384-Well Tilted-Bottom Microplate

Reduced sample volumes and improved sensitivity for Octet assays

Key features

- Assay with volumes as low as 40 µL per well
- Reduces variation in baseline signals, and improves assay sensitivity
- Use with Octet® HTX, RED384 and QK384 systems
- 384-well, black, polypropylene microplate
- Standard SBS format

The 384TW microplate is a black, polypropylene, 384-well tilted-bottom plate designed for ForteBio’s Octet family of label-free biomolecular interaction analysis instruments. It can be used on the Octet QK384, RED384 and HTX instruments, all of which accommodate the 384-well microplate format. The 384TW microplate enables use of samples as small as 40 µL in Octet assays and reduces variability in the background signal, thus improving assay sensitivity, particularly beneficial for peptide, small molecule and fragment analysis.

Assays using limited sample volume

When assaying precious samples, the ability to limit the volume of sample per experiment is critical. The 384TW microplate provides the ability to perform assays using as little as 40 µL per well. This is a 3-fold decrease over the volume needed in a standard 384-well plate and a 5-fold decrease over the sample volume needed in a 96-well plate. The assay performance (dynamic range, precision, accuracy) for kinetic characterization and quantitation in the 384TW microplate are equivalent to that in a black, polypropylene 384-well standard flat-bottom microplate.

Figure 1 shows results from a Protein A biosensor-based quantitation assay to detect human IgG (hIgG) in solution. The assay was run on Octet RED384 system using the standard Basic Quantitation settings with Regeneration module, available in the Octet Data Acquisition software.

Table 1 compares data obtained in a 96-well flat-bottom microplate (Figure 1) using 200 µL of sample to that obtained in a 384TW microplate using only 40 µL of sample shows that assay performance is equivalent in both microplates.

<table>
<thead>
<tr>
<th>Test concentrations</th>
<th>Measured avg. concentrations (µg/mL)</th>
<th>Percent CV</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>96-well</td>
<td>384TW</td>
</tr>
<tr>
<td>2000</td>
<td>2000.00</td>
<td>1999.98</td>
</tr>
<tr>
<td>1500</td>
<td>1508.78</td>
<td>1503.58</td>
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<td>1000</td>
<td>1015.90</td>
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<tr>
<td>700</td>
<td>700.20</td>
<td>700.70</td>
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<tr>
<td>500</td>
<td>503.03</td>
<td>502.53</td>
</tr>
<tr>
<td>300</td>
<td>300.50</td>
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<tr>
<td>0.5</td>
<td>0.50</td>
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</table>

Table 1: Measured concentrations and CVs show good dynamic range, recovery and precision using the 384TW vs. 96-well standard microplate.

Figure 1: Calibration curve across the dynamic range of 0.5–2000 µg/mL hIgG; results from 384TW microplates compares very well to those from standard flat-bottom 96-well microplates.
Improved sensitivity in small molecule and peptide analyses

In high-sensitivity screening and kinetic applications, the limit of detection is often defined as 3 times the standard deviation of the baseline signal. The 384TW microplate decreases system artifacts when used on the Octet QK384, RED384 and HTX systems, decreasing the variation (and thus the standard deviation) of the baseline signal, allowing smaller signals to be resolved from baseline noise.

Table 2 contains data obtained on the Octet RED384 system using Super Streptavidin (SSA) biosensors. It includes the average signal and standard deviation of the baseline of these biosensors. The averages and standard deviations for each data point are calculated from 1920 replicates. The lower limit of detection shown is defined as 3 times the standard deviation over the average baseline. For many screening campaigns, this value defines the smallest signal contribution by a positive binding sample. In this experiment the 384TW microplate was found to be advantageous, having a 2-fold lower limit of detection than a standard 384-well flat-bottom plate.

Instructions for use

The 384TW microplate is used on the Octet HTX, RED384 and QK384 systems in the same way as a flat-bottom 384-well microplate with the following exceptions:

- Minimum sample volume is 40 µL.
- Maximum sample volume is 100 µL.
- When pipetting into the wells, avoid trapping bubbles of air in them. Bubbles in the well can result in spikes in the assay data. An effective method for removing any bubbles that occur is to centrifuge the filled plate in a swinging plate rotor at 1000 rpm for 2 minutes prior to the assay.
- Assays should be set up with the biosensor offset at 3 mm.
- Set the “Delay experiment start” setting available in the Octet Data Acquisition software to a minimum of 10 minutes (600 seconds) to allow the plate to equilibrate inside the instrument prior to the first data point.
- Check the “Shake sample plate while waiting” option in the Octet Data Acquisition software. Shaking the plate during the equilibration time helps to eliminate trapped air.

<table>
<thead>
<tr>
<th>Specification</th>
<th>Details</th>
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<tbody>
<tr>
<td>Applications</td>
<td>Low volume and high sensitivity applications</td>
</tr>
<tr>
<td>Instrument compatibility</td>
<td>Octet HTX, RED384 and QK384 systems</td>
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<tr>
<td>Material</td>
<td>Polypropylene</td>
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<tr>
<td>Color</td>
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<td>Dimensions</td>
<td>SBS standard</td>
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<tr>
<td>Run time on Octet RED384, QK384 and HTX systems</td>
<td>Screening applications: &lt;10 hours at 40 µL (biosensor offset 3 mm) Kinetics &amp; quantitation applications: &lt;4 hours at 40 µL (biosensor offset 3 mm)</td>
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<tr>
<td>Evaporation rate</td>
<td>Non-linear rate of 30% loss at 6 hours for initial volume of 40 µL of PBS</td>
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<tr>
<td>Maximum volume</td>
<td>100 µL</td>
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<tr>
<td>Minimum volume</td>
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<td>Sterilization</td>
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Ordering information

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<th>UOM</th>
<th>Description</th>
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<tbody>
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<td>18-5080</td>
<td>Pack</td>
<td>384-well microplate, black, tilted-bottom, polypropylene, 10 per pack. Enables low volume samples, lowers background for peptide, small molecule and fragment analysis applications.</td>
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<tr>
<td>18-5076</td>
<td>Case</td>
<td>384-well microplate, black, tilted-bottom, polypropylene, 100 per case. Enables low volume samples, lowers background for peptide, small molecule and fragment analysis applications.</td>
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</table>

* All specifications are subject to change without notice.