

# Maintaining HTS compound library integrity using an automatable microplate lid that provides an environmental barrier seal

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## Abstract

An important driver in the drug screening industry is to reduce costs through assay miniaturization and improvements in small volume liquid handling. Acoustic dispensing, for example, allows dispensing of nanoliter amounts of compounds directly into assay plates, eliminating the need for intermediate dilution plates. At volumes this small, however, compounds are much more sensitive to being impacted by the environment. Consistent generation of valid data from HTS screens requires that libraries are protected from both evaporation and from hydration of DMSO by atmospheric moisture. To realize the benefits of small volume dispensing while still generating consistent, reliable HTS data, screeners are evaluating multiple approaches to maintaining the integrity of their samples that are stored in 1536 well source plates. To this end, we compared the protective capacity of a novel microplate lid with other currently available microplate sealing methods. This ANSI compliant lid, which is compatible with manual or automated methods, consists of a gasket and four latches that are used to seal the well array of the plate. For our testing, four microliters of dry DMSO were dispensed into 1536 well plates, and the plates were surveyed for % DMSO and average height in the wells using a Labcyte<sup>®</sup> Echo<sup>™</sup> 550. We have determined that utilizing the locking lid method resulted in an average %DMSO of above 94 at four weeks.

## Use of locking Storage Lid for assay-ready compound plates

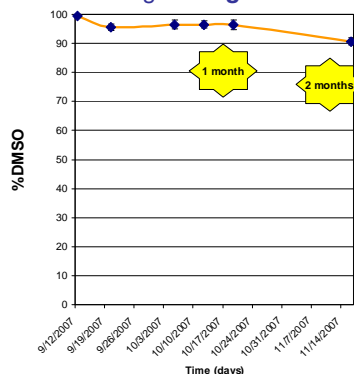
- A Beckman Coulter FRD was used to dispense 4ul of dry DMSO into three Corning Echo Qualified 1536 plates (Cat# 3730).
- Microplates were covered with a prototype locking storage lid, or with a conventional tape seal (bar graphs).
- Plates were surveyed on the Labcyte Echo 550 to determine %DMSO at time 0 weeks.
- Covered plates were then stored at room temperature and surveyed at indicated times for up to two months.
- Plates were also surveyed on the Labcyte Echo 550 to determine average height of the liquid in each well over a one month period (graphs to the right).

- DMSO absorbed <10% over a 2 month period (upper left).
- DMSO evaporated <15% in one week and <40% in one month.
- Locking Storage Lid performed similarly to adhesive tape seals.

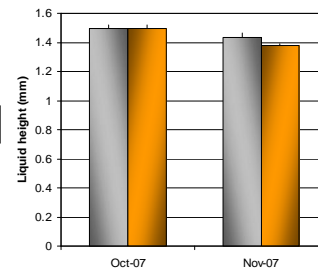
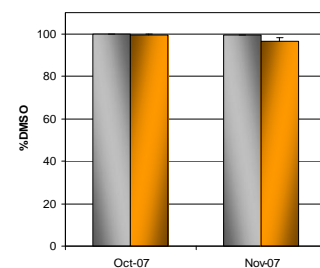
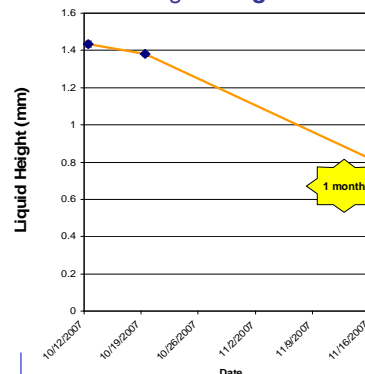
n=3 plates, performed 1X, SD indicated



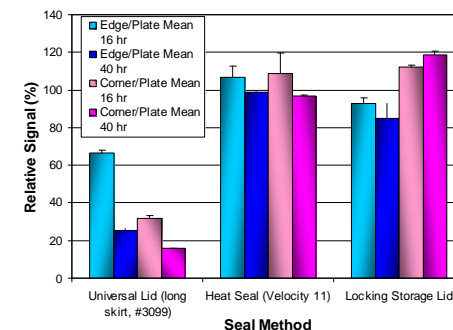
## Hydration of 100% DMSO Using Storage Lid



## Evaporation of 100% DMSO Using Storage Lid



## Aqueous Evaporation Using Storage Lid

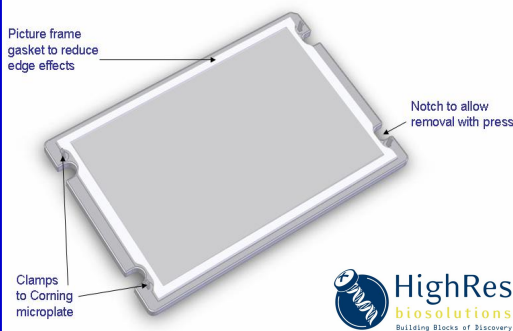


## Use of locking Storage Lid for assay-ready reagent plates, or long incubations

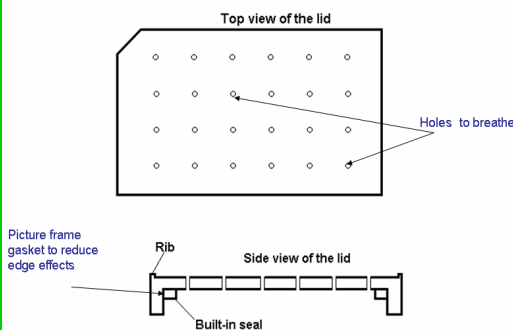
- A Perkin Elmer FlexDrop was used to dispense 4ul of Cell Titer-Glo (Promega) + ATP into Corning Echo Qualified 1536 plates.
- Microplates were covered with either a long-skirt lid, heat seal tape or a prototype locking storage lid.
- Plates were stored on the benchtop overnight (16hr) or for 40 hr.
- Luminescence was read on a Perkin Elmer Viewlux at indicate times.
- Mean signal from edge and corner wells were plotted relative to total plate mean signal.
- Aqueous edge and corner effect was severe using standard lids.
- Aqueous solutions in plates using the Locking Storage Lid only evaporated ~10% more than adhesive tape seals, even over 40 hr.
- The Locking Lid reduced variation due to personal handling of plates to remove heat sealed tape (and is automatable).

n=3 plates, performed 1X, SD indicated

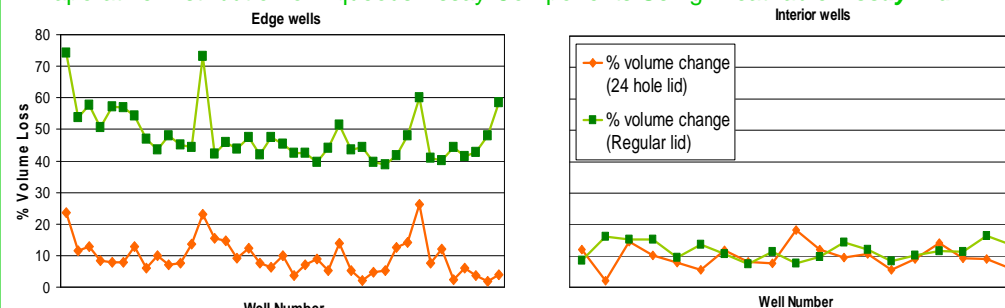
## Storage Plate Lid



## Assay Plate Lid



## Evaporative Distribution of Aqueous Assay Components Using Breathable Assay Lid



## Use of breathable Assay Lid for cell-based assays

- 96 well plates were loaded with 100 ul PBS by hand.
- Microplates were covered with either a long-skirt lid or a prototype 24-hole assay lid.
- Plates were stored on the benchtop for 24 hr.
- Volume changes from edge and selected interior wells were plotted relative to total plate mean volume.

- Aqueous solutions showed a significant edge evaporation (up to 75%) from outside wells (top, bottom, and side wells) using standard lids.
- The Assay Lid did not demonstrate significant edge effect (compare orange lines in left and right graphs).
- Internal wells showed comparable relative volume loss between the two lids.

n=3 plates, performed 1X

## Conclusions

- Interaction between compounds or assay components with the atmosphere can significantly impact screening results.
- The Locking Storage Lid protects compounds from atmospheric hydration.
- Evaporation of compounds is also alleviated using the Locking Storage Lid.
- Heat seal tape and the Locking Storage Lid performed similarly over the time frame of these preliminary studies, and the latter is automatable.
- The Locking Storage Lid can also be used to eliminate evaporation of aqueous reagents (biochemical assays) for up to 40 hrs.
- A breathable Assay Lid is useful for cell-based assays to reduce edge effect by distributing atmospheric exchange evenly across a microplate.

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