Leveraging computational design, miniaturization, and microfluidics to accelerate the discovery and engineering of therapeutic proteins

Mark Ibrahim on behalf of Merck Biologics Discovery and Engineering

Affiliations: Merck & Co., Inc., Boston, MA, USA

June 25, 2024

Merck Research Labs Mission

To translate breakthroughs in fundamental biomedical research into meaningful new therapeutics and vaccines that improve and extend the lives of people, worldwide.

We're conducting R&D to address some of the world's most urgent global health challenges.

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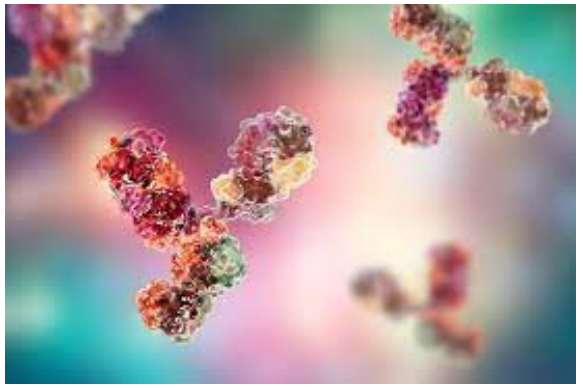
Discovery Biologics – Mission Statement

Deliver therapies that span from conventional biologics to emerging platforms in bioconjugates, engineered proteins, and cell therapies.

Utilize the **deep scientific expertise** and **differentiated capabilities** across our network to **collaboratively invent impactful medicines** for patients.



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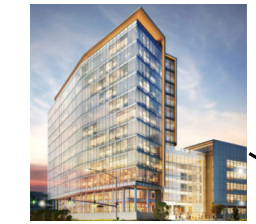
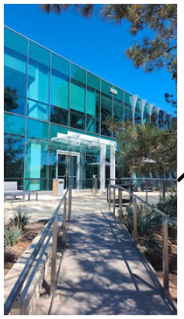
Discovery Biologics Locations



San Francisco, CA

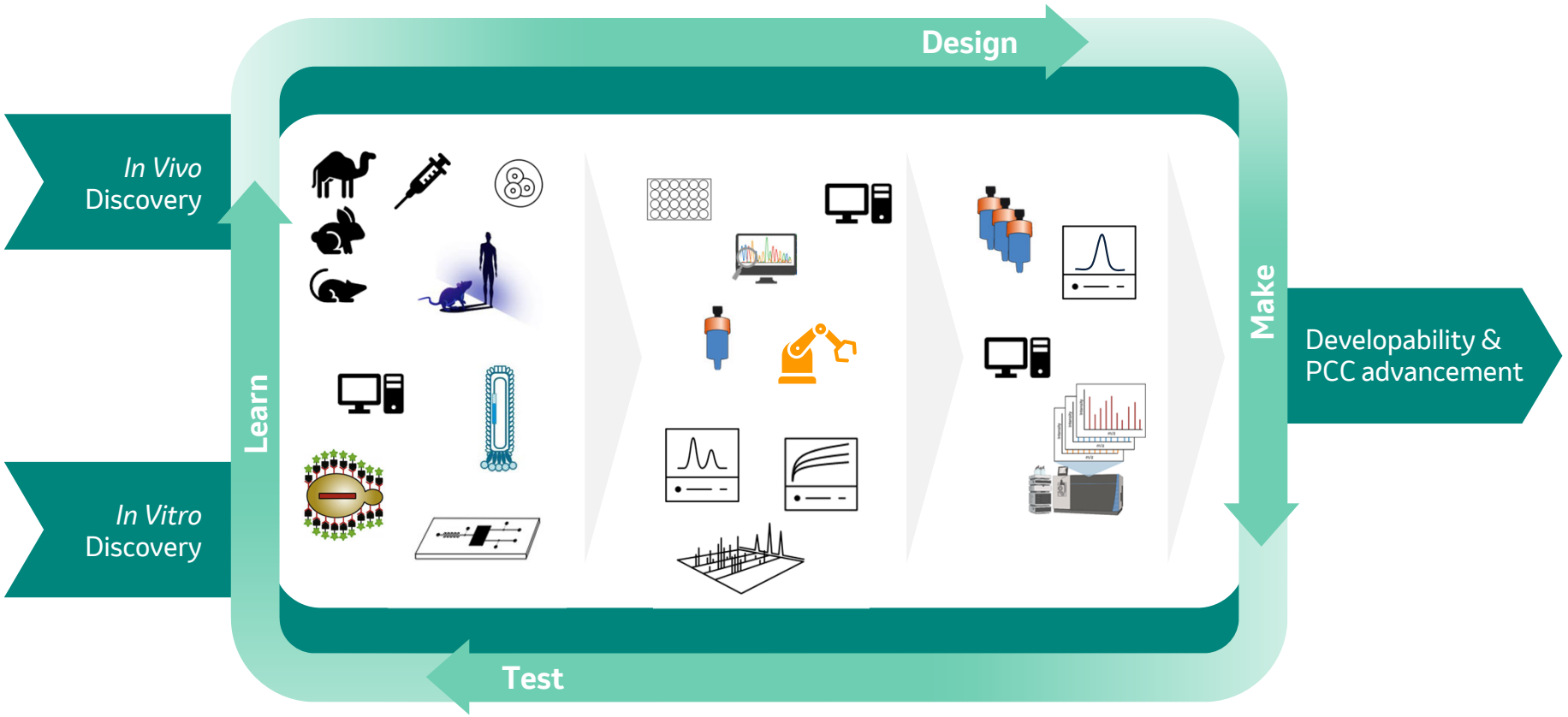


San Diego, CA

Boston, MA

Cambridge, MA

Discovery Biologics Research in Practice



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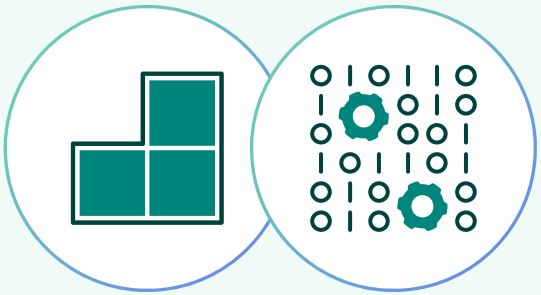
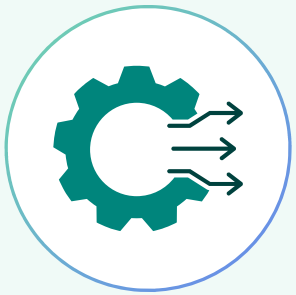
Enhancing our discovery process using three connected priorities



Generate

Build new and enhance current material and data generation capabilities



Integrate

Bring together internal and external data to enable seamless data access for all

Predictive Design

Using predictive modeling to inform and de-risk our decision-making

Physical and digital automation supports all three priorities!!

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Public

Accelerating the Discovery Biologics end-to-end workflow

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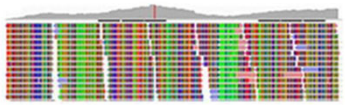
uHTP/HTP function & binding assays



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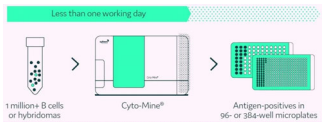

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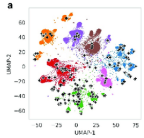

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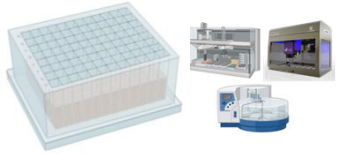

Micro-scale production

HTP characterization

φ BioPhi

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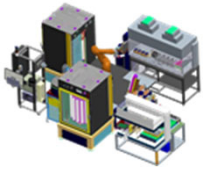
Lead Engineering
1 - 10 sequences



Mid/large scale production

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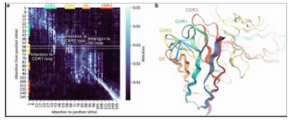
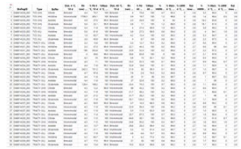


ML-guided liability correction, engineering, humanization



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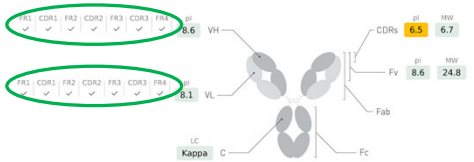

PCC!

Physical and digital automation to enable all of these efforts

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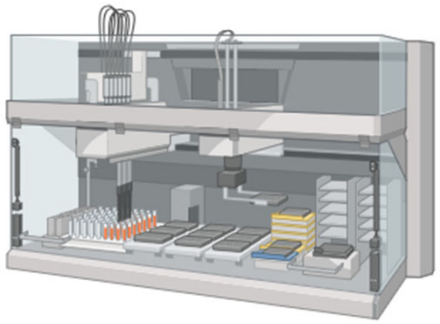
Plated HTP workflow can express and purify 100s of proteins in 5-days

D 0
- DNA Mixing

TECAN

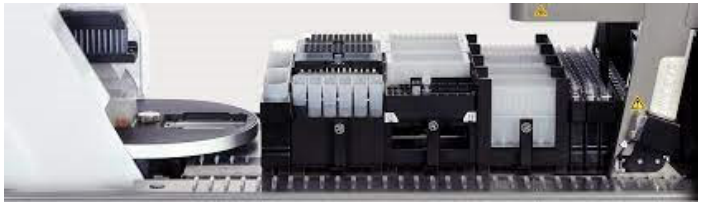


D 0 / D 1
- Transfection / Feed

LYNX

D 5
- Purification

KingFisher / Hamilton



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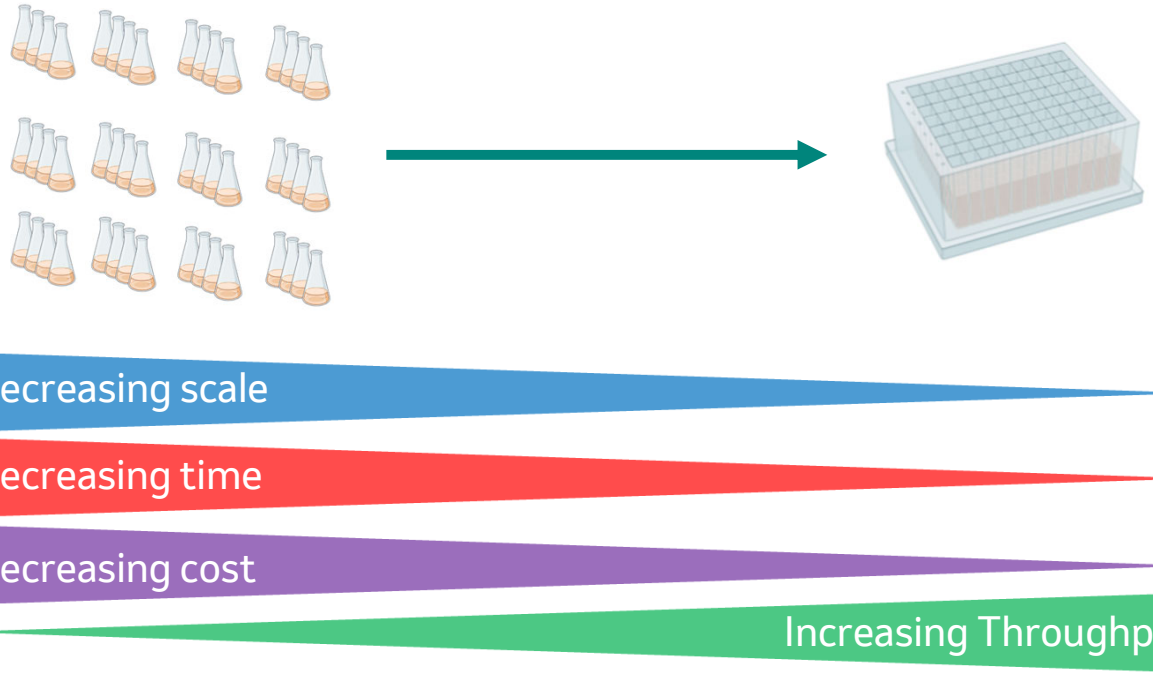
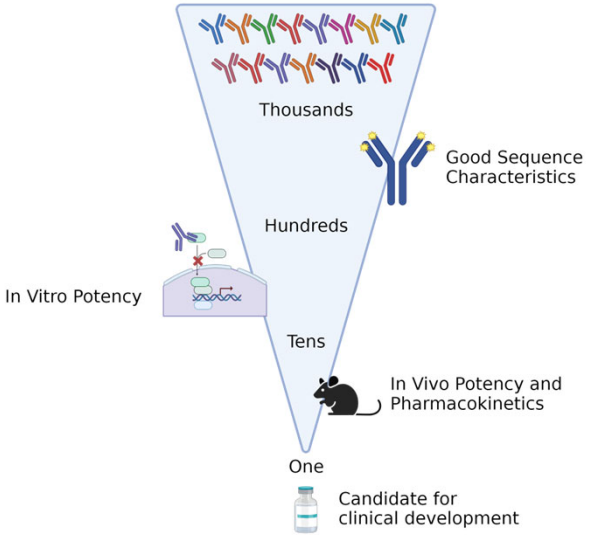
Decreasing scale increases throughput while saving time and money





Microfluidic technology to enable uHTP binding

Biacore T200
3/cycle
High-sensitivity

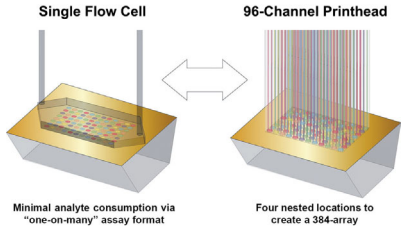
Biacore 8K
8/cycle
Med/high-sensitivity

Octet HTX
96/Cycle
Low-sensitivity

Carterra LSA
384/Cycle
Med/high-sensitivity

Printhead uses flow micro-spotting technology that enables:

- Screening of up to 384 binding interactions simultaneously
- Binding results in substantially less time
- Limited sample consumption



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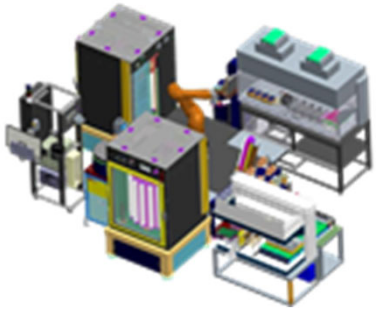
HTP binding-Translation across scale and purity

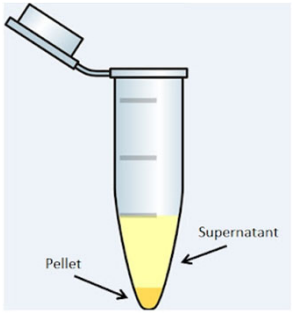
- Improvements to **upstream** workflows require improvements to **downstream** workflows:
  - Characterize **hundreds of molecules** for binding
  - **Limited** sample consumption
  - **Generate accurate** kinetic data in “screening mode”

Large-scale purification



Small-scale purification

Crude mAbs in supernatant



- High-throughput binding technology was leveraged to compare binding affinities of ~150 antibodies produced in different expression systems

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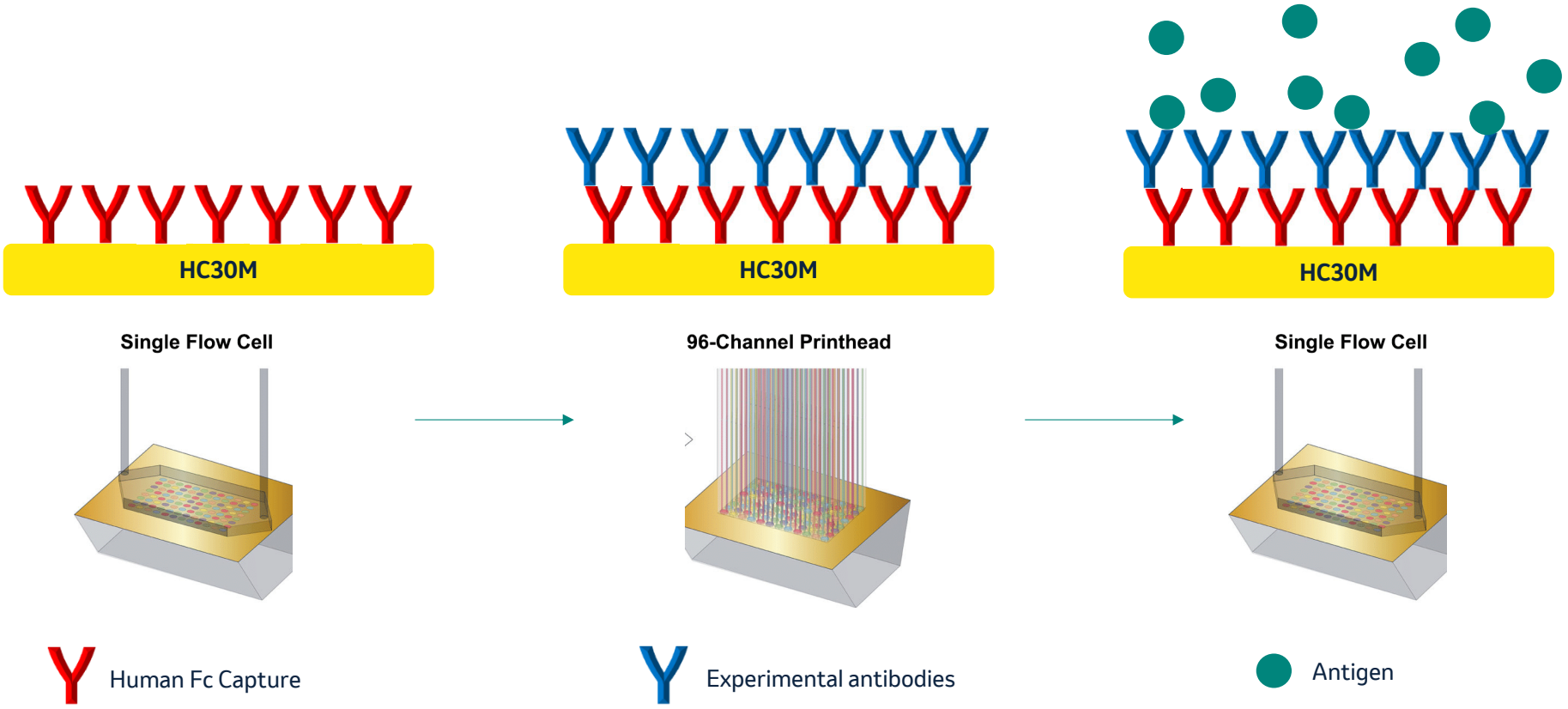
HTP binding method enabled through use of 96-channel printhead



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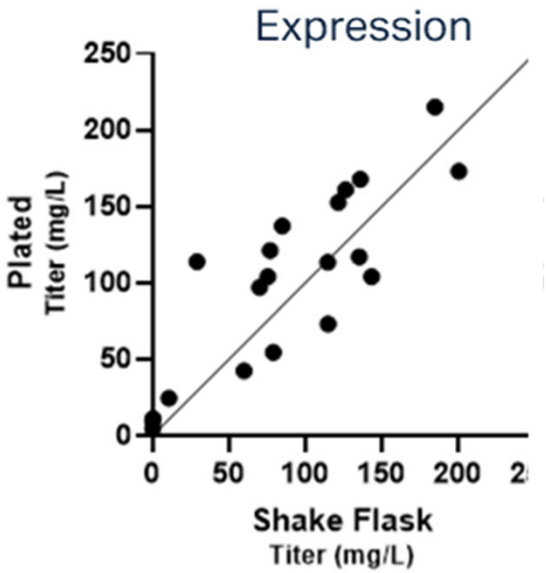
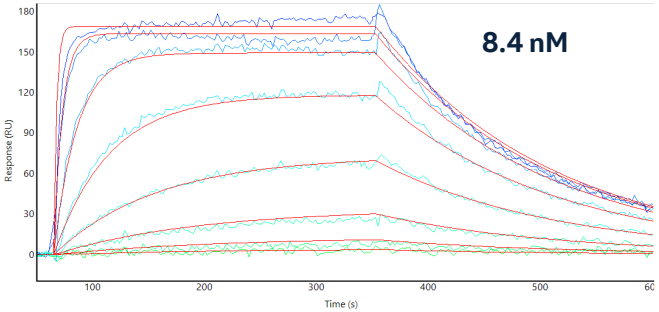
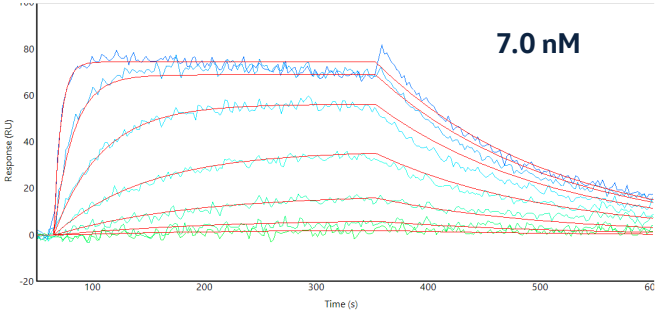
HTP binding is comparable across scale and purity

- Antibodies exhibited comparable expression levels between plate-based production and shake-flask production
- Evaluated ~150 mAbs, ~>90% comparability in binding between expression formats
- Entire experiment used ~3 ug per mAb and ~30 ug antigen
- 24-hour experiment run time





Large-scale purified clone



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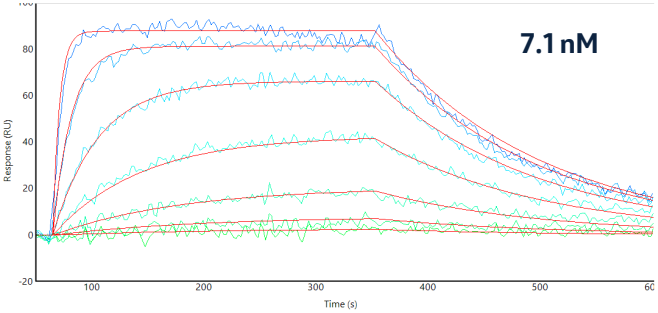

Supernatant

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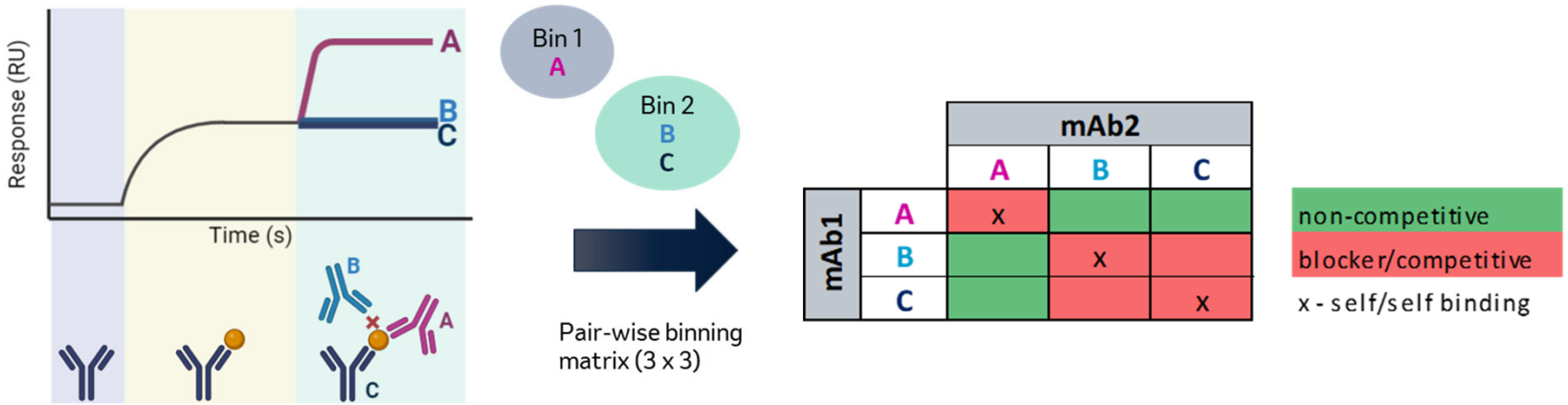
HTP epitope binning-Finding the right binders

- Epitope binning provides crucial information regarding antibody diversity
- Specific “bins” might be more desirable due to antibody functionality to that epitope
- Ability to select antibodies that bind to desired bins early in screening process greatly improves workflow
- High-throughput binning was leveraged to characterize **~200** VHH-Fcs for binding to functional epitope



HTP SPR used to support 203 x 203 binning in single experiment

Octet HTX
203 x 203 binning

- >1mg / mAb (500nM)
- >1mg antigen (200nM)
- >80 expts, 3-month instrument run-time
- >50 trays of sensors
- Daily setup required
- Highly complicated plate setup

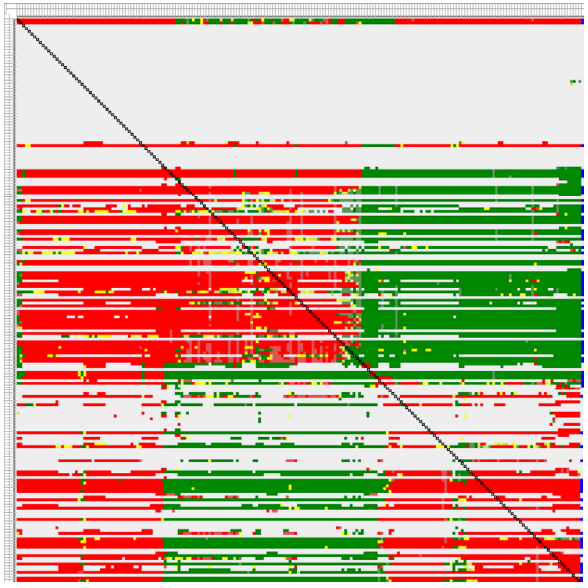
Carterra LSA
203 x 203 binning

- 15ug / mAb (200nM)
- 370ug antigen (200nM)
- 1 single experiment, ~6 days continuous instrument time
- 1 sensor chip
- Walkaway
- Simple setup – a single 384-well sample plate

Process Improvement

- ~70-fold less antibody used
- ~3-fold less antigen used
- ~data generated 15X faster

Epitope binning heat map



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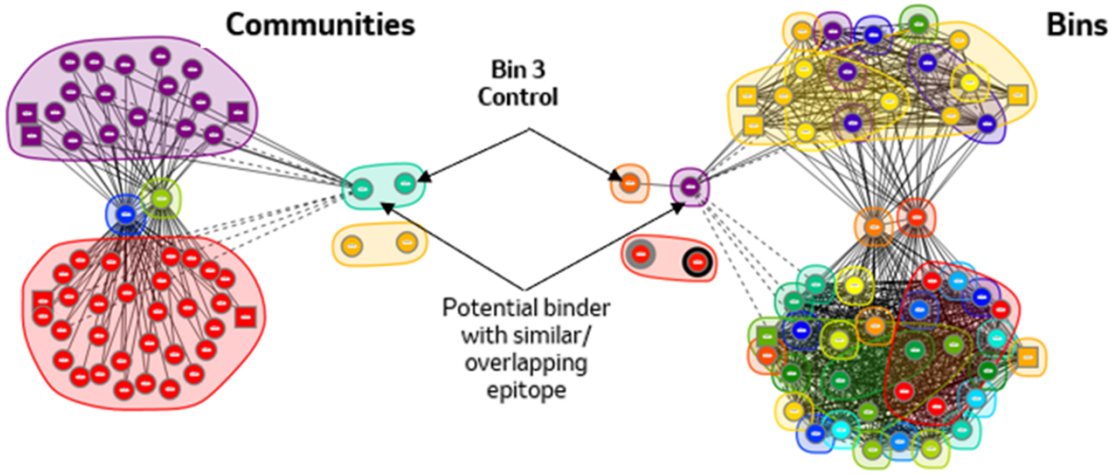
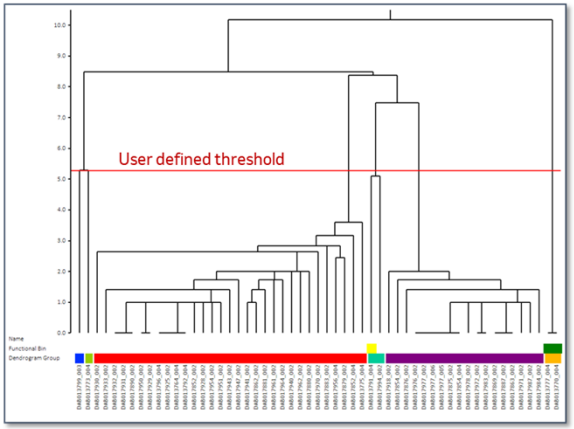
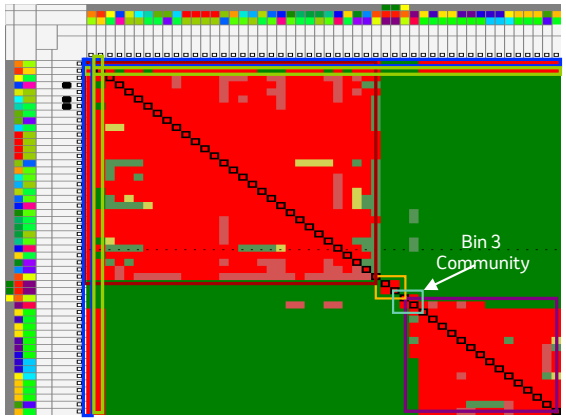
Experiment identified potential new functional binders in a difficult to target epitope







6 communities identified

40 unique bins identified

Microfluidic technology enabled the identification of *potential* new functional binders for a rare epitope bin in <1 week with minimal setup time and minimal sample consumption

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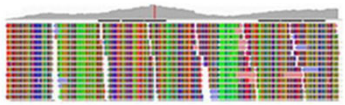

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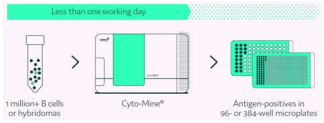

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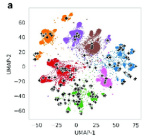

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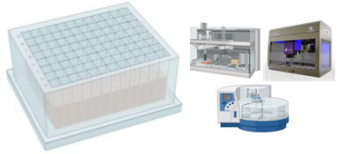

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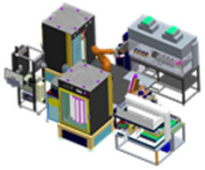

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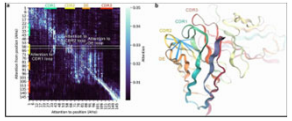

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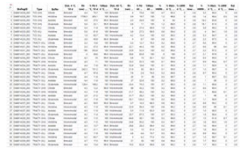

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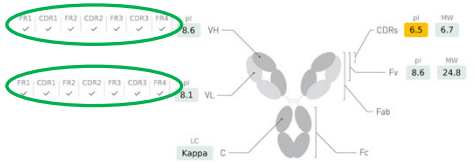

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