DRUG PARAPHERNALIA

The Evaluation of Drug Paraphernalia—Recovery and Analysis of DNA from Crack Pipes Using the PowerPlex® 16 System

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INTRODUCTION

The Mansfield Division of Police Laboratory supports the largest decentralized drug task force in the state of Ohio. During an investigation, a suspected drug dealer fled a vehicle, leaving behind a bag of "crack" cocaine. Officers observed the suspect "spit out" the sample during the foot pursuit. The suspect was not apprehended, and the drug sample was delivered to the laboratory for analysis. Based on the circumstances of the offense, DNA analysis using the PowerPlex® 16 System (1) was initiated on the plastic bag. A complete DNA profile was developed and uploaded to CODIS. The profile resulted in a hit to a Minnesota Bureau of Criminal Apprehension Laboratory convicted offender database. The results of this analysis were not a surprise based on the DNA source, timely collection and proper packaging of the sample. This case initiated a new source of potential requests and additional questions with respect to drug paraphernalia.

The goals of this project are to develop an extraction process to recover DNA from crack pipes, determine if CODIS-eligible profiles can be generated from crack pipes, and evaluate the results to determine if this process is worth the time and resources.

MATERIALS AND METHODS

We evaluated 34 crack pipes, which were submitted as drug paraphernalia evidence. The submitted samples were analyzed for the presence of cocaine residue by washing the inside of the pipe with ethyl alcohol, followed by UV and gas chromatography/mass spectrometry analyses.

DNA extraction was then performed using a modified organic extraction procedure. The crack pipe was placed in a 16 × 100 mm culture tube with the oral end in 1 ml of Stain Extraction Buffer [10 mM Tris-HCl (pH 7.5), 10 mM EDTA (pH 8.0), 100 mM NaCl, 2% SDS]. Proteinase K (25 µl of a 20 mg/ml stock) and dithiothreitol (12.5 µl of a 62 mg/ml stock) were added, and the sample was mixed on an orbital shaker at 100 rpm for 1 hour. The extraction buffer was removed and incubated at 56 °C for a minimum of 2 hours. After incubation, the buffer was extracted with 1 ml of UltraPure™ phenol:chloroform:isoamyl alcohol (Invitrogen Cat.# 15593-031; 2) followed by Microcon® YM-100 (Millipore Corporation) concentration and recovery in 20–100 µl of nuclease-free water.

DNA samples were quantified by real-time PCR using the Applied Biosystems 7500 Real Time PCR System and Quantifiler® Human DNA Quantification Kit (3). As required, DNA was diluted with nuclease-free water to 0.4 ng/µl. DNA (2.5 µl) was amplified using the PowerPlex® 16 System[(d–g)] and a GeneAmp® 2400 PCR System using a 30-cycle run as directed by the manufacturer. A target range of 0.5–1.0 ng is required for amplification, so for samples with a lower DNA concentration, 3–5 µl of extract was added to bring the DNA amount within this range.

Table 1. Quantitation and Amplification Results.

Sample Name	Total DNA Recovered	Result
CP22	2.28 ng	Mixture profile, 10+ loci
CP37	2.65 ng	Partial profile (not CODIS-eligible)
CP28	4.20 ng	Complete profile
CP35	5.31 ng	Partial profile (not CODIS-eligible)
CP25	7.70 ng	Partial profile (not CODIS-eligible)
CP36	7.71 ng	Partial profile, 10+ loci
CP31	8.34 ng	Mixture profile (complete)
CP15	8.95 ng	Mixture profile (complete)
CP11	11.7 ng	Complete profile
CP27	11.8 ng	Complete profile
CP32	15.6 ng	Mixture profile (complete)
CP33	18.4 ng	Partial profile (not CODIS-eligible)
CP29	34.6 ng	Partial profile, 10+ loci
CP10	36.5 ng	Complete profile
CP23	44.5 ng	Complete profile
CP34	98.6 ng	Complete profile
CP20	213 ng	Complete profile
CP7	1,030 ng	Complete profile
CP5	16,568 ng	Complete profile

DRUG PARAPHERNALIA

RESULTS

Each quantitation result was examined to determine if the sample was eligible for DNA analysis. Nineteen of 34 samples (56%) yielded sufficient amounts for DNA analysis. The total amount of DNA recovered was in the range of 2.28–16,568 ng (Table 1). CODIS-eligible DNA profiles (Figure 1) were developed in 79% of samples; this included mixture profiles with a distinguishable major contributor (Figure 2). Fifteen profiles were sufficient for CODIS entry, with 9 complete profiles, 3 complete mixture profiles and 3 partial profiles containing greater than 10 loci (Table 1). The remaining 4 partial profiles were insufficient for CODIS.

DISCUSSION

The modified extraction process described here is adequate to recover DNA from crack pipes and involves only an increase in extraction reagent volumes to accommodate the size of the specimen.

No laboratory has the time or resources to analyze every drug paraphernalia case for DNA; however, our results indicate that DNA can be recovered from over 50% of crack pipes submitted. This analysis may assist the investigation of homicides, sexual assaults or other crimes routinely examined in the laboratory.

REFERENCES

1. Krenke, B. *et al.* (2002) Validation of a 16-locus fluorescent multiplex system. *J. Forensic Sci.* **47**, 773–85.
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3. *Quantifiler® Kits User Manual* (2005) Applied Biosystems.

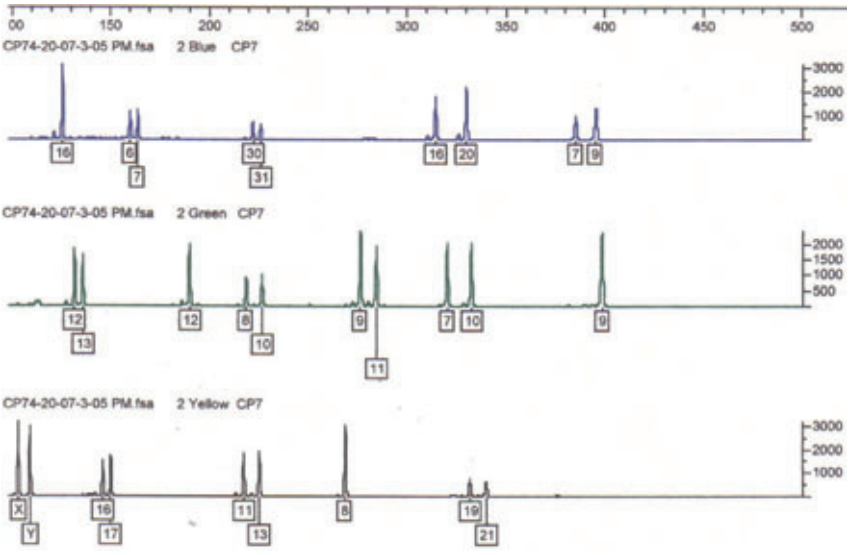

Figure 1. A complete profile obtained from a crack pipe. DNA was extracted as described, quantitated and amplified using the PowerPlex® 16 System, a GeneAmp® 2400 PCR System and a 30-cycle run. One microliter of amplified product was combined with 0.5 µl of Internal Lane Standard 600 and 24 µl of Hi-Di™ formamide, then denatured for 3 minutes at 95 °C and snap-cooled in an ice block for 3 minutes. The samples were analyzed using an ABI PRISM® 310 Genetic Analyzer and a 3-second injection. Additional injections of 6 and 9 seconds were performed as required. GeneScan® software version 3.7, Genotyper® software version 3.7 and PowerTyper™ Macros were used to complete the data analysis. A minimum peak height threshold of 150 RFU was required for allele calls.

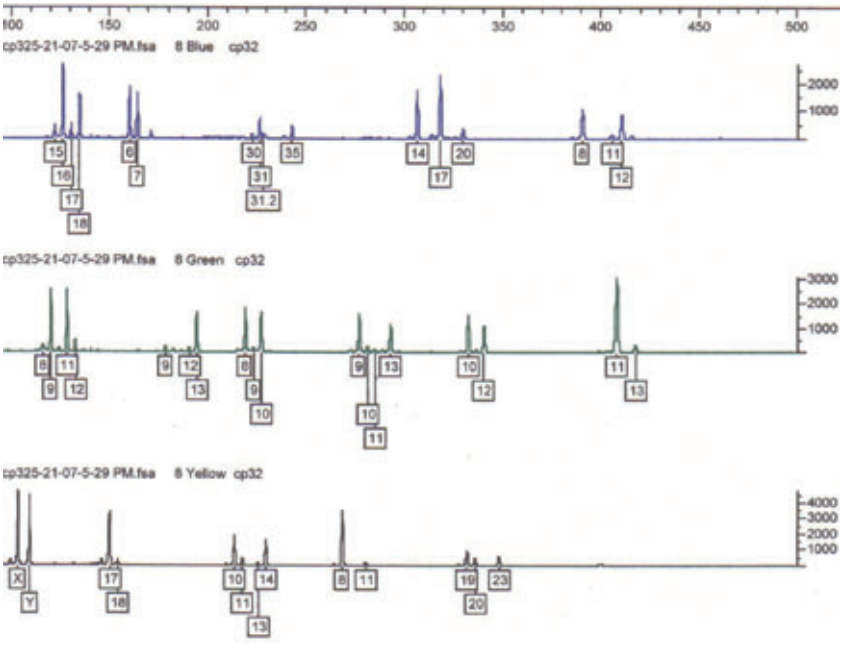

Figure 2. A mixture with a major profile suitable for CODIS submission. DNA was extracted, and samples were amplified and detected as described in Figure 1.