



Rapid DNA maturity assessment



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ABSTRACT

Two fully integrated rapid DNA platforms were tested as a part of a rapid DNA maturity assessment in the fall of 2014. The assessment was conducted with sets of blinded single-source reference samples to gauge the typing success of the current rapid DNA typing technology. Samples were provided to participants for testing on the individual rapid platforms, and data was returned to the National Institute of Standards and Technology (NIST) for review and analysis. Both automated and manual review of the data sets were conducted to assess the success of genotyping the CODIS 13 core STR loci. Genotype results from the multiple platforms, participating laboratories, and STR typing chemistry was combined into a single analysis. The current assessment of the maturity of rapid DNA technology was focused on genotyping success, peak height ratios, and stutter artifacts across two platforms and multiple STR kit chemistries.

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1. Introduction

Integration of the extraction, amplification, separation, and detection processes for forensic DNA typing is a challenging goal. Several parallel efforts have been made to integrate the forensic workflow and utilize a simple swab in, answer out process within a single platform [1–3]. The ANDE (accelerated nuclear DNA equipment) device, developed by NetBio, Inc. (Waltham, MA), provides users with a fully integrated device to generate full STR profiles using the PowerPlex 16 chemistry within 84 min [1]. IntegenX Inc. (Pleasanton, CA) developed the RapidHIT 200 integrated device, which utilizes the PowerPlex 16 HS chemistry denoted 'PowerPlex 16 RapidHIT' and produces profiles in less than 90 min [2], and supports the GlobalFiler Express chemistry which produces profiles in less than 120 min [3]. The purpose of the 2014 Rapid DNA Maturity Assessment was to assess the current status of rapid DNA typing technology for the CODIS core STR loci in support of laboratory and future external (non-laboratory based) rapid DNA instrumentation implementation. Integrated (swab in – profile out) instruments capable of genotyping the core CODIS 13 STR markers were eligible for this study.

2. Materials and methods

Two fully integrated rapid DNA platforms were involved in the testing; the ANDE and the RapidHIT 200. The PowerPlex 16 chemistry (Promega, Madison, WI) was tested on both the ANDE and RapidHIT 200 platforms, and the GlobalFiler Express chemistry (Thermo Fisher Scientific, Waltham, MA) was additionally tested on

the RapidHIT 200 platform. NIST provided 20 single-source buccal swabs to seven laboratories which spanned across U.S. Federal, State and private laboratories. Eleven independent instruments were tested (5 ANDE platforms, 6 RapidHIT 200 platforms). 160 PowerPlex 16 samples were run between the ANDE and RapidHIT 200 platforms, and 120 samples with the GlobalFiler Express chemistry were run on the RapidHIT 200, for a total of 280 samples run across all instruments and chemistries. All data was exported from both platforms was returned electronically to NIST for data analysis.

Data was first parsed for genotype success with the provided analysis files and .png files (specific to ANDE) per platform. Genotyping success was assessed using two analysis methods: Rapid DNA Analysis and Modified Rapid DNA Analysis. These methods are described in the SWGDAM Quality Assurance Standards Addendum [4]. Profile interpretation by Rapid DNA Analysis does not involve human intervention, whereas Modified Rapid DNA Analysis requires manual interpretation of the profile [4]. Further analysis of peak height ratios and stutter percentages was performed using GeneMapper/D-X v1.3 for both the ANDE and the RapidHIT 200 platforms by importing the .fsa files. Bins and panels were created independently for both platforms and chemistries for the analysis.

3. Results and discussion

3.1. Genotyping

Success was measured by complete and concordant genotypes produced by the integrated rapid DNA devices when compared to genotypes obtained from traditional laboratory methods. Success was reported for the CODIS 13 core STR loci, PowerPlex 16 loci, and

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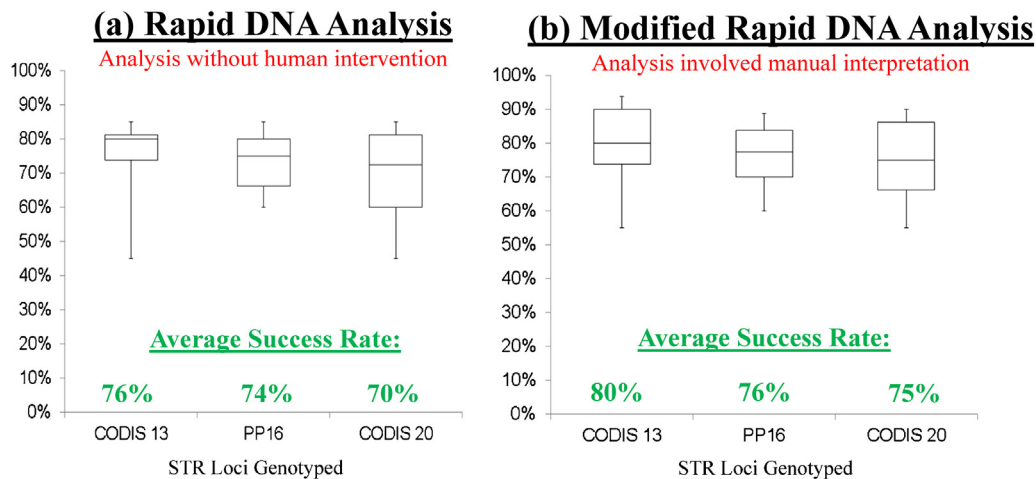


Fig. 1. Genotyping success for Rapid DNA Analysis (a), and Modified Rapid DNA Analysis (b). Success rates indicated the average success for each STR locus group genotyped. The minimum and maximum success rates observed within individual participating laboratories is represented by the whiskers of the boxplot.

the new CODIS 20 core loci. For Rapid DNA Analysis success was 76%, 74%, and 70% for the CODIS 13 core loci, PowerPlex 16 loci, and the new CODIS 20 core loci, respectively. Slight increases in success were observed for the Modified Rapid DNA Analysis with rates of 80%, 76%, and 75% for the CODIS 13 core loci, PowerPlex 16 loci, and the new CODIS 20 core loci, respectively (Fig. 1).

Success per locus for the PowerPlex 16 data (ANDE and RapidHIT 200 data combined) was above 80% across all loci. The success per locus for the GlobalFiler Express data (RapidHIT 200) was above 80% for all loci except the Y-indel (77%) and D8S1179 (78%).

3.2. Peak height ratios and stutter analysis

Peak height ratios were calculated for all complete profiles for the combined ANDE and RapidHIT 200 PowerPlex 16 data ($n = 118$). The mean peak height ratios ranged from 81% to 93% across all PowerPlex 16 loci. For the RapidHIT 200 GlobalFiler Express ($n = 67$) data set the mean peak height ratio ranged from 79% to 92% across all loci. Stutter percentages calculated for both the combined PowerPlex 16 dataset and the GlobalFiler Express dataset were within the observed developmental validation range for both PowerPlex 16 and GlobalFiler Express using conventional laboratory techniques [5,6].

4. Conclusions

Two fully integrated platforms (ANDE and RapidHIT 200) were included in the 2014 Rapid DNA Maturity Assessment. A total of 11 instruments were tested within 7 laboratories, for a total of 280 samples examined. Data for the Maturity Assessment was generated from October 2014 to December 2014, and returned to NIST for all data analysis and interpretation. It should be noted that further updates to instrumentation, manufacturing, and software may have taken place since this maturity assessment was completed. Observed success for the CODIS 13 core loci ranged from 76% employing Rapid DNA Analysis to 80% employing Modified Rapid DNA Analysis (8 additional profiles were deemed fully concordant). Peak height ratios and stutter percentages were within the observed developmental validation ranges for both the PowerPlex 16 and GlobalFiler Express within each dataset.

The implementation of fully integrated platforms aims to provide the ability to greatly reduce the overall forensic DNA typing process for single-source reference samples. This could be valuable for laboratories that need to produce a profile in a time-sensitive case for single-source samples. Generating DNA profiles in an automated fashion with the use of integrated platforms has future applications at booking stations and in other field locations such as airports or

border crossings. The ability to generate a DNA profile in less than 2 h allows for many potential point-of-collection and STR typing scenarios.

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Conflict of interest

None.

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