Science & Justice 59 (2019) 580–584

Contents lists available at ScienceDirect

Science & Justice

journal homepage: www.elsevier.com/locate/scijus

Technical Note

Collection and direct amplification methods using the GlobalFiler™ kit for DNA recovered from common pipe bomb substrates

Esiri Tasker[a,*], Carrie Mayes[a], Bobby LaRue[a], Sheree Hughes-Stamm[a,b]

[a] Department of Forensic Science, Sam Houston State University, Huntsville, TX, USA
[b] School of Biomedical Sciences, University of Queensland, Brisbane, QLD, Australia

ARTICLE INFO

Keywords:
Forensic science
Forensic biology
"Touch" DNA
Polymerase chain reaction
Direct amplification
Short tandem repeats

ABSTRACT

When analyzing DNA from exploded pipe bombs, quantities are often in trace amounts, making DNA typing extremely difficult. Amplifying minute amounts of DNA can cause stochastic effects resulting in partial or uninterpretable profiles. Therefore, the initial DNA collection from "touch" evidence must be optimized to maximize the amount of DNA available for analysis.

This proof-of-concept study evaluated two different swab types with two direct amplification strategies to identify the most effective method for recovering DNA from common pipe bomb substrates. PVC and steel pipes, electrical tape, and copper wire spiked with epithelial cells were swabbed with cotton or microFLOQ® Direct Swabs and amplified directly or via a pre-treatment prior to STR amplification.

Not only was the microFLOQ® Direct Swab protocol the quickest method with the least risk of contamination, but in combination with direct amplification, the microFLOQ® Direct Swabs also generated the most complete STR profiles.

1. Introduction

Pipe bombs are explosive devices that are relatively easy to construct. As a result, they are often the chosen method by domestic and international terrorists to cause harm and destruction in communities [1]. After an explosive event, identifying a suspect in a timely manner is critical. While there are several approaches to identify a potential suspect [1,2], few methods are as individualizing as DNA identification. Unfortunately, any trace DNA left on post-blast fragments is unlikely to be high in quantity or quality, which can impede downstream DNA analysis. In addition, DNA is often lost during DNA processing (collection, extraction, and quantification), further decreasing the likelihood of producing a quality DNA profile for generating an identification or investigative lead [3].

Newer and alternate sample collection and amplification methods may be more effective than traditional casework protocols when processing "touch" and other challenging DNA samples, and therefore warrant investigation. Direct amplification of DNA in a forensic context was first demonstrated in 2010 [4]. Since then, several studies have explored various direct amplification methods; however, few studies have applied this approach to substrates commonly used to manufacture improvised explosives [5–7]. Direct amplification bypasses the DNA extraction and quantification steps by placing the collected sample directly into the PCR [4,8,9]. This reduces DNA loss, increases the amount of starting template available for amplification, and increases the likelihood of generating more complete profiles [9]. This approach holds great potential for crimes involving weapons and improvised explosives, as high quantities of DNA are unlikely to be recovered. However, direct amplification from the swab does not allow for DNA quantitation and may also consume the entire sample depending on the type of swab used.

STR kits specifically designed for direct amplification include the GlobalFiler® Express (Thermo Fisher Scientific), PowerPlex® 18D (Promega, Madison, WI), and the Investigator® 24plex GO! (Qiagen®, Valencia, CA). However, these kits are intended for processing blood or buccal reference samples with ample amounts of high quality DNA [10]. Some traditional casework STR kits also provide alternate protocols for direct PCR [11,12]. However, many laboratories may prefer to use the same STR kit for all samples (casework and reference). Therefore, using a standard case-working STR kit for all sample types (including a direct amplification approach) may be more practical and cost effective for forensic laboratories by eliminating the need to purchase and validate additional specialty STR kits (i.e. direct amplification kits).

* Corresponding author at: Department of Forensic Science, College of Criminal Justice, Sam Houston State University, Chemistry and Forensic Science Bldg., 1003 Bowers Blvd., Office 221H, Huntsville, TX 77340-2525, USA.
E-mail address: ett006@shsu.edu (E. Tasker).

https://doi.org/10.1016/j.scijus.2019.06.002
Received 9 February 2019; Received in revised form 6 May 2019; Accepted 2 June 2019

While cotton swabs are commonly used as collection devices and undergo a full DNA extraction prior to amplification, microFLOQ® Direct Swabs (Copan Italia, Bresica, Italy) were developed to be placed directly into a PCR to decrease the likelihood of DNA loss and contamination [13]. Unlike the tightly woven fibers of cotton swabs, the small swab head consists of nylon-flocked fibers treated with a proprietary lysing agent which helps release DNA into solution [14]. While this potentially increases the risk of amplifying impurities and inhibitors (by eliminating purification steps), sample processing time is shortened as fewer steps are employed. In addition, these swabs can be used in a subsampling manner to triage evidence and reduce sample consumption [13]. There are few studies that have examined the utility of microFLOQ® Direct swabs [13,15–17]. However, none to date have compared the performance of the swabs to what is commonly used in forensic laboratories.

This study investigates two different collection devices and two direct PCR processing methods which aim to decrease DNA loss and capture more genetic information from handled items. Common pipe bomb substrates were spiked with controlled amounts buccal cells and collected with cotton or microFLOQ® Direct Swabs. Traditional DNA workflows and two direct amplification methods were tested, and completeness of profiles was used to evaluate overall success.

2. Materials and methods

2.1. Epithelial cell suspension preparation

To control the number of cells being deposited experimentally, an epithelial cell suspension was prepared from buccal swabs obtained from a single male donor in accordance with Sam Houston State University International Review Board Guidelines (#2016-09-31948). Using a modified method from University of Illinois, Chicago (personal communication), a single buccal swab was placed into a 2 mL microcentrifuge tube with 1 mL of 1× PBS (VWR, Radnor, PA). The tube was incubated at room temperature (24 °C) with shaking at 700 rpm for 30 min. After incubation, the tube was inverted several times and centrifuged in a spin basket at maximum speed (approx. 13,000 ×*g*) for 1 min to pellet the cells. The swab and spin basket were discarded, and the liquid was decanted. Then, 1 mL of fresh 1× PBS was added and the tube inverted to resuspend the cells.

2.2. Pipe bomb substrate preparation

Four common pipe bomb substrates were selected: PVC pipe, galvanized steel pipe, electrical tape, and copper wire. PVC and galvanized steel pipes were washed with Alconox® detergent (Alconox, Inc., White Plains, NY) and sterilized using 20% bleach and 70% reagent alcohol. The ends of insulated copper wire were stripped (approx. 1 cm) and cleaned with 70% reagent alcohol. All four substrates were then UV-treated in a cross-linker (UVP, LLC., Upland, CA) for 20 min.

Ten aliquots (10 μL each) of diluted cell suspension (approx. 200 pg or 30 cells) were placed onto the surface of each substrate (adhesive side for electrical tape) and dried overnight. Dried cell spots were swabbed for 30 s with either a cotton swab (Puritan, Guilford, ME) treated with 30 μL of 2% SDS or a microFLOQ® Direct Swab moistened with 1 μL of dH_20. Swab blanks were also taken from each substrate and processed through each method to ensure that cleaning was sufficient. All swabs were dried for at least 24 h after swabbing before proceeding with extraction or amplification. Cotton swabs were air dried and microFLOQ® Direct swabs were placed back into the transport tube.

2.3. Sample processing

2.3.1. Traditional sample processing (controls) and direct amplification

For traditional DNA processing, cotton swabs (N = 10 per substrate) were extracted using the PrepFiler Express™ Forensic DNA Extraction Kit (Thermo Fisher Scientific) on the AutoMate Express™ Forensic DNA Extraction Instrument (Thermo Fisher Scientific) using the "body fluids on swabs (buccal and other body fluids)" protocol [18]. In addition, a neat epithelial cell suspension (30 μL) was extracted in triplicate to determine the degree of DNA loss.

2.4. DNA quantification

To estimate cell concentration of the suspension, a hemocytometer was used to visualize the cells using a 1:1 ratio of suspension and methylene blue to ensure intact cells are present, and cell counting was performed using standard counting methods [19]. The DNA concentration was also determined using the QuantiFiler® Trio DNA Quantification Kit (Thermo Fisher Scientific, Waltham, MA). Triplicate aliquots of neat suspension (2 μL) was added directly to the qPCR mix and rigorously vortexed to shear the cells and release the DNA. The cell suspension was then diluted based on the qPCR quantitation values to approximately 0.04 ng/μL (6 cells/μL) and quantified using the previously described method to verify the DNA concentration.

The QuantiFiler® Trio DNA Quantification Kit was also used to assess DNA concentration, DNA degradation, and inhibition for the extracted swabs [20]. Replicates were averaged, and recovery was calculated by dividing the average DNA concentration of each substrate by the concentration of the epithelial cell suspension (0.04 ng/μL). The maximum volume of extract (15 μL) was then added to the GlobalFiler® reaction mix for amplification.

2.5. Direct amplification

For direct amplification, a small portion of each cotton swab (approx. 1.2 mm) was sampled using a Whatman™ Uni-Core™ Punch (Fisher Scientific, Pittsburg, PA) or one microFLOQ® Direct Swab head was added directly to a 0.2 mL PCR tube containing 25 μL of GlobalFiler® reaction mix.

A pretreatment method for direct PCR was also explored to prevent consuming the entire sample and allow for DNA quantification and resampling if necessary. This is particularly important for the microFLOQ® Direct Swabs as the entire swab is used for amplification via the recommended protocol. Swab pretreatment was adapted from the GlobalFiler® Express direct amplification kit (Thermo Fisher Scientific) [21]. Swabs were incubated in either 400 μL of TE at 90 °C (cotton swabs) or in 40 μL of TE at room temperature (microFLOQ® Direct Swabs) for 20 min. The maximum volume of lysate (15 μL) was then directly added to 10 μL of GlobalFiler® master mix (25 μL total reaction volume).

2.6. DNA amplification and analysis

All samples were amplified using the GlobalFiler® PCR Amplification Kit (Thermo Fisher Scientific) using the standard 29 cycles on a ProFlex™ PCR System [22]. Amplified products were separated and detected on a 3500 Genetic Analyzer (Thermo Fisher Scientific) using a 36 cm capillary array with POP-4™ polymer (Thermo Fisher Scientific), and a 5 s injection time at 1.2 kV. STR analysis was conducted using GeneMapper® ID-X v 1.4 (Thermo Fisher Scientific) and in-house Excel workbooks with validated analytical and stochastic thresholds of 150 and 600 RFUs, respectively. Non-parametric univariate ANOVA with a Games-Howell post hoc and Welch t-tests were performed in RStudio [23] and graphed in Microsoft Excel. A confidence level of 0.05 ($p < 0.05$ = "*") was used for all statistical analyses.

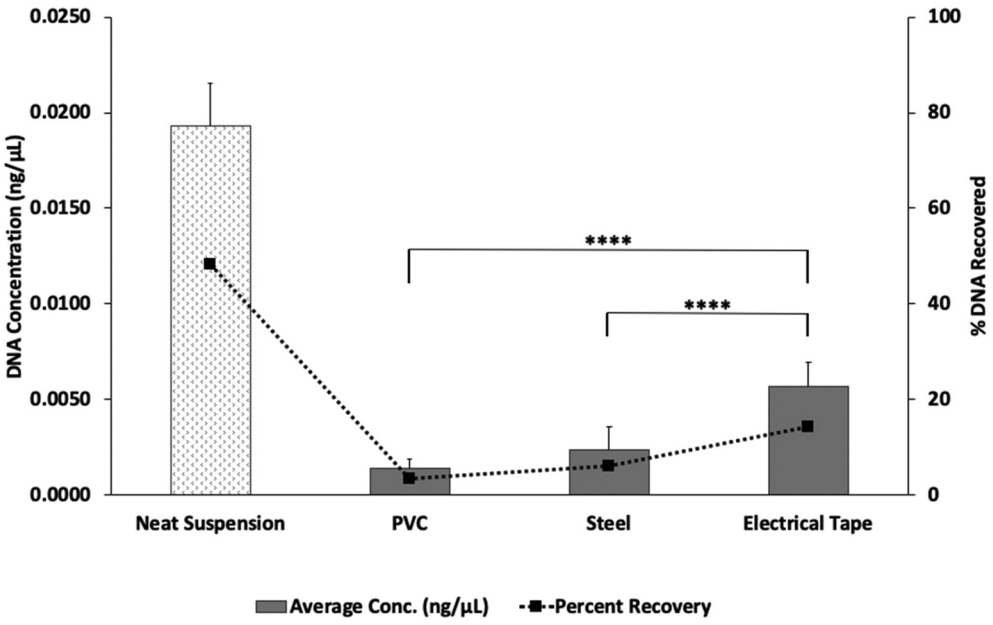

Fig. 1. Average DNA concentration for extracted cotton controls (N = 10 per substrate) and neat cell suspension (N = 3) extracted using the PrepFiler *Express*™ DNA Extraction Kit on the AutoMate *Express*™ extraction instrument. Statistical significance determined by ANOVA and Games-Howell post hoc test. Error bars represent standard deviation.

3. Results

3.1. Traditional processing – cotton controls

Neat cell suspension extracts (controls) and cotton swab extracts (N = 10 per substrate) were quantified to determine DNA concentration and DNA quality. The Quantifiler® Trio kit did not indicate the presence of PCR inhibitors, and no alleles were observed in STR profiles generated from all copper wire samples. Therefore, the copper substrate was removed from the study.

The percent DNA recovery was low for the remaining three substrate types (Fig. 1). More than half (52%) of the DNA from the neat epithelial cell suspension (control) was lost during the full extraction process. As may be expected, DNA loss was even more severe when collected from the PVC pipe, steel pipe, and electrical tape using cotton swabs, which resulted in 3%, 6%, and 14% DNA recovery, respectively. A one-way ANOVA and a Games-Howell post hoc test revealed that electrical tapes resulted in significantly higher DNA quantitation values compared to PVC ($p < 0.0001$) and steel pipes ($p < 0.0001$). No significant difference in DNA concentration between PVC and steel pipes was observed.

The STR profiles produced from all substrates resulted in less than half of the expected number of alleles being called, with electrical tape having the most alleles reported (Fig. 2). Not only did electrical tape samples yield the highest DNA concentrations, but also the most complete STR profiles and highest peak heights compared to PVC ($p = 0.0001$) and steel pipes ($p < 0.01$). No alleles were observed in any of the substrate negative controls, and no spurious drop-in alleles were observed in the experimental samples or any no template controls.

3.2. Comparison of sample processing strategies with both swab types

Direct PCR and swab pretreatment strategies were tested with both cotton and microFLOQ® Direct Swabs to determine if sample processing time could be reduced while preserving (or increasing) the number of reportable alleles. Each swab type was processed with both methods and STR results were compared. As with the cotton control samples, all copper wire samples failed to amplify with both the direct and pretreatment methods and therefore were also removed from the study.

When DNA was collected using cotton swabs from all substrates, more alleles were recovered when swabs underwent a full extraction compared to the direct PCR and pretreatment methods (Suppl. Fig. 2). Direct amplification of cotton swabs collected from PVC pipes failed to produce any STR results; therefore, only full automated extraction and swab pretreatment methods were compared. When compared to the extracted cotton swabs, a Welch *t*-test revealed significant differences for direct PCR and swab pretreatment ($p < 0.05$) for PVC pipes. A Games-Howell post hoc test also revealed significant differences for electrical tape ($p = 0.0001$), specifically between extraction and direct PCR, and extraction and swab pretreatment ($p < 0.001$ for both). Though the full DNA extraction provided more alleles for DNA from steel pipes than the other processing method, this increase was not statistically significant.

Overall, samples collected using the microFLOQ® Direct Swabs showed the most complete STR profiles for the three substrate types regardless of the DNA processing method used ($p < 0.0001$) (Fig. 3), with direct amplification of the microFLOQ® Direct Swabs being the most successful method. When microFLOQ® Direct Swabs were placed directly into the PCR amplification more alleles were reported for the PVC ($p < 0.001$) and steel ($p < 0.0001$) pipes compared to pretreatment, with comparable results for the electrical tape. As with the cotton swabs, pretreating the microFLOQ® Direct Swabs diluted the DNA concentration and therefore reduced the amount of DNA for amplification.

Directly amplifying microFLOQ® Direct Swabs from electrical tape did not result in a significant difference in reportable alleles compared to the pretreatment method. It should be noted that nylon fibers from the swab head adhered and remained attached to the adhesive surface of the tape after collection, but this did not seem to impact STR results. Average peak heights were the highest when microFLOQ® Direct Swabs were directly amplified ($p < 0.01$; Suppl. Fig. 3). The success with electrical tape could be attributed to the adhesive surface with the epithelial cells likely clinging to the adhesive that was then collected by the swab head.

4. Discussion

Large quantities of DNA was lost using traditional extraction methods. This loss was not unexpected, as other studies have documented significant DNA loss after DNA extraction [3,24,25]. Specifically, one study also observed sufficient DNA loss on the AutoMate Express™ for trace DNA quantities below 0.05 ng/µL [26]. Despite the expected DNA loss, in our study more DNA was recovered, and more alleles were reported from the electrical tape samples compared to the

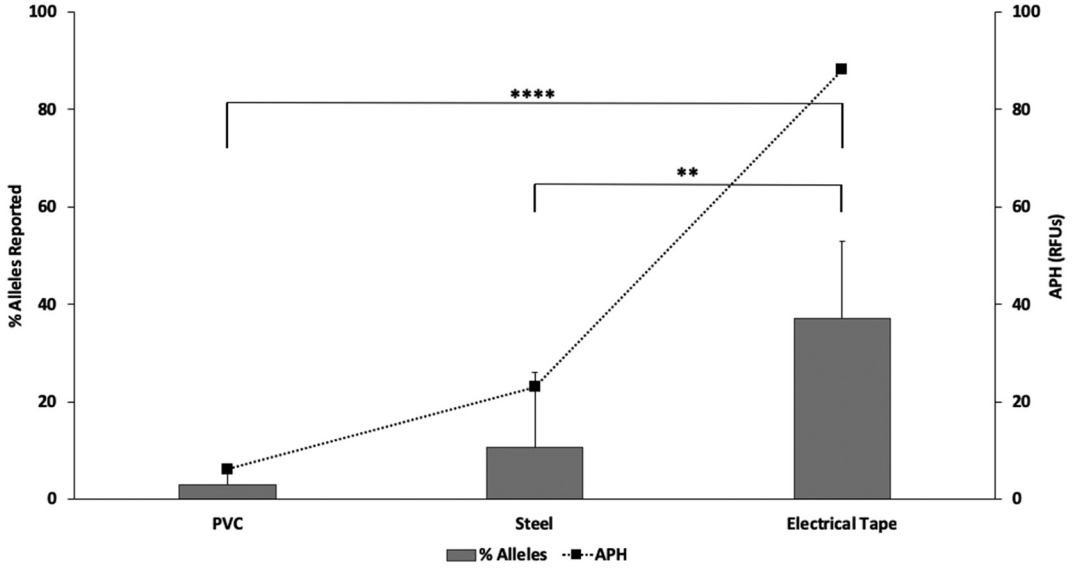

Fig. 2. Average percent of alleles reported and average peak height for control extracts (N = 10) per substrate. Statistical significances determined by ANOVA and Games-Howell post hoc test. Error bars represent standard deviation.

PVC and steel pipes. These data are consistent with a study by Mattayat et al. also demonstrating that electrical tape is a viable source of DNA from improvised explosives [27]. These results also suggest that electrical tape may be a rich source of DNA when touched, and therefore could be prioritized in relevant forensic casework. However, the relatively low DNA recovery and STR success rates overall demonstrate that these traditional DNA extraction methods may not be the best approach for "touch" samples.

A similar study conducted by Phetpeng et al. explored the effectiveness of various DNA collection devices and moistening agents to recovery DNA from pipe bomb substrates, including electrical tape [28]. However, the overall DNA recovery was relatively low for electrical tape (0–58%) regardless of the collection device or moistening agent used, which the authors attributed to cells (from buffy coat) adhering to the substrate. While that study did investigate several cotton collectors and one nylon swab, all of the collection devices underwent traditional DNA processing. Therefore, it is possible that their low recovery was also attributed to substantial DNA lost during extraction in addition to cells adhering to the electrical tape. This may also explain the reduced recovery for DNA collected from electrical tape when extracted via traditional methods (37%).

Direct amplification with microFLOQ® Direct Swabs produced the most complete STR profiles of all the methods tested. While studies utilizing these swabs are limited, high STR success has been observed [13,17]. However, both direct amplification and pretreatment methods with cotton swabs were less successful. While our swabbing technique ensured that only the tip of the swabs was used for collection, it is possible that the small portion (approx. 1.2 mm) sampled for direct

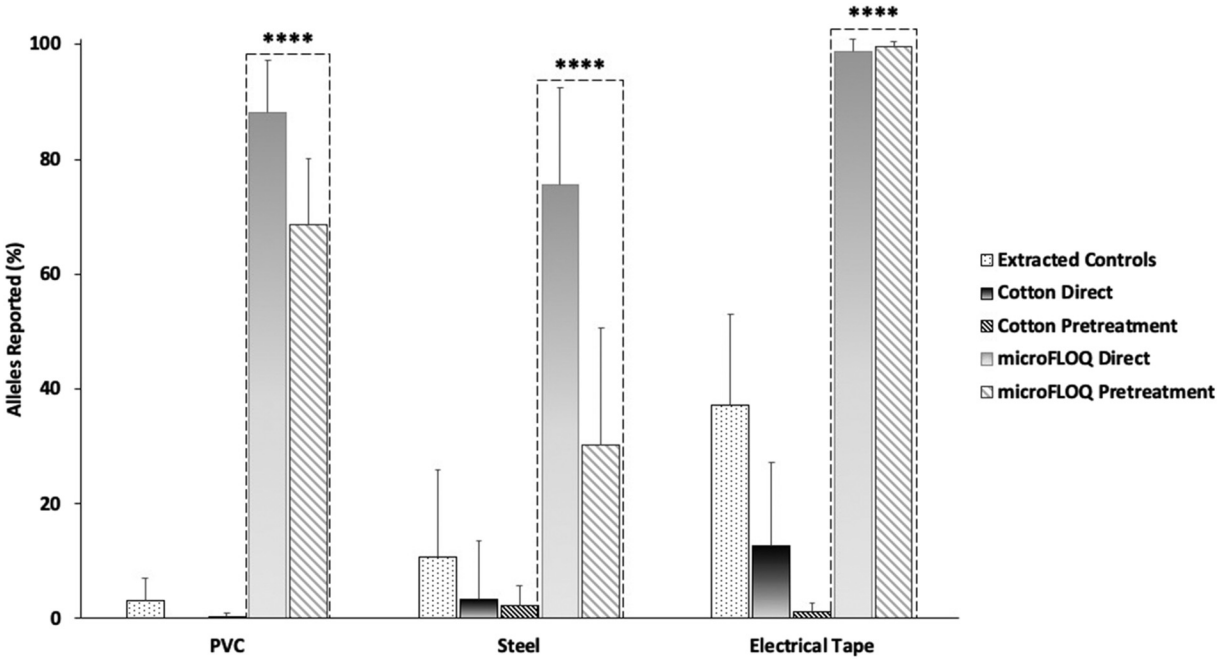

Fig. 3. Comparison of automated extraction, direct amplification, and pretreatment methods for both cotton and microFLOQ® Direct Swabs (N = 10 per method and substrate). Controls were cotton swabs extracted using the PrepFiler *Express*™ Forensic DNA Extraction Kit (N = 10 per substrate). Statistical significance determined by ANOVA and Games-Howell post hoc test. Error bars represent standard deviation.

amplification did not contain a sufficient amount of DNA. Although only a small portion was taken per swab in this study, the swabs could be re-sampled in an attempt to produce more complete profiles. For the pretreatment method, 400 μL of the incubation liquid was needed to adequately cover the swab head. This large volume also diluted the DNA recovered. Additional steps such as using a spin basket or concentrating the DNA, could be taken in order increase the amount of DNA available for amplification. However, this added step will increase processing time, any PCR inhibitors, and the risk of contamination.

DNA collected from copper wires failed to produce results regardless of the method used. Although the QuantiFiler® Trio kit was used to access the quality of DNA extracts, it failed to indicate any inhibition for the copper wire extracts. Copper is a known PCR inhibitor [29] and was likely the cause of the failed profiles for all samples. However, considering 2 μL of extract is used for quantitation and 15 μL is amplified for STR typing, the level of inhibition was likely high enough in the STR reaction to prevent amplification despite not being detected during quantitation. Similar results were observed in other studies [28,30] which further supports that PCR inhibition is a reasonable explanation for the results observed in this study.

5. Conclusion

The results of this study provide further support that traditional DNA processing may not be the most effective method for processing "touch" and low-template DNA samples for STR analysis. Collection of biological evidence from touched items using microFLOQ® Direct Swabs and processed via direct amplification resulted in the most complete STR profiles regardless of IED substrate type (copper wires excluded). In addition, direct amplification was successful using the routine casework GlobalFiler® kit, thereby eliminating the need to use a specific direct PCR kit for this type of evidence. This study also demonstrates that this faster and more streamlined method may be advantageous for processing challenging DNA samples, including those recovered from pipe bomb fragments and other weapons.

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scijus.2019.06.002.

Funding

None.

Disclaimers

None.

Declarations of competing interests

None.

Acknowledgements

The authors would like to acknowledge Thermo Fisher Scientific for providing kits and reagents, and Copan Italia for providing the microFLOQ® direct swabs.

References

[1] T.W. Bille, C. Cromartie, M. Farr, Effects of cyanoacrylate fuming, time after recovery, and location of biological material on the recovery and analysis of DNA from post-blast pipe bomb fragments, J. Forensic Sci. 54 (2009) 1059–1067.
[2] A. Berti, F. Barni, A. Virgili, C. Colozza, F. Maiorino, M. Tocca, The recovery of DNA profiles from saliva and touch evidences after postal bomb explosion, Forensic Sci. Int. Genet. Suppl. Ser. 3 (2011) e471–e472.
[3] R.A.H. van Oorschot, D.G. Phelan, S. Furlong, G.M. Scarfo, N.L. Holding, M.J. Cummins, Are you collecting all the available DNA from touched objects? Int. Congr. Ser. 1239 (2003) 803–807.
[4] A. Linacre, V. Pekarek, Y.C. Swaran, S.S. Tobe, Generation of DNA profiles from fabrics without DNA extraction, Forensic Sci. Int. Genet. 4 (2010) 137–141.
[5] P. Tonkrongjun, P. Thanakiatkrai, S. Phetpeng, W. Asawutmangkul, S. Sotthibandhu, T. Kitpipit, Touch DNA localization and direct PCR: an improved workflow for STR typing from improvise explosive devices, Forensic Sci. Int. Genet. Suppl. Ser. 6 (2017) e610–e612.
[6] B. Martin, R. Blackie, D. Taylor, A. Linacre, DNA profiles generated from a range of touched sample types, Forensic Sci. Int. Genet. 36 (2018) 13–19.
[7] S. Dangsriwan, P. Thanakiatkrai, W. Asawutmangkul, S. Phetpeng, T. Kitpipit, Direct PCR improves STR profiles from substrate of improvised explosive device, Forensic Sci. Int. Genet. Suppl. Ser. 6 (2017) e507–e509.
[8] Y.C. Swaran, L. Welch, A comparison between direct PCR and extraction to generate DNA profiles from samples retrieved from various substrates, Forensic Sci. Int. Genet. 6 (2012) 407–412.
[9] J.E. Templeton, A. Linacre, DNA profiles from fingermarks, BioTechniques 57 (2014) 259–266.
[10] S. Flores, J. Sun, J. King, B. Budowle, Internal validation of the GlobalFiler™ Express PCR Amplification Kit for the direct amplification of reference DNA samples on a high-throughput automated workflow, Forensic Sci. Int. Genet. 10 (2014) 33–39.
[11] S.E. Cavanaugh, A.S. Bathrick, Direct PCR amplification of forensic touch and other challenging DNA samples: a review, Forensic Sci. Int. Genet. 32 (2018) 40–49.
[12] Promega, Powerplex® Fusion 6c System For Use On The Applied Biosystems® Genetic Analyzers, in (2017).
[13] A. Ambers, R. Wiley, N. Novroski, B. Budowle, Direct PCR amplification of DNA from human bloodstains, saliva, and touch samples collected with microFLOQ® swabs, Forensic Sci. Int. Genet. 32 (2018) 80–87.
[14] R.J. Brownlow, K.E. Dagnall, C.E. Ames, A comparison of DNA collection and retrieval from two swab types (cotton and nylon flocked swab) when processed using three QIAGEN extraction methods, J. Forensic Sci. 57 (2012) 713–717.
[15] K.W.Y. Chong, Y. Wong, B.K. Ng, W.S.H. Lim, A.R. Rosli, C.K.C. Syn, A practical study on direct PCR amplification using the GlobalFiler™ PCR Amplification Kit on human bloodstains collected with microFLOQ™ Direct swabs, Forensic Sci. Int. 300 (2019) 43–50.
[16] S. Hubac, M. Mori, R. Dominguez, G. Pierrini, GendSAG/MicroFLOQ™: an innovative DNA collection tool on crime scene for direct and fast analysis, International Symposium on Human Identification, Minneapolis, Minnesota, 2016.
[17] M. Habib, A. Pierre-Noel, F. Fogt, Z. Budimlija, M. Prinz, Direct amplification of biological evidence and DVI samples using the Qiagen Investigator 24plex GO! Kit, Forensic Sci. Int. Genet. Suppl. Ser. 6 (2017) e208–e210.
[18] Thermo Fisher Scientific, PrepFiler Express™ and PrepFiler Express BTA™ Forensic DNA Extraction Kits User Guide, (2017).
[19] K.S. Louis, A.C. Siegel, Cell viability analysis using trypan blue: manual and automated methods, in: M. Stoddart (Ed.), Mammalian Cell Viability: Methods and Protocols, Humana Press, 2011, pp. 7–12.
[20] Thermo Fisher Scientific, Quantifiler® HP and Trio DNA Quantification Kits User Guide, (2015).
[21] Thermo Fisher Scientific, GlobalFiler™ Express PCR Amplification Kit User Guide, (2016).
[22] Thermo Fisher Scientific, GlobalFiler™ PCR Amplification Kit User Guide, (2015).
[23] RStudio Team, RStudio: Integrated Development for R, RStudio, Inc, Boston, MA, 2015http://www.rstudio.com/.
[24] A. Barbaro, N. Staiti, P. Cormaci, L. Saravo, DNA profiling by different extraction methods, Int Congr Ser, Elsevier, 2004, pp. 562–564.
[25] R. Kishore, W. Reef Hardy, V.J. Anderson, N.A. Sanchez, M.R. Buoncristiani, Optimization of DNA extraction from low-yield and degraded samples using the BioRobot® EZ1 and BioRobot® M48, J. Forensic Sci. 51 (2006) 1055–1061.
[26] M. Stangegaard, B.B. Hjort, T.N. Hansen, A. Hoflund, H.S. Mogensen, A.J. Hansen, N. Morling, Automated extraction of DNA from biological stains on fabric from crime cases. A comparison of a manual and three automated methods, Forensic Sci. Int. Genet. 7 (2013) 384–388.
[27] D. Mattayat, T. Kitpipit, S. Phetpeng, W. Asawutmangkul, P. Thanakiatkrai, Comparative performance of AmpFLSTR® Identifiler® Plus PCR amplification kit and QIAGEN® Investigator® IDplex Plus kit, Sci. Justice 56 (2016) 468–474.
[28] S. Phetpeng, T. Kitpipit, P. Thanakiatkrai, Systematic study for DNA recovery and profiling from common IED substrates: from laboratory to casework, Forensic Sci. Int. Genet. 17 (2015) 7.
[29] C.D. Matheson, T.E. Marion, S. Hayter, N. Esau, R. Fratpietro, K.K. Vernon, Removal of metal ion inhibition encountered during DNA extraction and amplification of copper-preserved archaeological bone using size exclusion chromatography, Am. J. Phys. Anthropol. 140 (2009) 384–391.
[30] L.I. Moreno, B.R. McCord, Understanding metal inhibition: the effect of copper (Cu2+) on DNA containing samples, Forensic Chem. 4 (2017) 89–95.