

**A Forensic Study of Unnatural Deaths in Kuwait:
Epidemiological, Virtual Autopsy and DNA
Investigations**

by

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Declaration

I declare that while registered as a candidate for the degree for which this submission is made that I have not been a registered candidate for another award by any other awarding body. No material contained in this thesis has been used in any other submission for an academic award.

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Type of Award: (PhD)

School: School of Forensic and Investigative Sciences

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Abstract

Forensic science is growing rapidly in the world today. During the past decade, medico-legal investigations have been highly expanded to include all areas of forensic science. The present study investigated three important aspects of forensic biology. First, this present project investigated, a total number of 5,703 reported medico-legal cases diagnosed as un-natural deaths by The Forensic Department in Kuwait, during the year 2003-2009. The results show that accidental, homicidal and suicidal deaths accounted for 86%, 8% and 6%, respectively. The results showed that most people who died of unnatural deaths were more predominant in the age group 20-29 years (third decade). Road Traffic Accidents accounted for 65% of accidental deaths, and 4% out of them were related to alcohol consumption. The results also illustrated that the highest rate of homicide in Kuwait was due to stab wound injuries (38%) compared to the lower rate of homicidal pattern for infanticides (3%). Similarly, the study showed that the most common method of suicide in Kuwait was death by hanging and this accounted for (60%). This study further demonstrated the effectiveness of virtual autopsy technique as a new tool in forensic investigations to determine various un-natural death causes. A total of thirty (30) male forensic cadavers were employed in this project. The cases were RTA (11), firearm injuries (10), drowning (4), head injuries (3) and lastly strangulation (2). All these cases were compared to the findings of traditional autopsy. The results show similar findings for virtopsy compared to traditional autopsy. This study clearly revealed that virtopsy could be an effective alternative in certain situation, being noninvasive and rapid. The present project also investigated 28 samples of human blood, saliva or semen. The experiments were done at four different temperatures (55°C, 37°C, 24°C and 4°C) and four different humidity ranges (41%, 55%, 58% and 61%), respectively. The results showed that, DNA quantity in blood, saliva and semen samples remained more or less the same at temperatures of 4°C, 24°C and 37°C compared to values for day one with all other days. In contrast, when the temperature was raised to 55 °C, the DNA started to degrade with time until it reached zero at day 12 for saliva and day 15 for blood, but not for semen. The results clearly show that DNA in saliva and blood samples is extremely sensitive to heat compared to semen. In conclusion, the study reveals the different causes of unnatural deaths, the value of virtual autopsy and the need for early DNA measurement in Kuwait.

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Nadiah AL-Kandari

DEDICATION

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Abbreviations

| | |
|-------|---|
| ANOVA | : Analysis of Variance |
| B.C | : Before Crisis |
| CDCP | : Center for Disease Control and Prevention |
| CIA | : Central Intelligence Agency |
| CODIS | : Combined DNA Index System |
| CR | : Crown-rump |
| CSO | : Central Statistics Organization |
| CT | : Computerized Tomography |
| DNA | : Deoxyribonucleic Nucleic Acid |
| DTT | : Dithiothretol |
| FBI | : Federal Bureau of Investigation |
| FIT | : Forensic Investigation Team |
| FSS | : Forensic Science Service |
| FMD | : Forensic Medicine Department |
| GDCE | : General Department of Criminal Evidence |
| IMR | : Infant Mortality Rate |
| KIDL | : Kuwait Identification DNA Laboratory |
| KW | : Kuwait |
| NCHS | : National Center of Health Statistics |
| MD | : Medical Degree |
| MSCT | : Multislice Computed Tomography |
| MOI | : Ministry of Interior |
| MOSF | : Multi Organ System Failure |
| MRI | : Magnetic Resonance Imaging |
| NKW | : Non Kuwaiti |
| NLR | : National Literacy Rate |
| NMR | : Nuclear Magnetic Resonance |
| NVDRS | : National Violent Death Reporting System |
| PCR | : Polymerase Chain Reaction |
| PK | : Proteinase K |
| RDS | : Respiratory Distress Syndrome |
| Rh | : Rhesus factor |
| RTA | : Road Traffic Accidents |
| Sec | : Second |
| SIDS | : Sudden Infant Death Syndrome |
| SGM | : second Generation multiplex |

STRs : Short tandem repeats
SUD : Sudden Unexpected Death
UAE : United Arab Emirates
USAD : United State American Delivery
UNICEF : United Nation Children Fund organization
VNTRs : Variable number tandem repeats
WHO : World Health Organization

CHAPTER 1

INTRODUCTION

1.1 Introduction to Research

Death may occur naturally, un-naturally, or as the result of a combination of both originating from oneself or another person. In medico-legal aspects, the responsibility of the Forensic Pathologist is to utilize a comprehensive investigative study of the circumstances of the cause leading to and surrounding death (Kuruc et al., 2009). Death is considered un-natural when it is caused prematurely against the order of nature by injury due to either trauma, physical or chemical agents, or other means of violence (Serafettin et al., 2009). Many cultural and socio-economic factors of a country are usually related to the causation of un-natural deaths (Palmiere et al., 2010). Un-natural deaths happen almost everywhere in the world. Data on un-natural deaths in a particular geographical area can give an indication of the nature of such communities, and the health standards and legislation policies (Arun et al., 2010).

Un-natural deaths are categorized into three groups as either accidental, homicidal or suicidal (Sjögren et al., 2000). Death may also occur in situations where loss of life has occurred on a larger scale and many individuals have died, such as during military action or natural disasters (Kuruc et al., 2009). Traditionally, either autopsies or dissections of deceased individuals are performed by Medical Examiners and Forensic Pathologists. These professionals take samples from target organs and look for signs of trauma and stress in the soft and hard tissues of the human body. A report is then generated by Forensic Scientists as to overall condition of the individual and whether any signs of trauma, physically or chemically induced, self-

inflicted or in what every way it might have administered are apparent (Palmiere et al., 2010). These reports are then submitted to a branch of law enforcement for potential cases in culpability.

In Kuwait, medico-legal cases are transferred to the (FMD) to investigate the cause of death, which can be established via a variety of investigations including examination of the crime scene, autopsy and chemical analytical tests. Investigating these cases is the responsibility of the Police and Forensic Scientists (Al-Kandari, 2007; Al-Kandari, 2008). However, with each case, vital statistics are amassed by the FMD, including the identity of the dead person, the manner of death, the ethnicity, the socio-economic status, nationality and other demographic data. The collection and dissemination of such information becomes vital to any preventive measures which the Government may want to undertake. One of the aims of this study was to explore the effectiveness of the (FMD) in Kuwait as it currently exists.

1.2 The Skewed Records of Death

Systems for determining the causes of death differ from country to country and often reflect the general level of prosperity and medical and health care in the country (Khandekar et al., 2008). In many countries, there is no autopsy activity and information about the causes of deaths is based purely on the clinical history or other information about the circumstances surrounding the death (Yen et al., 2005; Brådvik et al., 2009). The frequency of autopsy is quite variable in different countries and sometimes, it is difficult to obtain information about autopsies. In many countries in Asia, certification of death is based on the external examination of corpses (Palmiere et al., 2010). This is

often the reason for inaccuracies in the statistics on the causes of deaths. A medico-legal system for determining the causes of deaths exists in most countries of the world, where the police are duty-bound to investigate certain kinds of death (O'Grady, 2010). The cases reported to the police are very similar in many countries and they include violent deaths—homicides, suicides, accidents, occupational accidents, malpractice, deaths in custody and in jails and sudden deaths. They also involve those cases in which a medical examiner cannot establish the cause of death by external examination of the body or by the history of diseases. In addition, it includes cases in which the doctor has not met his or her patient during a certain period of time before the death. In this circumstance, a decision about an autopsy is normally taken.

In China, medico-legal examination has to be done in cases where there is a suspicion of a dangerous contagious disease (Palmiere et al., 2010). Decisions about autopsy are taken by different authorities in different countries. For instance, in Finland and in Denmark the police order autopsies, while in Belgium and France, the decision is made by the law court and in the State of Virginia in US this is made by coroners (O'Grady, 2010). While in Panama, with approximately 2.4 million inhabitants, the medico-legal requirements are met by appointed Physicians (Bertolote, 2005). The record of death is skewed globally because nations (i.e. national Government and their subsidiaries, etc.) devote different resources according to the rates of unnatural deaths and the socio-political priorities of national budgets. The lack of trained professionals and under-funded allotments to branches of Government and educational institutions in forensic science as a field of endeavor are also another constraint (Sauvageau and Boghossian, 2011).

1.3 Kuwait, the Study Area

According to the Central Statistics Organization (CSO), the midyear population of Kuwait as of July 2011 was estimated to be 2,868,000 people. Of these numbers, 975,000 residents were Kuwaitis and 1,893,000 residents were non-Kuwaitis (Health Kuwait, 2010). The population of Kuwait is a mixture of Arabs, Persians, Asians, and Westerners. Kuwaitis represent 35% of the population, while Arabs constitute 22%, South Asians 30%, Iranians 4%, and other nationalities 9%. Kuwaiti citizens are represented to their maximum concentration in the Capital Governorate (Kuwait City), and Non-Kuwaitis in Farwania Governorate. The lowest concentration of Kuwaitis can be found in the Jahra Governorate, and of Non-Kuwaitis in the Mubarak Al-Kabeer Governorate. The ratio of males to females in the total population is 1.7:1 (CIA World Fact Book, 2010). Kuwait is divided into six main Governorates. Farwania Governorate, with a population of 766,425 residents, is the most heavily populated region; Mubarak Al-Kabeer, with 186,254 inhabitants, is the least populated (CIA World Fact Book, 2010).

Arabic is the official language, but English is widely spoken. Kuwait is a highly developed welfare state with a free market economy and is one of the wealthiest countries in the world. Education is free and compulsory for all Kuwaiti Citizens. Health services are organized into primary, secondary, and tertiary health care, including high technology medicine. Health services are provided free of charge to all Kuwaiti citizens and there are nine to ten health centers in each Governorate. Each health center serves the population according to their catchments area and with the goal of making geographical access to health services equitable (Health Kuwait, 2010).

Kuwait is at the crossroads of Asia. Kuwait is situated Northeast of Saudi Arabia at the Northern end of the Arabian Gulf and South of Iraq. It comprises a total area of 6,880 square miles (17,819 square kilometers) and it is mainly a low-lying area dominated by barren desert (CIA World Fact Book, 2010). Kuwait, like other Arab Gulf States, attracts large populations of various ethnic groups who live and work as expatriates. These immigrants do not usually mix with the indigenous local population. They retain a diversity of their native socio-cultural practices in Kuwait. Figure 1.1 and table 1.1 show a map of the State of Kuwait and the population of Kuwait, respectively.

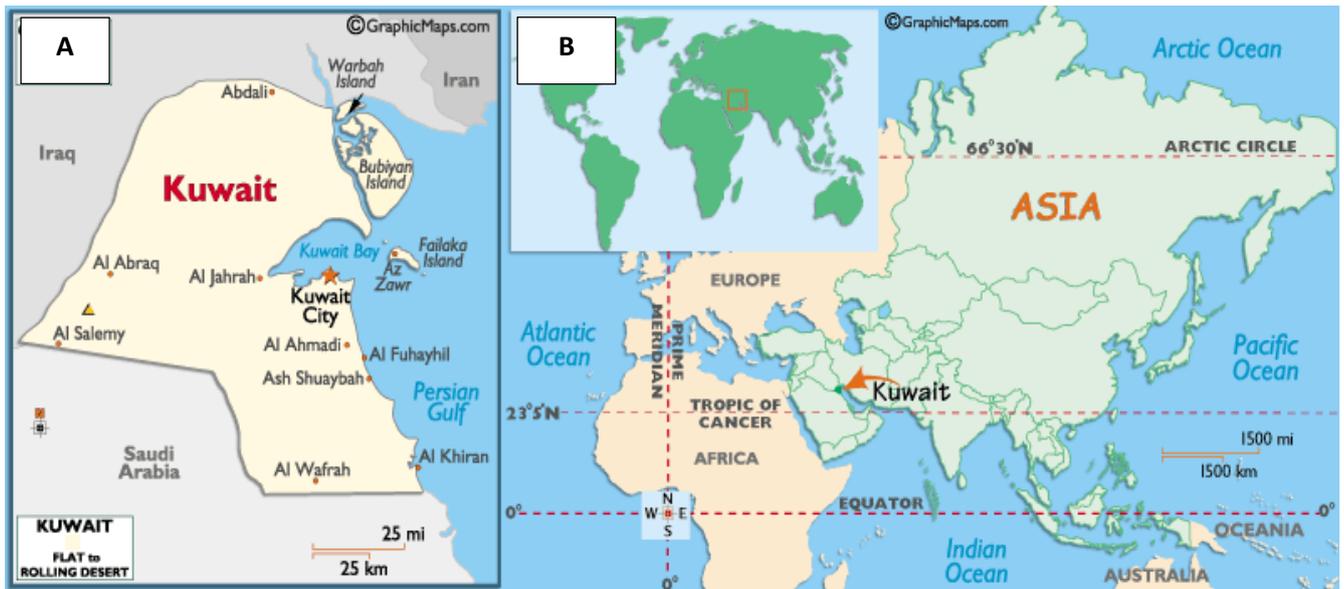


Figure 1.1: Map showing the State of Kuwait (A) and its relation to other countries in the Middle East (B)

Table 1.1: Table showing the Kuwait population in the middle of February 2011 according to Governorates and the area of each Governorate (source; Kuwait Central Statistical Office, 2011). Data will be discussed in chapter 2, Epidemiology of unnatural deaths related to governorates.

| Kuwait Governorates | A-L (KM ²) | Population |
|---------------------|------------------------|-------------------|
| Al-Ahmadi | 5,120 | 480,927 |
| Al-Farwaniyah | 190 | 740,935 |
| Hawalli | 84 | 562,127 |
| Al-Asimah- Capital | 200 | 354,503 |
| Mubarak Al-Kabeer | 94 | 275,244 |
| Jahra | 11,230 | 422,915 |
| Total | 17,818 | 2, 836 644 |

1.3.1 An overview of the General Department of Criminal Evidence (GDCE) in Kuwait

GDCE is considered one of the most specialized Departments in the Ministry of Interior in Kuwait. Employing a variety of highly trained scientists and professionals, this department processes cases that come from various investigative authorities such as Public Prosecutors, Hospitals, Investigation Departments and Courts (Al-Kandari, 2008). Reports issued by the GDCE are significant to the investigative process and the final outcomes of legal rulings in Kuwait. The role of the GDCE not only extends to the legal field, but it also renders services such as retrieval of certain vital data required for both the Government and the private sector jobs.

1.3.2 Historical view of the GDCE

The GDCE has gone through different stages in its 50 years or more long history since it was first established in 1954. This development is examined over six historical periods as follows: Period One (1954-1960); Period Two (1961-1973); Period Three (1974-1984); Period Four (1985-1990); Period Five (1990 Iraqi Invasion); and Period Six (Post Iraqi-invasion). These periods are distinguishable because of the Kuwaiti Government's desire to establish and structure effective branches within itself. Thus, the issuance of legislation governing the practices and goals of the Kuwait Ministries and the overall socio-cultural trends in unnatural deaths in Kuwait would refine the role of the GDCE (Al-Dousery, 1994).

The role of the GDCE, during Period one (1954-1960) was primarily to identify the populace and to assist with establishing a national identity.

Beginning in 1954, the Kuwait Government determined that it was necessary to establish a special unit equipped to deal with fingerprinting and personal identification (Al-Dousery, 1994). The goals of this unit were:

- To file fingerprinting cards of those resident government employees who previously had been convicted of crimes.
- To identify criminals and suspects based on their fingerprint classification.
- To collect and gather fingerprints found at scenes of crime.
- To identify unknown bodies by fingerprints.
- To issue good conduct certificates to applicants.

The Government of Kuwait established the Justice System on 19th December, 1959. It was the first legislation that organized and controlled the Judicial System. In 1960, new legislation governing criminal law and procedures was passed and approved. In 1961, a ministerial decree established the Forensic Medicine Department in Kuwait (Al-Dousery, 1994). Thus, in Period Two, (1961- 1973), the GDCE began to take its shape as we know it to exist today, a support for the court system and the entity responsible for the medic-legal aspect of criminal investigations.

Period three (1974-1984) could be defined as the national Government's desire to streamline its branches. At the beginning of 1974, the organizers in the Ministry of Interior decided that it was necessary to arrange the structure of the Ministry in a more scientific manner, and that it should be more aligned with the functions and duties of police departments. Therefore, those Departments with similar duties were unified. Four Departments were

formed, including the Fingerprints and Identification Department, the Forensic Science Laboratory, the Forensic Medical and Personal Identification Departments (Al-Dousery, 1994).

By 1984, the Criminal Laboratory and Forensic Medicine Department (FMD) and the Crime Scene Officers Department were combined to form what was later called the Central Department of Criminal Laboratory and Forensic Medicine. Period four (1984-1990) also marked by another streamlining of departmental functions was accompanied by the addition of a pathology unit. In 1985, a pathology section was added to this Department and it was subsequently renamed as the General Department of Criminal Evidence (GDCE). It consisted of the following Departments:

1. Central Department of Personnel Identification.
2. Central Department of Forensic Science Laboratories and Forensic Medicine.
3. Central Department of Crime Scene Officers.

Period five (Iraqi invasion), began on August 2, 1990 when Kuwait was invaded by 200,000 Iraqi troops. This invasion destroyed the entire structure of Forensic Departments in Kuwait. The present Period Six (post-invasion) is defined as a reconstruction period. After the liberation of Kuwait, the main goal of the Kuwaiti Government was to rebuild most important Departments in Kuwait (Al-Dousery, 1994). In 1997, The General Department of Criminal Evidence was reorganized to consist of the following:

- Criminal Laboratory Department

- Crime Scene Officers Department
- Forensic Medicine Department (FMD)

In 1999, The Support Services Department and the Operation Section were established. In March 2001, specialized clinics were set up within the Forensic Medicine Department (FMD), consisting of orthopedic, dental, ophthalmology, and ultrasound clinics. In September 2003, the Kuwaiti Government launched the Kuwait Identification DNA Laboratory (KIDL), the first of its kind in the Arabian Gulf Region and the Middle East. In October 2001, two sections were established that included:

- Research Development and Training Section
- Quality Control Section.

The Facial Reconstruction Unit was also launched in the Department of Forensic Medicine at this time.

1.3.3 Current structure of the organization

The current structure of the GDCE consists of the following departments;

- Department of Crime Scene
- Department of Forensic Science
- Department of Criminal Laboratories
- Department of Personal Identification and Automated Fingerprints
- Department of Forgery and Counterfeit Prevention
- Department of Support and Service Department
- Department of Quality Control

- Department of Operation
- Department of Research, Development, and Training.

As shown in figure 1.2, the following Departments, Sections and Divisions are part of the current organizational structure of the GDCE in Kuwait.

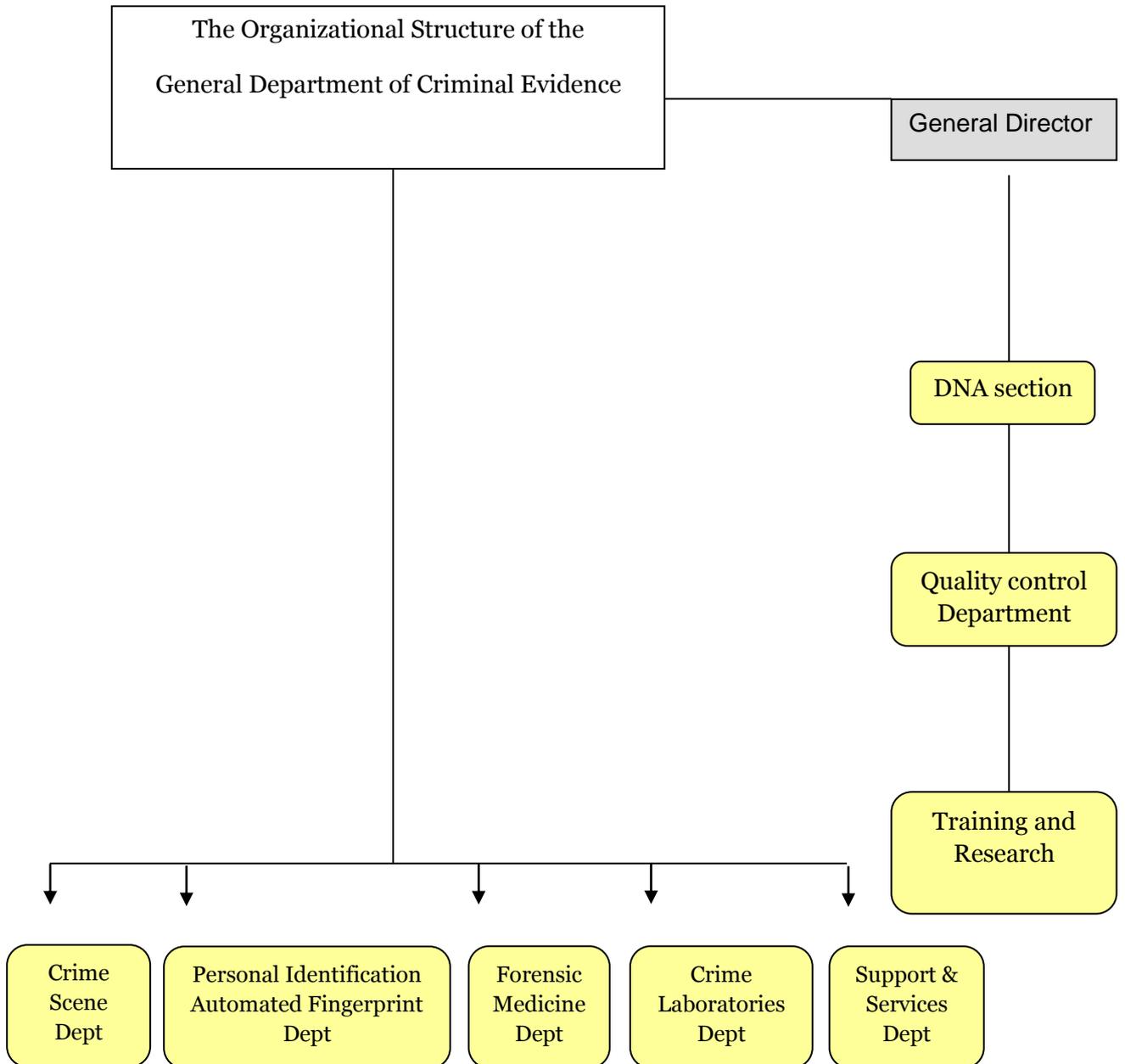


Figure 1.2: A flow diagram showing the organizational structure of the General Department of Criminal Evidence (GDCE).

Since this study is focused mainly on the Department of Forensic Medicine, an overview of the sub-divisions of this department will also be addressed. They include the Forensic Medicine Examination Section, Pathology Laboratory Section, the Morgue, the Legal Nurses section, blood extraction clinic, facial reconstruction unit, and X-Ray section (see figure 1.3).

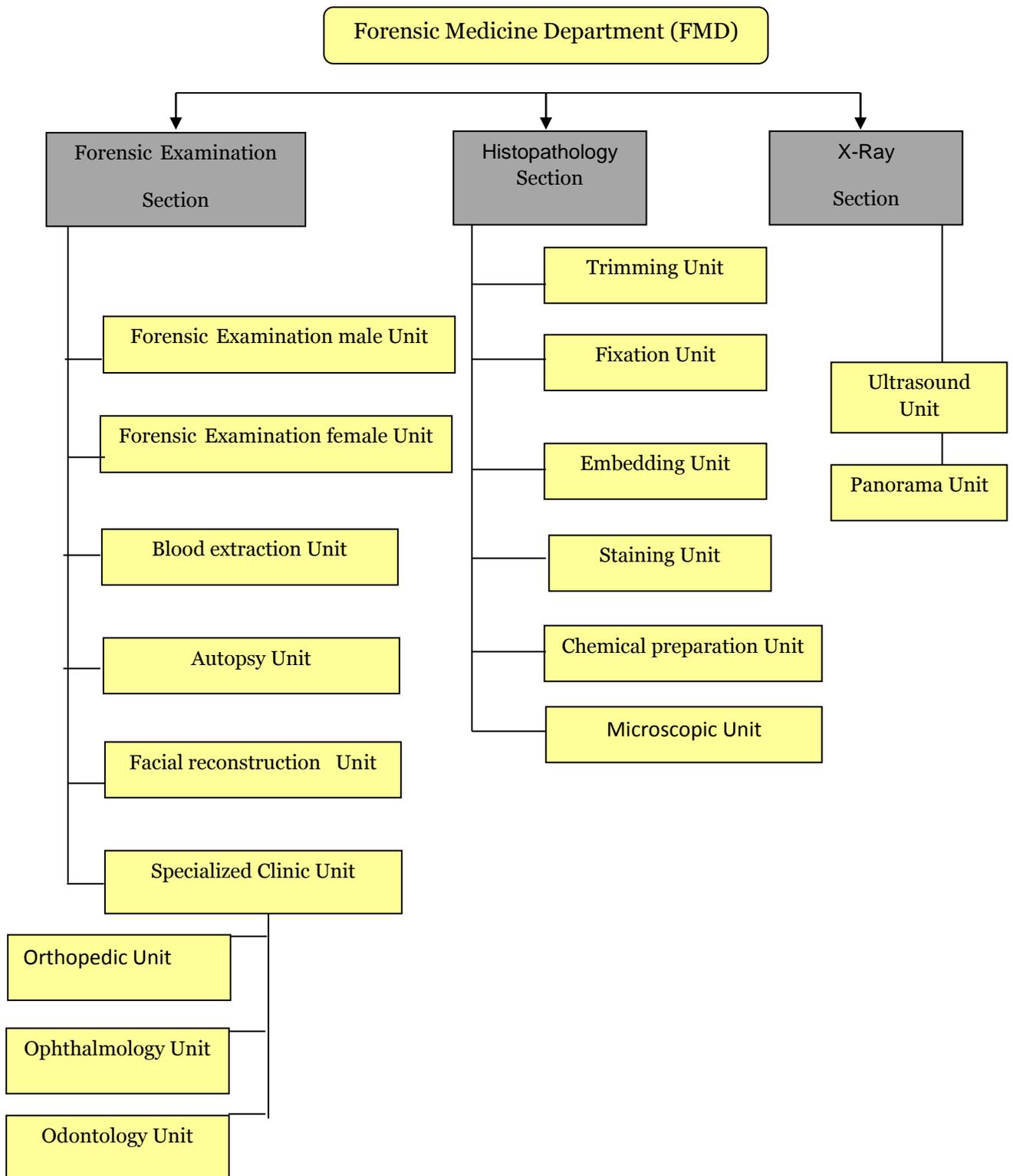


Figure 1.3: A flow diagram showing the current structural organization (sections and sub-sections) of the FMD in Kuwait

1.3.4 Personnel of the GDCE, Kuwaiti Laws and Forensic Practice

Currently, the GDCE employs a highly specialized, professional staff of 604 individuals. They are distributed as follows: 65 staff in the General Director office, 230 in the Department of Personal Identification, and 104 in the Forensic Science Laboratory, (of whom about two-thirds are Scientists), 97 in the Department of Forensic Medicine, and 108 staff in the Department of Crime Scene, of whom about two-thirds are police officers (AL-Kandari, 2007).

The FMD of GDCE in Kuwait receives death cases and living subjects from Federal Agencies (such as police stations) and health organizations (Hospitals, Clinics, etc.) representing medical examiners. With regards to death cases and as mentioned earlier, those organizations and others are obliged by law to investigate all Road Traffic Accidents (RTA), unexpected, unexplained and suspicious deaths of individuals, to ensure that they are due to natural causes and to exclude other explanations. This is done through several investigations involving the police. Parents are usually relieved that everything was done to discover the causes of death. At the time of the autopsy, family members are expected to answer all questions of interest to the best of their ability, so that the autopsy can be performed legally, efficiently, quickly and effectively. The Forensic Section in Kuwait reviews any cases of death that are referred to the department whether it was of natural or un-natural cause and the department is also responsible to investigate any disturbance of pregnancy and before spontaneous or medically-induced deliveries.

1.4 Medico-legal investigation of unnatural deaths in the Middle East

The most frequently cited reasons for lack of research commitment in the Middle East were time constraints, lack of academic institutional support for research in forensic pathology, and isolation from academic institutions. Other reasons were regulations covering confidentiality, poorly standardized data acquisition and information technology, and lack of Federal research support (Koronfel, 2002).

Little attention has been paid to unnatural deaths in the Middle East. Epidemiological and psychological cases of unnatural deaths are very scarce; it is a very sensitive issue, few studies have been found regarding suicide, and people are usually suspicious of questions and they do not understand the importance of statistics (Al-Kandari, 2008). In Kuwait, there have been very few studies concerning unnatural death cases. In 1982, a study was conducted for the first time to investigate suicide in Kuwait from 1978 to 1981 (Fido and Mughaiseeb, 1989). The study revealed that 96 individuals died from suicide within the 30-39 age group and constituted the majority of the deaths (35.5% of total unnatural deaths). The number per 100,000 for the total population was 3.1. The study also revealed that the majority of people who committed suicide were Arabs, although the majority of the population in Kuwait are foreign workers (Fido and Mughaiseeb, 1989).

In Syria, as in all other Muslim countries, the religion forbids suicide, but suicide and suicide attempts are not punishable by Syrian law. More suicidal acts were reported in large urban centres like Damascus than in rural provinces (Maziak et al., 2006). The differences were attributable not only to the concentration of people in the cities, but also to the lesser degree of

concealment in areas where religious and traditional forces were weaker. Poisons and pills as a method of suicide were used by 80.3 % of the total reported cases of suicide. Moreover, 66% of the suicide death cases were females and the largest number of suicidal acts took place before the age 25, particularly between 15 and 24 (Maziak et al., 2006).

According to official statistics in Egypt in the year 2007, the incidence of suicide in Egypt was 0.2 per 100,000, the lowest among the twenty six countries survived by the World Health Organization (WHO, 2008). The highest incidents for suicide were amongst the age group, 15-24 years and they constituted 60% of the whole. Moreover, professional people ranked the lowest with 6% with the most prevalent means by way of drug overdose (Okasha and Lotaif, 2007).

It is generally agreed that current statistics on the incidence of unnatural deaths concerning homicide, suicide and accidental deaths are grossly inadequate and that their comparisons based on the figures available are at best inaccurate and often misleading (Daradkeh, 1989; Koronfel, 2002).

1.4.1 Death by Suicide

According to WHO definition, suicide is the act of deliberately or intentionally taking one's own life (WHO, 2006). Suicide has become an increasing social challenge in today's stressful world. WHO report in 2010, indicates that about every 40 seconds, somebody dies by suicide (WHO, 2010). This is an alarming matter for the whole world to consider. Suicide is viewed in varying ways among different cultures, religions, spiritual traditions, legal, and social systems of the world. Elderly people (over 65 years) have a higher suicide

rate within the general population, and on an average, one old person killed himself or herself every one hour and 41.4 minutes (WHO, 2009). These statistics suggest that suicide is a major public health problem. Suicide is considered a sin or immoral act in many religions and as a social stigma in some cultures.

Suicides must be competently investigated by both law enforcement officers and the medical examiner to eliminate any issues relating to homicide and to solve the actual cause of death (Dogan et al., 2010). During an early investigation, an incorrect determination might be made, so the forensic pathologist plays a significant role in considering all of the evidence concerning death (Leenaars et al., 2010). According to WHO estimates for the year 2020 and based on current trends, approximately 1.5 million people will die every year from suicide, and 10 to 20 times more people will attempt suicide worldwide (WHO, 2010).

1.4.2 Suicidal Risk Factors

Suicide is the result of an interaction of risk and protective factors both external and internal to the individual (Allampalli et al., 2010; Kaslow et al., 2011). Several research studies have focused on identifying the risk and protective factors associated with suicide. While there is wide acknowledgment of risk factors associated with an individual, some risk factors may vary with age, gender, or ethnic group and may occur in combination or change over time (Hejna and Šafr, 2010). Sometimes people, families and others may underestimate the impact of the risk factors on suicidal behaviours (Mostaque and Das, 2009). Research has shown that suicides are associated with several factors including:

- Depression and other mental disorders or a substance-abuse disorder (family history of mental disorder or substance abuse). More than 90 percent of people who die by suicide have these risk factors.
- Prior suicide attempt and availability of suicide methods.
- Family history of suicide and family history of child maltreatment.
- Family violence, including physical or sexual abuse.
- Availability of firearms in the home.
- Incarceration.
- Exposure to the suicidal behavior of others, such as family members, peers, or media figures.
- Losses (financial, education, romance, etc...)
- War and political events
- Cultural and religious beliefs, in which suicide is believed to be a noble resolution to a personal dilemma.
- Isolation, a feeling of being cut off from other people

1.4.3 Epidemiology of Suicide

Incidences of suicide are under-reported due to both religious and social pressure and possibly completely unreported in some areas. Nevertheless, from reported cases, certain trends are apparent. Since the data are skewed, comparing suicide rates between nations is statistically unsound. Suicide rates vary widely by nation. The latest figures available from the World Health Organization (WHO, 2010) indicate that nearly a million people take their own lives every year, more than those murdered or killed in

wars. Suicide rates are highest in Europe, where around 40 people per 100,000 die by suicide each year, second in line is in the Sub-Saharan Africa where 32 people per 100,000 die from suicide each year (Levy et al., 2007). The lowest rates are found mainly in Latin America and a few countries in Asia. Research on socio-demographic factors suggests that important variables such as; gender, age, and racial differences exist in the risk for suicide. Membership in certain groups increases the risk for suicide (Hakko et al., 2007).

1.4.4 Gender and Suicide

One of the most commonly reported differences in suicide between males and females is the method of suicide which they select to kill themselves. Men tend to choose more violent methods in suicide attempts (Pritchard, 2007). Deliberate self-harm (DSH) is usually higher in males than females; thus, males are more likely to use lethal suicide methods, such as hanging, breathing vehicle's exhaust gas, asphyxiation and firearms (Hakko et al., 2007). Females, on the other hand, are more likely to choose self-poisoning and less harming procedures (Allampalli et al., 2010). In the United States, males are four times more likely to die by suicide than females (Brunel et al., 2010). WHO reported in 2006 that male suicide rates are higher than females in all age groups (the ratio varies from 3:1 to 10:1), while in other Western Countries, males are also much more likely to die by suicide than females (usually by a factor of 3–4:1) (WHO, 2006). On the contrary, an analysis of suicide was performed within the People's Republic of China and the results have found that the profile was the opposite to that reported in the

rest of the world, as more female subjects kill themselves than males, particularly younger women (Pritchard, 2007).

1.4.5 Age and Suicide

In terms of age, youth (15-24) and elderly persons (above 65) have been identified as two high suicide risk groups (O'Grady, 2010). It is noteworthy that although these two age groups have been identified as high suicide risk groups, the rates of completed suicide are higher for the elderly than for the youth. In 2004, of 100,000 people, 14.3 elderly individuals died by suicide versus 8.2 adolescents (aged 15 to 19) and 12.5 young adults (aged 20 to 24) (WHO, 2006). Among youth, alcohol use beginning in preteens, social and educational disadvantage, childhood and family adversity, psychopathology, individual and personal vulnerabilities, exposure to stressful life events, circumstances and adverse social, cultural and contextual factors have all been identified as suicide risk factors (Kivisto et al., 2009). The presence of a physical illness (against psychiatric symptoms in young adults), physical pain, depression, functional impairment, lack of future orientation, and the death of partner have been identified as the major risk factors in suicide among the elderly (Kivisto et al., 2009; Sheikazadi et al., 2010).

1.4.6 Ethnicity and Suicide

Ethnicity and cultural background can be major influences on suicidal behaviour (Arun et al., 2010). A study of young people of Asian origin in the UK has found that the suicide rate of 16-24 year old women was 3 times that of white British women of similar age group (Borrill et al., 2006). This contrasts

sharply with the suicide rates of young Asian men who appear to be far less vulnerable to suicide than young men from white British backgrounds (Mckenzie et al., 2003).

Asian women's groups have linked the high suicide rates amongst young Asian women to cultural pressure, conservative parental values and traditions such as arranged marriages which may clash with the wishes and expectations of young women themselves (Borrill et al., 2006; Tewksbury et al., 2010). By ethnicity, Caucasians are nearly 2.5 times more likely to kill themselves than people of Native American descent and Hispanics in the United States (Arun et al., 2010). Whites and Native Americans (especially adolescents) have the highest suicide rates than any other ethnic group in US (Baeza et al., 2000). African Americans are more exposed to violence, and expected to choose violent methods of suicide. Recent studies have found an increase in the rate of suicide among young African American males is increasing steadily (Stack et al., 2005). Incarceration, police brutality, and the urban socio-economic landscape impact African American males more severely.

In both Asian and Pacific-Island countries, the numbers of reported suicides are growing every year (Elfawal, 1999). A study conducted in Saudi Arabia in 1999 revealed that Asians accounted for (70%) of the whole cases that are received by the Medical Examiner Office- moreover Indians- demonstrated the highest suicidal rates (43%) among the overall data. The method of suicide included hanging, followed by jumping from heights and self-inflicted gunshot injuries (Elfawal, 1999).

1.4.7 Marital status and Suicide

Marital status has a strong association with the rates of suicide. The rates are higher in divorced and widowed individuals than in single individuals, who in turn have higher suicide rate than married people (Mckenzie et al., 2003). Living alone and being single both can increase the risk of suicide. Marriage is associated with lower overall suicide rates and on the other hand, divorced, separated and widowed people are more likely to direct the individual to commit suicide. These associations fit well with the classic sociological theory of suicide proposed by Emile Durkheim (1897) (Hanzick et al., 2002). Durkheim proposed that suicide would be common where the level of social integration (the extent to which the members of a society are bound together in social networks) is low (Hanzick et al., 2002).

1.4.8 Socio-Economic Factors

Researchers had theorized that poverty would be a significant risk factor for suicide (Liem and Roberts, 2009). The theory was based on the hypothesis that being poor could make one feel depressed, desperate or ashamed at sometimes (Lorant et al., 2007). This is not entirely wrong, but research has shown that both the lowest-low and the highest-high incomes are more strongly associated with rates of suicide than other income levels (Lorant et al., 2007). In other words, it is the extremes of either poverty or wealth that are associated with higher suicide rates (Strand et al., 2010). Moreover, suicide risk is higher among individuals who are unemployed (Kaslow et al., 2011).

1.4.9 Methods of Suicide

Suicide methods vary within individuals, communities and several factors are related to the availability of methods and to gender (Lorant et al., 2007). Individuals with suicidal feelings may consider different methods or any means by which a person can purposely kill him or herself. Suicidal methods can be classified into two categories either physical or chemical (Kaslow et al., 2011). Physical suicidal processes act by incapacitating the respiratory or the central nervous system whereas, chemical suicidal processes focus on interrupting biologically significant processes such as cellular respiration or diffusion capacity (Khandekar et al., 2008). While chemical methods of suicide produce latent evidence of action, physical methods often provide direct evidence on the manner of death (Arun et al., 2010). There are many different ways by which people commit suicide. Some of the most common ones are described below:

1.4.9.1 Suicide by hanging

Hanging is a process where by an individual uses either a rope, cord, or any kind of ligature, placed around the neck and then dislodges him or herself from a chair or similar object. It is the prevalent means of suicide in pre-industrial societies and is more common in rural than in urban areas (Dogan et al., 2010). With this technique, the actual cause of death depends upon the type of hanging. In a short drop, the victim may die from strangulation in which the death may result from pressure on the carotid sinus leading to reflex cardiac inhibition and mechanical asphyxia. In addition, acute respiratory acidosis may occur, and the jugular vein may be compressed

sufficiently to cause cerebral ischemia and a hypoxic condition in the brain which will contribute to death (Bertolote et al., 2005). In the case of a sufficiently long drop, the individual is likely to suffer injuries to the vertebrae and spinal cord, a fracture dislocation of the 2nd and 3rd and/or 4th and 5th cervical vertebrae which may cause paralysis or death (Hakko et al., 2007). Forensic Pathologists examine the rope marks on the neck of an individual to determine if the edges are inflamed, a sign that the victim was alive (Tewksbury et al., 2010). The rope marks on the victim's neck are examined to check that they match the rope found at the scene of crime (Stack et al., 2005).

1.4.9.1.1 Crime Scene Investigation in Hanging

Inspecting the crime scene is considered an important part in order to solve the case. All death investigations should be handled as homicide cases until the facts are proven differently. The majority of these cases occur in the person's own home or garden (Pritchard, 2007). A small minority occurs in prisons or psychiatric wards. Either rope or cord is the most common ligature (Levy et al., 2007). In almost half of the cases, individuals are not fully suspended off the ground. The rope employed in hanging is generally a slender cord (e.g., clothes-line, belt, suspenders, and towels); also, in the Middle East individuals may use scarves (Shields et al., 2005). Crime scene inspection should be carried out very carefully to ease the investigations. Moreover, the following points should be noted in the crime scene and should also be considered as being suspicious circumstances:

- Sometimes the presence of dirt, leaves, or anything that is not present in the scene of hanging.
- The rope marks on the victim's neck are examined to check that they match the rope found at the scene of crime (appendix 4, colour plate 1).
- Noticing if the knots and nooses are formed in such a way that it is doubtful whether the dead person could have made them.
- Observations can be made if the victim is right-handed. He or she usually places the noose on the right handed-side of the neck and left-handed ones on the left. Reversal of any of these positions is suspicious (Spitz, 2006).
- The usual suicidal hanging shows a mark on the neck which does not run around the full circumference of the neck because the junction between the noose and the vertical part of the rope pulls away from the skin, thus not marking it (Eren et al., 2007).
- In hanging, the ligature mark on the neck usually rises to a peak pointing to the junction (teardrop shaped), this being distinguishing feature from the strangulation ligature. However, exceptions occur if the noose is very tight (Eren et al., 2007).
- To further differentiate strangulation from ligature, the mark on the neck is usually below the level of the thyroid cartilage, while in suspicious hanging, it is usually above thyroid cartilage (Hejna and Safr, 2010).

1.4.9.1.2 Forensic Investigation in Suicidal Hanging Cases

Medical Examiners must be able to distinguish between hanging and asphyxiation caused by other means such as manual strangulation, ligature

strangulation and smothering (Spitz, 2006). Much depends on observations made in the scene investigations.

- Neck marks are common, but may not be present if a soft material like a scarf or a sheet has been used. The marks run diagonally across the neck in hanging, but are commonly horizontal in manual strangulation.
- An exhaustive examination of the ligature material, placement of the ligature mark over the neck and other associated findings go a long way in deciding the issue. Usually in strangulation, the ligature mark is transversely placed in the lower aspect of neck below the level of thyroid cartilage, whereas in hanging it is often obliquely placed in the upper aspect of the neck (Thierauf et al., 2010).
- Toxicology testing for either drugs or alcohol plays an important role in the investigation of hanging. A suicidal victim may take drugs to get up courage to perform the act where as a murder victim may be subdued or made unconscious in the case of a homicide, despite the fact that this is an extremely difficult task. On the other hand, homicidal hanging of a fully conscious able-bodied person is virtually impossible. Suicidal strangulation by a ligature is rare, but not impossible, and must be ruled out when a victim has been strangled (Parroni et al., 2002).

1.4.9.2 Suicide by Drowning

In this situation, the resolution of the mode of death in drowning is based on a series of factors that eliminate both homicide and accident (Spitz, 2006). Drowning is considered among the least common methods of suicide, typically accounting for less than 2% of all reported suicides in the United

States (Borrill et al., 2006). Bodies recovered from water are examined to find out whether water is actually present in lungs and stomach of the victim (Athanaselis et al., 2002). If the latter signs are apparent, then the victim did actually die due to drowning; although whether it was murder or accidental will be left for investigators to determine (Mckenzie et al., 2003). Further examinations will reveal if bleeding occurred in the lungs, suggesting that there was a struggle for breathing during the drowning (Pritchard, 2007). Moreover, to reveal whether bodies were either alive or dead upon entering the water, an analysis of Diatoms is performed, which are unicellular algae (Tewksbury et al., 2010). Comparison of diatoms found in the body with those found in the water is carried out and if these samples match, then the body was alive before entering the water. Otherwise, the body was dead upon entering the water and the Forensic Examiner will continue searching for other injuries that may point to murder (Azmak, 2006). Findings of asphyxiation due to drowning, includes presence of foam in the airways (which is formed due to the mixing of mucus and water as the victim struggles to breathe), haemorrhage of the middle ears, and electrolyte imbalance is also found (Tewksbury et al., 2010).

In addition to the physiological observations of the corpse, Investigators would consider whether the individual was clothed, the presence or absence of a suicide note, and the depth of the water in which the individual was found. Drowning in shallow water could indicate either an accident or murder, especially if the victim is undressed. A killer will remove the victim's clothing to make it look like an accident. If the body is fully clothed, it is most likely a suicide since the victim does not want to be found in the nude (Browne,

2004). The presence of a suicide note usually indicates a suicide, but could be faked. This suicidal note should be considered in every suicide case (Stack et al., 2005). If the victim shows evidence of gunshot wounds, bruises or cuts, it is most likely to be a murder (Spitz, 2006). The discovery of either alcohol or drugs at the scene could suggest an accidental drowning (Spitz, 2006).

1.4.9.3 Suicide by Suffocation

Suicide by suffocation is the act of inhibiting one's ability to breathe or limiting oxygen uptake while breathing, causing hypoxia and eventually asphyxia. (WHO, 2006; Boghossian et al., 2010). Suffocation may be achieved with different items, such as: plastic bags, masks or more complicated apparatus involving some other kinds of head covering. Sometimes, victims use anaesthetics, other gases or volatile substances e.g. glue. However, using plastic bag suffocation as a method of suicide remains unusual (Serafettin et al., 2009). Other commonly used in suicides by suffocation is Helium, Argon and Nitrogen. While breathing, the gas enters the lungs very quickly and it renders a person unconscious and may cause death within minutes. Forensic examination in cases of suffocation shows prominent organ congestion, petechial hemorrhages and a remarkable pulmonary edema (Athanaselis et al., 2002). In these cases where a Forensic Pathologist is called upon to try and determine the cause of death, he or she should observe a number of characteristics.

First, one of the initial tasks the Forensic Examiner will do, if there are no discernible wounds or marks, is to check the deceased's eyes (Spitz, 2006). In case of either suffocation or smothering, the eyes of the deceased will be

protruded, congested (blood shot); with tiny haemorrhagic spots on the conjunctiva or even ecchymosis, which indicates anoxia of the tiny capillaries. These findings could be a positive sign for reaching an accurate diagnosis (Hejna and Šafr, 2010).

In addition, crime scene investigation in cases of suffocation can reveal that most individuals usually choose a private or secluded place for them to commit suicide. When carried out at the victim's home, the activity is usually confined to an out-of-the-way place (e.g. basement) at a time when the home is vacated by other family members (Parroni et al., 2002). Sometimes this practice is intended to induce sexual pleasure and not suicide, the so called sexual asphyxia. Hence, the Forensic Investigator must report any evidence of sexual activity (Prakash et al., 2008). Often the body may be found either nude, partially nude, or with the penis projecting through an open fly, and the hand touching the genital area, with the presence of some pornographic materials (Delmonte and Capelozzi, 2001).

Also, in case of carbon monoxide poisoning, where the individual tries to use the gas stove as a method of suicide, the Forensic Scientist must prove whether or not the victim was conscious at the time of the fire starting or if indeed the person had already been deceased (Sauvageau and Boghossian, 2011). This process is measured by the amount of carbon monoxide in the deceased's blood. To diagnose suffocation by smothering, the examiner should look for bruising around the nose and mouth, more over looking for trace evidence such as hair and fibers around the nose and mouth of the deceased (Spitz, 2006). In addition, histological screening for increase air

trapping and evidence of acute rupture of inter alveolar septum, as well as possibility of increase in blood concentration of carbon dioxide (blood gases) may also add positive findings to reach the diagnosis (Bennett et al., 2006).

1.4.9.4 Suicide by Electrocutation

Injuries and deaths by electric current are not uncommon. Most deaths by electric current are accidental and are caused by contact with broken or non-insulated wires in faulty domestic or industrial appliances (Spitz, 2006). Cases of suicide electrocution involves using a lethal electric current to kill oneself have been recorded, while electric homicide although rare, is possible when an electric shock may be given in malice (WHO, 2009).

One of the difficult cases that may face the Pathologist is when a person is killed by poisoning or throttling, then electrical burns may be caused on his finger so as to make it appears like death from electrical lesions. In such cases it is difficult to differentiate ante mortem from postmortem electric burns even with microscopic examination (Khandekar et al., 2008).

Forensic examination reveals petechial haemorrhages of internal organs called "electrical petechiae" that represent a nonspecific finding of electric burns. (Spitz, 2006). A typical finding in electrocution irrespective of the mechanism leading to death, also with autopsy a blackish linear mark on the pleura of the inner side of the thoracic cavity (Khandekar et al., 2008). Electrical burns have a characteristic appearance and pathologic findings. Electrocutation includes streaming of the epidermal nuclei at the point of contact with the current (Eren et al., 2007). Higher voltage, typically electrifies jewelry

and can cause significant secondary burns where these meet the skin. The tissues in the proximity of a necklace, watch or ring will also be severely damaged in many cases (Khandekar et al., 2008).

1.4.9.5 Suicide by Fall from Heights

There is very little data on suicidal falls from heights and there is very little information available in the forensic literature or on web sites concerning fatal falls (Pritchard, 2007). The Forensic Pathologist has an important role in cases of fall from a height death. It is often very difficult to determine whether the fall resulted from an accident or whether it was a suicide or homicide, especially if there were no witnesses and no suicide note or any other indications of a potential suicide (Hakko et al., 2007). The Investigators, along with the Forensic Examiners, have to identify the physics of the problem, in which they have to make calculations of the trajectory during the fall (Pritchard, 2007). Some studies have indicated that subjects who attempt suicide by jumping from a height have different profiles of mental state, personality function, and psychiatric diagnosis, which are very crucial as determinants of the method used in suicide (Moore and Robertson, 1999). In determining the heights, the Forensic Scientist has to be able to study the following points in to consideration: the height of the building, the horizontal distance travelled during the fall, the vertical slope of the building, and the primary and subsequent points of impact (Spitz, 2006).

First, in order for the report to be of a highly scientific investigation, it is very important to calculate the height of a building, although it may not be easy to measure it correctly (Bertolote et al., 2005). Forensic Pathologists can

usually determine the cause of death from the nature of the injuries, and can usually tell whether the deceased landed on either his head first, then feet or feet first then head (Adamec et al., 2010). It is very crucial to measure the distance travelled during the fall (Adamec et al., 2010). It might involve one team abseiling down a cliff and another team taking distance and angle measurements (Thierauf et al., 2010). Furthermore, the vertical slope of the building is also significant. The deceased person may have bounced or slid to the point where they were found. If the fall is into water, then police or navy divers may need to assist with measurements close to the edge of a cliff (Türk and Tsokos, 2004; Thierauf et al., 2010). Finally, the primary and subsequent points of impact must be determined by the Forensic Scientist (Spitz, 2006). The duty of the Forensic Expert is to identify the initial impact point and compare this with the death scene. It should be marked with paint and photographed for later examination, even if the initial investigation points to suicide. It might later on turn out to be a homicide (Adamec et al., 2010).

1.4.9.6 Suicide by Fire Arms

Throughout the world, deaths due to firearm weapons have increased tremendously due to the availability of firearm weapons and other factors such as drugs, media, and mental disorders, all play an important role in increasing the statistics of fire arm fatalities (Hussain et al., 2006). Both males and females might select a firearm as the preferred method of suicide, and they choose it because it is quick and relatively painless (Sheilds et al., 2005). Firearms are used in both homicide and suicide as a method of killing and they are used is differently from one area to another in the world. In England and Wales, the most frequent firearms are shotguns, both in cases of

homicide and suicide. While in United State, there are more than 25,000 people who die every year by injuries which are caused by firearms (Tewksbury et al., 2010). In the United States, the most frequent method of killing in cases of homicide and suicide is by means of firearms (Hejna and Safir, 2010).

The appearance of firearm wounds depends on several factors which include:

- Type of weapon used.
- Nature of the projectile fired (may be shots, pellets, slugs or bullets).
- Distance of firing and the velocity of the bullet.
- The direction of firing.

Differentiating between suicide and homicide in firearm cases can be determined by the site of the injury, distance of firing, direction of fire, number of wounds, and presence of gunpowder on the hands of the individual. In suicide, there are three preferred sites; the right temple, the roof of the mouth and the left side of the chest. Wounds located anywhere else may be homicidal or accidental (Hejna and Šafr, 2010). Most suicide injuries are at contact or near contact range, causing a burn mark around the wound and leaving gunpowder residue (Spitz, 2006). A crater or star-shape wound is usually produced in contact shot, especially if the gun is fired over a bone, such as the skull or the sternum, anything further away is likely homicide (Okoro et al., 2005). Sometimes suicide shots are angled slightly upward. After one firing, even if a suicide victim is not dead, the individual would likely be unconscious or physically unable to fire a second time. Multiple shots usually indicate homicide (Shields et al., 2005).

Finally, in suicide, it is crucial to inspect the powder residue from unburned carbon on the hand that fired the gun. Shooting through clothing sometimes suggests homicide. A suicide victim will rarely shoot through clothing. If he shoots himself in the chest, which is unusual, he usually opens his shirt to expose the skin (Tewksbury et al., 2010). If the victim left a suicide note, or was known to have personal problems, or if there was an evidence of drug use or drinking, suicide is likely to be the cause of death (Leenaars et al., 2010). Forensic Investigators usually study the association of gun availability and cases of homicide and suicide victimization (Hejna and Safr, 2010). Most articles predict that the storage of a firearm at home predicts an increased rate of a violent death. Other study indicates that increased gun prevalence goes with increases the risk of homicide and suicide rate (Okoro et al., 2005).

1.4.10 Suicide in the Middle East

There are very little data available in the literature regarding suicide in the Middle East Region. One previous study was conducted at the Medico-legal Centre in Eastern Province of Saudi Arabia (Elfawal, 1999). Within this study, the largest racial group was South Asians. To evaluate the influence of racial and cultural factors on suicidal rate, the study concluded that among this group, Indians outnumbered others by 43%. Suicide by hanging was the most popular method and accounted for 63% (Elfawal, 1999). Another research study conducted in United Arab Emirates revealed that hanging was the commonest method for committing suicide. Both Females-non-Indian expatriates choose jumping from a height and self-poisoning as a method of suicide that was triggered by depression and un-employment (Koronfel,

2002).

1.5 Homicide

1.5.1 Introduction

Violence takes its toll on individuals, families, and communities all over the world. Another form of violence beside suicide is homicide. In suicide the violence is directed towards the own self while in homicide it is directed outwards on others (Boghossian et al., 2010). The precise term for the killing of one person by another is homicide (Levy et al., 2007). Murder is a form of criminal homicide that has a precise legal meaning (WHO, 1978). In 2002, the Center for Disease Control and Prevention (CDCP) established the National Violent Death Reporting System (NVDRS) in United State. NVDRS collects data on violent deaths from a variety of sources, to provide a comprehensive picture of the circumstances surrounding homicide or suicide in order to provide some insights into ways to improve efforts to prevent violent deaths.

To date, most of the studies have been carried out on data from Western Europe/North America and non-Muslim Countries. This study is concerned with the un-natural deaths in Kuwait. Thus, emphasis is placed on culture and religion which could play an important role in the pattern of violent deaths. It is equally important to study the death issues in the context of Middle East Region.

1.5.2 Investigation of Homicide

All homicides should be treated equally. No distinction is made in investigative technique with regard to the various motives (Adi et al., 2010).

The general stages of investigation protocols may be similar, but the emphasis and direction of investigation will vary according to the particular motive for the homicide. For example, if a homicide involves the shooting of a wife by her husband, then the husband should not be investigated in the same way as a case in which the nude body of a woman is found on the highway.

1.5.3 Law and Homicide

In general, homicide means killing of one human being by another (WHO, 2009). There are different legal variations of murder, known as degrees. Degrees of murder vary by the gravity (seriousness) of the offense (usually measured by the intent of the perpetrator) and the sentence assigned to that offense (WHO, 2009). For example, first-degree murder carries the sternest sentences and is usually reserved for murders committed with premeditation or extreme cruelty. Justifiable homicide is accepted in circumstances that led to death of the victim. For example, if homicide is caused unintentionally, by an act done in good faith, in self or family defense, homicide committed by the insane, or during surgical operation by a doctor (Grabherr et al., 2010).

1.5.4 Motives for Homicide

In order to study the incidence of homicide and set up some remedial strategies within any area in the world, motives that led to homicide should be recalled and investigated. Although some will end a life of another person without apparent motive, individuals who kill without motive are more likely to have: (1) history of alcohol abuse, (2) a recent release from prison, (3) claims of amnesia for the crime, and (4) denial of the crime. They also tend to exhibit

psychotic behaviour following the crime and to be assessed as not guilty of the crime due to mental illness (Scolan et al., 2004).

The motive is defined as the alleged primary causal factor that precedes and often leads to the events, the outcome of which is the death of the victims. Motives for homicide include:

- Revenge
- Involving a lover or other lover's quarrels
- Money to buy drugs
- Alcohol-related arguments
- Emotionally-based, typically anger
- Financial gain (Robbery)
- Sexual gratification and apparently sex-connected homicides
- Domestic which includes jealousy, desertion/termination of relationship, and other domestic altercation.

1.5.5 Relationship between victims and offenders

A five-category description of the relationship between victims and offenders including: strangers, acquaintances, friends, relatives, and those romantically linked, should be taken into consideration by the death investigators (Ambade et al., 2007). Previous studies have suggested that as age increases, homicides are more likely to involve persons in intimate relationships and family members. Although most homicides tend to occur in residential premises, those involving youth often have a fair chance of occurring in pubs, on the streets or other open areas (Grabherr et al., 2010).

1.5.6 Homicide using firearms

Throughout the world deaths due to firearm weapons have increased tremendously (Levy et al., 2006). In the United States, the most frequent method of killing in cases of homicide and suicide is by means of firearms. More than 25,000 people die every year in the USA by injuries caused by firearms. In England and Wales, the most frequent firearms are shotguns (Hussain et al., 2006). In the Middle East, there are a few studies that are conducted to evaluate the problem of homicide in the region. In 1997, a research study was performed in the Eastern Province of Saudi Arabia during the 10-year period from 1985 to 1994. The study showed that 48% of unnatural deaths were homicide and hand guns were responsible for 56% of homicides (Elfawal and Awad, 1997).

Gunshot wounds may resemble stab wounds in external examination (Wilson, 2010). Therefore, the forensic examination team assists in providing clues to the circumstances under which they occurred. Basically, when a firearm is discharged, four events occur:

1. Flame is emitted from the barrel.
2. Smoke then follows this flame.
3. The bullet emerges from the barrel.
4. Burnt and un-burnt gunpowder follows the bullet out of the barrel.

All of the above points should be considered in any firearm death (Marshall et al., 2009). Moreover, external examination of the cadaver should be conducted as early as possible in the crime scene prior to the transferred of

the body to the mortuary (See appendix 2, colour plate 2). In addition, the injuries must be numbered, charted and the size, shape and exact site of the firearm injury related to a fixed anatomical landmark must be documented (O'Grady, 2010). All the diagnostic characteristics of firearm injuries must be noted too. The injuries must be examined by the naked eye as well as a magnifying glass lens. In addition, internal examination must also be performed of the viscera and organs in all the main cavities. The track of the projectile must be followed and the extent of internal injuries to different viscera and organs must be determined (Wilson, 2010).

1.5.7 Kuwait Firearms Unit

Kuwait Firearms Unit is constituted as one part of Criminal Laboratory Department, which examines all cases that are transferred from Forensic Medicine Department that are assigned to assess firearms for operability, compares fired evidence bullets and cartridge cases to test fired ammunition, and makes determinations of firing distance.

The general examination of the firearm includes determination of loaded or unloaded status, rendering the firearm safe for handling and examination, and a determination of operability. Fired bullets and cartridge cases are examined in the unit with the aid of microscopes to determine what type of firearm may have been used. Striations (scratch marks) on fired bullets are unique to each firearm, as they are caused by imperfections in the barrel. Based on a finding of sufficient similarity between the questioned and test fired bullet, the Criminalist can reach the positive conclusion "this firearm fired this bullet." Similarly, fired cartridge cases can be identified to the firearm

in which they were fired, through microscopic comparison of marks left on the cartridge case by the firing pin, chamber, extractor, ejector or breech face of the firearm.

Firing distances are determined in the ballistic unit by examination of target surfaces for gunpowder, lead, and nitrites. The pattern of these depositions gives information as to the probable firing distance (Spitz, 2006). Trajectories of bullets are determined to aid in the reconstruction of events at a shooting scene. Obliterated serial numbers on firearms may be restored through special chemical processing of the obliterated surface (Wilson, 2010).

1.5.7.1 Homicide by Stab wound

Penetrating injuries on the body may be homicidal, suicidal or accidental in nature. There are certain features that are quite reliable pointers to the nature of these injuries (Bennet et al., 2006). However, differentiation is difficult particularly when the characteristics of stabbing are perplexed and information related to circumstances of the incident is misleading. In medico-legal practice, the Forensic Examiner has to give his opinion about the homicidal or suicidal nature of death (Scolan et al., 2004). Critical analyses are made by expert for various characteristic features of the stab wound may help to determine the manner of death due to either suicide or homicide. The site of the wound has particular importance in determining the manner of death (see appendix 2, colour plate 2). A vertically placed stab wound on the chest at the level of left anterior axillary line implies homicide, while, in a case of suicide, the direction of the wound is normally horizontal (Vij, 2002).

One weapon can cause many different types of injuries (i.e. incision wounds and stab wounds). In a knife case, the Forensic Examiner can never match the weapon, to the assailant, with 100% certainty, but residual blood or other biological evidence from the victim on it or if the tip of a broken knife is found as in the case of a gun (Spitz, 2006). The Forensic Examiner can, however, eliminate certain weapons. There are several factors to be taken into consideration when determining if a death is homicide or suicide especially with cuts.

- **Defensive cuts:** In a homicide, there will be defensive cuts on the palms of the victim's hands and on the underside of her arms as s/he tries to fight off the attacker (Vij, 2002).
- **Number of wounds and their location:** A murder usually involves multiple stab wounds to the side, back or stomach. In a suicide, while there may be additional cuts across the wrist and tentative test stabbings to see if it will hurt, or to work up courage, there will usually only be one wound and most likely in the chest .
- **Location of the murder weapon:** In a suicide, the weapon will be at the scene with the victim's fingerprints on it. In a homicide, the weapon is often missing.
- **Presence of a note:** A suicidal note should be considered, victims will almost always leave a note before committing suicide (Serafettin et al., 2009).

1.5.7.2 Stab Wounds in Homicides

These are commonly found on the trunk, neck, upper arms and head. There are regularly multiple wounds which are widely spread. There are also defensive injuries involving incised wounds of the palms and fingers, that are common features in stabbings (Abdullah et al, 2003). This indicates an attempt to grab the weapon or to protect against it and shows that the victim has had time to react.

1.5.7.3 Homicide by Strangulation

Application of force to the neck by the hands or a ligature is called strangulation (See appendix 2, colour plate 2). In strangulation by hand (throttling), death sometimes occurs immediately from reflex cardiac inhibition, but usually the squeezing of the neck arteries is incomplete, so death results from interruption of air supply to the lungs (Athanaselis et al., 2002; Boghossian et al., 2010).

1.5.7.4 Homicide by using blunt force

One of the most common cases encountered by Forensic Pathologist, is deaths resulting from blunt force trauma (Krienert and Walsh, 2010). Whereas other forms of traumatic deaths (e.g. gunshot wounds, sharp force injuries) occur under a relatively limited number of circumstances. Deaths resulting from blunt force trauma occur in a variety of scenarios (Spitz, 2006). Accidental blunt force includes several injuries such as transportation fatalities, motor vehicle collisions, pedestrians being struck by vehicles, airplane crashes, and boating incidents—as well as jumping or falling from

heights (Wilson, 2010). Blunt force trauma may cause contusions and lacerations of the internal organs and soft tissues, as well as fractures and dislocations of bony structures which can be detected through autopsy (Marshall et al., 2009).

1.5.7.4.1 Inspection of Crime Scene in Blunt Force Injury

Scene findings often play an important role in the death investigation process. Examples include blood spatter evaluation and DNA analysis in homicide cases involving multiple blows with a blunt object, such as a baseball bat, and scene reconstruction following motor vehicle collisions (Gilligan and Lennings, 2010).

1.5.7.4.2 Inspection for Trace Evidence

In certain cases, it may be necessary to examine the skin surface and wounds for trace evidence. This may involve looking for paint, metal, or glass fragments on a body that was struck by a hit-and-run motor vehicle. Such trace evidence may be used by forensic science technicians along with the Forensic Examiner to help in identifying the vehicle or weapon that caused the injuries (Gilligan and Lennings, 2010).

1.5.7.4.3 Signs of Blunt Force Trauma

There are several different signs of blunt force trauma and they are include: 1) bruising which is a good indicator that there are broken blood vessels beneath the surface of the skin; 2) abrasions are cuts, grazing of the skin, or friction burns which can be caused by the victim being beaten, dragged or kicked (Spitz, 2006). These wounds can sometimes indicate that a

victim hit against something or was hit with something and it can also be used to measure how much of a struggle the victim put up against his or her attacker; and 3) lacerations, the tearing of tissue underneath the skin. An individual may be beaten severely or have sustained a severe bump against a stationary object and underneath the injury, there may be severe damage caused to tissue and organs.

1.6 Infanticide

Infanticide means an unlawful destruction of a newly born child and this is regarded as murder by the law (Levy et al., 2007). It is necessary for the Medical Examiner and Forensic Pathologist to prove whether the baby was born alive or still (Macgregor, 2003). Filicide, neonaticide, and infanticide are terms that have been used interchangeably in the literature to describe child homicide. Filicide is the murder of a child by a parent (Brunel et al., 2010). Neonaticide refers to killing of a child within the same day of birth. On the other hand, infanticide applies mainly to the killing of a child under the age of 12 months by a mother who suffers from mental distress (Goldenberg, 2004).

1.6.1 Medico Legal Aspects

In any cases of unexpected infant deaths, or when a discovered dead body of an infant is found under suspicious circumstances, especially if it is a body of newly born infant, a medico-legal protocol must be followed by the Forensic Pathologist to determine the following (Krous et al., 2002). First, the alleged mother should be examined for signs of recent delivery, such as: dilated cervix, perineal tear, sub involutes of uterus, passage of a puerperal vaginal discharge called lochia, engorged breasts with discharge of milk, and

recent stria gravidrum (Bennet et al., 2006). In addition, a psychological examination for her mental condition should be taken into consideration, including an assessment for the surrounding circumstances around her pregnancy and labour. It must be determined whether it is legal or illegal pregnancy. The social, traditional, cultural, and financial condition of the mother must also be considered.

Also, regarding the dead foetus, the Pathologist has to answer the following medico legal questions:

1. Was the foetus stillborn, dead born or live born?
2. Is it mature (full term) or pre term (premature)?
3. If pre term (premature), what was the viability of the foetus?
4. If born live, how long did the child live?
5. What is the cause of death? What are the identifying relevant features?
6. Time of survival (in living birth).
7. The type of delivery, whether it was normal, difficult or precipitated
8. Time passed since death occurred (from post mortem changes).

A detailed discussion for each point should be submitted. New born infants found dead are not necessarily victims of infanticide. Those stillborn or naturally dying or from unnatural lack of care may be hidden or neglected. Also, the cause of in such cases may be for the purpose of birth conferment from another for any cause (Gilligan and Lennings, 2010). A cooperative effort must also be undertake from the medico legal doctor and from the police research. These including identification of the examined child as regard his/her ethnic group, blood grouping, DNA genetic typing etc., all of

which will help in elimination or confirmation of consanguinity with the putative mother (Gilligan and Lennings, 2010).

1.7 Accidental Deaths

An accidental death is as the term would suggest, the death of an individual by means other than natural death, murder or suicide (Singh et al., 2005). Accidental death can also be categorized as death by misadventure; thus, the victim has died by accident either in general recklessness or by taking risks that would put him or her in mortal danger (Palmiere et al., 2010). In this manner of death, some degree of human negligence may be involved in accidental deaths. For example, a pedestrian killed in an intersection by a sober driver, not speeding or running a red light, would be reasonably considered a victim of accidental death (Spitz, 2006). In addition, a lot of extreme sports participants have died and their deaths have been classified as death by misadventure because of the extreme nature of their pastimes. Generally speaking, any event, occurring suddenly, unexpectedly and inadvertently under unforeseen circumstances is considered as accidental death (Ahmet et al., 2005). These cases range from fatal car accidents to slip and fall accidental deaths and they also include motorcycle, trucking, aviation, railroad, boating accidents and crashes. Accidental deaths also occur due to fire, electric shock and drowning. The following are some kinds which are considered as accidental deaths: Road traffic accidents (RTA), occupational accidents, or domestic accidents (See appendix 2, colour plate 3).

Since accidental death is of great concern in Middle East and Kuwait specifically, RTA, occupational, and domestic accidents are discussed

thoroughly in the next few paragraphs. RTA is one of the most common causes of un-natural deaths below the age of 50 years (Singh et al., 2005). The pattern of injury varies considerably depending upon whether the victim is a vehicle passenger, a motorcyclist or a pedestrian (Kuruc et al., 2009). Accidents are the third leading cause of deaths for males in the United States, but only the seventh leading cause of unnatural death for women. Men are more vulnerable to accidental death than women due to the type of occupation and other risk factors (Singh et al., 2005).

Incidences of RTA have been increasing at an alarming rate throughout the world. In Kuwait, RTA death victims are transferred to the GDCE, (FMD) where they are examined for injuries causing death and laboratory blood test are routinely carried out to check for alcohol consumption. RTA accidents in the Middle East are considered as the main cause of accidental deaths (Middle East High Way Report, 2010). Conditions have become so bad, that the Indian Government recently issued warnings to its nationals about hazardous driving conditions in Saudi Arabia and other Gulf Countries (Kuruc et al., 2009; Ahmet et al., 2005). In Saudi Arabia, over 5,000 Saudis died in traffic accidents in 2009, most were motorists who ignored basic traffic safety rules, such as using a signal when turning, maintaining safe speed, or using lights at night (Middle East High Way Report, 2010). Kuwait's Ministry of Health reported that the number of accidents in Kuwait, for the first three quarter of the year 2010, have increased in comparison to the previous year (Health Kuwait, 2010). Studies have shown that collisions at 20 mile per hour result in a 5% risk of death, while, those at 85 mile result in an 85% risk of death. Road traffic accident is now

acknowledged to be a global phenomenon in virtually all countries of the world (Middle East Highway Report, 2010).

In Kuwait, whenever a car travels a given distance, the possibility of one RTA death is 40 times greater than in United State and the number of deaths related to the number of injured is 9 times than those in the US (Kuwait Central Statistic, 2011). In Kuwait, about 70% of accidents are due to ignorance, carelessness and speeding, and the remaining 30% are as a result of mechanical problems and poor road conditions in very far desert areas. Therefore, the situation in Kuwait is more serious and getting even worse year by year, especially exacerbated by the progressing density of traffic with annual increase of 80.000 vehicles per year (Al-Hassan, 2011).

The term occupational accident refers to work-related mishaps (WHO, 2006). In such category, accidents that injure employees are the responsibility of the employer and are covered by workers compensation insurance. Recently, the term has been expanded to include job-related long-term exposure to hazardous substances such as asbestos, silicosis, hospital infections for medical staff and hospitals workers that result in occupational diseases, and such emotional injuries as nervous breakdowns and even heart attacks that all lead to death. The responsibility of the Forensic Examiner is to identify in computing the degree and percent of disability in cases of living person and cause of death for the purpose of compensation in cases of incidents leading to death (Spitz, 2006). In occupational deaths, individuals die from several areas concerning occupational injury related to death and they include;

- Machinery, falling objects
- Gas station attendant
- Environmental contamination
- Falls
- Fires/explosions
- Infections acquired in a hospital

Death can occur from any electric sources that carry enough current to cause death (Glover et al., 2004). Small currents (40 A-700 A) usually trigger fibrillation in the heart which is reversible via a defibrillator. However, large currents (>1 A) can cause permanent damage via burns, and cellular damage and the heart is most devastated by foreign electricity as well as the brain (Eren et al., 2007). Most accidental deaths are caused by aerial power lines (Wick et al., 2006). The most common place for an electrical injury is a railway area or residential property (Palmiere et al., 2010). Study done by Lindström in 2006, analyzing accidental fatalities caused by electricity—at work and during leisure time, revealed that most deaths were caused by aerial power lines (Lindström et al., 2006).

1.7.1 Death by drug or medical over dose toxicity

Drugs come in many forms both legal as prescribed and illegal. Illegal drugs will obviously be seized as evidence of criminal activity based on the fact that even possession of such substances is against Kuwait law and other judicial system (International Constitutional Law, 1992). Death by drug overdose often leaves no visible marks on the body other than heavy congested lungs (Spitz, 2006). For the Forensic Pathologist who cannot find

any cause of death in or on the body, the identification by a Forensic Toxicologist's of the drugs present may lead to a conclusion of drug overdose by exclusion.

Deaths by drug over dose may leave confusion over whether the deaths were suicidal or accidental. There are 30 to 50 times as many attempted suicides as completed suicides (Neale, 2000). Four times as many males complete suicide than females, but adolescent females attempt suicide twice as frequently as male teenagers (Owens et al., 2002). Statistics also show that kids from high-income families kill themselves as often as those from lower or middle-class families (Jenkins et al., 2002). Due to the availability of painkillers medication everywhere, they are among the most popular means that are used in suicide (Brådvik et al., 2009). Accidental drug overdose contributes substantially to mortality among multi-drug users. Multi-drug use has been documented as a key risk factor in overdose and overdose mortality. Research findings on suicidal death due to overdose indicate that opiates, cocaine and alcohol were the three drugs most commonly attributed as the cause of accidental overdose deaths (Coffin et al., 2003). A study was carried out to investigate changes in the rate of multi-drug combination to single drug overdose, revealed that death due to multi-drug combination accounted for most of the overdose death rates, whereas single drug overdose death rates remained relatively stable (Owens et al., 2002).

1.7.1.1 Type of Drug Evidence

Drugs may be defined as any type of substance- whether natural or synthetic and which are used to produce certain psychological,

pharmacological or physiological effects on the body (Spitz, 2006). Drug evidence may be classified into the following categories:

- Opiate or narcotics (Drugs that reduce pain and other sensation and lead to a sleepy state).
- Stimulants (drugs that stimulate the sympathetic nervous system and cause agitation, high blood pressure, increased heart rate and rapid breathing).
- Hallucinogens (Drugs that alter one's perceptions and may lead to disturbing hallucinations).
- Depressants (i.e. drugs, alcohol, etc.) that depress the central nervous system and result in loss of motor coordination and reduced respiration. Some examples of drugs in these categories are found in Table 2.2.
- Drug overdose may also be performed by mixing medications with one another or with alcohol or illicit drugs. Because of this the toxicology laboratory is an important tool to the medical examiner in the investigative section.

Therefore, the Kuwait medico-legal system has a very well-equipped toxicology laboratory, which plays a significant role in determining the causes and manner of death and whether it will be erroneous in many cases. Forensic Toxicologists examine the effects of chemicals on the body and analyze blood, urine, bile and eye fluid. They may perform simple drug tests as described in table 1.2 or use sophisticated methods

such as gas chromatography and mass spectrometry. They look for poisons, prescription, non-prescription drugs and alcohol.

Table 1.2: Table showing commonly used drugs, screening reagents and colour reaction

| DRUGS | REAGENTS | COLOUR REACTIONS |
|-------------------------|--|---|
| Amphetamine | <ul style="list-style-type: none"> • Mandelin • Marquis | <ul style="list-style-type: none"> • Gray – green • Red-brown |
| Cocaine | <ul style="list-style-type: none"> • Cobalt Thiocyanate • Mandelin | <ul style="list-style-type: none"> • Blue precipitate orange |
| Codeine | <ul style="list-style-type: none"> • Mandelin • Marquis | <ul style="list-style-type: none"> • Green • Purple |
| Heroin | <ul style="list-style-type: none"> • Mandelin • Marquis • Nitric Acid | <ul style="list-style-type: none"> • Brown • Purple • Yellow-green |
| LSD | <ul style="list-style-type: none"> • Marquis • Erlich • Van Urk | <ul style="list-style-type: none"> • Dark purple • Pink-purple • Blue-purple |
| Marijuana | <ul style="list-style-type: none"> • Modified Duquenois | <ul style="list-style-type: none"> • Blue |
| Methadone | <ul style="list-style-type: none"> • Cobalt Thiocyanate • Marquis | <ul style="list-style-type: none"> • Blue • Yellow-pink |
| Morphine | <ul style="list-style-type: none"> • Mandelin • Marquis • Nitric Acid | <ul style="list-style-type: none"> • Orange-brown • Purple • Orange |
| Psilocin and Psilocybin | <ul style="list-style-type: none"> • Weber Test | <ul style="list-style-type: none"> • Red to blue |
| Benzodiazepines | <ul style="list-style-type: none"> • Janovsky | <ul style="list-style-type: none"> • Orange |

1.7.1.2 Kuwait Drug Laboratories (KDL)

When investigating drug related cases, Forensic Chemists use utmost caution and care not only in safety guidelines but also in preserving evidence. Forensic Chemists use many different methods to analyze drugs in the forensic laboratories. This is due to the wide variety of different drug substances and chemical components that make up the drug. In almost all Forensic Drug Testing Laboratories, the examination requires both qualitative and quantitative analysis of the drug. Kuwait Forensic Drug testing Laboratories have some limitation in the analysis of drugs. The major limitation is the inability of the laboratory to determine the concentrations of drugs or poisonous substances and their metabolites in body fluids (except for alcohol). KDL only analyzes the qualitative data of the drugs and this in fact because the liquid phase extraction method is used instead of the more sophisticated solid phase method.

1.7.1.3 Drugs and Alcohol Combination

Several studies have indicated that a combination of two or more drugs, for example, alcohol and legal drugs can result in a 'third drug' (synergistic effect) that can harm the body more than one or the other drug when used individually (Spitz, 2006). Researchers have demonstrated that in the presence of cocaine and ethanol, the liver metabolizes cocaine to its ethyl homologue, called Coca-ethylene (Allampalli et al., 2010). This new drug is a neurologically active compound that provides the same feelings of euphoria and well-being as cocaine but is more intense and for longer periods. It is more addictive than the parent drug, and it is certainly more lethal. Death due

to an acute overdose of alcohol is uncommon; indirectly, ethanol plays a significant role contributing to too many causes of deaths. In some violent death cases, alcohol is the major contributing factor in motor vehicle accidents in modern countries. To date, no studies have been done in Kuwait to investigate the effect of alcohol consumption on death cases related to motor vehicle accidents, although, several death cases have been documented in Kuwait Forensic Laboratory reports of alcohol-related RTA. The use of both alcohol and drugs are forbidden in Islamic religion and death sentences are often passed for trafficking in drugs (Christen, 2006). Kuwait has a per capita income of about \$20,000, which is a ready market for drug-smuggling network. Recent statistics released from Kuwait Ministry of Interior indicate that drug addiction is growing in Kuwait. In recent years, about 20,000 cases have been reported annually where people are caught drinking and trafficking in drugs (MOI Report, 2011). In Kuwait, (FMD) usually classifies the manner of death due to drugs as accidental and not as suicidal. The rationale behind this, 1) there is no intention of death; 2) deaths occur without a real overdose in the most common type of abuse which is narcotics (heroin and related substances).

1.8 Domestic Accidents

Many environmental threats to an individual's health are aggravated by persistent poverty, conflicts, natural and man-made disasters, and social inequity. Children are worst affected in the developing world but there are many children in the more developed countries also at risk. The study of domestic accidents including those in and around the home and in institutions, are of increasing importance (Egge et al., 2010). They include:-

- Poisoning
- Falls from height
- Drowning
- Fire, burns and smoke inhalation
- Firearm injuries
- Plastic bag suffocation

Based on WHO report, it became obvious that there was a surprisingly high presentation rate of significant injuries in children. These injuries were presented by the parents or care givers (domestic servants) as being accidental (WHO, 2008). Children and elderly people are particularly vulnerable to immersion accidents due to their physical characteristics. Differences in social class (occupational status) are significant as predisposing influences, with a high concentration of swimming pool accidents in upper social class families, bath-tub accidents occur almost exclusively in lower social class homes. Children within single-parent families are not particularly at risk. Evidence is presented which indicates that socio-demographic patterns differ significantly between fatal and non-fatal cases (Macgregor, 2003). These injuries were attributable to falls. For example, children may fall from bed, a pram or buggy as kids grow older. Furthermore, there are numerous inquiries which are related to scalds and burns. A study in the USA showed burns to be the fourth leading cause of deaths due to injuries in children. Ninety per cent of these occurred at home and they all resulted from simple domestic accidents that were preventable (Ghaffar et al., 2008).

1.9 Kuwaiti System in dealing with cases of death

In Kuwait, the forensic specialist team usually works in collaboration with all other Forensic Departments in Ministry of Interior. The Forensic Personnel in this Ministry includes the Forensic Medical Examiners, the Forensic Pathologists, the Laboratory technicians, the Photographer and the crime scene officer. The working environment of Forensic Examiners is rather stressful. They are under constant pressure to work quickly under difficult circumstances to establish the cause of death and to facilitate justice to a speedy end. Establishing culpability in many crimes depends, to a large extent, on the scientific evidence gathered by investigation team. Scientific progress has widened the scope of modern day investigation in un-natural death cases.

In Kuwait, the Forensic Medicine Examiners and the Crime Scene Officers work hand in hand with the rest of Forensic experts. Their job is to determine the mechanism used for death. For example, in cases of strangulation, they must look for usual features of bruises and abrasions on the neck. Furthermore, they must inspect the materials used for strangulation such as a towel, rope, scarf or any piece that is used.

1.10 Types of cases received

Not all cases are referred to the GDCE for Forensic Examination, but, whenever any of the following circumstances exist, the cases are referred for investigations:

1. A sudden or violent death whether apparently suicidal, homicidal or accidental, including but not limited to deaths apparently caused accidentally or contributed to by thermal, traumatic, chemical, electrical or radiation injury or a complication of any of these or by drowning or suffocation.
2. If death occurs at home, and the patient is not under medical care, it must be reported to the police sector in the area; in turn, they will contact Forensic Medicine Department for a Forensic Team to be made available at the scene. The Forensic Medical Examiner evaluates all the circumstances concerning the body and determines the causes of death based on a history obtained from whoever can provide the most detailed information.
3. In either maternal or foetal death due to an abortion, pregnancy, perinatal (prenatal, natal, postnatal) death and or any death due to or related to a sex crime.
4. A death where circumstances are suspicious, obscure, mysterious or otherwise unexplained or where, in the written opinion of the attending physician, the cause of death is not determined.
5. A death where alcohol or any drug abuse may have been a contributory cause.
6. Deaths due to industrial disease or related to the deceased employment.
7. Death due to neglect by others, including some cases of infant deaths and child abuse.
8. Allegation of and/or medical malpractice and mismanagement

9. Death during police custody

1.11 Laws of Kuwait

As mandated by Kuwait law, the Office of the Medical Examiner is responsible for investigating the death of any person who dies within the whole area of Kuwait (Al-Dousery, 1994). All deaths include results of criminal violence, or neglect, or by casualty or by suicide, or suddenly when in apparent health, or when unattended by a physician, when a person is confined in a public institution other than a hospital, infirmary or nursing home, or in any suspicious or unusual manner. Full forensic autopsies are performed that routinely include an initial investigation, external and internal examination, toxicology and histology, and for these deaths to be reported to the criminal evidence sector in the (FMD). The Forensic Examiner informally reports the results of the autopsies of all cases (Al-Dousery, 1994). In all death cases, a formal report is available to legal authority and others after laboratory results are returned. This may take four to six weeks. Death sentences in Kuwait and other Gulf States are carried out by hanging, and they must be signed by the Emir (President of Kuwait) to be implemented. The laws of Kuwait regarding killing, injuring, battery, and harming are referred to in the present study (See Appendix 1, Laws of Kuwait). Any crime committed in Kuwait against an infant or a child is considered the same as killing or injuring an adult. The laws regulating such offenses and crimes outline definitions of unnatural death and penalties for anyone culpable.

The medico-legal autopsies are performed by the Medical Examiners and by the Forensic Pathologists. The Medical Examiners control the whole system of death investigation and it is the responsibility of the Kuwait GDCE to

generate death certificates as to cause and manner of death and issues a letter to a specified municipal office to obtain permission to bury the body. The GDCE-FMD, in conjunction with Kuwait University-Faculty of Medicine, is responsible for the education of 5th- 7th year students and specialists. The Forensic Department also participates in training programme for Medical Faculty resident Pathologists. In addition, programmes are provided for several scientific colleges in Kuwait and out abroad.

1.12 Aims of the research

1. The primary aim of this study was to investigate the medico-legal cases of un-natural deaths in Kuwait and to identify the patterns of suicide, homicide and accidents from the period of 2003 and 2009.
2. The study focused on such non-invasive techniques as alternative procedures compared to the traditional autopsy to determine the causes of un-natural deaths (either via suicide, homicide or accident) employing virtual autopsy via MRI and CT scanning. This was to establish the usefulness of these novel techniques and the feasibility of either replacing or adding them to the Forensic laboratories in Kuwait.
3. To determine the length of DNA survival in Kuwait as characterized by hot weather in the summer and cold in the winter.
4. To examine forensic demographic data of all categories of violent deaths and to describe any emerging epidemiological patterns of these cases.

5. To distinguish between age groups, gender, regional governments, marital status, religion and nationalities on the distribution of reported medico legal cases of unnatural deaths in Kuwait.
6. To compare and contrast the pattern of suicides, homicides and the pattern of accidental deaths in Kuwait compared to other Middle East countries.
7. To investigate the success rate reported for medico-legal cases of unnatural deaths using the virtual autopsy technology, to compare the current procedures involving autopsy examination by the (FMD), and to help adopt a reliable international system for death registration, data collections, storing and statistical analysis, as well as publication of the data.

CHAPTER 2

Epidemiology of Un-natural Deaths In Kuwait during 2003-2009

2.1 Introduction

As mandated by Kuwait law, the Office of the Medical Examiner is responsible for investigating the death of any person in Kuwait as a result of criminal violence, neglect, casualty, suicide, or sudden death, whether in apparent health, unattended by a physician, confined in a public institution other than a hospital, infirmary or nursing home, or in any suspicious or unusual circumstances (Al-Dousery, 1994). Complete forensic examinations are performed that routinely include an initial investigation, external and internal postmortem examinations, toxicology and histology for these deaths to be reported to the criminal evidence sector in the (FMD) in the (GDCE) in Kuwait. This proposed study investigated the pattern of un-natural deaths in Kuwait, one of the Gulf States which incorporate a wide variety of multi-ethnic communities between 2003 to 2009. In this chapter, the effects of all demographic factors on each aspect of unnatural deaths in Kuwait, including homicidal, suicidal and accidental causes, are studied. Moreover, the rate of unnatural deaths in the Middle East region in general was also reviewed and compared with other parts of the world. The ultimate goal is to demonstrate that there are public health concerns that have not been addressed by the Region's Governments due to the poor accountability of un-natural deaths in the Middle East.

2.2 Materials

The materials of this study had been obtained from three sources. They included the following:-

1. General Department of Traffic which is one sector in Ministry of Interior, Statistic Section. These data included all registered death cases documented in the registration files of traffic death cases in Kuwait. All Road Traffic Accidents in Kuwait are normally transferred to FMD for Medico-legal investigations.
2. The second and the main source of the research data were collected from the (FMD), an affiliate of the (GDCE) in which cases were referred by Justice, Police Authorities and from Hospital Investigator to (GDCE). These cases were examined by Forensic Medical Examiners.
3. The third group of materials included cases that had been referred to Pathology Department for gross and microscopic examinations of specimens that were taken from cadavers in order to find underlying aetiological factors related to the cases and asserting causes of death. All other materials and facilities were available at the GDCE in Kuwait.

In addition, records of the Crime Scene Department were reviewed for each case to obtain demographic data. This data included a summary of the case, a brief account of the circumstances and all the available recorded data on individual Involved. Forms were completed for all cases to record the details of each death.

2.3 Methods

2.3.1 Research design

This descriptive study was conducted over the period of seven years from 2003 to 2009. This study involved all the un-natural deaths in all the six Governorates in Kuwait. As described, the data were collected from the (FMD), an affiliated section of the (GDCE) Head Quarter, Ministry of Interior, Kuwait. This study had a relevant clearance from the Ethics Committees from GDCE and University of Central Lancashire to conduct the investigations.

The manner of death was classified as follows: accident, homicide and suicide. Manner of death was classified according to “A Guide for Manner of Death Classification” issued by the National Association of Medical Examiners (Hanzlick et al., 2002).

2.3.2 Data collection and preparation

The material of this study constituted a total of 5,703 reported un-natural deaths referred for Medico-legal examination during the period of January 2003 through December 2009. In order to identify the cause of death in each case, a full review of the data was made. This included epidemiological data, scene examination, radiographic investigations and a complete autopsy study which entailed histo-pathological examination and toxicological screening to identify each cause of death.

2.3.3 Statistical Analysis

This was a retrospective research study for the period 2003 to 2009, and the data were related to all reported medico-legal cases of un-natural deaths in Kuwait covering the six Governorates (Ahmady, Jahraa, Hawaly, Mubarak Al-Kabeer, Farwaniya and Al-Asema "Capital"). The data were analyzed using manner of death, age, gender, governorate. The manner of death was classified as follows: accident, homicide and suicide.

The following definitions are verbatim following (Hanzlick et al., 2002):

- Accident applied when an injury or poisoning causes death and there was little or no evidence that injury or poisoning occurred with the intent to harm or cause death. In essence, the fatal outcome was unintentional.
- Suicide resulted from an injury or poisoning due to an intentional, self-inflicted act, committed to inflict self-harm or cause the death of oneself.
- Homicide occurred when death results from a violent act committed by another person to cause fear, harm or death. Figure 2.1 described the sub-classification of unnatural death categories.

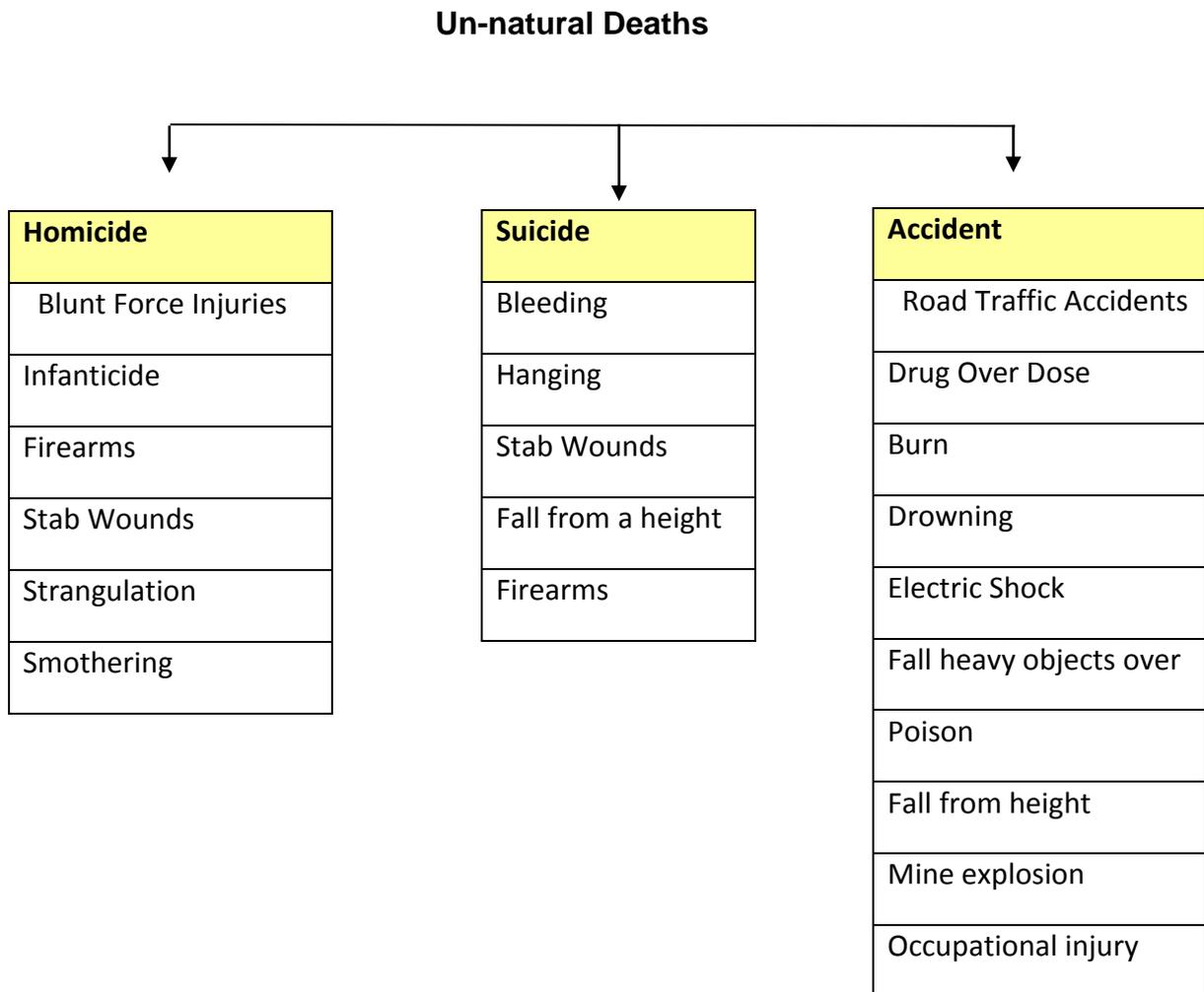


Figure 2.1: A flow diagram showing categories of un-natural deaths

The data were retrieved from the Archives of the Department of (FMD), (GDCE) in the Interior Ministry of Kuwait. These data consisted of autopsy records, clothes examination and general physical examination of the body as well as the pathological, toxicological and crime scene investigations. Demographic information regarding governorate, gender, age, religion, marital status, occupation and death category were collected.

All definitions employed in this study were those previously used by WHO and Center for Disease Control, USA. The Regional Ethical Review Board of General Department of Criminal Investigations in the Interior Ministry of Kuwait and the Ethics Committee in the School of Forensic and Investigative Sciences at the University of Central Lancashire approved all the procedures used, provided the confidentiality of each case was maintained.

In this study, data were analyzed using SPSS 17.0 (Chicago, USA). Statistical analyses involved computation of descriptive statistics for all demographic factors such as age, sex ratio in the sample, ethnic background, occupational level, and religion. Descriptive statistics were used to show the percentage of reported medico legal cases in different categories according to each year. ANOVA Test was used to compare more than two groups depending on the normality of the variables. A value of $p < 0.05$ was taken as significant.

2.4: Results

2.4.1 Reported natural and un-natural death cases in the General Department of Criminal Evidence during the period 2003-2009

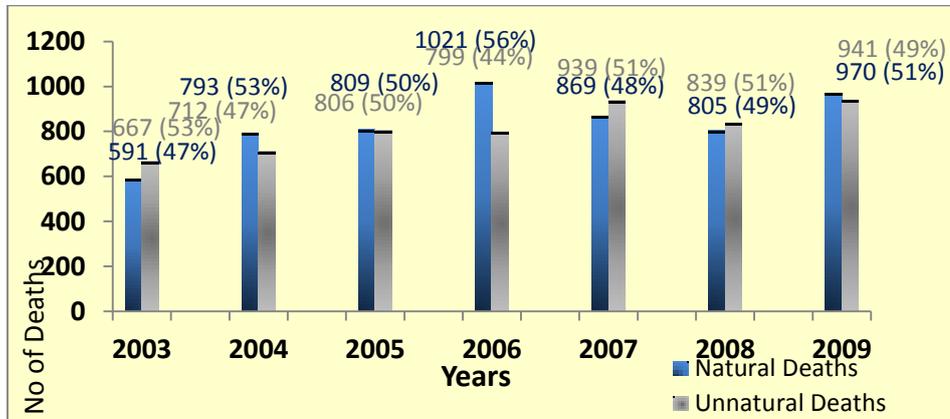


Figure 2.2: Bar charts showing the number and percentage of reported medico-legal natural and un-natural deaths cases in Kuwait between 2003 and 2009.

The results in figure 2.2 reveal that the highest number of natural deaths was reported in year 2006 accounting for 1021 (56%), whereas, in year 2009, 941 (49%) cases of un-natural deaths were reported. The lowest cases of natural and un-natural deaths were reported in 2003 accounting for 591 (47%) and 667 (53%) cases, respectively.

2.4.2 Total number and percentage of reported unnatural death cases in the GDCE during the period 2003-2009

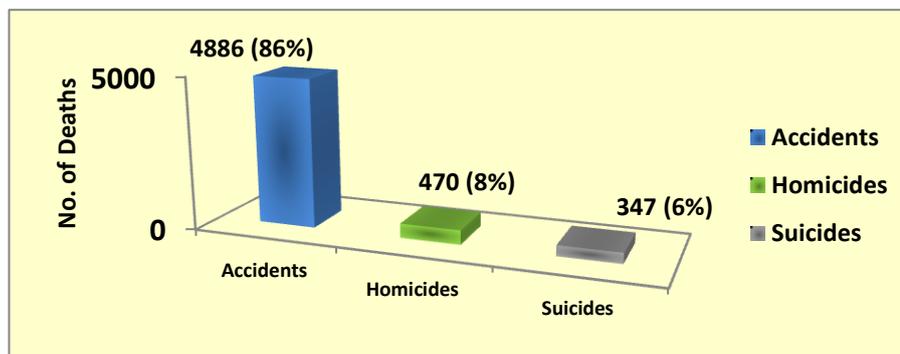


Figure 2.3: Bar charts showing the total number and percentage of reported unnatural causes of deaths in Kuwait between 2003 and 2009.

The results in figure 2.3 show that accidental deaths in Kuwait are the major cause of un-natural deaths. Accidents constituted 4,886 cases of deaths within the 7 years of the study and accounted for 86%. This was followed by homicidal deaths which constituted 470 (8%) cases while suicidal deaths account for only 347 (6%) cases of the total reported cases of un-natural deaths.

2.4.3 Total number and percentage of reported un-natural death cases according to Kuwait Governorates from 2003 to 2009.

Table 2.1: Frequency table showing the total number and percentage of reported medico-legal cases of un-natural deaths

| Governorate | Un-natural Death Cases | | |
|-------------------|------------------------|-----------|------------|
| | Homicide | Suicide | Accident |
| Jahra | 80 (17%) | 54 (16%) | 934 (19%) |
| Farwania | 176 (37%) | 137 (40%) | 1418 (29%) |
| Hawally | 76 (16%) | 36 (10%) | 739 (15%) |
| Ahmady | 50 (11%) | 68 (20%) | 930 (19%) |
| Mobarak Al-Kabeer | 24 (5%) | 14 (4%) | 239 (5%) |
| Capital | 64 (14%) | 38 (11%) | 626 (13%) |

The results in table 2.1 shows that Farwania Governorate ranks first with 1418 in cases of accidental deaths. In the homicidal death category. Mubarak Al Kabeer Governorate demonstrated the lowest rate of homicidal deaths (5%). There were significant ($p < 0.000$) differences in the percent of death categories in various Governorates.

2.4.4 Total number and percentage of reported un-natural death cases in Kuwait according to gender during the period 2003 to 2009.

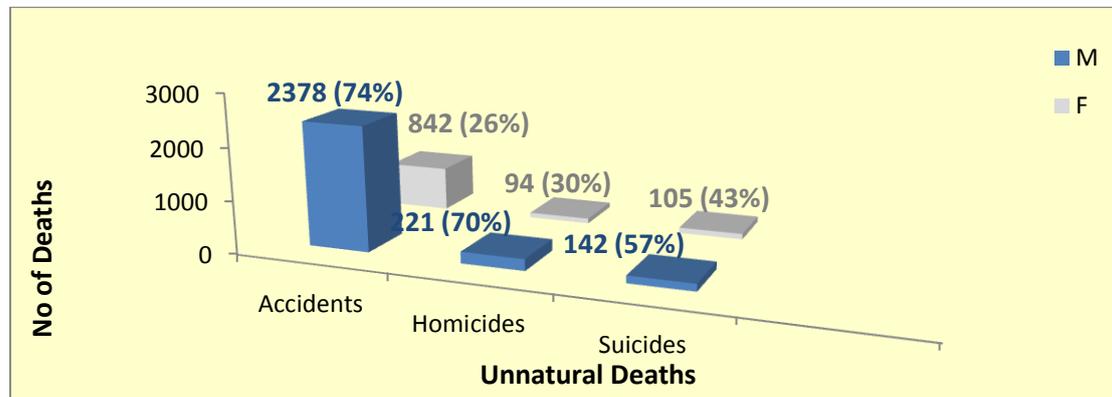


Figure 2.4: Bar charts showing the number and percentage of reported medico-legal cases of un-natural death. (The data was corrected according to 1.7: 1 male to female ratio). The results in figure 2.4 show that males outnumber females in all categories. In accidental death cases, there were 2378 males (74%), compared to 842 females (26%). In the suicidal category, the number of males was 142 (57%) cases compared to female (105 cases 43%). The results also show a significant difference ($P < 0.001$) in all death categories of males compared to females.

2.4.5 Total number and percentage of reported un-natural death cases in Kuwait according to age group between 2003 and 2009

Table 2.2: Frequency table according to different age groups during 2003 and 2009 showing the total number and percentage of reported medico-legal cases due to accidental, homicidal and suicidal causes.

| Age Group | Un-natural Death Cases | | |
|-----------|------------------------|-----------|------------|
| | Homicide | Suicide | Accident |
| 0-9 | 58 (12%) | 0 (0%) | 503 (10%) |
| 10-19 | 22 (5%) | 8 (2%) | 493 (10%) |
| 20-29 | 124 (26%) | 126 (36%) | 1288 (26%) |
| 30-39 | 131 (29%) | 149 (43%) | 1186 (24%) |
| 40-49 | 84 (18%) | 43 (13%) | 784 (16%) |
| 50-59 | 35 (7%) | 18 (5%) | 350 (8%) |
| 60+ | 16 (3%) | 3 (1%) | 280 (6%) |

The results in table 2.2 reveal that the highest rate (or number and percentage) in accidental deaths was in 20-29 year old age group who accounted for 1,288 cases (26%), followed by the 30-39 year old age group who accounted for 1186 cases (24%). The lowest number of accidental deaths was registered in the age group of over 60 years (280 cases or 6%). In the suicidal category, the 30-39 year old age group ranks first (149 cases or 43%), followed by the 20-29 year old age group (126 cases or 36%) and the 40-49 year old age group (43 cases or 12%). There was no suicidal case for the 0-9 year old age group. In contrast the over 60 year old age group accounted for occurred in 3 cases (1%) of suicidal deaths. Interestingly, the 30-39 year old of age group ranks first in homicidal deaths (131 cases or 29%), followed by 20-29 year old age group (124 cases or 26%) and by the 40-49 year old age group (84 cases or 18%). The least number of homicidal deaths was registered among age group over 60 years (16 cases or 3%).

2.4.6 Total number and percentage of reported un-natural death cases in Kuwait according to nationalities between 2003- and 2009

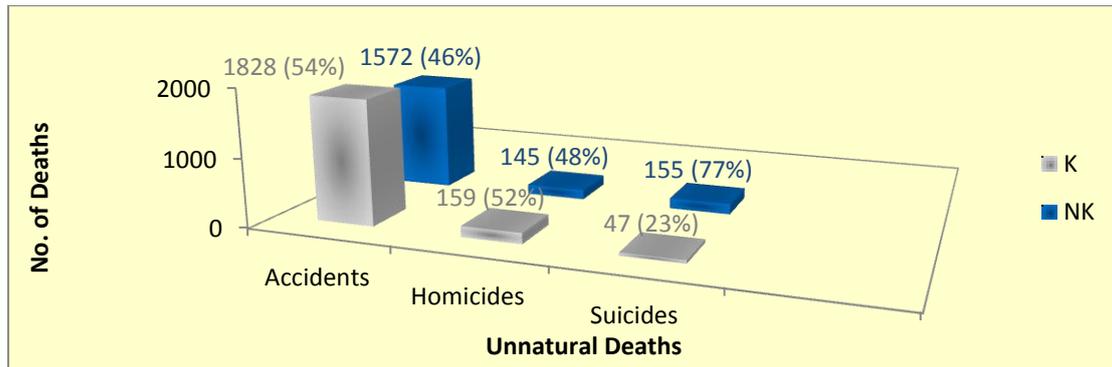


Figure 2.5: Bar charts showing the total number and percentage of reported medico-legal cases of un-natural death cases according to nationalities in Kuwait between 2003 and 2009. (Data was corrected according to Kuwaiti and Non-Kuwaiti ratio of 1:1.94)

The results presented in figure 2.5 reveal that more Kuwaitis died in deaths due to accidents and homicides during the duration of this study. Kuwaitis accounted for 1828 cases (54%) of accidental deaths, 159 cases (52%) of homicidal deaths and 47 cases (23%) of suicidal deaths.

2.4.7 Reported medico-legal cases of unnatural deaths in Kuwait to the GDCE according to the years from 2003 to 2009.

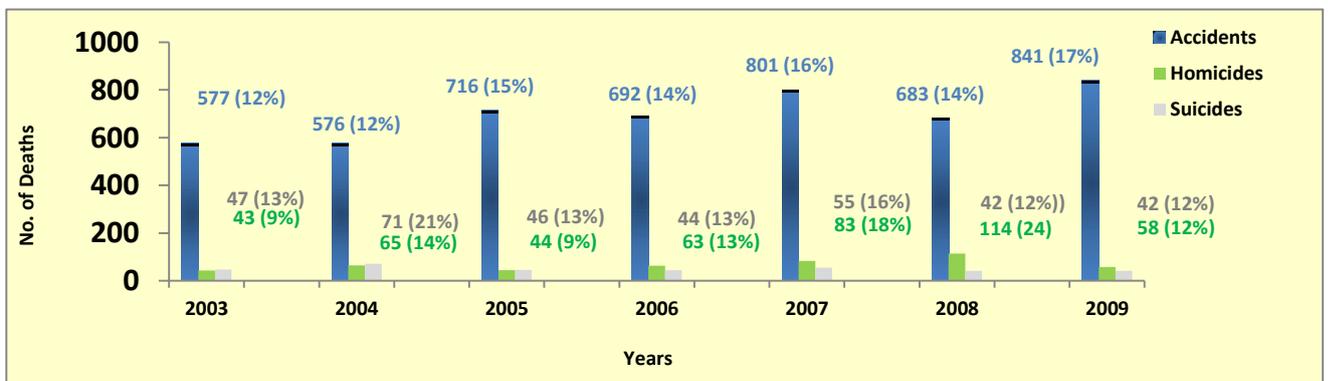


Figure 2.6: Bar charts showing the total number and percentage of reported medico-legal death cases of un-natural death categories in Kuwait between 2003 and 2009.

The results in figure 2.6 reveal that accidental deaths reached their highest peak in 2009 accounting for 841 cases (17%) and lowest rate in 2003 and 2004 accounting for 577 (12%) and 576 cases (11%), respectively. Homicides demonstrated the highest value in 2008 accounting for 114 cases (24%) and lowest value in 2003 accounting for 43 cases (9%). Suicide showed the highest rate in 2004 accounting for 71 cases; (21%) and lowest rate in 2008 and 2009 accounting for 42 cases (12%) for each year, respectively.

2.4.8 Causes, number and percentage of reported medico-legal cases of homicidal deaths in Kuwait between 2003-2009

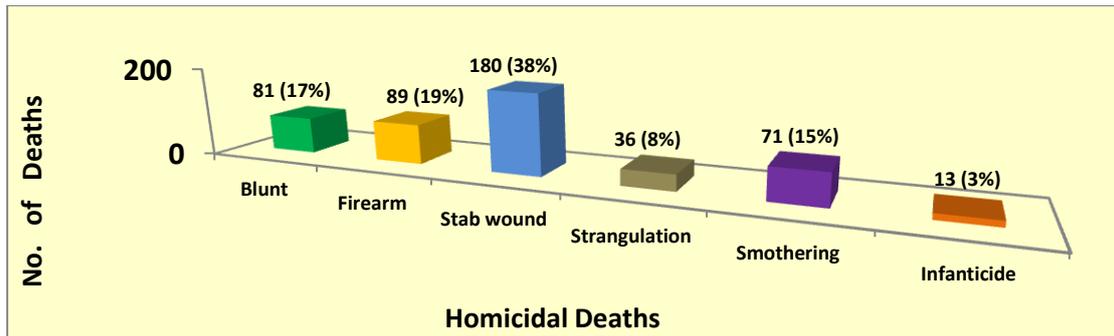


Figure 2.7: Bar charts illustrating the number and percentage of reported un-natural deaths concerning homicide.

The results in figure 2.7 show that stab wound injuries were the leading cause of homicidal death. They accounted for 180 cases (38%) of the total homicidal cases, followed by deaths caused by firearm injuries (89 cases or 19%) and blunt force injuries (81 cases or 17%). The least cases reported were for infanticide, totaling only 13 cases (3%) of the total homicidal deaths, whereas, strangulation accounted for 36 cases (8%) of homicidal deaths.

2.4.9 Causes, percentage and total number of reported medico-legal cases of Homicide in Kuwait according to nationalities between 2003 and 2009

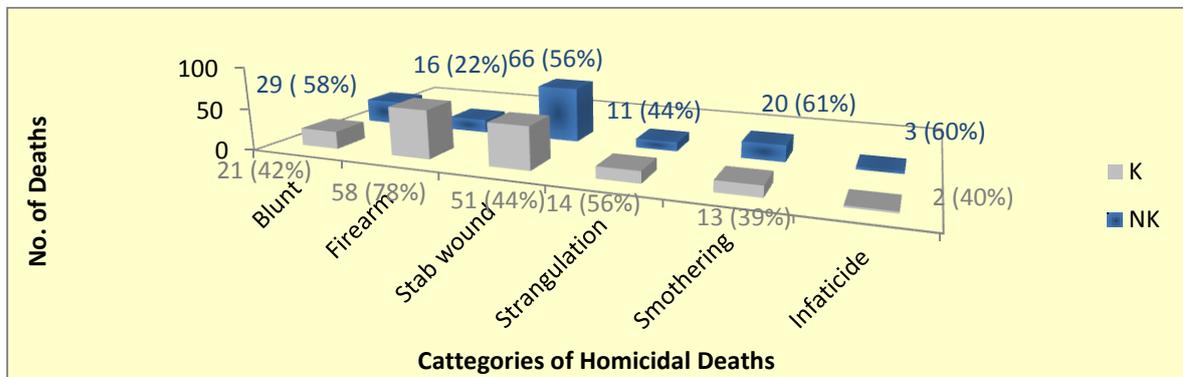


Figure 2.8: Bar charts showing the number and percentage of reported medico-legal cases of homicide in Kuwait according to nationalities (The data was corrected according to 1.94: 1 Non-Kuwaiti to Kuwaiti ratio).

The results in figure 2.8 show that Kuwaiti nationals accounted for the lowest percentage of homicide in all categories compared to non-Kuwaiti nationals, except for deaths due to firearms and strangulation. Typically, Kuwaiti residents accounted for 58 (78%) cases due firearm injuries and 14 (56%) cases for deaths due to strangulation.

2.4.10 Total number, percentage and causes of reported medico-legal cases of Homicidal deaths in Kuwait according to gender.

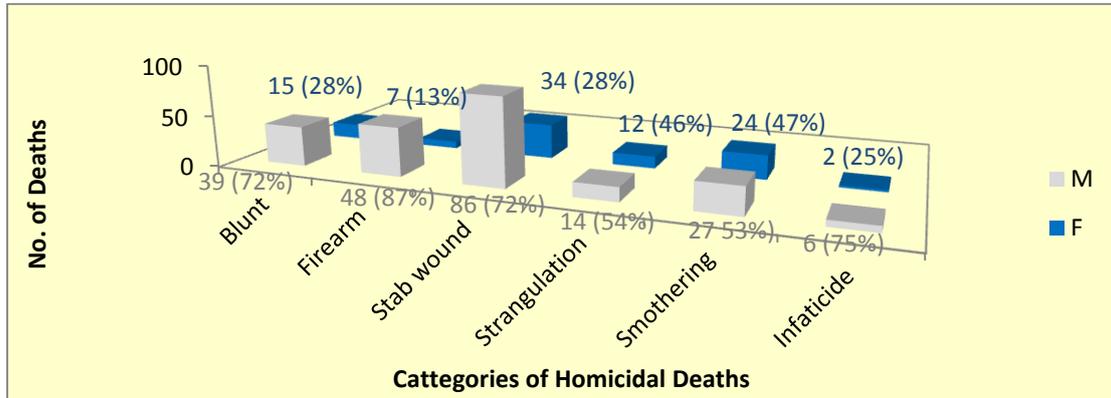


Figure 2.9: Bar charts showing the reported medico-legal cases of un-natural death cases due to homicide in Kuwait according to gender. (Data was corrected according to male to female skew ratio 1.7: 1).

The results presented in figure 2.9 show that males outnumbered females in all homicidal categories. Males showed the highest rate in deaths due to stab wound (86 cases or 72%) compared to females (34 cases or 28%). The results also show a significant difference ($P < 0.001$) in death categories of males compared to females.

2.4.11 Reported medico-legal cases of homicidal deaths due to blunt force injuries that were reported to the GDCE in Kuwait according to age group

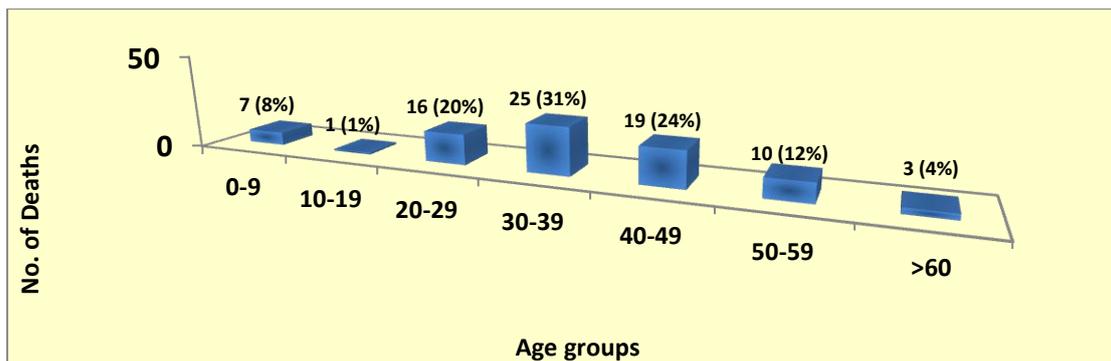


Figure 2.10: Bar charts showing the number and percentage of reported medico-legal cases of homicidal deaths due to blunt force injuries in Kuwait.

The results in figure 2.10 revealed that there was a significant difference ($p < 0.001$) using ANOVA test in the total number of homicidal deaths due to blunt force injuries according to age groups. The major age group that had higher mortality rate of homicide was 30-39 years of age accounting for 25 cases (31%).

3.4.12 Reported homicidal deaths by the use of firearm injuries that were reported to the GDCE in Kuwait according to age group between 2003 and 2009

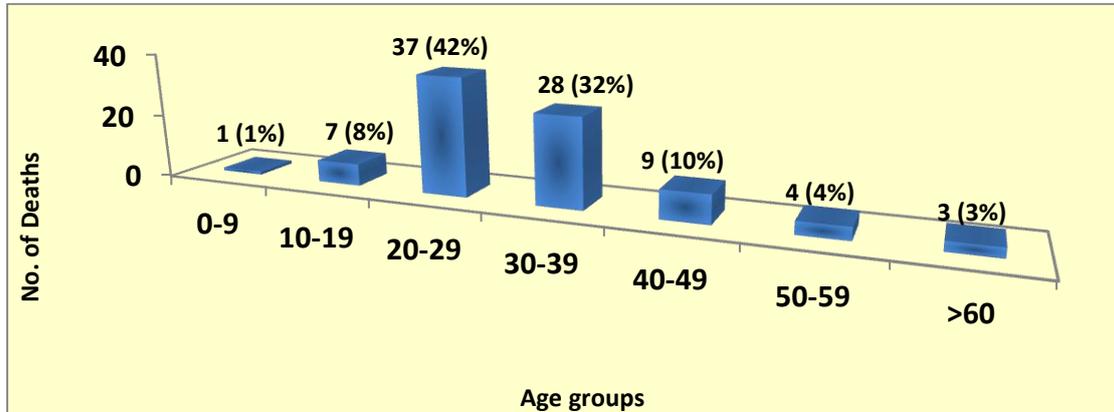


Figure 2.11: Bar charts showing the reported homicidal deaths by the use of firearm injuries in Kuwait as classified by different age groups.

The results in figure 2.11 have revealed that there was a significant difference ($p < 0.001$) using ANOVA test in different age groups. The major age group that had highest mortality rate was 20-29 years of age accounting for 37 cases (42%), followed by age group 30-39 years who accounted for 28 cases (32%).

2.4.13 Reported medico-legal homicidal deaths due to stab wound injuries that are reported to the GDCE in Kuwait according to age group

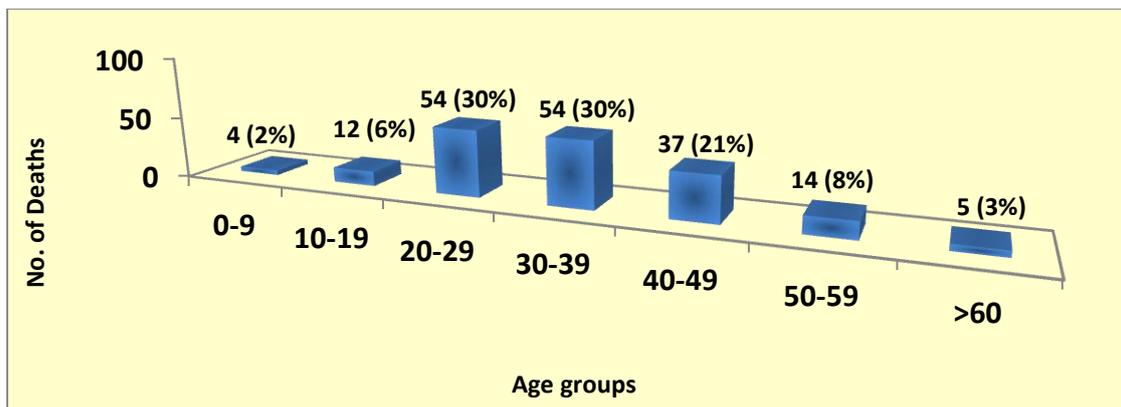


Figure 2.12: Bar charts showing the reported medico-legal homicidal deaths due to stab wound injuries in Kuwait as classified by different age groups.

From figure 2.12 it can be seen that similar results were found in the age groups 20-29 years and 30-30 years and they accounted for 54 (30%) cases each. The lowest number was reported for the age group 0-9 years who accounted for only 4 (2%) cases due to stab wound injuries.

2.4.14 Reported medico-legal cases of homicidal deaths due to strangulation that were reported to the GDCE in Kuwait according to age group

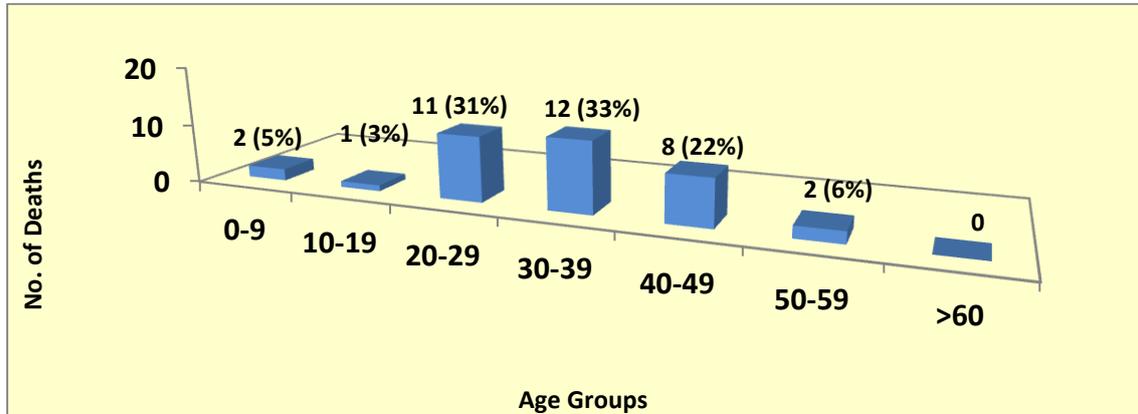


Figure 2.13: Bar charts showing the reported homicidal deaths due to strangulation in Kuwait between 2003 and 2009 as classified by different age groups.

The results in figure 2.13 reveal that, the highest mortality rate of homicide was among the age group 30-39 years old and they accounted for 12 (33%) cases. In contrast, no case was reported in age group 60 years over.

2.4.15 Reported medico-legal cases of homicidal deaths due to smothering injuries in Kuwait according to age group.

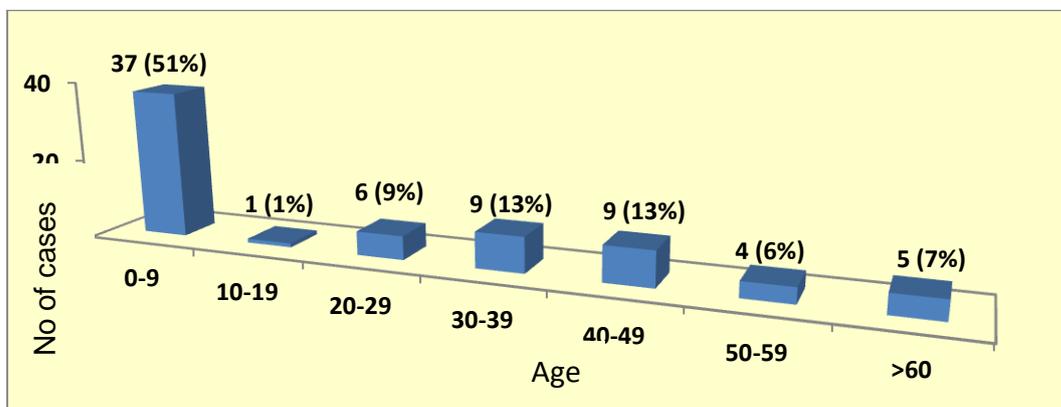


Figure 2.14: Bar charts showing the reported medico-legal cases of homicidal deaths due to smothering injuries in Kuwait as classified by different age groups.

The results in figure 2.14 show that, the major age group that had highest mortality rate of homicide was 0-9 year old accounting for 37 cases (51%). The least cases were reported for age group of 10-19 years accounting for 1 case (1%) of total homicidal cases due to smothering.

2.4.16 Areas of the body involved in fatal injuries of homicidal deaths

Table 2.3: Table showing different parts of the body involved in fatal injuries.

| Areas involved | Types of injuries | | |
|----------------|-------------------|----------|----------|
| | Firearm | Sharp | Blunt |
| Head | 24 (23%) | 50 (29%) | 51 (62%) |
| Neck | 5 (10%) | 64 (36%) | 12 (15%) |
| Chest | 42 (47%) | 42 (23%) | 4 (5%) |
| Abdomen | 10 (17%) | 18 (10%) | 2 (3%) |
| Upper limbs | 03 (3%) | 4 (2%) | 5 (6%) |
| Lower limbs | 05 (6%) | 2 (1%) | 7 (9%) |

The results in table 2.3 reveal that, the head is the major and most frequent area involved in blunt force injuries accounting (63%). In firearm injuries, the chest was the most involved area accounting for 42 cases (47%).

2.4.17 Total number, percentage and modes of strangulation

Table 2.4: Table illustrating the total number and percentage of reported medico-legal cases of unnatural deaths due to strangulation between 2003 and 2009.

| | Ligature strangulation | | Manual strangulation | | Total |
|-----------------------------|------------------------|----------|----------------------|---------|-------|
| | Male | Female | Male | Female | |
| Conjunctival petechiae | 26 (72%) | | 10 (28%) | | 36 |
| | 9 (35%) | 17 (65%) | 8 (80%) | 2 (20%) | |
| fractures of the hyoid bone | 12 (67%) | | 6 (33%) | | 18 |
| | 8 (67%) | 4 (33%) | 4 (67%) | 2 (33%) | |
| Total | 38 | | 16 | | |

2.4.18 Total number, percentage and causes of accidental medico-legal death cases that were reported to the GDCE in Kuwait.

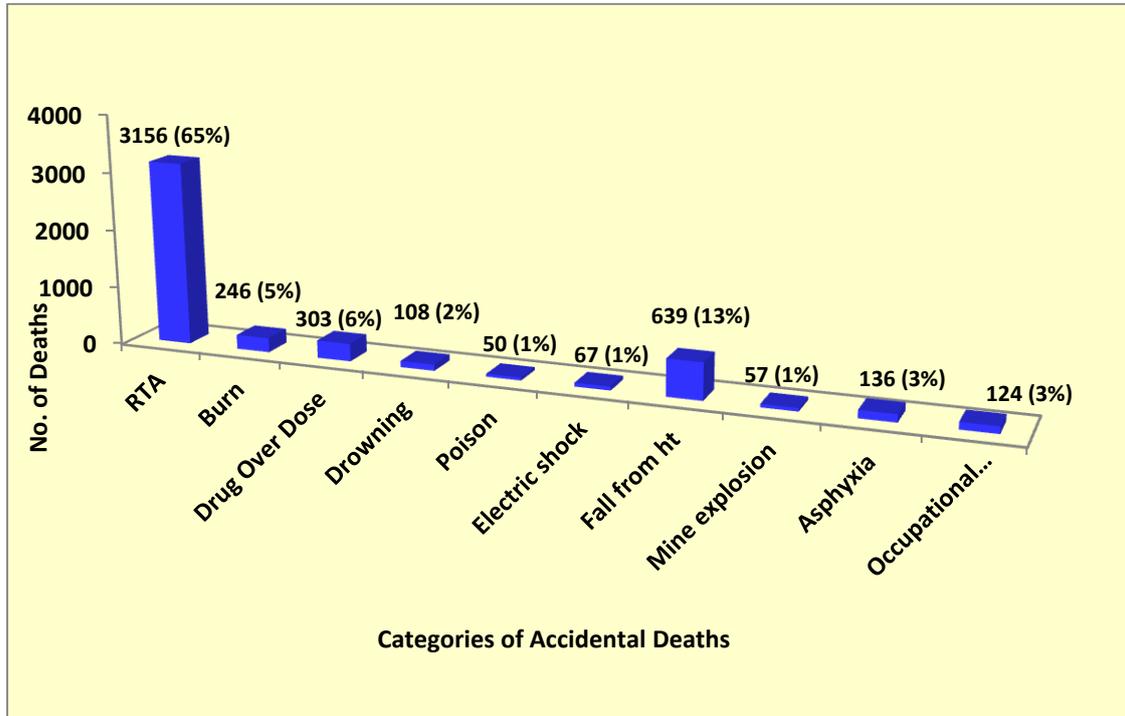


Figure 2.15: Bar charts showing both the number and percentage of reported medico-legal cases of un-natural deaths due types of accidents.

The results in figure 2.15 show that road traffic accidents were the leading cause of accidental deaths in Kuwait accounting for 3,156 cases (65%) of the total reported accidental deaths. Deaths due to accidental burn injuries were 246 cases (5%) of the total accidental cases. The lowest reported cases were for deaths due to poisoning and they accounted for 50 (1%) reported cases.

2.4.19 Reported medico-legal cases of accidental deaths according to nationalities between 2003 and 2009.

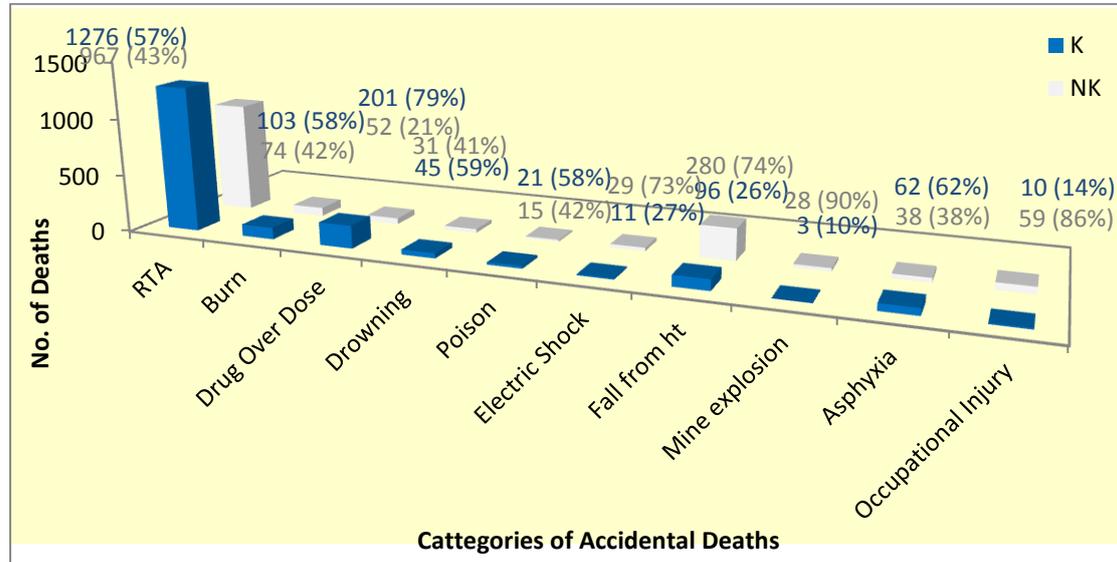


Figure 2.16: Bar charts illustrating the total number, percentage and causes of medico-legal cases of un-natural deaths due to accidents that were reported to the (GDCE) in Kuwait according to nationalities between 2003 and 2009. (Data were corrected for skew ratio of Kuwaiti and Non Kuwaiti 1:1.94)

The results presented in figure 2.16 show that Non-Kuwaiti Nationalities had the highest incidence of mortality in accidental deaths due to electric shock, fall from height, mine explosion and occupational injury. Deaths due to RTA in Kuwaiti accounted for 1276 (57%) cases, burn injuries accounted 103 (58%) cases, drowning accounted for 45 (59%) cases, poison accounted for 21 (58%) cases, asphyxia shocks accounted for 62 (62%) cases, accounted for 201 (79%) cases, mine explosions accounted for 54 (95%) cases and asphyxia accounted for 74 (54%) cases. There were significant differences $p=0.000$ using ANOVA test in all categories of accidental deaths according to the two nationalities.

2.4.20 Total number, percentage and causes of medico-legal accidental death cases that were reported to the GDCE in Kuwait according to gender

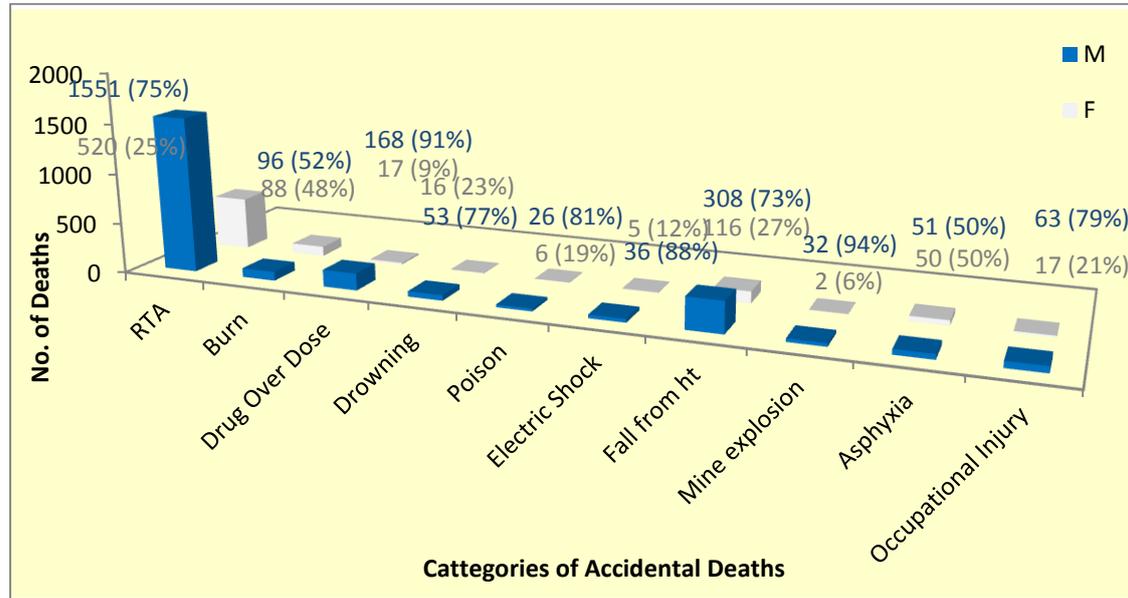


Figure 2.17: Line graphs showing the number, percent and causes reported unnatural deaths due to accidents according to gender. (Data was corrected based on male to female ratio of 1.7: 1)

Figure 2.17 shows the number and percentage of the reported medico-legal cases of un-natural deaths due to accidents to the GDCE in Kuwait according to gender between 2003 and 2009. The results show that male victims outnumbered female victims. Males showed a higher rate (incidence) in death cases due to RTA. Typically, males accounted for 1551 (75%) cases compared to females who accounted for 520 (25%) cases. In the cases of accidental deaths due to burn injuries males accounted for 96 (52%) cases and females accounted for 88 (48%) cases. The same is also true for deaths due drug over dose males who accounted for 168 (91%) cases compared to females who accounted for 17 (9%) cases. Similarly, males accounted for a higher rate in deaths due to drowning and poisoning, accounted for 53 (77%) cases and 26 (81%) cases, respectively. Also, males accounted for a higher rate in deaths due to falls from heights (308 cases or 73%). Equally, deaths due to mine explosions occurred predominantly among male victim, accounting for 32 (94%) cases. The results clearly show a significant difference ($p < 0.000$) in death categories of male compared to female victims.

2.4.21 Reported medico-legal accidental death cases due to occupational injuries

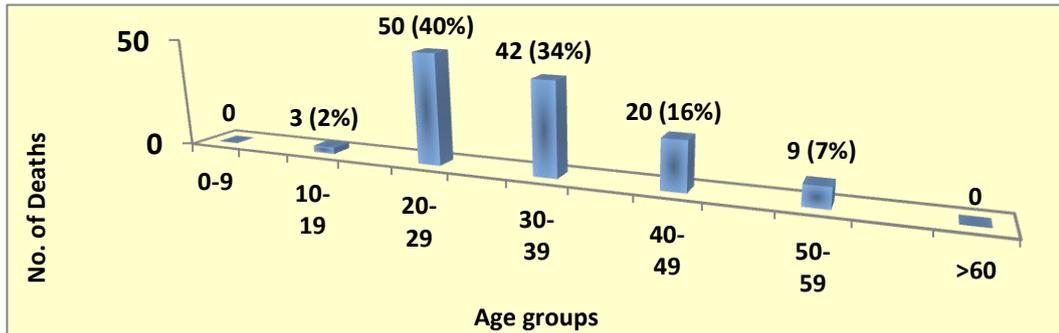


Figure 2.18: Bar charts showing deaths due to occupational injuries in Kuwait according to age group between 2003 and 2009.

The results in figure 2.18 reveal that the highest number of reported cases due to occupational deaths was in the 20-29 year old age group accounting for 50 (40%) cases. This was followed by the 30-39 year old age group who accounted for 42 (34%) cases.

2.4.22 Total number and percentage of medico-legal cases of accidental deaths due to mine explosion reported to the GDCE in Kuwait according to age groups

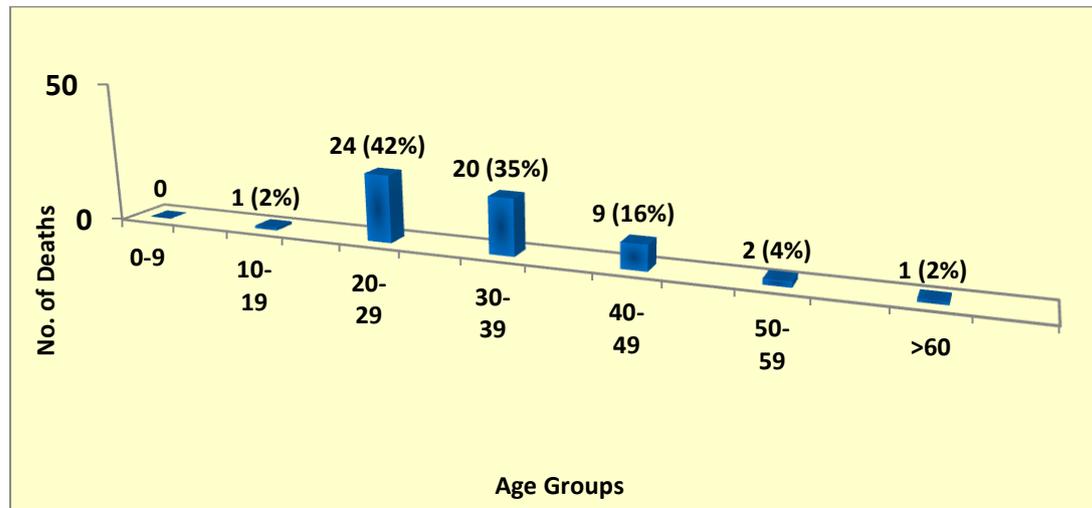


Figure 2.19: Bar charts showing the reported medico-legal cases of un-natural deaths due to mine explosions in Kuwait according to different age groups

The results in figure 2.19 reveal that the highest number of deaths occurred in the age group 20-29 years accounting for 24 (42%) cases. The lowest number of deaths due to mine explosions was reported in both age groups of 10-19 years and 60 years and over accounting for only 1 (2%) case. The age group of 0-9 years had no accidental deaths due to mine explosions.

2.4.22 Total number and percentage of medico-legal cases of accidental deaths due to electrocutions in Kuwait according to different age groups

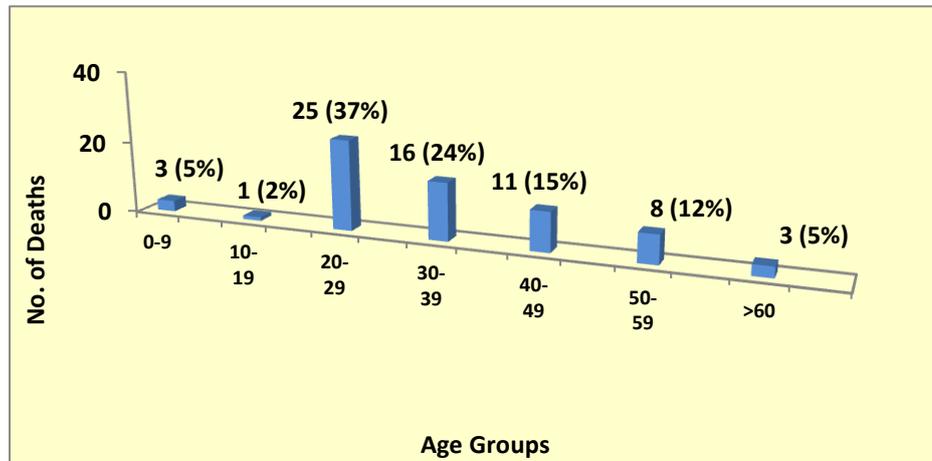


Figure 2.19: Bar charts showing the total number and percentage of reported medico-legal cases accidental deaths due to electrocution and according to different age groups.

The results in figure 2.19 reveal that most deaths from electrocutions occurred in the age group 20-29 years accounting for 25 (37%) cases. The lowest number of reported death cases occurred in age group 10-19 years accounting for only 1 (2%) case.

2.4.23 Total number and percentage of medico-legal cases of accidental deaths due to poisoning in Kuwait according to age groups

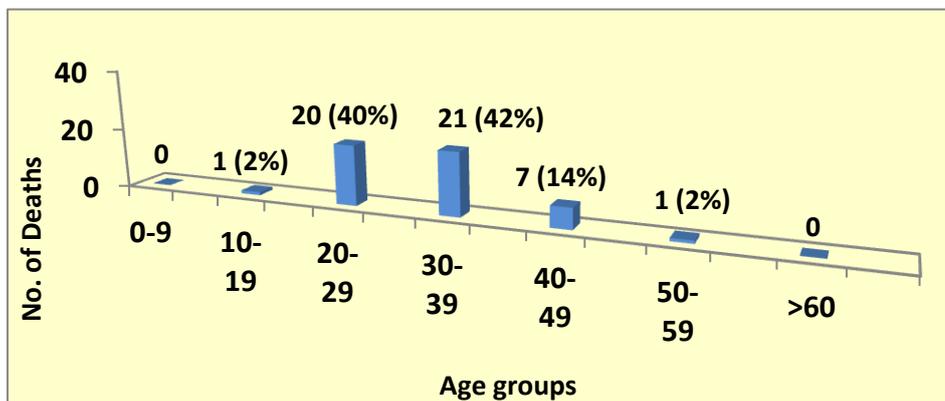


Figure 2.20: Bar charts showing the reported medico-legal accidental cases of unnatural deaths due to poisoning in Kuwait according to different age groups.

The results in figure 2.20 reveal that most victims died in the age group 30-39 years accounting for 21 (42%) cases. Similar results were registered for age groups 20-29 years and 50-59 years accounting for 1 (2%) case each. No case of accidental death due to poisoning was recorded in age groups 0-9 years and over 60 years.

2.4.24 Medico-legal cases of accidental deaths due to drowning in Kuwait according to age groups

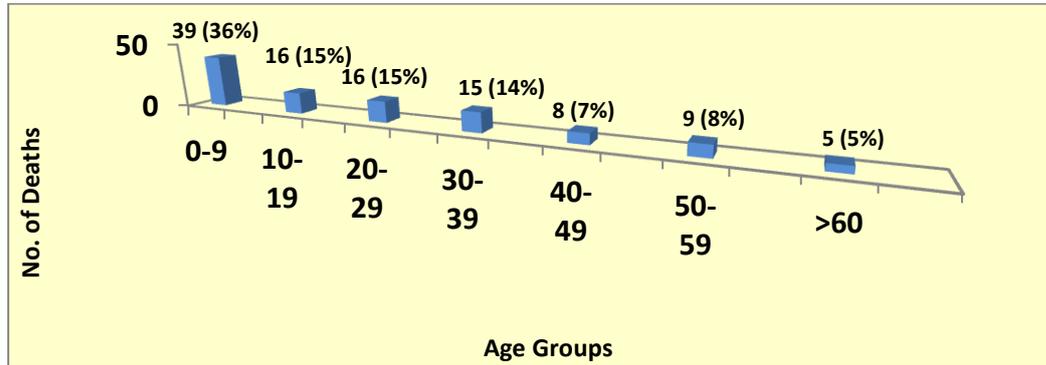


Figure 2.21: Bar charts showing the reported medico-legal cases of un-natural accidental deaths due to drowning in Kuwait according to different age group.

The results in figure 2.21 show that the highest number of reported deaths occurred in age group 0-9 years accounting for 39 (36%). The age groups 30-39 years accounted for 15 (14%) cases, while, the least cases were registered for age groups over 60 years accounting 5 (5%) cases.

2.4.25 Medico-legal cases of accidental deaths due to drug over dose in Kuwait according to age group

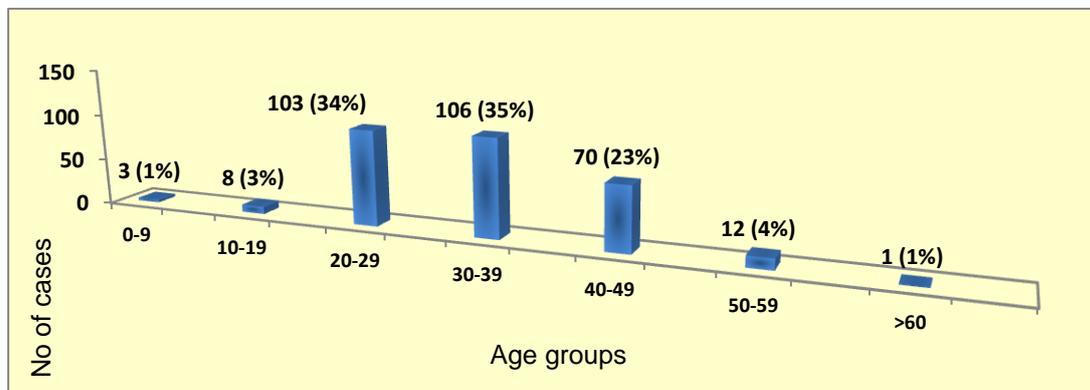


Figure 2.22: Bar charts showing the total number and percentage of reported medico-legal cases of un-natural deaths due to drug over dose in Kuwait according to different age groups between 2003 and 2009.

The results in figure 2.22 reveal that the highest number of deaths occurred in age group 30-39 years accounting for 106 (35%) cases. The results also show that 3 (1%) cases were reported in age group 0-9 years for drug over dose.

2.4.26 Total number and percentage of medico-legal cases of accidental deaths due to burning in Kuwait and according to age group

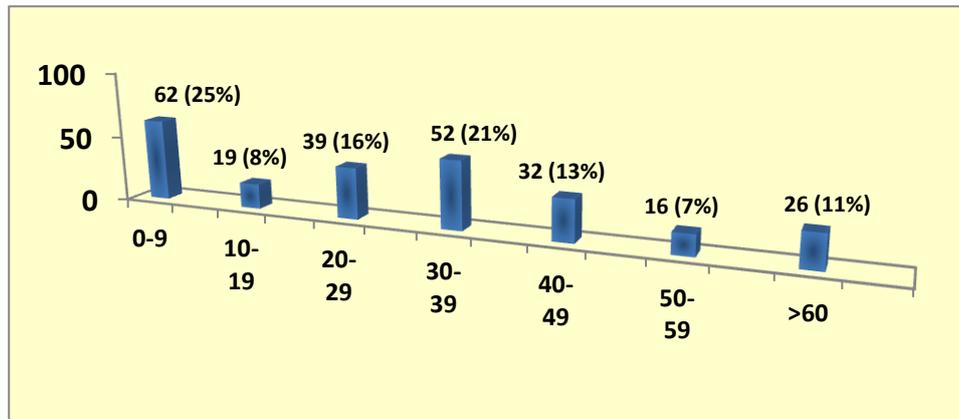


Figure 2.23: Bar charts showing the reported medico-legal cases of un-natural deaths due to burning in Kuwait according to different age groups.

The results in figure 2.23 reveal that the highest number of accidental deaths occurred in age group 0-9 years accounted for 62 (25%) cases. The results also show that the age group 50-59 years accounted for 16 (7%) cases of accidental deaths by burning.

2.4.27 Medico-legal cases of accidental deaths due to road traffic accidents in Kuwait according to age group

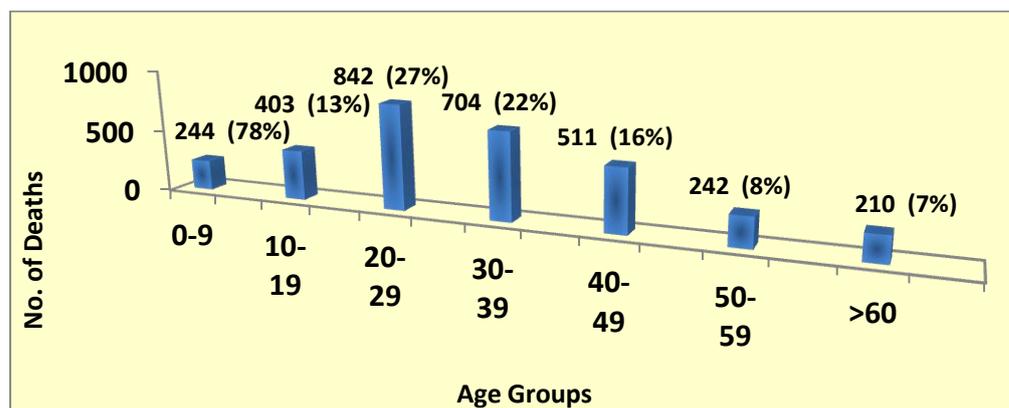


Figure 2.24: Bar charts showing the reported medico-legal cases of un-natural deaths due to road traffic accidents according to different age groups.

The results in figure 2.24 reveal that the highest number of deaths due to RTA was in the age group 20-29 years accounting for 842 (27%) cases, followed by age group 30-39 years accounting for 704 (22%) cases. The least number of RTA was registered in age group over 60 years accounting for 210 (7%) cases.

2.4.28 Medico-legal cases of accidental deaths due to falls from heights in Kuwait according to different age group

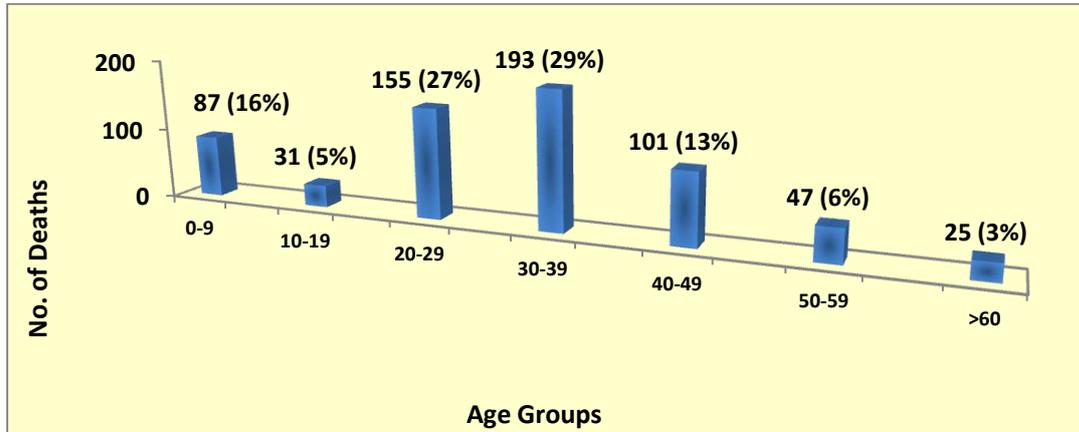


Figure 2.25: Bar charts showing the reported medico-legal cases of accidental deaths as a result from falls from heights and according to different age group.

The results in figure 2.25 reveal that the highest number of accidental deaths due to falls from heights occurred in age group 30-39 years accounting for 193 (29%) cases followed by age group 20-29 years accounting for 155 (27%) cases. The least number of deaths due to falls from heights was registered in the age group over 60 years accounting for 25 (3%) cases.

2.4.29 Total number and percentage of medico-legal cases of accidental deaths due to asphyxia according to different age group

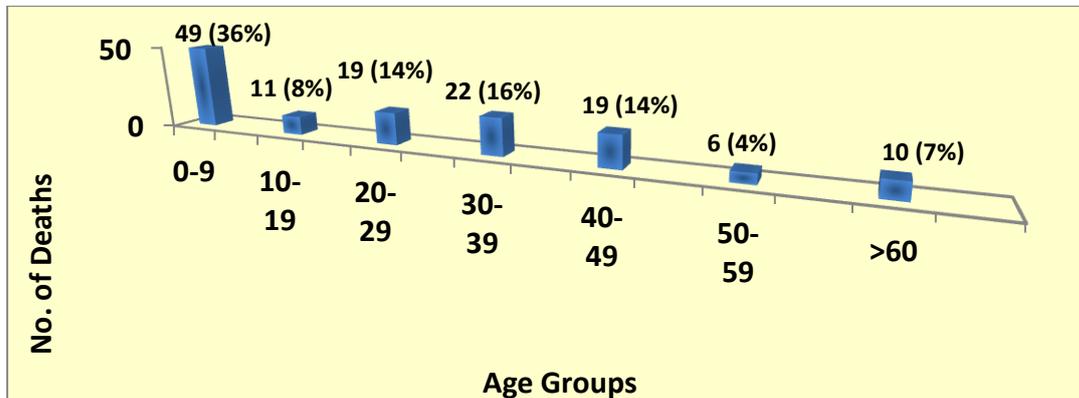


Figure 2.26: Bar charts showing the number and percentage of reported medico-legal cases of un-natural deaths due to asphyxia according to different age group.

The results in figure 2.26 show that the highest number of deaths due to asphyxia was in the age group 0-9 years accounting for 49 (36%) cases, followed by the age group 30-39 years accounting for 22 (16%) cases. The results also reveal that the least number of deaths due to asphyxia was registered in the age group 50-59 years accounting for 6 (4%) cases.

2.4.30 Total number, percentage and causes of medico-legal cases due to suicidal deaths reported to the GDCE in Kuwait

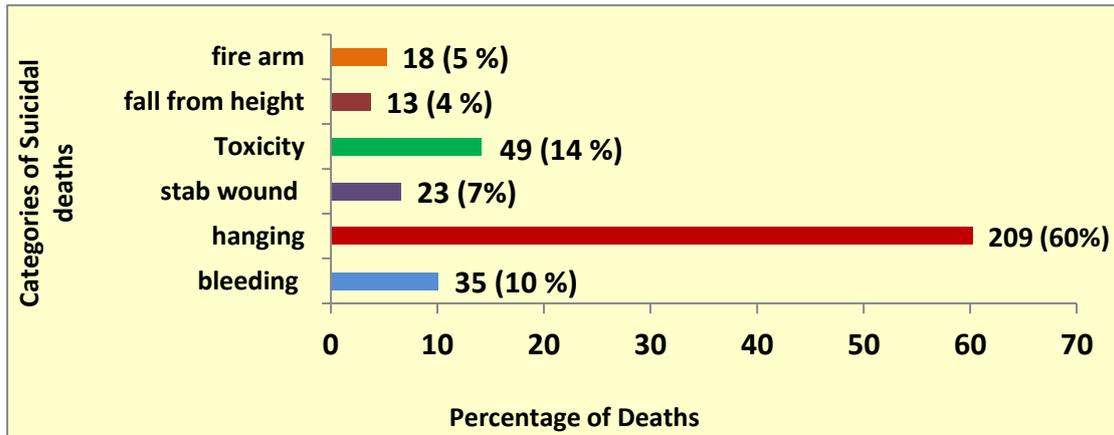


Figure 2.27: Bar charts showing the reported medico-legal cases due to suicidal deaths reported to the GDCE in Kuwait between 2003 and 2009.

The results presented in figure 2.27 show that suicide by hanging accounted for 209 (60%) cases. Suicide by using of firearm injuries and stab wounds were 18 (5%) and 23 (7%) cases, respectively. The results also showed that falls from heights were the least suicidal method employed for killing accounting for only 13 (4%) cases.

2.4.31 Medico-legal suicidal death cases in Kuwait according to nationalities

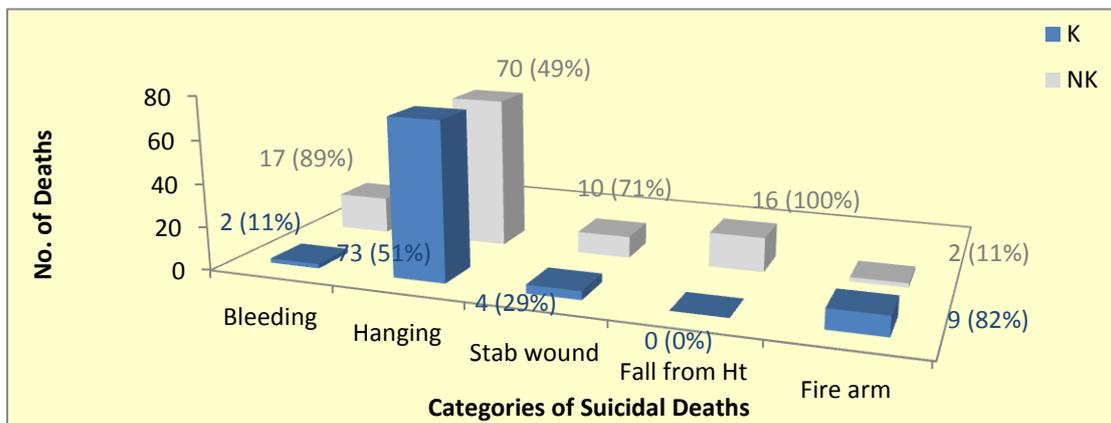


Figure 2.28: Bar charts showing the total number, percentage and causes of medico-legal suicide cases in Kuwait reported to GDCE according to nationalities from 2003 to 2009. (Data was corrected based on Kuwaiti and Non Kuwaiti ratio of 1: 1.94)

The results in figure 2.28 show the different causes of suicides and they indicate that all patterns of suicide were higher in the Non-Kuwaiti population with the exception of suicide by the use of firearm as a method of suicide which was higher in Kuwaiti residents accounting for 9 (82%) compared to 2 (11%) in non-Kuwaiti nationals and also due to suicide by hanging in which Kuwaiti nationals accounting for (51%) compared to (49%). (p value= 0.000; df=5).

2.4.32 Total number, percentage and causes of medico-legal cases due to suicidal death cases reported to the GDCE in Kuwait according to gender between 2003 and 2009

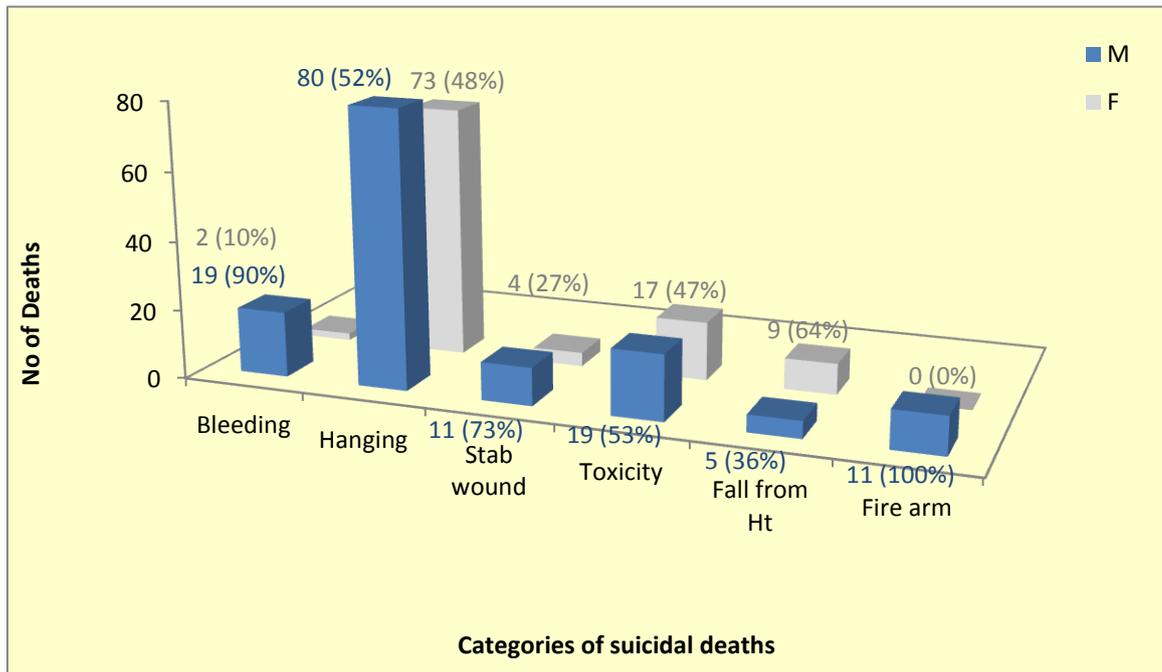


Figure 2.29: Bar charts showing the total number, percentage and causes of reported medico-legal cases of un-natural deaths due to suicide in Kuwait according to gender during 2003 and 2009 (Data was corrected based on male to female ratio of 1.7: 1).

Figure 2.29 shows the total number, percentage and causes of reported medico-legal cases of un-natural death cases due to suicide to the GDCE in Kuwait according to gender between 2003 and 2009. The results showed that significantly ($p < 0.05$) more males died from suicide compared to females in all types (causes) of suicides except in falls from heights, in which males ranked (5; 36%) cases compared to females who accounted for (9; 64%) cases. Males ranked first in suicidal death cases due to bleeding. Typically, (males accounted for (19; 90%) cases compared to females who accounted for (11; 10%) cases. The same was also true for suicidal death due to hanging where males accounted for (80; 52%) cases compared to females who accounted for (73; 48%) cases. The results also showed that in suicidal deaths due to stab wound injuries males accounted for (11; 73%) cases whereas, female accounted for (4; 27%) cases. In suicidal deaths by the use of firearm, male accounted for (11; 100%) cases of all firearm cases whereas, no case was registered for female (P Value = 0.000, DF = 5).

2.4.33: Categories of suicidal Deaths according to governorates between 2003 to 2009.

Table 2.5: Frequency table showing the total number and percentage of different categories of suicide.

| Years | Categories of Suicidal Death | | | | | |
|----------------------|------------------------------|------------------|---------------------|-------------------|---------------------------------|-------------------|
| | Bleeding N (%) | Hanging N (%) | Stab wound N (%) | Toxicity N (%) | Fall from height N (%) | Fire arm N (%) |
| Jahra | 7 (20) | 34 (15) | 3 (13) | 7 (14) | 1 (8) | 2 (11) |
| Farwania | 9 (26) | 89 (43) | 9 (39) | 19 (40) | 3 (21) | 8 (44) |
| Hawally | 5 (14) | 19 (9) | 2 (9) | 5 (10) | 5 (39) | 0 (0) |
| Ahmady | 9 (26) | 39 (19) | 4 (17) | 9 (18) | 2 (15) | 5 (28) |
| Mobarak Al-Kabeer | 1 (3) | 10 (5) | 0 (0) | 3 (6) | 0 (0) | 0 (0) |
| Capital | 4 (11) | 18 (9) | 5 (22) | 6 (12) | 2 (15) | 3 (17) |
| Total | 54 | 137 | 36 | 68 | 14 | 38 |

The results in table 2,5 shows that Farwania Governorate ranks first in all causes of suicidal deaths except in death due to fall from height. The results also showed that Mobarak Al-Kabeer Governorate recorded the least cases of all categories of suicidal deaths.

2.4.34 Medico legal cases of suicidal deaths due to bleeding according to age groups

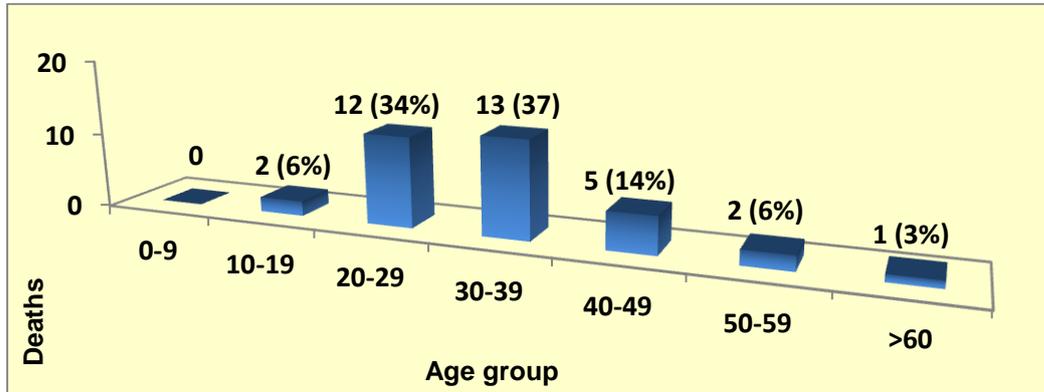


Figure 2.30: Bar charts showing the suicidal deaths due to bleeding in Kuwait between 2003 and 2009 as classified by different age groups.

The results in figure 2.30 show that there was a significant difference ($p < 0.001$) using ANOVA test, between age groups due to suicide by bleeding. The age group 30-39 years accounted for 13 (37%) cases, followed by the age group of 20-29 years who accounted for 12 (34%) cases.

2.4.35 Medico legal cases of suicidal deaths due to hanging according to age groups

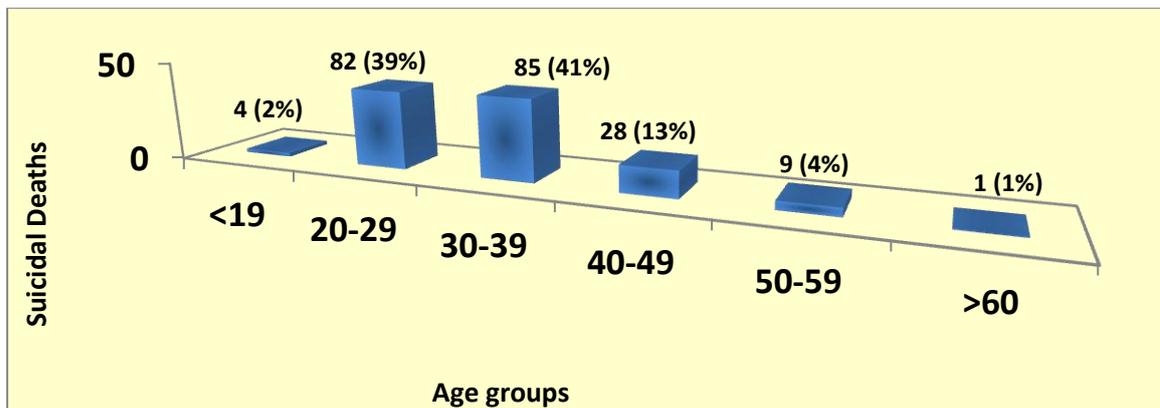


Figure 2.31: Bar charts showing the reported medico-legal cases of suicidal deaths due to hanging in Kuwait as classified by different age groups.

The results in figure 2.31 show that there was a significant difference ($p < 0.001$) using ANOVA test between the different age groups with the highest number reported among age groups 30-39 years and 20-29 years compared to the other groups accounting for 85 (86%) and 82 (39%), respectively.

2.4.36 Medico legal cases of suicidal deaths due to toxicity (poisoning) in Kuwait according to age groups

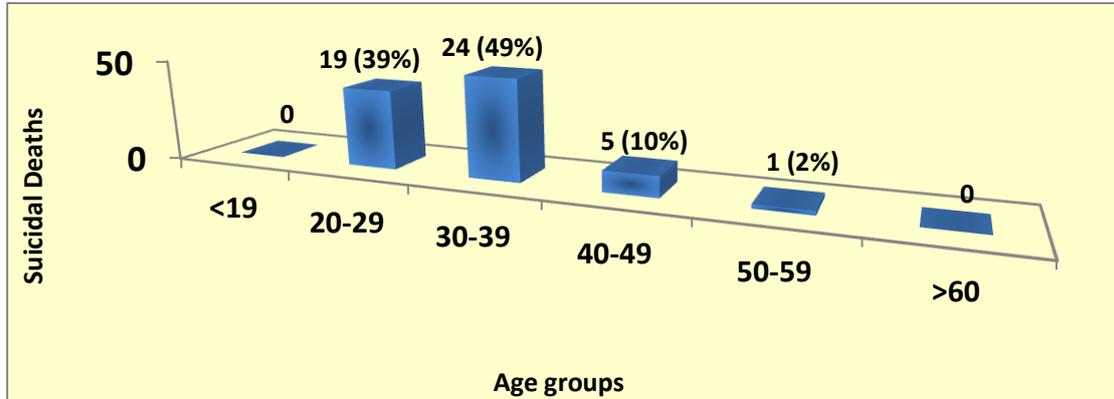


Figure 2.32: Bar charts showing the reported medico-legal cases of suicidal deaths due to toxicity (poisons) in Kuwait as classified by different age groups.

The results presented in figure 2.32 reveal that there was a significant difference ($p < 0.001$) using ANOVA test, in the number of reported suicidal deaths among the different age groups. The highest number of suicidal deaths due to toxicity was reported in age groups 30-39 years and 20-29 years compared with the other age groups accounting for 24 (49%) cases and 19 (39%) cases, respectively.

2.4.37 Medico-legal cases of suicidal deaths due to stab wound injuries according to age groups

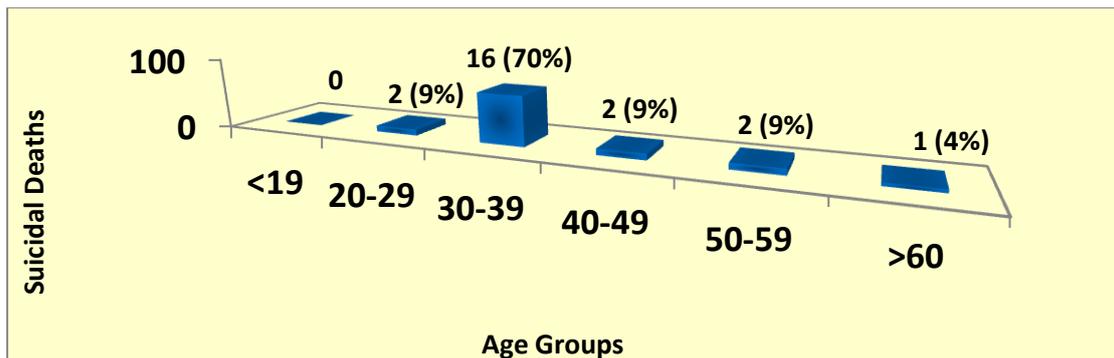


Figure 2.33: Bar charts showing the total number and percentage of reported medico-legal cases of suicidal deaths due to stab wound injuries as classified by different age groups.

2.4.38 Reported medico-legal cases of suicidal deaths due to falls from height injuries in Kuwait according to age groups

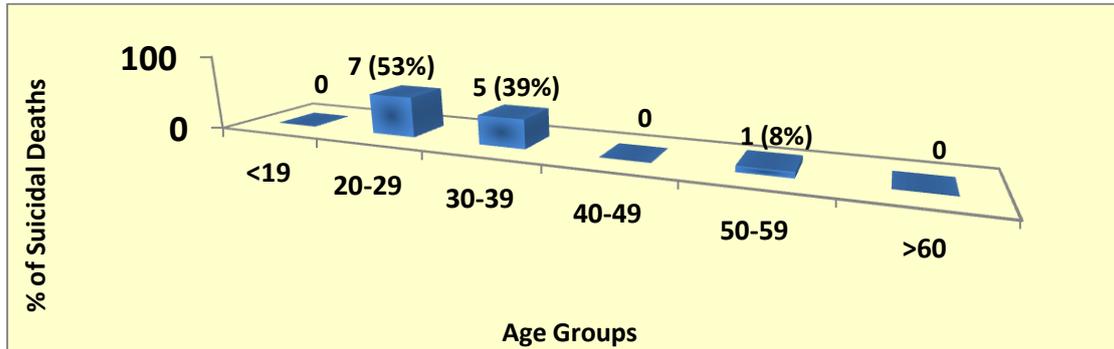


Figure 2.34: Bar charts showing the reported medico-legal cases suicidal deaths due to falls from height injuries in Kuwait as classified by different age groups.

The results in figure 2.34.reveal that there was a significant difference ($p < 0.001$) in the number of reported medico-legal suicidal cases due to falls from height injuries in age groups 20-29 and 30-39 years compared with the other age groups. No reported case was accounted for in age groups 0-9 years, 40-49 years and 60 years over.

2.4.39 Reported medico legal cases of suicidal deaths due to firearm injuries in Kuwait according to age groups between 2003 and 2009

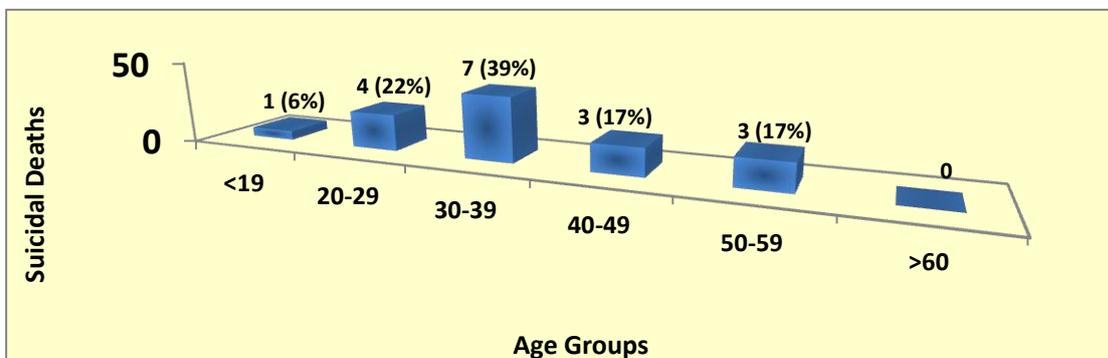


Figure 2.35: Bar charts showing the reported suicidal deaths due to firearm injuries as classified by different age groups.

The results in figure 2.35 show that the highest number of reported death cases due to firearm injuries occurred in age group 30-39 years, accounting for 7 (39%) cases, followed by the age group 20-29 years which accounted for 4 (22%) cases.

2.4.40 Comparison between suicide rates in Middle Eastern Countries (per 100,000) by country, year and gender for 2002.

Table 2.6: Table showing a comparison between suicide rates in Middle Eastern Countries (per 100,000), by year (2002) and gender. Data are derived from the World Health Organization (WHO).

| Serial no | Country | Year | Males | Female | Total |
|-----------|--------------|------|-------|--------|-------|
| 1 | ISRAEL | 2002 | 10.5 | 2.6 | 13.1 |
| 2 | BAHRAIN | 2002 | 4.9 | 0.5 | 5.4 |
| 3 | EGYPT | 2002 | 0.1 | 0.0 | 0.1 |
| 4 | IRAN | 2002 | 0.3 | 0.1 | 0.4 |
| 5 | JORDAN | 2002 | 0.0 | 0.0 | 0.0 |
| 6 | KUWAIT | 2002 | 1.6 | 1.6 | 3.2 |
| 7 | SAUDI ARABIA | 2002 | 2.6 | 1.4 | 1.2 |
| 8 | QATAR | 2002 | 0.9 | 0.4 | 0.5 |
| 9 | JORDAN | 2002 | 0.3 | 0.8 | 1.1 |
| 10 | SYRIA | 2002 | 0.2 | 0 | 0.2 |
| 11 | YAMEN | 2002 | 1.2 | 0.4 | 1.6 |

The data in table 2.6 shows that suicide rates are given for both male and female population per 100000 and the total number. The total rate of suicides is based on the total number of suicides divided by the total population in a country. The data show that in year 2002, the least suicide rate was recorded in Syria accounting for 0.2, followed by Qatar and Jordan which accounted for 0.5 and 1.1, respectively. The highest rate within the Gulf Countries was recorded for Bahrain accounting for 5.4. Whereas, the highest rate in the Middle Eastern Countries was recorded in Israel. Kuwait had a suicidal rate of 3.2 which is considered to be high compared to a neighboring Saudi Arabia with a suicidal rate of 1.2.

2.4.40 Comparison between Homicide Rates in 2010 for Middle Eastern Countries with other areas in the World (per 100,000) by country, year and gender.

Table 2.7: Table showing a comparison between homicide rates in Middle Eastern Countries with other countries around the world in 2010 (per 100,000), by year and gender (WHO, 2010).

| No | Country | Year | Rate |
|----|-------------------------|------|------|
| 1 | Bahrain | 2010 | 1.27 |
| 2 | KUWAIT | 2010 | 1.38 |
| 3 | United Arab Emirates | 2010 | 0.92 |
| 4 | Qatar | 2010 | 0.93 |
| 5 | Saudi Arabia Kingdom | 2010 | 1.04 |
| 6 | Egypt | 2010 | 1.24 |
| 7 | Morocco | 2010 | 1.37 |
| 8 | Jordan | 2010 | 1.74 |
| 9 | Israel | 2010 | 2.1 |
| 10 | Yemen | 2010 | 4 |
| 11 | United Kingdom | 2010 | 1.17 |
| 12 | United State of America | 2010 | 5 |
| 13 | Canada | 2010 | 1.8 |
| 14 | Iran | 2010 | 3.0 |
| 15 | India | 2010 | 3.4 |
| 16 | Turkey | 2010 | 3.8 |
| 17 | Libya | 2010 | 2.9 |
| 18 | Brazil | 2010 | 23 |
| 19 | Mexico | 2010 | 15 |
| 20 | Pakistan | 2010 | 7.3 |

Table 2.7 shows a list of homicide rates for Middle Eastern Countries with other countries in the world according to data most recent available year 2010 from the World Health Organization (WHO). The reliability of "homicide" differs among countries. Homicide may or may not include infanticide, assisted suicide or euthanasia. Moreover, they may also be underreported for political reasons. Nevertheless, the results show that high rate of homicide was recorded in Brazil accounting for 23. Whereas, the lowest homicide rate was recorded in United Arab Emirates accounting for 0.92. Kuwait has a homicidal rate of 1.38, considered as the highest rate within the Middle Eastern Countries.

2.5 Discussion

This study was designed to investigate the pattern, including total number, percentage and causes of the reported medico-legal cases of unnatural deaths examined at the Forensic Department of the Ministry of Interior in Kuwait between the periods from January 2003 to December 2009. This study is first of its kind in Kuwait and possibly in the whole of the Middle East Region comparing the different parameters (age, gender, religion, drugs, nationalities, marital status and Governorates) with the number, percentage and causes of reported medico-legal cases of unnatural deaths. The results will help the Kuwaiti Government to tackle the problems and to reduce the number of unnatural deaths in the country.

The results presented in this study highlight a number of interesting details regarding unnatural deaths including the trends or causes of homicide, suicide and accidents in Kuwait paying particular attention to gender, nationalities, age and Governorates. Moreover, the discussion will briefly address the relationship between unnatural deaths and religion in Kuwait. The study will also investigate the influence of alcohol abuse on reported unnatural death cases. Finally, the discussion will also focus briefly on the epidemiology of unnatural deaths in some Middle East Countries mainly for comparison.

2.5.1 Prevalence of un-natural deaths in Kuwait

The reported medico-legal cases of un-natural deaths in Kuwait were divided into three groups namely, suicide, homicide and accidental deaths for further analysis. The results showed that accidental deaths were the major cause of un-natural deaths in Kuwait over the seven years during the period

of (2003-2009) of this study and they accounted for 4,886 (86%) reported cases. Out of 5,703 un-natural death cases reported during the study, there were 470 (8%) cases of homicidal deaths and 347 (6%) cases of suicidal deaths (Figure 2.3). The modes of deaths established in this study indicated that accidental deaths reached a peak value of 841 cases (17%) in 2009, followed by 801 cases (16%) in 2007, 716 cases (14%) in 2005, 692 cases (14%) in 2006, 683 cases (14%) in 2008, 577 cases (12%) in 2003 and 576 cases (11%) in 2004, respectively (Figure 2.2). The lowest incidence of accidental deaths was reported in the years 2003 and 2004. During the period of this study, the year 2008 showed a slight decrease in the incidence of accidental fatalities. The highest rate was reported in the year 2009 with an increase of 5% compared to the year 2003.

2.5.2 Geographical distribution of reported medico-legal cases of un-natural death in Kuwait

The present study has shown that Farwania Governorate had the highest incidence of accidental deaths (1418 cases or 29%), followed by Jahra which accounts for 934 cases or 19.1%. Similarly, Ahmady accounted for 930 cases or 19%. Hawally Governorate accounted for 739 cases or 15%. The results also show a lower incidence of accidental deaths in Mubarak Al Kabeer and Kuwait City (the Capital) Governorate. The figures of the current study have shown a highly significant difference ($p=0.000$ $df= 10$) in the three categories of un-natural deaths among various Governorates (Table 2.1).

According to Kuwait Central Statistical Office in mid of February-2011, Kuwait population was 2,836,644 million. The population is estimated to rise from 2.9 million in 2011 to 3.2 million by the end of 2015 per capita per Governorate (Kuwait Central Statistics, 2011). According to the data presented by the Kuwait Central Statistics Office, the largest Governorate area is Al-Jahra (11,230 km²), with a population of 422,915 (see table 1.1). Taking these data into consideration with the results obtained from this present study, only 19% of the total cases accounted for accidental deaths in Al- Jahara Governorate. In addition, Al-Jahra Governorate also accounted for 16% of suicidal deaths, and 17% of homicidal deaths. Surprisingly, there was a decreasing trend in the total cases of un-natural deaths in the Mubarak Al-Kabeer (MK) Governorate. A possible explanation is that MK is considered as the second smallest Governorate in terms of both population and in size compared to other Governorates.

The analysis of this study regarding reported medico-legal cases of suicidal deaths in Kuwait Governorates revealed that Farwania Governorate showed the highest incidence (40%), followed by Ahmady Governorate (20%), Jahra Governorate (16%) and Hawally (10%). The lowest number of suicidal deaths was recorded in Mubarak Governorate (4%). Among the homicidal death category, Farwania showed the highest incidence (38%), followed by Jahra Governorate (17%), Hawally (16%) and Capital (14%). However, Mubarak Al-Kabeer Governorate showed the lowest incidence of homicidal deaths (5%). There were significant ($p < 0.000$ $df = 10$) differences in the incidence of reported medico-legal cases of homicidal deaths among the various Governorates.

2.5.3 Distribution of reported medico-legal cases of un-natural deaths according to the gender in Kuwait

The data of un-natural deaths was corrected according to male to female ratio of 1.7:1. The findings in this study have shown that males outnumbered females in reported medico-legal cases of accidental deaths accounting for 2378 (74%) cases compared with 842 (26%) cases for females. Similarly, males outnumbered females in deaths due to suicide accounted for 142 (57%) cases. Moreover, in homicidal deaths, the number of males was higher (221 cases; 70%) than these of females (94 cases; 30%). There were significant differences ($P=000$, $df= 2$) in all three death categories of un-natural deaths comparing males with females. Similar results were obtained from a study carried out in India to detect the incidence of un-natural deaths in a three years study (2000-2003), which revealed that a male predominance was observed among the overall percentage of un-natural deaths, with the male: female ratio being 2.2:1. This study reported a male: female ratio of various modes of un-natural deaths of 2:1 for suicides, 3:1 for accidents and 4:1 for homicides (Singh et al., 2005).

2.5.4 Relationship between age distribution and the different categories of un-natural deaths in Kuwait

The present study showed that young adults belonging to the age group of 20-29 years constitute the majority of reported medico-legal cases of un-natural deaths accounting for 26%. This was followed by the age group 30-39 years who accounted 24% (Table 2.2). These two age groups are in their prime and productive years of life and this in turn makes them more prone and vulnerable to un-natural deaths. A study in Eastern Nepal showed

that the maximum percentage (47%) of un-natural deaths belonged to the age group of 21-30 years (Jha et al., 2011). Similarly, another study in Pakistan also shows that the highest percentage (28-40%) of un-natural deaths was reported for the same age group and with of all reported homicidal cases occurred in the age bracket of 20-29 years (Mohanty et al., 2005). Other studies in Egypt and India have also reported that this age group is most vulnerable to accidents and suicides (Gupta et al., 2004; Shalaby et al., 2010). In contrast, a study in USA show that the highest percentage of reported medico-legal cases for accidents and suicides occurred in the age group 10-25 years. This difference could be due to the fact that in the USA individuals start a more independent life at an earlier age, thus exposing themselves to all sorts of gang violence at schools, colleges, surrounding neighboring environment and even accidents which occurred at work place (Zafar et al., 2006).

2.5.5 Relationship between un-natural deaths and nationalities

The data of un-natural deaths was corrected according to Kuwaiti and Non-Kuwaiti nationals ratio of 1:1.94. The present results show that Non-Kuwaiti residents outnumbered Kuwaiti residents only in deaths due to suicide. Accidents reported for Kuwaiti residents accounted for 54% compared to 46% for Non-Kuwaiti residents. Similarly, reported medico-legal cases of homicidal deaths for Non-Kuwaiti residents accounted for 48% compared to 52% for Kuwaiti residents. In relation to suicides, Non Kuwaiti residents accounted for 77% compared to 23% for Kuwaiti residents. These results clearly show that more Non Kuwaiti residents died during the study period compared Kuwaiti residents for deaths due to suicide. Whereas,

Kuwaitis outnumbered Non-Kuwaiti. There was a significant difference in all three categories of un-natural death ($P=0.000$ $df= 2$ using ANOVA test). Similar results have been found in Saudi Arabia and Jordan (Daradkeh, 1989; Elfawal, 1999).

2.5.6 Pattern of homicides in Kuwait

A study of the patterns of homicides in a society is one of the first steps in developing strategies to prevent it. During the 7 years period of the current study, FMD at GDCE, Kuwait, conducted a total of 8,385 forensic autopsies. Among these, 17% cases were due to blunt force, 19% of the cases owing to the use of firearm injuries; 38 % because of stab wounds and 8 % were due to strangulation. This study also showed that 15% of homicidal cases were due to smothering while infanticide accounted for 3%. These figures showed a significant ($p<0.05$) difference in various categories of homicidal fatalities. The number of expatriates in Kuwait is almost double compared to the indigenous Kuwaiti population in which non-Kuwaiti residents outnumber Kuwaiti residents by a ratio of 1.94:1. In addition, following the Iraqi invasion of Kuwait in 1990, reports have shown that psychological distress was in the increase (Fido, 2009). Not surprisingly, the availability of firearms has also increased significantly.

2.5.6.1 Trends of homicidal deaths in Kuwait according to gender

Homicide is one of the oldest forms of crime in human civilization (Nordentoft and Bull, 2007). During the period of this present study, the trends of reported medico-legal cases of homicidal deaths revealed that males outnumbered females in all types of homicidal deaths (Figure 2.9). The major

types of homicides reported in this study included stab wounds, fire arms, blunt force and smothering. Together, strangulation and infanticide were less than 10%. Comparative study of homicide and suicide done in India found that males outnumber females in homicides and the major methods of deaths due to homicides were blunt trauma, sharp trauma, burning and strangulation (Ambade et al., 2007; Calce and Rogers, 2007). The involvement of gender with regard to crime has largely been ignored and pushed aside in criminological and sociological studies (Ingoldsby and Callagy, 2010). Deaths from violence were always more common among males than females, since males tend to deal with things in a violent manners. The majority of the studies relating to reported medico-legal cases of deaths due to homicides have shown a male dominance (Sjögren et al., 2000; Strand et al., 2010; Sauvageau and Boghossian, 2011). However, some studies from the West show contrasting trend of female dominance among reported medico-legal cases of homicides, especially homicidal asphyxia, thereby demonstrating both geographical and societal variations in the involvement of victims of crimes (Baeza et al., 2000; Cowell, 2009).

2.5.6.2 Geographical distribution of homicidal deaths in Kuwait

The trends of homicidal deaths in Kuwait were analyzed in detailed according to the geographical regions. The results show that Farwania Governorate ranked first in blunt force injury deaths (33%), followed by Hawally Governorate (24%), Jahra Governorate (15%), Capital Governorate (14%), and Mubarak Al Kabeer Governorate (10%) (Table 2.1). The least reported medico-legal cases were recorded in Al Ahmady Governorate accounting for only 5%. Homicidal deaths due to the use of firearm were most

common in Jahra Governorate accounting for 26%, followed by similar number of reported cases of deaths (25%) for both Farwania and Capital Governorates. The Mubarak Governorate reported the least homicidal deaths due to firearm injuries (1%) and this was probably attributed to the fact that Mubarak Governorate has 83 % non-Kuwaiti residents compared to 17% of Kuwaiti residents. Normally, it is a crime for a non- Kuwaiti resident to own firearms. With regards to the high percentage of homicidal deaths in Farwaniya Governorate, this could be attributed to the density of the population. Most people live in this Governorate compared to the six other Governorates of Kuwait.

The results of this study show that expatriates form 74% of the Kuwaiti population (see table 1.1) and most of them live under extreme pressure, stress and tension due to hard work, low salary, ill health, uninhabitable social conditions and homesickness, all of which may play a crucial role in homicidal activities.

The most reported medico-legal cases of homicidal deaths due to stab wounds were in Farwania Governorate (49%), followed by Hawally (18%) and Jahra (12%). Al Ahmady and Kuwaiti Capital Governorates reported similar homicidal deaths accounting for 10% each. Mubarak Al Kabeer reported the least medico-legal cases of deaths (1%) (Table 2.1).

Reported medico-legal cases of homicidal deaths in Kuwait by strangulation were found to be the highest in Jahra (28%), followed by Mubarak Al Kabeer (25%) (Table 2.1). A similar number of medico legal death cases were reported for Hawally and Al Ahmady Governorate (11%). However, Kuwaiti

Capital Governorate reported the minimum number of homicidal deaths due to strangulation (3%). Strangulation is considered to be a form of mechanical asphyxia. For example, in Africa, rape was the motive in 66% of the female victims of ligature strangulation and 52% of those were due to manual strangulation (Egge et al., 2010). The highest percentage of strangulation was reported among the age group of 20 to 39 years (Egge et al., 2010). With relation to infanticide, the present results showed that Farwania Governorate ranked first (39%), followed by Capital (23%) and Jahra (15.4%). A similar number of cases for infanticide were reported for Hawally, Al Ahmady and Mubarak.

The present results show a significant difference in the number of medico-legal cases of deaths in the different categories that were reported according to Governorates (Table 2.1). These are due to several socio-economic and health-related factors in the different Governorates. For example, Farwaniya Governorate is over populated and it is mostly inhabited with expatriate residents with low-income. Most of the residents work in shops, factories, as taxi-drivers, labourers, house maids and servants and with cleaning companies. Such workers are normally lured into the Kuwait with promises of good work and high salaries. Upon their arrival, they find themselves on the streets and out of jobs. These are the same individuals, who sometimes out of frustration and for survival, are forced to commit crimes or become part of a gang that provides them with extra income. The present study has illustrated that Farwaniya Governorate demonstrated the highest incidence (percentage) of reported medico-legal deaths due to blunt force, stab wounds, smothering and Infanticide. According to the annual report of

crimes in Kuwait (Ministry of Interior Report, 2011), it has been noticed that the crime rate has increased markedly, particularly in some areas of Farwaniya Governorate. This region is known for several incidences of raids for the illegal production of home-made liquor, kidnapping, sex slavery and prostitution (Ministry of Interior Report, 2011). There are some areas in Farwaniya Governorate in which criminals were arrested for manufacturing an average of sixty bottles of home-made liquor per day (illicit). These places are very popular among Asians and to some extent Arabs who have been seen purchasing illicit alcohol during weekends. Both purchasing and the drinking of alcohol are illegal by Kuwait law. It is also noteworthy, that most of Farwaniya buildings and rooms are sub-let by tenants in order to earn extra income. The overcrowding of these places may play a crucial part in causing crimes.

The results in this study showed that Jahra Governorate reported the highest incidence in medico-legal cases of homicidal deaths due to firearms, and strangulation. To further explain the reason for the wide spread of firearms in Jahra, it is important to describe the location of the area. Al-Jahra Governorate is located in the North West of Kuwait, bordering Iraq. The Iraqi invasion of Kuwait played an important role in providing illegal firearm weapons. These weapons are hidden inside their houses. The Ministry of Interior in Kuwait established a Committee for illegal weapon collections from people. Officials of the Ministry are allowed by law to search for these weapons in some houses where they have definite information regarding the ownership of unlicensed firearms.

Homicide is a common endpoint of many different behavioral pathways which can lead to deaths (Krienert and Walsh, 2010). It may be as a result of arguments between acquaintances, domestic violence, robberies, drug addiction and terrorism and several others. In this study, the results show that the highest number of homicides occurred in 2008, accounting for 24%, followed by 18% in 2007, 14% in 2004, 13% in 2006, 12% in 2009, 9% in 2005 and 9% in 2003 (Figure 2.6). However, the year 2008 demonstrated the highest incidence of reported medico-legal cases of homicidal deaths compared to 2003 when the lowest homicidal death cases were reported during the study period between 2003 and 2009. The number of reported medico-legal cases of homicidal deaths was elevated to more than twice in the year 2008 compared with 2003. However, it is interesting to note that in 2009, it had declined again to 9%. Further in-depth study is required in order to ascertain the root causes of decline of homicidal deaths. The results also show a significant difference in the number of reported medico-legal cases of deaths within the three groups of death categories comparing one with another ($P=0.000$, $df = 12$ using ANOVA test).

2.5.6.3 Pattern of homicidal deaths according to nationalities

The relationship between race and crime has been a topic of public controversy and scholarly debated for more than a century (Linehan et al., 2006; Crowell et al., 2008). Statistics from the Department of Justice in United States in 2004 delineated that African Americans were seven times more likely than white Americans to commit homicides and six times more likely than white Americans to be murdered (Sauvageau and Yesovitch, 2010). A close examination at the trends of reported medico-legal cases of un-natural

deaths due to homicides reveals that the percentage of deaths among expatriates in Kuwait was higher than indigenous Kuwaiti residents in all categories of homicidal deaths except for strangulation and firearm fatalities. The data of homicidal deaths was corrected according to Kuwaiti and Non-Kuwaiti ratio of 1:1.94. Moreover, Non-Kuwaiti residents dominate Kuwaiti Nationals such fatalities as death due to blunt force, stab wound injuries, smothering and infanticide. In contrast, significantly more medico-legal cases of deaths due firearm fatalities and deaths due to strangulation were reported for indigenous Kuwaiti residents (78%) compared to expatriates (22%) in fire arm fatalities and (56%) for Kuwaitis and (44%) in Non-Kuwaiti nationals in deaths due to strangulation. Expatriates make up around 74% of the Kuwaiti's population, ranking it fourth worldwide in a ratio between nationals and expatriates (CIA, 2010). The expatriates are the low income group in the country with a very low salary scale. Moreover, they are not allowed to purchase houses in Kuwait so they pay high rent until they leave the country. Because of their low salary and lack of money, they tend to be involved in many disputes petty crimes and problems in the areas where they live. These low income groups are not allowed to bring their families and as a result, they indulge in illegitimate relationships which may end in disputes and quarrels. This is the main reason for a higher percentage of homicides among expatriates in Kuwait.

2.5.6.4 Age group distribution of homicidal deaths due to firearms

Based upon the analysis of the present study, the results show that young adults belonging to the age group 20-29 years constituted the majority of homicidal deaths due to firearm injuries, accounting for 42%, followed by

the age group 30-39 years accounting for 32% of all reported medico-legal cases (Figure 2.11). These results are in accordance with a study published in Pakistan where a higher incidence was found among the age group 20-29 years for both males and females (Zafar and Zahid, 2010). Reports from India and Turkey have also shown similar findings that people from the age group 20-29 years are most vulnerable in homicidal activities (Gupta et al., 2004; Mohanty et al., 2005). Studies in USA have indicated that the highest number of homicidal deaths occurred at an earlier age of between 10 to 25 years. This difference in age group could be explained in view of American adolescents being more independent at an earlier age than adolescents from in Middle Eastern societies (Mohanty et al., 2005). Adolescents from the USA are more often exposed to all sorts of violence even at Schools, moreover, they have more access to guns compared to Middle Eastern Countries (Gupta et al., 2004).

Although according to Kuwait law, individuals are required to possess a special license to own a firearm. The current study has shown an increase in the incidence of firearm injuries. This might be attributed to the Gulf War, since after the liberation of Kuwait, Iraqi troops left many weapons behind. The easy availability of these firearms may have had a great impact on the firearm violence in Kuwait. Another reason that may have led to the increase incidence of firearm injuries among Kuwaitis is that some tribes, especially Bedouin, use firearms to shoot in the air as part of their wedding ceremonies. Unfortunately, this traditional Bedouin culture causes some fatalities among innocent victims in the area. The Government recently established a legislation requiring permits to fire guns at weddings especially those held in

tents and on public land. The rationale is to decrease the firing of illegal guns as part of the celebration. Levels of gun violence vary greatly across the world, with very high rates in South Africa, Mexico and Colombia, as well as high incidence in Thailand, Guatemala, and some other developing countries (Zafar and Zahid, 2010). Rates of gun violence are low in Singapore, Chile, New Zealand compared to the United States which has the highest rate among developed countries (The Seventh United Nations Survey on Crime Trends, 2011).

2.5.6.5 Blunt force trauma

Traumatic episode of blunt force trauma caused to the body by a blunt object would result in severe damage (Zafar and Zahid, 2010). During this period of study, blunt force injuries in Kuwait were the third cause of the total reported medico-legal cases of homicidal deaths accounting for 17 %. The results of the present study have revealed that adults belonging to the age group 30-39 years constituted the majority for reported medico-legal cases due to deaths as a result of blunt force injuries accounting for 31% compared to extreme age groups of 0-9 years, 60 and over years and 10-19 years who showed less than 10% of reported deaths. Similar studies in India have also reported that this age group (30-39 years) was the most vulnerable regarding blunt force injuries (Sharma et al., 2006). The analysis of current data also concluded that non-Kuwaiti Nationals outnumbered Kuwaiti Nationals in deaths due to blunt force injuries (Figure 2.8).

3.5.6.6 Incidence of stab wound injuries

In Forensic Medicine, stabbing has always been a very common means of homicide. In the present study, the incidence of un-natural deaths due to stabbing injuries in Kuwait was studied according to different age groups between 2003 and 2009. The results showed that the leading cause of homicidal deaths was due to stab wound injuries constituting 38% of the total homicidal deaths. Similar results were found in the age groups 20-29 years and 30-39 years accounting for 30% each, followed by age group 40-49 years accounting for 20%. Homicide due to stab wound injuries as a method of choice among the age group 10-19 years accounted for 7%. The least number of cases were reported for the age group 0-9 years accounting for 2%. In India, one of the most frequent ways of committing homicide is by inflicting injuries with a sharp cutting or stabbing weapon. Precise examination of such injuries may reveal a sizeable number of clues which may be of paramount importance in the reconstruction and interpretation of the whole events (Brunel et al., 2010).

2.5.6.7 Incidence of strangulation

Strangulation is a type of mechanical asphyxia produced by constriction of the neck by the force applied other than by the weight of the body (Tsung-Hsueh et al., 2008). Literature on homicide prominently describes strangulation as one of the leading causes of homicides, especially in the developing world (Zafar et al., 2006). During the 7 years of the present study, among the 450 medico-legal cases of homicidal deaths that were reported in Kuwait, 8% of cases were due to strangulation. Based upon the present analysis, the incidence of strangulation was low and came only as the second lowest of homicidal deaths. A large percentage (64%) of victims were

young subjects in their prime and productive years of life between the ages of 20 and 39 years. Incidence of deaths due to strangulation in Kuwait was above 60% in both the age group 20-29 years and 30-39 years. Deaths by strangulation in the age group 30-39 years were almost double accounting for 33.3% (Figure 2.13) compared to the percentage reported in India for similar age group accounting for 20% (Gupta et al., 2004). The lowest number of fatalities due to strangulation was reported among age group 10-19 years accounting for 3% of total homicidal cases. In contrast, no medico-legal case was reported in homicidal deaths due to strangulation for the age group of over 60 years. The male to female ratio of all the strangulation cases in the present study were almost similar, where males accounting for 54% compared to 46% for females. The present results also showed that the highest percentage of medico-legal deaths due to strangulation occurred among Kuwaiti residents and they accounted for 56% compared to 44% for Kuwaiti residents (Figure 2.8). This increased prevalence of male involvement truly reflects the overall male dominance in violent asphyxias and especially among homicides worldwide (Gupta et al., 2004; Hejna and Šafr, 2010). However, some studies from the West show contrasting trend of female dominance among cases of homicidal asphyxia (Serafettin et al., 2009).

2.5.6.8 Area of the body involved in fatal injuries

During the time course of the present study, an analysis was made on the different parts of the body involved in various types of un-natural deaths including firearm injuries, stab wounds and blunt force injuries. The results revealed that the majority of cases had several body injuries involving multiple sites of the body. The maximum number of injuries was inflicted on the chest

for firearm injuries (42/89), whereas the most common area for stabbing was the neck (cutting the throat) accounting for 64/180. In deaths due to blunt force injuries, the greatest number of injuries was in the head accounting for 51/81. The upper limbs and lower limbs sustained minimum number of fatal injuries. Several studies have indicated that the skull is the target in the majority of cases of homicidal actions specifically in blunt force injuries (Sauvageau and Boghossian, 2011). Several other studies have reported similar findings, pointing the skull as the main target in majority of cases especially in blunt force injuries. In contrast, fatal injuries were most common on the neck and chest (Mohanty et al., 2005).

2.5.7 Pattern of reported medico-legal cases of suicide in Kuwait

Suicide is one of the leading causes of death in the world and suicidal attempts constitute a major public health concern globally (Brunel et al., 2010). An analysis of the pattern of reported medico-legal cases of deaths by suicides in this study showed that out a total of 5,703 cases there were 347 (6%) suicidal cases (Figure 2.3). The results show that suicidal deaths due to hanging were the leading cause accounting for 60%. Meanwhile, poisoning was the second cause accounting for 14%, followed by cut wounds accounting for 10%. Suicides by firearm injuries and stab wounds accounted for 5% and 7%, respectively. The results also show that falls from heights were the least common suicidal method and accounted for only 4%. This could be explained that falls from heights constitute a horrible experience for the person committing suicide. Generally, suicide itself is a stressful case and can be viewed and noticed by others outside. However, suicide by hanging

has some private features although the mechanism of death is complex (Elfawal and Awad, 1997).

The present study has illustrated that Kuwait has a low rate of suicide and is also considered of having a lower rate of homicides and accidental deaths. The literature has limited reports on suicide in the Middle East. Reports of suicide rate in Bahrain in 1988 show similar values to that in Kuwait (5.4%) (WHO, 2006). Most Arabic families of suicidal victims would never admit the actual cause of death to anyone, and always state that their son or daughter had died suddenly. Saudi Arabia views suicide in as a disgrace and shameful more than in other Arab World (Al-Barraq and Farahat, 2011). Similar results have been found in United Arab Emirates in which the annual rate of suicide in Dubai between 1992 and 2000 was 6.2/100,000 (Benomran and Hassan, 2007). The low rate of reported medico-legal cases of suicides in Kuwait compared to other Arab Countries may be attributed to people's commitment to Islamic teachings which prohibit suicide, as well as to social and economic stability in the country.

2.5.7.1 Gender and Suicide

The pattern of medico legal suicide cases in Kuwait according to gender showed that males outnumbered females in all patterns of suicidal deaths except in falls from heights in which the percentage of females was greater 64% (Figure 2.29). Males constituted the majority of reported medico-legal cases of suicidal deaths due to cut wounds accounting for 73%. The same is also true for suicidal deaths due to hanging where males accounted for 52% compared to 48% for females. The results also show that in stab

wound injuries, 73% were males and 27% were females. In firearm injuries, all victims were males. The main explanation for this observation is that Kuwait is a Muslim country where women have limited freedom and due to strict regulation of owning a gun, therefore possession of guns may be limited to men only.

The present results are comparable to those reported cases in Saudi Arabia where 82 % of people who committed suicide were males and females accounted for only 18% (Koronfel, 2002). In Bahrain, the report of deaths by suicide is extremely difficult due to a combination of social, legal and religious factors. Moreover, there are limited data in Bahrain to analyze especially since, some Middle Eastern countries do not offer protection to domestic workers under its labour laws (Al-Ansari et al., 2007).

The most common reported medico-legal cases of deaths due to suicide in this and similar studies in the Middle East is hanging. In contrast, in a Scottish study, poisoning was clearly the method most commonly used to commit suicide (Arun et al., 2010). Rates of suicide in most countries in the world, including Denmark, are higher among males than in females (Spicer and Miller, 2000; Yong, 2009). China is an important exception, because it has a very high suicide rate among females, especially young women in rural areas (Chui et al., 2002). This is further supported by a previous study carried out in 2006 by Dirnhofer who reported that illiterate, low income and unmarried females living in rural areas are more prone to suicidal thoughts and planning (Dirnhofer et al., 2006).

2.5.7.2 Suicidal deaths according to nationalities

The results in this study have clearly shown that all categories of suicide were higher among the Non-Kuwaiti population, except for hanging and firearm which accounted for 51% among the Kuwaiti population compared to 49% among the Non-Kuwaiti Nationals in deaths due to hanging and 18% among Non-Kuwaiti and 82% in deaths due to firearm among Kuwaiti nationals (Figure 2.28). Meanwhile, among Kuwaiti residents, hanging as a method of choice was 51% of total hanging fatalities. Suicide by cut wounds was equally higher among non-Kuwaiti residents accounting for 89% compared to 11% Kuwaiti residents. Whereas, in suicide by falling from no cases were reported among Kuwaiti nationals and non-Kuwaiti accounting for 100%. An in depth study is urgently required to investigate the underlying causes of suicidal deaths in Kuwait. Similar results were reported in Bahrain, in which the mean suicide rate was 0.6 per 100,000 for the Bahraini nationals and 12.6 per 100,000 for expatriates. Indian immigrants in Bahrain constituted the highest suicidal rate (Al-Ansary et al., 2007). The same is true in Saudi Arabia, where more people from the expatriate population (65%) committed suicide compared to Saudi citizens who accounted for 35% compared to non-Saudi expatriates (Elfawal, 1999).

2.5.7.3 Suicide according to Governorates

The present results show that the reported medico-legal cases of suicidal deaths in various Governorates of Kuwait were similar to those reported for other modes of deaths, i.e. accidents and homicides. The results show that Farwania Governorate ranked first with 44% in suicidal deaths by

the use of firearm, followed by Ahmady Governorate which accounted for 28% and Capital (Kuwait city) which accounted for 17%. No case of suicide by firearm was reported in Mubarak Governorate. The results also revealed that Hawally Governorate ranked first in suicide due to falls from heights accounting for 39% followed by Farwania Governorate which accounted for 23% (Table 2.5). In reported medico-legal cases due to poisoning, Farwania Governorate accounted for 39% and ranked first. This was followed by Mubarak Al Kabeer Governorate which accounted for 18%. The least percent of reported suicidal deaths due to stab wounds was reported in Hawally Governorate accounted for 9%, whereas, in Al-Ahmady and Farwania Governorates suicide cases due to cut wounds accounted for 26% each. Farwania and Hawally Governorates are heavily populated with expatriates and mostly inhabited by labourers who work in factories and cleaning companies. Most of these areas have rented buildings and rooms that have been sub-let by tenants in order to earn more or to cover their own expenses. The overcrowding of these places makes it difficult for authorities to monitor the area. As a result, suicidal deaths are higher in these Governorates. Data provided by Ministry of Interior in Kuwait in March, 2010, revealed that Farwania and Hawaly Governorates have high rates of crimes (Ministry of Interior Report, 2011). It is possible that most individuals who live in these Governorates retaliate by committing crimes or they become part of criminal gangs who earn extra income via crimes. It is now well known that these depressed areas in Kuwait are believed to be true fortress for many organized crimes and as a result many people are stressed out and they are under severe pressure due to the gang crimes. This in turn leads to suicide.

2.5.7.4 Age distribution of suicidal deaths due to cut wounds

The results show that significantly more people from the age group 30-39 years committed suicide by bleeding accounting for 37% (Figure 2.30). The age group 20-29 years came second with 34%. This was followed by the age groups 40-49 and 50-59 years who accounted for 14% and 6%, respectively. The least cases recorded for suicidal deaths due to bleeding were in the age group of over 60 years and they accounted for 3%. In general, the victims of suicide were mostly young people in their prime and productive years of life. Around 71% of these individuals were between the ages of 20 and 39 years. The extremes of ages were the least vulnerable to suicide with 6% up to 19 years of age and 3% above the age of 60. In general, this trend varies with different age groups and a clear suicide peak was found in younger people as indicated in the previous group. Similar results were found in India in 2001 by Gupta et al who highlighted that the commonest age group of the suicidal victims was 21-30 years (Gupta et al., 2004). Whereas in United States, a study in 2009 showed that suicide was the third leading cause of deaths for young people between the ages of 15 to 24 years for every 100,000 young people (Egge et al., 2010).

The present study was also designed to examine the relationship between age and suicide in Kuwait due to hanging. The results have revealed that victims from the age group 30-39 years were responsible for the highest mortality rate for suicide by hanging, accounted for 41%, followed by age group of 20-29 years who accounted for 39% and while, age group of 40-49 years accounted for 13% (Figure 2.31). In the age group of 50-59 years suicide by hanging accounted for 4%, followed by age group of 0-19

accounted for 2%. The least percent of medico-legal cases for suicidal deaths due to hanging was reported in the age group of over 60 years and they accounted for 1%. Hanging was the most common method of suicide by male victims. The result also revealed that almost one-third of all male victims who committed suicides came from the age group of 30 and 39 years, while in female the mean age of suicide due to hanging was 49 years.

2.5.7.5 Age distribution of suicidal deaths due to toxicity

Through time poison (toxicity) has been one of the most common forms of suicide or homicide. In this study, suicidal deaths due to toxicity in Kuwait according to different age groups revealed that the age group 30-39 years accounted for 49% of all reported medico-legal cases of deaths due to suicide (Figure 2.32). This was followed by age group of 20-29 years who accounted for 39%. Suicide by toxicity as a method of choice among the age groups 40-49 years and 50-59 years accounted for 10% and 2%, respectively. In contrast, no case of suicide due to poison was reported for age groups 0-19 and over 60 years. A study conducted in Egypt revealed that young females between the ages of 13-19 years are at higher risk of self-poisoning (Shreed et al., 2011). Deliberate self-harm is a major problem in the developing world and with the availability and easy access of poisons make it easier for the vulnerable to commit suicide (Linehan et al., 2006). This is quite often seen in cases of drug overdose which is another method of committing suicide (Hunt et al., 2007). A study was conducted in an addiction treatment center in a psychiatric hospital in Kuwait to review cases of poisoning in order to identify the important poisons used for self-harm in this region (Fido and Mughaiseeb,

1989). The results showed that pesticides were the most prevalent types used as a form of suicide, followed by paracetamol consumption (acetaminophen) and both of these are very common and available in any grocery stores.

A Kuwaiti Government report published in 2009 showed that most medico-legal cases of suicide were committed by ingesting of pesticides and paracetamol (acetaminophen) (Toxicological Division report, 2009). These drugs have become the most popular method of self-harm due to the free availability in the housing areas especially among people from Bangladesh community who live in Kuwait. In India, self-poisoning with medicines such as benzodiazepines and antidepressants were most commonly used by victims (Gupta et al., 2004). These data add to a body of evidence suggesting that restrictions are strongly required in order to combat the blatant abuse of some drugs which are easily available over the counter in shops, pharmacy and in supermarkets (Alexander et al., 2008).

2.5.7.6 Age distribution of suicidal deaths due to firearm injuries

The results of this study show that most victims died in the age groups 30-39 and 20-29 years due to suicide by firearm injuries, accounting for 39% and 22%, respectively (Figure 2.35). Similar results were recorded for age groups 40-49 and 50-59 years accounting for 17%. Suicide by firearm injuries as a method of choice among the age group of 0-19 years accounted for 6%. In contrast, no medico-legal case was reported among age group over 60 years.

Guns are one of the most lethal means in committing suicide and they are far more prevalent than overdoses, strangulation or cutting oneself (Wilson, 2010). Given the high rate of gun ownership in the USA, it is logical to expect a direct link between increased access to guns and a high incidence of suicide. However, international data show that higher levels of gun ownership do not always correlate with higher levels of suicide (Wilson, 2010). For example, personal gun ownership is more prevalent in Switzerland than in USA, but suicide rates are higher in the United States (Tewksbury et al., 2010). The actual cause of selecting fire arm as a means of death is unclear. In fact, there are some mediating factors that contribute for fire arm suicide. Kuwaitis have guns in their possessions after the gulf war in 1990. People consider this as an honorable and quick way to deal with personal troubles and disappointments. Easy access to dangerous weapons is also an important risk factor.

2.5.8 Patterns of accidental deaths in Kuwait

During the period of this present study, the results show that the leading cause of accidental deaths in Kuwait was road traffic accidents (RTA) (Figure 2.15). These accounted for 65% of the total reported medico-legal accidental cases. RTA was followed by deaths due to falls from heights which accounted for 13%, followed by deaths due to drug over dose which accounted for 6%. In contrast, deaths due to accidental burn injuries were 5% of the total accidental cases. Almost similar results were found in deaths due to asphyxia (3%), occupational injuries (3%) and drowning (2%). The least medico-legal cases reported were for deaths due to poisoning, totaling only

(1%) of the total accidental death cases. Death due to electrocution accounted for (1%) of accidental death cases.

Road traffic deaths are a major global health and developmental problem, not only in the Middle East Region but also worldwide (Baldwin, 2010). An understanding of the existing burden of road traffic deaths in Kuwait population is necessary for developing effective preventive strategies. Kuwait has the world's highest traffic accident death rate, and the country is spending \$95 billion (USAD) in tackling the issue (WHO, 2010). Despite its small total population of around three million inhabitants, Kuwait has witnessed an increasing rate of road accidents and injuries. In Kuwait, most accidents can be attributed to errors by the drivers (reckless or irresponsible driving). Road designs are critical in addressing the problem of traffic density and congestions. These are due to the lack of an effective traffic routing model in Kuwait. This is one main reason for the traffic congestion that has plagued the country (Al-Hassan, 2011). Speeding is another problem as well as pedestrians crossing the roads without paying attention to incoming vehicle resulting in their deaths.

Another important issue in accidental deaths is falls from heights. In this study, falls are responsible for 16% of the total reported medico-legal cases of accidental deaths in Kuwait. In Turkey, 89% of reported medico-legal cases due to falls from heights were accidental deaths and 11% were due to suicidal death. The majority of falls were from balconies and rooftops due to the tendency of residents to either sit or sleep on these places during the hot seasons (Kuruc et al., 2009). Little is known about the epidemiology of

falls from heights in the Middle Eastern populations. In particular, there are no published reports from any of the Arab Countries. A study was conducted at a specialized orthopedics hospital in Kuwait which provides services to the public in three Governorates which represent about 62% of the total population of Kuwait. The data show that 85% of the reported medico-legal cases occurred among non-Kuwaiti nationals (Figure 2.16).

2.5.8.1 Gender distribution in reported medico-legal accidental death cases

Throughout this study, males outnumbered females in all categories of accidental deaths (Figure 2.17). Males rank first in death cases due to RTA (males 75% compared to females 25%). In accidental deaths due to burn injuries, males accounted for 52% compared to 48% for females. The same is also true for deaths due to drug over dose where males accounted for 91% and females accounted for 9%. Similarly, males rank first in deaths due to drowning and electrocutions accounting for 77% and 88%, respectively. The results also revealed that males rank first in deaths due to falls from heights accounting for 73%. Deaths due to mine explosion were 94% among males. The results show a significant difference ($p < 0.000$) in death categories for males compared to females. This is in accordance with a study conducted in Malaysia showing that accidental death rates among males are significantly higher compared to females (Cai-Lian, 2011). The marked differences in the percentage of reported medico-legal cases of deaths due to accidents among males compared to females may be due to the lack of attention and negligence of safety protocols among males especially since the major portion of responsibility weighs on their shoulders in Kuwait. These results are more

or less similar to the percentage of reported cases among males who died due to falls from heights especially in occupational injuries. This occurs mainly among window cleaners and construction workers. Similar findings were reported in United State where younger males are most vulnerable to accidental deaths in the construction industry (Sheikhazadi et al., 2010).

2.5.8.2 Accidental deaths according to nationalities

During the period of this study in Kuwait, the reported medico-legal cases for Kuwaitis showed a significantly higher percentage of mortality in all categories of accidental deaths except in the cases of electric shock, fall from height, mine explosion and occupational injury (Figure 2.16). In deaths due to RTA, the rate was 57% among Kuwaiti residents compared to 58% for deaths due to fire, 59% for drowning, 58% for poisoning, 62% for asphyxia, while non-Kuwaiti reported higher mortality in deaths due to electrocution accounting for 73%, 74% for falls from heights, 90% for mine explosions, and 86% for occupational injuries (Figure 2.16). The results show that there was a significant difference ($p=0.000$) using ANOVA test in all categories of accidental deaths classified according to nationalities comparing Non Kuwaiti Nationals with Kuwaiti Nationals. One possible explanation for the difference is that Kuwait is a Welfare State so Kuwaiti citizens can easily obtain these drugs without paying attention to the costs. A previous study in USA, reported that both un-productivity and psychological distress are responsible for the high incidence of drug overdose among nationals (Kirsi et al., 2004). Similar findings have been found in Saudi Arabia, where reported medico-legal cases

of deaths due to drug overdose were most common among Saudi nationalities (Al Jahdali et al., 2004).

2.5.8.3 Age distribution among road traffic accidents victims

The present study revealed that the majority of the victims who died from RTA were young people in their productive years of life between 20-29 years representing 27% of reported medico-legal cases (Figure 2.24). In Kuwait, most accidents can be attributed to speeding and careless driving, and traffic rules are rarely obeyed and often resulting in horrific accidents (Christen, 2006). Children sit in the front seat, and this also makes it a major risk factor for RTA. The lack of an effective traffic routing model in Kuwait is also another main reason for the traffic congestion that has plagued the country (Al-Hassan, 2011). All these factors contribute to road accidents mortality.

2.5.8.4 Effect of age distribution in fire (burn) deaths

Deaths by fire are responsible for a significant number of mortality and morbidity worldwide and they are among the most devastating of all injuries leading to physical impairments, disabilities and to emotional and mental stress and consequences (Gupta et al., 2004). In Kuwait, the present results have revealed that the highest age group in deaths due to fire was reported for the 0-9 years accounting for 25%, followed by the age groups 30-39 years accounting for 21%, 20-29 years accounting for 16% and 40-49 years and 10-19 years accounting for 13% each (Figure 2.23). From the reported medico-legal cases, the results show that in Kuwait, there was a low incidence of deaths due to fire from burn injuries. Survival rate of fire victims is

improving constantly due to recent advances in treatment. However, prevention remains an issue of concern. This situation is now becoming grimmer due to the lack of burn prevention programmes and organized burn care units at the primary and secondary health care levels. In addition, most caregivers or house servants are uneducated in fire prevention because they come from countries where little or no fire training is available. Similarly, their employers do not provide any training of safety guidelines (Porter and Gavin, 2010). Deaths due to burns being higher in the 0-9 age groups could be due to the young age of the victims and inadequate knowledge and care by caregivers/ servants. Though Kuwait is a rich country, it is highly dependent on house keepers and baby sitters. Caregivers are mostly uneducated (no training) and they often possess limited knowledge in dealing with fire and fire-related incidences. Given these drawbacks, standard quality care is obviously not possible. Attention is needed to reduce burn incidence not only in Kuwait, but worldwide (Liao and Rossignol, 2000; Atiyeh et al., 2009).

In many countries like India with low income and high poverty levels, the rates of accidental deaths due to burning have increased, whereas in developed countries like USA, UK, France and Germany, there has been a decrease in deaths due to fire (Sauvageau and Yesovitch, 2010). An epidemiological survey is important since it will form the basis for a preventive programme in Kuwait as an extremely heterogeneous society.

2.5.8.5 Reported accidental deaths due to drug over dose

The present results show that the reported medico-legal accidental deaths due to drug overdose in Kuwait between 2003 to 2009 revealed that

males accounted for 91% compared to 9% for females (Figure 2.17). During this study, a total of 201 Kuwaiti residents died from drug overdose compared to 52 reported cases among Non-Kuwaiti residents accounting for 79% in Kuwaiti nationals. (Figure 2.16). The highest age group in accidental death due to drug overdose was reported in younger individuals between the 20 to 39 years accounting for 69%. The age group 40-49 years accounted for 23% of the total deaths due to drug overdose while the age group 50-59 years accounted for 4% (Figure 2.22). In contrast, the age groups 10-19 years and over 60 years accounted for 3% and 3%, respectively (Figure 2.22). The age group 0-9 showed the lowest number with 1% of deaths due to drug overdose. Despite the steady efforts made by Kuwait Ministry of Interior to combat drug trafficking at their source, the figures still reflect the continuous loss of lives among young individuals. Family awareness is needed first, followed by community alertness to eliminate bad habits in order to save lives. Strict law and religious doctrine in Kuwait forbid drugs, providing a significant cultural deterrent and moreover, there are also strong criminal legislations against the use of illicit substances. Together, these may be responsible for the decrease in the rate of drug abuse in Kuwait.

2.5.8.6 Reported accidental deaths due to drowning

Drowning is a common cause of accidental deaths not only among children and teenagers but also among adults and the elderly. In Kuwait, most deaths due to accidental drowning involved accidents in swimming pools, bathing tubs, boating or other water sporting activities. The reported medico-legal cases of accidental deaths in Kuwait due to drowning revealed that the highest age group of accidental deaths was reported among children in the

age group 0-9 years accounting for 36%, followed by similar results in the age group 10-19 years and 20-29 years who accounted for 15% each (Figure 2.21) This decline is more than two-fold. There is a prevalence of higher incidence among the younger age group. The age group 30-39 years accounted for 13.9% compared to age groups 40-49 years and 50-59 years who accounted for 7% and 8% of reported deaths, respectively. The lowest incidence of drowning was reported in the age group 60 years and over accounting for 5%.

In Kuwait, about 72% of the houses have a built-in swimming pool either indoor or outdoor (Ministry of Planning Report, 2011). Children are left to swim alone without proper supervision by an expert swimmer. Normally, a baby sitter or a house keeper has to keep an eye on the children but because of neglect and improper training, children usually drown. Similar findings were reported by Quan and Cummings in 2003. In 2010, a UNICEF report estimated that almost half a million deaths worldwide are caused by drowning (UNICEF, 2010). It was the second leading cause of injury-related deaths with 57% of these being children up to 14 years of age. Quan and Cummings (2003) found that drowning was a frequent cause of accidental deaths and hospital admission in 26 of the world's richest nations (Quan and Cummings, 2003). In United States, several studies which have investigated deaths due to un-intentional drowning during 1998 through 2008 found more children died in the lower age group (UNICEF, 2010). These studies have recommended that all children should be taught to swim after 3 years of age or even earlier. Although it seems obvious that better swimmers would be less likely to drown, the relationship between swimming lessons, swimming ability and the risk of

drowning is unknown. Comparisons of practices in regions with varied drowning rates might lead to new insights for prevention. In recent years, the incidence of accidental drowning of infants and children in Kuwait is on the rise especially in the age group from 1-4 years accounting for 42% compared to other age groups. Complete and consistent documentation of the circumstances surrounding drowning would greatly facilitate these efforts (Quan and Cummings, 2003; Adi et al., 2010).

2.5.8.7 Reported deaths due to poisoning according to age group

The reported medico-legal cases of accidental deaths due to poisoning according to different age groups revealed that the highest incidence occurred in the age group 30-39 years and they accounted for 42%, followed by the age group 20-29 years who accounted for 40% (Figure 2.20). Deaths due to poison declined from 42 % to 14 % in the age group 40-49 years. Similar results were registered in the age groups 10-19 years and 50-59 years age groups accounting for 2% each. No case of accidental death due to poisoning was reported in the age groups 0-9 years and 60 years and over in the present study. These findings are contradictory to the results reported by another study in which Kivisto et al (2009) reported that the risk of deaths from both intentional and unintentional poisoning persisted in young adults (Kivisto et al., 2009). However, Kivisto et al (2009) reported no accidental death cases due to poisoning in the age group 0-9 years and 2% less deaths in the age group 10-19 years in Malaysia (Kivisto et al., 2009).

2.5.8.8 Reported deaths due to electrocution

Electrical burns which are usually preventable with simple safety measures are associated with significant morbidity and mortality worldwide (Eren et al., 2007). A recent study has reported that the majority of deaths due to electrocution were accidental and the remaining cases were suicidal in nature (Sheikhazadi et al., 2010). The reported number of medico-legal cases of accidental deaths due to electrocution in Kuwait between 2003 and 2009 has been classified according to different age groups. The results revealed that the highest rate was reported in the age group 20-29 years accounting for 37%, followed by the age group 30-39 years who accounted 24%. The age groups 40-49 years and 50-59 years accounted for 16% and 12%, respectively. The age groups 0-9 years and 60 years over accounted for only 5% each. The lowest incidence of deaths due to electrocution was reported in the age group 10-19 years and they accounted for 2% (Figure 2.19).

The present results have shown that Non-Kuwaiti residents were more vulnerable to accidental deaths due to electrocution and they accounted for 73% compared to Kuwaiti nationals who accounted for 27% (Figure 2.16), Males outnumbered females in deaths due to electrocution accounting for 88% (Figure 2.15). Employment category was not included in this study due to lack of information, though many cases of occupational injuries and deaths were received in GDCE. These reported cases consisted mostly of male who had little or no professional training in handling electricity and in simple safety procedures. These accidents and deaths occurred mainly at homes and also at work especially in electrical construction occupation. Construction workers are still about four times more likely to be electrocuted at work than workers in

all the other industries combined (Eren et al., 2007). A recent study has shown that electrocution continues to be a significant cause of occupational deaths. Although crude fatality rates showed a falling trend, electrocutions were ranked as the second leading cause of deaths among construction workers, accounting for an average of 15% of traumatic deaths in the industry from 1980 to 1991 (Brunel et al., 2010). The results of this study have indicated that the workers most at risk of electrical injuries are male electricians, structural metal workers and labourers, similar to those reported by other study (Brunel et al., 2010).

Electrocution is not an un-common cause of deaths (WHO, 2006). A study in Gauteng, South Africa between 2001 and 2004 has reported a higher number of electrocution-related fatalities in the region compared with the rest of the world (Blumenthal, 2009). Similarly, the highest rates also occurred among American Caucasians and Indians who worked in the construction, mining, agriculture, forestry and fishing industries and also in trades associated with these industries (Taylor et al., 2002). Previous studies conducted in Australia have indicated that deaths due to electrocution were the lowest among accidental deaths and this is explained by the high awareness by individuals to safety procedures (Eren et al., 2007; Khandekar et al., 2008). In order to overcome this problem, public awareness programmes on electrical safety procedures in both the domestic and public (occupational) environment need to be introduced in order to prevent fatalities. Workers need to be provided with safety training and employers, in particularly smaller employers, need to be persuaded to provide safety training for their employees (Ingoldsby and Callagy, 2010).

2.5.8.9 Reported deaths due to falls from heights

Falls from heights are frequent and in homogeneous events which can occur at all ages (Glover et al., 2004). During the period of the present study, 29% of such cases were reported among the age group 30-39 years, followed by the age group 20-29 years who accounted for 27% (Figure 2.25). Likewise, the age groups 40-49 years, 0-9 years and 50-60 years accounted for 13%, 16% and 6%, respectively. The results also revealed that the age groups 10-19 years and 60 years and over accounted for the lowest incidence of 5% and 3%, respectively. The results also showed that males (73%) outnumbered females (27%) (Figure 2.17). The major cause of falls from heights was due to window cleaning both at homes and at work, especially from large buildings (Ministry of Interior Report, 2011). The deaths usually occurred because the workers failed to observe simple safety precautions during climbing. It is particularly noteworthy, that poor accident records and reporting systems tend to hide the extent of the construction safety problems and issues in Kuwait. In addition, many people at management level are unaware of accident-related costs and the effectiveness of a safety programme in reducing project costs. A study was conducted in Kuwait University, Department of Civil Engineering, presenting an analysis of construction-related accidents in Kuwait along with the causes of accident injuries that were fatal. The research confirmed that construction was the most hazardous industry in Kuwait, with accidents accounting for 48% of work-related-deaths in 1996 (Ministry of Planning, 1997). This rate is considered very high and can be compared to construction accident statistics in the U.S.A. which accounted for only 14% of all work-related deaths in the same year. Based on that study, falling from a height

appears to be the major cause of construction injuries and fatalities in Kuwait (Thierauf et al., 2010).

2.5.8.10 Reported accidental deaths due to mine explosion according to age group

The reported medico-legal cases of accidental deaths due to mine explosion in Kuwait during this study period showed a significant ($p < 0.000$) difference using ANOVA test in males compared to females, with males accounted for 94% of such fatalities (Figure 2.17). Most reported cases of deaths occurred among the age group 20-29 years accounted for 42%. Similarly, the age groups 30-39 years, 40-49 years and 50-59 years accounted for 35%, 16% and 4%, respectively (Figure 2.19). It is noteworthy, that accidental deaths that were reported due to mine explosions were mainly as a result of Iraqi invasion to Kuwait in 1990 resulted in mining a sizable part of the country. The Iraqi regime occupied the State of Kuwait for a period of seven months. During this period, the Iraqi Armed Forces brought massive quantities of army materials into Kuwait. Since liberation, 11 years ago, about seven million landmines have been identified in Kuwait by the force of the mine explosion. These run across Northern side of the border to Southern Kuwait, and in all over the desert area, each varying in depth from 60 to 150 meters. Casualties in Kuwait since that period were high and they reached hundreds of cases and especially both civilians and military personnel close to the Northern border. This is the area where there are many farms and many shepherds (mainly expatriates) who take their sheep around the farms. As a

result, they end up dead because of stepping on one of the mines hidden by the Iraqi troops. Cases of mine explosions are widespread in countries that suffer from war including Iraq, Iran and Afghanistan which have about 10 deaths due to mine explosions every week (Dogan et al., 2010).

2.5.8.11 Reported deaths due to asphyxia according to age group

In this study, several forms of asphyxia were combined because of the small number of cases. These include suffocation, choking, positional asphyxia/wedging in children and suffocative asphyxia due to different gases and other accidental types. The results for the study period from 2003 to 2009 revealed that the highest number of reported medico-legal cases of deaths due to asphyxia was in the age group 0-9 years accounted for 36%, followed by age group 30-39 years who accounted for 16% (Figure 2.26). Of the total number of reported death cases from asphyxia, the age groups 20-29 years, 10-19 years and 60 years over accounted for 14%, 8% and 7%, respectively (Figure 2.26).

The results revealed that the lowest death rate due to asphyxia was reported in the age group 50-59 years who accounted for 4% compared to the highest number in the age group 0-9 years who accounted for 36%. The increase in the number of reported cases in the very young age group may be due to the fact that young children between 0-6 years are at an increased risk and more vulnerable to asphyxia. Similar findings have been reported in USA (Egge et al., 2010). Young children are particularly vulnerable to asphyxia due to choking on food because of their small size and immature anatomy during early developmental stage in life and moreover, their inability to bite and chew

food, as well as the smaller diameters of their airways (Boghossian et al., 2010).

The majority of reported medico-legal cases to the GDCE during the duration of this study came from different housing areas in Kuwait which are prone to accidental deaths. During the present study, a number of accidental deaths (14 cases) due to gas poisoning (phosphine) were reported. The basements in these houses were rented to Companies that used the basement as a store for commercial items. The Companies also used insecticide to prevent insects from penetrating there products. As a result, phosphine gas could spread easily from the basement to flats and floors above where the families live. Following these reported incidents, the Ministry of Planning in Kuwait banned the use of basements for the storage of insecticides. Previous studies have found that most accidental deaths due to asphyxias were very common in young males and this could be easily prevented if these people were properly educated on preventative measures and moreover, if they had receive the necessary rehabilitation following an asphyxia insult (Belviso et al., 2003; Azmak, 2006).

2.5.8.12 Comparison of reported deaths in the Middle East Countries

The present study also compared the rate of suicide and homicide within the Middle Eastern Countries. The recent available data were obtained from WHO for the year 2002 for suicidal deaths, whereas, the data for homicidal deaths were obtained for the year 2010. The results show that the least suicide rate was recorded in Syria accounted for 0.2 per 100000

followed by Jordan and Qatar which accounted for 0.5 and 1.1 per 100000, respectively (Table 2.6). The highest rate within the Gulf Countries was recorded for Bahrain accounting for 5.4 per 100000. Whereas, the highest rate in the Middle Eastern Countries was recorded in Israel. Kuwait had a suicidal rate of 3.2 per 100000 which is considered to be high compared to neighboring Saudi Arabia with a suicidal rate of 1.2. The study also compared the homicidal rate in year 2010, which revealed that the highest rate of homicide was recorded in Brazil accounted for 23. In contrast, the lowest homicidal rate within the Middle Eastern Countries was recorded in United Arab Emirates which accounted for 1.38. In general, insufficient resources have been devoted to the field of forensic death investigations in the Middle East as a region and Kuwait as an independent nation. For example, in Iraq, it was very difficult to get a clear picture of reported unnatural deaths from official sources (WHO, 2008). Presently, the socio-political climate in Iraq has translated into a loss of data. Furthermore, information is not registered properly in the system and the authorities do not compile statistical data in general about unnatural death cases. No data sets were available to correlate suicides with different age groups. Moreover, there was no statistical analysis of any available as to the sex of the persons who committed suicide. All the reported suicides were committed in the urban areas, especially in Baghdad (Koronfel, 2002). Whereas, in Bahrain, an investigation done in year 2000 on suicide cases from the years 1955 to 1975 indicated that eight of the twelve suicides were committed by persons 18 to 30 years of age (Al-Mahroos, 2000).

It is generally agreed that current statistics on the incidence of unnatural deaths concerning homicide, suicide and accidental deaths are grossly inadequate and that their comparisons based on the figures available are at best inaccurate and often misleading (Daradkeh, 1989; Koronfel, 2002).

The results presented in this chapter of the thesis have clearly highlighted the causes of reported medico-legal cases of un-natural deaths due to suicide, homicide and accidents to the GDCE in Kuwait during the 7 years of the study. The study also examined a number of important parameters which are associated with the reported death figures to ascertain the human and environmental factors involved with the deaths. These include age groups, Governorates, gender, nationalities and other parameters. This is the first report of its kind in Kuwait and Middle Eastern societies in investigating the causes and patterns of all unnatural deaths. It is anticipated that the data and recommendations can be used to prevent such unnecessary deaths in Kuwait.

2.6 Conclusions

1. The present study has identified homicide, suicide and road traffic accidents to be more prevalent in the over-populated Farwania Governorate which is inhabited mainly by low income foreign labourers. Generally, it is a deprived area of Kuwait lacking educational and social services as well as health cares.
2. Furthermore, the results revealed that young adults belonging to the age group of 20-29 years constituted the majority of reported medico-legal cases of un-natural deaths.
3. The study has demonstrated that both the pattern and rate of reported homicidal deaths in Kuwait are alarming, with very high percentages of homicides due to stab wound injuries and more so, by firearms.

Moreover, the study revealed that levels of gun violence are higher in Kuwait as compared to other Middle Eastern countries. This may be attributed to the availability of firearm after Iraqi war. Nevertheless, these findings are of particular interest to the Government and they have to take urgent measures in reducing the ownership of legal as well as illegal firearms and knives.

4. The results show that hanging was the most preferred suicidal mode of deaths in Kuwait in both Kuwaiti and Non-Kuwaiti nationals and the present study concluded that Kuwait has the lowest rate of suicide compared to other Middle Eastern countries. Moreover, falls from heights were more common among males compared to females and this could be the easiest and most available method of deaths.
5. The results show that the leading cause of accidental deaths in Kuwait was by road traffic accidents. Despite its small population, RTA in Kuwait ranked first compared to other Middle Eastern countries.
6. In conclusion a close analysis of the present study showed that accidental deaths were the major cause of the reported cases of unnatural deaths in Kuwait over the stipulated study period. Moreover, the accidental death rate in Kuwait is considered very high when compared to other Middle Eastern and Western countries.

CHAPTER 3

A Comparative Study of the Usage of Virtual Autopsy and Conventional Autopsy in Detecting Causes of Un-Natural Deaths:

A Forensic Investigative Study

3.1 Introduction

3.1.1 Scope of this chapter

The traditional means of postmortem investigations to detect the cause of death represents invasive "body-opening". The importance of autopsy is well known and recognized as a valuable source of information. The decreasing trend in the frequency of autopsies has become a serious issue in forensic examination (Aghayev et al., 2008; Thali and Viner, 2010). To overcome this issue, deployment of the latest radiological imaging techniques known as "Virtual Autopsy" or "Virtopsy" could be of benefit to the forensic science field (Alfakih et al., 2004). With the 'virtopsy' technique, a scalpel never has to touch the victim's body as it is being examined (Ross et al., 2008; Thali and Viner, 2010). Deeper layers of tissues can be looked at and studied. In this way, bullet pathway, stab wound patterns, skull indentations and other forensic evidence can be carefully examined and documented without worry of crucial evidence being destroyed. The focus of this chapter is to assess the effectiveness of such procedure (Virtual Autopsy) as a tool in the forensic investigations of un-natural deaths Fig 3.1.



Fig 3.1: A photograph showing the forensic equipment for the CT scanner.

3.2 A review of virtual autopsy compared to conventional autopsy

The term 'virtual' is derived from the Latin word 'virtus' which means useful and efficient. The term 'autopsy' is a combination of the Greek term 'autos' (self) and 'opsomei' (I will see). Thus, autopsy means 'to see with one's own eyes' (Alfakih et al., 2004). At times, the subjectivity implied by the term 'autos' are eliminated and, the terms 'Virtual' and 'Autopsy' are merged, deleting 'autos', to create the term 'Virtopsy' (Bruschweiler et al., 2003).

The U.S. armed forces have been performing a version of a virtual autopsy on all service men and women killed in Iraq and in Afghanistan since 2004 (Thali and Viner, 2010). The armed forces are using both virtual and traditional autopsies for comparison to help Pathologists to create a huge database to enable them investigate the condition of the body for clues to the cause and manner of death. (Thali and Viner, 2010). In Washington D.C, the institutional review board Armed Forces Institute of Pathology conducted a study on 13 soldiers who died from gunshot injuries. Virtual autopsy was performed prior to the traditional autopsy. Images were evaluated for lethal wounds, number and location of wound tracks, injured structures, and metal fragment location. These, in turn, were compared to the findings and interpretations obtained from traditional post-mortem examination (Levy et al., 2006).

In 2003, Thali and his colleagues performed more than 100 Virtopsies in Kolkata. Every virtual autopsy was followed by a traditional autopsy. The study confirmed that virtual autopsy visualization enabled Forensic Pathologists to observe conditions of organs that were difficult or impossible to detect by traditional means (Thali et al., 2003). At present, there are only a few institutions worldwide that have recognized the possible impact of cross-sectional imaging in postmortem investigation and have invested efforts in its implementation (Ross et al., 2008; Thali and Viner, 2010).

A study was conducted in Switzerland, the main country using virtual autopsy, to evaluate the usefulness of this procedure in cases of hanging and manual strangulation. The results have revealed that strangulation signs were concordant with the forensic pathology findings. The main radiological findings in the neck and cervical spine region were thyroid and hyoid fractures, in addition to ruptures of the ligaments of the cervical spine. In manual strangulation, the primary findings were accurately depicted, with the exception of one slight haemorrhage. All frequent collateral signs could be diagnosed radio-logically, apart from one vocal cord haemorrhage. Traumatic lymph node haemorrhages were found in all manual strangulation cases (Grabherr et al., 2007; Grabherr et al., 2010).

3.3 Historical Background

'Virtual autopsy' concept of post-mortem imaging derives its roots from Japan and Switzerland following a series of high profile murders (Jeffery, 2010). In the middle of the 19th century, Virchow introduced microscopic examination to classic pathology, thereby helping to establish modern pathology, a discipline that has since exerted a great influence over the medical and forensic world (Levy et al., 2007). In 1996 Michael Thali at the University of Bern, Switzerland, developed the virtopsy, combined his reconstructions of bodies, with 3D reconstructions of crime scenes and

objects or weapons that may have caused the injuries or deaths. In doing so, he was able to obtain a clearer picture of the sequence of events that caused the injuries (Thali et al., 2003; Levy et al., 2007).

In most of the cases, traditional autopsy can be replaced by a non-invasive virtual autopsy and when necessary, minimal invasive image-guided tissue sampling from one body-location can be made. Virtual autopsy has the potential to gain high acceptance within the population compared to the traditional or conventional autopsy, making it possible to maintain high quality control in forensic and traditional medicine (Ross et al., 2008). New possibilities became available for Forensic Pathologists, with the advance of medical technologies such as computed tomography (CT) introduced by Hounsfield and Cormack in the early 1970s (Aghayev et al., 2008). In Forensic Science, a CT scan was first used on victims of gunshot injury to the head as early as in 1977 (Yen et al., 2005). A growing number of countries, including UK, Australia, Switzerland, USA, and Japan, are implementing virtual autopsy as an alternative to opening dead bodies in suspicious circumstances (Levy et al., 2007).

3.4 Main aim of the study

The main aim of this study was to evaluate the usefulness of virtual autopsy technique in establishing the causes of deaths in different categories of un-natural deaths, including firearm injuries, RTA, drowning and violent asphyxia. By assessing its performance as an aid in post-mortem imaging for forensic evaluation, the introduction of such technique might be of immense importance for forensic investigation in Kuwait to limit the medico-legal cases that undergo traditional autopsy.

3.4.1 Specific aims

* To validate this new approach by systematically comparing the radiological and surface scanning findings with those obtained at traditional autopsy in different un-natural causes of deaths.

* To speed the forensic examination process in cases of RTA, drowning and other un-natural causes of death cases.

* To locate the entrance and exit of firearm wounds.

* To implement radiological imaging to benefit forensic sciences in cases of homicide, when law enforcement needs a quick results of the causes of deaths

3,5 Materials and Methods

3.5.1 Materials and Tools used in the study

The main tools employed in this study were already being implemented in the daily forensic practice for virtual autopsy:

- 3D photogrammetry-based optical surface scanning.
- MSCT and MRI.
- Shandon Tissue processor.
- Shandon Tissue embedding.
- Microtome.
- Shandon Auto-strainer.
- Gas chromatography/mass spectrometry

3.5.2 Methods

This study was prospectively conducted during the period March 2010 through April 2011 at the CT scan section, Selfridge Base, Michigan, USA with collaboration between the Radiology and the Forensic Medicine Departments of Macomb County. The project had the relevant ethical clearance from the Ethics Committee from UCLAN, GDCE in Kuwait and Forensic Medicine Department in Macomb County, USA.

3.5.3 Study group

The study population included a total of thirty (30) male bodies with age range of 18-65 years, (mean age being $42.3 \pm$ years). These cases were received as routine forensic cases that underwent routine autopsy. The manner of death for the thirty bodies included 10 cases of homicide by firearm injuries, 11 cases of road traffic accidents (RTA), 2 cases of strangulation and 4 cases of accidental drowning, one case blunt force injury and 2 cases of fall from height. The identity and circumstances of all the cases were kept confidential for the purpose of this study. Cases were transferred from Selfridge air base to the Macomb County Medical Examiner Office wrapped in a special forensic case bag with a red tag to conserve the identity of each cadaver and differentiate them from other bodies received during the study period (Figure 3.2).

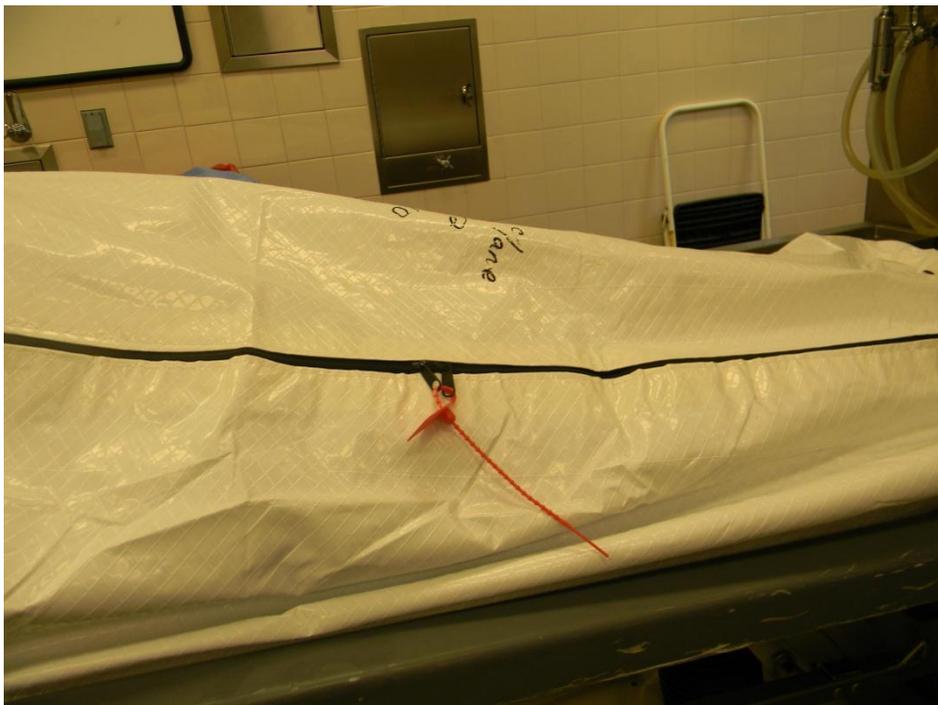


Figure 3.2: A photograph showing the forensic bag and the red tag on the cadaver for the purpose of the present study.

3.5.4 Post-mortem Radiological imaging (MSCT and MR imaging)

All the 30 deceased cases were imaged prior to autopsy with total-body multi detector CT scan (Light- Speed 16; GE Medical Systems, Milwaukee, Wis) within a few hours after the deceased bodies were received at the Macomb Medical Examiner's Office. Thereafter, full body MRI and CT scans of the vital body regions (head, neck, thorax, abdomen and pelvis) were carried out in all cases. The total MRI and CT scan durations for each case were between 20 to 45 minutes. MSCT and MR imaging data were transferred to a virtual autopsy table (Leonardo, Siemens medical systems, Erlangen, Germany). The scanning protocol was the standard plain scan protocol for every region with reference to pitch, exposure and other technical factors.

3.5.5 Image Interpretation and data analysis

The findings were reviewed by two qualified Radiologists (D. R. Macenzy and Dr. S. Wilkowski) and other experts in CT scanning at the Selfridge Air Base. This was a blinded study for the whole team. Each case was analyzed for the following findings: soft tissue injuries of the skin, subcutaneous fatty tissue; post-mortem alterations (e.g., heat induced injuries, signs of decomposition), temporal muscles injuries, fractures of the skull and skull base, the facial bones and cervical vertebrae 1 and 2, intracranial hemorrhage, extra and intra-axial haemorrhage, brain contusions and lacerations; injuries of the cerebellum and brain stem; lacerations of the dura mater and foreign bodies all over the cadaver. Following the scan, each image was interpreted for each cause of death in the present study.

A: Image interpretation in firearm injuries

The images were evaluated for firearm injuries to determine the lethal gunshot wound and the number and location of all gunshot wound tracks. Wound tracks in firearm injuries in the present study were identified by several indications and observations including: the presence of gas, metal distribution in soft tissues and

haemorrhage. The direction of bone, metal fragments and the beveling of bone defects were used to determine the bullet trajectory. Once a track was identified, it was visually connected to a possible entry and exit defects in the skin. Entry wounds were also suggested when there were bone or metal fragments driven in beyond the skin or bone defect. The finding of gas alone within vascular spaces was excluded because intravascular gas can be a normal decomposition finding in the postmortem state.

B: Image interpretation in drowning case

In order to diagnose drowning as a cause of death, the whole Respiratory System was evaluated to determine either the presence or absence of fluid and/or sediment in the para-nasal sinuses, frothy fluid and fluid in the mastoid air cells. The lungs were evaluated for parenchymal consolidation and pleural spaces were assessed for evidence of effusion. The stomach was evaluated for evidence of fluid and gastric distention (Yen et al., 2005). While during autopsy, complete dissection and gross examination of the intracranial contents, oral cavity, neck, chest, heart, mediastinum, abdomen and pelvis were performed in each subject. The final determination of drowning as the cause of death was made on the basis of data derived from the scene investigation and a combination of supportive autopsy findings. These included cerebral oedema, laryngeal oedema, hyper-inflated lungs, congested or oedematous lungs, presence of fluid and sediment in the subglottic airways, frothy fluid in the airways, fluid and sediment within the sphenoidal sinus, and evidence of swallowed water (Levy et al., 2006).

C: Image interpretation in strangulation cases

Images were interpreted according to presence of soft-tissue emphysema below the strangulation mark and finding of air ascends from a pneumomediastinum. These were caused by a rupture of alveoli during the attempts of breathing against occluded airways, this finding served as a vital sign. In addition, the presence of

subcutaneous and intramuscular haemorrhage in manual strangulation was also investigated.

D: Image interpretation in RTA cases

Images in the present study were interpreted according to the findings of either damage or fracture to the head, neck, thorax, abdomen, and other part of the body and findings of internal haemorrhage. Injuries were interpreted according to the detect of subcutaneous cavity is formed in which blood and liquefied fat collect also the present of fat contusion at the site of impact.

E: Image interpretation of blunt force injuries

In these cases images were visualized for head, spine, and pelvic injuries, fracture, haematomas, internal haemorrhage and air-filled cavities. Images were also interpreted for cerebral contusions in the head. Low attenuation oedema was visualized adjacent to contusions. Similarly, the presence of epidural haematomas, intracranial lesions, haemorrhages and subdural haematomas were also investigated.

3.5.6 Forensic autopsy

The author of this thesis performed the autopsies (upon receiving permission from both the Macomb County Sherriff Department and Wayne State University) under the supervision of the Chiefs Forensic Pathologist, Dr. Werner Spitz and Dr. Daniel Spitz and anatomical dissection was carried by the same forensic experts. All three body cavities (cranium, thorax and the abdomen) were examined in each case.

3.5.7 Toxicological analysis

In all 30 cases of the present study, a comprehensive toxicological examination was performed. The samples were analysed in the toxicological laboratory of the Department of Forensic Medicine, Faculty of Medicine at Wayne

State University, Michigan. Toxicological examination was done to determine whether common drugs, alcohol or other intoxicants had been used. The samples taken for each case were femoral blood, urine, and vitreous humour for ethanol analysis, stomach contents and liver for basic drugs and blood for carbon monoxide. Estimation of the levels of alcohol was quantitated mainly by computerized dual column gas chromatography method from the blood samples (Kentaro et al. 2006; Bhullar, 2011).

3.5.8 Microscopic examination

In all cases, as routine work in Macomb Medical Examiner Office in Michigan, tissue samples were taken from different part of the body depending on the case. These included brain, lungs, bronchi, bronchial lymphatic nodes, larynges, heart, thyroid gland, liver, spleen, pancreas, suprarenal glands and kidneys. Samples were processed as follow:

- All samples were dissected to about 5 mm thickness by Chief Forensic Pathologist Dr. Daniel Spitz then all following steps of tissue processing were performed by the author of the present study.
- All the samples were fixed with the buffered formalin for at least 24 hour except for brain tissues were fixed by formalin 20% were used. In cases were bones and cartilage being examined are fixed with 10% formic acid for 17 hour (Figure 3.3).



Figure 3.3: A photograph showing dissection of heart tissue in the gross laboratory after it was fixed for 24 hour in buffer formalin.

- Samples were inserted in capsules and an identification tag was inserted to each capsule for case number and type of specimen (Figure 3.4).



Figure 3.4: A photograph showing heart tissues which were inserted in cassettes and identification paper was inserted with each specimen including case number and type of specimen.

- Once each tissue was fixed, it was processed into automatic tissue processor for 24 hours for dehydration and clearing. (Figure 3.5).



Figure 3.5: A photograph showing tissue processor which contained tissues for processing over a period of 24 hours.

- Tissues were embedded in paraffin then sectioned using Microtome at anywhere from 3 to 10 microns and inserted into water bath at 36 C and lifted onto a slide (Figure 3.6)



Figure 3.6: A photograph showing tissues trimming using Microtome.

- The sections were stained with automated staining using the routine Hematoxylin and Eosin stain. Thereafter, the slides were observed by the doctors performing the autopsies. Slides were examined for histological diagnosis.

3.5.9 Correlation of autopsy and MSCT/MRI results

Autopsy and imaging results were compared and correlated. In this evaluation and validation of injuries, the question specifically addressed was whether MSCT and MRI predicted the relevant classical autopsy findings of all the different causes of deaths in the 30 cases analyzed. Final autopsy reports were written with input from the whole team.

3.5.10 Data analysis

Data analysis was based on a direct correlation between autopsy findings and radiological imaging. A retrospective analysis of the radiology data was performed with regard to the relevant forensic findings that had mostly been unknown to the evaluating Radiologists and Pathologist. Radiological findings constituted MRI and CT scanning findings, in which specific findings was evaluated for each cause of death that was considered in this present study. Data were obtained by calculating the percentage and presented as numbers and percentages.

3.6 Results

3.6.1 Comparison of traditional autopsy and virtual autopsy (CT scanning and MRI imaging) in firearm injuries

A total of 10 cases of firearm injuries were examined for the detection of several findings and comparing these data with CT and MRI with conventional autopsy. All of these cases were males, with age range of 21-56 years, with the mean age of 41.1 years. Rigor mortis was present in all 10 cases causing difficulty in true positioning of the body particularly of lower limbs. These findings are presented in Table 3.1.

Table 3.1: Table showing pathological findings for comparison of autopsy and virtual autopsy (CT scanning and MRI imaging) for firearm injuries in 10 different cases with age range of 21-56 years.

| Pathological findings detected | Findings detected in autopsy (%) | Findings detected in CT (%) | Findings detected in MRI (%) |
|-----------------------------------|----------------------------------|-----------------------------|------------------------------|
| Air embolism in the heart | 3 (30%) | 10 (100%) | 10 (100%) |
| Air embolism in the blood vessels | 0 (0%) | 10 (100%) | 10 (100%) |
| Blood aspiration to the lung | 10 (100%) | 8 (80%) | 10 (100%) |
| Wound entrance | 10 (100%) | 10 (100%) | 10 (100%) |
| Wound exit | 8 (80%) | 10 (100%) | 10 (100%) |

| | | | |
|--|-----------|-----------|-----------|
| Bullets location | 10 (100%) | 10 (100%) | 10 (100%) |
| Fracture pattern | 9 (90%) | 10 (100%) | 10 (100%) |
| Gunshot residues deposited within and under the skin | 10 (100%) | 9 (90%) | 10 (100%) |

The results of the present study presented in table 3.1 reveal that Multi-detector CT and MRI aided in the correct prediction of all 10 cases in identifying the gunshot wound tracks and the certainty of entrance and exit of the wounds as well as the detection of wound tracks were identified in all 10 cases. There were a total of 48 wound tracks identified at autopsy. The number of wound tracks identified in Multi-detector CT were 42/48 wounds. Similarly, MRI also aided in the correct prediction of 40/48 of the gunshot wound tracks. The remainder of wound tracks that were not identified prospectively at multi-detector CT and MRI include those in which they were located in the upper extremity of the body and may be missed out due to perceptive errors. The findings of the directions of the gunshot wound paths were accurately detected radiologically in both MRI and CT scan in all 10 cases.

The results of the present study showed that radiological imaging in both MRI and CT revealed a correct evaluation of the bullets location in all 10 cases accounting for 100%. Properties of the projectile, size and shape of the fragment, location in the body were also detected in both MRI and CT scanning prior to the autopsy as shown in (Figure 3.7). Whereas, with conventional autopsy, only 8 cases identified the exit of the bullet and this may be due to lead fragments that are smaller than a millimeter and which cannot be visualized by the naked eyes. Gunshot residues deposited within and under the skin were detected in all 10 cases during autopsy and MRI, whereas, they were found in only 9 cases using CT scanning. Finding of fracture pattern in the body was detected in 9 cases by traditional autopsy and in all cases using both CT and MRI (Figure 4.8).

The results of this study also show that CT and MRI can document vital reaction to the gunshot by demonstrating findings of air emboli in the heart and blood vessels in all 10 cases, accounting for 100%. In contrast, during conventional autopsy air embolism in the heart was detected in only 3 cases. In addition, air embolism in the blood vessels was not seen at all in all 10 cases by traditional autopsy.

With regards to the finding of classic pattern of blood aspiration to the lung, this was identified in all 10 cases in both traditional autopsy and MRI. However, during radiological imaging by CT scanning blood aspiration was detected in only 8 cases accounting for 80%.



Figure 3.7: A photograph showing transverse CT image with the entry of gunshot in the chest as shown by the arrow on upper left side.

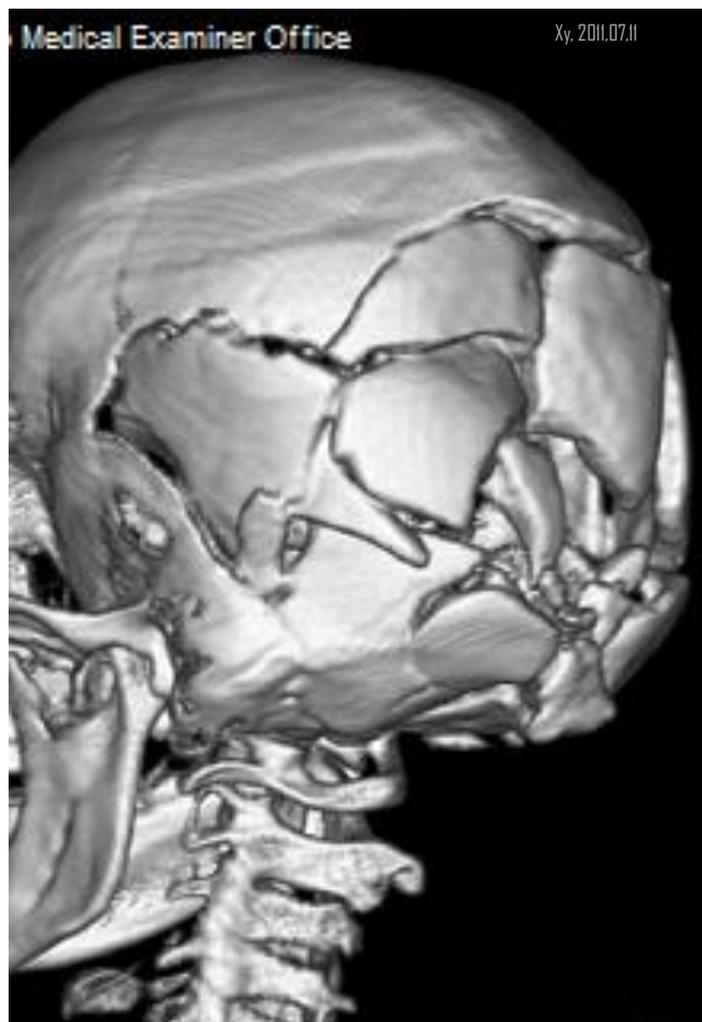


Figure 3.8: A photograph showing a three-dimensional volume-rendered image of gunshot wound to the skull with multiple fractures of the posterior skull.

3.7.2 Postmortem computed tomography findings in RTA cases

A total of 11 cases of road traffic accidents were examined for the detection of several findings and comparing these findings with CT and MRI with conventional autopsy. All of these cases were males, with age range of 18-65 years, with the mean age of 35.18 years. These findings are presented in Table 3.2.

Table 3.2: Comparison of pathological findings between virtual autopsy and conventional autopsy in investigating accidental deaths due to road traffic accidents in 11 cases between ages of 18-65 years.

| Pathological findings | Autopsy | CT | MRI |
|-------------------------------------|-----------|-----------|-----------|
| Subdural and extradural Haemorrhage | 11 (100%) | 11 (100%) | 4 (36%) |
| Skull and maxillary fracture | 11 (100%) | 11 (100%) | 2 (18%) |
| Oedema and heomarrhage | 11 (100%) | 8 (72%) | 11 (100%) |
| Soft-tissue emphysema | 11 (100%) | 11 (100%) | 10 (90%) |

| | | | |
|----------------------------------|-----------|-----------|-----------|
| Chest injury | 11 (100%) | 11 (100%) | 11(100%) |
| Hip injury | 11 (100%) | 11 (100%) | 11 (100%) |
| Spinal cord injury | 10 (90%) | 11 (100%) | 6 (54%) |
| Injury in other part of the body | 11 (100%) | 11 (100%) | 8 (72%) |

Table 3.2 shows correlation of pathological findings between virtual autopsy finding and conventional autopsy for un-natural death cases due to RTA. The results revealed that findings of subdural and extradural haemorrhage were present in all 11 cases of RTA in both conventional autopsy and CT scanning. In contrast, these were clear only in 4 cases of 11 using MRI. Similarly, skull and maxillary fracture were present in both conventional autopsy and CT imaging, whereas, only 2 cases out of 11 demonstrate skull and maxillary fracture. Findings of oedema and haemorrhage were present in both conventional autopsy and MRI and found only in 8 cases out of 11 constituting 72%.

The present study showed that tissue emphysema was detected in both conventional autopsy and CT scanning and was detected in 10 cases of 11 with MRI. Chest and hip injury was detected in all diagnostic studies and conventional autopsy in all 11 cases. Injury of the spinal cord was present in 10 cases of RTA by conventional autopsy, while, it was clear in all 11 cases using CT scanning as shown in figure 3.9 In contrast, MRI only detected 6 cases of spinal cord injury.

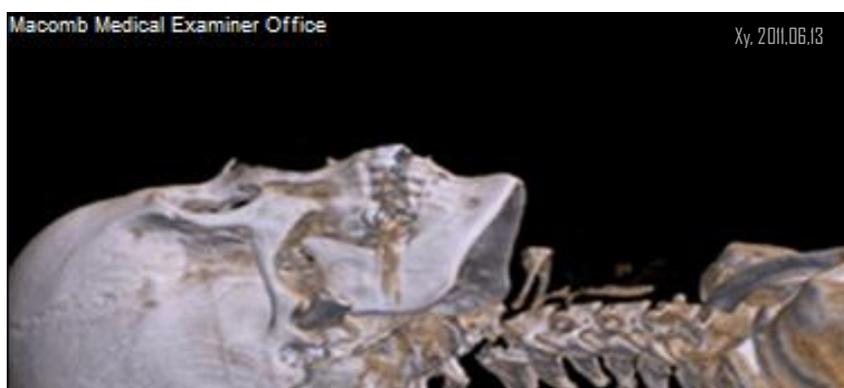


Figure 3.9: An 18 year old male died of RTA showing 3D post-mortem anatomy of a broken neck.

3.7.3 Postmortem computed tomography findings in cases of asphyxia

A total of 2 cases of homicidal death due to strangulation were examined for the detection of several findings and comparing these findings with CT and MRI with traditional autopsy. These two cases were for males, with ages of 30 and 41 years, with the mean age of 35.5 years. These findings are described in Table 3.3.

Table 3.3: Comparison of traditional autopsy and imaging findings in manual strangulation (n=2).

| Findings | Autopsy | CT | MRI |
|-------------------------------|----------|----------|----------|
| Subcutaneous haemorrhage | 2 (100%) | 2 (100%) | 2 (100%) |
| Intramuscular haemorrhage | 2 (100%) | 1 (50%) | 2 (100%) |
| Lymph node haemorrhage | 2 (100%) | 0 (0%) | 1 (50%) |
| Thyroid haemorrhage | 2 (100%) | 0 (0%) | 2 (100%) |
| Fractures of the hyoid bone | 1 (50%) | 2 (100%) | 2 (100%) |
| Soft-tissue emphysema | 2 (100%) | 0 (0%) | 2 (100%) |
| Fracture of thyroid cartilage | 1 (50%) | 1 (50%) | 2 (100%) |
| Vocal cord haemorrhage | 1 (50%) | 0 (0%) | 0 (0%) |

Table 3.3 shows a comparison of the CT scan and MRI findings with traditional autopsy for two homicidal death cases due to manual strangulation. During traditional autopsy there was externally evident of superficial injury on the neck of both cases due to pressure of finger nails. The results shows that subcutaneous haemorrhage was found in both traditional autopsy and radiological imaging (MRI and CT scanning) accounting for 100%. Similarly, intramuscular haemorrhage, was detected in 1 case out of 2 in CT scanning accounting for 50% and in MRI Intramuscular hemorrhage finding was detected. Lymph node haemorrhage finding was detected in traditional autopsy in both cases and was found in only one case of strangulation by CT scanning accounting for 50%. Interestingly, this finding was clear in both cases by MRI.

The results also showed that CT scanning and MRI imaging have both proven the finding of fractures of the hyoid bone in the two cases of strangulation (Figure 3.10) and this was detected only in one case by traditional autopsy. On the other hand, traditional autopsy detected the finding of vocal cord haemorrhage in one case only of the two and this was not detected in both radiological imaging (CT scanning and MRI). Postmortem magnetic resonance imaging (MRI) showed thyroid cartilage fracture in both cases. In contrast, (MSCT) and traditional autopsy shows these in both cases. Both findings of soft-tissue emphysema and thyroid cartilage fracture were present using MRI and traditional autopsy and these were not clear in CT scan.



Figure 3.10: A photograph showing CT scan of 41 years old male exposed to pressure neck injury showing hyoid bone fracture on its right limb.

In the laryngeal cartilages, mineralisation occurred of the larynx, in which superior cornu of the larynx meets in a point with the lamina of the thyroid cartilage making it highly vulnerable to fracture. The most intense effects of mechanical forces are to be expected here. For histological examination all larynges of the two cases of strangulation were fixed for at least 24 hours in buffered formalin and dissected into 8 mm. Thereafter, the tissues were incubated with 10% formic acid for 17 hour. After washing for more than 24 hour, the specimens were embedded in paraffin and cut into 4 mm thick sections. The results revealed that lesions were located in the peri-vascular space deep inside the cartilage showing erythrocytes in cartilage canals of the larynx (see figure 3.11)



Figure 3.11: Microscopic picture of 30 year old male showing peri-vascular erythrocytes in cartilage canals of larynx as a result of fractures of the mineralized superior cornua commonly occur in cases of strangulation.

3.7.4 Identification of drowning cases using virtual autopsy

A total of 4 cases of accidental deaths due to drowning were investigated in the study. Radiological Imaging by both MRI and CT scanning were performed and compared with traditional autopsy findings. The 4 cases were for males, with age range of 19 to 55 years, with the mean age of 36.75 years. The 4 cases were drowned in Great Lake of Michigan, USA, while fishing on frozen lake. These findings are described in Table 3.4.

Table 3.4: Table showing the correlation of virtual autopsy findings and autopsy findings in 4 cases of accidental deaths due to drowning.

| Findings | Autopsy | CT | MRI |
|--|----------|----------|----------|
| Frothy fluid in the airways or lungs | 4 (100%) | 1 (25%) | 4 (100%) |
| Hyper-inflated lung | 4 (100%) | 2 (50%) | 4 (100%) |
| Fluid and high-attention material in the paranasal sinuses | 4 (100%) | 4 (100%) | 4 (100%) |
| Fluid in the stomach | 4 (100%) | 1 (25%) | 4 (100%) |
| Gastric distention | 4 (100%) | 1 (25%) | 4 (100%) |
| subglottic trachea and bronchi fluid | 4 (100%) | 2 (50%) | 4 (100%) |
| Sphenoidal and ethmoidal sinus fluid | 1 (25%) | 2 (50%) | 4 (100%) |

| | | | |
|-------------------------------|----------|---------|---------|
| Congestion and edematous lung | 4 (100%) | 1 (25%) | 2 (50%) |
|-------------------------------|----------|---------|---------|

Table 3.4 shows radiological imaging of CT and MRI findings in 4 medico-legal cases of accidental deaths due to drowning in Great Lake in Michigan. The results revealed that all drowning cases had upper airway and sinus due to drowning. All drowning subjects had evidence of fluid in the para-nasal sinuses in all four cases (100%) in both traditional autopsy and radiological imaging. Two cases (50%) had fluid in the subglottic trachea and bronchi detected in CT scanning. In contrast, they were found in all four cases by MRI imaging. Hyper-inflated lungs were detected in 4 cases using MRI and traditional autopsy (Figure 3.12). Whereas, it was found only in two cases using CT scanning. Fluid in the stomach and gastric distention were detected in all 4 cases in traditional autopsy and in two cases using MRI, whereas it was only present in one case by using CT imaging accounting for 25%.

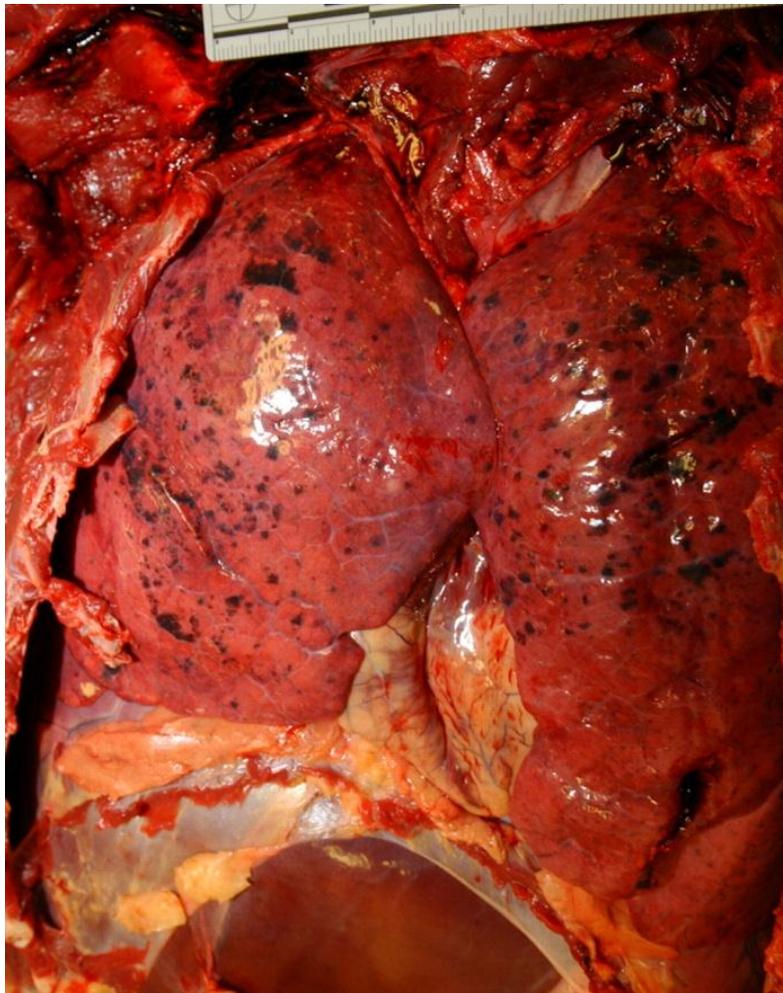


Figure 3.12: Image at conventional autopsy shows lungs of 40 year old male drowned in a lake. Showing lung congestion, oedema and hyperinflation with extension of the lungs across midline and black anthracotic pigment is also present.

3.7.4.1 Microscopical findings (lungs)

Classical histological examination of lung tissues using (Hematoxylin and Eosin staining) shows obvious intra-alveolar edema and dilatation of the alveolar spaces with secondary compression of the septal capillaries (Figure 3.13).

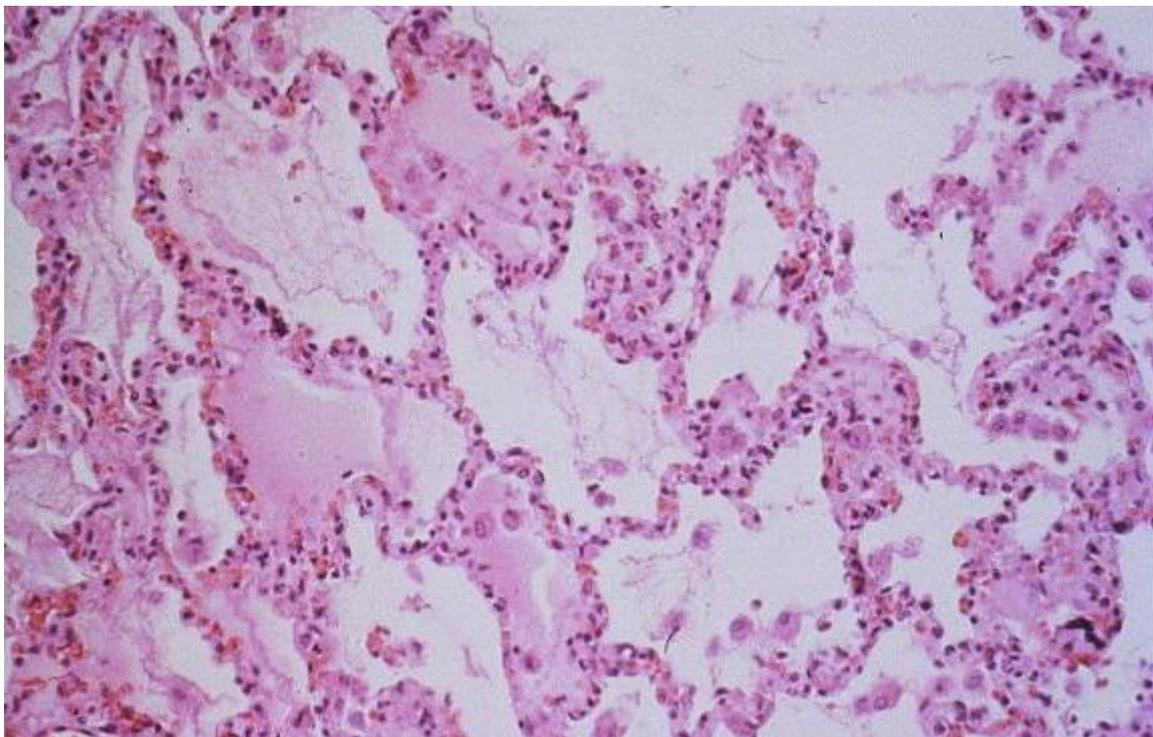


Figure 3.13: Microscopic slide of lung tissue showing obvious intra-alveolar oedema and dilatation of the alveolar spaces with secondary compression of the septal capillaries and lung parenchyma due to asphyxia as a result of drowning.

3.7.5 Head injuries identification by virtual autopsy

In this study, a total of 3 cases of head injuries were investigated using virtual autopsy. Radiological Imaging by both MRI and CT scanning was performed and compared with traditional autopsy findings. The 3 cases were for males and with an age range from 32 to 42 years, with the mean age of 38.6 years. All the cases were as follow:

- Case (1): A 42 old male, death due to blunt force injury using a hammer.
- Case (2): A 32 old male, death was due to accidental death owing to fall from height (fall from the roof of house).
- Case (3): A 42 old male, death due to head injury owing to backwards fall, shown in figure 3.14.

| Findings | Autopsy | CT | MRI |
|---------------------------------|----------|-----------|----------|
| Superficial lesions | 2 (66%) | 0 (0%) | 0 (0%) |
| Subdural and epidermal hematoma | 3 (100%) | 1 (33.3%) | 3 (100%) |

| | | | |
|------------------------------------|-----------|-----------|-----------|
| Oedema | 3 (100%) | 3 (100%) | 3 (100%) |
| Fracture of the occipital skull | 3 (100%) | 3 (100%) | 1 (33.3%) |
| Brain and cerebral contusions | 3 (100%) | 0 (0%) | 0 (0%) |
| Intracranial bleeding | 3 (100%) | 1 (33.3%) | 3 (100%) |
| Spinal cord injury | 1 (33.3%) | 0 (0%) | 2 (66%) |
| Injuries to other part of the body | 3 (100%) | 3 (100%) | 3 (100%) |

Table 3.5: Table showing the pattern of head injuries identified by traditional autopsy and radiological imaging in 3 cases with age range from 32 to 42 years.

Table 3.5 shows the pattern of head injuries due to different causes (homicidal and accidental) resulting in death. In case number 1 (a blunt force injury of a 42 years old male), the results showed local impression and ring fracture of the occipital skull was present in both traditional autopsy and CT scan and this was detected in only one case by MRI accounting for 33%. In the same case of the 42 years old male, the contusion and superficial lesions were missing in both MRI and CT while they were detected in only 2 cases during traditional autopsy. MSCT of the upper head of (case 2) of a 42 years old male, who had fallen backwards from a roof of his house showed subcutaneous tissue swelling and haemorrhage in the left occipital region (see figure 3.15).

The study also showed that subdural and epidermal haematoma was detected in both MRI and traditional autopsy accounting for 100%. In contrast, it was found only in one case of accidental fall from height accounting for 33% as shown in figure 4.16. In addition, the results showed that, cerebral contusions was found in all 3 cases of head injuries by traditional autopsy examination and was missing in both radiological

imaging CT and MRI. The finding of intracranial bleeding was detected in both traditional autopsy and MRI and was present in only one case in CT scanning accounting for 33%. Moreover, oedema was found in both traditional autopsy and radiological imaging in all 3 cases.



Figure 3.14: A photograph showing 42 year old male (case#3) in Macomb Medical Examiner morgue who died from falling backwards in which the skull shows no abnormalities from the outside.

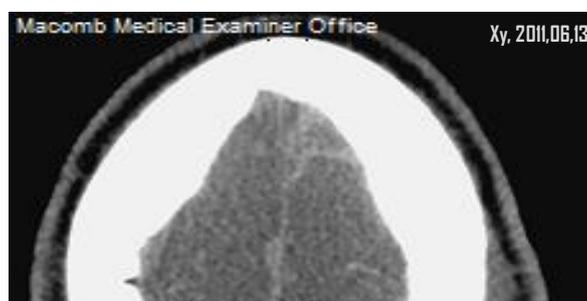


Figure 3.15: An image showing MSCT of the upper head (case 2) of a 42 years old male, who had fallen backwards from a roof of his house. The subcutaneous tissue shows swelling and haemorrhage in the left occipital region.

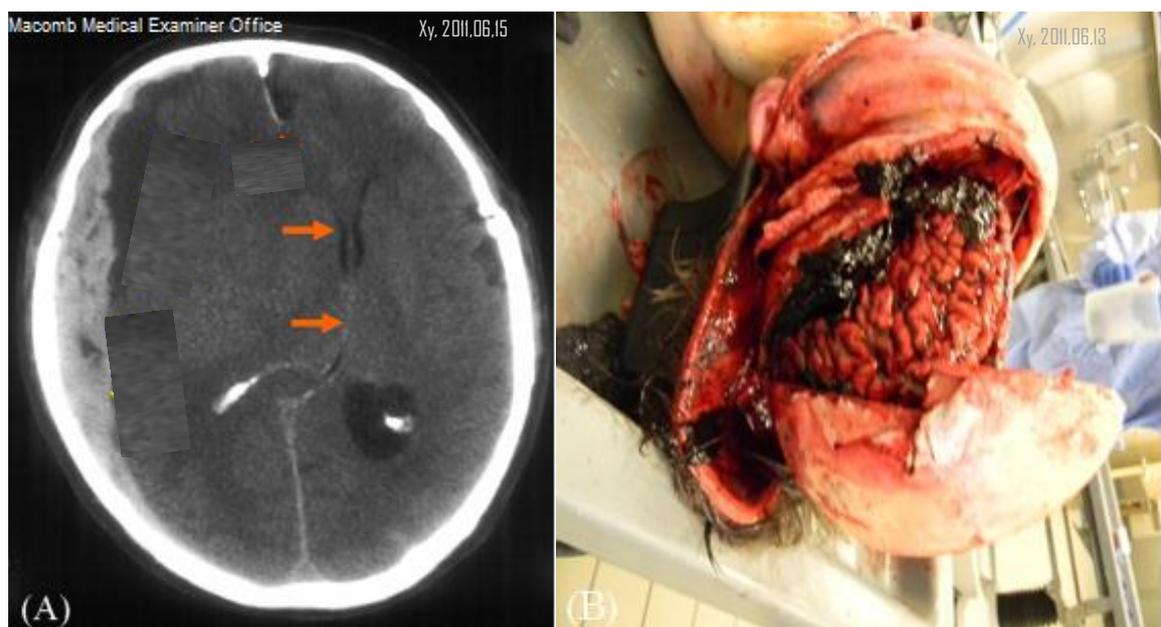


Figure 3.16: (A) MSCT image showing the same 42 years old male (case #3) and (B) conventional autopsy. Both images show subdural haematoma on the right side representing separation of the blood components and massive swelling of the brain with compression of the ventricles. Note that the midline shifting is easily detected in (A) this MSCT slice. (B) Similarly, the autopsy (B) findings show a good correlation to the imaging data in this case.

3.7.5.1 Toxicological findings

Toxicological analysis of all 30 cases of this present study revealed that alcohol was detected in 8 out of the 30 subjects accounting for 26.7%. The highest level of alcohol detected in the urine samples was 3.14 mg/100 ml, ranging from 1.5 to 3.14 mg/100 ml, with a mean value of 2.42 mg/100 ml. In death due to firearm injuries, they represent 20%. Two of these cases had alcohol in their urine. The ages of the males who had alcohol in their samples were between 21 and 26 years of age. The alcohol levels varied between 1.8 and 2.97 mg/100 ml. In cases of death related to RTA, alcohol was detected in 4 out of 11 subjects, accounting for 36.4% of the total RTA ranging from 1.5 to 3.14 mg/100 ml. No alcohol was found in cases of strangulation. In deaths due to drowning, alcohol was detected only in one case accounting for 25%.

The alcohol level was 2.1 mg/100ml in the subject. In death due to fall from height, only one subject had alcohol level of 3.02 mg/100ml. All together, the results clearly show that alcohol plays a major role in the deaths of these reported cases.

3.8 Discussion

Virtual autopsy is a novel, quick and unique technique which can be used to aid forensic investigation (Bolliger et al., 2010). The aim of this study was to investigate the effectiveness of this technique and also to assess whether it is possible to introduce this technique in Kuwait. Once established in Kuwait, this will be the first of its kind in Middle Eastern countries. The results show a very good agreement with conventional autopsy method compared to virtual autopsy. Although virtual autopsy was described in the literature since about 12 years ago, the use of it for forensic investigation has not been fully established worldwide (Cannan and Friedrich, 2010; Jeffery, 2010).

Although the present results were obtained from a small number of subjects, they still showed that virtual autopsy can reliably aid identification of several unnatural causes of deaths whether they were due to firearm injuries, accidental deaths

as a result of drowning and road traffic accidents. The present results showed that virtual autopsy was very helpful in identifying the cause of deaths in different circumstances. Moreover, this technique is very rapid without even using the blade, which is an important issue to be considered especially in Muslim Faith.

3.8.1 Identifying the causes of death due to head injuries

Virtual autopsy is useful to visualize and reconstruct blunt injury patterns compared to conventional autopsy. Moreover, it can facilitate the understanding of the mechanism of injury on the head, spine, and other injuries in the body. The present study investigated 3 cases of un-natural deaths due to head trauma. The results indicated that both radiological techniques showed findings of oedema, bleeding, hematoma and fractures. But it failed to identify superficial lesion and contusion. Similar study conducted by Jeffery (2010), concluded that MRI and CT scanning identified head trauma injuries in 3 cases out of a total of 9 deaths due to head trauma. (Jeffery, 2010). In contrast, Thali et al (2003) illustrated the effective use of both multi-detector computed tomography (MSCT) and magnetic resonance imaging (MRI) in locating head trauma injuries (Thali et al., 2003).

3.8.2 Identifying Drowning Cases

Investigation of a body recovered from water can be challenging. The determination of drowning as a cause of death requires a full investigation on the part of the police and Pathologist (Jeffery et al., 2011). The investigation should include a detailed account of the circumstances surrounding the death, and a full autopsy including microscopic examination and toxicological findings. Data that support the diagnosis of drowning include frothy fluid in the airways or lungs and fluid in the para-nasal sinuses or stomach. All of these findings were present in both virtual autopsy and conventional autopsy as shown in this study. CT and MRI results have indicated that all of the drowning subjects investigated in the present study had evidence of fluid in the Para-nasal sinuses in all four cases (100%). Subglottic trachea and bronchi

findings were present in all 4 cases in both conventional autopsy and MRI. While, it was present in 50% of the cases by CT scanning, (25%) had fluid in the stomach. Anatomic findings at autopsy supported the diagnosis of drowning which include the presence of frothy fluid in the airways and lungs, hyper-inflated and congested lungs, fluid in the para-nasal sinuses, watery fluid in the stomach, and dilated and engorged right-sided cardiac chambers and great vessels. Gastric distention was clearly found in MRI imaging in all 4 cases and only in 2 cases that were present by the use of CT scanning. Similar results were obtained by Jackowski in 2006 examining 28 male drowning victims (Jackowski et al., 2006). In 2005, Yen reported the findings of bilateral patchy or diffuse of ground-glass opacity in the lungs of drowning subjects using thin-section computed tomography (CT) (Yen et al., 2005). In 2010, Thali and Viner (2010) published findings of deaths by drowning in two subjects and described the spectrum of paranasal sinus fluid, patchy airspace disease, right atrial enlargement, and upper gastrointestinal fluid (Thali and Viner, 2010). In another study conducted by Levy and colleagues (2007) in The Armed Forces (Institute of Pathology, Washington, DC, USA), they reported similar findings on drowning cases (Levy et al., 2007).

3.8.3 Virtual autopsy in firearm injuries

Investigating un-natural causes of deaths related to firearm injuries can be very easily conducted using the virtual autopsy technique. Gunshot wounds in the head and neck were identified by the presence of gas and metallic fragments along the bullet paths connecting to entry and exit defects that were present along the skin surface. When the bullet passed through bone, spicules were embedded in the bullet path. The results have shown that gas, bone and metal were present in the brain parenchyma in those subjects with tracks through the brain.

Although the results of this study were obtained from a small number of firearm injuries, they nevertheless show that virtual autopsy can reliably aid identification of

firearm injuries. The detection of wound channels, as well as the certainty of entrance and exit wounds from the 3D MSCT data, can provide essential information for the investigation of the cause of death. CT features and MRI findings of the present study, accordance to gunshot wound tracks, were typically linear tissue defects that contain gas and metallic fragments.

The present results also show that virtual autopsy allows correct prediction of all cases of firearms wound tracks in gunshot victims up to 100% accuracy for both gunshot wound paths and entrance and exit of the pullet compared to 80% detection of the bullet exit during traditional autopsy. These findings are in complete agreement with other studies in which their data supported the present results (Grabherr et al., 2010). However, these other studies encountered some limitations to the identification of the correct gunshot wound path passing through extremities (Levy et al., 2007). These sites were not included in the field of view of the scan which may be attributed to the fact that the cases have sustained multiple gunshot wounds which made interpretation of the correct path impossible at CT (Levy et al., 2007). Also in other study conducted by same authors (Levy et al., 2006), they concluded that MRI and CT scanning did not show all paths of firearm injuries due to rigor mortis which affected the positioning of several victims. This in turn contributed to some gunshot wound paths that were missed in the elbow region. However, in the present study, rigors mortis did not affect the result of the study (Levy et al., 2006; Levy et al., 2007). Virtual autopsy would enhance the finding of the bullets making it much easier and faster for the Forensic Pathologist to identify the exact spot, instead of cutting the body in several places to find the bullet. With virtual technique, Pathologists could make a single incision (Grabherr et al., 2007; Grabherr et al., 2010). According to Thali and Viner (2010), a bullet hole is an obvious entry point, but once the lead enters the body, a bullet can ricochet off bones to rest in unexpected places (Thali and Viner, 2010).

3.8.4 Virtual autopsy of deaths due to strangulation

Although conventional autopsy is the best method used in strangulation cases, the 3D reconstruction of CT and MRI data can verify vital signs that are caused by strangulation. The 3D images can visualize strangulation marks that are of forensic importance and in reproducing the strangulation process (Jacobsen et al., 2008). The results of the present study showed correct correlation between conventional autopsy findings and MSCT. Both techniques demonstrate subcutaneous haemorrhage and fracture of hyoid bone and these were detected in both cases of strangulation.

Since there were a limited number of death cases due to strangulation in this study, more prospective studies will be needed to determine the real value of imaging in post-mortem forensic casework to identify the value of radiological imaging when determining the causes of death. In all cases of strangulation in this present study, MRI findings were superior to conventional autopsy in identifying fracture of thyroid cartilage. With regards to vocal cord haemorrhage, this was not detected in both CT and MRI screening since vocal cords were situated just between two sections and therefore could not be evaluated by MRI.

However, as yet, both CT and MRI with conventional autopsy cannot provide all the information that is expected by the Criminal Justice System in forensic cases involving strangulation. Further information has to be obtained by crime scene investigation. It is particularly noteworthy that one major advantage of CT and MRI application is that it is a very quick process and it takes as little as 30 minutes to perform, whereas a traditional autopsy can take anywhere from two hours to several days depending on the case (Spitz, 2006).

3.8.5 Limitation of Traditional Autopsy

Traditional autopsy has several limitations and they include the following:

1. Practically, it is a time intensive and an extremely messy procedure, energy consuming and moreover, it requires a lot of experience, patience and tolerance.
2. It is difficult to obtain information relating to the whole body during autopsy techniques. Dissections and investigations of some areas of the body may be difficult to perform by the Forensic Pathologist. Some of these areas include the entire examination of the spinal cord, pineal body, pituitary gland, bone of vertebral column and skin system. This difficulty can lead to inaccurate information and diagnosis of the precise cause of death of the victims.
3. Samples are taken for histological, microbiological, chemical and biochemical studies. Thus, normally forensic findings are documented in a subjective way (observer-dependent) and the findings that have been documented may be very hard to retrieve.
4. The incision runs from shoulder to shoulder, meeting in the mid chest region, and extending down to the pubic area (creating a Y-shape). This in turn will affect the family to see their loved ones in this way.
5. Sometimes, autopsy performance is rejected or not accepted by family members especially in the Arabic culture, in which families prefer to bury their loved ones very soon following death. This procedure has been recommended by Prophet Mohammad.
6. In Muslim cultures, when a person dies, his own family prefers to wash his/her body and not to let anyone else doing this. Therefore, when they see cuts of autopsy, from shoulder to shoulder, meeting in the mid chest region, and extending down to the pubic area (creating a Y-shape), this will psychologically hurt them for long period of time and make the washing process very difficult knowing that their loved ones have undergone such a procedure.
7. Traditional autopsy can take anywhere from a two hours to half a day depending on availability of Forensic Pathologist as well as by his/her experience.

3.8.6 Advantages of Virtual Autopsy

The appliance of full-body radiography in virtual autopsy in forensic investigation has several advantages as described below:

- It is considered as non-invasive or minimally invasive approach envisioned by post-mortem surface scanning.
- It plays an important role in the identification process of unidentified bodies in a very fast time.
- Virtopsy takes as little as 30 minutes to perform.
- Radiologists can create full 3D visualizations of the deceased, which allow Forensic Pathologists, Coroners and Medical Examiners to examine the condition of bones, tissues, organs and blood vessels for clues to the causes and manner of death (Bruschweiler et al., 2003).
- This technique gives precise, objective and clear documentation of forensic findings for the court and police authorities and this procedure does not damage or destroy key forensic evidence, as can happen during a conventional autopsy.
- This technique also provides quality assurance through digital data archivation and retrieval of the data at any time needed.
- The virtual autopsy helps to reduce any psychological trauma for the next-of-kin compared to conventional in which the body is dissected.
- There is also improved judicature in cultures with low autopsy acceptance.
- The technology can significantly help forensic and law enforcement efforts to quickly and accurately pinpoint the chain of events in any un-natural death case and also after a bomb blast or other terrorist attack. It also can be used to determine the causes of the death of a victim of a natural disaster event when dead bodies are already badly decomposed (Jacobsen et al., 2008).

- The Virtual Autopsy Table is a powerful teaching tool for educational purposes in a wide range of environments such as schools, universities, museums and Science and Technology centre (Thali et al., 2003).
- The advantage with virtual autopsy is that it does not destroy key forensic evidence which may be damaged during conventional autopsy as well as, Forensic Department has a permanent 3D record of all data which is not possible once the body is dissected.

3.8.7 Drawbacks of the virtual autopsy

- The major disadvantage of this technique is that it is very expensive to establish due to the cost of the equipment.
- It is very expensive to train an expert in using the equipment.
- It has some drawbacks in diagnosing toxicological deaths
- It requires the necessity of highly professional team to interpret the findings including both Radiologists and Pathologists.
- Not all Radiologists are used to interpreting postmortem radiological data.
- Virtopsy cannot tell you the colour of organs and it is important when trying to find inflammation.
- Similarly, virtopsy cannot distinguish the difference between pre and post death wounds (or artifacts found in the body post death),

3.9 Conclusion

Virtual Autopsy is a newly developed automated procedure that will facilitate and possibly overtake traditional autopsy, giving it the capacity to achieve more reliable results in Forensic investigation. At present, there are only a few institutions worldwide that have recognized the feasibility and possible impact of cross-sectional imaging in postmortem investigation. It is clear that the introduction of this high-tech

automated method can have a big impact on Forensic Medicine in general and also in Kuwait GDCE. Implementing this strategy for forensic investigation will improve the completeness and reliability of forensic investigation, it will shorten and speed up all the procedure for comforting the family in two ways: Firstly, by not cutting their loved one and secondly, by not delaying the burial of the body in the cemetery.

Current study provides evidence as to whether virtual autopsy is a good alternative to be used compared to conventional autopsy in determining the causes of death in trauma victims. These applications of virtual autopsy in forensic field have suggested that this technique could be used for postmortem data acquisition in cases of mass casualties such as airplane crashes or natural disasters (eg, the recent tsunami catastrophe in Asia and the mass murder of 212 females and children in Kuwait during wedding ceremony). In these incidents, identification of bodies is the major issue to be addressed, and CT can be of crucial value in disaster victim identifications.

Although a small number of cases were investigated in this present study, the results show the following:

- The CT scanning times are short (whole-body documentation takes 5–10 minutes), depending on the section thickness and the volume to be covered.
- Virtual autopsy can provide strong visual evidence for use any time and can be retrieved for courtroom proceedings since the digitally stored data may be recalled at any time to provide fresh, intact topographic and anatomic-clinical information.
- It will help in visualization of fracture patterns at the exit of the bone defects.
- It can also help in bullet track documentation and wound tract analysis and also the entrance and exit of firearm wound injuries.
- MR imaging has a greater impact in demonstrating soft-tissue injury as well as organ trauma for example as in suicidal, homicidal and accidental cases.

- Virtual autopsy is of great value as an additional method allowing perfect documentation of the gross findings without disfiguring the body as well as reducing, but not omitting the necessity of partial or targeted autopsy.

In conclusion, it is possible to recommend to the Kuwaiti Government the use and implementation of virtual autopsy in Kuwait. This technique can be combined with conventional autopsy to speed up the process.

CHAPTER 4

Time-Dependent Effects of Temperature and Humidity on Quantity of DNA in Samples of Human Saliva, Blood and Semen in Kuwait

4.1 Introduction

During the past two decades numerous advances have been made in the forensic science field and the use of DNA (deoxyribonucleic acid) evidence to either exonerate or convict people and to solve crimes worldwide (Kaiser et al., 2008). The availability of new and improved technology makes DNA analysis critical in the field of forensic science (Schneider, 2007; Hedman et al., 2010). The DNA of a person is unlike that of any other human, except in the case of identical twins (Walsh et al., 2010). Therefore, analysis of DNA evidence is extremely valuable and it is the best way to determine the identity of the person who left behind evidence at the scene of a crime in any form including body fluids such as blood, saliva, semen, sweat and such tissues as body teeth, skin, hair roots and bones (Castriciano et al., 2010; Dissing, 2010). It has also proven to be valuable for the identification of human remains. DNA has been important in revolutionizing the entire field of forensic

sciences. This impact is felt within the criminal justice system and contributes to the accurate safeguarding of society (Schneider, 2007). This chapter of the thesis investigated the time-dependent effects of temperature and humidity in the Kuwaiti environment on DNA degradation especially since Kuwait is considered as a very hot country.

4.2 DNA profiling in investigation of crimes

DNA analysis is useful for a number of different purposes in solving a wide variety of criminal investigative cases. This may involve cases such as homicide, sexual assault, physical assault, hit and run incidents, missing person investigations, identification of human remains, determination of paternity and several others (Castriciano et al., 2010).

Specifically, DNA can aid the investigations in the following circumstances (Dissing, 2010):

- Identifying the source of biological evidence found at a crime scene.
- Redirecting the investigation in a new direction.
- Linking serial crimes together.
- Identifying the number of assailants.
- Identifying additional victims.
- Exonerating people who have been wrongfully convicted of a crime they did not commit.
- Several others.

4.3 DNA polymorphisms

For the purposes of forensic science, DNA polymorphisms are specific sites in

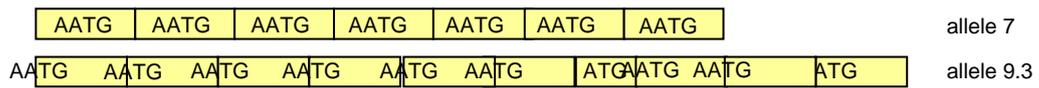
the genome where the precise sequence of DNA tends to differ in unrelated individuals. In forensic geneticists, there are regions within the genome that are hyper-variable and these have been the target for most forensic analysis (Holt et al., 2002).

Commonly used regions include sequences that are repeated tandemly. The original DNA 'fingerprinting' analysed mini-satellites are often referred to as variable number tandem repeats (VNTRs) (Butler et al., 2003). Short tandem repeat (STR) (also known as microsatellites) technology is currently used for DNA profiling where variability in alleles can be used to distinguish one DNA profile from another (Alonso et al., 2004). The odds that two individuals will have the same specific STR regions profile is about one in a billion or even higher (Piacenza and Grimme, 2004).

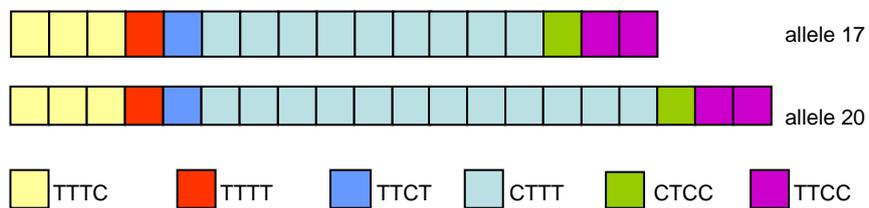
4.3.1 Short tandem repeats (STRs)

The human genome is full of repeated DNA sequences (Hedman et al., 2010). These repeated sequences come in various sizes and are classified according to the length of the core repeat units, the number of contiguous repeat units, and/or the overall length of the repeat region (Schneider, 2007). DNA regions with short repeat units (usually 2-6 bp in length) are called short tandem repeats (STR). STRs can be classified according on their structures as simple, compound, complex and complex highly variable (Lee et al., 2011) (See figure 4.1). STRs have proven to have several benefits that make them especially suitable for human identification (Kaiser et al., 2008).

THO1 – Simple repeat with a non-consensus allele



FGA – Compound repeat



D21S11 – Complex repeat sequence

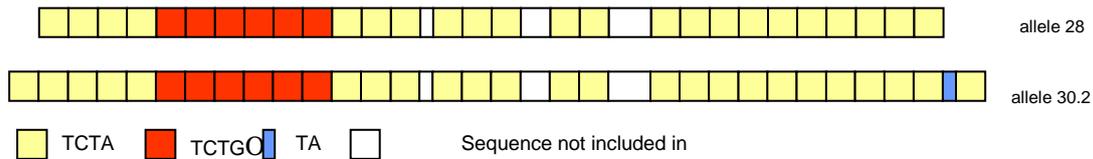


Figure 4.1: The structure of three commonly used STR loci, THO1, FGA and D21S11. The THO1 locus has a simple repeat with a non-consensus allele; in the example the 9.3 allele is missing the first A from the seventh repeat. The FGA locus is a compound repeat composed of several elements. The D21S11 allele is an example of a complex repeat; the three regions not included in the D21S11 nomenclature are an invariant TA, TCA and TCCATA sequence (Taken from Goodwin et al., 2007).

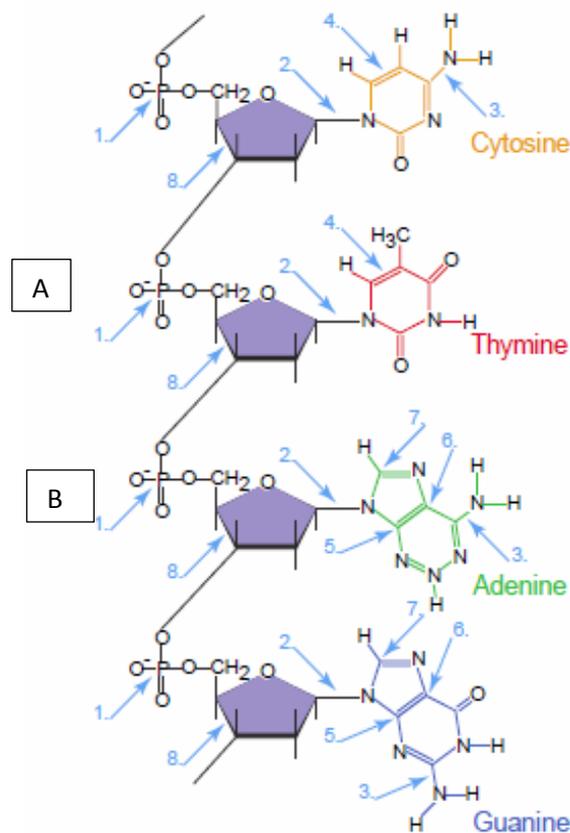
In 2001, Peter Gill, working in Forensic Science Service (FSS[®]) in the UK, developed and introduced the first multiplex which is widely used in forensic analysis (Gill, 2001). Further efforts have been made by the FSS[®] resulted in the development of the second Generation multiplex (SGM). This in turn incorporated six polymorphic STRs (TH01, VWA, FGA, D8S1179, D18S51 and D21S11) and the amelogenin marker (Sullivan et al., 1993) (See Table 4.1). Currently, the SGMPlus kit with 10 STRs is used in the UK, while the Federal Bureau of Investigation (FBI) uses a standard set of 13 specific STR regions for CODIS. The use of STR technique in forensic science is highly powerful due to several properties. First, it produces profiles that are highly discriminating. Second, the profiles of the STR technique are relatively easy

to generate. Third, the profile can be compared between laboratories. Finally, by using this technique it is possible to obtain results from degraded samples. In this study, the AmpFISTR® Identifiler® kit (Applied Biosystems) was used to generate DNA profiles from samples.

Table 4.1: The development of STR systems. Two STR systems, the quadraplex and SGM Plus were developed by the Forensic Science Service in the UK. The AmpFISTR® SGM Plus® became commercially available in 1998 and has been adopted by a large number of laboratories for routine forensic casework. The AmpFISTR® Identifiler® and PowerPlex® 16 both analyze 15 STR loci, including the 13 CODIS loci that are required to be analyzed for forensic casework in the USA. The two kits are used widely worldwide, particularly for kinship testing (Taken from Goodwin et al (2007)).

| | | | | |
|-------------|------------|------------------|---------------------|----------------------|
| QUAD | SGM | SGM Plus® | Identifiler® | PowerPlex® 16 |
|-------------|------------|------------------|---------------------|----------------------|

| Vwa | Amelogenin | Amelogenin | Amelogenin | Amelogenin |
|-------|------------|------------|------------|------------|
| THO1 | vWA | D3S1358 | D3S1358 | D3S1358 |
| F13A1 | D8S1179 | vWA | vWA | vWA |
| FES | D21S11 | D16S359 | D16S359 | D16S359 |
| | D18S51 | D8S1179 | D8S1179 | D8S1179 |
| | THO1 | D21S11 | D21S11 | D21S11 |
| | FGA | D18S51 | D18S51 | D18S51 |
| | | THO1 | TH01 | TH01 |
| | | FGA | FGA | FGA |
| | | | D13S317 | D13S317 |
| | | | CSF1PO | CSF1PO |
| | | | D7S820 | D7S820 |
| | | | TPOX | TPOX |
| | | | D5S818 | D5S818 |
| | | D2S1338 | D2S1338 | Penta D |
| | | D19S433 | D19S433 | Penta E |



C

D

Figure 4.2: A photograph showing the hydrolytic processes responsible for sugar backbone damage: (A) depurination responsible for base losses. (B) Deamination of cytosine, adenine and guanine. (C) Oxidative processes attack the double bond of C5 and C6 carbons of cytosine and thymine and the C4, C5 and C8 carbons of purines. (D) Oxidative attack of the sugar backbone can also occur (8). (Taken from Hebsgaard et al., 2005).

4.4 DNA evidence recovered from crime scenes

DNA evidence has become a standard forensic technique for investigating a wide spectrum of crime types ranging from burglary to murder (Bond and Hammond, 2008). DNA evidence found in the crime scenes often treated as crucial evidence by Forensic scientists to solve cases. If the evidence is successfully detected and analyzed then the identity of the individual can be determined.

Substantial DNA evidence can be recovered from crime scenes. The largest source recovered from crime scenes is mainly saliva via drinking vessels and cigarette ends (Barbaro et al., 2008). Whilst treated as a different source, blood is the next largest source recovered. Both of these sources are typical of DNA recovery at the crime scene that is used by an offender (Bond and Hammond, 2008). Other DNA evidence encountered in crime scenes including epithelial cells semen (sperm), urine, faeces, bone, hair shaft and other tissues. In forensic analysis, the quality of DNA evidence recovered from a crime scene is affected by several environmental factors that can lead to a highly degraded DNA resulting in a poor PCR amplification (Chung et al., 2004).

4.5 Factors affecting DNA degradation

DNA degradation can occur when samples have been exposed to several environmental insults and chemical factors. These include the following:

- Light (UV) (Dissing, 2010).
- Humidity has more of an effect on the quality of the DNA, rather than the quantity (Yang and Watt, 2005).
- Elevated temperatures and moisture will degrade DNA and will make it difficult to obtain a profile (Piacenza and Grimme, 2004).
- Fungal contamination (Alonso et al., 2004).
- Length of the postmortem interval (Holt et al., 2002).

The survival of DNA depends on several factors and environmental conditions, hence, the purpose of the present study was to investigate mainly the effect of various temperatures and humidity on DNA survival over time.

4.5.1 Effect of temperature on DNA degradation

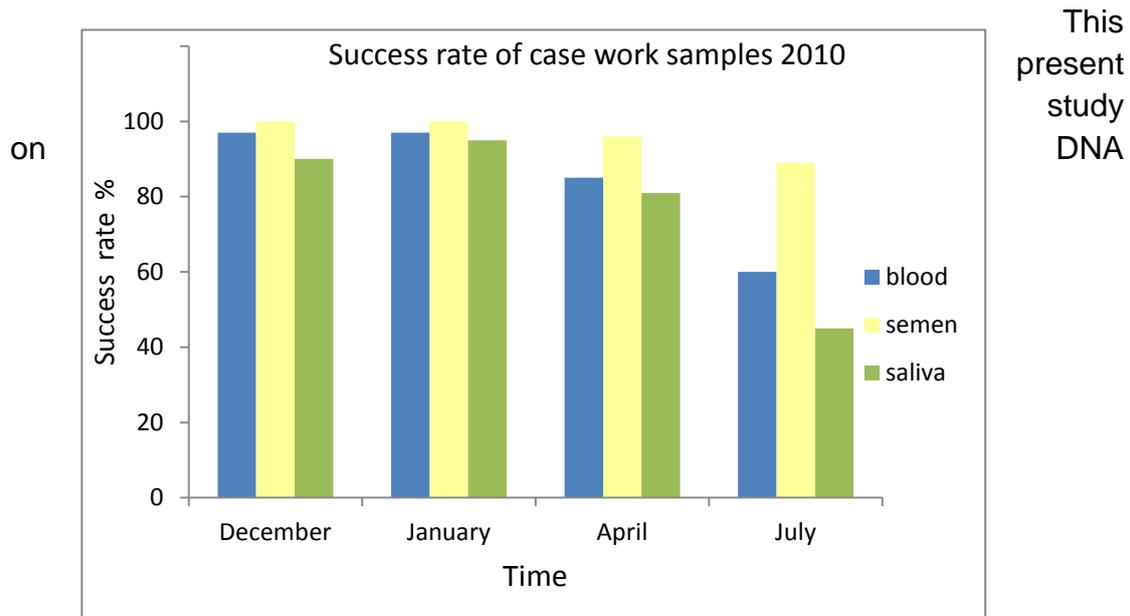
DNA can be rapidly degraded post-mortem, initially from the enzymes released as the cell dies losing its structural integrity and then later from environmental conditions (Clayton et al., 1995; Budowle et al., 2005). Temperature plays an important role in both DNA degradation and survival. DNA degradation depends both on its amount and type of damage that accumulates over time. This depends greatly on the conditions and integrity of the biological material when it is found. However, as temperature increases, damages can take place rapidly and this damage can accumulate with time (Alonso et al., 2004).

4.5.2 Effect of humidity on DNA survival

Weather forecast normally refers to humidity as relative humidity (RH). Relative humidity is defined as the amount of water molecules present in the atmosphere and the amount of water that the air can sustain at a certain temperature (Dissing, 2010). Exposing biological materials to an external environment of high quantity of water in the air and with elevated temperature can lead to the growth of microorganisms, such as bacteria and fungi (Davoren et al., 2007). The degradation of DNA often parallels the degradation of proteins and many of the processes that degrade DNA depend on the presence of water (Fisher et al., 1993; Hill et al., 2007). High relative

average humidity climates with 100% humidity, will enhance the appearance of fungal/microbial growth causing rapid decay of DNA at high relative humidity (Ovchinnikov et al., 2001; Palo et al., 2007).

4.6 Study which led to this investigation



measurement was conducted at Kuwait DNA Laboratory at the GDCE. Currently, Kuwait Identification DNA laboratory (KIDL) is facing a huge load of cases to be identified. For example, more than 1000 cases of paternity testing for DNA and a similar number relating to crime scenes are done annually. The Kuwaiti Government has also invested more than 750 thousand Kuwaiti Dinar (£1=0.44 KD) for the analysis of DNA samples annually and moreover, to solve cases urgently. To date, a lot of time and effort had been spent on DNA analysis at KIDL without initially investigating factors which could influence either DNA degradation or survival and whether the DNA was indeed present in each sample. In addition, most samples normally came from different parts of Kuwait and moreover, they were exposed to different temperatures and environmental conditions (insults) including humidity over time. Figures 4.3 and 4.4 show the percentage of successful rates of DNA survival from generation of DNA profiles during the months of December, January, April and July for 2010 and 2011, respectively. The data depicted in figures 4.3 and 4.4 clearly showed that successful profile generation was markedly decreased in July when the Kuwaiti weather was very hot and more success was obtained in December–January when the weather was cold. In many cases, it could be a waste of time, money and labour if no DNA was detected in a sample due its degradation. As such, it has now become apparent that initial studies must be done in order to investigate a number of parameters (eg temperature, time, humidity etc) on DNA degradation/survival in samples taken from human subjects. As such, this chapter of the thesis was designed specifically to undertake a feasible study in order to help the KIDL to minimize the cost, labour and effort in examining these numbers of cases. Such studies have never been undertaken before in KIDL.

Figure 4.3: Bar charts showing success rate of case work samples of human blood (blue), semen (yellow) and saliva (green) for DNA profiling in 2010.

The data in figure 4.3 illustrate the percentage profiles generated) from a total of 100 cases received in December, January, April and July for either blood, semen or saliva. Note that the successful rate of DNA measurement was markedly decreased in July (hot temperature) compared to the other months especially December (cold temperature).

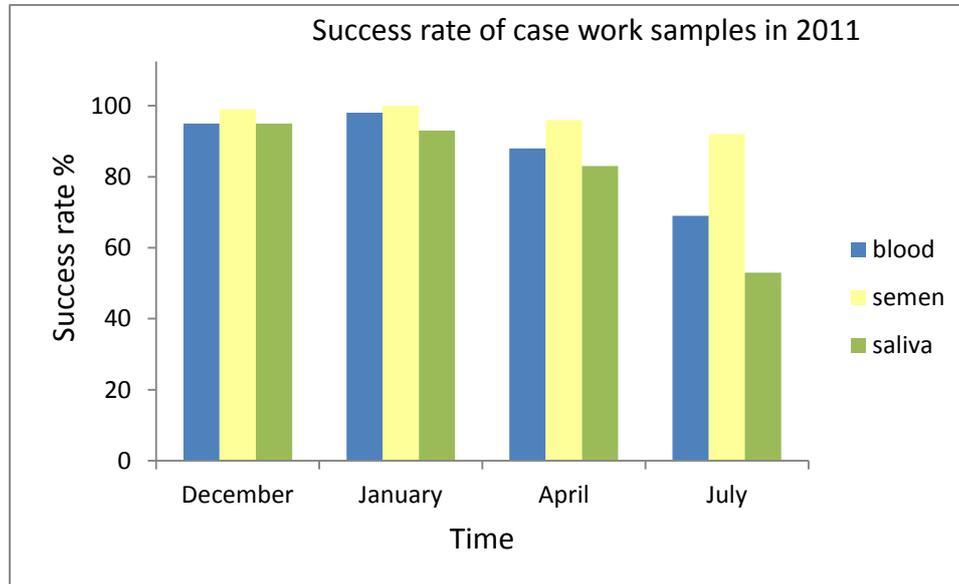


Figure 4.4: Bar charts showing success rate of case work samples of human blood (blue), semen (yellow) and saliva (green) for DNA profiling in 2011.

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markedly decreased in July (hot temperature) compared to the other months especially December (cold temperature).

4.7 Climate in Kuwait

This present study was conducted in Kuwait, a Country which is located at the Northwest corner of the Arabian Gulf and lies 30.05 degrees North of the Equator. Kuwait shares borders with the Kingdom of Saudi Arabia from the South and to the North and West, it shares borders with Iraq. Kuwait is Located in the desert geographical region famous for its very hot climate. In Kuwait, summer starts from the month of April and continues until September. The summer months are extremely hot and while the average normal day temperature is 37 °C (99°F) and this can rise up to an average of 55 °C (131°F) during the months of June, July and August. Another feature of Kuwait climate is that it rarely rains during the summer months and from June to September which are the driest months. Frequent dusty storms often occur during the summer months. Likewise in winter the temperature in Kuwait can drop to an average of 4 °C. The average highest humidity in Kuwait occurs in the month of December reaching 64% and the average lowest humidity is in July accounting for 41%.

4.8 Aims of the study

The main aim of this study was to determine the time course of DNA survival from such different DNA sources as blood, saliva and semen, which are usually found in crime scenes. A duration of 28 days was selected to give adequate time for DNA survival. These assimilated experiments were designed to show the length of DNA survival in four different temperatures (4,

24, 37 and 55 °C). In addition, at the same time, the study investigated how changes in the humidity (61, 58, 55 and 41%) could affect DNA degradation in tandem with the same corresponding temperatures, respectively (4, 24, 37 and 55 °C). These numbers represented the most apparent temperature and humidity changes in Kuwaiti weather in both summer and winter. Since the study was conducted in Kuwait which is characterized by a very hot weather in the summer and cold in the winter, it is crucial to verify the validity of DNA profiling for characterizing the genetic makeup of post-mortem biological evidence in such common temperatures and percentage of humidity in the area. In addition, the results may help in reducing cost for DNA measurements and analysis at the KIDL in the General Department of Criminal Evidence in Kuwait.

4.9 Materials and Methods

4.9.1 Materials

4.9.1.1 Samples

- a) Human blood
- b) Human saliva
- C) Human semen

4.9.1.2 Reagents

- a) Sterile water
- b) Alcohol (70, 80 and 95%)
- c) 0.5% Sodium hypochlorite
- d) DTT Dithiothreitol
- e) PK Proteinase K

4.9.1.3 Consumables

- a) 1.5 ml tubes
- b) Tips (10, 100, 200 and 1000 μ l) and Gilson pipettes.
- c) Microcon® -30 filter
- d) Cotton swabs
- e) SecurSwab S.I.T. Collector (bode tech)

4.9.1.4 DNA extraction and Quantification Kits

- a) DNeasy® Blood and Tissue Kit (Qiagen)
- b) QIAamp DNA Investigator Kit (Qiagen)
- c) Quantifiler™ Human DNA Quantification kit (Applied Biosystems)

4.9.1.5 Equipments

- a) Laminar flow hood
- b) CL-1000 UV cross linker
- c) Incubators
- d) Centrifuges
- e) ABI PRISM® 7500 Real-Time PCR system (Applied Biosystems)
- f) Fridges
- g) Freezers
- h) Temperature and humidity incubator
- i) Thermo-mixer

4.9.1.6 Contamination controls and Laboratory

Much of the work undertaken as part of this research involved the PCR amplification of DNA, often from a low copy number template. Precautions were taken throughout the study to minimise the probability of introducing contamination. Where possible, the study employed criteria that were used for analysing ancient DNA (Yang and Watt, 2005). Extraction blanks were included with all DNA extractions (1 blank per 15 extractions). All human samples were extracted in triplicates, with the extractions taking place at different times. The profiles were checked against a staff DNA database to detect any contamination introduced from laboratory staff. The study had the relevant ethical clearance from the Ethics Committee in GDCE and University of Lancashire.

4.9.1.7 Laboratory Design

DNA samples were extracted in the laboratories of the DNA Identification Laboratory in the State of Kuwait. The laboratories were organized to ensure the unidirectional flow of work also to be sure that no contamination may occur (See Figure 4.5).

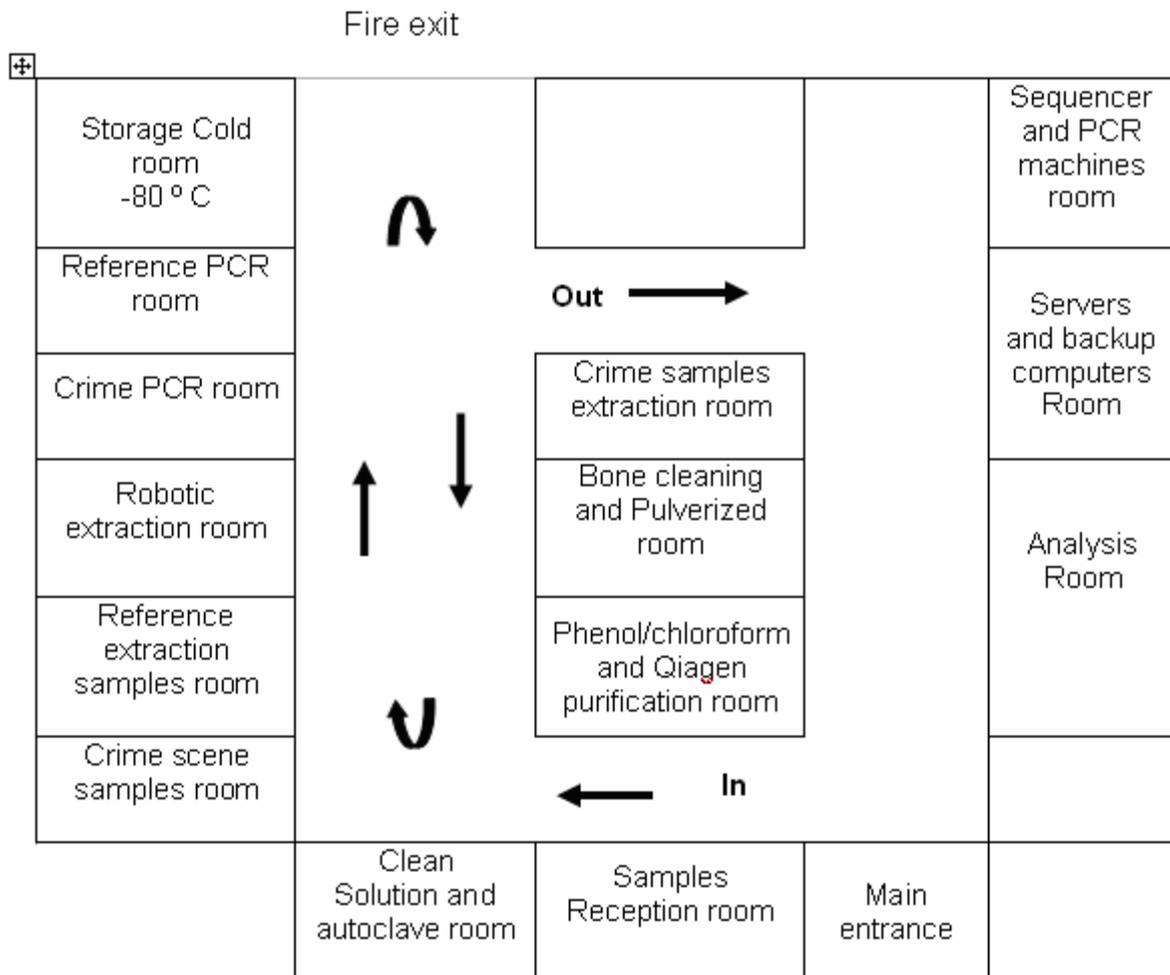


Figure 4.5: Design of the Forensic DNA Laboratory in the State of Kuwait. The laboratory was set up in an arrangement to minimize the contamination of samples (Taken from Al-Enizi, 2009).

4.10 Methods

In this study one sample of either liquid blood, semen or saliva were collected from one human volunteer and split into 28 fractions. The liquid saliva, semen and blood were collected in 1.5 ml tube. The experiments were done at four different temperatures (55°C, 37°C, 24°C and 4°C) and four different humidity ranges (41%, 55%, 58% and 61%), respectively, which are similar to most Kuwaiti weather conditions in summer, spring and winter times. All samples were exposed to the various temperatures and humidity ranges over a duration of 28 days of exposure. Each extraction was quantified in triplicate for each sample in order to calculate the mean and the standard deviation bar.

4.10.1 Sample preparation

All the samples were prepared as follow. A volume of 50 µl of blood was added on SecurSwab S.I.T. Collector (bode tech) in a total of 28 samples. A volume of 50 µl of saliva was added on buccal swab in total of 28 samples. A volume of 1 µl pure semen was diluted with 990 µl distilled H₂O and then the dilution was divided into two 500 ml tubes and to each 200 µl of 20 mg/ml PK was added. The liquid was incubated for 2 h at 56 °C. This was done to break down the epithelial cells. Following cell lysis, the sample was centrifuged at 5,000 g for 3 min. The supernatant was transfer to another tube and the pellet (sperms) was diluted with 400 µl distilled H₂O. A volume of 5 µl of sperms liquid was added to the swabs to give about 50 – 55 ng DNA in each swab. All the swabs were used for extraction immediately after the application of the materials. Another 28 swabs were used as negative controls at each temperature and humidity range. On each day, starting from day 1 (control),

one of the negative swab was extracted side by side to the positive one. The samples were labelled as follow started from day 1 to day 28 (see Figure 4.6).

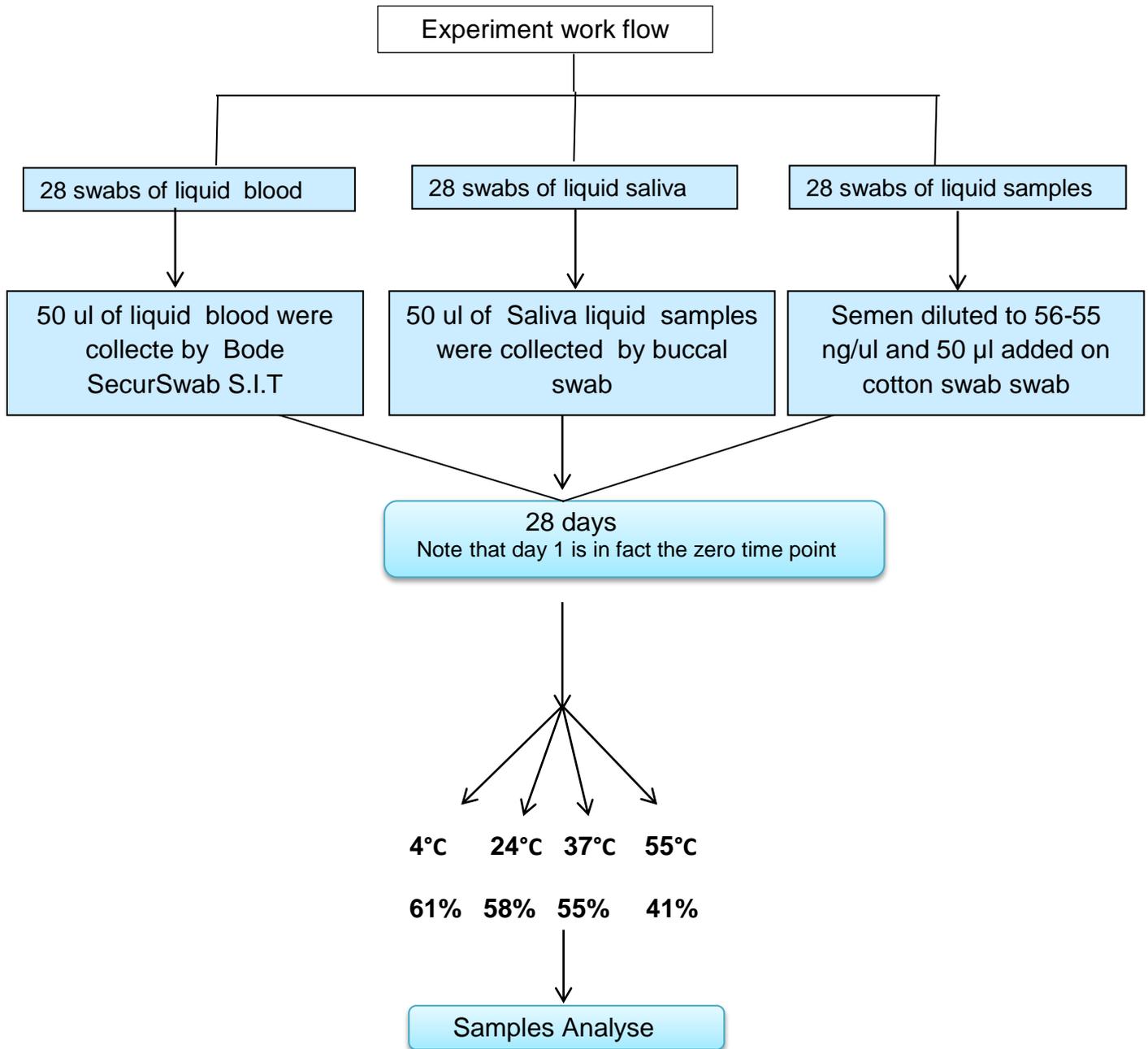


Figure 4.6: Experimental design work flow showing the protocol for evaluating different DNA persistence in different temperature and humidity settings.

Dilution of semen was performed as follow: Each sperm contained 3 ng DNA. The sperm count of the volunteer was about 21 million spermatozoa per ml, with an average of 21,000 spermatozoa/ μl . Based on previous fact it was possible to make dilution and calculate the DNA roughly. A volume of 1 μl pure semen was diluted with 990 μl dH₂O and then the dilution was divided into two 500 ml tubes and to each a volume of 200 μl 20 mg/ml PK was added. The liquid was incubated for 2 h at 56 °C in order to lyse the epithelial cells. The sample was centrifuged at 5,000 g for 3 min. The supernatant was transferred to another tube and the pellet (sperms) was diluted with 400 μl dH₂O. A volume 5 μl of sperms liquid was added to the swabs (appx 50 – 55 ng DNA in each swab).

4.10.2 Temperature and humidity measurements

The swabs were placed in swabs holders and incubated in temperature and humidity incubator as illustrated in Table 4.2;

Table 4.2: Table showing the different ranges of temperature and humidity for the present work study during four different months of the year. The effects of temperature and humidity were monitored on a day by day basis.

| No of samples | Temperature °C | Humidity (Rh) % |
|---------------|----------------|-----------------|
| 28 | 4 | 61 |
| 28 | 24 | 58 |
| 28 | 37 | 55 |
| 28 | 55 | 41 |

4.11 DNA extraction

4.11.1 Blood and saliva

The DNeasy® Blood and Tissue Kit (Qiagen) were used in this study to measure DNA in each sample. The cotton part of the swab was cut by using sterile blade and inserted in sterile 1.5 ml tubes. The blood and saliva stain extractions were done side by side. To the cotton, volumes of 0.5 ml ATL buffer, 100 µl PK (20 mg/ml) and 10 µl of 1 M DTT (Sigma Aldrich) were added. The mixture was placed in a rotary incubator at 55 °C, for 24 h. The supernatant was then removed to another 1,5 ml tube to which 0.5 ml of AL buffer was added. This was mixed gently and incubated at 70 °C for 30 min. A volume of 0.5 ml of absolute ethanol was then added and the solution was mixed gently and then transferred to a DNeasy® mini spin column and centrifuged at 8000 g, for 1 min. The flow through was discarded and the step was repeated until all the extraction mixture had passed through the spin column. The spin column was placed in a new collection tube and 500 µl AW1 buffer was added. After centrifugation for 1 min at 8000 g, the wash step was repeated with 500 µl AW2 buffer. The spin column was then centrifuged for 1 min at 13000 g to remove any residual ethanol from the membrane. DNA was eluted by adding 30 µl AE buffer to the membrane and after incubation for 5 min at room temperature, the spin column was centrifuged at 8000 g for 1 min. The elute was subsequently collected in a clean 1.5 ml tube.

4.11.2 Semen

The QIAamp DNA Investigator Kit (Qiagen) were used in this study following the manufacturer's protocol. The cotton part of the swab was cut by using sterile blade and inserted in sterile 1.5 ml tubes. To the cotton, 20 µl proteinase K and 500 µl Buffer ATL were added to the sample. The tube was mixed by pulse-vortexing for 10 sec, then placed in a thermo mixer incubator at 56°C with shaking at 900 rpm for at least 1 hr. Briefly, the tube was centrifuged to remove drops from the inside of the lid then the solid material was removed to another 1.5 ml tube. The tube was centrifuged for 5 min at 20,000 g at full speed and then the supernatant was transferred to a new tube leaving about 30 µl of the supernatant above the pellet without disturbing the pellet. The pellet was subsequently re-suspended in 500 µl buffer ATL and mixed by pulse-vortexing for 10 s. The tube was then centrifuged for 5 min at 13,000 g at full speed. The supernatant was carefully aspirated and discarded leaving only about 30 µl of the supernatant to prevent any disturbance to the pellet. Thereafter, the washing steps were repeated at least three times with the ATL buffer. Thereafter, 280 µl buffer ATL, 10 µl proteinase K and 10 µl 1 M DTT were added to the pellet. The tube was capped and the contents mixed by pulse-vortexing for 10 s. The tube was then placed in a thermo-mixer and incubated at 56°C with shaking at 900 rpm for at least 1 hour. It was briefly centrifuged to remove drops from the inside of the lid. A volume of 300 µl buffer ATL was added to the tube which was then capped and mixed by pulse-vortexing for 10 sec. Thereafter, the tube was placed in the thermo-mixer or in a heated orbital incubator and incubated at 70°C with shaking at 900 rpm for 10 min. The tube was then centrifuged at full speed (20,000 x g)

for 1 min, and the supernatant was carefully transferred to a new 1.5 ml tube to which a volume of 150 μ l ethanol (96–100%) was added. The tube was then capped and it was mixed by pulse-vortexing for 15 s. Thereafter, it was centrifuged to remove drops from the inside of the lid. Following centrifugation, the entire lysate was carefully transferred from the previous step to the QIAamp MinElute column. The lid of the column was closed and it was centrifuged at 6000 x g (8000 rpm) for 1 min. Thereafter, the QIAamp MinElute column was transferred into a clean 2 ml collection tube and the collection tube containing the flow-through was discarded. The QIAamp MinElute column was carefully opened and a volume of 500 μ l buffer AW1 was added. The column was closed and centrifuged at (6000 x g) for 1 min.

Following centrifugation, the QIAamp MinElute column was placed in a clean 2 ml collection tube and the collection tube containing the flow through was discarded. The QIAamp MinElute column was carefully opened and a volume of 700 μ l buffer AW2 was added. The lid was closed and the column was centrifuged at (6000 x g) for 1 min. Thereafter, the QIAamp MinElute column was placed into a clean 2 ml collection tube and the collection tube containing the flow-through was discarded. The lid of the QIAamp MinElute column was carefully opened and a volume of 700 μ l of ethanol (96–100%) was added. Thereafter, the column was capped and it was centrifuged at (6000 x g) for 1 min. After centrifugation, the QIAamp MinElute column was placed in a clean 2 ml collection tube and the collection tube containing the flow-through was discarded. The tube was centrifuged at full speed (20,000 x g) for 3 min to dry the membrane completely.

Thereafter, the QIAamp MinElute column was in a clean 1.5 ml micro-centrifuge tube and the collection tube containing the flow-through. was discarded. Thereafter, the lid of the QIAamp MinElute column was opened carefully and incubated at room temperature (15–25°C) for 10 min or at 56°C for 3 min. Subsequently, a volume of 20–50 µl buffer ATE was added to the center of the membrane. The lid of the column was closed and it was incubated at room temperature (15–25°C) for 1 min and then centrifuged at full speed (20,000 x g) for 1 min.

4.12 Quantification

Human DNA in the extracts was quantified using the Quantifiler™ Human DNA Quantification kit (Applied Biosystem, Foster City, CA, USA) following the manufacturer's protocol, except that half volume (12.5 µl) reactions were used containing 1 µl of template DNA, 5.25 µl of Quantifiler Human Primer Mix and 6.25 µl Quantifiler PCR Reaction Mix. The reactions were run using a ABI PRISM® 7500 Real-Time PCR system (Applied Biosystems). See figure 5.4 for details.

4.13 Data Analysis

Statistical analyses were performed and analyzed by SPSS version 17 for Windows (SPSS Inc., Chicago, Illinois). The temperature data were in four groups of 4, 24, 37, 55 °C and analysis of variance (ANOVA) was performed, which showed that DNA values decreased with increase in temperature. A value of $p < 0.05$ was taken as significance. The data are presented as tables and also graphically in the results section of this chapter.

4. 14 Results

4.14.1 Effect of 55°C and 41% humidity on DNA degradation in saliva samples

Table 4.3: Table showing DNA quantification following extraction of human saliva,

| Days | Samples | Temp °C- RH % | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | Mean | SD |
|------|---------|---------------|---------------------|---------------------|---------------------|-------|------|
| 1 | Saliva | - | 31.09 | 31.12 | 31.1 | 31.10 | 0.02 |
| 2 | Saliva | 55 , 41 | 25.01 | 25 | 25.03 | 25.01 | 0.02 |
| 3 | Saliva | 55 , 41 | 19.12 | 19.14 | 19.1 | 19.12 | 0.02 |
| 4 | Saliva | 55 , 41 | 14.13 | 14.13 | 14.12 | 14.13 | 0.01 |
| 5 | Saliva | 55 , 41 | 8.19 | 0 | 8.23 | 5.47 | 4.74 |
| 6 | Saliva | 55 , 41 | 1.1 | 1.11 | 1.1 | 1.10 | 0.01 |
| 7 | Saliva | 55 , 41 | 1.09 | 1.1 | 1.12 | 1.10 | 0.02 |
| 8 | Saliva | 55 , 41 | 0.9 | 0.91 | 0.9 | 0.90 | 0.01 |
| 9 | Saliva | 55 , 41 | 0.81 | 0.76 | 0.78 | 0.78 | 0.03 |
| 10 | Saliva | 55 , 41 | 0.61 | 0.5 | 0.55 | 0.55 | 0.06 |
| 11 | Saliva | 55 , 41 | 0.42 | 0.39 | 0.39 | 0.40 | 0.02 |
| 12 | Saliva | 55 , 41 | 0.29 | 0.25 | 0.25 | 0.26 | 0.02 |
| 13 | Saliva | 55 , 41 | 0.16 | 0.15 | 0.15 | 0.15 | 0.01 |
| 14 | Saliva | 55 , 41 | 0.09 | 0.1 | 0.11 | 0.10 | 0.01 |
| 15 | Saliva | 55 , 41 | 0.05 | 0.04 | 0.04 | 0.04 | 0.01 |
| 16 | Saliva | 55 , 41 | 0.01 | 0 | 0.01 | 0.01 | 0.01 |
| 17 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 18 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 19 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 20 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 21 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 22 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 23 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 24 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 25 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 26 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 27 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 28 | Saliva | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |

incubated at temperature of 55 °C and humidity 41% over a duration of 28 days.

Table 4.3 shows that each sample was analyzed in triplicate at 55 °C. Note that day 1 is the zero time point (the sample with no treatment). The results were obtained from real-time PCR and they showed that the DNA in saliva samples started to degrade in quantity after the second day until reaching zero level at day 17. The results also show significant ($p < 0.05$) decreases in DNA level from day 2 onwards.

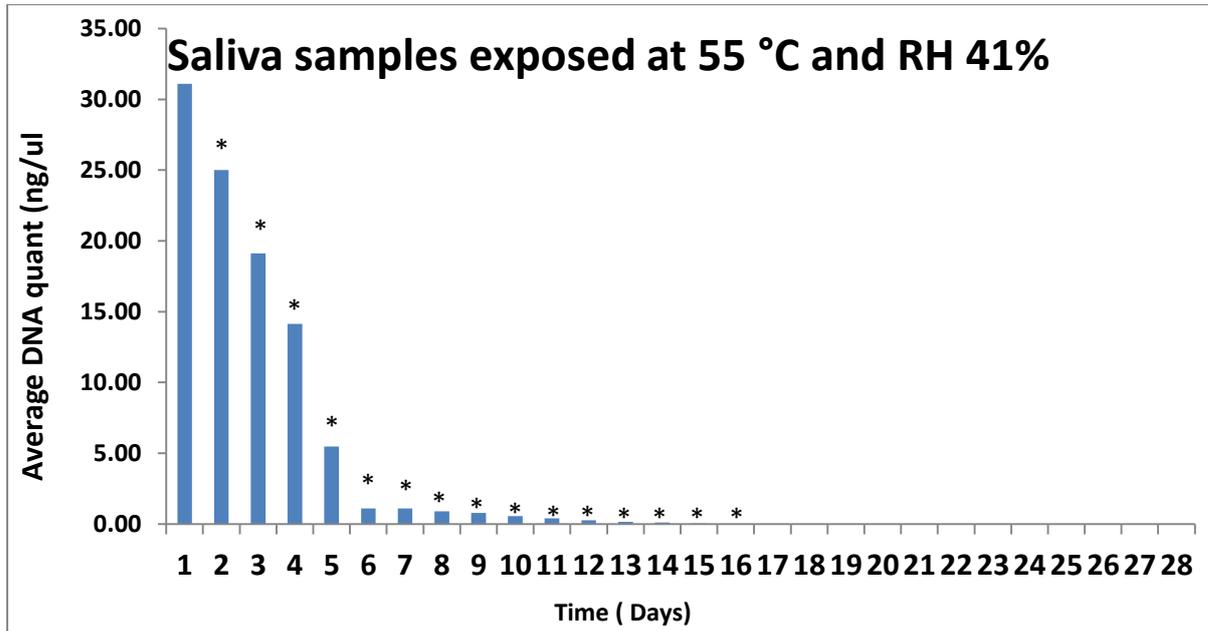


Figure 4.7: Bar charts showing the time-course average of DNA recovered from triplicate DNA quantification of 50 μ l saliva samples stained on buccal swab. The results were obtained from real-time PCR at 55 °C and at a humidity of 41%. Note that day 1 is the zero time point (the sample with no treatment). All data are mean \pm -SD; $p < 0.05$.

The results in figure 4.7 show that high temperature had a profound and significant ($p < 0.05$) effect on DNA degradation which started within one day of exposure at 55 °C and at a humidity of 41%. The DNA continued to degrade significantly ($p < 0.05$) at days 2-16 compared to day 1. These results clearly show that DNA was completely degraded in saliva samples at 55 °C and at a humidity of 41%.

4.14.2 DNA quantification of human blood samples following extraction at 55 °C and at a humidity of 41%

| Day | Sample | Temp °C and RH % | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | Mean | SD |
|-----|--------|------------------|---------------------|---------------------|---------------------|-------|------|
| 1 | Blood | - | 45.2 | 45.22 | 44.22 | 44.88 | 0.57 |
| 2 | Blood | 55 , 41 | 44.22 | 44.21 | 44.2 | 44.21 | 0.01 |
| 3 | Blood | 55 , 41 | 37.7 | 37 | 37.4 | 37.37 | 0.35 |
| 4 | Blood | 55 , 41 | 31.1 | 31.13 | 31.01 | 31.08 | 0.06 |
| 5 | Blood | 55 , 41 | 28.2 | 28 | 27.9 | 28.03 | 0.15 |
| 6 | Blood | 55 , 41 | 24.55 | 24.57 | 24.34 | 24.49 | 0.13 |
| 7 | Blood | 55 , 41 | 18.88 | 18.38 | 17.9 | 18.39 | 0.49 |
| 8 | Blood | 55 , 41 | 12.23 | 11.95 | 12 | 12.06 | 0.15 |
| 9 | Blood | 55 , 41 | 7.65 | 7.6 | 6.9 | 7.38 | 0.42 |
| 10 | Blood | 55 , 41 | 3.44 | 3.34 | 3.01 | 3.26 | 0.23 |
| 11 | Blood | 55 , 41 | 1.01 | 1 | 0.98 | 1.00 | 0.02 |
| 12 | Blood | 55 , 41 | 0.5 | 0.1 | 0.13 | 0.24 | 0.22 |
| 13 | Blood | 55 , 41 | 0.03 | 0.03 | 0.02 | 0.03 | 0.01 |
| 14 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 15 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 16 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 17 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 18 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 19 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 20 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 21 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 22 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 23 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 24 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 25 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 26 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 27 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |
| 28 | Blood | 55 , 41 | 0 | 0 | 0 | 0.00 | 0.00 |

Table 4.4: Table showing the time course of DNA quantification following extraction of 50 μl of human blood, incubated at temperature of 55 °C and RH of 41% over a period of 28 days. Table contains original and mean data with SD;n=3.

Table 4.4 shows the time course of DNA quantification at temperature of 55 °C and RH of 41% over a period of 28 days. Note that day 1 is in fact the zero time point (the sample with no treatment). The results showed that at 55 °C and at a RH of 41%, the DNA in blood samples start to degrade significantly ($p < 0.05$) in quantity after the third day compared to day 1 until reaching zero at day 14.

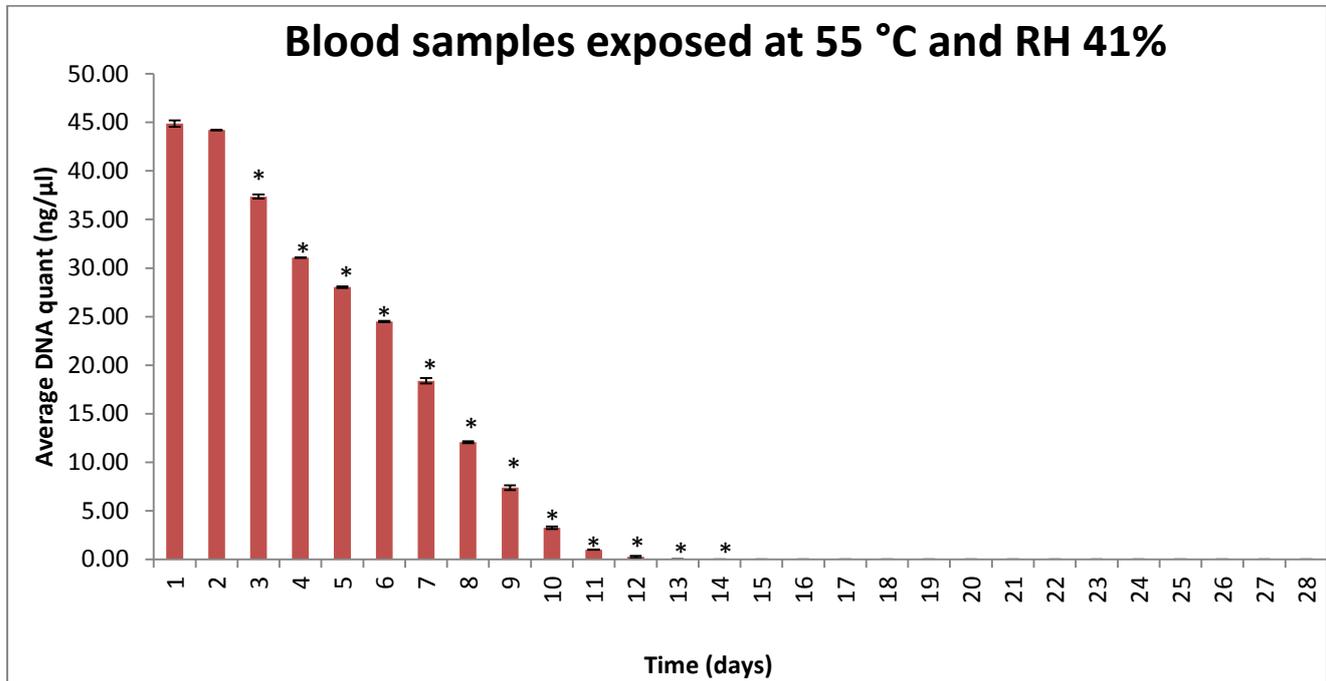


Figure 4.8: Bar charts showing the time-course average of DNA recovered from triplicate DNA quantification of 50 μ l blood samples stained on buccal swab. The result obtained from real-time PCR showed the data at 55 $^{\circ}$ C and at a humidity of 41%. Data are mean \pm SD, n=3. * $p < 0.05$ for day compared to the other days. Note that all the DNA was completely degraded by day 14. Note that day 1 is in fact the zero time point (the sample with no treatment).

The results presented in figure 4.8 show that DNA in blood samples started to degrade gradually at day 3 and onwards until it was completely degraded at day 12. These results are particularly interesting since they show that high temperature at 41% humidity had profound and significant ($p < 0.05$) effect on DNA degradation. The results also indicate that particular caution must be taken when samples are collected for forensic DNA examination.

4.14.3 DNA quantification of human semen following extraction at 55 $^{\circ}$ C and at a humidity of 41%

| Day | sample | Temp°C And RH | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | Mean | st.dev |
|-----|--------|---------------------|------------------------|------------------------|---------------------------|-------|--------|
| 1 | Semen | - | 55.01 | 55.36 | 55.47 | 55.28 | 0.24 |
| 2 | Semen | 55 , 41 | 56.1 | 54.68 | 55.24 | 55.34 | 0.72 |
| 3 | Semen | 55 , 41 | 55.12 | 55.64 | 55.57 | 55.44 | 0.28 |
| 4 | Semen | 55 , 41 | 55.46 | 54.58 | 55.04 | 55.03 | 0.44 |
| 5 | Semen | 55 , 41 | 55.02 | 54.37 | 54.57 | 54.65 | 0.33 |
| 6 | Semen | 55 , 41 | 55 | 55.07 | 55.38 | 55.15 | 0.20 |
| 7 | Semen | 55 , 41 | 54.11 | 55.04 | 54.75 | 54.63 | 0.48 |
| 8 | Semen | 55 , 41 | 55.25 | 54.02 | 55.28 | 54.85 | 0.72 |
| 9 | Semen | 55 , 41 | 53.44 | 54.93 | 54.58 | 54.32 | 0.78 |
| 10 | Semen | 55 , 41 | 56.01 | 53.55 | 54.19 | 54.58 | 1.28 |
| 11 | Semen | 55 , 41 | 53.76 | 55.27 | 55 | 54.68 | 0.81 |
| 12 | Semen | 55 , 41 | 55.33 | 55.01 | 54.57 | 54.97 | 0.38 |
| 13 | Semen | 55 , 41 | 56.34 | 55.12 | 55.58 | 55.68 | 0.62 |
| 14 | Semen | 55 , 41 | 55 | 0 | 53.58 | 36.19 | 31.35 |
| 15 | Semen | 55 , 41 | 54.89 | 54.11 | 56.59 | 55.20 | 1.27 |
| 16 | Semen | 55 , 41 | 55.36 | 53.44 | 54.1 | 54.30 | 0.98 |
| 17 | Semen | 55 , 41 | 56 | 53.76 | 55.21 | 54.99 | 1.14 |
| 18 | Semen | 55 , 41 | 54.68 | 56.34 | 53.4 | 54.81 | 1.47 |
| 19 | Semen | 55 , 41 | 54.78 | 55.36 | 56.27 | 55.47 | 0.75 |
| 20 | Semen | 55 , 41 | 55.64 | 56.47 | 53.71 | 55.27 | 1.42 |
| 21 | Semen | 55 , 41 | 55.27 | 55.27 | 54.62 | 55.05 | 0.38 |
| 22 | Semen | 55 , 41 | 54.58 | 55.57 | 54.72 | 54.96 | 0.54 |
| 23 | Semen | 55 , 41 | 53.55 | 55.47 | 55.63 | 54.88 | 1.16 |
| 24 | Semen | 55 , 41 | 54.37 | 55.02 | 55.27 | 54.89 | 0.46 |
| 25 | Semen | 55 , 41 | 54.93 | 54.14 | 54.52 | 54.53 | 0.40 |
| 26 | Semen | 55 , 41 | 55.07 | 55.27 | 55.01 | 55.12 | 0.14 |
| 27 | Semen | 55 , 41 | 54.02 | 53.43 | 55.11 | 54.19 | 0.85 |
| 28 | Semen | 55 , 41 | 55.04 | 56.76 | 54.1 | 55.30 | 1.35 |

Table 4.5: Table showing DNA quantification following extraction of 50 μl of human semen, after dilution and stained, incubated at temperature of 55 °C over a duration of 28 days. Data are original and mean values with SD;n=3.

Table 4.5 shows the DNA quantification at 55°C and at a RH of 41%. The extractions were done using the QIAamp DNA Investigator Kit (Qiagen). Note that at day 14 there was human error. Note that day 1 is the zero time point (the sample with no treatment) and the data show no significant ($p>0.05$) degradation of DNA quantity in semen samples was obtained comparing levels at day 1 with day 28 that at 55 °C and at a RH of 41%.

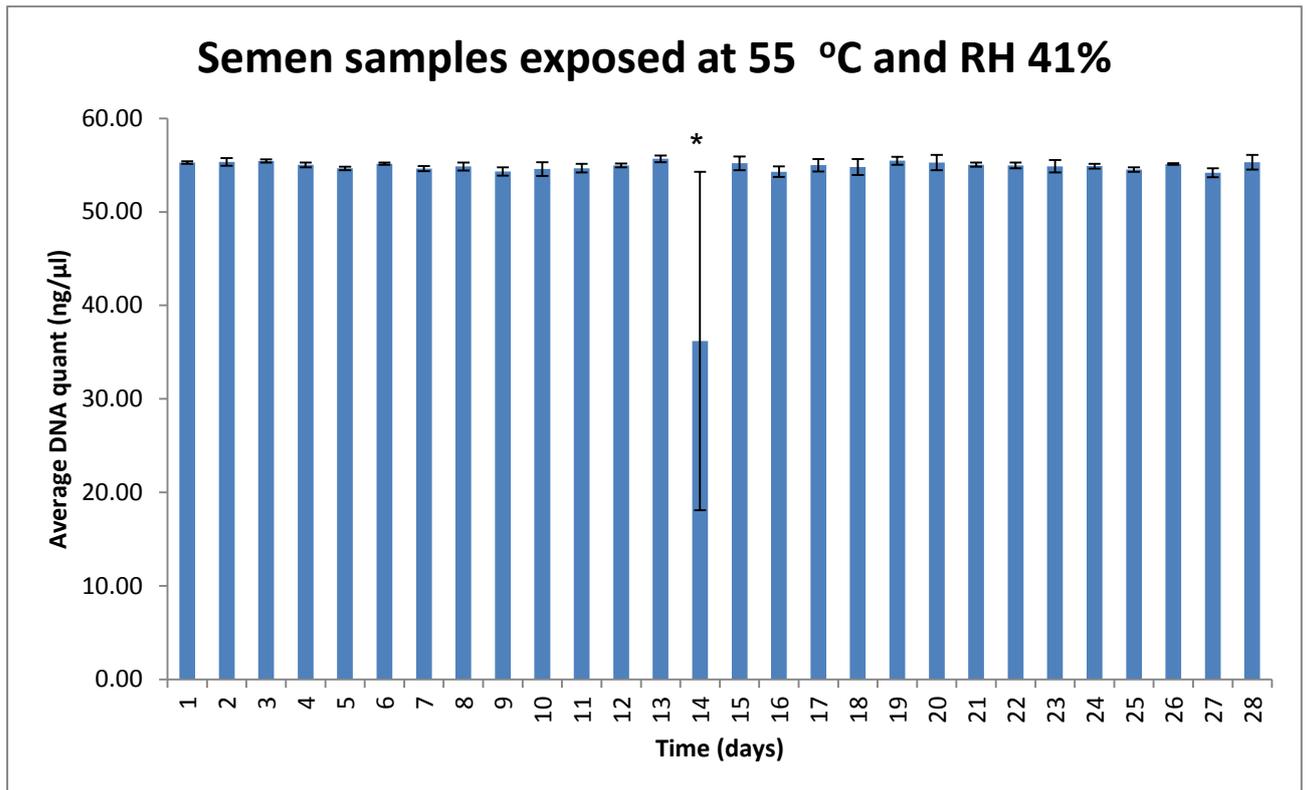


Figure 4.9: Bar charts showing the time-course average of DNA recovered from triplicate DNA quantification of 50 μ l semen after dilution. The results obtained from real-time PCR showed at 55 $^{\circ}$ C and at a humidity of 41%. The extractions were done using the QIAamp DNA Investigator protocol. Data are mean \pm SD, n=3. * Note that $p > 0.05$ for day 1 compared to all other days. Note that at day 14 DNA value was very little in one test due to human error. Note also that day 1 is the zero time point (the sample with no treatment).

The results in figure 4.9 show that at 55 $^{\circ}$ C and at a RH of 41%, the DNA quantity in semen samples were more or less similar between values obtained to day 1 compared to day 28. The results show clearly that DNA in semen is more resistant to degradation following environmental insults suggesting that the sperms have special protection mechanism in preserving DNA content.

4.14.4 DNA quantification in human saliva samples following extraction at 37 $^{\circ}$ C and at a humidity of 55%

| Days | Samples | Temp °C and RH 55% | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | Mean | SD |
|------|---------|--------------------|---------------------|---------------------|---------------------|-------|-------|
| 1 | Saliva | - | 45.01 | 45.06 | 45.02 | 45.03 | 0.03 |
| 2 | Saliva | 37, 55 | 45.45 | 45.34 | 45.45 | 45.41 | 0.06 |
| 3 | Saliva | 37, 55 | 45.34 | 45.3 | 45.29 | 45.31 | 0.03 |
| 4 | Saliva | 37, 55 | 45.55 | 45.34 | 45.46 | 45.45 | 0.11 |
| 5 | Saliva | 37, 55 | 45.27 | 45.45 | 45.34 | 45.35 | 0.09 |
| 6 | Saliva | 37, 55 | 44.02 | 45.66 | 45.57 | 45.08 | 0.92 |
| 7 | Saliva | 37, 55 | 44.87 | 44.67 | 44.37 | 44.64 | 0.25 |
| 8 | Saliva | 37, 55 | 44.67 | 44.45 | 44.56 | 44.56 | 0.11 |
| 9 | Saliva | 37, 55 | 44.07 | 44.12 | 44.11 | 44.10 | 0.03 |
| 10 | Saliva | 37, 55 | 43.92 | 44.66 | 45.36 | 44.65 | 0.72 |
| 11 | Saliva | 37, 55 | 44.27 | 44.59 | 44.78 | 44.55 | 0.26 |
| 12 | Saliva | 37, 55 | 43.87 | 43.68 | 43.18 | 43.58 | 0.36 |
| 13 | Saliva | 37, 55 | 43.78 | 43.81 | 43.77 | 43.79 | 0.02 |
| 14 | Saliva | 37, 55 | 43.12 | 43.32 | 43.66 | 43.37 | 0.27 |
| 15 | Saliva | 37, 55 | 10.22 | 43.32 | 43.01 | 32.18 | 19.02 |
| 16 | Saliva | 37, 55 | 40.22 | 41.22 | 41.35 | 40.93 | 0.62 |
| 17 | Saliva | 37, 55 | 40.01 | 40.38 | 40.25 | 40.21 | 0.19 |
| 18 | Saliva | 37, 55 | 40.01 | 39.69 | 39 | 39.57 | 0.52 |
| 19 | Saliva | 37, 55 | 39.72 | 39.28 | 39.29 | 39.43 | 0.25 |
| 20 | Saliva | 37, 55 | 39.01 | 39.11. | 39.38 | 39.20 | 0.26 |
| 21 | Saliva | 37, 55 | 38.56 | 38.22 | 38.23 | 38.34 | 0.19 |
| 22 | Saliva | 37, 55 | 38.02 | 0 | 20.22 | 19.41 | 19.02 |
| 23 | Saliva | 37, 55 | 37.88 | 37.86 | 37.01 | 37.58 | 0.50 |
| 24 | Saliva | 37, 55 | 37.4 | 37.2 | 37.1 | 37.23 | 0.15 |
| 25 | Saliva | 37, 55 | 36.67 | 36.46 | 36.67 | 36.60 | 0.12 |
| 26 | Saliva | 37, 55 | 36.01 | 36.22 | 36.67 | 36.30 | 0.34 |
| 27 | Saliva | 37, 55 | 35.92 | 35.2 | 34.99 | 35.37 | 0.49 |
| 28 | Saliva | 37, 55 | 34.79 | 34.68 | 34.67 | 34.71 | 0.07 |

Table 4.6: Table showing time course of DNA quantification following extraction of 50 μl of human saliva and incubated at temperature of 37 °C and at a RH of 55% over a duration of 28 days. Data are original and mean values with SD;n=3.

Table 4.6 shows the time course of DNA at 37 °C and at a RH of 55% over a duration of 28 days. Note that day 1 is the zero time point (the sample with no treatment). The results showed that the DNA in saliva degraded slowly starting from day 12 to day 28 but these values were not significantly different from day 1. The results also show a human error on day 22 in which DNA quantity was zero.

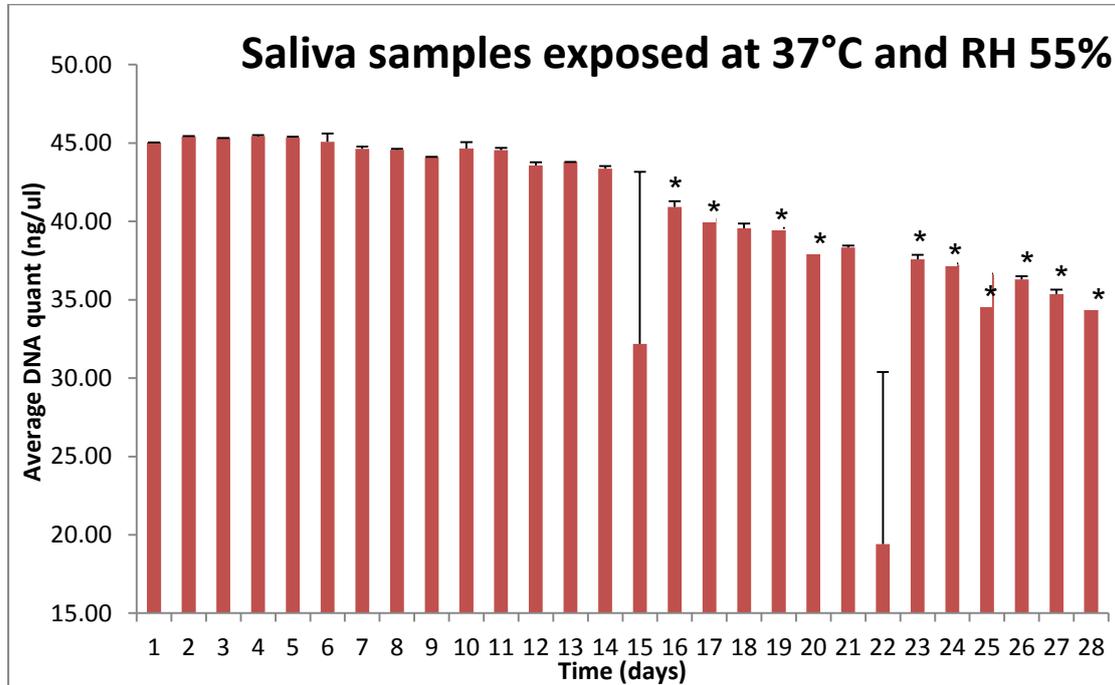


Figure 4.10: Bar charts showing the mean (\pm SD) time-course of DNA quantification following extraction of 50 μ l of human saliva material at temperature of 37 °C and at a RH of 55%. Note that day 1 is the zero time point (the sample with no treatment). * $P < 0.05$ for days 1 compared to day 16 and onwards. Note that samples for day 15 and 22 showed standard deviation error related to the differences on DNA quantity due to human error. (Data taken from table 5.6).

The results presented in figure 4.10 show that the DNA in saliva degraded slowly and gradually starting from day 12 to day 28. DNA quantity during all the days were almost close to each other and at the same time DNA quantity from day 23 to day 28 seem different from day 1 ($p < 0.05$).

4.14.5 DNA quantification from human blood samples following extraction at 37 °C and at a RH of 55%.

| Days | Samples | Temp °C- RH % | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | Mean | SD |
|------|---------|------------------|------------------------|------------------------|------------------------|-------|-------|
| 1 | Blood | - | 46.2 | 45.9 | 46.32 | 46.14 | 0.22 |
| 2 | Blood | 37, 55 | 45.8 | 45.7 | 45.34 | 45.61 | 0.24 |
| 3 | blood | 37, 55 | 45.67 | 45.87 | 45.69 | 45.74 | 0.11 |
| 4 | blood | 37, 55 | 45.34 | 45.6 | 45.2 | 45.38 | 0.20 |
| 5 | blood | 37, 55 | 45.7 | 45.23 | 45.21 | 45.38 | 0.28 |
| 6 | blood | 37, 55 | 45.02 | 45.1 | 45.02 | 45.05 | 0.05 |
| 7 | blood | 37, 55 | 45.01 | 45.01 | 44.9 | 44.97 | 0.06 |
| 8 | blood | 37, 55 | 44.89 | 44.78 | 44.9 | 44.86 | 0.07 |
| 9 | blood | 37, 55 | 44.36 | 44.69 | 44.37 | 44.47 | 0.19 |
| 10 | blood | 37, 55 | 44.78 | 44.16 | 44.01 | 44.32 | 0.41 |
| 11 | blood | 37, 55 | 44.82 | 44.53 | 44.24 | 44.53 | 0.29 |
| 12 | blood | 37, 55 | 44.58 | 44.89 | 44.24 | 44.57 | 0.33 |
| 13 | blood | 37, 55 | 44.03 | 44.05 | 44.17 | 44.08 | 0.08 |
| 14 | blood | 37, 55 | 43.79 | 43.89 | 43.47 | 43.72 | 0.22 |
| 15 | blood | 37, 55 | 43.46 | 43.67 | 43.17 | 43.43 | 0.25 |
| 16 | blood | 37, 55 | 43.45 | 43.21 | 43.01 | 43.22 | 0.22 |
| 17 | blood | 37, 55 | 43 | 43.12 | 43.03 | 43.05 | 0.06 |
| 18 | blood | 37, 55 | 42.22 | 42.79 | 43.78 | 42.93 | 0.79 |
| 19 | blood | 37, 55 | 41 | 40.45 | 41.7 | 41.05 | 0.63 |
| 20 | blood | 37, 55 | 41.2 | 41 | 41.03 | 41.08 | 0.11 |
| 21 | blood | 37, 55 | 40.89 | 40.78 | 40.19 | 40.62 | 0.38 |
| 22 | blood | 37, 55 | 40.01 | 39.79 | 39.69 | 39.83 | 0.16 |
| 23 | blood | 37, 55 | 39.21 | 39.1 | 39.71 | 39.34 | 0.33 |
| 24 | blood | 37, 55 | 38.33 | 38.01 | 37.92 | 38.09 | 0.22 |
| 25 | blood | 37, 55 | 8.1 | 0 | 37.8 | 15.30 | 19.90 |
| 26 | blood | 37, 55 | 37.01 | 37.23 | 37.1 | 37.11 | 0.11 |
| 27 | blood | 37, 55 | 36.89 | 36.88 | 36.89 | 36.89 | 0.01 |
| 28 | blood | 37, 55 | 36.13 | 36.02 | 36 | 36.05 | 0.07 |

Table 4.7: Table showing time course change of DNA quantification following extraction of 50 μl of human blood samples, incubated at temperature of 37 °C and at a RH of 55% over a duration of 28 days. Data are original and mean values with SD.

Table 4.7 shows the time course change of DNA quantification at 37 °C and at a RH of 55% over a duration of 28 days. Note that day 1 is in fact the zero time point (the sample with no treatment). The results showed that the DNA in blood samples slowly degraded starting from day 14 but this value was only significant from day 18 and onwards compared to day 1. *P<0.05. The results also show a human error on day 25 in which DNA quantity was zero.

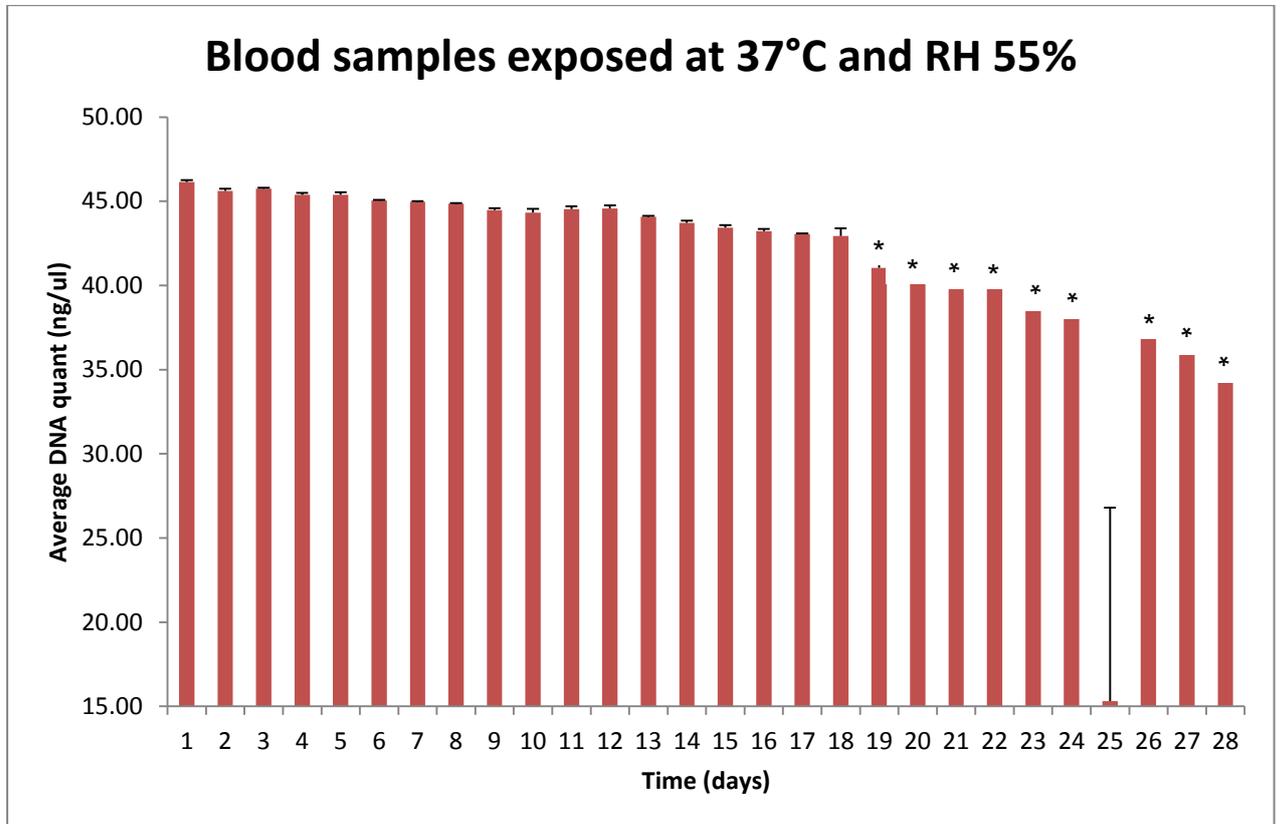


Figure 4.11: Bar charts showing the mean (+SD) time course of DNA quantification following extraction of 50 μ l of human blood stain material incubated at temperature of 37 °C and RH 55% over a duration of 28 days. Note that at day 25 the values are very low probably due to human error and day 1 is the zero time point (the sample with no treatment). The results showed that at 37 °C and at a RH of 55%, the DNA in blood degraded slowly starting from day 14 to day 28 but these values were not significantly different from day 1. However, the data show significant differences comparing days 24, 26, 27 and 28 with day 1. (* $p < 0.05$).

4.14.6 DNA quantification following extraction of human semen samples at a temperature of 37 °C and at a humidity of 55%.

| Days | Samples | Temp °C And RH % | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | mean | st. dev |
|------|---------|---------------------|------------------------|------------------------|------------------------|-------|---------|
| 1 | Semen | - | 55.01 | 56.1 | 55.36 | 55.49 | 0.56 |
| 2 | Semen | 37 , 55 | 55.12 | 55.46 | 55.64 | 55.41 | 0.26 |
| 3 | Semen | 37 , 55 | 55.02 | 55 | 54.37 | 54.80 | 0.37 |
| 4 | Semen | 37 , 55 | 54.11 | 55.25 | 55.04 | 54.80 | 0.61 |
| 5 | Semen | 37 , 55 | 53.44 | 56.01 | 54.93 | 54.79 | 1.29 |
| 6 | Semen | 37 , 55 | 53.76 | 55.33 | 55.27 | 54.79 | 0.89 |
| 7 | Semen | 37 , 55 | 56.34 | 55 | 55.12 | 55.49 | 0.74 |
| 8 | Semen | 37 , 55 | 54.89 | 55.36 | 54.11 | 54.79 | 0.63 |
| 9 | Semen | 37 , 55 | 56 | 54.68 | 53.76 | 54.81 | 1.13 |
| 10 | Semen | 37 , 55 | 54.78 | 55.64 | 55.36 | 55.26 | 0.44 |
| 11 | Semen | 37 , 55 | 40.27 | 54.58 | 55.27 | 50.04 | 8.47 |
| 12 | Semen | 37 , 55 | 53.55 | 54.37 | 55.47 | 54.46 | 0.96 |
| 13 | Semen | 37 , 55 | 54.93 | 55.07 | 54.14 | 54.71 | 0.50 |
| 14 | Semen | 37 , 55 | 54.02 | 55.31 | 53.43 | 54.25 | 0.96 |
| 15 | Semen | 37 , 55 | 55.1 | 55.6 | 56.17 | 55.62 | 0.54 |
| 16 | Semen | 37 , 55 | 53.48 | 54.39 | 55.92 | 54.60 | 1.23 |
| 17 | Semen | 37 , 55 | 55.11 | 55.34 | 54.68 | 55.04 | 0.34 |
| 18 | Semen | 37 , 55 | 54.68 | 54.9 | 55.11 | 54.90 | 0.22 |
| 19 | Semen | 37 , 55 | 55.17 | 55.23 | 54.57 | 54.99 | 0.36 |
| 20 | Semen | 37 , 55 | 54.37 | 55.13 | 55.07 | 54.86 | 0.42 |
| 21 | Semen | 37 , 55 | 55.27 | 54.14 | 55.27 | 54.89 | 0.65 |
| 22 | Semen | 37 , 55 | 56.11 | 53.25 | 56.34 | 55.23 | 1.72 |
| 23 | Semen | 37 , 55 | 55.22 | 55.13 | 55.57 | 55.31 | 0.23 |
| 24 | Semen | 37 , 55 | 55.44 | 55.19 | 30.27 | 46.97 | 14.46 |
| 25 | Semen | 37 , 55 | 54.68 | 55.68 | 53.44 | 54.60 | 1.12 |
| 26 | Semen | 37 , 55 | 55.6 | 54.28 | 54.68 | 54.85 | 0.68 |
| 27 | Semen | 37 , 55 | 55.23 | 53.68 | 55.64 | 54.85 | 1.03 |
| 28 | Semen | 37 , 55 | 55.36 | 54.68 | 55.31 | 55.12 | 0.38 |

Table 4.8: Table showing DNA quantification following extraction of 50 μl of human semen, after dilution and incubated at temperature of 37 °C over a duration of 28 days. Data are original and mean values with SD; n=3.

Table 4.8 shows DNA quantification at 37 °C over a duration of 28 days. Each sample was analyzed in triplicate at 37 °C and at a RH of 55%. Note that day 1 is in fact the zero time point (the sample with no treatment). The results showed no significant ($p>0.05$) degradation of DNA quantity in semen samples was obtained, comparing levels in day 1 with all the other days including day 28 t at 37 °C and at a RH of 55%.

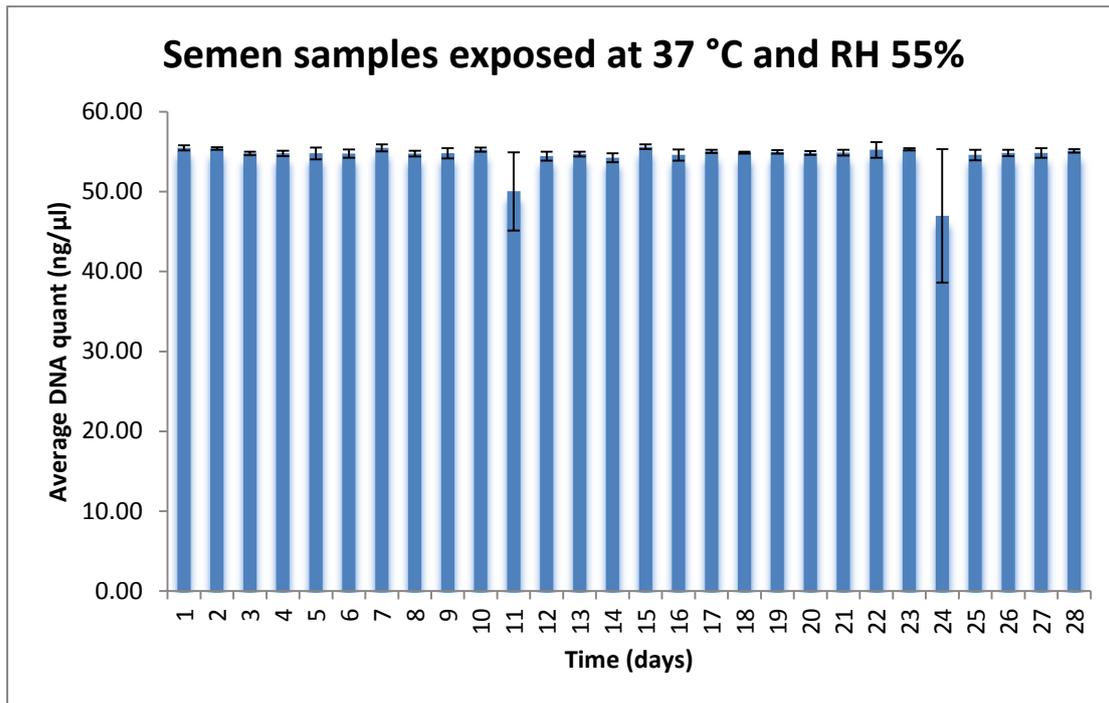


Figure 4.12: Bar charts showing the time course of DNA quantification following extraction of 50 μl of human semen stain samples incubated at temperature of 37 °C and RH 55% over a duration of 28 days. Data are mean ± SD, n=3. ($p > 0.05$ for day 1 compared to all the other days. Note that at day 11 and 24 DNA value was reduced in one test due to human error (Data taken from table 4.8).

The results in table 4.12 showed that at 37 °C and RH 55%, the DNA quantity in semen samples were more or less similar but these values were not significantly ($p > 0.05$) different from day 1.

4.14.7 DNA quantification following extraction of human saliva samples at temperature of 24 °C and at a humidity of 58%.

| Days | samples | Temp °C – RH 58% | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | Mean | SD |
|------|---------|------------------|---------------------|---------------------|---------------------|-------|-------|
| 1 | Saliva | - | 32.70 | 32.72 | 32.69 | 32.70 | 0.02 |
| 2 | Saliva | 24 , 58 | 32.80 | 32.81 | 32.81 | 32.81 | 0.01 |
| 3 | Saliva | 24 , 58 | 32.63 | 32.59 | 32.62 | 32.61 | 0.02 |
| 4 | Saliva | 24 , 58 | 32.67 | 32.70 | 32.64 | 32.67 | 0.03 |
| 5 | Saliva | 24 , 58 | 32.76 | 32.73 | 32.73 | 32.74 | 0.02 |
| 6 | saliva | 24 , 58 | 32.41 | 32.30 | 32.39 | 32.37 | 0.06 |
| 7 | saliva | 24 , 58 | 32.51 | 32.44 | 32.51 | 32.49 | 0.04 |
| 8 | saliva | 24 , 58 | 32.50 | 32.54 | 32.50 | 32.51 | 0.02 |
| 9 | saliva | 24 , 58 | 32.40 | 32.56 | 32.49 | 32.48 | 0.08 |
| 10 | saliva | 24 , 58 | 32.39 | 32.41 | 32.37 | 32.39 | 0.02 |
| 11 | saliva | 24 , 58 | 32.45 | 32.42 | 32.42 | 32.43 | 0.02 |
| 12 | saliva | 24 , 58 | 32.55 | 32.49 | 32.49 | 32.51 | 0.03 |
| 13 | saliva | 24 , 58 | 32.26 | 32.20 | 32.30 | 32.25 | 0.05 |
| 14 | saliva | 24 , 58 | 32.30 | 32.36 | 32.31 | 32.32 | 0.03 |
| 15 | saliva | 24 , 58 | 32.28 | 32.27 | 32.30 | 32.28 | 0.02 |
| 16 | saliva | 24 , 58 | 32.11 | 32.10 | 32.12 | 32.11 | 0.01 |
| 17 | saliva | 24 , 58 | 32.16 | 32.14 | 32.10 | 32.13 | 0.03 |
| 18 | saliva | 24 , 58 | 32.10 | 32.11 | 32.13 | 32.11 | 0.02 |
| 19 | saliva | 24 , 58 | 32.09 | 32.08 | 32.10 | 32.09 | 0.01 |
| 20 | saliva | 24 , 58 | 32.11 | 32.10 | 32.10 | 32.10 | 0.01 |
| 21 | saliva | 24 , 58 | 31.90 | 31.91 | 31.80 | 31.87 | 0.06 |
| 22 | saliva | 24 , 58 | 31.70 | 31.71 | 0 | 21.14 | 18.30 |
| 23 | saliva | 24 , 58 | 31.68 | 31.69 | 31.70 | 31.69 | 0.01 |
| 24 | saliva | 24 , 58 | 31.50 | 31.51 | 31.48 | 31.50 | 0.02 |
| 25 | saliva | 24 , 58 | 31.66 | 31.67 | 31.63 | 31.65 | 0.02 |
| 26 | saliva | 24 , 58 | 31.40 | 31.41 | 31.42 | 31.41 | 0.01 |
| 27 | saliva | 24 , 58 | 31.29 | 31.27 | 31.26 | 31.27 | 0.02 |
| 28 | saliva | 24 , 58 | 31.30 | 31.41 | 31.32 | 31.34 | 0.06 |

Table 4.9: Table showing the time course of DNA quantification incubated at temperature of 24°C and at a RH of 58 % over a duration of 28 days. Data are original and mean values.

Table 4.9 shows the time course of DNA at 24°C and at a RH of 58 % over a duration of 28 days. Note that at day 22, DNA quantity was zero and this was related to human error and day 1 is in fact the zero time point i.e. the sample with no treatment. The results showed no significant ($p > 0.05$) degradation of DNA quantity in saliva samples at temperature of 24°C and at a RH of 58%, comparing levels in day 1 with day 28.

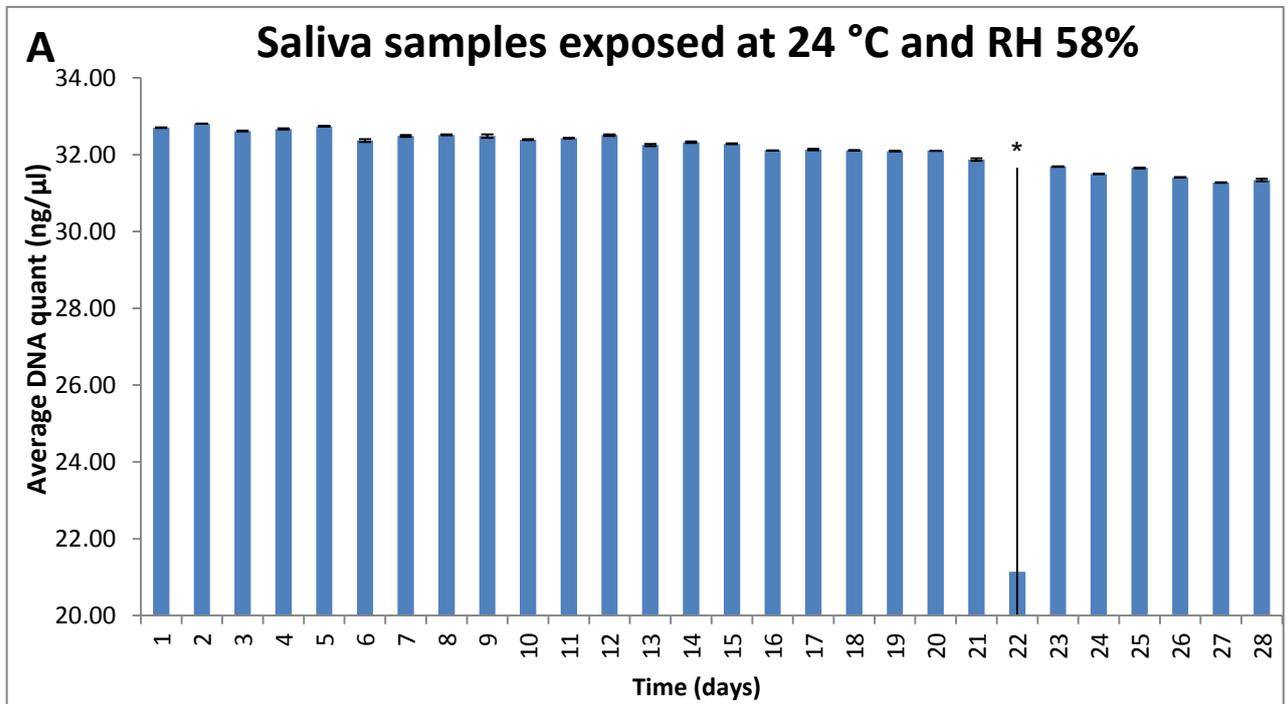


Figure 4.13: Bar charts showing the time course of DNA quantification following extraction of 50 μl of human saliva stain material incubated at temperature of 24 °C and at a RH of 58% over a duration of 28 days. Data are mean \pm SD, $n=3$. $P > 0.05$ for day 1 compared to all the other days. Note that at day 22 DNA value was very little in one test due to human error. (Data taken from table 4.9).

The results in figure 4.13 show that at 24°C and RH 58%, the DNA quantity in saliva samples were more or less similar but these values were not significantly ($p > 0.05$) different from day 1.

4.14.8 DNA quantification from human blood samples following extraction at 24 °C and at a humidity of 61%

| Days | Samples | Temp °C and RH% | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | Mean | SD |
|------|---------|-----------------|---------------------|---------------------|---------------------|-------|-------|
| 1 | Blood | - | 46.3 | 45.9 | 46.34 | 46.18 | 0.24 |
| 2 | Blood | 24 , 58 | 45.9 | 45.01 | 45 | 45.30 | 0.52 |
| 3 | Blood | 24 , 58 | 45.01 | 45.5 | 45.07 | 45.19 | 0.27 |
| 4 | Blood | 24 , 58 | 45.28 | 45.22 | 45.27 | 45.26 | 0.03 |
| 5 | Blood | 24 , 58 | 45.46 | 45.41 | 45.44 | 45.44 | 0.03 |
| 6 | Blood | 24 , 58 | 45 | 45.3 | 45.5 | 45.27 | 0.25 |
| 7 | Blood | 24 , 58 | 44.06 | 44.1 | 44.09 | 44.08 | 0.02 |
| 8 | Blood | 24 , 58 | 44.2 | 44.34 | 43.9 | 44.15 | 0.22 |
| 9 | Blood | 24 , 58 | 44.03 | 44.4 | 44.13 | 44.19 | 0.19 |
| 10 | Blood | 24 , 58 | 44.2 | 44.24 | 44.66 | 44.37 | 0.25 |
| 11 | Blood | 24 , 58 | 44.1 | 0 | 44.12 | 29.41 | 25.47 |
| 12 | Blood | 24 , 58 | 44.01 | 44.71 | 44.61 | 44.44 | 0.38 |
| 13 | Blood | 24 , 58 | 43.9 | 42.9 | 43.09 | 43.30 | 0.53 |
| 14 | Blood | 24 , 58 | 44.1 | 44.33 | 44.14 | 44.19 | 0.12 |
| 15 | Blood | 24 , 58 | 43.5 | 43.67 | 43.03 | 43.40 | 0.33 |
| 16 | Blood | 24 , 58 | 43.6 | 43.12 | 43.23 | 43.32 | 0.25 |
| 17 | Blood | 24 , 58 | 43.4 | 43.01 | 43.41 | 43.27 | 0.23 |
| 18 | Blood | 24 , 58 | 43.55 | 43.51 | 43.53 | 43.53 | 0.02 |
| 19 | Blood | 24 , 58 | 43.23 | 43.02 | 43 | 43.08 | 0.13 |
| 20 | Blood | 24 , 58 | 43.01 | 43.1 | 43.09 | 43.07 | 0.05 |
| 21 | Blood | 24 , 58 | 43.03 | 43 | 43.06 | 43.03 | 0.03 |
| 22 | Blood | 24 , 58 | 43 | 43.04 | 43.02 | 43.02 | 0.02 |
| 23 | Blood | 24 , 58 | 43.02 | 43.1 | 43.09 | 43.07 | 0.04 |
| 24 | Blood | 24 , 58 | 42.9 | 42.92 | 42.77 | 42.86 | 0.08 |
| 25 | Blood | 24 , 58 | 42.7 | 42.56 | 42.75 | 42.67 | 0.10 |
| 26 | Blood | 24 , 58 | 42.9 | 42.78 | 42.83 | 42.84 | 0.06 |
| 27 | Blood | 24 , 58 | 42.7 | 42.9 | 41.7 | 42.43 | 0.64 |
| 28 | Blood | 24 , 58 | 42.8 | 42.86 | 42.76 | 42.81 | 0.05 |

Table 4.10: Table showing the time course of DNA quantification incubated at temperature of 24 °C and at a RH of 58% over a duration of 28 days. Data are original and mean values.

Table 4.10 shows the time course of DNA quantification at 24 °C and at a RH of 58% over a duration of 28 days. Data are mean ± SD, n=3. Note that at day 11 DNA value was zero due to human error and day 1 is in fact the zero time point i.e. the sample with no treatment. The results show no significant degradation of the DNA samples comparing day one with day 28. ($p > 0.05$).

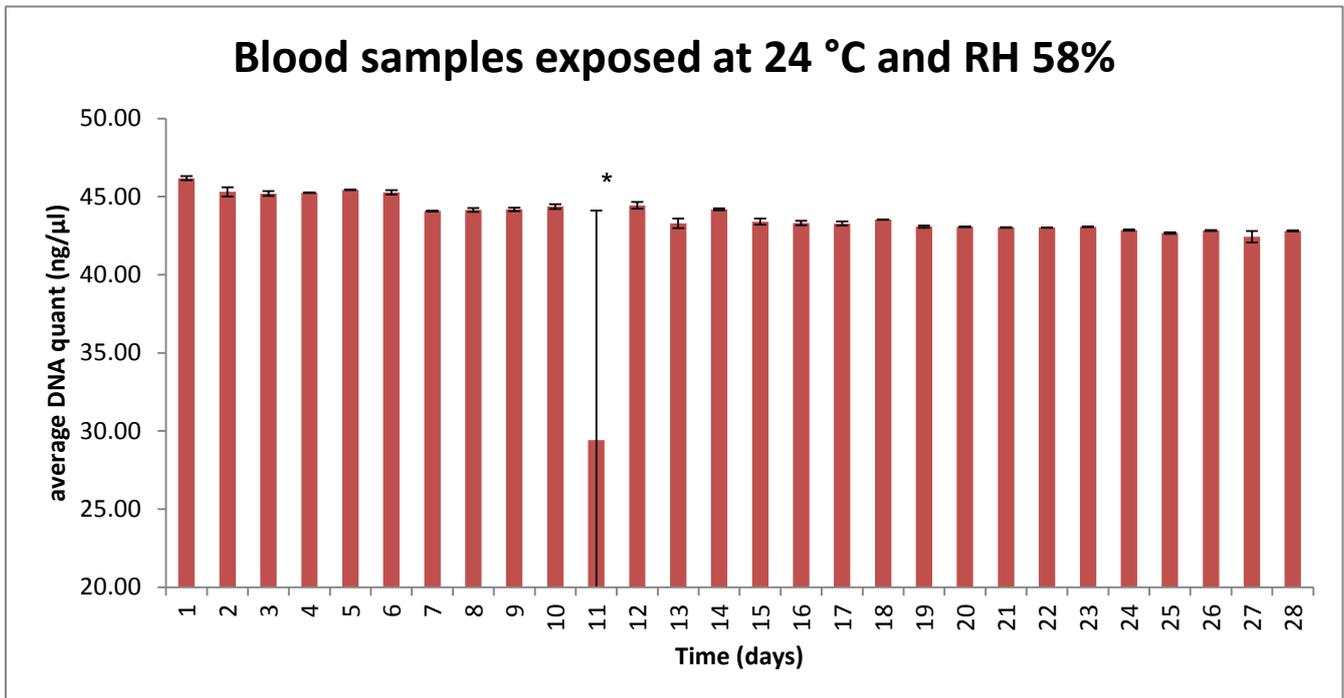


Figure 4.14: Bar charts showing time course of DNA quantification following extraction of 50 μ l of human blood stain samples incubated at temperature of 24 °C and at a RH of 58% over a duration of 28 days. Data are mean \pm SD, n=3. Note that at day 11, the DNA value was zero reading in one sample probably due to human error.

The results in figure 4.14 show that at 24 °C and at a RH of 58%, the DNA quantity in blood samples showed no significant ($P>0.05$) degradation of DNA comparing day one with day 28.

4.14.9 DNA quantification from human semen samples following extraction at 24 °C and at 58% humidity

| Days | Samples | Temp °C and RH % | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | mean | SD |
|------|---------|------------------|---------------------|---------------------|---------------------|-------|-------|
| 1 | Semen | - | 55.28 | 55.31 | 55.1 | 55.23 | 0.11 |
| 2 | Semen | 24 , 58 | 54.19 | 54.61 | 53.01 | 53.94 | 0.83 |
| 3 | Semen | 24 , 58 | 54.54 | 55.6 | 56.01 | 55.38 | 0.76 |
| 4 | Semen | 24 , 58 | 53.24 | 54.11 | 53.48 | 53.61 | 0.45 |
| 5 | Semen | 24 , 58 | 56.37 | 54.39 | 55.32 | 55.36 | 0.99 |
| 6 | Semen | 24 , 58 | 54.11 | 55.18 | 56.12 | 55.14 | 1.01 |
| 7 | Semen | 24 , 58 | 55.21 | 55.34 | 55.11 | 55.22 | 0.12 |
| 8 | Semen | 24 , 58 | 53.41 | 54.17 | 54.8 | 54.13 | 0.70 |
| 9 | Semen | 24 , 58 | 56.22 | 54.9 | 55.3 | 55.47 | 0.68 |
| 10 | Semen | 24 , 58 | 53.28 | 53.16 | 54.89 | 53.78 | 0.97 |
| 11 | Semen | 24 , 58 | 54.63 | 55.23 | 54.68 | 54.85 | 0.33 |
| 12 | Semen | 24 , 58 | 54.64 | 55.48 | 55.27 | 55.13 | 0.44 |
| 13 | Semen | 24 , 58 | 55.61 | 55.13 | 54.37 | 55.04 | 0.63 |
| 14 | Semen | 24 , 58 | 55.2 | 55.11 | 55.17 | 55.16 | 0.05 |
| 15 | Semen | 24 , 58 | 54.51 | 54.14 | 54.68 | 54.44 | 0.28 |
| 16 | Semen | 24 , 58 | 55.28 | 53 | 55.64 | 54.64 | 1.43 |
| 17 | Semen | 24 , 58 | 55.11 | 53.25 | 54.37 | 54.24 | 0.94 |
| 18 | Semen | 24 , 58 | 54.11 | 56.33 | 54.93 | 55.12 | 1.12 |
| 19 | Semen | 24 , 58 | 54.37 | 55.13 | 55.57 | 55.02 | 0.61 |
| 20 | Semen | 24 , 58 | 55.01 | 56.69 | 55.27 | 55.66 | 0.90 |
| 21 | Semen | 24 , 58 | 55.22 | 55.19 | 55.17 | 55.19 | 0.03 |
| 22 | Semen | 24 , 58 | 54.02 | 55.68 | 55.01 | 54.90 | 0.84 |
| 23 | Semen | 24 , 58 | 54.89 | 55.68 | 56.11 | 55.56 | 0.62 |
| 24 | Semen | 24 , 58 | 56.23 | 20.01 | 55.11 | 43.78 | 20.60 |
| 25 | Semen | 24 , 58 | 54.6 | 54.28 | 55.22 | 54.70 | 0.48 |
| 26 | Semen | 24 , 58 | 54.68 | 55.68 | 55.1 | 55.15 | 0.50 |
| 27 | Semen | 24 , 58 | 55.57 | 53.68 | 55.44 | 54.90 | 1.06 |
| 28 | Semen | 24 , 58 | 55.22 | 56.71 | 54.51 | 55.48 | 1.12 |

Table 4.11: Table showing DNA quantification following extraction of 50 μl of human semen after dilution and incubated at temperature of 24 °C and at a humidity of 58% over a duration of 28 days. Data are original and mean values with SD; n=3.

Table 4.11 shows DNA quantification at 24 °C and at a humidity of 58% over a duration of 28 days. Note that day 1 is in fact the zero time point i.e. the sample with no treatment. The results showed no significant ($p>0.05$) degradation of DNA quantity in semen samples comparing levels in day 1 with day 28 at 24 °C and at 58% humidity.

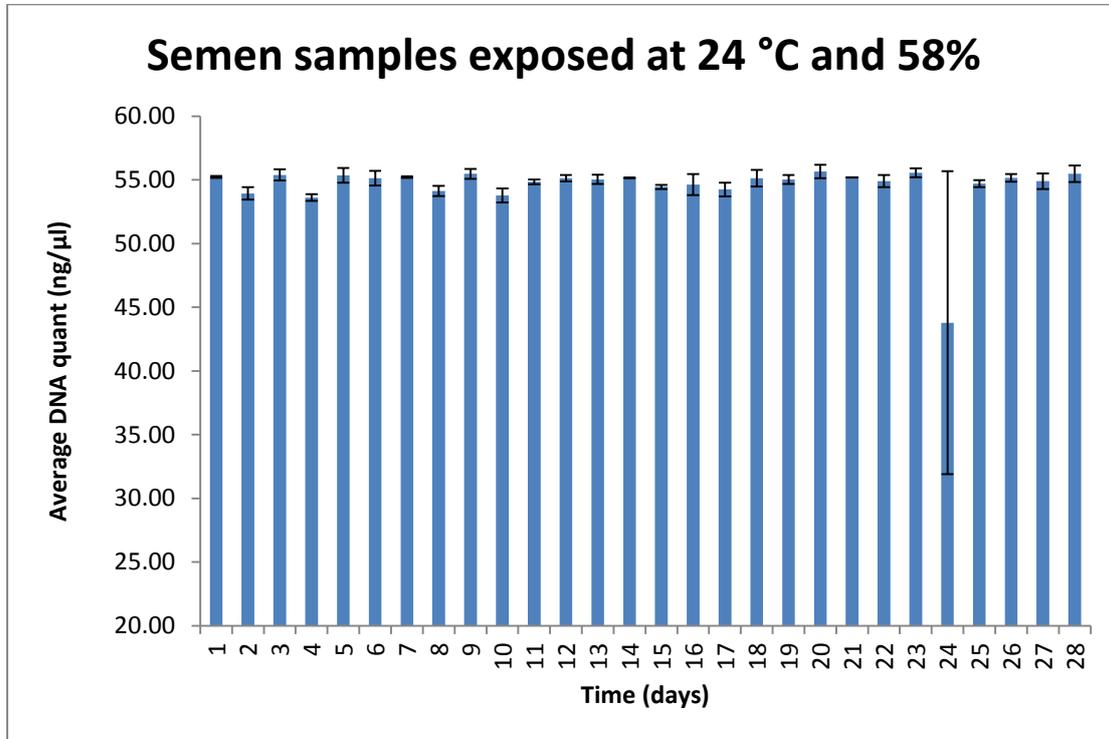


Figure 4.15: Bar charts showing the time course of DNA quantification following extraction of 50 ng/μl of human semen stain samples incubated at temperature of 24 °C and at a humidity 58% over a duration of 28 days. The extractions were done using the QIAamp DNA Investigator protocol. Data are mean \pm SD, n=3. $P > 0.05$ for day 1 compared to all other days. Note that at day 24 DNA value was little in one test due to human error. (Data taken from table 5.11).

The results in figure 4.15 show that at 24 °C and at a RH of 58%, the DNA quantity in semen samples were more or less similar but these values were not significantly different comparing day 1 with all other values up to day 28. ($p > 0.05$).

5.15.10 DNA quantification following extraction of human saliva material at a temperature of 4 °C and at a humidity of 61%.

| Days | Samples | Temp°C and RH % | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | Mean | SD |
|------|---------|-----------------|---------------------|---------------------|---------------------|-------|------|
| 1 | Saliva | - | 32.40 | 32.46 | 32.49 | 32.45 | 0.05 |
| 2 | Saliva | 4 , 61 | 32.50 | 32.44 | 32.50 | 32.48 | 0.03 |
| 3 | Saliva | 4 , 61 | 32.51 | 32.34 | 32.51 | 32.45 | 0.10 |
| 4 | Saliva | 4 , 61 | 32.41 | 32.30 | 32.39 | 32.37 | 0.06 |
| 5 | Saliva | 4 , 61 | 32.76 | 32.73 | 32.73 | 32.74 | 0.02 |
| 6 | Saliva | 4 , 61 | 32.67 | 32.70 | 32.64 | 32.67 | 0.03 |
| 7 | Saliva | 4 , 61 | 32.63 | 32.59 | 32.62 | 32.61 | 0.02 |
| 8 | Saliva | 4 , 61 | 32.80 | 32.81 | 32.81 | 32.81 | 0.01 |
| 9 | Saliva | 4 , 61 | 32.70 | 32.72 | 32.69 | 32.70 | 0.02 |
| 10 | Saliva | 4 , 61 | 32.73 | 32.73 | 32.76 | 32.74 | 0.02 |
| 11 | Saliva | 4 , 61 | 32.79 | 32.65 | 32.68 | 32.71 | 0.07 |
| 12 | Saliva | 4 , 61 | 32.75 | 32.72 | 32.74 | 32.74 | 0.02 |
| 13 | Saliva | 4 , 61 | 32.69 | 32.70 | 32.71 | 32.70 | 0.01 |
| 14 | Saliva | 4 , 61 | 32.40 | 32.46 | 32.49 | 32.45 | 0.05 |
| 15 | Saliva | 4 , 61 | 32.30 | 32.39 | 32.41 | 32.37 | 0.06 |
| 16 | Saliva | 4 , 61 | 32.73 | 32.73 | 32.76 | 32.74 | 0.02 |
| 17 | Saliva | 4 , 61 | 32.71 | 32.64 | 32.67 | 32.67 | 0.04 |
| 18 | Saliva | 4 , 61 | 32.58 | 32.62 | 32.63 | 32.61 | 0.03 |
| 19 | Saliva | 4 , 61 | 32.83 | 32.81 | 32.80 | 32.81 | 0.02 |
| 20 | Saliva | 4 , 61 | 32.74 | 32.69 | 32.70 | 32.71 | 0.03 |
| 21 | Saliva | 4 , 61 | 32.70 | 32.75 | 32.73 | 32.73 | 0.03 |
| 22 | Saliva | 4 , 61 | 32.71 | 32.70 | 32.73 | 32.71 | 0.02 |
| 23 | Saliva | 4 , 61 | 32.32 | 32.30 | 32.32 | 32.31 | 0.01 |
| 24 | Saliva | 4 , 61 | 32.46 | 32.47 | 32.44 | 32.46 | 0.02 |
| 25 | Saliva | 4 , 61 | 32.53 | 32.54 | 32.56 | 32.54 | 0.02 |
| 26 | Saliva | 4 , 61 | 32.53 | 32.55 | 32.56 | 32.55 | 0.02 |
| 27 | Saliva | 4 , 61 | 32.43 | 32.44 | 32.43 | 32.43 | 0.01 |
| 28 | Saliva | 4 , 61 | 32.39 | 32.40 | 32.43 | 32.41 | 0.02 |

Table 4.12: Table showing time course of DNA quantification following extraction of 50 μl of human saliva and incubated at temperature of 4 °C and RH 61% over a duration of 28 days. Data are original and mean data with SD; n=3.

Table 4.12 shows the time course of DNA at 4 °C and RH 61% over a duration of 28 days. Note that day 1 is in fact the zero time point i.e. the sample with no treatment. The showed no significant ($p > 0.05$) degradation of DNA in saliva samples comparing all days with day 1.

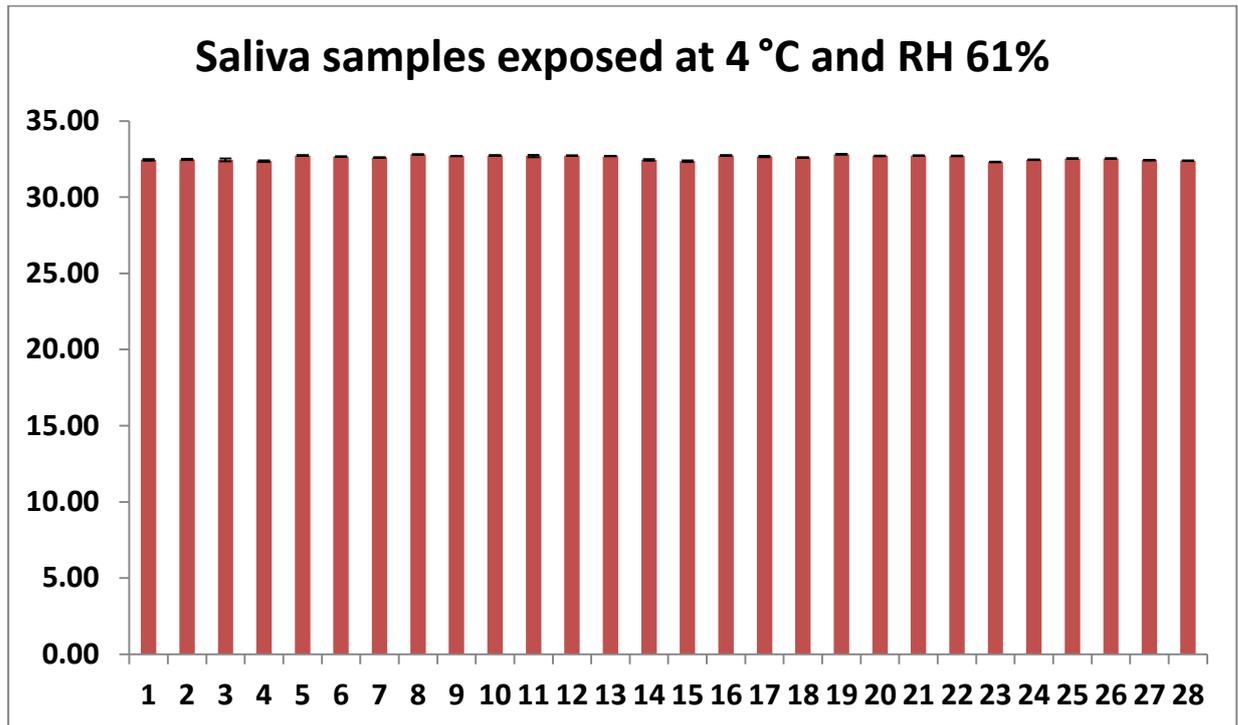


Figure 4.16: Bar charts showing the time course average of DNA measurement recovered from triplicate extractions in saliva samples. DNA was extracted from 50 μ l human saliva samples that subjected to 4 °C and at RH of 61% for 28 days. Data are mean (\pm SD), $n=3$. $P > 0.05$ for day 1 compared to all other days.

The results in figure 4.16 show that exposure of salivary DNA at 4 °C and at a RH of 61%, over a duration of 28 days had no significant ($p > 0.05$) effect on salivary DNA degradation comparing all the other days with day 1.

4.14.11 DNA quantification from human blood samples following extraction at 4 °C and at 61% humidity

| Days | Samples | Temp °C and RH % | DNA quant 1 (ng/µl) | DNA quant 2 (ng/µl) | DNA quant 3 (ng/µl) | Mean | SD |
|------|---------|------------------|---------------------|---------------------|---------------------|-------|-------|
| 1 | Blood | - | 45.22 | 45.09 | 45.32 | 45.21 | 0.12 |
| 2 | Blood | 4 , 61 | 45.21 | 45.26 | 45.37 | 45.28 | 0.08 |
| 3 | Blood | 4 , 61 | 45.32 | 45.33 | 45.42 | 45.36 | 0.06 |
| 4 | Blood | 4 , 61 | 45.33 | 45.54 | 45.67 | 45.51 | 0.17 |
| 5 | Blood | 4 , 61 | 45.36 | 45 | 45.36 | 45.24 | 0.21 |
| 6 | Blood | 4 , 61 | 45.12 | 45.34 | 45.15 | 45.20 | 0.12 |
| 7 | Blood | 4 , 61 | 45.57 | 45.34 | 45.67 | 45.53 | 0.17 |
| 8 | Blood | 4 , 61 | 45.57 | 45.34 | 45.96 | 45.62 | 0.31 |
| 9 | Blood | 4 , 61 | 45.47 | 45.17 | 45.01 | 45.22 | 0.23 |
| 10 | Blood | 4 , 61 | 45.82 | 45.49 | 45.78 | 45.70 | 0.18 |
| 11 | Blood | 4 , 61 | 45.36 | 0 | 45.16 | 30.17 | 26.13 |
| 12 | Blood | 4 , 61 | 45.68 | 45.74 | 45.78 | 45.73 | 0.05 |
| 13 | Blood | 4 , 61 | 45.92 | 44.9 | 45.39 | 45.40 | 0.51 |
| 14 | Blood | 4 , 61 | 45.37 | 45.29 | 45.33 | 45.33 | 0.04 |
| 15 | Blood | 4 , 61 | 45.67 | 45.34 | 45.56 | 45.52 | 0.17 |
| 16 | Blood | 4 , 61 | 45.2 | 45.26 | 45.09 | 45.18 | 0.09 |
| 17 | Blood | 4 , 61 | 45.21 | 45.26 | 45.03 | 45.17 | 0.12 |
| 18 | Blood | 4 , 61 | 45.09 | 45.11 | 45 | 45.07 | 0.06 |
| 19 | Blood | 4 , 61 | 45.19 | 45.05 | 45.12 | 45.12 | 0.07 |
| 20 | Blood | 4 , 61 | 45.38 | 45.29 | 45.65 | 45.44 | 0.19 |
| 21 | Blood | 4 , 61 | 45 | 45.07 | 45.02 | 45.03 | 0.04 |
| 22 | Blood | 4 , 61 | 45.31 | 45.4 | 45.29 | 45.33 | 0.06 |
| 23 | Blood | 4 , 61 | 45 | 45.06 | 45.22 | 45.09 | 0.11 |
| 24 | Blood | 4 , 61 | 45.66 | 45.28 | 45.48 | 45.47 | 0.19 |
| 25 | Blood | 4 , 61 | 45.45 | 45.27 | 45.44 | 45.39 | 0.10 |
| 26 | Blood | 4 , 61 | 45.45 | 45.87 | 45.09 | 45.47 | 0.39 |
| 27 | Blood | 4 , 61 | 45.05 | 45.11 | 45.18 | 45.11 | 0.07 |
| 28 | Blood | 4 , 61 | 45.26 | 45.33 | 45.43 | 45.34 | 0.09 |

Table 4.13: Table showing time course of DNA quantification following extraction of 50 µl of human blood and incubated at temperature of 4 °C and at a RH of 61%. Data are original as well as the mean ± SD values, n=3.

Table 4.13 shows the time course of DNA quantification at 4 °C and at a RH of 61%. Data are mean ± SD, n=3. Note that at day 11 DNA value was zero due to human error and day 1 is in fact the zero time point i.e. the sample with no treatment. The results showed no significant ($p>0.05$) degradation of DNA in blood samples over a duration of 28 days comparing values from day one to day 28.

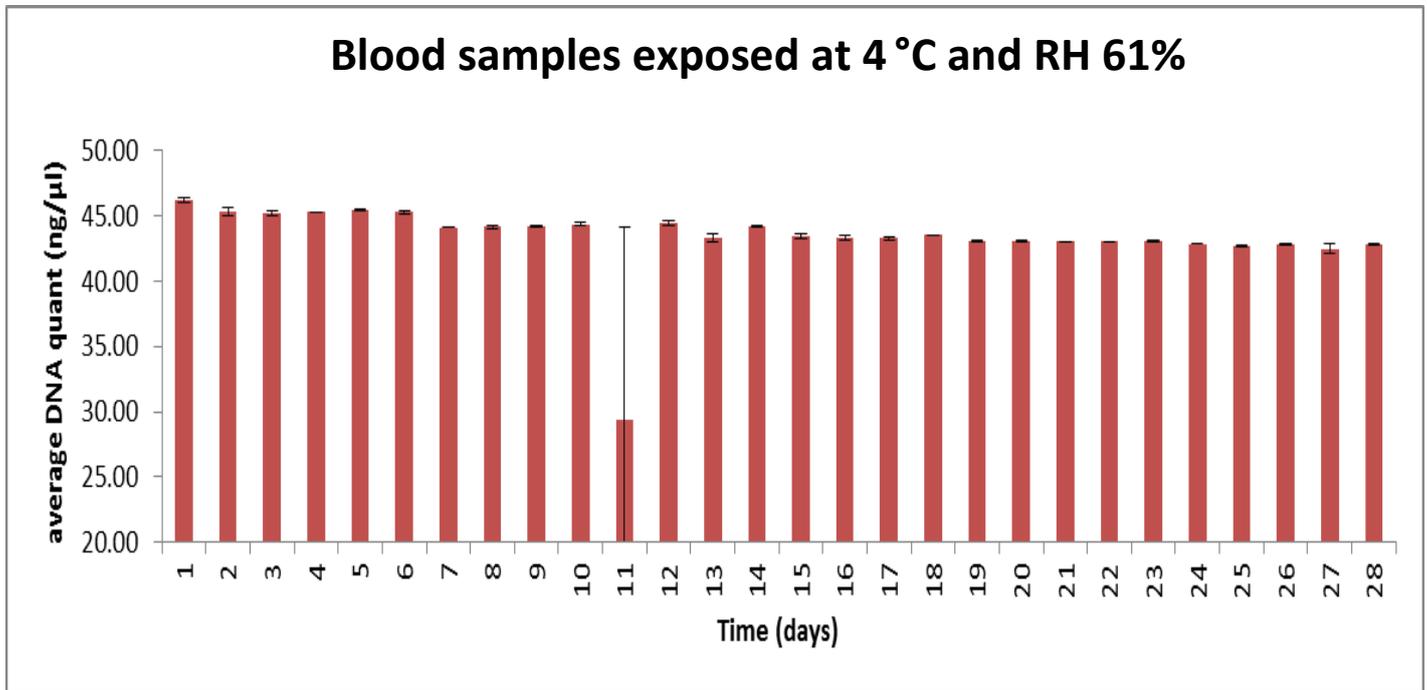


Figure 4.17: Bar charts showing the time-course average of DNA recovered from triplicate measurements in human blood samples at temperature 4 °C and RH 61% for a period of 28 days. The extractions were done using the DNeasy® protocol and all values are expressed as mean \pm SD. $P > 0.05$ for day 1 compared to all other days. Note that large SD in day 11 due to human error in one of the readings and day 1 is in fact the zero time point i.e. the sample with no treatment.

The results in figure 4.17 reveal that there was no significant ($p > 0,05$) DNA degradation in blood samples comparing day one to day 28.

4.14.12 DNA quantification of human semen samples following extraction at 4 °C and at 61% humidity

| Days | Samples | Temp °C and RH% | DNA quant 1 (ng/μl) | DNA quant 2 (ng/μl) | DNA quant 3 (ng/μl) | mean | SD |
|------|---------|-----------------|---------------------|---------------------|---------------------|-------|-------|
| 1 | Semen | - | 55.28 | 54.28 | 55.36 | 54.97 | 0.60 |
| 2 | Semen | 4 , 61 | 55.2 | 55.19 | 54.6 | 55.00 | 0.34 |
| 3 | Semen | 4 , 61 | 56.17 | 54.54 | 55.64 | 55.45 | 0.83 |
| 4 | Semen | 4 , 61 | 55.32 | 53.24 | 55.1 | 54.55 | 1.14 |
| 5 | Semen | 4 , 61 | 54.8 | 56.27 | 55.2 | 55.42 | 0.76 |
| 6 | Semen | 4 , 61 | 56.22 | 54.12 | 55.07 | 55.14 | 1.05 |
| 7 | Semen | 4 , 61 | 55.48 | 55.11 | 55.04 | 55.21 | 0.24 |
| 8 | Semen | 4 , 61 | 54.68 | 53.11 | 56.12 | 54.64 | 1.51 |
| 9 | Semen | 4 , 61 | 55.92 | 56.2 | 54.68 | 55.60 | 0.81 |
| 10 | Semen | 4 , 61 | 55.01 | 53.47 | 53.55 | 54.01 | 0.87 |
| 11 | Semen | 4 , 61 | 54.1 | 30.2 | 55.27 | 46.52 | 14.15 |
| 12 | Semen | 4 - 61 | 54.28 | 54.28 | 55.01 | 54.52 | 0.42 |
| 13 | Semen | 4 , 61 | 55.12 | 55.48 | 55.12 | 55.24 | 0.21 |
| 14 | Semen | 4 , 61 | 55.46 | 55.29 | 55.02 | 55.26 | 0.22 |
| 15 | Semen | 4 , 61 | 54.68 | 54.38 | 54.11 | 54.39 | 0.29 |
| 16 | Semen | 4 , 61 | 56.81 | 55.69 | 53.44 | 55.31 | 1.72 |
| 17 | Semen | 4 , 61 | 54.62 | 55.18 | 53.76 | 54.52 | 0.72 |
| 18 | Semen | 4 , 61 | 54.35 | 54.1 | 56.34 | 54.93 | 1.23 |
| 19 | Semen | 4 , 61 | 55.62 | 54.58 | 55.36 | 55.19 | 0.54 |
| 20 | Semen | 4 , 61 | 55.11 | 55.58 | 56.47 | 55.72 | 0.69 |
| 21 | Semen | 4 , 61 | 55.46 | 55.69 | 55.27 | 55.47 | 0.21 |
| 22 | Semen | 4 , 61 | 55.47 | 54.59 | 55.57 | 55.21 | 0.54 |
| 23 | Semen | 4 , 61 | 55.27 | 54.79 | 55.47 | 55.18 | 0.35 |
| 24 | Semen | 4 , 61 | 54.57 | 56.89 | 55.02 | 55.49 | 1.23 |
| 25 | Semen | 4 , 61 | 55.57 | 54.39 | 54.14 | 54.70 | 0.76 |
| 26 | Semen | 4 , 61 | 54.13 | 54.93 | 55.27 | 54.78 | 0.59 |
| 27 | Semen | 4 , 61 | 55.27 | 55 | 53.43 | 54.57 | 0.99 |
| 28 | Semen | 4 , 61 | 55.02 | 55.17 | 56.76 | 55.65 | 0.96 |

Table 4.14: Table showing DNA quantification following extraction of 50 μl of human semen samples and incubated at temperature of 4 °C and at a RH of 61% over a duration of 28 days. Data are original and mean values.

Table 4.14 shows the DNA quantification at 4 °C and at a RH of 61% over a duration of 28 days. Each sample was analyzed in triplicate at 4 °C. Note that day 1 is in fact the zero time point i.e. the sample with no treatment. The results showed no significant degradation of DNA quantity in semen samples at temperature of 4 °C and at a RH of 61%, comparing levels in day 1 with day 28 at 4 °C and RH 61%.

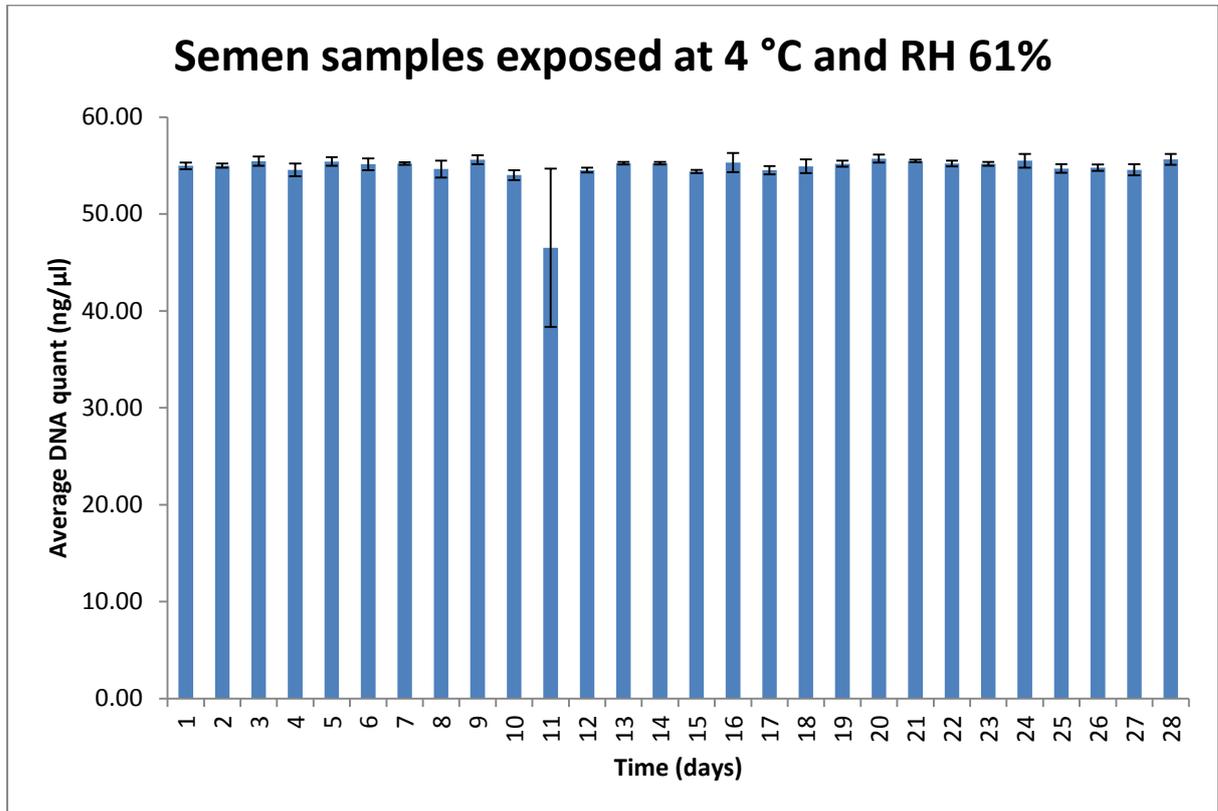


Figure 4.18: Bar charts showing the time course of DNA quantification following extraction of 50 ng/μl of human semen stain material incubated at temperature of 4 °C and RH 61% over a duration of 28 days. Data are mean ± SD, n=3. $P > 0.05$ for day 1 compared to all other days. Note that at days 24 DNA value was reduced in one test due to human error.

The results in figure 4.18 show that the DNA quantity in semen samples remained more or less same at 4 °C and RH 61% and for all 28 days, but these values were not significantly ($p > 0.05$) different from day 1.

4.15 Discussion

The experiments presented in this chapter of the study were done to simulate circumstances of a crime scene in which a body may be found in different seasons in Kuwait. The techniques employed in this study for DNA quantification provided the best markers with the DNA preservation in the samples used in this investigation. The DNA is normally degraded rapidly post-mortem, initially from the enzymes released as the cell dies losing its structural integrity and then later from environmental insults (Dissing, 2010). For this purpose, blood, saliva and semen samples were exposed in the present study to four ranges of temperature (55°C, 37°C, 24°C and 4°C) and four ranges of relative humidity (41%, 55%, 58% and 61%), respectively at different time points starting from day 1 to day 28. The aim of this study was to ascertain how DNA can persist following changes in both temperature and humidity. The rationale was to mimic a natural environmental condition normally occurring in Kuwait during different season.

The results show that at a low a temperature of 4°C and at a RH of 61% the DNA was not significantly degraded either in blood, saliva or semen samples for the whole period of the study (28 days) comparing values for day one with all other days. Typically, the mean values of 45.21 ± 0.12 ng/ μ l, 32.45 ± 0.05 ng/ μ l and 54.97 ± 0.60 ng/ μ l were obtained for blood, saliva and semen, respectively at day 1 compared to values of 45.34 ± 0.09 ng/ μ l, 32.41 ± 0.02 ng/ μ l and 55.65 ± 0.96 ng/ μ l at day 28, respectively, These data revealed no significant change in DNA quantity at a low temperature of 4°C and at a humidity of 58%. It is well known that low temperature tends to preserve DNA

better in the samples (Butler et al., 2003; Dissing, 2010). Similarly, at temperature of 24°C and at a RH of 58%, the quantity of DNA in blood samples was 46.3 ± 0.24 ng/ul at day 1 and this decreased slightly to 42.81 ± 0.05 ng/ul at day 28. Likewise, the DNA quantity found in saliva samples was 32.70 ± 0.02 ng/ul at day 1 and then decreased slightly to 31.34 ± 0.06 ng/ul at day 28. Similarly, the DNA quantity obtained from semen samples in 24°C and at a RH of 58% at day 1 was 55.23 ± 0.11 ng/ul compared to 55.48 ± 1.12 ng/ul. at day 28. These results showed no significant degradation of DNA comparing DNA level at day 1 to day 28 at 24°C. A temperature of 24°C and a humidity of 58% are physiological ranges for DNA persistence.

The present results have also shown that the quantity of DNA from human blood samples incubated at 37°C and at a humidity of 55% decreased as the time points increased. These results show that the relative humidity of 55% and temperature of 37°C did not play any significant role in DNA degradation at least up to day 20 (Figure 4.6, 4.7 and 4.8). As time increased, DNA started to degrade gradually becoming significant after day 19 and onwards. This slow degradation was probably due to the fact that the humidity was probably not high enough. Typically, DNA quantification of human blood following extraction at 37°C was 46.14 ± 0.22 ng/ul at day one then it started to decrease reaching 36.05 ± 0.07 ng/ul at day 28. The same is true for the result obtained at 37°C in saliva samples. The DNA quantity was 45.03 ± 0.03 ng/ul on day 1 and reduced to 34.71 ± 0.07 ng/ul at day 28. Surprisingly, in semen samples the DNA quantity at day 1 was 55.49 ng/ul ± 0.56 and remained almost the same at day 28 at a mean value of 55.12 ± 0.38 ng/ul

(Figure 4.10, 4.11 and 4.12). These results have indicated that the quantity of DNA in both blood and saliva samples showed slow degradation which started from around days 15-19 and this was probably due to both microorganisms effect of temperature and humidity (Alonso et al., 2004). In contrast, in semen samples the quantity of DNA remained almost the same with no sign of degradation during the whole period.

In contrast, the result obtained from this study have shown that when the temperature was raised to 55°C, the DNA started to degrade immediately and rapidly in both blood and saliva samples with time until it reached zero at day 14. The quantity of DNA in blood at day 1 was 44.88 ± 0.57 ng/ μ l. This amount then started to degrade in quantity after the third day until it reached zero at day 14. Similarly, with temperature increased to 55°C and 41% RH, the results obtained from saliva samples revealed that DNA quantity was 31.10 ± 0.02 ng/ μ l on day 1 and this then started to degrade with time until it reached zero at day 17. These results for both saliva and blood samples show more or less the same pattern for DNA degradation over time at 55°C. In contrast, the quantity of DNA obtained from semen sample at day 1 was 55.28 ± 0.24 ng/ μ l compared to 55.30 ± 1.35 ng/ μ l at day 28. The results have clearly indicated that semen samples seem to be resistant to degradation at high temperature even when exposed over 28 days.

Temperature and humidity are known to play both physiological and an evident role in DNA degradation and Kuwait is a typical country in the Middle East in which the temperature is liable to be variable due to seasonal changes. Although, relative humidity in Kuwait is not high to affect the DNA degradation, several studies have concluded that elevated humidity provides

the best growth conditions for microorganism (bacteria and fungi), which participate in the degradation process (Fisher et al., 1993; Hill et al., 2007). Similar findings were obtained by Zhang et al (2010) on bone marrow specimens that were preserved in temperature of 24°C for 2 months.

The technique of DNA profiling is expensive and, moreover, it is a time consuming procedure. The data obtained in this study have indicated that DNA survival could be found in both saliva and blood samples until day 13 and 17 respectively, when the temperature reached 55°C. The results showed significant increases in the level of DNA degradation along with increase in both time and temperature (55°C). In contrast, the results show that exposure of sperms (semen) to a high temperature of 55°C and humidity of 41% had no significant effect on DNA degradation comparing day 1 with all the other days. The results of this study have also shown no significant changes in DNA degradation in the blood and saliva samples exposed at either 24°C or 4 °C. It is now well known that low temperatures have the ability to preserve the quantity of DNA in biological samples (Alonso et al., 2004; Dissing, 2010). Firstly, the results have clearly indicated that, if samples of either blood, saliva, semen or similar materials were transferred to the DNA laboratory after 12-15 days and the crime was committed in a very hot season such as July (55 °C or above), then there is no chance to find survival DNA and moreover, there may be no need to waste time, materials and manpower in assessing the samples for DNA. Second, the results have also indicated that DNA can be obtained from semen evidence even during the hot season because it can resist elevated temperature, probably because of the structure of the sperm head (Zhang et al, 2010).

The success of Forensic Scientist in investigating a crime scene for evidence is the ability to profile samples for DNA survival successfully (Utsuno and Minaguchi, 2004). When a DNA sample has been degraded by any means, the average size of a DNA fragment might be reduced to less than 300 bp leading to insufficient DNA template, resulting in an inability to identify the individual.

Several studies have been done to investigate factors affecting biological evidence that are recovered from a crime scene (Alaeddini et al., 2010). These evidences are usually exposed to different environmental insults or conditions such as heat, humidity, chemical and microorganisms (bacteria and fungi) and others (Dissing, 2010). These factors can affect the stability and survival of the DNA (Coble and Butler, 2005). Thus, to ensure that the recovered DNA can support the casework or not, the effect of environmental conditions on DNA survival should be investigated.

A similar study was done by Alaeddini et al (2010) who investigate the environmental factors affecting DNA degradation process that were exposed to a high temperature of 48°C. These results revealed that a temperature of 48°C can cause a high degree of degradation on blood stain samples (Alaeddini et al., 2010). In another study conducted by Rebecchi et al (2009), on the effect of temperature over 21 days of exposure, their results revealed that high temperatures reaching 50°C can change significantly the state of DNA stability thus reducing its survival (Rebecchi et al., 2009).

The influence of the temperature of the surrounding environment on the stability of DNA was also investigated by Dissing (2010), who observed no

microbial growth at higher temperatures (45–65°C) and at 100% humidity. He also observed that DNA could be amplifiable after eight months at a temperature of 45°C and 100% humidity, but only survived for one month at 55 °C accompanied with humidity of 100% (Dissing, 2010). The evidence of the present study and those in the literature have clearly indicated that all biological evidence should be dealt with immediately after being in crime scene or when a body is transferred to the Morgue. Moreover, all samples must also be carefully collected. Surprisingly, the DNA testing commonly employed in forensics laboratories are often time consuming, and require the consumption of a substantial percentage of the precious samples (Alaeddini et al., 2010). Together, the results of the present study are in close agreement with other similar studies, that DNA is extremely sensitive to elevated temperatures over time.

4.16 Conclusion

The main and general purpose of forensic science is to serve justice and to solve the forensic case work. The biggest concern for DNA laboratory is the stability of DNA from forensic evidence, in which biological evidences are exposed to various environmental factors and insults. These factors play crucial roles in DNA degradation and this indeed represents a challenge for Forensic Scientists. It is well recognised that DNA often persist for months or years as ambient and untreated dry stains. At the same time, it is also well known that various parameters can affect the persistence of DNA and increase the level of DNA degradation resulting in a failure to solve a forensic case.

The present work in this thesis was designed especially to investigate the effect of environmental insults, as in temperature changes accompanied by the correspondence humidity, on DNA samples in human saliva, blood and semen samples exposed over a period of 28 days. The data show that at an environmental temperature of 55°C, there was no DNA survival in either blood or saliva after 13 and 17 days of exposure. However, at 37°C, DNA degraded slowly in both samples reaching significant value after 19-25 days of exposure. In contrast, exposure of both samples at 4°C and 24°C had no significant effect on DNA quantity throughout the whole experimental period. Furthermore, DNA in semen samples were resistant to degradation either at low or high temperature. From the present results, it is tempting to conclude that if a crime was committed in the summer in Kuwait with a temperature of 55°C or higher, then there will be no need to perform a DNA analysis if the samples of blood and saliva were exposed for more than 15 days. Second, since this procedure and test are costly and time consuming, then there is no need to analyze the samples for DNA after about 15 days of exposure.

Further work should be done using short amplicon as miniSTR kit to check the level of degradation in the DNA markers by time. Furthermore, it is equally important to determine the suitability of a DNA before it is used. In addition, these experiments should also be done side by side with different environmental factors such as humidity and UV, indoor and outdoor environmental insults to determine the relation between these factors and DNA degradation.

CHAPTER 5

GENERAL DISCUSSION, CONCLUSIONS AND SCOPE OF FUTURE STUDIES

5.1 Patterns of accidental deaths in Kuwait

The data presented in this thesis showed that accidental deaths were the major cause of unnatural deaths in Kuwait over the seven years of study during the period 2003-2009 accounting for 86% of all reported cases. This finding is similar to a study done in Qatar in 1992 which found that accidental deaths constituted a high proportion of un-natural deaths representing 76% (Al-Barraq and Farahat, 2011). The results of the present study revealed that road traffic accident (RTA), one aspect of accidental deaths was the common mean of un-natural deaths constituting 65% of the total accidental cases. The majority of RTA victims in the current study were in the age group 20-29 years and they represented 27%. According to International Road Federation (IRF) in 2010, around 200 Kuwaitis are killed and 6,000 are injured annually in traffic accidents, giving Kuwait, the Northern Arabian Gulf Country, the world's top ranking in the number of deaths and injuries resulting from road traffic accidents (IRF, 2010). The World Health Organization estimates that by the year 2020, road traffic injuries will be the third leading cause of disability and life lost worldwide (WHO, 2010). The analysis of the International Road Federation's world road statistics found that five countries in the Middle East (Kuwait, Oman, Saudi Arabia, Qatar and United Arab Emirates) are among the highest road traffic death rates in the world (Al-Hassan, 2011). All these five countries had more than 18 deaths per 100,000 people in year 2000 (Al-Hassan, 2011). In the United States, RTA is considered as the ninth cause of deaths (Ivers et al., 2009). This comparison between Kuwait, as a small country, and the United States, as the largest modernized country in the world is very worrying. The rate of car accidents and deaths from road traffic

accidents, in Kuwait have double compared to the number of car accidents and deaths from road traffic accidents in the US (Al-Hassan, 2011). Leading experts have concluded that intensive management should be taken into consideration with regards to this issue by the implementation of scientific measures (IRF, 2010). Research into car accidents in various countries has found that the rates of seat belt wearing are substantially lower in total collisions than the general average rate (Mobeireek et al., 2008). For example, only around 55% of drivers in total crashes in Finland wore seatbelts compared to 35% in Sweden (Shalaby et al., 2010). In Kuwait several measures have been taken to assure the safety of the individual. These include penalty of not wearing seat belt, speeding and for using the cell phone while driving, but still the RTA remain high. The present results reflect how the problem of RTA is a source of social and health concern throughout the world generally and particularly in Kuwait.

During the period of the present study, males outnumbered females in accidental deaths accounting for 74% compared to 26% among females. The differences in the rate of accidents among males compared to females were probably due to a lack of attention, and negligence of safety protocols, speeding and not taking precautions. Second, more males drive cars in Kuwait compared to females. Similarly, the results show that more males died due to falls from heights especially in occupational injuries in which safety precaution are lacking and moreover, where little or no compliance with quality assurance within their working area and environment by employees. Second, more males are employed in those dangerous occupations compared to females. Similar findings were also reported in other studies in

Turkey for occupational injuries where the number of deaths among males is much higher compared to females (Christen, 2006).

5.2 Patterns of suicide in Kuwait

Suicides must be competently investigated by both law enforcement and by the office of Medical Examiner to eliminate any issues relating to homicide and to solve the actual cause of death (Dogan et al., 2010). During the period of the present study, 6% of all deaths reported for autopsy at the GDCE were determined to be suicidal in manner. The present study revealed that hanging was the commonest method employed for suicide accounting for 60% followed by poisoning 14%. The results also showed that falls from heights were the least common suicidal method and accounted for only 4%. To explain this, hanging is considered the easiest available means to commit suicide. This may be attributed to the high population of migrant workers in Kuwait in which special groups are often driven to suicide by stress, harsh living and working conditions and loneliness due to home sick (Fido, 2009). Similar findings have been reported in Saudi Arabia, where hanging is a very common method of suicide among the immigrant workers (El-Fawal, 1999). The report also pointed that cultural, religious and economic influences may account for the differences and they also indicated that long period of separation from families may explain such findings (El-Fawal, 1999). In addition, poisoning has been shown as the second preferred manner of suicide in Kuwait. This could be attributed by the easy availability of hazardous substances and medicines in the society, in which medicines that contain paracetamol and ibuprofen and other allergy medication are easily

available over the counter with no need for a prescription from practising Physicians in Kuwait (Fido, 2009).

In addition, the study has provided strong evidence to show that suicide is mostly abundant in Non-Kuwaitis, in which they constituted 77% compared to 23% for Kuwaiti Citizens. The same is also true in Saudi Arabia, where Expatriates have greater suicidal death rates compared to Saudis Nationals. They accounted for 65% when compared to 35% for non-Saudi Nationals (El-Fawal, 1999). This could be due to the fact that Expatriates are suffering from stress, depression and over working, as well as leaving their families behind in poverty stricken countries. Because of these adverse conditions they mostly end up by taking their own lives via suicide, a form of death (El-Fawal and Awad, 1997).

During the period of this study, the results have revealed that male subjects outnumbered female subjects in all patterns of suicidal deaths except in falls from heights, in which the number of females was greater accounting for 64% compared to 36% for males. This can be explained that, falls from heights, especially from a high building, is the easiest available method of suicide for female house maids. Several cases have been reported to GDCE for attempted suicides (GDCE Annual report, 2010). Recent epidemiological studies in Saudi Arabia highlighted the fact that psychiatric disorders, marriage, masculine dominance and home sickness are among the most important interrelated reasons in increasing the likelihood of Expatriates being victims of suicide (Benomran and Hassan, 2007). The present finding is in contrast to the results obtained from China, in which the number of females committing suicides is higher compared to males by a 3:1 ratio, making it the

only country in the world where women commit suicide more than men (Eren et al., 2007). This may be due to several factors including violence against women and girls, discrimination in education and employment, the traditional preference for male children to play a crucial role, birth-limitation policies, and other societal factors which contribute to the high female suicide rates (Eren et al., 2007).

The present study has also indicated that all categories of suicide were higher in Non-Kuwaiti population except in deaths due to hanging and firearm which accounted for 51% and 82% respectively, among Kuwaiti population. Results on deaths due to suicide by firearm were for 82% for Kuwaitis and 11% for Non-Kuwaitis. Moreover, Expatriates are not allowed to own a firearm in Kuwait. As explained earlier, Expatriates face obstacles with law regulations while living away from their countries, loneliness and the fact that many have psychological distress due to home sick. Recent epidemiological studies in Saudi Arabia highlighted the fact that psychiatric disorders, loneliness, masculine dominance and home sickness are among the most important interrelated reasons in increasing the likelihood of Expatriates being victims of suicide (Benomran and Hassan, 2007). In contrast, deaths due to firearm injuries were much higher in Kuwaitis and this could be attributed to the fact that there is a high rate of gun possession in the area under study especially after Iraqi invasion of Kuwait in 1990 (Fido, 2009).

5.3 Prevalence of homicide in Kuwait

During the period of this study, homicide constituted 8% of the total unnatural death cases that were referred to the GDCE. These results were

similar to the figures found in Pakistan (Gupta et al., 2004). The highest homicide rate reported in the world was in South Africa (WHO, 2010). Both Columbia and Estonia have a rate of 23 per 100,000 population per year (Hepburn and Hemenway, 2004). This could be due to the high rate of gun possession in these two countries especially since automatic weapons are easily available to the public. Another reason could be a lack of confidence in the Judicial System, in which people tend to take the law into their own hands for settling differences and disputes. The results of the present study also showed that the major cause of homicidal deaths was due to stab wound injuries constituting 38%, followed by death due to firearm injuries. In United State, the most common mode of homicide is death due to firearm injuries in all States except in Staten Island's, where the murderers preferred to use knives or fists instead of guns and bullets (Ambade et al., 2007).

The incidence of homicide reported in this study is low compared to other parts of the world (Gupta et al., 2004). This perhaps, could be attributed to cultural and religious background. The present study has also indicated that 70% of homicidal cases were males and 30% were females. This is similar to that reported in Saudi Arabia (1.8:1) but far lower than that reported in Israel (4:1) (WHO, 2010) (Table 2.7). This can be explained by the structure of Kuwait society and other areas with high male to female ratio where females are primarily confined to their homes and are therefore protected from being involved in violence. This is in contrast to the more industrialized places like USA and Europe where females are more actively involved in workplaces and moreover, they play relatively proactive roles, thereby exposing themselves to a pattern of violence and similarly homicide experienced by males (Ahmet et

al., 2005). In contrast, Russia has a higher homicide rate in female than male accounting for 2:1. The study explained that this is due to the break-up of the Soviet Union and contradictory status of women in Russia which may contribute to these findings (Ambade et al., 2007).

In addition, the present study also indicated that the most vulnerable age of homicides seemed to occur in the age group between 30-39 years of age, with 28% in the third decade of life. Other studies in Qatar also give the highest occurrence of homicide in the same age group, with 40% of all homicides being in the age bracket of 30-39 years (Daradkeh, 1989). Studies in India and Turkey have also reported this age group to be the most vulnerable to commit homicide (Humayun et al., 2009; Brunel et al., 2010). In contrast, studies in U.S.A indicated that the highest rates of homicide were at an earlier age of 10-25 years (Wilson, 2010). This difference could be because individuals start a more independent life at an earlier age in U.S.A, thus exposing them to all sorts of violence, something also reflected by the increasing incidence of juvenile offenders and school violence.

Responsibility for the prevention of violence in the society does not account only on the law enforcement personnel, but also on the population-based attitudes. In addition, other human service agencies must join and assist in preventing primary violence as they have done to prevent other major causes of morbidity and mortality.

5.3.1 Incidence of infanticide in Kuwait

Throughout this study, the results have shown that infanticide was responsible for the least homicidal cases accounting for 3%. Males

outnumbered females constituting 75% and 25%, respectively with a ratio of 5.5:1. Meanwhile, infanticide among expatriates was reported to be 71% more than those reported in indigenous Kuwaitis 29%. The highest incidence of infanticide was reported in Farwania Governorate which is known to house inhabitants from low socioeconomic backgrounds. This may be due to strict legislations in preventing Expatriates to settle with members of their families (International Constitutional Law: Kuwait, 1992). Female house-maids are not allowed to bring their families (spouse and children) when they immigrate to Kuwait. This causes stress which in turn leads to illegitimate relationship and hidden prostitution resulting in unwanted pregnancies. This might force them to criminally terminate the pregnancy or committing infanticide when the illegitimate baby is born. In general, such policies lend to illicit relationships that are considered non-social and illegal (Al-Yacoub, 1995). Similar findings were obtained in Saudi Arabia, Bahrain and United Arab Emirates (Koronfel, 2002; Sauvageau and Yesovitch, 2010). Perhaps, the Kuwaiti Government should reconsider its policy and allow spouses to join their partners. In addition, Kuwaiti officials should be well informed about how such prohibited policies can lead to the prevalence of the existing trend on infant mortality and overall unnatural deaths in the country. Moreover, an in-depth study is urgently needed to identify the root cause of infanticide.

From the results obtained in this study, the analysis of un-natural deaths in Kuwait showed that accidental deaths were the major cause of deaths in Kuwait over the stipulated study period. The present study also identified that homicide, suicide and accidents are highly concentrated in the

overpopulated Farwania Governorate which is mostly inhabited by low income foreign labourers.

This study also compared homicide in different Middle East Countries according to data from the World Health Organization (WHO), most recent available year 2010. The reliability of "homicide" differs among countries. Homicide may or may not include infanticide, assisted suicide or euthanasia. Moreover, they may also be underreported for political reasons. The results shows that high rate of homicide was recorded in Brazil accounting for 23. In contrast, the lowest homicide rate was recorded in United Arab Emirates accounting for 0.92. Kuwait has a homicidal rate of 1.38, considered as the highest rate within the Middle Eastern Countries (WHO, 2010). In addition, the study also compared suicide rates in 2002 for some Middle Eastern Countries according to data from the World Health Organization (WHO). The total rate of suicides is based on the total number of suicides divided by the total population in a country. The data show that in year 2002, the least suicide rate was recorded in Syria accounting for 0.2, followed by Qatar and Jordan, accounted for 0.5 and 1.1, respectively. The highest rate within the Gulf Countries was recorded for Bahrain accounting for 5.4. Whereas, the highest rate in the Middle Eastern Countries was recorded in Israel. Kuwait had a suicidal rate of 3.2 which is considered to be high compared to a neighboring Saudi Arabia with a suicidal rate of 1.2 (WHO, 2010).

5.4 Virtual autopsy study

This part of the study evaluated the effectiveness of radiological imaging techniques MRI and CT scanning in post-mortem examinations in

several cases in order to determine the causes of un-natural deaths. This technique is currently entering the field of forensic medicine (Jackowski et al., 2006; Jackowski et al., 2008). Implementing virtual autopsy technique in forensic medicine not only offers new possibilities in forensic diagnosis, it also provides an excellent learning tool for different professions for medical students, forensic scientists, law enforcement officers and other trainees. The aim of the present study was to evaluate the effectiveness of virtual autopsy technique in identifying injuries in several un-natural cases of deaths. The rationale is to employ this novel autopsy protocol as either an alternative procedure or an additional technique to determine the causes of deaths due to either firearms, blunt force injuries, strangulation, falls from heights and deaths due to RTA. Currently, there is no other authorized morgue in Kuwait for investigating dead persons whether they have died from natural or un-natural causes except for the morgue which is located in the (GDCE). Thus, the (FMD) morgue at GDCE in Kuwait has a very heavy work load, constituting average of 25 autopsy cases on a daily bases. This number involves all cases of RTA, hospital and house deaths which are referred to the FMD. The implementation of this new virtual autopsy technique will facilitate and speed up the autopsy process, thus reducing the efforts made by the Forensic Pathologists in identifying the causes of deaths. Although, the present results were obtained from a small number of subjects, they clearly show that virtual autopsy can reliably aid the identification of the sites of any lethal injuries inflicted on the victims with a fairly reasonable amount of certainty and accuracy.

5.5 Time course effect of temperature and humidity on human DNA quantity at crime scenes in Kuwait

The present study also investigated an important field of forensic science which is DNA analysis (Zubakov et al., 2008). In recent years, the ability to recover and analyse minute amounts of DNA from biological material has revolutionized forensic science (Lee et al., 2011). The DNA science is very unique and at the same time very sensitive, in which the DNA itself can be degraded rapidly by several environmental factors/insults including humidity, microorganisms, chemicals and temperature (Hernandez and Mondala, 2009). Problematic samples containing only degraded DNA are obstacles to the resolution of practical forensic cases that involve trace amounts of evidence under extreme conditions.

Initial studies in the Forensic Laboratories at GDCE in Kuwait demonstrated the trend of DNA degradation in either blood, saliva or semen obtained from human samples in the months of December, January, April and July for 2010 and 2011. The average temperatures during these months represent an average 20-25 °C in November-December, 0-5 °C in January-February, 30-40 °C in April and 40-50 °C in July each year. On rare occasions, the temperature can rise to 55 °C in July and August. These samples were exposed in the present study to the four chosen ranges of temperature (4°C, 24°C, 37°C and 55°C) with their correspondence relative humidity (RH) ranges that were present in the same months of the year, at different time points starting from day 1 to day 28. The climate in Kuwait is considered as a very hot weather with dust storms in the summer, in which the average maximum temperature In July and August is about 45-55 °C and

RH of 41%, whereas, in January, the average temperature is 4 °C with RH around 61%.

The results of this study have shown that when samples of either blood, saliva or semen were exposed to temperature of 4 °C (RH 61%) and 24 °C (RH 58%), DNA survival remained more or less the same indicating that DNA can survive at low temperature for at least 28 days, the duration of the study. The values obtained at day 1 for either blood, saliva or semen samples were more or less similar to the values obtained on the other 27 days. This is probably due to the fact that low temperatures are favorable for DNA preservation and survival (Zubakov et al., 2008). Whereas, the quantity of DNA from both blood and saliva samples incubated at 37 °C decreased slowly with time. The results show that DNA can degrade in saliva and blood samples slowly from day 11 in saliva samples and from day 7 in blood samples and this DNA degradation was probably due in part to microorganism growth or directly to exposure to 37 °C. In contrast, the DNA quantity in semen samples remained more or less the same during the whole period of study at 37°C and RH 55% and this may be due to the fact that the sperms in semen have high levels of glucose, protein and electrolytes (Dissing, 2010). Furthermore, this study investigated the effects of high temperature reaching 55°C and RH of 41% (in the month of July in Kuwait) on DNA survival. The results revealed that, the mean level of DNA quantity in blood samples started to degrade immediately with time until it reached zero at day 14. The same is also true for saliva samples. When temperature was increased to 55°C, DNA quantity in saliva was 32.08 ng/μl on day 1 then this started to degrade with time until it reached 0 on day 17. The results obtained in the present study

showed a significant association between the temperature and the quantity (ng/ μ l) of DNA in each sample over time. Several studies have indicated that such multi-environmental factors/insults as heat, cold, sunlight, bacteria and mould can all induce lethal effects on DNA making it unusable and unstable (Kaiser et al., 2008; Hernandez and Mondala, 2009). There is much evidence in the literature that sperms can survive during adverse conditions including temperature changes (Hemandez and Mondala, 2009). Several studies have also reported that high temperature can denature DNA, causing the double helix to split resulting in 2 single stranded DNA molecules. Another study on DNA degradation indicated that storage of blood samples for 1 month at 50 °C is equivalent to sample stability for 6 months at room temperature (Zubakov et al., 2008). A similar study has been conducted confirming the strong effects of hot environment on DNA survival (Lee et al., 2011). Other previous studies show that if DNA samples have been degraded by any means, the average size of DNA fragments might be reduced to less than 300 bp. This in turn can lead to insufficient DNA template, which eventually can alter the process of identification (Zubakov et al., 2008; Lee et al., 2011).

The result of this study are very interesting for Forensic Specialists who work in the medico-legal field of forensic investigation. The study has clearly indicated that, if a body is discovered after 12-15 days at temperatures of 55°C or above, and samples of blood or saliva were transferred to the DNA laboratory, then there may be no need to analyze the samples for DNA. The high temperature would have already degraded the DNA sample. This in turn can save time for the Forensic Scientist and cost of valuable materials, as well as manpower due to the absence of DNA. At the same time, the study

concluded that semen samples were the best evidence that could resist DNA degradation due to high temperature.

5.6 Conclusions

In conclusion, the results of this study have shown the following:

- I. The results have demonstrated that different causes of un-natural deaths and which claimed a substantial number of lives in Kuwait between 2003-2009, the period of this study and the manner of death was categorized as either homicidal, suicidal or accidental deaths.
- II. The study highlighted that accidental deaths were the major cause of unnatural deaths in Kuwait over the seven years of study. The leading cause of accidental death in Kuwait was road traffic accidents (RTA), followed by falls from heights. Moreover, the study showed that more males died compared to females in all three death categories and non-Kuwaitis ranked first in suicidal deaths while Kuwaiti Citizens ranked first in deaths due to accidents and homicide.
- III. The present study has identified that homicide, suicide and accidents are highly concentrated in the over-populated Farwania Governorate which is mostly inhabited by low income foreign labourers.
- IV. The study show that homicidal deaths are the second mode of un-natural deaths in Kuwait and stab wound injuries are the common mode of homicidal deaths followed by firearm injuries.
- V. The study also investigated the effectiveness of virtual autopsy technique in forensic examination in 30 cases of un-natural deaths including RTA, strangulation, head injuries, drowning and firearm

injuries. The study concluded that the results from virtual autopsy can match closely with the findings of traditional autopsy in identifying the causes of deaths among the 30 victims. Moreover, the results have demonstrated that both radiological imaging (MRI and CT scanning) technique were much superior to the traditional autopsy technique in the detection and examination of the trauma. In addition, the study has identified the track record of injuries in various cases.

- VI. The results also demonstrated that virtual autopsy should be used initially as a protocol to ascertain the causes of deaths of the victims, in order to help the Forensic Pathologists to decide which victims should have an autopsy or to determine whether the autopsy should be limited or complete. It is possible that virtual autopsy may diminish the need for traditional autopsy in cases of RTA and firearm injuries as well as in cases of falls from heights.
- VII. This present study concluded that virtual autopsy technique can enhance quick postmortem investigations of the deceased subjects within 30 to 45 minutes as compared to several hours by traditional autopsy. This helps with rapid screening of each case and moreover, it helps in deciding whether to go ahead in using traditional autopsy. This rapid handling of each case may enhance the burial process of the deceased victim. In the Islam religion, burying the body after death must be done within 24 hours. Moreover, using this technique will ease the pressure on the family in seeing their loved ones in good shape without any scars on the body.

VIII. The present study investigated the effect of various temperature and humidity changes on DNA samples in human saliva, semen and blood. The results show that different environmental temperatures (55, 37, 24 and 4 °C), had no significant effect on DNA degradation on human semen samples. In contrast, exposure of either blood or saliva samples to a temperature of 55 °C and humidity of 41% over 28 days resulted in rapid degradation of DNA with complete degradation within 14-17 days. However, at 37°C DNA degraded slowly in blood and saliva samples reaching significant value after about 20-25 days of exposure. In contrast, exposure of saliva and blood samples to 4°C and 24°C had no significant effect on DNA quantity throughout the whole experimental period. The results show that the best biological sample that can resist environmental insults due to both temperature and humidity is semen.

Together, the results presented in this thesis have provided new recommendations for the Kuwait Ministry of Interior in reducing the number of deaths due to unnatural causes. Moreover, the results also recommend the installation of MRI and CT scanning to support traditional autopsy to investigate the causes of deaths. Finally, the results provided important information on DNA testing in biological samples under harsh environmental conditions.

5.7 Recommendations

From the findings of this study it is possible to make the following recommendations:

- Review and evaluate annually the number of un-natural deaths in Kuwait supported by recommendation for the national campaign for community awareness programme.
- The documentation and registration of all cases of un-natural deaths in Kuwait are essential by law and these have to be done via collaboration among different authorities concerned particularly Kuwait Ministry of Health and Ministry of Interior. The authority must follow the same protocols as WHO when registering epidemiological data.
- Identify the risk groups who are most susceptible to un-natural deaths implementing further improvement of labour laws in Kuwait and applying of strict traffic rules regarding accidental causes. In turn, these can decrease the number of unnatural deaths in Kuwait.
- Undertaking similar studies on patterns of unnatural deaths in other Middle Eastern Countries for comparison.
- Installing of virtual autopsy technique at GDCE in Kuwait. This in turn will make Kuwait a leading country of 'virtopsy' in the Middle East. It is important to validate the legality of 'virtopsy' imaging technique for admissibility of imaging evidence to be accepted in court, through concerned authorities, particularly Ministry of Interior and General Department of Criminal Evidence with implementation of traditional autopsy replacement by virtopsy in certain circumstances.
- Introduce the applications of investigative techniques including endoscopies, laparoscopic and needle core biopsy for pathological specimens.

- Develop special training courses in virtopsy technique for forensic personnel particularly radiologists who are involved in virtopsy technique.
- Introduce proper forensic DNA sampling collections with innovation of new protocols regarding environmental insults on the samples. Such protocols are to be conducted through collaboration between the DNA laboratory and Forensic Medicine Department.

5.10 Scope for Future work

The results from the present study have clearly demonstrated the importance of a similar prospective study in the field of Forensic Medicine as regards to the epidemiology of un-natural deaths, virtopsy and DNA degradation.

- Similar DNA studies need to be performed using another set of genetic markers with the same hypothesis involving single nucleotide polymorphisms (SNPs). SNPs offer several advantages including a higher recovery of data on degraded DNA and possess mutation rates about 100 thousand lower than STR (Huff et al., 2011).
- Undertake similar DNA recovery studies using both hard and soft tissues as bone, teeth, hair, vitreous fluid and muscles and varying the environmental insults to includes humidity, temperature, microorganisms, sun light, chemicals etc.
- The quality and efficiency of the virtual autopsy procedures need to be developed as a designated protocols in handling forensic cases when investigating the causes of deaths.

- To start using the virtopsy technique in all RTA and firearm cases in Kuwait in order to minimize the heavy work load on Forensic Pathologists there by speeding up the process of Forensic investigation.
- Studies should be undertaken on the epidemiology of un-natural deaths in the Middle Eastern countries with over all collaboration with WHO records, comparing the data from one country to another.

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CHAPTER 7

APPENDICES

Appendix 1

Kuwait Law

7.1 Kuwait Law

- **Article number 149 of the law of Kuwait**
Anyone who kills a person on purpose is punished either by the death penalty or is imprisoned for life. A fine of not more than 1,025 KD may be added to the sentence.
- **Article number 150 of the law of Kuwait**
Murder is punishable by death if combined with premeditation.
- **Article number 151 of the law of Kuwait**
Premeditation is deliberate intention to commit the act before it was implemented well in advance in order for effective deliberation in a calm, active surveillance is waiting for his victim in a place guess the relevance of the implementation of the act Were surprisingly.
- **Article number 152 of the law of Kuwait**
Anyone who wounded or beaten, or deliberately gave him narcotics, without intend to kill him, but his action lead to death, will punishable by imprisonment for a term no exceeding ten years and may be added a fine not exceeding ten thousand KD.
- **Article number 153 of the law of Kuwait**
Anyone who caught and surprised by his wife, his sister, his daughter, with a strange man, through intercourse, and killed her or him, will be punished by imprisonment for a term not exceeding 3 years and a fine not exceeding 3 000 or one of these finalities.
- **Article number 154 of the law of Kuwait**
Anyone who kills by mistake or causes the killing of non-intentional, was caused by reckless or negligence or lack of attention or non-observance of the regulations shall be punished by imprisonment for a term not exceeding 3 years and a fine not exceeding 3000 or one of these finalities.
- **Article number 155 of the law of Kuwait**
When a neonate is born alive, he/she is considered as a person that can be killed, without consideration of whether he/she breathes or not, whether

he/she has an independent blood circulation or not, and whether his/her umbilical cord is cut or not.

- **Article number 156 of the law of Kuwait**

Man is not considered that he killed another human being if the victim did not die within one year from the occurrence of the cause of death, and the day is counted from the first time the crime which occurred when another illegal act resulting in death.

- **Article number 157 of the law of Kuwait**

The man is consider for the cause of death for another person, whether in direct reason or indirect reason of death or the only reason for death in these occasions:

- If the offender left the victim injury necessitated surgery or treatment and resulted in death of the victim as long as the process or treatment has been done with the experience and care according to medical principals.
- If the offender left the victim injury and did not cause death, and the victim did all necessary precaution and health issues.
- If the offender let the victim to do something that cause of his death either through violent or by threatening him.
- If the victim is suffering from a disease that leads to his death and the offender by his action accelerates and speeds up death.

Appendix 2

Original Photographs Taken for Victims of Unnatural Deaths

Colour plate I: Suicide cases



Suicidal Hanging



Suicidal Cut Throat

Colour plate 2: Homicide cases



Homicidal Multiple Stab Wounds



Homicidal Multiple Stab Wounds

Colour plate 2: homicide cases



Homicidal Gunshot



Homicidal Strangulation

Colour plate3: Accident cases



Accidental Road Traffic Accident



Accidental Explosive Death