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## Potential Use of Touch DNA in Terrorism Cases: A Report of Four Cases



الاستخدام المحتمل للحمض النووي التلامسي في القضايا الإرهابية: تقرير عن أربع حالات

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### Abstract

The Kingdom of Bahrain is one of the few countries that has significant numbers of terrorist investigations, which has allowed our scientists to develop expertise in the forensic examination of post and pre-blast explosive exhibits. This paper presents a review of forensic investigations into improvised explosive device (IED) cases in the Kingdom of Bahrain from 2011.

A total of four IED investigations were reviewed (i.e. Directionally focused charges (DFC), Directional Focused Fragmentation Charge (DFFC) and Explosively formed penetrator/projectiles (EFP). DNA recovery utilized different collection methods, such as swabbing, tape lifting, wiping and direct cutting of certain separated parts of the IEDs. Samples were extracted and purified with magnetic beads chemistry and quantified. Low copy DNA extracts were subjected to different concentration steps, and DNA extracts were amplified and processed for detection to obtain reliable results. Using the results of the study, we have developed the concept of Forensic DNA Intelligence, which involves the extraction of human cells deposited in low copy number in challenging areas within the evidence which can lead to significant results.

This article will be very useful and informative to assist the forensic community in terrorism cases applications worldwide. Continued efforts must be made to re-evaluate standard operating protocols with empirical studies.

**Keywords:** Forensic Science, IED, Intelligence, DNA, Kingdom of Bahrain, Recovery, Terrorism.

### المستخلص

تعتبر مملكة البحرين واحدة من الدول القليلة التي لديها أعداد كبيرة من التحقيقات الإرهابية، والتي سمحت للأخصائيين بتطوير خبراتهم في الفحص الجنائي للمعروضات المتفجرة قبل وبعد الانفجار. تعرض هذه الورقة مراجعة لتحقيقات جنائية لحالات عبوات ناسفة بدائية في مملكة البحرين تعود لعام 2011.

وتمت مراجعة ما مجموعه أربع تحقيقات حالات عبوات ناسفة بدائية (الشحنات الموجهة بشكل مباشر (DFC)، والشحنات المتجزأة الموجهة بشكل مباشر (DFFC) والمتفجرات المخترقة / المقذوفات (EFP).

وقد استخدمت طرق جمع مختلفة للحصول على الحمض النووي، مثل المسح والأشرطة اللاصقة والمسح والقطع المباشر لبعض الأجزاء المنفصلة من العبوات الناسفة. وتم استخلاص العينات وتنقيتها بواسطة كيمياء الحبيبات المغناطيسية ثم أجري لها تقدير كمي، وتم إخضاع العينات التي تحتوي على نسخ قليلة من الحمض النووي لخطوات تركيز مختلفة، وتم تكثير الحمض النووي المستخلص ومعالجته للكشف عنه وذلك للحصول على موثوقية أكثر.

وباستخدام نتائج هذه الدراسة قمنا بتطوير مفهوم تحقيق للحمض النووي الجنائي. والذي يشمل استخلاص الخلايا البشرية الموجودة في الأدلة المضبوطة بعدد منخفض في مناطق صعبة وتشكل تحدي حقيقي، والذي يمكن أن يؤدي إلى نتائج مهمة تفيد التحقيقات.

وستكون نتائج هذه الورقة العلمية ذات فائدة علمية وتطبيقية لمساعدة خبراء الأدلة الجنائية في الحالات الإرهابية في جميع أنحاء العالم، ويجب مواصلة الجهود في هذا المجال للوصول إلى بروتوكولات التشغيل القياسية والتي تم تطويرها ببيانات من الدراسات التجريبية.

**الكلمات المفتاحية:** علوم الأدلة الجنائية، العبوات الناسفة البدائية، الحمض النووي، مملكة البحرين، الاستخلاص، الإرهاب.

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## 1. Introduction

Most of the traces in crime scenes are comprised of blood spots, semen and sperm secretions, tool marks, trace evidence and human remains. With the advances in forensic technology, this evidence is increasingly recovered from crime scenes, using the correct equipment, processes and expertise [1]. If visible stains are present, a cotton swab can be used to collect and process evidence to assist in solving the case. Since the unrest period in Bahrain in 2011 we have encountered hundreds of terrorism cases involving IEDs (improvised explosive devices), modified devices and phones, detonators and DFCs, as shown in Figure-1.



**Figure 1-** Some of the processed IEDs of different terrorism cases.

These Terrorism cases have caused many casualties, increasing fear in the population and sometimes death among citizens and first responding police officers. In assessing these forensic cases, the scientist requires a different approach and intellectual way of thinking, one must enter the criminal's mind, visualize the way the terrorist bomb maker assembled the IED and how they deployed it. The traditional "simple swabbing" procedure is no longer ef-

fective. Many cases were misreported neither negative or simply had very high contributors in DNA mixture because of the excessive swabbing area. IED are used by terrorists worldwide. Touch DNA from bomb assemblers usually yields low success rate due to the inability locate the areas where DNA has been left [2].

However, as was learned from Locard's exchange principle "every contact leaves a trace" [3], trace in the guise of DNA usually must be there, somewhere and somehow, in the form of hidden touch DNA.

With this principle in mind, we processed hundreds of terrorism cases. The investigations revealed that most suspects who were identified using DNA evidence had significantly more serious criminal histories than those identified through traditional property crime investigations [4].

We have defined Forensic DNA Intelligence as human cells deposited in low copy number in challenging areas within the evidence which leads to significant results. This concept was achieved by processing many terrorism cases with different types of samples collected from 2015 to the present.

Before dealing with securing evidence in terrorist cases, dedicated trained and professional forensic investigation teams must be established with the following skills:

- Crime scene examiner
- DNA expert
- Fingerprint expert
- Ballistics expert
- Forensic electrical engineer
- Forensic photographer

Each forensic specialist has a significant role in identifying the forensic leads which would assist in the identification of how IEDs and components were manufactured or modified and how they were used. To proceed with the forensic collection, several

steps must be carried out to achieve the maximum recovery of results:

- Obtain Good Laboratory Practice (GLP), to protect the evidence from contamination or cross-contamination
- Initiate safety-first protocols: Ensure the IED and/or components (improvised detonators) are made safe and modified devices by personnel with explosive ordnance disposal (EOD) expertise
- Complete a detailed description including overall photos of the IED
- Separate the components of the IED (i.e. wires, devices, detonators, etc.)
- Unseal locked/closed devices
- Take detailed photos after opening the IED device

## 2. Materials and Methods

### 2.1 Samples Collection, Extraction and Normalization

For this study, a total number of four IEDs cases were identified for detailed analysis from various terrorist incidents (i.e. DFC, DFFC and EFP) in the kingdom of Bahrain in year 2015. IEDs components were separated and unsealed to reveal the internal parts for processing along with the external parts of the IEDs. Separation of samples is essential to obtain reliable and beneficial results, and to obtain an accurate number of profiles.

In the first case, we have received homemade IED with claymore and phones which was deployed near to a sport club. Second case it was a series of IEDs containing black devices linked with wires. Third case we have received an explosion case in Muharraq area involving a real IED with claymore and phones. Last case it was a hoax IED containing bundle of tapes and batteries.

Single or double cotton swabs (moistened with DNA grade purified water) were used to collect possible human

cells from handles of cylinders, aluminum foils, internal parts of devices and modified phones [5]. Tape lifts and Single or double nylon swabs (moistened with DNA grade purified water) were used to collect from adhesive samples, such as long sticky tape. Moistened wipes (locally sterilized from Kimwipes™ by UV sterilization) were used to collect touch DNA from long wires and large exhibits. Direct cutting of samples was done to sample small pieces of tape endings, small fuses, left over tissues and wire twists.

Touch DNA was extracted and purified using magnetic beads chemistry (i.e. EZ1® Advanced XL [6] – Qiagen and AutoMate Express™ DNA Extraction System [7] – Thermo fisher Scientific ) with increase time of incubation time in EZ1 to 30 minutes at 56 °C using 485 µl of undiluted G2 buffer and 15 µl PK. Quantification was done thru Investigator Quantiplex Hyres® Kit – Qiagen [8] using a 7500 Real Time System (Applied Biosystems) following the manufacturer protocol. Most of the samples were subjected to various concentration steps using Amicon® Ultra-0.5 centrifugal filters [9] or/and vacuum dry technique [10] (i.e. Concentrator® Plus – Eppendorf), to obtain a reliable quantity for a successful PCR.

### 2.2 Amplification and Detection

DNA extracts were amplified using AmpFℓSTR® Identifier® Plus PCR Amplification Kit - Thermo Fisher Scientific following the manufacturer protocol [11]. Cycles conditions were as the following:

- 95 °C for 11 minutes
- 29 Cycles of:
  - 94 °C Denaturation for 20 Seconds
  - 59 °C Annealing/ extending for 3 minutes
- 60 °C Final Extension for 10 minutes
- Final hold for 4 °C

If required for some degraded amplicons, they were



supplemented with AmpF $\ell$ STR $\text{\textcircled{R}}$  MiniFiler $\text{\textsuperscript{TM}}$  PCR Amplification Kit by following the manufacturer protocol.

Previously genotyped female control with commercially available amplification kit along with negative controls were used during amplification. The amplicons were analyzed by capillary electrophoresis by an ABI 3500xL Genetic Analyzer for fragment length determination. Samples were prepared by adding 1 $\mu$ l of the PCR product or allelic ladder to the corresponding well on the CE plate which contained a mixture of formamide and size standard (8.5  $\mu$ l formamide and 0.5  $\mu$ l GeneScan LIZ600 size standard, v2.0, of Life Technologies).

The plate was prepared and denatured at 95  $^{\circ}$ C for 3 min and then placed on ice for 3 min before loading for capillary electrophoresis. PCR products were separated and detected using POP-4 $\text{\textsuperscript{TM}}$  polymer and an ABI 3500xL Genetic Analyzer (Life Technologies). The data was analyzed using GeneMapper ID-X (GMID-X) software v1.4 (Life Technologies).

### 2.3 Data Analysis

Data was captured by 3500 Series Data Collection v3.1. The raw data was then analyzed using GeneMapper ID-X v1.4. RFU values were obtained through in-house validation of the AmpF $\ell$ STR $\text{\textcircled{R}}$  Identifiler $\text{\textcircled{R}}$  Plus PCR Amplification Kit for 29 cycles. Single source samples were checked into the local-made oracle base Bahrain DNA database containing ~ 50,000 DNA STR profiles. DNA mixtures of 2-3 contributors are supported with Likelihood values using LRmix Studio available online [12, 13].

## 3. Results and Discussion

From 2011 until 2014, most of the DNA samples collected from the external parts of the IEDs were either negative or generated mixtures containing many numbers of

contributors and hence unfit for proper interpretation. During that period only, a small numbers of academic articles had been published regarding the retrieval of touch DNA in terrorism cases, some of these academic papers were focusing upon touch DNA retrieval from methods of transferring IEDs in bags or backpacks [14]. Some of the references applied mtDNA for the exploded IED parts [15]. however, due to the loss of individuality power and the long processing time of the mtDNA testing, this test is used mainly for the purpose of research studies.

In the Kingdom of Bahrain, we received and processed 2 blasts in 2011, 48 IEDs in 2012, 65 IEDs in 2013, 381 IEDs in 2014, 375 IEDs in 2015, 242 IEDs in 2016 and 76 IEDs in 2017 (up to April 2017). We were averaging one IED sample per day (c2014), which highlights the need for the establishment of advanced protocols to properly investigate these terrorism cases. Due to the high number of terrorism incidents, terrorism activities across the Kingdom receive a high priority from the state and are actively investigated by the police and security forces as shown in Table-1, which shows the number of processed IEDs up to April 2017.

**Table 1-** *Estimated number of IEDs in the Kingdom of Bahrain.*

Year	No. of Processed IEDs
2011	2
2012	48
2013	65
2014	381
2015	375
2016	242
2017 (Up to April 2017)	76



In 2015, and with the continuous experience of investigating these IEDs and modified terrorism exhibits, we have followed the above described method using a dedicated and experienced investigative forensics team. Based on our experience with numerous IED devices, we employed our newly developed workflow in 2015 with our dedicated forensic intelligence team. The quantity of extracted touch DNA ranged from 0.025-0.09ng using 7500 Real Time System (Applied Biosystems) prior to DNA concentration and amplification. Any value less than 0.025ng was considered negative and unfit for further analysis. The average RFU values obtained using the GeneMapper ID-X (GMID-X) software v1.4 (Life Technologies) were estimated to be about 400-1000. RFU values of 200-300 were supported with AmpF $\Phi$ STR<sup>®</sup> MiniFiler<sup>™</sup> PCR Amplification Kit, especially for degraded samples.

In the four IED case studies analyzed, two of the cases had negative results from external parts; one DFC case having a claymore, wires and modified phones and the other case of DFFC with modified transmitter devices and wires;

however, while processing the internal parts of the IEDs, it generated a single DNA profile from the modified phone linked with the DFC (piece of facial tissue) and the other from a piece of tape hidden inside the wire cover linked with the DFFC with modified transmitter devices as shown in Table-2.

The other cases processed revealed one EFP case with many contributors to the DNA mixture in a tape bundle and the other DFC case of two single DNA profiles from the tape on the claymore and the other profile generated from the keyboard of the modified phone. On one of the cases, we found a fuse with bite marks and it generated a single male profile and the other DFC case, we processed the internal parts of the phone (where a DNA profile from the keyboard was developed) and found a small tissue paper which generated another single male profile as indicated in Table-3.

We were very successful in generating DNA profiles from items that were presumably touched during manufacture with single source DNA profile or two to three-person

**Table 2-** Description of results obtained from the first and second processed IEDs.

IED Case	Type	Samples Collected	Results	IED Case	Type	Samples Collected	Results
1	DFC	Swabs from external claymore and modified phone and wipe from wires.	-ve	2	DFFC	Swabs from external parts of the modified transmitters and wipes from wires	-ve
		Internal part of phone: small left over facial tissue	+ve (fit)			Direct cut of the discovered hidden tape under the wire cover	+ve (fit)



**Table 3-** Description of results obtained from the third and fourth IEDs.

IED Case	Type	Samples Collected	Results	IED Case	Type	Samples Collected	Results
3	DFC	Direct tape from clay-more and swabs from keyboard of modified phones	+ve (fit)	4	EFP	Swabs from tape bundle	+ve (high mix – unfit)
		Internal part of phone: small left over facial tissue	+ve (fit)			Direct cut of a fuse with bite marks	+ve (fit)

mixtures allowing positive forensic interpretation from matches in the DNA Database. Establishing the principle of Forensic DNA Intelligence, has led us to conclude that DNA evidence is central to understanding both who was involved but also what their role was in the design and manufacture of the IED by separating the components of IEDs, unsealing electrical devices and focusing upon modified items:

- Is DNA found in internal or external parts of the evidence?
- Is the terrorist a distributor?
- Is the terrorist an assembler of IED?
- Is the terrorist an electrician?

Once we learn about the role of the suspect (s), it will assist the criminal investigators in their investigation. Where the forensic investigation secures numbers of single unknown profiles, this also assists the investigation in estimating the member size of the terrorist group or organization. Many IEDs recovered from crime scenes are complex hoax devices; however, these devices cause the same impact of fear among citizens. Nevertheless, same procedure

must be followed for any type of IEDs received as many of the hoax IEDs are used in conjunction with live explosive devices. Whilst police responders are dealing with a hoax IED, the live device detonated as a secondary incident, designed to kill the police first responders.

In addition, this article will be very useful and informative to assist the forensic community in terrorism case applications worldwide. We have developed many DNA profiles, generated from terrorism related exhibits. Some of them are still unknown to the police investigators while others are arrested and referred to Bahrain Law Courts. The findings of this study emphasize the need to continuously re-evaluate standard operating protocols with empirical studies for these types of cases.

#### **Conflict of interest**

None

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