

Research article

# A comparison of three automated DNA purification methods in Forensic casework

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## Abstract

Manual Chelex<sup>®</sup>-100 and organic extractions (phenol/chloroform) are used as routine methods at the Swedish National Laboratory of Forensic Science, SKL. The aim of this study was to find an automated DNA purification system to replace the organic method. The following methods were evaluated and compared to each other and to the organic method used routinely; BioRobot<sup>®</sup> EZ1 with EZ1 DNA Investigator Kit and Card (Qiagen), iPrep<sup>™</sup> Purification Instrument with iPrep<sup>™</sup> ChargeSwitch<sup>®</sup> Forensic Kit and Card (Invitrogen), Magnatrix<sup>™</sup> 1200 Workstation with the Magnatrix<sup>™</sup> gDNA Blood Kit Forensic and two different protocols; Forensic protocol A and B (Magnetic Biosolutions). Blood on fats, cotton swabs, moist snuff, paper towels and leather, post-mortem blood and muscle tissue were extracted with the different methods. DNA concentration and quality of the electropherograms were examined. Individual comparisons between the four extraction methods showed that iPrep<sup>™</sup> and Magnatrix<sup>™</sup> 1200 gave significantly lower mean quantities compared to BioRobot<sup>®</sup> EZ1 and the organic extraction method ( $p < 0.05$ ). There were no significant differences between the latter two. BioRobot<sup>®</sup> EZ1 generated the best results and is in the process of being validated for routine analysis at SKL.

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**Keywords:** Organic extraction; BioRobot<sup>®</sup> EZ1; iPrep<sup>™</sup>; Magnatrix<sup>™</sup>; Magnetic bead; Magtration<sup>®</sup> technology

## 1. Introduction

Manual Chelex<sup>®</sup>-100 [1] and organic extractions (phenol/chloroform) [2] are used as routine methods at the Swedish National Laboratory of Forensic Science, SKL. The organic method is time consuming and involves hazardous chemicals. The aim of this study was to find an automated DNA purification system to replace the organic method.

Organic extraction is used for complicated samples such as moist snuff, paper towels and leather, which include many PCR inhibitors. It is important that a new method is able to purify the sample from inhibitors and at the same time give a satisfactory yield and quality of DNA.

## 2. Materials and methods

The following methods were evaluated and compared to each other and to the organic method used routinely; iPrep<sup>™</sup> Purification Instrument with iPrep<sup>™</sup> ChargeSwitch<sup>®</sup> Forensic

Kit and Card (Invitrogen), Magnatrix<sup>™</sup> 1200 Workstation with the Magnatrix<sup>™</sup> gDNA Blood Kit Forensic and Forensic protocol A and experimentally programmed protocol B (Magnetic Biosolutions) and BioRobot<sup>®</sup> EZ1 with EZ1 DNA Investigator Kit and Card, Tip Dance protocol (Qiagen). All systems use Magtration<sup>®</sup> technology [3] and are magnetic bead based systems.

Blood ( $4.1 \times 10^9$  white blood cells per litre blood) on fats, cotton swabs, moist snuff, paper towels and leather, post-mortem blood and muscle tissue were extracted with the different methods.

The DNA analysis was performed using ABI 7300 Real Time PCR System with Quantifiler<sup>™</sup> Human DNA Quantification Kit, GeneAmp<sup>®</sup> PCR System 9700 with AmpFISTR<sup>®</sup> SGM Plus<sup>®</sup> PCR Amplification Kit, Applied Biosystems 3130xl Genetic Analyzer and GeneMapper<sup>™</sup> ID v 3.1 (Applied Biosystems).

## 3. Results and discussion

Fig. 1 shows the quantification results from stained leather and kitchen paper. Individual comparisons between the four

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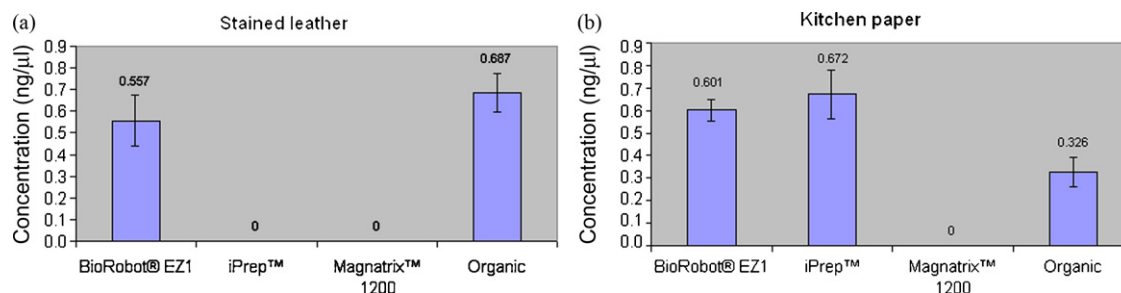


Fig. 1. Quantification results of samples made from leather (a) and kitchen paper (b), with 5 μl whole blood extracted with different methods. Each column in the diagram represents the mean DNA concentration of four replicates. The vertical lines mark the standard deviation.

extraction methods showed that iPrep™ and Magnatrix™ 1200 gave significantly lower mean quantities compared to BioRobot® EZ1 and the organic extraction method ( $p < 0.05$ ). There was no significant difference between the latter two.

iPrep™ produced complete DNA profiles from blood on cotton swabs, paper towels, fats and muscle tissue, but failed to generate satisfactory results from blood on leather, moist snuff and post-mortem blood. A possible explanation for these results can be that the materials affect the pH changes that the ChargeSwitch® technology depends on.

Magnatrix™ 1200 did not generate any DNA results except for some of the muscle tissue samples. The blank results can be deduced to the newly programmed Forensic B protocol and that the chemistry was run on a larger scale than recommended. A sensitivity test (0.05–1.0 μl whole blood) was run according to the volume recommendations and protocol A, but only blank results were produced (data not shown).

BioRobot® EZ1 generated good results for all the tested materials.

#### 4. Conclusion

BioRobot® EZ1 generated the best results on these types of samples and is currently being validated for routine analysis at SKL. After validation we anticipate that it will replace organic extraction for most types of samples.

#### Conflict of interest

None.

#### References

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