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Positive Pressure, Clean Environments in Automated Workflows on NIMBUS through Clean Air Protection (CAP) System with HEPA Filtration

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Introduction

High Efficiency Particulate Air (HEPA) filters are used across a wide range of industries and applications to protect materials in a working environment from contaminating airborne particulates down to 0.3 µm with 99.97% efficiency. In a HEPA filtration system, air is drawn in from the room environment and through the HEPA filter where particulates are trapped. The resulting clean air continuously flows over the work environment, creating positive pressure to prevent unwanted air from entering the enclosure.

Many options are available for HEPA filtration in clean workbenches, however, as they are intended for use with manual workflows, several issues arise when this equipment is applied to applications with increased throughput. The working area may be too small to comfortably integrate a robotic system and may prevent additional manual work from being performed within the space. A dedicated cabinet to house a robotic system is expensive and encompasses precious laboratory space. Additionally, the sheer volume of space required by the robotic system may compromise proper air flow in a dedicated cabinet and thereby increase risks of contamination.

The CAP (Clean Air Protection) system from Hamilton Robotics is an efficient alternative for those who require high throughput sample processing in a clean, positive-pressure environment. CAP is a HEPA-filtered hood that may be integrated with any standard, enclosed Microlab® NIMBUS Personal Pipetting Workstation. Air intake fans at the top of the unit pull air in and through polyurethane open cell foam pre-filters to catch large particles and protect the HEPA filter. The pre-filters have a 520-hour life span — equivalent to approximately three months of use — and may be washed, dried and reused, or replaced by users without the use of tools. The air then passes through the HEPA filter, which is 99.99% effective at removing particulates down to 0.3 µm and has a 2080-hour life span — equivalent to approximately twelve months of use. The HEPA filter is also user-replaceable without requiring tools. The HEPA filtered air flows onto the deck, maintaining positively pressured, clean air flow via an integrated flow sensor, even when the NIMBUS enclosure door is open. CAP features variable fan speed control, user-derived alarm settings and maintenance alerts, and real-time temperature and humidity levels are tracked and displayed on the unit. Additionally, CAP meets ISO 14644-11 standards, and verification reports are generated during installation and annually as part of preventative maintenance, which may be combined with NIMBUS preventative maintenance plans for added convenience. The integrated system may be controlled manually, via computer interface, or through user-driven scheduled events.

Benefits-Based Highlights

- Convenience of an integrated system for clean, high-throughput applications without the space limitations, bulky footprint, or expense of a biological safety cabinet.
- No need for separate preventative maintenance plan to address air filtration system.

Materials and Methods

Particle Size Testing

An Airy Technology model P311 handheld particle counter (Airy Technology, Stoughton, MA) was used to test for the presence of any 0.3 µm, 0.5 µm, and 5 µm particles contained within a Microlab NIMBUS equipped with a CAP system. Using aseptic technique, the decontaminated particle counter was placed onto one of eight areas on the NIMBUS deck, denoted as blue, lettered rectangles in Figure 1, and the NIMBUS enclosure deck was closed. The CAP fan speed was then set to 10% (950 RPM, 12.5 Watts, 0.26 Amps, 27 CFM), and the flow of air equilibrated for three minutes. After equilibration, 2.83 L of air was pulled through the particle counter over 60 seconds, and particle counter readings were recorded in particle counts per cubic meter. This process was repeated for each of the blue, lettered positions indicated in Figure 1.

Microbial Testing

The inside of a Microlab NIMBUS equipped with a CAP system was disinfected with Microcide (Microcide, Inc., Sterling Heights, MI) followed by 70% isopropyl alcohol (Thermo Fisher Scientific, Kalamazoo, MI). After disinfection, the NIMBUS enclosure door was closed, and the CAP fan speed was set to run at 10% (950 RPM, 12.5 Watts, 0.26 Amps, 27 CFM) overnight. The next day the door was opened to perform air sampling and the fan automatically ramped to the default of 80% (1980 RPM, 96 Watts, 1.9 Amps, 123 CFM) in order to maintain consistent air flow.

Using aseptic technique, a decontaminated Bioscience SAS 180 microbial air sampler (Bioscience International, Rockville, MD) was introduced into one of five designated areas on the NIMBUS deck denoted as green, numbered circles in Figure 1 and loaded with a sterile 55 mm contact plate (Bioscience International). The air sampler was noted as fully compliant with national and international standards for environmental air monitoring, including those set by ISO14698, EU GLP-GMP, EN45001, ACGIH, USP1116, and USP797, with capabilities of 100% sampling efficiency for particles down to 1 µm. The sampling volume on the air sampler was set to 1000 L, and testing commenced. Upon completion, the contact plate was removed from the sampler and labeled with deck location, date, and platform configuration. The test was repeated under identical aseptic conditions for each of the remaining deck positions shown as green, numbered circles in Figure 1, as well as a location on the benchtop outside the NIMBUS unit and exposed to ambient room conditions, which served as a positive control.

All contact plates were inverted immediately after sampling and placed in a Heratherm™ Advanced Protocol Microbiological Incubator (Thermo Fisher Scientific, Kalamazoo, MI) where they were maintained at 30 – 35 °C for 72 hours. Incubator conditions were then set to 20 – 25 °C and the plates were further incubated for an additional 72 hours. The plates were then visually inspected for evidence of contamination.

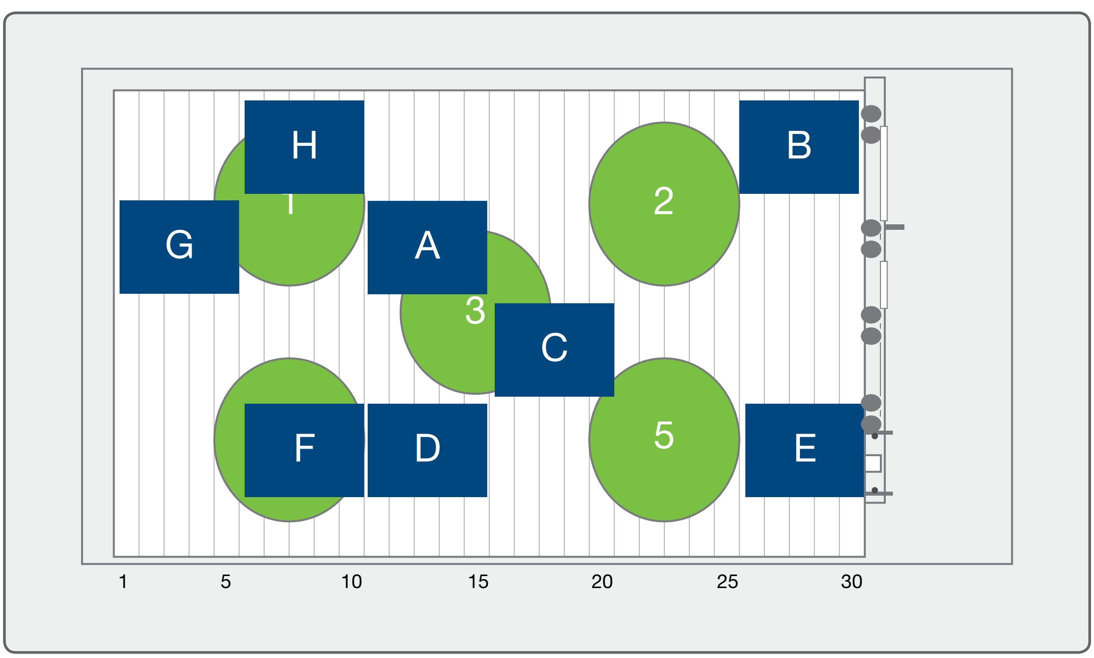

Figure 1: Representation of the NIMBUS deck, showing placement of the particle counter pedestal in eight air sampling locations (blue rectangles A – H) and placement of contact in five air sampling locations (green circles 1 – 5).

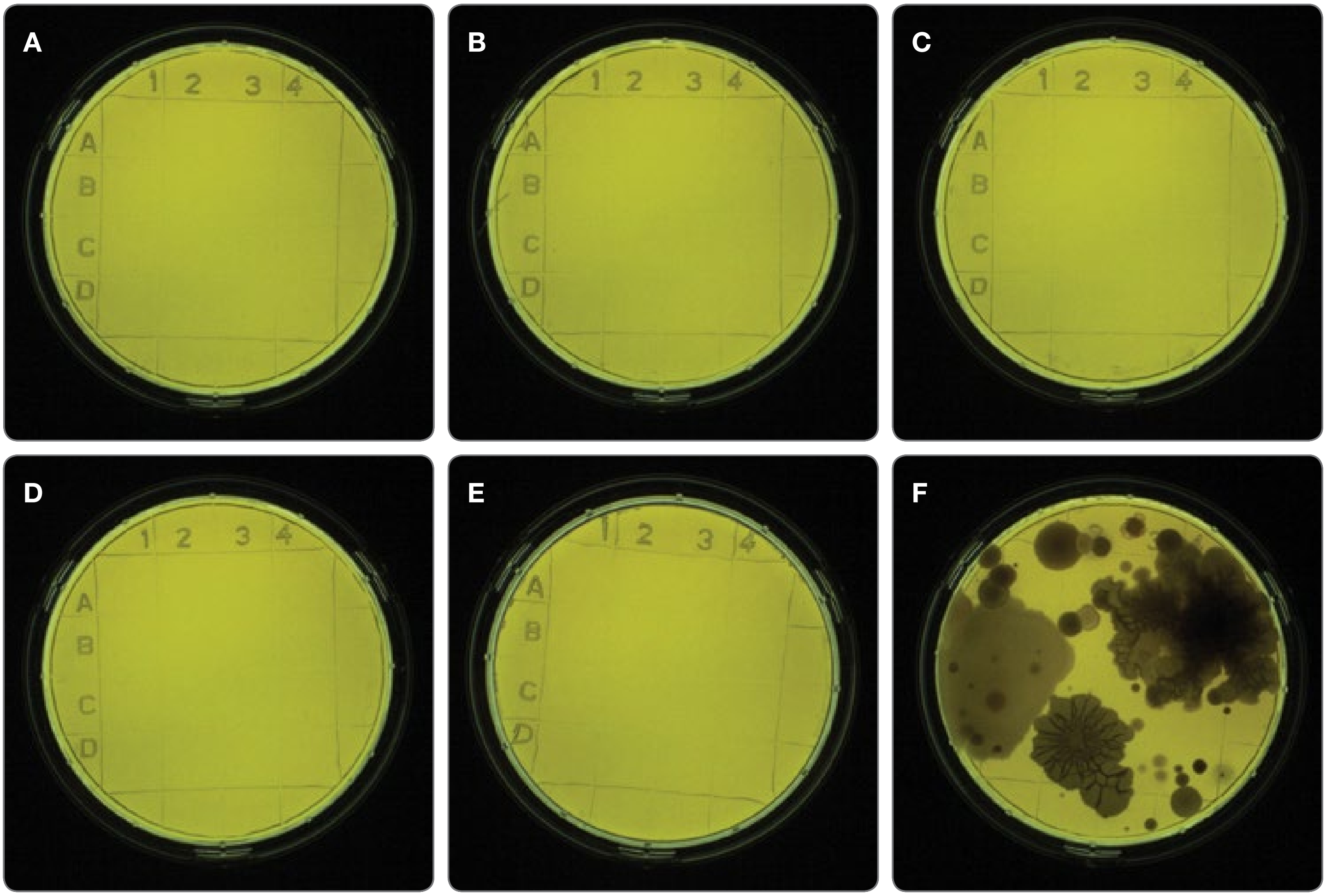

Figure 2: Air sampling results after incubation. Contact plates at NIMBUS deck air sampling locations (A) 1, (B) 2, (C) 3, (D) 4, and (E) 5 are free from contaminated growth, while (F) positive control contact plate contains multiple biological contaminants.

Results and Discussion

No particles of any size, including 0.3 µm which constitutes the most penetrating particle size (MPPS), were detected at any of the eight NIMBUS deck collection sites. Therefore, the CAP system is effective at particle filtration, and is compliant to ISO 14644-1 Class 5 standards for air cleanliness.

Visual examination of the incubated contact plates after air sampling of 1000 L of air indicates a lack of contamination of the environment surrounding each of the contact plates on the NIMBUS deck (Figures 2A – E), while contamination exists on the plate exposed to ambient room conditions (Figure 2F).

Conclusion

The CAP System, integrated with a standard, enclosed Microlab NIMBUS, provides a clean environment for high-throughput applications where sample/workflow integrity is paramount, without the need for the bulk or expense of biological safety cabinets.

Lit. No. AN-1803-29-01 v.1.0 — 04/2018

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