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ASX Announcements
4th Floor
20 Bridge Street
Sydney, NSW, 2000

Dear Sir,

Market Update

- **FRG "proven to be effective in a real world environment"**
- **Second stage MTB trials to continue at the Bhaskar Medical Centre**
- **Commercial engagement with industry continues to advance**

1. Tuberculosis Trials

PuriflOH Limited ("PO3" or "Company") is pleased to provide the first stage report recently received from the Government Chest Hospital in Hyderabad, India. Dr Anil reports on the protocols and outcomes associated with the testing of PO3's Free Radical Generator (FRG) air purification system, a series of trials targeting the elimination of air borne *Mycobacterium tuberculosis* (MTB) bacteria.

The Company has previously reported on these trials to the ASX (30 April 2019).

The site of the first stage of the trial was a dedicated ward for respiratory diseases, that was both large in scale and open to outside air, where the ward treated patients suffering from tuberculosis.

As previously announced, the environment in which the initial trials were conducted was complex and it ultimately proved difficult to isolate baseline MTB levels. However, the overall results of the first stage tests have reinforced the Company's confidence the FRG will be effective in reducing airborne bacteria levels and the second stage of testing has now commenced as outlined below.

The report provides a detailed analysis of the tests which include, by way of overview:

"The Bluemist™ air purification system has successfully demonstrated the ability to destroy bio-aerosols in a real world setting."

The high levels of airborne contamination within the ward overpowered the presence of MTB making it difficult to establish meaningful baseline MTB levels. This level of background contamination did however provide an opportunity to assess the ability of the system to perform in a real world environment. The report provides positive validation of this performance as there was a significant reduction in bioaerosol levels during the treatment period including reductions in pathogenic species of mold such as *Aspergillus fumigatus and Aspergillus flavus* .

This is the first time that the FRG system has demonstrated it's effectiveness in eliminating pathogenic organisms in a hospital environment, an important outcome achieved in spite of the challenges involved with the high ventilation levels in the ward.

Issues identified during the testing phase which aid planning of the next trials were as follows:

A. The patients within the ward were in an advanced stage of treatment and consequently not releasing significant levels of MTB into the ward environment. High levels of other airborne contaminants, particularly bacteria and mold, dominated the sample cultures with the presence of MTB being too low for meaningful baseline results to be established.

B. The ward had a great deal of air flow variability due to a high rate of ventilation as:
   - it was open to the outside to provide ventilation; and
   - had a large number of health care workers moving in and out of the ward.

C. The tuburculosis ward itself was too large (93m2) for the unit that has been configured for a target room size of 30 m2.

The resultant continuous air exchange in B above, and the ward size in C above, was far in excess of the air treatment volume of the FRG unit designed for the trials. This reduced the impact of the system on the internal environment.

Various changes to the sampling methodology were trialed - ultimately the environment was deemed unsuitable to accurately prove the desired outcomes.

The trials have now moved to the Bhaskar Medical College and Hospital. The information from the initial trials clearly determines the need for a controlled trial environment which allows for stricter control of the test zone. This has meant extensive discussions with the Hospital Management to ensure that patient care is paramount, whilst providing conditions suitable for a valid test program.

The current status is that:

- Protocols have been agreed with the Hospital;
- The Hospital Ethics Committee has approved the trial;
- A 11.2 m^2 room with controlled ventilation has been readied for the trials to commence; and
- Sampling for the purposes of baseline measurement will commence a few weeks.

Results from these trials are now anticipated during the September quarter.

2. OEM Commercial Update

Ongoing due diligence ("DD") testing trials with Original Equipment Manufacturers ("OEM") have been primarily focused on the air purification system. They are now also extending to water treatment applications – separate to recreational water applications - at the request of the OEMs. Initial engagement, establishment of relationships and the exhaustive DD processes are all positive developments and resulted in important feedback from our market place about how best to integrate the PO3 technologies into existing OEM products.

The Company is pleased with ongoing development and relationship work and remains confident of final commercial outcomes with a number of groups with global recognition.

End

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Reduction in bio-aerosol levels through Bluemist™ intervention in a respiratory health isolation ward

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Abstract

The Bluemist™ air purification system (Somnio Global LLC, USA) has demonstrated great efficacy in destroying bacterial, viral and fungal bio-aerosols in controlled and targeted efficacy trials [1]. Kill performance over 4 log was achieved in a single-pass for all bio safety level 1 organisms tested and a 5-log net reduction was achieved in as little as 60-90 minutes. The present study was designed to examine the impact of the Bluemist™ system in a real world application. An isolation ward for respiratory diseases at the Government Chest Hospital in Hyderabad, India was chosen as the site for this study. Baseline sampling of the air in the ward was done over a 3 week period and revealed the presence of high levels of airborne mold and some bacteria. The Bluemist™ system was subsequently installed and run 24/7 over a sampling period of 3 weeks. The samples collected during the intervention period showed a statistically significant reduction in overall bio-aerosol levels in the isolation ward compared to the baseline.

1. Background

The working of the Bluemist™ air purification system is based on a unique single-stage cold plasma channel that destroys microbes, VOCs and other airborne contaminants. Operating in closed loop mode the contaminated air from an occupied room is pulled into the device and passes through the cold plasma wherein the chemicals and microbes are destroyed. Additionally, submicron particles are charged within the plasma chamber enabling the filter to trap them with great efficiency. The effectiveness of the Bluemist™ system has been previously demonstrated in controlled efficacy trials [1].

The aim of this study is to demonstrate the effectiveness of the Bluemist™ system in a real world setting. An isolation ward in the Government Chest Hospital in Hyderabad, India was chosen as the location for this study. The isolation ward typically houses 8-10 patients with respiratory disease at any given time. The location was chosen for its severity of microbial activity, including the possibility for presence of *Mycobacterium*.

The real world setting brings with it some additional challenges that are not seen in controlled testing. First, the micro-organisms in a hospital setting are commonly of the pathogenic type. Secondly, the study is conducted without any intrusion on or obstruction to patient care. In this particular example, this meant the presence of a high rate of ventilation through the isolation ward and movement of health care workers in and out of the treatment space. This movement of air and personnel has a significant impact on the levels of micro-organisms in the isolation ward and hence on distinguishing the effectiveness of the air purifier.

2. Study Methodology

2.1 Isolation Ward Setup

The isolation ward was installed with the Bluemist™ system in a central location to allow for homogenous treatment of the room air. The sampling points were distributed at 4 different locations in the ward to get the best approximation of the distribution of the bio-aerosol load within the ward.

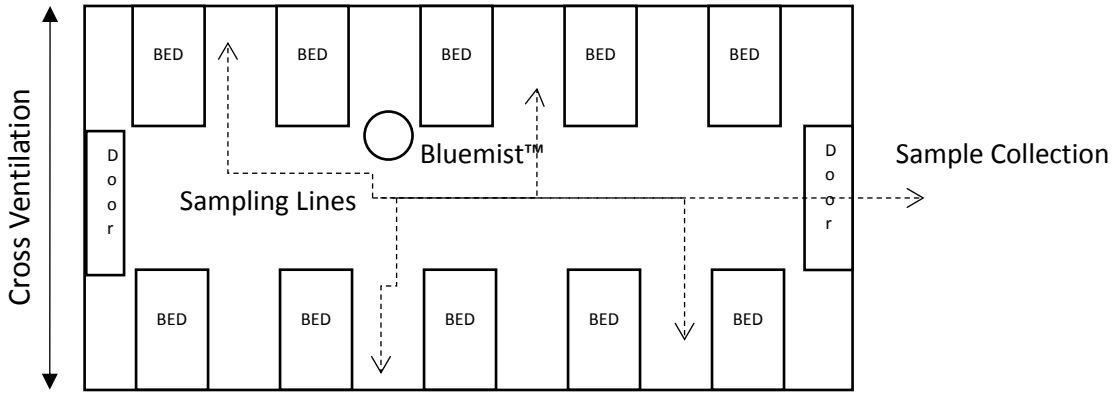

FIG 1. Isolation Ward Setup

The sampling lines were setup away from foot traffic. No changes were made to the ventilation within the isolation ward. The ward had mesh screened windows running along its entire length that were kept at least partly open throughout the study period, as per the patients' convenience. The door at the far side of the room, away from the sample collection point, was kept closed for the most part, aside from when health care workers moved through it. The sample collection point was just outside the ward, past a moveable solid screen door.

Some pertinent information with regards to the ward and its use as a treatment space are listed in table 1 below.

Isolation Ward Area	93 m^2
Isolation Ward Volume	425 m^3
Max. Patient Capacity	10
Estimated Ventilation Rate [2]	5-7 ACH

Table 1. Isolation Ward Information

A single Bluemist™ system was used for the study. Typically, a system of the size used here is intended for room sizes up to 30 m^2 in area. With the isolation ward area so large and the abundance of natural ventilation, the test setup was not ideal to test the efficacy of the device. Still, an attempt was made to test the effectiveness of the system under these conditions.

Table 2 shows some important parameters of the Bluemist™ system.

System Flow Rate	212 m^3/hr
Air Exchange Rate	0.5 per hr

Table 2. Bluemist™ System Information

2.2 Sampling Station Setup

The bio-aerosol sampling was performed using a single-stage viable cascade impactor (SKC Inc, USA). Impactors work by directly impacting air onto media-filled petri dishes that can then be incubated and cultured. This direct transfer approach for sampling offers higher sensitivity (typically 1-1.5 log) compared to other air sampling techniques such as liquid impingers.

Owing to the high rate of air exchange with the outdoor environment, the levels of the micro-organisms in the ward, as a result of patient emission, were expected to be low. The use of impactors for sampling hence, increases the chance of gathering representative ward bio-aerosol levels.

The sampling setup is setup as described below in figure 2.

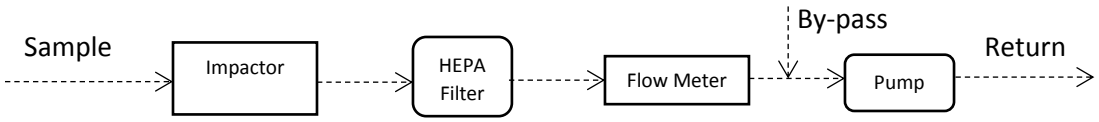

FIG 2. Sampling Setup

The air inside the isolation ward was drawn through the sampling lines into the impactor using a suction pump. The flow rate for impactor sampling is set at 28.7 lpm as per manufacturer specifications for the instruments using a flow meter connected just before the pump. A HEPA filer is installed between the impactor and the flow meter to remove any bio-aerosols not trapped in the collection media (typical efficiency of impactor is over 99%). The sampled air is then returned from the pump back into the isolation ward.

Pre-poured media plates of Middlebrook 7H10 type (100 mm) were used for the impactor sampling. The sampling time was set to 5 mins.

2.3 Study Procedure

The isolation ward at the Government Chest Hospital houses 8 to 10 patients at any given time. The patient turnover for this particular ward is also highly regular – about one week. The only variable is the stage of treatment, with some patients at the beginning of their treatment cycle and some towards the end of treatment. However, given a long enough sampling timeline, this factor is normalized. Based on this, the study was designed to be performed in two stages.

First, baseline sampling was performed, without Bluemist™ intervention, for 7 sampling days over a 3 week period. Each day, 3 samples were collected at different time points – morning, noon and afternoon.

Second, the Bluemist™ system was installed and run. Samples were collected for 8 sampling days over a 3 week period. As with the baseline phase of testing, 3 samples were collected each day, one each in the morning, noon and afternoon.

There were no changes made to the ward or the sampling lines between the two phases of testing. A total of 20 samples were collected during the baseline period and 23 during the intervention period.

2.4 Sample Processing

Impactor samples were collected on pre-poured Middlebrook 7H10 plates. These plates were stored under refrigeration till the sampling day and then transferred to a cooler with an ice-pack for transport between the testing facility and the culturing lab.

The plates were handled aseptically, as per the manufacturer's instructions and samples were numbered and packed individually in re-sealable bags immediately following sampling.

The impactors were disinfected using 90% isopropyl alcohol between each sample, as per the manufacturers' recommendation. At the end of each sampling day, the impactors were sterilized in a steam autoclave.

All samples were processed at Dr. Iravatham's Laboratory (Hyderabad, India) a NABL (National Accreditation Board for Testing and Calibration Laboratories) accredited laboratory that follows ISO/IEC 17025 Standard for quality assurance.

The plates were incubated at a temperature of 37 degrees Celsius in 5-10% CO_2 environment. The plates were enumerated after a 72 hr period of incubation. The enumerated plates were incubated further to check for the slow dividing *Mycobacterium*.

3. Results and Discussion

The colony counts enumerated during the baseline phase of the study revealed high levels of airborne mold in the isolation ward. Particularly, colonies of *Aspergillus niger*, *Aspergillus flavus* and *Aspergillus fumigatus* were observed. In addition, some instances of colonies from the *Mucor* genus were also seen.

Some samples also showed growth for both gram –ve and gram +ve cocci. However, after 6 weeks of incubation, no *Mycobacterium* growth was observed. As mentioned previously, all the patients here were on medication during their stay in the isolation ward and this may have suppressed the viability of *Mycobacterium* in bio-aerosol form – a known phenomenon.

The results discussed here are presented as total colony forming units per unit volume (TCFUs/m^3) and include all micro-organisms counted from those listed above.

During the intervention phase of the study, mold colonies of A. *niger*, A. *flavus*, A. *fumigatus* and *Mucor* were observed again, albeit at distinctly lower concentrations.

No bacterial growth was observed on samples collected during this phase.

Period	No. of Samples	All Samples Mean (CFU/m^3)	No. of Samples Below Mean	Percentage below Mean (%)
Baseline	20	43.5	9	45
Intervention	23		17	74

Table 3. Summary of Results Presented on Sample Mean

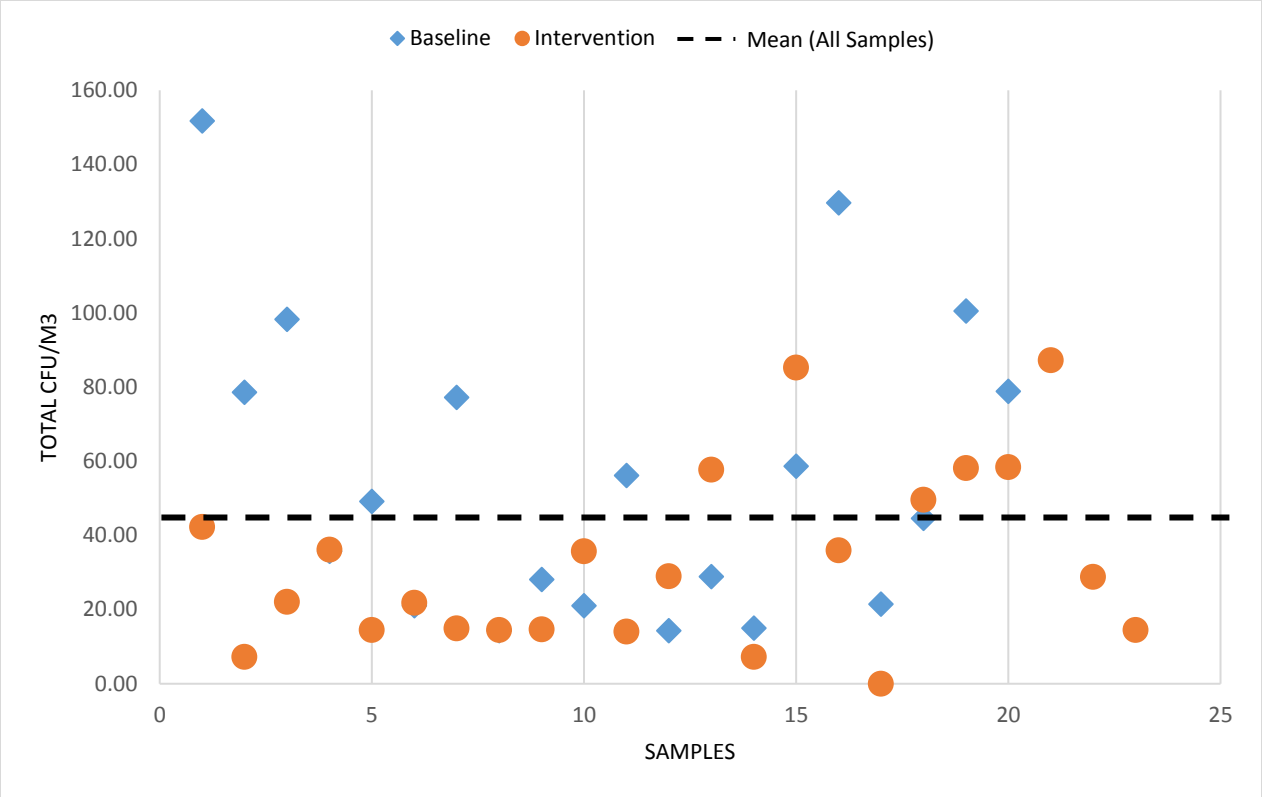

FIG 3. Total CFUs/m^3 from baseline and intervention samples

The data from Table 3 and Figure 3 show the difference in total bio-aerosol concentrations between the baseline period and the intervention period. What is immediately clear from the data is that the average sample collected during the intervention period falls much below the average concentrations noticed during both periods of study. In fact, 74% of all samples collected during the intervention period are lower than the study mean. Furthermore, the mean for the baseline period registers, higher, at 56.1 CFUs/m^3. The percentage of samples during the intervention period that fall below the baseline mean is 78%.

Fig 4 shows the distribution of the samples collected during both periods of the study. The median colony count during the intervention period is nearly 40% lower that of the median for the baseline period of study – 29 to 46 TCFUs/m^3. The data also shows that the two sample sets are significantly different with a t-stat of 2.28, with t-critical and p-critical values (two-tailed) of 2.04 and 0.029 respectively, at a significance level of 5% ($\alpha = 0.05$).

The above demonstrates with statistical significance the difference in bio-aerosol levels between the baseline and intervention periods of the study. Given that the rate of air exchange in the isolation ward due to natural cross-ventilation is significantly higher than the air exchange in the ward through Bluemist™ action alone (by a factor >10), these results are demonstrative of Bluemist™'s suitability for real world applications.

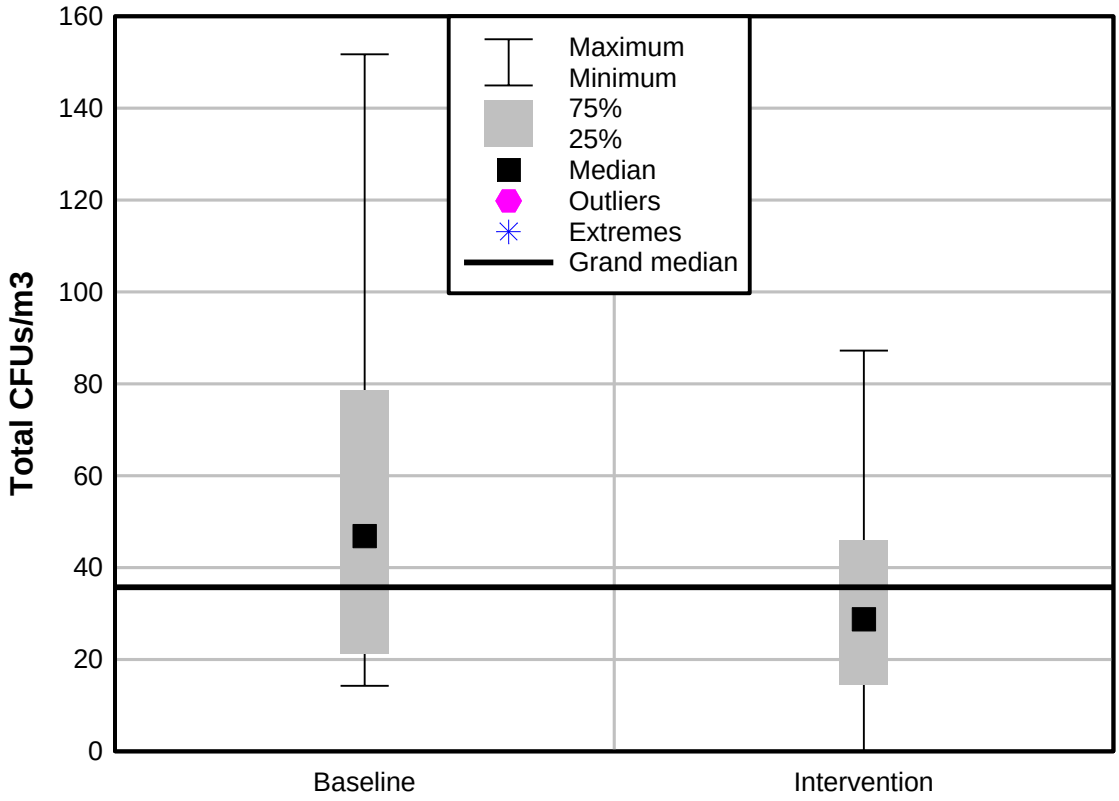

FIG 4. Box and Whiskers Plot of Sample Population for Baseline and Intervention Periods

4. Conclusion

The Bluemist™ air purification system has successfully demonstrated the ability to destroy bio-aerosols in a real world setting. The treatment space included the presence of pathogenic species of mold such as *Aspergillus fumigatus* and *Aspergillus flavus* and also other species of mold and bacteria. Despite the effect of natural ventilation processes and the large size of the isolation ward, the Bluemist™ system was able to significantly lower the overall bio-aerosol load in the treatment space. Based on this evidence, it is reasonable to conclude that the effectiveness of the Bluemist™ system will be greater still when used in a more appropriately sized treatment space.

5. References

[1] Efficacy of Somnio Global's BluemistTM System against Various Bioaerosols and Particulates. Jamie Balarashti, Zach Conley - Aerosol Research and Engineering Laboratories Inc. Olathe KS

[2] 2009 ASHRAE® Handbook, American Society for Heating, Refrigeration, and Air-Conditioning Engineers, Inc., ISBN 978-1-933742-54-0