Chelex DNA Extraction from Epithelial Cells

(FOR AMYLASE POSITIVE STAINS OR SWABS, CIGARETTE BUTTS, SCRAPINGS)

Sample sizes for this Chelex extraction should be approximately a 5x5mm cutting or 50% of the scrapings recovered from an item.

1. Review batch setup.
2. Remove the samples from the refrigerator. Extract either evidence or exemplars.
3. Obtain two tubes for the extraction negatives and label them.
4. 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.
5. Have a witness confirm the order of the samples.
6. To each tube add: 200 µL of 5% Chelex (from a well-resuspended Chelex solution).
   1 µL of 20 mg/mL Proteinase K
   (Note: For very large cuttings, the reaction can be scaled up to 4 times this amount. This must be documented. Scaling up any higher requires permission from the supervisor and/or IA of the case. The final extract may need to be Microcon concentrated.)
7. Mix using pipette tip.
8. Incubate at 56°C for 60 minutes.
9. Vortex at high speed for 5 to 10 seconds.
10. Incubate at 100°C for 8 minutes using a screw down rack.
11. Vortex at high speed for 5 to 10 seconds.
12. Spin in a microcentrifuge for 2 to 3 minutes at 10,000 to 15,000 x g (13,200 rpm).
13. 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.

14. As needed, pipette aliquots of neat and/or diluted extract (using TE⁴) into microcentrifuge tubes for real-time PCR analysis to determine human DNA concentration [refer to the DNA quantitation procedure(s) in the STR manual].

15. Store the remainder of the supernatant at 2 to 8°C or frozen.

16. Ensure all required fields in the test batch have been filled out and review the assay.