

Automated 96 well plate pH measurement on Big Kahuna

Introduction

One of the major challenges in drug development is the need for rapid formulation of biologic drug candidates. Unchained Labs has addressed this challenge in vaccine and protein formulation with high-throughput research systems, namely, the Big Kahuna system configured for biologics formulation (Figure 1). These systems enable drug development teams to explore much broader experimental conditions and analytical samples more efficiently than by conventional methods.

Identifying the bottlenecks

The Big Kahuna system was developed to automate lab workflows and increase throughput by removing analytical measurement and processing “bottlenecks.” One significant bottleneck is pH measurement of formulations.

Measuring pH is critical for evaluating formulation stability. Traditional pH measurements are performed with glass pH probes; however, these

devices have limitations that make measurements labor intensive, time consuming and error prone. Specifically, glass pH probes require long equilibration times and frequent calibrations. They are also fragile and risk damage if not carefully handled and stored.

Improving pH measurement

To overcome the limitations of glass pH probes while also improving the throughput and reproducibility of pH measurements, Unchained Labs developed automated, multi channel pH probes (MpH, Figure 2) for use in high-throughput formulation development of biologics.

Unchained Labs' MpH

Unchained Labs' MpH is an automated component on the Big Kahuna system. The MpH has measurement resolution of 0.1 pH units and offers the following advantages over traditional pH probes:



Figure 1: The Big Kahuna system configured for biologics formulation.



Figure 2: Multi channel pH probes (MpH).

- Significantly faster equilibration time
- Greater probe durability
- Ability to be stored wet or dry

The performance of the MpH was evaluated in a direct comparison with measurements taken with a manual glass pH probe in the experiment described below.

Materials and method

- The MpH was calibrated with pH 4, 7 and 10 standards.
- A 96-well microtiter plate was prepared with 250 µL of a pH calibration standard in each well. Half of the wells contained pH 4 calibration standard, and the other half contained pH 10 calibration standard.
- The 96-well plate was placed on the Big Kahuna system, and the pH values of the unstirred cali-

bration standards were measured with a glass pH probe and then with the MpH element.

- Between each measurement, the probes were automatically washed and blown dry with air.
- The pH measurements and total time required for all measurements were recorded.

Results

Table 1 and Table 2 show the pH measurements obtained from the MpH and glass probe.

A direct comparison of the performance of the MpH and the glass pH for the measurement of 96 samples probe is shown in Table 3.

The MpH significantly increased the speed and precision of the measurements compared to the traditional glass probe method.

Unchained Labs' automated MpH results												
Total measurement time of 96-well microtiter plate: 24 min												
	1	2	3	4	5	6	7	8	9	10	11	12
A	4.00	4.00	9.90	9.90	4.00	4.00	9.90	10.00	4.00	4.00	10.00	10.00
B	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00
C	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00
D	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00
E	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00
F	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00
G	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00
H	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00

■ pH 4 □ pH 10

Table 1: Measurements using Unchained Labs' MpH.

Manual glass pH probe results Total measurement time of 96-well microtiter plate: 273 min												
	1	2	3	4	5	6	7	8	9	10	11	12
A	4.40	4.20	11.10	10.40	4.40	4.00	10.10	10.00	4.00	4.00	10.00	10.00
B	4.00	4.00	8.70	10.00	4.00	4.00	10.00	10.00	4.00	4.00	10.00	10.00
C	4.00	4.00	10.10	10.00	4.10	4.00	10.00	10.00	4.00	4.00	10.20	10.10
D	4.00	4.00	10.30	10.20	4.00	4.00	9.10	8.30	4.10	4.30	10.20	10.30
E	4.00	4.00	10.70	10.10	4.00	4.20	10.20	10.10	4.00	4.00	10.20	10.20
F	4.10	4.00	10.20	10.10	4.00	4.00	10.10	10.20	4.00	4.00	10.30	10.30
G	4.10	4.00	10.20	10.10	4.00	4.00	10.10	10.10	4.00	4.00	10.30	10.20
H	4.00	4.10	10.10	10.20	4.10	4.00	10.10	10.30	4.10	4.00	10.20	10.20

pH 4
 pH 10

Table 2: Measurements using a manual glass pH probe.

		MpH	Glass pH probe
pH 4 buffer	Average	4.10	4.10
	Standard deviation	0.03	0.10
	Range	4.00–4.10	4.00–4.40
pH 10 buffer	Average	10.00	10.10
	Standard deviation	4.00	0.40
	Range	9.90–10.00	8.30–11.10
Measurement time for 96 samples (min)		24	273

Table 3: Performance comparison between Unchained Labs' MpH and glass pH probe.

- The MpH demonstrated a six-fold increase in throughput: the time to read the 96-well plate was 273 minutes with the glass probe and 24 minutes with the MpH.
- Standard deviations were much lower with the MpH than with the glass pH probe: The standard deviation for the MpH measurements was 0.03 for pH 4 samples and 0.01 for pH 10 samples.

Conclusion

Unchained Labs' MpH was evaluated as a high-throughput pH measurement device to further enhance the efficiency of the Big Kahuna system configured for biologics formulation. This evaluation involved performance testing on pH 4 and pH 10 calibration standards compared with a standard glass pH probe.

The MpH was found to have better accuracy and reproducibility than manual glass probes. In addition, the measurement of 96 samples using the MpH was completed eleven times faster. Overall, the MpH significantly increased the throughput of pH measurements, while retaining high accuracy and reproducibility.

Integrating Unchained Labs' MpH into the Big Kahuna system configured for biologics formulation can effectively remove the bottleneck of pH measurement in drug development, leading to dramatic gains in productivity and accelerating the formulation process.



Unchained Labs
6870 Koll Center Parkway
Pleasanton, CA 94566
Phone: 1.925.587.9800
Toll-free: 1.800.815.6384
Email: info@unchainedlabs.com

© 2018 Unchained Labs. All rights reserved. Big Kahuna is a trademark and Unchained Labs is a registered trademark of Unchained Labs. All other brands or product names mentioned are trademarks owned by their respective organizations.