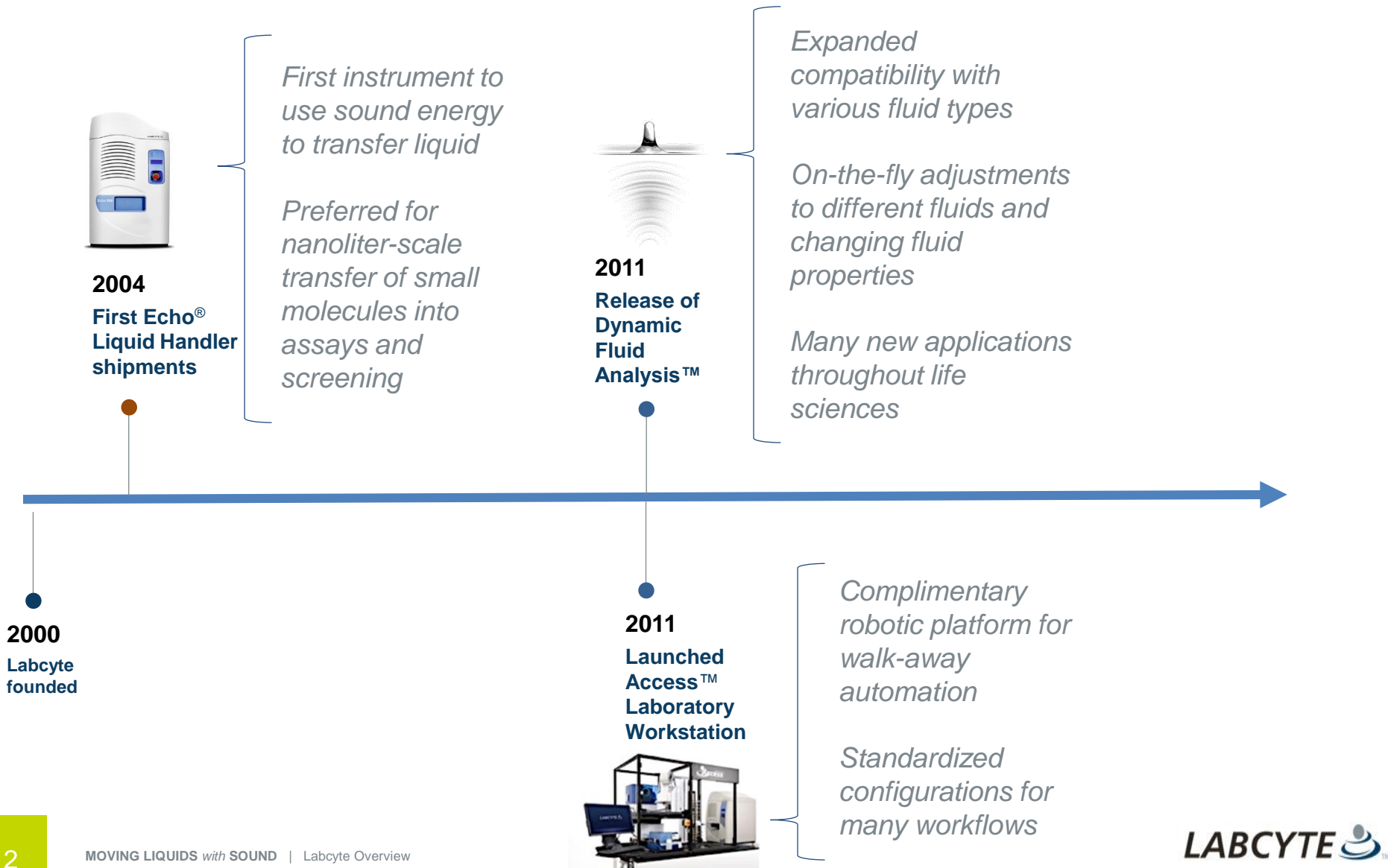


Use of Echo[®] Liquid Handlers to Improve the Efficiency and Reliability of Bioassay Laboratories

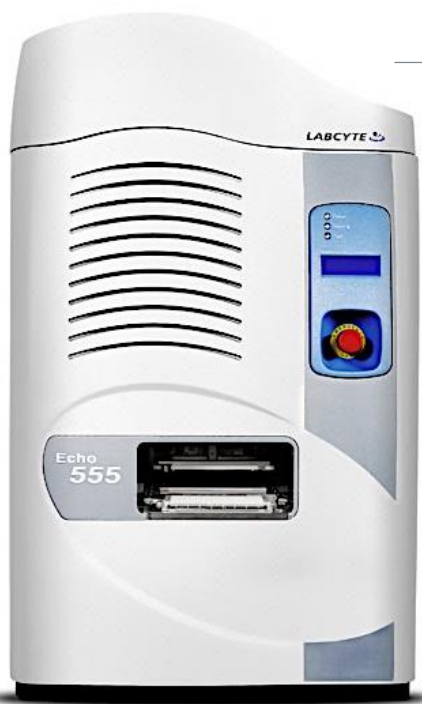
Tim Allison,
May 8, 2017

Corporate History



Echo[®] Liquid Handler Models

Non-contact, Contamination-free Acoustic Liquid Handling



Echo[®] 555 / 550 Liquid Handler

- ▶ 2.5 nL increment
- ▶ Flow rate:
0.25 – 1 μ L/sec
- ▶ Ideal for reactions of
0.05 – 1 μ L
- ▶ Supports 384 and
1536-well source
plate formats
- ▶ ***Used for transfer of
small molecules for
high throughput
screening***



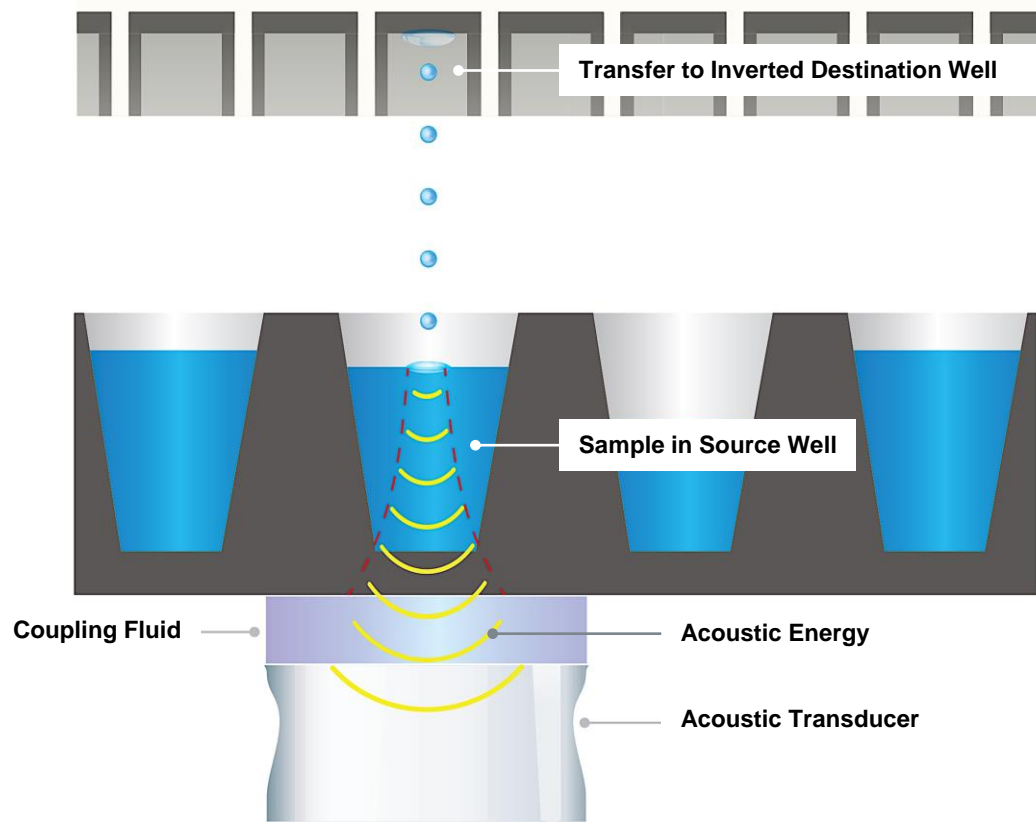
Echo[®] 525 Liquid Handler

- ▶ 25 nL increment,
aqueous only
- ▶ Faster flow rate:
3 – 6 μ L/sec
- ▶ Ideal for reactions
of 0.2 – 10 μ L
- ▶ Supports 384-well
source plate format
- ▶ ***Used for transfer
of samples and
reagents for
genomic
applications***

Echo Liquid Handling Technology

Moving Liquids *with Sound*

- ▶ **Rapid and Flexible**
 - 200 – 500 droplets / second
 - Any well to any well
 - Automatically adjusts when fluids change
- ▶ **No physical contact**
 - Low energy transfer
 - Perfect sample integrity
 - Consistent drop size, 2.5 nL or 25 nL
- ▶ **Revolutionary**
 - Improve accuracy and precision
 - Reduce volumes and costs
 - Eliminate washing and pipette tips
 - Eliminate potential for cross-contamination



Echo Liquid Handling Technology

A View Inside the Echo



Key Features and Benefits



Small Volumes
Typical CVs <5%, Spec 8%



Cost Savings
Assay Miniaturization
Reduction of tips and plates
Conserve precious samples/libraries

Non Contact
Does not lyse cells,
denature proteins, damage
DNA



Improved Data Quality
Tighter CVs observed in assays
No potency loss due to adherence to tip
No cross contamination (qPCR)
No carryover error

Any Well To Any Well
No fixed tip constraints



Workflow Optimization
Direct Dilution vs. Serial Dilution
High Speed Cherry Picking
High Speed Normalization and Pooling
Minimize intermediate assay steps
Avoid positional artifacts by optimizing
placement of controls & replicates

Broad Range of Applications

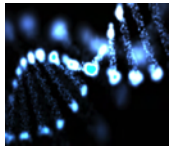
Used in life sciences, research, drug discovery and personalized medicine

Genomics

Translational

Drug Discovery

Synthetic Biology



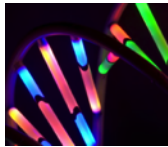
Increase efficiency and speed while reducing costs

Sequencing



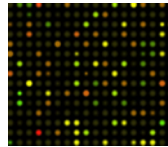
Low cost, highly efficient library preparation

Epigenetics



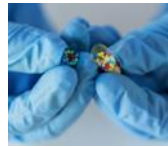
Enabling high-throughput epigenetic screening

Gene Expression



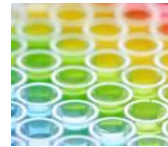
Cost-effective, high-throughput RT-qPCR

Precision Medicine



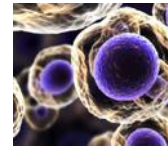
From personalized medicine towards precision medicine

HTS / Secondary Screening



Discover the right drugs with improved transfer performance

Cell-based Assays



Biologically relevant assays with unmatched data quality

Biochemical Assays



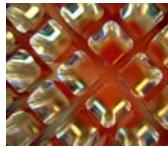
Simplify assay workflows with precise reagent transfers

ADME-Tox



Enable cost-effective, earlier safety screening

Combination Screening



Increase efficiency and speed while reducing costs

► Results

- Better experimental data at a fraction of the cost
- New drugs discovered
- Cost effective approaches to patient sample targeted therapy
- More efficient diagnostic development and testing programs

Echo® Acoustic
LIQUID HANDLING

The Future of Science is
Sound



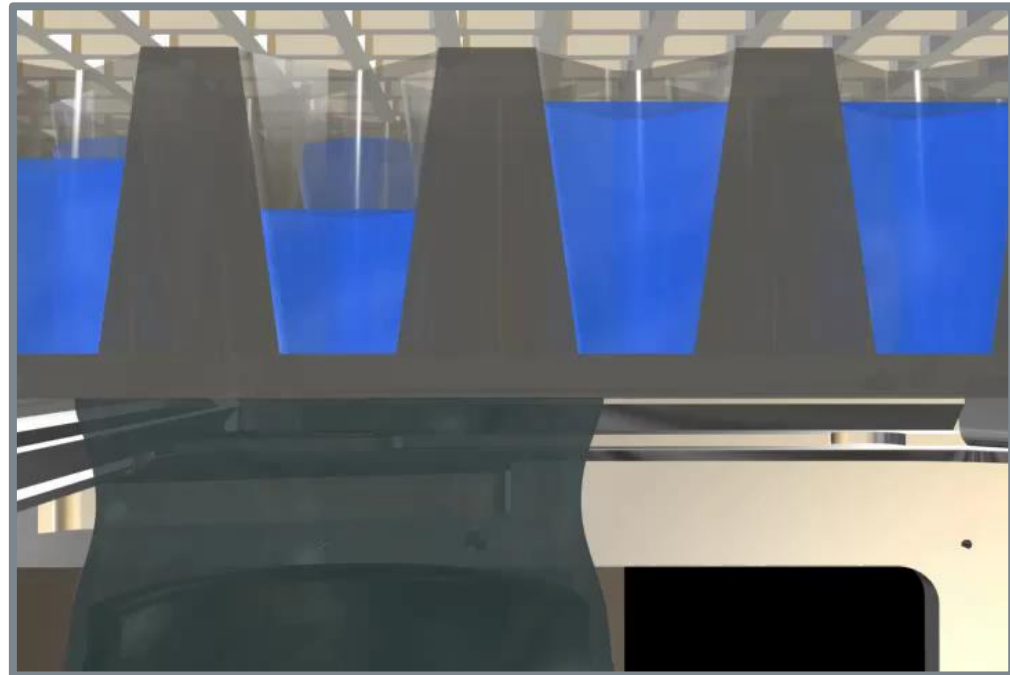
Unique Capabilities

of Echo Liquid Handlers

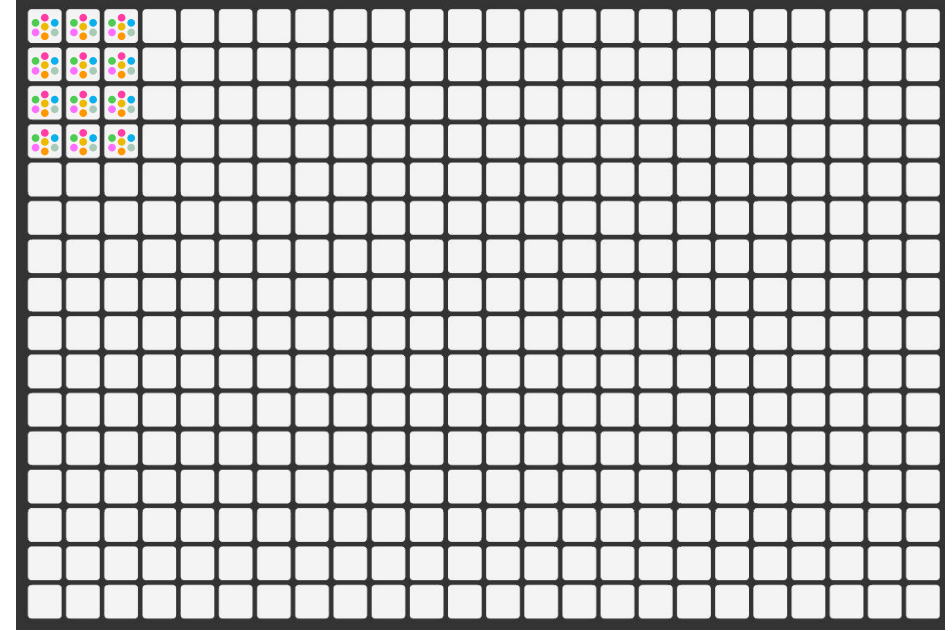
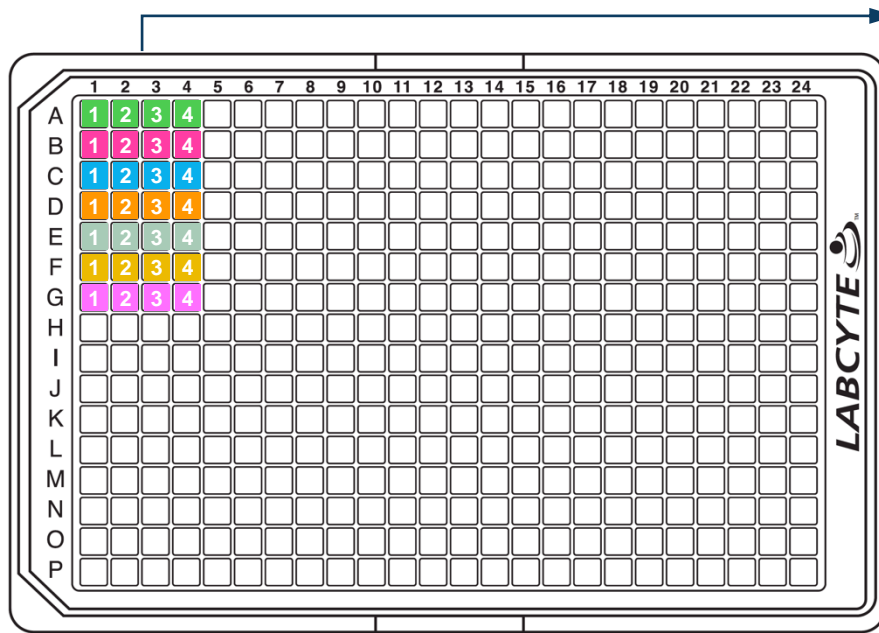
Acoustic Dynamic Fluid Analysis™

On-the-fly analysis of Fluid Properties to Ensure Precision, Accuracy

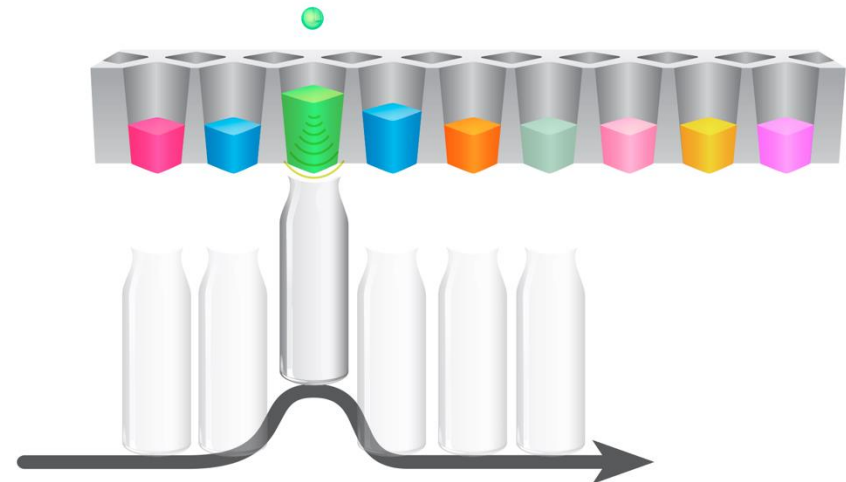
- ▶ **Automatically adapts to fluid properties and adjusts in real-time**
- ▶ **Handles a wide range of fluids, including:**
 - Cell lysates
 - Various reagents and concentrations
 - Glycerol storage solution
 - DNA, RNA, proteins
- ▶ **No operator intervention, no user calibration**
- ▶ **Reliable fluid transfer enables miniaturization with no loss of data quality**
 - Lower cost
 - Higher throughput
 - More accurate data



Any-Well to Any-Well Acoustic Transfer



- ▶ **50-100X Faster vs. Tips**
- ▶ **Many to One: Pooling**
 - NextGen Sequencing, Combination Screening
- ▶ **One to Many: Precious Samples or Reagents**
 - qPCR Primers, Assay Development / DOE



The Future of Science is
Sound



Bioassay Applications

for Echo Liquid Handlers

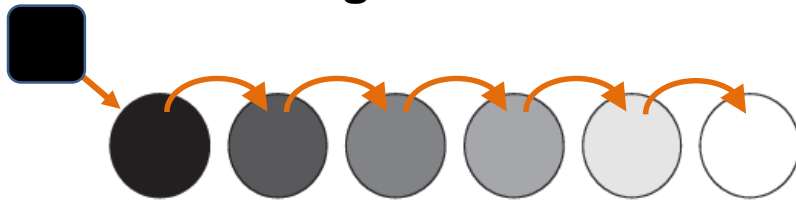
Improve Assay
Precision
Reproducibility
Repeatability

Acoustic Direct Dilution

Eliminate errors associated with serial dilution

Serial Dilution

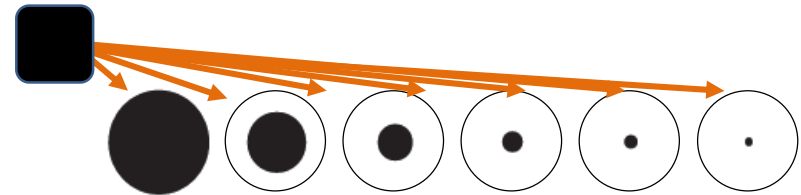
- ▶ Transfer same volume of decreasing concentrations



- ▶ Sequential dilution can allow for error propagation
- ▶ Many “touches” with tips
- ▶ Significant potential for carryover or leachates

Direct Dilution

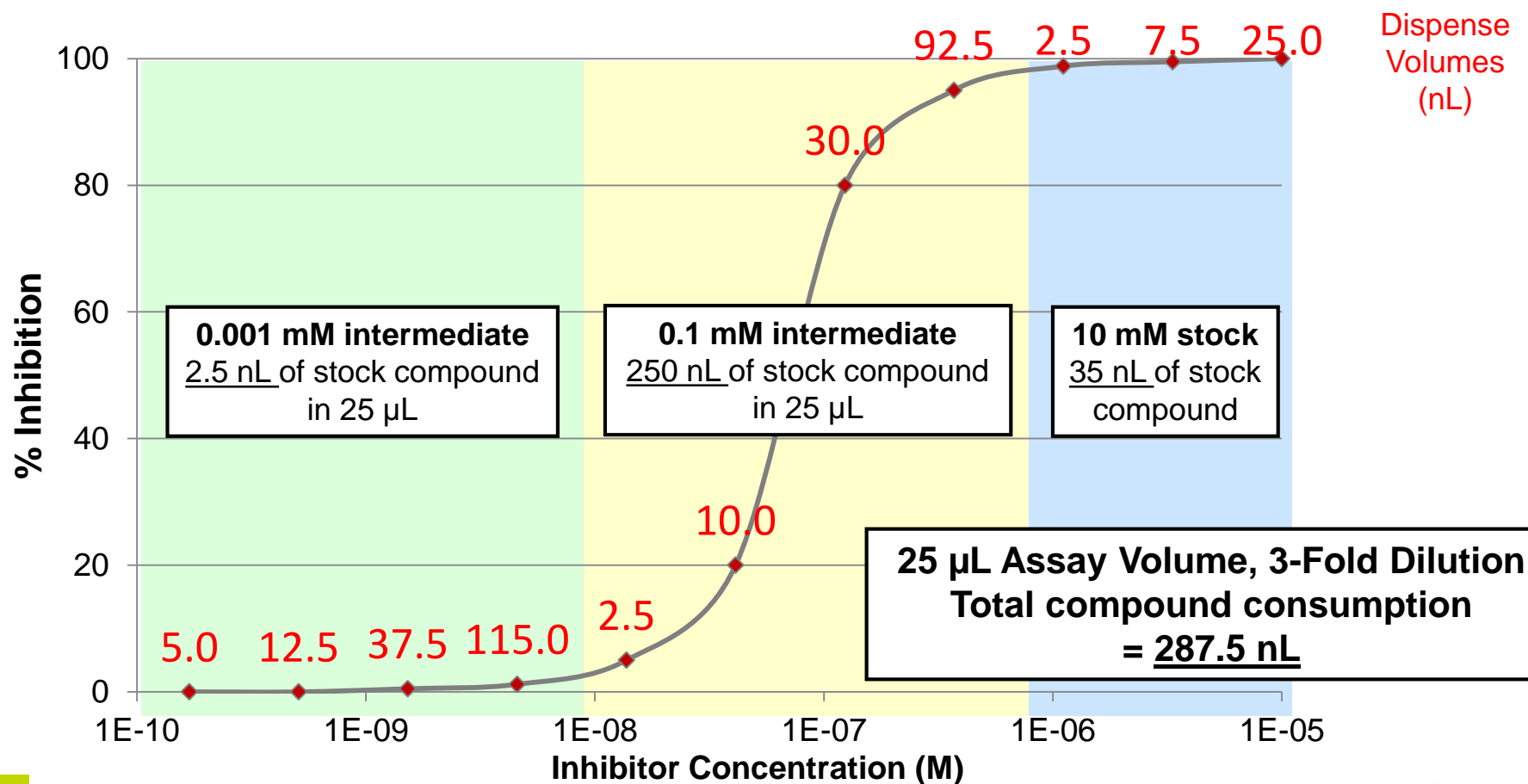
- ▶ Transfer decreasing volumes of same concentration



- ▶ Fewer dilution steps needed with greater precision
- ▶ Touchless—no carry-over, leachates or binding
- ▶ No solute lost

Potency Assays with Direct Dilution

11-point dose-response curve with < 300nL of sample



Potency Assays with Direct Dilution

Customer Example

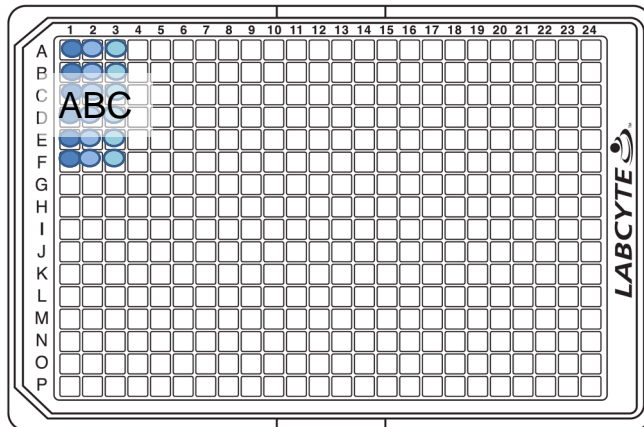
See Poster P119 from Amgen:
Cameron Cunningham, Jill Crouse-Zeineddini

Comparison of manual vs Echo-based dilutions

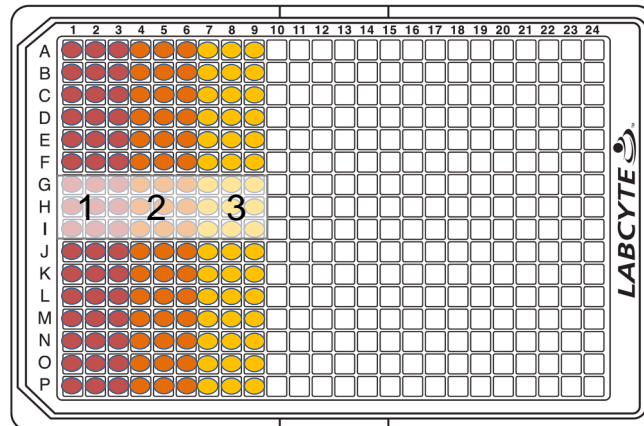
Fractional Factorial DOE Conditions

Optimization of cell-medicated cytotoxicity reporter gene assay

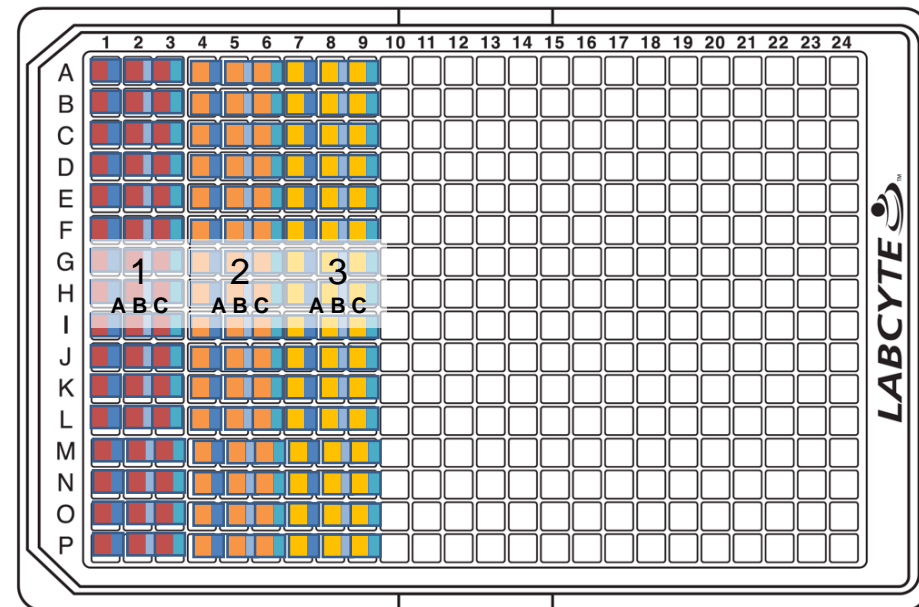
Source Plate: Three concentrations of **target cells**



Assay Plate: Three concentrations of **effector cells**



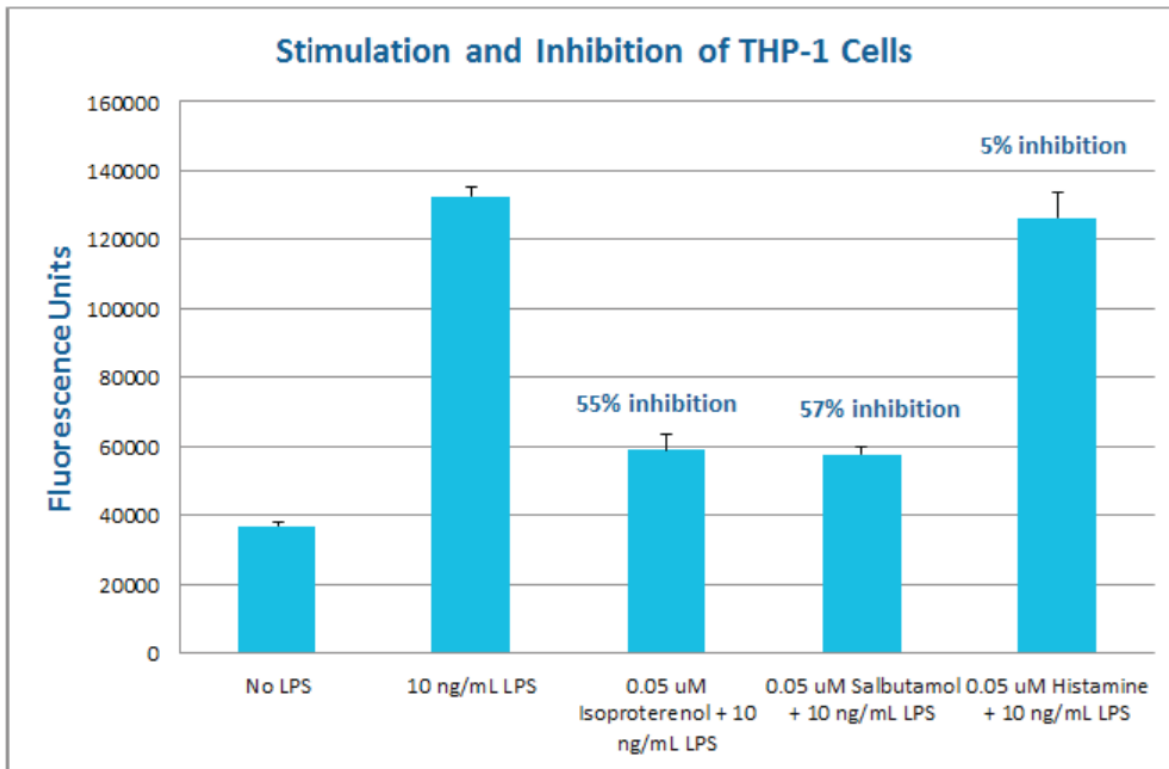
Final Assay Plate



Multiple copies can be made to evaluate target and effector incubation times if they're included in the factorial design.

AlphaLISA Assays: TNF- α Production

25-Fold Reduction of Assay volumes and Beads



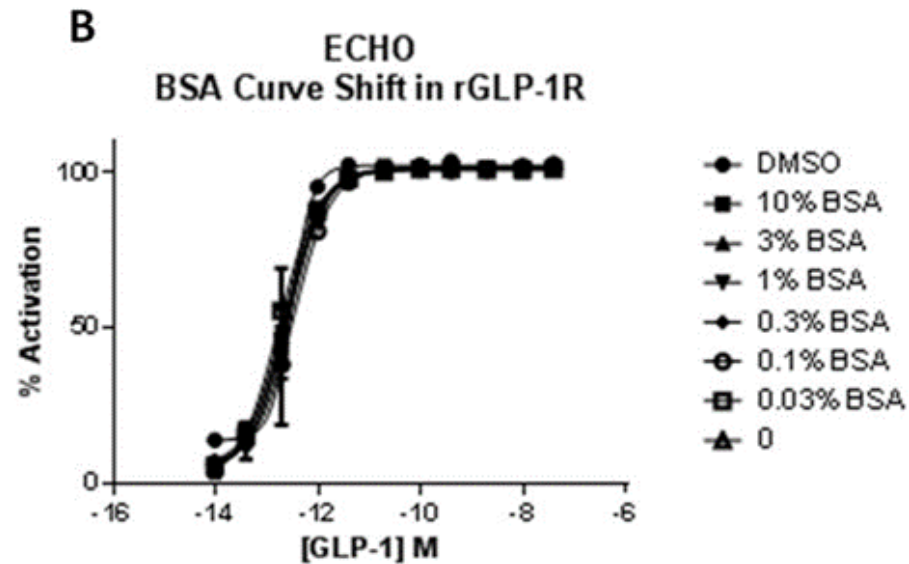
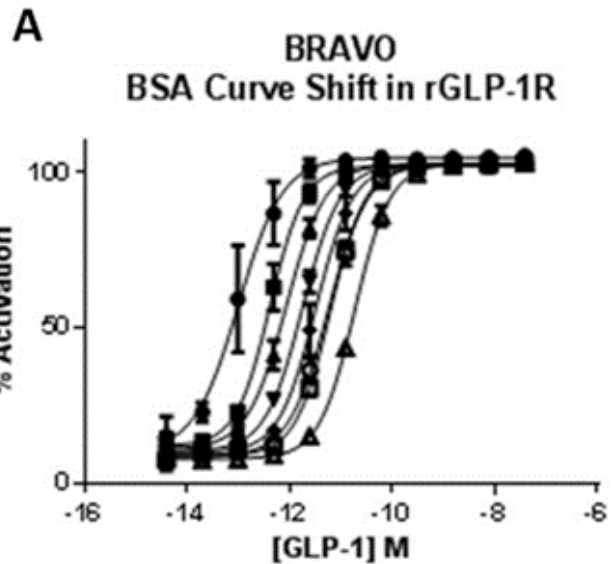
Tip Based 50 μ L Assay	Acoustic 2 μ L Assay
5 μL analyte	1.6 μL analyte
20 μL acceptor beads/anti- analyte mix (2.5x)	200 nL acceptor beads/anti- analyte mix (10x)
25 μL donor beads (2x)	200 nL donor beads (10x)

Case Study - Medimmune

Acoustic Dispensing Preserves the Potency of Therapeutic Peptides Throughout the Entire Drug Discovery Workflow

Jacqueline Naylor, Alessandra Rossi, and David C. Hornigold

Journal of Laboratory Automation, 2211068215587915, first published on May 22, 2015



A changing concentration of BSA in the solution changes the apparent EC50 when using a pipette.

No such shift is seen with the Echo system.

Microsampling for PK/PD studies

Use Less Compound and Fewer Animals with Miniaturization

▶ **Use as little as 1 μ L of plasma to conduct studies**

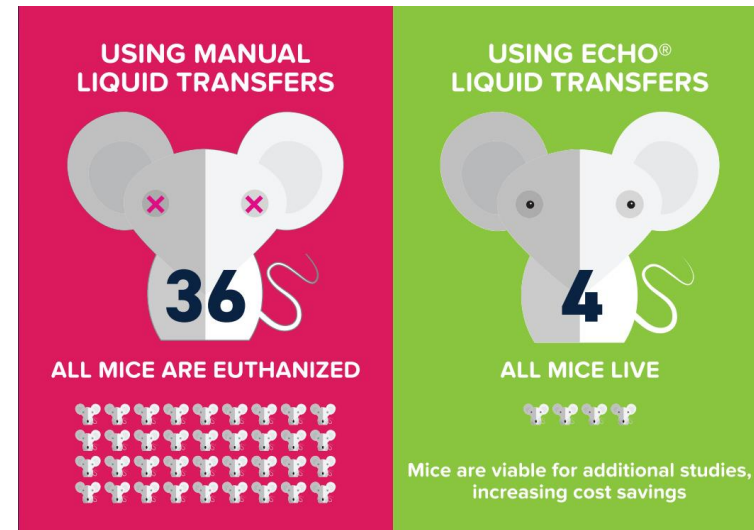
- May allow less invasive methods for blood collection
- Use fewer animals
- Extend animal life

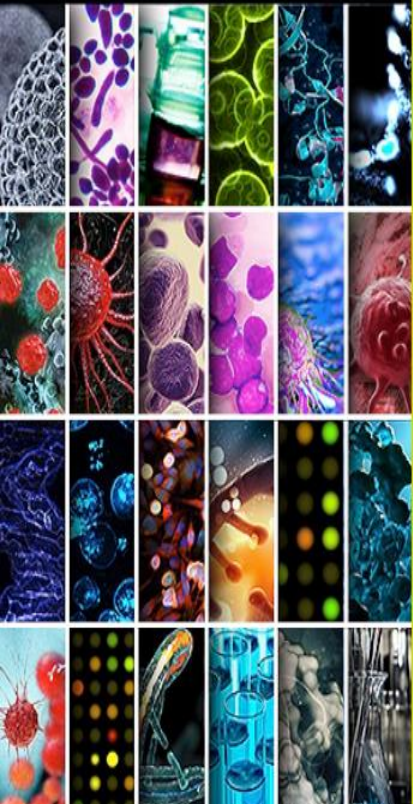
▶ **Use 75% less compound**

- Lower overall study costs

▶ **Get better data**

- Removes animal to animal variability
- Avoid stress-related artifacts
- Use same animals for PK and PD studies

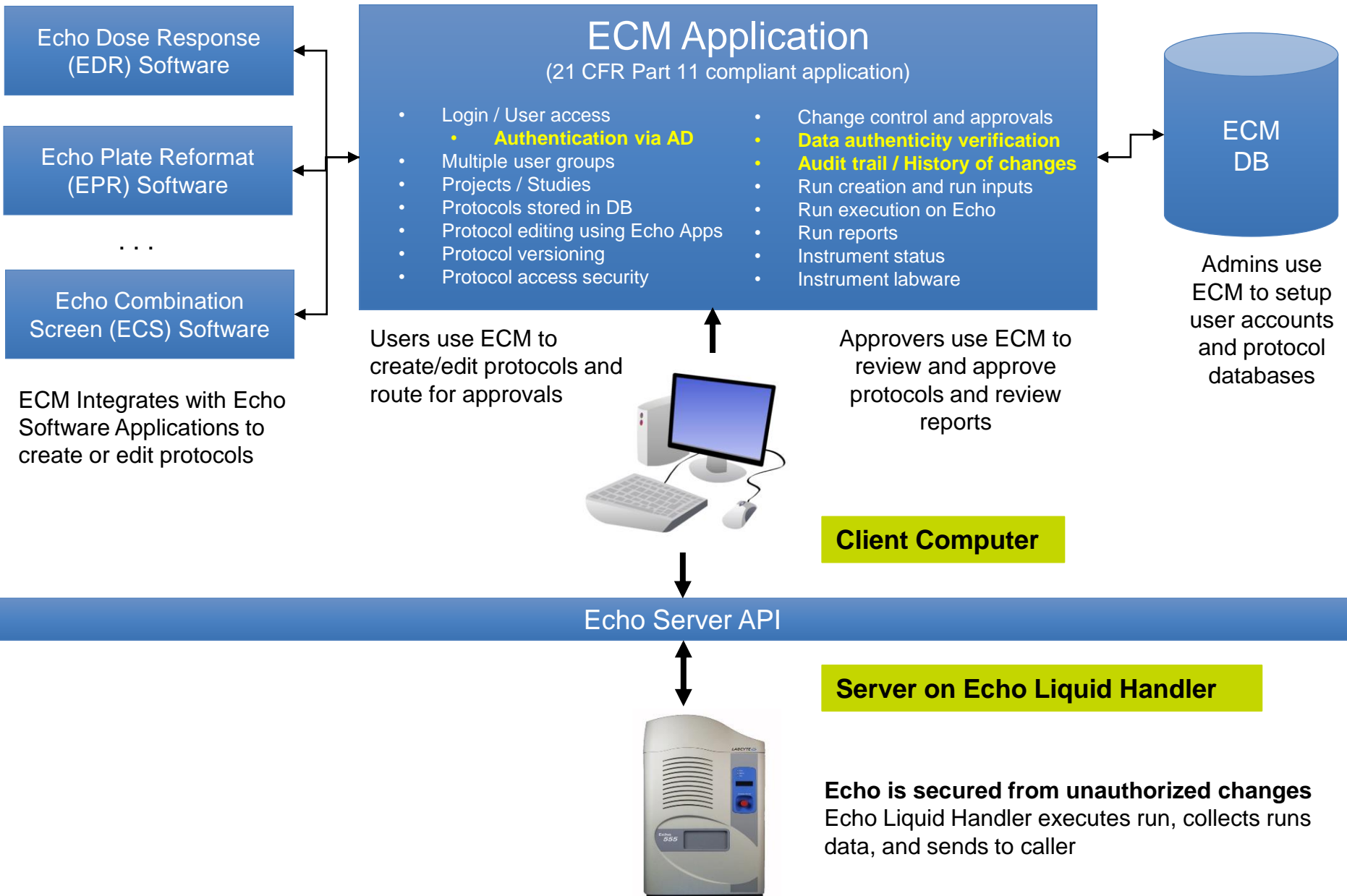




Echo® 21CFR11 Compliance Manager (ECM)

In Development for 2018

How Echo Compliance Manager works...



ECM User Workflow

Login and view list of projects

Log in

The screenshot shows a login dialog box titled "Echo Compliance Manager - User Login". It contains a "Login information" section with a "Username" field containing "joemontana" and an empty "Password" field. Below this is a "Database" dropdown menu set to "MAIN_db". At the bottom are "Login" and "Cancel" buttons.

View draft and approved protocols by project

The screenshot shows the "Echo Compliance Manager - Main Form" window. The "Current Information" section displays "User: joemontana" and "Database: MAIN_db". Below this are tabs for "Projects", "Approvals", and "Project NCT02783573". A table lists projects with columns for "Project Code", "Project Name", and "Status".

Project Code	Project Name	Status
NCT02783573	A Study of LY3314814 in Participants With Mild Alzheimer's Disease Dementia (DAYBREAK-ALZ)	✓

User Account Levels

- ▶ **Guest**
- ▶ **User**
- ▶ **Power User**
- ▶ **Approver**
- ▶ **Admin**

Create, edit, copy, execute, and route protocols for approval from one interface

The screenshot shows the "Echo Compliance Manager - Main Form" window with the "Project NCT02783573" tab selected. The "Current Information" section shows "User: joemontana" and "Database: MAIN_db". Below this, the "Project Code" is "NCT02783573" and the "Project Title" is "A Study of LY3314814 in Participants With Mild Alzheimer's Disease Dementia (DAYBREAK-ALZ)". The "Status" is "Active". The "Protocols" tab is selected, showing buttons for "New...", "Copy...", "Edit...", "Route for Approval...", and "RUN >".

ECM User Workflow

Configurable Audit Trail

Access and report information from the ECM database

- ▶ User Names
- ▶ Approver Names
- ▶ Protocol Names
- ▶ Protocol Types
- ▶ Versions
- ▶ Modifications
- ▶ Reasons for Changes
- ▶ Date of Changes
- ▶ Date of Approval Request
- ▶ Date of Approvals
- ▶ Run Dates

Protocol Change Log

Protocol Name:	8-pt DR 10mM 3 to 1 dilution
Protocol Type:	epr
Version:	2.1 DRAFT
Modifications:	Changed Transfer Volume from 100.0 nL to 75.0 nL
Reason for Change:	Assay too sensitive at 100 nL, backing off to 75 nL.
Protocol Name:	8-pt DR 10mM 3 to 1 dilution
Protocol Type:	epr
Version:	2.0 Approved
Modifications:	Changed Region A1:P24 to B2:O23
Reason for Change:	Eliminating outer rows and columns to reserve for solvent addition for hydration management.
Protocol Name:	8-pt DR 10mM 3 to 1 dilution
Protocol Type:	epr
Version:	1.0 Approved
Modifications:	Changed Transfer Volume from 50.0 nL to 100.0 nL
Reason for Change:	Needed to change volume to 100 nL because assay is not sensitive enough at 50 nL

OK

Summary

An Exciting Opportunity for Bioassay Development and Execution

- ▶ The any-well-to-any-well acoustic dispensing of compounds, reagents and samples afforded by Echo Liquid Handlers greatly facilitates bioassay development.
- ▶ The ability to dispense in nanoliter volume increments with high precision and accuracy yields improved data quality and reproducibility.
- ▶ Assay miniaturization is also possible, resulting in significant cost savings and minimal expenditure of precious samples
- ▶ Echo 21CFR11 Compliance Manager manages the creation and approval of all Echo system protocols. All information is databased for auditing.