

# TECHNICAL INFORMATION

*BioRobotics*

## DRY SPOT PIPETTING WITH THE BIOMEK<sup>®</sup> FX 96-CHANNEL PIPETTING HEAD (200 $\mu$ L) AND AP96 P20 DISPOSABLE TIPS

*Edwin Dario*

*Beckman Coulter, Inc.*

### Introduction

The Biomek<sup>®</sup> FX is the newest liquid handler offered by Beckman Coulter, Inc. What makes the Biomek FX unique is its versatility and expandability to meet the ever-changing needs in Drug Discovery. The Biomek FX offers Smart Solutions<sup>™</sup> for a variety of specific applications including plate replication, nucleic acid preparation and high throughput screening. Utilizing single or dual bridge systems configured with various pipetting heads, and on-deck devices provides the user with both flexibility and throughput.

A common liquid handling application for the Biomek FX system in both library management and high-throughput screening is dry spot pipetting small aliquots of compounds (typically DMSO solutions) into assay plates. This bulletin provides details that characterize this pipetting technique at 1  $\mu$ L or less using the Biomek FX 96-Channel Disposable Tip Pipetting Head (200  $\mu$ L). This head was designed to use the existing Biomek AP96 P20 and P250 disposable tips—the same tips currently utilized by the Multimek<sup>™</sup> 96 liquid handler, employing the industry standard of 9-mm center spacing within the tip rack for use with 96-well microplates.

### Manufacturing Criteria

Beckman Coulter has established criteria that each head must meet in a manufacturing test prior to shipment. For the Biomek FX 96-Channel Disposable Tip Pipetting Head (200  $\mu$ L) these criteria are a dispense of 200  $\mu$ L at <1.00% C.V. and a dispense of 5  $\mu$ L at <2.00% C.V. using AP96 P20 tips. The tests are performed as follows: 5- $\mu$ L or 200- $\mu$ L of Eosin Y dye solution is

dispensed into three 96-well microplates and read on the microplate reader for an immediate read at 510 nm and 650 nm (to account for background). All three plates' C.V.s must fall below the current given specification to qualify. In addition, a DMSO (dimethylsulfoxide) leak test is performed to ensure that all the mandrels are leak-free.



*Figure 1. Biomek FX*

### Testing Beyond the Specifications

It is understood that the Biomek FX is used in the laboratory to pipette a wide variety of liquids at lower volumes; far too many to include in the manufacturing test. Thus, the experiments described in this bulletin were carried out to characterize the performance of the systems under more lifelike conditions.



The study conducted tests on three randomly selected 200  $\mu\text{L}$  heads at 1.0  $\mu\text{L}$ , 0.50  $\mu\text{L}$ , and 0.25  $\mu\text{L}$  in DMSO. This evaluation was performed on a single-bridge Biomek<sup>®</sup> FX equipped with the 96-channel disposable tip pipetting head (200  $\mu\text{L}$ ). Biomek AP96 P20 tips were used in this study.

## Solutions and Techniques Used

For the purpose of trying to emulate dry spot pipetting in the laboratory, Eosin Y dye in DMSO was the matrix used. A Biomek FX method was created that dispenses Eosin Y Dye/DMSO solution into three Beckman Coulter flat 96-well microplates. One dye stock solution was created by adding ~1.35 gm/L of Eosin Y dye (Sigma, catalog #11983-0) to DMSO (Sigma, catalog #15493-8), respectively. At this concentration, pipetting 1.0-, 0.5-, and 0.25- $\mu\text{L}$  gives absorbance readings at 510 nm of approximately 1.2 OD, 0.60 OD, and 0.30 OD, respectively. For each 200  $\mu\text{L}$  dispense head, three plates were dispensed at each volume. The C.V.s of the plates from the three heads were averaged, and the average of nine plates was reported.

The same stock solution was used for all three volumes, and each microplate was normalized to 60  $\mu\text{L}$  with deionized distilled water.

For example, the 1- $\mu\text{L}$  assay (see Figure 2) was performed by dry spotting 1.0  $\mu\text{L}$  of DMSO/dye to the first of three Beckman Coulter flat 96-well microplates. Without changing tips, the tips were washed in a separate reservoir containing deionized distilled water for a 30  $\mu\text{L}$  x 3 cycle wash/mix. Then, 30  $\mu\text{L}$  x 2 of deionized distilled water was dispensed into plate one. A new rack of tips was used for each subsequent plate for a total of three racks used per volume. The plates were then manually transported to the SAGIAN<sup>™</sup> Plate Shaker for a two-minute shake, then taken to the Molecular

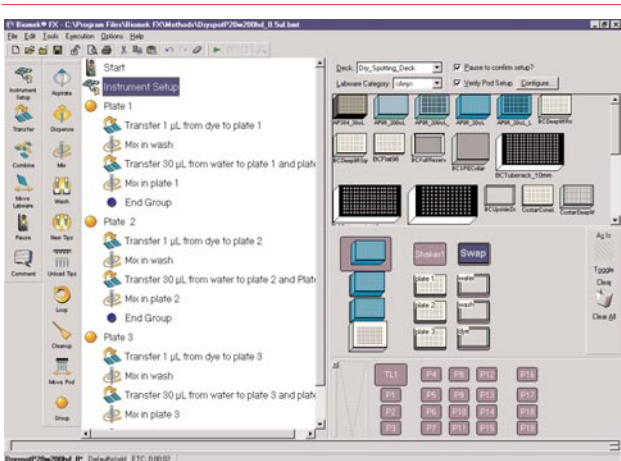


Figure 2.

Devices SPECTRAMax<sup>®</sup> PLUS plate reader for an immediate read at 510 nm and 650 nm (to account for background).

The Biomek FX method incorporated the following example parameters:

- Trailing Airgap None
- Aspiration Speed 25%
- Dispense Speed 25%
- Blowout 25  $\mu\text{L}$
- Move within the well @ 10%
- Aspiration, Dispense, and Tip Touch delays of 500 ms
- Pre-Mix
- Tip Touch
- Follow Liquid

It is highly recommended that a custom Technique and Liquid Type template are used for dry spotting assays (see Figures 3 and 4). Doing so will ensure that the assay being performed is optimized to its maximum capabilities.

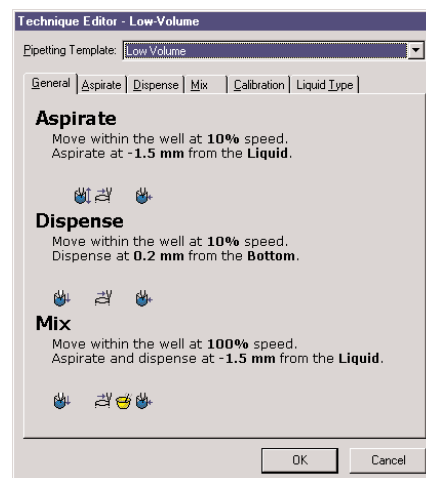


Figure 3.

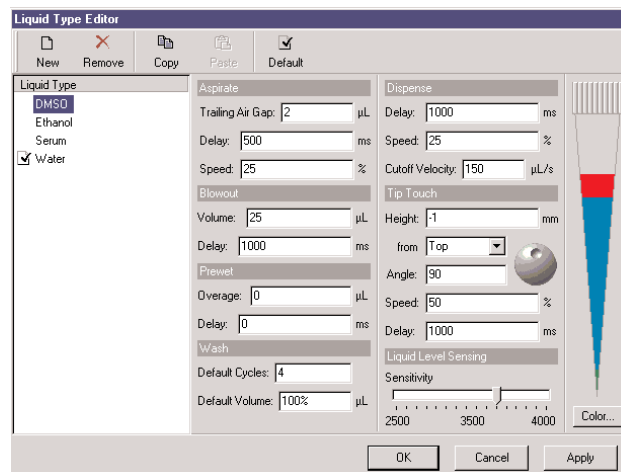


Figure 4.

## The Results of This Study

Three experiments were run on each of three separate 96-channel disposable tip pipetting heads (200 µL) for this study. The values reported in Table 1 represent the the average %C.V. of nine plates for each volume.

## Conclusion

This bulletin shows the characteristic pipetting performance of the Biomek® FX 96-channel disposable tip pipetting head (200 µL) for dry spot pipetting using AP96 P20 tips under a set of typical laboratory conditions. End-user results will likely vary when using reagents and labware other than those described. However, the Biomek FX software is rich in pipetting control features that should enable most laboratories to obtain similar results for their respective application.

**Table 1: AP96 P20 Dry Spot Pipetting Using Three Biomek FX 96-Channel Disposable Tip Pipetting Heads (200 µL)**

	<b>DMSO</b>
0.25 µL	5.93%*
0.50 µL	4.58%
1.0 µL	3.21%

\* Because of the natural “wicking” and chemical attributes of DMSO, the 0.25 µL dispense value (average %C.V.) for DMSO was obtained utilizing a customized pipetting template and technique within the Biomek FX software. Parameters such as the slope and offset were manipulated on a trial-and-error basis until an accurate volume of ~0.25 µL was obtained prior to assessing the repeatability. This demonstrates the flexibility of the Biomek FX software which allows the user to create, customize, and manipulate techniques and other variables within their method to ensure accuracy as well as precision.

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Beckman Coulter, Inc. • 4300 N. Harbor Boulevard, Box 3100 • Fullerton, California 92834-3100  
Sales: 1-800-742-2345 • Service: 1-800-551-1150 • Telex: 678413 • Fax: 1-800-643-4366 • www.beckmancoulter.com

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