FORENSIC BIOLOGY PROTOCOLS FOR FORENSIC STR ANALYSIS

Recovery of Spotted DNA Extracts from Whatman FTA Elute MicroCards				
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Recovery of Spotted DNA Extracts from Whatman FTA Elute MicroCards

1 Procedure:

- 1.1 This procedure will be documented on a paper form and attached to the casefile. See The Recovery from FTA Cards Extraction Sheet
- **1.2** Clean a 3mm hole puncher by punching a blank FTA Card 3 times, followed by wiping the puncher with a lint free wipe that has been prewet with 70% Ethanol.
- **1.3** Use the 3mm hole punch to punch out sample into a labeled 1.5 mL tube based on the chart below:

Original Quantitation Value	Recommended # of 3 mm punches for Recovery
1 ng/uL and above	At least 8 punches
100 pg/uL to 999 pg/uL	At least 10 punches
Less than 100 pg/uL	Punch entire stain

A maximum of ten punches per 1.5mL tube is suggested for enhanced recovery. If more than ten punches are needed, separate the punches into multiple 1.5mL tubes. The extract from these samples can be combined during the microcon.

- 1.4 Label 1 tube as the extraction negative.
- 1.5 Heat the tubes with the punches at 80°C for 20 min.
- 1.6 Wash Step: Add 500 µL of sterile/ultrapure water to each tube, vortex for 5 seconds.
 - 1.6.1 Note: Make sure the punches are moving around during the vortex step to ensure all punches are being washed. You may need to add more water if there are a large number of punches. Otherwise, there is the potential for PCR inhibitors to be left behind.
- 1.7 Briefly centrifuge the tubes, remove and discard the supernatant.
- 1.8 Add sterile/ultrapure water to each tube, 50 uL per hole punch. Briefly centrifuge the tubes to ensure the punches are all immersed in water.
- 1.9 Place tubes in thermomixer at 95°C for 30 minutes, shaking at 500 rpm. At the end of the incubation step, remove the tubes from the heat block and vortex for 60 seconds.

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- 1.10 Briefly centrifuge the tubes.
- 1.11 Collect the eluted DNA solution with a pipette and transfer to a labeled 1.5 mL tube. Discard the tube containing the discs/hole punches.
- 1.12 Microcon all samples down to 25 uL. See <u>Microcon DNA Fast Flow DNA Concentration and</u> <u>Purification</u>.
 - 1.12.1 Filter a maximum of 400µL at one time. For samples with more than 400µL, transfer the microcon membrane to a new collection tube as needed. If samples were separated during step 2.1, combine the samples using the same microcon membrane filter.
 - 1.12.2 Samples can be microconed to concentrate, however, for samples where color from the card is left behind (usually samples with a greater number of punches), microcon to clean and concentrate.
- 1.13 After re-extraction, if sample testing will continue in Forensic Biology, and the sample(s) is not already in the LIMS, the sample(s) will need to be entered to continue with processing. See <u>Creating Ad Hoc Samples</u> in the LIMS Process Manual. A quantitation of the sample is recommended post re-extraction from the FTA Elute Card.
- 1.14 If samples will be transferred outside of Forensic Biology, refer to <u>QC710 Locating and</u> <u>Processing of Retained and/or Spotted Extracts</u>
- 1.15 Attach the Extraction sheet to the associated casefile.

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