

Artel MVS[®] as a Tool For Measuring Liquid Mixing Efficacy in Verification Plates

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ABSTRACT:

You wouldn't bake a cake without mixing the ingredients first, yet when important assays are being conducted the liquids dispensed into a microplate are often assumed to have mixed through simple diffusion or after only a cursory agitation of the plate.

For assay results to be truly trustworthy, every part of the methodology employed needs to be assessed for efficacy and repeatability. This application note describes how the Artel MVS[®] Multichannel Verification System can be used as a tool for measuring the efficacy of liquid mixing protocols.

In addition, the data presented herein highlights the importance of ensuring that effective mixing of the solutions dispensed into a microplate has occurred during every step of the assay procedure. This is particularly poignant as experiments conducted within the Artel laboratory show that diffusion-based mixing can take as long as 24 hours, and even a plate shaker set at 2200 RPM can take close to 10 minutes to achieve complete mixing if the shaker protocol has not been optimized.

INTRODUCTION:

Homogeneity of solutions in Verification Plates plays a crucial role in assay effectiveness, yet is an area of assay method development and verification that is often overlooked. Without ensuring that solutions have been effectively mixed, it is possible for non-homogenous samples to be used for subsequent assay steps leading to aspiration of very concentrated or very dilute aliquots.¹

In this study, the MVS was used to measure the absorbance of dye solutions in microplate wells at multiple time points. The methods discussed herein maybe applied to any mixing method including diffusion, aspirate and dispense steps with liquid handling instruments or other mixing methods.

Complete mixing of two solutions following a wet dispense protocol is particularly difficult to achieve. In order to mimic this challenging mixing scenario, studies involving the wet dispensing of a small volume of sample into a much larger volume of Diluent were performed.

For each measurement set, the average, standard deviation and %CV were calculated for the absorbance values for the control and the test data sets. The control solution was pre-mixed and therefore should have displayed no change in absorbance, regardless of the number of mix cycles performed. Therefore, the %CV values for the control samples should remain constant. Conversely, the test samples typically exhibited large initial absorbance readings due to a concentrated area of sample solution in the well resulting from the wet dispense. The %CV is also high due to lack of homogeneity of the samples in the wells prior to mixing. As the mixing action achieves homogeneity of the samples and diluent in the wells, the %CV is reduced, ultimately becoming constant from measurement to measurement.

REFERENCES

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