Human Hair in Personal Identification and Documenting Drug and Substance Abuse

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ABSTRACT Human hair has played a significant role in forensic anthropology for decades. The morphological characteristics of human hair have long been utilized for personal identification in forensic investigations. With the development of better analytical techniques, hairs samples are also being used to understand and solve instances of poisoning, drug and substance abuse. This communication presents a brief outline of the structure of various types of hair, its formation and growth, along with its various types. The issues of hair as an evidence, its transfer, utility and significance of its morphological variables are discussed. The importance of hair in documenting exposure to drug and substance abuse is also highlighted. The main advantages and disadvantages of using hair as evidence are also discussed.

INTRODUCTION

Human hair has a significant potential in forensic anthropology and has been extensively used in forensic investigations. Investigators believe that their presence can associate a suspect to a victim or a suspect/victim to a crime scene primarily because hairs can be transferred from the suspect to the victim and/or vice-versa during physical contact. The types of hair recovered, the condition and number of hairs found in the scene of crime all have great relevance as evidences in criminal investigations. The forensic anthropologist routinely compares the morphological characteristics of the hair samples in question to known hair samples to determine a transfer. Human hair has also been successfully used to assess drug and substance abuse as drugs of abuse cannot be often detected in body fluids. Using very sophisticated analytical techniques such as immunoassay and gas chromatography, investigators are now extensively using human hair to solve cases of poisoning and drug and substance abuse.

Hairs may be defined as slender filamentous outgrowths of the skin and are primarily composed of keratin. It differs from one animal species to another in the basis of length, color, shape, root appearance and morphological characteristics. There is also a considerable deal of variability in the types of hairs that are found on the body of a particular animal. In humans, hairs are distributed on the head, pubic region, arms, legs, and other body areas.

HAIR STRUCTURE

Hair is a biological polymer with over ninety percent of its dry weight being made up of keratin protein. Keratins are cystine-containing proteins, which forms two large groups called Intermediate filament proteins and Intermediate filament-associated proteins (Powell et al. 1991) that are almost equally abundant in most hairs. Of the several types of bonds that stabilize the keratin molecule, the most unique is –S-S-. This –S-S- linkage is formed by two cystine residues contained in adjacent polypeptide chains and it is this disulfide bond that is mainly responsible for keratin’s resistance to destruction. Research work on the elucidation of genes encoding structural proteins of the human hair follicle has advanced rapidly during the last decade and significant developments were made in both the characterization of human hair keratins, as well as the hair keratin associated proteins. Rogers et al. (2006) provides a pertinent review of the keratin proteins found in human hair.

The human hair follicle is a structure, which is formed as a result of epithelio-mesenchymal interactions initiated around the third month of fetal development. It is very complex, consisting of more than twenty different cell types distributed into six main areas. These areas are
the connective tissue sheath, the dermal papilla, the outer root sheath, the inner root sheath, the shaft and the sebaceous gland. This complex appendage behaves in a unique pattern in mammals as, after a hair production phase, it involutes in situ before entering a resting phase after which it renews in a cyclical but stochastic fashion. The hair follicle thus is a fully auto-nomous skin appendage with its own hormonal control, its own autocrine and paracrine network and its own cycle, appearing as an incredibly complex and stable structure that summarizes the main rules of tissue homeostasis (Bernard 2005). As a result, studies on the histo-physiology of the hair follicle are now gaining a great importance (Barinov and Sulaeva 2004).

Structurally, hair consists of: a) an inner cortex comprising spindle-shaped cells, and b) an outer sheath called the cuticle. Each cortical cell consists of many fibrils. In between the fibrils a softer material called the matrix is present which grows from a hair follicle. The fibrils run to form the fiber axis. The cuticle consists of scale-shaped layers and is responsible for much of the mechanical strength of the hair fiber. The cuticle is made up of a number of layers, which varies from one species to another. Typical human hair has six to eight layers of cuticle.

**HAIR FORMATION AND GROWTH**

Hair formation begins in the third month of fetal life. Each hair grows through a follicle and is made up of epidermal cells that grow under the dermis. Initially the epidermis thickens and cells begin to grow down into the dermis. This down growth forms a cap over some of the connective tissue to create papillae whose cells multiply to form the hair. As these cells are pushed up the central canal of the hair shaft and away from their source of nourishment, they become impregnated with keratin. The morphogenesis of most hairs follow a cyclic pattern of cell proliferation and differentiation initiated in mid to late embryonic development and repeated throughout life (Orwin 1979). As hairs undergo a cyclical growth, intermediate and resting phases, the visible morphological characteristics under the microscope are sufficient to determine the phases of growth of the hair.

**The Various Stages of the Hair Cycle**

Each hair passes through three distinct stages called anagen, catagen and telogen. Anagen is the period of active follicle differentiation and hair growth. At the end of this phase, there is a short transitionary phase called the catagen which then leads to the dormant stage referred to as telogen. During the telogen phase, the fully formed hair is retained in the follicle as club hair. In humans, the anagen phase lasts for three to five years, the catagen phase for a few weeks and the telogen phase for two months (Stene 2004). However, although the structure and histology of hair are well known, the molecular events controlling the hair cycle remain unclear (Bernard 1994). Nevertheless, a great deal of research work is currently being undertaken in the various aspects of the hair cycle (e.g. Bayer-Garner et al. 2002; Paus and Foitzik 2004).

The morphological study of the root area of the hair determines the growth phase. About eighty to ninety percent of the hair follicles of the head hair of a healthy individual are in the anagen phase, while only two percent are in the catagen phase and the rest ten to eighteen percent are in the telogen phase. The anagen phase of a new hair starts at the moment it begins to grow. During this time there is very active growth in the hair bulb and materials are deposited in the hair shaft by cells found in the follicle. The medulla, cortex, cuticle, and accompanying root sheath are formed by the metabolically active dividing cells above and around the dermal papilla of the follicle that grow upward during this phase. Hair melanin is produced in the hair bulb throughout this phase of the hair cycle. A short resting phase known as the catagen phase follows the anagen phase. No pigment is made during this phase and the follicle stops producing hair. In the telogen phase, the follicle is dormant. This is the time when a new hair begins to grow from the hair follicle. As it grows upwards the old hair will be shed naturally or may be pulled out, which happens easily and painlessly. These old hairs are the ones that come out when an individual combs the hair. At any one time, around one in ten of the follicles on an individual’s head are in the shedding phase. Once the hair reaches the telogen phase, the follicles have achieved a mature, stable stage of quiescence and are anchored in the follicle only by the club-shaped root. The next generation of an anagen hair is formed by the germ cells below this root. The replacement of human head hair does not have any apparent seasonal pattern, rather it occurs in a scattered mosaic fashion. The
average time period of growth for scalp hair is approximately one thousand days with the resting phase lasting for about hundred days. Around ten percent of the head hairs of a human are, therefore, in the quiescent telogen phase, and a minimal amount of force—such as that from combing—is required to dislodge the hairs from the dormant follicle. The new hair emerges from the same opening at the surface of the skin as the old one, and the hair cycle begins afresh.

Factors Controlling Hair Growth

Hair grows roughly at an average speed of 10 cm/yr (0.3 mm/day). A general good health and nutrition are important for healthy hair growth. Malnutrition, under-nutrition and anemia have strong influences on hair growth. Certain minerals, vitamins, hormones and trace elements are also particularly important for normal hair growth. The levels of androgens are another important factor regulating hair growth (Itami and Inui 2005). Estrogens, on the other hand, slow down hair growth during the growing period. A current review by Conrad and Paus (2004) provides an insight to the effects of estrogens on hair follicle cycling, cutaneous expression of estrogen receptors, and potential functions of estrogens in hair biology. At the onset of puberty the immature vellus hairs on the bodies change to terminal hairs. This change is due to both adrenal and gonadal androgens that exert their biological effects via the androgen receptor, a nuclear transcription factor modulating a specific transcription regulation of largely unknown genes (Hiort 2002).

Types of Hair

The hair follicle present in humans is quite distinct from hair types found in animals. However, even though the basic human hair follicle structure remains the same, the type of hair that is produced can be quite different. Humans have several different types of hair that can be classified depending on their body position and form.

Generally three different hair types are described. They are based on the length and diameter of the hair follicles in the skin and called lanugo, vellus and terminal. Lanugo is the hair type produced in the very first cycle of hair growth when a hair follicle enters shortly after it develops in the embryo. These hairs are fine and soft, and they grow all over the baby’s body and its main function is to retain body heat. They all grow at the same rate and so are of the same length. Around the eighth month of development this hair is usually shed and often a second generation of lanugo hairs then starts growing and lasts until the first three or four months of extra uterine life are completed. These are then replaced by vellus hair. Sometimes incorrectly referred to as lanugo hair, vellus hair is short, fine, “peach fuzz” unpigmented hair. It is very soft, much softer than lanugo and grows in most places on the human body in both sexes. It is usually less than two centimeters long and the follicles are not connected to sebaceous glands. Vellus hair is also present among pre-adolescents (Tanner stage 1).

Terminal hair is developed hair, which is generally longer, coarser, thicker, and darker than vellus hair. The phases of growth in terminal hair are more apparent than in vellus hair and the former generally has a longer anagen phase. Terminal hair contains a large hair follicle and sometimes, a medulla. During puberty many hair follicles in the pubic region, armpits, legs, chest and face, the latter two in case of males, transform from vellus hair to terminal hair under the influence of hormones. Terminal hairs can be further subdivided into different types depending on their nature and/or position of growth on the body. The different types of terminal hair types include hair of the eyebrows, eyelashes, head hair, beard hair, pubic hair and peri-anal hair.

Types of Terminal Hair

Eyebrow hair is a protective patch of hair above the eye. It keeps the eyes free from sweat and other fluids and helps reduce any excessive glare from sunlight entering the eyes. The hair fiber contains a medulla and is curved and coarse. Eyebrow hair is considered to have one of the slowest growth rates of any hair follicle type found on the human body. Eyelash hair is very similar to eyebrow hair. Its growth rate is about the same as for eyebrows. Eyelash hair is very important in protecting the eye from dust and debris. Head hair is generally comprised of terminal hair which grows in a clockwise whorl pattern on the top of the head and merging of this pattern into hair angled downwards and away from the face around the ears and lower back of the scalp. Growth rates are possibly slightly faster in females than males. Head hairs are usually the
longest hairs on the human body and are characterized as having a uniform diameter. The beard develops in response to testosterone/steroidal hormones during puberty in males and is thick, coarse with a triangular cross section. Heavy shouldering or troughs in the hair are observed under magnification. Other characteristics include a wide medulla and a razor-cut tip. The hair follicles of the beard are some of the fastest producers of hair. Limb hairs are shorter in length, arc-like in shape, and often abraded or tapered at the tips. The pigment in limb hair is generally granular in appearance, and the medulla is trace to discontinuous. Pubic hair is a kind of terminal hair that is usually pigmented, short, thick, coarse and kinky. Pubic hair is quite different from head hair as instead of forming a round shape, the hair is oval. They possess a considerable diameter variation or buckling and often have a continuous to discontinuous medulla. The growth period for pubic hair is short and within six months, the hair follicle dies and the hair falls out. Hence, pubic hair never gets a chance to grow longer. Though not much is known about peri-anal hairs, it is generally agreed that these are terminal hairs surrounding the anus. Peri-anal hairs are distinguished from other hair types in the vicinity by their size and their unusually large associated sebaceous glands.

HAIR AS EVIDENCE

It has been recognized for a number of decades that the presence of hairs can associate a suspect to a victim or a suspect/victim to a crime scene. The types of hair recovered, their condition and number of hairs all have a great impact as evidence in a criminal investigation. Comparisons of the morphological characteristics of questioned hairs to known hair samples are usually done to ascertain whether a transfer may have occurred.

Hair Transfer

Physical contact may result in the transfer of hairs. The transfer can directly be from the region of the body where they are growing (primary transfer) or they can transfer from the clothing of individuals (secondary transfer). An individual sheds approximately hundred head hairs each day on clothing and on items in the environment. Contact between a victim and a suspect’s environment can easily cause a secondary transfer of hair. Hairs that are found on the clothing of suspects or victims and appear to have fallen out naturally may be the result of primary or secondary transfer. Hairs that have been forcibly removed may suggest a violent confrontation and this can often happen in cases of sexual assault. A study has reported higher transfer of pubic hair from females to males than from males to females during sexual intercourse (Exline et al. 1998). This prevalence of female-to-male pubic hair transfers suggests the importance of collecting pubic hair combings from the male suspects as well as from female victims, provided the time interval is not extreme. The type of fabric also has a major role in hair transfer (Dachs et al. 2003). It is recognized that generally, hairs persist longer on rougher fabrics. Moreover, wool gives the best chance of recovering samples of hair as the speed at which hair was lost from fabrics decreased in the order polyester, cotton/acrylic, polycotton, cotton, smooth wool, rough wool. In their study on fibre transfer, Palmer and Banks (2005) observed that the greatest degree of secondary transfer occurred with cotton, then acrylic, then wool. The very importance on hair transfer can be gauged from the fact that there have been scientific papers published based on specific individual investigations (e.g. Taupin 1996)

Hair Morphological Studies

The examination of human hairs in the forensic laboratory is typically conducted through the use of light microscopy. The forensic anthropologist compares the various morphological characteristics of the hair samples such as medullation, diameter, form and cross-section. Methods of analyzing these characteristic features were developed decades ago. The initial methodology for understanding hair morphology was developed by Hausman in the 1920s. Subsequently a number of notable studies on hair morphological characteristics were initiated. Such studies include those of Wynkoop (1929) on hair diameter, Fiala (1930) on hair cross-section, Trotter et al. (1956) on hair form, Hrdy (1973) on hair medulla and Das Chaudhuri (1977) on hair cross-section. With some modifications, these methods are still being used today. For instance, Kempson et al. (2002) developed a simple technique is presented for the longitudinal sectioning of hair samples without the need for any embedding medium.
The morphological examination routinely involves a two-step process involving a comparison microscope — (i) the identification of questioned hairs and (ii) the comparison of questioned and known hairs. During comparison, full length of the hairs as well as a maximum number of microscopic features must be considered. The purpose for conducting this examination is to ascertain whether two or more individuals could have come into contact or whether one or more individuals could have come into contact with an object. This associative evidence is particularly useful in crimes of violence where physical contact may have occurred, such as homicide and sexual assault. Crimes such as burglary and armed robbery typically involve the recovery of articles and clothing, which may contain hairs that are useful for the identification of suspects. However, the value and reliability of this evidence is closely related to the variability of hair characteristics between individuals in the population.

The main steps used by the forensic anthropologist in the morphological study of human hair are outlined below:

a) **Body Area Determination**

The body area from which a hair originated can be determined by general morphology. Length, shape, size, color, stiffness, medullar appearance, curliness, and microscopic appearance all contribute to the determination of body area. There is a wide range of interpersonal variation in head and pubic hairs as a result of which, the majority of work in forensic anthropology has been in comparing and differentiating hairs from the head and pubic regions.

Head hairs can appear uncut, with tapered tips but are more often cut with scissors and razors. In general these hairs are subject to more alteration than hairs from other body areas. Alterations can be attributed to the use of chemicals (hair dyes, rinses, permanents and frosts) and also to the environment (exposure to excessive sunlight, wind and dryness). As such, it is recommended that head hair samples be obtained as soon as possible from suspects, victims of crime and possibly from other individuals (elimination samples). Head hair samples obtained years after a crime are generally not suitable for meaningful comparison purposes. The known sample usually contain a random sampling of hair from different areas of the scalp. The number of hairs required for a meaningful comparison may vary depending on the uniformity of characteristics present in the hairs from an individual but it is recommended that at least twenty-five full-length hairs be obtained, including both plucked and combed hairs, packaged separately. Pubic hairs are not subject to as much change as head hairs over time, as a result of which, a sample can still be collected months after a crime for the purpose of suitable and meaningful comparisons. However, it is recommended that a known pubic hair sample be obtained as soon as possible after a crime and should contain at least twenty-five full-length hairs taken from different areas of the pubic region. Pubic hairs commonly may possess tapered tip, but they also be cut with scissors and razors. Presence of facial hairs, more commonly called beard hairs or moustache hairs, on the clothing of a suspect or victim may help establish contact between these individuals. While these hairs may be compared morphologically with the help of a microscope, the significance of the association may not be as great as head hair and pubic hair associations. While limb hairs are not routinely compared in a forensic laboratory, they can differ in appearance between individuals. However, these differences are not considered sufficient to allow limb hairs to be of value for meaningful comparison purposes. The presence of leg or arm hairs on certain items of evidence only corroborates other investigative information.

b) **Racial Determination**

Forensic anthropologists routinely try to establish the racial affinities of the head hair samples. However, it must be cautioned here that it is difficult to assess the racial affinities of head hairs of infants. Moreover, hair samples from individuals of mixed racial affinities may yield microscopic features attributed to more than one racial group.

The main features of hair that are studied are shape of the longitudinal cross section, hair pigmentation, shape of the cuticle, hair texture, medullation and hair form. In case of Caucasoids, the cross section varies from round to oval, with an evenly distributed pigment granules, a colour ranging from blonde to brown to black, medium cuticle, fine to medium coarseness, discontinuous medulla and a wavy hair form. The Negroids exhibit a flat cross section, a dense pigment, a
thin cuticle, an absence of medulla and a curly/woolly/kinky hair form. Negroid hair pigment granules are larger than those found in other racial groups and the shaft exhibits variation or apparent variation in diameter as it is often flattened nature and the manner in which it lies on the microscope slide. Twisting of the hair shaft (buckling) may be present, and the hair shaft may be frequently split along its length. The hairs of Mongoloids show a round cross section and a straight hair form, having a characteristic reddish appearance or tinge. These hairs are often coarse having a wider diameter than those of the other racial groups. The cuticle is usually thicker than that of Negroid and Caucasian hairs, and the medulla is continuous. The hair shaft or cortex of Mongoloid hair contains pigment granules that are generally larger in size than the pigment granules of Caucasian hairs.

c) Age and Sex Determination

The age of an individual is difficult to determine by a microscopic examination. But the microscopic characteristics hair of infants and elderly individuals may provide a general indication of age. It may be mentioned here that hairs of infants are generally finer and less distinctive in microscopic appearance. With the advancement of age, hair may lose pigments, turn gray and also become much finer and more variable in diameter. In many cases, instances of gray hair in head and pubic hair samples were utilized for a general assessment of age. In a significant study, Haga et al. (1995) investigated the appearance age and possible appearance age of gray hair in pubic hair in an attempt to ascertain age of individuals and formulated a five-stage classification. However, the sex of an individual is difficult to determine from microscopic examination of hair. Women generally possess longer, treated hairs than men. Sex can be determined from a forcibly removed hair (with tissue), but this is not routinely done. The staining of sex chromatin in the cells found in the follicular tissue is a better option. With the advent of better techniques, DNA tests can also provide more specific information.

d) Determination of Hair Treatment and Forceful Removal of Hair

Morphological examination can identify whether a hair sample had undergone artificial treatment such as use of dyes. Moreover, as the growth rate of head hairs is known, the investigator can assess the approximate time of this treatment by measuring the length of untreated area of the hair. This is done by a direct, side-by-side comparison of the color of the questioned and known artificially treated hairs. A microscopical study of the hair root determines whether the hair was forcibly removed from the body or shed naturally. Hairs that fall out naturally exhibit a club-shaped root, while a forcibly removed hair will be stretched and may have tissue attached to it. The manner in which a hair was removed has considerable forensic value, especially when there is a possibility of violent contact between a suspect and a victim. Microscopic examination can also be used to establish whether the hair was burned, cut or crushed. The microscopical study of changes in hair roots has been often utilized to solve murder cases (Tafaro 2000).

Conclusions and Significance of the Morphological Study of Human Hair

Forensic anthropologists provide a range of opinions regarding the importance of hair examinations. Some believe that hair cannot be comparable to the specificity of dermatoglyphics and opine that the probability of establishing personal identification from hair is the same as the probability of determining identification using the ABO blood group system. Others believe that hair from two individuals is distinguishable and that no accidental or coincidental matches can take place. If such accidental or coincidental matches occur, these would be referred to as only a relatively rare event. Lamb and Tucker (1994) pointed out that the interpretation of human hair comparisons is highly subjective and there tends to be some caution when evaluating positive evidence. This, they opined was mainly due to a lack of background information relating to the interpretation of the results obtained. Their study was significant because they concluded that comparisons of Afro-Caribbean hairs were possible and a significant proportion of individuals can be excluded by such tests. There are three possible conclusions that can be reached from the microscopic examination and comparison of human hairs. It may be concluded that:

a) The questioned hair and the hairs in the known hair sample exhibit the same characteristics and, accordingly, is associated with the same source of the known hairs.
b) There are differences between the questioned hair and the known hair sample and, accordingly, the questioned hair cannot be associated to the same source of the known hairs.

c) Similarities exist between the questioned hair and hairs in the known hair sample, along with some differences. Hence, it is not definitely possible to determine whether the questioned hair and the known hairs originated from the same source. In such cases, the forensic anthropologist may make a general statement concluding that since it is unusual to find hairs from two different individuals sharing same characteristics, such a microscopic association or match is the basis for a strong association.

While trying to establish a particular association between the hair sample and the victim/accused, the probability that the association (or elimination) was due to coincidence or the probability that the association (or elimination) was due to examiner error and the probability that there is an alternative explanation for the evidence can both be influencing factors. This alternative explanation can be secondary transfer, contamination, or deliberate planting.

Specific case situations can also play an important role. In investigations dealing with sexual offences and rape, it is always important to put a patient at ease prior to making an examination for the collection of seminal stains, hair samples and a general gynaecological examination (Huntington 1976). The broad protocol goals are to minimize the physical and psychological trauma to the victim while maximizing the probability of collecting and preserving physical evidence for potential use in the legal system. Furthermore, there are also established procedures that the hospital staff needs to follow in cases of sexual assault. The procedure followed by the Sexual Assault Nurse Clinician (SANC) in Emergency Departments in Minneapolis, Minnesota (Ledray 1992) may be mentioned here. Similar national hospital/community model protocols for the forensic and medical examination of victims of sexual assault exist in various cities and hospitals in the United States (Young et al. 1992; Sievers et al. 2003). However, in case of sexual assault of children, the guidelines for collection of forensic specimens like hair are not suited, even though the American Academy of Pediatrics recommend-
consistency in examiner discrimination, routine hair classification is not feasible. They have also shown that with the application of rigid selection criteria, the frequency of coincidental matches in forensic science hair comparisons is low.

The importance of hair evidence is also greatly dependent upon its association with a number of compared characteristics, length of compared hair, hair treatment, racial group and most importantly, the number of known hairs with which the sample was matched. A positive hair comparison is strengthened by the presence of two or more mutually dissimilar hairs that are similar to a known sample or by hairs with unusual characteristics or by two-way transfers. Additional examinations of confirmation, such as DNA and sex typing also lay credence to a positive hair comparison. A hair comparison is weakened if the known sample has too few hairs, unrepresentative hairs, incomplete hairs, and a significant temporal difference between the offense and the collection of the known sample. Negative hair comparison conclusions are also weakened by the presence of incomplete questioned hairs and by similarities and differences within the hair sample.

HAIR ANALYSIS IN DOCUMENTING DRUG AND SUBSTANCE ABUSE

Background

It is generally accepted that chemical testing of body fluids is the most objective means of diagnosis and documenting exposure to drug and substance use. However, it has often reported that drugs of abuse cannot be detected in body fluids and as such hair becomes important as a tissue material for analysis of the same. With the development of different analytical methods in recent years, hair began to be utilized in solving poisoning cases and cases of drug and substance abuse. In a very recent review on this subject, Balikova (2005) has pointed out that for over the last twenty years hair analysis for drugs abuse has been gaining increasing attention and recognition in forensic identification, doping control of banned substances and clinical diagnostics in health problems. Hair is also a unique material for the retrospective investigation of chronic drug consumption, intentional or unintentional chronic poisoning in criminal cases, gestational drug exposure or environmental exposure to pollutants and adulterants. Moreover, as hair growth is uniform, segmental hair analysis can provide the information about the time course of the substance use or exposure. Advantages of analyzing hair samples also include their easy, non-invasive collection, the small sample size required for analysis, and their easy storage at room temperature (Daniel et al. 2004). Hair samples have also been collected from dead bodies at the time of postmortem and analyzed for drug content (Drummer 2004). Among the drugs involved, heroin, cocaine, amphetamine, and cannabis are the ones that are most frequently involved in judicial inquiries. The detection of cocaine in hair has been the Federal Bureau of Investigation’s first priority in hair testing for drugs of abuse because of its prevalence.

Techniques and Advantages of Using Hair in Documenting Drug and Substance Abuse

The analysis of hair for drugs of abuse is a powerful tool that can be very useful here in solving such cases (Moeller 1996). In recent years, remarkable advances in sensitive analytical techniques have enabled the analysis of drugs in hair. The main analytical methods that are used to analyze drugs in hair are immunoassay, gas chromatography and mass spectrometry. Improved chromatographic-mass spectrometric techniques with increased selectivity and sensitivity and new methods of sample preparation have now improved detection limits from the ng/mg range to below pg/mg (Pragst and Balikova 2005). Moreover, as hair growth is continuous, the window of drug detection is dramatically extended to weeks, months or even years. A lock of hair the width of a pen tied at the root end is all that is required for a comprehensive drug test and profile (Flanagan et al. 2005). There is a recent review on the current status of hair analysis in drug abuse (Kintz 2004). This review by Kintz (2004) is worth mentioning as in an earlier paper Kintz and Mangin (1995) pointed out that hair was still a seldom used specimen in most laboratories for documenting drug abuse mainly because the scientific community still had reservations about the validity of hair analysis. They had further argued that the areas of agreement and consensus needed to be worked out. Miller et al. (1997) also reported that the Federal Bureau of Investigation only resorts to hair analysis when other information pointed towards drug use and also
when hair analysis could remove a person from suspicion or associate them with criminal activity.

**Kinds of Drugs and Substance Analyzed**

The use of hair for analyzing exposure to substance abuse started in the 1990s. Many employers in the United States started programs to deter drug use by their employees and these programs included the collection and analysis of hair samples (Bost 1993). It may be mentioned here that in Greece, hair analysis for drug abuse has been in use since 1993 and this was in addition to the psychiatric or other forensic clinical examinations necessary for the confirmation of a person’s use of drugs (Tsatsakis 1998). The importance of hair analysis towards the understanding of drug and substance abuse can be gauged from the fact that there have been also a number of research publications on specific cases of such abuse that had been solved by hair analysis. In this context, the studies of Strano Rossi et al. (1998) on heroin abuse, Selavka et al. (1995) on fentanyl consumption, Tagliaro et al. (1998) on morphine abuse, Tassiopoulos et al. (2004) on cocaine abuse, Klys et al. (2004a) on levomepromazine poisoning, Villain et al. (2004) on bromazepam cases, Berankova et al. (2005) on methamphetamine abuse and De Giovanni et al. (2005) on buprenorphine intake may be referred to. Cases related to complex and multiple drug poisoning has been reported by Klys et al. (2004b) and Musshoff et al. (2006).

**Disadvantages of Using Hair in Documenting Drug and Substance Abuse**

In a review, Wennig (2000) pointed out that it is rather difficult to interpret parent drug or metabolite concentrations in hair. This is mainly due to differences in hair growth rate, anatomical region, age, gender, ethnicity and inter-individual variability. Furthermore, as drug incorporation mechanisms into hair matrix is not yet fully understood, it is difficult to extrapolate details on time and dose from hair segment analysis. Cosmetic hair treatment, natural and artificial hair colour, differences in hair structure and specificity of analytical methodology may represent other bias sources affecting concentrations of drugs in hair. It is also opined that the frequency of drug consumption and time intervals between multiple consumption or lag time between consumption and appearance in the hair has not been fully investigated and needs further research. Harkey (1993) pointed out that a basic understanding of hair biology is important necessary so as to interpret the results of hair analysis tests in cases of drug abuse. It is well recognized that hair grows from follicles present within the skin, which has multiple layers of tissue, glands whose secretions bathe hair, and multiple vascular systems. All these he opined are capable of transferring drugs to hair at many levels along the length of the hair shaft.

Moreover, it is difficult to interpret to what extent test results from different laboratories are comparable and how far their results are consistent. For this, in the United States, proficiency testing of crime laboratories began in the mid-1970s and presently assumes an important role in quality assurance programs within most forensic laboratories. Peterson and Markham (1995a) reviewed the origins and early results of this testing program and also highlighted the progress of proficiency testing. They also reviewed the success of laboratories in the identification and classification of some common forensic evidences and concluded that laboratories have more success in determining the origin of finger and palm prints, metals, firearms and footwear and only moderate success in determining the source of bloodstains and hair. In yet another review, Peterson and Markham (1995b) highlighted laboratory proficiency in determining if two or more evidences shared a common source. Furthermore, hair reference materials containing recommended concentrations of drugs that are generally used during the process of determination are also lacking. Very few laboratories have developed such reference materials (Welch et al. 1993). Similarly, there also has to be consensus on the establishment of cut-off values. The cut-off values proposed by Kintz and Mangin (1995) using 30-ng hair samples may be cited here - 0.5 ng/mg in the case of heroin abuse and 1 ng/mg of cocaine in the case of cocaine abuse, which can be decreased to 0.5 ng/mg when use is supported by other evidence of drug intake.

**CONCLUSION AND FUTURE DIRECTIONS**

Hair consists of an inner cortex and a cuticle, and is basically made up of cystine containing keratin protein. Within this protein many bonds are present, the most important of which is a disulfide bond. This disulfide bond that is mainly responsible for keratin’s resistance. The hair follicle is a very complex structure and has its
own sebaceous network and cycle. Hair formation begins in the third month of fetal life and follows a cyclic pattern of growth. This hair growth is influenced by a number of factors such as nutrition and hormones. There are three types of hair in humans - lanugo, vellus and terminal. All these types have their own characteristic features.

Hair has a great role in forensic anthropology as its presence can associate a suspect to a victim or a suspect/victim to scene of crime. One of the main aspects of this investigation is to understand whether any transfer (primary or secondary) had occurred. Then comes the process of comparing the microscopic features of the known and unknown hair samples. The hairs that are routinely compared are head and pubic hairs the hairs. The purpose for conducting this examination is to ascertain whether two or more individuals could have come into contact or whether one or more individuals could have come into contact with an object. For this a maximum number of microscopic features need to be considered. The forensic anthropologist also tries to ascertain the body area of the unknown sample, along with the racial type and age and sex. It is also determined whether the hair sample had undergone artificial treatment and whether the hair had been forcefully pulled out. Forensic anthropologists give a wide range of opinions with regards to hair microscopic examinations. For some, the probability of identification is the same as that obtained by ABO blood groups, while for others it is almost certain.

However, method of hair collection, the evidence processing techniques employed for the examination of the victim and the accused, the methodology used to study hair morphological features and the experiences of the hair examiner are all confounding factors in the final analysis. The other conflicting parameters are the number of compared characteristics, length of compared hair, hair treatment, racial group and the number of known hairs with which the sample was matched. Moreover, recent advances in DNA technology have questioned hair examinations, future studies need to use both hair and DNA technology to arrive at a conviction or exoneration. The utility of hair analysis for drugs and substances of abuse has been well-established. Future studies need to be done to understand the process of incorporation of drug and substances into the hair matrix. The areas of individual variations in hair growth rate, anatomical region, age, gender and ethnicity also need to be addressed. There need to be close cooperation and linkage between different laboratories as questions are often raised regarding the reliability consistency of the results. Reference materials and cut-off values of the discussed variables required with respect to the specific outcomes.

There is the need to develop universally accepted methods of hair collection from the scene of the crime and the evidence processing techniques employed. Similarly the methodology used to study hair morphological features should also need to be looked into. Hair treatment is another area which needs to be addressed. As recent advances in DNA technology have questioned hair examinations, future studies need to use both hair and DNA technology to arrive at a conviction or exoneration. The utility of hair analysis for drugs and substances of abuse has been well-established. Future studies need to be done to understand the process of incorporation of drug and substances into the hair matrix. The areas of individual variations in hair growth rate, anatomical region, age, gender and ethnicity also need to be addressed. There need to be close cooperation and linkage between different laboratories as questions are often raised regarding the reliability consistency of the results. Reference materials and cut-off values of the discussed variables need to be developed.

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REFERENCES


