

Sensitivity of the sbeadex® forensic kit





Aim

The study set out to evaluate the suitability of the sbeadex® forensic kit for use in extracting DNA from forensic crime stains. The first stage of the evaluation was to test the sensitivity of the sbeadex® forensic kit at recovering DNA from blood and saliva dilution series' processed on a liquid handling robotic workstation.

Experimental design

A saliva sample dilution series and blood dilution series were prepared from donors with known genotypes in the concentration range of 2.5 pg/ μ L to 2000 pg/ μ L (Table 1).

Saliva sample concentrations	Number of replicates per plate	
2.5 pg/μL	8	
5 pg/μL	8	
10 pg/μL	8	
25 pg/μL	8	
35 pg/µL	8	
50 pg/μL	8	
Total	48	

Each 96-well extraction plate contained 48 saliva samples, 44 blood samples and four negative samples. Two plates were processed using the sbeadex® forensic kit and protocol on a liquid handling robot. The resulting DNA extracts were quantified using the Quantifiler™ Human assay (Applied Biosystems). 25 μL PCRs were performed with 1 ng of DNA added per PCR sample if sufficient levels of DNA were present using SGM Plus® PCR kits (Applied Biosystems). The PCR products were run on an Applied Biosystems 3130xl Genetic Analyser. The STR profiles were then analysed using GeneMapper™ ID v3.2 analysis software.

Blood sample concentrations	Number of replicates per plate	
50 pg/μL	4	
100 pg/μL	8	
250 pg/μL	8	
500 pg/μL	8	
750 pg/µL	4	
1000 pg/μL	4	
1500 pg/μL	4	
2000 pg/μL	4	
Total	44	

Table 1: Details the sample concentrations included in the dilution series

Sample type	Concentration	% of expected alleles observed – plate 1	% of expected alleles observed – plate 2
Saliva	2.5 pg/µL	31	6
	5 pg/μL	53	12
	10 pg/μL	60	25
	25 pg/μL	74	61
	35 pg/μL	82	68
	50 pg/μL	80	63
Blood	50 pg/μL	100	100
	100 pg/μL	100	100
	250 pg/μL	100	100
	500 pg/μL	100	100
	750 pg/μL	100	100
	1000 pg/μL	100	100
	1500 pg/μL	100	100
	2000 pg/μL	100	100

Table 2: Shows the percentage number of expected peaks observed in the STR profiles

Results

Full profiles were obtained for all blood samples across the range of DNA tested 50 pg/ μ L to 2000 pg/ μ L. Partial profiles were obtained for all saliva samples across the range of DNA concentration tested 2.5 pg/ μ L to 50 pg/ μ L (Table 2).

The mean peak heights for the blood samples varied from 499 rfu to 1726 rfu for the range of DNA tested. The mean peak heights for the saliva samples varied from 39 rfu to 144 rfu for the range of DNA tested (Figure 1).

Comparison of peak heights

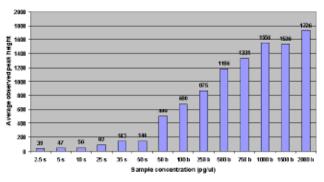


Figure 1: Shows the average observed peak height by DNA Figure 1: concentration and sample type. Key: s=saliva, b=blood

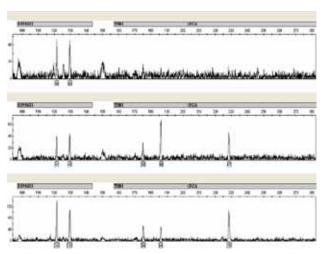


Figure 3: Example STR profiles generated from saliva samples Figure 3: with DNA concentrations of 5 pg/µL, 10 pg/µL and

Figure 3: 50 pg/µL respectively

Conclusion

The investigation has shown that the sbeadex® forensic kit and protocol is able to isolate and purify DNA from samples with a starting concentration of 2.5 pg/µL. Therefore it can be concluded that the sbeadex® forensic kit is suitable for use on forensic crime stain samples where there is only a low level of DNA deposited.

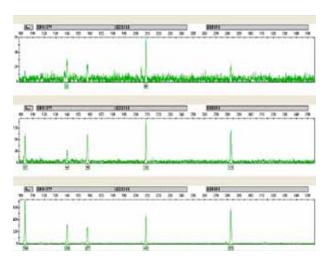


Figure 2: Example STR profiles generated from saliva samples Figure 2: with DNA concentrations of 5 pg/ μ L, 10 pg/ μ L and 35 pg/ μ L respectively

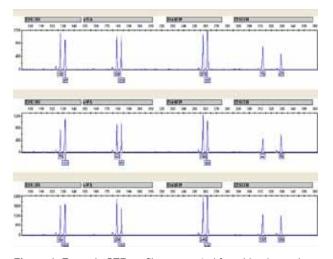


Figure 4: Example STR profiles generated from blood samples **Figure 4:** with DNA concentrations of 50 pg/μL, 250 pg/μL and **Figure 4:** 750 pg/μL respectively



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