Balwant Rai Jasdeep Kaur

Evidence-Based Forensic Dentistry



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Preface

Why this book, *Evidence-Based Forensic Dentistry*? This specialized branch of dentistry has the potential to solve the legal issues with oral-dental structures and related issues encountered during forensic investigations. The research and experimentation in this field can all lead to improvements in dentistry. This book draws on our own studies and related studies of other researchers—particularly over the last 9 years.

Identification of unknown individuals and the determination of their age, race, and sex are among the most important functions of forensic odontology. Fundamental applications of forensic dentistry include mass disaster investigations, evaluating bite marks, bite mark evidence in death investigations, and child abuse investigations. The field of forensic odontology has come up with a long approach in recent years. New and unique discussions offer information that will benefit professionals faced with many of the current aspects of the science. This book covers all standard examination procedures of dental evidence, including identification of unknown individuals (age, race, sex). It also includes special chapters on the proper handling of records; writing a legal report about dental age, sex, bite mark, etc.; forensic dental radiography; new methods of dental age estimation; jurisprudence and legal issues; oral fluid in forensic odontology; teeth and oral fluid in toxicology; real cases of dental identification; dental age estimation; bite marks and child abuse, etc.; technological advances in forensic odontology; and oral-dental autopsy. The aim of this book is to include comprehensive, step-by-step instructions on how to practice each stage of forensic odontology and dental toxicology.

The editors and contributors have endeavored to appear objective and rational about the development and future of forensic odontology and dental toxicology and other closely linked forensic and dental disciplines. We can be good forensic experts if well-proven scientific principles are applied from the initial step, and continued throughout, which is important to ensure a successful result. Whether you are a dental examiner, a dentist, a pathologist, a law enforcement officer, or a legal professional who needs to know about the proper handling and evaluation of dental evidence, a legal or police science professional who needs to know how to deal with the proper presentation of dental findings in a court of law, or a dentist who wants to use one's own training and experience in an exclusive, interesting, and challenging way, this book is for you investigators.

We would like to thank our family, especially our parents, and our professors. Finally, we thank the publishers for supporting our venture.

Copenhagen, Denmark Copenhagen, Denmark Balwant Rai Jasdeep Kaur

Contents

1	Forensic Odontology: History, Scope, and Limitations	1
2	Orodental Identification	9
3	Dental Age Estimation	35
4	Craniofacial Identification	65
5	Sex Determination Methods in Forensic Odontology	73
6	Tooth Morphology	81
7	Bite Marks: Evidence and Analysis, Part 1	87
8	Bite Mark Analysis, Part 2: Adobe Photoshop®	101
9	Cheiloscopy in Identification: Forensic Odontology	109
10	Palatal Rugoscopy: An Important Tool in Forensic Odontology	115
11	Forensic Orodental Radiology	119
12	Denture Marking: Forensic Odontology Aspects	127
13	Different Types of Light in Forensic Photography	131
14	Advanced Technologies in Forensic Odontology	141
15	Forensic Odontology in the Management of Bioterrorism	149
16	Management of Mass Disasters: Forensic Odontology	153
17	DNA Technology and Forensic Odontology	163
18	Pink Teeth: An Important Aspect for Forensic Science	169
19	Role of Dentist and Forensic Odontologist in Child Abuse Cases	175
20	Dental Jurisprudence and Ethics	185
21	Oral Fluids and Teeth in Toxicology	189
22	Forensic Odontology Report	201
Ind	lex	205

Forensic Odontology: History, Scope, and Limitations

1

2

2

3

3

3

4

4

4

5

5

6

6

6

1

Contents

1.1	Introduction				
1.2	History				
1.3	Dental Identification				
1.4	Dental Record as a Legal Document				
1.5	Mass Disaster Identification				
1.6	Age Assessment				
1.7	Bite Mark Evidence				
1.8	Child Abuse				
1.9	Facial Reconstruction and Superimposition				
1.10	Sex Determination				
1.11	Cheiloscopy and Palatoscopy				
1.12	DNA Profiling				
1.13	Limitations of Forensic Odontology				
References					

1.1 Introduction

Forensic odontology involves the correct collection, management, interpretation, evaluation, and presentation of dental evidence for criminal or civil legal proceedings: a combination of various aspects of the dental, scientific, and legal professions (Hinchliffe 2011). Forensic dentistry may be defined as the specialized branch of dentistry that applies dental knowledge to civil and criminal problems. The JBR Group in forensic odontology, India (leader and founder, Dr. Balwant Rai) has simply expanded this definition to include the unique needs of the forensic odontologist in medicolegal and other services. While it is true that the primary mission is to support requests for aid in forensic dental identification, we must understand that dental identification is only one of a number of major areas in forensic dentistry. These include dental identification, age estimation, sex determination, cheiloscopy and palatoscopy, molecular biomarkers, bite mark analysis, human abuse and neglect, dental malpractice and negligence, and dental anthropology and archaeology. The forensic odontologist helps legal authorities by examining dental evidence in different conditions. The subject can be roughly divided into three major fields of activity: civil or noncriminal, criminal, and research (Cameron and Sims 1974).

1.2 History (Frness 1970; Keiser-Neilsen 1968, 1980; Valerie and Souviron 1984)

The establishment of forensic odontology as a unique discipline has been attributed to Dr. Oscar Amoedo (considered the father of forensic odontology), who identified the victims of a fire accident in Paris, France, in 1898. The following timeline highlights key moments in the history of forensic odontology:

1453: First reported case of dental identification: The Earl of Shrewsbury, who fell in the Battle of Castillon, was identified.

1775: Dr. Paul Revere, the first forensic odontologist, identified the remains of a victim based on the retrieval of a prosthesis he had constructed.

1849: The first conviction based on dental evidence occurred; the evidence was crowns from charred remains of the victim.

1850: In Boston, Dr. John Webster was convicted of murder based on dental evidence. He was later hanged.

1884: R. Reid, a dentist, read an important paper to the British Dental Association meeting in Edinburgh about the applications of dental science in the detection of crime.

1887: Godon in Paris recommended the use of teeth in the identification of missing persons, depending on accurate records being kept by dentists.

1897: One hundred twenty-six Parisian socialites were burned to death in a few minutes in the Bazar de la Charité. At the request of the consul, who knew many of victims, Dr. O. Amoedo (a Cuban dentist who worked in Paris), aided by two French dentists, Drs. Devenport and Brault, examined and identified many of the bodies. This incident was published as the first text in forensics dentistry on a mass disaster.

1898: Dr. Amoedo wrote his thesis on the value of dentistry in medicolegal affairs; he is universally recognized as the father of forensic odontology.

1932: Edmond Locard recommended the use of lip prints in identification.

1937: A murder trial ended in a conviction based on bite mark evidence for the first time. 1946: Welty and Glasgow devised a computerized program to sort 500 dental records.

1963: Hands, eyes, ears, scalps, and filled teeth had been removed after death to conceal their identity by J. Taylor.

1967: Linda Peacock had a bite mark along with other evidence that led to a young man's murder conviction.

1967: According to Keiser-Nielsen (1967), forensic dentistry is defined as the "proper handling and examination of the dental evidence, in the interests of justice, so that the dental finding may be properly presented and evaluated."

1972/1973: The International Reference Organization in Forensic Medicine and Sciences (I.N.F.O.R.M.) published a compendium of 1,016 references concerning dental identification and forensic odontology compiled diligently by Dr. William and covering over 120 years.

1973: The basic practice of forensic odontology has changed little except that advances in dental materials and laboratory techniques and improvements in scientific and photographic technology have established proof and presentation much closer to forensic science as defined by Harvey. Harvey demonstrate that the fact that the external physical appearance of bitemarks changed with time, and the causative factors precipitating these changes were largely unknown. It will be examined later that the dramatic realization of the possible value of bite marks has assumed a great importance in identification in the last 10 years.

1.3 Dental Identification

Dental identification assumes a main role in the identification of remains when there are postmortem changes, tissue injury, and a lack of fingerprints or other identifying methods. The identification of dental remains is of key significance in cases where the deceased person is decomposed, skeletonized, or burned. The main advantage of dental evidence is that it is frequently preserved after death and not affected by adverse conditions. The basic principles of dental identification are those of comparison, of exclusion, and of making a profile (Rai et al. 2006; Spitz 1993). In spite of the method used to identify a individual, the results of the comparison of antemortem and postmortem data lead to one of these four conditions (American Board of Forensic Odontology 1986):

- Positive identification: Comparable items are sufficiently distinct in the antemortem and postmortem databases; no major differences are analyzed.
- Possible identification: Commonalities exist among the comparable items in the antemortem and postmortem databases, but enough information is missing from either source to prevent the establishment of a positive identification.
- Insufficient identification evidence: Insufficient supportive evidence is available for comparison and definitive identification, but the suspected identity of the decedent cannot be ruled out. The identification is then deemed inconclusive.
- 4. Exclusion: Unexplainable discrepancies exist among comparable items in the antemortem and postmortem databases. Sometimes explainable discrepancies are present, such as changes in restorations related to the passage of time, avulsion of a tooth or teeth secondary to the trauma at the time of death, or additional treatments by a second party that were not registered in the antemortem record. In all these cases, the discrepancies can be explained and identification can still be made.

1.4 Dental Record as a Legal Document

The dental record is a legal document owned by the dentist and all dental professionals and consists of subjective and objective information about the patient. It contains a physical examination of the dentition and supporting oral and surrounding structures, results of clinical laboratory tests, study casts, photographs, and radiographs. It should be kept for 5–30 years. Corrections in the record should not be erased, just corrected with a single line drawn through the incorrect material. Computer-generated (i.e., digital) dental records are becoming more common for dental records. Thus, proper dental documentation is needed for forensic odontology.

1.5 Mass Disaster Identification

Different mass accidents form the majority of cases in which dental identifications are required, mainly aircraft accidents, fires in and collapse of heavily occupied buildings, among others. The forensic odontologist is generally a member of the investigating team, the composition of which varies depending on the nature of the disaster. Usually, teeth and restorations are resistant to heat if they are exposed directly to flame. Preservation is therefore possible in most cases (Rai and Anand 2007a).

Dental identification has been considered one of the main members of the INTERPOL (INTERPOL 2008) disaster victim identification protocol. The orodental structures and dental restorations may be the only parts of the body not affected. The definite establishment of identity of a body essentially comes from a detailed comparison and matching of tangible antemortem records and postmortem findings. It is rarely the case that the two match in all aspects, so some judgment is required.

1.6 Age Assessment (Rai et al. 2009, 2010; Willems 2001)

There are a number of medicolegal reasons and criminal cases necessitating the estimation of an individual's age. Orodental structures can provide useful indicators as to the individual's chronological age. The age of children can be determined by the examination of tooth development and a subsequent comparison with development charts, usually to an accuracy of approximately 2.3 years. The use of attrition and the development of third molars have been suggested as means of aging those individuals over Fig. 1.1 Bite marks



18, but both are unpredictable. Advanced techniques like aspartic acid racemization and translucent dentine have been proposed and have proven to be highly accurate in adult age assessment. tooth impressions caused by the conical shape of these teeth (Spitz 1993). As the introduction of molecular biology to dental identifications, the use of DNA in bite mark cases was found in an effort to eliminate the subjectivity associated with conventional analysis (Pretty and Sweet 2000).

1.7 Bite Mark Evidence

Bite mark evidence may be present in cases of sexual or physical assault by an adult on a child; in rapes or attempted rapes, where bites are likely to be noted on the breasts; in violence between homosexuals; and in family or domestic violence (Pretty and Sweet 2000; Sweet and Pretty 2001). The marks, single or multiple in nature, may be of varying degrees of severity, ranging from a mild marking of the tissues to a deep perforation of the epidermis and dermis, and may be found on breasts, face/head, abdomen, shoulders, upper extremities, buttocks, female genitalia, male genitalia, legs, ear, nose, and neck (Pretty and Sweet 2000; Sweet and Pretty 2001; Vale and Noguchi 1983). Its examination is the one aspect of forensic odontology requiring an immediate response by the forensic odontologist. Sometimes nonhuman bite injuries are found on victims. Animal bites are usually distinguished from human bite injuries (Fig. 1.1) by differences in arch alignments and specific tooth morphology. Dog bites, perhaps the most common nonhuman bite, are characterized by a narrow anterior dental arch and consist of deep tooth wounds over a small area. Cat bites are small and round, with pointed cuspid

1.8 Child Abuse

Abuse of a child by a caretaker is defined as physical force that is outside the acceptable norm of child care. The head and facial areas are regularly injured in such cases (Ambrose 1989). Human bite marks are often seen in abuse cases, frequently attended by other injuries. Those found in infants tend to be on different locations from those in older children or adolescents, and this is unclear because most injuries to children result from punitive measures. The marks may be ovoid or semicircular.

1.9 Facial Reconstruction and Superimposition

If the postmortem profile does not obtain the provisional identity of the deceased, it may be important to reconstruct the person's appearance during life. This is the responsibility of forensic artists and forensic odontologists, who utilize the dental parameters to aid with facial reconstruction (Gatliff 1984) (Fig. 1.2). Antemortem photographs have been used to allow facial

Fig. 1.2 Facial reconstruction

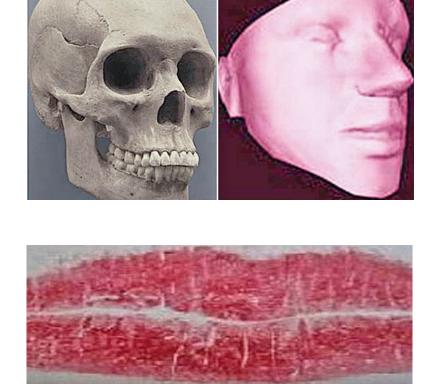


Fig. 1.3 Lip prints

superimposition of skeletal and teeth fractures in cases of identification. This technique requires the availability of appropriate antemortem photographs that show views of the individual's teeth (Austin-Smith and Maples 1994).

1.10 Sex Determination

The dimorphism of the canines, the mandibular canine index, and the BR (Balwant Rai) formula have been used as valid tools in the forensic and legal identification of an individual (Rai et al. 2004). Different molecular techniques have been used for sex determination from teeth. Sex determination from pulpal tissue depends on the presence or absence of an X-chromosome. Sex determination from human tooth pulp in individuals is possible up to a period of 4 weeks after death. But teeth can also serve as an excellent source of genetic material, and nonrestored teeth are even more appropriate for DNA analysis than restored teeth (da Silva et al. 2007).

1.11 Cheiloscopy and Palatoscopy (Caldas et al. 2007; Rai and Anand 2007b)

Lip prints (Fig. 1.3) and palatal rugae patterns (Fig. 1.4) are considered to be unique to an individual and hence hold potential for identifying an individual. Cheiloscopy is appropriate in identifying the living, since lip prints are usually left at crime scenes and can provide a direct link to the suspect. Palatal rugae, also called plica palatinae transverse and rugae palatine, refer to the ridges on the anterior part of the palatal mucosa, each side of the median palatal raphe and behind the incisive papilla. Catastrophic accidents involving plane crashes, fires, and explosions can destroy the fingerprints, but, interestingly, palatal rugae

Fig. 1.4 Palatal rugae patterns



patterns are preserved. Once the palatal rugae are formed, they do not undergo any changes except in length, due to normal growth, remaining in the same position throughout an entire person's life. They help in the identification of an individual.

1.12 DNA Profiling (da Silva et al. 2007)

Different biological materials may be employed for the isolation of DNA to be used in laboratory tests for human identification, including teeth, bone tissue, hair bulbs, biopsy samples, saliva, blood, and other body tissues. Many varied techniques were applied to identify thousands of victims of the 2004 South Asian tsunami disaster, such as forensic pathology, forensic dentistry, DNA profiling, and fingerprinting.

1.13 Limitations of Forensic Odontology

Although there are few shortcomings associated with the various methods used in forensic odontology, the inconsistencies associated with them are to be weighed cautiously to make forensic odontology a more accurate, reliable, and reproducible investigatory science.

 Palatal rugae cannot be used in cases of edentulous mouths, when antemortem records are not available, when palatal pathology exists, and with fire, decomposition, and skeletonization since the palatal rugae are often destroyed.

- Lip prints cannot be used 20 h after the time of death, with lip pathologies like mucocele and clefts, or with any postsurgical alteration of the lips, scars, etc.
- 3. Bite marks cannot be used 3 days after the time of death or with a decomposed or burned body.
- 4. Errors may develop while taking radiographs and photographs, resulting in low-quality documents. Errors may develop in sample collection, processing, and interpretation. Any bacterial contamination and another person's DNA can alter the interpretation.

Conclusions

The contribution of a forensic odontologist in medicolegal proceedings can be of great importance.

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Orodental Identification

Contents

2.1	Introduction	9
2.2	Common Reasons for Identification	10
2.3	Dental Identification	10
2.3.1	Comparative Identification	12
2.3.2	Principles of Dental Identification	25
2.4	Postmortem Profiling Construction	25
2.4.1	Age Estimation	25
2.4.2	Ethnic Group Determination	28
2.4.3	Stature	28
2.4.4	Heredity	29
2.4.5	Oral Manifestations	
	of Systemic Diseases	29
2.4.6	Habit and Identification Markers	29
2.4.7	Other Identification Methods	
	Such as DNA Analysis	30
2.4.8	Bite Marks	31
2.4.9	Facial Reconstruction	31
Refer	ences	31

2.1 Introduction

Forensic odontology is a specialized branch of dentistry that studies the proper handling and examination of orodental evidence as well as the evaluation and presentation of orodental findings in the interests of justice and for medicolegal purposes. Dental identification is an establishment of the individuality of a person either dead or living. Dental identification may be required in living persons in the case of absconding criminals, soldiers, missing persons, impostors, escaped prisoners, etc. Identification may be essential where unclaimed dead bodies are found, when bodies are decomposed beyond recognition, and when grossly mutilated bodies or skeletal remains are found. The use of the unique features of the human dentition to aid in personal identification is well established within forensic odontology. The typical comparative dental identification utilizes postmortem and antemortem dental records to determine positive and negative identification, although in a number of cases, the identification of the individual is unknown because antemortem records cannot be found. In such a case, a dental profile of the individual is developed to aid in searching for the individual's identity by dental indicators of age, ethnicity, habits, professional status, and gender.

2.2 Common Reasons for Identification

Verifying the identity of the deceased is not only important for the family and relatives of the deceased from an emotional standpoint, but it is also a medicolegal requirement. Dental identification of human beings is an important tool in criminal investigations, for social rituals such as burial, and for the identification of individuals missing for prolonged periods. Identification plays an important role in civil cases like insurance claims, matrimonial disputes, property disputes, impersonation, and the issuance of passports and various licenses (Pretty and Addy 2002). The common reasons for identification are as follows (Pretty and Sweet 2001):

- Marriage: Individuals from many religious backgrounds cannot remarry unless their partners have been confirmed dead.
- *Criminal*: Classical forensic investigations into a homicide cannot begin until the victim has been positively identified.
- *Closure*: The identification of individuals missing for prolonged periods can bring sorrowful relief to family members.
- *Monetary*: The payment of life insurances and other government or private benefits relies upon positive confirmation of death.
- *Burial*: Many religions necessitate that a positive identification be made before burial in sacred sites.
- *Social*: Society's duty to preserve human rights and dignity beyond life begins with the basic principle of an identity.

2.3 Dental Identification

Dental identification is very vital tool because teeth may be the only body part that remains intact. Teeth are the hardest substance in the body and can endure extremely adverse conditions (Table 2.1), including high temperatures up to 1,600°C (Andersen et al. 1995). Teeth are the components of the body that often survive severe fires because of their highly resistant composition (Fig. 2.1) and they are protected

Temperature (°C)	Result
100	Whiter to resemble mottled teeth
	Enamel rods dentive altered and root light yellow
200	Crown and root orange
300	Crown yellow brown, root destroyed, cracks in enamel and root dark brown
400	Multidirectional cracks in black-brown crown
500	Crown and root grayish-white; multiple cracks and enamel exfoliates
150	No alteration
175	Longitudinal fissures in incisors and canines
215	Carbonization of pulp
250-300 fibers	Centrilobular destruction of enamel and carbonization of tome
400	Crown of healthy teeth split
800	Reduction in volume of root and carbonization of dentin
1,100	Dentin and enamel retain their narrow canals tome fibers
640	Heat of household furnace
915	Silver amalgam may reach this temperature intact
915–1,090	Gold alloy melts synthetic porcelain faces
1,090	Porcelain teeth may survive
Carrier Transmith	(1072)

Source: From Luntz and Luntz (1973)

by the soft and hard tissues of the face and other materials (Delattre 2000). The macroscopic color of unrestored teeth varies depending on the temperature rise and time of application, from a natural color to black, brown, blue, gray, white, and, finally, pink (Endris and Berrsche 1985). The prismatic structure of the enamel was difficult to identify above 1,100°C, while the dentin tubules were identifiable at 1,150°C (Merlati et al. 2004). Dental restorations and prostheses are also extremely durable and can aid in identification (Røtzscher et al. 2004). It has been reported that glass ionomers were decomposed at 200°C, while the compomers and three different composite materials were deeply altered at 200-500°C (Robinson et al. 1998). A macroscopic

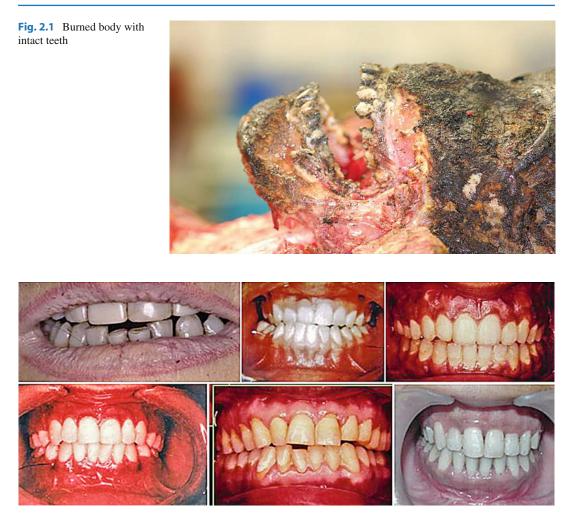


Fig. 2.2 Uniqueness of dentition

observation could identify composite fillings up to 800°C and amalgam fillings up to 1,000°C (Savio et al. 2006). Each dentition is unique to an individual (Pretty and Sweet 2001). The human adult dentition typically consists of 32 teeth, each with five surfaces, thus providing 160 possibilities for individual variations in surface anatomy and dental restorations in configuration, size, shape, material, and wear patterns (Fig. 2.2). This does not even take into account, and is not limited to, such factors as decay, missing and extra teeth, alignment of the dental arches, individual tooth positioning, and prosthetic appliances.

Norrlander (1997) classified body burns in five categories: (1) superficial burns; (2) destroyed

epidermis areas; (3) destruction of the epidermis and dermis and necrosis areas in underlying tissues; (4) total destruction of the skin and deep tissues; and (5) burned remains. Because the destruction of burned victims of the third, fourth, and fifth categories is extensive, remains cannot be identified by common methods. In these cases, forensic odontologists are called in to assist the identification by comparing the postmortem records of the burned, charred, or incinerated individual teeth with the antemortem dental clinical history (Delattre 2000).

Dental identification takes on two main forms:

- Comparative identification,
- Construction of postmortem profile



Anti-mortem record

Post-mortem records

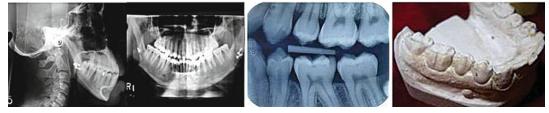


Fig. 2.3 Postmortem and antemortem dental records

2.3.1 Comparative Identification (American Board of Forensic Odontology 2011)

The most commonly performed examination is a comparative identification. This includes the comparison of postmortem dental evidence with antemortem dental records (Fig. 2.3). Generally, when a body or human remains are found, it is reported to the police, who then initiate the request for identification. If this cannot be done by normal forensic methods of identification, then the police or a judge requests that a forensic odontologist perform a dental examination. Often a presumptive or tentative identification of the remains is evident, perhaps from identification cards in the wallet or from photos on a missing persons list, or the antemortem dental records can be located. The forensic odontologist should construct a postmortem record, recording all extraoral and intraoral features, charting all teeth and restorations and pathology findings. If the dentist has antemortem radiographs, photographs, and other dental evidence, then the forensic odontologist should try to compare these with the postmortem evidence.

2.3.1.1 Dental Records

Dental records can be located from family dentists or oral physicians. Thus, it is important for dentists to keep records. The records should be complete, including proper dental charting, radiographs, and photographs from the first visit through follow-ups.

Dental charting: The dental record is considered legal, admissible evidence in the arbitration of contended negligence or malpractice. In many instances, the dental record is the only means of identifying a deceased person. Dentists should have recorded all the information on charts and possible X-rays and models.

Four-quadrant system: Zsigmondy (Zsigmondy 1874) adopted the old system of dividing the teeth into four quadrants and using a system of angles:

8	7	6	5	4	3	2	1	2	3	4	5	6	7	
	1 R	ight						8 L	eft					
8	7	6	5	4	3	2	1	2	3	4	5	6	7	
1 Right							8 L	eft						

1: central incisor

2: lateral incisor

- 3: canine
- 4: first premolar
- 5: second premolar
- 6: first molar
- 7: second molar
- 8: third molar

Universal system: The universal system is very simple in concept: Just number the 32 permanent teeth from 1 to 32. Starting with the third molar in the upper-right quadrant (tooth #1), the teeth are numbered around the arch, so the maxillary left third molar is tooth #16. One then goes down to the mandibular left third molar (#17) and numbers the teeth around the lower teeth, finishing with the mandibular right third molar (#32). The 20 primary teeth are coded alphabetically from A through T. The system must be memorized by rote. If using this system infrequently, it helps to remember that A, J, K, and T are the second molars (at the distal ends of the quadrants) and that E, F, O, and P are the central incisors. Since there are only five teeth per quadrant, one can generally visualize the other tooth codes.

FDI system: Most dentists are right-handed, so quadrant 1 (maxillary right) is closest to the dentist when examining a patient and is scored first, then the upper-left quadrant, then one down to the lowerleft quadrant, finishing with teeth in the lower-right quadrant (Keiser-Nielsen 1971, p. 105). This means the upper-right side (quadrant 1) is the patient's Fédération upper-right side. The Dentaire Internationale's (FDI) description also suggests how to verbalize the system, namely, "The digits should be pronounced separately; thus, the permanent canines are teeth one-three, two-three, threethree, and four-three" (Keiser-Nielsen 1971, p. 1034). So much dental practice attention is spent on the permanent teeth that they are coded as quadrants 1 through 4. The principle is to use numbers 5 through 8 to code the four primary quadrants even though they develop first. This numerical oddity was the subject of considerable discussion by the FDI committee, but it was reasoned that "mainly because deciduous teeth function for such a short time in comparison with permanent teeth that the bulk of dental data to be collected and computerized in the future would obviously concern permanent teeth" (Keiser-Nielsen 1971, p. 1035).

Permanent teeth

18 17 16 15 14	21 22 23 24 25
13 12 11	26 27 28
48 47 46 45 44	31 32 33 34 35
43 42 41	36 37 38

Deciduous teeth

55	54	53	52	51	61	62	63	64	65	
85	84	83	82	81	71	72	73	74	75	

Photography (Hinchliffe 2007): Forensic odontologists take photos to provide evidence of clinical findings in dental identification, abuse, bite marks, and similar cases. It allows reconstruction for forensic odontologists and assists in illustrating findings to juries and dentists. It can be used to explore unsuspected findings by infrared, ultraviolet, and alternative light source photography. Photographs should be sharp, labeled, and with the ABFO No. 2 ruler (Fig. 2.4). Dental evidence photographs should be taken by forensic odontologists because they are specially trained in taking close-ups and collecting all evidence for dental identification.

Radiography (American Board of Forensic Odontology 2011): Radiographs are of principal importance in dental identification. The comparison of antemortem and postmortem radiographs is a major method of dental identification. Radiographs should be of high quality (Fig. 2.5). Digital radiography is an accepted method, whose image quality is equal to or better than conventional X-rays. With digital radiography technology, a digital sensor replaces the conventional film (Dölekog'lu et al. 2011). Three types of sensor systems are commonly used in dentistry. Two types are known as charged-coupled devices (CCD), and the other one is known as a complementary metal oxide semiconductor sensor (CMOS). CCD systems are wireless systems connected to a computer. These technologies allow images to be loaded and viewed on a computer immediately. CMOS is an indirect system using a phosphor plate. It uses the plate to transfer the image from the subject to the plate-scanning device. These screens are reusable. Scanning electron microscopy and energy dispersion X-ray spectroscopy are useful in the recognition and discrimination of tooth materials from other materials. They are mainly useful in decomposed, incinerated, and bite mark analysis (Bush et al. 2008).

Sources of antemortem identification (American Board of Forensic Odontology 2011) include ante-mortem records such as dental records, written

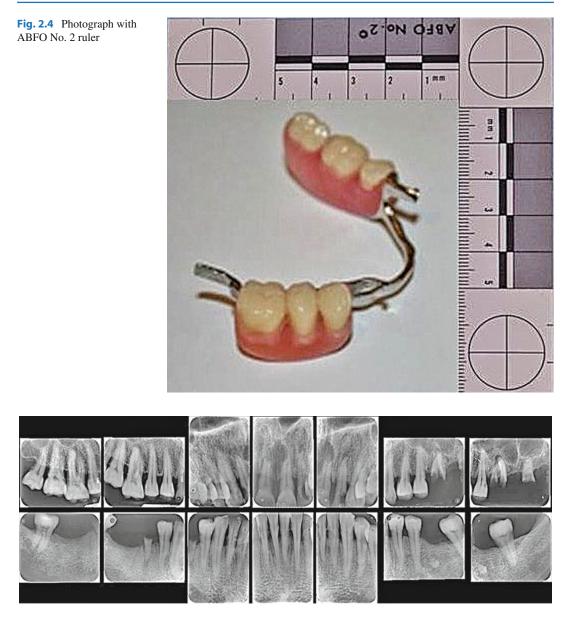


Fig. 2.5 Full mouth radiographic profile

records, and models. Photographs are then obtained from the dentist or oral physician. Collecting antemortem records is the responsibility of the investigating agency that has access to missing and other reports at the local, national, and international levels.

2.3.1.2 Charting, Records, and Databases (American Board of Forensic Odontology 2011)

Dental charting solves the problem of dental identification by giving the forensic odontologist antemortem charts, which can easily and quickly

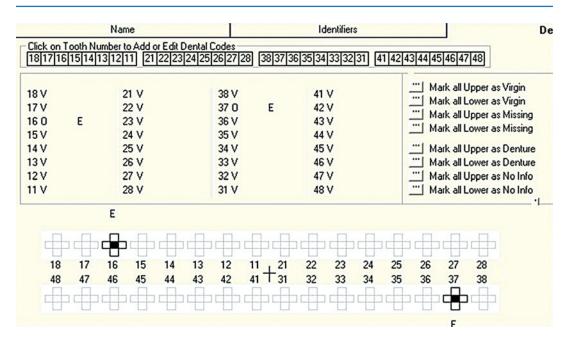


Fig. 2.6 WinID (antemortem)

be interpreted into WinID, CAPMI, and Plassdata. Dentists' shorthand, incomplete documentation, and different charting techniques are big problems for forensic odontologists when comparing antemortem and postmortem data. WinID (Fig. 2.6), CAPMI, or Plassdata charting (Figs. 2.7 and 2.8) should be used to solve this problem.

Clinical records: Complete dental records must be maintained. The ability of clinical practitioners to produce and maintain good dental records is essential for good-quality patient care as well as being a legal obligation. Dentists have a role to play in keeping accurate dental records and providing all important information so that legal authorities may recognize malpractice, negligence, abuse, and identification of victims. In brief, a patient's record includes a complete history, physical examination, diagnosis, treatment, and care (Lawrey 1998). The record may consist of different elements, including written notes, radiographs, study models, referral letters, consultants' reports, clinical photographs, results of special investigations, drug prescriptions, laboratory prescriptions, patient identification information, and a comprehensive medical history. It is essential that a practitioner maintains this information in an easily accessible manner (Collins 1996). The production, retention, and release of clear and accurate patient records is an essential part of a dentist's professional responsibility (Rai et al. 2006).

Content of patient's record: The patient's record consists of the history, physical examination, diagnosis, treatment, and care. The established minimum information in written notes should include (Lawrey 1998):

- *Identification data*: Name, date of birth, address, phone numbers, and emergency contact information;
- *Medical history*: Thorough investigation, to include a minimum of
 - Name and phone number of general physician

Family name Ray MISSIN	
Forename(s) G	C Bex
Date of birth: 09 Day 06 Month 1964 Year	@ Male C Female
86 Dental findings	
R 11 int	int P21# L
12 int	int 22
13 int	int 23
14 int	int 24
15 int	int 25
16 col 0	int 26
18 int	int 28
18 17 16 15 14 13 12 11	21 22 23 24 25 26 27 28
48 int	int 38
47 int	cot O 37
46 int	int 36
45 int	int #351
29 Page 12 - Case no. Ant 1 - D Hondt, Herbert - (Gender: Male	
44 int	int 34
43 int 42 int	int 33
41 int	int 32
	liw Tara
87 Specific data Crowns, bridges and dentures	12 12 12
88 Further data	
Occlusions, attrition, anomalies, smoker, periodontal status, etc.	in the second se
	<u>e</u>
89 X-rays available	
Type, region and year	2
90 Further material	
	24 15
91 Age at time of disapp.	

Fig. 2.7 Plassdata (antemortem)

- Dentist's own evaluation of patient's general health and appearance
- List of systemic diseases, such as diabetes, rheumatic fever, hepatitis, etc.
- Any ongoing medical treatment
- Any bleeding disorders, drug allergies, smoking, and history of alcohol consumption
- Any cardiac disorders
- Relevant family medical history
- Pregnancy
- Physical and emotional tolerance for procedures
- Dental history
- Clinical examination, to include an accurate charting
- Diagnosis
- Treatment plan
- Documentation of informed consent How to create and maintain a patient's record:

Lawrey (1998) describes a simple ten-step

procedure to ensure that patients' records are adequate. A modified and expanded version, appropriate for the National Health Service, UK, is given below:

- 1. Use a consistent style for entries: The appearance of the record is enhanced by using the same color and type of pen and the same abbreviations and notifications, etc.
- Date and explain any corrections: It may be a fatal error in a malpractice case if records appear doctored in any way. These unexplained corrections can undermine the credibility of the entire record and of the treating dentist.
- 3. Do not use correction fluids: Not only is this messy, but it is conspicuous and may indicate that there has been an attempt to hide information.
- 4. Use ink: Pencil can fade and opens up the question of whether or not the records have been altered.

a Plassdata	DEAD BODY	
Nature of disaster :	1 🖉 No:	1
Place of disaster:	India	to:
Date of disaster:	23 12 2010 Year Sex	C Female C Unknown
Page F1 - Case no. TEST 4 - 84 Material	(Gender: Unknown Age: 0/0 Height: 0/0 Weight: 0/0)	
01 Jaws present?	Upper Lower 1 1 2 17	Specimen taken ?
02 Fracmentary remains	Upper Lower 1 T 2 T	Specimen taken ?
03 Single teeth	4 pieces	Specimen taken ?
04 Other		Specimen taken ?
05 Location of specimen		
85 Supplementary de	tails	
Condition of the body		1
Condition of the jaws		
Injuries to - oral soft tissue. - jaws. - teeth.	Intact mandible with two second molar and	d two premolar
Possible cause(s) of injuries.		
Other details		
	- (Gender: Unknown Age: 0/0 Height: 0/0 Weight: 0/0)	-
ragerz caseno. rest 4	(Gender, Onknown Age: 0/0 Height: 0/0 weight: 0/0)	
36 Dental findings		
B6 Dental findings	mis	21
	mis mis mis	21 22 23
R 11 mis	mis	22
R 11 mis 12 mis 13 mis 14 mis 15 mis	mis mis mis mis	22 23 24 25
R 11 mis 12 mis 13 mis 13 mis 14 mis 15 mis 16 mis	mis mis mis mis mis	22 23 24 25 26
11 mis 12 mis 13 mis 14 mis 16 mis 17 mis	mis mis mis mis	22 23 24 25 26 27
11 mis 12 mis 13 mis 14 mis 16 mis 17 mis 18 17 16	mis mis mis mis mis mis mis 15 14 13 12 11 <u>5</u> 21 22 23 24	22 23 24 25 26 27 20 4 25 26 27 28
R 11 mis 12 mis 13 mis 14 mis 16 mis 16 mis 17 mis 18 mis 18 17 16 X X X 2	mis mis mis mis mis mis 15 14 13 12 11 21 22 23 24	22 23 24 25 26 27 28 4 25 26 27 28
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11 mis 12 mis 13 mis 14 mis 15 mis 16 mis 17 mis 18 17 18 17 18 17 18 17 48 47 46 amt 46 amt 46 amt 46 amt 46 amt 44 mis	mis amt VO cca MV mis amt VO cca MV mis amt VO cca MV	22 23 24 24 26 26 27 28 4 25 26 27 28 4 25 26 27 28 4 35 36 37 38 30 37 36 37 36 37 38
11 mis 12 mis 13 mis 14 mis 15 mis 16 mis 17 mis 18 17 18 17 18 17 48 47 46 amis 46 amis 46 amis 43 mis	mis amt VO cca MV mis mis mis mis mis mis mis	22 23 24 26 26 27 28 4 25 26 27 28 4 25 26 27 28 4 35 36 37 38 38 39 37 36 37 36 37 38 37 36 37 38
11 mis 12 mis 13 mis 14 mis 15 mis 16 mis 17 mis 18 17 18 17 18 17 48 47 46 amt 46 amt 46 amt 43 mis 42 mis	mis	22 23 24 24 25 26 26 27 28 4 25 26 27 28 4 25 26 27 28 4 35 36 37 38 38 37 36 37 36 37 36 37 38 37 36 37 38
11 mis 12 mis 13 mis 14 mis 15 mis 16 mis 17 mis 18 17 18 17 18 17 48 47 46 amis 46 amis 46 amis 43 mis	mis amt VO cca MV mis mis mis mis mis mis mis	22 23 24 26 26 27 28 4 25 26 27 28 4 25 26 27 28 4 35 36 37 38 38 39 37 36 37 36 37 38 37 36 37 38
11 mis 12 mis 13 mis 14 mis 15 mis 16 mis 17 mis 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 18 17 16 16 41 mis	mis mis mis mis mis mis mis mis 15 14 13 12 14 13 12 11 21 22 23 24 245 44 44 42 45 44 45 44 46 42 47 31 32 33 345 44 45 44 46 42 12 31 32 33 345 34 45 44 46 43 47 31 32 33 34 34 13 32 33 34 mis mis mis mis mis mis mis mis	22 23 24 24 25 26 26 27 28 4 25 26 27 28 4 25 26 27 28 4 35 36 37 38 38 37 36 37 36 37 36 37 38 37 36 37 38
11 mis 12 mis 13 mis 14 mis 15 mis 16 mis 17 mis 18 17 18 17 18 17 48 47 46 amt 46 amt 46 amt 43 mis 42 mis	mis mis mis mis mis mis mis mis 15 14 13 12 14 13 12 11 21 22 23 24 245 44 44 42 45 44 45 44 46 42 47 31 32 33 345 44 45 44 46 42 12 31 32 33 345 34 45 44 46 43 47 31 32 33 34 34 13 32 33 34 mis mis mis mis mis mis mis mis	22 23 24 24 25 26 26 27 28 4 25 26 27 28 4 25 26 27 28 4 35 36 37 38 38 37 36 37 36 37 36 37 38 37 36 37 38
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Fig. 2.8 (a, b) Plassdata (postmortem)

- 5. Use single line cross-out: This preserves the integrity of the record and shows that you have nothing to hide.
- 6. Write legibly: An illegible record may be as bad as no record at all. Entries that are difficult to read can lead to guesswork by others, and this may not be favorable to you.
- 7. Express concerns about patient needs: By doing this, one is documenting that one has listened, empathized, understood, and acted upon the wishes of the patient. It also enables an explanation to be given should a patient's wishes be unobtainable or unrealistic and can help instantly diffuse a malpractice case.
- Never write derogatory remarks in the record: Superfluous entries only serve to convey a feeling of unprofessionalism and may give doubts to the overall credibility.
- 9. Use only accepted abbreviations for treatments: This is helpful both in a malpractice situation and also when transferring records to a dentist for referral, to obtain prior approval, or to change the dentist of record.
- Maintain a chronological order: The use of a hole punch and metal retainer clips in the top of the record may be helpful to keep loose sheets organized.

Storing radiographs: The most common technique for radiograph storage is in a small envelope with the patient's details, type of radiograph, and data listed on the front. The patient's record can quickly become filled with these envelopes, and establishing a timeline can be difficult and confusing, especially when endodontic films become commingled with diagnostic films. A mounting method can be a more effective solution to radiograph storage. As well as the need for accurate, well-stored and documented radiographs, the frequency of radiographic examination is important. A patient's record that is lacking up-to-date radiographs may jeopardize a malpractice case and is contrary to the patient's best interest (Plunkett 1997).

Use computers to maintain records: Paper records and radiographs can simply be scanned into a personal computer using a desktop scanner.

The most commonly used programs in forensic investigations are Adobe Photoshop 4.0 and Microsoft Program. By using these techniques, the dentist can protect him- or herself from malpractice claims without using valuable physical storage space.

The law concerning the length of time records must be stored varies from country to country, between 2 and 20 years, with 5-10 years in India, for example. Thus, each dentist has a responsibility to understand the forensic implications associated with the practice of his or her profession. This understanding should include more than ethics and jurisprudence, which were traditionally the only aspects of a dentist's knowledge of the law. An appreciation of forensic dental problems permits clinicians to maintain legally acceptable records and to assist legal authorities in the identification of victims of disasters and crimes. Success in this task will assist the dentist should a medico-legal claim be made and can assist the police and crime detectors in the correct identification of individuals.

2.3.1.3 Types of Antemortem Records (American Board of Forensic Odontology 2011; Pretty and Sweet 2001)

Antemortem dental records consist of X-rays and dental charts, described in this section.

X-rays: X-rays are the most vital antemortem records. These are recovered and submitted in forensic investigations. The dentist should submit copies of the X-rays and keep the duplicates. The original film should be a better-quality X-ray than the duplicate. Bitewing, periapical, a full mouth series of X-rays (consists of periapical and bitewing X-rays), a Panoramic X-ray or lateral skull X-ray, CT scan, cone beam CT, and so forth are important tools. Scanned images of dental X-rays or a digital X-ray, saved as bitmap files, can be transmitted electronically when required.

Dental charts: The original dental chart should be obtained if possible. It can be very important, but the forensic odontologist should keep in mind that the original dentist may have

	Negroid	Mongoloid	Caucasoid
Cranial shape	Long	Broad	Medium
Nose shape	Broad	Medium	Narrow
Nasal bone size	Medium/small	Small	Large
Sagittal view	Highly variable, post bregmatic depression	High and globular	High and rounded
Nasal view	Straight to concave	Concave	Straight
Incisor shape	Blade	Blade	Shoveled
Facial profile	Extreme prognathism	Moderate prognathism	Reduced prognathism
Mandible	Gracile, oblique gonial angle	Robust	Medium
Chin shape	Median	Median	Bilateral

 Table 2.2
 Ethnic differences among three groups

Source: From Blumenfeld (2000)

made mistakes when filling out the chart. Therefore, X-rays are a much improved means of making a comparison.

If the dentist has kept dental models or casts, they can be very important in bite mark cases. Additionally, dentists commonly take photographs of their patients' teeth to document their condition. Family snapshots, award receptions, graduations, weddings, or other ceremonial pictures may expose a smile. Dental appliances, such as partial dentures, bite splints, mouthguards, orthodontic retainers, or full dentures are also part of a forensic investigation.

The antemortem forensic record should be recorded in a layout that accurately shows the latest known status of that patient's dental status (Table 2.2).

2.3.1.4 Procedures for Postmortem Dental Record Collections (American Board of Forensic Odontology 2011; R\ptzscher

et al. 2004)

This section describes the appropriate procedures for postmortem dental record collection.

Examination of the oral tissues: A forensic odontologist can help find and identify dental remains that are decomposed, charred, or traumatically mutilated. A fragment of a tooth, a single tooth, or a jaw fragment may be vital to the identification. Postmortem head and neck X-rays (full body X-rays, CT scan) may find dislodged

teeth or fragments. X-ray assessment of muck or charred debris may also help in finding the fragmentary evidence. Depending on the condition of the body, the precise procedures used for the identification may vary. A forensic odontologist may utilize the following techniques in processing a dental identification:

- 1. Normal condition or visually identifiable.
 - (a) A dental examination should be conducted if
 - 1. There are no reports of a missing individual or suspicions as to the identity.
 - 2. No personal effects are found.
 - 3. No fingerprints are done.
 - (b) Dental examination should carry on with no mutilation to the face, that is, no resection or removal of the jaws.
 - (c) Dental examination would comprise photographs of the teeth, a full mouth series of standard dental X-rays with a portable dental X-ray machine, dental charting, and dental impressions and cast construction (if possible).
- 2. Completely decomposed or skeletonized.
 - (a) Simple and easy to work with.
 - (b) Some of the teeth will dislocate from sockets because the periodontal ligaments have been damaged in the breakdown process. All loose teeth should be recovered and replaced in their sockets.
 - (c) Dental examination would comprise photographs of the teeth, a full mouth series of standard dental X-rays with a portable

dental X-ray machine, dental charting, jaw articulation and occlusal analysis, and preservation of remains (if possible).

- 3. Partially decomposed.
 - (a) Difficult to examine due to the odor and presence of insects (e.g., maggots, flies, beetles, etc.).
 - (b) Jaws need to be resected (Fig. 2.9).
 - (c) Dental examination would comprise photographs of the teeth, a full mouth series of standard dental X-rays with a portable dental X-ray machine, dental charting, and preservation of remains (if possible).
- 4. Burned.
 - (a) Access to the teeth is very difficult, as the tissues are very rigid and fragile.
 - (b) Jaws need to be resected.
 - (c) Caution is recommended in resecting seriously burned or calcined jaws because they are extremely fragile. Fixation with clear acrylic spray is recommended.
 - (d) Dental examination would comprise photographs of the teeth, a full mouth series of standard dental X-rays with a portable dental X-ray machine, dental charting, and preservation of remains (if possible).
- 5. Mutilated or traumatized.
 - (a) Teeth and jaws may be fragmented and distorted.
 - (b) Recovery of all the teeth and teeth segments may be difficult.
 - (c) Dental evidence may be imbedded in other areas of the body or scattered about the area.
 - (d) Resection of both jaws.
 - 1. Generally, in cases of complex decomposition, severe mutilation, and bodies that are burned or burned beyond recognition, the jaws must be detached.
 - 2. Resection of the jaws must be accomplished if there is no open viewing of the body.
 - The forensic odontologist can accomplish a more thorough and complete examination, and acquire better-quality X-rays and photographs, by resection of the jaws.

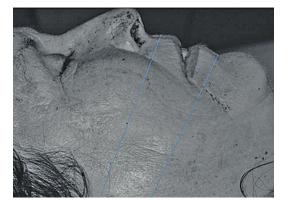


Fig. 2.9 Extraoral incision

4. Jaws should be preserved in 10% formalin if necessary.

2.3.1.5 Forensic Odontology Kit

The forensic odontology kit consists of the following items:

- 1. SLR or digital camera, flash (color and black and white), cheek retractors, mouth mirror, video camera, different types of light sources and filters for UV, IR, and ALI light photography and ABFO No. 2 scale
- 2. Personal protective devices, disposable gowns, face masks, latex and nitrile gloves, eye protection, soap, septula
- 3. Bleach, cynoacrylate, disclosing solution, formalin
- 4. Instruments: mouth probes and mirrors, toothbrushes, explorers, periodontal probes, scalpel handles, hand saw, mallet, millimeter rule
- 5. Radiography: radiography machine, NOMAD, film of various sizes, sensor of radiography, dental X-ray film envelopes, film mounts, processor chemicals, automatic film processor
- 6. Disposables: gauze, cotton roll, cotton swabs, clay
- 7. Software's: Plassdata, WinID, Adobe Photoshop, age estimation
- 8. Documents: antemortem and postmortem Interpol forms, pens, pencils, labels, dental charts, reference materials

9. Other: wire, batteries, computer and printer, magnifying glass, clipboards, plastic denture bags

2.3.1.6 Preparation of Postmortem Records

When taking photographs for postmortem records (Fig. 2.10a–c), the following are needed:

- 1. Full face (frontal and lateral profile): with and without ABFO No. 2 scale
- 2. Close-up of the anterior teeth: with and without ABFO No. 2 scale
- 3. Right and left lateral views of the teeth in occlusion or their proper bite: with and without ABFO No. 2 scale

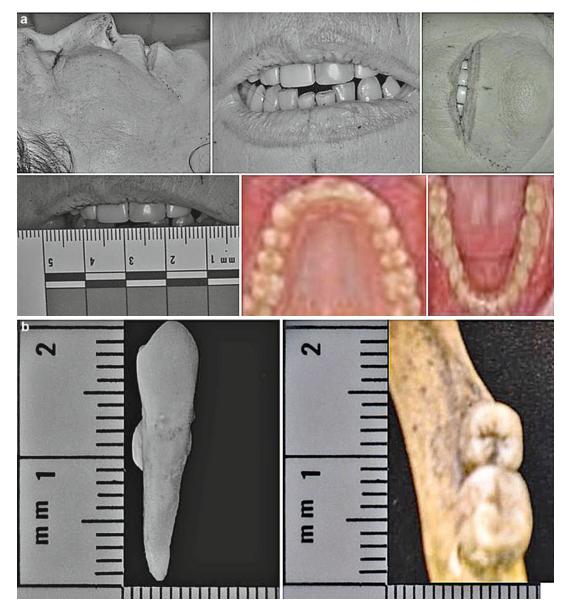


Fig. 2.10 (a–c) Postmortem records

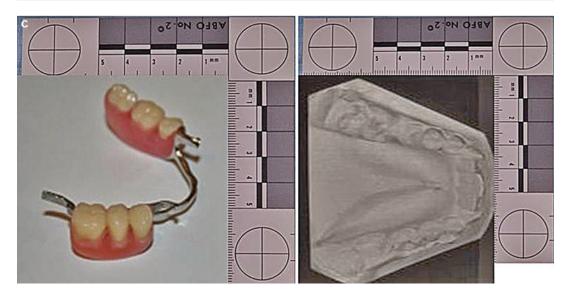


Fig. 2.10 (continued)

- 4. Views of the occlusal of the teeth in both jaws: with and without ABFO-2 scale
- 5. Close-up photography of any additional features that may be important

Radiographic projections useful in identification include periapical, bitewing, occlusal, panoramic, lateral jaw and skull, anterior posterior skull, and temporomandibular joint. At least a series of periapical (whole profile of teeth) of all teeth should be taken. The exposure time should be reduced by a third for resected jaws and by 50% for skeletonized jaws. An X-ray examination is mandatory for estimation of age and identification of the victim.

Notes and Charts

- 1. A dental chart (Interpol or other charting document) should be prepared indicating all significant information. The postmortem forensic record should be recorded in a lay-out that accurately shows the latest known status of that victim's dental status (Table 2.2).
- 2. This information should include, but is not limited to:
 - (a) Present or missing teeth
 - (b) Type of filling and restored teeth
 - (c) Root canals
 - (d) Any prosthetic, orthodontic, or other appliances and implants

- (e) The location and size of the decayed surfaces of the teeth
- (f) Chipped or fractured teeth
- (g) Any malpositions or rotations of teeth
- (h) Unusual anatomy or shapes or anomaly of teeth
- (i) Explanation of an occlusion
- (j) An evaluation of the periodontal profile, including calculus and different types of staining
- (k) Descriptions of any oral-dental pathology or other anatomic features that may be of possible importance
- (1) Sinus morphology
- (m) Pulp stones and pulp anatomy
- (n) Dilacerated roots
- (o) Trabecular patterns, endostoses, and exostoses

Study models: Study models should be taken and models (casts) constructed if indicated. They are indicated not only for teeth but also for abnormal soft tissues. Impressions using silicone (ADA recommendation) can be taken of the victim's teeth and then casts are made by using dental stone. Two sets of casts are made for each impression; each cast should be labeled with the date, forensic odontologist's ID number, and case number. The consulting forensic odontologist will decide if this procedure is necessary.



Fig. 2.11 Important finding in postmortem records

2.3.1.7 Comparison of Antemortem and Postmortem Records

Keep in mind that radiographs and photographs are more consistent compared to written records by dentists because of the possibility of human error in charting notations and the tendency for some dentists only to record the areas requiring attention and not all existing conditions. In some cases, there will be insufficient information in the antemortem records to make a clear comparison. Today digital dental radiographic and photographic superimposition using Adobe Photoshop, WinID, etc. can be used for identification, allowing comparison of the spatial relationships of specific structures of the teeth in antemortem and postmortem records (Pretty and Sweet 2001). Antemortem and postmortem records (Fig. 2.11) must be compared in a methodical and systematic way, examining each tooth (Fig. 2.12) and their surrounding structures. The following features are examined during the comparative identification:

Teeth: The following features of teeth are examined during the comparative identification:

- *Teeth present:*
- (a) Erupted
- (b) Unerupted
- (c) Impacted
- Missing teeth:
- (a) Congenitally
- (b) Lost antemortem
- (c) Lost postmortem

Tooth type:

- (a) Permanent
- (b) Deciduous
- (c) Mixed
- (d) Retained primary
- (e) Supernumerary
- Tooth position:
- (a) Malposition
- (b) Rotated
- Crown morphology:
- (a) Size and shape
- (b) Enamel thickness
- (c) Contact points
- (d) Racial variation
- Crown pathology:
- (a) Caries
- (b) Attrition, abrasion, erosion
- (c) Atypical variations
- (d) Dentigerous cysts
- Root morphology:
- (a) Size
- (b) Shape
- (c) Number
- (d) Divergence
- (e) Dilacerations
- (f) Fracture
- Pulp morphology:
- (a) Size
- (b) Shape
- Pulp pathology:
- (a) Stones and calcification

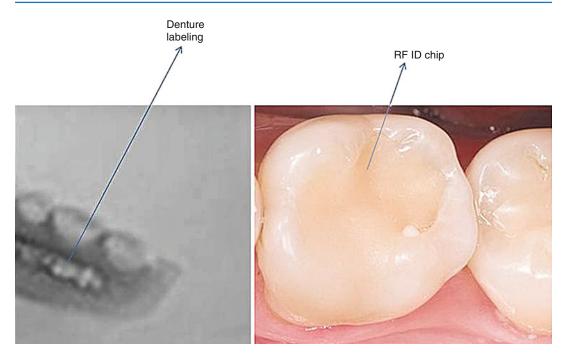


Fig. 2.12 Intact postmortem finding

- (b) Root canal treatments
- (c) Apicectomy

Periapical pathology:

- (a) Abscesses and cysts
- (b) Cementosis
- Dental restorations:
- (a) Metallic
- (b) Full or part coverage
- (c) Implants
- (d) Bridges
- (e) Prostheses

Periodontal tissues: The following features of *periodontal tissues* are examined during the comparative identification:

Gingival morphology/pathology:

- (a) Contour, recession
- (b) Color
- (c) Plaque/calculus

Periodontal ligament morphology/pathology:

- (a) Thickness
- (b) Widening
- (c) Cysts
- Alveolar process and lamina dura:
- (a) Height, contour, and density
- (b) Thickness of interradicular bone

- (c) Exostoses and tori
- (d) Pattern of lamina dura
- (e) Bone loss
- (f) Trabecular bone pattern
- (g) Residual root fragment

Anatomical features: The following features of different anatomical variations are examined during the comparative identification:

Maxillary sinus:

- (a) Size, shape, and cysts
- (b) Foreign bodies and fistula
- (c) Relationship to teeth
- Anterior nasal spine:
- (a) Incisive canal
- (b) Median palatal suture
- Mandibular canal:
- (a) Mental foramen
- (b) Relationship to adjacent structures
- Coronoid and condylar processes:
- (a) Size and shape
- (b) Pathology

Temporomandibular joint:

- (a) Size and shape
- (b) Hypertrophy
- (c) Atrophy

- (d) Ankylosis
- (e) Fractures
- (f) Arthritis
- Other pathologies:
- (a) Developmental cysts
- (b) Salivary gland pathology
- (c) Reactive and neoplastic
- (d) Metabolic bone disease
- (e) Radiopacities
- (f) Evidence of surgery
- (g) Trauma

2.3.2 Principles of Dental Identification

The basic principles of dental identification are those of comparison and exclusion. For example, dental identification is used when antemortem records for the assumed deceased person are available, circumstantial evidence suggests the identity of the decedent, and antemortem records of other suspicious, unidentified persons are accessible and must be ruled out. Identification needs a list of the possible persons involved so that appropriate antemortem records can be placed. The accessibility and accuracy of these records establish the achievement of identification. In spite of the method used to identify a person, the results of the comparison of antemortem and postmortem data direct to one of these four situations (American Board of Forensic Odontology 1986):

- Positive identification: Comparable evidence is sufficiently distinct in the antemortem and postmortem databases; no major differences are observed.
- Possible identification: Commonalities exist among the comparable evidence in the antemortem and postmortem databases, but enough information is missing from either source to prevent the establishment of a positive identification.
- Insufficient identification evidence: Insufficient supportive evidence is available for comparison and definitive identification, but the suspected identity of the decedent cannot be ruled out. The identification is then deemed inconclusive.

 Exclusion: Unexplainable discrepancies exist among comparable items in the antemortem and postmortem databases.

To reach a conclusion, the forensic odontologist must note all similarities and discrepancies (Pretty and Sweet 2001). The discrepancies can either be explained (i.e., relate to the time elapsed) or unexplained (e.g., central incisors not present antemortem but present postmortem).

2.4 Postmortem Profiling Construction

When dental records are unavailable and other methods of identification are not possible, the forensic odontologist can often construct a profile of the general features of the individual (Pretty and Sweet 2001). This process is known as *postmortem dental profiling*. A dental profile will typically provide information on the deceased's age, ethnicity, gender, socioeconomic status, occupation, dietary habits, habitual behaviors, and dental or systemic diseases.

The information that a forensic odontologist can take from dental remains is considerable. The typical information will be described next.

2.4.1 Age Estimation

Different dental age estimation methods have been proposed based on changes in dental structures as follows (Pretty 2003):

The appearance of tooth germs Earliest detectable trace of mineralization Degree of completion of the unerupted tooth Rate of formation of enamel and formation of the neonatal line Clinical eruption Degree of completion of roots of erupted teeth Degree of resorption of deciduous teeth Attrition of the crown Formation of physiologic secondary dentin Formation of cementum Transparency of root dentin Gingival recession

Root surface resorption

Discoloration and staining of teeth

Changes in the chemical composition of teeth

2.4.1.1 Dental Age Estimation Methods in Children

Methods for dental age estimation in children can be divided based on approach (Cameriere et al. (2004), Fishman (1982), Gleiser and Hunt (1955), Haavikko (1970), Kvaal and Solheim (1989), Maples and Rice (1979), Maupome and MacEntee (1998), Olze et al. (2004), Rai et al. (2006a, b, 2007, 2010a, b), Schulz et al. (2008), Stott et al. (1982), Sweet and DiZinno (1996), Ten Cate (1998)).

Atlas techniques: These methods are based on tooth formation and mineralization. The most commonly used dental age estimation atlas techniques are Schour and Massler (1941), Moorrees and colleagues (1963), Gustafson and Koch (1974), and Anderson et al. (1976) techniques. These techniques are based on morphologically different stages of mineralization of all teeth (Willems 2001). Schour and Massler (1941) proposed 20 stages of teeth development, from 4 months until 21 years of age, while another technique (Moorrees et al. 1963) proposed dental maturation of permanent teeth (maxillary and mandibular incisors) divided into 14 stages, from initial cusp formation to apical closure for both genders. Gustafson and Koch's technique proposed tooth development from mineralization, completion of crown formation, eruption, to completion of root formation (1974).

Scoring techniques: Scoring techniques are also based on tooth formation and mineralization staging. The most commonly used dental age estimation scoring techniques are Demirjian et al. (1973), Demirjian and Goldstein (1976), Willems et al. (2002), and Chaillet et al. (2004). Demirjian et al.'s technique proposed eight stages (A–H) of mandibular left teeth. Another method (Nolla 1960) is based on ten stages of teeth in both genders (Rai and Anand 2006; Rai 2008).

Quantity parameters-based technique: This technique is based on open and closed apex of teeth (Cameriere et al. 2006).

Eruption of teeth method or visual methods: The 20 teeth of the primary dentition erupt when the infant is between about 5 and 32 months of age (ADA 2005; Rai et al. 2008).

2.4.1.2 Dental Age Estimation in Subadults

Different techniques have been used for dental age estimation based on third molar staging in subadults (Demirjian et al. 1973; Harris and Nortje 1984; Kohler et al. 1994; Kullman et al. 1992; Mesotten et al. 2002; Mincer et al. 1993; Moorrees et al. 1963; Nortjé 1983; Rai 2009; Solari and Abramovitch 2002).

2.4.1.3 Dental Age Estimation Techniques in Adults

These methods are based on postformation changes in teeth, such as gross anatomical and histological and biochemical changes in teeth. Dental age estimation methods in adults can be divided depending on approach. Various morphologically based methods have been proposed for dental age estimation in adults, including Gustafson (1950), Dalitz (1962), Joanson (1971), Rai et al. (2006), Maples (1978), Bang and Ramm (1970), Solheim (1993), Lamendin et al. (1992), Prince and Ubelaker (2002), the cementum annulations method (Zander and Hürzeler 1958), Kvaal et al. (1995), Cameriere et al. (2007), and Kvaal and Solhiem (1994). Kvaal and Solhiem's method includes both morphological and radiological parameters such as

- 1. Apical translucency in mm (T)
- 2. Periodontal ligament retraction in mm (P)
- 3. Pulp length (PL) (radiograph)
- 4. Radiographic root length on mesial surface (RL)
- 5. Pulp width at cement–enamel junction on radiographs (PWC)
- 6. Radiographic root width at cement–enamel junction (RWC)
- 7. Radiographic pulp width at mid-root (PWM)
- 8. Radiographic tooth width at mid-root on radiographs (RWM)

Biochemical method of age estimation (Ohtani et al. 2003): This method is based on L-enantiomer,

which is transformed into D-enantiomer in enamel, dentin, and cementum.

2.4.1.4 Recommendations for Dental Age Estimation

The American Board of Forensic Odontology (2011) proposed recommendations for dental age estimation as follows:

Children: Recommendations for children are divided into two categories:

- 1. Living: nonextraction, that is, radiographs, such as Moorrees, Fanning, and Hunt, and shed tooth (biochemical analysis), such as aspartic acid racemization
- 2. Deceased: extraction of tooth (biochemical analysis), such as aspartic acid racemization Adolescents:
- 1. Living: nonextraction (radiographs), such as Moorees et al. and Mincer et al., and extracted tooth (biochemical analysis), such as aspartic acid racemization
- 2. Deceased: extraction of tooth (biochemical analysis), such as aspartic acid racemization Adults:
- Living: nonextraction (radiographs/morphological), such as Kvaal and Solhiem, and extracted tooth (biochemical analysis), such as aspartic acid racemization
- 2. Deceased: extraction of tooth (biochemical analysis), such as aspartic acid racemization, and postformation changes (Johanson sectioning, Lamendin et al., Bang and Ramm)

2.4.1.5 Quality Assurance in Dental Age Estimation

The following recommendations are proposed by these authors.

The purpose of the age examination is to

- 1. Estimate the most likely age of the individual
- 2. Make reference to the methods used

All agreed on these steps. We should never give any wrong figures in our reports, especially when it comes to statistics. However, the figures for the distribution of the data must only be taken as indicative for the real variation in the actual population.

As optional steps are that the purpose also is to 3. Describe the likelihood of an official age, if it exists 4. Describe the likelihood of an alternative age if it exists

We recommend that we have to take the official age and the alternative age into consideration, instead of just saying something about the standard deviation. We should express the likelihood of these two ages and if one can be excluded. If we do not do this, it is left to lawyers to argue without proper statistical understanding.

Also optional is the way of arriving at the final estimate by using

- 1. The expert's own assessment
- 2. The person's living conditions and diseases
- 3. The results from scientific statistically reliable methods

As some dentists are not interested in background information, collecting it is also optional, which affects the dental development. There was only agreement on checking the identity of the examined person during the clinical examination. Evaluating the oral mucosa and describing the dentition as far as occlusion, teeth present in the mouth, individual characteristics of the teeth, degree of attrition, color of the teeth, staining and calculus, periodontal conditions, and visual age assessment, solely based upon the teeth, are optional.

It was agreed to carry out a radiographic examination, which should include radiographs that may permit the age estimation methods decided upon and which should describe the dentition and individual characteristics of the teeth. It was also agreed to use as many appropriate parameters as possible, to use methods as originally described in the literature, and to use as many teeth as possible. Using at least two independent statistical methods was left as an option. Finally, the conclusion should end with a complete assessment of the most likely chronological age.

As some of the participants would not make a clinical assessment nor ask for background factors, they naturally had difficulty with taking these factors into consideration in the final assessment of the age. The optional factors were as follows:

- 1. Assess if the methods are appropriate in relation to the individual.
- 2. Assess factors that may have influenced tooth development or ageing.
- 3. Assess especially if pathologic factors or others may have influenced the findings.

2.4.2 Ethnic Group Determination

Ethnic group determination (Table 2.2) is also an important marker of identification. Ethnic group can be evaluated by examining the facial skeleton and comparing the features with the main characteristics of the three racial groups: Mongoloid, Negroid, and Caucasoid (Blumenfeld 2000). Some racial characteristics follow (Blumenfeld 2000; Pretty and Sweet 2001; Rai et al. 2006):

Attrition: Australian Aborigines, American Indians, Greeland Ekimos, Dust Bowl dwellers. Arch

- 1. Regular teeth and large arch: Bushmen and Bantus, Lapp
- 2. Narrow arch and crowding: Europeans

 Wide arch: Chinese in Liverpool Cusp of para molar prostylid: Prima Indians Cusp of cara belli: Australian Aborigines, Pecos

Indians, mixed Greenland Eskimos, Japanese Midline diastema: South African Australoids,

Boskopoids, and Australian Aborigines

Enamel pearls between roots: Pure Greenland Eskimos, Australian Aborigines

Enamel extension between roots of molars: Ait chison, Chinese

Enamel pearls occlusally in premolars: Mongoloids

Early Eruption: Third molars in ligandans, permanent teeth in Zealanders compared with English, maxillary canine in earlier Japanese than Caucasians

Incisors missing in mandible: Mongoloids

Icisors shovel-shaped: American Indians, Finns, Chinese, Japanese, Tibetans, East Greenland Eskimos, Bantus, and Bushmen *Large size of teeth*: American Indians, Australians, and Lemanesians

Mandibular formen shape: In Japanese, Yugoslavs, and Indians, oblique V-shaped fissure

Mandibular angle wide: Maoris, Bushmen, Bantus

Mandibular lingular absent in half-mandibles: Bushmen, Indians, Japanese

Third molars with five cusps: Chinese

Molars with four cusps: Europeans

Third molar missing: Australian Aborigines, Mongoloids

Three-rooted deciduous molars and third molars: Chinese

Third molars in maxilla with three cusps— Bantus, Australian Aborigines

Paramolar: Australians, Negroes, Bushmen *Mylohyoid canal*: Negroes, Italians, Indians *Occlusion*: Edge to edge: Australian

Aborigines; class II: British, Bushmen

Palate V-shaped paraboloid: Japanese

Palate U-shaped: Negroes, Australians

First Mandibular Premolar with second lingual cusp: Negroes

Prognathism: Bantus, Bushmen, Australian Aborigines, Aleuts

Premolar wider mesiodistal and buccolingual compression: Caucasians

Small teeth: Lapps, Bushmen

Torus palatinus: Lapps, Tristans, Greenland Eskimos, Icelanders, Australian Aborigines

Torus mandibularis: Chinese, Aleuts, Icelanders

Taurodont molar: South African peoples, Europeans

2.4.3 Stature

In the identification of unknown human remains, stature estimation (height of a person in the upright posture) is a preliminary investigation (Chiba and Terazawa 1998). It has been reported that a good correlation exists between skull and

Parameters	Gender	Regression equation	S.D. (p value)
Mesiodistal width	Unknown	14.098x+876.312	3.67 (0.005)
of maxillary incisors	Males	9.145x+1245.756	3.89 (0.003)
	Females	-0.234x + 1678.234	3.94 (0.003)
Skull diameters	Unknown	5.999x+678.324	1.23 (0.0002)
	Females	0.567x+1567.879	1.87 (0.003)
	Males	2.456x+1423.256	1.56 (0.002)
Head circumference	Unknown	2.345x+235.167	1.23 (0.05)
	Males	1.359x+987.234	1.12 (0.002)
	Females	0.123x+1458.765	1.09 (0.001)

 Table 2.3
 Regression equation of stature estimation from skull and odontometric parameters from North Indian population

jaw dimensions and stature (Chiba and Terazawa 1998). Rai and Kaur (2011) proposed a regression formulae of stature determination from teeth and skull measurements (mesiodistal crown width of the maxillary anterior teeth (fronto-occipital head circumference, height, and diameter of the skull) on a North Indian population, as shown in Table 2.3.

2.4.4 Heredity

Heredity helps in identification and paternity cases. Amelogenesis imperfecta, taste blindness, Carabelli tubercle in Japanese children, pegshaped lateral teeth, cleido-cranial dysostosis, and osteogenesis imperfecta produce classical heredity deformities.

2.4.5 Oral Manifestations of Systemic Diseases (ABFO 2011; Neville et al. 2010)

Pits in the tips of upper and lower right canine teeth of a young age develop due to hyperplasia or hypocalcification. Marked erosion of the palatal surfaces of maxillary incisors and premolars shows in cases of gastric ulcer, anorexia nervosa, and chronic alcohol abuse subjects. Green-tobrownish yellow discoloration of teeth can be detected in subjects with neonatal jaundice and congenital porphyria. Hutchinson's incisors and mulberry molars can be detected in cases of congenital syphilis.

2.4.6 Habit and Identification Markers (ABFO 2011; Neville et al. 2010)

Musicians damage not only the soft tissue of lips and gums but also the teeth. In pipe smokers and cigarette smokers, lesions are mostly found at the angle of the mouth and are distinctly in the form of a diamond-shaped or irregular gap when the jaws are closed. Partial dentures that are loosely attached to remaining natural teeth with clasps or wires will wear down some parts of teeth. Nervous habits such as chewing or biting on objects, fingernails, or lips can be detected; a special group is typists and secretaries, all of whom may bite on pens. In intensive smoking, a black stain is deposited by cigarette smokers or pipe smokers, but in different sites. With pipe smoking, the stain is accentuated usually on the lingual or palatal surfaces of the front teeth, while in cigarette smokers the stain is more commonly seen on the labial surface of anterior teeth. Drinkers of strong tea often have black stains on their teeth and dentures. Loosening of certain teeth can be found in almost all those who habitually bite, quite often done by people in stressful occupations like airplane pilots, bus drivers, and truck drivers. The labial enamel and later the

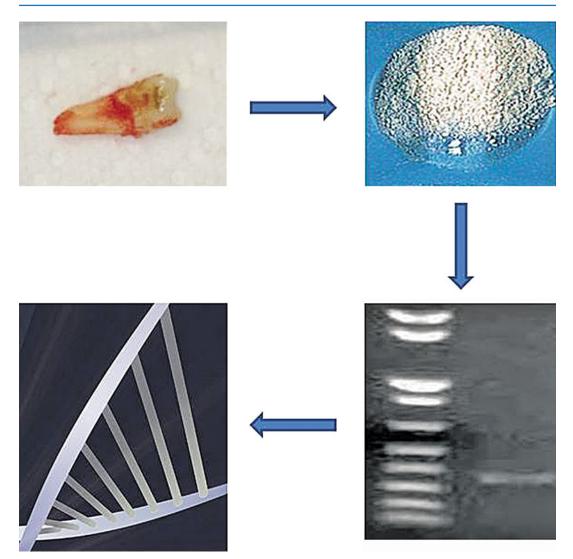


Fig. 2.13 DNA extraction from teeth

exposed dentition are simply dissolved by chemical action such as sulfuric, nitric hypochloride phosphoric acid, and after prolonged exposure nothing remains, so that sharp incisal edges are created with the lingual surfaces still intact. Excessive chewing of acidic foods such as lemons and oranges causes extensive erosion of almost every surface since the acids are held in the mouth for long periods and are well distributed within it. Lead, iron, phosphorus, arsenic, and iodine produce certain characteristic colors of the teeth and gums well known to dental pathologists. Copper causes a green color, while lead and silver deposit a black stain particularly at the neck of teeth or at the marginal part of gums. Bismuth and aniline give a bluish color.

2.4.7 Other Identification Methods Such as DNA Analysis

Teeth are an excellent box of genetic material for proving identity. Their resistance to adverse conditions mentioned before provides us with a second option for dental identification when other dental identification methods fail (Pretty and Sweet 2001). The genetic material obtained from teeth is genomic DNA; this is found in the nucleus of each cell. The DNA is taken from the calcified tissues of the tooth via a grinding technique (Fig. 2.13) (Pretty and Sweet 2001). Forensic odontologists may be required to provide samples for DNA analysis in many medicolegal cases. The sources include saliva, mucosal swabs, and teeth (Malaver and Yunis 2003). These methods consist of the detection, quantification, and analysis of DNA from the nucleus and mitochondria. Nuclear DNA is a marker of both paternal and maternal inheritance. Mitochondrial DNA (mtDNA) is derived from the ovum and so is purely maternal. In teeth particularly, the dentin consists of cellular extensions rich in mitochondria. Dentin powder is therefore presumably a good source of mtDNA (Malaver and Yunis 2003).

2.4.8 Bite Marks

A bite mark expresses a mark sourced by teeth alone or the teeth in combination with other oral cavity parts such as the tongue. These bite marks may be established in the flesh of a victim or in foodstuffs. It may be made in a variety of ways; it can result from direct pressure from the teeth, from tissue being pressed against the teeth by the tongue, or by teeth scraping over tissue. Bite marks are complex injuries, and their recognition and interpretation depend upon an understanding of the mechanisms involved. Bite mark analysis methods have evolved over the years to give more reliable and reproducible results (Hinchliffe 2011).

2.4.9 Facial Reconstruction

Facial reconstruction is a method used in forensic anthropology and odontology to assist in the identification of skeletal remains. The reproduction of the facial features of an individual is based upon the soft tissue thicknesses over various anatomical sites of the skull and jaws (Krogman and Iscan 1986) and is duplicated using modeling clay and computerized.

Conclusions

Forensic odontology plays an important role in the identification of those individuals who cannot be identified visually or by other advanced technologies. The unique nature of orodental features and restorations affords a certain precision when the techniques are correctly employed.

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Dental Age Estimation

Contents

3.1	Introduction	35
3.2	Dental Age Estimation Methods	36
3.3	Factors Used for Age Determination Using Dentition	36
3.4	Basic Principles of Dental Age Estimation	36
3.4.1 3.4.2	Development of Teeth Rate of Formation of Incremental	36
3.4.3	Structures in the Tooth Crown Changes in the Pulpodentinal Complex	37 37
3.4.4	Changes in Chemical Composition	38
3.4.5 3.4.6 3.4.7	Fluorescence of Dental Hard Tissues Epidemiological Criteria Attrition	38 38 39
3.5	Methods of Dental Age Estimation	39
3.5.1	Children	39
3.5.1 3.5.2 3.5.3	Children Subadults Adults	
3.5.1 3.5.2	Children Subadults	39 40
3.5.1 3.5.2 3.5.3	Children Subadults Adults Combination of Morphological	39 40 46
3.5.1 3.5.2 3.5.3 3.5.4	Children Subadults Adults Combination of Morphological and Radiological Parameters' Methods Selection of Methods Other Skeletal Methods of Age	39 40 46 57 58
3.5.1 3.5.2 3.5.3 3.5.4 3.6 3.7	Children Subadults Adults Combination of Morphological and Radiological Parameters' Methods Selection of Methods Other Skeletal Methods of Age Estimation	39 40 46 57 58 59
3.5.1 3.5.2 3.5.3 3.5.4 3.6	Children Subadults Adults Combination of Morphological and Radiological Parameters' Methods Selection of Methods Other Skeletal Methods of Age	39 40 46 57 58
3.5.1 3.5.2 3.5.3 3.5.4 3.6 3.7 3.7.1	Children Subadults Adults Combination of Morphological and Radiological Parameters' Methods Selection of Methods Other Skeletal Methods of Age Estimation Greulich and Pyle's Method	39 40 46 57 58 59 59
3.5.1 3.5.2 3.5.3 3.5.4 3.6 3.7 3.7.1 3.7.2	Children Subadults Adults Combination of Morphological and Radiological Parameters' Methods Selection of Methods Other Skeletal Methods of Age Estimation Greulich and Pyle's Method Fishman's Method	39 40 46 57 58 59 59 59

3.1 Introduction

Identification of the living and the dead is of paramount importance in routine forensic practice. Age estimation is one of the prime factors employed to establish identity (Rai et al. 2007). In cases of unknown bodies, age estimation becomes necessary if there is no antemortem information available and a personal profile has to be reconstructed. Age estimation is of broader importance in forensic odontology, not only for identification purposes of deceased victims, but also in connection with crimes and accidents. In addition, age is used in most societies for school attendance, social benefits, employment, adoption, political asylum, and marriage (Willems 2001). Teeth are the strongest parts in the human body and are therefore very resistant to external influences, such as extreme temperatures, explosions, and other extreme conditions, which make them available for extensive postmortem periods. In addition, teeth are good indicators of people's age. These two facts allow us to use human teeth for age estimation in forensic work. Furthermore, dental age estimation in the living is mostly based upon noninvasive methods, which evaluate the timing and sequence of defined growth stages of the developing dentition and the sequence or modification of traits in the mature dentition and the surrounding tissues. Therefore, recommendations for age estimation of living persons include a dental status and a panoramic radiograph, a general physical examination, and an X-ray examination of the hand (Schmeling et al. 2004).

3.2 Dental Age Estimation Methods (Willems et al. 2002)

Various methods are utilized to determine age from dentition. These may be described in four categories, namely, clinical, radiographic, histological, and physical and chemical analyses:

- 1. *Clinical or visual method*: Visual observation of the stage of eruption of the teeth and evidence of changes due to function such as attrition can give an approximate estimate of age.
- 2. *Radiographic method*: Radiography can provide the gross stage of dental development of the dentition.
- 3. *Histological method*: Histological methods require the preparation of the tissues for detailed microscopic examination, which can determine the stage of development of the dentition more accurately. This technique is more appropriate for postmortem situations. It is also significant in estimation of age of early development of dentition.
- 4. *Physical and chemical analyses*: The physical and chemical analyses of dental hard tissues to determine alterations in ion levels with age have been proposed. While these techniques are not yet of great value to the forensic odontologist, future developments might provide an adjunctive means of collecting evidence of value in the dental context.

3.3 Factors Used for Age Determination Using Dentition (Pretty 2003)

The factors used to determine age using dentition are as follows:

- The appearance of tooth germs
- Earliest detectable trace of mineralization
- Degree of completion of the unerupted tooth
- Rate of formation of enamel and formation of the neonatal line
- · Clinical eruption
- Degree of completion of roots of erupted teeth
- · Degree of resorption of deciduous teeth
- Attrition of the crown

- · Formation of physiologic secondary dentin
- Formation of cementum
- Transparency of root dentin
- Gingival recession
- Root surface resorption
- Discoloration and staining of teeth
- Changes in the chemical composition of teeth

3.4 Basic Principles of Dental Age Estimation

Dental age can be estimated according to developmental traits such as mineralization, gingival emergence, the quantification of cementum layers or decreasing pulpal space, degenerative changes such as dental attrition or periodontal recession, the fluorescence intensity and density of dentin, the racemization of aspartic acid, or dentin sclerosis, among others.

3.4.1 Development of Teeth

Tooth formation is a composite process that begins with a gradual reorganization and a shift in the phenotype of embryonic cells. Deciduous teeth start to develop in the 6th to 8th gestational week; permanent teeth develop in about the 20th week of gestation (Ten Cate 1998). Data on the timing of human dental development are based on radiographic, histological, and morphological studies. It has been reported that histological criteria allow an individual's age to be estimated from the 7th week of gestation up to 3 years of age (Calonius et al. 1970). Hillson (1996) reported that gestational age and the height of deciduous tooth germs were also found to be correlated. The tables of Schour and Massler (1941) have become a typical example of the commonly used atlas approach for children's age estimation. Schour and Massler's data represent 22 stages of dental development, starting from 5 months in utero until 35 years of age. Gleiser and Hunt (1955) described detailed stages of the calcification of the mandibular first molar based on radiographs in the first follow-up study design in this field. Moorrees et al. (1963) derived the age of attainment for 14 developmental stages, from "initial cusp formation" to "apical closure complete," for the maxillary anterior teeth and all eight mandibular teeth. These tables were updated by Anderson for all teeth, including the wisdom teeth (Anderson et al. 1976). Another atlas approach involves the charts of Gustafson and Koch (1974), which are based on the beginning of mineralization, the completion of the crown, the eruption of the respective tooth, and the completion of the root(s). Demirjian et al. (1973; Demirjian 1976) defined four developmental crown and four developmental root stages, which were based on the appearance of the radiological visible tooth germ. In this approach, a scoring system was used for the formation of seven left mandibular teeth. Every stage was assigned a certain dental score. Adding up all scores gave the Dental Maturity Score. Third molars are the teeth with the highest variability concerning anatomy, agenesis, and age of eruption. Age estimation for medicolegal and criminal case purposes by means of third molar development is used for ages between 14 and 21 years when all other permanent teeth have finished their formation. Using Demirjian's developmental stages for third molar mineralization analysis (Schmeling et al. 2004) has been recommended, while using modified Gleiser and Hunt stages for third molar mineralization analysis (Rai et al. 2010a, b; personal experience with the Indian legal system) can also be beneficial. Other groups have suggested that modified Demirjian stages have the advantage of being clearly defined with radiographs, diagrams, and written criteria. This was also confirmed by a direct comparison of five common classification systems on orthopantomograms of females aged12–25 years (Olze et al. 2005).

3.4.2 Rate of Formation of Incremental Structures in the Tooth Crown

Boyde (1963, 1989) observed that age could be estimated from prism cross-striation counts. Prisms are bundles of hydroxyapatite crystallites that form mature enamel. An additional

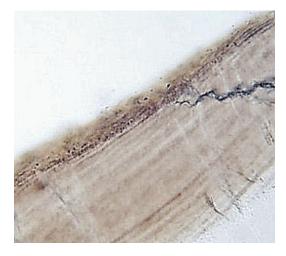


Fig. 3.1 Retzius lines

dental feature visible in crown enamel is known as Retzius lines (Fig. 3.1), which run from the enamel-dentin junction (EDJ) in the occlusal direction. Retzius lines can be found in permanent teeth as well as in the deciduous dentition, but only in postnatal enamel (Pantke 1985). Thus, the neonatal line, which constitutes a borderline between pre- and postnatal formed enamel (and dentin), is frequently used to estimate age at death. Perikymata are small, visible lines on the enamel surface. Brown striae of Retzius and perikymata groove counts have been used to estimate the age at death (Dean and Beynon 1991).

3.4.3 Changes in the Pulpodentinal Complex

The secondary dentin is built physiologically and is formed slowly by cells lining the pulp chamber. Secondary dentin formation is initiated subsequent to dentinogenesis (Costa 1986). The continuous formation of secondary dentin is due to the biological response to masticatory stress and fluctuation of temperature (Rösing and Kvaal 1998). It has been reported that the apposition of secondary dentin is correlated with chronological age (Bodecker 1925). The apposition of secondary dentin leads to a gradual reduction in the size of the pulp chamber and can affect the obliteration of the root canal (Kvaal et al. 1995; Zilberman and Smith 2001). Measurements quantify the amount of secondary dentin indirectly via the assessment of the decrease in size of the pulp cavity (Rösing and Kvaal 1998). Kvaal and Solhiem (1994) reported a method where the pulp width and length are calculated in relation to the tooth width and length. Reports with canine teeth have examined the pulp/tooth area ratio on orthopantomograms (Cameriere et al. 2004), periapical X-rays (Cameriere et al. 2007), and individual periapical X-ray (Rai et al. 2010a, b). Root dentin transparency was first introduced in 1950 for age estimation by Gustafson as one of his proposed six criteria. Bang and Ramm (1970) reported an association between age and the length of the dentin transparency and concluded that feasible results can be obtained for sectioned and unsectioned teeth specimens up to 75 years of age. Lamendin and colleagues observed an approach for age determination for single teeth that used periodontosis height and root transparency as parameters for age prediction (Lamendin et al. 1992). The quantification of age-related changes in cementum determined that the cementum's layered appearance is due to structural differences in the mineral phase, an optical phenomenon that is possibly related to altered mineral crystal orientations (Cool et al. 2002), and reflects a cyclic annual formation pattern. Gustafson first introduced cementum for human age estimation (Gustafson 1950). Stott and co-workers were the first to publish a study on the use of cementum layer counting for age estimation in humans (Stott et al. 1982). Rai and colleagues reported that cementum apposition in impacted teeth can be used for age determination (Rai et al. 2008, 2009, 2010a, b). Increasing discoloration is due to the degradation of organic components in the dental hard tissues, the deposition of external substances in enamel and dentine, mineralization, and associated alterations in the refractive index (Bang 1989). Biedow (1963) suggested that tooth color should be added to Gustafson's six-criteria system for age estimation (1950) instead of the root resorption parameter.

3.4.4 Changes in Chemical Composition

A chemical component that changes with age is the nitrogen content, which was found to increase with advancing age. It has been reported that the calcium content of teeth is higher in the coronal concentration than apical and also increases with advancing age (Bang and Monsen 1968). Another useful concept of age-related change in the chemical composition of dental tissues is the racemization of aspartic acid. In the living body, proteins are normally composed of the L-form of amino acids, turning polarized light toward the left. As age advances, L-aspartic acid will change into D-aspartic acid. An as-yet rare technique in dental age estimation is the analysis of the fluoride concentration in dentin.

3.4.5 Fluorescence of Dental Hard Tissues

The tooth roots were sectioned longitudinally and scanned in a transverse direction at the midroot area (Kvaal and Solheim 1989). Partial correlation coefficients demonstrated that an increase in fluorescence intensity is strongly associated with the deepening of dentin color even after correction for age and extraction reason. The fluorophores responsible for the respective emission peaks in the visible spectrum as well as the underlying cause for the deepening of dentified (Kvaal and Solheim 1989). The color changes in the cementum and dentin are caused by an infusion of decomposition products from erythrocytes (Fig. 3.2).

3.4.6 Epidemiological Criteria

An example of an epidemiological criterion for age estimation is the Decayed, Missing, Filled, Teeth (DMFT) index. The DMFT index can be used for an orientation but does not have strong significance in human age estimation because of high interindividual variation and the influence of dietary habits and caries prophylactic measures (Endris 1979).

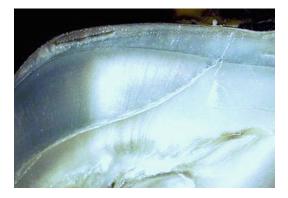


Fig. 3.2 Fluorescence (LASER) of dentin and enamel

3.4.7 Attrition

Miles presented a method that calibrated the rate and pattern of attrition in molars with eruption (Miles 1962). Extensively used is a method based on British Neolithic to medieval skeletal material, which was subdivided into four age subclasses (Brothwell 1963). Brothwell's table represents the expected dentine exposure in the permanent first three molars. Rai et al. (2010a, b) proposed a new classification of dental age estimation from first molars for the Indian population.

3.5 Methods of Dental Age Estimation (Willems 2001)

3.5.1 Children

Dental age estimation methods in children can be divided into different categories depending on the approach.

3.5.1.1 Atlas Techniques

The most commonly used dental age estimation atlas techniques are those of Schour and Massler (1941), Moorrees and colleagues (1963), Gustafson and Koch (1974), and Anderson et al. (1976). These techniques are based on morphologically different stages of mineralization of all teeth (Willems 2001). Schour and Massler (1941) proposed 20 stages of tooth development from 4 months until 21 years of age, while Moorrees et al. (1963) proposed dental

Tooli Tooli Tormation s	lages
Stage	Coded symbol
Initial cusp formation	Ci
Coalescence of cusp	Cco
Cusp outline complete	Coc
Crown 1/2 complete	Cr1/2
Crown 3/4 complete	Cr3/4
Crown complete	Cr-c
Initial root formation	Ri
Initial cleft formation	Cli
Root length 1/4	R1/4
Root length 1/2	R1/2
Root length 3/4	R3/4
Root length complete	Rc
Apex 1/2 closed	A1/2
Apical closure complete	Ac
	(10(2))

Source: From Moorrees et al. (1963)

maturation of permanent teeth (maxillary and mandibular incisors) divided into 14 stages, from initial cusp formation to apical closure for both genders (Table 3.1). Gustafson and Koch's (1974) technique proposed tooth development from mineralization, completion of crown formation, eruption, to completion of root formation.

3.5.1.2 Scoring Techniques

The most commonly used dental age estimation scoring techniques are Demirjian et al. (1973), Demirjian and Goldstein (1976), Haavikko (1970), Willems et al. (2001), and Chaillet et al. (2004). Demirjian et al.'s technique proposed eight stages (A-H) of the mandibular left teeth. Finally, outcome-based on statistical analysis, they were able to assign a maturity score for each of these seven teeth to almost each of the eight stages for both genders, as can be seen in Tables 3.2 and 3.3. Final results were calculated by adding eight scores, as shown in Tables 3.4 and 3.5. Willems et al.'s technique is based on Demirjian et al.'s staging but uses different statistical analysis method and adding scores directly gives the age, as shown in Tables 3.6 and 3.7 (Willems et al. (2001)). Another method (Nolla 1960) is based on ten stages of teeth in both genders, as can be seen in Table 3.8. As per Nolla (1960), norms by dental age were calculated for the Indian population, as shown in Tables 3.9 and 3.10 (Rai and Anand 2006; Rai 2008).

 Table 3.1
 Tooth formation stages

	А	В	С	D	Е	F	G	Н
31				0	1.9	4.1	8.2	11.8
32			0	3.2	5.2	7.8	11.7	13.7
33			0	3.5	7.9	10	11	11.9
34		0	3.4	7	11	12.3	12.7	13.5
35	1.7	3.1	5.4	9.7	12	12.8	13.2	14.4
36			0	8	9.6	12.3	17	19.3
37	2.1	3.5	5.9	10.1	12.5	13.2	13.6	15.4

Table 3.2 Individual maturity scores for boys for each of the developmental stages

Source: From Demirjian et al. (1973)

 Table 3.3
 Individual maturity scores for girls for each of the developmental stages

	А	В	С	D	E	F	G	Н
31				0	2.4	5.1	9.3	12.9
32			0	3.2	5.6	8.0	12.2	14.2
33			0	3.8	7.3	10.3	11.6	12.4
34		0	3.4	7.5	11.8	13.1	13.4	14.1
35	1.8	3.4	5.4	10.6	12.7	13.5	13.8	14.6
36			0	4.5	6.2	9.0	14.0	16.2
37	2.7	3.9	5.9	11.1	13.5	14.2	14.5	15.6

Source: From Demirjian et al. (1973)

3.5.1.3 Quantity Parameters-Based Technique

This technique is based on open and closed apices of teeth (Cameriere et al. 2006). According to this technique, the seven permanent mandibular teeth were assessed. The number of teeth with complete root development and completely closed apical ends of roots (N_0) were calculated. Teeth with incomplete root development, and therefore with open apices, were also examined. For teeth with one root, the distance A_i , i=1, ..., 5, between the inner side of the open apex was measured. For teeth with two roots, A, i=6, 7, the sum of the distances between the inner sides of the two open apices was evaluated. In order to take into account the effect of possible differences in magnification and angulation among X-rays, measurements were normalized by dividing by tooth length $(L_i, i=1, ..., 7)$. Lastly, dental maturity was evaluated using the normalized measurements of the seven permanent left mandibular teeth $(x_i=A/L_i, i=1, ..., 7)$, the sum of the normalized open apices (s), and the number (N_0) of teeth with completed root development (Fig. 3.3):

Age = 7.083 + 0.493G + 0.931 x 3-0.854S + 0.693N0 - 0.185S N0 (Cameriere et al. 2006),

where C is a dummy variable equal to 0 for the center or north of India and 1 for the south,

Age=9.402 - 0.879C + 0.663N0 - 0.711s - 0.106sN0

(Rai et al. 2010a, b)) for the Indian population, where C is a dummy variable equal to 0 for center or north of India and 1 for South India.

3.5.1.4 Eruption of Teeth Method or Visual Methods

The 20 teeth of the primary dentition erupt when the infant is between about 5 and 32 months of age, as shown in Tables 3.11 and 3.12 (ADA 2005; Rai et al. 2008). The modal eruption sequence is i1–i2–m1–c–m2 in both arches. A statistically significant male precedence compared to females has been reported (ADA 2005).

3.5.2 Subadults

Different techniques have been used for dental age estimation based on third molar staging in subadults (Demirjian et al. 1973; Harris and Nortje 1984; Köhler et al. 1994; Kullman et al. 1992;

Age	Score	Age	Score	Age	Score	Age	Score
3	12.4	6.4	39	9.7	87.7	13.0	95.6
3.1	12.9	6.5	39.2	9.8	88.2	13.1	95.7
3.2	13.5	6.6	40.6	9.9	88.6	13.2	95.8
3.3	14	6.7	42	10	89	13.3	95.9
3.4	14.5	6.8	43.6	10.1	89.3	13.4	96
3.5	15	6.9	45	10.2	89.7	13.5	96.1
3.6	15.6	7.0	46	10.3	90	13.6	96.2
3.7	16.2	7.1	48.3	10.4	90.3	13.7	96.3
3.8	17	7.2	50	10.5	90.6	13.8	96.4
3.9	17.6	7.3	52	10.6	91	13.9	96.5
4.0	18.2	7.4	54.3	10.7	91.3	14	96.6
4.1	18.9	7.5	56.8	10.8	91.6	14.1	96.7
4.2	19.7	7.6	59.6	10.9	91.8	14.2	96.8
4.3	20.4	7.7	62.5	11.0	92	14.3	96.9
4.4	21	7.8	66	11.1	92.2	14.4	97
4.5	21.7	7.9	69	11.2	92.5	14.5	97.1
4.6	22.4	8	71.6	11.3	92.7	14.6	97.2
4.7	23.1	8.1	73.5	11.4	92.9	14.7	97.3
4.8	23.8						
4.9	24.6	8.2	75.1	11.5	93.1	14.8	97.4
5.0	25.4	8.3	76.4	11.6	93.3	15.0	97.6
5.1	26.2	8.4	77.7	11.7	93.5	15.1	97.7
5.2	27	8.5	79	11.8	93.7	15.2	97.8
5.3	27.8	8.6	80.2	11.9	93.9	15.3	97.8
5.4	28.6	8.7	81.2	12.0	94	15.4	97.2
5.5	29.5	8.8	82	12.1	94.2	15.5	98
5.6	30.3	8.9	82.8	12.2	94.4	15.6	98.1
5.7	31.1	9	83.6	12.3	94.5	15.7	98.2
5.8	31.8	9.1	84.3	12.4	94.6	15.8	98.2
5.9	32.6	9.2	85	12.5	94.8	15.9	98.3
6.0	33.6	9.3	85.6	12.6	95	16	98.4
6.1	34.7	9.4	86.2	12.7	95.1	10	2011
6.2	35.8	9.5	86.7	12.8	95.2		
6.3	36.9	9.6	87.2	12.9	95.4		

 Table 3.4
 Final maturity scores for boys

Source: From Demirjian et al. (1973)

Mesotten et al. 2002; Mincer et al. 1993; Moorrees et al. 1963; Nortje 1983; Rai 2009; Solari and Abramovitch 2002).

3.5.2.1 Demirjian et al.'s Technique

This technique is based on eight stages of the third molar (Fig. 3.4), as follows (Demirjian et al. 1973):

- A. Cusp tips are mineralized but have not yet coalesced.
- B. Mineralized cusps are united so the mature coronal morphology is well defined.
- C. The crown is about half-formed; the pulp chamber is evident and dentinal deposition is occurring.

- D. Crown formation is complete to the dentinoenamel junction. The pulp chamber has a trapezoidal form.
- E. Formation of the interradicular bifurcation has begun. The root length is less than the crown length.
- F. The root length is at least as great as the crown length. Roots have funnel-shaped endings.
- G. Root walls are parallel, but apices remain open.
- H. Apical ends of the roots are completely closed, and the periodontal membrane has a uniform width around the root.

Rai (2008) has proposed the dental age estimation in North Indian subadults based on Demirjian et al. (1973), as shown in Table 3.13 and graph 3.1.

Age	Score	Age	Score	Age	Score	Age	Score
3	13.7	6.4	42.5	9.7	90.7	13.0	97.3
3.1	14.4	6.5	43.9	9.8	91.1	13.1	97.4
3.2	15.1	6.6	45.2	9.9	91.4	13.2	97.6
3.3	15.8	6.7	46.7	10	91.8	13.3	97.9
3.4	16.6	6.8	48	10.1	92.1	13.4	97.7
3.5	17.3	6.9	49.5	10.2	92.3	13.5	97.8
3.6	18	7.0	51	10.3	92.6	13.6	98
3.7	18.8	7.1	52.9	10.4	92.9	13.7	98.1
3.8	19.5	7.2	55.5	10.5	93.2	13.8	98.2
3.9	20.3	7.3	57.8	10.6	93.7	13.9	98.3
4.0	21	7.4	61	10.7	93.9	14	98.3
4.1	21.8	7.5	65	10.8	94	14.1	98.4
4.2	22.5	7.6	68	10.9	94.2	14.2	98.5
4.3	23.2	7.7	71.8	11.0	94.5	14.3	98.6
4.4	24	7.8	75	11.1	94.7	14.4	98.7
4.5	24.8	7.9	77	11.2	94.9	14.5	98.8
4.6	25.6	8	80.2	11.3	95.1	14.6	98.9
4.7	26.4	8.1	80.5	11.4	95.3	14.7	99
4.8	27.2						
4.9	28	8.2	81.2	11.5	95.4	14.8	99.1
5.0	28.9	8.3	82.2	11.6	95.6	15.0	99.1
5.1	29.7	8.4	83.1	11.7	95.8	15.1	99.2
5.2	30.5	8.5	84	11.8	96	15.2	99.3
5.3	31.3	8.6	84.8	11.9	96.2	15.3	99.4
5.4	33	8.7	85.3	12.0	96.3	15.4	99.4
5.5	33.5	8.8	86.1	12.1	96.4	15.5	99.5
5.6	34	8.9	86.7	12.2	96.5	15.6	99.6
5.7	35	9	87.2	12.3	96.6	15.7	99.7
5.8	36	9.1	87.8	12.4	96.7	15.8	99.8
5.9	37	9.2	88.3	12.5	96.8	15.9	98.9
6.0	38	9.3	88.8	12.6	96.9	16	100
6.1	39.1	9.4	89.3	12.7	97		
6.2	40.2	9.5	89.8	12.8	97.2		
6.3	41.3	9.6	90.2	12.9	97.3		

Table 3.5 Final maturity scores for girls

Source: From Demirjian et al. (1973)

 Table 3.6
 Individual maturity scores for boys for each of the developmental stages

	А	В	С	D	Е	F	G	Н
31	0.00	0.00	1.68	1.49	1.50	1.86	2.07	2.19
32	0.00	0.00	0.55	0.63	0.74	1.08	1.32	1.64
33	0.00	0.00	0.00	0.04	0.31	0.47	1.09	1.90
34	0.15	0.56	0.75	1.11	1.48	2.03	2.43	2.83
35	0.08	0.05	0.12	0.27	0.33	0.45	0.40	1.15
36	0.00	0.00	0.00	0.69	1.14	1.60	1.95	2.15
37	0.18	0.48	0.71	0.89	1.31	2.00	2.48	4.17

Source: From Willems et al. (2001)

31 0.00 0.00	1.83	2.19	0.24			
		2.1)	2.34	2.82	3.19	3.14
32 0.00 0.00	0.00	0.29	0.32	0.49	0.79	0.70
33 0.00 0.00	0.60	0.54	0.62	1.08	1.72	2.00
34 -0.95 -0.15	0.16	0.41	0.60	1.27	1.58	2.19
35 -0.19 0.01	0.27	0.17	0.35	0.35	0.55	1.51
36 0.00 0.00	0.00	0.62	0.90	1.56	1.82	2.21
37 0.14 0.11	0.21	0.32	0.66	1.28	2.09	4.04

 Table 3.7
 Individual maturity scores for girls for each of the developmental stages

Source: From Willems et al. (2001)

Table 3.8 Nolla's developmental stages

Stage 10: apical end of root completed
Stage 9: root almost complete; open apex
Stage 8: two thirds of root completed
Stage 7: one third of root completed
Stage 6: crown completed
Stage 5: crown almost completed
Stage 4: two thirds of crown completed
Stage 3: one third of crown completed
Stage 2: initial calcification
Stage 1: presence of crypt
Stage 0: absence of crypt
Source: From Nolla (1960)

 Table 3.9
 Sum of stages of 7 maxillary and mandibular

 teeth and sum of 14 teeth belonging to boys aged 3–15

Age (years)	Sum of stages seven maxillary teeth	Sum of stages seven mandibular teeth	Sum of stages of both
3	26.80	29.70	57.10
4	33.10	33.90	65.20
5	37.80	40.90	75.60
6	43.10	44.20	86.05
7	49.10	52.10	100.10
8	52.30	55.50	106.20
9	56.30	58.90	113.20
10	58.50	60.40	116.30
11	62.80	62.80	125.60
12	65.00	65.00	130.00
13	68.00	69.00	137.00
14	70.00	69.00	139.00
15	70.00	70.00	140.00

Source: From Rai (2006)

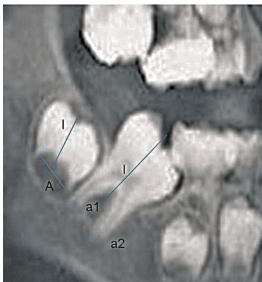
Fig. 3.3 Parameters for Cameriere et al.'s method: left mandibular teeth ($x_i = A_i L_i$, i = 1, ..., 7), the sum of the normalized open apices (*s*), and the number (N_0) of teeth with completed root development

 Table 3.10
 Sum of stages of seven maxillary and mandibular teeth and sum of 14 teeth belonging to girls aged 3–15

Age	Sum of stages seven maxillary	Sum of stages seven mandibular	Sum of stages of
(years)	teeth	teeth	both
3	29.50	28.60	57.30
4	33.30	34.96	67.90
5	37.95	38.85	75.30
6	45.20	46.90	90.05
7	50.60	54.30	102.20
8	54.08	57.50	110.40
9	56.35	62.80	116.30
10	59.50	64.35	124.60
11	62.90	65.60	129.40
12	67.60	69.10	134.20
13	69.00	69.50	137.60
14	69.60	69.90	139.10
15	70.00	70.00	140.00
а Б	D : (2004)		

Source: From Rai (2006)

Cameriere et al method



	Maxillary	Mandible
Tooth	Mean (S.D.)	Mean (S.D.)
i1	7.07 (2.45)	6.14 (1.12)
i2	9.98 (4.56)	12.14 (4.76)
С	17.09 (6.56)	17.67 (5.23)
M1	16.78 (4.67)	16.03 (4.52)
M2	28.97 (6.45)	27.89 (4.32)

 Table 3.11 Mean age (in months) for emergence of primary teeth

Source: From Rai et al. (2008)

Table 3.12 Range for emergence and shed of primary teeth

Tooth	Maxillary eruption (months)	Maxillary shed (years)	Mandible eruption (months)	Mandible shed (years)
i1	8-12	6–7	23-31	10-12
i2	9–13	7–8	14-18	9-11
С	16-22	10-12	17–23	9–12
M1	13–19	9–11	10-16	7-8
M2	25-33	10-12	6–10	6–7

Source: From American Dental Association (2005)

Moorrees et al. (1963) used data from the famous Fels Longitudinal Study in combination with their incisor radiographs. The authors derived the age of attainment for 14 developmental stages (Table 3.1), from "initial cusp formation"

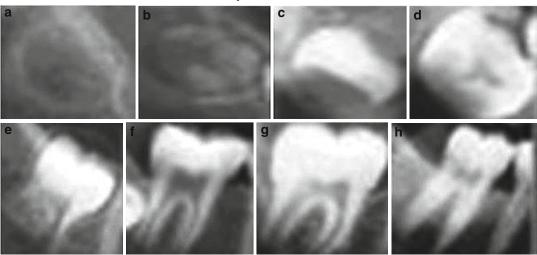
to "apical closure complete" (Moorrees et al. 1963) for the maxillary incisors and all eight mandibular teeth. A well-known study on third molar development was published by the American Board of Forensic Odontologists (Mincer et al. 1993), which used Demirjian's developmental stages to assess the mean ages of attainment. Unfortunately, the study was weakened by the diversity of its sample, which suggests that these data should not be used in practice. Gleiser and Hunt's (1955) technique based on ten stages of the third molar is shown in Table 3.14. Rai and Kaur (2011) prepared regression equations for age estimation using Gleiser and Hunt's (1955) stages for third molars (Table 3.15). The Haavikko technique is based on 12 stages of a tooth, such as Moorrees' stage Coc collapsed into Haavikko C1/2, Ri into Cc, Cli into R1/4, and A1/2 into Rc, Solari stages F1 and F combined into Demirjian stage F, and G1 and G into Demirjian G. Gustafson and Koch (1974) only provided a verbal description and defined the following four-stage classification:

Stage 1: commencement of mineralization

Stage 2: completion of crown

Stage 3: eruption when the cusp(s) penetrate the gingiva

Stage 4: completion of root(s)



Demirjian et al method

Fig. 3.4 Stages of Demirjian et al.'s method

		-		-	-
Ages	Sex	Maxillary right third molar (18)	Maxillary left third molar (28)	Right mandibular third molar (38)	Left mandibular third molar (48)
С	Male	15.2	15.0	14.6	14.7
		(14.1–16.2)	(14.1–15.9)	(14.1–15.1)	(14.0–15.3)
	Female	15.3	15.2	14.7	14.9
		(14.2–16.4)	(14.2–16.1)	(14.1–15.3)	(14.2–15.5)
D	Male	16.1	16.0	15.9	16.2
		(15.4–16.7)	(15.1–16.3)	(15.5–16.3)	(15.6–16.7)
	Female	16.2	16.2	16.3	16.4
		(15.6–16.8)	(15.7–16.7)	(15.7–16.9)	(15.8–16.9)
Ε	Male	16.7	17.5	17.4	17.2
		(16.1–17.3)	(16.8–18.1)	(16.9–17.9)	(16.8–17.6)
	Female	17.2	17.9	17.5	17.4
		(16.6–17.8)	(16.8–18.9)	(16.9–18.0)	(16.9–17.9)
F	Male	18.3	18.6	18.2	18.1
		(17.3–19.4)	(17.9–19.3)	(17.5–18.9)	(17.2–18.9)
	Female	18.6	19.0	18.5	18.3
		(17.5–19.6)	(18.1–19.9)	(17.7–19.2)	(17.4–19.2)
G	Male	19.9	19.8	19.3	19.4
		(19.8–20.0)	(19.0-20.2)	(18.7–19.9)	(19.0–19.8)
	Female	20.4	19.3	19.3	20.1
		(20.1-20.9)	(19.2–20.9)	(19.0–21.2)	(19.1–20.9)
Н	Male	21.4	21.4	20.6	20.8
		(21.0-21.7)	(20.9–21.9)	(20.1–21.1)	(19.6–20.6)
	Female	21.5	22.4	21.2	20.9
		(21.0-21.9)	(20.8–22.0)	(20.3-22.0)	(19.8–22.0)

Table 3.13 Mean ages (in years) (95 % confidence intervals) of Demirjian's stages assumed from North Indian juveniles

p<0.001

Table 3.14	Gleiser and	l Hunt's	staging
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Staging	Description
1	1/2 crown
2	3/4 crown
3	Crown complete
4	Just root formation
5	1/4 root formation
6	1/2 root formation
7	3/4 root formation
8	Root formation complete
9	Root apex half-closed
10	Apex closed

Soure: From Gleiser and Hunt (1955)

A detailed study on ethnical differences in third molar mineralization was published by Olze et al. (2004), who evaluated more than 3,000 conventional orthopantomograms. It was shown that Japanese, German, and South African individuals differ up to several years in reaching the respective developmental stage according to Demirjian. Therefore, using population-specific data in age estimation is recommended to guarantee for optimal reproducibility and feasibility of the evaluation.

3.5.2.2 Eruption of Permanent Teeth

There are mainly two methods of dental age assessment: radiographically and by clinical visualization of eruption of teeth. By radiographic methods it is possible to follow the formation of crowns and roots of teeth and their calcification. The clinical method to assess dental age is based on emergence of teeth in the mouth. This method is more suitable since it does not require any special equipment or expertise and is more economical. Permanent teeth eruption in North Indians is shown in Tables 3.16 and 3.17.

Sex	Third molar	Regression formula	S.D.
Males	UR (upper right)	12.456+0.976UR	1.89
	UL (upper left)	12.422+0.875UR	1.78
	LL (lower left)	13.435+0.745LL	1.85
	LR (lower right)	13.456+0.567LR	1.84
Females	UR (upper right)	13.589+0.823UR	1.74
	UL (upper left)	13.617+0.821UR	1.72
	LL (lower left)	14.342+0.645LL	1.56
	LR(lower right)	14.682+0.642LR	1.47
Unknown	UR (upper right)	12.995+0.768UR	1.67
	UL (upper left)	12.231+0.934UR	1.92
	LL (lower left)	13.764+0.9835LL	1.78
	LR (lower right)	13.785+0.745LR	1.65

Table 3.15 Regression formulas for dental age estimation

 Table 3.16
 Mean ages (in years)±standard deviation

 (S.D.) of maxillary permanent teeth in North Indians

Boys	Girls	Both genders
7.73 ± 0.23	7.61 ± 0.21	7.64 ± 0.22
8.53 ± 0.27	8.42 ± 0.26	8.33 ± 0.25
11.94 ± 0.27	11.12 ± 0.21	11.53 ± 0.24
12.12 ± 0.26	12.10 ± 0.22	12.06 ± 0.24
12.68 ± 0.24	12.14 ± 0.23	12.37 ± 0.22
6.93 ± 0.24	6.82 ± 0.21	6.91 ± 0.21
13.04 ± 0.27	12.91 ± 0.23	12.97 ± 0.25
	8.53 ± 0.27 11.94 ± 0.27 12.12 ± 0.26 12.68 ± 0.24 6.93 ± 0.24	7.73 ± 0.23 7.61 ± 0.21 8.53 ± 0.27 8.42 ± 0.26 11.94 ± 0.27 11.12 ± 0.21 12.12 ± 0.26 12.10 ± 0.22 12.68 ± 0.24 12.14 ± 0.23 6.93 ± 0.24 6.82 ± 0.21

p < 0.001

 Table 3.17
 Mean ages (in years)+standard deviation

 (S.D.) of mandibular permanent teeth in North Indians

Tooth (FDI)	Boys	Girls	Both genders
31, 41	6.94 ± 0.23	6.39 ± 0.21	6.72 ± 0.22
32, 42	7.94 ± 0.24	7.62 ± 0.27	7.78 ± 0.25
33, 43	11.34 ± 0.21	10.84 ± 0.25	11.01 ± 0.23
34, 44	11.83 ± 0.26	10.92 ± 0.26	11.47 ± 0.26
35, 45	12.34 ± 0.23	12.69 ± 0.27	12.43 ± 0.26
36, 46	6.94 ± 0.27	6.43 ± 0.25	6.62 ± 0.26
37, 47	12.69 ± 0.24	11.94 ± 0.26	12.23 ± 0.25
- <0.001			

p < 0.001

3.5.3 Adults

Methods for the dental age estimation of adults can be divided into different groups depending on approaches. Various morphologically based methods have been proposed for dental age estimation in adults.

3.5.3.1 Gustafson's Method

Gustafson's (1950) method is based on measuring physiological changes in teeth, such as occlusal attrition, secondary dentin formation, loss of periodontal attachment, apposition of cementum, apical resorption, and transparency of root. It uses six parameters that are assigned different scores on a scale from 0 to 3. After collecting the data and calculating the total score, it calculates the estimated age using the Gustafson formula (Gustafson 1950) (Figs. 3.5 and 3.6). Attrition (A):

A0: No attrition

A1: Attrition limited to enamel level

A2: Attrition limited to dentine level

A3: Attrition up to pulp cavity

Periodontal disease (P):

P0: No obvious periodontal disease

P1: Beginning of periodontal disease but no bone loss

P2: Peridontal disease more than 1/3 of the root

P3: Peridontal disease more than 2/3 of the root Secondary dentine (S):

S0: No secondary dentine formation

S1: Secondary dentine up to upper part of pulp cavity S2: Secondary dentine up to 2/3 of the pulp

cavity

S3: Diffuse calcification of entire pulp cavity Root translucency (T):

T0: No translucency

T1: Beginning of translucency

T2: Translucency more than 1/3 of the apical root

T3: Translucency more than 2/3 of the apical root

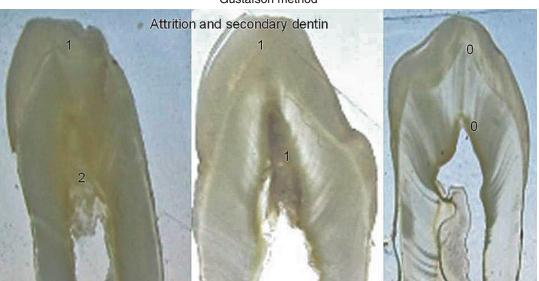
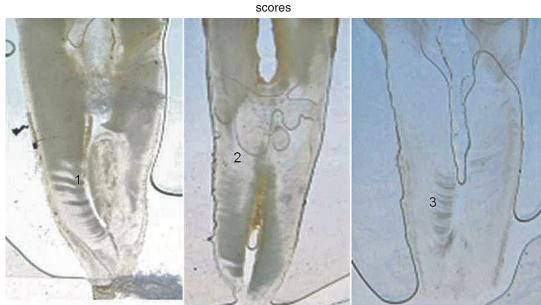


Fig. 3.5 Attrition and secondary dentine scores



Root translucency scores

Fig. 3.6 Root translucency

Cementum apposition (C):

C0: Normal cementum

C1: Thickness of cementum more normal

C2: Abnormal thickness of cementum near the apex of the root

C3: Generalized abnormal thickness of cementum throughout the apex of the root Root resorption (R):

R0: No resorption

R1: Spotted resorption

R2: Resorption limited to cementum

R3: Extensive resorption of the cementum and dentin both

Gustafson's linear regression formula for dental age estimation was

Age = 11.43 + 4.56X and S.d. = 3.63 years (Gustafson 1950)

$$Age = 4.6696x + 10.381$$
 (Rai 2008).

Measurement of parameters: The tooth was extracted by extraction forceps and preserved in formalin until the ground section was prepared. The ground section was prepared by hand grinding, which was done first with a lathe and then with rough carborundum stone until a 1-mm section was obtained; at this thickness, the root translucency was noted. Grinding was further done using a fine stone until a 25-mm-thick section was left. Finally, the cleaned and dried section was mounted on a slide and viewed under a microscope for secondary dentine, cementum apposition, and root resorption.

3.5.3.2 Dalitz's Method (1962)

In investigating the Gustafson method, Dalitz found that cementum apposition and root resorption were poorly related to age. As a consequence of these findings, he omitted them from further work and went on to develop a weighting of the remaining criteria based on their relative correlation to age, using regression of his results to derive a predictive formula for the estimation of age. It is also known as a 5-point system:

3.5.3.3 Johanson's Method

Johanson (1971) used basically the same age indicators as Gustafson but decided that intermediate stages of severity could be detected reliably, resulting in a system of seven ordinal stages for each of the six variables, as opposed to Gustafson's original four. Instead of Gustafson's crude summation of total points for a given individual followed by linear regression of total point score against age, Johanson used a multivariate regression against age, which is stated to have yielded a standard error of 5.16 years for any given age estimation. It is worth noting at this stage that neither Johanson nor Gustafson differentiated between tooth locus in their regression analysis. Using this method, Johanson reported a mean error of ± 5.16 years for the 65 % confidence interval:

Age =11.02 + 5.14A + 2.30S + 4.14P + 3.71C + 5.57R + 8.98T

3.5.3.4 Rai et al.'s Method (2006)

Rai et al. used basically the same age indicators as Gustafson except for epithelial attachment, but the individual changes were classified using a 13-point scale (0, 0.25, 0.5, 0.75, 1, ..., 3) in their method. The ground sections were examined four times at low magnification. The formulas for age calculation as a relationship between the sum of point values (SPV) and without transparency regarded (SPV-T) and age using regression analysis were estimated as follows:

Age =
$$(SPV\% + 2.27) / 1.12$$

Age =
$$((SPV-T\%)+3.42) / 1.16$$

3.5.3.5 Maples' Method (1978)

A study by Maples in 1978 attempted to assess which criteria worked best in conjunction with each other using multiple regression analysis (Maples 1978). The most effective combination involved all criteria except root resorption when specifically used on the second permanent molar, and gave an error of ± 5 years (Maples and Rice (1979)). However, the combination of just secondary dentine apposition and transparency gave the best overall result when applied to all teeth. He proposed the different regressions based on position of the tooth (Table 3.18).
 Table 3.18
 Formulas for Maples' method based on position of tooth

Position formulas for ST
Age=3.89S+14.23T+15.28 (9.1SE) Position 1
6.51S+12.55T+25.16 (9.6SE) Position 2
18.67S + 11.72T + 21.94 (11.0SE) Position 3
2.82S+15.25T+19.65 (12.2 SE) Position 4
4.79S+15.53T+17.99 (7.6) Position 5
11.28S+5.32T+10.86 (11.1SE) Position 6
6.99S + 10.86T + 19.31 (6.8SE) Position 7
4.71S+12.30T+24.57 (12.0SE) Position 8
Position formulas for APST
Age=4.23A-4.18P+2.98S+8.63C+18.15T-3.35 (10.1 SE) Position 1
3.47A+6.78P+13.67S+4.21C-0.99T+12.73 (9.6SE) Position 2
11.02A+7.35P+18.52S+0.35C+7.54T-1.14 (10.1SE) Position 3
20.16A-0.85P-3.01S+2.65C+13.23T+5.96 (7.1SE) Position 4
15.88A+3.23P+2.46S-0.53C+14.73T+1.79 (10.1SE) Position 5
2.09A+3.33P+9.08S+1.47C+4.38T+6.32 (11.0SE) Position 6
8.91A+2.23P+7.64S+0.45C+6.99T+12.10 (5.0SE) Position 7
5.55A-0.92P+4.30S-0.80C+13.77T+24.76 (13.7SE) Position 8
Formulas weighted for all positions
Age=6.54S+10.88T+16.08+position value (9.1SE)
Position values
Position 1=0.00
Position 2=11.24
Position 3=13.18
Position 4=4.39
Position 5=5.21
Position $6 = -5.37$
Position 7=3.73
Position 8=8.04
Age=7.09A+2.56P+5.20S+0.26C+9.14T+2.32+position value (9.3SE)
Position values
Position 1=0.00
Position 2=15.59
Position 3 = 16.07
Position 4=10.4
Position 5=11.69
Position 6=1.98
Position 7=11.65
Position 8 = 18.44

3.5.3.6 Bang and Ramm's Method (1970)

Bang and Ramm devised a method based on measuring one parameter such as the length of the apical translucent zone in mm of the given tooth for different positions of a tooth, side, intact, and tooth section: If transluscency zone $< 9 \text{ mm} (\text{Age} = \text{B0} + \text{B1x} + \text{B2x}^2)$

If transluscency zone > 9 mm (Age = B0 + B1x)

Regression constants are taken from the reported tables (Table 3.19).

gression constant and the regression coefficient as reported by Bang and Ramm (1970). Differentiation was made on the level of substrate (intact or section	ngth of the translucent zone (<9 mm and >9 mm) (<i>m</i> mesial, <i>d</i> distal, <i>p</i> palatal, <i>r</i> root)
Table 3.19 Regression constant	th of the tran

					mmd Jimm	60001 1 6				
Tooth	<9 mm						>9 mm			
	Intact roots			Tooth sections			Intact roots		Tooth sections	
	B 0	B1	B 2	B0	B1	B2	B0	B1	B0	B1
11	20.30	5.74	0.000	21.02	6.03	-0.060	20.34	5.74	22.36	5.39
21	24.30	6.22	-0.119	26.84	6.00	-0.155	26.78	4.96	30.18	4.30
12	18.80	7.10	-0.164	23.09	7.04	-0.197	22.06	5.36	25.55	5.23
22	20.90	6.85	-0.223	24.62	5.18	-0.077	25.57	4.38	25.90	4.39
13	26.20	4.64	-0.044	21.52	649	-0.171	28.13	4.01	28.01	4.23
23	25.27	4.58	-0.073	24.64	5.22	-0.143	27.59	3.65	29.41	3.32
14/24	23.91	3.02	0.203	29.98	2.73	0.107	18.42	5.40	28.44	3.81
15	23.78	5.06	-0.064	24.76	4.81	0.000	25.33	4.28	24.75	4.01
25	25.95	4.07	-0.067	22.34	7.59	-0.393	26.92	3.37	26.21	4.03
41	9.80	12.61	-0.711	13.63	12.11	-0.683	29.00	4.23	31.78	4.19
31	23.16	9.32	-0.539	26.46	8.79	-0.511	37.56	2.94	37.89	3.08
42	26.57	7.81	-0.383	21.77	10.19	-0.581	38.81	2.81	38.49	3.03
32	18.58	10.25	-0.538	22.22	9.07	-0.444	33.65	3.53	35.19	3.49
43	23.30	8.45	-0.348	24.34	8.38	-0.358	37.56	3.50	40.32	3.05
33	27.45	7.38	-0.289	23.88	8.76	-0.388	41.50	2.84	42.07	2.73
44	24.83	6.85	-0.237	21.54	8.63	-0.395	30.83	4.05	33.10	3.66
34	29.17	5.96	-0.173	26.02	7.00	-0.234	34.97	3.74	32.79	4.11
45	29.42	4.49	-0.065	14.90	9.93	-0.451	30.68	3.76	27.46	4.17
35	18.72	5.79	-0.082	23.87	5.50	-0.098	20.87	4.79	25.60	4.41
16/26mr	30.25	3.23	-0.018	28.22	4.82	-0.101	30.56	3.00	30.03	3.48
36/46mr	27.39	6.25	-0.239	33.42	5.18	-0.302	30.32	3.66	35.27	2.78
16/26dr	34.73	0.67	0.211	20.43	6.09	-0.182	29.49	3.32	26.89	3.55
36/46pr	30.21	3.32	-0.181	29.91	4.97	-0.102	31.46	3.77	30.31	4.22
16/26pr	27.43	3.64	0.039	25.15	4.34	-0.032	26.81	4.07	25.83	3.95

Table 3.20 Multiple regression formulas with age as the dependent variable for each tooth type (parameters that were strongly correlated with age were used in the regression formulas)

Color and gender include
Maxillary
1. Age=24.3+8.7CEST+5.2TD-2.3CAP-4.3SEX
2. Age=38.7-126ST+4.7CEST+4.2TD+0.05C1
3. Age=10.1+2.3TID+4.4SJ+6.1CEST
4. Age=8.0+7.3CEST+4.1SJ+1.4TID
5. Age=6.1+9.1CEST+3.3AJ+7.3LPMEAN+1.4TID
Mandibular
1. Age=-21.8-55.3SC+32.8LC1-10.3SEX+2.6TID
2. Age=-24.5+4.9CEST+2.1TID-7.0SEX+20.1LC 1+2.4AJ
3. Age=19.2+1.7TID+5.1CEST+3.5SJ
4. Age=-28.1+3.0TID+0.6ARA+24.1LC1-5.6SEX +7.3LPMPEAN
5. Age=7.5+2.7TID+4.9SJ+4.9ARS
Color and gender excluded
Maxillary
1. Age=25.3+7.1TID-3.1CAP+5.3SRS-7.5EX3+ 0.02CI
2. Age=46.7-142ST=6.5TD+0.05C1
3. Age = 12.1 + 2.9TID + 4.9SJ + 3.9SRS
4. Age=14.6+6.3SJ+2.5TID
5. Age = 14.2 + 2.5TID + 4.1AJ + 8.9LPMEAN + 3.0SJ
Mandibular
1. Age=-32.1-52.5SC+31.1LC1+1.9TID+4.6SRS
2. Age=37.1+2.7TID=5.9SRS-46.3SC
3. $Age = 27.5 + 2.6TID + 4.4SJ$
4. Age=-26.9+3.2TID+0.5ARA+22.3LC1+7.1LPM EAN
5. Age=7.5+2.7TID+4.9SJ+4.9SRS
(SE=8.7–10.8 years)

3.5.3.7 Solheim's Method (1993)

Solheim's technique includes those parameters showing a strong correlation with age. Table 3.20 shows the multiple regression equations with age as the dependent variable and the age changes, including color and sex versus exclusion of color and sex, as independent variables to be measured.

The following parameters are taken into account:

CEST: color estimation (Fig. 3.7)

SEX: male (1), female (0)

EX3: extraction for caries (1) or other (0)

SRS: surface roughness score (Fig. 3.8)

TD: translucency according to Dalitz (1962) (Fig. 3.9)

TID: length of translucent zone measured in dry intact tooth

CAP: crown area of the pulp (Fig. 3.10)

LPMEAN: Log10(PMEAN)

PMEAN: mean loss in mm of periodontal attachment (V, M, P, D)

ARA: area of attrition in mm²

AJ/SJ: attrition/secondary dentine according to Johanson (1971)

SC: pulp diameter divided by root diameter measured at cementoenamel junction (Fig. 3.11)

ST: sum of pulp diameters divided by sum of root diameters (measured at cementoenamel junction, coronal 1/4, midroot, and apical 1/4) (Fig. 3.12)

C1: Sum of cementum thickness on vestibular and lingual surface 1/3 root length from apex (Fig. 3.13)

SC: pulp diameter divided by root diameter measured at cemento-enamel junction

ST: sum of pulp diameters divided by sum of root diameters (measured at cementoenamel junction, coronal 1/4, midroot, and apical 1/4)

CEST: Color estimation

Scores	Trubyte
1	59
2	56
3	69
4	81
5	82

SRS: Surface roughness score

0: almost completely smooth surface

1: almost even surface with microscopic roughnesses $(10 \ \mu m)$ and only a few larger roughnesses and no external resorption defects

2: less even surface with a number of larger roughnesses but with no or only a few resorption defects

3: uneven surface with both smaller and larger roughnesses and some resorption defects

4: a very uneven surface with large roughnesses and resorption defects

3.5.3.8 Rai's Method (2009)

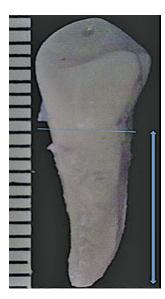
Rai's method is based on the coronal displacement of the cementum that occurs with aging in impacted teeth. The teeth were rinsed in running water and were placed in formalin solution for 17 days. The buccolingual ground sections were prepared from each specimen. The distance

Fig. 3.7 Color estimation

COLOR ESTIMATION	Co	lor	estimation
------------------	----	-----	------------

Scores	Biodent
1	10
2	21
3	27
4	31
5	32





SRS score

2



Translucency

According to Dalitz method score = 2

Fig. 3.9 Translucency according to Dalitz's method (score: 2)

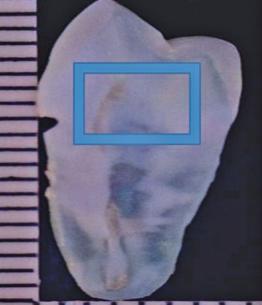
$$(x+439)/22.4.$$

Fig. 3.8 Surface roughness score

between the enamel and cementum, or the amount of cementum overlapping the cervical region of the ground sections of teeth, was measured by means of a micrometer attached to a light microscope. The measurement distance (μ m) between the edges of enamel and cementum in the impacted teeth is *x* (Fig. 3.14), and we apply the BR regression equation as follows:

3.5.3.9 Lamendin et al.'s Method (1992)

Lamendin et al.'s method is based on translucency of the tooth root and periodontosis. In a French sample, Lamendin et al. (1992) related these parameters to the overall height at the root and applied multiple regression analyses to generate the following equation for determining age at death independently of ancestry or sex: **Fig. 3.10** Crown area of the pulp



Crown area of the pulp (mm2)

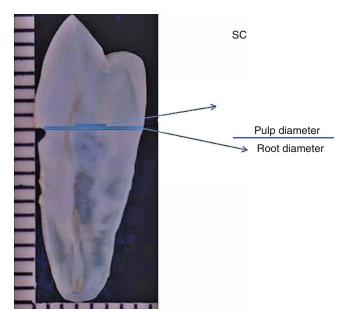
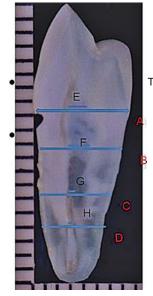


Fig. 3.11 SC (pulp diameter divided by root diameter measured at cementoenamel junction)

Fig. 3.12 ST (sum of pulp diameters divided by sum of root diameters) [measured at cementoenamel junction, coronal 1/4, midroot, and apical 1/4]



ST

Total pulp diameters / root diameters

C1

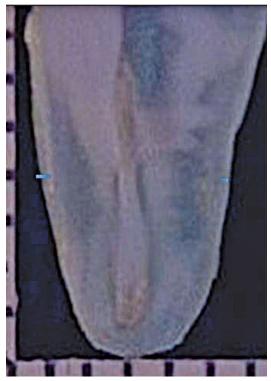


Fig. 3.13 C1 (sum of cementum thickness on vestibular and lingual surface 1/3 root length from apex)

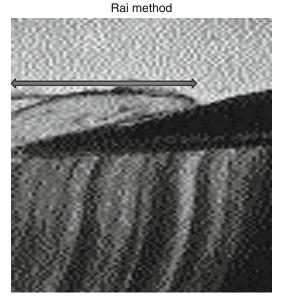


Fig. 3.14 The measurement distance (μm) between the edges of enamel and cementum in impacted teeth

 $A = 0.18 \cdot P + 0.42 \cdot T + 25.53,$

where A = age in years, P = periodontosisheight \cdot 100/root height, and T = translucencyheight \cdot 100/root height.

3.5.3.10 Prince and Ubelaker's Method (2002)

Prince and Ubelaker's method has created specific formulas for different subpopulations, classified as black females, black males, white females, and white males. The authors also included root height in the multiple regression analysis for the equations they developed:

White males (A = $[0.16 \cdot RH] + [0.29 \cdot P] + [0.39 \cdot T] + 23.17$) and white women (A = $[1.10 \cdot RH] + [0.31 \cdot P] + [0.39 \cdot T] + 11.82$),

where RH=root height.

3.5.3.11 Cementum Annulations Method

It has been reported that cementum is potentially a better age-estimating tissue due to its unique location in the alveolar process. It has been hypothesized that these incremental lines in the tooth cementum can be used as a more reliable age marker than any other morphological or histological traits in the human skeleton (Zander and Hürzeler 1958). These authors derived a regression equation for age estimation based on cementum annulations. A longitudinal ground section of tooth was prepared and examined under light microscope and polarized microscope. In each section, the area at the junction of the apical and middle third of the root and the area where the lines were easiest to count, disregarding whether the cementum was cellular or acellular, were selected for counting (Fig. 3.15). These areas were photographed and images were transmitted from the microscope to a computer (Adobe Photoshop), and counting was done with the help of image analysis software. First, the width of the cementum from the dentinocemental junction to the surface of the cementum was measured in an area where the lines seemed to run approximately parallel. Then the measurement was made of the width occupied by the two adjacent incremental lines, which were most easily recognizable, and the number of incremental lines in the total cementum width was calculated:

Number of incremental lines (n) = X/Y

where X is the total width of cementum (from DEJ to cementum surface) and Y is the width of cementum between the two incremental lines;

Age = number of incremental lines (0.987) + eruption age of tooth

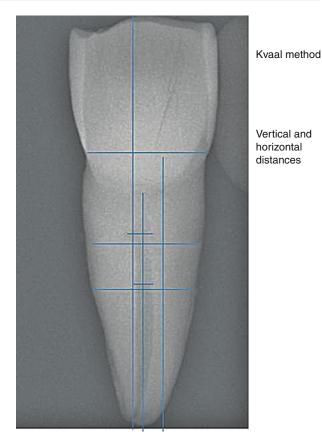
This equation is known as the JBR equation, following the authors' names.



Cemental annuations

Fig. 3.15 Cementum annulations method

Fig. 3.16 Parameters for Kvaal's method



These techniques are suitable for dental age estimations in living persons because teeth do not need to be extracted.

3.5.3.12 Kvaal et al.'s Method (1995)

Kvaal et al.'s method is based on measurements of the size of pulp observed on periapical radiographs of six types of teeth, including maxillary central and lateral incisor and second bicuspid. The age estimation depends on gender and measurements of different length and width ratios (Fig. 3.16) in order for magnification and angulation of original tooth image on the radiograph such as pulp/root length (P), pulp/tooth length (R), tooth/root length (T), pulp /root width at cement-enamel junction (A), pulp/root width at midpoint between levels C and A, pulp/root width at midroot length (C), mean value of all ratios expect T(M), mean value of width ratios B and C (W), and mean value of length ratios P and R (L)(Willems 2001). The results of equation analyses with age as dependent variable and two predictors and gender as independent variables in the formula for age estimation of the mandibular lateral incisors having values sex (male=1 and female=0) are shown in Table 3.21.

3.5.3.13 Cameriere et al.'s Method (2007)

The method Cameriere et al. used was similar, containing three formulas: for the lower canine, upper canine, and both canines. Dental maturity was evaluated by measuring the pulp/tooth area ratio on upper (x1) and lower (x2) canines:

When only the lower canine is considered, the regression equation may be written as follows:

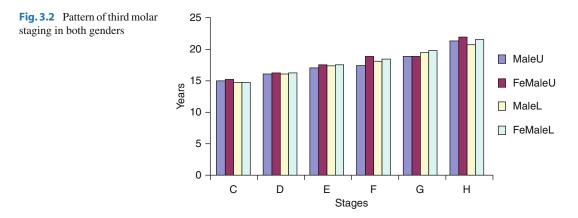


Table 3.21 Multiple regression equations for dental age estimation based on radiological parameters

Teeth	Equation	R2
11/21, 12/22, 15/25, 32/42, 33/43, 34/44	Age = 129.8 - 316.4(M) - 66.8(W - L)	0.76
11/21, 12/22, 15/25	Age = 120.0 - 256.6(M) - 45.3(W - L)	0.74
32/42, 33/43, 34/44	Age = 135.5 - 356.8(M) - 82.5(W - L)	0.71
11/21	Age = 110.2 - 201.4(M) - 31.3(W - L)	0.70
12/22	Age = 103.5 - 216.6(M) - 46.6(W - L)	0.67
15/25	Age = 125.3 - 288.5(M) - 46.3(W - L)	0.60
32/42	Age = 106.6 - 251.7(M) - 61.2(W - L) - 6.0(G)	0.57
33/43	Age = 158.8 - 255.7(M)	0.56
33/44	Age = 133.0 - 318.3(M) - 65.0(W - L)	0.64

Source: From Kvaal et al. (1995)

When only the upper canine is included, the equation becomes

Age = 99,937 - 532,775x2

3.5.4 Combination of Morphological and Radiological Parameters' Methods

3.5.4.1 Kvaal and Solheim's Method (1994)

Kvaal and Solheim's method includes both morphological and radiological parameters, such as

- 1. Apical translucency in mm (T)
- 2. Periodontal ligament retraction in mm (P)
- 3. Pulp length (PL) (radiograph)
- 4. Radiographic root length on mesial surface (RL)
- 5. Pulp width at cementoenamel junction on radiographs (PWC)

- 6. Radiographic root width at cementoenamel junction (RWC)
- 7. Radiographic pulp width at midroot (PWM)
- 8. Radiograhic width at midroot on radiographs (RWM)
- 9. FL (PL/RL)
- 10. FWC (PWC/RWC)

Put values in regressions shown in Table 3.22.

3.5.4.2 Biochemical Method of Age Estimation (2003)

Ohtani et al. proposed a biochemical method of age estimation based on L-enantiomer being transformed into D-enantiomer in enamel, dentin, and cementum:

Central incisor: 756.63x - 22.20Lateral incisor: 780.70x - 19.70

Tooth	Equation
11/21	Age=71.2-133.3FWM-56.0FWC
12/22	Age=69.3-14.5FWM-63.0FWC
13/23	Age=120.2-62.5FL
14/24	Age=82.0-95.9FWC+2.0T+1.7P-50.6FL
	Age = 112.6 - 85.0FWC + 2.4P - 116.3FWM - 64.8FL (without translucency)
15/25	Age = 30.8 + 2.5P - 96.0FWC + 3.7T
	Age = 36.9 + 2.9P – 102.9FWC (without translucency)
31/41	Age = 40.3 - 122.4FWC + 4.4T
	Age=68.5-124.4FWC (without translucency)
32/42	Age=72.1-173.6FWC
33/43	Age=43.8-139.6FWC+3.8T
	Age = 75.9 – 174.7FWC (without translucency)
34/44	Age=75.5-185.9FWC-105.4FWM+1.4P
35/45	Age=54.0-107FWM-97FWC+2.4T
	Age=80-192.7FWM-96.6FWC (without translucency)

Source: From Kvaal and Solhiem (1994)

Canine: 806.23x - 25.49First premolar: 782.58x - 23.79Second premolar: 677.96x - 11.75First molar: 686.57x - 16.37Second molar: 640.76x - 9.01Total teeth: 696.20x - 13.84

3.6 Selection of Methods

In the child and juvenile stages, the use of morphological methods based on radiological examination of a combination of dental and skeletal development is recommended (Ritz-Timme et al. 2000). Programs developed by official institutions and NGOs have established protocols of good practice, which include the elements corresponding to age assessment (UNHCR 1997). In accordance with the guidelines proposed by Ritz-Timme et al. (2000) for an age assessment method to be considered acceptable, it must fulfill the following requirements:

- The method must be transparent and provable, presented to the scientific community, as a rule by publication in peer-reviewed journals.
- 2. Clear information concerning the accuracy of age assessment by the method should be available.

- 3. The method needs to be sufficiently accurate to fulfill the specific demands of the single case to solve the underlying questions.
- 4. In cases of age assessment in living individuals, principles of medical ethics and legal regulations have to be considered, especially if medical intervention is involved.

Reference material used must fulfill certain requirements (Solari and Abramovitch 2002):

- 1. It should have an adequate simple size. The number of subjects of each sex and age group should be ten times the number of examined features.
- 2. The age indicated by the subjects should be verified.
- 3. It should have an even distribution of subjects across all age groups.
- 4. All data have to be collected separately for both sexes.
- 5. The time of the examination should be recorded.
- 6. The examined features should be defined unmistakably.
- 7. The technique used in the examination should be described specifically.
- Information on genetic-geographic origin, socioeconomic status, and health of the reference population is essential.
- The simple size, mean value, and statistical parameters of deviation should be provided for every feature examined.
- Information on inter- and intra-observer error is less in acceptable limit.

It has been recommended (Ritz-Timme et al. 2000) that the published data that fulfill the requirements listed above in childhood and adolescence are those based on the examination of dental and/or skeletal development by skilled personnel. In childhood (0–14 years), the radiological examination of dental development includes all types of teeth. In adolescence (14–21 years), third molars are the only teeth undergoing maturation, resulting in a lesser degree of accuracy. In both cases, sex and race influence tooth development, so those factors have to be taken into account.

The German age study group considers that age diagnosis examination should include (AGFAD 2001) a physical examination that also records anthropometric data, signs of sexual maturation, and any age-relevant developmental disorder, an X-ray examination of the left hand, and a dental examination that records dentition status and evaluates an orthopantomogram. It also recommends that these methods are used in combination to increase accuracy in age assessment and to facilitate the identification of age-relevant developmental disorders (Schmeling et al. 2004).

3.7 Other Skeletal Methods of Age Estimation

3.7.1 Greulich and Pyle's Method

Greulich and Pyle's studies were the base for a long-term investigation of human growth and development. A large number of children of different ages were enrolled in the study. In total the study ran from 1931 until 1942. These children had radiographs taken of their left shoulder, elbow, hand, hip, and knee. In the first postnatal year, an examination was conducted every 3 months. From 12 months to 5 years old, they were examined each 6 months, and they were examined annually thereafter. Todd's Atlas was used as a standard reference until it was revised by W. W. Greulich and S. I. Pyle in 1950, who used it to compile their well-known Radiographic Atlas of Skeletal Development of the Hand and Wrist, published in two editions in 1951 and 1959. It is still extensively used today. The Greulich and Pyle series was based on large samples of healthy middle-upper class North American children. Fundamentally, the method assesses a "mean" bone age by matching an X-ray against the bones of a standard atlas, and normality estimates based on a range of results are made using standard deviation values.

3.7.2 Fishman's Method (1982)

Fishman's method evaluates the stage of skeletal maturation of each hand/wrist radiograph, with the following parameters:

- MP₃: The middle phalanx of the third finger; the epiphysis equals its diaphysis.
- S stage: The first mineralization of the ulner sesamoid bone.
- $MP_{_{3Cap}}$: The middle phalanx of the third finger; the epiphysis caps its diaphysis.
- MP_{5Cap}: Capping of epiphysis seen in middle phalanx of fifth finger.
- DP_{3u}: The distal phalanx of third finger, complete epiphyseal union.
- MP_{3u} : The middle phalanx of third finger, complete epiphyseal union.

3.7.3 Age Estimation in the Clavicle

To solve the medical-legal case of estimation of whether a person has reached the age of 18, it is predominantly useful to evaluate the ossification status of the medial epiphysis of the clavicle, because all other examined developmental systems may already have completed their growth by that age.

Universal classification systems differentiate among four stages of clavicle development:

- Stage 1: ossification center not ossified
- Stage 2: ossification centre ossified, epiphyseal plate not ossified
- Stage 3: epiphyseal plate partially ossified Stage 4: epiphyseal plate fully ossified

Also, Schmeling et al. (2004) divided the stage of total epiphyseal fusion into two extra stages as follows:

- Stage 5: epiphyseal plate fully ossified, epiphyseal scar visible
- Stage 6: epiphyseal plate fully ossified, epiphyseal scar no longer visible

A further improvement of age diagnostics based on clavicular ossification was the subdivision of stages 2 and 3 by Kellinghaus et al. (2010a, b). The subclassification stages were distinct, as follows:

- Stage 2a: The lengthwise epiphyseal measurement is one third or less compared to the widthwise measurement of the metaphyseal ending.
- Stage 2b: The lengthwise epiphyseal measurement is greater than one third but less than two thirds the widthwise measurement of the metaphyseal ending.

- Stage 2c: The lengthwise epiphyseal measurement is over two thirds compared to the widthwise measurement of the metaphyseal ending.
- Stage 3a: The epiphyseal–metaphyseal fusion completes one third or less of the former gap between the epiphysis and metaphysis.
- Stage 3b: The epiphyseal-metaphyseal fusion completes more than one third up to two thirds of the former gap between the epiphysis and metaphysis.
- Stage 3c: The epiphyseal–metaphyseal fusion completes over two thirds of the former gap between the epiphysis and metaphysis.

Stage 3c first appeared at age 19 in both sexes. If stage 3c is found, it is therefore possible to confirm that an individual has already reached the legally important age threshold of 18 years.

Another radiation-free method to evaluate the ossification stage of the medial clavicular epiphyses is ultrasonography (Schulz et al. 2008). Classification of the medical end of the clavicle is as follows:

- Stage 1: The medial end of the clavicle is configured acute-angled. A bony center of ossification is not representable.
- Stage 2: The medial end of the clavicle is separated from the bony center of ossification by a sound gap.
- Stage 3: Both an ultrasound gap with a bony center of ossification and a fully ossified epiphyseal plate with a convex-curved end of the clavicle are representable.
- Stage 4: The medial end of the clavicle is convexcurved. A bony center of ossification is not representable.

The earliest ages at which the respective ossification stages were observed were 17 years for stage 2, 16 years for stage 3, and 22 years for stage 4.

3.8 Age Estimation Report

According to the recommendations of the Study Group on Forensic Age Diagnostics (Schmeling et al. 2008), the collected findings and the determined stages are to be accessible in detail in the expert's report. The background of the case as given in the report should be able to stand alone and be understood without requiring other documents to be read (Schmeling et al. 2008).

The material:

In case of dead:

When did you obtain the material and how did you obtain it?

What did the material consist of?

How did you store it?

Did you score the material in any way?

Did you revisit the material or do you keep it?

In case of living person:

- Control the identity of the person.
- Make sure he or she understands the language.

Give background information relevant to the case. Describe your role.

Obtain consent to the examination.

The stage classifications and reference studies used need to be mentioned (Schmeling et al. 2008). Reference studies used for forensic age estimation should meet the following requirements:

Adequate sample size

Proven age of subjects

Even age distributions of subjects

Analysis separately for both sexes

Information on the time of examination

- Clear definition of the examined features
- Detailed description of the methods
- Data on the reference population regarding ethnicity, socioeconomic status, state of health data on the sample size, mean value, and range of scatter for each examined feature

To this end, the court's case files of the persons originally examined for age estimation purposes at the institute were asked to see whether the actual age of these persons was established during the court proceedings. In all cases where the age of the person concerned could be verified beyond uncertainty, the deviation between the estimated and actual ages ranged between ± 12 months (Schmeling et al. 2003). Furthermore, the expert's report should give the degree of probability that the stated age is the actual age or that the individual's age is above the relevant age limit. For this purpose, the following probability ratings can be used (Schmeling et al. 2003):

 "Almost certain probability (beyond reasonable doubt)": probability >99.8 % (this probability refers to the threefold standard deviation)

- 2. "Very probable" or "high probability": probability >90 %
- 3. "Probable": probability >50 %
- 4. "Undecided": probability =50 %
- 5. "Less probable" or "improbable": probability <50 %
- 6. "Very improbable": probability <10 %
- 7. "Almost certainly improbable": probability <0.2 %

The following phrasing gives an example of an adequate conclusion:

Summarizing all test results, the following can be established: There is a very high probability that the given date of birth is not correct but that an earlier date can be assumed. There is a very high probability of an age of above 13 years and a high probability of one above 15, there is but very low probability that the 18th year of age has been reached, and there is almost certain probability that the 20st year of age has not been reached.

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Craniofacial Identification

Contents

4.1	Introduction	65
4.2	Anthropometry	65
4.3	Photo-anthropometry	66
4.4	Morphology	66
4.5	Superimposition	66
4.6 4.6.1	Facial Reconstruction Traditional Method or Plaster Scalp Reconstruction	67 68
4.6.2	Three-Dimensional Facial Reconstruction via Computer Graphics	68
4.7	Biometric System of Facial Identification	70
Refer	ences	71

4.1 Introduction

Craniofacial identification is a forensic process where a photograph of a missing person is compared with a skull to determine its identity. There are different methods to sculpture a face onto the skull, all of which are based on the reproduction of a possibly recognizable face using the photos (Phillips and Smuts 1996). Facial reconstruction is a method used in forensic investigations to assist in identification. This chapter is focused on the role of craniofacial identification in forensic investigations.

4.2 Anthropometry (Bromby 2003)

The use of facial parameters as a form of evidence is new, and its reliability in medicolegal cases is debatable. Identification from photographs using facial anthropometry has been used in present-day medicolegal court cases (Bromby 2003). An investigative medicolegal report will usually contain multiple comparisons (Bromby 2003). According to Bromby, "Facial mapping is defined as the process of identifying which individual through the facial features and can include video/photo superimposition, anthropometric measurements and morphological comparisons" (2003, p. 302).

4.3 Photo-anthropometry

Photo-anthropometry is defined as the analysis of anthropometric landmarks, dimensions, and angles to quantify facial characteristics and proportions from a photograph (Iscan 1993). The main objective of this concept is to get information concerning the real objects illustrated in a photograph from this two-dimensional image. It is frequently used in the examination of traffic accidents, major disasters, and crime scenes.

4.4 Morphology

When the morphological characteristics of an individual are documented, the height, weight, and body type are the noted first. Hair, racial, and ethnic facial appearances, the color of the individual's skin, and any distinctive individuality such as tattoos, surgical scars, and other scars, congenital deformities, and so forth are documented if feasible. It has been reported that camera angles and camera distance can all affect the appearance of the individual and make identification more difficult. It has been reported that emotions such as anger, fear, happiness, and surprise can significantly change facial features (Ïscan 1993). Physiological and pathological changes such as disease, aging, and exposure to the elements also affect morphology (Donofrio 2000).

4.5 Superimposition

Superimposition is a method of identification that involves photographic and video superimposition. If no medical or dental records are available, but one has an idea about the identity of the deceased person, then a skull and a photograph can be superimposed as well as two photographs. Before the person's identity has been formally determined, dental and other records, if available, should be examined for further verification. Video superimposition on dead persons is better used to exclude someone rather than to provide positive identification because it can certainly be stated that a skull and photograph may not be of the identical individual (Vanezis and Brierley 1996). In order to ascertain a positive identification, the two images should be taken with the same viewpoint or at least from a very similar position (Vanezis and Brierley 1996). When both images are from living individuals, as opposed to one being an image of a skull, landmarks vary according to the position from which the image was obtained (Shahrom et al. 1996). Landmarks include the nasal cavity, bite line, and eye sockets on a skull, which are usually aligned (Yoshino et al. 2000). It has been reported that special attention is paid to the curves of the forehead, the depth of the nasal bridge, the shape and projection of the nasal bones, the lower face and chin prominence, and the height of the vault, when comparing a skull to a lateral photograph, while attention is focused on the orbital size and shape, the breadth of the nasal bridge, the width of the nasal aperture, the total facial length and width, the ratio of mid-face to upper or lower face length, and the mandibular shape with a frontal photograph. As the images are in position, one image is superimposed on top of the other, keeping the horizontal, vertical, or diagonal alignment (Austin-Smith and Maples 1994).

The skull landmarks (George 1993) that are often used follow:

Craniometric landmarks (Fig. 4.1):

Dacryon (Da): the point of junction of the frontal, maxillary, and lacrimal bones on the lateral wall of the orbit

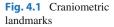
Frontomalar temporal (Fmt): the most lateral point of junction of the frontal and zygomatic bones

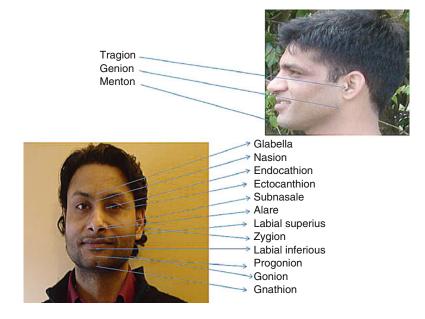
Glabella (G): the most prominent point between the supraorbital ridges in the midsagittal plane

Gnathion (Gn): a constructed point midway between the most anterior (Pog) and most inferior (Me) points on the chin

Gonion (Go): a constructed point; the intersection of the lines tangent to the posterior margin of the ascending ramus and the mandibular base, or the most lateral point at the mandibular angle

Nasion (N): the midpoint of the suture between the frontal and the two nasal bones





Nasospinale (Ns): the point where a line drawn between the lower margins of the right and left nasal apertures is intersected by the midsagittal plane (MSP)

Pogonion (Pog): the most anterior point in the midline on the mental protuberance

Prosthion (Pr): the apex of the alveolus in the midline between the maxillary central incisor Zygion (Zy): the most lateral point on the zygomatic arch

Cephalometric landmarks

Alare (al): the most lateral point on the alar contour

Ectocanthion (Ec): the point at the outer commissure (lateral canthus) of the palpebral fissure just medial to the malar tubercle (of Whitnall) to which the lateral palpebral ligaments are attached

Endocanthion (En): the point at the inner commissure (medial canthus) of the palpebral fissure

Glabella (g'): in the midline, the most prominent point between the eyebrows

Gnathion (gn'): the point on the soft tissue chin midway between the Pog and Me

Gonion (go'): the most lateral point of the jawline at the mandibular angle

Menton (Me): the lowest point on the MSP of the soft tissue chin

Nasion (n): in the midline, the point of maximum concavity between the nose and forehead; frontally, this point is located at the midpoint of a tangent between the right and left superior palpebral folds

Pogonion (pog'): the most anterior point of the soft tissue chin

Labiale inferius (Li): the midpoint on the vermilion line of the lower lip

Labiale superius (Ls): the midpoint on the vermilion line of the upper lip

Subnasale (sn): the midpoint of the columella base at the angle where the lower border of the nasal septum meets the upper lip

Tragion (t): point in the notch just above the tragus of the ear; it lies 1-2 mm below the spine of the helix, which can be palpated

Zygion (Zy'): the most lateral point of the cheek (zygomaticomalar) region

4.6 Facial Reconstruction

Facial reconstruction is an identification method used to assist in building a "living" face out of skeletal remains. The reproduction of facial characters is based upon the soft tissue thicknesses over the underlying bony structure of the skull (Philips et al. 1996). An anthropologist can identify race, sex, and age from skull. Before any facial reconstruction is attempted, a correct anthropological assessment of the skull to determine sex, age, and race is of crucial significance. This method of identification is employed depending on the available evidence. Facial reconstruction is used as a last alternative when other identifying aspects such as DNA or dental records are not available.

4.6.1 Traditional Method of Plaster Scalp Reconstruction

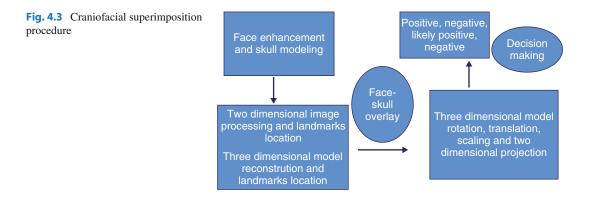
The traditional method of facial reconstruction is very simple. Its tools consist of clay modeling tools and a packet of cocktail sticks. It also needs the eyes and hands of an artist and the specialized skill of face anatomy. It starts with a cast of the skull made with alginate. It's ideal to cast the skull and mandible together and fit the fake eyes before casting. As the two halves of the mould are opened and the alginate peeled off, the replica of the skull comes out. Then the position of the skull should be determined (Lane 1992). Pegs that are 2-3 mm in diameter are fixed at the distance according to the thickness of the soft tissues based on the age, sex, and ethnic group. The medial and lateral canthi of the eye are noticeable with copper pin pegs projected from the nasal aperture. Muscles are constructed by using special clay known as cornish pot clay with fine grog from the surface of skull outward (Lebedinskaya et al. 1986). The sequence of muscle construction is temporal muscle, masseter, buccinator, and orbicularis oris. The width of the mouth is estimated by the outer borders of the canine teeth. If the teeth are missing, then the width is same as the distance between the inner borders of the iris. Expression muscles such as the levator anguli oris, levator labii superioris, zygomaticus major and minor, depressor labii inferioris, and depressor anguli oris are constructed. A further procedure is the design of the general size and shape of the nose; the width of the nasal aperture in the skull is equal to three fifths of the overall nose's width. The angle at which the eye incline can be estimated from the skull. Now the neck can be constructed. The next step is to wrap up the whole sculpture with a layer of clay to imitate the outer layers, known as wide strips. The ears and mouth are the most provisional features. The mouth has to be created in harmony with the rest of the face. Ears are not very important. Reconstructions are usually broadcast on TV news, displayed by other media, and printed on posters. The average success is considered to be between 30% and 70% (Lebedinskaya et al. 1986).

4.6.2 Three-Dimensional Facial Reconstruction via Computer Graphics

Facial reconstruction has become an important tool in forensic investigations. This method has provided an answer to basic limitations of the classic craniofacial reconstruction methods using clay, such as its being time-consuming, highly subjective, and requiring artistic capacity (De Greef and Willems 2005). In this field, computers were commonly used for many years to store and retrieve images from facial feature databases. Recently, new software has proposed altering the occurrence of facial features of a given reference facial image by means of three-dimensional (3D) digitized images, graphical modeling, and animation (Quatrehomme et al. 1997). These methods utilize computer programs to convert laserscanned 3D skull images into faces. Also, the results are more reproducible, but subjectivity could remain in the "pegging" of a complex facial image onto the digitized skull matrix (Fig. 4.2). A database of head models (characteristics such as skulls and faces) and soft tissue depth, with their personal characteristics (age, sex, race, and nutritional status), is obligatory. It uses the same relationship between soft tissue depth and the underlying bone -as used in traditional construction (Tyrell et al. 1997). The remains of the dead are first freely examined by a team of a forensic pathologist (postmortem examination), a forensic anthropologist (osteological study such as age, sex, race, ethnicity, and nutrition), a forensic radiologist (radiographs, laser, MRI, and CT scanning), and a forensic odontologist (dental analysis). The same 40 landmarks are available to



Fig. 4.2 Computer-aided three-dimensional facial reconstruction



be placed on the image of the face. The operator aligns the skull with the face and the markers are rechecked. The 3D image of the skull is superimposed on the face (Fig. 4.3). The peg markers are matched and moved manually on the average face. The computer then reconstructs an image of the face on the skull. The final image can be printed out or photographed directly from the monitor and can be saved for future reference. The information given by the analysis is developed by the model in order to decide the suitable skull and soft tissue templates (Tyrell et al. 1997). A color laser scanner and graphical software are used to generate a depiction of the scanned skull as a matrix made up of a number of latitudes and longitudes. The skull is placed on head holder. As the skull turns on the stand, the longitude changes and the "radius" is determined for each latitude. So a "wireframe" of 256×256 radii is created. The wireframe matrix of the skull must be transformed using tissue depth measurements to generate the base of the facial reconstruction. Digitized images of facial features, not calculated by the skull contours such as nose, eyes, and

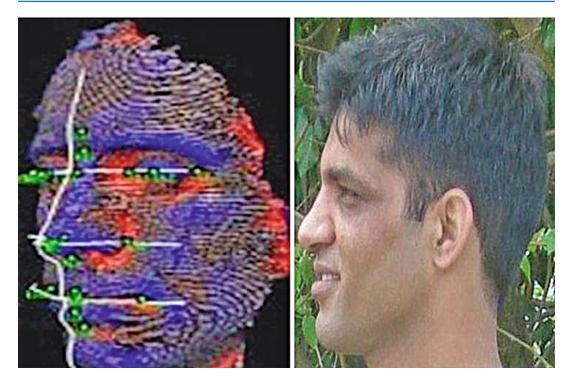


Fig. 4.4 Biometric system of facial identification

mouth, should then be additional with separate means to generate a wireframe face, onto which color and texture can later be provided. So the generated image can be obtained in many ways (Miyasaka et al. 1995). CT and MRI are the latest scanning methods used to precisely detect the margins of bone and soft tissues of various kinds and to make accurate measurements of tissue depths (De Greef and Willems 2005).

4.7 Biometric System of Facial Identification

Biometric systems of facial identification are automated facial recognition systems to compare offender and suspect images. They are an important tool in identification in different places, such as airports and banks. Such a system is based on the biometric analysis of facial characteristics. A biometric system can be divided into two components, such as the enrollment module and the identification module (Pankanti et al. 2000). The enrollment module is the component of the system that trains it to identify a definite individual, while the identification module is the component of the system that recognizes the person. For example, in the enrollment stage of a facial recognition system, the person's face would be scanned and a template would be produced from a digital representation. Incorporated in the template may be the size and positions of the nose, mouth, and eyes (Fig. 4.4). Templates of all individuals included would be stored in a database. During the identification phase, the biometric sensor scans the person and acquires the biometric used for identification, in this case the face, and the data are transferred into the same type of template used in the prior phase. This template is compared alongside each of the earlier stored templates to determine if there is a match. A big issue facing many private citizens is the effect of biometric identifications on an individual's personal privacy.

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Sex Determination Methods in Forensic Odontology

Contents

5.1	Introduction	73
5.2	Determining Sex from Craniofacial Morphology and Dimensions	73
5.3	BR Regression Equation for Sex Determination	74
5.4	Sex Determination Using Canine Dimorphism	75
5.4.1	Dental Index	75
5.4.2	Odontometric Difference	76
5.5	Sex Determination Using Barr Bodies	77
5.6	Sex Determination Using DNA	78
5.7	Sex Determination from the Enamel Protein	78
Refere	nces	79

5.1 Introduction

Determining the sex of unknown human remains is the important step in the triad of building a dental profile. Forensic odontology plays an important role in establishing the sex of the victims with bodies damaged beyond recognition due to major mass disaster. Sex can be determined based on data from the morphology of the skull and mandible, soft tissues, and metric features, as well as by DNA analyses of teeth.

5.2 Determining Sex from Craniofacial Morphology and Dimensions

The use of morphological features of the skull and mandible is a common approach used by anthropologists in sexing (Sweet 2001). A number of features are known to show variation between the sexes. The use of multiple features tends to be more accurate when used with young adults and the middle-aged. Williams and Rogers (2006) achieved 96% success in determining the sex using different features of the skull and the mandible. They also observed that using a constant six traits-mastoid, supraorbital ridge, size and architecture of skull, zygomatic extension, nasal aperture, and mandibular gonial angle-the accuracy was 94%. This indicates that craniofacial morphology can be used to determine the sex of skeletal specimen with a high degree of precision. The mandible is the largest and hardest facial bone and keeps its shape better than other bones in the forensic

Discrimination of ge		
Traits	Male	Female
General size	Large	Small
Architecture	Rugged	Smooth
Supraorbital ridge	Medium to large	Small to medium
Mastoid processes	Medium to large	Small to medium
Occipital area	Muscle lines and protuberance marked	Muscle lines and protuberance not marked
Frontal eminence	Small	Large
Parietal eminence	Small	Large
Orbits	Squared, lower, relatively smaller, with rounded margins	Round, higher, relatively larger, with sharp margin
Forehead	Steeper, less rounded	Rounded, full, infantile
Cheekbones	Heavier, more laterally arched	Lighter, more compressed
Mandible	Larger, higher symphysis, broader ascending ramus	Small, with less corpal and ramus dimensions
Palate	Larger, broader, tends to be U-shaped	
Occipital condyle	Larger	Small
Teeth	Larger, lower M1 more often cusped	Small, more often 4-cusped
	(100()	

 Table 5.1
 Discrimination of gender

Source: From Krogman and Iscan (1986)

and dental anthropologic fields. It has been reported that of 13 nonmetric items of the mandible, the characteristic that best permitted the sexes to be distinguished was the contour of the lower border of the mandible: Rocker-shaped mandibles predominated in males (68.1%), whereas most females (84.6%) exhibited a straight mandible. In addition, the mental region was shaped differently between the sexes: The shape of the chin in most males was generally bilobate or square (91.7%), whereas the chin in females was either square (45.5%) or pointed (54.5%). In this study, the positive predictive values of male and female were 92.5% and 73.7%, respectively (Hu et al. 2006). Five variables were developed from the complete cranium, vault, face, mandible, and bizygomatic breadth. Dimensions from the complete cranium provided the best accuracy. In the mandible, bigonial breadth was the most dimorphic of the measurements taken. Average accuracies ranged from 80% (bizygomatic breadth alone) to 86% (cranium). Diagnostic accuracy, however, was lower than that obtained from the femur and tibia (Steyn and Iscan 1998). Rai et al. (2007) proposed that five BR criteria of sex determination from mandible parameters such as intercanine distance, intercondylar distance, interlingula (covering inferior alveolar foramen), mesiodistal diameter of mandibular canines, and mental foramen to lingula (covering inferior alveolar fora-

men). BR criteria for determination of sex reveal that the probability of male sex is 95% if three or more of the dimensions are greater than their deviation values. Krogman's cranioscopy for sex determination is based on 14 characteristics of the skull (Table 5.1). On the other hand, a modified Krogman's cranioscopy trait by grading was used to simplify gender classification since it was based on a four-point grade (i.e., 0, smooth; 1, small or rough; 2, medium or rougher; and 3, large or very rough) of only three external characteristics of the skull: supraorbital torus, glabella region, and external occipital protuberance (Krogman and Iscan 1986). These authors conducted a study to evaluate the sexual dimorphism in the cranium and mandible of North Indians using Krogman's cranioscopy and the modified Krogman's cranioscopy trait by grading (Table 5.2).

5.3 BR Regression Equation for Sex Determination

Teeth may be used for differentiating sex by measuring their mesiodistal (MD) and buccolingual (BL) dimensions. Various studies have shown significant differences between male and female permanent and deciduous tooth

Characteristics	Male (%)	Female (%)	<i>p</i> value
Supraorbital torus		× /	1
Smooth	6.2	71.3	
Small	23.3	20.0	
Medium	35.4	6.0	0.05
Large	34.3	2.7	
Glabella region	0	0	0.001
Smooth	1.4	68.7	
Rough	6.2	23.9	
Moderately rough	13.2	8.7	
Very rough	80.0	6.7	
External occipital protuberance		0	0.001
Smooth	2	37.3	
Rough	13.3	51.3	
Moderately rough	29.3	6.7	
Very rough	56.4	4.7	

 Table 5.2 Discrimination of gender using modified Krogman's cranioscopy trait by grading in North Indian population

crown dimensions. In the majority of studies, the canines have consistently shown the maximum sex difference. It has been reported that sex can accurately be determined from maxillary and mandibular canines and mandibular second molar in 77% of cases (Iscan and Kedici 2003). Canines showed the greatest univariate sex dimorphism, followed by the buccolingual (BL) dimension of maxillary first and second molars in Nepalese (Acharya and Mainali 2007). The mandibular canines are considered to demonstrate high sexual dimorphism among teeth in their mesiodistal width. Maxillary first molars (BL diameter), especially the right side, have a higher sexual dimorphism (8.49% casts, 8.27% intraorally) as compared to the left side and hence help in forensic dentistry during an impacted canine (Rai et al. 2008).

Rai et al. (2004) proposed a regression for sex determination from teeth as follows:

Sex =1.528 (Maxillary central incisor B-L) – 1.322 (Premolar B-L) + 1.94 (Maxillary first molar B-L) -0.97 (Canine M-D) + 0.837 (Canine B-L)

This formula is known as the BR formula. This division point in males is greater than 16.932 and in females less than 16.392. Note that such a function would vary among populations because of variations in the absolute size of the teeth. Even if

a similar coefficient worked, the duration point would be different.

5.4 Sex Determination Using Canine Dimorphism

A study by Anderson and Thompson (1973) observed that the mandibular canine width and intercanine distance were greater in males than in females and permitted a 74.3% correct classification of sex. It has been reported that sexual dimorphism by measuring the mesiodistal width of canine teeth in different ethnic groups. They concluded that the magnitude of canine teeth sexual dimorphism varies among different ethnic groups. Furthermore, the mandibular canine showed a greater degree of sexual dimorphism than the maxillary canine (Garn et al. 1973). Rao et al. (1988) reported that the mesiodistal width of mandibular canines was significantly greater in males than in females. Rai et al. (2006) reported that the mesiodistal and buccolingual widths of mandibular canines were significantly greater in males than in females.

5.4.1 Dental Index

In relation to absolute tooth size, tooth proportions have been suggested for differentiating between males and females. Aitchison presented the "Incisor index" (Ii), which is calculated by the formula

where MDI 2 is the maximum mesiodistal diameter of the maxillary lateral incisor and MDI1 is the maximum mesiodistal diameter of the central incisor (Rai et al. 2006). Another index, the "mandibular canine index" (Fig. 5.1), proposed by Rao et al. (1988), gives an accurate indication of sex in an Indian population as follows:

Canine Canine

[(Mean m-d canine dimens(Mean m-d canine dimension in female + S.D.) in males–S.D)] /2

Fig. 5.1 Mandibular canine index

[(Mean m-d canine dimension + (Mean m-d canine dimension in female + S.D.) in males – S.D)] / 2

The value obtained using this formula was 7.1; that is, 7.1 mm is the maximum possible mesiodistal dimension of mandibular canines in females. The same dimension is greater in males. The success rate of determining sex using the above formula was close to 89%.

5.4.2 Odontometric Difference

The odontometric difference between males and females is generally explained as a result of greater genetic expression in males, as shown in Tables 5.3 and 5.4 (Rai et al. 2006). The canines followed by premolars showed the highest sex dimorphism compared to other teeth. However, in the buccolingual dimension, the mandibular canine showed more dimorphism and in the mesiodistal dimension, the maxillary canine had more diphormism (Fig. 5.2, Table 5.3). The distolingual–mesiobuccal dimension of canines showed more dimorphism than other teeth. The distolingual–mesiobuccal dimension of maxillary and mandibular premolars showed greater sexual dimorphism (Table 5.4).

Table 5.3 Mean values and standard deviation of different diameter (MD, BL) in mm of different teeth of both genders(FDI system followed for tooth designation)

	MD		BL	
	(Mean+S.D.)	(Mean+S.D.)		
Tooth	Males	Females	Male	Females
11	8.76+0.89*	8.36+0.82*	7.23+0.84	7.22+0.62
12	6.93+0.67	6.64 + 0.57	6.37+0.89	6.25+0.87
13	7.55+0.87	7.23+0.93	7.63+0.69*	6.79+0.72*
14	6.72+0.92	6.69+0.69	9.21+0.42	9.11+0.89
15	6.56+0.53	6.23+0.52	9.72+0.52	9.19+0.79*
16	11.57+0.92	11.43+0.52	11.14+0.60	10.84+0.59
17	11.00+0.63	10.57 ± 0.72	11.12+0.72	10.78+0.79
41	6.42+0.55	6.38+0.53	6.29+0.78	6.12+0.84
42	6.32+0.49	6.30 ± 0.42	6.52+0.82	6.42+0.93
43	7.54+0.22*	6.32+0.31*	7.55+0.87	7.32+0.82
44	7.12+0.42	6.99 ± 0.47	7.78 ± 0.84	7.52+0.82
45	7.82+0.63	7.72+0.69	8.73+0.52	8.13+0.62
46	11.52+0.66	10.74 ± 0.62	10.47 ± 0.84	10.19+0.68
47	10.44+0.62	10.22+0.79	10.32+0.75	9.97+0.73

Source: Rai et al. (2006) **p* <0.01

Mandibular canine index

	DB-ML		DL-MB	
	(Mean+S.D.)		(Mean+S.D.)	
Tooth	Males	Females	Male	Females
11	7.52+0.89	7.46+0.82	8.42+0.69*	7.92+0.62*
12	6.59 ± 0.62	6.37+0.82	$6.92 \pm 0.72^*$	6.32+0.73*
13	7.62+0.56*	7.21+0.52*	8.32+0.59*	7.23+0.52*
14	8.54+0.89	8.23+0.93	8.34+0.52	8.32+0.57
15	8.57+0.57*	8.32+0.53*	9.12+0.73	9.08+0.53
16	11.53+0.84	11.52+0.94	12.78+0.54*	12.22+0.59*
17	11.07+0.52*	9.54+0.54*	11.89+0.39*	10.98+0.62*
41	5.79 ± 0.87	5.72+0.62	5.82+0.42	5.72+0.53
42	6.12+0.73	6.02+0.82	6.32+0.57	6.22+0.54
43	6.82+0.59*	6.42+0.53*	7.68+0.66*	7.19+0.53*
44	7.13+0.87	7.03+0.63	7.84+0.52*	7.23+0.57*
45	8.13+0.82	7.99+0.82	8.52+0.62*	7.53+0.63*
46	11.58+0.42*	$11.17 \pm 0.47*$	11.74+0.57*	11.32+0.52*
47	11.39+0.53*	10.84 + 0.57*	11.62+0.62	11.52+0.62

Table 5.4 Mean values and standard deviation (mean + S.D.) in mm of tooth diameters (DB-ML and DL-MB) between male and female diameters (FDI system followed for tooth designated)

Source: Rai et al. (2006) **p*<0.01

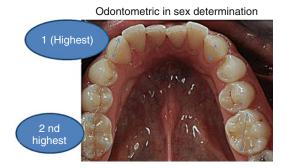


Fig. 5.2 Sex dimorphism in dentition

5.4.2.1 Measurement Methods

Each tooth was measured in four different dimensions: buccolingual (BL), mesiodistal (MD), distobuccal-mesiolingual (DB-ML), and distolingualmesiobuccal (DL-MB) (Rai and Anand 2007):

- The buccolingual crown diameter is the greatest distance between the buccal surface and lingual surfaces of the crown, measured with a vernier caliper held at a right angle to the mesiodistal crown diameter of the tooth.
- The mesiodistal crown diameter is the greatest distance between the approximal surfaces of the crown measured with a vernier caliper held parallel to the occlusal and vestibular

surfaces of the crown. If a tooth was malpositioned or rotated in relation to the dental arch, mesiodistal measurements were taken between the contact points of the crown.

- The distobuccal-mesiolingual crown diameter is the greatest distance between the distobuccal and mesiolingual surfaces of the crown, creating a diagonal in relation to the tooth and with the vernier caliper held parallel to the occlusal surface of the crown.
- The distolingual-mesiobuccal crown diameter: is the greatest distance between the mesiobuccal and distolingual surfaces of the crown, creating a diagonal in relation to the tooth and with the vernier caliper held parallel to the occlusal surface of the crown.

5.5 Sex Determination Using Barr Bodies

The presence or absence of an X-chromosome can be studied from buccal smears, a skin biopsy, blood, cartilage, a hair root sheath, and tooth pulp (Rai 2010). After death, it perseveres for variable periods depending upon the humidity and temperature in which tissue has remained. The

Fig. 5.3 Barr body (female)

Barr body

Barr body (saliva from female)

X-chromatin and intranuclear structure is also known as the Barr body (Barr et al. 1950). It is present as a mass usually lying against the nuclear membrane in the females. It has been shown that up to a period of 4 weeks after death, we can determine the sex accurately from the study of X- and Y-chromosomes, keeping in mind the variations in temperature and humidity. It has been reported that sex determination from necrotic pulp tissue stained by quinacrine mustard using a fluorescent Y-chromosome test for maleness and claimed that up to 5 weeks after death, sex determination can be done with a high degree of accuracy (Whittaker et al. 1975). Duffy et al. (1991) have shown that Barr bodies (Fig. 5.3) and F bodies of Y-chromosomes are preserved in dehydrated pulp tissues up to 1 year and that pulp tissues retain sex diagnostic characteristics when heated up to 100°C for 1 h.

5.6 Sex Determination Using DNA

Polymerase chain reaction (PCR) is a method of amplifying small quantities of relatively short target sequences of DNA using sequence-specific oligonucleotide primers and thermostable Taq DNA polymerase (Tsuchimochi et al. 2002). A procedure utilizing Chelex 100, chelating resin, was adapted to extract DNA from dental pulp. The procedure was simple and rapid, involved no organic solvents, and did not require multiple tube transfers. The extraction of DNA from dental pulp using this method was as efficient, or more so, than using proteinase K and phenol-chloroform extraction (Tsuchimochi et al. 2002). The determination of sex from blood and teeth by PCR amplification of the alphoid satellite family using amplification of X- (131 bp) and Y- (172 bp) specific sequences in males and Y-specific sequences in females has been reported. It was shown to be a useful method in determining the sex of an individual (Hanaoka and Minaguchi 1996).

5.7 Sex Determination from the Enamel Protein

Amelogenin, or AMEL, is a major matrix protein found in the human enamel. It has a different signature (or size and pattern of the nucleotide sequence) in male and female enamel. The AMEL gene that encodes for female amelogenin is located on the X-chromosome, and the AMEL gene that encodes for male amelogenin is located on the Y-chromosome. The female has two identical AMEL genes or alleles, whereas the male has two different AMEL genes. This can be used to determine the sex of the remains with very small samples of DNA (Hanaoka and Minaguchi 1996).

Conclusions

Determining sex using skeletal remains presents a great problem to forensic investigators, especially when only fragments of the body are recovered. Forensic odontologists can assist other experts to determine the sex of remains by using teeth and the skull. The durability of teeth in a fire, during bacterial decomposition, and when exposed to environmental factors makes them helpful for identification and sex determination.

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Tooth Morphology

6

Contents

6.1	Introduction	81
6.2	Incisors	81
6.2.1	Upper Incisor	81
6.2.2	Upper Lateral Incisor	82
6.2.3	Lower First Incisor	82
6.2.4	Mandibular Lateral Incisor	82
6.3	Canine Teeth	82
6.3.1	Upper Canine	82
6.3.2	Lower Canine	82
6.4	Premolars	82
6.4.1	Upper First Premolar	82
6.4.2	Upper Second Premolar	83
6.4.3	Lower First Premolar	83
6.4.4	Lower Second Premolar	83
6.5	Molars	83
6.5.1	Upper First Molar	83
6.5.2	Maxillary Second Molar	83
6.5.3	Maxillary Third Molar	83
6.5.4	Lower First Molar	83
6.5.5	Lower Second Molar	83
6.5.6	Lower Third Molar	84
6.6	Deciduous Teeth	84
6.6.1	Upper Central Incisor	84
6.6.2	Upper Second Incisor	84
6.6.3	Lower Central Incisor	84
6.6.4	Lower Lateral Incisor	84
6.6.5	Lower Canine	84
6.6.6	Upper First Molar	84
6.6.7	Deciduous First Molar	84
6.6.8	Upper Second Deciduous Molar	85
6.6.9	Lower Second Deciduous Molar	85
Refere	ences	85

6.1 Introduction

During the examination of dentition, the forensic odontologist plays a vital role in identifying an individual as accurately as possible from the natural teeth present in mouth. The morphological details of teeth can be used for identification.

It is difficult to identify the tooth morphology of the permanent molar, upper premolars and lower central incisors, deciduous canines, and some other deciduous teeth (Ash and Nelson 2002; Rai 2010).

6.2 Incisors

6.2.1 Upper Incisor

- The labial surface
 - The labial surface, bordering toward the root in a convex line, widens slowly toward the incisal edge.
 - The mesial border joins the edge at almost a right angle.
 - The distal corner is markedly rounded.
 - The labial surface is quadrilateral.
- The lingual surface
 - It is deeply concave.
 - The dental tubercle is present in the cervical third.
 - The marginal parts after the lingual surface run as slight ridges toward the incisal edge, forming a horseshoe-shaped elevation with the tuberculum.

- The proximal surface
 - It is roughly triangular.
 - The line of the cementoenamel junction on the proximal surface is V-shaped.
- Root
 - The root is roughly cone-shaped.

6.2.2 Upper Lateral Incisor

- The upper lateral incisor is similar to the first incisor.
- The crown is more slender and only rarely grooved on its labial surface.
- The lingual surface is more deeply concave compared to the first incisor.
- The root of the upper second incisor is slender and more compressed in the mesiodistal direction and a groove is present.

6.2.3 Lower First Incisor

- The crown is chisel-shaped.
- The labial surface is slightly convex; its mesial and distal borders meet the incisal edge at right angles.
- The lingual surface is convex in its cervical part and flat concave in its greater central and incisal parts.
- The proximal surface is triangular.
- The root of the lower first incisor is markedly flattened in the mesiodistal dimension.
- The lower first incisor is the smallest tooth of permanent human dentition.

6.2.4 Mandibular Lateral Incisor

- The second mandibular incisor shape resembles the first but is slightly larger.
- The most important difference between them is the enhanced divergence of the mesial and distal surfaces in the second incisor.
- The mesial surface is nearly vertical.
- The distal surface deviates toward the incisal edge.

Note: The side of the lower first incisor can be difficult to identify.

6.3 Canine Teeth

6.3.1 Upper Canine

- The upper canine has a pointed cusp.
- The mesial edge is shorter and not as steeply inclined as the longer distal edge.
- The labial surface is convex.
- The lingual surface is concave.
- The proximal surface is triangular and has a broad base as the triangle is convex occlusally.
- The root of the upper canine is the longest and strongest of the human dentition; the cross section of root is triangular, oval at the labial border, being broader and plumper than the lingual border.

6.3.2 Lower Canine

- The lower canine has a higher, narrower crown with a less pointed cusp.
- The slenderness of the lower tooth is responsible for a less divergent position of the mesial and distal surfaces.
- The enamel on the labial surface of the lower canine extends further apically than that on the lingual surface.

6.4 Premolars

6.4.1 Upper First Premolar

- The occlusal surface is oval.
- The proximal surface is roughly rectangular and slightly converse, the distal being a stronger converse.
- The proximal surface converges lingually.
- The buccal surface of the upper first premolar is strikingly similar to that of the canine.
- The upper first premolar has marked concavity on the mesial surface at the cervical margin.
- The upper first premolar sometimes has two roots.
- The two cusps are roughly cone-shaped, the buccal cusp always being larger in circumference and higher than the lingual cusp.

6.4.2 Upper Second Premolar

- The upper second premolar has smaller cusps of almost equal size, often set closer together.
- The crown appears slightly compressed in a buccolingual direction.
- It has a flat mesial surface.
- As seen from the occlusal surface, the crown of the second premolar is more symmetrically shaped than that of its mesial neighbor.

6.4.3 Lower First Premolar

- The occlusal surface is circular.
- It has a large buccal cusp and a very small lingual cusp.
- The lower first premolar often has a fissure running from the mesial pit over onto the flattened mesial surface.
- The lingual cusp is usually skewed slightly to the distal.
- The labial surface of the lower first premolar is inclined so lingually that the tip of the buccal cusp lies almost above the center of the cervical cross section of the tooth.
- The lingual surface is slightly narrower.
- The lingual surface is lower than the labial surface.

6.4.4 Lower Second Premolar

- The crown of the lower second premolar is larger than that of the first.
- There is greater development of the lingual cusp.
- It has one or two lingual cusps more closely approximating the buccal cusp in height.
- The lingual surface is slightly narrower and slightly lower than the buccal surface and is often asymmetrical in shape if the lingual cusp has shifted mesially; the distolingual groove is almost always present and is often deep.
- The root of the lower premolar is stronger than that of the first in cross section, nearly circular.

6.5 Molars

6.5.1 Upper First Molar

- The upper molar has three to four cusps.
- It consists of three roots.
- It has a diamond-shaped occlusal surface.
- It has four well-developed cusps, and the tubercle of Carabelli is mesiolingual.
- It has long roots that are well shaped and well separated.

6.5.2 Maxillary Second Molar

- It has a triangular-shaped occlusal surface.
- There is a varying degree of reduction of the distolingual cusp, which may be slightly or considerably reduced.
- It has short roots that are more irregular and may be completely fused.

6.5.3 Maxillary Third Molar

- Occlusal surface varies.
- Roots vary.

Note: If the distolingual cusp is present, the molar may be easily identified on the basis of the feature of the oblique ridge connecting the distobuccal and mesiolingual cusps.

• It has a flatter mesial surface, a more convex distal surface, and a larger mesiobuccal cusp compared with the distobuccal cusp.

6.5.4 Lower First Molar

- It consists of two roots.
- It usually has five cusps.
- The crown is regular and well shaped.
- Roots are long, well shaped, and separated.

6.5.5 Lower Second Molar

- It usually has four cusps.
- Crown usually regular and well shaped.
- Roots are shorter and more irregular and can be fused.

6.5.6 Lower Third Molar

- It may have four to five cusps.
- Crown is usually irregular in shape.
- Roots vary.

Note: If the molar has five cusps, it can be examined by the position of the smallest cusp at the distobuccal corner.

• It has a flatter mesial surface, a more convex distal surface, a curved buccal surface, and a vertical lingual surface.

6.6 Deciduous Teeth

6.6.1 Upper Central Incisor

- The mesial corner is sharp and almost at a right angle.
- The distoincisal corner is well rounded.
- The lingual tubercle is always well developed.
- The root of the upper central deciduous incisor is slightly compressed in the labiolingual direction.

6.6.2 Upper Second Incisor

The upper lateral incisor duplicates the shape of its mesial neighbor much more closely in the deciduous dentition than in the permanent dentition.

6.6.3 Lower Central Incisor

- It is smaller than the lateral incisor.
- Its incisor shape is similar to the lateral incisor.

6.6.4 Lower Lateral Incisor

- It is larger than the central incisor.
- It shows a well-rounded distoincisal corner.
- As compared to permanent teeth, the roots of the deciduous mandibular incisors are far less flattened in the mesiodistal direction.

6.6.5 Lower Canine

- The labial surface is strongly convex.
- Convexity is strongest at the cervical border.
- As a rule, the tubercle on the lingual surface is well developed.
- The root of the upper deciduous canine is triangular in cross section.
- The lower deciduous canine, as a whole, looks narrower and therefore appears to be more slender.

6.6.6 Upper First Molar

- The occlusal surface of the upper first deciduous molar is irregularly quadrilateral.
- The distal border runs in a straight buccolingual direction and therefore joins the buccal and lingual borders at right angles.
- The mesial border is oblique in a mesiobuccal to distolingual direction.
- The buccal surface of upper first deciduous molar is wider in its mesial part than in its distal part because the enamel in the mesial part of the tooth reaches toward the root than distally.
- The upper first deciduous molar has three roots that are in a position similar to that set up in the permanent molars of the upper jaw.
- The molar tubercle of Zuckerkandl is always found at the mesiobuccal corner of the cervical margin.

6.6.7 Lower Deciduous First Molar

- The occlusal surface of the lower first deciduous molar is oval with a longer mesiodistal diameter.
- The mesial cusp is always larger than the distal one.
- The two buccal cusps are separated from the lingual part of the crown by a zigzagging mesiodistal groove that ends at the mesial and distal marginal ridges.
- The labial surface of the tooth is steeply inclined lingually, which accounts for the

relative narrowness of the occlusal surface in the buccolingual direction.

- The buccal cingulum is well developed also on the lower first deciduous molar, and here, too, a molar tubercle may be present in the mesiocervical part of the labial surface.
- The two roots of tooth, mesial and distal, are flat in the mesiodistal direction, especially the mesial root.

6.6.8 Upper Second Deciduous Molar

- The crown of the upper second deciduous molar is smaller than that of the first permanent molar, but otherwise it is almost identical in all particulars, even being the smallest.
- A Carabelli's tubercle may also be found on the mesial half of the lingual surface; in fact, it is more prominent in this tooth than in the first molar.
- The root of this tooth also closely resembles that of the first permanent molar, but their divergence, as a rule, is more pronounced.

6.6.9 Lower Second Deciduous Molar

- It is a slightly reduced replica of the first permanent molar.
- The only differences between these two teeth are a greater prominence of a buccal cingulum and a stronger convexity of proximal surfaces, which cause a conspicuous constriction of the cervical part of the deciduous tooth.
- The roots are always strongly divergent in their cervical half.

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Bite Marks: Evidence and Analysis, Part 1

7

Contents

7.1	Introduction	87
7.2	Definitions	88
7.3	Teeth Marks and Bite Marks	88
7.4	Class and Individual Characteristics	88
7.5	Additional Features	90
7.6	Locations of Bite Marks on Humans	90
7.7	Multiple Biting Episodes	91
7.8	Estimating the Age of Bite Marks	91
7.9	Concept of Uniqueness	91
7.10	Stepwise Process of Bite Mark Investigation	91
7.11	Responsibility of Forensic Odontologist in Bite Mark Cases	92
7.12	Collection of Evidence from Victim	92
7.12.1	Saliva Swabs of the Bite Sites	92
7.12.2	Photographic Documentation of the Bite Injury	93
7.12.2 7.12.3 7.12.4	Photographic Documentation of the Bite Injury Impressions Removal of the Bite Mark	93 94 95
7.12.2 7.12.3 7.12.4 7.12.5 7.13 7.13.1	Photographic Documentation of the Bite Injury Impressions Removal of the Bite Mark First Aid Evidence Collection from the Suspect History and Dental Records	93 94 95 95 95 95
7.12.2 7.12.3 7.12.4 7.12.5 7.13 7.13.1 7.13.2 7.13.3	Photographic Documentation of the Bite Injury Impressions Removal of the Bite Mark First Aid Evidence Collection from the Suspect History and Dental Records Photographs Extraoral and Intraoral Examination	93 94 95 95 95 95 95 96
7.12.2 7.12.3 7.12.4 7.12.5 7.13 7.13.1 7.13.2	Photographic Documentation of the Bite Injury Impressions Removal of the Bite Mark First Aid Evidence Collection from the Suspect History and Dental Records Photographs Extraoral and Intraoral Examination Impressions and Study Casts Test Bites	93 94 95 95 95 95 95
7.12.2 7.12.3 7.12.4 7.12.5 7.13 7.13.1 7.13.2 7.13.3 7.13.4 7.13.5	Photographic Documentation of the Bite Injury Impressions Removal of the Bite Mark First Aid Evidence Collection from the Suspect History and Dental Records Photographs Extraoral and Intraoral Examination Impressions and Study Casts	93 94 95 95 95 95 96 96 96 96

7.16	Forensic Physical Comparison of Exhibits	98
7.17	Reporting Conclusions and Opinions	98
Refere	nces	98

7.1 Introduction

Bite mark analysis is a vital area within this highly specialized field and constitutes the commonest form of dental evidence presented in criminal court. It is well known that assailants in sexual attacks, including sexual homicide, rape, and child sexual abuse, often bite their victims as an expression of dominance, rage, and animalistic behavior. The first formally reported case of dental identification was that of the 80-year-old English warrior John Talbot, Earl of Shrewsbury, who fell in the battle of Castillon in 1453 (Swanson 1967). The earliest recorded criminal case involving bite mark analysis was an American case (State of Ohio v. Robinson) in 1870, where in an individual named Robinson was charged with the murder of his mistress. Though there appeared to be a match between bite marks on the victim's arms and his teeth, Robinson was acquitted of the charge (Pierce 1996). In 1906, colliers was charged with breaking and entering

into a store followed by stealing some goods. During the examination of the premises, a block of cheese was discovered from which a piece had been bitten out, leaving teeth marks. Bite mark analysis and comparison led to his conviction (Pierce 1996). The scientific basis of bite mark analysis is rooted in the principle of the uniqueness of the human dentition, the belief that no two humans have identical dentitions in regard to size, shape, and alignment of the teeth (Heras et al. 2005). This declared uniqueness is transferred and recorded in the injury produced by the teeth during biting. The main aim of the analysis of bite mark cases is to connect the biter to the teeth pattern present in an object or the skin and to find out whether it is in any way related to the crime or event. The human skin has the ability to register sufficient detail of the teeth of the biter; however, it is quite variable. A number of studies have shown that the physical nature of the skin causes distortion of the bite marks. Moreover, the process of healing and decomposition produces changes in the bite marks that are left in the skin of dead or living individuals. Tooth markings may also be found in inanimate objects that might be associated with the crime, such as foods like chocolates, vegetables, chewing gums, Styrofoam cups, and cigarette butts, and even on the steering wheel of a car (Aboshi et al. 1994).

7.2 Definitions

According to authors of this chapter, a bite mark is defined as a patterned injury on the skin or other surface caused by the biting surfaces of human or animal teeth with a minimum amount of force. The ABFO manual defines a bite mark as "(1) a physical alteration in a medium caused by the contact of teeth, and (2) a representative pattern left in an object or tissue by the dental structures of an animal or human" (Freeman et al. 2005).

7.3 Teeth Marks and Bite Marks

The teeth can also leave marks without the act of biting, as is seen when the skin or the objects contact the teeth instead of the biter intentionally closing his jaws. So the teeth marks are reflexive, as there is no intentional, active, or reflexive jaw movement. These teeth marks are different from the bite marks where the muscles of the jaws are active, causing the teeth to move into the bitten substrate, which can be skin or any other object (Sweet and Pretty 2001).

7.4 Class and Individual Characteristics (Sweet and Pretty 2001)

Human bite marks are mainly found on the skin of victims, and they may be found on almost all parts of the human body. Females are frequently bitten on the breasts and legs during sexual attacks, while bites on males are usually seen on the arms and shoulders (Pretty and Sweet 2000; Vale and Noguchi 1983). A typical representative human bite (Fig. 7.1) is depicted as an elliptical or circular injury that records the specific characteristics of the teeth (American Board of Forensic Odontology 1995). It may be composed of two U-shaped arches (Fig. 7.2) that are separated at their bases by an open space. The diameter of the injury typically ranges from 20 to 45 mm (Rai B 2011). Frequently, a central area of bruising can be seen within the bite marks from the teeth. In the center of the bite mark injury, due to the pressure created by the biting teeth and by the negative pressure created by the tongue and suction, there is extravascular bleeding, which causes bruising. The color of these bruises changes over a period of time and the color also changes as the injury undergoes a healing process in the skin of a living individual. The tooth class characteristics and bite mark characteristics are the two types of class characteristics. In a bite mark, the front teeth, which include the central incisors, lateral incisors, and cuspids, are the primary biting teeth according to tooth class characteristics. There are 12 teeth markings in a bite mark that has both the maxillary and mandibular front teeth. Each type of tooth in the human dentition has class characteristics that differentiate one tooth type from the others. The two mandibular central incisors and the two mandibular lateral incisors are almost uniform in width, while the

Fig. 7.1 Human bite marks





Fig. 7.2 U-shaped arches (human bite marks)

mandibular cuspids are cone-shaped. When compared to the maxillary central incisors, the maxillary lateral incisors are narrower and the maxillary cuspids are cone-shaped. The upper jaw is wider than the lower jaw. The bite mark characteristics help us in determining which marks were made from the maxillary teeth and from the mandibular teeth. According to bite mark characteristics, the maxillary central incisors and lateral incisors make rectangular marks whose centrals are wider than the laterals, and the maxillary cuspids produce round or oval marks. The mandibular central incisors and lateral incisors also produce rectangular marks, but these are almost equal in width, whereas the mandibular cuspids produce round or oval marks. Sometimes the suspect may have a missing tooth/teeth, or due to breakage the tooth is shorter in size, or there may have been some clothing that prevented the tooth/teeth from contacting the skin. All these factors may lead to gaps that can be seen between the marks. Large carnivore or other animal bite marks such as dog bite wounds can be remarkable in their depth and amount of damage to the skin and underlying muscle. Use, misuse, and abuse of the teeth lead in exclusive features such as fractures, rotations, attritional wear, and congenital malformations, which are referred to as accidental or individual traits. When these are recorded in the injury, it may be possible to compare them to identify the specific teeth that caused the injury. These animals have extremely long canines and a complement of six incisors, plus the two canines, for a total of eight.

Note: The authors have witnessed blunt punch injuries that have resembled bite marks. These could be differentiated by the absence of class characteristics caused by human teeth in each case.

7.5 Additional Features (Bowers 2004)

Bite marks can have the following additional features:

- 1. Ecchymosis:
 - (a) Central ecchymosis: The negative pressure formed by the tongue and suction and the positive pressure created by closing

of the teeth cause extravascular bleeding due to interruption of the small blood vessels, producing central ecchymosis.

- (b) Linear abrasions, contusions, or striations: These are produced by slipping of the teeth against the skin or by imprinting of the lingual surfaces of the teeth on the skin.
- (c) Double bite: It is also called *bite within a bite* and is produced when, during initial contact with the teeth, the skin slips and the teeth contact again for the second time with the skin.
- (d) Weave patterns of interposed clothing.
- (e) Peripheral ecchymosis: It is produced when there is excessive and confluent bruising.
- 2. Partial bite marks:
 - (a) One arched: Also called half-bites.
 - (b) One or a few teeth.
 - (c) Unilateral marks: They are produced when the dentition is incomplete or when there is irregular pressure during biting.
- 3. Faded bite marks:
 - (a) Fused arches: In this there are no individual tooth marks.
 - (b) Solid: It is produced when the erythema or the contusion fills the entire central area of the bite mark. In this the bite mark does not show a ring pattern, but instead there is a discolored circular mark.
 - (c) Closed arches: In this the maxillary and the mandibular arches are joined at their edges.
 - (d) Latent: Seen with special imaging techniques.
- 4. *Superimposed bites:* Two bite marks superimpose each other.
- 5. *Avulsive bites:* In these the tissue is bitten off the victim during biting.

7.6 Locations of Bite Marks on Humans (Bowers 2004)

In case of sexual assault, females show evidence of bite marks on breasts, nipples, abdomen, thighs, and pubis, while males show bite marks mainly on the back, shoulders, and penis. In case of *self-defense*, individuals being attacked can receive bite marks from their attacker on their

	Adelson (1974)	Rentoule and Smith (1973)	Camps (1976)	Spitz et al. (1980)	Rai and Kaur (2011)
Initial colour	Red /blue	Violet	Red	Blue/red	Red
1-3 days	Blue/brown	Dark blue	Purple, black	Dark purple	Dark purple
1 week	Yellow/green	Green	Green	Green/yellow	Yellowish green
8-10 days		Yellow	Yellow	Brown	Green
2 weeks		Normal	Normal	Normal	Brown
20 days					Normal

Table 7.1 Ageing of bruises and bite marks

Table 7.2 Summary of documentation of bite mark and patterned injuries with photography (Rai and Kaur 2011)

Photography	Age estimation (Estimate of time to recover image)
Colour photography (400–700 nm)	12–16 days
Black and white (400–700 nm)	10–14 days
UV light (300-375 nm)	10-46 days
ALI light (350 nm)	5-9 days
IR light (750-900 nm)	7-13 days

forearms and hands. Initial animal attacks on humans focus on the legs and then advance to hands, arms, and the head and neck. On female victims of violence, these sites include the breasts, thighs, abdomen, pubis, and buttocks, while defensive wounds on the hands and forearms of a victim must also be considered an option.

7.7 Multiple Biting Episodes

Bruises of differing colors can point out a series of separate biting actions. Disappearance of skin injuries may be difficult to see without close examination under various types of light, such as ultraviolet, alternative, and infrared lights (as described in Chap. 14).

7.8 Estimating the Age of Bite Marks

Bruises in the skin of humans change color as healing takes place. These color changes are different from person to person. Age estimation of bite marks is neither a scientific nor an accurate process. Ageing of bruises and bite marks can be used by changing in colour (Table 7.1) and can be detected using by different photography (Table 7.2).

7.9 Concept of Uniqueness (Sognnaes et al. 1982; Van der Velden et al. 2006)

Bite mark analysis is based on the concept that the biting surfaces of the anterior dentition, typically the six or eight most anterior maxillary and mandibular teeth, are sufficiently distinctive that welltrained forensic odontologists can differentiate the incisal portion of one person's anterior teeth from another's (Fig. 7.3). But there is no study of large populations to solidify this argument. There is no conclusive demonstration of the distinctive nature of a single bite pattern. Most forensic odontologists suppose that bite patterns are characteristic and original, but this is not scientifically documented. Many bite marks are incomplete registrations of the involved dentition. The theoretical studies promoting the concept of uniqueness generally include multiple teeth in both arches, which may have little relevance to actual cases in which only small portions of the dentition or only parts of the incisal edges are recorded.

7.10 Stepwise Process of Bite Mark Investigation (Bowers 2004; Rai et al. 2007)

The flow of a bite mark case involves the following steps:

- 1. Recognition, initial assessment
- 2. Documentation such as photographs and dental charting



Fig. 7.3 Uniqueness of dentition

- 3. Evidence collection and preservation (DNA and physical evidence), including swabs of skin, preservations of skin
- 4. Measurements, drawings
- 5. Physical dental profiling of the questioned evidence (bite mark)
- 6. Physical dental profiling of the known evidence (suspect)
- 7. Physical comparison of (5) and (6), which produces

A common connection or

No connection or

An inability to determine because of poor quality of the evidence

- 8. DNA profiling bite mark salivary swabbing evidence and suspect's DNA
- 9. Statement of results to authorities and legal counsel

7.11 Responsibility of Forensic Odontologist in Bite Mark Cases

The forensic odontologist has three main roles in evidence collection: (1) to physically document and collect evidence related to the patterned injury, (2) to perform an analysis of the injury and, if appropriate, determine from class and individual characteristics a dental profile, and (3) to obtain as much scene and other information from the investigative authorities as possible (Dorion 2005).

7.12 Collection of Evidence from Victim

The collection of evidence associated with the bite mark is very imperative and fundamental to investigating the injury. Healing of the bite mark occurs in a living victim and degradation occurs in a deceased victim; hence, it is important to analyze and document the injury pattern over time, that is, follow up (Wright and Dailey 2001).

7.12.1 Saliva Swabs of the Bite Sites

Saliva will have been deposited on the skin during biting or sucking, and it should be collected and analyzed. About 0.3–0.05 ml of saliva is deposited while making a bite; saliva is one of the various body fluids from which DNA can be extracted. This can be used as a source of salivary DNA for investigational purposes in criminal and legal cases. Salivary DNA activity is accelerated due to the ambient temperature of the skin of a living victim, so there is degradation of the salivary DNA with the advance of time. Exfoliated cells of the oral mucous membrane can also be used as a very



Fig. 7.4 Saliva swab of the bite site

good source of DNA. Different methods and techniques have been described to collect the saliva from the bite site, but the most commonly used is Sweet's double-swab technique, which uses two cotton swabs to collect the saliva (Sweet and Pretty 2001). First, a cotton swab moistened with distilled water is employed to wash the surface that was contacted by the tongue and lips using light pressure and circular motions (Fig. 7.4). Second, a swab that is dry is used to collect the remaining moisture that is left on the skin by the first swab. Both swabs are thoroughly air-dried at room temperature for at least 45 min before they are released to police authorities for testing. Both swabs should be kept dry and cool in order to prevent degradation of the salivary DNA and to prevent the contamination of the saliva by the growth of other micro-organisms. If there is a lot of time has elapsed from the collection of the salivary swabs until they are sent for analysis, then the cotton swabs should be stored properly in a box that allows air to circulate between the swab tips.

7.12.2 Photographic Documentation of the Bite Injury

Taking photos is the universal method used to document and preserve the bite mark evidence. Hence, one should take care that the photographs are of the highest quality. Currently, there are numerous manufacturers of quality digital and conventional 35-mm cameras available for forensic investigations. A main criterion in the selection of a camera is its ability to provide high-quality, 1:1 close-up macro images. It is suggested the photography be in black and white as well as color. Other important considerations are field of view, appropriate shutter speeds, f-stop settings for depth of field, and control of light intensity, source, and angulation. Field of view should show the area of interest centered and millimeters scale, preferably an ABFO No. 2 (Fig. 7.5) (American Board of Forensic Odontology 1997), close to but not obscuring any portion of the injury. The computer can change the same image into a black-and-white image, hence eliminating the step of taking a separate black-and-white photograph. The photographs have to be taken both before and after the DNA procedure. Photographs taken before the DNA protocol record the unique condition of the bite injury, and the ones taken after the DNA protocol record the condition of the bite injury after the bite injury has been cleansed of all blood and other stains if present. The color changes of bite marks that take place improve the detail of the bite mark. Hence, photographic documentation of the injury should also be after an interval of a few days. Whenever the photographic documentation is carried out, a reference scale such as a ruler should be placed in the same plane as the bite injury. This reference scale should be visible in the photographs, as it can be used for future measurements of the bite marks. The ABFO No. 2 scale is the most commonly used reference scale. A bite injury in the skin may also have depth or three-dimensional features, which can be recorded by the use of side lighting and by low-level positioning of the flash. The human body has a few flat surfaces, and the muscle activity of the human body makes the skin surfaces curved or uneven. Photographs of such curved surfaces create shapes that are distorted from real life. However, this incorrectness can be overcome by using incremental positioning of the camera over the curved surfaces. In many conditions, due to the curvature of an injury area, it is essential to divide the pattern into two portions and photograph each as a separate image, thus avoiding a common source

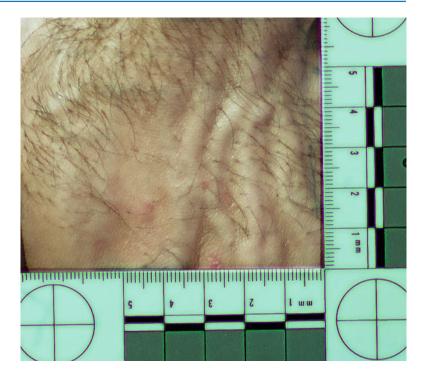


Fig. 7.5 Photograph of bite mark (ABFO No. 2 scale)

of distortion. The camera should be oriented 90° to the surface. Attention to this matter will usually provide an accurate view without angulation-related distortion. It has been suggested that the use of four samples gives light source angulations. This issue is especially significant with situations where there is a depth or third dimension to the injury area. The light source and angulation will determine the existence or position of shadows and highlights. It is recommended that the photographer take several pictures using the same camera orientation but varying the light source position, as illustrated (Fig. 7.5). If the light-generating equipment has intensity controls, that feature may also be used to enhance the image quality. Light control angulations render a specific variety of highlights and shadows to a bite mark, especially if it has a depth dimension.

There are four types of photographic distortions.

Type I distortion: when the scale and the bite mark are on the same plane, but the camera is not parallel to them.

Type II distortion: when the scale is not on the same plane as the bite mark.

Type III distortion: when one leg of the twodimensional scale has a perspective distortion, but the other leg does not.

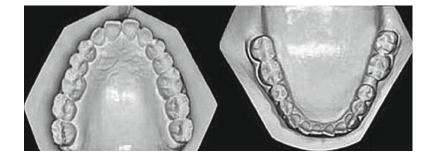
Type IV distortion: when scale is itself bent or twisted.

Different light photography can also be used, including alternative light (ALI), ultraviolet photography, and infrared photography (see additional details in Chap. 14).

7.12.3 Impressions

Construct an accurate impression of the bitten surface to record any irregularities produced by the teeth, such as cuts, abrasions, etc. (American Board of Forensic Odontology 1995; Sweet and Pretty 2001; Pretty 2008). Impressions should be taken only after taking the photographs and swabs of the bite mark site. With the help of these impressions, dental stone models can be fabricated, which are then compared to the suspect. Various dental materials have been used for taking the impressions of the bite marks, but the most commonly used is polyvinyl siloxane because it can be poured

Fig. 7.6 Dental casts



numerous times if there is an error. Also, alginate can be used for taking the impressions although the main disadvantage in using alginate as an impression material is that it has to be poured within 1-2 h to prevent contraction. Prepare the site for impression taking by removing all the hair; the area should be washed and dried. The impression material is then applied to the area and allowed to set. Care must be taken to reinforce the impression material by a durable and rigid material, such as acrylic, orthopedic mesh, dental stone, or silicone putty, before the impression material is removed from the skin. This strengthening will prevent inaccuracies from being developed in the impressions due to physical distortion. These impressions are then used to create dental stone models (Fig. 7.6). Two stone models should be prepared because one is used for analysis purposes and the second cast is preserved for presentation in court. This whole process of taking an impression and fabricating a dental stone model should be appropriately photographed.

7.12.4 Removal of the Bite Mark

Tissue incision is done in the case of diseased individuals. In this method the skin and the underlying fatty tissue are removed from the diseased victim. The bite mark and adjacent tissue are attached to a plastic ring to maintain the orientation of the injured tissue and excised. Various fixatives can be used for the proper preservation of this tissue, but the most commonly used fixative is 10% formalin. The excised tissue is stored in a plastic bag. The excised skin is then visualized by a transillumination method, in which a bright light is placed behind the excised skin (American Board of Forensic Odontology 1995; Sweet and Pretty 2001; Pretty 2008).

7.12.5 First Aid

Timely medical attention should be provided for the living victim since human bites have a higher potential for infection than animal bites (Pretty et al. 1999).

7.13 Evidence Collection from the Suspect

The collection of dental exhibits for forensic odontology uses has been supposed to be an invasive procedure. In the authors' experience, suspects are usually uncooperative during the collection of physical exhibits. The evidence required from a suspected biter may include dental history and examination, photographs, dental impression, bite records, and biological evidence, including saliva and blood samples from bite sites (American Board of Forensic Odontology 1995).

7.13.1 History and Dental Records

Dental records play a very imperative role in establishing the identity of the suspect (American Board of Forensic Odontology 1995; Sweet and Pretty 2001; Pretty 2008). These dental records can be obtained from the suspect's dentist. These dental records are tremendously useful in those cases where the suspect has deliberately had the front teeth altered after leaving a bite mark.

7.13.2 Photographs

Taking photographs of the suspect is imperative. Both intraoral and extraoral photographs should be taken. The extraoral photographs should include both profile and full face photographs. The intraoral photographs should include the frontal, lateral, and occlusal views of both arches. Photographs of the maximum interincisal opening with and without a scale in place should also be taken. A reference scale to facilitate measurements to be taken from the photographs should be included in the same plane of the teeth.

7.13.3 Extraoral and Intraoral Examination

It is imperative to examine the suspect's oral cavity intraorally and extraorally (American Board of Forensic Odontology 1995; Sweet and Pretty 2001). Biting is a forceful process that is influenced by a number of factors, including both soft tissue and hard tissue factors like the functioning of temporomandibular joints, activity of the muscles of mastication, and facial asymmetry. Different types of disorders affecting the temporomandibular joints may cause variation of the lower jaw while biting. The maximum opening of the oral cavity should be measured and noted, and divergences of the jaw during opening and closing should also be noted. Injuries or surgical interventions sometimes leave scars on the face, which should be noted during extraoral examination. The suspect may have a missing tooth due to trauma or due to caries, which has to be noted along with any misalignment of the teeth if present. Poor oral health may lead to caries for which the suspect might have had his teeth restored by various restorative materials, or the suspect may have a poor periodontal condition leading to mobility of his teeth. All these findings should be properly recognized. The size of the tongue may vary; the suspect may have an enlarged tongue or may have a small tongue, which may affect the functioning of the tongue. The size and function of the tongue should also be documented.

7.13.4 Impressions and Study Casts

Taking an impression and fabricating study casts of the dentition of the suspect are a very vital part of the examination of bite marks (American Board of Forensic Odontology 1995; Sweet and Pretty 2001; Pretty 2008). Care should be taken to include all the physical traits and the characteristics of the dentition. Various impression materials are available to take the impressions of the suspect's dentition, but the most commonly used are vinyl polysiloxane and polyether due to their high degree of accuracy and dimensional stability. After the impressions have been taken, they should be poured in dental die stone to fabricate two sets of study casts. Care should be taken that the soft tissue extensions of the impression in the master casts should be trimmed.

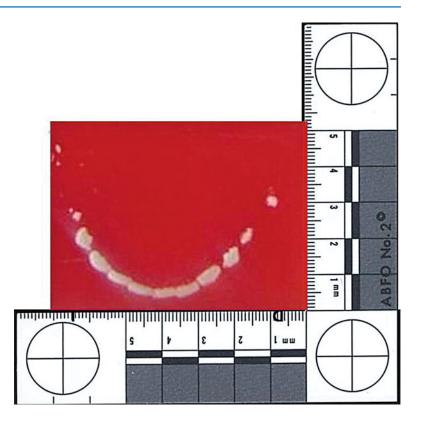
7.13.5 Test Bites

Test bites are very useful for investigating the position, shape, and alignment of the incisal edges of the biter's teeth (Fig. 7.7; American Board of Forensic Odontology 1995; Sweet and Pretty 2001; Pretty 2008). Various materials can be used for this purpose, but the most frequently used are baseplate wax, Aluwax, and Coprwax. Silicone putty can be used to take the sample bites of the suspect in centric occlusion. These sample bites have to be photographed for future comparison.

7.13.6 DNA Samples

As already described, saliva is one the sources from which DNA can be extracted. Nevertheless, we can also use the suspect's whole blood to extract his or her DNA. This is an invasive procedure, so we can use saliva as a source of DNA, which can be collected by noninvasive procedures. Buccal swabs are also a valuable source of DNA. These samples collected from the suspect can then be used for comparison with any biological evidence at the crime scene

Fig. 7.7 Test bites



that might be thought to originate from the suspect (American Board of Forensic Odontology 1995; Sweet and Pretty 2001; Pretty 2008).

7.14 Classifications of Bite Mark Injury

Bite mark injuries were classified (Pretty 2007) to create a standardized system of classification of importance for forensic odontologists, as follows:

- 1. Visibly mild; no individual tooth marks present; diffuse arches visible, may be caused by something other than teeth—low forensic significance.
- 2. Obvious bruising with individual, discrete areas associated with teeth; skin remains intact—moderate forensic significance.
- Very obvious bruising with small lacerations associated with teeth on the most severe aspects of the injury; likely to be assessed as definite bite mark—high significance.

- 4. Numerous areas of laceration, with some bruising; some areas of the wound may be incised; unlikely to be confused with any other injury mechanism—high forensic significance.
- 5. Partial avulsion of tissue; some lacerations present, indicating teeth as the probable cause of the injury—moderate forensic significance.
- 6. Complete avulsion of tissue; possibly some scalloping of the injury margins suggested that teeth may have been responsible for the injury. May not be an obvious bite injury low forensic significance.

7.15 JBR Classification

These authors (Rai and Kaur) also proposed a new classification system based on the depth of the injury. This classification is known as the JBR classification.

Mild: No bleeding, but bite marks are easily recognized—low forensic significance.

Moderate: Injury of only soft tissues (i.e., bleeding) is taking place—high significance. Severe: Involves the soft tissues of body parts and injury of hard tissues like bone—low forensic significance.

7.16 Forensic Physical Comparison of Exhibits

Once the process of fulfilling documentation requirements has been completed for both the bite mark and the dentition of the suspect, a comparison analysis is begun. Chapter 8 on bite mark analysis describes the entire comparison process. The most common methods to establish if the suspect's teeth caused the bite mark include techniques to compare the specific features of the teeth (shape, size, position of teeth, etc.) with similar traits and characteristics present in life-sized photographs of the injury using transparent overlays. These overlays have been produced using various methods (Pretty 2008). It has been reported that the most accurate technique is computer-based (Sweet et al. 1998). Other methods of comparison include the direct comparison of the suspect's study casts with photographs of the bite mark, comparison of test bites produced from the suspect's teeth with the actual bite mark, and the use of radiographic imaging and scanning electron microscopy (Rawson et al. 1979). Because biting is a dynamic process comprised of multiple-component movements by the perpetrator and the victim, every episode of contact is a unique event, and the same dentition can produce bite marks with variations in appearance. This is one of the reasons for the complexity of bite mark analysis and emphasizes the need to apply objective techniques and incorporate movement in the analysis. These challenges can be met with a 3D approach to bite mark analysis. It has been reported that a 3D approach to bite mark analysis is the best technique while using the same parameters as in other methods.

7.17 Reporting Conclusions and Opinions

The American Board of Forensic Odontology (1995) has provided a range of conclusions to describe whether or not an injury is a bite mark. These conclusions are

Exclusion: The injury is not a bite mark.

Possible bite mark: An injury showing a pattern that may or may not be caused by teeth, could be caused by other factors, but biting cannot be ruled out.

Probable bite mark: The pattern strongly suggests or supports origin from teeth but could conceivably be caused by something else.

Definite bite mark: There is no reasonable doubt that teeth created the pattern.

Different words are used in bite mark reports, such as "reasonable dental/medical certainty," "probable," "exclusion," and "inconclusive." They are listed in the order of the highest positive correlation between biter and bitee to the lowest positive correlation. In our point of view, DNA terminology can be used.

Conclusions

Conclusions from the analysis of bite mark evidence can aid the legal system in answering crucial questions about potential suspects and victims.

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Bite Mark Analysis, Part 2: Adobe Photoshop®

Contents

8.1	Introduction	101
8.2	Advantage of the Digital Analysis of Bite Marks	102
8.3	Ideal Physical Evidence	102
8.4	Stepwise Bite Mark Analysis by Taking Digital Images	102
8.5	Digital Comparison of Bite Marks	105
Refe	rences	107

8.1 Introduction

Bite mark (Fig. 8.1) evidence found on the skin of a living person or a dead body and on objects may be of great importance in criminal and legal investigations (Vale 1986). The method for comparing the bite marks is well recognized and includes measurement and analysis of the pattern, size, and shape of teeth against similar characteristics observed in an injury on skin or a mark left on an object (ABFO 1995). The most frequent analysis methods are used to produce life-sized comparison overlays from a suspect's teeth to detect similarities or differences with the bite mark. Various methods exist to produce these overlays; in most techniques, the perimeter of the biting edges of the suspect's teeth are hand-traced directly from

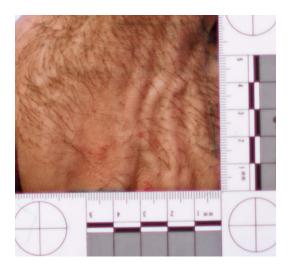


Fig. 8.1 Human bite marks

dental study casts or from wax bite exemplars, or indirectly from xerographic images produced with office photocopiers that are calibrated to produce life-sized final images. But these methods cannot avoid the bias innate in observer subjectivity, and significant errors incorporated in the overlays may make it difficult to reach conclusions with a high degree of confidence in court and medical legal proceedings. Therefore, the AFBO has proposed an objective method using Adobe Photoshop® (ABFO 1995). Since biting is a forceful procedure involving three moving systems-the maxilla, the mandible and the victim's reaction-most recent research has focused on analyzing how these factors affect the bite mark with the application of 3D procedures (Thali et al. 2003; Martin-de las Heras et al. 2005). The functional tools within Photoshop[®] can be used to detect and correct for certain angular distortions. This is an extremely important step, as it forms the foundation for the comparison procedures that follow (Johansen and Bowers 2003).

8.2 Advantage of the Digital Analysis of Bite Marks (Johansen and Bowers 2003)

Digital analysis of bite marks has a much faster processing speed, which saves time. Also, it is easy to distribute digital information. It is also easy to store a large number of images. Finally, it is a more objective method, with minimal bias.

8.3 Ideal Physical Evidence (Johansen and Bowers 2003)

As bite marks are photographed or dental radiographs are used as evidence, forensic odontologists should attempt to obtain a precise image of the bite marks. Thus, a tripod should be used even though off-angle distortion is really impossible to avoid. Ideally, photos of bite marks of two- and three-dimensional evidence should have the following criteria:

- 1. ABFO No. 2 scale and bite marks should be in the same plane.
- 2. The direction of the camera and the scale should be parallel.

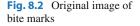
- 3. The bite mark should be photographed perpendicular to the ABFO No. 2 scale (Fig. 8.2).
- 4. Avoid creating a parallax distortion of the scale caused by placing the scale away from the plane of the bite mark.

8.4 Stepwise Bite Mark Analysis by Taking Digital Images

The following steps explain how to use Adobe Photoshop to take a digital image of a bite mark:

- 1. Import the image of the bite mark by clicking File; then click Open and open the Images file (Adobe Photoshop CS5®).
- 2. Click on File, and save images as PSP.
- 3. Prepare bite mark image by using the measure tool and rotation tool (click measurement tool and draw a line along one leg of the ABFO No. 2 scale).
- 4. A rotating angle appears.
- 5. Click Image of Adobe Photoshop.
- 6. Click Rotate canvas.
- 7. Angle appears.
- 8. Press OK.
- 9. Click the crop (crop the image according to size).
- 10. Save the image (name it).
- 11. Change the background color by clicking the Color tool.

Note: At this point, it is important to determine and correct any angular distortions based on the size and shape of the ABFO No. 2 scale present in the bite mark image. If the scale illustrates no distortion, then the image does not need to be adjusted. The sides of the scale should be parallel. The incremental lines must be perpendicular to these sides and equally spaced; if present, check for shapes of scale circles by drawing a circle (i.e., the scale circle must be round). Next, measure the angle of rotation: The forensic odontologist assessing the bite mark photograph should refer to the circular reference shapes present on the scale. An elliptical shape indicates the camera-positioning angle was incorrect. The distortion must be accurate before the bite mark photograph is resized and analyzed. There are four types of distortions:





Type I distortion: The scale and bite mark are on the same plane. Peripheral scale inaccuracies can be discounted. In this case use only the area subsequent to the bite mark for the resizing and analysis. Do not use the entire scale.

Type II distortion: When the ABFO No. 2 ruler and bite mark are not on the same plane, correcting the scale will adversely affect the proportions of the bite mark.

Type III distortion: This occurs when one leg of ruler has an angular distortion but the other leg does not.

Type IV distortion: The scale itself may be bent or skewed.

12. Determine the image's linear distortion using a grid. Click on View, Show, Grid, Preferences, Guide, Grid and slices.

Note the following:

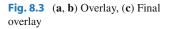
- (a) The legs of the scale should be perpendicular.
- (b) The legs of the scale should be equal in width.
- (c) The sides of the legs should be parallel.
- 13. Determine the angular distortion by selecting ruler circles or elliptical marquee. Click

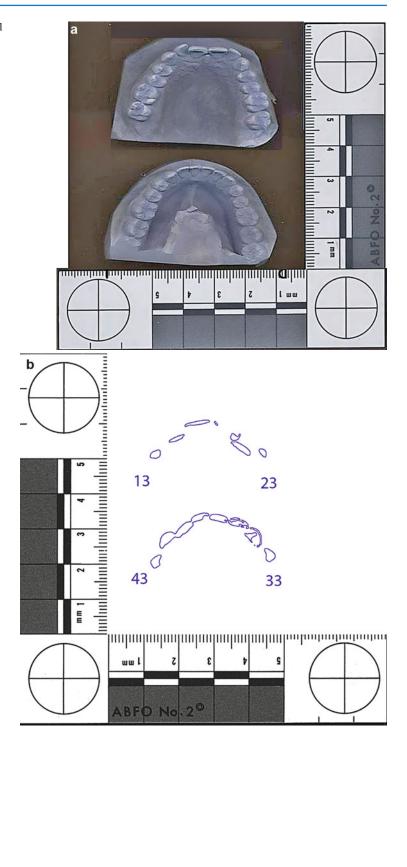
Elliptical marquee tool. Draw a circle. Click Edit, Stroke, OK. Click Free transformation. The ruler circle should match the reference circle. If they match, click OK.

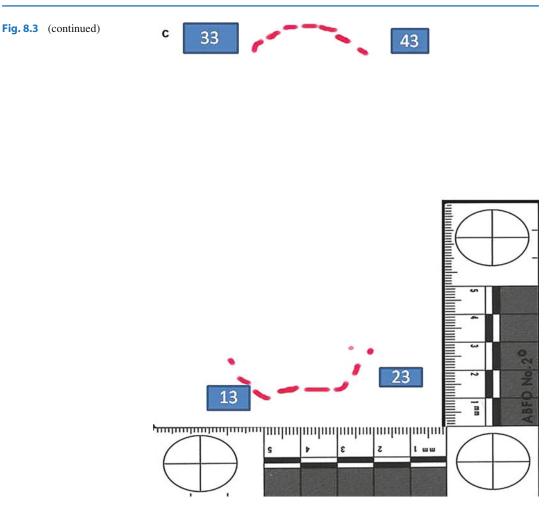
- 14. Resize the bite mark image:
 - (a) Import a standard size photograph using the ABFO No. 2 scale and compare it with the bite mark image scale.
 - (b) Activate the Snap to guide function by clicking View, Snap to guide.
 - (c) Apply the following formula:

The real size of the scale = scale size (actual) Resize ratio = scale size (actual)/scale size (image)

- (d) Click Image, image. Enter the value calculated using the above formula. Click OK, final image.
- 15. Print the image: Click Image, Mode, CMYK color (recommended).
- Add text: Click Image, Canvas size. Enter value. Click OK.
- 17. Add text: Click T tool. Enter desired text. Click OK.







18. Scan the cast:

Scan both casts with the ABFO No. 2 scale using a high-quality scanner, and save the image. Overlay the fabrication (Fig. 8.3a–c). In 1996, Dr. Heidi Christensen developed a method of scanning dental models on a flatbed scanner, then generating solidand hollow-volume overlays using Adobe Photoshop (Christensen and Alder 1996). The final process is to compare the overlay with the bite mark evidence and assess the physical correspondence between the two. The forensic odontologist uses the computer program to select the dental biting edges instead of using hand-drawn tracings of the suspect's plaster models.

 Prepare image casts. Rotate maxillary cast: Select Free transfer tool and rotate the maxillary cast. Deselect the image. Select the tooth edges by selecting Magic wand tool. Select Modify, Smooth, OK. Save as a layer (name it).

 Add ABFO No. 2 scale: Select ABFO No. 2 scale image. Click Edit, Copy, New layer, Paste, OK. Finally, the overlay has been fabricated (Fig. 8.3c).

8.5 Digital Comparison of Bite Marks

Method 1. Nonmetric analysis (Fig. 8.4): Import the overlay and bite mark image into software as described above.

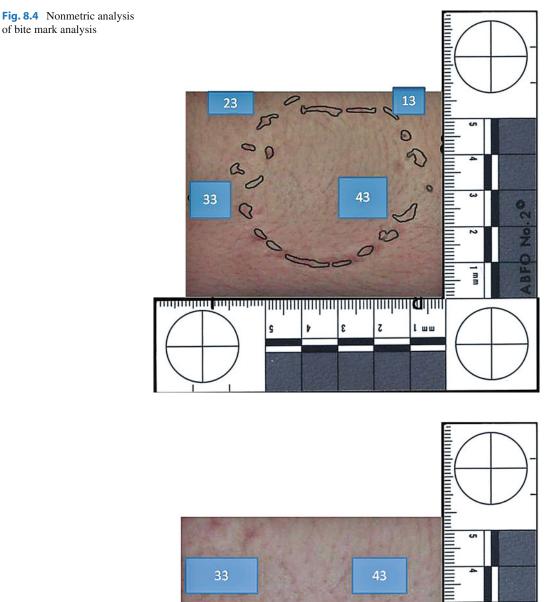
- 21. Select Window tool, Documents, Tile.
- 22. Check resolution: Click Image, Image size. Check the size of both images.
- 23. Ensure the proper orientation of the overlay: Click Image, Rotate canvas. Flip canvas horizontally or vertically (based on condition).

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Fig. 8.5 Mertic analysis of bite mark analysis

of bite mark analysis

24. Select Rectangular marquee tool (overlay image). Move tool. Move overlay on bite mark image. Select Free transform. Try to match bite mark.

Method 2. Nonmetric analysis (Fig. 8.5): Use spatial polygon and polygon lines. Click the Line tool (overlay image). Make a pyramid. Move tool. Move overlay on bite mark image. Select Free transform. Try to match bite mark.

Metric analysis of bite mark: The forensic odontologist needs to measure physical data in bite mark cases. The application of certain Photoshop tools and functions offer forensic odontologists physical evidence data that will provide linear and angular parameters that are useful in analysis, such as the shape of the dental arch, the arch's width, the distance between the cuspid to cuspid labiolingual position, the rotational position, intertooth spacing, and so forth.

Example:

Metric comparison:

1. Arch width:

In patient's mouth, it is 3.72 cm.

In the resized image of the bite, it is 3.78 cm.

2. Intercuspal width:

3.01 cm in patient's mouth (maxillary).

3.03 cm in the image of bitemark on skin (maxillary).

2.26 cm in patient's mouth (mandibular).

2.36 cm in the image of bitemark on skin (mandibular).

- 3. Shape of dental arches: round.
- 4. Mesiodistal width of teeth as follows: 11=0.45 21=0.48 41=0.30 31=0.34
- 5. Measure angles between the teeth: The angle between the two yellow lines is 160.7°.

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Cheiloscopy in Identification: Forensic Odontology

9

Contents

9.1	Introduction	109
9.2	Background	109
9.3	Lip Print Classification	110
9.4	Analyzing and Recording Lip Prints	111
9.5	Recording Lip Prints	112
9.5.1	Photography	112
9.5.2	Recording Lip Prints Using Lipstick or	
	Another Transferring Medium	112
9.5.3	Using a Finger Printer	112
9.5.4	Processing and Development of	
	Lip Prints	113
9.5.5	Developing Latent Lip Prints	113
9.6	Use in Forensic Odontology	113
9.6.1	Identification	113
9.6.2	Sex Determination	113
Refer	ences	113

9.1 Introduction

Establishing a person's identity can be a very complicated process. Dental, fingerprint, and DNA comparisons are almost certainly the most common techniques used in this pursuit. However, there are many well-known implanted methods of human identification. One of the most interesting emerging methods of human identification originating from criminal and forensic odontology is human lip recognition. Lip prints are unique and do not change during the life of a person (Tsuchihashi 1974). The external surface of the lip has numerous elevations and depressions that form a characteristic pattern, referred to as lip prints. Lip prints can be obtained at crime scenes from clothing, cups, glasses, cigarettes, windows, and doors (Williams 1991).

9.2 Background

Cheiloscopy (from the Greek words *cheilos*=lips, e skopein; see Molano et al. 2002) is the name given to lip print studies (Perper and Menges 1990). The biological phenomenon of systems of furrows on the red part of human lips was first noted by anthropologists. R. Fischer was the first to describe it in 1902 (Thomas and van Wyk 1988). The use of lip prints in personal identification and criminalization was first recommended in France by Edmond Locard (Thomas and van Wyk 1988). In 1950 Synder was the first person who suggested the idea of using lip prints for identification. He had conducted an investigation of a traffic accident and established that the characteristics of lips formed by lip grooves are as individually distinctive as the ridge characteristics of fingerprints (Snyder 1967). Until 1950, however, anthropology merely mentioned the existence of the furrows without suggesting a practical use for the phenomenon. Since 1950 the Japanese have carried out extensive research in the area. In the period between 1968 and 1971, two Japanese scientists (Suzuki and Tsuchihashi 1970) examined 1,364 persons at the Department of Forensic Odontology at Tokyo University. Based upon that research, it was established that the arrangement of lines on the red part of human lips is individual and unique for each human being. This statement led to the conclusion that it is possible to use the arrangement of furrows (on a trace, in a linear form) on lips for the identification of a person. In further research, the Japanese scientists examined the principles of the heredity of furrows on the red part of lips.

In Poland (Suzuki and Tsuchihashi 1971), the interest in lip prints started in 1966 when a lip print was revealed on window glass at the scene of a burglary. Research was carried out, and its results were comparable to those achieved in Japan and Hungary. The research was only of preliminary nature and did not allow for practical application of results as yet. A project aimed at that objective was launched in 1982, in the Forensic Institute of Warsaw University Criminal Law Department, in cooperation with the former Forensic Institute of Militia in Warsaw. The material for study was collected in the former Military Training Center at Minsk Mazowiecki. Lip prints were collected from 1,500 persons (including 107 women), coming from various locations around the country. The volunteers ranged in age from 5 to 60 years. Altogether more than 7,000 traces of the red part of the lips were examined. As a result of the examination, the individuality of lines in the red part of lips and their unchangeability within the limits practicable for identification were proven. Since 1985 in Poland, the methods of finding and recovering lip traces, recovering comparative material, and the techniques employed to carry out that expertise have been introduced into casework of the Fingerprint Department of the Central Forensic Laboratory of Police in Warsaw. During the years 1985–1997, cheiloscopic techniques were used in 85 cases,

including 65 burglary cases, 15 cases of homicide, and 5 cases of assault. In 34 cases the identification was positive, which means that cheiloscopic techniques were equal in value to other types of forensic evidence. It was also included in evidence presented in court.

9.3 Lip Print Classification

Lip prints can classified using any of the following methods:

- 1. *Martín Santos classification* (Santos 1967): This system divides the lip grooves into two groups: (1) *simple*, when they are formed only by one element; this element can be a straight line (R-1), a curve (C-2), an angular form (A-3), or sinusoidal (S-4); (2) *compound*, when they are formed by several elements; in this case, they can be bifurcated (B-5), trifurcated (T-6), or anomalous (An-7).
- 2. *Renaud classification* (Renaud 1973): The lips are considered in halves (left and right), and every groove, according to its form, has a number (Table 9.1). According to Renaud's formula, capital letters are used to describe the upper lip left (L) and right (R) sides, and small letters classify each groove. In the lower lip, it is done the other way around, using capital letters to classify the grooves, and small letters to separate the left from right sides.
- 3. *Suzuki and Tsuchihashi classification* (1971): Their system is based on six different types of grooves, as seen in Table 9.2.

Table	e 9.1	Renaud's (197	3) lip print classification

Classification	Groove type
А	Complete vertical
В	Incomplete vertical
С	Complete bifurcated
D	Incomplete bifurcated
E	Compete branched
F	Incomplete branched
G	Reticular pattern
Н	X or coma form
Ι	Horizontal
J	Others forms (ellipse, triangle)

 Table 9.2
 Suzuki and Tsuchihashi's (1971) lip print classification

Classification	Groove type
Type I	Complete vertical
Type Ia	Incomplete vertical
Type II	Branched
Type III	Intersected
Type IV	Reticular pattern
Type V	Irregular



Fig. 9.3 Type III



Fig. 9.1 Type I



Fig. 9.2 Type II

- 4. *Suzuki classification* (1970): This system is based on four types of natural lip marks/fissures:
 - Type I: vertical, comprised of complete (end-to-end) longitudinal fissures/patterns (Fig. 9.1).
 - Type II: branching Y-shaped pattern (Fig. 9.2).
 - Type III: criss-cross pattern (Fig. 9.3).
 - Type IV: reticular, typical fence-like checkered pattern (Fig. 9.4).

9.4 Analyzing and Recording Lip Prints (Caldas et al. 2007)

Lip prints can link a subject to a specific site if found on clothes or other objects, such as glasses, cups, or even cigarette butts (Ponce et al. 2003). Visible and invisible lip prints are significant (Ponce et al. 2003). The vermillion border of the lips has minor salivary and sebaceous glands, which, together with the moisturizing done by the tongue, direct to the possibility of the existence of latent lip prints (Ball 2002). The identification of latent print evidence is frequently considered the important tool in solving a crime (Ball 2002). It has been reported that latent prints can be easily seen using fluorescent dyes (Ponce et al. 2003). When dealing with lip prints from persistent lipsticks, one must always remember that persistent lipsticks have minimal oil content; therefore, their development using conventional powders might not be effective, so lysochromes can be used (Castelló et al. 2002). Latent lip prints should always be considered when processing a crime scene, even if there are no traces

of lipstick. Surveillance should be the first step when processing lip prints by using white and ultraviolet light sources (Castelló et al. 2004). Photographs should be made prior to any processing in order to protect the evidence (Caldas et al. 2007), as per FBI guidelines. Latent prints should be photographed individually with an identification label and a scale; each step in the processing series must be photographed (Caldas et al. 2007). Even the invisible lip prints can be used and can be lifted using aluminum and magnetic powder. If lip prints are located on a nonporous surface, they can be photographed and zoomed out (Ball 2002). Another way is to make overlays by using transparent overlays (Ball 2002). In some situations, lip prints can be covered with substances allowing direct observation and photography (Pueyo et al. 1994).

9.5 Recording Lip Prints

Lip prints can be recorded in different ways:

- 1. The suspect's lips can be photographed (Williams 1991).
- On a nonporous flat surface such as a mirror, they can be photographed, enlarged, and overlay tracings made of the grooves (Ball 2002).
- Lipstick, lip rouge, or another suitable transfer mediums can be applied to the suspect's lips. Then the suspect will be asked to press his or her lips to a piece of paper or cellophane tape or similar surface (Tsuchihashi 1974).
- Lip prints can be recorded using a finger printer, preferably a roller finger printer (Suzuki and Tsuchihashi 1970).
- 5. The subject can press his or her lips (without lipstick or other recording medium) against a suitable surface. These prints can be processed with either conventional fingerprint developing powder or with a magna brush and magnetic powder (Williams 1991).

9.5.1 Photography

When the lips are photographed, proper lighting should be focused on the lips at an angle that emphasizes the contrast between the white and dark areas. The resulting lip print photographs should be of fairly accurate natural size. This can be accomplished by placing a measuring device such as ABFO No. 2 scale (Williams 1991).

9.5.2 Recording Lip Prints Using Lipstick or Another Transferring Medium

Williams (1991) recommended that after lipstick is applied to the lip, multiple records or several "sets" of lip prints should be taken. Each "set" is taken by applying a large amount of the transfer medium and then having the individual press his or her lips against the recording medium, such as paper, glossy cards, piece of glass, and so on, in a series of lip prints until all of the transfer medium is exhausted. To ensure that all parts of the lips are recorded, several "sets" of prints should be taken.

9.5.3 Using a Finger Printer

Suzuki and Tsuchihashi (1970) proposed a "roller finger printer" (made by Ollister Co., USA) to collect lip prints. This was an accepted method for recording fingerprints because it could be used to take fingerprints clearly and simply without staining the fingers. This method was therefore adopted to obtain lip print records. The special paper rolled onto the roller finger printer is applied directly to the lips to record the pattern of the lip print. The lip prints obtained by these methods are traced onto cellophane paper and examined with a magnifying glass. The traces of a lip print can be obtained using a fingerprinting roller (Kasprzak 1990). The examined person covers his or her lips with a thin layer of skin care cream. After about 3 min, a strip of paper 120 mm long and 45 mm wide mounted on a specially profiled roller is lightly pressed to the lips. The impression is subsequently visualized with the use of ferromagnetic powder used in developing latent fingerprints, and then fixed on a transparent foil. The lip prints are on the flushed part, or the zone of transition, of the lips, which are extremely mobile, so smudging of the prints can occur because of excessive or uneven pressure usually noticed in subjects with a prominent upper and/or lower lip when other methods, such as using a cellophane tape or paper roller, roller printer, or dabbing of the lips against the paper, are used to collect upper and lower lip prints together. When the subject is asked to press his or her lips against the folded paper, it is possible that only the central area will come in contact with the paper. In that case, the rest or the relaxed position of the lips is not achieved, which leads to distortion of the prints.

9.5.4 Processing and Development of Lip Prints

Provided the lip print is left on a suitable medium, it can be developed using a number of different powders (Ball 2002) and photographed.

9.5.5 Developing Latent Lip Prints

Williams (1991) suggests a powdering method using magna brush and magnetic powder. These magnetic powders and magna brush are costly compared to the cost of conventional powders.

9.6 Use in Forensic Odontology

9.6.1 Identification

Research suggests (Ball 2002) the conclusive evidence that lip prints are suitable for the successful comparison, analysis, and identification of a person involved in a crime. In fact, there have been convictions of perpetrators who were positively identified via the analysis of their known lip prints to those found at the crime scene. There is a need to develop one cohesive cheiloscopy system, practicable in forensic odontology. Apart from identifying and evidential use, lip prints may also be used in detection work, being the source of tactical and criminalistic information (Ponce et al. 2003). A lip print at the scene of a crime can be a basis for conclusions as to the character of the event, the number of the people involved, sexes, cosmetics used, habits, occupational traits, and the pathological changes of lips themselves.

9.6.2 Sex Determination

The authors of this chapter reported (Rai et al. 2011) that Type I patterns were found to be dominant in females, while Type III and Type IV patterns were dominant in males (Fig. 9.1).

Conclusions

A lip print may be depicted as a surface with visible elements of lines representing the furrows. This unique characteristic pattern helps to identify the individuals. Individual identification of human being based on this trace is extremely difficult in case of distortion of lines.

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Palatal Rugoscopy: An Important Tool in Forensic Odontology

10

Contents

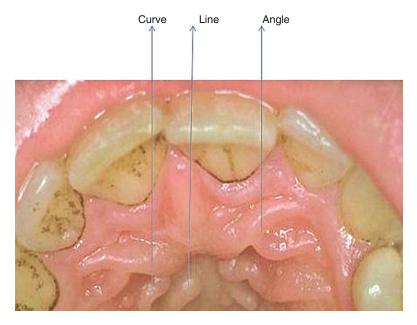
10.1	Introduction	115
10.2	Classification of Palatal Rugae	115
10.3 10.3.1	Analyzing and Recording Palatal Rugae New Method of Palatal Ruga Analysis	
10.4	Shortcomings of Rugoscopy	117
10.5 10.5.1 10.5.2 10.5.3	Use in Forensic Odontology Identification Ethnic Groups Sex Determination	118 118
10.5.4	Difference Between Edentulous and Dentate	118

10.1 Introduction

Establishing a person's identity can be a very difficult process in forensic identification. Dental, fingerprint, and DNA comparisons are the most common techniques used in this context, allowing fast and secure identification. However, since they cannot always be used, sometimes simple techniques can be used successfully in human identification, such as palatal rugoscopy, which is the study of the palatal rugae (Caldas et al. 2007). Palatal rugoscopy was first proposed in 1932 by the Spanish investigator Trobo Hermosa. Palatoscopy, or palatal rugoscopy, is the name given to the study of the palatal rugae in order to establish a person's identity. Palatal rugae have been compared with fingerprints and are unique to an individual (English et al. 1988). They can be of particular significance in edentulous cases and also in certain conditions where there are no fingers to be studied, such as burned bodies or bodies that underwent severe decomposition (Caldas et al. 2007). The uniqueness, postmortem resistance, overall stability, and, in addition, low utilization cost make palatal rugae an ideal forensic identification parameter (Kapali et al. 1997).

10.2 Classification of Palatal Rugae

The first system of classification was developed by C. Goria in 1911 and was rudimentary. The rugae pattern was categorized in two ways: specifying the number of rugae and specifying the extent of **Fig. 10.1** Different types of rugae



the rugal zone relative to the teeth. In this system, compound rugae of two or more branches were counted as one, whether they were V- or Y-shaped. Goria further distinguished two types: simple or primitive and more developed. L. Lysell's (English et al. 1988) classification in 1955 is the most important, and it has been used widely in research involving rugae. It is comprehensive and includes the inter rugae (IR). Rugae are measured in a straight line between the origin and termination and are grouped into three categories:

- 1. Primary: ≥5 mm
- 2. Secondary: 3-5 mm
- 3. Fragmentary: 2-3 mm

Rugae smaller than 2 mm are disregarded. This is a rather simplified picture of the intricate form that rugae usually present. Therefore, Thomas and Kotze (1983) further detailed the various patterns of primary rugae. These include branched, unified, cross-linked, annular, and papillary, among others. Their classification is based on the length of rugae:

- 1. Primary rugae: >5 mm.
- 2. Secondary rugae: 3–5 mm.
- 3. The shapes of individual rugae were classified into four major types: curved, wavy, straight, and circular (Fig. 10.1).
- 4. Unification occurs when two rugae are joined at their origin or termination. Rugae are considered *diverging* if two rugae had the same

origin but immediately branched, whereas rugae with different origins that joined on their lateral portions are considered *converging*.

The Cormoy system of classification (Pueyo et al. 1994) is based on the length of the palatal rugae:

- 1. Principal rugae: >5 mm
- 2. Accessory rugae: 3–4 mm
- 3. Fragmental rugae: <3 mm

An additional classification is that of Martins dos Santos (Perrella et al. 2000), which is based on the form and position of each palatal ruga. This classification indicates and characterizes the following:

- 1. One initial ruga: The most anterior one on the right side is represented by a capital letter.
- 2. Numerous complementary rugae: The other right rugae are symbolized with numbers.
- One subinitial ruga: The most forward one on the left side is symbolized with a capital letter.
- Numerous subcomplementary rugae: The other left rugae are represented by numbers. Finally, the Basauri classification (Pueyo et al.

1994) is a very easy classification to use. It discriminates between the principal ruga, which is the more anterior one (labeled with letters), and the accessory rugae, which concern all the remaining rugae (labeled with numbers), as seen in Table 10.1. The rugogram is begins from the right side of the palate.

10.3 Analyzing and Recording Palatal Rugae (Pueyo et al. 1994)

There are numerous ways to analyze palatal rugae (Fig. 10.2). Intraoral examination is probably the most commonly used way and also the easiest and the cheapest. Nevertheless, it can create difficulties if a future comparative exam is required. A detailed and exact study, as well as the need to preserve evidence, may validate oral photography or oral impressions. The overlay print of palatal rugae in a maxillary cast can be used in order to perform comparative analysis. By using stereoscopy, for example, one can obtain a three-dimensional image of the palatal ruga anatomy. It is based on the analysis of two pictures taken with the same camera, from two different points, using special equipment. Another technique is stereophotogrammetry, which, by using a special device called the Traster Marker, permits a precise determination of the length and position of every single palatal ruga. However, due to its sim-

Table 10.1	Basauri	palatal rugae	classification
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Principal rugae classification	Accessory rugae classification	Rugae anatomy
А	1	Point
В	2	Line
С	3	Angle
D	4	Sinuous
Е	5	Curve
F	6	Circle
Х	7	Polymorphic

Source: Pueyo et al. (1994)

plicity, price, and reliability, the study of maxillary dental casts is the most common technique.

10.3.1 New Method of Palatal Ruga Analysis

A special software was designed called Palatal Rugae Comparison Software (PRCS Version 2.0). The initiation and termination points of rugae were marked on all images of clinical photographs using MS Paint version 5.1 software. A strict protocol was undertaken for the order in which points were plotted: First, the tip and base of incisive papilla, then each ruga was plotted at the medial and lateral ends, working from anterior to posterior. Left-sided rugae were plotted before right. These plotted points were processed by the software and the information sequentially stored corresponding to pixel position. All the photographs were stored in the software. Later same photographs were loaded into the software one by one. After marking the points in the second set of photos, the match command is given in the software. The software will search for its match from previously loaded photographs.

10.4 Shortcomings of Rugoscopy

Postmortem identification is not possible without antemortem records. Complex rugae patterns can lead to intra- or interobserver errors. It has been observed that denture wear, tooth

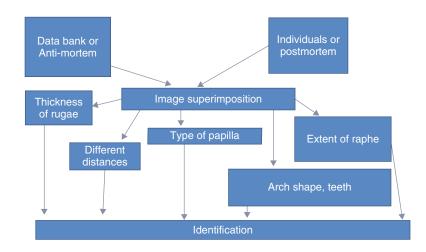


Fig. 10.2 Rugae identification process

10 Palatal Rugoscopy: An Important Tool in Forensic Odontology

malposition, and palatal pathology can cause alterations in rugae patterns (Kapali et al. 1997). Rugae patterns are genetically determined, and so they can be used in population differentiation as well as in individual identification (Caldas et al. 2007). In circumstances involving fire, palatal rugae are regularly destroyed. Also, because decomposition and skeletonization can occur in less than 6 weeks in summer and 4 months in winter, rugoscopy does not have application after this specific period (Caldas et al. 2007).

10.5 Use in Forensic Odontology

10.5.1 Identification

Thomas and van Wyk (1987) successfully identified a severely burned body by comparing the rugae to the pattern on the victim's old denture.

10.5.2 Ethnic Groups

A significant association is evident between rugae forms and ethnicity. Kapali and colleagues (Kapali et al. 1997) studied the palatal rugae patterns in Australian Aborigines and whites. They observed the number, length, shape, direction, and unification of rugae. The authors concluded that the mean number of primary rugae in Australian Aborigines was higher than that in whites, although whites had more primary rugae that exceeded 10 mm in length. The most common shapes in both ethnic groups were wavy and curved forms, while straight and circular forms were uncommon. Japanese children had more primary rugae than did Indian children, but both groups had the same number of transverse palatine rugae (Kashima 1990). Straight rugae are common in the North Indian population (Rai and Anand 2007).

10.5.3 Sex Determination

The incidences of straight and forwardly directed rugae were higher among females than males, while those of wavy and backwardly directed rugae were higher among males (Rai and Anand 2007).

10.5.4 Difference Between Edentulous and Dentate

The reduced number, shorter lengths, lesser complexity and perplexity, and more anterior and lateral position are more dominant features in older compared to young dentate individuals (Rai and Anand 2007).

Conclusions

Palatal rugea acts as a reference landmark in various dental treatment modalities. Palatine rugae are unique to individual, so it is acting as identification of individual in forensic odontology investigation. Thus, analysis of palatal rugae pattern combined to the other methods is an important aid technique for human identification, providing a significant contribution in cases of forensic investigation.

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Forensic Orodental Radiology

Contents

11.1	Introduction	119
11.2 11.2.1 11.2.2 11.2.3	Intraoral Radiographic Techniques Periapical Radiographs Bitewing Radiographs Occlusal Radiographs	120 120 120 121
11.3 11.3.1 11.3.2 11.3.3 11.3.4 11.3.5	Extraoral Techniques Panoramic Radiograph Lateral Oblique Jaw View Cephalometric View Temporomandibular Joint View Cone Beam Computed Tomography	121 121 121 121 121 121 122
11.4	NOMAD	122
11.5 11.5.1 11.5.2 11.5.3 11.5.4 11.5.5	Identification Process by Radiographs Age Estimation Sex Determination Racial Determination Socioeconomic Status Estimation Evaluation of Death and Cranial Injury	123 123 123 123 124 124
11.6	Comparative Methods of Identification	124
11.7 11.8	Facts of Exposure in Forensic Odontology Specimens Parameters and Density	124 124
11.9	Precautions to Take During Intraoral X-Rays	124

11.1 Introduction

Recently, forensic radiology has been based almost exclusively on X-rays and images captured on the radiograph. The advanced modalities, such as computed tomography (CT), cone beam computed tomography (CBCT), and magnetic resonance imaging (MRI), are being added to the forensic toolkit. The X-ray fingerprint method known as X-ray fluorescence radiography (XFR) has been revived in the recovery of latent prints from surfaces. The XFR method is employed when prints are suspected to rest on problematical materials such as multicolored documents, cloth, polythene, wax, cardboard, hardboard, varnished and untreated wood, rubber pigskin, and the skin of human corpses or nonvital separated body parts. This method is not used commonly because it needs special tools, and the lead dust can expose forensic investigators to a health risk (American College of Radiology 2011).

The forensic odontologist identifies and characterizes the teeth of unidentified bodies and essentially works expansively with radiographs. The dental charts of missing individuals can then be compared with the forensic odontologist's report to identify the body. If the body is decomposed, skeletonized, fragmented, burned, or mutilated for any other reason, it is extremely common that the dentition will be intact and will provide a valuable tool for the identification process (Kvaal et al. 1995; Lichtenstein et al. 1988). This is predominantly true for victims of fires and mass disasters (Ligthelm 1983). Thus, despite the abundance of possible techniques, those used in forensic dentistry are extremely valuable for this purpose (Sainio et al. 1990). Thus, dental records and radiographs are the most useful tool for identifying such mutilated remains (Fischman 1985). This chapter will highlight the scope of oral-dental radiology in forensic odontology.

11.2 Intraoral Radiographic Techniques

Two of the basic principles of radiography are that (1) the central beam must pass through the area to be examined, and (2) the X-ray film must be placed in position so as to record the image with the least amount of image distortion (Dunn and Kantor 1993).

11.2.1 Periapical Radiographs

The principle of the intraoral periapical (Fig. 11.1) examination is to acquire a view of



Fig. 11.1 Periapical radiograph

the tooth and its surrounding structures. Two exposure techniques may be employed for periapical radiography: the paralleling technique and the bisecting angle technique. The paralleling technique is the most common method. This technique gives less image distortion and reduces excess radiation exposure to the patient. The bisecting technique can be applied for subjects unable to accommodate the positioning of the paralleling technique. Shortcomings to the bisecting technique comprise image distortion and excess radiation due to increased angulations involving the eye and thyroid glands. The paralleling technique is so named because the tooth, receptor, and end of the position-indicating device are all maintained on parallel planes. The principle is based on the fact that image sharpness is primarily affected by the focal-film distance, object film distance, motion, and the effective size of the focal spot of the X-ray tube. It requires the use of film holding devices to maintain the relatively particular positioning needed. A variety of film holders are available: simple, complex, light, heavy, reusable, disposable, autoclavable, and non-autoclavable. A few of the more common include XCP with localizing rings, Snap-a-ray, and Stabe Bite Blocks. In the bisecting technique, the film touches the teeth at the incisal edge or the lingual-occlusal surface and then the rest of the film is angled depending on the anatomy of the area. The X-ray beam is directed perpendicular to the bisecting line between the angle of the long axis of the tooth and the angle of the film. Supporting the film pack with the patient's forefinger is not recommended.

11.2.2 Bitewing Radiographs

Bitewing X-rays (Fig. 11.2) show the size of the pulp chamber. They are a useful adjunct to evaluate periodontal conditions, offer a good view of the septal alveolar crest, and, in addition, permit changes in bone height to be accurately assessed by comparison with adjacent teeth. They do not provide information about the apices of the tooth.



Fig. 11.2 Bitewing radiograph



Fig. 11.3 Occlusal radiograph

11.2.3 Occlusal Radiographs

Occlusal radiographs (Fig. 11.3) allow a complementary radiographic examination intended to provide a more extensive view of the maxilla and mandible. They are very useful in determining the buccolingual extension and displacement of fractures of the mandible and maxilla. They also aid in localizing unerupted teeth, retained roots, foreign bodies, and calculi in the submandibular and sublingual salivary glands and ducts.

11.3 Extraoral Techniques

Various extraoral techniques have been proposed, which this section reviews (Fischman 1985).

11.3.1 Panoramic Radiograph (Hatcher 2010)

A panoramic X-ray (Fig. 11.4) shows the dentition and surrounding structures, the facial bones and condyles, and parts of the maxillary sinus and nasal complexes. The panoramic radiography equipment comprises a horizontal rotating arm that holds an X-ray source and a moving film mechanism (carrying a film) arranged at opposed extremities. The subject's head sits between the X-ray generator and the film. The X-ray source is collimated toward the film, to provide a beam shaped as a vertical blade having a width of 4–7 mm (Hatcher 2010) when arriving on the film, after crossing the subject's skull. Also, the height of that beam covers the mandibles and the maxilla regions. The arm moves; its movement may be described as a rotation around an instant center that shifts on a dedicated trajectory. It can be used in identification processes such as age, sex, and race determination.

11.3.2 Lateral Oblique Jaw View

The lateral oblique jaw view (Fig. 11.5) is a useful view of large areas of the jaw. The X-ray beam is directed at a vertical angle of -10° to -15° , with the cone head perpendicular to the film.

11.3.3 Cephalometric View

The cephalometric view (Fig. 11.6) is used to measure and assess the patient's profile, including soft tissues,

11.3.4 Temporomandibular Joint View

A submentovertex view shows the temporomandibular joint (TMJ) and other structures from underneath the chin. Another view is known as a tomograph, which is an X-ray for TMJ diagnosis.







Fig. 11.5 Lateral oblique jaw view



Fig. 11.6 Cephalometric view

11.3.5 Cone Beam Computed Tomography

Cone beam computed tomography (CBCT) permits the construction in "real time" of images in the axial plane as well as two-dimensional images in the coronal, sagittal, and even oblique or curved image planes known as multiplanar reformation. It provides three-dimensional information (Scarfe et al. 2006, 2010) and can be used in identification processes by scanning postmortem dental and skull fragments and comparing them to the threedimensional antemortem image of a victim's skull image also obtained from a CT scan.

11.4 NOMAD

A NOMAD portable x-ray system is a handy device for human use in dental offices without additional shielding for radiographers or forensic odontologists. It permits access to victims and specimens in inadequate spaces. The battery life is long, and it has a rechargeable battery. It was the only portable X-ray unit used by the forensic odontologists of DMORT at the Hurricane Katrina sites. It was used by Hans-Peter Kirsch of the Reserve Major Medical Corps, German Academy of Forensic Odontostomatology, to identify 15 victims of a plane crash at the Lukla airport in Nepal (NOMAD 2011).

11.5 Identification Process by Radiographs

The process of human identification can be classified into two major categories: (1) no antemortem group, in which there is no antemortem record from which documents may be obtained. The fundamental aim of the process used in this category is to construe a profile of the person from the remains, such as sex, age, race, occupation, stature, etc. (2) The comparative group is that in which there is antemortem record from which documents may be obtained. The matching set of antemortem records comprises a proof of identity such as photographs and radiographs.

The procedures for using orodental radiographs in reconstructing a profile for identification are discussed in this section.

11.5.1 Age Estimation

Estimating the age of unknown remains is an important step toward identification. Numerous methods have been reported for age estimation based on the chronology of third molar eruption, dentition development, and sutures (Willems 2001). Deciduous eruption, crown and root mineralization (i.e., tooth staging), tooth/pulp chamber area ratio, and dental erosion were other parameters for age estimation (Scheurer et al. 2011). It is a well-known fact that by analyzing the developmental stages of primary/permanent teeth observed on a panoramic radiograph and then classifying according to the table of dental mineralization, chronology can be used to estimate the age in children. Age estimation in adult individuals can be accomplished by a radiological method estimation of the reduction in pulp cavity size resulting from secondary dentin deposition, which is proportional to the age of an individual (Zaher et al. 2011). Kvaal et al. (1995) estimated the age by determining the radiological reduction in the size of the dental pulp cavity, caused by deposition of secondary dentin (by measuring the ratios between the lengths pulp/ root, pulp/tooth, tooth/root width, and pulp/root at three different levels). The maxillary sinus as

seen in a posterior-anterior roentgenogram reaches its maximum size during the third decade of life and does not increase thereafter, becoming triangular in form (Hanihara 1959).

11.5.2 Sex Determination

It has been reported that a posterior-anterior view to measure the total craniofacial height, mastoid height, bicondylar width, and mandibular width can be used for sex determination (Nortjie and Harris 1986). Sex determination was reported in 88.6% of 35 Japanese skulls by measuring nine parameters, including maximum length, breadth of the skull, height of the skull, facial breadth, upper facial height, mandibular breadth, symphysial height, condylar height, and ramal breadth (Hanihara 1959), while in another study, four diameters—total craniofacial height, mastoid height, bicondylar width, and mandibular width—were measured to determine gender (Sedwick 1934).

11.5.3 Racial Determination

Determination of race by radiographs is quite a difficult task, and very few classical features have been found to be important for determination of race. The pulp cavity in the Mongoloid race is exceptionally wide and deep. Mongolic populations have an extra root in mandibular molars (Lasker and Lee 1957). Shovel-shaped maxillary incisors are visualized radiographically and can be seen in 90% of Asians and Native Americans. Caucasians have a cusp of Carabelli, which is an accessory cusp found on the mesiolingual cusp of the maxillary first molar (Brogdon 1998). Cephalofacial radiographic parameters such as skull length, skull breadth, skull height, sagittal contour, face breadth, face height, orbital opening, nasal opening, lower nasal margin, facial profile, and palate shape can be used in the determination of race (Krogman 1955). Racial variation in the size and configuration of the sella turcica and skull outline between Caucasian and Black children was determined by means of lateral radiographs (Royster and Moriarty 1930). Racial differences in the maxillary sinuses in Caucasians and Indians were determined by measuring their vertical height; the average height was lower in the Indians (Sedwick 1934).

11.5.4 Socioeconomic Status Estimation

Dental caries and periodontitis without any dental intervention suggest lower socioeconomic status, while crowns, bridges, dental implants, or root canal fillings are indicative of well-cared-for dentitions (Brogdon 1998).

11.5.5 Evaluation of Death and Cranial Injury

Postmortem radiography of the skull is complex to complete because of anatomical superimposition. Modified Parma radiographs can give an excellent view of calvaria fractures in which the collimator of the dental unit is detached and the depart port of the dental X-ray unit is positioned on the contralateral side of the fractures (Wong et al. 1997).

11.6 Comparative Methods of Identification

When attempting an identification, the investigator's work is centered around finding an antemortem record of the supposed victim and matching postmortem records from the deceased and the remains. It is further divided into two types.

Type I includes cases where antemortem records exist although they were not used for purposes of identification. Radiography has become a regular process in dental, medical, and health centers and hospitals. It is generally available from dentists, physicians, or hospitals for 10 years. The deceased's remains will have been X-rayed under similar conditions as in the antemortem views. The comparison of the antemortem and postmortem radiographs leads to an absolute positive or negative identification in Type I cases. Type II

includes cases in which an antemortem record was mainly taken, coded, and filed for the particular purpose of identification. In comparative identification, antemortem records are compared to those of the postmortem record. Recovered antemortem radiographs can be any types of radiographs; they are compared with postmortem radiographs by looking at different parameters such as the number and position of teeth, shape of teeth (crown and roots), different anatomic landmarks, any pathology, sinuses, caries or periodontal pathology, and dental restoration, among others.

11.7 Facts of Exposure in Forensic Odontology Specimens

It has been reported that the exposure energy must be reduced when working with forensic remains due to a lack of normal tissues. There are two ways to do this: by reducing exposure time or mA parameters by one third in most cases and by reducing them in half in cases of burned and skeletonized remains (American Board of Forensic Odontology 2011).

11.8 Parameters and Density

The density of the X-ray image is controlled by four factors: kilovoltage, exposure time, milliamperage, and target-film distance. As the kilovoltage is increased, there will be an increase in density. Safety protection is very important. Forensic odontologists or radiographers must be able to stand at least 2–3 m away from the source of the X-ray scatter and out of the primary beam. If the radiographers cannot stand 2–3 m away, a protective barrier or apron must then be provided (White 1992).

11.9 Precautions to Take During Intraoral X-Rays

Taking an intraoral X-ray in the dentition of deceased persons, whose soft tissues lose elasticity and undergo rigor mortis, the insertion of the film as well as their retention in the correct position between the tongue and the lingual surface of teeth frequently presents trouble (Oliveira et al. 1999). The body becomes extremely fragile in deceased individuals. Therefore, the use of force when inserting the film can destroy the teeth, with a subsequent loss of fundamental information. The dental X-rays should preferably be taken after the autopsy, conducted by a pathologist, who should remove the larynx through a cut in the pharynx, thus leaving a free entry into the oral cavity for the X-ray film.

Conclusions

Different views and types of orodental radiographs make them important tools in forensic odontology. Dentists, oral physicians, and other dental practitioners have a responsibility to understand the forensic odontology suggestions associated with the practice.

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Denture Marking: Forensic Odontology Aspects

12

Contents

12.1	Introduction	127
12.2	Inclusion Methods	128
12.2.1	ID-Band	128
12.2.2	T-Bar	128
12.2.3	Laser Etching	128
12.2.4	Electron Microchips	128
12.2.5	RFID Tags	128
12.2.6	Denture Barcoding	129
12.3	Surface Methods	129
References		129

12.1 Introduction

Marking of all dentures is recommended by most international dental associations. In some countries and certain states in the United States, the labeling of dentures is regulated by legislation (Borrman et al. 1999). As a component of the responsibility of the dental profession, a dentist needs to maintain careful dental records of his or her patients. This would include documenting the identity of dentures in forensic odontology. It has been reported that marking dentures can be significant in identifying patients with dementia or those who perished in a disaster (Alexander et al. 1998). Also, it becomes easier to identify the person if dentures were uniquely coded or marked. Positive identification of the denture is usually done with a minute, unique identification code that is affixed in the denture base. Dental prostheses labeled with at least the patient's name and unique identifier markers such as gender, phone number, address, occupation, and national identity number may play an important role in forensic casework (Stenberg and Borrman 1998). The importance of placing identification labeling on dentures has long been recognized by dentists even though no standard method has been developed (Stavrianos et al. 2007). The standard necessities for denture labelings are that they should be biocompatible, inexpensive, easy and quick to apply, possible to recover after an accident, and resistant to acid, extreme temperatures, cleansing, and disinfectants (Borrman et al. 1999). The labeling should be aesthetically acceptable, visible,

and durable without compromising the strength of the prosthesis agents. The recommended areas for labeling are therefore the posterior regions of the lingual flange and the palate (Borrman et al. 1999). Various methods of denture labeling have been reported, but there are two main methods in labeling dentures: inclusion systems and surface methods (Stavrianos et al. 2007). Denture labeling systems can generally be divided into inclusion systems or marking systems. Most of them are time-consuming, are aesthetically unpleasant, or use equipment not readily available in most dental laboratories, particularly in developing countries, and if the denture needs relining, the denture label becomes invisible (Richmond and Pretty 2006).

12.2 Inclusion Methods

Inclusion methods are more enduring as opposed to the relatively simple surface marking methods, but these methods require a set of skills and are time-consuming. The marks are made by using metallic or nonmetallic materials, microchips, and micro labels that are enclosed in the denture at the packing stage. Occasionally, a dislocation, crumpling, or slash can happen, which is a disadvantage as an identification method in forensic odontology (Stavrianos et al. 2007).

12.2.1 ID-Band

Dentures may be labeled with a stainless steel metal band. The most commonly used fireresistant materials are titanium foil and Ho Matrix Band containing an identifiable coding system representing patient details (Stavrianos et al. 2007). Stainless steel has a good biocompatibility. It has been reported that the Swedish ID-Band has now become the international standard among ID bands. It is resistant to heat, cost-effective, legible, radiopaque, cosmetically attractive and requires no special skills to use (Stenberg and Borrman 1998).

12.2.2 T-Bar

A T-bar is a T-shaped clear poly(methyl methacrylate) (PMMA) resin bar constructed by cutting baseplate wax, which is then flasked, packed, processed, and completed with clear PMMA. A printed identification label is affixed beside the flat section of the bar. It is then surface polished to create a clear gap presenting the ID label (Ryan et al. 1993).

12.2.3 Laser Etching

A copper vapor laser (CVL) can label the cobaltchromium components of dentures easily and reduce the font size of the data. A personal computer controls the movement of the scanner and the firing of the CVL. Nevertheless, this method is not only expensive but also requires specialized equipment and skills to do the procedure (Ryan et al. 1993).

12.2.4 Electron Microchips

The patient's information is imprinted onto a chip measuring $5 \times 5 \times 0.6$ mm. It is resistant to acid and heat, is radioopaque, and bonds well with acrylic resin. However, the main disadvantage of the chip is that it can be adorned only by the manufacturer, and not by the dentist (Richmond and Pretty 2006).

12.2.5 RFID Tags

The inclusion of radio-frequency identification (RFID) tags within dentures is a cosmetic, effective labeling method permitting quick and consistent identification of the wearer (Ryan et al. 1993). It is preferred because of their small size and the large amount of denture user data that can be stored. No special training is required to set the tag in the denture. The chip is resistant to disinfectants, solutions, and heat. Unfortunately, RFIDs are not extensively used due to the high cost of manufacture and data incorporation (Raymond and Pretty 2009).

12.2.6 Denture Barcoding

A barcode applicable to dentures consists of a machine-readable code of a series of bars and spaces printed in definite ratios. A monotonous technique translates that denture barcode so that a number code is printed on paper. The paper is photographed, a negative is made, and then it is transferred onto a piece of silk. An illustration of the barcode shows on a prepared faience, by a machine that forces the paint through the silk, when heated to 860 °C for 30 min in an industrial porcelain oven (Agülolu et al. 2009). The barcode is read with a reader, incorporated onto the denture, and sealed with acrylic resin. It can also be used for crowns and other tooth parts as well.

12.3 Surface Methods

In these methods, the marks are placed on one of the denture's surfaces and can be completed by "scribing" or "engraving" the denture itself (Stavrianos et al. 2007). This engraving can cause detrimental effects such as food debris getting stuck, leading to infection.

Conclusions

We concluded that the value of labeling dentures is immense for identifying the individual during forensic investigations. Hence, a suitable structure within dental education is required to ensure that dentists and dental technologists are exposed to denture-marking methodologies. There is a need to offer patients an aesthetically suitable denture-marking system that is also inexpensive and permanent. Dental associations should find more efficient ways of promoting the practice of denture labeling within the dental profession and the community. The dentist should always notify the patient of the benefits of denture labeling and motivate the patient to request one.

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Different Types of Light in Forensic Photography

Contents

13.1	Introduction	131
13.2	Basic Concept of Light and Spectrum of Light	132
13.3	Electromagnetic Spectrum and Skin	132
13.4	Basic Concept of Photography,	
	Inflammation, and Repair	132
13.5	Different Parameters of Photography	133
13.5.1	Lens Aperture	133
13.5.2	Depth of Field	134
13.5.3	Shutter Speed	134
13.5.4	Film Speed	134
13.5.5	Focus	134
13.5.6	Lenses (Angle)	134
13.6	Equipment for Forensic Photography	134
13.7	Digital-Based Photography	135
13.8	Types of Forensic Dental Photography	136
13.8.1	Visible Light Photography	136
13.8.2	Alternate Light Photography	137
13.8.3	Ultraviolet Light Photography	137
13.8.4	Infrared Light Photography	138
Referen	1ces	139

13.1 Introduction

Physical evidence documentation through forensic photography remains one the most vital features of crime scene investigation. The subsequent analysis of photographs will often yield clues investigators can use to recreate the stages of an event, or may provide the proof necessary to gain a conviction at trial. Conventional photography records images in the visible light spectrum and typically will record on film or in a digital file that the human eye can see. Forensic odontogist photographers are often confronted with evidence where traditional photographic techniques are unsuccessful at documenting the evidence necessary to settle the facts of a legal case. For years forensic photographers have had a variety of specialized techniques available for documenting evidence under challenging situations. Infrared, ultraviolet (UV), and alternate light imaging (ALI) photographs can be used in a variety of these situations to gain results that could not be obtained by photographing in the visible light spectrum. A forensic light source is a crime scene investigator's and lab technician's tool for enhancing observation, photography, and collection of evidence, including latent fingerprints, body fluids, hair and fibers, bruises, bite marks, wound patterns, shoe and foot imprints, gunshot residues, drug traces, questioned documents, bone fragment detection, and so on (Wright and Dailey 2001; Wright and Golden 1997). Thus, it is essential that the forensic odontologist

Fig. 13.1 Spectrum of light

accountable for the evidence collection understands the capabilities of full spectrum digital photography. Different light forensic photography is very important in cases involving dental identification, human abuse, and bite marks. The purpose of this chapter is to review the application of forensic light sources in dental legal cases and other cases.

13.2 Basic Concept of Light and Spectrum of Light

The concept of color and photoluminescence emission stands for the observation of physical phenomena due to the interaction of light with material. Light consists of electromagnetic energy, including microwaves, radio waves, etc. The visible waves, which make up white light, form only a part of the electromagnetic spectrum. The capability of our eyes to see visible light is called color vision and is similar to a filter that transmits only the radiation between approximately 400–700 nm (Fig. 13.1). Radiation at 420 nm is observed as blue, at 450 nm as alternate light or yellow, at 550 nm as green, and at 650 as red. The mixture of electromagnetic rays between 400 and 700 nm is seen as white light.

13.3 Electromagnetic Spectrum and Skin

Photographic techniques utilizing these four types (visible light, ALI light, UV light, and IR light) of the light spectrum give very different looks into pattern injuries on skin. Because the human eye is unable to see beyond the visible light spectrum, these special types of photographic techniques are applied to produce images in the nonvisible zones of electromagnetic radiation such that they can be seen with the human eye. When light hits human skin, different types of processes take place, including reflection, absorption, fluorescence, and scattering of the light. Reflection occurs as the shorter wavelengths of light strike the surface of the skin. Depending on different parameters of radiation, more than 60% of short wavelengths do not penetrate the surface of the skin and are reflected back (Anderson and Parrish 1982; Rai and Kaur 2010), while the longer wavelengths of light (700-900 nm) can penetrate the skin up to 2-3 mm (Anderson and Parrish 1982; Rai and Kaur 2010). When light rays strike the skin, a molecular-level excitation within the skin occurs that increases the resting state energy of the molecules within the skin, known as fluorescence. This phenomenon remains for about 100 ns. A special technique known as alternative light imaging is needed for the human eye to see and the camera to record this type of fluorescence (Wright 1998). This technique is named for Prof. G. G. Stokes, who reported in 1853 that the remitted wavelength of a predominant color of light is of a different frequency than the illuminating source. A part of the energy of the light at a particular frequency is absorbed by the subject matter it strikes. Once that energy is absorbed in the form of electrons, it creates a molecular excitation that looks for a return to its rest position. The return of the electrons to their resting position releases that energy as fluorescence (Stokes 1853).

13.4 Basic Concept of Photography, Inflammation, and Repair

It is most important for forensic photographers to understand the physiological changes that take place during a skin injury. These changes from a normal state to an injured state to a healing state permit us to differentiate among the contusions illuminated by light sources of various wavelengths. A skin injury leads to inflammation and repair (Wright and Golden 1997, 2010). Inflammation leads to vascular changes at the site of the injury, as the body starts to transport blood in response to tissue damage and to mediate the framework for healing. The damaged tissues further lead healing through the process of repair or scarring. The tissues heal by repair via cell replication, while if damage is too large for repair, the tissue will heal by scarring. Inflammation is the strongest soon after the injury occurs, with repair becoming the prevalent event as the tissue begins to heal. The chemical composition of the injured skin is very different from the surrounding normal, healthy skin. Damaged tissues respond a different way to electromagnetic radiation than the adjacent normal, healthy skin because of differences in photoactive agents, including hemosiderin, melanin, hemoglobin, beta-carotene, etc. (Wright and Golden 1997).

When the pattern injury in the skin shows signs of injury on the epidermis, it is best to use the UV range to photographically illustrate the surface damage because it allows more absorption and has a shallower depth of penetration (Wright and Golden 1997, 2010). UV photography requires a special armamentarium for taking UV photographs. If a skin injury shows signs of an injury on the dermis, it would require using longer wavelengths of light that could go through the skin to the level of the bruising, namely, longer wavelengths of light in the IR range. It is very important to distinguish between the healthy adjacent skin and the injured skin. It would require using a special type of photography known as fluorescent photography techniques (ALI) (Wright and Golden 1997). All photography requires a totally dark room. The forensic photographer can employ any or all of the photographic techniques to take and preserve the evidence.

Note: Visible light photography creates images of the injuries as seen by the unaided eye. UV photography shows details of the damaged epidermis surface of the skin, while IR photography exhibits the tissue injury at the dermis and below. ALI photography will record the difference between the uninjured skin adjacent to the injured skin using fluorescence (Fig. 13.2). It is important to understand that all techniques will create useful images at all times (Rai and Kaur 2010).

13.5 Different Parameters of Photography (Redsicker DR 1994)

The light can be controlled three ways:

- 1. Lens aperture (f-stops), e.g., f/2 (maximum light), f/2.8, f/4, f/5.6, f/8, f/11, f/16 (minimum light)
- Shutter speed, e.g., 1/(maximum light) ... 1, 2, 4, 8, 15, 30, 60, 125, 250 (minimum light)...
- Speed of film (ISO/ASA), e.g., 64 (less light), 100, 200, 400, 800, 1,600 (more light)

Digital cameras are becoming entirely automatic. This does not make them foolproof; it just means they are more complicated to use appropriately. Be familiar with your camera—read your *manual*—have your *manual* with you refer to it.

13.5.1 Lens Aperture

The lens aperture ring limits the amount of light allowed through the lens. This limiting is selected with f-stops—numerical representations of the aperture, where the number increases as the aperture (opening) decreases:

- Notes
- (a) Large opening (small no.), small opening (large no.).
- (b) Light requires less light/more light?
- (c) Depth of Field: narrow/wide?
- (d) Place your subject at a far enough distance so that your lens indicates that the maximum depth of field extends to infinity. Therefore, your "sharpness" extends from your minimum range of depth of field (designated on your lens) to infinity; thus, you achieve maximum depth of field.

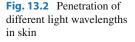
Good to expert (Symes SA, personal communication)

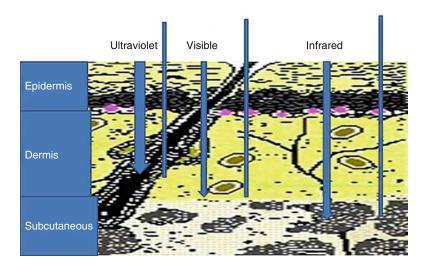
Keep camera out of the sun.

Keep camera clean.

Carry some sort of lens cleaning kit so it is handy and usable.

Keep lens cap on when not in use or at least have a skylight filter on the end of the lens for protection.





Velcro cap to camera if necessary, or put it in your pocket.

Always check lens before shooting.

Clean only with lens paper or lens brush. Do not use eyeglass cleansers.

13.5.2 Depth of Field

Depth of field is also known as "depth of sharpness" (Ralph 1975). It is controlled by two methods:

- Increase depth of field by reducing the lens aperture—stopping down—higher f-stop.
- Move farther from photographer's subject;
 e.g., move to your infinity (∞) on lens. From this point on, everything is in focus.

13.5.3 Shutter Speed

In terms of setting the shutter speed, keep in mind that a faster stop means the most movement but requires more light.

13.5.4 Film Speed

Faster film takes less light. For instance, ISO 100 takes twice as much light as ISO 200, and 200 twice as much as 400, and so on.

13.5.5 Focus

Focus involves considering a focal length. Focusing a lens is simply adjusting the distance of the lens to the film. A distance of 50 mm is considered normal for 35-mm film, while 35 mm is common for digital non-SLR. Focus is the most often abused aspect of photography.

13.5.6 Lenses (Angle)

A wide angle has the advantages of bringing more into the field close to infinity, a smaller focus length, and a greater depth of field.

13.6 Equipment for Forensic Photography (Wright and Golden 1997; Rai and Kaur 2010)

The forensic odontologist should have the following equipment when preparing to take photographs:

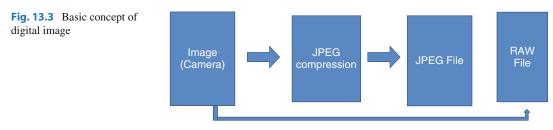
 Camera body: The 35-mm single-lens reflex camera has been in common use in forensic dentistry. Others such as TTF film plane flash metering, DX coding, auto film advance, data back, auto focusing, etc., have also been used in forensic odontology.

- Lenses: such as quartz lens and other. Focal length: 100, 105 mm. Aperture range: f/22, f/32.
- 3. Electronic flash: Different companies such as Adolf Gasser, Washington Scientific, and Trojan Camera offer a portable and powerful electronic flash.
- 4. Light sources such as ALI, UV, IR, etc.
- Film: ISO scaling of 25–160 are recommending for film used in forensic odontology photography.
- 6. Copy stand.
- 7. 50- or 55-mm macro lens.
- 8. 35–70 zoom lens.
- 9. Sturdy tripod.
- 10. Second 35-mm SLR camera.
- 11. Camera bag.
- 12. Extra batteries.
- 13. Extra film.
- 14. Green and yellow filter.
- 15. UV filter.
- 16. 18 A or 18B W filter and other filters.
- 17. ABFO No. 2 scale.
- 18. ID tags, marking pens, tapes.
- 19. Background material such as cardboard, etc.
- 20. Mirrors and cheek retractors.
- 21. Point-and-shoot 35-mm rangefinder.
- 22. Auto-focus camera with built-in flash.
- 23. Polaroid instant camera.

13.7 Digital-Based Photography

Digital image taking is different from film in that a specialized computer chip in the camera reads the light coming through the lens and electronically saves the image on magnetic media, eliminating the need for film. The image capture devices used today include the charge-coupled device (CCD) and complementary metal oxide semiconductor (CMOS). CCD sensors are arranged with geometric green, red, and blue areas known as pixels that are sensitive to their corresponding colors of light. Typically, there are two green pixels for every red or blue pixel in an arrangement known as the Bayer pattern image (Wright and Golden 2010). There are a number of suggested practices for the correct use of digital forensic photography. First and foremost, it must be understood that precise photographic techniques are a must for quality results. Most point-and-shoot digital cameras today still fall below the minimum resolution requirements, as established by SWIGIT (Scientific Working Group on Imaging Technologies). Most agencies have used this resolution as the norm because of the large number of images that can be stored at that level. A misconception about the lower resolution is that it "looks just fine" on the computer monitor. Ruler placement is of utmost importance with digital photography. To make this method easier, make sure to place ABFO rulers along the horizontal or vertical edge of the camera viewfinder area, if possible. This will allow for accurate image orientation and allow for easy cropping of the image. It is needless to print out the entire image area during analysis, so a cropped image will allow for a much faster printing time from the computer to the printer.

Currently, digital cameras with a 5-mega-pixel resolution capability or higher are working at an acceptable level. It requires a digital camera with approximately 10-mega-pixel capabilities to be equivalent to 35-mm film. Because of the current high cost of these cameras, it still isn't economically feasible for forensic odontology. The other vital parts of the digital pipeline include a compatible computer system with the appropriate amounts of memory, storage media, and output devices, such as CD-R writers and printers. It is a good idea to establish an agency-wide standard operating procedure, in writing, that sets the required guidelines. Be consistent with the production of your digital imaging files. Another very key point that must be covered is the use of JPEG files and TIFF files. The JPEG file format is probably the most widely used photographic format today. In fact, this is what allows the less expensive cameras on the market to shoot so many images on a storage card or disc. The JPEG file format is a compression format. Every time an image is saved as a JPEG, you will continue to lose information. On the other hand, the TIFF file format is a lossless type. TIFF files are generally



of a much larger size and higher resolution, so it is difficult for a forensic odontologist to shoot TIFF images at the scene. For those who still use cameras that use the JPEG file format, it has been suggested that once the images have been taken, download your images to your computer and save them in a file folder, under the current case number, as TIFF files (Fig. 13.3). This will guarantee that there will be no more image information loss during copying, moving, or saving. The computer and the output devices are just as imperative as the camera. A highly acceptable scenario for a total digital system would include a laptop computer with at least 128-256 megabytes of RAM memory, 40-gigabyte hard drive, and an on-board CD-R writer. That way, you could download your images directly to a computer hard drive and burn them to an archival CD-R disc. There are also external CD-R writers available that will plug right into the computer. Only those images needed for a court proceeding would need to be printed on conventional photo paper. It is not cost- or time-effective to try to print out large numbers of images on an ink-jet printer. Photo-quality papers are still higher in price and the printers themselves cannot keep up with the high print-output pace of a sophisticated chemistry-based photographic printer.

13.8 Types of Forensic Dental Photography

There are basically four types of forensic dental photography, as follows:

- 1. Visible light photography
- 2. ALI light photography

3. UV light photography

4. IR light photography

13.8.1 Visible Light Photography

Visible light penetrates deeper into the skin than UV light and is sufficient to document most bruises and pattern injuries. Visible light photography equipment is specifically designed to have an optimal performance in the 400 to 760nm range of the electromagnetic spectrum (Golden and Wright 2005). The equipment of choice again is a 35-mm SLR with a fast lens. Most of the shots are at the minimum focusing position on the lens and available light as opposed to flash. If the room has fluorescent lights, be sure to use a correction filter such as the Cokin A.036. Special coatings on the lens or filters in front of the digital capture device of the camera allow only visible light to pass (Golden and Wright 2005). The standard technique in visible light photography should include a series of photos from wide-ranging orientation images up to and including close-up images, taken with and without a photographic ruler. Be careful when using a white ruler or measuring tape in close-up pictures. The meter may take the light reading of the scale, and the skin tones and injury will be too dark. This can be avoided by keeping the scales at the edge of the photo since most light meters are centerweighted. The ruler is used as a reference tool because the images can be rendered to life-sized proportions at some future time during an identification process. Finally, images should be saved as TIFF files.

13.8.2 Alternate Light Photography

Alternate light photography (ALI) has been used in the detection of fingerprints, blood, semen, saliva, types of ink, false documents, bruises or other pattern injuries, and tooth color restorations (Golden 1994; Stoilovic 1991; Rai and Kaur 2010; Ray 1992; Redsicker DR 1994). It is based on the 450-nm-wavelength light (Golden and Wright 2005). Special type #15 gel or glass (yellow) band-pass filters and light sources having 450-nm (blue) monochromatic light is used (Golden 1994). ALI photography should be completed in total darkness with no other light. ALI photography is based on the principle that when monochromatic light strikes the skin, the longer wave-fluorescing energy is observable. It has longer exposure times, so it requires a tripod. According to ABFO recommendations, the ruler is placed adjacent to the wound or bruise or bite mark in the same plane as the face of the lens as the photograph is taken. It has been reported that the images are bracketed from f4-f16, with times ranging from 1/2 s to 2 or more seconds (Golden 1994). The 100 ISO film, exposure range 1/4 to 2 s at f-stop settings of f/4-f/5.6 apertures, and light source and camera distance 12-18 in. from subjects have been proposed to the ASFO (Golden and Wright 2005) (Figs. 13.4 and 13.5). Various factors can affect the photographic protocol and parameters. Special eyewear (goggles) must be worn to protect the eye.

13.8.3 Ultraviolet Light Photography

The ultraviolet spectrum consists of wavelengths shorter than those of visible light; because of this, UV photography has special applications for the forensic odontological investigation. UV penetrates only minimally into the epidermal tissue, where it is either absorbed or reflected by various biochemical compounds that are a part of the healing process in the skin (Barsley et al. 1990). Since UV light only penetrates a few microns into the surface of skin, the use of this type of photography captures the details associated with the surface of skin and further highlights the surface aberrations related to the injured skin. UV photography has long been used to identify the presence of specific compounds in the tissues by means of their unique fluorescent properties, as identified in the case of fingerprints, semen, blood, trace metals, shoe prints, subclinical bruises, tooth-colored restorations, and gunpowder residue (Vogeley et al. 2002).

13.8.3.1 Ultraviolet Light Sources

Sunlight, fluorescent tubes, mercury vapor lights, flash units, Wood's lamps, and special types of UV LED lights are the main sources of UV light (Golden and Wright 2005).

13.8.3.2 Focus Shift

Focus shift is defined as the distance between the visible focus and either the infrared or ultraviolet focus (Nieuwenhuis 1991). A few (generally old and single-coated) general-purpose lenses do transmit usable amounts of UV. However, virtually all of them suffer from a problem not frequently discussed in photography forums. These lenses are corrected in the visible range but exhibit varying amounts of *focus shift* between visible and UV. This phenomenon is comparable with the focus shift between visible and IR, which is a much more widely known phenomenon.



Fig. 13.4 Tooth-colored filling (ALI light)

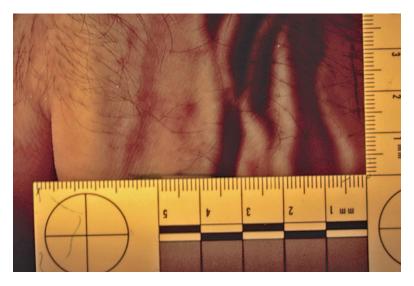


Fig. 13.5 Human bite marks (ALI light)

In practice, this means that focusing in visible light produces an out-of-focus picture in the UV or IR. Many lenses have a red dot on the focusing scale, used for manually compensating for focus shift in IR photography. There is no guarantee that focus shift in the UV is in the same direction, or by the same amount, as in the IR. The shifting focus for UV photography is in the same direction and amount done in infrared photography (Nieuwenhuis 1991). The authors of this chapter have found that focus shift is required for ultraviolet photographs when using lenses designed specifically for UV and IR. They have also found that the shifting focus for UV photography is in the opposite direction and amount as in infrared photography.

13.8.3.3 UV Photography Protocol (Herschaft et al. 2007)

Working with UV photography requires a particular type of equipment and techniques. The armamentaria for UV photography are a 35-mm SLR camera body, UV light source, UV specific band-pass filter such as 18A, digital electronic sensor sensitive to UV light. It is also recommended to secure a non-flourite-coated or quartz lens so that the UV wavelengths can pass through to the sensor. Even though quartz lenses do have a distinct advantage over conventional glass lenses, some glass-based color video lenses can capture about a 50% transmission in the near-UV band (Golden and Wright 2005). The exposure time remains standard at 1/60–1/90 of a second, and the flash output should be set to manual for maximum brightness. ISO 400 and bracketing the image exposures from f/5.6 to f/8 with exposure times from 1/60th s to 1 s parameters were proposed by researchers (Golden and Wright 2005). An ABFO No. 2 ruler should be positioned adjacent to the injury in parallel to the front surface of the lens.

13.8.4 Infrared Light Photography

Infrared light (IR), between 760 and 1,500 nm, is not only a very powerful tool for forensic science but also an underused means of detecting latent evidence. IR has been used to examine questioned documents, blood stain patterns, and the age of bite marks and pattern injuries of the skint (Santacroce 1995). Infrared light is capable of penetrating up to 3 mm below the surface of the skin and a diameter of 0.5–2 mm (Raymond and Hall 1986).

13.8.4.1 Infrared (IR) Light Sources

Flash units, tungsten lamps, quartz-halogen lamps, and special-wavelength IR LED lights are the main sources of IR light (Golden and Wright 2005). IR light requires a particular type of equipment and techniques.

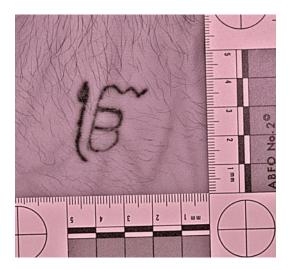


Fig. 13.6 Tattoo (IR light)

13.8.4.2 IR Photography Protocol (Herschaft et al. 2007)

The equipment needed for IR photography includes a 35-mm SLR camera body, IR light source, IR specific band-pass 87, and digital electronic sensitive to IR light. ISO 100-1600, aperture F/8–f/16, exposure time 1/125th–1 s parameters have been proposed by researchers (Golden and Wright 2005). An ABFO No. 2 ruler should be positioned adjacent to the injury, parallel to the front surface of the lens (Fig. 13.6).

Conclusions

Scientific forensic photography is critical to document and preserve images when conducting identifications, bite mark investigations, and other odontologically related forensic activities. It is important to uphold the reliability of the evidence in terms of its original form and reproducibility. Photography is one of the most vital tools used in the practice of forensic dentistry. Developing the skills necessary to competently document these injuries with different types of light is one of the great challenges in forensic dentistry.

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Advanced Technologies in Forensic Odontology

14

Contents

14.1	Introduction	141
14.2	NOMAD	141
14.3	Three-Dimensional Computer-Aided Stereophotogrammetry	142
14.4	Magnetic Resonance Imaging	142
14.5	Cone Beam Computed Tomography	142
14.6	Capillary Electrophoresis	143
14.7	Orthograde Entrance Methods	143
14.8	Energy-Dispersive X-ray Spectroscopy	144
14.9	WinID	145
14.10	Adobe Photoshop [®]	145
14.11	Mideo Systems' CASEWORKS	145
14.12	Automated Dental Identification	
	System	146
Referen	ices	146

14.1 Introduction

Mass disaster and medicolegal cases create a big problem in today's society. With the increasing rate and complexity of crime, it's becoming more difficult for the collection of feasible and suitable evidence and appropriate comparison methods. Equally, however, there has been incredible research that has led to technological advancements in forensic odontology on a global scale.

14.2 NOMAD

The NOMAD hand-held X-ray unit (Fig. 14.1) is very useful in mass disasters and medicolegal investigations. It is unfeasible to move the bodies to X-ray units to take radiographs in medicolegal cases and mass disasters. Thus a portable, handheld, lightweight, battery-operated investigative X-ray unit was developed to take the images on a big scale (Danforth et al. 2009). Called NOMAD, it is powered by a 14.4-V rechargeable battery. It is designed with a reduced weight and size for easy operator handling. The minimum intrinsic filtration in the X-ray beam is at least 1.5 mm. The source-to-skin distance is a minimum of 20-30 cm (Turner et al. 2004). The radiographic parameters of peak potential and anode current are both fixed at 60 kV and 2.3 mA, respectively. The only worker-adjustable parameter is the irradiation time, which varies from 0.01 to 0.99 s (Turner et al. 2004). It has many advantages over



Fig. 14.1 NOMAD portable X-ray device

conventional radiography, including that it reduces the radiation dose to the patient, reduces the radiation leakage from the tube, and has less backscatter radiation. The backscatter radiation is absorbed in a lead-filled acrylic shield attached at the end of the exit cone. This shield has a leadequivalent of 0.5-mm thickness and protects the operators' hands, face, and gonads from backscattered radiation.

14.3 Three-Dimensional Computer-Aided Stereophotogrammetry

In three-dimensional computer-aided stereophotogrammetry, twin image pairs that have been adopted in the field of photography to permit the production of 3D stereoscopic images by taking two or more pictures of the same object at subtly different angles. A computerbased program was developed to construct 3D images of real-time objects. It utilizes the common Microsoft Windows operating system to provide an easy-to-use graphical user interface to capture the stereo image pair. It also uses a mathematical equation to build the wireframe 3D model as a data file (Mac 2000).

14.4 Magnetic Resonance Imaging

Magnetic resonance imaging (MRI) is a noninvasive imaging technique of mapping the internal structure and certain aspects of function within the body. It uses nonionizing electromagnetic radiation and does not expose the subject to radiation. It uses radiofrequency (RF) radiation in the presence of controlled magnetic fields in order to create high-quality cross-sectional images of the body in different planes. The MR images are built by placing the subject inside a large magnet, which induces a relatively strong external magnetic field. This leads the nuclei of many atoms in the body, such as hydrogen, to support them with the magnetic field. With the subsequent application of the RF signal, energy is released from the body, detected, and used to build the MR images (Heiss 2009). It gives clear pictures of the body's interior, such as soft tissues, organs, and blood. MRI is very useful in crime scene investigation, especially in the analysis of bite marks. This analysis involves comparing the bite marks of the suspect on an apple with the bite mark on the victim's body part. The suspect is asked to bite an apple. Then impressions are taken from the apple and from the victim's bite mark, followed by making casts (Mac 2000). The collected subject material, apple or other fruit casts and dental stone casts, is sectioned into 2-cm³ samples. The images are captured and compared. Its advantages include being painless and noninvasive, having no radiation exposure, and differentiating among various soft tissues.

14.5 Cone Beam Computed Tomography

Cone beam computed tomography (CBCT) is accomplished by using a rotating framework to which an X-ray source and detector are fixed



Fig. 14.2 Cone beam computed tomography

(Hatcher 2010). A divergent cone-shaped source of ionizing radiation is directed through the middle of the area of interest onto an area X-ray detector on the opposite side of the subject. The X-ray source and detector rotate around a fixed fulcrum within the region of interest (ROI). During the exposure sequence, hundreds of planar projection images are acquired of the field of view (FOV) in an arc of at least 180°.

Based on the available or selected scan volume height, the use of units can be designated as follows (Choi and Mah 2010) (Fig. 14.2):

- Localized region (also referred to as focused, small field, or limited field): approximately 5 cm or less
- 2. Single arch: 5–7 cm
- 3. Interarch: 7–10 cm
- 4. Maxillofacial: 10-15 cm
- 5. Craniofacial: >15 cm

It has been reported that CBCT pulp-tooth volume was used to estimate the dental age from monoradicular teeth (Star et al. 2011).

14.6 Capillary Electrophoresis

Capillary electrophoresis is used for organic and elemental identification. It has the exceptional important tool of separation and resolution, rapid analysis time, simplicity, versatility, low mass limits of detection, economy of reagents, and minimal sample requirements. Also, the possibility of direct sample injection without complex sample pretreatments makes capillary electrophoresis an attractive methodology to forensic odontologists when the sample matrix may be extremely completed, as in a victim's different dental restorations, denture material, gunshot wound in the oral cavity, and a number of other endogenous compounds that have to be determined from the area of interest (Cruces-Blanco et al. 2007). The system's main components are a sample vial, source and destination vials, a capillary, electrodes, a high-voltage power supply, a detector, and a data output and handling device. The source vial, destination vial, and capillary are full with an electrolyte. To introduce the sample, the capillary inlet is positioned into a vial containing the sample and then revisited to the source vial. The analytes divide and migrate due to their electrophoretic mobility and are detected near the outlet end of the capillary. The output of the detector is sent to a data output and handling device such as an integrator or computer.

14.7 Orthograde Entrance Methods (Alakoc 2009)

The tooth is the most important source to extract DNA. It is a preserved package of DNA protecting it from adverse environmental conditions. The DNA can be collected by perforating the apex of the tooth. In the reverse root canal technique, it is more likely that sterilizing agents pull off into the pulp canal and cavity through cement layers and dentinal tubules, which may damage DNA. The fact that enamel is less permeable, dense tissue than the layered root cement makes the orthograde entrance method more favorable, rendering more severe operations applicable during decontamination.

14.8 Energy-Dispersive X-ray Spectroscopy

Energy-dispersive X-ray spectroscopy, abbreviated EDS or EDX, is an analytical technique used for the elemental analysis or chemical characterization of a sample. It is one of the variants of X-ray fluorescence spectroscopy, which relies on the investigation of a sample through interactions between electromagnetic radiation and matter, analyzing X-rays emitted by the matter in response to being hit with charged particles. Its potential descriptions are due in large part to the fundamental principle that each element has a unique atomic structure allowing X-rays that are characteristic of an element's atomic structure to be identified uniquely (Ubelaker et al. 2002). Scanning electron microscopy with energy-dispersive X-ray spectroscopy (SEM/EDS) (Fig. 14.3) is a technique that produces high-resolution images as well as an X-ray spectrum that characterizes an elemental profile of that product. These methods were used in the generation of spectral databases for dental resins with the intent of simplifying identification of these materials (Bush et al. 2007). Once an individual has been incinerated, restorations or debris of (adherence to dentin or enamel) restorations can be found (Bush et al. 2008). If unknown materials are collected from a debris field or crime scene site, then the identification of important evidence cannot be difficult because these materials must be compared with a known standard automated database generated by Fourier transform infrared spectroscopy and Raman spectroscopy based on organic composition and inorganic elemental composition using EDS spectra derived from SEM/EDS analysis (Ubelaker et al. 2002). The major difference between XRF (X-ray fluorescence) and EDS is the excitation radiation. XRF uses an X-ray beam to

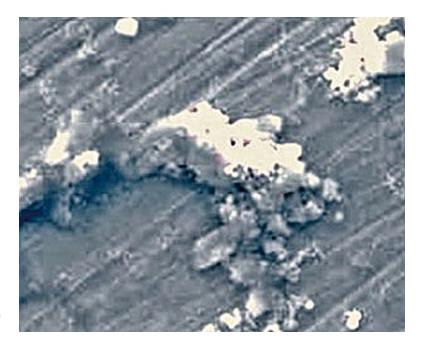
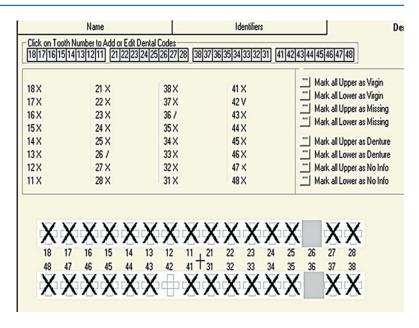


Fig. 14.3 Energy-dispersive X-ray spectroscopy of tooth





generate characteristic X-rays, while EDS uses an electron beam. The characteristic X-rays constitute the spectra. XRF has the capability to detect major, minor, and trace levels of an element, while EDS is limited to major and minor elemental concentrations (Schweitzer et al. 2005).

14.9 WinID (McGiveney 2011)

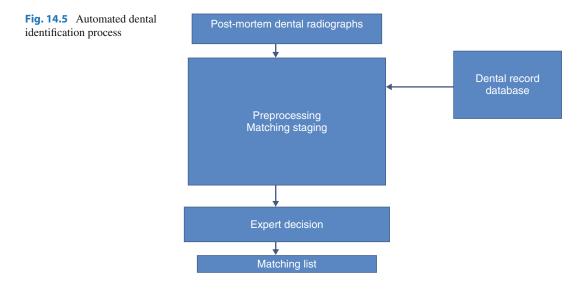
WinID is computer software that helps match missing persons to unidentified human remains by containing information about restored dental surfaces, physical descriptors, and pathological and anthropologic findings (Fig. 14.4). It makes use of dental and anthropometric characteristics to rank possible matches. WinID and Access provide extensive data filtering and data sorting capabilities. WinID is used by dentists, forensic odontologists, pathologists, forensic investigators, and personnel in medicolegal systems to identify the unknown. It is available in different languages, including English, French, German, Italian, Portuguese, and Spanish. It has proved useful in mass disaster situations and in the creation and maintenance of missing person databases.

14.10 Adobe Photoshop[®] (Bowers and Johansen 2002)

Adobe Photoshop is an important software program that permits a huge number of editing features, functions, enhancements, and metric (distances and angles) analysis. Once a photograph is scanned and imported into Photoshop, the initial working image can be enlarged using the zoom tool. A very high-resolution image (300 dpi) is required to avoid fuzziness of the magnified picture, which is Photoshop's main limitation. It is used to compare antemortem and postmortem evidence (see Chap. 8 for more details using Photoshop on in forensic investigations).

14.11 Mideo Systems' CASEWORKS

Mideo Systems' CASEWORKS is software that facilitates all areas of digital evidence in forensic cases, from the analysis and optimization of, communication about, and output of images in identification, bite mark, and other forensic cases.



14.12 Automated Dental Identification System

Biometrics is a key tool in forensic identification, including in forensic odontology. As advancing technology increased the number of medicolegal cases that forensic specialists are required to investigate, the automation of forensic identification became predictable. Dental evidence is the best marker for postmortem identification. An automated dental identification system (ADIS) may offer automated search and matching potential for radiographs and photographic images. Researchers proposed an ADIS having the following different components (Fahmy et al. 2005; Jain et al. 2003; Zhou and Abdel-Mottaleb 2005):

- The Potential Matches Search component: Mainly responsible for the archiving and recovery of dental records based on high-level dental features. These features include the number, position, and shape of teeth and other features. Recognition of a component requires implementing sufficient techniques for dental film classification, teeth segmentation, extraction of teeth contour, and feature indexing.
- The Image Comparison component: Responsible for low-level comparison of the radiographs of a case against those of suspected cases. Recognition of this component follows a pyramidal architecture for image matching.

Image comparison is carried out in the following steps:

- (a) Preprocessing of images (enhancement, segmentation, and alignment are accomplished to correct for possible geometric and intensity transformations)
- (b) Decision making (low-level features are extracted)
- The Digital Image Repository: This is the image and feature database component. Figure 14.5 outlines the ADIS flowchart.

Conclusions

Advances in technology ensure the incorporation of evidence in medicolegal cases related to forensic odontology. It's imperative to use and assess these new approaches to assist in the identification of individuals in crime scenes and medicolegal cases to ensure a correct identification.

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Forensic Odontology in the Management of Bioterrorism

15

Contents

15.1	Introduction	149
15.2	Bioterrorism Attack	149
15.3	Bioterrorism Agents	150
15.4	Classification of Bioterrorism Agents	150
15.5	Role of Forensic Odontology in the Management of Bioterrorism	151
References		152

15.1 Introduction

The threat of bioterrorism, long ignored and denied, has increased over the past few years. Bioterrorism involves the purposeful release of viruses, bacteria, or other agents used to cause illness or death in people, animals, or plants (Hamburg 1999). These agents classically are found in nature, but it is possible that they could be changed to increase their ability to cause disease, to be resistant to current medicines, or to be spread into the environment. Biological agents can be spread through the air, through water, or in food (Hamburg 1999; Miller 2001). Terrorists may use biological agents because they can be extremely difficult to detect and do not cause illness for several hours to several days. Some bioterrorism agents, such as the smallpox virus, can be spread from person to person, while others, such as anthrax, cannot.

15.2 Bioterrorism Attack

A bioterrorism attack is the deliberate release of viruses, bacteria, toxins, or other harmful agents used to cause illness or death in people, animals, or plants. These agents are typically found in nature, but it is possible that they could be mutated or altered to increase their ability to cause disease, make them resistant to current medicines, or increase their ability to be spread into the environment. Biological agents can be spread through the air, water, or food. Terrorists tend to use biological agents because they are extremely difficult to detect and do not cause illness for several hours to several days. Some bioterrorism agents, like the smallpox virus, can be spread from person to person, and some, like anthrax, cannot (CDC 2001, 2011).

15.3 Bioterrorism Agents (Miller 2001)

Bioterrorism agents comprise bacteria, viruses, fungi, and other microorganisms as well as biotoxins formed by microorganisms, plants, and animals that can kill or debilitate. Because they can reproduce, biological agents have the sole potential to make an environment more dangerous over time. If used for aggressive purposes, any disease-causing microorganism could become a weapon. For the reasons of warfare, the specific uniqueness of certain agents makes them more likely to be used than others. Some potential warfare agents can make their victims very sick without necessarily killing them. Examples include the microorganisms that cause tularemia, Q fever, and yellow fever. After suffering debilitating illness, victims of these diseases often recover, though not always. Other agents are more likely to be lethal. The bacteria that cause bubonic plague and the virus that causes smallpox can kill large numbers of untreated people. Early management regularly cures plague victims, and smallpox vaccinations before exposure to the virus can prevent the disease.

15.4 Classification of Bioterrorism Agents

The Centers for Disease Control (CDC 2001; Rotz et al. 2002) separates bioterrorism agents into three categories, depending on how easily they can be spread and the severity of illness or death they cause:

Table 15.1 Category A biological agents

Agent	Disease caused
Bacillus anthracis	Anthrax
Clostridium botulinum	Botulism
Yersinia pestis	Plague
Variola major	Smallpox
Franscisella tularensis	Tularemia
Ebola, Lassa, Machupo, and Marburg viruses	Viral hemorrhagic fever

Source: From CDC (2011) and Rotz et al. (2002)

Table 15.2	Category B	biological agents
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Agents	Disease caused
Brucella species	Brucellosis
Salmonella species	Food safety threats
Burkholderia mallei	Glanders
Chlamydia psitasi	Psittacosis
Coxiella burnetti	Q fever
Rickettsia prowazekii	Typhus fever
Vibrio cholera	Water threats

Source: From CDC (2011) and Rotz et al. (2002)

- *Category A* agents are organisms or biotoxins and are considered the highest risk to the public and national security because they can easily be spread or transmitted from person to person, they result in high death rates and have the potential for major public health impact, they might cause public panic and social disruption, and they require special action for public health preparedness. Diseases caused by such agents include anthrax, botulism, plague, smallpox, tularemia, and viral hemorrhagic fevers (Table 15.1).
- Category B agents are moderately easy to spread and result in moderate illness rates and low death rates. Example agents in this category comprise Burkholderia mallei, ricin toxin (a poison made from castor beans), and Escherichia coli (a bacterium that in some forms can cause disease, though it normally lives in the intestines of humans and other animals) (Table 15.2).

• *Category C* is made up of emerging infectious diseases such as the Nipah virus and the Hantavirus.

15.5 Role of Forensic Odontology in the Management of Bioterrorism

In order to make a protocol for dentists to follow in the management of terrorist attacks, the American Dental Association organized a meeting, known as the Workshop on the Role of Dentistry in Bioterrorism (Han et al. 2003). Many problems would need to be solved in the event of a bioterrorism attack, in particular, sample collection, handling, and preservation protocols since the material could be very harmful; microbial diversity; databases of genomes, signatures, and methods; effects of decontamination on evidence; systematic procedures for selecting and ordering analytical methods; and determining host immune responses and pharmacokinetics of prophylactic drugs (Budowle et al. 2005). The general consensus has been that dentistry can contribute valuable assets, both in personnel and in facilities, to the preparation for, and in the immediate response to, a bioterrorist attack and its aftermath (Fig. 15.1). These contributions can make a significant difference in the outcome (Guay 2002).

Forensic dentistry (Guay 2002; Han et al. 2003) can

- Correct misinformation.
- Serve as a surveillance resource, as dental offices are located across the community.
- Help controlling the terror attack.
- Calm public fears.
- Work as physicians by helping with vaccinations, first aid, etc.

According to the results of the 2003 ADA workshop, due to their education, dentists (Guay 2002) are able to help treat cranial and facial injuries. They also can

- Provide or assist in the administration of anesthetics
- Start intravenous lines
- Perform appropriate minor surgery and suturing
- · Aid in shock management
- · Help in stabilizing patients
- Collect samples
- Take medical histories
- · Provide cardiopulmonary resuscitation

In the case of a bioterrorism attack, hospital organizations may be beset and external areas for the provision of health care must be found. In these cases, dentists can contribute by applying their medical knowledge and professional experience. Furthermore, as dental or forensic odontology practitioners, there is much dentists can offer, but there is still a limit to what can and cannot be

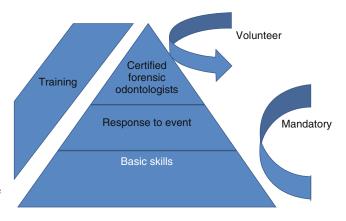


Fig. 15.1 Forensic odontologists' role in the management of bioterrorism

achieved due to training and experience and, to a lesser degree, personality and attitude. The expectation is not for the forensic odontologist to suddenly become a top-class trauma surgeon, but rather that he or she will provide a level of general medical care beyond that expected of a normal person. The challenge therefore is to combine existing skills and perform them well and to develop and extend capabilities by further training, which does not need to be at a specialist level. Advanced life support courses run by local emergency medical services help facilitate further training and professional development and would increase the forensic odontologist's level of skill to where he or she can be regarded by other allied health professionals as important and vital members of the emergency medical team.

Conclusions

Terrorism with biological weapons is likely to remain very uncommon. However, in the event of a bioterrorist attack, dentists may be relied upon to accomplish numerous functions such as education, risk communication, diagnosis, surveillance and notification, treatment, distribution of medications, decontamination, sample collection, and forensic dentistry (Guay 2002). Forensic odontology societies should develop a plan for the dental response to potential bioterrorist attacks that can be included into each community's mass disaster response plan.

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Management of Mass Disasters: Forensic Odontology

Contents

16.1	Introduction	153
16.2	Types of Mass Disasters	154
16.3	Difficulties During Mass Disasters	154
16.4	Disaster Site/Crime Scene	
	Management	154
16.4.1	Surveillance	154
16.4.2	Referral of Patients	155
16.4.3	Diagnosis and Monitoring	155
16.4.4	Triage	155
16.4.5	Immunization and Medications	155
16.4.6	Infection Control	155
16.4.7	Definitive Treatment	155
10.4.7	Demintive fredunent	155
16.5	Dental Team Organization	
16.5	Dental Team Organization	
16.5	Dental Team Organization Forensic Odontology and Identification	155
16.5 16.6	Dental Team Organization Forensic Odontology and Identification Procedures General Disaster Management	155
16.5 16.6	Dental Team Organization Forensic Odontology and Identification Procedures	155 156
16.5 16.6 16.6.1	Dental Team Organization Forensic Odontology and Identification Procedures General Disaster Management Considerations	155 156
16.5 16.6 16.6.1	Dental Team Organization Forensic Odontology and Identification Procedures General Disaster Management Considerations Identification Steps by Disaster	155 156 156
16.5 16.6 16.6.1 16.6.2	Dental Team Organization Forensic Odontology and Identification Procedures General Disaster Management Considerations Identification Steps by Disaster Victim Identification Teams	155 156 156 156

16.1 Introduction

The world has experienced an excess of mass disasters in recent years: acts of terrorism, bombings, earthquakes, hurricanes, typhoons, air crashes and other transportation mishaps, not to mention armed conflicts and migrants drowned in various bodies of water. A major disaster may be defined as any event that occurs with little or no warning causing death or injury, damage to property or the environment, and disruption of the community, and the effects of which are of such a scale that they cannot readily be dealt with by local services and authorities as part of their everyday activities (McLay 2009). Typically, emergency medical services (EMS) will be the first to respond and mainly comprise the ambulance service, local hospitals, and civil defense, with further extension. These services would be easily accessible, however, in the event of a large-scale mass casualty or disaster circumstances such as the Asian tsunami in December 2004, which affected hundreds of thousands of people in India, Thailand, Sri Lanka, and Indonesia, among other countries very difficult to manage and implement properly (Hinchliffe 2011; Rai and Anand 2007). The terrorist attack on the Twin Towers in New York City on September 11, 2001, heralded a new era of urban warfare on a truly global scale. Whereas the urban conflict in Northern Ireland in the 1970s and 1980s was more or less localized to the United Kingdom, "9/11" showed that terrorism was worldwide and could strike anywhere and at anytime (Hinchliffe 2011). The Bali Bombings in 2003 and the London bombings on

July 7, 2005, bear terrible witness to this. Situations like these would place considerable strain on EMS resources in different countries and would be an opportunity for general dentists and forensic odon-tologists as health professionals to expand their capabilities in times of emergency and disaster. This chapter reviews the current literature and highlights potential areas in which the general dental practitioner and forensic odontologist may contribute during mass disasters.

16.2 Types of Mass Disasters (Levinson and Granot 2002)

Mass disasters typically are one of the following types:

- Natural disasters: These include hurricanes, tornadoes, floods, volcanoes, earthquakes, tsunamis, and any other natural phenomena that cause destruction.
- Transportation accidents: This type includes accidents involving airplanes, trains, as well as passenger ships.
- Terrorism: The most common form of terrorism seen in current times involves some kind of explosive device used to kill and wound large numbers of people.

Additionally, according to Hinchliffe (2011), additional classifications of mass disaster are the following:

- Closed type: Identifying information or a potential list of those involved is available.
- Open type: At first, there is no idea who may be involved.
- Mixed: This is a combination of open and closed.

16.3 Difficulties During Mass Disasters

Medicolegal personnel will likely encounter the following issues when coping with a mass disaster:

- 1. Large numbers of humans whose remains are fragmented, commingled, and burned
- 2. Difficulty in determining who was involved in the disaster

- 3. Difficulties acquiring useful medical and dental records and radiographs
- 4. Legal, jurisdictional, organizational, and political issues
- 5. Internal and external documentation and communication problems
- 6. Issues in the application of universal human forensic identification codes

16.4 Disaster Site/Crime Scene Management (Colvard et al. 2006; Guay 2007; Markarian et al. 2006)

The prime purpose of engaging the support of dentists or forensic odontologists in responding to mass disasters is to allow catastrophe managers to use scarce physician resources in the most effective manner possible by having some services they would ordinarily provide be successfully provided by dentists or forensic odontologists where possible. There are several general areas of response activity in which dentists or forensic odontologists can be helpful, as described next.

16.4.1 Surveillance

Many mass disasters are discrete entities easily recognized and of easily defined duration and effects on a population, while some disasters, predominantly bioterrorism attacks and pandemics, often have relatively indistinguishable beginnings and ends and unpredictable effects on a population. Because of the different incubation periods of infectious agents, the time of exposure can be estimated only after disease has manifested (Guay 2007; Markarian et al. 2006). It also may take valuable time to determine that a population-wide problem actually exists. Dentists or forensic odontologists can be part of a successful surveillance network because they are scattered throughout a community much as the general population is. Observations of intraoral or dental lesions when they are present and the notification of health authorities on these observations may assist in the early detection of a bioterrorism attack or the spread of a pandemic infection. Early detection of an infectious agent in a population may help reduce the number of casualties by the prompt initiation of preventive and therapeutic intervention.

16.4.2 Referral of Patients

Patients who show early signs or symptoms of infectious disease, have suspicious oral or cutaneous lesions, or are suspected of having such disease may be referred to a physician for a definitive diagnosis and appropriate treatment if necessary (Guay 2007).

16.4.3 Diagnosis and Monitoring

After an infectious disease that causes mass disasters has been identified, dentists or forensic odontologists who are able to identify the sign and symptoms of that disease may be identifying afflicted patients. Dentists can collect samples such as salivary and nasal swabs or other specimens when appropriate for laboratory processing, which may provide valuable diagnostic information or indicate the progress of the treatment (Guay 2007; Markarian et al. 2006).

16.4.4 Triage

In the effective response to any mass disaster events, a system must be recognized to prioritize treatment among casualties. Dentists or forensic odontologists are able to help in this important function with additional training (Guay 2007).

16.4.5 Immunization and Medications

To prevent the spread of infectious agents, whether from a natural pandemic, a deliberate bioterrorism attack, or contamination as a result of a local event, immunization of great numbers of individuals may be required in a short amount of time. Forensic odontologists or dentists can participate in mass immunization programs and prescribe some medications with a minimum of additional training and may be a critical factor in the success of urgent programs (Guay 2007; Markarian et al. 2006).

16.4.6 Infection Control

Dentists or forensic odontologists perform sound infection-control procedures in their offices on a daily basis. They are well versed and well practiced in infection control and can bring their expertise to mass casualty situations. Dentists or forensic odontologists who are familiar with disaster mortuary activities can be useful in managing the remains of victims whose death is a result of the event, particularly infectious events (Guay 2007; Markarian et al. 2006). These remains most likely will be contaminated and will require careful management to prevent further disease spread.

16.4.7 Definitive Treatment

Forensic odontologists have training and experience in many areas that may be a part of casualty care in mass casualty events, such as treating oral, facial, cranial injuries; providing cardiopulmonary resuscitations; obtaining medical histories; taking samples; assisting with anesthesia; starting intravenous lines; suturing and performing appropriate surgery; assisting in stabilizing patients; and assisting in shock management (Guay 2007; Markarian et al. 2006).

16.5 Dental Team Organization

Basically, a specific outline of the dental team organization is needed. It becomes understandable that many issues must be taken into account in order to organize a team of specialists that would be able to act fast and adequately in an emergency such as a mass disaster. The basic requirements include the following:

- 1. Team experts: Odontologists and members of forensic odontology boards
- Training standards: Training centers, continuing education, special expertise (oral pathology,

oral surgery), helping the team to be physically able to handle complex situations

- 3. Considerations of complex legal issues
- 4. Establishment plan in case of emergency, identification of team members, team structure, supplies, safety of team members and facilities, serve for the adequate mobilization and for the protection of the team

16.6 Forensic Odontology and Identification Procedures

After survivors have been removed from the disaster site, concentration can be focused on recovery and identification of the deceased. An examination of the deceased is conducted for the following purposes (Hinchliffe 2011; Wallace et al. 1994):

- To legally confirm a death
- To estimate the time of death
- To provide identification for families and legal purposes
- To identify the cause and nature of death
- To obtain forensic evidence when criminal involvement is alleged
- To collect evidence that may be needed for criminal investigations

16.6.1 General Disaster Management Considerations (Hinchliffe 2011; Wallace et al. 1994)

General disaster management considerations include:

- Protection and safety of the disaster areas.
- Recovery or collection of bodies, body parts, and belongings.
- Transportation and storage of the remains: Temporary mortuaries consist of an Internet connection with computer networks, communications, water supply, examination tables, radiography equipment, protective clothing, containers, cafeterias, toilets, etc.
- Appropriate team for the collection of antemortem evidences, identification process, body release and other procedures: pathologists,

forensic odontologists, forensic photographers, fingerprint and DNA experts, police, IT specialists, and others.

- Teams of social workers and other personnel trained to deal with hysterical relatives and friends of the deceased.
- Personnel trained to communicate with the media.
- · Quality assurance.

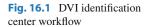
16.6.2 Identification Steps by Disaster Victim Identification Teams (Schuller-Gotzburg and Suchanek 2007; Perrier et al. 2006; Rai and Anand 2006)

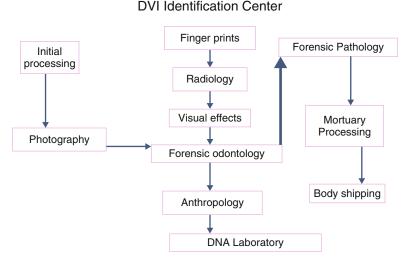
Disaster victim identification (DVI) teams (Fig. 16.1) work in a series of steps in the process of identifying victims. The steps consist of four main stages: body tagging and bagging; finger-printing; forensic pathology; and forensic odon-tology. The bodies are refrigerated, both before and after the procedure.

- Body tagging and bagging: Each body is labeled with an identifying number and then placed in a body bag.
- Fingerprinting: Done where possible, but it is impossible in cases of highly decomposed bodies, which almost invariably show extensive postmortem skin desquamation.
- Forensic pathology: Each body is examined by a four-member team. The team consists of a forensic pathologist, a forensic death investigator, a mortuary technician, and a forensic photographer.
- Forensic odontology: Oral cavity (Oral-dental) is examined by expert forensic odontologist.

16.6.3 Steps in Identifying Bodies

When a DVI team is at work identifying bodies, they take the steps outlined here to identify bodies. Bodies are transported to the mortuary from the fingerprinting section. The scribe receives and signs the tracking form. The pathologists and score



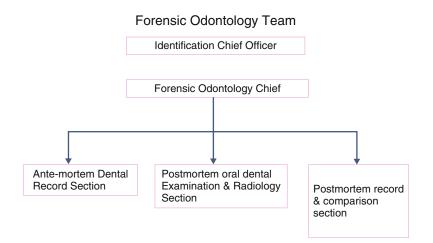


confirm the body number using the pink PM DVI form. The body number is photographed. The technician removes and rinses or washes the clothes to display their respective brand, size, color, and designs. The attire is then photographed and recorded. Attire is then placed in a sealed bag, which, in turn, is placed within the body bag. An external examination of the body is carried out. The sex, height, estimated age, tattoos, scars, physical abnormalities, and other characteristics are recorded. A midline abdominal incision is made to check for the presence or absence of the gallbladder, appendix, female internal genitalia, and for evidence of any other previous operations. Other incisions are made where necessary. Proof of any other identifiable disease is required and evidenced. The technician removes the mandible to facilitate the subsequent forensic dental examination. The body is finally sent for dental examination.

16.6.3.1 Forensic Odontology

The report of the SS *Noronic* disaster in 1949 gives a clear account of the amount of information that could be derived from dental and oral findings (Grant et al. 1952). As the report described, a DVI team of paired odontologists, supervised by a senior odontologist (Fig. 16.2), worked in these sections, for dental examination. Facial dissection was performed. The skin and underlying tissue were then reflected upward over the face to expose the maxilla and mandible. The photographs were taken and labeled. The dental investigator then proceeded to produce the postmortem dental record. The dentist carried out the dental examination and called out the finding for every tooth. The dentist inscribe charted them in the pink DVI form using the Interpol dental charting system.

Profiling has gained importance in human identification following mass disasters in the last decade (Bud Imlija 2003; Hsu et al. 1999). The Interpol dental charting system has been applied as the World Dental Federation tooth numbering system. During examination, teeth that might have received root canal treatment are identified for radiographic investigation. An untreated tooth, preferably a molar, provides a source of genomic DNA for profiling, but if teeth are not available, then a segment of femur shaft bone is removed instead by pathologists or anthropologists. One dentist X-rays the dental remains while the other labels each X-ray film. Two bitewing radiographs, one for each side of the jaw, and some additional radiographs are taken. Any information revealed by radiographs will be recorded in the pink DVI form. The many combinations of missing teeth, caries, fillings, and prostheses involving these 160 surfaces greatly decrease the likelihood of any two adults having identical mouths. It has been shown that even identical twins do not have identical



dentition. Dental structures are the hardest and most resilient tissues of the body. No matter the manner of death, dentition can survive most postmortem events that can disrupt or change other body tissue (Sweet and DiZinno 1996). Infection control and the welfare of attending workers are also duly considered and attended to in the DVI process. Postmortem data are recorded on Interpol forms and matched with antemortem data.

16.6.3.2 Forensic Odontological Identification Process

Mass disaster victims' bodies may be in different conditions, such as traumatized, drowned, burned, decomposed, and skeletonized (Wright 1995). The condition of burned bodies ranges from somewhat reddened skin to incinerated remains. The final state of decomposition is skeletonization. In the conditions described above, different methods are used during the process of identification (Wright 1995):

- 1. Easily identifiable body
 - (a) Photographs
 - (b) Radiographs
 - (c) Dental charting
 - (d) Dental impressions
 - (e) Preservation of oral structures
- 2. Decomposed, traumatized, or incinerated body
 - (a) Photographs
 - (b) Radiographs
 - (c) Dental charting
 - (d) Preservation of remains

- 3. Skeletonized remains
 - (a) Photographs
 - (b) Radiographs
 - (c) Dental charting
 - (d) Jaw articulation and occlusal analysis
 - (e) Preservation of remains

Access to the Oral Cavity

(Fereira et al. 1997; Stavrianos et al. 2010)

Forensic odontology adds to the identification process by estimating age and habits and determining sex and race of the bodies. Bite marks or pattern injuries as well as rugoscopy, photography, and radiography may also serve as useful material.

A method for the approach comprises cutting the soft tissues through the fat and muscles from the angle of the mandible to the midline. The incision continues to the lateral surfaces of the mandibular base (Whittaker and McDonald 1989). The masseter muscles and the vestibular attachment are incised. The next steps involve the removal of the mandible and maxilla. It is known as a Stryker autopsy saw (Sperber 1999; Whittaker and McDonald 1989). In most cases where postmortem rigor (rigor mortis) is present, the admittance is possible either by approaching and pushing on the retromolar pad of the mandible or by following a transmandibular resection. In other cases, the removal of the tongue and larynx is another possible approach (Whittaker and McDonald 1989).



Fig. 16.3 Perioral incision

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Perioral incision
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for reflecting extra-oral tissue over teeth

Other Techniques for Dissections

Additional techniques used for dissections are as follows (Sperber 1999):

- 1. Mallet and chisel method: According to this approach, in which a Le Fort I fracture is induced, position the chisel below the zygomatic arch, high on the maxillary sinus walls bilaterally.
- 2. Pruning shears method: The blade of the scissors is placed within the nares and forced back into the maxillary sinus. The cut is made superiorly to the apices of roots of the maxillary teeth.

In case of badly disfigured cadavers, attention must be given to the possibility of tooth loss during the process of head dissection. It is important to carry out the radiographic examination prior to the relocation of the cadaver. Extraoral incisions, such as bilateral incisions from the oral commissures to the body of the ramus, on a plane parallel with the plane of occlusion, may need to be made (Fig. 16.3). An inframandibular incision applies to dissection of the skin inferior and medial to the mandible in a direction from the ear across the midline to the opposite ear. Jaw resection can be performed using a Stryker saw in order to make a cut on the ramus of the mandible and most superior-posterior part of the maxilla, avoiding the area of the root of the teeth. Furthermore, the tongue and the floor of the mouth can be dissected with a standard knife (Whittaker and McDonald 1989). After adequately identifiable photographs are taken, in order to record the conditions under which the cadaver was found, the following incisions are made (Fereira et al. 1997):

- 1. Superior incision: incision from one side of tragus to other tragus of the ear, including the anterior nasal spine
- 2. Inferior incision: from the mental eminence of the jaw, at the base of the alveolar process sideways to the body of the mandible, parallel to its inferior edge, crossing the ramus and arriving at its back edge, sectioning the masseter muscle
- Lateral incisions: two, one on each side, joining the two aforementioned sections

After all of the above processes have occurred, prostheses and other appliances are removed if applicable, anomalies are noted, and the occlusion is charted, followed by occlusal radiographs and other radiography being taken. In cases where DNA profiling is a method of identification, untreated teeth with large pulp are maintained for the laboratory (Rai and Anand 2007). Basically, this approach isn't the method of choice, and as in the other identification methods, it requires comparative data, which means antemortem records. Dental teams will usually be involved with AM (antemortem) and PM (postmortem) dental information collection, input, interpretation, clarification, and comparisons. While remains are being recovered, general and dental antemortem information will be collected. Family members are contacted, usually by police, for information on the whereabouts of any dental records. Antemortem dental teams can then begin to translate the records, wrestling with terminology, handwriting, abbreviations, and various world charting systems, so that the most up-to-date dental status, at the time of death, is known. The AM team may be based in their home area/country with information relayed electronically to the incident site, or at the incident site. The findings are recorded in a suitable format for comparison with the dental postmortem examination findings (Hinchliffe 2011). The antemortem data should include dental records such as radiographs and photographs.

16.7 Antemortem Data Procedures (Whittaker and McDonald 1989)

The AM team will perform the following tasks:

- 1. Set up a possible victim list.
- 2. Establish and procure antemortem records.
- 3. Organize for delivery of original medical/dental files to disaster center (e-mail).
- 4. Develop a composite antemortem form for each potential victim from evidence supplied.

Conclusions

Every dentist, forensic odontologist, or physician should maintain antemortem records. Local communities should establish adequate manpower strength available for instant activation. This should include local personnel, law enforcement agencies, transportation and traffic companies, as well as specialists such as forensic odontologists, forensic pathologists, forensic anthropologists, fingerprint specialists, etc., with good teamwork. Updated equipment and supplies should be maintained in a ready condition at all times. The material should be periodically checked, and outdated materials should be replaced. The education and training of the disaster team and related group should be done periodically to ensure readiness of the team and to update and refine the protocol for better performance. If resources and comparative data are available, simple methods can be supplemented by forensic techniques, namely, dental, DNA analysis, and fingerprinting. A good team should have a formal procedure to coordinate the management of dead bodies.

Finally, no country has sufficient capacity to respond to very large disasters, and networks of countries, forensic institutes, and international agencies such as WHO and Interpol are needed to provide assistance and funding for the management of the dead following future mass disasters.

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DNA Technology and Forensic Odontology

Contents

17.1	Introduction	163
17.2	Guidelines for Obtaining Dental DNA	164
17.3	Source of DNA	164
17.3.1	DNA from Teeth	164
17.3.2	Common Method of DNA Extraction	
	from Teeth	165
17.3.3	DNA from Saliva and Oral Mucosa	165
17.3.4	Salivary DNA from Bite Mark Sites	165
17.3.5	DNA from Buccal Mucosa	165
17.4	Handling of Samples, DNA Extraction, and PCR Amplification	165
17.4 17.5		165
	and PCR Amplification Genomic and Mitochondrial DNA	165 166
	and PCR Amplification	
17.5	and PCR Amplification Genomic and Mitochondrial DNA in Forensic Dentistry	166
17.5 17.5.1	and PCR Amplification Genomic and Mitochondrial DNA in Forensic Dentistry CODIS	166 166

17.1 Introduction

The well-known significance of forensic dentistry for human identification, mainly when there is little remaining material to perform such identification (e.g., in fires, explosions, decomposing bodies or skeletonized bodies), has led forensic odontology investigators to become more familiar with advanced molecular biology technologies. Mass disaster victim identification traditionally relies on the teamwork of different experts, such as police, forensic odontologists, physicians, and pathologists, where antemortem information from the missing persons is compared with postmortem data of the deceased persons (Olaisen et al. 1997). In most cases, forensic odontology investigations may fail due to a lack of proper antemortem records (Pretty 2007). If antemortem data are unavailable, then the precise identification becomes complicated, and only DNA profiling systems can expose the exact identity of a person. Because of the resistant nature of dental tissues to environmental assaults, such as incineration, immersion, trauma, mutilation, and decomposition, teeth represent an excellent source of DNA material (Schwartz et al. 1991; Rai et al. 2004). The DNA extracted from the teeth of an unidentified individual will be compared with DNA isolated from known antemortem samples, such as stored blood, toothbrush, hairbrush, clothing, collection of buccal cells with help of DNA-SALTM, cervical smear, biopsy, or DNA of a parent or sibling (Sweet and DiZinno 1996).

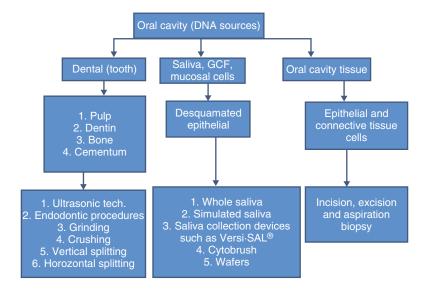


Fig. 17.1 Orodental source of DNA

17.2 Guidelines for Obtaining Dental DNA

The following guidelines outline how to obtain dental DNA:

- 1. Determine if any soft tissue or blood is adherent to the tooth that should be sampled.
- 2. Debride that tooth of any plaque or calculus with a curette, and wash thoroughly with hydrogen peroxide followed by ethanol.
- If the tooth is intact and is supposed to have been removed from the alveolus recently, a conventional endodontic access and instrumentation can be performed.
- 4. Sectioning the tooth provides greater access to pulp.
- Once the tooth is opened, the walls of the pulp chamber can be curetted or instrumented with a slow rotary burr. Then pulp tissue can be collected in a wide open sterile tube.
- 6. In case of dried specimens, the pulp may be mummified parchment-like. After instrumentation, the chamber is best irrigated with buffer. Subsequent ultrafiltration of the liquid at the lab will remove the cellular material needed for analysis.
- 7. Finally, crushing the tooth may be required (Smith et al. 1993).

17.3 Source of DNA (Fig. 17.1)

17.3.1 DNA from Teeth

Teeth are acknowledged to survive most postmortem events, including natural phenomena such as decomposition and autolysis, as well as environmental insults, such as water immersion, burial, and fires to as hot as 1,100°C (Schwartz et al. 1991; Rai et al. 2004). In the tooth, dentin and pulp are rich sources of DNA, which can be successfully extracted (Shiroma et al. 2004; Sweet and Sweet 1995). It has been evident that a sufficient quantity of DNA can be extracted from the crown body, root body, and root tip, although the root body is the region that yields the highest quantities of DNA (Gaytmenn and Sweet 2003). Tsuchimochi et al. (2002) analyzed the Chelex 100 chelating resin to extract DNA from the dental pulp for subsequent application on PCR analysis. They found that extraction of DNA from the dental pulp using this resin is appropriate for obtaining high-quality DNA samples for PCR amplification (Tsuchimochi et al. 2002). The pulp created the strongest PCR amplification signals, while dentin and cementum signals were very similar to each other (Malaver and Yunis 2003). In another study, a DNA profile was obtained in four of five tested samples composed of $250 \ \mu$ l of saliva deposited on the skin (Anzai et al. 2005).

17.3.2 Common Method of DNA Extraction from Teeth

In an adapted horizontal sectioning procedure, the tooth is circumferentially scored 1 mm below the cementoenamel junction with a long-shanked round burr, leaving a 3- to 4-mm wide isthmus of intact tooth structure on the buccal surface. The crown is then physically separated from the root. A large round burr is subsequently used to remove as much coronal and root dentin as possible. Reestablishing the shape of the tooth back to its presampled state is possible by reapproximating the crown and root portions of the tooth at the isthmus.

17.3.3 DNA from Saliva and Oral Mucosa

The forensic odontologist should also be asked for recommendations on issues concerning the biological character of oral mucosal and salivary DNA. Salivary DNA can be found from a wideww range of dead objects, including clothing, foods, tobacco products, oral hygiene devices, drink containers, dental prostheses, stamps, and envelopes. DNA from the oral mucosa or buccal mucosa is the objective of the buccal swab technique.

17.3.4 Salivary DNA from Bite Mark Sites

Sweet et al. (1997) proposed the double-swabbing technique in 1997, which truly established the standard of practice for salivary DNA collection from skin. The technique is as follows:

- 1. Systematically moisten the head of a cotton swab in sterile, distilled water.
- 2. Roll the head of this swab over the area of the saliva stain while using moderate pressure and a constant circular motion.
- 3. Permit this first swab to air-dry in a contamination-free environment for at least 30 min.

- 4. Within 10 s of completing the first swab, roll the tip of the second, dry swab crossways through the now moist area of the stain.
- 5. Use a circular motion and light pressure to absorb the moisture from the skin into the second swab.
- 6. Allow the second swab to air-dry in a contamination-free environment for at least 30 min.
- After drying, both swabs are packaged together, sealed, and marked with unique sample and case numbers.
- The chain-of-document is completed and samples are presented to the laboratory.

17.3.5 DNA from Buccal Mucosa

The use of a buccal swab as a rapid, noninvasive alternative to blood collection is becoming increasingly routine in forensic odontology (Walker et al. 1999). Rai (2011, personal communication) used DNA·SALTM (Oasis Diagnostics, USA) to collect DNA from buccal mucosa and found this device reliable.

17.4 Handling of Samples, DNA Extraction, and PCR Amplification (Silva et al. 2007; Walker and Rapley 1999)

Various protocols are used for DNA extraction and analysis, and there is no standard methodology. The PCR technique has been the standard choice for investigating the frequencies of short tandem repeats (STRs) (Anzai et al. 2005). This technique permits amplification of limited regions of the human genome, those associated with genomic hybridization. It has been evident that the PCR method enables differentiation of one individual from another, with a high level of reliability and with about 1 ng (1 one billionth of a gram) of the target DNA (Sweet 2001). It should be mentioned that DNA extraction is a process composed of three different stages: cell rupture or lysis (which allows the use of several techniques for effective rupture of the cell membranes); protein denaturation and inactivation (by chelating agents and proteinases in order to inactive elements, such as proteins); and finally, DNA extraction itself (Vogel et al. 2000). The techniques of DNA extraction most often employed in forensic dentistry are the organic method (composed of phenol-chloroform and used for high-molecular-weight DNA, with a higher likelihood of errors, given the use of multiple tubes), Chelex 100 (the fastest with the lowest risk of contamination, yet very expensive), FTA paper (composed of absorbent cellulose paper with chemical substances, which speed up its use), and isopropyl alcohol (containing ammonium and isopropanol, which is less expensive and also an alternative to the organic method) (Butler 2005).

17.5 Genomic and Mitochondrial DNA in Forensic Dentistry (Silva et al. 2007)

It has been reported that the analysis of mitochondrial DNA for forensic purposes is restricted to ancient tissues, such as bones, hair, and teeth, in which the nuclear DNA cannot be analyzed (Silva and Passos 2002). However, this examination is performed by direct sequencing of its nitrogenous bases, which is a very expensive technique because it employs a highly specialized technology. Also, mitochondrial DNA is exclusively matrilineal and hence less informative (Silva et al. 2007). In forensic odontology samples, the study of DNA (genomic and mitochondrial) is usually performed by STR analysis, which can be defined as hypervariable regions of DNA that present consecutive repetitions of fragments that have two to seven base pairs (bp). The VNTR (variable number of tandem repeats) testing, which may present short repeated sequences of intermediate size (15-65 base pairs), is infrequently used in forensic analyses due to the poor-quality DNA provided with this method. The most valuable STRs for human identification are those that present a greater polymorphism (greater number of alleles), smaller size (in base pairs), higher frequency of heterozygotes (higher than 90%), and low frequency of mutations (Silva et al. 2007).

17.5.1 CODIS

CODIS stands for Combined DNA Index System. It was established and funded by the Federal Bureau of Investigation (FBI) and has become the core of the national DNA database in the United States. It gives information as a central database of the DNA profiles from all user laboratories (Combined DNA Index System 2011). The database uses two distinct indexes.

17.6 Identification and Sex Determination in Forensic Odontology

DNA profiling is used not only for identification but also for gender determination. Sex has been identified from the dental pulp DNA through the analysis of the peaks of X and Y loci by capillary gel electrophoresis (Komuro et al. 1998).

Conclusions

The advance method DNA finger printing has revolutionized the concept of identification in forensic investigations. It is evident that the teeth represent an excellent source of DNA, which is protected by epithelial, connective, muscular and bone tissues in case of incineration, injury and chemical exposure. Forensic odontologist experts working on the field of Forensic odontology should incorporate these new technologies in their work, as several methods are available for DNA extraction from oral-dental materials.

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Pink Teeth: An Important Aspect for Forensic Science

Contents

18.1	Introduction	169
18.2	Review of the Literatures	169
18.2.1	Forensic Pink Teeth	169
18.2.2	Archaeological Pink Teeth	171
18.2.3	Other Causes of Pink Teeth	172
Referen	1ces	172

18.1 Introduction

The appearance of pink teeth after death is a common phenomenon in forensic dentistry. The emergence of pink teeth and evaluation of their causes in forensic dentistry following death are important aspects. Although the phenomenon of pink teeth has been known since 1829, when it was first described by Bell (1829), its application in forensic medicine as well as in forensic dentistry has been inadequate. Recently, however, attention was again focused on pink teeth in legal cases. The medicolegal implications ruled out the use of pink teeth as a "possible marker" in estimating the cause of death.

18.2 Review of the Literatures

We can divide the pink teeth literature into three parts:

- 1. Forensic pink teeth
- 2. Archaeological pink teeth
- 3. Other causes of pink teeth

18.2.1 Forensic Pink Teeth

Thomas Bell first observed the pink teeth phenomenon in 1829 (Fig. 18.1). He illustrated that a pink coloration in teeth had appeared in drowned or strangled bodies. It was the consequence of an increased intrapulpal pressure, which caused the



Fig. 18.1 Forensic pink teeth

formation of pink teeth (Bell 1829). Miles et al. (1953) stressed the occurrence of this phenomenon in cases of strangulation and carbon monoxide intoxication in connection with the Christie murder case. On examination of the tooth's anatomical features, including alveolar bone, periodontal ligament, and the pulp, marked hyperemia was observed more specifically in root dentin than in coronal dentin. Van Wyk (1987) described that a red-pink coloration was observed in the root of the teeth, which, on reaching predominantly deeper areas toward the cementoenamel junction, usually fades off, but the pink color was still visible beneath the enamel. The pulp area in fractured and sectioned teeth was depicted as being filled with a deep red or pink gelatinous material that extended into the dentin, waning in intensity toward the enamel or cementum. In addition, he also commented upon the variability between even adjacent teeth within the same jaws, where some of them displayed a marked "pinkness," while others were relatively unaffected. Van Wyk also performed a histological identification for the causative pigment and recommended that the tubules played a vital role in pigmentation formation, as the "larger their diameter, the greater is the chance of pigmentation of the dentin" (van Wyk 1987, p. 13). He found that the heme group is accountable for the presence of colored pigments that show the phenomenon of postmortem pink teeth; this heme group is present in hemoglobin, the porphyrins, hemosiderins, bile, and its related pigments. Of these, the most important is hemoglobin. Hemosiderin is composed of a protein framework combined with ferric acid and is a breakdown product of hemoglobin, but it only crops up when breakdown is due to macrophages. Hemoglobin was found to be the most likely pigment responsible for the postmortem pink staining. It occurs naturally after death in many forms, i.e., oxyhemoglobin, reduced hemoglobin, meth-hemoglobin, hematin, and hemochromogen (van Wyk et al. 1987; van Wyk 1987, 1988a, b, 1989). Strangling, suffocation, and hanging positions of the body markedly increase the prevalence of pink teeth formation. If there had been an increase in hemosiderin levels soon after death, it possibly would be due to autolysis in the tissues and observation of marked congestion in the head region, which would bring about further congestion within the pulp, followed by hemorrhage and diffusion of blood into the pulp chamber (van Wyk et al. 1987). Miller (1957) found that this phenomenon could also occur in living subjects most frequently associated with infectious disease (e.g., typhoid fever). However, the causes are different from those of postmortem pink teeth. Camps (1953) reported that the formation of carbon monoxide complexes with the heme group occurs as a result of blood vasodilatation, and this would penetrate the pulpal tissues, creating the prerequisites for the pink teeth. An ample amount of blood must be present in the pulp chamber in order to release enough hemoglobin or hemoglobin derivatives from hemolysis to penetrate into the dental hard tissues. Kirkham et al. (1977) also observed that incisors, canines, and premolars seem to stain more intensely than the other teeth. It has also been observed that healthy teeth are more prone, as they rapidly turn into pink, compared to decayed teeth due to an initial reduction in pulpal volume and a subsequent reduced amount of blood in the pulp chamber. The general consensus states that there was no obvious connection between the occurrence of pink teeth and the cause of the death, but the condition of the surroundings (especially humidity) ought to certainly play an important role in the development of the pink tooth phenomenon. The phenomenon is more pronounced in younger individuals due to little age-related changes of the root canals, with a lesser amount of secondary dentin deposition, which makes it more penetrable to the pigment, further accounting for the trouble-free postmortem pink staining. By a histochemical method and autofluorescence, hemoglobin and its derivatives have been identified as the most likely pigments responsible for a postmortem process that can be considered analogous to postmortem lividity. The pigmentation is more prominent in the teeth with single roots rather than in posterior teeth with multiple roots (Campobasso et al. 2006).

Van Wyk (1987) suggested that the phenomenon of the presence of sclerosed and/or secondary dentin is attributable to its impenetrability to hemoglobin and hence free from the pink pigmentation. It has been observed that the phenomenon depends on some special anatomical features, such as the existence of porous structures protected by a dense material, consequently explaining the existence of pink teeth and pink fingernails (Campobasso et al. 2006). Moody and Southam (1982) demonstrated that the pulp of the tooth is second only to the uterus in its fibrinolytic activity. This hypothesis states that the extra fibrinolytic activity of the pulp in certain situations facilitates the rapid breakdown of coagulated blood or prevents postmortem coagulation in the pulp with the breakdown of erythrocytes, which promotes the diffusion of hemoglobin into the dentinal tubules. The situation appears to be critical and might result in "sudden death." The intrapulpal blood vessels become markedly dilated postmortem in such an order that the blood diffuses into the surrounding pulpal tissues, which become pink in color. It also depends on the gravity and subsequent livor mortis (hypostasis) of the blood. The dental hard tissues are more likely to be stained in moist, humid conditions because of extravasations of erythrocytes from the pulpal capillaries undergoing subsequent hemolysis, releasing hemoglobin and its derivatives. Fibrinolysis likely promotes the hemorrhage in the pulp chamber. Pink teeth in drowning cases are observed under increased hypotonic pressure, thus resulting in hemolysis. Extensive burns also result in increased hemolysis. Carbon monoxide intoxication produces marked hyperemia, diffuse hemorrhages, hemolysis, and edema in the tissues. It is possible that congested blood vessels followed by hemolysis will rapidly produce a large amount of hemoglobin within the

pulpal tissue (Beeley and Harvey 1973). Reportedly there exists no significant connection between the pink teeth phenomenon and the cause of death (Xu et al. 2006). It has also been suggested that age as calculated from the pink teeth was probably underestimated, but more accurate age estimation could be obtained after adequate washing (Ohtani et al. 1999; Ritz-Timme et al. 1998; White 1970). It was recently reported that blood congestion and autolysis were prerequisites, but the occurrence of a pink discoloration in certain body regions may be due to special anatomical conditions and may be largely independent of the cause of death. Consequently, the nonspecificity of congestion events and the possibility of postmortem congestion genesis or reinforcement by exogenous factors did not allow the pink phenomenon to be used as specific forensic evidence (Ohtani et al. 1999).

18.2.2 Archaeological Pink Teeth

The description of a pink color in ancient teeth was attributed to the tunneling hyphal penetration in the dentinal tubules as well as small amount of iron, but it was reported that the iron was not responsible for pinkteeth formation. Archaeological teeth had tunneling changes in the dentin because of some fungi such as actinomycetes where the enamel was intact (Soggnaes 1950; Werelds 1962). These tunnels were irregular in path and had no mineral deposition associated with them. Linear longitudinal tunnels were more regular in path than the Wedl tunnels, following the osteon structure of the bone, but displayed a certain amount of redeposited mineral material. Some authors depicted that the budded tunnels were wider than Wedl tunnels or linear longitudinal tunnels, and were described as sometimes having a pinkish tinge (Poole and Tratman 1978). Lamellate foci were seen to follow the osteon structure of bone and were accompanied by a heavy loss of bone mineral and collagen. The microscopic appearance of postmortem changes seen in bone from a medieval ossuary in Gottingen, Germany, was similar to dentin, in which much of the destruction consists of tunnels,

18 Pink Teeth: An Important Aspect for Forensic Science

which gives the impression of having a relationship with the Haversian system (Poole and Tratman 1978). The fungi responsible were cultured and found to be Stachybotrys cylindrospora, Doratomyces stemonitis, genus Pythium, and genus Rhizoctonia. In all the stained areas, a yellowish-green fluorescence was seen in ultraviolet microscopy using a stimulating wavelength of 350-450 nm; blue-white fluorescence stimulated by 350 nm and 300 nm ultraviolet light was associated with collagen cross-links. The dentin in the modern control displayed blue-white fluorescence as exhibited by the nonpink areas of dentin from the Chichester tooth. The pink areas displayed no fluorescence, indicating that collagen cross-linking was no longer present. No yellowish-green fluorescence was observed (Dumser and Türkay 2008). The rate and mode of decomposition of human bodies in the deep sea vary considerably and were mainly influenced by the local faunal composition.

18.2.3 Other Causes of Pink Teeth

Invasive cervical resorption, characterized by its cervical location and its invasive nature, furthermore results in a resorptive process leading to progressive destruction and usually loss of the tooth structure, the clinical features of which often resemble internal resorption ("pink tooth") (Matthews 2000). Primary teeth turned pink as the result of being treated with arsenic and resorcinol. Reasons for the teeth to appear pink other than resorcinol staining include incidence of pulp polyp, internal resorption, caries, and blood extravasations into the dentin after trauma (Zheng et al. 2003). Some restorations such as ceramic teeth may appear pink. Pink discolorations are also associated in patients with lepromatous leprosy, trimipramin intoxication, hypothermia, and pneumonia. It has also been reported that pink teeth are due to barbiturate poisoning. The color of teeth may also turn pink due to thermal damage, but it further depends on the temperature and duration of heat, as demonstrated in burn cases (Dhatterwal and Rai 2006; Endris and Berrsche 1985; Hale 2001).

Conclusions

Pink teeth come about from different causes, such as trimipramin intoxication, hypothermia, pneumonia, systemic diseases, oral diseases, drug poisons, dental materials, complications due to dental treatment, implant materials, fungi, or other stated causes depending upon the type of samples. The mechanism of this phenomenon might be the same as autolysis or blood congestion, which can only favor the occurrence of a pink discoloration. The pink phenomenon depends on special anatomical features such as the existence of porous structures protected by a dense material, which explains the incidence of a pink discoloration in teeth except in dental materials and implants, where it may be explained due to the material's color. Further studies are thus required to differentiate among different types of pink teeth. At present, this issue is partly ignored; only a few publications are available on this matter to provide a thorough understanding about the role of this phenomenon in death cause investigations in forensic dentistry.

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Role of Dentist and Forensic Odontologist in Child Abuse Cases

19

Contents

19.1	Introduction and Background	175
19.2 19.2.1 19.2.2	Child Abuse Scenario Child Abuse Across the Globe Child Abuse in India	176 176 176
19.3 19.3.1	Child Maltreatment Definitions	177 177
19.4	Causes of Death	178
19.5	Risk Factors	178
19.6	Identification	178
19.7	Typical Features of Physical Abuse	178
19.8	Typical Features of Sexual Abuse	180
19.9	Identification of Emotional Abuse	180
19.10	Identification of Neglect	181
19.11	Dental Staff's Involvement	181
19.12	Documentation and Intervention	181
19.13	Reporting Suspicions	182
19.14	Advocacy for the Child Patient	182
19.15	Forensic Odontologists and Child Abuse	182
Referen	nces	183

19.1 Introduction and Background

Maltreatment of infants and children has been traced far back in history, and, tragically, it is still too prevalent in our "modern" world. Good efforts have been made in the different parts of the world, including India, in recent decades in the areas of child abuse recognition and prevention. Many dedicated people today work diligently and tirelessly to educate not only mandated reporters of child abuse but also the general public as well. Child abuse is defined as those acts or oversights of care that divest a child from the opportunity to fully develop his or her unique potentials as a person physically, socially, or emotionally. It can also be defined as any nonaccidental trauma, failure to meet basic needs, or abuse inflicted upon a child by the caretaker that is beyond the acceptable norm of childcare in our culture (Kenney and Spencer 1995; Misawa 2001). Abuse may cause serious injury to the child and may even cause death. To date, the incidence of child abuse is not really clear. Statistical data do not show the actual rate because of the unreported cases. Forensic dentists and oral physicians are in a strategic position to recognize and report children being abused because they often see the child and parents interacting during multiple visits and over a long period of time.

19.2 Child Abuse Scenario

19.2.1 Child Abuse Across the Globe

The United Nations' Secretary-General's Study on Violence against Children (Pinheiro 2006) has given the following overview of the situation of abuse and violence against children across the globe:

- 1. The World Health Organization (WHO) estimates that almost 53,000 child deaths in 2002 were due to child homicide.
- 2. In the Global School-Based Student Health Survey carried out in a wide range of developing countries, between 20 % and 65 % of school-going children reported having been verbally or physically bullied in school in the previous 30 days. Similar rates of bullying have been found in industrialized countries.
- An estimated 150 million girls and 73 million boys under 18 have experienced forced sexual intercourse or other forms of sexual violence involving physical contact.
- 4. UNICEF estimates that in sub-Saharan Africa, Egypt, and Sudan, three million girls and women are subjected to female genital mutilation (FGM) every year.
- 5. The International Labor Organization (ILO) estimates that 218 million children were involved in child labor in 2004, of whom 126 million were engaged in hazardous work. Estimates from 2000 suggest that 5.7 million were in forced or bonded labor, 1.8 million were in prostitution and pornography, and 1.2 million were victims of trafficking.
- 6. Only 2.4 % of the world's children are legally protected from corporal punishment in all settings.

19.2.2 Child Abuse in India (India Committee of the Netherlands 2007)

This section describes child abuse specifically within India.

19.2.2.1 Physical Abuse

The following data have been reported regarding physical abuse of children in India: General

- 1. There is very little research on physical abuse in India. Only two earlier studies are mentioned; two out of three children were physically abused.
- 2. Of 69 % children who were abused, 54.68 % were boys.
- 3. Over 50 % of children in all 13 sample states were subjected to one or more form of physical abuse.
- 4. Most children did not report the matter to anyone.
- 5. The states of Andhra Pradesh, Assam, Bihar, and Delhi have almost consistently reported higher rates of abuse in all forms compared to other states.
- 6. In different age categories, the highest percentage of physical abuse was reported among younger children (5–12 years).

Family

7. Of those children physically abused in family situations, 88.6 % were physically abused by parents.

School

- Sixty-five percent of schoolchildren reported facing corporal punishment; that is, two out of three children were victims of corporal punishment in public and private school.
- 9. Sixty-two percent of the corporal punishment occurred in government and municipal schools.
- 10. NGO-run schools also reported a high percentage of corporal punishment.

Working Children

11. Boys and girls were being abused equally and run a high risk of abuse.

Institutions

- 12. The percentage of abuse of boys in correctional institutions was very high, at 56.37 %.
- 13. Physical abuse of girls in institutions was also very high.

Street Children

14. Physical abuse was reported by 66.8 % of street children.

19.2.2.2 Sexual Abuse

The India Committee of the Netherlands (2007) also found that

- More than half the children surveyed (53.22 %) reported having faced one or more forms of sexual abuse.
- 2. Andhra Pradesh, Assam, Bihar, and Delhi reported the highest percentages of sexual abuse among both boys and girls.
- 3. Severe forms of sexual abuse were reported by 21.90 % of child respondents, while 50.76 % reported other forms of sexual abuse.
- 4. Of the child respondents, 5.69 % reported being sexually assaulted.
- 5. Children on the street, children at work, and children in institutional care reported the highest incidences of sexual assault.
- 6. Half of the reported abusers are persons known to the child or in a position of trust and responsibility.
- 7. Most children did not report the matter to anyone.

19.2.2.3 Emotional Abuse and Girl Child Neglect

Every second child reported facing emotional abuse, and an equal percentage of both girls and boys reported facing emotional abuse. In 83 % of the cases, parents were the abusers. Finally, 48.4 % of girls wished they were boys.

19.2.2.4 Child Labor in India

The SRO is united national based authority which offers technical support to countries in Eastern, Southern and Anglophone West Africa for the development of policy responses and action plans, strengthening the knowledge base, and building institutional capacity for effective and sustainable impact. Recently it is also focused on the design and implementation of comprehensive national action plans aimed at eliminating the worst forms of child labour within the shortest possible time (SRO, 2000). According to (SRO 2000), there were 1104000000 child labour in India, and the world's highest number of working children is in India.

19.3 Child Maltreatment

Child maltreatment is defined as all forms of physical and/or emotional ill-treatment, sexual abuse, neglect or negligent treatment, or commercial or other exploitation, resulting in actual or potential harm to the child's health survival, development, or dignity, that occurs in the context of a relationship of responsibility, trust, or power.

Child maltreatment can be of four types:

- 1. Physical abuse
- 2. Sexual abuse
- 3. Emotional and psychological abuse
- 4. Neglect

19.3.1 Definitions

Physical abuse is exacting physical injury upon a child. This may include hitting, slapping, kicking, beating, or otherwise harming a child.

Sexual abuse is inappropriate sexual behavior with a child. It includes fondling a child's genitals, making the child fondle an adult's genitals, sexual assault (intercourse, incest, rape, sodomy), exhibitionism, and pornography. To be considered child abuse, these acts have to be committed by a person responsible for the care of a child or related to the child (for example, a babysitter, parent, neighbor, relative, extended family member, peer, older child, friend, stranger, or a daycare provider).

Emotional and psychological abuse (also known as verbal abuse, mental abuse, and psychological maltreatment) includes acts or the failure to act by parents, caretakers, peers, and others that have caused or could cause serious behavioral, cognitive, emotional, or mental distress/trauma.

Child neglect is an act of omission or commission leading to the denial of a child's basic needs. Neglect can be physical, educational, emotional, or psychological. Physical neglect entails denial of food, clothing, appropriate medical care, or supervision. It may include abandonment. Educational neglect includes failure to provide appropriate schooling or special educational needs. Psychological neglect includes lack of emotional support and love.

19.4 Causes of Death (Schwartz et al. 1976; Vale 1997)

The following have been reported as causes of death resulting from child abuse:

- 1. Intracranial injuries such us subdural hematoma, subarachnoid bleeding, brain contusion
- Traumatic shock such us widespread hypodermal or intramuscular bleeding
- Suffocation from oronasal block, choking, or drowning
- 4. Weakness from malnutrition
- 5. Pneumonia

19.5 Risk Factors

Child abuse can occur in all cultural, ethnic, and income groups, in rich households and poor ones. About 95 % of victims know their perpetrators. People (1) with a history of child abuse in their own childhood or of abuse against other children, (2) with problems with alcohol or drug abuse, (3) with anger management problems, (4) with especially poor parenting skills, and (5) with poor coping skills, especially related to problem solving and making or having choices, have risk factors for being abusive (Vale 1997). Children born prematurely have been shown to have more risk of being abused (Kenney and Spencer 1995). Children of low-income parents have been shown to have more risk of being abused (Rai 2010, personal view). Stress on families can contribute to the maltreatment of children. Typical problems include financial stress, family separation, illness, substance abuse, unemployment, and overcrowded housing. Some mothers are simply not fulfilled by the insensitivity and lack of feedback from an infant. Stress inside a family due to various reasons (e.g., isolation, a crisis, drug or alcohol abuse, teenaged parents, unmarried mothers, etc.), financial worries, bad residential environments, and children's defiant attitudes are all contributing factors to abuse (Misawa 2001).

19.6 Identification

It has been reported that in order to avoid suspicion, an abusive parent or caregiver may bring a child to various physicians or hospitals over a period of time for treatment, but will visit the same dental clinic or medical center repeatedly (Mouden 1994). Clinical signs of child maltreatment are shown in Table 19.1 (Tsang and Sweet 1999). The history is the most important piece of information and thus should be recorded in detail. Abuse or neglect should be considered when the history reveals the following:

- 1. The child has a history of multiple injuries.
- The family presents an explanation that is not in line with the nature of the injury (i.e., if the dental injuries resulted from a fall, one would usually expect also to find bruised or abraded knees, hands, or elbows).
- 3. There is a delay in seeking care for the injury.
- The child or parent avoids discussing the injury.
- 5. The parent refuses to consent with the planned course of treatment or refuses to be separated from the child, such as during X-rays, photographs of lesions, etc.
- 6. The parent refuses to consent to diagnostic studies for the child.
- 7. The parent or family member brings the child from office to office or from one hospital emergency room to another, so as to avoid the chances of recognition.
- 8. The parent persists in presenting symptoms unrelated to the obvious condition of the child.

19.7 Typical Features of Physical Abuse

Physical abuse may result in several types of injuries, including contusions, ecchymosis, abrasions, lacerations, fractures, burns, bites, hematomas, retinal hemorrhaging, traumatic alopecia, and dental trauma.

Head injuries, including subdural hematomas (which cause more serious injuries and deaths than any other form of abuse), traumatic alopecia, subgaleal hematomas, and bruises behind the ears,

Table 19.1 Clinical signs of child maltreatment

At reception

- 1. Routinely observe children for strange behavior. Evaluate hygiene, outward signs of proper nourishment, and general health. Is the child's clothing appropriate for the present weather?
- 2. Are there any wounds or bruises on the child's face or body?
- 3. How does the child respond to others? Abused children may act aggressively by showing inappropriate anger and loss of control, or they may be sullen, stoic, or withdrawn

Extraoral examination

- 1. Examine the head and neck for asymmetry, swelling, and bruising; inspect the scalp for signs of hair pulling; check the ears for scars, tears, and abnormalities
- 2. Look for bruises and abrasions of varying color, which indicate different stages of healing. Check for distinctive pattern marks on skin left by objects such as belts, cords, hangers, or cigarettes
- 3. Examine the middle third of the face for bilateral bruising around the eyes, petechiae (small red or purple spots containing blood) in the sclera of the eye, ptosis of the eyelids, or a deviated gaze, a bruised nose, deviated septum, or blood clot in the nose
- 4. Check for bite marks, which may be the result of uncontrollable anger by the adult or another child. Bite marks in areas that cannot be the result of self-inflicted wounds are never accidental

Intraoral examination

- Burns or bruises near the commissures of the mouth may indicate gagging with a cloth or rope. Scars on the lips, tongue, palate, or lingual frenum may indicate forced feeding. Oral manifestations of sexually transmitted diseases may indicate sexual abuse
- 2. A torn labial frenum (Fig. 19.1) is an intraoral finding that may indicate abuse. Remember that a child's age is an important consideration since a frenum tear in a young child who is learning to walk is not unusual
- 3. The cause of hard tissue injuries due to trauma, such as fractured (Fig. 19.2) or missing teeth or jaw fractures, should be investigated

Source: From Tsang and Sweet (1999)



Fig. 19.1 Labial frenum injury



Fig. 19.2 Blunt injury in submental mandibular injury

retinal hemorrhage, ptosis, and periorbital bruising, bruising of the auricle and tympanic membrane damage, nasal fractures, or an injury resulting in clotted nostrils (Casamassimo 1986; Kotch et al. 1995; Macintyre et al. 1986) have been reported. Orofacial injuries, including lip injuries such as lacerations, burns, abrasions (Fig. 19.3), or bruising; mouth injuries such as labial or lingual frenum tears, burns or lacerations of the gingiva, tongue, palate, or floor of the mouth; maxilla or



Fig. 19.3 Fracture of maxillary incisors



Fig. 19.4 Bite marks

mandible injuries, such as past or present fractures to facial bones, condyles, ramus, or symphysis of mandible, have been reported (Davis et al. 1979).

Bite mark injuries are usually associated with physical or sexual abuse. It has been reported that many times bite marks are misdiagnosed as simple childhood bruises. Special characteristics of bite marks are oval or circular configuration or a hemorrhage, representing a "suck" or "thrust" mark (Fig. 19.4). It has been reported that marks may occur anywhere on a child's body; the most common sites are the cheeks, back, sides, arms, buttocks, and genitalia (Stechey 1991). Gags applied to the mouth may leave bruises, lichenification, or scarring at the corners of the mouth (McNeese and Hebeler 1977). Multiple injuries, injuries in different stages of healing, injuries inappropriate for the child's stage of development, or a discrepant history should arouse suspicion of abuse.

19.8 Typical Features of Sexual Abuse

Although the oral cavity is a frequent site of sexual abuse in children, visible oral injuries or infections are rare (Folland et al. 1977). While dentists are not as involved as other health professionals in the diagnosis of sexual abuse, orofacial manifestations have been reported, including gonorrhea (erythema to ulcerations and from vesiculopustular to pseudomembranous lesions), condylomata acuminata (appear as single or mulpedunculated, cauliflower-like tiple raised, lesions), syphilis (a papule on the lip or dermis at the site of inoculation), herpes simplex virus [type 2 (HSV-2) presents as an oral or perioral painful, reddened area with a grape-like cluster of vesicles (blisters) that rupture to form lesions or sores], and erythema and petechiae (trauma at the junction of the hard and soft palate may indicate forced oral sex).

19.9 Identification of Emotional Abuse

The World Health Organization (WHO) has defined emotional abuse as follows:

Emotional abuse includes the failure to provide a developmentally appropriate, supportive environment, including the availability of a primary attachment figure, so that the child can develop a stable and full range of emotional and social competencies commensurate with her or his personal potentials and in the context of the society in which the child dwells (p. 13). (WHO 1999)

It may also include acts such as restriction of movement, patterns of belittling and denigrating, scapegoating, threatening, scaring, discriminating, ridiculing, or other nonphysical forms of hostile or rejecting treatment toward the child that cause or have a high probability of causing harm to the child's health or physical, mental, spiritual, moral, or social development (WHO 1999). Although difficult to diagnose, a child enduring emotional abuse may exhibit behavioral and physical indicators such as lack of self-esteem, poor social skills, often antisocial feelings, developmentally delays, passive/aggressive tendencies, behavioral extremes, pronounced nervousness that is often manifested in habit disorders such as sucking and rocking, and self-inflicted injuries such as lip or cheek biting (Erickson et al. 1984).

19.10 Identification of Neglect

Dental neglect, as defined by American Academy of Pediatric Dentistry, is intentional neglect by the parents or caregiver toward the child's oral cavity and dental health or a neglect that prevents the child from getting the necessary dental and oral treatment in order to achieve an oral health level for adequate function. Dental neglect can be seen as the appearance of caries, periodontal disease, and other oral cavity diseases (Kellog 2005). Caries, periodontal diseases, and other oral conditions, if not treated, can lead to pain, infection, and loss of oral function that can affect communication, nutrition, learning activities, and other children's activities needed for normal growth and development (Kellog 2005). The failure to get good dental treatment can be caused by various factors, such as family isolation, poor financial status, parent's neglect, and a lack of parental appreciation for the value of oral health. The point at which to consider a parent negligent and to begin intervention occurs after the parent has been properly alerted by a health care professional about the nature and extent of the child's condition, the specific treatment needed, and the mechanism of accessing that treatment (Sirotnak and Grigsby 2004).

19.11 Dental Staff's Involvement

All dentists should be familiar with the signs and symptoms of child abuse and should be familiar with the reporting laws of their respective country. In the fight against child abuse, the early recognition of the problem is absolutely crucial so that effective intervention can be undertaken (Kenney and Spencer 1995). If the dentist suspects physical abuse with a young patient, then he or she should have another dental staff member also witness the injuries and assist in their documentation. It is important to understand that all dentists have a unique opportunity and an ethical obligation to assist in the struggle against child abuse, and that's because a high proportion of abused children suffer injuries to the face and head, including the oral and perioral regions. These injuries may be observed during the course of dental treatment and in some cases even before the child is seated in the dental chair (Kenney and Spencer 1995). Oral physicians can be expected to perform their duty to help protect our children only after receiving appropriate education about their role in identifying and reporting suspected cases of maltreatment. Dentists must become more aware of their moral, legal, and ethical responsibilities in recognizing and reporting child abuse and neglect. Dentistry must do its part to manage and treat the pain, suffering, and death that result from children's maltreatment; it has been said that victims of child abuse and neglect fall into only two categories: those who lived through it and those who did not (Mouden 1998).

19.12 Documentation and Intervention

The dentist or oral physician should routinely question the child and the parent separately about what caused any observed injuries, and a staff member should be present to act as a witness. Open-ended questions should be asked to avoid harsh implications. Injuries that are contradictory with the described history should be treated as suspicious and documented in the patient's chart. Adequate documentation of suspected cases includes photographs (if possible with different types of light photography) and radiographs of the structures involved. Photographs should include a ruler placed beside the injury to help record its size. Detailed written notes should be made in the dental chart with respect to the location, appearance, severity, and distribution of the injuries. If doubt exists as to whether or not to submit a report, a consultation with the patient's physician, a social worker, or local authorities is recommended. Other practitioners in contact with the child may have similar suspicions and may be willing to discuss them. Consultation with a dental colleague who has experience in child abuse cases may also be helpful.

19.13 Reporting Suspicions

Dentists who report cases of suspected child abuse often ask if the parents should be informed. Evidently, it can be very difficult to tackle a parent with whom a professional relationship has been established. Parents or caretakers do not have to be told when a report is filed. Generally, it is suggested that parents should not be informed because this may lead to an angry confrontation that could potentially place the dentist, the office personnel, or the child at risk. Additionally, the dentist may be convinced not to report the event if it is discussed with the parents (Spencer 1995). The dentist who initially filed the report can ask to be kept informed as to the progress of the investigation.

19.14 Advocacy for the Child Patient

Pamphlets on abuse and resource materials from community agencies can be displayed in the waiting room. Participation by dentists and dental office personnel in organizations concerned with ending family violence can help raise community awareness and improve the profession's public image. All members of the office staff should become familiar with the signs of abuse and be encouraged to pursue continuing education on the subject.

19.15 Forensic Odontologists and Child Abuse

Forensic odontologists may be called upon to assess pattern injuries in child abuse. Usually these injuries will be bite marks inflicted upon the child. The odontologist may have the opportunity to examine the child, living or deceased. More often the odontologist will be supplied with photographs and asked for an opinion as to the presence of bite marks, the quality of the marks, and whether an adult or child may have inflicted the marks. Too frequently the supplied photos are of poor quality, taken at too great a distance from the injury, out of focus, or without the presence of a scale (ABFO ruler). An experienced odontologist will take quality photographs when he or she has the opportunity to see the child, whether in the hospital, clinic, or morgue, etc. Bite marks on a child's body, unless observed soon after the bite was imposed, often present as a diffuse bruising of ovoid or elliptical shape with little or no definition of individual teeth (Chiodo and Rosenstein 1984). Often, the odontologist can only determine if it is a human bite mark and perhaps if it was inflicted by an adult or young child. Proper photography of pattern injuries (bite marks) in the dentist's office, the hospital emergency room, or at the police station can greatly aid in the subsequent analysis and potential comparison of the marks. In cases of child abusehomicide, the forensic odontologist will take photographs of the suspected pattern injuries. If there is any third dimension to an injury, the odontologist will take an impression with a dental impression material in order to have a model of the injury to use for potential comparison to dental models of any suspected biters. It also is recommended that the odontologist cut the tissue involving the bite mark and properly preserve it for later transillumination. Good forensic evidence collection with pattern injuries in child abuse can be of great assistance to the law enforcement investigator and the prosecutor. In some cases, the defendant has pled guilty just prior to trial or there was a stipulation as to the bite mark evidence rather than having the odontologist testify.

Conclusions

This chapter has discussed the prevalence of child abuse in India and worldwide as well as the importance of the dentist's recognizing physical abuse and the methods of documenting suspected injuries. The forensic odontologist may be called upon to document and analyze pattern abusive injuries. It is important that dentists realize their comfort level and competence in treating children and refer the young patient to a specialist when indicated. The dental office should maintain complete and legible records on their pediatric patients for potential forensic utilization.

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Dental Jurisprudence and Ethics

Contents

20.1	Introduction	185
20.2	Presenting Evidence in Court	185
20.2.1	General Principles of Presenting	
	Evidence in Court	185
20.2.2	Evidence Collection and Presentation	
	in Court	186
20.3	Forensic Odontologist as Expert	
	on Evidence	186
20.4	Evidence in Courts	186
20.5	Professional Expectations of Forensic	
	Odontologist in Court Cases	187
Refere	1068	188

20.1 Introduction

Professionals in law enforcement and medicolegal disciplines should know how, in a layperson's way, to present their findings regarding forensic odontology. Dental identification has a legal record with little doubt. The forensic odontologist must follow guidelines proposed by ethical committees and law enforcement authorities. This is vital in medicolegal issues both for the acceptance of forensic dental evidence and to protect the dentist against claims of malpractice, manipulation, and other liabilities. The American Board of Forensic Odontology (ABFO) has published manuals to its diplomates detailing the standard guidelines for such procedures (Hampl 2010).

20.2 Presenting Evidence in Court

20.2.1 General Principles of Presenting Evidence in Court

A forensic odontologist must follow these general principles while presenting evidence in court:

- 1. Attend court punctually and produce all the documents asked for. Attire should be professional.
- 2. Be very familiar with the details of the case, anticipate likely questions, and study the literature.
- 3. Never attempt to remember. Speak slowly and clearly, with confidence.

- Use simple language (layperson's language), avoiding superlatives and exaggeration. Do not fumble while reporting on records or literature. Avoid discrepancies between the record and the testimony.
- 5. Be pleasant and polite to the counsel or the accused. Do not try to evade a question and do not lose your temper.
- 6. Keep an open mind. Be honest, impartial, and truthful.
- 7. Avoid long discussions.
- 8. Textbooks or respected journals can be offered as evidence. Before answering a question with these materials, read the complete passage aloud, as the lawyer might have read only the statements that favor his or her position.
- 9. A dentist does not have the professional privilege to recuse him- or herself from testimony and thus must answer all questions.
- 10. Volunteer information if unfairness would result if this information is not revealed. The response should not exceed the expert's knowledge.

20.2.2 Evidence Collection and Presentation in Court (Bowers 2002)

It is important to follow proper legal steps in the field and laboratory for evidence that will be presented in court proceedings. The characteristic criminal proceeding in the court has the prosecution presenting evidence that either directly or indirectly involves someone in the commission of a crime. The best defense for such accusations of guilt is to persuade the court to refuse to accept such evidence at trial. The argument of inadmissibility is the defense counsel's strongest argument before the trial even begins. The defense may introduce forensic evidence supporting its arguments. The legal basis for a motion to reject evidence at examination is that the supporter violated the defendant's legitimate right to protection against an illegal search or arrest by the police. Evidence resulting from this illegal search or seizure is also subject to exclusion. The general rules regarding this are important for the investigator to understand. The least contentious means of collecting evidence is to get the subject's written permission to obtain evidence or to have a court order permitted for the exact type of evidence required, such as swab, saliva, dental impressions, biopsy, etc.

20.3 Forensic Odontologist as Expert on Evidence (Bowers 2002)

The courts believe an expert to be an individual whose knowledge, training, and experience produce an understanding of facts beyond the abilities of the average individual. This skill must be related to the question being asked in court, such as, "Is this injury a dog bite mark?" or "Is this a human or dog bite mark?" This skill has to help the judge and the jury in reaching a decision. The forensic odontologist, once certified, is allowed to make an opinion on matters that occurred outside his or her presence. It is an important tool in criminal or medicolegal cases, where quite often both the prosecution and defense counsel have their own experts whose opinions do not agree. Experts actually reconstruct, to the best of their ability, the events that occurred during an act related to a crime. Certainly, regarding dental evidence, the best information for a court is from a certified forensic odontologist.

20.4 Evidence in Courts (Bowers 2002)

The physical evidence available in a child abuse case is challenging and needs the forensic odontologist to use extreme care in forming his or her opinion. Courts consider it a new science and subject to review. The law dictates that before evidence from a new scientific method can be acceptable, the contributor of the evidence must demonstrate that the relevant scientific community proves the technique reliable. Different courts have stated that to meet the standard of acceptability, the contribution must have met the following criteria:

- 1. The method has been deemed reliable.
- The witnesses providing testimony are appropriately qualified by a forensic odontologist to give an opinion.
- 3. Correct scientific procedures were used.
- 4. The error rate of the method used and results has been determined. Errors in forensic odontology can be either a misidentification from teeth or a rejection of the true identity.
- 5. The appropriate scientific community has approved the method.
- 6. The method underwent appropriate peer review and publication processes.

20.5 Professional Expectations of Forensic Odontologist in Court Cases

During court proceedings, expert witnesses should be prepared to answer the following questions:

- 1. What are the expert's qualifications in the scientific community?
- 2. Can other experts replicate the same method and come up with the same results?
- 3. Can the method and its results be clarified adequately so the judge and jury can understand its simple meaning?

All factors are measured, independent determinations by the court and do not have to be assembled by the expert. They may influence the weight or value of the expert's testimony. The ABFO code of ethics reads as follows:

To maintain the uppermost quality of individual and professional conduct of ABFO members, the code of ethics was transmitted, which is authorized by all diplomates of the ABFO, and similar rules are followed by a majority of forensic odontology organizations around the world. Every fellowshall avoid doing any material misrepresentation of education, training, or area of expertise; shall refrain from any material misrepresentation of data upon which an expert opinion or conclusion is based; must provide a signed and ethics statement adopted by the executive committee; should maintain his/her professional competency through existing programs of continuing education; may submit formal written allegations of contraventions concerning a fellow with proof and correct interpretations; shall provide technically correct statements in all written and oral reports, testimony, public addresses, or publications, and should avoid any misleading or inaccurate claims (p. 20). (Hampl 2010)

The summary of scientific reasoning is as follows:

- 1. Collect all related evidence and information desired.
- Do not selectively overlook evidence or place unnecessary authority in shaky or irrelevant information.
- 3. Do not permit one's judgment to be affected by bias.
- 4. Use skill when making interpretations, drawing inferences, or publicizing an opinion.

When examining a suspect, the forensic odontologist must be sure to obtain a replica of the relevant legal document, such as a court warrant, for the examination of the suspect (dental records such as detailed extraoral and intraoral examination with photographs and radiographs, case history, DNA sample). *Written consent is mandatory except under specified conditions*.

The contents of a forensic odontology report must be arranged in a standard format and include the following (Hampl 2010):

- Qualification(s) of the analyst
- List of items examined (dental records including radiographs, photographs, and biochemical test reports)
- Results, statistical analysis, and conclusions, with standardized references

ABFO Ethics Committee supervises all forensic dental experts and conducts relevant proceedings relating to all issues in this regard. If all guidelines are firmly adhered to, the forensic dental expert will be observed as a skilled authority and will be protected from any liability. If not followed, the forensic dental expert will face disciplinary action by the peer board as well as civil and criminal lawsuits. Anyone can help the courts and judges formulate a decision and with proper qualifications can be disclosed as an expert. It is the importance that testimony given by a certified forensic dental expert should be accepted by Jury. Simple dental identification cases of unknown deceased individuals require skills any licensed forensic odontologists should possess. Dental identification cases need an interdisciplinary approach to be solved.

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Oral Fluids and Teeth in Toxicology

Amal Abd El-Salam El-Bakary

Contents

21.1	Introduction	189
21.2	Physiology and Biochemical Aspects	190
21.3	Regulation of Salivary Gland Secretions	190
21.4	Sample Collection	191
21.4.1	Collection Methods	191
21.4.2	Collection Devices	191
21.5	Analytical Methodology for the	
	Measurement of Drugs in Saliva	191
21.5.1	Preanalytical Manipulations	191
21.5.2	Immunological Methods	192
21.5.3	Chromatographic Methods	192
21.6	The Use of Saliva for Drug Monitoring.	192
21.6.1	Pathophysiology	192
21.6.2	Drugs Monitored in Saliva	193
21.6.3	Drugs of Abuse	193
21.6.4	Doping Drugs	194
21.7	Advantages and Drawbacks of Use	
	of Saliva	194
21.7.1	Advantages of Use of Saliva	194
21.7.2	Safety	194
21.7.3	Drawbacks of Use of Saliva	194
21.8	Teeth in Toxicology	195
21.8.1	Heavy Metals	195
21.8.2	Tobacco	195
21.8.3	Polyhalogenated Aromatic Hydrocarbons.	195
21.8.4	Fluoride	196
21.8.5	Tetracyclines	196
21.8.6	Other Drugs	196
Referen	ices	197

21.1 Introduction

Whole saliva is a mixture of oral fluids and includes secretions from salivary glands in addition to several constituents of nonsalivary origin (Navazash 1993). Oral fluid is an alternative biological matrix that might have advantages over urine for drug analysis in treatment programs. Whole saliva can be collected noninvasively and by individuals with limited training. In recent years, saliva has attracted much attention, in particular among people interested in the determination of drug concentrations. This suggests that saliva might be substituted for plasma in the areas of pharmacokinetic studies and drug monitoring because of the growing interest in noninvasive procedure (Vindenes et al. 2011).

Oral fluid is an exciting alternative matrix for monitoring drugs of abuse in the workplace, clinical toxicology, criminal justice, and DUI (driving under the influence of drugs) programs. In 2004, the U.S. Substance Abuse and Mental Health Services Administration (SAMHSA) proposed recommended guidelines for oral fluid collection, point-of-collection testing devices, low-cutoff concentrations, and screening and confirmation methods (Bush 2008).

Human teeth can be adversely affected by environmental chemicals, such as heavy metals and dioxin (Billings et al. 2004), and drugs, including tetracycline, anticonvulsants, and those used in chemotherapy (Alpaslan et al. 1999). Mercury in teeth amalgam was reported to become absorbed from teeth to cause systemic toxic effects (Mutter 2011).

21.2 Physiology and Biochemical Aspects

Whole saliva (mixed saliva) is a mixture of oral fluids and includes secretions from both the major and minor salivary glands in addition to several constituents of nonsalivary origin, such as gingival cervicular fluid, expectorated bronchial and nasal secretions, serum and blood derivatives from oral wounds, bacteria and bacterial products, viruses and fungi, desquamated epithelial cells, other cellular components, and food debris (Sreebny 1989).

The human salivary glands produce serous and mucinous saliva containing minerals, electrolytes, buffers, enzymes and enzyme inhibitors, growth factors and cytokines, immunoglobulins, mucins, and other glycoproteins (Tabak 1995). Mucins are a group of glycoproteins that contribute to the viscoelastic character of the mucosal secretions (Schenkels et al. 1995).

Proteins that are found in saliva, such as lactoferrin, lysozyme peroxidase, defensins, and histatins, can destroy or inhibit the growth of microorganisms in oral cavity (Xu et al. 1991; Schenkels et al. 1995). In addition, saliva has lubricating functions and aids in the digestion of food (Mandel 1987).

Saliva is alkaline with a pH between 6.4 and 7 (Navazash 1993), a specific gravity of 1.007, and a viscosity of 99.5% water and 0.5% solid contents (40% inorganic constituents/60% organic constituents). Organic constituents are mucin, enzymes, amylase, lysozyme, albumin, globulin, urea, uric acid, cholesterol, vitamins, and phospholipids (Kaufman and Lamster 2002).

Salivary composition depends on many factors: stimulation, diet, age, time of day, disease, etc. Lower pH values occur more frequently among caries-susceptible individuals. The amount of saliva secreted by an adult in 24 h varies between 1,000 and 1,500 ml. In the absence of obvious external stimuli, the rate of salivary secretion in adults is 0.1–0.25 ml/min; values <0.1 ml/min should be considered abnormal. The stimulated flow rate varies between 1 and 2 ml/min, and values <0.5 ml/min should be considered abnormal. The resting saliva reflects the basal flow rate. Stimulated saliva is present in our mouths for up to about 2 h of the day (Langel et al. 2008).

21.3 Regulation of Salivary Gland Secretions

Secretion of saliva is governed by the central nervous system along with the autonomic nervous system. Reflex salivary flow occurs at a low "resting" rate and for short periods of the day. A more intense taste or chewing stimuli evoke up to tenfold increases in salivation. All salivary glands are supplied by cholinergic parasympathetic nerves, which release acetylcholine that binds to muscarinic receptors, evoking the secretion of saliva by acinar cells. Most salivary glands also receive a variable innervation from sympathetic nerves, which release noradrenalin, causing a greater release of stored proteins (Proctor and Carpenter 2007).

Stimulated secretion occurs via nervous reflexes. Neural mechanoreceptors and chemoreceptors in the oral cavity respond to dryness of mucosa, chewing chemicals in foods, and texture of the food. Afferent impulses are integrated in the medulla, and the salivary center receives inputs from the cortex, amygdala, and hypothalamus. Salivary gland secretions may be inhibited temporarily with infections or drugs. Permanent inhibition occurs in irradiation of the head and neck (Rai 2007).

A number of drugs can affect the secretion of oral fluid. These include amphetamines, cannabis, sedating antihistamines, antipsychotic drugs, anticholinergic drugs, and a number of antidepressants. There are less commonly used drugs that increase flow, including clonidine, pilocarpine, and beta-2 stimulants (Aps and Martens 2005).



Fig. 21.1 An example of an oral fluid device consisting of a swab and a collecting vial

21.4 Sample Collection

21.4.1 Collection Methods

A variety of methods are available for collecting saliva. Some involve stimulating saliva production, while others target the collection of unstimulated saliva. According to Navazash (1993), unstimulated saliva can be collected by the draining method, which is performed by allowing saliva to drip from the mouth into a collection container. Several techniques may be used to collect stimulated saliva. The simplest involves tongue, cheek, or lip movements without the use of an external stimulus. Chewing paraffin wax, Parafilm, Teflon, rubber bands, gum base, or chewing gum are usually referred to as mechanical methods of stimulating saliva production. A lemon drop or citric acid can be placed in the mouth to provide a gustatory stimulus for saliva production. Following stimulation by one or more of these methods, saliva can be spit, suctioned, or swabbed from the mouth (Hold et al. 1995). The collected saliva is then put into a vial, centrifuged at 3,000 rpm for 5-10 min, and supernatant fraction is sealed following the complete chain of custody (Bosker and Huestis 2009).

There are several potential problems associated with stimulating saliva production. Parafilm absorbs some drugs and therefore gives erroneous results when saliva is tested for drugs or drug metabolites. Also, paraffin contains compounds that may affect chromatographic analyses—again affecting drug testing accuracy. Some salivary stimulants may change the salivary composition and, therefore, affect the saliva-drug concentration. For example, citric acid may change saliva pH and consequently alter drug concentrations in the saliva. Citric acid and cotton have also been shown to alter immunoassay drug test results (Hold et al. 1995; Gallardo and Queiroz 2008).

21.4.2 Collection Devices

Oral fluid-collection devices are now used for large-scale testing applications. Many devices collect 1 ml of oral fluid. They have names such as Oral Diffusion Sink and Salivette (Shipley et al. 1992), Proflow (Sonesson et al. 2008), and Orasure (Fritch et al. 2011) (Figs. 21.1 and 21.2).

Some oral fluid-collection devices have builtin volume adequacy indicators, facilitating appropriate collection, whereas other manufacturers report only approximate amounts collected. Another approach is gravimetric determination by weighing the device before and after oral fluid collection (Dickson et al. 2007).

21.5 Analytical Methodology for the Measurement of Drugs in Saliva

21.5.1 Preanalytical Manipulations

Once samples have been collected, it is important that they be properly stored (at $-20 \degree C \text{ or } -80 \degree C$)

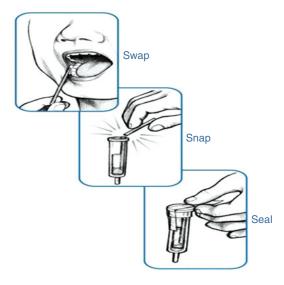


Fig. 21.2 The method of oral fluid collection using the Intercept collecting device

unless analyses are to be performed immediately. Long-term storage of salivary samples at room temperature requires cortisol (Cone and Weddington 1989).

Most authors use the centrifugate, either by extracting the centrifugate with an organic solvent at a desirable pH or by using the centrifugate directly in the analytical process without sample pretreatment (Hold et al. 1995).

21.5.2 Immunological Methods

Immunological methods of analysis have been widely used for monitoring drugs in saliva and other body fluids, mainly because of their relative simplicity of use, requiring little or no extractive operations, their application to large batch analyses, and their sensitivity (Caddy 1984). However, they do not always possess the specificity required for distinguishing metabolites from the parent drug (Paxton and Donald 1980).

Radioimmunoassay (RIA) techniques have been used especially for the analysis of hormones in saliva and some drugs as cocaine, cannabinoids, haloperidol, theophylline, and cotinine. An enzymemultiplied immunoassay technique (EMIT) may be used, but it is less sensitive (Hold et al. 1995).

21.5.3 Chromatographic Methods

Thin-layer chromatography (TLC) has the advantage of simplicity and allows the simultaneous determination of several samples. However, the major limiting factor is the detection of the respective spots on the thin-layer plate at low drug levels in saliva (Hold et al. 1995).

After the initial screen test, confirm suspect positive test results by gas chromatography/ mass spectrometry (GC/MS) or liquid chromatography–mass spectrometry/mass spectrometry (LC-MS/MS). These confirmation methods provide the most sensitive "fingerprint" identification available for the target drug. LC-MS/MS permits the simultaneous analysis of multiple, nonvolatile, labile, polar, and/or high-molecular-weight compounds in a limited oral fluid volume (Samyn et al. 2007).

21.6 The Use of Saliva for Drug Monitoring

21.6.1 Pathophysiology

There are several ways by which serum constituents that are not part of the normal salivary constituents (i.e., drugs and hormones) can reach saliva. Within the salivary glands, transfer mechanisms include intracellular and extracellular routes. The most common intracellular route is passive diffusion (across a concentration gradient), although active transport has also been reported. Ultrafiltration, which occurs through the tight junctions between the cells, is the most common extracellular route (Haeckel and Hanecke 1993).

A fundamental prerequisite for this diagnostic application of saliva is a definable relationship between the concentration of a therapeutic drug in blood (serum) and the concentration in saliva. The presence of a drug in saliva is influenced by the physicochemical characteristics of the drug molecule and its interaction with the cells and tissues of the salivary glands, as well as by extravascular drug metabolism. Factors such as molecular size, lipid solubility, and the degree of ionization of the drug molecule, as well as the effect of salivary pH and the degree of protein binding of the drug, are important determinants of drug availability in saliva (Siegel 1993).

Generally, smaller molecules diffuse more easily than larger ones and lipophilic molecules diffuse more easily than lipophobic molecules (Haeckel and Hanecke 1996). Drugs that are not ionizable, or are not ionized within the pH range of saliva, are the most suited to salivary drug monitoring. The unbound (usually the pharmacologically active) fraction of the drug in serum is available for diffusion into saliva (Haeckel 1993).

Weak bases are detected in higher concentrations and for longer times in oral fluid than in plasma because of oral fluid ion trapping, as blood pH is 7.4 and that of oral fluid is 4–6 (Bosker and Huestis 2009).

21.6.2 Drugs Monitored in Saliva

Anticonvulsant drugs can be easily monitored using saliva, including phenobarbital, phenytoin, carbamazepine, lamotrigine, oxcarbazepine, topiramate, levetiracetam, and gabapentin, providing an approximation of the serum-free level. For highly protein-bound medications such as phenobarbital, phenytoin, and carbamazepine, saliva has the advantage of providing an approximation of the serum-free, active moiety level (Baumann 2007). Carbamazepine levels were found to be 38% of serum carbamazepine levels (Rosenthal et al. 1995). In another study, salivary levels of phenobarbital and phenytoin demonstrated excellent correlations (r=0.98 and 0.97, respectively) with their serum levels (Kankirawatana 1999).

Saliva may be used for monitoring patient compliance with psychiatric medications (El-Guebaly et al. 1981). A significant correlation (r=0.87) exists between the salivary and serum lithium levels in patients receiving lithium therapy (Ben-Aryeh et al. 1980, 1984). A lower correlation (r=0.68) was found between salivary and total serum levels of cyclosporine. Salivary levels of this drug reflect the serum levels of free cyclosporine. Therefore, salivary cyclosporine levels may correlate better with serum levels of free, rather than total, cyclosporine (Coates et al. 1988).

Similarly, the salivary theophylline concentration demonstrated a better correlation with the serum concentration of free theophylline (r=0.85) than with the serum concentration of total theophylline (r=0.85) (Kirk et al. 1994). Stimulated saliva predicted both the total and unbound serum theophylline concentrations within ±1 mcg/ml in 62.5% and 92.9%, respectively, of the samples examined. The therapeutic range for saliva, which corresponds to the accepted total serum concentration range of 10–20 mg/l, was found to be approximately 5.6–11.3 mg/l (Blanchard et al. 1991).

Van Oosten et al. (1986) detected metronidazole in saliva. Measuring chloroquine concentrations, the saliva-to-total plasma concentration ratio was found to be approximately constant (Brown-Awala et al. 1989). Also, saliva may also be used for monitoring levels of anticancer drugs such as irinotecan (Takahashi et al. 1997). Paraquat has been detected in oral fluids after exposure to acute polychlorinated biphenyls (PCBs) and polychlorinated-dibenzo-p-dioxins (Ogawa et al. 2003).

21.6.3 Drugs of Abuse

Following drug use, the appearance of the drug in saliva follows a time course that is similar to that of serum. In contrast, drugs appear at a later time point in urine. The presence of illicit drugs, and not their concentration, is usually sufficient for forensic purposes (Kaufman and Lamster 2002). The LC-MS/MS method has been used for screening the most commonly abused drugs and their metabolites in preserved oral fluid to detect a relatively recent use of drugs (Badawi et al. 2009; Vindenes et al. 2011).

The salivary ethanol concentration may be used as an index of the blood ethanol concentration, provided that the salivary sample is obtained at least 20 min following ingestion. This will allow for the absorption and distribution of alcohol and prevent a falsely elevated reading due to the oral route of consumption (McColl et al. 1979). The saliva-to-serum ratio of ethanol is generally about 1 (Penttila et al. 1990).

Oral fluid testing is useful for monitoring codeine exposure, with a detection window of 7–21 h for single doses, depending on cutoff concentrations (Kim et al. 2002).

Other drugs can be identified in saliva, such as amphetamines, barbiturates, benzodiazepines, phenyclidine, cocaine, and opioids (Kidwell et al. 1998; Øiestad et al. 2007). Also, hypnotics, sedatives, and a muscle relaxant were identified (Gjerde et al. 2011). Saliva can be used to detect recent marijuana use for at least 4 h after marijuana is smoked (Isutsu et al. 1981) by means of radio immunoassay (Gross et al. 1985), ultraperformance LC-MS/MS (Badawi et al. 2009), and enzyme immunoassay (ELISA) (Schwope et al. 2010).

Saliva can be used to monitor tobacco smoking and exposure to tobacco smoke. Salivary cotinine levels were found to be indicative of active and passive smoking (Repace et al. 1998).

21.6.4 Doping Drugs

The most frequently used biological specimen for the determination of drugs in doping control is urine, since only a noninvasively obtained sample is acceptable for routine collection. Yet, even the acceptability of a urine sample is being disputed in view of the potential invasion of privacy, especially if a directly observed collection is advisable to prevent adulteration or substitution of sample (Schramm et al. 1992).

That happens, for instance, when athletes try to escape detection by using urine from someone else. Another major disadvantage of urine is the variability in the renal clearance of drugs and their metabolites, which is largely due to fluctuations in the flow rate and pH of urine. Not all drugs are excreted in the urine; for instance, the lipid-soluble β -blocking drugs tend to be rapidly eliminated by various metabolism systems in the liver (McDevitt 1987). At present, saliva is not used as a biological fluid for doping control (Rai 2007).

21.7 Advantages and Drawbacks of Use of Saliva

21.7.1 Advantages of Use of Saliva

In a clinic or lab, saliva is relatively easy to collect in sufficient quantities for analysis, the costs of storage and shipping tend to be lower than those for serum and urine requires no special handling. Saliva is easy to obtain, with less invasion of privacy and ease of adulteration compared to urine. Salivary sampling protocols are advantageous as they make for frequent and easy collection of samples by noninvasive, needle-free, stress-free techniques (Choo and Huestis 2004). The risk of infection is lower than that for blood (Langel et al. 2008).

21.7.2 Safety

For forensic scientists, saliva tests are safer than blood tests, which are more likely to result in exposure to HIV or hepatitis. For the patient, the noninvasive collection techniques for saliva can dramatically reduce anxiety and discomfort, thereby simplifying collection of serial samples for monitoring of drugs (Rai 2007).

21.7.3 Drawbacks of Use of Saliva

There are several drawbacks to using saliva in drug testing:

- The concentration of drugs tested is usually higher in blood and urine than in saliva.
- Forensic toxicologists know little about saliva and are understandably reluctant to use it as a drug determinant fluid.
- Forensic laboratories now have automated settings for testing blood and urine. Setting them up for saliva will be required.
- It is essential that calibrators and controls be prepared in the same matrix as authentic samples, especially with LC-MS.
- Drugs may reduce salivation, limiting sample volume and necessitating sensitive analytical methods to quantify multiple analytes in one assay.

- Contamination may occur from ingested food and beverages and unknown adulteration techniques.
- Drugs that are smoked, inhaled, insufflated, or taken orally or sublingually also may contaminate the oral mucosa and oral fluid, increasing detection, but reducing correlation with blood concentrations for 30–60 min (Crouch 2005; Verstraete 2005; Bosker and Huestis 2009).

21.8 Teeth in Toxicology

Many environmental chemicals, drugs, or physical agents can adversely affect human teeth during their embryonic development and after their eruption into the oral cavity.

21.8.1 Heavy Metals

It has long been known that lead crosses the placenta and has the potential to affect the development of different organ systems (Goyer 1990), including teeth (Pearl and Roland 1980). Evidence of a cause-and-effect relationship between embryonic and early childhood exposure to lead and an enhanced susceptibility to dental caries is lacking. Some studies reported that lead significantly diminished salivary flow rates and subsequently increased caries levels (Watson et al. 1997).

Lead and cadmium can be detected in tooth enamel in persons exposed to them (Arruda-Neto et al. 2010). The concentrations of chromium, cobalt, lead, and cadmium were significantly higher in primary and permanent teeth with cariesthaninthehealthyones(Gierat-Kucharzewska and Karasi ski 2006). Cadmium chloride induced discoloration of the incisors in ovariectomized rats and destruction of the enamel organ (Katsuta et al. 1996). An individual was reported to have green surface staining of the cervical margins of his teeth 10 months after starting work in a brass foundry (Donoghue and Ferguson 1996). Cadmium was reported to cause defects in crystal formation and impairment in the mineralization of bone and teeth (Kakei et al. 2009).

Although the toxic properties of mercury have been well understood for many years, there is no known association between mercury and tooth development or between mercury and dental health. Mercury is an integral component of dental amalgam used as a restorative material in the treatment of dental caries. Both the American Dental Association and the U.S. Public Health Service have concluded that other than rare allergic reactions, mercury from dental amalgam restorations should not be considered harmful to human health when used in the prescribed manner (Langan et al. 1987). However, Mutter (2011) concluded that dental amalgam is by far the main source of human total mercury body burden. This was proven by autopsy studies that found 2-12 times more mercury in the body tissues of individuals with dental amalgam. These autopsy studies have shown consistently that many individuals with amalgam have toxic levels of mercury in their brains or kidneys.

21.8.2 Tobacco

Investigators have specifically linked cigarette smoking with periodontal disease in adults (Johnson and Slach 2001). Maternal but not paternal smoking is a significant risk factor for predicting caries in young children (Williams et al. 2000).

21.8.3 Polyhalogenated Aromatic Hydrocarbons

The individual congeners of this group of toxins vary substantially in their toxicity depending on the number and position of the chlorine groups. Oral hard and soft tissues may be susceptible to the adverse effects of dioxins, although epidemiologic studies on accidental exposure to dioxins and related compounds have not produced consistent findings. Overall, they seem to suggest that both prenatal and postnatal exposure to these compounds may cause oral soft tissue abnormalities and mineralization defects in children's teeth, such as an increased prevalence of natal teeth, gingival hypertrophy, intraoral hyperpigmentation, tooth chipping, and dental caries (Rogan et al. 1988). Kiukkonen et al. (2002) found that both the mesenchymal and, to a lesser extent, epithelial elements of the forming tooth were affected dose-dependently at relatively high doses of 2,3,7,8-tetrachlorodibenzo-p-dioxin. It induced color defects, pulpal perforation, and arrest of dentin formation.

21.8.4 Fluoride

Fluoride was introduced to dentistry over 70 years ago and is now recognized as the main factor responsible for the dramatic decline in caries prevalence that has been observed worldwide. However, excessive fluoride intake during the period of tooth development can cause dental fluorosis (Buzalaf et al. 2011). The white opaque appearance of fluorosed enamel is caused by hypomineralized enamel subsurface. With more severe dental fluorosis, pitting and a loss of the enamel surface occurs, leading to secondary brown staining. Dental fluorosis is a dose-response condition (Denbesten and Li 2011). In general, fluoride intake during critical periods of tooth development and maturation from approximately birth to 8 years of age, in the range of 0.03-0.1 mg F/kg body weight per day, can cause dental fluorosis (Grobler et al. 1986).

21.8.5 Tetracyclines

It has been known for many years that tetracycline has the potential to cause discoloration of teeth (Kutscher et al. 1966) that is both time- and dosedependent (Grossman et al. 1971). Tetracycline ingestion during periods of enamel calcification either by the mother during gestation (Genot et al. 1970) or by the child up to approximately age 5 (Harcourt et al. 1962) will result in discoloration of the enamel ranging from yellow to gray-brown. Chronic ingestion of tetracyclines may cause tooth discoloration, but the antimicrobial effects prevent dental caries, as evidenced by earlier observations in children with cystic fibrosis (Billings et al. 2004).

21.8.6 Other Drugs

There have been many reports over the past several years of prenatal and postnatal administration of anticonvulsants and chemotherapeutic drugs that have an adverse effect on teeth and oral tissues (Alpaslan et al. 1999). The severity of dentofacial-developmental and tooth-related abnormalities secondary to chemotherapeutic therapy is related to the age of the child, dosage, and duration of treatment. Dental abnormalities include tooth agenesis, arrested root development, microdontia, and enamel disturbances. Salivary flow rate decreased significantly after radiation therapy for head and neck cancer that was aggravated by the increase in irradiation dosage, which carried an increased risk for dental caries (Sun et al. 2010).

Drugs that are used for the treatment of asthma, including inhaler medications such as corticosteroids and β 2 agonists, have not been shown to have adverse effects on tooth development. However, their association with an increased susceptibility or resistance to dental caries has been equivocal, with some studies reporting an increased susceptibility to caries with the use of antiasthma medications (Milano 1999) and others reporting no increased susceptibility (Shulman et al. 2001).

Conclusions

Research involving the use of saliva sampling, for drug analysis, as noninvasive qualitative and quantitative techniques has become increasingly important. Saliva offers distinctive advantages over serum because it can be collected noninvasively by individuals with modest training. Saliva may be used in drug analysis and in therapeutic and toxicological monitoring. Although highly sensitive methods of detection are required, most drugs can be detected in salivary secretions.

Many chemicals and drugs can adversely affect human teeth, including lead, fluoride, and tetracycline, or become absorbed from teeth to cause systemic toxic effects, such as mercury in amalgam.

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Forensic Odontology Report

Contents

22.1	Introduction	201
22.2	Identification Cases	201
22.2.1	Basic Information	201
22.2.2	Before Postmortem Examination	202
22.2.3	The Postmortem Examination	202
22.2.4	Investigation of Antemortem Material	202
22.2.5	Comparison of Antemortem	
	and Postmortem Records	202
22.2.6	Conclusions	202
22.3	Dental Age Estimation Cases	203
22.3 22.4	Dental Age Estimation Cases Bite Mark Cases	
	0	203
22.4	Bite Mark Cases	203 203
22.4 22.4.1	Bite Mark Cases Evidence from Victim	203 203
22.4 22.4.1 22.4.2	Bite Mark Cases Evidence from Victim Evidence from Suspect	203 203 203

22.1 Introduction

Writing an ideal forensic odontology report is as much as art as it is a science. This chapter focuses on writing an exemplary legal report.

22.2 Identification Cases

The steps outlined in this section should be followed when working on an identification case (American Board of Forensic Odontology 2011).

22.2.1 Basic Information

The report should include at a minimum the following elements:

- 1. Requirements of report.
- 2. The forensic investigation should explain orodental findings in details that can be used for identification (age, sex, ethnic group, and occupation) (if possible).
- 3. The report must explain if the death could be due to trauma or any family or other violence that led to any injuries or trauma to the teeth, jaws, and any other oral and maxillofacial part. The possible causes should be given, too, if possible.
- 4. The forensic odontology identification report should be understandable to a general audience (i.e., it should be written using layperson's language).

22.2.2 Before Postmortem Examination

The following parameters should be taken in account before the postmortem examination:

- 1. The date and by whom you were given direction to do investigation
- 2. The date when and condition and position in which the body was found
- 3. Possible cause of death according to police and family members or others
- 4. Date and location of mortuary where autopsy was performed
- 5. Possible cause of death and main findings (according to the forensic pathologist or chief forensic investigator)
- 6. Date and place of the odontology examination
- 7. Name of the forensic investigators, other forensic experts, and police officers
- 8. Type of material for forensic odontology investigations

22.2.3 The Postmortem Examination

The report should include the following information about the postmortem exam:

- 1. Materials and condition of materials provided for forensic odontology investigations
- 2. Injuries, trauma, or deformity to the teeth, oral and maxillofacial regions
- 3. Details describing how material for forensic odontology examination was assessed
- 4. Descriptions of the types of the material
- Descriptions of each tooth (the clinical profile, such as intact tooth, carious, filled, type of filling crown, root, periodontal condition, calculus, staining, and other findings)
- 6. Details about prosthetic, orthodontic, or other types of appliances
- 7. Radiographs and photographs (with and without ABFO No. 2 scale) taken and characteristics shown
- 8. Other specialized findings

9. An estimate of the age, determination of sex, assessment of ethnic group, and the methods used (with references)

22.2.4 Investigation of Antemortem Material

The report should include a detailed description of material available for forensic odontology investigations, including

- 1. The quality of the material
- 2. Demographical information, such as full name, age, sex, date of birth, address
- 3. The details of when the person was reported missing and dental information
- 4. Information about the dentist or oral physician (name, address, telephone, e-mail) from whom the clinical records were obtained
- 5. Details for each tooth (the clinical profile, such as intact tooth, carious, filled, type of filling crown, root, periodontal condition, calculus, staining, and other findings)
- 6. Details about prosthetic, orthodontic, or other types of appliances
- 7. Radiographs and photographs (taken by family or dentists) and characteristics shown
- 8. Other specialized findings
- 9. Age at time of disappearance

22.2.5 Comparison of Antemortem and Postmortem Records

When comparing the ante- and postmortem records,

- 1. Look for nonconcordant findings.
- 2. Find compatible features.

22.2.6 Conclusions

The comparison should end in a conclusion about the probability of identity. Three forensic odontologists should agree upon the conclusion and sign the report.

22.3 Dental Age Estimation Cases (American Board of Forensic Odontology 2011)

The forensic odontologist should include the following information in reports of dental age estimation cases.

- Background information: describe demographical data, such as sex, family status, any diseases, any disorders and trauma, any dental problems, oral hygiene, and nutritional status.
- 2. *Clinical examination:* Describe the clinical exam in both living and dead individuals as detailed in Sect. 22.2.3, "The Postmortem Examination."
- Radiographic examination: A detailed description should be given regarding any radiographic examination.
- 4. *Methods of dental age estimation:* When estimating the dental age, the forensic odontologist should:
 - (a) Apply the maximum suitable techniques using the maximum number of teeth possible.
 - (b) Apply methods as originally described in the scientific journals and refer to the methods used.
 - (c) Calculate the inter- and intraobserver analysisfor each applied method if possible.
- 5. *Estimation:* When completing the dental age estimation report,
 - (a) Evaluate if the methods are appropriate in relation to the particular cases.
 - (b) Study all factors, like nutrition, ethnic group, etc., and pathological factors that may have influenced tooth development.
 - (c) Prepare an evaluation of the most appropriate chronological age.

22.4 Bite Mark Cases (American Board of Forensic Odontology 2011)

This section describes the steps to follow in assessing bite mark cases.

22.4.1 Evidence from Victim

- Document (more subjective part) how it happened, take informed consent for examination, take a saliva sample from victim as well as a medical history.
- 2. Describe the bite mark, age of mark, whether it is a human or animal bite mark, site, take photographs with and without ABFO No. 2 scale, and take an impression (for making model).
- 3. In case of deceased person, remove the bite mark sites.

22.4.2 Evidence from Suspect

- 1. Document (more subjective part) and take informed consent for examination, take salivary sample, and take medical history.
- 2. Describe the bite marks, age, human or animal bite marks, sites, take photographs with and without ABFO No. 2. Make a bite registration and take impression (for making model).
- 3. The bite mark comparison will be made pursuant to standards.
- 4. Finally, make a conclusion.
- 5. Three forensic odontologists should agree upon the conclusion and sign the report.

22.5 Basic Components of Forensic Odontology Report

The forensic odontology report should contain the following parts:

 General: A forensic odontology report is legal evidences and thus must be written according to legal guidelines and the questions it was intended to answer. It should be on legal paper and include the institution, name, qualification(s), address, telephone, e-mail address of the forensic odontologist, the date, an answer to the person or institution who made the request, the reference number(s), and a detailed description of the material examined.

- 2. *Methods used for assessment*: Describe the methods used with references cited.
- 3. *Results*: Describe positive and negative findings, answer the questions, and provide results of supplementary investigations.
- 4. *Discussion*: The discussion section should include details on the expert's interpretation of the findings and results.
- 5. *Conclusions*: The conclusions should be clear and contain no confusing or ambiguous statements.

6. *Signature block*: Full name, date of the report, signature, and professional affiliation.

Reference

American Board of Forensic Odontology (2011) ABFO manual. http://www.abfo.org/abfo_manual.htm. Accessed 20 Mar 2011

Index

A

ABFO, 13, 14, 20-22, 29-30, 88, 93, 94, 101-103, 105, 112, 135, 137-139, 182, 185, 187, 202, 203 ABFO no.2 scale, 20, 21, 93, 94, 102, 103, 105, 115, 135, 202, 203 Abrasion, 23, 90, 94, 178, 179 Abuse, 1, 4, 13, 15, 29, 87, 90, 132, 175-183, 186, 189, 193-194 Accidents, 2, 3, 5, 35, 66, 109, 127, 154 Adobe photoshop, 18, 20, 23, 55, 101-107, 145 Age estimation, 1, 20, 25-28, 35-61, 91, 123, 171,203 Alternate light, 131, 132, 137 Antemortem, 3-5, 9, 11-16, 18-20, 23-25, 35, 117, 122-124, 145, 158, 160, 163, 202 Anthropology, 1, 31, 110 Aperture, 66-68, 73, 133-135, 137, 139 Autopsy, 125, 158, 195, 202

B

Biochemical, 26, 27, 57–58, 137, 187, 190 Bioterrorism, 149–152, 154, 155 Bite Marks, 1, 2, 4, 6, 13, 19, 31, 87–98, 101–107, 131, 132, 137–139, 142, 145, 158, 165, 179, 180, 182, 186, 203 Bite wing, 18, 22, 120–121, 157 BR, 5, 52, 74–75 Burn, 11, 136, 171, 172, 178, 179

С

Camera, 20, 66, 93, 94, 102, 117, 132–139 CBCT. *See* Cone beam computed tomography (CBCT) Charged-coupled devices (CCD), 13, 135 Charting, 12, 14–20, 22, 23, 92, 157, 158, 160 Child abuse, 4, 175–183, 186 Child neglect, 177 Clinical records, 15, 202 Computer software, 145 Cone beam computed tomography (CBCT), 18, 119, 122, 142–143

D

Deciduous, 13, 23, 25, 28, 36, 37, 74, 81, 84–85, 123
Decomposition, 6, 20, 38, 79, 88, 115, 118, 158, 163, 164, 172
Dental age, 25–28, 35–61, 143, 203
Dental evidence, 1, 2, 12, 13, 20, 87, 146, 185, 186
Dental identification, 1–4, 6, 9–11, 13, 14, 19, 25, 30, 87, 132, 146, 185, 188
Dental Jurisprudence, 185–188
Digital, 3, 13, 18, 20, 23, 70, 93, 102–107, 131–136, 138, 139, 145, 146
DNA, 4–6, 30–31, 68, 73, 78, 92, 93, 96–98, 109, 115, 143, 144, 156, 157, 160, 163–166, 187
Dog bites, 4, 90, 186
Drug, 15, 16, 131, 151, 172, 178, 189–196

Е

- Emotional, 10, 16, 175, 177, 178, 180-181
- Equipment, 45, 94, 117, 121, 128, 134–136, 138, 139, 156, 160 Ethic, 18, 58, 185–188
- Expert witness, 187
- Extraoral incisions, 20, 159

F

- Facial reconstruction, 4–5, 31, 65, 67–70
- Fatality, 6
- Film, 13, 18, 20, 120, 121, 124, 125, 131, 133–135, 137, 146, 157

Fingerprinting, 6, 112, 156, 160

Forensic odontologist, 1–4, 6, 11–15, 18–20, 22, 25, 31, 36, 44, 68, 79, 81, 91, 92, 97, 102, 105, 107, 119, 122, 124, 131, 134, 136, 143, 145, 151, 152, 154–156, 160, 163–166, 175–183, 185–188, 202, 203

G

- Genetic, 5, 30, 31, 58, 76, 118
- Guidelines, 58, 112, 134, 164, 185, 187, 189, 203

Н

Habits, 9, 25, 29–30, 38, 113, 158, 181 Hand held, 141 Histological, 26, 36, 55, 170 Homicide, 10, 87, 110, 176, 182 Human identification, 6, 109, 115, 123, 132, 157, 163, 166

I

Imaging, 90, 97, 119, 131, 132, 135, 142
Infrared photography, 94, 138
Injury, 2, 88, 90, 92–94, 97, 98, 101, 124, 132, 133, 136, 138, 139, 153, 175, 177–179, 181, 182, 186
Investigations, 10, 15, 18, 19, 28, 59, 65, 68, 91–93, 101, 109, 129, 131, 137, 139, 141, 142, 144, 145, 156, 157, 163, 166, 172, 182, 201, 202, 204

J

Jaw resection, 159 JBR, 1, 55, 97–98 Jurisprudence, 18, 185–188

K

Katrina, 122

L

Lateral skull, 18 Legal, 1, 3, 5, 12, 15, 18, 37, 58, 59, 92, 101, 102, 131, 132, 154, 156, 169, 181, 185–187, 201, 203 Living individual, 58, 66, 88

М

Malpractice, 1, 12, 15, 16, 18, 185 Mass disaster, 2, 3, 73, 120, 141, 145, 152–160, 163 Medicolegal, 1–3, 6, 9, 10, 18, 31, 37, 65, 141, 145, 146, 154, 169, 185, 186

Ν

Neglect, 1, 177, 178, 181 NOMAD, 20, 122, 141–142

0

Occupation, 25, 29, 113, 123, 127, 201

Р

PCR. See Polymerase chain reaction (PCR)
Personal effects, 19
Photographic distortion, 94
Photography, 13, 20, 22, 93, 94, 112, 117, 131–139, 142, 158, 181, 182
Physical abuse, 176–182
Polymerase chain reaction (PCR), 78, 164–166
Possible identification, 3, 25

Postmortem, 2–4, 9, 11–13, 15, 17, 19–31, 35, 36, 68, 115, 117, 122, 124, 145, 146, 156–158, 160, 163, 164, 170, 171, 202, 203 Preparedness, 150

Q

Quality, 13, 27-28, 92-94, 135, 156, 182, 187, 202

R

Radiography, 13, 20, 36, 119–121, 124, 142, 156, 158, 160 Record, 2, 3, 6, 9, 11–25, 59, 66, 68, 88, 93–95, 112, 117, 120, 123, 124, 127, 131–133, 146, 154, 157–160, 163, 181, 183, 185–187, 202 Restorative materials, 96, 195

S

Saliva, 6, 31, 92–93, 95, 96, 137, 165, 186, 189–196 Salivary DNA, 92, 165 SEM, 144 Sex determination, 1, 5, 73–79, 113, 118, 123, 166 Sexual abuse, 87, 177–180 Skeletonized, 2, 19, 22, 119, 124, 158, 163 Software, 20, 55, 68, 69, 105, 117, 145 Suspects, 5, 70, 90, 92, 94–98, 101, 105, 112, 142, 181, 187, 192, 203

Т

Tattoos, 66, 139, 157 Three dimensional (3D), 68–70, 93, 102, 117, 122, 142 Trauma, 3, 19, 25, 96, 152, 163, 172, 175, 177–180, 201–203

U

Ultraviolet photography, 94 Unidentified persons, 25 Uniqueness, 11, 88, 91, 92, 115, 150

V

Verbally, 176 Victims, 2–4, 6, 10, 11, 15, 18, 22, 31, 35, 73, 87, 88, 90–92, 95, 98, 102, 118, 119, 122, 124, 142, 143, 150, 155, 156, 158, 160, 163, 176, 178, 181, 203 Video, 20, 65, 66, 138

W

WinID, 15, 20, 23, 145 Wounds, 4, 90, 91, 97, 131, 137, 143, 154, 179, 190

Х

X ray, 12, 13, 18–20, 22, 35, 38, 40, 59, 119–122, 124–125, 141–145, 157, 178