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TPP TubeSpin® Bioreactor 50: Conical vs Round Tube Bottom

The TPP TubeSpin® bioreactor 50 mL is available with either a round "U" (#87050) or conical "V" (#87056) bottom. The choice of bioreactor bottom can affect the cultivation of cells as well as subsequent steps such as harvesting cells or supernatant by centrifugation.

TPP recommends a shaking diameter of 50 mm because the oxygen transfer rate (OTR) at constant speed (RPM) is greater for larger diameters (see: TechDoc available from TPP). If it is not possible to select the recommended 50 mm diameter, TPP recommends testing and optimizing the cell culture conditions.

In a study, the influence of the selected tube bottom, the influence of media volume, inclination angle and shaking speed were tested by culturing Chinese hamster ovary (CHO) cells for 14 days with a shaking diameter of 25 mm and a speed of 210 rpm. The results show a correlation between shaking speed, media volume and selected inclination angle.

CHO cells cultivated in 15 mL medium

CHO cells were cultured in 15 mL of media for 14 days in 50 mL TPP TubeSpin® bioreactors with either round "U" or conical "V" bottom in a test tube holder with adjustable inclination angle. The incubator shaker had a shaking diameter of 25 mm and the shaking speed was set at 210 rpm (Table 1). The number of viable and total cells was determined before subcultivation using Vi-Cell XR. CHO cells were inoculated into TPP TubeSpin® bioreactors at an initial cell density of 5×10^5 viable cells/mL on Monday and Wednesday and 3×10^5 viable cells/mL on Friday.

Table 1: Shaking parameter used with TPP TubeSpin® bioreactor 50 mL round “U” and conical “V” bottom

TPP TubeSpin® Bioreactor 50	Conical “V” (order-no.: 87050) Round “U” (order-no.: 87056)
Shaking speed rpm	210
Shaking diameter mm	25
Working volume mL	15
Inclination angle	0° 35°

CHO cells in 15 mL culture media showed no differences in cell growth and viability between conditions with or without inclination (35° or 0°) and between TPP TubeSpin® bioreactor 50 mL with either a round "U" or conical "V" bottom (Figure 1).

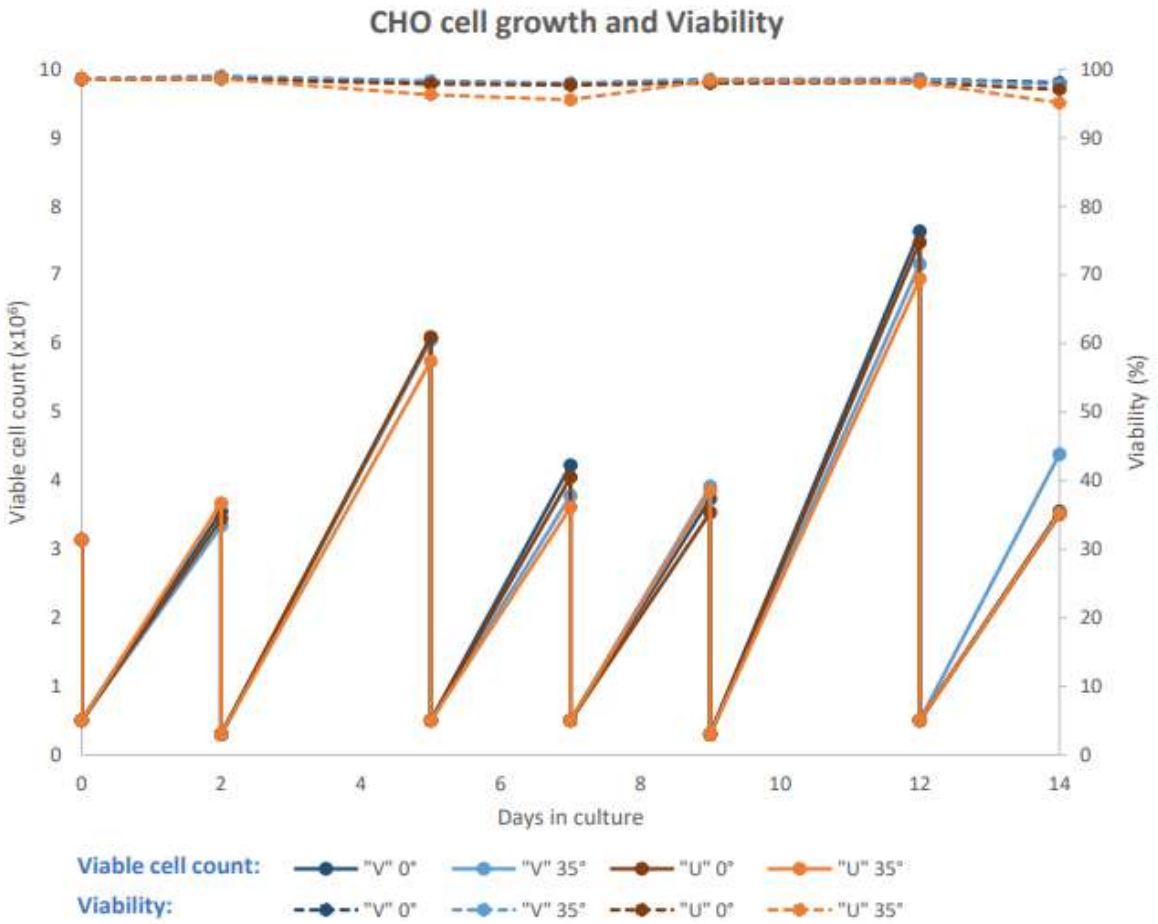

Figure 1: Either CHO cells were cultured in TPP TubeSpin® bioreactor 50 mL with a round "U" or a conical "V" bottom with an angle of inclination of 0° and 35° alternatively. CHO cells were cultured in 15 mL of media for 14 days at a shaking diameter of 25 mm and a speed of 210 rpm. Vi-Cell XR was used to determine the number of viable cells and viability. The subcultivation schedule was Monday-Wednesday-Friday.

1. CHO cells cultivated in 30 mL medium

CHO cells were cultured in 30 mL of media for 14 days in 50 mL TPP TubeSpin® bioreactors with either round "U" or conical "V" bottom in a test tube holder with adjustable inclination angle. The incubator shaker had a shaking diameter of 25 mm and a shaking speed of 210 rpm (Table 2). The number of viable and total cells was determined before subcultivation with Vi-Cell XR. CHO cells were inoculated into TPP TubeSpin® bioreactors at an initial cell density of 5×10^5 viable cells/mL on Monday and Wednesday and 3×10^5 viable cells/mL on Friday.

Table 2: Shaking parameter used with TPP TubeSpin® bioreactor 50 mL either with round “U” or with conical “V” bottom

TPP TubeSpin® Bioreactor 50	Conical “V” (order no: 87050) Round “U” (order no: 87056)
Shaking speed rpm	210
Throw mm	25
Working volume mL	30
Inclination angle	0° 35°

CHO cells in 30 mL of culture medium at 0° inclination settled in the bottom of a 50 mL TPP TubeSpin® bioreactor 50 mL with either a round "U" or conical "V" bottom. This resulted in reduced viability and slower cell growth. The conical "V" bottom showed earlier cell settling. In contrast, cultivation at a 35° slope showed equivalent results with no loss of viability and the expected cell growth (Figure 2).

Further testing showed that increasing the shaking speed to 230 rpm allowed cultivation in 30 mL at 0° inclination (data not shown).

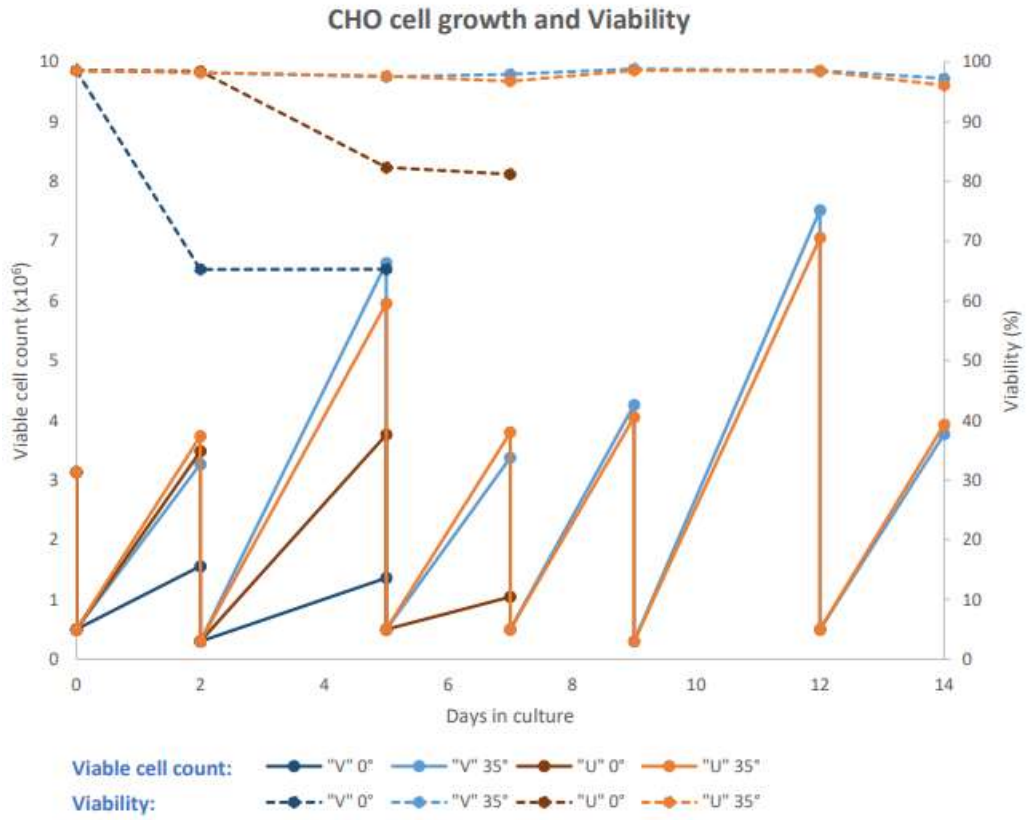

Figure 2: CHO cells were cultivated in TPP TubeSpin® Bioreactors 50 with either round “U” or with conical “V” bottom and an inclination angel of 0° and 35°. CHO cells were cultivated in 30 mL media volume for 14 days, with a shaking diameter of 25 mm and a speed of 210 rpm. Vi-Cell XR determined viable cell count and the viability. The passage schedule was Monday-Wednesday-Friday.

Summary of the Results:

CHO cells were grown in 15 mL of medium at 0° and 35° inclination, 210 rpm shaking speed and 25 mm shaking diameter in 50 mL TPP TubeSpin® Bioreactors 50 mL with either a round "U" or conical "V" bottom:

- ✓ Expected performance and equivalent cell growth and viability results

CHO cells were cultured in 30 mL of medium at 0° inclination, 210 rpm shaking speed, and 25 mm shaking diameter in 50 mL TPP TubeSpin® Bioreactors 50 mL with either round "U" or conical "V" bottoms:

- ✓ Cells settle to the bottom of the tube
- ✓ Conical bottom conditions lead to earlier cell settling
- ✓ Loss of viability and slower cell growth
- ✓ Cultivation in 30 mL at 0° inclination without loss of viability is possible by increasing the in-shaking speed from 210 rpm to 230 rpm.

CHO cells were grown in 30 mL of medium at 35° inclination, 210 rpm shaking speed and 25 mm diameter in TPP TubeSpin® Bioreactors 50 mL with either round "U" or conical "V" bottom:

- ✓ No cell settling, normal cell growth and viability
- ✓ No difference between conical and round bottom tubes

Technical Data TPP TubeSpin® bioreactor 50 mL

Materials

Screw cap	PE
Membrane	PTFE
Tube	PP

Measurements	**87050**	**87056**
Volume graduation mL	50	50
Length mm	115	115
Diameter mm	30	30
Max. RCF x g	15‘500	15‘500
Form	“V” conical	“U” round
Optimal filling volume mL	1 – 35	1 – 35
Shaker recommendations: Shaking diameter (orbit / throw) mm	50	50
Shaker recommendations: Speed rpm	180	180