

Applications

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Eppendorf Plate® Deepwell 96 and 384: g-Safe®

Investigating stability during centrifugation of Eppendorf Plate Deepwell

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Abstract

Eppendorf Plate Deepwell 96 and 384 were developed specifically for the demanding requirements of centrifugation and, therefore, have high inherent stability. This application note shows that all plate formats in Standard quality as well as DNA and Protein LoBind quality have stability during centrifugation of up to 6,000 x g. The sterile product variations of Eppendorf Plate Deepwell can be centrifuged up to 5,000 x g. Our studies also show that Eppendorf Plate Deepwell 96 and 384 are suitable for centrifugation over a broad temperature range in both swing-bucket and drum rotors.

Introduction

If we look at developments in laboratory routine over recent years, there is a clear trend towards ever-increasing sample throughput – with sample processing increasing in high-throughput screening in the pharmaceutical industry and in the research fields of genomics and proteomics, as well as in routine food analysis. In order to be able to achieve this high sample throughput, multi-well plates have to be used. On the one hand, such plates need to be the proper size for use by automated systems and to conform to particular standard dimensions (SBS standard, Society for Biomolecular Screening). On the other hand, it should be possible to execute both sample preparation/treatment steps and storage and transport of samples conveniently, efficiently and safely. Eppendorf developed a new generation of deepwell plates to take account of this trend – offering users a unique design, specific well geometry, high-precision manufacture (SBS standard) and maximum stability during centrifugation.

Many sample preparation methods – such as pelleting a bacterial culture (e.g., 6,000 x g, 0 °C), nucleic acid extraction (e.g., 4,300 x g, 20 °C; 6,000 x g, 4 °C) or plasmid isolation (e.g., 2,500 x g, 4 °C) – include centrifugation steps that can become a problem when deepwell plates are used, especially at elevated g-forces. Under the effect of strong forces, such as those that may occur during centrifugation, plates tend to twist, tear or even split open. It is particularly important for users that deepwell plates withstand these forces. In other words, wells may not split open and deepwell plates may not tear or become so deformed that they can no longer be used in liquid handling machines, for example. The experiments described below are used to examine the centrifugal forces that Eppendorf Plate Deepwell can withstand while maintaining defined stability criteria.

Materials and methods

Eppendorf Plate Deepwell and deepwell plates from other suppliers were centrifuged and then rated with regard to their durability in relation to defined stability criteria.

Stability criteria

- **Durability**
No split wells or cracks may occur.
- **Deformation**
Bases of wells, webs or side walls were permitted deformation only within the confines of the SBS standard. Footprint dimensions as per SBS standard had to be maintained.

Deepwell plates used

Eppendorf Plate Deepwell 96/1000 µl
Eppendorf Plate Deepwell 96/2000 µl
Eppendorf Plate Deepwell 96/500 µl
Eppendorf Plate Deepwell 384/200 µl
Corresponding deepwell plates from competitors

Eppendorf Plate Deepwell and deepwell plates from other suppliers were centrifuged in the centrifuges/rotors listed below at 40 °C, 20 °C, 4 °C and 0 °C for 90 minutes/ 10 minutes at the maximum possible centrifugal force of the centrifuge and rotor used. All the wells of the plates were filled to the maximum possible working volume with an aqueous solution, with 70 % ethanol or with DMSO, respectively. The stability of Eppendorf Plate Deepwell, including DNA LoBind, Protein LoBind and the sterile variants, was likewise tested as described. The plates were tested in various centrifuges without interruption for the specified time and under the conditions listed.

Centrifuges and rotors used

Eppendorf 5804 R; Rotor A-2-DWP
Eppendorf 5810 R; Rotor A-4-81-MTP/Flex and A-4-62-MTP/Flex
Eppendorf 5430; Rotor A-2-MTP
Multifuge® 3L; Rotor *6445 and Highplate®
Sigma 4-15; Rotor 11150
Sigma 4K15C; Rotor 09100

Results and discussions

Eppendorf Plate Deepwell 96 and 384 were centrifuged in various centrifuges with the associated swing-bucket rotors at their maximum possible force for 90 minutes at 40 °C. The wells of the plates were filled with an aqueous solution. Assessment criteria for evidence of centrifugation stability were the durability of the plates (no cracks or split wells) and maintenance of SBS dimensions.

Centrifugation in Eppendorf centrifuges

In these trials, it was shown that all plate formats could be centrifuged in Eppendorf centrifuges/rotors at the maximum permitted speed in each case without cracks occurring or wells splitting. No deformation in excess of the SBS standard was found (**Tab. 1**). By way of example, corresponding deepwell plates from four other suppliers were also tested. These plates also proved durable at the centrifugal forces that occur in Eppendorf centrifuges.

The effect of centrifuge and rotor

Stability to centrifugation is heavily dependent on the centrifuge and the rotor used. All four Eppendorf Plate Deepwell formats proved to be fully durable and inherently stable at 3,364 x g in the Multifuge 3L and swing-bucket rotor *6445. One of the four competing plates tested was highly deformed following centrifugation, to the extent that its dimensions were outside those of the SBS standards (**Fig. 1**).

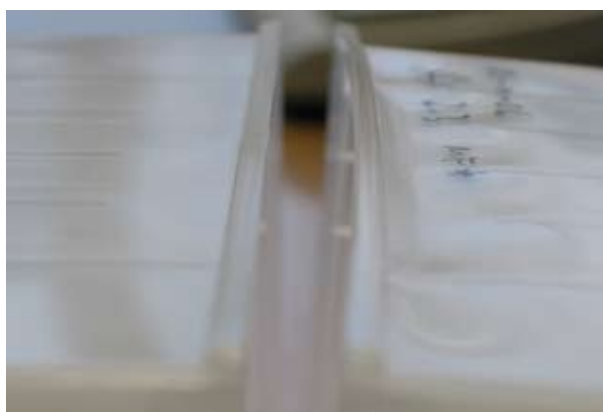


Fig. 1: Deformation of a competitor's plate following centrifugation in the Multifuge 3L Rotor *6445 (non-refrigerated) at 3,364 x g for 90 min. Left: Before centrifugation; Right: After centrifugation

The centrifugal forces act differently on the plates depending on the design of the rotor, so different degrees of deformation can result. Lateral deformation of the base of all plates occurred even at a relatively low load in the

Highplate rotor (5,084 x g). However, no cracks or split wells were found. In the Centrifuge Sigma 4–15/Rotor 11150 (4,256 x g), on the other hand, Eppendorf Plate Deepwell were durable and inherently stable (**Tab. 1**).

Table 1: Results of centrifuging Eppendorf Plate Deepwell at the maximum possible force of the centrifuge and rotor at 40 °C for 90 minutes.

- ● = plates durable (no split wells or cracks) and deformations not outside SBS standard dimensions
- ▲ = plates durable (no split wells or cracks) but deformations outside SBS standard dimensions, n.d. = not determined

40 °C, 90 minutes	Durability/Deformation			
	Eppendorf Plate Deepwell			
	384/200 µl	96/500 µl	96/1000 µl	96/2000 µl
Eppendorf 5804 R Rotor A-2-DWP 2,250 x g	● ●	● ●	● ●	● ●
Eppendorf 5810 R Rotor A-4-81-MTP/Flex 2,916 x g	● ●	● ●	● ●	● ●
Eppendorf 5810 R Rotor A-4-62-MTP 2,750 x g	● ●	● ●	● ●	● ●
Eppendorf 5430 Rotor A-2-MTP 2,204 x g	● ●	● ●	n.d.	n.d.
Multifuge 3L 4x-swing-bucket rotor *6445 3,364 x g	● ●	● ●	● ●	● ●
Multifuge 3L Rotor Highplate 5,084 x g	● ▲	● ▲	● ▲	● ▲
Sigma 4-15 Rotor 11150 4,256 x g	● ●	● ●	● ●	● ●

The effect of temperature

Centrifugation temperature also has a considerable effect on the stability of multi-well plates. At low temperatures, for example 0 °C or 4 °C, polypropylene is much more brittle than at room temperature. This makes the material more fragile so that it tears more easily. At higher temperatures, e.g. 40 °C, polypropylene is softer and more flexible. Under the effects of centrifugal forces, therefore, polypropylene plates tend to deform to a greater extent. To test how Eppendorf Plate Deepwell behave over a broad temperature

range, all plate formats were centrifuged at 0 °C (10 min), 20 °C (90 min) and 40 °C (90 min) in the Sigma centrifuge 4K15C/rotor 09100 at the maximum centrifugal force of 6,189 x g. The results show that Eppendorf Plate Deepwell can be centrifuged in a broad temperature range at both low (0 °C) and elevated temperatures (20 °C; 40 °C) without the risk of wells splitting (Tables 2 and 3). The plates from most competitors were durable only at higher temperatures (20 °C and 40 °C) (**Tab. 3**).

Table 2: Results of centrifuging Eppendorf Plate Deepwell in the Centrifuge Sigma 4K15C/Rotor 09100 at various temperatures: 0 °C for 10 min, 20 °C for 90 min, 40 °C for 90 min.

● = plates durable (no split wells or cracks)
▲ = plates cracked or wells split

Sigma 4K15C Rotor 09100 6,189 x g	Durability			
	Eppendorf Plate Deepwell			
	384/200 µl	96/500 µl	96/1000 µl	96/2000 µl
0 °C; 10 minutes 6,189 x g	●	●	●	●
20 °C; 90 minutes 6,189 x g	●	●	●	●
40 °C; 90 minutes 6,189 x g	●	●	●	●

Table 3: Comparison of centrifuging Eppendorf Plate Deepwell 96/2000 µl and 96/1000 µl and the relevant competitors' plates in the Centrifuge Sigma 4K15C/Rotor 09100 (6,189 x g) at various temperatures: 0 °C for 10 min, 20 °C for 90 min, 40 °C for 90 min.

● = plates durable (no split wells or cracks)
▲ = plates cracked or wells split

Sigma 4K15C Rotor 09100 6,189 x g	Durability		
	0 °C, 10 minutes	20 °C, 90 minutes	40 °C, 90 minutes
Eppendorf Plate Deepwell 96/2000 µl	●	●	●
Competitor A	▲	●	●
Competitor B	●	●	●
Competitor C	▲	●	●
Competitor D	●	●	●
Eppendorf Plate Deepwell 96/1000 µl	●	●	●
Competitor A	▲	●	●
Competitor B	●	●	●
Competitor D	▲	▲	▲

The effect of solvents

Centrifugation steps in the presence of solvents are an integral component of many methods. For nucleic acid precipitation, for example, it has to be possible to centrifuge deepwell plates in the presence of 70 % ethanol at 4 °C and 6,000 x g without wells splitting or cracks forming. However, solvents can reduce the centrifugation stability of tubes and plates made of polypropylene. To test the durability of the Eppendorf Plate Deepwell in the presence of frequently used solvents, all plate formats were filled with 70 % ethanol or 100 % DMSO and centrifuged at 4 °C, 90 min in the Centrifuge Sigma 4K15C/Rotor 09100 at 6,185 x g. Under the selected conditions, all Eppendorf Plate Deepwell proved durable; no split wells or cracks were observed. Competitor D plates, on the other hand, did not withstand this load (**Fig. 2**). Table 4 shows a comparison of Eppendorf Plate Deepwell with various competitors' plates with regard to durability.



Fig. 2: Fragmented competitor's plate following centrifugation with 70 % ethanol (4 °C, 90 min) at 6,189 x g in the Centrifuge Sigma 4K15C, Rotor 09100.

Eppendorf Plate Deepwell LoBind and Sterile

The Protein LoBind and DNA LoBind plates behaved just the same as the Eppendorf Plate Standard quality in terms of durability and can be centrifuged up to 6,000 x g (data not shown). On the other hand, the tests on sterile plates had different results, as they proved stable to centrifugation up to 5,000 x g (40 °C, 90 min) (Sigma 4K15C/Rotor 09100) (data not shown). Eppendorf Plate Deepwell are sterilized by irradiation. This makes the material more brittle than the standard material, and its centrifugation strength is consequently somewhat reduced.

Stability to centrifugation of Eppendorf Plate Deepwell	
Standard	6,000 x g
Protein LoBind	6,000 x g
DNA LoBind	6,000 x g
Sterile	5,000 x g

Table 4: Results of centrifugation of a 70 % ethanol solution at 4 °C for 90 min in the Centrifuge Sigma 4K15C, Rotor 09100. Eppendorf Plate Deepwell 96/2000 µl and 96/1000 µl and corresponding deepwell plates from other suppliers were tested.

- = plates durable (no split wells or cracks)
- ▲ = plates cracked or wells split

Sigma 4K15 Rotor 09100 6,189 x g, 4 °C, 90 minutes, 70 % Ethanol	Durability
Eppendorf Plate Deepwell 96/2000 µl	●
Competitor A	●
Competitor B	●
Competitor C	●
Competitor D	▲
Eppendorf Plate Deepwell 96/1000 µl	●
Competitor A	●
Competitor B	●
Competitor D	▲

Ordering information

Eppendorf Plate Deepwell 384/200 µl*

Name	Quality	Color**	Packaging	Order no. international	Order no. North America
Regular package	Standard	red	40 plates (5 bags of 8)	0030 521.129	951031046
	Sterile	red	40 plates (5 bags of 8)	0030 522.125	951031143
	DNA LoBind (also for RNA & other nucleic acids)	red	40 plates (5 bags of 8)	0030 523.121	951031241
	Protein LoBind	red	40 plates (5 bags of 8)	0030 524.128	951031348
Large package	Standard	red	120 plates (10 bags of 12)	0030 525.124	951031445
	Sterile	red	120 plates (10 bags of 12)	0030 526.120	951031542
	DNA LoBind (also for RNA & other nucleic acids)	red	120 plates (10 bags of 12)	0030 527.127	951031640
	Protein LoBind	red	120 plates (10 bags of 12)	0030 528.123	951031747

Eppendorf Plate Deepwell 96/500 µl*

Name	Quality	Color**	Packaging	Order no. international	Order no. North America
Regular package	Standard	red	40 plates (5 bags of 8)	0030 501.128	951031844
	Sterile	red	40 plates (5 bags of 8)	0030 502.124	951031941
	DNA LoBind (also for RNA & other nucleic acids)	red	40 plates (5 bags of 8)	0030 503.120	951032042
	Protein LoBind	red	40 plates (5 bags of 8)	0030 504.127	951032140
Large package	Standard	red	120 plates (10 bags of 12)	0030 505.123	951032247
	Sterile	red	120 plates (10 bags of 12)	0030 506.120	951032344
	DNA LoBind (also for RNA & other nucleic acids)	red	120 plates (10 bags of 12)	0030 507.126	951032441
	Protein LoBind	red	120 plates (10 bags of 12)	0030 508.122	951032549

*All Deepwell plates are available with bar code upon request.

**Available in five frame colors (white, yellow, red, green, blue).

Ordering information**Eppendorf Plate Deepwell 96/1000 µl***

Name	Quality	Color**	Packaging	Order no. international	Order no. North America
Regular package	Standard	red	20 plates (5 bags of 4)	0030 501.225	951032646
	Sterile	red	20 plates (5 bags of 4)	0030 502.221	951032743
	DNA LoBind (also for RNA & other nucleic acids)	red	20 plates (5 bags of 4)	0030 503.228	951032841
	Protein LoBind	red	20 plates (5 bags of 4)	0030 504.224	951032948
Large package	Standard	red	80 plates (10 bags of 8)	0030 505.220	951033049
	Sterile	red	80 plates (10 bags of 8)	0030 506.227	951033146
	DNA LoBind (also for RNA & other nucleic acids)	red	80 plates (10 bags of 8)	0030 507.223	951033243
	Protein LoBind	red	80 plates (10 bags of 8)	0030 508.220	951033341

Eppendorf Plate Deepwell 96/2000 µl*

Name	Quality	Color**	Packaging	Order no. international	Order no. North America
Regular package	Standard	red	20 plates (5 bags of 4)	0030 501.322	951033448
	Sterile	red	20 plates (5 bags of 4)	0030 502.329	951033545
Large package	Standard	red	80 plates (10 bags of 8)	0030 505.328	951033642
	Sterile	red	80 plates (10 bags of 8)	0030 506.324	951033740

*All Deepwell plates are available with bar code upon request.

**Available in five frame colors (white, yellow, red, green, blue).

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