Comparative Evaluation of RapidHit™ ID and ANDE™ 6C

JIN, Soulbee, KILMURY, Sara, DZIAK, Renata, and KOMONSKI, Diane*
Centre of Forensic Sciences - Biology Section, Toronto, ON; Ministry of the Solicitor General
**email: Soulbee.jin@ontario.ca*

Introduction

- Total of 107 and 104 samples were processed on RapidHIT™ ID (RHID; Applied Biosystems) and ANDE™ 6C (ANDE; ANDE Corporation), respectively
- A wide variety of common forensic sample types were tested at various sensitivity levels
- Profiles were evaluated for quality and concordance based on
  - Rapid analysis: Run Result produced by the expert system accepted without further review
  - Modified Rapid analysis: profiles were technically reviewed (RHID only)
- Overall laboratory workflow & user experience from sample preparation to data review were compared, and results will be used to inform implementation strategies

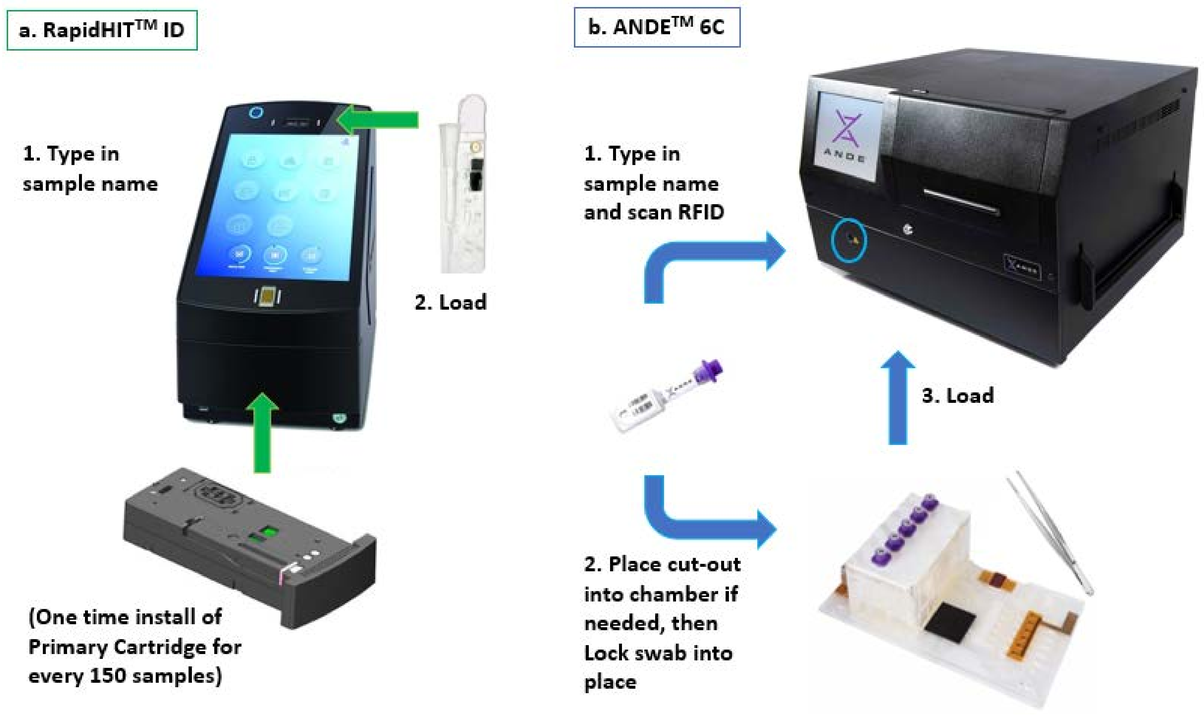

Figure 1. Workflow comparison between RapidHit™ ID (a) and ANDE™ 6C (b). All reagents from extraction to capillary electrophoresis (CE) are contained on an ANDE Chip whereas on RHID, CE reagents are contained separately on the Primary Cartridge.

Methods

- Sample sets were prepared in near identical manner for each system to allow for a direct comparison
- A-Chip (ANDE) & Ace cartridge (RHID): high yield blood & buccal swabs only (blood not recommended by ANDE, ANDE Corporation)
- I-Chip (ANDE) & Intel cartridge (RHID): low yield blood & buccal swabs, and other forensic sample types (sampling and pre-treatment as per manufacturer guidelines)
- For RHID data review, GeneMarker® default settings were used for both cartridge types
- On ANDE, minimum number of passing loci was set to 20 and 10 for A-Chip and I-Chip, respectively

	RapidHit™ ID	ANDE™ 6C
Chemistry	GlobalFiler™ Express (22+2 Y-STR)	FlexPlex™ (24 +3 Y-STR)
Processing time	1 sample in ~90 min	5 (A)/4 (I) samples in ~90 min
Pre-treatment of samples	None	Yes, for FTA and bones
Method for low yield sample processing	Decreased lysis volume + increased PCR cycle	Ultrafiltration
Sample recovery	Yes	No
Required run frequency	Run 1x per week	None
Expert system Passing criteria	Full profiles without QA flags	Full and partial profiles*
Manual data interpretation within the Rapid System	Possible	No
Import of external profiles into software	No	Yes

Table 1. Contrasting features of the two Rapid systems in sample processing, instrument and chip function, and data analysis. Minimum number of passing loci on ANDE was set to 20 and 10 for A- and I-Chip, respectively (*).

Results

- While run time was quicker on ANDE, RHID workflow per sample was less laborious (ie. plug-and-go, no pre-treatment; see Figure 1, Table 1).
- Many challenging samples were chosen for this evaluation. Percentage of successful profiles generated by RHID and ANDE in Figure 2 allows for direct comparison between the systems and is not a reflection of their overall success rate outside of this study.
- Using the Rapid Analysis, ANDE generated a higher amount of passing data which included both full and partial profiles. On average, first pass success rate for full profiles was 35% for both systems (Figure 2a).
- There were six (6) instances of discordant genotypes with ANDE, compared to two (2) with RHID. In these cases, discordance only occurred in one (1) locus per profile (Figure 2a).
- Using the Modified Rapid Analysis on RHID, the number of full profiles increased by 11% and 33% using Ace and Intel Cartridges, respectively, compared to the Rapid Analysis. Using the Modified Rapid Analysis, RHID produced a higher number of full profiles compared to ANDE by 22% after review (Figure 2b).
- Run Results are categorized by sample type in Table 2. Stain cut-outs performed better on RHID; for all others ANDE generated better results using the Rapid Analysis, RHID generated better results using the Modified Rapid Analysis.

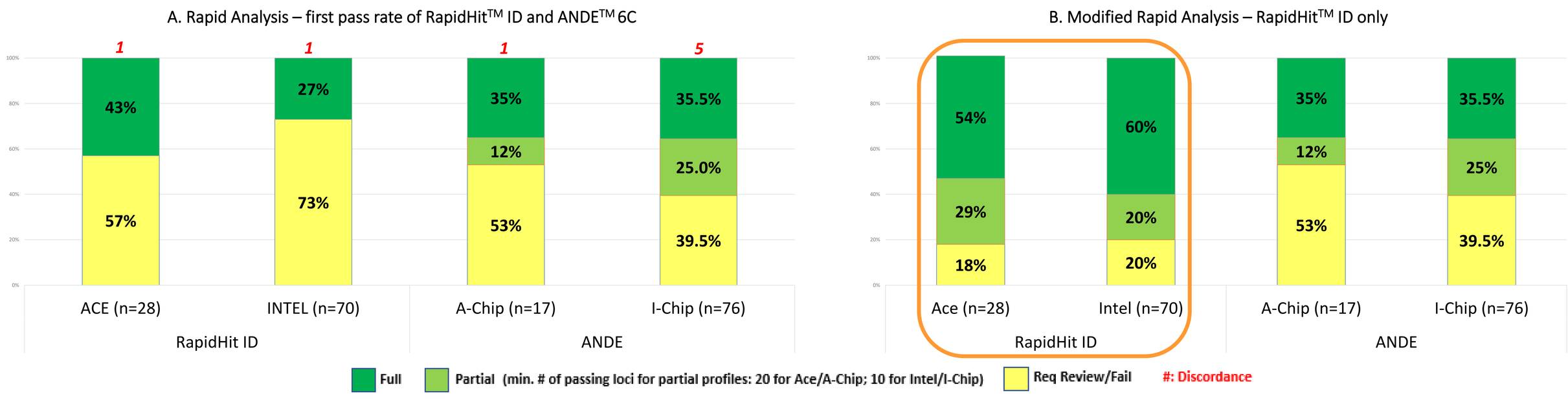

Figure 2A. Success rate of profiles generated by RapidHit™ ID and ANDE™ 6C using the Rapid Analysis where the Run Result generated by the expert system is accepted without further review. With ANDE, passing profiles include both full and partial profiles that meet the minimum number of pass loci (set as 20 and 10 for A- and I-Chip, respectively). On RHID, only full profiles without artefacts are marked as Passed. Figure 2B. Success rate of profiles generated by RapidHit™ ID and ANDE™ 6C using the Modified Rapid Analysis for RHID only (Modified Rapid Analysis not possible with ANDE).

Samples – sensitivity range or substrate types tested	Sampling recommendation based on profile quality obtained	
	RHID	ANDE
Buccal swabs (Touch to 40 swipes; 1 direction = 1 swipe)	A: 20-40 I: Touch-20	A: 20-40 I: Touch-10
Blood swabs (0.05 – 100 µl)	A: 20-100 I: 4-20	A: 20 I: 4-20
Mock casework sample types	I: drinking container swabs, marijuana cut outs, toothbrush swabs, chewed gum, blood/saliva on tissue, used utensil	
UHR (bone and tissue)	Muscle swab, organ cut out, fresh bone (scraping/granules)	
Cut-outs (buccal, blood on FTA, S&S, cotton)	A: 2x(3x3mm), 6x6mm I:3x3mm, 2x(3x3mm)	I:2x(3x3mm) Low success rate

Table 2. Sample types tested during this study and sampling recommendations based on the quality of profiles obtained. "A" indicates Ace (RHID) or A-Chip (ANDE); "I" indicates Intel (RHID) or I-Chip (ANDE). Duplicate testing was done for buccal and blood swabs; replicate testing was done for mock casework sample types if available;

Conclusion

- Users must be trained on correct sample collection procedures to obtain the best possible profile
- Implementation strategy should be used to inform the choice of a Rapid System
- ANDE is better suited in cases where access to scientific interpretation is minimal to none.
- ANDE is well-suited in scenarios where a large number of samples require a quick turnaround time (ie. mass disaster scenarios).
- RHID can generate a high amount of usable data where scientific interpretation is readily available. If setting new analytical thresholds, validation is recommended.
- RHID is useful in cases where sporadic priority samples demand a quick turnaround time (ie. investigative aid) or where a steady level of samples can be expected overtime that would benefit from the quick processing.
- CFS is following the FBI and SWGDAM guidelines for the validation and implementation of ANDE for forensic casework, starting with a pilot project through a collaboration with the Office of the Chief Coroner & Ontario Forensic Pathology Service.

*Acknowledgements: Jason Werking & Amie Ingold from ThermoFisher Scientific, Melissa Schwandt and Julie French from ANDE have provided excellent training and on-going support for the two Rapid Systems.