ASX Announcement
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ASX Announcements
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TECHNICAL UPDATE

- Independent laboratory testing of the new FRG+ Air Purifier proves the competitive advantages of boosting air purifiers with PuriflOH's Free Radical Generator (FRG).
- Adding a low power FRG module accelerates the removal of airborne microbes, ultimately resulting in the lowering of infection risk from respiratory or other airborne illnesses in occupied indoor spaces, creating a safer and more productive environment.
- Results from the Independent Laboratory, Co-Labs of Melbourne, prove that the FRG+ air purifier is:
    - 150% more effective at risk mitigation than a standard HEPA air purifier; and
    - safe to operate in occupied spaces.
- The new FRG format is cost effective with improved efficacy and is easily adaptable through retrofit to existing residential or commercial air purifiers.

Mr Vigneswaran Appia, the Senior Technical Manager of PuriflOH Australia, previously employed with Somnio Global in Detroit, has been overseeing the validation testing of the Air Conditioning Environmental Remediation Treatment (ACERT) system and the new air purifier upgrades. He said today that: "*A new generation of FRG+ Air Purifiers can decrease the risk of transmission of airborne diseases by impacting the speed of removal of ambient pathogenic germs. The successful transmission of any respiratory illness is dependent upon the ambient microbial load and the time required for an individual to interact with the germs. The FRG can reduce ambient microbial levels and can increase the rate of reduction of these germs compared to a stand-alone HEPA based air purifier. I am confident due to internal testing conducted by myself that we can continue to improve the device and achieve better results. Our internal testing has shown even faster reductions when compared with standard HEPA based filtration*."

This technical update provides perspective and explanation to the results of independent performance testing conducted on behalf of PuriflOH Limited ("PuriflOH", "PO3" or "Company") on its new generation of neutralizer free Air Purifiers. The work was conducted by Co-Labs of Brunswick, Victoria. Co-Labs is a Bio-safety Level 2 Laboratory that has recently installed a 75 cubic meter bio-aerosol testing chamber and is capable of testing various microbial organisms on surface or air using internationally accepted methods and equipment.

The Co-Labs report is attached to this ASX Release.

Significance of the new approach

Thus far, PuriflOH's approach on air purification has been to provide a single stage technology that can eliminate all air pollutants such as particles, odour, Volatile Organic Compounds (VOCs) and air-borne microbes. PuriflOH , initially through its research partner Somnio Global, and now operating through its own auspices, have continued to develop the Free Radical Generator (FRG)-based air purifiers. The new generation is now more powerful with a higher single-pass kill rate – the factor of destruction of microbes between outlet and inlet of an air purifier. Existing products require 3-6 stages of filtration to achieve the same effectiveness. This is a significant and unique improvement over current air purification solutions.

Since 2018, Somnio and PuriflOH have engaged with OEM companies in order for them to integrate versions of the Free Radical Generator (FRG) into existing designs to significantly enhance product performance. This OEM engagement has lead to useful market feedback and design changes. For example, the high concentration of radicals needed to significantly remove VOCs and other contaminants efficiently has necessitated neutralizer filters, which add to the overall size of the end product and cost as an air purifier.

A result of the Covid-19 pandemic has been that market priorities have shifted to highlight microbial decontamination above all else. This has led to the company focusing its efforts on the Air Conditioner Remediation and Environmental Treatment (ACERT) device as its first commercial product and re-evaluating the design of its air purifiers.

Recently, PuriflOH has been actively developing a low-power FRG addition to Air Purifiers, FRG+, that are simple to implement whilst providing significant benefits to the final product. The result of this approach is a **new generation** of Air Purifiers empowered by the FRG.

By modifying the design of the FRG and emphasizing hydroxyl production over ozone production, PuriflOH is able to add an FRG module to existing HEPA based air purifiers without making any significant changes to their size, volume flow rate or power consumption.

This newly designed FRG module has the following performance features:

- An improvement of **at least 150%** in the speed and efficacy of air purification over current market leading devices.
- Safe operation in **occupied spaces** without the need for **neutralizer filters**.
- Only **10W** of added power consumption.
- **Reducing noise levels** in existing air purifiers while improving efficacy.

Independent testing and results

PuriflOH has concluded its first independent test on this new FRG+ version of an Air Purifier, conducted at Co-Labs Melbourne, which houses the world's largest and Australia's first Bio-aerosol testing chamber. The test was conducted with *Staphylococcus epidermidis*, a vegetative bacterium that is analogous to MRSA, a common hospital-acquired infection frequently used in testing disinfectant efficacies.

PuriflOH's previous aerosol testing results were obtained from ARE Labs in the USA (ASX Release 17 April 2018 "*Aerosol Results*") and performed in a chamber sized 15 cubic metres. The airflow rate of the "Bluemist" device provided for more than 12 air changes per hour in this previously tested chamber. In the test conducted at Co-labs, the chamber volume was 5 times larger (75 m3) and the flow rate allowed for only 5 air changes every hour. Smaller air changes typically increase the time taken to achieve total disinfection. This air exchange rate of 5 per hour is similar to how ventilation and air purification systems are configured in the real world.

Despite the larger chamber size and the lower exchange rate, the new FRG+ device was able to **achieve 99% reduction in airborne bacteria in approximately 30 mins, at least 1.5 times faster than a standard HEPA based air purifier acting under the same conditions**. Moreover, ozone levels in the chamber were tested to remain well within acceptable values, allowing the device to be used in occupied spaces.

Real world impact of faster speed of reduction

Air purifiers are typically limited in their effectiveness by the air exchange rate – no matter how effective the filtration, air needs to pass through the filters to be cleaned. Further, air flow rate is limited by

considerations such as noise and comfort. In fact, most air purifiers are certified at their maximum flow rate, but seldom run at that flow rate.

With the FRG boosting performance and being independently effective at removing air-borne microbes, a new generation of FRG based air purifiers can prove to be highly effective infection control tools, acting to rapidly disinfect occupied spaces and significantly reduce transmission of air-borne diseases even when other measures are insufficient.

The combined action of FRG+ air purification in combination with the periodic air conditioning disinfection provided by the ACERT helps create a highly effective risk mitigation plan against air-borne diseases and can play an essential role in disease prevention in spaces such as hotels, restaurants, work spaces, mining camps, aged-care centres and medical and other clinics.

Commercial impact of new design

This new format of the FRG has been designed to incorporate a simple manufacturing and installation process that should increase chances of commercial success. The new design also creates new opportunities for PuriflOH and removes existing obstacles as:

- This design is highly **cost effective** and **significantly upgrades** the efficacy of air purifiers.
- The design is suitable for businesses, offering a highly effective solution with low operational expenses such as maintenance and power consumption.
- The FRG is now a **retrofit solution** to existing air purifiers.
- The design is **easily adaptable** to any existing residential or commercial air purifier, thereby making it a very easy OEM solution.

Next Steps

PuriflOH will continue to seek internal and external validation of FRG+ Air Purifiers on a roster of challenge organisms including viruses and fungal/bacterial spores, while driving the development towards improved performance in occupied spaces without the need for neutralizer filters.

The company will also seek to validate the ACERT system with various challenge organisms including those prescribed by the Australian Therapeutic Goods Administration and other regulatory bodies and is in the process of engaging Co-Labs along with other laboratories around the world to perform independent testing.

This release is authorised and approved by the Chairman of PuriflOH Limited.

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Determination of the Disinfection Efficacy of the PuriflOH Air Purifier Against Aerosolized *S. epidermidis*

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Background: This in-vitro study characterised the disinfection efficacy of the PuriflOH Air Purification device against aerosolised *Staphylococcus epidermidis*. The device's performance was also compared against a standard HEPA-based air purifier. A single and identical flow rate was used for both tests, and they only differed by adding PuriflOH's Free Radical Generator (FRG) to one of the tests. A natural decay test was also conducted to understand the natural decline of air-borne microbes under current conditions. Across the tests, samples were collected from the bio-aerosol chamber at various time points.

Methods: *Staphylococcus epidermidis* was aerosolised into a 75 m^3 bioaerosol testing chamber via a medical nebuliser. The test device was placed at the chamber's centre along with a mixing fan to simulate normal recirculation. AGI-30 impingers were used for sampling via eight evenly spaced points at six different time points. All impinger samples were serially diluted, plated and enumerated in duplicate to yield viable bioaerosol concentrations to determine the disinfection efficacy of viable bioaerosol. Three tests were conducted in total - (1) baseline natural decay, (2) Off the shelf standard HEPA air purifier, and (3) the same air purifier at the same flow rate with an FRG boost.

Results: Results showed that the viable microbial decline was significantly faster in the third test, where the air purifier was boosted with the Free Radical Generator when compared with the standard HEPA air purifier. Additionally, ozone sensors placed at various points in the chamber also maintained Ozone values below limits specified by the National Air Quality Standards of Australia.

Overview

This study was conducted to evaluate the effectiveness of the PuriflOH Air Purifier device developed by PuriflOH Limited. The PuriflOH air purification device is a single-stage cold plasma-based reactor intended to destroy a host of airborne contaminants, including disease-causing microbes as well as Volatile Organic Compounds. *Staphylococcus epidermidis* was chosen as the challenge organism for this study due to its resilience to aerosolisation and its general acceptance across various disinfectant studies and standards.

Testing was conducted in a chamber designed for bioaerosol challenge constructed at Co-Labs Melbourne. The efficacy of the device was assessed via sampling from 8 separate points in the chamber at a given time point and measuring bioaerosol concentration (CFU/ml). A baseline natural decay test was conducted as a control, and two effectiveness tests were then conducted on a standard HEPA-based air purifier and compared against a similar air purifier boosted with PuriflOH FRG technology. An initial concentration of at least 10^6 viable organisms per cubic metre of air was targeted in all tests.

PuriflOH Free Radical Generator

PuriflOH's Free Radical Generator is a device that uses Streamer Discharge to produce Free Radicals from ambient air that can destroy ambient airborne contaminants such as microbes and harmful Organic Compounds. In the version used for testing here, the

production of Hydroxyl radicals is boosted over the production of other oxygen-based radicals. This ensures the safe usability of the device in occupied spaces while significantly increasing the rate of destruction of ambient microbes. The modified FRG has been retrofitted into a standard Air Purifier to boost its performance against airborne pathogens.

Bioaerosol Testing System

The testing system used was built at Co-Labs and designed to generate and hold over 10^6 viable microbes per cubic metre of air in a sealed $75m^3$ chamber **Figure 1**. The size of the chamber built at Co-Labs makes it the largest Bio-aerosol testing chamber in the world, over 2-5 times as large as those present in other laboratories across the world. This size better represents commercial spaces such as offices with a 5 m x 6 m x 2.5 m room. The system features eight sampling points split across two different heights. Four sampling points are suspended 1m from the ceiling, while another 4 are suspended 1.5m from the ceiling. All sampling points are spaced evenly from the corners to ensure conformity in the sampling location. Sampling points are connected via a manifold leading to an impinger external to the chamber for operation. Air is drawn from a vacuum pump housed in the chamber at a flow rate of 12L/pm through an AGI-30 impinger and then back into the chamber. A floor fan is used to help keep the air within the chamber homogeneous at all times. The device undergoing efficiency testing sits within the chamber. After each test, bleach is aerosolised within the space to ensure sterility post-test and then purged through an exhaust system with HEPA filtered air. All devices within the chamber are remotely operated through the use of external switches.

Bioaerosol Generation

Staphylococcus epidermidis was aerosolised using a medical nebuliser for 10 minutes to generate over 10^6 viable cells per cubic meter of chamber air.

Bioaerosol Sampling

SeveralAGI-30 impingers were used to collect air samples across seven different time points at a flow rate of 12L/pm. Each impinger was filled with 30ml of sterile Phosphate Buffered Solution to collect bioaerosols. After sample collection at a given timepoint, samples were serially diluted and plated for enumeration.

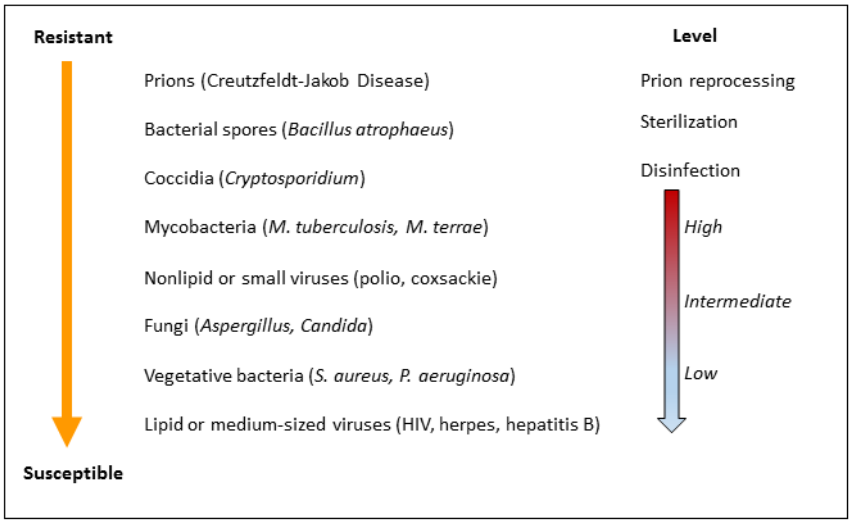

Figure 1: ***Decreasing order of resistance of microorganisms to disinfection and sterilisation and the level of disinfection or sterilisation (CDC, 2016)***

Organism Selection

Staphylococcus epidermidis, a gram-positive bacterium, was chosen as the organism of choice due to its genus being commonly associated with numerous contamination events in a variety of environments (Lee et al., 2019). The vegetative bacterium is also considered to be less susceptible to disinfectants than coronaviruses (CDC 2017).

The innoculum was cultured in LB Broth overnight from glycerol stocks to obtain a healthy culture for testing the following day.

Testing Methods

Before testing, an overnight culture of *Staphylococcus epidermidis* was prepared to ensure a consistent cell count and viable bioaerosol nebulisation. All sample vials, solutions, and impingers are sterilised. Prior to nebulisation, the mixing fan is turned on to allow for continuous homogenisation during nebulisation. After nebulising a dilution of overnight culture, the first sample is drawn. The next four samples are drawn at 15-minute intervals, with the sixth sample drawn after an additional hour. Each sample is drawn for 10 minutes.

After the first sample was collected to establish the starting bioaerosol concentration, the standard HEPA purifier device and the same device with integrated FRG were turned on for the remainder of their respective tests, with samples taken at intermittent time frames. After test completion, the respective device was deactivated, and sterilisation began.

Additionally, ozone sensors placed in the chamber during the test found the readings to be within the limits prescribed by the National Air Quality Standards.

Plating and Enumeration

Plating and Enumeration were conducted by plating 1ml of the sample (either diluted or undiluted) on Petrifilm™ Aerobic Count Plates Supplied from Thermofisher Scientific. After plating, samples were incubated for 18-24 hrs as per the supplier's instructions.

Decontamination

Post-testing, a solution containing 10% bleach was aerosolised for 30 seconds and allowed to permeate throughout the chamber for 20 minutes.

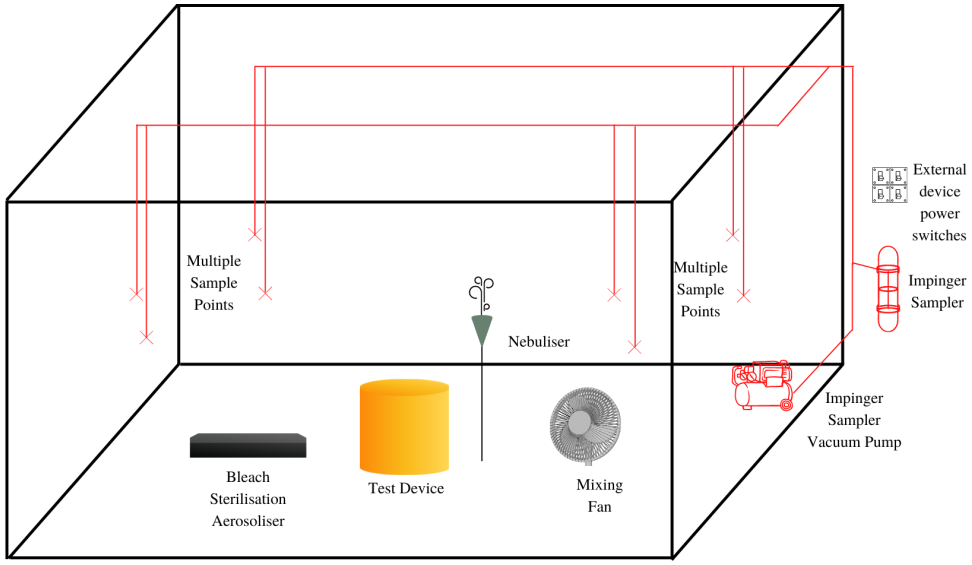

Figure 2: Bioaerosol Testing Chamber Configuration for PuriflOH Air Purifier

Results

Table 1 - results - actual microbial counts

	Remaining viable organisms		
Duration(mins)	Natural Decay	HEPA Air Purifier	FRG boosted Air Purifier
0	5052000	2000000	1962000
15	Not measured	435000	156000
30	3415000	126000	27000
60	2522000	9250	1500
120	1138636	375	0

Table 3 - Logarithmic representation

	Remaining viable organisms		
Duration (mins)	Natural Decay	HEPA Air Purifier	FRG boosted Air Purifier
0	0	0	0
15	NA	-0.6625407	-1.099574
30	-0.170072634	-1.2006595	-1.861335
60	-0.30171826	-2.3348883	-3.116608
120	-0.647078431	-3.7269987	-6

Table 2 - Percentage reduction of viable microbes

	Remaining viable organisms		
Duration (mins)	Natural Decay	HEPA Air Purifier	FRG boosted Air Purifier
0	100.00%	100.0000%	100.0000%
15	NA	21.7500%	7.9511%
30	67.60%	6.3000%	1.3761%
60	49.92%	0.4625%	0.0765%
120	22.54%	0.0188%	0.0000%

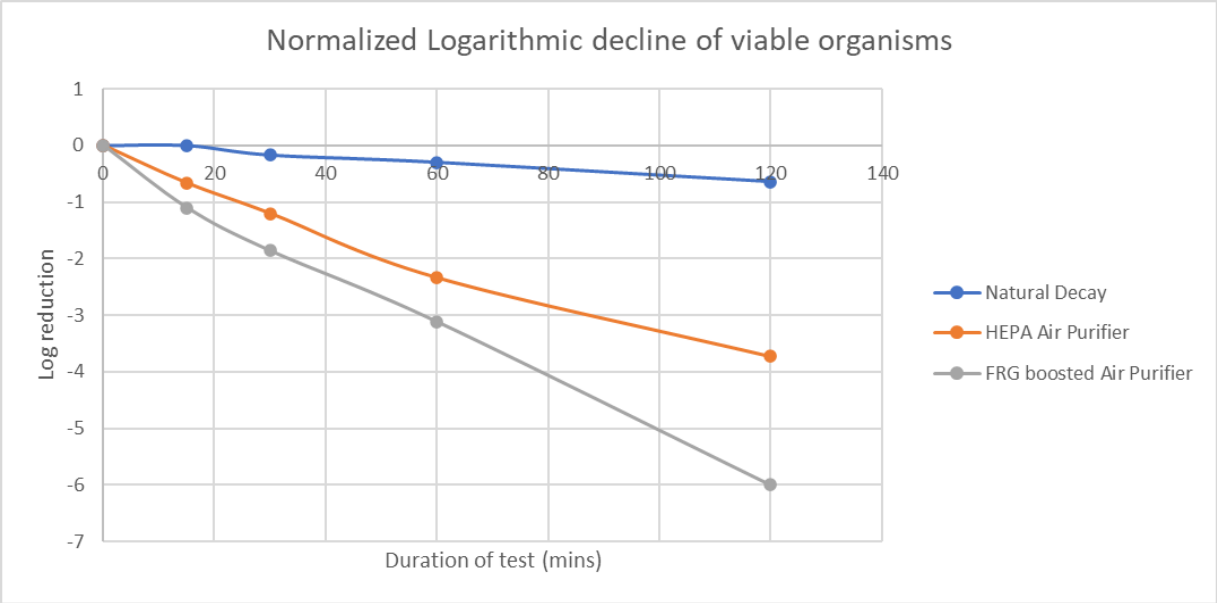

Figure 2 - Decline of viable organisms

Summary of findings

According to the results, the PuriflOH boosted air purifier was significantly faster at disinfecting airborne microbes than the standard version of the Air Purifier. It was also found to be safe to use in occupied spaces for the time period tested. Over 90% reduction was observed in 15 minutes of operation. Across the different time points, the FRG boost provided over 1.5 times faster reduction in viable microbes when compared with the standard HEPA air purifier. This could offer a significant advantage in real-world settings by decreasing the risk of transmission of air-borne diseases even more than a traditional air purifier.

References

CDC (2017) *Guideline for Disinfection and Sterilization in Healthcare Facilities (2008)*, Centers for Disease Control and Prevention, viewed 20 August 2022, <https://www.cdc.gov/infectioncontrol/guidelines/disinfection/tables/figure1.html>

Lee, Jung Hoon et al. (2019) "Assessment of air purifier on efficient removal of airborne bacteria, Staphylococcus epidermidis, using the single-chamber method." *Environmental monitoring and assessment* vol. 191,12 720. 6 Nov. 2019, doi:10.1007/s10661-019-7876-3

Co-Labs Bioaerosol Testing Facility

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