Developmental validation of the Yfiler® Plus PCR amplification kit

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ABSTRACT

Ychromosomal markers have proven useful in solving investigations where low levels of male DNA are recovered along with high levels of female DNA. An intrinsic limitation of Y-STRs compared with autosomal STRs is a reduced power of discrimination due to a lack of recombination throughout most of the Y-chromosome. Thus, in an effort to increase the power of discrimination we have developed a new 6-locus, 27-plex Y-STR system that includes the 17 markers from the AmpFSTR Yfiler® and Yfiler Direct kits plus 10 additional highly polymorphic Y-STR markers (DYS556, DYS627, DYS456, DYS516, DYS557, DYS449, DYS481, DYS3851a/b and DYS533). These ten new loci include 7 rapidly mutating Y-STR loci which allow for improved discrimination of related individuals. The new multiplex shows improved performance in inhibited samples and advanced female and male sample sets at ratios 1000:1, better discrimination between male-male mixture samples in high female DNA background, and faster time to results. Additionally, no reproducible cross-reactive products were obtained from bacteria or commonly encountered animal species. The haplotype diversity and discriminatory capacity calculations for several population groups will be presented, as well as father-son studies and validation studies demonstrating improved performance with challenging samples.

RESULTS

Figure 3: Yfiler® Plus Kit Power of Discrimination

<table>
<thead>
<tr>
<th>Number of Males</th>
<th>Yfiler® Plus (27 loci)</th>
<th>Yfiler® (17 loci)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>832</td>
<td>813</td>
</tr>
<tr>
<td>2</td>
<td>16</td>
<td>24</td>
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<tr>
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<td>1</td>
</tr>
<tr>
<td>5</td>
<td>1</td>
<td>1</td>
</tr>
</tbody>
</table>

Figure 6: Mixture amplification with Yfiler® Plus Kit

A) Profiles shown in the panels from top to bottom: 1 ng of male DNA with 1 µg female DNA, 50 pg male DNA with 1 µg female DNA, 1 ng of male DNA, 1 µg female DNA, 250 pg male DNA with 1 µg female DNA, 1 µg female DNA. B) Mixtures amplified with the Yfiler® Plus Kit produce high ICB at each dye channel when compared to the Yfiler® Kit. C) Yfiler® Plus Kit produces high first pass success rates with direct amplification of database samples (Database), comparable to Yfiler® Direct Kit.

CONCLUSION

• The Yfiler® Plus Kit is a dual application kit designed for amplification of extracted DNA (casework samples) as well as direct amplification of multiple database sample types (Blood and Buccal samples on treated paper). Blood samples on untreated paper with addition of Swim-in-Go® buffer, and Buccal swab lysates treated with Prep-n-Go® buffer.
• The Yfiler® Plus Kit contains 27 markers including 10 new highly for improved discrimination of related individuals.
• The data demonstrates optimal peak heights, as well as good intracorder balances for all sample types tested.
• The Yfiler® Plus Kit shows improved performance over Yfiler® with amplification of male-female mixture samples.
• The improved master mix formulation enables better tolerance to high levels of inhibitors, resulting in full profiles compared to no amplification with the Yfiler® Kit.
• PCR reaction time is >90 mins for a 30 cycle amplification protocol.

REFERENCES


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TRADEMARKS/LICENSING

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