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DNA Extraction Using forensicGEM™ Storage Card (Saliva)

Zygem Quick-Start Guide

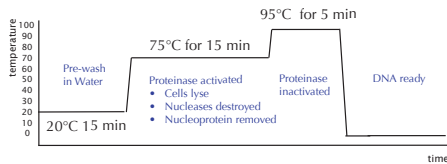
forensicGEM™ Storage Card Saliva

forensicGEM™ is specifically formulated and validated for forensic DNA extractions. Validation data can be obtained from www.zygem.com

The following method is recommended for extracting DNA from saliva deposited on storage cards.

- All manipulations should be performed in a clean-room or a PCR hood.
- Use only certified DNA-free tubes and reagents.
- The Harris punch and cutting mat should be wiped with 0.05% hypochlorite bleach and rinsed with DNA-free water between samples.

Procedure Outline



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- forensicGEM™ Storage Card Saliva is a preparative method for DNA extraction from most types of storage card. The forensicGEM™ method lyses cells and removes nucleoproteins from the DNA. Extracted DNA can be used for many types of genotyping including SNP and STR analysis as well as quantitative, multiplex and end-point PCR.
- Storage cards contain preservatives that can inhibit Tag DNA polymerase. The pre-soak step is to remove these inhibitors and is essential.
- DNA extracted using forensicGEM™ is largely single-stranded because of the 95°C heat step.
- For accurate yield assessment, a qPCR is recommended. The DNA produced by forensicGEM™ is approximately 90% single-stranded. If standard fluorescent chelating dyes are to be used for quantification, then this factor should be taken into consideration.
- As with any preparative method for nucleic acid extraction, best results are obtained when samples are handled at 4°C, or on ice, before and after extraction.
- For long term storage of the extracted DNA, add TE buffer to 1x (10 mM Tris, pH 7.5, 1 mM EDTA) and store at -20°C.
- The forensicGEM™ reagents are stable at 4°C but after tubes have been opened and for longer term storage, reagents should be stored at -20°C.

Technical tips for sample management

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Preparation

1. Remove one or two 1.2 mm discs from the card-stored sample and place into a thin-walled PCR tube or a 96-well tray.

Uneven application of the swab onto the storage card results in DNA yield variations. For the best results, punch in the centre of the area where the sample was applied.

2. Wash the disk in 100 µl of DNA-Free water by incubating at room temperature for 15 min. Aspirate the water from the disc and discard.

Extraction

1. Add:
 - 5 µl of 10x Buffer **SKY BLUE**
 - 44 µl of DNA-free water
 - 1 µl forensicGEM™

2. Incubate:
 - 75°C for 15 minutes
 - 95°C for 5 minutes

A thermal cycler can be used for this step

3. Pipette the solution to a new tube
The DNA is in this solution - not the punch

The sample is now ready for quantification and analysis. Typically, 2 - 5 µl should be used in PCR

To order forensicGEM™ Storage Card Saliva visit www.zygem.com or email: info@zygem.com or contact your local distributor

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