Does automation improve cell-based potency assay development?

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Challenges with cell-based potency assay development

- Developing precise, accurate and reproducible potency assays in a GMP environment presents multiple challenges.
- Potency assay are laborious to set up, requiring on average 2-4 hours of hands-on time (HoT) for a less complex assay, with many analysts reporting repetitive motion issues.
- A single plate potency assay involving a reference and three samples in duplicate involves approximately 200 liquid transfers, and many potency assays require three plates.
- In a QC environment, traceability of pipetting steps is non-existent, making troubleshooting anomalous results challenging.
- Method transfer into a QC environment requires several training assays and has varying levels of success dependent on analyst experience.
- Repeated method transfer is often required due to analyst turnover and availability.
- All the above factors contribute to high assay failure rates and a lack of reproducibility, which can lead to drug development delays and increased cost.

Can automation facilitate potency assay development?

- Implementation of automation can reduce the number of manual liquid transfers by 75%, reducing repetitive motion and risk of analyst error.

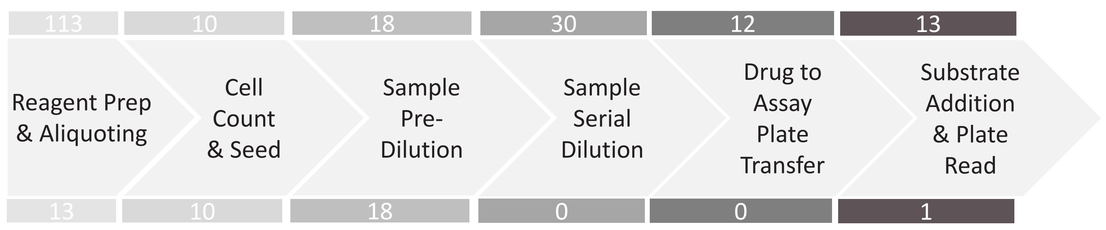


Figure 1. Schematic representation of the main steps in a cell-based potency assay. Numbered rectangles highlight the number of liquid transfers; top line represents a fully manual setup; bottom line represents a semi-automated assay setup performed using the Hamilton STAR Microlab liquid handler.

- During 2023, analyst error accounted for 44% of all non-chargeable assays across Chemistry Manufacturing and Controls.
- Reduced analyst input can facilitate method transfer, improve assay reproducibility and minimize the number of failed, non-chargeable assays.

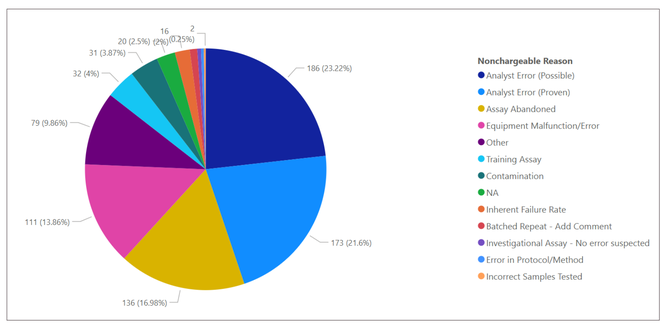
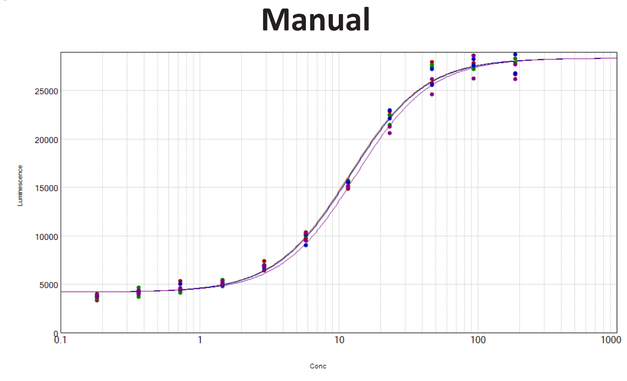
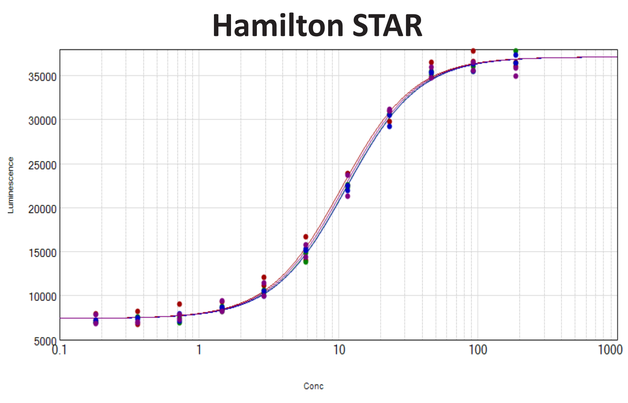


Figure 2. Pie chart showing the number and percent of non-chargeable failed assays and the reason for the assay failure.

- Automation can increase assay throughput.
- Automation improves data integrity as audit trails and error logs are provided for all automated liquid handling steps.
- Automation enables more complex assay formats such as complete randomization eliminating plate position effects.

Manual and automated methods give comparable relative potencies

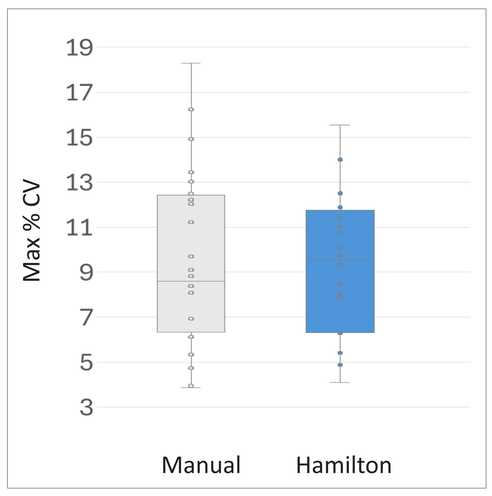




Target Potency	Manual	Hamilton
100% (n=6)	104	107
	105	107
	98	77
	98	93
	98	92
	89	96
50% (n=2)	54	47
	50	46
71% (n=4)	74	65
	65	66
	72	73
	70	73
141% (n=3)	144	135
	135	146
	156	155
200% (n=3)	208	225
	197	216
	209	212

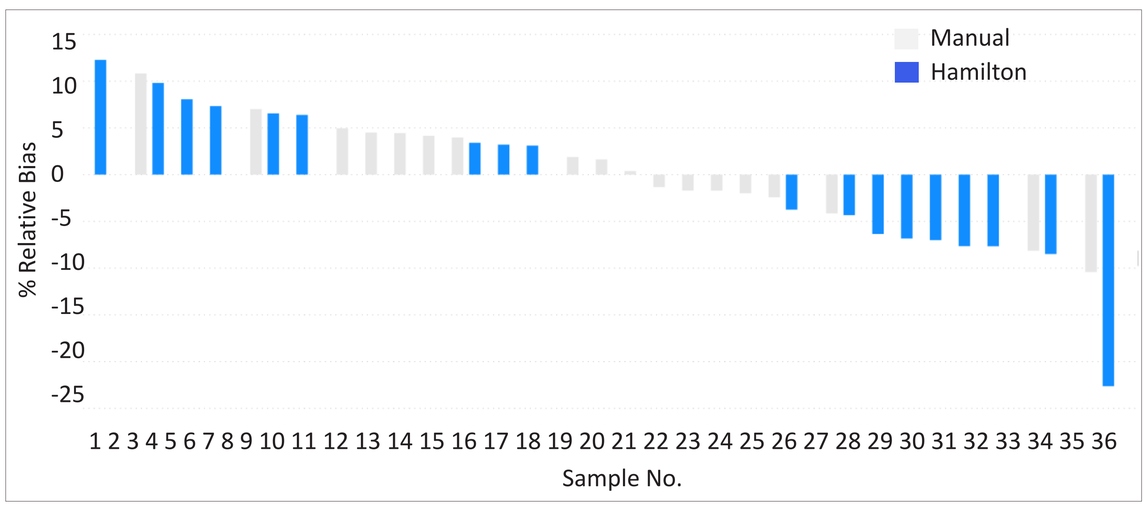
Manual and automated methods have equivalent precision

- Maximum %CV for reference and sample material was used to monitor assay precision (n=24).
- The manual method and automated method has assay precision in the small data set; however, assessment over a larger data set including several analysts may identify differences.
- The average Max %CV for the manual method was 9.5% ± 3.9 and for the automated method was 10.2% ± 5.6.



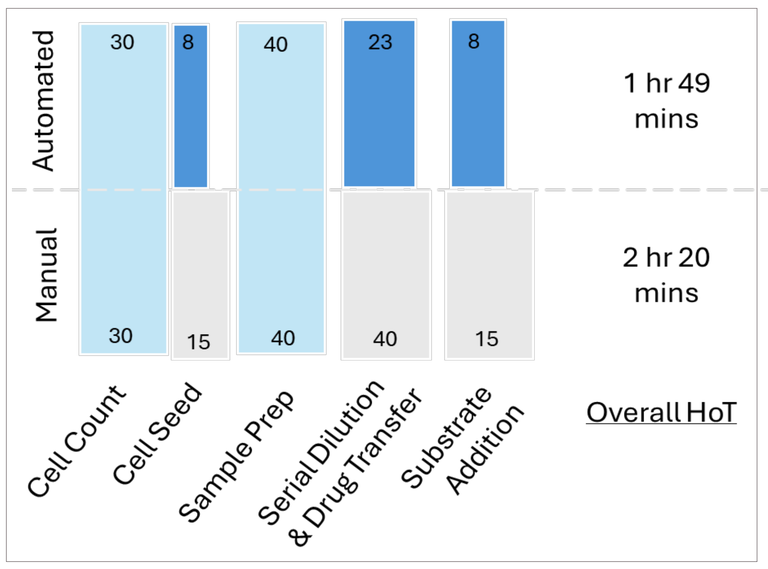
Manual and automated methods have equivalent accuracy

- The accuracy of both the manual and automated methods were assessed using target potencies of 50, 71, 100, 141 and 200% and the percent relative bias was calculated for each sample (n=18).
- The relative bias for the manual method was between -8% and +11%, and for the automated method between -6% and +12% (excluding a single relative bias reading of -23% appeared anomalous).



Conclusions

- Automation reduced the number of manual handling steps by 75%.
- The automated cell-based potency assay produced equivalent assay accuracy, precision and repeatability compared to the manual method.
- Automation reduced the average HoT by 30 minutes, but due to assay incubations, did not shorten the total assay time.



- Specific consumables were required for the automated method, but the overall cost difference per assay was negligible. Larger volumes of reagents were also required to allow for dead volumes on the Hamilton Microlab STAR Liquid Handler robot.

Consumable	Quantity per assay	Unit Cost (£)	
		Manual	Automated
Dilution Plate	1	£1.65 (Dilution Plate)	£3.80 (96 Micronic Tubes)
Sterile Trough	4	£1.68 Reagent Reservoir	£6.00 (Integra Sterile Trough)
Filter Tips	Manual 3 Boxes Automated 4 Boxes	£14.20 (Rainin)	£8.25 (Hamilton)
Total Assay Cost	n/a	**£51**	**£60**

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