

FORENSIC BIOLOGY PROTOCOLS FOR FORENSIC STR ANALYSIS

Chelex Extraction from Soft Tissue (e.g. Fetus Samples)

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Chelex Extraction from Soft Tissue (e.g. Fetus Samples)

Sample sizes for this Chelex extraction should be approximately a 3x3mm cutting of tissue.

1. Remove the extraction rack from the refrigerator. Extract either evidence or exemplars. Obtain tubes for the extraction negatives and label them. Have a witness confirm the order of the samples.
2. Have a witness confirm that the tube label and entire LIMS input sample ID match for each sample and that the samples are in the correct order.
3. Obtain reagents and record lot numbers.
4. Pipette 1 mL of sterile or UltraPure deionized water into each of the tubes in the extraction rack. Mix the tubes by inversion or vortexing.
5. Incubate at room temperature for 15 to 30 minutes. Mix occasionally by inversion or vortexing.
6. Spin in a microcentrifuge for 2 to 3 minutes at 10,000 to 15,000 x g (13,200 rpm).
7. Carefully remove supernatant (all but 30 to 50 μ L).
8. To each tube add: 200 μ L of 5% Chelex (from a well-resuspended Chelex solution).
1 μ L of 20 mg/mL Proteinase K
9. Mix using pipette tip.
10. Incubate at 56°C for 60 minutes.
11. Vortex at high speed for 5 to 10 seconds.
12. Incubate at 100°C for 8 minutes using a screw down rack.
13. Vortex at high speed for 5 to 10 seconds.
14. Spin in a microcentrifuge for 2 to 3 minutes at 10,000 to 15,000 x g (13,200 rpm).
15. Place the LIMS output sample labels on the proper tubes. Confirm that the tube label and entire LIMS output sample ID match for each sample.
16. As needed, pipette aliquots of a neat, 1/100 dilution and a 1/10,000 dilution (using TE⁻⁴) into microcentrifuge tubes for real-time PCR analysis to determine human DNA concentration (refer to Section 4 of the STR manual).
17. Store the extracts at 2 to 8°C or frozen.
18. In the LIMS system, navigate to the Data Entry page, assign the samples to a storage unit (cryobox), and indicate which samples are completed.

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