Supplementary Technical Notes for Biology Casework SFR

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Purpose	This document provides general technical information that is relevant to the reporting of biology/DNA findings via the SFR process. This set of agreed, supplementary notes allows the SFR to be concise, whilst referencing further guidance about the methods used and their interpretation, in support the case management process.
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1. Introduction

These guidance notes are intended to be used to supplement the production of an MG22 (SFR) form, such that the forensic result can be reported in the most clear and succinct way. The relevant version and section(s) of these supplementary notes should be quoted within the MG22 document produced by the Forensic Service Provider.

1.1. Definitions & Abbreviations

Abbr.	Meaning
LCV	Leuco Crystal Violet
DNA	Deoxyribonucleic Acid
SFR	Streamlined Forensic Reporting
NDNAD	National DNA Database
AP	Acid Phosphatase
STR	Short tandem repeat
Shall	indicates a requirement
Should	indicates a recommendation
May	indicates a permission
Can	indicates a possibility or a capability

2. Blood

2.1. Examination for Blood

Searching for blood typically involves the combination of careful visual inspection aided by oblique lighting, low power microscopy, and a chemical screening test (presumptive test).

Bloodstains are usually red-brown in colour, however this can depend on the colour of the surface on which they are deposited and the conditions to which they have been subjected.

Likely bloodstains are tested using a chemical presumptive test for blood. If the stain contains blood then a colour change of the chemical reagent is observed. Therefore, if a visible red-brown stain that has the appearance of blood gives a positive reaction to a presumptive test for blood, the stain is identified as blood.

2.2. Interpretation of Blood Pattern/Distribution (General)

When a person is bleeding, their blood may be transferred onto nearby surfaces. This can occur by a number of different methods, involving either direct contact or airborne transmission. The nature and distribution of the resultant bloodstain(s) on an item can help in evaluating how the blood staining came to be deposited onto the surface.

Direct contact with a source of wet blood will result in a contact bloodstain or smear, whereas, spots of blood usually indicate airborne transfer. Small spots of blood may be produced in a number of ways including from an impact into wet blood. Small spots of blood can also be seen when blood is expirated from the airways such as the mouth or nose.

The amount and distribution of blood transferred will depend upon factors such as the nature and duration of the contact, the proximity of the people/objects involved, the amount of blood that was shed at the time of the assault and the amount of force applied.

2.3. Interpretation of Blood Pattern/Distribution (Assault)

During the course of a physical assault, if a person sustains an injury that bleeds, blood may be transferred onto the clothing of those involved, onto surrounding surfaces and onto any weapon(s) used. The appearance of a bloodstain and the distribution of groups of bloodstains may be used to ascertain how an item became blood stained.

When items stained with wet blood come into contact with other items a contact bloodstain results. If there is movement at the time of contact or whilst the blood is still wet then the resultant stain will be present in the form of a smear.

Large blood spots / drips of blood are formed due to blood falling freely under their own weight whereas smaller spots are formed where energy has broken up the blood and dispersed it as small airborne blood drops of varying size, for example, when an object strikes a surface wet with blood, when blood is expirated from a bloody nose or mouth, or when blood drips onto a surface already wet with blood. So when force is applied to wet blood, for example, if an injured area is repeatedly struck by a weapon or by punching or kicking, it is separated into small airborne drops which are dispersed away from their source and land on any items that lie in their path. The blood spots formed by an object striking liquid blood are also known as impact spatter. The

shape of the spots may be used to determine the approximate area from which they had originated.

The amount and distribution of blood transferred will depend upon such factors as the nature and duration of the contact, the proximity of the people/objects involved, the number of parties involved in the incident, the availability of any blood to transfer and the degree of force applied.

2.4. Blood Patterns on Clothing

Kicking into an area wet with blood may produce a bloodstain pattern on the shoes and lower legs of the trousers of the kicker which may consist of contact bloodstains with associated airborne bloodstains radiating from the contact bloodstains. If kicks occur into areas that are not blood stained, blood may still be transferred but not in any particular pattern or it may be that no blood is transferred.

Punching into an area wet with blood may produce a bloodstain pattern on the sleeves and chest of upper garments. This consists of contact bloodstains around the cuff area (for long-sleeved garments) with associated airborne bloodstains radiating from the contact bloodstain(s). If punches occur into areas that are not blood stained, blood may still be transferred but not in any particular pattern or it may be that no blood is transferred.

In some cases the forensic scientist may deem such bloodstain patterns to be indicative of the action causing the blood pattern observed (such that there may be few other ways in which the pattern observed could have been caused); It should be noted however, that such patterns are not commonly deposited onto assailant's clothing during a punching or kicking assault, particularly in cases where there has not been repeated punching or kicking into an already wet bloodstained surface(s).

2.5. Blood Patterns on Weapons

If a weapon, such as a baseball bat, had been used to strike a surface heavily stained with wet blood, contact bloodstaining and possibly blood spots radiating away from the contact bloodstain on the surfaces of the weapon may result. However, if the weapon struck a surface which is not bloodstained at the time then it is possible that no blood would transfer onto its surfaces. If the weapon is repeatedly used to strike into wet blood then this can give rise to further types of bloodstains which are caused when a weapon that is heavily wet with blood is swung and strikes a surface (e.g. percussive staining).

When a weapon such as a knife is used to injure an individual then blood is sometimes seen on the blade in a smeared pattern which, depending on certain factors such as the area injured on the body (and whether this is through clothing), may include fatty deposits, skin and/or hair;

although this is not always the case and depending on the circumstances of its use and what happens to the knife afterwards, there could be little or no visible blood on the blade.

2.6. Absence of Blood

It is possible for someone to be the perpetrator of a physical assault and for no blood to transfer to them or their clothing. This could be because the attack took place at a time when there was no blood available for transfer (for example the injuries started to bleed after the assailant left, or the blood was absorbed by the injured party's clothing preventing a transfer) or the attack took place to areas that were not blood stained (for example the victim was kicked about the body when it was a head injury that was bleeding).

2.7. Altered Blood Stains

Blood may undergo a change prior to forming a pattern which can affect its appearance, for example it may clot or become mixed with other body fluids such as saliva or urine. It may also become mixed with other non body fluids or attempts may be made to clean the blood by washing or wiping.

2.8. Cast-Off

Small airborne drops of blood can be produced if a wet blood stained object is moved, for example when a weapon that is wet with blood is swung. The blood is caused to leave the object due to its motion and this can produce a linear series of blood spots on the surface(s) on which they land. Blood staining of this type is referred to as cast-off blood staining.

2.9. Expirated Blood

Blood can be propelled from in or around the openings of the airway, for example, by coughing, sneezing or wheezing. The resulting pattern generally consists of a number of spots of blood of varying size some of which may be directional and some may have air bubbles present. The blood may also become mixed with other body fluids such as saliva or lung surfactant and can have a dilute or 'stringy' appearance. However, the absence of air bubbles, dilution or any stringy appearance does not necessarily indicate that a blood pattern is not expirated blood and the pattern produced is dependent on the amount of blood available for transfer and the degree of exhaling force applied.

2.10. Projected Blood

Blood can be projected from severed arteries and veins, often with some force, and can be deposited on nearby surfaces such as walls and clothing. This can result in the formation of a projected pattern of blood.

If an individual receives an injury that breaches an arterial or venous blood vessel, patterns of bloodstains can arise which are indicative of that type of injury. This may involve large volumes of blood being ejected with some force and can be referred to as a projected pattern. Dependent upon whether the injured person is moving at the time of bleeding, projected patterns can arise which appear as circular bloodstains, in-line or in a zigzag or wave pattern. Projected patterns occur as a result of the blood being forced out of the breached vessel under intermittent pressure reflecting the injured person's heartbeat.

2.11. Drips and Blood Trails

When blood falls onto a surface due to gravity alone, a large spot of blood (a drip stain) is formed. If the source of blood is moving, a drip trail is formed. If the source of blood is stationary, then several drops of blood will fall in one place. This can result in a pooling of blood and secondary spatter (small drops of blood being ejected away from the area of the drips).

2.12. Chemical Enhancement of Blood

Chemical reagents such as Luminol and BlueStar can be used to detect the presence of non-visible blood staining. These are presumptive tests; the reagent is applied as a spray which reacts with blood to produce a blue-green glow that can be seen in darkened environments. Luminol and BlueStar are extremely sensitive and can detect very small amounts of blood that are not visible to the naked eye, or blood that has been diluted. A positive reaction obtained from an area where no visible blood staining is present indicates, but does not confirm, the presence of blood. Additional chemical testing can therefore be conducted in an attempt to further investigate the possible presence of blood. Following Luminol examination, items present a health and safety risk which should be considered before any defence examination or return to owner. Items should not be re-opened unless under laboratory conditions, and should undergo professional cleaning or be disposed of appropriately. Safety Data Sheets and further advice can be provided if required.

Hydrogen Peroxide can also be used for the detection of very small or non-visible bloodstains, and enhancement of visible blood staining on dark items. The chemical reagent, used as a presumptive test, is applied as a spray which reacts with blood to produce a visible white reaction. The area can be further tested using other chemical screening tests for blood and/or DNA analysis.

Leuco Crystal Violet ('LCV') is also used to enhance marks in blood and other areas of blood staining, including instances where a clean up has been attempted. LCV reacts with the blood producing a purple/violet colour, improving the visualisation of blood on many surfaces. LCV can be especially useful for resolving fine details. An LCV positive reaction obtained from an area where no visible blood staining is present indicates, but does not confirm, the presence of blood.

The presumptive tests are not specific tests for human blood and will give positive results with animal blood. Further immunological tests and/or mitochondrial DNA can be used to identify species, if required.

3. Damage Assessment and Interpretation

Areas of evidential textile damage are examined visually and using a low power microscope to observe the characteristics of the damage, such as dimensions, shape and appearance as well as to observe the fibre ends at the damaged site. This detailed examination aids in determining whether the damage present has been caused by a ripping or tearing action or by a cutting action from a sharp implement such as a knife. The appearance of the damage may also indicate whether it is 'recent'. The term 'recency' does not designate a time period, but rather, it indicates that the damaged item has not been washed or worn extensively since the damage occurred.

It should be noted that in some cases, where there is heavy blood staining associated with the area of damage, this may obscure any characteristic features at the edges or margins of the damage and may also alter the dimensions of the damage feature, particularly once the blood has dried.

3.1. Cuts

Cuts to clothing may be broadly categorised into stab cuts, slash cuts or scissor cuts. Stab cuts are penetration of the clothing by a cutting implement in a thrusting manner. Slash cuts are cuts produced in a sweeping or slashing manner and may not involve penetration. An area of damage caused by a blunt implement being thrust into and penetrating a garment can be referred to as puncture damage.

In assessing fabric damage, many variables need to be taken into consideration. The process of stabbing can be a very dynamic situation, as individuals will generally move in response to avoid being stabbed. As such, clothing may become bunched and folded during the entry and exit of a weapon, such as a knife. This can affect the overall shape and dimensions of the area of damage, such that it may not reflect the true <u>size</u> or shape of the blade of the implement.

All of these factors need to be considered when assessing and interpreting damage evidence, and in forming conclusions as to the size and shape of the implement used. Furthermore, these

findings may need to be considered in conjunction with injury interpretation provided by a pathologist or other medical specialist.

3.2. Tears

Tearing actions will tend to cause distortion and fraying of fabric and can be distinguished from cut or slash damage, which normally displays straight or more uniform edges.

The process of tearing an item of clothing can be a very dynamic situation, particularly if the individual is not wearing the item in the correct orientation at the time of the incident and/or the individual wearing the item is involved in a struggle. This can result in damage being present in areas or locations that might not normally become exposed and/or damaged. Due to the movement of the individuals concerned, it can often be difficult to reliably assess the level of force required to create a particular damage feature.

All of these factors, as well as the general condition of the item and how likely it is to display damage, need to be considered when assessing and interpreting damage evidence.

NOTE: Other types of damage, in addition to cuts and tears, are encountered more rarely in forensic casework including abrasion damage, thermal damage, chemical damage and environmental damage. Where these are encountered, they will be described in more detail within the content of the MG22 report.

3.3. Test Damage

In an attempt to assess whether a particular implement could produce the textile damage observed on an item, an implement of interest (or other, similar implements) can be used to produce areas of test damage on the item in question or on items of similar construction. The areas of test damage are examined in detail and then compared with the characteristics of the evidential damage. It is also possible to create ‘test tears’ to a garment, with a view to addressing whether or not the evidential damage could have been caused in the manner alleged.

3.4. Physical Comparison of Items and Physical Fit

When an item is broken, cut, torn or otherwise separated into two or more pieces, there will be, in many instances, a unique characteristic profile formed between the opposing surfaces of each of the pieces of the item in question. It is therefore possible to carry out a detailed examination of the irregularly formed damaged edges as well as any features of the items, such as colour patterns, to determine whether or not they were once joined, i.e. they physically fit together. If a physical fit can be determined, it may be possible to conclude that they were originally joined, demonstrating that the pieces of the item were once a single item. Such a unique physical fit is

made possible by the involvement of random processes and the particular separation would not be expected to occur in exactly the same way more than once.

In some instances, it may not be possible to 'fit' the damaged edges of the item together precisely, for example when pieces are missing or when the broken edges have become distorted due to the forces applied which caused the break. However, in these cases, the forensic scientist may still be able to form an opinion as to whether they once could have formed part of the same item.

4. Semen

4.1. Semen Persistence

Following a sexual act in which semen is ejaculated, the individual components of seminal fluid will remain detectable for different lengths of time. Two components are of significance in determining the presence of semen: one is a chemical called Acid Phosphatase ('AP'), and the other is the spermatozoa or sperm cells.

When semen is deposited in a body orifice such as the vagina/anus/mouth during sexual intercourse, it can subsequently be recovered during a medical examination by sampling using cotton swabs. The likelihood of detecting semen on vaginal/anal/mouth swabs (and mouth washings) is dependent on the delay between an act of sexual intercourse with ejaculation of semen, and the taking of the samples at medical examination.

The current guidelines for sampling during the medical examination have been produced by the Faculty of Forensic and Legal Medicine and can be viewed/downloaded using the following link:

http://fflm.ac.uk/publications/recommendations-for-the-collection-of-forensic-specimens-from-complainants-and-suspects-3/

These guidelines are reviewed every 6 months to take into account new information and advances in forensic recovery and analysis. It should be noted that these guidelines are for the collection of samples only, taking into consideration the information available at the time of the medical examination. Any requirement for laboratory testing of the samples will be considered in light of the full circumstances of the case, using information provided on the medical examination forms and laboratory (MG21) submission form. It is not necessarily the case that all samples taken will need to be examined in order to progress the specific issues identified.

Following an act of vaginal intercourse involving internal ejaculation, AP is likely to be detected on vaginal swabs taken up to 24 hours afterwards.

Following ejaculation during oral/vaginal/anal intercourse, spermatozoa (sperm cells) are gradually lost from the mouth/vagina/anus by various actions such as degradation, drainage and washing/wiping. Spermatozoa can be detected:

- on mouth washings/mouth swabs taken within 12 hours, and more rarely up to 2 days after oral intercourse;
- on vaginal swabs taken within 3 days of vaginal intercourse, and more rarely up to 7 days and beyond;
- on anal swabs taken within 2 days of anal intercourse, and more rarely up to 3 days after intercourse.

In certain circumstances the spermatozoa may be absent or reduced in number and only the seminal fluid is detected. Vasectomy, medical conditions or natural causes may produce this state and in the latter two it can be permanent or transitory. In the absence of spermatozoa, the presence of seminal fluid may be investigated further using the 'Florence Iodine' test which detects the presence of the chemical choline which is present in seminal fluid. Choline may be detected on vaginal swabs taken up to 24 hours after sexual intercourse.

If vaginal intercourse occurs with no internal ejaculation, then semen may not be found on the swabs taken. However, in the absence of ejaculation, it is possible for traces of residual semen or cellular material on the skin of the penis to transfer to the vagina. These traces could persist for up to, and sometimes beyond, 48 hours.

Material transferred to the vagina/mouth/anus during such acts will also be gradually lost by various mechanisms such as degradation, drainage and washing/wiping.

In circumstances where semen could be mixed with other cells/body fluids, such as blood, or epithelial cells that line the vagina/mouth, attempts can be made to separate the spermatozoa (into a 'seminal' fraction) from the other cellular material (into a 'cellular' or 'epithelial' fraction) prior to DNA profiling. This is an attempt to avoid the production of mixed DNA results. However, this procedure does not always result in a complete separation, particularly if the proportion of semen in the sample is very low compared to the proportion of other cells. An incomplete separation of cells types can result in mixed DNA profiles being obtained. It should be noted that semen contains both spermatozoa and other cells from the male donor.

4.2. Examination for Semen on other items

Semen deposited into the vagina/anus during sexual intercourse will subsequently drain from these orifices onto items worn next to the genitals such as underwear. Semen can also be deposited on to an item of clothing in a number of other ways. These include direct ejaculation onto the item or by being transferred from a wet semen stained object or body part, or to a lesser extent, a dry semen stained object or body part.

When semen is deposited on to an item it is usual for the AP and the spermatozoa to remain in detectable amounts for an indefinite period of time. This is reliant on the clothing not being washed.

Semen stains on items such as fabric may sometimes appear as off-white, occasionally crusty staining, but are not always visible. The process of identifying semen on items of clothing or other such exhibits is a two staged process. In the absence of any visible staining, in order to locate them the item is initially screened for AP. If positive chemical reactions are obtained the area can be further extracted and the extract can be examined microscopically for the presence of sperm cells, which will confirm the presence of semen. However, it should be noted that positive reactions to the AP test may be obtained from other biological materials such as vaginal material, yeast and bacteria. Therefore, the AP test result is carefully considered alongside the nature/condition of the item and the presence/levels of sperm cells detected in the corresponding (and sometimes surrounding) area in order to fully interpret the scientific findings.

4.3. Semen and Washing

If semen is deposited onto an item of clothing/bedding and the item is subsequently washed, obtaining a positive chemical reaction for AP would be unexpected. However, it is known that sperm cells can be retained on an item after one or more wash cycles.

Furthermore, if a previously non semen stained item is washed with other items that are stained with semen, sperm cells may be transferred to the non-semen stained item and may be detected. Therefore, the presence of trace levels of sperm cells on an item that has been washed may not be as a result of any sexual activity.

4.4. Penile/Digital Penetration

If a penis/fingers penetrates the mouth/vagina/anus it is possible that saliva/vaginal material/faeces may be transferred to the surface of the penis/fingers. In addition, if any of these orifices are bleeding or have semen in them it is also possible that blood and/or semen from them may be transferred to the surface of the penis/fingers.

Cellular material/blood/saliva/semen/faecal material transferred to the penis can persist for up to 24 hours and more rarely up to 3 days. This transfer of material may also occur to fingers in digital penetration cases, although its persistence would likely be for a shorter period of time. Material transferred during such acts will be gradually lost for example via contact with items such as underwear, washing/wiping and further sexual or other activity.

4.5. Condom Use

If a condom is worn correctly during vaginal/oral/anal intercourse with ejaculation, then cellular material would be expected to transfer from the vagina/mouth/anus to the outside surface of the condom and for semen to be deposited on the inside. This semen/cellular material may persist on the surfaces of the condom for a considerable amount of time, depending on the conditions the condom is exposed to.

It is possible that whilst the condom is being removed and in any subsequent contact with it that semen and cellular material on the inside/outside surfaces may be re-distributed onto all surfaces of the condom.

5. Saliva

5.1. Detecting saliva

The presence of saliva on an item or swab can be determined using a presumptive chemical test that detects an enzyme normally found in high levels in saliva called alpha (α) amylase, then extracting the stain to recover cellular material which could have originated from the mouth. There are no reliable tests to say specifically whether or not the cellular material has originated from the mouth, however, DNA profiling tests may help to determine from whom the cellular material could have originated.

Varying levels of alpha (α) amylase can also be present in other body fluids, but these are mostly at lower levels than in saliva with the exception of vaginal secretions and faeces. Therefore, the detection of alpha (α) amylase is not a conclusive test for saliva.

Furthermore, some individuals produce amylase at undetectable levels in their saliva and, as such, a negative result does not necessarily mean that no saliva is present.

5.2. Saliva transfer and persistence

Licking, sucking, kissing or biting may result in the transfer of saliva. The amount of saliva deposited on the recipient surface will depend on such factors as the level of saliva production during this activity and the amount and duration of contact between the mouth and the surface.

However, whilst a strong chemical reaction may suggest high levels of amylase, this does not necessarily correlate with the amount of cellular material transferred. Saliva will persist on the skin for a short period of time as the staining will gradually be removed, e.g. by subsequent transfer onto the clothing being worn or by the action of washing or wiping. Saliva transferred onto clothing may persist until the item is thoroughly washed.

5.3. Airbags

Airbags, whilst not manufactured in 'DNA-free' sterile conditions, are usually installed into a vehicle as a folded, sealed unit. This means that prior to the airbag being deployed, it is unlikely that substantial amounts of DNA from drivers or passengers in the vehicle would be detectable on the surface of the airbag.

It should be noted that a road traffic collision is often a chaotic and dynamic event, and the movement of individuals within a vehicle, particularly if not restrained by a seat belt, may not be readily predictable. Bearing that in mind, it is still the case that when a vehicle is involved in a collision and an airbag deploys, any individuals travelling in the vehicle may be propelled internally, such that their head/face makes contact with the steering-wheel or dashboard mounted airbag. When this occurs, DNA from the surface of their skin or saliva from the individual's mouth can be deposited onto the airbag. If an individual has a bleeding injury, as a result of the collision for example, then blood may also be transferred onto the airbag. Hairs and fibres may also be transferred between the occupant(s) and the surfaces of the airbag.

These expectations relate largely to steering wheel and dashboard airbags. Other forms of airbag carry different expectations for the transfer of skin cells and body fluids and these will be dependent on the nature and duration of contact during the road traffic collision.

It must also be considered that if an individual has close contact with an airbag after it deploys but before it is recovered, then there may be additional opportunities for DNA/body fluids to transfer onto the surface of the airbag. Examples would include a person returning to or entering a vehicle to retrieve an item or help another individual, or if they need to pass through another seat or area to exit the vehicle due to damaged or blocked doors or due to a 3-door layout. If a vehicle has partly or completely overturned, this will also affect movements and thereby expectations.

It may be necessary to examine more than one airbag to obtain a more complete picture before evaluating the findings to assist in addressing the likely location of a given individual within the vehicle at the time of the collision, particularly if there were multiple occupants of a vehicle at the time of a collision.

Similarly, interpretation of hair and/or fibre transfer to an airbag is dependent on the nature of the collision, and subsequent activity within the vehicle before the airbags have been recovered. ‘Fibre-fusions’ i.e. localised areas of softened / melted fibrous material that has been transferred from the surfaces of the garment(s) being worn to the airbag, may be observed as a result of friction between these surfaces.

If someone has been in a vehicle before, particularly if they are a regular driver/passenger, there may be a build-up of their DNA within the vehicle. This means that small amounts of their DNA may transfer onto an airbag during or after a collision even if they are not present. However, there is a low expectation that this DNA would transfer at a comparable level to someone impacting an airbag during a collision, particularly where body fluids have been detected.

6. Faeces

When an item is examined for the presence of faeces, a visual examination is carried out for the presence of brown staining with a characteristic smell, and if visible, samples can be microscopically examined for the presence of undigested foodstuffs. A presumptive chemical test for the presence of urobilinogen, a chemical in faeces, is also carried out. Positive results for these tests suggest the presence of faeces, but are not conclusive; however, the presence of visible staining which has the appearance and odour of faeces and gives a positive result when tested for urobilinogen is usually confirmatory. Negative test results can occur with low levels of faeces and therefore a negative result does not exclude the possible presence of faeces.

7. Urine

When an item is to be examined for the presence of urine, a visual examination is carried out for yellowish "tidemark" staining that may also exhibit stiffening, and noting whether or not the item has a characteristic smell. A presumptive chemical test(s) is also used to test potential urine stains. It should be noted that this test(s) can give positive reactions with other substances and therefore a positive reaction to this test(s) does not specifically identify the presence of urine. In addition, a negative reaction to this test(s) does not necessarily mean that urine is not present on the item. The presence of a characteristic smell, visible staining and a positive reaction to the chemical test(s) can be used to provide support that urine is present; where only some of these results are obtained, this will only provide a possible indication of the presence of urine.

8. Hairs

Hairs are examined visually, both with and without a microscope, to assess several features of the hairs. From such observations, it may be possible to determine a number of characteristics,

including: whether a hair originated from an animal or human; the region of the body from which a hair originated; the growth stage of a hair; ethnicity of the donor and any damage features. Macroscopic and microscopic morphological features of the hairs that are noted include their length, colour (including whether or not there are indications that the hair has been dyed) and whether or not each hair has a root. By examining the hair and its root, it may be possible to determine how the hair was removed.

Loose hairs are naturally shed from the scalp during normal grooming activities such as washing of the hair, combing or brushing. Any root present on a naturally shed hair will normally be small and dry. However, if an actively growing hair was to be forcefully pulled from the scalp it will often have a stretched appearance and bring sheath material lining the hair follicle with it because the root is normally deeply embedded inside the follicle during this stage.

A hair removed from the scalp by cutting will have no root and a relatively uniform straight end. Burnt hairs can exhibit colour changes, become brittle and distorted in appearance. Hair shafts which have been crushed, for example by blunt force, can exhibit a splintered and fibrous appearance.

The majority of DNA in human hairs that is suitable for the standard DNA analysis procedure is located in the hair root, especially in the root sheath material that surrounds the root. If a hair does not have a root, or has a root with a very small amount of root sheath material, the chances of obtaining a meaningful DNA profile are reduced. By grouping a number of hair roots from visually similar hairs together, it may be possible to increase the amount of DNA recovered such that a complete DNA profile can be obtained. Consideration should also be given to the presence of DNA on the hair shaft deposited as a result of contact such as touching which may or may not relate to the donor of the hair themselves.

Other techniques are available for the DNA analysis of human hairs, if there is insufficient root material present, such as mitochondrial DNA testing.

8.1. Hair Comparisons

Hairs recovered from items can be compared with respect to their length, colour and microscopic appearance with a reference sample of hairs from a given individual. If the recovered hairs are indistinguishable from the reference sample or fall within the range of variation in the reference sample, then it is possible to conclude that they could have come from the same source. If the recovered hairs do not match the reference sample, then it is possible to conclude that either the hairs are not from the same source or that the reference sample of hairs that was provided was not representative of the complete range of hairs from that person. There can be considerable variation in hair length and colour across the head of any one person.

9. Wearer and Contact DNA

Cellular material may be deposited on the surface of an item when it is worn or handled. The amount of cellular material deposited will vary from person to person and also depends on other factors such as the cleanliness of the person's skin, the duration of contact and the nature of the item or surface. It is possible to sample this cellular material from areas that would be expected to have been in close contact with the wearer or handler, for example by using specialist adhesive tape to sample the collar of a jacket, or by swabbing the handle of a screwdriver. The selected sample can then be submitted for DNA profiling analysis.

By comparing the DNA profiles obtained from the cellular material with those from reference samples, it may be possible to identify a potential wearer or handler of the item. In many cases this will be the regular wearer or handler of the item.

If an individual wears an item of clothing or handles an item for a short period of time they may leave little or no cellular material behind, and as a result an incomplete DNA profile or no DNA profile of that person may be obtained. However, if no DNA profile is obtained, it does not mean that a nominated individual did not or could not have worn or handled an item. Furthermore, cellular material may also be removed by washing.

It is usually not possible to determine when DNA was deposited, or if a nominated individual was the last person to wear or handle an item. Therefore obtaining the DNA profile of an individual from an item cannot necessarily be related to a specific time or sequence of events; however, there has been some research into the likelihood of detectable levels of DNA being transferred under various scenarios and it may therefore be possible to provide some assistance in considering the significance of results obtained in relation to any given account.

10. DNA(STR) Profiling

10.1. Introduction

A technique called STR ('Short Tandem Repeat') profiling has been used in this case. This is a form of DNA analysis. DNA is a complex chemical found in most cells of the human body. It carries genetic information that determines the physical characteristics of a person and controls the functioning of their body. The information is carried in coded form and half is inherited from each parent. Each person's total DNA complement is unique, although DNA profiling does not enable us to analyse an individual's total DNA. Instead, we look at certain areas of DNA which are known to have high levels of variability between people. In almost all instances, each person's DNA is the same in all of their cells so DNA recovered from blood cells will be the same as DNA from hair roots, saliva or semen.

10.2. DNA analysis systems

DNA profiling uses an amplification technique to target and copy specific areas of DNA. In this case one or more of the following DNA analysis systems were used:

- DNA-17

Seventeen different areas of DNA are amplified. Sixteen of these areas contain STR regions. These are called D10S1248, VWA, D16S539, D2S1338, D8S1179, D21S11, D18S51, D22S1045, D19S433, THO1, FGA, D2S441, D3S1358, D1S1656, D12S391 and SE33. The seventeenth area, known as amelogenin, indicates the sex of the donor of the DNA. Together the results from these regions are called the DNA-17 profile.

- SGM Plus

Eleven different areas of DNA are amplified. Ten of these areas contain STR regions. These are called D3S1358, VWA, D16S539, D2S1338, D8S1179, D21S11, D18S51, D19S433, THO1 and FGA; these areas are the same as the corresponding areas in a DNA-17 STR profile. The eleventh area, known as amelogenin, indicates the sex of the donor of the DNA. Together the results from these regions are called the DNA (SGM Plus) profile. All of the SGM Plus loci can be compared with their corresponding DNA-17 loci, but the additional information in the DNA-17 profile is not used.

10.3. Attributing a DNA profile to a particular body fluid

When an identifiable body fluid is subjected to DNA profiling tests, it is usually the case that the DNA profile obtained, or certain DNA components within a mixed DNA profile, can be attributed to the identified body fluid. In some instances it is not possible to reliably attribute a DNA profile to a particular body fluid in this way, especially in weak samples.

10.4. Indirect Transfer of DNA

It is feasible for DNA from an individual to be transferred onto an object via an intermediary surface or person, a process called indirect or secondary transfer. For example, if individual ‘A’ has deposited DNA onto either the surface of an object or another person's skin and that DNA is not removed by processes such as washing, degradation or transfer, the DNA from individual ‘A’ may then be transferred onto another surface when that surface comes into contact with the intermediary. DNA from individual ‘A’ is now present on a surface with which they have had no direct contact.

There are numerous factors that influence whether or not secondary transfer would occur and whether or not DNA transferred by secondary transfer would persist in sufficient quantities to provide a detectable DNA profile. These factors include:

- how readily a person sheds DNA, as some individuals tend to shed more DNA than others, as well as that person's level of cleanliness;
- which biological materials are available for transfer, in particular body fluids such as saliva which have higher levels of DNA than the outer skin surface;
- the length of time the person who is the source of the DNA is in contact with the intermediate surface;
- the level of contact pressure applied;
- various characteristics of the intermediate surface in relation to how readily it will pick up and retain any DNA that could be transferred.

The amount of DNA subject to the secondary transfer is also affected by what happens to the intermediate surface prior to touching the final surface. These factors include whether or not the surface is washed or wiped, time frames and environmental conditions such as light, heat and humidity.

As only a portion of the available DNA can be transferred in each contact, indirect DNA transfers involving three or more transfers (tertiary or higher transfer) will result in even lower expected levels of DNA on the final surface, and a correspondingly lower expectation that a detectable DNA profile will be obtained.

Although it is generally acknowledged that secondary transfer of DNA can occur, it is not inevitable that it will or that if it does, sufficient DNA will be transferred to produce a DNA profile.

In general, if a high level of DNA matching an individual is detected on an object, it would more readily be explained if it had been deposited as a result of direct contact rather than indirect transfer of ‘touch’ DNA (from the surface of the skin). High levels of DNA could also be explained by direct or indirect transfer from a rich source of DNA such as a body fluid. If only low levels of DNA are detected, the possibility of indirect DNA transfer should be considered more pertinent.

10.5. DNA comparison

A DNA profile obtained from a human body fluid such as blood or cellular material can be compared with the DNA profile of a given person. If the profiles are different then the DNA could not have originated from that individual. If they are the same, then that person, or any other person who has the same profile, can be considered as a possible source of the DNA. The significance of finding such a match may then be assessed.

10.6. DNA Mixtures

A DNA profile which has originated from one individual (a single source DNA profile) usually consists of a maximum of two components for each region of DNA examined. The appearance of more than two components at any one of the DNA regions examined usually indicates the presence of DNA from more than one individual. DNA mixtures can comprise DNA from any number of contributors and may do so in any proportion. The more contributors there are to a mixed DNA profile the greater the number of components present. As the number of contributors increases the more likely it is that coincidental matches will occur. In order to statistically evaluate the significance of matching DNA components, the most likely number of contributors to the mixture must be taken into account.

If a sample consists of more DNA from one contributor than the other(s) then the mixed DNA profile generated from it would include components at a higher level which represent the DNA from the major contributor and lower level components which represent the contribution(s) of DNA from the minor contributor(s); in such a case the DNA profile of the major contributor can be determined. When the proportion of DNA from two or more contributors to a mixture is approximately equal then it may not be possible to determine the DNA profiles of the individual contributors.

As with a single source profile, a mixed DNA profile can be compared with the DNA profile of a known individual, but this will be a more complex process. The complexity of this process will depend on factors such as the number of contributors to the mixture and the amount of DNA that is present. If a person is considered to be a potential contributor to a mixed DNA profile, a statistical method is applied to determine the significance of this finding, by calculating an estimate of the 'likelihood ratio'. The likelihood ratio approach involves considering two propositions, one which includes the named individual as a contributor of DNA to the sample and one which does not include the named individual as a contributor. The resulting likelihood ratio will be an expression of how much more likely the DNA profile is if one of these propositions is true rather than the alternative. A number of different computer programs have been developed to carry out these calculations using sophisticated ‘probabilistic genotyping’ software.

It should be noted that statistical analyses of DNA profiling results, taken in isolation, relate only to the potential source(s) of DNA in a sample, and do not directly address how, when, in what order or from what cellular type the various contributions of DNA originate.

For very weak and or complex DNA profiling results, it may not be possible to make any useful comparison with reference profiles and therefore results of this type may not provide assistance in addressing whether or not an individual could have contributed.

10.7. Additional low-level DNA components

The sensitivity of current DNA profiling processes are such that it is not uncommon to detect additional traces of DNA in samples at a very low level. In any environment occupied by humans, surfaces within that space can acquire traces of human DNA over time. This can be acquired through touching, through contact with other bare skin surfaces or due to the shedding of particulate debris from the human body such as skin flakes and hairs. If a sample of biological material is taken from an object or surface within such an environment, for example by means of swabbing or taping, then it is possible that traces of DNA from the person using that object or occupying that environment will also be recovered along with it.

This is often referred to 'background DNA' as it emanates from pre-existing DNA on a surface which was present before the deposition of the targeted material but recovered along with it.

These traces may be considered by the Reporting Scientist to not have any bearing on the matter under investigation scientifically.

10.8. Issues arising from the use of different STR kits in forensic investigations

The introduction of the DNA-17 short tandem repeat (STR) chemistries into forensic DNA testing in the UK has resulted in the use of several different STR kits. This, together with the continued use of legacy SGM and SGM Plus profiles, has raised some issues which should be considered in cases where profiles generated by different STR kits are being compared. The potential issues are listed here:

- **Incomplete comparison**

The DNA-17 systems test 16 STR loci (plus a sex marker), which include all ten of the loci tested in the previously used SGM Plus system. In cases where an SGM Plus profile needs to be compared to a DNA-17 profile, all of the SGM Plus loci can be compared with their corresponding DNA-17 loci, but the additional information in the DNA-17 profile is not used. Hence, the DNA comparison can be considered to be incomplete. If the source of the DNA is contested then it would be appropriate to attempt a more complete comparison, if possible.

- **Discordance**

All DNA-17 kits test the same set of STR positions (loci), ten of which are also shared with the SGM Plus system. However, there is a small risk that two different kits may give a different result for the same sample at one (or very rarely, two) of the loci tested. This risk is well understood

and has been assessed in a large study carried out by the National DNA Database (NDNAD) using all test kits prior to their approval for NDNAD use.

a) Apparent mismatch caused by discordance ('false negative'): This form of discordance will most commonly be observed when a crime stain profile and a reference profile from the same individual have been tested either with two different DNA-17 kits or with a DNA-17 kit and the SGM Plus system. In these cases, discordance may be observed as a difference between the two profiles at one or (very rarely) two loci even though they originated from the same source. Such close matches between profiles will be identified, either by the NDNAD or by a scientist carrying out a direct comparison, and retesting of one of the two samples with the alternative kit will confirm whether this is a discordance event, or a genuine mismatch.

b) Apparent match caused by discordance ('false positive'): The potential for discordance between kits also raises the possibility, albeit very remote, that a subject could have a profile which appears to match a crime stain profile but does so only as a result of the different kits used. This form of discordance would require that all other loci in both the subject and crime stain profiles match by chance and so this possibility is, in almost every case, extremely remote. In fact, it is estimated that the overall risk of a chance match occurring, whether due to a random individual having the same profile as the subject, or due to a discordance event, is less than 1 in one billion (where full profiles are concerned).

11. Y-STR Profiling

Standard DNA analysis (for example using DNA-17 or SGM Plus DNA profiling methods) provides a profile that reflects the DNA inherited by the individual from both parents. Y-STR analysis is a sensitive DNA profiling technique that provides a profile which reflects the DNA inherited only from the individual's father and therefore, detects male DNA only. The technical term for this inherited set of male Y-STRs is a haplotype.

Y-STR analysis is useful for selectively targeting and characterising male DNA in a very large background of female DNA. For cases involving digital penetration of the vagina, or penile penetration without ejaculation, or where a male assailant may have touched a female in an area that may be stained with her blood, the number of cells deposited by the male will be relatively small in comparison to the number of cells present from the female (e.g. vaginal cells and/or blood from an injury). However, given that the female contribution is essentially invisible to the

Y-STR test, Y-STR testing offers the potential to be able to detect these low quantities of male cells.

Y-STR profiles are produced using a test kit which amplifies 23 different regions of DNA on the Y-chromosome (only found in males), usually resulting in individual components at each area tested, represented as a series of peaks (the haplotype). The positions in which these peaks appear in the profile can be measured and have been found to vary widely between unrelated individuals. The combination of Y-STR DNA components found in one individual is normally inherited without change from father to son. This means that all closely related paternal-line male relatives (e.g. father, brothers, paternal grandfather, paternal uncles, cousins and nephews related through the paternal line, etc.) are likely to share the same Y-STR profile. However, occasional mutation events can happen, which means an individual may occasionally pass on a slightly different Y-STR profile to his son which is then passed on to subsequent generations.

A Y-STR profile prepared from biological material can be compared with the Y-STR profile obtained from a reference profile from any male. If a male's Y-STR profile is different from that of the biological material, then that male can be excluded as a source of the material. If the Y-STR profiles are the same, then that male, together with anyone else who has the same Y-STR profile can be considered as a potential source of the material. The significance of any matches obtained can be assessed by reference to an on-line global Y-STR database called the 'Y Chromosome Haplotype Reference Database' (https://yhrd.org), which is composed of information collected internationally and reflects diverse population groups. The number of matching Y-STR profiles observed in a relevant population dataset on the YHRD is used to provide an estimate of the frequency of that Y-STR profile in that population.

As individuals with the same male lineage are expected to have the same or very similar Y-STR haplotype, clusters of Y-STR haplotypes can form within a given geographical area. The particular circumstances of the case may therefore have a bearing on the significance of any matching Y-STR profile.

12. Criminal paternity

Using standard autosomal DNA techniques, at each area of DNA tested, up to two DNA markers are observed in an individual's DNA profile. One marker will have been inherited from the mother and the other marker will have been inherited from the biological father. By comparing the DNA profile obtained from the child to the DNA profile of the mother, it is possible to determine the child's maternally inherited markers. The remaining markers must be the child's paternal markers inherited from the biological father. If the alleged father is the biological father of the child then he would be expected to match the paternal marker in each DNA test performed. If this is the case, then a statistical evaluation can be undertaken. Alternatively, if several of the paternal

markers are missing from the DNA profile of the putative father, that individual can be excluded as the biological father of the child.

Based on the inheritance patterns of alleles, other questioned relationships can also be considered and evaluated. For example, it is possible to statistically evaluate the DNA profiles of individuals in order to determine the likelihood of them being related as siblings, as opposed to being unrelated. More distant family relationships (e.g. half-siblings, cousins, aunt/uncle to nephew/niece, grandparent to grandchild) are generally too distant for standard autosomal DNA techniques to reliably detect, therefore in these circumstances either reference samples from more closely related individuals should be sought or consideration given to the use of alternative DNA techniques with more direct inheritance patterns i.e. Y-STR analysis, Mitochondrial sequence analysis.

13. Interpretation and evaluation of scientific evidence

13.1. Forensic interpretation and evaluation

The forensic expert can have two different roles to assist in an inquiry; an investigator or an evaluator.

Investigative opinion is usually given by the expert when at least one version of events is missing (e.g. unconscious complainant, 'no comment' interviews etc). In these cases, the expert is unable to provide any weight or strength of support for any given proposition, normally due to the absence of sufficient information provided to them about the case circumstances and/ or the absence of sufficient information or detail about what each party states occurred. Occasionally, possible explanations may be generated by the expert, using their experience and expertise to attempt to account for observations/ test results, or to lead the investigation in a certain direction. The expert will not provide an exhaustive list of possible explanations, but, where appropriate, may describe what they believe to be a scenario which may be likely to generate the same or similar findings as seen in that particular case.

Evaluative opinion is that based on case specific propositions, usually one of which is provided by the Prosecution and one provided by the Defence, and taking into account clear background information. With sufficient information, the expert may be able to provide a weight or strength of support that the findings provide for one version of events over another.

13.2. Evaluation of the evidence

To help evaluate the weight of the scientific findings, the usual procedure is for the expert to consider a pair of relevant propositions based on the prosecution and defence positions and the issues in the case.

To evaluate the weight of any forensic result, the questions that the expert would address would be typically:

a) What is the probability/likelihood of obtaining this result (or finding) if the prosecution version of events were true?
b) What is the probability/likelihood of obtaining this result (or finding) if the defence version of events were true?

The ratio of these probabilities or likelihoods gives guidance on whether the scientific findings support one version of events over the other, and by how much.

It can be seen therefore, that, if either version is missing or is incomplete, the evaluation cannot be carried out, as only half the ratio would exist. Similarly, if the prosecution or defence version of events changes or if details are added, the expert's expectation of obtaining the findings may change and hence the expert will need to re-evaluate the findings using the new versions of events.

13.3. Scale of support

In expressing the evidential significance of the findings, the following scale is commonly used:

- No
- Limited
- Moderate
- moderately strong
- strong
- very strong
- extremely strong support.

14. Supporting Documentation

List of all supporting documentation referred to within this document:

Document name	Document reference)
MG22A	SFR MG22A July 2020
MG22B	SFR MG22B July 2020
MG22C	SFR MG22C July 2020
MG22D	SFR MG22D July 2020
SFR Annex	SFR2 Annex July 2020
Case Management Risk Form	SFR Case Management Risk Form July 2020
National Guidance for Streamlined Forensic Reporting	FCN-SP-MGT-GUI-0003