

FORENSIC BIOLOGY PROTOCOLS FOR FORENSIC STR ANALYSIS

Zygem One-Step Sperm Lysis of Sexual Assault Stains or Swabs		
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Zygem One-Step Sperm Lysis of Sexual Assault Stains or Swabs

Approximately 1/4 of a swab or a 3x3mm cutting of a stain should be used. The Zygem One-Step Sperm Lysis 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.

1 Procedure

- 1.1 Retrieve the following reagents and record the lot numbers.

Allow reagents to thaw before use:

Acrosolv
Orange + Buffer
forensicGEM™ enzyme
Ultrapure water

- 1.2 After the reagents have thawed, vortex briefly and spin down for a few seconds.
- 1.3 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+1 samples and two extraction negatives, using the following ratio:

Acrosolv -	10 µL
Orange+ Buffer -	10 µL
forensicGEM™ enzyme -	2 µL
Ultrapure water-	78 µL

- 1.4 Retrieve the sample cuttings in 0.2mL tubes.
- 1.5 Obtain two 0.2 mL tubes for the extraction negatives. Label each tube followed by the last three or four digits of the output sample id.
- 1.6 Have a **witness** confirm that the tube labels are correct.
- 1.7 Ensure that all sample cuttings are lightly pushed to the bottom of the tubes (a new pipette tip for each sample can be used for this.)
- 1.8 Add 100 µL of master mix to each sample tube and extraction negative.
- 1.1 **For thermal cycler usage see [the Using the Mastercycler X50s manual](#).**

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1.9 The Zygem program is as follows:

52°C for 5 minutes
75°C for 3 minutes
95°C for 3 minutes
4°C for 2 minutes

- 1.10 Briefly centrifuge the tubes to spin down any condensation.
- 1.11 The samples can then be aliquoted for Quantifiler Trio.
- 1.12 Store the extracts at 4°C.
- 1.13 Reagent Storage:
 - 1.13.1 After tubes have been opened, the enzyme and the Acrosolv should be stored at -20°C.
 - 1.13.2 The Orange Plus buffer can remain at 4°C for convenience.