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From Theory to Practice: The Biological Findings

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Abstract It is about the biological samples from the scene, and its extraction and significance on forensic science.

Keywords: Biological samples, Scene, Blood, Saliva, Sperm, Forensic science.

1 Site inspection: techniques and technologies

What is a physical test? As it can be recorded, collected and preserved? As can be gleaned the information from it? How you should interpret the information obtained?

The judicial inspection consists in the inspection and description of a location where it was committed a crime or a crime, he aims to establish the existence and type of crime, the means and manner of execution of the same, when, how and by whom the offense was committed (articles 348 and 359 of the Italian criminal procedure Code).

The purpose of scientific investigation is to answer three key priorities: "fix" the crime scene, to reconstruct the circumstances of the crime, collect useful in identifying those responsible. On this basis it is understood that, even without having read the specialized narrative or multiple television hit series, the inspection is the most important phase of the whole activity of the forensic geneticist, who must play its role in concert with police forces. An

examination of the scene with light or inexperience leads in the first case to ignore or underestimate the valuable biological material and quickly degradable, and in the second produces, even more seriously, the scenario being compromised or worse, the biological contamination of the existing tracks. This premise is a must to remember that the work of the experts at this stage will be summed up in strictly scientific conclusions so that he can then have probative value.

The complex of scientific investigation activities so begins the so-called survey on crime scenes. We speak in the plural because more often within the same crime is necessary to inspect a variety of environments, both open and closed, to collect the necessary information to the reconstruction of that happened and recover as many elements for subsequent biological investigations.

In this regard it is good to make a note on a problem that emerges about the murders and suicides or assumed. The role of the medical and paramedical staff comes sooner in place is to revive the subjects less than obvious certain death signs: decapitation, state of

advanced decomposition, presence of hypostatic or rigor mortis spots. It is clear however that in most cases you do not reveal these features, the 118 operators endeavor of bodies already cadaverous manipulating them, often in a noninvasive, and altering the scene of the crime; so investigators and forensic scientists do not look more realistic picture of the crime, and the reconstruction of the event, the determination of the times, the recovery of residues and traces on or near the body is difficult and often impossible. Certainly in these cases you can not refer to inexperience, since both the first aid providers both investigators claim the right to carry out their specific skills; is rightful concern, however, the awareness of this problem in our country because they establish guidelines or rules designed to answer that question.

There is no regulation or standardization of the survey in Italy. Rather, they follow the general guidelines based on the expertise and experience of practitioners. The scientific departments of the police in this regard, are the reference point for what concerns the management

of the crime scene, also because of the possibility of using the latest technology. The specific competence in the techniques and knowledge of forensic science, the guarantee to ensure high quality standards of their work, knowledge of safety standards and cooperative spirit with all the investigative staff members are the basic prerogatives because you can challenge themselves in the survey.

1.1 On the scene

First, the scene is frozen with planimetric measurements of the rooms, photographs and video footage of the whole, and then more and more detailed.

The scene has been looking for as much evidence and clues you can collect, and in a normal inspection may be several dozen artifacts. Many of them will prove to be not significant for investigation purposes, while others will cheer those who submit them for analysis, with the firm thought that he had solved the case. In this regard it should be pointed out that a physical



Figure 1. Blood trail on the edge of a plastic bucket; It turned out to belong to the victim of an assault with an ax and then burned alive. The papillary ridges drawn from the blood have identified the fingerprint of the alleged attacker.

Table 1. Indicative average content of DNA recover in some typical forensic biological samples. The amount of DNA is still influenced by environmental factors.

Type of specimen	Amount of DNA
whole blood	20.000-40.000 ng/ml
Track	250-500 ng/cm ²
Sperm	150.000-300.000 ng/ml
post-coital vaginal swab	10-3.000 ng
Training pilifera (with root)	1-750 ng / radice
Training pilifera fall	1-10 ng/radice
Saliva	1.000-10.000 ng/ml
oral tampon	100-1500 ng
Urine	1-20 ng/ml
Bone	3-10 ng/mg
Fabric	50-500 ng/mg

test, a find, can not always be associated with a person, place or object; ie they would not be "individualized". In most cases, in fact there may only be limited to "identify" a physical test, referred to then be able to confirm the maximum compatibility with a subject, and in any case not establish the association to one and only one subject but to a group or class. Find textile fibers, a fragment of paint or a trace of blood without being able to extrapolate a highly informative DNA profile are examples of identification. In contrast, a piece of plastic or tape with margin perfectly corresponding to a reference, complete a fingerprint, a DNA profile are individualized tests.

The inspection looking for biological evidence proceeds by chronological phases:

- 1.1.1. observation of the scene;
- 1.1.2. fixation with photographs and video reproductions;
- 1.1.3. execution of sketches and measurement environments;
- 1.1.4. recording and documentation of the location of physical evidence;
- 1.1.5. Research of minimum or latent tracks.

In particular, the search for physical evidence should not be carried out in a confused way or just in the vicinity of the victim. Every detail could be crucial for the reconstruction of the crime. For this reason, the search must be carried out in a systematic manner, using for example a spiral criterion, subdividing

the area into grids let you search by parallel lines or following a centrifugal criterion. Similarly, the inspection must cure before passing progressively larger objects into smaller ones. A methodical research approach thus reduces the amount of energy and maximizes the effectiveness in recovering even the smallest details.

Chi vanta esperienza nei sopralluoghi tecnici conosce molto bene l'importanza della precocità del primo accesso alla zona. Prima si interviene sulla scena, più probabile è che eventuali prove non vengano distrutte e che le prove biologiche presenti in minime quantità possano essere processate velocemente e con maggiore successo. Ciò nonostante è spesso necessario ritornare, anche più volte, sulla scena, ad esempio in seguito a nuovi indizi emersi durante gli esami autoptici, le prime analisi di laboratorio o indicazioni emerse dalle indagini.

At the crime scene may be present a wide variety of organic substrates: blood (Fig. 1), semen, hair, and a wide variety of epithelial cells isolated sources, such as saliva, dandruff, sweat, cigarette filters, dishes and glasses, urine, vomit, feces, or plantar digital fingerprints. The various media average guarantee amount of different cells and otherwise storable (Table 1). fresh tracks allow to obtain genetic profiles also as low as a few cells. On the contrary, from biological sources dated

or corrupted by physical or chemical agents (high temperatures, chemicals and inhibitors of Taq polymerase) is necessary to increase the amount of extracted DNA to increase accordingly the fraction of DNA used, and therefore not degraded, for get profiles. However, if the level of degradation is high they will not be possible to generate genetic profiles, although the biological trace is relatively recent.

The source of DNA that is found more often is bloody nature predominant in cases of violent crime. The saliva instead requires more detailed techniques to be detected, since it is not visible to the naked eye. You search on glasses, cutlery and crockery, bite marks; also it is valuable to identify the forms that its contamination draw in cases of choking, gagging and, typically, within hood of robbery suspects. Also important is the macroscopic analysis of semen traces, especially in cases of sexual violence or suspect that, even before his individualization through DNA. The same procedure is useful, with the techniques of which more later, to define areas soaked with sweat, such as those contained in the attacks in which you grab violently the victim. Finally, there is a wide range of exhibits from which to extrapolate isolated cell matrices. typically of cigarette butts we speak, glasses or cups, dandruff residues, cuffs, collars and underwear, toothbrushes and even fingerprints. Not breaking the hair formations, especially if torn, and then with the root and the hair bulb intact, are sources of large amounts of DNA.

Technologies increasingly purposes allow to increase from year to year, the threshold of sensitivity of the molecular DNA analysis. This is undoubtedly a great advantage for forensic geneticists, since it is now possible to get useful profiles even from minimal sources of biological material. However,

this potential can be a disadvantage since, in the same way of the tracks of interest, also the external contamination are enhanced by laboratory analysis. It becomes essential so that the protection, understood both as a protection of the scene both as self-protection of the operators in the survey. Not infrequently, in fact, the same investigators unknowingly waive their cells or, more often, their fingerprints.

1.2 Scene Safety

A safe scene must meet two requirements: to be preserved from people or things that may alter the conditions in which there was the crime itself and be insulated carefully to prevent the place itself can become source of danger to those present. Must be emphasized that safety concerns not only the contamination problems but above all personal safety.

Accidents in private buildings, industrial or public, air or boat, can expose hazardous, chemical or biological risk, or even a combination of several risks. In recent years, for example, the warning against the possibility of mass disasters caused by terrorism is very high. For this reason it should not be allowed to any forensic operator access, until after the

environment has been locked down and only with adequate protection.

1.3 Personal security

It prevents contamination from the operator himself, as we have said. Moreover, intervene on the scene of a crime, especially if it is a violent crime, it means potentially exposing themselves to risks: environmental, chemical and microbiological sometimes, more often organic; Prevention becomes essential, and should be implemented right from access outermost stage wearing sterile coveralls, boots and protective masks and obviously disposable gloves; the romantic image of the coroner in suits narrated in the style of Andrea Camilleri is exceeded (Rutty et al, 2003).

Particular attention and preparation should also be given in cases of suspected terrorist attack, potentially with chemical or bacteriological risk, a condition in which it is necessary the intervention of intervention units trained for such emergencies that adhere to international guidelines specially prepared.

Collect notes and findings at the crime scene is not enough, you must faithfully record what you observe to be able to document in court with



Figure 2. Example of reconstruction of an environment with the 3D rendering technique. These techniques allow you to view more carefully the dynamics of a crime and show more clearly the reconstruction of the event.

as much precision as possible. In this regard it can be made simplified diagrams of the location of objects, bodies and blood stains, even using the photo shoots (called sketching photo). Also today flock to the aid software that allow you to perform faithful reconstructions of the scene and victims based on technical CAD (Computer-Aided Drawing) and two is three-dimensional (rendering), that help for example to better understand the bullets trajectories or blood stains and the incident dynamics (Fig. 2).

2 Collection, storage and archiving of reports

The effectiveness of the presentation of evidence in court is also deeply influenced by the methods of collection and storage of artifacts. Their integrity, both scientifically and legally, must be preserved since the inspection stage. The specific collection methods depend on the state of preservation and the sample conditions. In general, a considerable amount of biological material should always be removed in order to make sure to recover a sufficient amount of DNA for subsequent genetic testing; nevertheless it is good practice to keep an adequate amount of material available to duplicate the analysis or to enable the other parties to be able to perform the same test, when authorized. During the step of collection of the sample it is also crucial to limit the removal of dirt, grease or other materials in the area of unknown nature, as they may prevent some subsequent genetic analysis.

The collection and storage of exhibits are crucial steps of the investigation carried out. In court, in fact, the admission of evidence may be questioned if the test does not fulfill the requirement of an accurate photographic documentation before collection of the specimen; Moreover,

the evidence he has collected or improperly influenced a find and the possibility of having it exposed to contamination can be used to discredit the results of DNA analysis.

Taking into account that today's extraction and PCR systems are quite sensitive, a significant problem can be represented by the phenomena of contamination, especially because they can lead to false exclusions or artificial mixed profiles rather than false inclusions.

Biological samples such as blood, semen, tissue, bones, hair, urine and saliva can be recovered directly from the bodies, from clothing, from the objects or areas of the crime scene. The body fluids are collected by making them adhere to specific cellulosic or synthetic supports (pads or special and sterile filter papers) or aspirated and deposited in test tubes if they are still in the liquid state. Once they have been deposited on a support become "tracks" biological. The finds no fluids, such as hair or tissue, can be removed by direct contact. Findings that are transferred from a person, an object or an environment through an intermediary (person or object) make up the so-called "secondary transfer". secondary transfers may, but not necessarily a direct link between subject and crime. Almost always these findings, also referred to as "micro-traces", contain small quantities of DNA and require more sensitive typifications (low copy number PCR, mtDNA, minisatellites).

In principle, all biological traces found on the scene have or may have as a result of a probative value. Many of them may be subjected to DNA analysis, but not for all will be necessary. Of a "pink" of blood splashing it is certainly not the genetic determinant result of each one, but rather the analysis of the size, the shape and the trajectory (Blood Pattern Analysis, BPA). The techniques and

new technologies now make it possible to bring in large amounts of evidence to the court. Paradoxically, in many cases, this does not help to define the dynamics of a crime. A significant amount of biological traces could burden its analysis and interpretation of results; also it might be a limiting factor, offering to critical defense and observations regarding the exchange of samples, contamination, deviations from the listed protocols, ambiguous interpretation of the results.

Classroom often discussed on a critical element linked to the traces of biological material: the age of the same. The information that provides a spot of blood or sperm, for example, is great but sometimes its meaning can be easily diminished since it is not possible to establish when it has been produced. For example, if during an inspection you can date a track and to show that it is closely associated with the crime in question, it can be dated assumptions the crime itself. Conversely, if you know the exact time of the crime, and you can date a track associated with it, the dating of biological trace itself could exclude the suspect from the accusations. Some efforts have been made in order to estimate the age of a track, especially of the blood spots (Anderson et al, 2005; Alvarez et al, 2006), but it is still too selective methods to be applied to most cases. Although in the near future you will be able to develop or improve techniques for the estimate in question, at present it remains extremely unlikely to run an age of a track evaluation.

3 Finding biological traces

3.1 Forensic light sources

The light is a form of electromagnetic energy of which only a small part of the whole spectrum consists of visible waves, and then white light. The human eye is able to

perceive the entire visible spectrum, from 400 to 700 nm, however, it shows greater sensitivity around 550 nm; the sensitivity is minimum in the violet, below 450 nm, and in the red region, above 650 nm.

The so-called forensic light sources are emission of light systems able to filter the same into individual wavelength bands. This filtration system allows to highlight the detection of evidence through interaction of luminous phenomena that include fluorescence, absorption and oblique light. The majority of biological fluids has natural fluorescence (light emitted only during excitation); if latent, their location, shape and intensity can only be disclosed with forensic light sources.

The first screening in the search for biological traces is performed with the aid of systems equipped with light-emitting lamps in the ultraviolet range

of the visible and capable of enhancing the observation, the photographic recording and collection of artifacts. These instruments (Crimescope CS16, Minicrimescope 400 or Polilight) allow the identification of digital and palm prints, footprints, body fluids (Fig. 3), hairy formations and fibers, contusions, bruises and skin lesions, traces of drugs and even documents or counterfeit money.

The instrument is equipped with a light source (metal halide lamp 400 C), a long liquid waveguide 2 meters wide and 10 mm; Subsequent filters allow you to select individual wavelengths, typically from 365 to 630 nm. Glasses with different filters (white > 400 nm, orange > 550 nm, red > 590 nm) also allow you to use the lamp without incurring damage.

3.2 Microscopy

After macroscopic observation with

the naked eye, the analysis of small traces of presumed biological nature can be significantly enhanced thanks to the use of microscopic techniques. In particular it is commonly employed the stereomicroscope. The main difference between a stereomicroscope and a common optical microscope compound is that, while the second shows the sample from only one direction, the stereomicroscope allows you to see the object from two slightly different angles, in a mode analogous to the human binocular vision. The vision of the objects is based mainly on the use of reflected light and its power typically ranges from 5 to 50X magnification, much less so than a standard optical compound microscope. The use of microscopy is of particular importance in the identification of the origin of hairy formations and their comparison.

3.3 Guidance testing and especially for blood, saliva and semen

A wide range of so-called "presumptive test", or guidance tests, is now available for the analysis of alleged biological traces. A difference of species tests are described further below, the guiding tests do not confirm with certainty the existence or the state the nature of a given biological sample; allow only to exclude the presence of a given substance, since a variety of compounds provides an equally positive result. Because it is not confirmatory tests but exclusion, all tests performed with guidance test must be confirmed by other methods.

Their usefulness for investigation purposes is important not only to skim the large number of non-organic traces that can be found on the scene, but especially for the reconstruction of the dynamics, providing important circumstantial evidence or probative.

These tests must be safe, simple and cheap to be carried out to interpret, the more sensitive as possible so as to minimize the amount of sample



Figure 3. Highlighting a sweat contamination through forensic light source (455 nm). The subsequent DNA analysis has allowed to obtain the genetic profile of the aggressor.



Figure 4. Evidenziazione with Luminol of the presence of latent blood on an apparently clean knife. Hemoglobin human nature was confirmed by immunoassay and DNA extracted from the blade coincided with that of the victim of aggression.

necessary for the test. Finally, the test should not affect subsequent analyzes of extraction and amplification of DNA.

4 Blood

4.1 Catalytic tests

The research methods of the blood traces benefit the activity of the heme peroxidase present in hemoglobin in red blood cells (in a microliter of blood are present up to 5,000 red blood cells).

On the individual nature of alleged blood traces are typically used test strips (Roche Combur Test[®], Hemastix[®]) impregnated with an organic hydroperoxide (dimethyl-dihydro-perossiesano) and a colorimetric indicator (tetramethylbenzidine), which turns from yellow to green-blue if there is hemoglobin which catalyzes the oxidation.

The test is very sensitive, so as to detect the presence of blood diluted up to one hundred thousand times. However there is a wide range of compounds, such as catalase and peroxidase animal or vegetable, detergents containing hypochlorites, metals (especially copper and iron) that have a similar peroxidase activity and can therefore produce false positives.

Alleged latent traces of blood is usually employed the Luminol test. The compound is an alkaline solution (pH 10.4-10.8) of luminol (3-aminoftalidrazina) and sodium carbonate in which the peroxidic component is given by sodium perborate or hydroperoxide (Fig. 4), the latter, however, limits the highlight of the source blood a few tens of seconds. The described solution is sprayed finely on the area (may also be treated very extended surfaces, such as integers domestic environments) and the reaction with hemoglobin produces a bright blue emission more visible in the dark environmental conditions;

Positive reactions can also be obtained if the bloodstains were washed (Fig. 5). As the benzidine test, even the Luminol test produces false positive results if there are peroxidase, hypochlorite and metal oxides. Nevertheless an expert eye can discern between the highly bright luminescence of the blood and the most glittering, uneven and most ephemeral of other substances. considerable limits of the technique are the toxicity of the solution, whose individual components are irritants, the shortness of the luminescent reaction, the difficulty of performing the test on smooth surfaces and on minimal traces that can be hopelessly diluted following the test.

There are also other methods for

the detection of latent blood; some employ fluorescein less sensitive reactions, most indiginose although more durable and feasible under normal light conditions (Tobe et al, 2007). The spread of these substances it should still at the lower impact on the operator's health than the Luminol, although recently has been demonstrated substantial harmlessness (Larkin et al, 2008).

4.2 Test immunocromatografia

The guidance catalytic tests offer the possibility to determine the possible presence of blood, or better of hemoglobin, without however being able to establish the kind of membership. Specific tests for the diagnosis of the human species of the

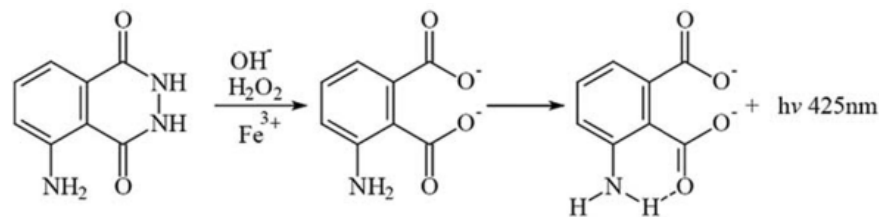


Figure 5 The reaction of luminol in the presence of hemoglobin.

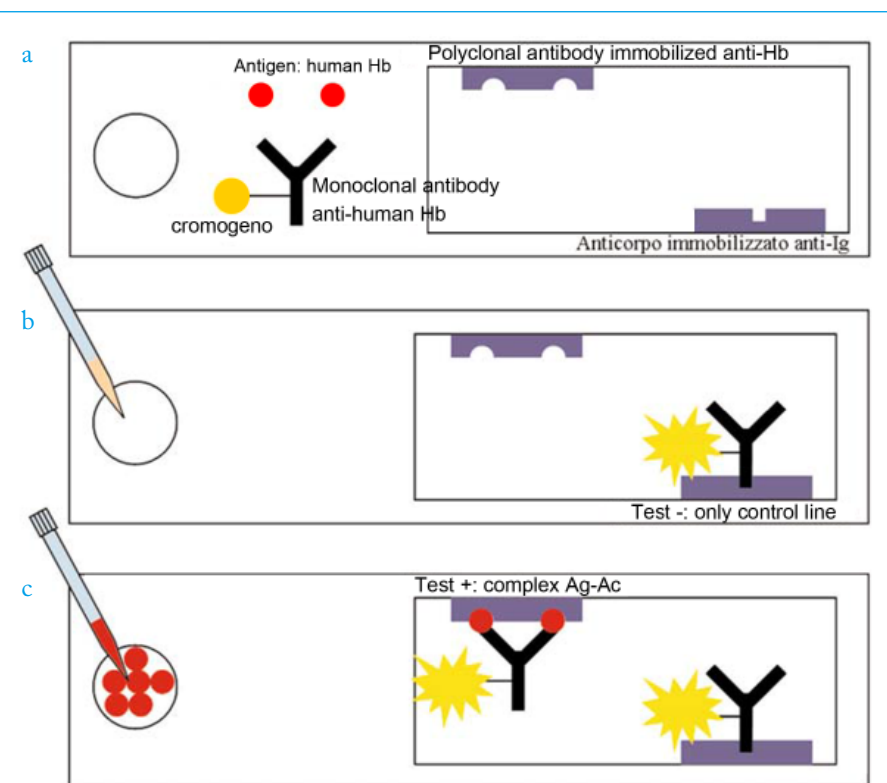


Figure 6 Principle of operation of a chromatographic immunoassay for the detection of human blood. Explanation in the text.

blood consist immunocromatografiche reactions routinely used for occult blood in the stool, and now widely distributed among the laboratories of scientific investigations.

The test utilizes monoclonal antibodies mobile anti-human hemoglobin conjugated with a chromogenic substance (Fig. 6a). After sowing a small aliquot of blood traces, if there is the human blood hemoglobin-antibody complex migrates along the membrane until it meets a test strip on which are immobilized antibodies polyclonal anti-human hemoglobin. The complex concentrates the particles of chromogen forming a colored line in the space of a few minutes (Fig. 6c). The verification that the reaction has proceeded correctly is given by mobile monoclonal antibodies not bound which, continuing the migration to a second test strip with immobilized anti-Ig antibodies, determine a second colored control band (Fig. 6b).

4.3 Histological analysis

The cell analysis of traces of blood may eventually provide information about the origin of the same, if necessary. For investigative purposes could be crucial to know whether it is likely to be blood epistassico (presence of epithelial cells of the nasal mucosa), menstrual blood (presence of the endometrium cells, the epithelium of the vaginal mucosa as well as bacteria) or rectal (mucinous epithelial cells). Recently finer are based on quantitative PCR assays for the analysis of expression profiles of tissue-specific genes tested methodologies to determine the origin of the biological traces.

5 Saliva

The saliva detection, even more the shape and size of the halos that it produces, it may be important for investigative purposes on clothing

(balaclavas, scarves), sheets and pillows, signs due to bites, tape or other items for 'gagging. A strong luminescence is emitted from salivary stains if you look at low wavelengths.

tests are only approximations for dell' α -amylase detection, a digestive enzyme that catalyzes the hydrolysis of α -1,4 glucoside bonds, producing simple sugars. In different isoforms, it is found in high concentrations in saliva (also called ptyalin) and pancreatic juice, but in small amounts can also be found in the sweat, blood, semen, urine and breast milk.

It is possible to evaluate the hydrolytic activity, and therefore the presence, amylase by measuring the optical density of the reaction products. Faster and less expensive, colorimetric and immunological tests are used in clinical chemistry to diagnose acute pancreatitis, and are used in forensics as guidance test. The first is based on the use of a solution containing a substrate, of starch microspheres purified conjugated to chromogenic, whose

hydrolysis with amylase operates in the track produces by-products with optical density such as to be observed with the naked eye, or detected with techniques spectrophotometric. The seconds, of at least two orders of magnitude more sensitive, are immunochromatographic assays with monoclonal antibodies anti-human α -amylase.

The technique allows to obtain very sensitive results, able to detect the presence of a few tens of ng / mcl of amylase, or a few nL of saliva. This represents an undoubted advantage in order to not consume valuable material for subsequent DNA analysis. As the guiding test for blood, also these assays do not allow to date to be able to distinguish a trace of human saliva from that of some animals, eg household rodents. In the market there are also more coarse and less sensitive systems consist of special filter papers already impregnated substrate and chromogen with which it is sufficient to buffer the saliva track to obtain a colorimetric result. The examination of

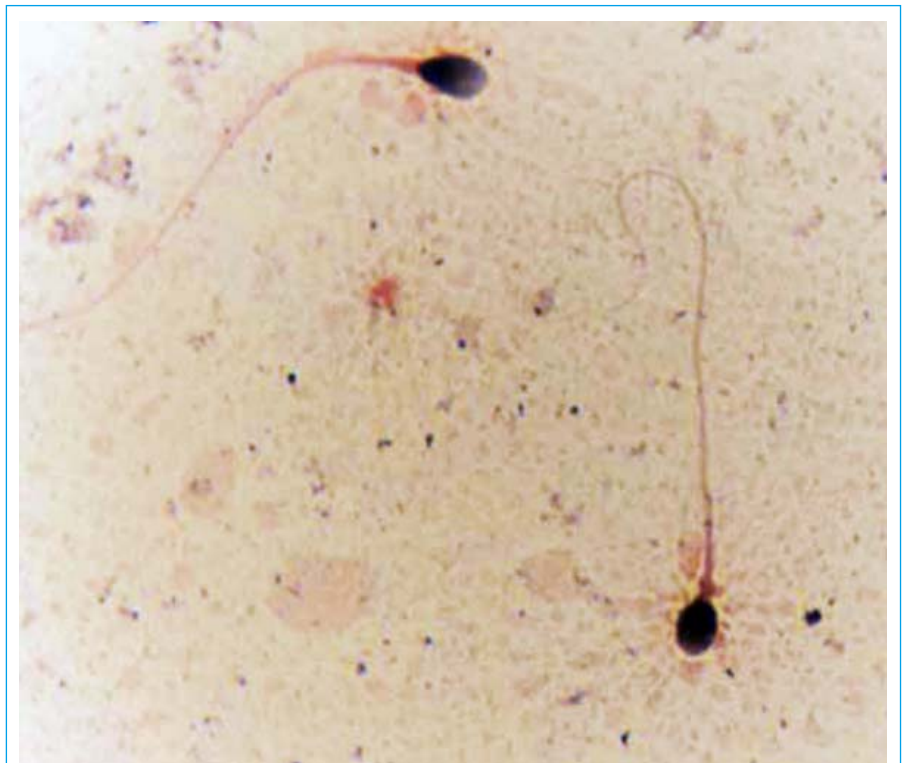


Figure 6 Microfotografia di due spermatozoi. Estratto da traccia su indumento risultata positiva sia all'osservazione con fonte di luce forense, sia al test immunocromatografico per la presenza di p30-, ematosilina-cosina, immersione 1000x.

the DNA can ultimately be considered the most stringent confirmatory test for the presence of human saliva.

6 Sperm

The semen analysis is crucial in cases of suspected sexual violence. Its composition can be simplified to two components, the seminal fluid and sperm. The first consists of a fluid rich in protein primarily produced by the prostate and the seminal vesicles. The seconds are male gametes, or sex cells, which some men produce in very limited quantities or can not produce at all because of birth defects, diseases, vasectomy interventions. For this reason, the semen analysis must always cover research analysis both of the seminal fluid is sperm.

The main source of research of traces of semen are the forensic light sources, since the sperm, along with the saliva, tends to emit more fluorescence than the other body fluids. The areas highlighted by the light source are then tested first with catalytic methods, so immunochromatographic and cytology.

The main screening test for the presence of semen is the detection of prostatic acid phosphatase (PAP) and prostate specific antigen (PSA), prostatic enzymes present in large quantities in semen; in amounts 50-100 times lower it is also present in the blood, saliva, urine and vaginal secretions. This test usually takes α -naphthyl phosphate and diazo blue as colorimetric agent. At pH 5.2 acid phosphatase catalyzes the hydrolysis of the ' α -naphthyl phosphate releasing α -naphthol which reacts with the chromogen salt; positivity is given by the color change to purple.

The samples positive results to the orientation analysis for the presence of seminal fluid may be subjected to specific tests to confirm the presence of sperm, by histological staining or

search for specific proteins of sperm.

There are various staining methods commonly used, although the most common are staining with hematoxylin-eosin (Fig. 7) and the specific staining "Christmas Tree" that uses the nuclear fast red staining (red, stains nuclei of epithelial cells) and picro indigo carmine (green \ blue, colors the cytoplasm). The limiting factors of the cytological detection of sperm are mainly the time elapsed from the time of the attack and the initial quantity of spermatic material, even if the staining "christmas-tree" seems to be more effective than the others.

It is also possible to prepare that immunohistochemical staining, using anti-human sperm monoclonal antibodies, allow to obtain a highly specific confirmatory tests, especially in the case of complex mixed tracks.

Because in rare cases the absence of sperm cytological analysis could not rule out the presence of sperm (eg in subjects oligo- or azoospermic), more specific confirmatory tests are represented by the search for the specific protein of human sperm PSA (prostate antigen specific), also known as p30 (also present in trace amounts in human breast milk and in any tumors of the breast) or semenogelina (Sg), secreted by the seminal vesicles (also present in minute traces in muscle, kidney, colon and in lung cancer).

For some time, they are commercially available immunochromatographic methods for the rapid detection which exploit the presence of immobilized anti-p30 or anti-Sg. These tests are quick (10 minutes), inexpensive and very sensitive (up to 2 ng / mL PSA, dilutions of 50,000 times for Sg).

References

1. Alessandrini F, Cecati M, Pesaresi M et al (2003) Fingerprints as evidence for a genetic profile: morphological study on

- fingerprints and analysis of exogenous and individual factors affecting DNA typing. *J Forensic Sci* 48(3):586-592
2. Allery JP, Telmon N, Mieuxsus R et al (2001) Cytological detection of spermatozoa: comparison of three staining methods. *J Forensic Sci* 46(2):349-351
3. Alvarez M, Ballantyne J (2006) The identification of newborns using messenger RNA profiling analysis. *Anal Biochem* 357(1):21-34
4. Anderson S, Howard B, Hobbs GR, Bishop CP (2005) A method for determining the age of a bloodstain. *Forensic Sci Int* 148(1):37-45
5. Barni F, Berti A, Rapone C, Lago G (2006) Alpha-amylase kinetic test in bodily single and mixed stains. *J Forensic Sci* 51(6):1389-1396
6. Bevel T, Gardner RM (2008) Bloodstain pattern analysis with an introduction to crime scene reconstruction, 3rd edn. CRC Press, Boca Raton, Florida
7. Fisher BAJ (2004) Techniques of crime scene investigation, 7th edn. CRC Press, Boca Raton, Florida
8. Goodwin W, Linacre A, Hadi S (2007) An introduction to forensic genetics. John Wiley & Sons Ltd, Chichester
9. Interpol bioterrorism incident pre-planning and response guide; disponibile online: www.interpol.int/Public/BioTerrorism
10. Jusola J, Ballantyne J (2007) mRNA profiling for body fluid identification by multiplex quantitative RT-PCR. *J Forensic Sci* 52(6):1252-1262
11. Larkin T, Gannicliffe C (2008) Illuminating the health and safety of luminol. *Sci Justice* 48(2):71-75
12. Lee HC, Ladd C (2001) Preservation and collection of biological evidence. *Croat Med J* 42:225-228
13. Mozayani A, Noziglia C (2006) The forensic laboratory handbook. Humana Press, Totowa, New Jersey
14. Pang BC, Cheung BK (2008) Applicability of two commercial available kits for forensic identification of saliva stains. *J Forensic Sci* 53(5):1117-1122
15. Ruttly GN, Hopwood A, Tucker V (2003) The effectiveness of protective clothing in the reduction of potential DNA contamination of the scene of crime. *Int J Legal Med* 117(3):170-174
16. Tagliabracci A, Domenici R, Pascali V, Pesaresi M (2007) Linea guida metodologico-accertativa criteriologico-valutativa. Indagini genetico-forensi di paternità e identificazione personale. Piccin, Padova
17. Tobe SS, Watson N, Daéid NN (2007) Evaluation of six presumptive tests for blood, their specificity, sensitivity, and effect on high molecular-weight DNA. *J Forensic Sci* 52(1):102-109. ■