the hair sample:
 - a. type of hair (e.g. head, pubic, chest, arm or leg hair)
 - b. original length of the hair specimen
 - c. length of the analysed hair segment
 - d. hair colour
 - e. cosmetic treatment
- Type of analysis performed (screening and/or confirmation)
- Analytical method applied
- List of drugs, metabolites or alcohol markers analysed and their detected concentrations
- Cut-offs values used
- Interpretation of the result (if requested):
 - a. Decision about negative or positive outcome

- b. Statement about the minimum and maximum time period represented by the investigated hair length or segment length.
- Name of the person who is authorised to declare the result of the analysis

9. Challenges to drug test results

In situations where there is a challenge to the results of a positive drug test result, the following guidelines must be used. Sample should be released for analysis to a drug testing laboratory accredited by a recognised external accrediting body and working to these guidelines. This release requires authorisation from both the customer/MRO and the donor.

The release must be supported by chain of custody procedures that can withstand legal scrutiny and include information about the findings of the original test (corresponding A sample) and the cut-offs used for the test.

The original laboratory must retain the residue of the original sample and its containers so that it can be compared with the B sample at a later date if required.

All laboratories that undertake B sample testing must be able to demonstrate that they can accurately determine the concentration of a drug or metabolite at 50% of the recommended confirmation cut-off concentration listed in Table 3 (or the cut-off used for the original test, whichever is the lower).

On receipt in the testing laboratory, the B sample should follow chain of custody procedures as outlined. It is recommended that the laboratory should carry out validity checks outlined prior to carrying out the confirmation analysis. Only those drugs identified for confirmation testing should be looked for. The purpose of the B sample analysis is to determine if the drug detected in the A sample is present in the B sample. No cut-off is to be applied to the B sample analysis and no comparison of the concentration of drug detected between the original A sample analysis and the B sample analysis should be made. If the B sample analysis does not detect evidence of the drug use detected in the A sample analysis, the final report should indicate that it is **inconsistent with the A sample analysis**. If the B sample analysis detects evidence of the drug use detected in the A sample analysis, the final report should indicate that it is **consistent with the A sample analysis**. Confirmation cut-off levels are not to be used as the determinant. There must be no comment on the final report that states whether the sample is positive or negative.

9.1 External contamination

The detection of metabolites is the main approach in hair testing to confirm drug use, and exclude external contamination, as metabolism requires ingestion into the body.

The following identification of metabolites are recommended for the exclusion of external contamination:

1. Ideally at least two metabolites, such as benzoylecgonine and/or norcocaine and/or cocaethylene and/or hydroxycocaines are used for the confirmation of cocaine consumption;
2. 6-acetylmorphine and morphine for the confirmation of heroin consumption;
3. Amphetamine for the confirmation of methamphetamine consumption;
4. THC-COOH for the confirmation of cannabis consumption;
5. MDA for the confirmation of Ecstasy (MDMA) consumption;
6. Norketamine for the confirmation of ketamine consumption;
7. EDDP for the confirmation of methadone consumption;
8. Norbuprenorphine for the confirmation of buprenorphine consumption.

The examination of the wash residues may aid the interpretation of the results. Decontamination of the hair samples before the analysis is a key step in the preparation of samples before extraction and analysis to remove and minimize the possibility of residue caused by external contamination and other potential assay interferents. It is not possible to know the efficiency of the hair washing and that it does not initiate the extraction of the drug in the hair sample. Most external contamination is due to own use of smokable drugs. By measuring the drug component present in the wash residue and comparing it with the amount in the hair samples it is possible to aid the interpretation of results and can lead to successful differentiation of external contamination from drug use in most cases.

9.2 Effects of Cosmetic Treatment

Every strong chemical, physical and mechanical influence could have harmful effects on the cuticle: perming, straightening, dyeing, bleaching, excessive washing, intensive illumination with ultraviolet radiation, excessive exposure to sunlight and heat. Bleaching, highlights, or lightening involve the irreversible destruction of melanin by oxidation, a

partial or even complete degradation of melanin is possible. When strong bleach is used the physical properties of hair will be altered (e.g. a higher porosity).

Hence cosmetic treatment must be considered and accounted for pre- and post-analysis since it can reduce the drug concentration below the limit of detection or cut-off value and hence cause false negative results.

Nonetheless, cosmetically treated hair is not automatically useless for drug and EtG detection; conversely, hair analysis is a powerful tool, often the only means to detect retrospective drugs and/or alcohol consumption even in dyed or bleached hair. In case of cosmetically treated head hair, a body hair specimen might be investigated at the same time.

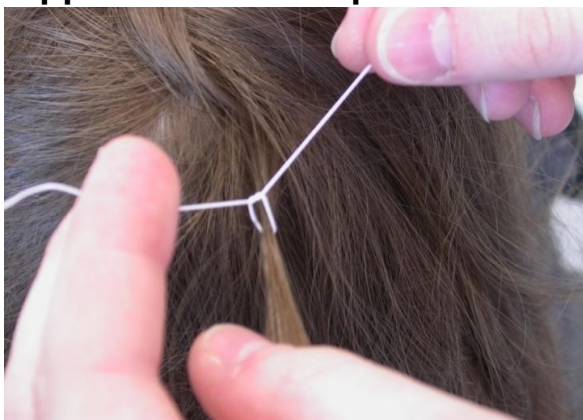
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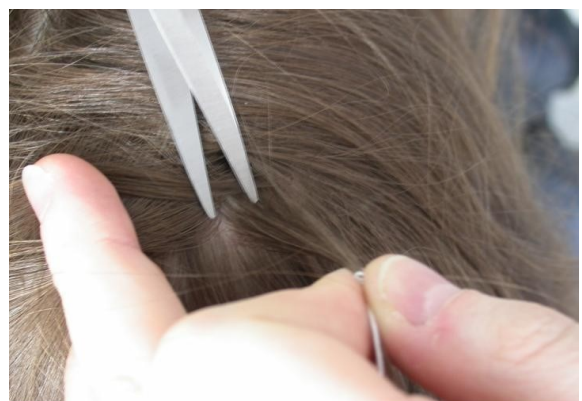
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Appendix A –Example of a hair collection technique



A lock of hair (or more, if necessary) about the width of a thin pencil are cut from the back of the head,



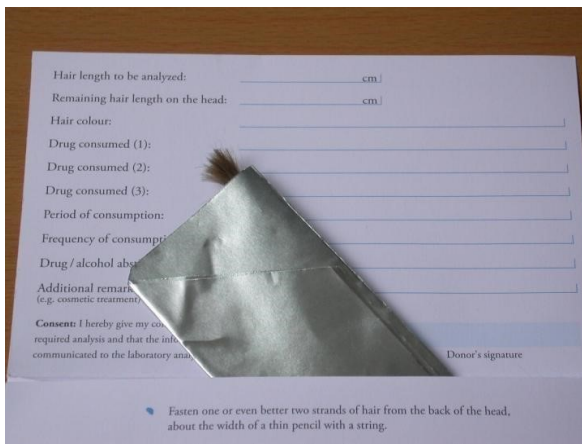
The hair is to be cut just above the skin, as close to the scalp as possible.



The hair lock(s) are put in separate aluminium foils provided with the root end exposed at the notched end.



Fill in the donor's particulars and required test. The donor should sign the declaration. Hair samples shall be sent to the laboratory in an envelope by post or by courier.



Insert the aluminium foil into the envelope provided. Seal the envelope.



Each aluminium foil is to be folded once as shown above.