APPLICATION NOTE

Safely Using a Centrifuge Near a Biological Safety Cabinet in Your Cell Culture Laboratory

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Key Words
Cell culture, biological safety cabinet, BSC, centrifuge, contamination

The placement of a general purpose centrifuge in relation to a biological safety cabinet (BSC) was analyzed to see if improvements could be made to optimize the ergonomics and workflow while maintaining the safety requirements for BSCs (EN12469[1]) and centrifuges (EN 61010-2-020[2]). The results showed that a Thermo Scientific™ 4-liter floor-standing refrigerated general purpose centrifuge[3] can be placed 30 cm from a Thermo Scientific™ Class II biological safety cabinet[4] aperture without interfering with containment in accordance to EN12469. This provides an outstanding option for improving ergonomics, reducing processing times and optimizing cleaning procedures.

Introduction

Centrifuges and biological safety cabinets are key equipment for cell culture laboratories. Traditionally, centrifuges are placed on laboratory benches well away from the BSC working area due to size and potential for airflow interference. This distance impacts the workflow efficiency, provides greater opportunity for contamination and can increase the time the cells are outside of the ideal growth conditions of a CO_2 incubator.

BSCs provide containment of hazardous biological aerosol within their sample chamber and prevent the entry of contamination into that same sample chamber by establishing an air barrier at the BSC front aperture. Traditional refrigerated centrifuges require large air movement through the refrigeration system at the side or the front of the centrifuge. The air currents caused by traditional refrigerated centrifuges could interfere with the air barrier at the BSC's front aperature, which could compromise safety and containment. Thermo Scientific 4-liter floor-standing refrigerated general purpose centrifuges have a special air flow design that takes air from the lower front of the centrifuge and then exhausts it to the lower rear.

A laboratory was set up to test and validate a Thermo Scientific Class II biological safety cabinet for containment, while using a Thermo Scientific 4-liter floor-standing refrigerated general purpose centrifuge in close proximity to the BSC opening. Standard KI-DISCUS™[5] test equipment was used to test containment, as per EN12469. The test generates a potassium iodide aerosol inside the BSC and escaping material is monitored with centripetal collectors outside of the BSC.

EN12469 requires that a Class II BSC demonstrate an Aperature Protection Factor (APF) of at least 1 x 10^5.

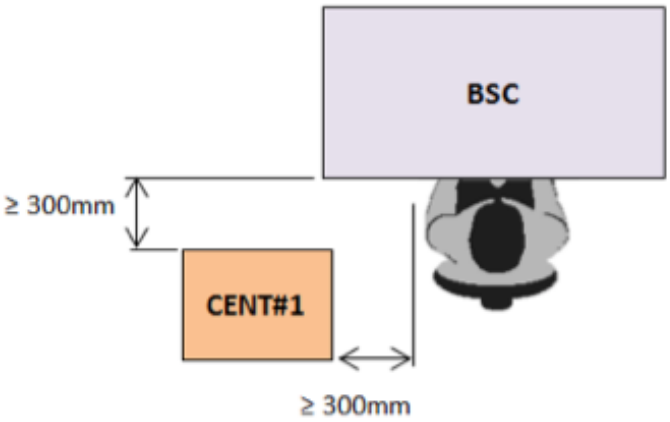
Methods

Using a Thermo Scientific™ MSC-Advantage™ Class II 1.2-meter biological safety cabinet, the device was set to the manufacturer's airflow velocity set points. Downflow and inflow were stabilized and velocities were measured and recorded as set out in EN12469.

The next step was to position the Thermo Scientific 4-liter refrigerated floor-standing centrifuge in close proximity to the BSC. In accordance to EN61010-2-020, the centrifuge was provided with a 300 mm safe zone around the centrifuge. The centrifuge was placed 300 mm from the BSC and 300 mm from an operator's elbow during a simulated use. See diagram 1 and diagram 2.

The centrifuge was set to 4° C to ensure the refrigeration system was operating at full power with maximum air movement.

Diagram 1: Centrifuge Position 1



Test 1 – Centrifuge Position 1 (Left Side)
We performed a full KI-DISCUS test for approximately 9 minutes.

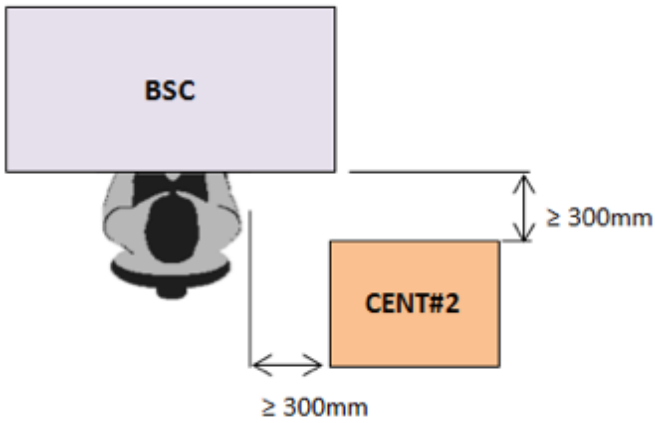
Test 2 – Centrifuge Position 1 (Left Side) with lid opened and closed. The BSC was in full operation and the KI-DISCUS test was running as detailed above.

The BSC was in full operation and the KI-DISCUS test was running as detailed above. Additionally, the centrifuge lid was fully opened and closed two times during the test to mimic normal working procedures with the centrifuge. If there were any disturbance to the BSC front airflow curtain from the centrifuge lid opening this would be detected in the KI-DISCUS test results.

The lid openings were powered by the centrifuge gas strut, which lifts to the fully open position; the lid was closed manually; slowly and steadily over 3 seconds.

Test 3 – Centrifuge Position 2 (Right Side) Procedure as per Test 1, but with centrifuge on right side of cabinet.

Diagram 2: Centrifuge Position 2



Test 4 – Procedure as per Test 2, but with centrifuge on right side of cabinet.

Table 1

Instrument	Serial number	Calibration date
Thermo Scientific MSC-Advantage Class II Biological Safety Cabinet, 1.2m	50123318	N/A
Anemometer	00110414	6/15
KI-DISCUS	K09940068	8/14
Dräger Air Flow Tester	N/A	N/A

Equipment used

- Anemometer from TSI™
- KI-DISCUS 2 from Containment Technology Ltd
- Dräger™ Air Flow Tester from Drägerwerk AG & Co. KGaA

BSC Set Points and Parameters

- The MSC-Advantage 1.2-meter biological safety cabinet airflows set out per EN12469 and as outlined in Table 2 and 4
- EN12469 states mean downflow to achieve product protection 0.25m/s – 0.50m/s
- Inflow velocity must be >0.40m/s

Table 2

100mm above edge of sash			
Downflow average = 0.31m/s			
0.30	0.30	0.31	0.31
0.32	0.32	0.31	0.30
Inflow = 0.45m/s			

Table 3

BSC Parameters	Setting
S1 – Downflow fan	68%
S2 – Inflow fan	79%
S3 – Downflow low alarm	52%
S4 – Inflow low alarm	60%

Testing conditions

All testing completed in the testing laboratory at Thermo Fisher Scientific, Langenselbold, Germany.

Results

Test 1 and 2 (Centrifuge position 1 – left side of cabinet)

The BSC passed the KI-DISCUS test for containment. Results from Test 1, which did not include opening or closing the centrifuge lid is shown in Table 4. Table 5 shows the dynamic testing involving lid opening and closing twice during test procedure.

Table 4

Centrifuge Left Side				
KI-DISCUS Test - Test Position - Central				
	Sample Head			
	X	**Y**	**X1**	**Y1**
Run 1	10	3	11	9
Run 2	4	6	3	8
Run 3	5	9	11	5
Run 4	5	9	4	4
Run 5	6	7	6	8
APF = 5.6×10^5 PASS				

Table 5

Centrifuge Left Side + Lid Openings				
KI-DISCUS Test - Test Position - Central				
	Sample Head			
	X	**Y**	**X1**	**Y1**
Run 1	7	5	8	9
Run 2	6	10	11	4
Run 3	9	7	14	5
Run 4	6	7	10	8
Run 5	4	3	8	12
APF = 4.4×10^5 PASS				

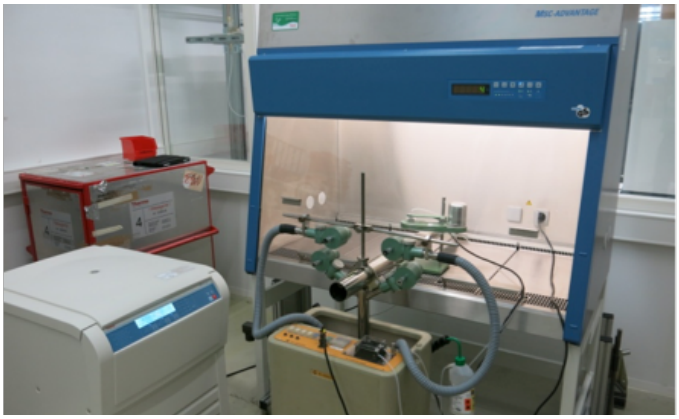


Figure 1

Test 3 and 4 (Centrifuge position 2 – right side of cabinet)

Table 6 shows KI-DISCUS containment results from test 1, which did not involve the lid of the centrifuge being opened and shut. Table 5 shows the dynamic testing involving lid opening and closing twice during test procedure.

The BSC passed the KI-DISCUS test for containment. Results from Test 1, which did not include opening or closing the centrifuge lid is shown in Table 6. Table 7 shows the dynamic testing involving lid opening and closing twice during test procedure.

Smoke testing using Dräger air flow testers was also completed. Smoke was introduced in and around the aperture to visualize any turbulence the centrifuge may cause by being located 300 mm from the BSC. No turbulence was witnessed and a good inflow of smoke was visually seen entering the BSC aperture. All smoke was captured in the BSC. See Figure 2.

Table 6

Centrifuge Right Side				
KI-DISCUS Test - Test Position - Central				
	Sample Head			
	X	Y	X1	Y1
Run 1	4	9	9	8
Run 2	5	3	7	4
Run 3	7	8	10	6
Run 4	2	5	6	2
Run 5	3	4	1	4
APF = 6.2×10^5 PASS				

Table 7

Centrifuge Right Side + Lid Openings				
KI-DISCUS Test - Test Position - Central				
	Sample Head			
	X	Y	X1	Y1
Run 1	5	8	9	4
Run 2	3	6	9	10
Run 3	4	3	8	7
Run 4	4	7	2	8
Run 5	7	7	8	2
APF = 6.2×10^5 PASS				

The smoke test and KI-DISCUS test results proved that an operating Thermo Scientific 4-liter refrigerated floor-standing centrifuge could be placed in Position 1 or 2 (close proximity to the BSC) without compromising BSC containment. The BSC met the containment test acceptance criteria provided in EN 12469. The testing also showed that slow and steady lid opening and closure did not impact the BSC containment.

Figure 2

Conclusion

The results show that Thermo Scientific 4-liter refrigerated floor-standing centrifuges can be positioned >300mm from a Thermo Scientific Class II biological safety cabinet and still provide containment in accordance with EN12469. This provides new opportunities for laboratory designers and cell culture personnel to improve ergonomics and workflow efficiencies within cell culture suites. Improving the workflow efficiency by minimizing movement to and from a BSC to a centrifuge will help to reduce the processing time of cells and help to reduce overall time that cells are outside of a BSC. This also reduces over time outside of the ideal growth environment inside the CO_2 incubator. Reducing this time is becoming increasingly important as more delicate cell types are investigated.

Moving the centrifuge off the bench eliminates hard to clean areas beneath and behind the centrifuge, which can harbor contamination that can spread around laboratories and eventually to cultures. Another benefit of floorstanding centrifuges is that they can be unlocked and wheeled around for quick and efficient cleaning.

There are also specific ergonomic benefits for technicians and researchers as benchtop centrifuges can be >1300mm tall and can be at a height difficult for some users to properly reach. The Thermo Scientific 4-liter floor-standing centrifuge has an ergonomic height of just 840 mm, which gives outstanding access even when sitting in a laboratory chair.

These results strictly apply to Thermo Scientific 4-liter refrigerated floor-standing centrifuges and Thermo Scientific Class II biological safety cabinets. Other combinations of centrifuges and BSCs will have differing air movement patterns (particularly from centrifuges), which could disrupt containment of a BSC. Airflow and containment disruption represents a safety risk for users and a contamination risk for cell cultures. Thus, the results presented here demonstrate improved ergonomics, safety and time saving for users. For cultured cells, the positioning of the floor-standing centrifuge close to the BSC results in reduced contamination risk and better growth conditions due to reduced handling and time outside the CO_2 incubator.

Reference

1. SEN12469: 2000 Biotechnology - Performance criteria for microbiological safety cabinets.

2. IEC 61010-2-020: 2016 Safety requirements for electrical equipment for measurement, control, and laboratory use - Part 2-020: Particular requirements for laboratory centrifuges.

3. Test applicable to Thermo Scientific™ Sorvall™ Legend™ XFR general purpose centrifuge, Thermo Scientific™ Heraeus™ Multifuge™ X3FR general purpose centrifuge and Thermo Scientific™ SL 40FR general purpose centrifuge.

4. Test completed on Thermo Scientific MSC-Advantage 1.2-meter biological safety cabinet. Due to identical airflows the test is also applicable to the Thermo Scientific KS12 and KSP12 biological safety cabinets and Thermo Scientific Safe 2020 1.2-meter biological safety cabinet.

5. Clark, Raymond P. KI-DISCUS operating instructions. Containment Technology Limited. Retrieved 5 January, 2016 from http://www.kidiscus.com.

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