Catalent BIOLOGICS

Automation Friendly Assay Design

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08May2017

DEVELOPMENT DELIVERY SUPPLY

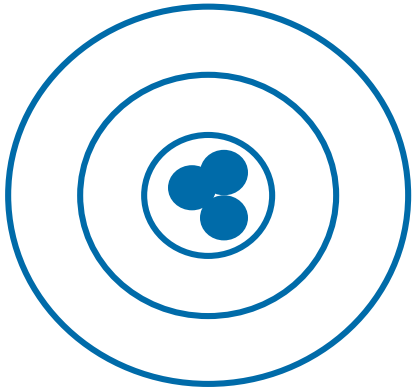
more products. better treatments. reliably supplied.™

Why Automation?

Automated instrumentation creates important advantages

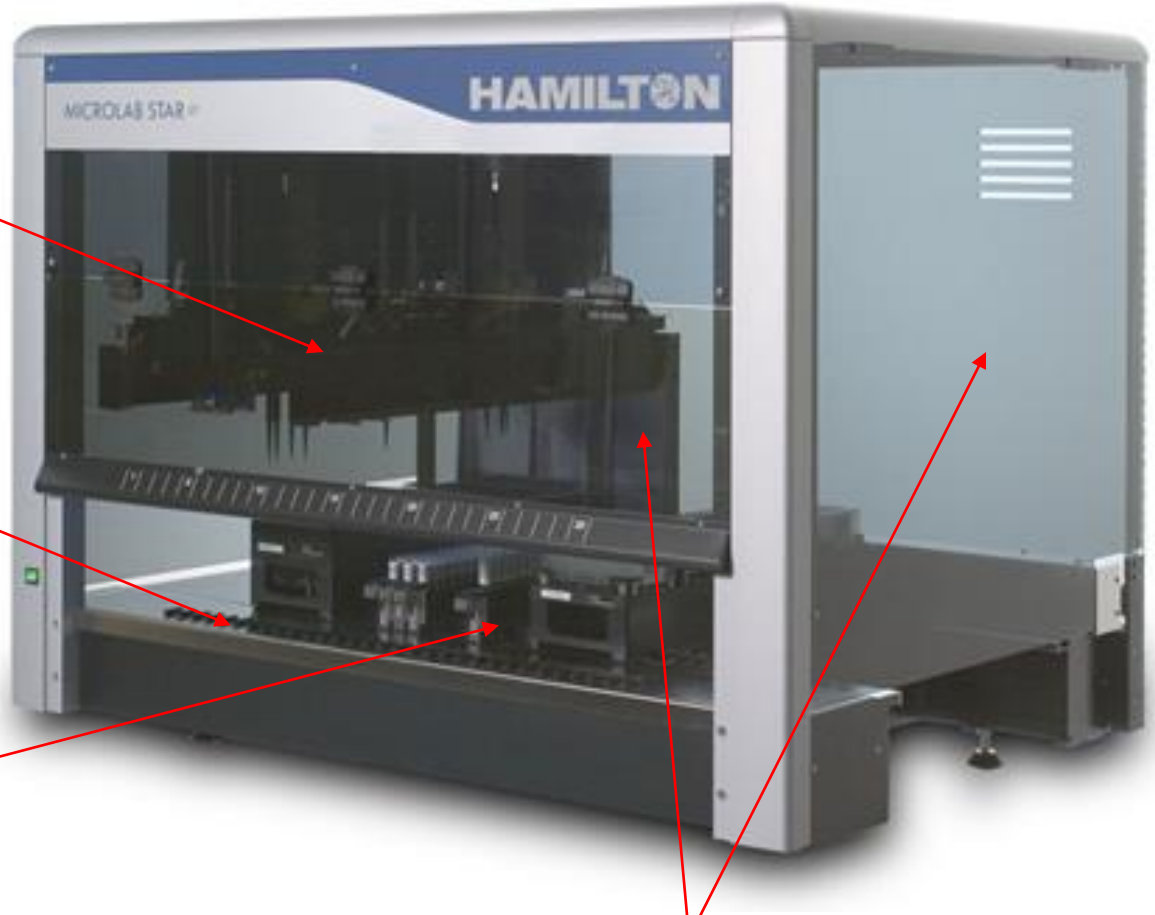
Reduced risk of repetitive motion injuries

Maximize:
Output
Minimize:
Time and Effort



Reliable, Accurate and Precise

Automated Liquid Handlers at Catalent



Pipetting arm

Deck

Carrier for tubes and assay plates

STAR^{let}

Front and side safety covers

Case Study

Product

- Naturally derived bioactive antagonist to virus infection/replication

Challenge

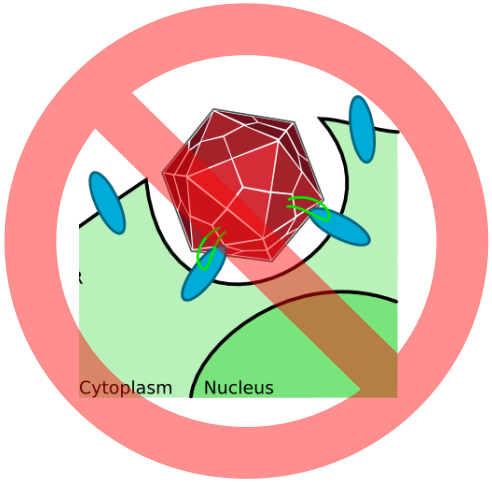
- Rapidly identify antagonist samples with high neutralizing activity

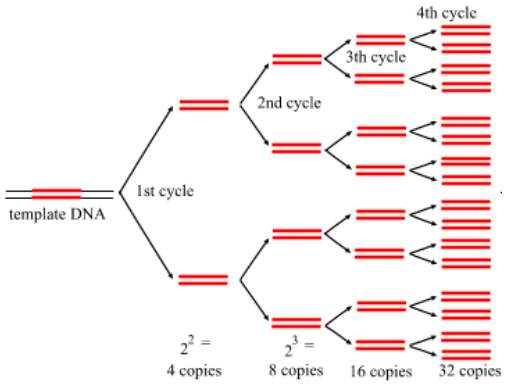
Current process

- Complex
- Cumbersome
- Very slow
- Low-throughput
- Prone to false positives and negatives

Conceptual Solution

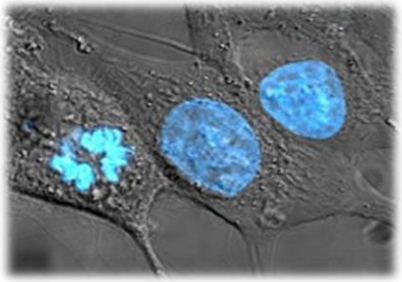
- A qPCR assay could quantitate viral replication
- Antagonist should block infection, depress replication





- Simpler and faster assay

Quantitative PCR



Cells

↓

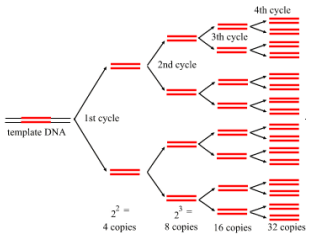
RNA extraction

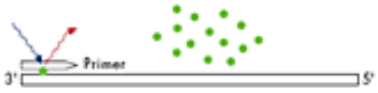
↓

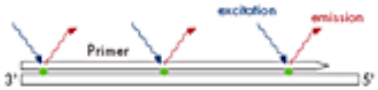
Reverse transcription

↓

qPCR







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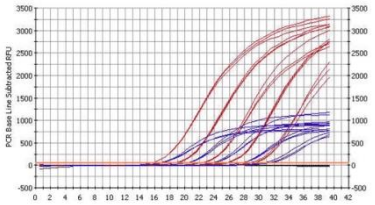
Fluorescent-label product

↓

Quantitate product



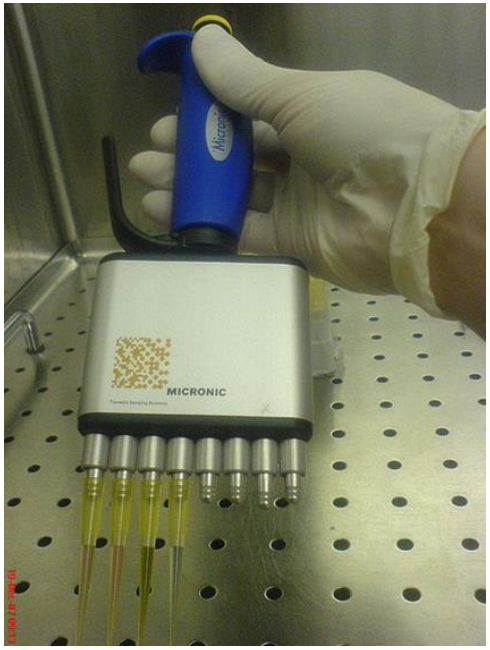
Modernization of 'Classical' Technique

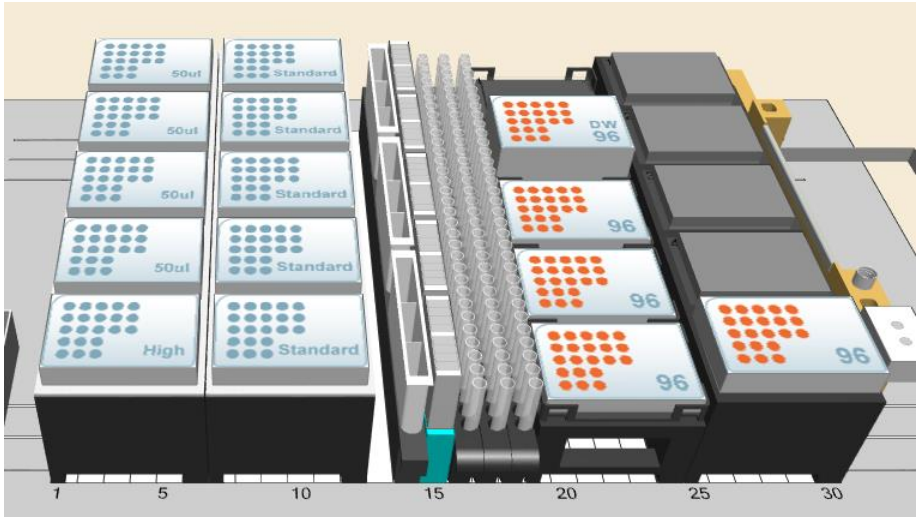
Old Assay

- 'Classical' but complex assay
- 8 Steps
- High resource

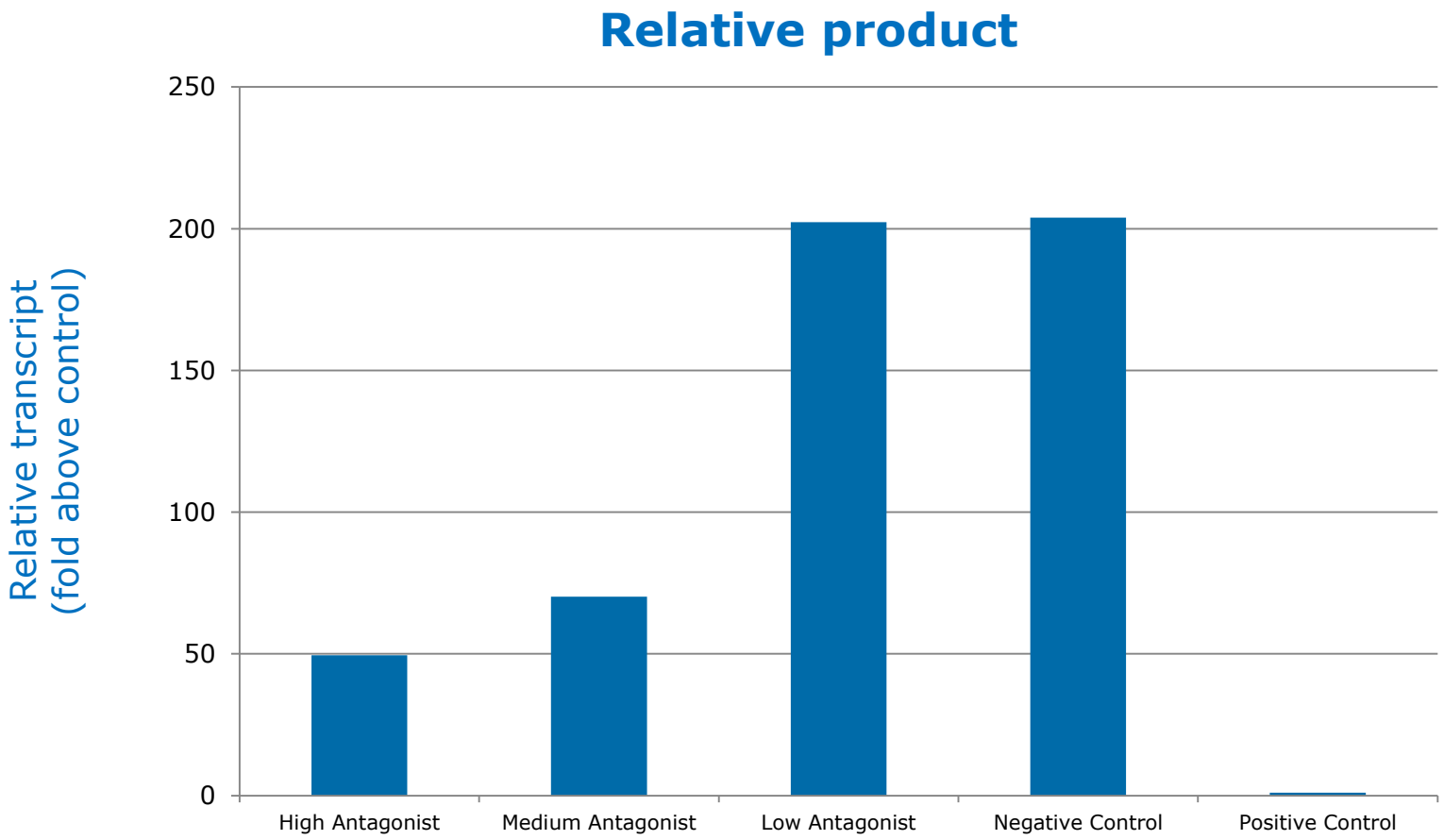
New Assay

- Simple modern assay
- 5 steps
- Lower resource



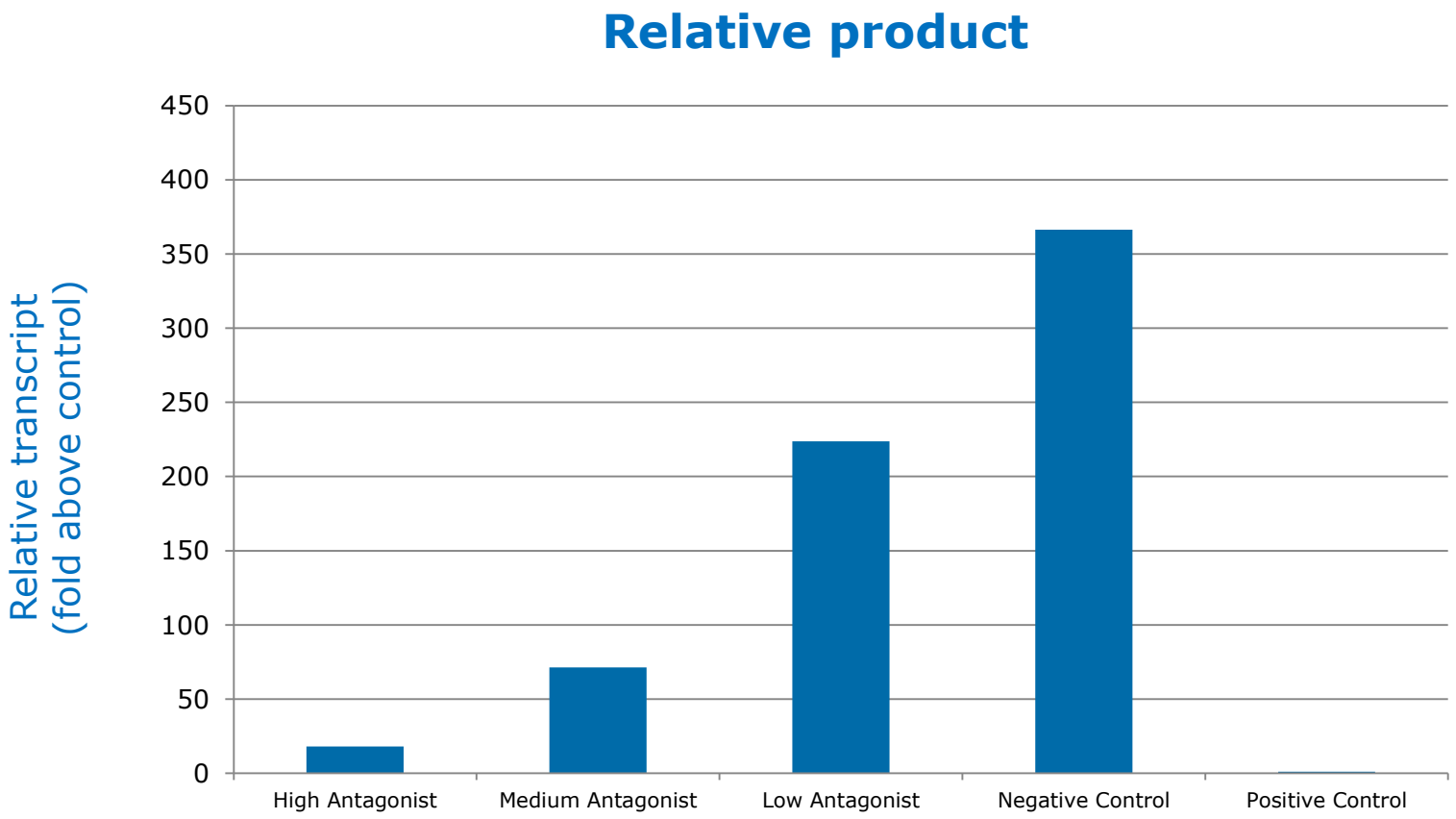


Manual qPCR Method Results



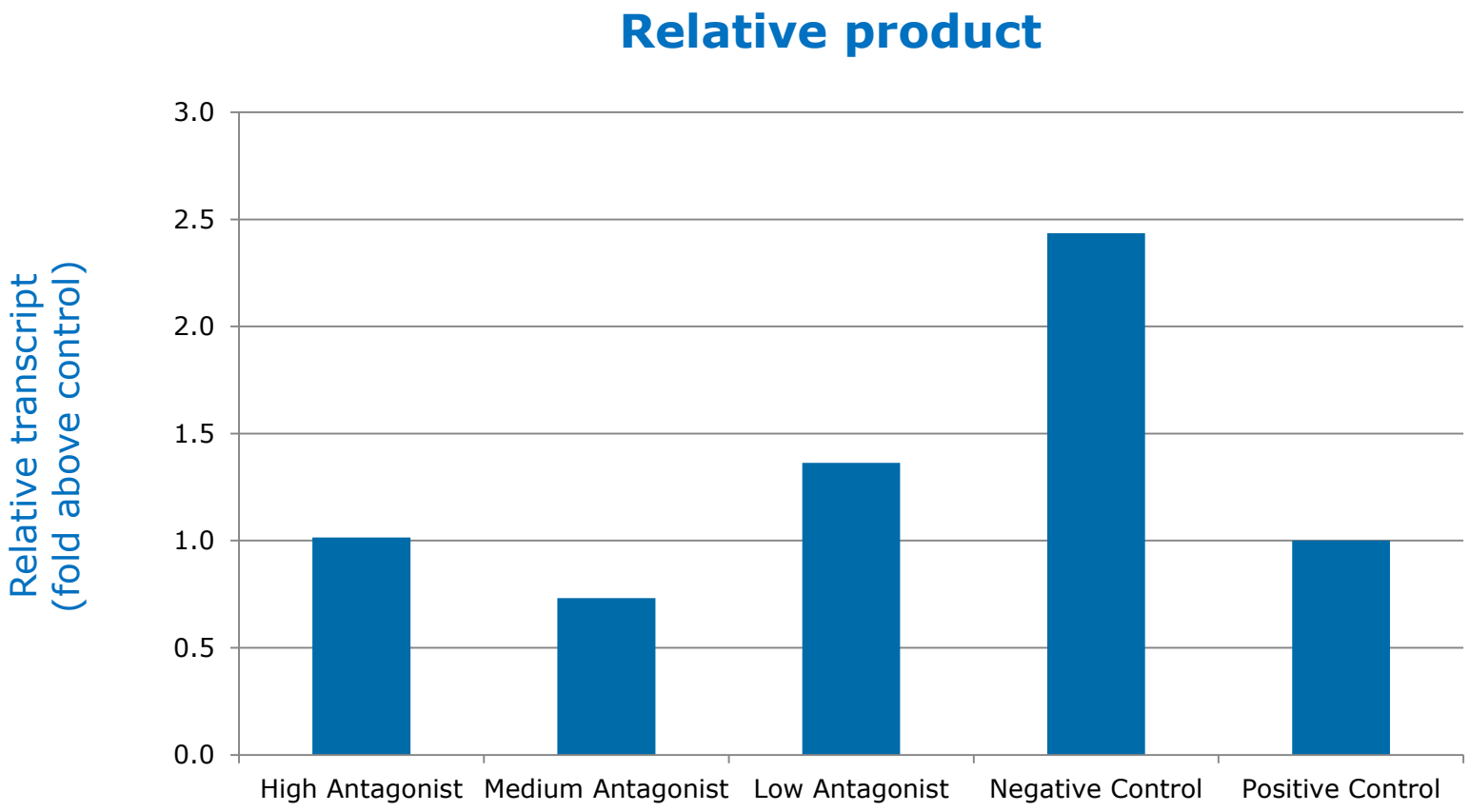
2 Analysts: 30 minutes to load 96 well plate with master mix and samples

Automated qPCR Method Results



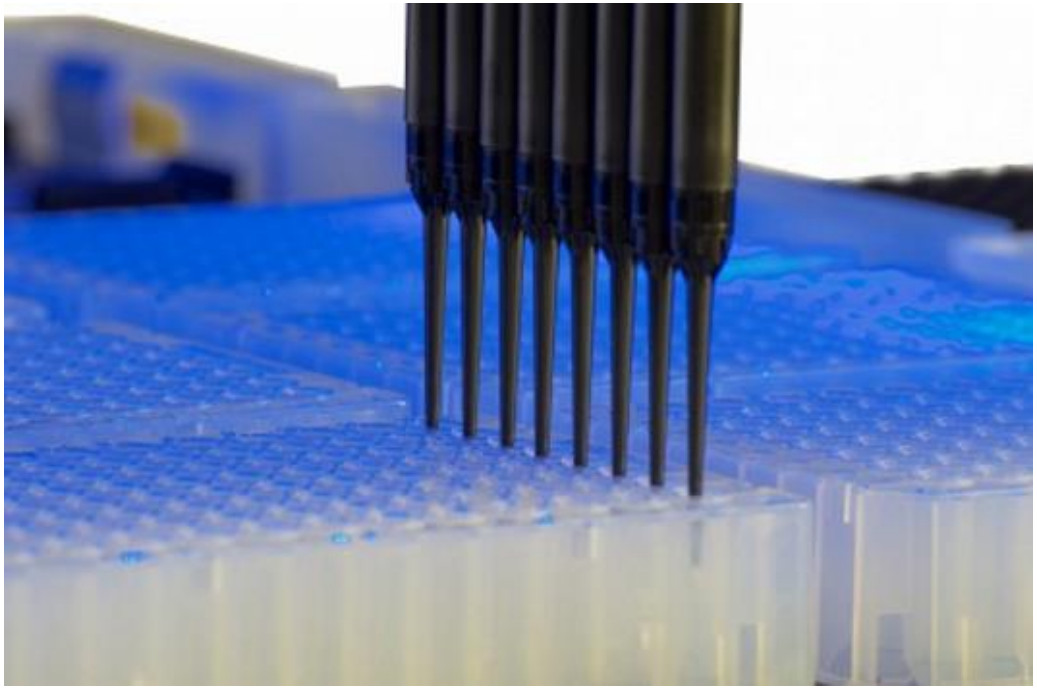
1 Analyst, 7 minutes to load 96 well plate with master mix and samples

First Attempt at Complete Automation



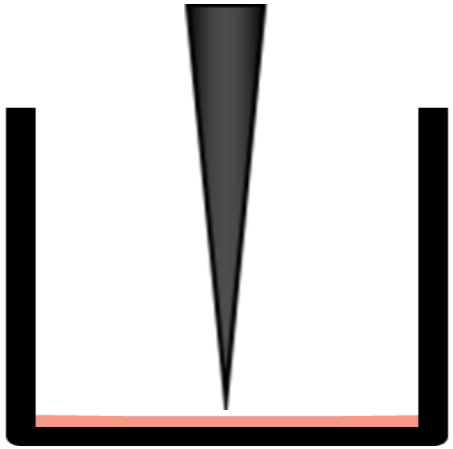
Speed Pitfalls

Use of the 96 well head for transfers from 96 well block to mixing plate and from mixing plate to cell plate

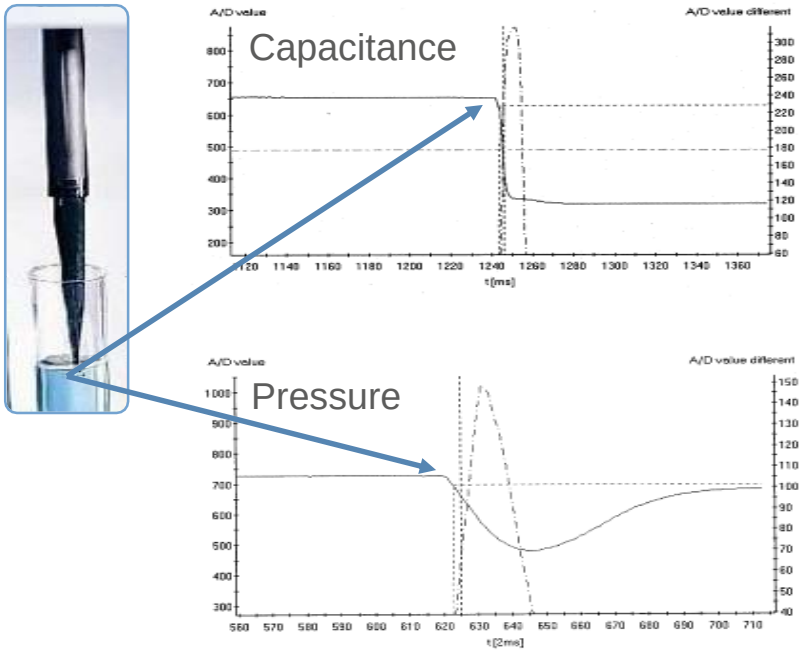


Larger volume tip for multi well dispensing when no mixing is required

Low Liquid Level Pitfalls



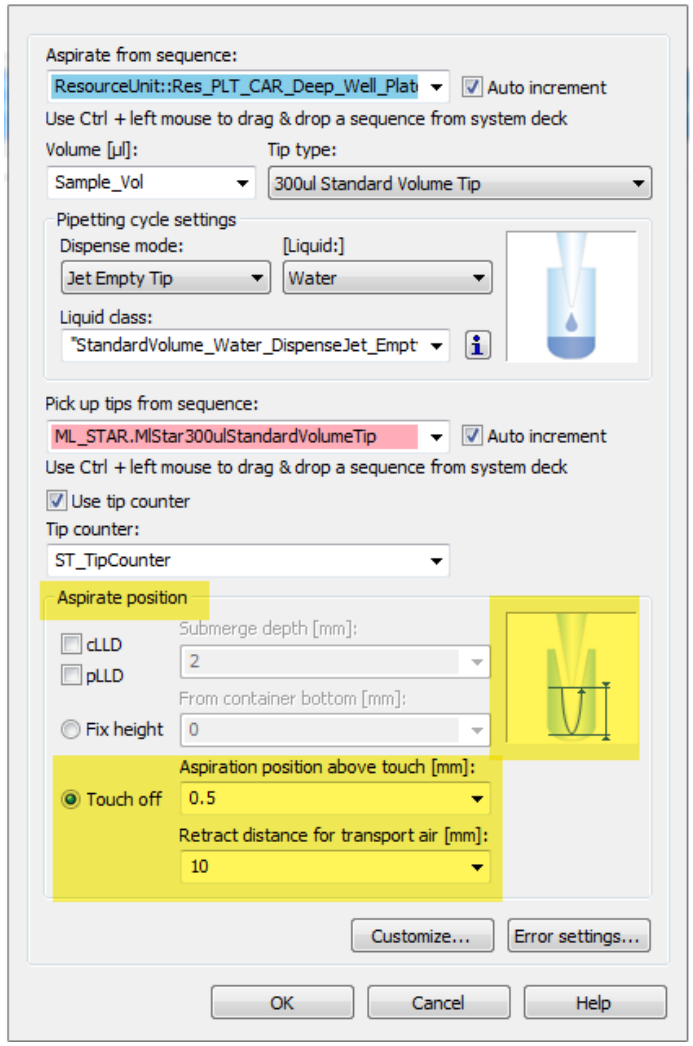
Insufficient volume to account for retained liquid on tips or reservoir/plate

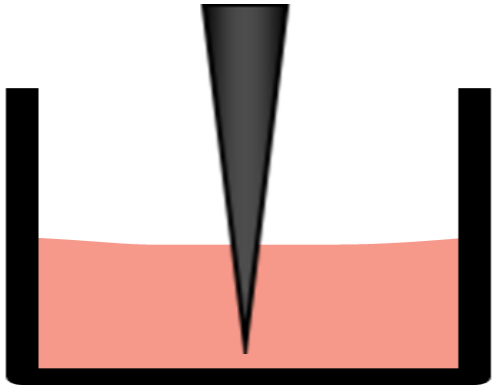


Conductivity detection errors through graphite tips to the reservoir/plate

Pressure sensing errors due to bubbles or low volumes

Low Liquid Level Corrections

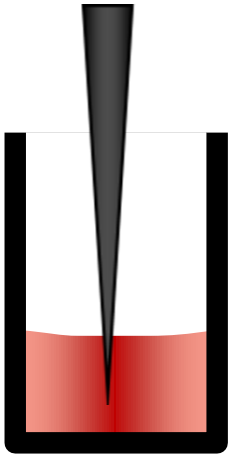




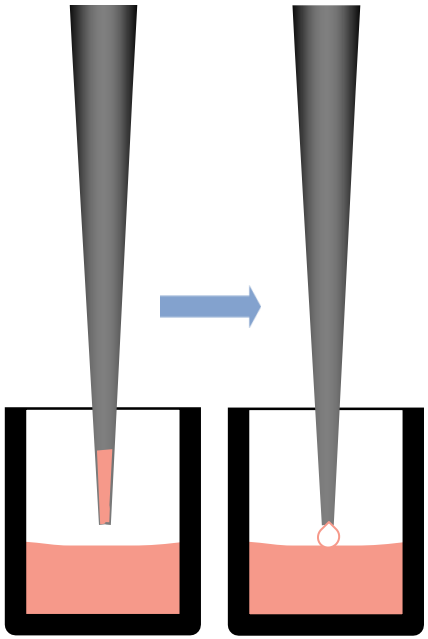
Adjusted 'dead volume'

Program "Touch off" to prompt the robot to touch the bottom of the well before pulling up the required volume

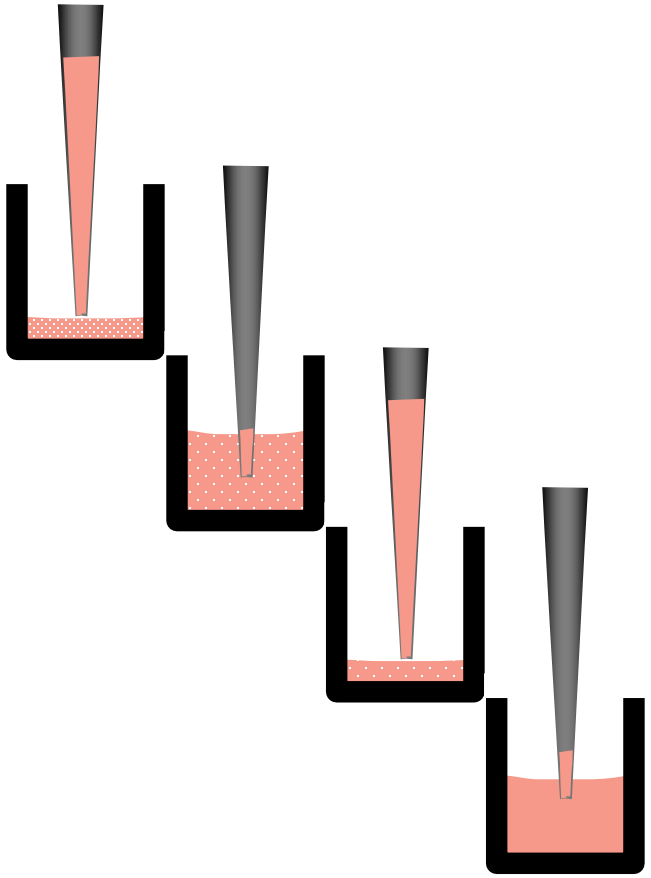
Pipetting Adaptations for Consistent Sample Preps



Slower aspiration when drawing up viscous fluid

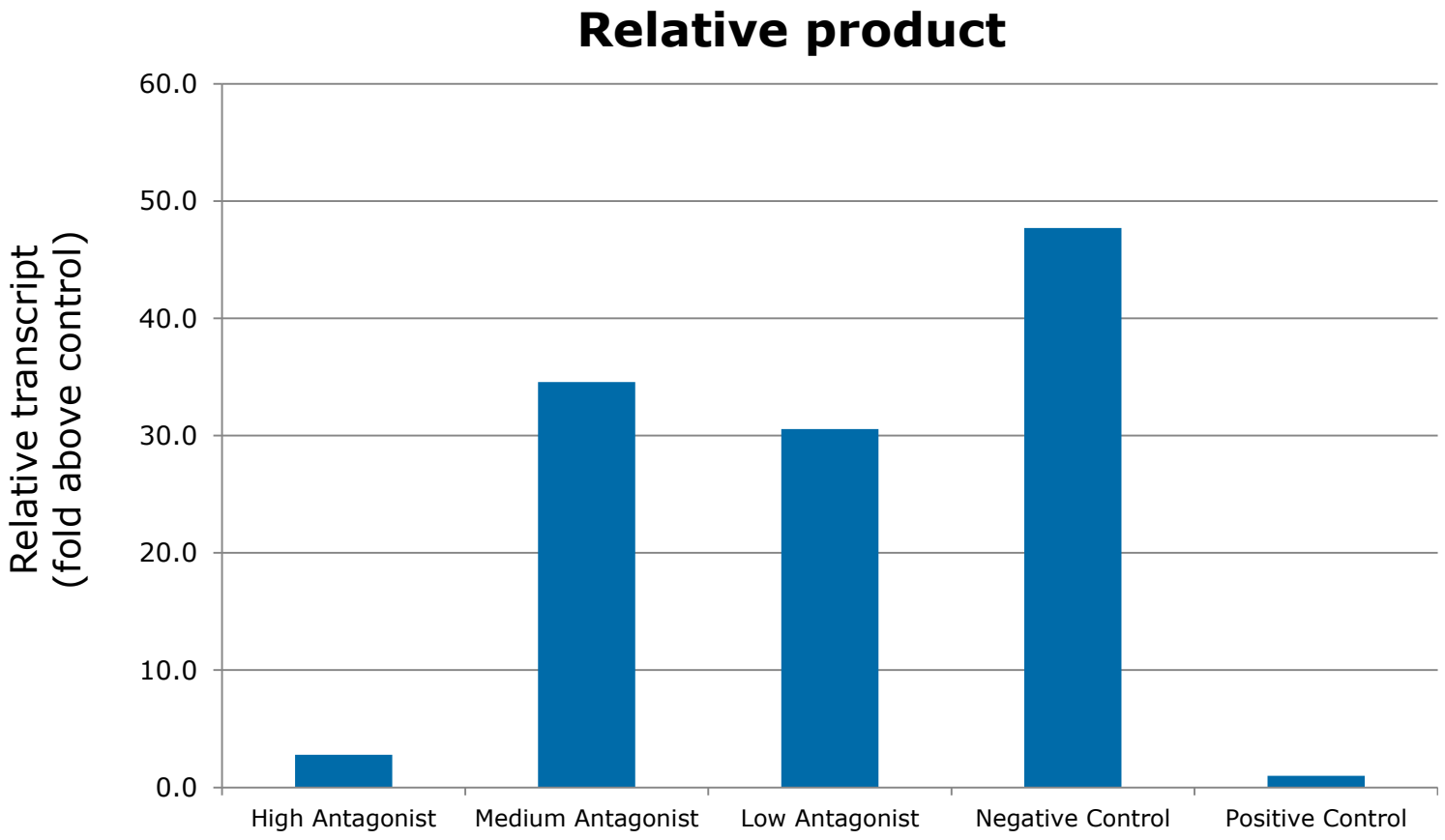


Program for 'blow out' of fluid when dispensing

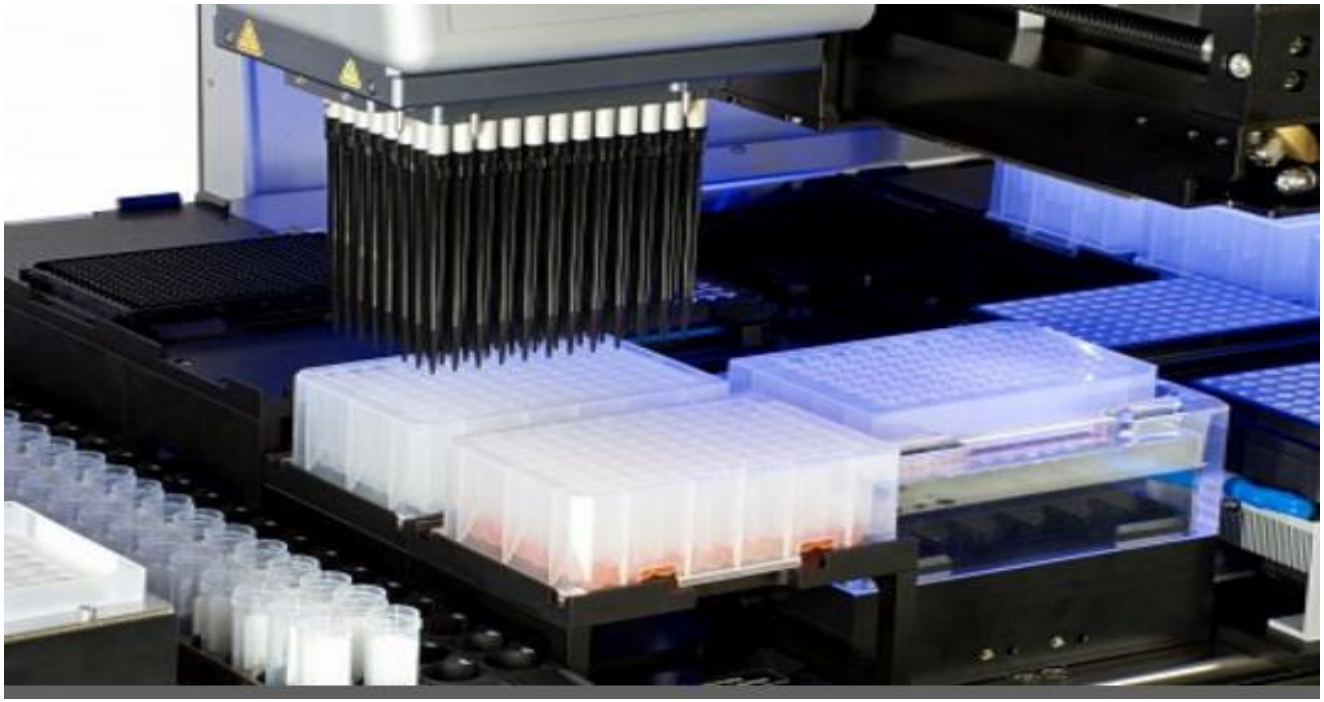


Additional mixing steps added

Second Attempt – Partial Success



Next Steps



- Aggregation challenges
- Mix immediately
- Tip Touch and pre-wetting

Case Study: Chemiluminescent cAMP Potency Assay

Cells

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Phosphodiesterase Inhibitor

↓

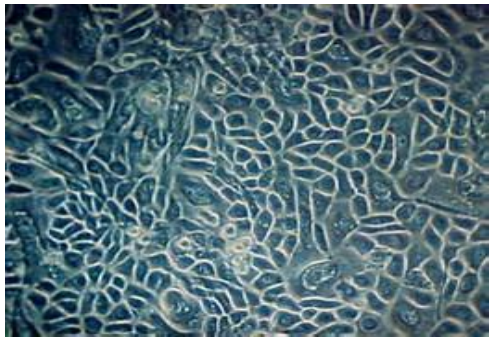
Standard/Sample dilutions

↓

Lysis Buffer

↓

Transfer to ELISA Plate



Add capture antibody

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Add detect antibody

↓

Incubate

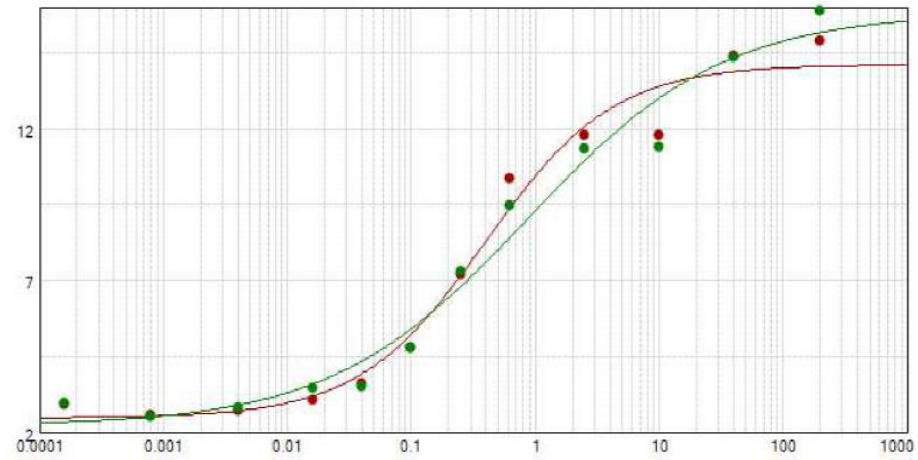
↓

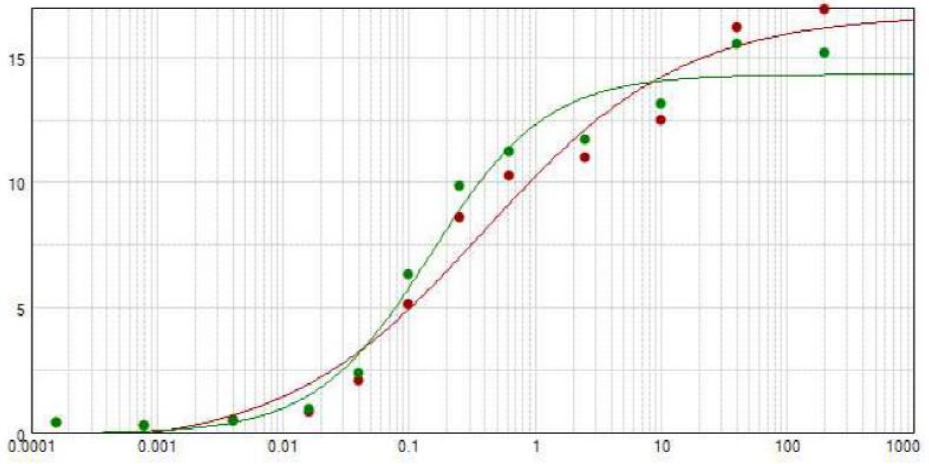
Wash 6x manually

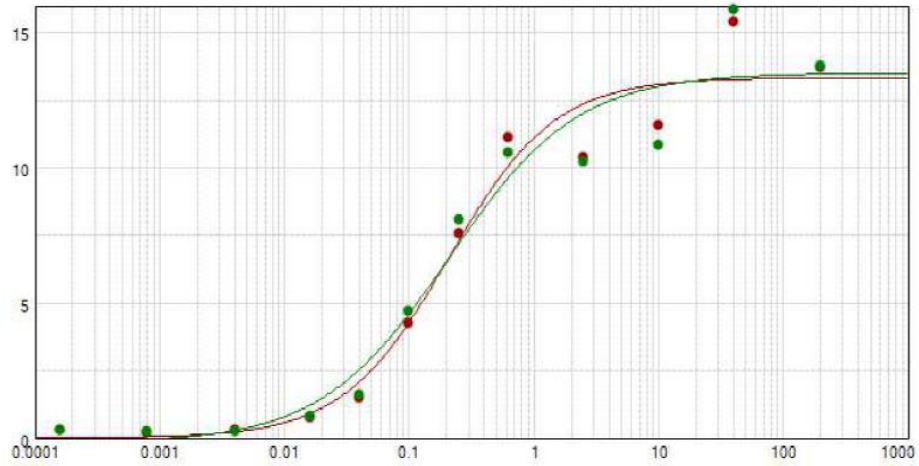
↓

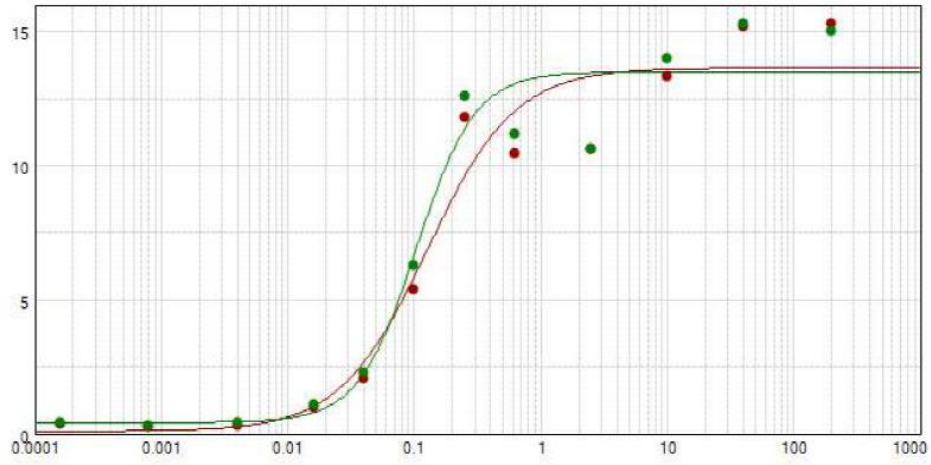
Add substrate

Manual Results







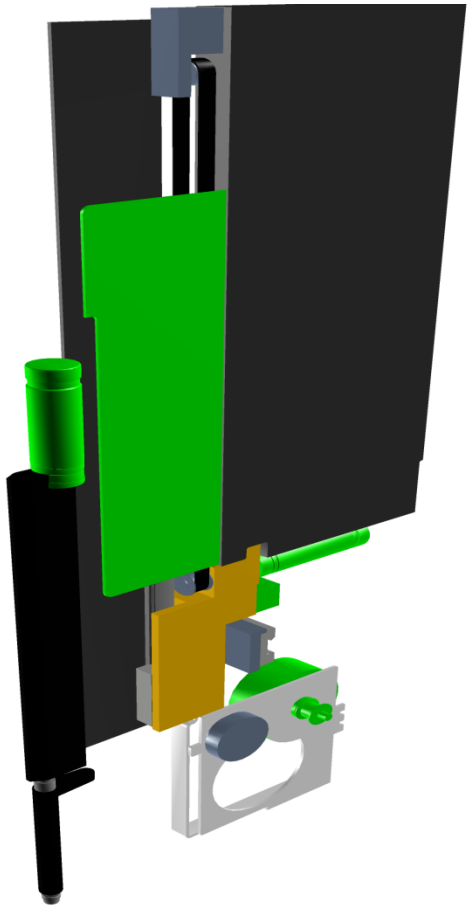


Difficult assay with manual transfers: 30% fail rates, and high sample load

Automation Friendly Assay Design: Can This Thing Be More Accurate and Precise?

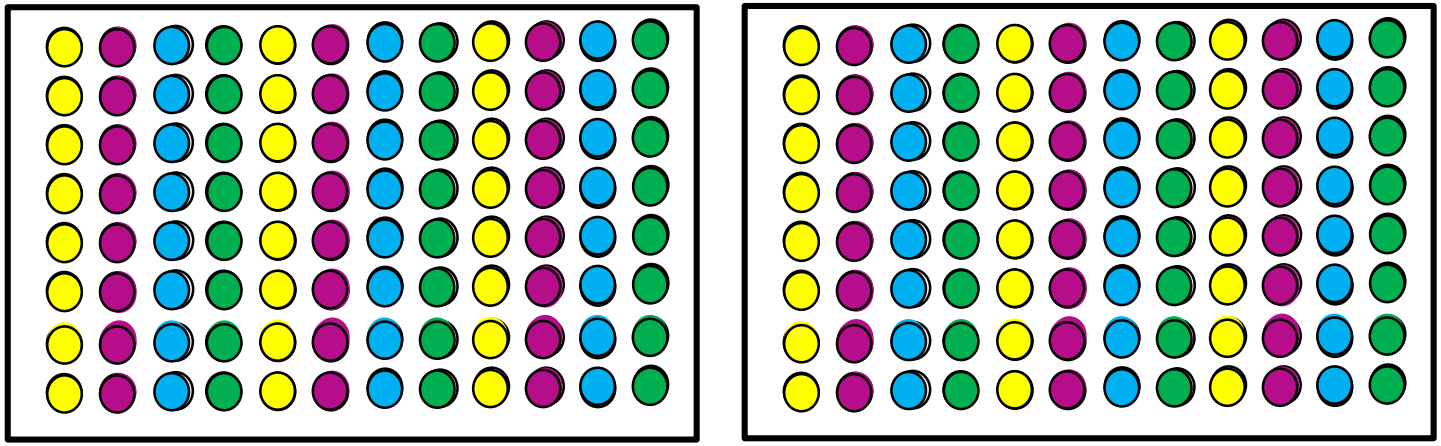
	1	2	3	4	5	6	7	8	9	10	11	12
A												
B	150 uL Smp	160 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	160 uL AB	160 uL AB
C	150 uL Ref	160 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	160 uL AB	160 uL AB
D	150 uL Smp	160 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	160 uL AB	160 uL AB
E	150 uL Ref	160 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	160 uL AB	160 uL AB
F	150 uL Smp	160 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	160 uL AB	160 uL AB
G	150 uL Ref	160 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	120 uL AB	160 uL AB	160 uL AB
H												

Single pipetting channel

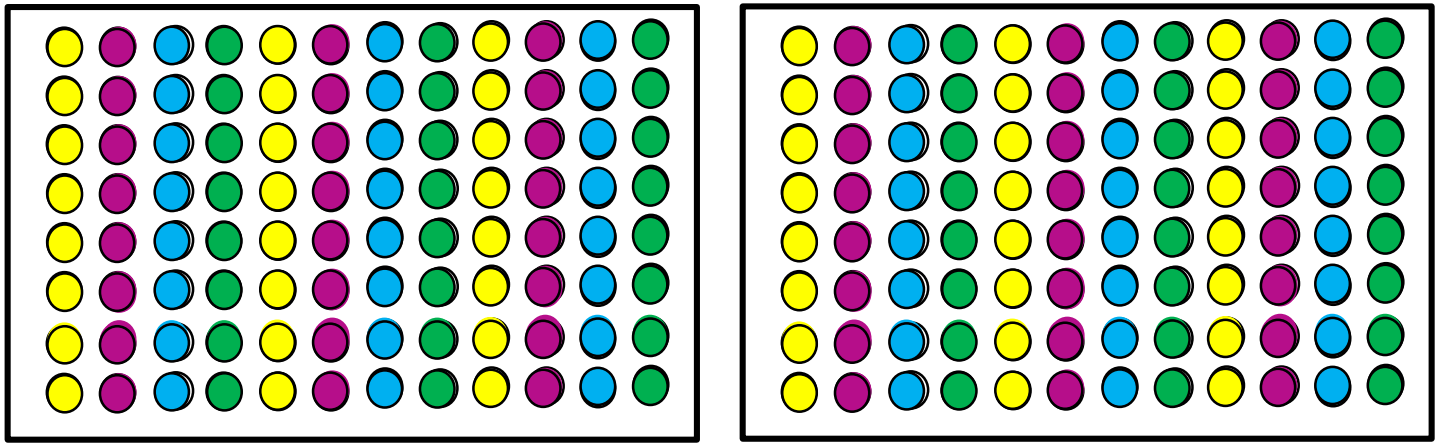


Automation Friendly Assay Design: Can This Thing Go Any Faster?

Column-to-column transfer (Manual or Robotic)

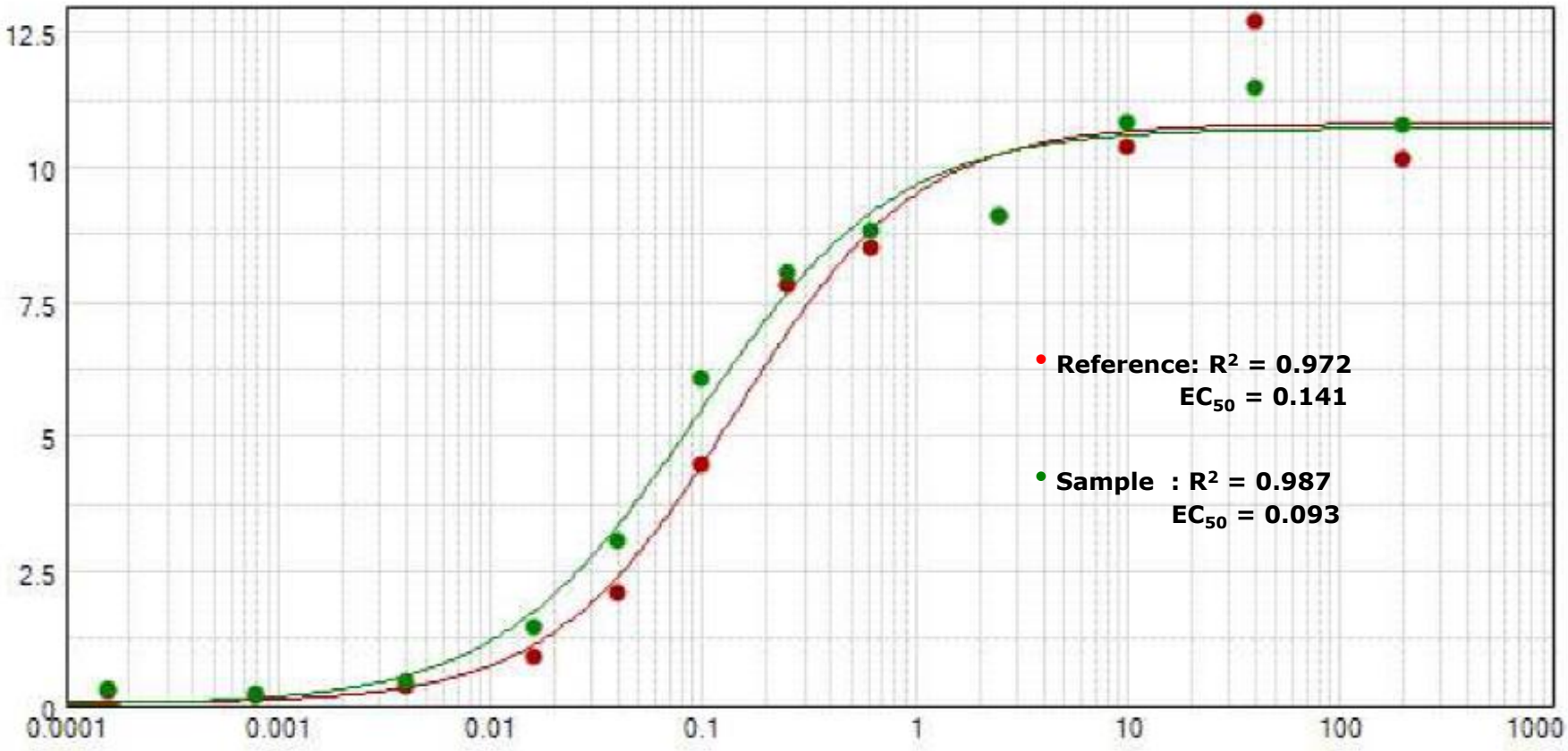


96-well Transfer (Robotic)



96 well
Probe Head

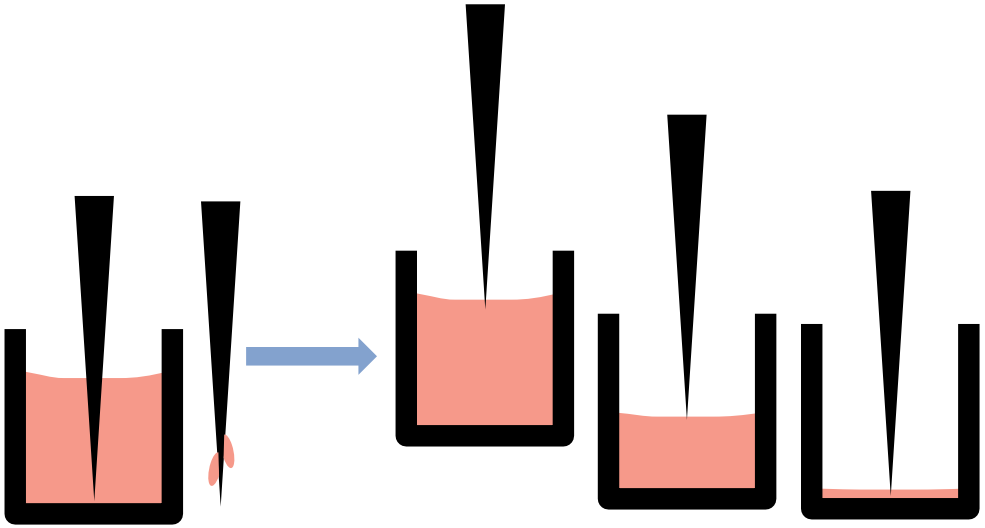
First Attempt



It’s all in the Delivery (and Removal)

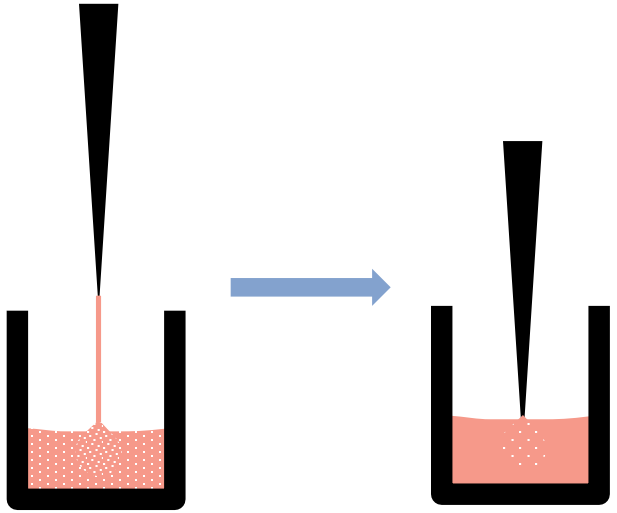
Variability and carryover

Switch to regular tips and aspirate to liquid level sensing

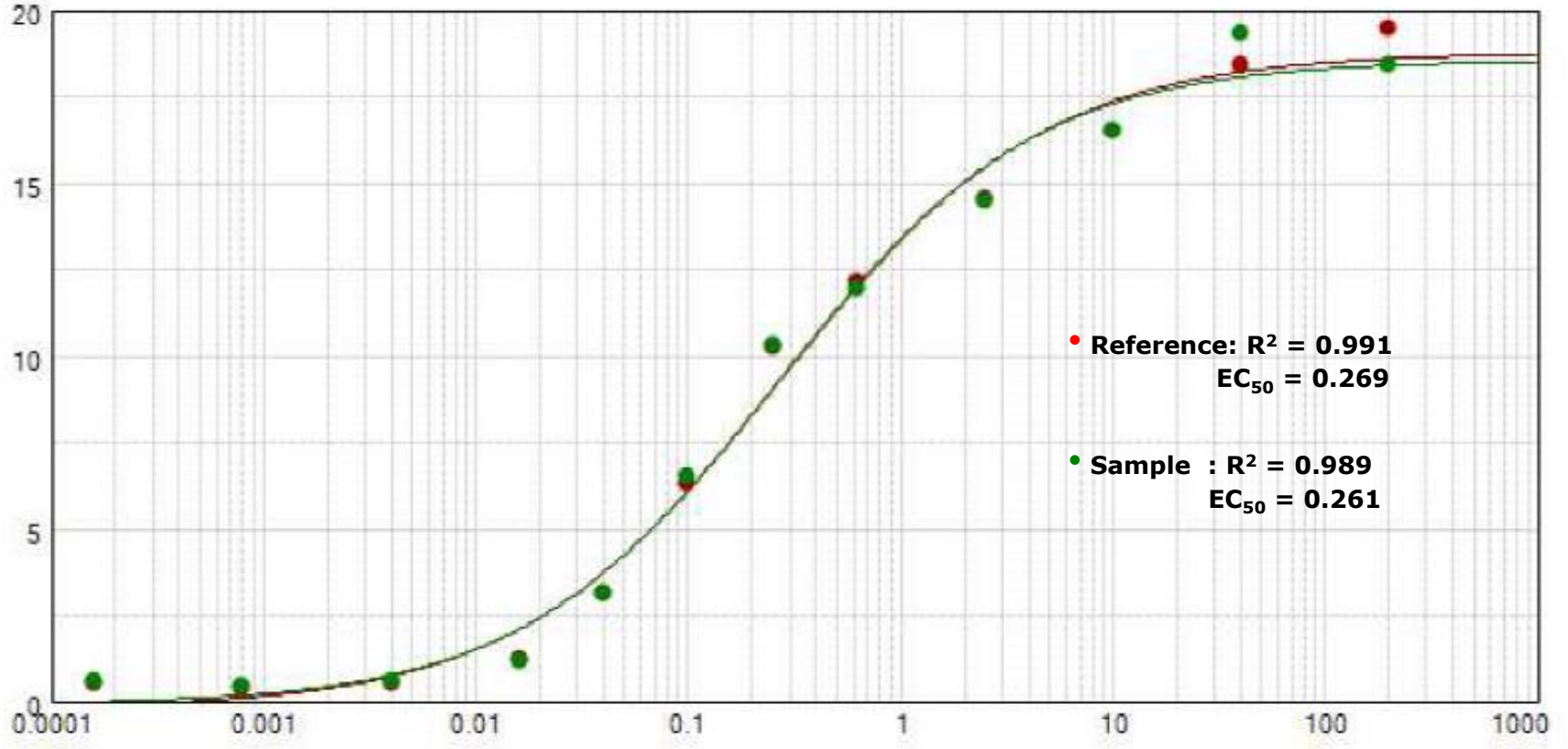


Bubbles in the plate

Adjusted addition speed and tip height



Current Process



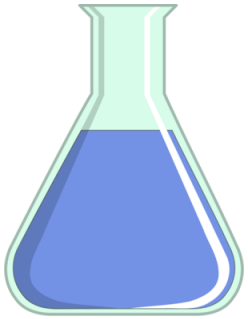
Next Steps

Discussions with Partner:

- Incorporate automation
- Method Revision
- Validation

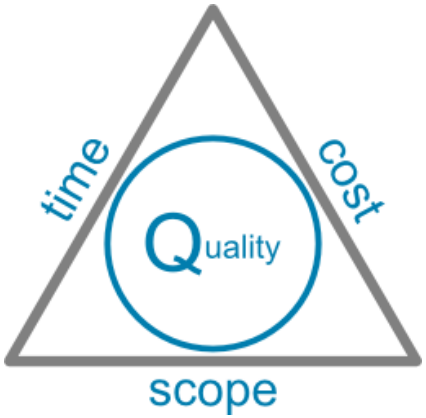
Lessons Learned

Engage early



Liquid Properties are Important

Aggregation, viscosity, density



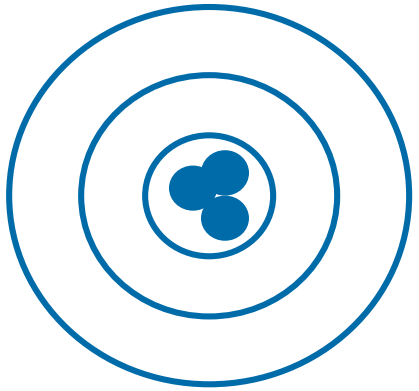
Time constraints

Conclusions

Automated instrumentation creates important advantages

Reduced risk of repetitive motion injuries

Maximize:
Output
Minimize:
Time and Effort



Reliable, Accurate and Precise

Acknowledgements

Catalent®

Catalyst + Talent.
Our name combines these ideas.

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