



A161 Comparison of PowerPlex®16 HS to Minifiler® and Identifiler® Amplification Kits

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After attending this presentation, attendees will be familiar with the advantages and disadvantages of using PowerPlex®16 HS over Minifiler® and Identifiler® for common forensic samples, including degraded, inhibited, and low copy DNA. The performance of each amplification kit was evaluated for sensitivity and overall quality of DNA profiles obtained, including the number of alleles obtained and peak height and balance.

This presentation will impact the forensic science community by providing an informative initial evaluation of PowerPlex®16 HS, along with its comparison to other common amplification kits. Currently, a number of commercially available short tandem repeat (STR) amplification kits are available for use in forensic DNA laboratories and new, potentially promising kits frequently emerge. It is time consuming and costly for a laboratory to investigate each and every new methodology that arises, which can result in a laboratory's decision to forego evaluations of new products due to the uncertainty of their benefits over current methods. Thus, the forensic community greatly depends on individual laboratories' evaluations of new products. This study will aid other laboratories in need of an alternative protocol for challenging samples by presenting an unbiased view of the performance of three amplification kits, thereby allowing attendees to decide whether pursuing the implementation of PowerPlex® 16 HS would be beneficial to their laboratory.

The PowerPlex® 16 HS kit was designed to reliably type problematic forensic samples, including degraded, inhibited and low copy nuclear DNA samples. An initial sensitivity study was performed to compare the ability of PowerPlex® 16 HS, Minifiler®, and Identifiler® to obtain profiles from 0.01-1.0ng of DNA from blood stains and buccal swabs. Typical low quality forensic samples prepared and/or collected for additional studies are as follows: blood degraded by heat, UV, environmental conditions, and treatment with active oxygen products; blood inhibited by dyes found in denim; high quality cell line DNA inhibited by common PCR inhibitors like hematin, tannic acid and humic acid; swabbings and cuttings from a variety of potential low copy DNA sources, ranging from telephones to worn clothing; and other common forensic or clinical samples, including cigarette butts and biopsy slides. Signs of degradation and/or inhibition for each sample were evaluated by comparing quantitation values obtained via the Quantifiler® Human and Duo Quantification Kits. In order to effectively evaluate and compare PowerPlex® 16 HS's performance to that of the FID's current methodologies, development of DNA profiles were attempted from each sample through amplification using PowerPlex® 16 HS and, at a minimum, Minifiler® (if enough DNA was available, Identifiler® was also used). Additionally, for the cell line DNA spiked with inhibitors, overall profile quality was evaluated using 28, 30, and 32 PCR cycles.

Minifiler® and PowerPlex® 16 HS performed better than Identifiler® in each study evaluated. The first full profile was obtained by Minifiler® at 0.10ng, followed by PowerPlex®16 HS at 0.25ng and Identifiler® at 0.50ng. Profiles obtained from Minifiler® and PowerPlex® 16 HS were comparable for the forensic samples, with both outperforming Identifiler®, but both also exhibiting gross over-amplification on samples that were not truly low quality. Examination of PCR cycle number for inhibited DNA revealed that PowerPlex®16 HS performed best using 30 cycles at 0.50ng, while Minifiler® peaked using 30 cycles with 0.25ng, and Identifiler® showed that a 28 cycle, 0.25ng DNA input was optimal. The presence of inhibitors may account for the lower than usual (0.50-1.0ng) optimal Identifiler® DNA input. Though Minifiler® and PowerPlex® 16 HS often exhibited comparable results, it is very important to note that Minifiler® is only capable of providing STR information for eight loci plus Amelogenin, whereas PowerPlex® 16 HS can potentially amplify all 13 CODIS loci plus Amelogenin and two additional penta repeats. Thus, a "full profile" from Minifiler® is not as informative as a full profile from PowerPlex® 16 HS.

PowerPlex®16 HS, Low Quality DNA, PCR Cycle Number