TECHNICAL BULLETIN : GENERAL INFORMATION

Impact of Class II Biological Safety Cabinet Downflow Velocity on Cross Contamination

Background

Over the last several years, Biological Safety Cabinet (BSC) manufacturers have lowered downflow velocity while maintaining inflow velocity in an effort to increase energy efficiency, reduce exhaust system requirements and lower BSC Noise and vibration levels. However, the lowering of downflow velocity may come with a consequence of greater lateral airflow movement within the workzone creating greater potential for cross contamination. The lowering of the downflow velocity does not compromise personnel and product containment performance. Why? Primarily, by maintaining inflow velocity above a certain level as required by the standard, typically 100 fpm (.51 mps). However, the standard does not require a specific downflow velocity and can be adjusted as needed based on the design parameters of the workzone supply diffuser and grill patterns (airflow distribution) to assure compliance to the personnel and product protection requirements of NSF/ANSI 49. NSF/ANSI 49 test procedures do test for cross contamination, but only at the workzone sidewalls, which doesn't necessarily translate into the lateral movement of airflow in the center of the workzone. The result of these lower downflow velocities may be a greater level of sample cross contamination using traditional work practice guidelines.

BMBL Guidelines

User work practice guidelines as stated in the BMBL 5th edition, Appendix A, Section V, Operation within a class II BSC states "Class II cabinets are designed so that horizontally nebulized spores introduced into the cabinet will be captured by the downward flowing cabinet air within 14 inches of travel. Therefore, as a general rule of thumb, keeping clean materials at least one foot away from aerosol-generating activities will minimize the potential for cross-contamination."

Historical Background

These guidelines were established in the 1970's when most all Class II BSC's operated with an average downflow velocity of 80 fpm (.41mps). This was a requirement for the NIH-03-112C Class II, type 1 BSC design and performance specification, which was the basis of BSC design and performance for all manufacturers at that time. In 1976 the first NSF standard 49 didn't state an explicit downflow requirement, but did state "The velocity of any single point cannot be below 45 fpm (.23 mps)", which means the lowest *average* downflow velocity could not be below 57 fpm (.29 mps). Most manufacturers maintained higher downflow velocities until the mid-1990's, and since began to reduce them for lower vibration and noise levels. Recently, over the last several years manufacturers have reduced downflow velocities for the improved energy efficiency. Today the mean average downflow velocity is 60 fpm (.30 mps) with some manufacturers going as low as 40 fpm (.20 mps). Lowering of the average downflow velocity seemed to provide the best design solution while maintaining containment performance to NSF/ANSI 49, but the question that needs to be answered, how low can a downflow average be, before it causes cross contamination according to industry standard work practice guidelines?

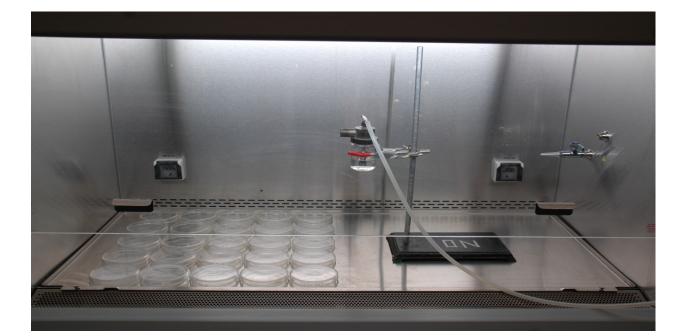
Cross Contamination Testing

In an effort to begin to determine the answer to the above question, the following testing was performed to evaluate the extent of lateral downflow air movement or potential cross contamination using a modified NSF/ANSI 49 cross contamination test method. The test modifications used placed the nebulizer position in the center of the workzone (on the front to rear airflow spilt line) and 14 inches above the work surface in an effort to simulate where most aerosols would be generated in normal cabinet use.

Test Materials: NuAire Model NU-480-400 Class II, type A2 BSC Collision CN-38 nebulizer with 5.0 x 10⁴ B. Subtilis spores Agar plates (100mm)

Test Method:

- 1. Set cabinet airflows for each test run as required.
- 2. Locate and identify downflow front to rear split line in center of cabinet.
- 3. Place nebulizer in workzone so the center of the nebulizer nozzle is located on front to rear split line facing right and 14 inches above the worksurface.
- 4. Place open agar settling plates on worksurface from front to rear 5 rows across starting with right edge of agar plates placed in the center of the cabinet.
- 5. Start nebulizer. After 5 minutes, stop nebulizer.
- 6. After 15 minutes, place covers on the agar plates. Incubate for 24 hours and record results.
- Run triplicate tests for each downflow set point of: 80fpm (.41mps), 70 fpm (.35mps), 60fpm (.30mps), 50fpm (.25mps), 40fpm (.20mps), 30fpm (.15mps).



Test Set-up

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Test Results:

Test 1 Inflow 105fpm (.53mps) Downflow 80fpm (.41mps)

Run 1					
Ø	Ø	Ø	Ø	Ø	
Ø	Ø	Ø	Ø	30	
Ø	Ø	Ø	Ø	≈ 100	
Ø	Ø	Ø	Ø	26	
Ø	Ø	Ø	Ø	4	

Γ.						
	Run 2					
	Ø	Ø	Ø	Ø	Ø	
	Ø	Ø	Ø	Ø	30	
	Ø	Ø	Ø	Ø	≈ 100	
	Ø	Ø	Ø	Ø	22	
	Ø	Ø	Ø	Ø	3	

Run 3					
Ø	Ø	Ø	Ø	1	
Ø	Ø	Ø	Ø	41	
Ø	Ø	Ø	Ø	≈ 100	
Ø	Ø	Ø	Ø	34	
Ø	Ø	Ø	Ø	Ø	

Test 2 Inflow 1	05fpm (.53mps)	Downflow 70fpm
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Run 1						
Ø	Ø	Ø	Ø	Ø		
Ø	Ø	Ø	Ø	1		
Ø	Ø	Ø	Ø	≈ 100		
Ø	Ø	Ø	Ø	15		
Ø	Ø	Ø	Ø	1		

s) Downflow 70fpm (.35mps)						
		Run	2			
Ø	ØØØØØ					
Ø	Ø	Ø	Ø	1		
Ø	Ø	Ø	Ø	>100		
Ø	Ø	Ø	Ø	≈ 50		
Ø	Ø	Ø	Ø	14		

Run 3						
Ø	Ø	Ø	Ø	Ø		
Ø	Ø	Ø	Ø	17		
Ø	Ø	Ø	Ø	>100		
Ø	Ø	Ø	Ø	88		
Ø	Ø	Ø	Ø	9		

Test 3 Inflow 105fpm (.53mps) Downflow 60fpm (.30mps)

		Run	1	
Ø	Ø	Ø	Ø	2
Ø	Ø	Ø	Ø	93
Ø	Ø	Ø	1	>100
Ø	Ø	Ø	Ø	24
Ø	Ø	Ø	Ø	10

Run 2					
Ø	Ø	Ø	Ø	Ø	
Ø	Ø	Ø	Ø	15	
Ø	Ø	Ø	Ø	>100	
Ø	Ø	Ø	1	82	
Ø	Ø	Ø	Ø	7	

Run 3						
Ø	Ø	Ø	Ø	10		
Ø	Ø	Ø	1	>100		
Ø	Ø	Ø	1	>100		
Ø	Ø	Ø	Ø	13		
Ø	Ø	Ø	Ø	3		

<u>Test 4</u> Inflow 105fpm (.53mps) Downflow 50fpm (.25mps)

Run 1						
Ø	Ø	Ø	Ø	28		
Ø	Ø	Ø	1	>100		
Ø	Ø	Ø	1	>100		
Ø	Ø	Ø	Ø	3		
Ø	Ø	Ø	Ø	1		

Run 2					
Ø	Ø	Ø	5	34	
Ø	Ø	Ø	1	>100	
Ø	Ø	Ø	2	>100	
Ø	Ø	Ø	2	10	
Ø	Ø	Ø	Ø	3	

	Run 3						
Ø	Ø	Ø	2	32			
Ø	Ø	Ø	5	>100			
Ø	Ø	Ø	4	>100			
Ø	Ø	Ø	Ø	9			
Ø	Ø	Ø	Ø	1			

Test 5 Inflow 105fpm (.53mps) Downflow 40fpm (.20mps)

Run 1						
Ø	Ø	Ø	2	101		
Ø	Ø	Ø	9	>100		
Ø	Ø	Ø	1	>50		
Ø	1	1	3	5		
Ø	Ø	Ø	Ø	1		

Run 2					
Ø	Ø	Ø	4	89	
Ø	Ø	Ø	1	>100	
Ø	Ø	Ø	1	>100	
Ø	Ø	2	Ø	12	
Ø	Ø	Ø	Ø	3	

Test 6 Inflow 105fpm (.53mps) Downflow 30fpm (.15mps)

			-			
Run 1						
Ø	Ø	Ø	1	52		
Ø	Ø	Ø	2	>100		
Ø	Ø	Ø	Ø	3		
Ø	Ø	Ø	Ø	Ø		
Ø	Ø	Ø	Ø	Ø		

Run 2					
Ø	Ø	Ø	Ø	6	
Ø	Ø	Ø	Ø	36	
Ø	Ø	Ø	Ø	7	
Ø	Ø	Ø	Ø	Ø	
Ø	Ø	Ø	Ø	Ø	

Run 3					
Ø	Ø	Ø	Ø	83	
Ø	Ø	Ø	2	>100	
Ø	Ø	Ø	Ø	92	
Ø	Ø	Ø	1	92	
Ø	Ø	Ø	Ø	Ø	

Run 3						
Ø	Ø	Ø	9	68		
Ø	Ø	Ø	2	>100		
Ø	Ø	Ø	Ø	5		
Ø	Ø	Ø	Ø	Ø		
Ø	Ø	Ø	Ø	Ø		

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Discussion of the testing results

The testing results in general indicate greater lateral airflow movement with lower average downflow velocities. At both 80 fpm (.41 mps) and 70 fpm (.35 mps) average downflow velocities, the B. subtilis lateral travel was limited to the first row of plates or 4 inches (102mm) of travel from the center of the work zone. At both 60 fpm (.30 mps) and 50 fpm (.25 mps) average downflow velocities, the B.subtilis lateral travel migrated to the second row or 8 inches (203mm) from the center of the work zone. At 40 fpm (.20 mps), the B.subtilis lateral travel migrated to the third row or 12 inches (305mm) from the center of the workzone (note at this point, there is a conflict with the BMBL work practice rule of one foot or 12 inches (305mm)). At 30 fpm (.15 mps) average downflow velocitinue to see even greater lateral migration. However, the B. subtilis lateral travel only migrated to the second row or 8 inches (203mm) from

To understand what was happening at the 30 fpm (.15 mps) average downflow velocity, we used smoke to visualize the airflow patterns of the test setup (see photo's below). What we found was as the B. subtilis airflow was leaving the nebulizer, the majority of the aerosolized B. subtilis was very gently being pushed down the nebulizer bottle then pulled into an eddy under the bottom of the bottle over and away from the agar plates. The velocity of the eddy was greater than the downflow velocity taking the nebulized B. subtilis with it and away from the agar plates. When evaluating the smoke patterns in general, there was also some vertical refluxing around the nebulizer where the nebulizer eddy was strong enough to overcome the normal downflow. The normal downflow in the open areas does push the air towards the work surface. When evaluating the smoke patterns just over the testing plates, there was considerable lateral movement across all five rows directly on the front to rear spilt line. Lastly, the smoke patterns indicated that if a standard cross contamination test was performed on the work zone sides with an average downflow of 30 fpm (.15 mps), it would pass. The smoke was easily pulled directly toward the cabinet sidewall with no reflux or change in direction.

Conclusions

As stated above, the testing results indicate greater lateral airflow movement with lower average downflow velocities. Average downflow velocities above 50 fpm (.25 mps) as tested document that a BSC will provide cross contamination protection to within industry accepted work practice guidelines. Average downflow velocities below 50 fpm (.25 mps) tested outside industry accepted work practice guidelines. As a result, work practice guidelines may have to be modified to minimize cross contamination risk. As an additional note, the above testing results are representative in a static test condition. Dynamic operator movement is also a consideration for any work practice modifications. Smoke pattern testing can aid to visualize airflow within the workzone and assure proper work practice guidelines and modifications for low downflow velocity cabinets.

Photos of Airflow Smoke Patterns at 30fpm (1.5mps)



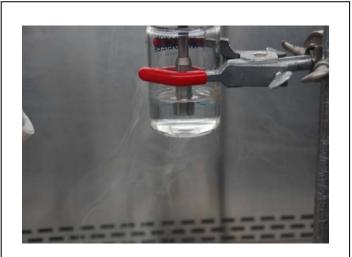
Downflow pushes smoke down towards the work surface



Lateral smoke movement across test plates



Airflow at sidewalls being pulled under work surface



Nebulizer Eddy smoke reflux