FORENSIC BIOLOGY PROTOCOLS FOR FORENSIC STR ANALYSIS

Qiagen Casework GO! Y Screen of Sexual Assault Stains or Swabs		
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Qiagen Casework GO! Y Screen of Sexual Assault Stains or **Swabs**

Approximately 1/4 of a swab or a 3x3mm cutting of a stain should be used. The Qiagen Casework GO! method should only be used for cases involving a male assailant on a female victim. Do not use for male on male cases, female on female cases, nor female assailant on male victim cases.

Procedure 1

1.1 Retrieve the following reagents and record the lot numbers

Allow reagents to thaw before use:

Casework GO! Lysis Buffer Proteinase K 1M DTT aliquot Ultrapure water, 15mL

- Turn on heating instruments. Set instruments with shaking to 65°C and instruments without 1.2 shaking to 85°C.
 - 1.2.1 More than one heating instrument per temperature may be used depending on the number of samples.
- 1.3 Retrieve sample cuttings in 1.5mL ClickFit tubes and spin baskets. Compare the tube tops to the input sample list in LIMS and confirm that you have the correct samples. The maximum number of samples per batch is 48 (47 samples + 1 extraction negative).
- 1.4 Obtain one 1.5 ClickFit tube with a Lyse and Spin basket for your extraction negative and label the tube top.
- 1.5 Fill out the performed by tab for Incubation. This will add the date and time to the extraction negative for the batch.
- 1.6 Print your output labels and label your samples and extraction negative.
- 1.7 Dilute 1M DTT. **NOTE**: 1M DTT aliquot must **NOT** be re-frozen.
 - 1.7.1 Pipette 5μL from the 1M DTT aliquot into a new 1.5 mL tube.
 - 1.7.2 Add 495µL of Ultrapure Water to 5µL of DTT. Vortex thoroughly.

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1.8 Under the reagents tab, select all reagents and click "Calculate Amount". Use the volumes listed in the "Needed Amount" column to prepare the master mix.

Manual calculation: n samples + one extraction negative x 1.1 using the following ratio.

Reagent	Per reaction	
Casework GO! Lysis Buffer	187 μL	
Proteinase K	7 μL	
DTT, diluted 1:100	6 μL	
Total volume	200 μL	

- 1.9 Vortex prepared master mix thoroughly.
- 1.10 Have a witness confirm the tube tops and output sample labels.
- 1.11 Add 200 µL of master mix to each sample tube including the extraction negative.
- 1.12 Incubate the sample tubes at 65°C for 25 minutes with shaking at 900 rpm. Record the incubation temperature and usage log for the instrument in LIMS.
 - 1.12.1 Record each heating instrument separately in LIMS, if more than one instrument is used.
- 1.13 Centrifuge the substrates in spin baskets at ~13,200 rpm to 15,000 rpm for 2 minutes.
 - 1.13.1 More than one centrifuge may be used depending on the number of samples.
 - 1.13.2 If liquid is still present in the spin basket of any samples, centrifuge only those samples for another 2 minutes.
 - 1.13.3 After the 2 minute spin, any liquid remaining in a spin basket can be manually pipetted from the basket to the sample tube.
- 1.14 Using a lint-free wipe, remove and discard the spin baskets (including the swab/substrate remains), taking care to avoid bubbles at the rim of the open tube. Close the tube.
- 1.15 Incubate the samples at 85°C for 5 minutes without shaking. Record the incubation temperature and usage log for the instrument in LIMS.
 - 1.15.1 Record each heating instrument separately in LIMS, if more than one instrument is used.
- 1.16 Store the extracts at 4°C.