Casework Experience with the GlobalFiler® STR Kits
Saint Louis County Police Crime Laboratory
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How to Use This Template

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Protocols Overview and Optimization

GlobalFiler® Express Kit:

- Add 2 μL of DNA to the reaction at 2x concentration (20 ng/μL, 10 ng/μL, and 5 ng/μL) to a total of 5 μL.
- Amplify for 45 minutes at 120°C with an initial denaturation for 2 minutes.
- Add 5 μL of DNA to the reaction at 4x concentration (20 ng/μL, 10 ng/μL, and 5 ng/μL) to a total of 10 μL.
- Amplify for 3 minutes at 94°C with an initial denaturation for 2 minutes.

GlobalFiler® Casework Kit:

- Add 2 μL of DNA to the reaction at 2x concentration (20 ng/μL, 10 ng/μL, and 5 ng/μL) to a total of 5 μL.
- Amplify for 45 minutes at 120°C with an initial denaturation for 2 minutes.
- Add 5 μL of DNA to the reaction at 4x concentration (20 ng/μL, 10 ng/μL, and 5 ng/μL) to a total of 10 μL.
- Amplify for 3 minutes at 94°C with an initial denaturation for 2 minutes.

Master Mix Additive (MMA) Update

- Customers reported seeing elevated NED peak heights as well as a reverse skew in the purple channel (ID®)
- Suspected cause is the old MMA formulation which used a phosphate buffer - phosphate likely forming a precipitate (MgPO4), which reduces the available Mg²⁺ in the reaction
- Higher input DNA samples (>4ng) have a more pronounced effect due to the balance of primers and the excess DNA using up even more of the Mg²⁺
- Updated kit uses BSA in a Tris buffer and MMA is no longer in a separate tube
- Only applies to the Casework kit, no changes to the Express kit

Performance check needed!

- Sensitivity Series:

<table>
<thead>
<tr>
<th>Sample ID</th>
<th>Plate</th>
<th>Plate</th>
<th>Adjusted Quant Value</th>
<th>Total DNA Input in React (μL)</th>
<th>Total DNA Input in React (μL)</th>
<th>Total DNA Input in React (μL)</th>
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</thead>
<tbody>
<tr>
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<td>0.45</td>
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<td>3.03236</td>
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</tr>
</tbody>
</table>

Known/non-probable:

- F1/F2/SEH
- SERP (30 μL + SA from 4042 (perf extraction))
- Semen/saliva on fabric
- NTS
- Bloodstain card extract
- Blood on FA
- KPR
- Whole blood (25 μL EDT extraction)
- CTA-57 (no differential)

GlobalFiler® GlobalFiler® 

- Performance Check Results – MMA Update
- No significant differences were noted between the two kit formulations
- All controls correct, no contamination indicated
- The "old" kit had been thawed 4 days prior to amplification
- Data indicates the level of precipitate formation may be related to the length of time the kit sits refrigerated after MMA is added

Interesting Case #1 – 1990 Homicide

Scenario:

- Female maintenance worker was found stabbed and strangled in the men’s restroom of a local seminary. Original suspect was the victim’s boss - a glove with a bloodstain (Q11) was located in a gym bag at the suspect’s residence
- Evidence: (Q4 not previously tested)
- Q11 - 2001 extract of RBS from glove located in suspect’s residence
- Q14 - swabs from restroom sink, back of fixture

GlobalFiler® was just being brought online (waiting for NDS approval)

These samples were the first run with GlobalFiler® once it came online

Q11:
- 13 year old extract amplified with GlobalFiler® (purple channel not shown)
- Full profile obtained
- Minor noted that was not previously detected with

Q14 - Identifier Results:

- Identifiler Results: Mix 2 (or more) contributors, male detected, looks promising...
- Q14 - GlobalFiler® results: Mix of at least 3 contributors; at least 2 males (Y indet), major contributor is the victim - Bummer!
- Although the new analysis did not “solve” the case, it was incredibly valuable to see a direct comparison of the Identifiler and GlobalFiler® kits on a casework sample
- The GlobalFiler® kit is far more sensitive than Identifiler

Interesting Case #2 – “Yahoo”

Scenario:

- Suspect broke into an unoccupied home (resident had recently passed away - relatives had been packing belongings). Suspect steals a TV and other items, then steals the victim’s car. He is found in the city passed out in the stolen car (high on heroin).
- Evidence:
- Q1 – swabs of blood from bathroom wall
- Q3 – swabs from mouth of “Yahoo” bottle

Possible Explanations:

- Officer collected swabs at same time (or used contaminated swabs)? – no. Per officer, he collected the Yahoo bottle swabs but the bucal swab was collected at the city jail the following day...
- Chimeras?
- CODIS hit to a multi-allelic offender solves the mystery. The suspect is a bone marrow recipient!
- Interesting that one saliva sample has a mixture ratio of 11:1 (buccal swab) and the other 2:1 (Yahoo)
- The MO Highway Patrol has 7 multi-allelic offenders (out of 263,755 offenders [0.0002%] as of 3/5/15)

Summary

- Overall we are very pleased with the GlobalFiler® kits.
- The significantly reduced processing/amplification times have improved our throughput and allowed for expedited analysis.
- The 15μL input volume allows for dramatically increased sensitivity.
- The additional loci provide more definitive information with respect to the number of contributors and the gender of contributors.
- The additional loci provide an amazing power of discrimination - Random Match Probabilities are routinely in the nonillion-decillion range (10⁻¹⁹⁻¹⁰)