

FORENSIC BIOLOGY PROTOCOLS FOR FORENSIC STR ANALYSIS

CHELEX DNA EXTRACTION FROM BLOOD AND BUCCAL SWABS		
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Chelex Extraction from Blood and Buccal Swabs

Sample sizes for Chelex extraction should be approximately 3 μ L of liquid blood or saliva, 1/3 of a swab, or a 3x3mm cutting of a bloodstain.

1. Review batch setup.
2. Remove the samples from the refrigerator. Extract either evidence or exemplars.
3. Have a witness confirm the tube label and entire LIMS input sample ID match for each sample and that the samples are in the correct order.
4. Have a witness confirm the names and order of the samples.
5. Obtain reagents and record lot numbers.
6. Pipette 1 mL of sterile or Ultrapure water into each of the samples.
7. Mix the tubes by inversion or vortexing.
8. Incubate in a shaker (at approx. 1000 rpm) at room temperature for 15 to 30 minutes.
9. Spin in a microcentrifuge for 2 to 3 minutes at 10,000 to 15,000 x g (13,200 rpm).
10. Carefully remove supernatant (all but 30 to 50 μ L). If the sample is a bloodstain or swab, leave the substrate in the tube with pellet.
11. Add 175 μ L of 5% Chelex from a well-resuspended Chelex solution using a P1000 μ L Pipetman.
12. Incubate at 56°C for 15 to 30 minutes.
13. Vortex at high speed for 5 to 10 seconds.
14. Incubate at 100°C for 8 minutes using a screw-down rack.
15. Vortex at high speed for 5 to 10 seconds.

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16. Spin in a microcentrifuge for 2 to 3 minutes at 10,000 to 15,000 x g (13,200 rpm).
17. 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.
18. Pipette aliquots of neat and/or diluted extract (using TE⁻⁴) into microcentrifuge tubes for real-time PCR analysis to determine human DNA concentration as needed (refer to the DNA quantitation procedure(s) in the STR manual).
19. Store the extracts at 2 to 8°C or frozen.
20. Ensure all required fields in the test batch have been filled out and review the assay.

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