



CASE STUDY: [REDACTED]

February 21, 2017

AirPHX Sports
1100 North Glebe Road, Suite 600
Arlington, VA 22201

Objectives

The treatment space is a large 306,000 cubic foot weight and equipment room at the [REDACTED], Virginia. The initial objective of the installation was to address odors in the treatment space. A secondary but related objective of the installation was to reduce the microbial “bio-burden” in the treatment space and provide a cleaner, more healthy space for members and staff.

Equipment

Four ceiling mounted PA2400 units were installed in late October 2016. Units were programmed to activate one hour after the gym closed and to shut down one hour prior to opening – for many nights, this schedule meant the units operated for only four hours from midnight to 4 am. To increase the effectiveness of the units, they can be run at longer intervals including 24/7 to achieve maximum effect.

Pre-treatment Testing (10/10/2016)

airPHX technicians took surface swabs at 20 locations in the treatment space. The swabs were subsequently “cultured” and analyzed by independent labs “Practical Air Testing Solutions” in Turlock, California. Results of this testing were provided in a report dated November 20, 2016 (following). The average level of contamination was 1,584 CFUs/cm² (“colony forming units per square centimeter”) – a very high level of contamination.

In-treatment Testing (02/07/2017)

airPHX technicians conducted in-treatment testing to evaluate effectiveness of the technology. In a report dated February 17, 2017, Practical Air Testing Solutions concluded:

- Surface swabs. 20 swabs were taken at approximately the same locations as the pre-treatment testing. The in-treatment testing indicated an average reduction in CFU/cm² of 96.8 percent in the swabbed area (including a reduction in the “front desk” of from 5,103 to 81 CFU/cm² – a total reduction of 98.4 percent).

Overall: 96.8% reduction in surface contamination

- Air Samples. 18 air samples were taken inside the gym and ancillary rooms. The average CFU/m³ (“colony forming units per cubic meter”) in the indoor air samples was 47 CFU/m³ compared to exterior contamination of 2,917 CFU/m³. Pre-treatment air samples were not taken, however, the ambient air in the facility is cleaner at all testing spots than outside air.

Overall: Indoor air 98.4% fewer contaminants than outside air

- Coupon testing. Perhaps the most important testing protocol, “inoculated” plastic and metal coupons were placed throughout the treatment space and were collected at 1, 2, 3 and 4 hour intervals. The coupons had a harmless surrogate organism to common gym pathogens (mRSA, norovirus, rhinovirus and other). The treatment resulted in almost total “kill” of these gym based pathogens in four hours.

<u>Hour</u>	<u>Log Reduction</u>	<u>Organism Reduction</u>
1	4.6	Over 10K reduced to 1
2	5.2	Over 100K reduced to 1
3	5.9	Approx 1M reduced to 1
4	6.5	Almost total “kill”

Overall: The coupon testing indicates that airPHX Sports equipment is essentially eliminating gym-based pathogens in the entire treatment space each night, including equipment, counters, and floors

Future Results

Continued use of airPHX Sports technology will have a cumulative sanitizing effect on the treatment space – surfaces will continue to get cleaner, the bioburden throughout the space will continue to decline, and odors and health benefits to members and staff will continue to increase.

Over time, the sanitizing effect of the equipment will have a positive impact on the entire [REDACTED] facility as treated air is circulated throughout the facility by HVAC ductwork.

Installed Unit



Testing Reports:
Practical Air Testing Solutions

Practical Air Testing Solutions



February 17, 2017

PhoenixAire
1100 North Glebe Road, Suite 600
Arlington VA 22201

Sanitation Capabilities of the airPHX System in a Typical Gym Environment – [REDACTED]

Conclusion

Contact swab, air samples as well as 5mm inoculated stainless steel and plastic coupons showed significant reduction after being exposed to airPHX ROS treatment.

- In-treatment contact (surface) swab results showed a 96.8% reduction from the pre-treatment findings, also given in the attached report.
- In-treatment air samples revealed a very low bioburden of 47 cfu/m³ which is < 100 cfu/m³ and is considered **clean and acceptable** per the Target Air Quality scale, see attached report.
- Inoculated stainless steel and plastic coupons were placed in five (5) different locations in the large workout room. An estimated 0.02 ROS concentration showed a 4.69-log reduction after one (1) hour and 5.24, 5.93 and 6.45-log reduction after two (2), three (3) and four (4) hours, respectively. These results were for location "A" which was the most difficult location for the ROS to reach.

Purpose

The subject trial was designed to demonstrate the benefits of using airPHX equipment to clean the air inside a large workout area by generating reactive oxygen species (ROS) from ambient air. The air was circulated in the room environment and exposed on *stainless steel (S)* and *plastic (P)* coupons, commonly used in gyms and exercise facilities. These coupons were inoculated with non-pathogenic *Enterococcus faecium* which is slightly more resistant to ROS and is an acceptable surrogate for *E coli O157:H7*, *Salmonella spp* as well as *Influenza A*, *mRSA*, *Norovirus*, and *Rhinovirus*.

As previously mentioned, this trial used a harmless, non-pathogenic surrogate for contaminants; this surrogate poses no food safety or health risk. This organism was selected since it acts similarly as pathogens of concern found in sports locker room, gyms and hospital facilities. The surrogate selected has the same growth characteristics and tolerances as the pathogens except, again, not being pathogenic.

Also, for complete safety purposes the selected non-pathogenic surrogate was dried for one hour at 22 C on the above mentioned coupons to prevent any of the tested materials from contacting room surfaces. This process took place before being brought into the test facility. As a further precaution each sample was isolated from the room surface by using a plastic dish 50mm by 15mm under each coupon, see pictures. Second, all rooms continued to be sanitized using the AirPHX system for several days following testing.

AirPHX ROS treatment has been demonstrated to be highly effective in reducing mold, pathogenic bacteria and viruses in a variety of indoor environments. In a growing number of commercial applications, these benefits have enabled the food industry, restaurants, hotels, office buildings and health clinics to reduce contamination in the air and surfaces thereby reducing operational risk and costs.

Bacterial culture

Enterococcus faecium, ATCC 12755; is a non-pathogenic surrogate, which were acquired from American Type Culture Collection (ATCC, Manassas, VA., USA) and maintained at 8°C on slants of tryptic soy agar (TSA, Hardy Diagnostics, Santa Maria, CA., USA).

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Cultured in tryptic soy broth (TSB, Hardy Diagnostics) at 26°C. Every 24h, the cultures were transferred to TSB by loop inoculation. Cells (approx. 1×10^8 to 1×10^9 CFU/ml) from a 24h static culture incubated at 26°C were used to inoculate a number, ~100 each, 5mm round stainless steel and plastic coupons. The inoculum suspension was enumerated by surface plating in duplicate samples on TSA and Bile Esculin Azide Agar (BEAA) after serial dilution in 0.1% peptone solution. Plates were incubated for 24h at 26°C.

Inoculation of the media surface

A 100µl droplet from the initial surrogate suspension was used to inoculate the external surface of the round coupons of stainless steel (S) and plastic (P) with the final inoculum level of approximately 8-log CFU/5mm. The inoculated samples were dried for 2h at 22°C prior to ROS treatment. The 2h drying will allow the inoculated cells to attach to the surface host.

airPHX Reactive Oxygen Species (ROS) treatment

ROS treatment was carried out using four (4) airPHX, PA 2400 suspended from the ceiling in opposing corners of large workout room and activated.

Approximately 100, 5mm stainless steel and plastic inoculated coupons (~50 each) were randomly placed (different locations) and was treated with airPHX system, see pictures attached. Samples were randomly harvested from each location after exposure of one (1) hour and then in one (1) hour increments for four (4) hours from the large workout room at ordinary maintained temperatures. Post treatment, all samples were subjected to enumeration by surface plating. The log reduction of the inoculum was evaluated with and without the consideration of resuscitation of injured cells after ROS treatment.

Three (3) different controls were prepared for ROS treatment. For a **positive control**, three (3) 5mm round coupons were inoculated with the surrogate's cells and not exposed to the ROS treatment. Three (3) **negative controls**, in which 5mm coupons were inoculated with 100 µl droplet of sterile water and dried for 2h. One (1) **negative control** was treated with ROS and the other will not be subjected to the ROS treatment. Each treatment sample was prepared in triplicate.

Recovery of surrogates from the various surface samples

After ROS treatment, each 5mm coupons were cleaned with 5ml sterile water and swabbed with a cotton swab to facilitate the release of the organism. The head of the swab was immediately removed and transferred into a 400ml stomacher bag (Fisher Scientific Inc., PA., USA) combined with 50ml sterile 0.1% peptone solution, and then blended with a AES Easy Mix Stomacher (AES Laboratories, Princeton, NJ., USA) for two (2) min at normal speed. Wash fluid will be serially diluted and surface plating for enumeration. A centrifugation method was used to recover low populations of ROS injured bacteria. The centrifugation method (Mossel and others 1991) was modified and used to concentrate the bacterial population in the wash fluid so that less than 250CFU/ml of the non-pathogenic samples can be enumerated by the surface plating.

Results

- a. ROS treatment of surface inoculation coupons, Table #1.
 - i. Large workout area with an estimated 0.02 ROS concentration showed:
 1. 4.60-log reduction after one (1) hour
 2. 5.24-log reduction after two (2) hours
 3. 5.93-log full reduction after three (3) hours
 4. 6.45-log or a full reduction after four (4) hours
 - ii. Traveling controls showed
 1. 8.53 to 8.55-log was recovered for all treatment times, no reduction was seen.

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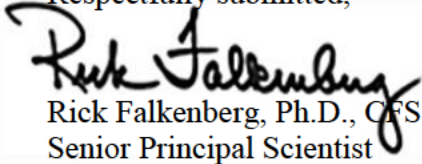


- b. Air Sampling, full report attached.
 - i. Various locations
 - 1. No pre-treatment air samples were taken.
 - 2. 02/07/17 in-treatment samples revealed a 47 cfu/m³ which is < 100 cfu/m³ and is considered **clean and acceptable** per the Target Air Quality scale.
 - c. Contact swabs, report attached.
 - i. Large workout area
 - 1. 10/20/16 pre-treatment showed an initial bioburden of 2,436 cfu/cm².
 - 2. 02/07/17 in-treatment samples showed a 98.2% reduction to 51 cfu/m³. Continued treatment will further reduce the bioburden ultimately to the < 5 cfu/cm² which is considered **clean and acceptable**.

AirPHX would like to express their appreciation for the [REDACTED] facility for supplying the large workout room for this evaluation.

Please contact me if there are questions or if further information is needed.

Respectfully submitted,


Rick Falkenberg, Ph.D., CFS
Senior Principal Scientist

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Table #1
In-Treatment - Inoculated Stainless Steel and Plastic 5mm Coupons

[REDACTED] - In-treatment						
Time	Coupon	Location	Reduction cfu/5mm	Reduction Log (cfu/5mm)	Recovered cfu/5mm	Recovered Log cfu/5mm
Hours	Type	A - E	Treatment*		Traveling Control*	
1	Stainless	A	45,500	4.66 ± 0.02		
	Plastic		39,800	4.60 ± 0.02		
	Stainless	B	58,200	4.76 ± 0.01		
	Plastic		53,300	4.73 ± 0.02		
	Stainless	C	49,900	4.70 ± 0.02	351,000,000	8.55 ± 0.01
	Plastic		48,000	4.68 ± 0.01	349,000,000	8.54 ± 0.01
	Stainless	D	47,000	4.67 ± 0.01		
	Plastic		47,100	4.67 ± 0.01		
	Stainless	E	52,000	4.72 ± 0.02		
	Plastic		51,500	4.71 ± 0.01		
2	Stainless	A	175,000	5.24 ± 0.01		
	Plastic		172,500	5.24 ± 0.01		
	Stainless	B	181,000	5.26 ± 0.02		
	Plastic		179,900	5.26 ± 0.01		
	Stainless	C	182,500	5.26 ± 0.01	348,000,000	8.54 ± 0.02
	Plastic		179,000	5.25 ± 0.02	347,000,000	8.54 ± 0.02
	Stainless	D	185,000	5.27 ± 0.01		
	Plastic		180,900	5.26 ± 0.01		
	Stainless	E	185,300	5.27 ± 0.02		
	Plastic		184,500	5.27 ± 0.02		

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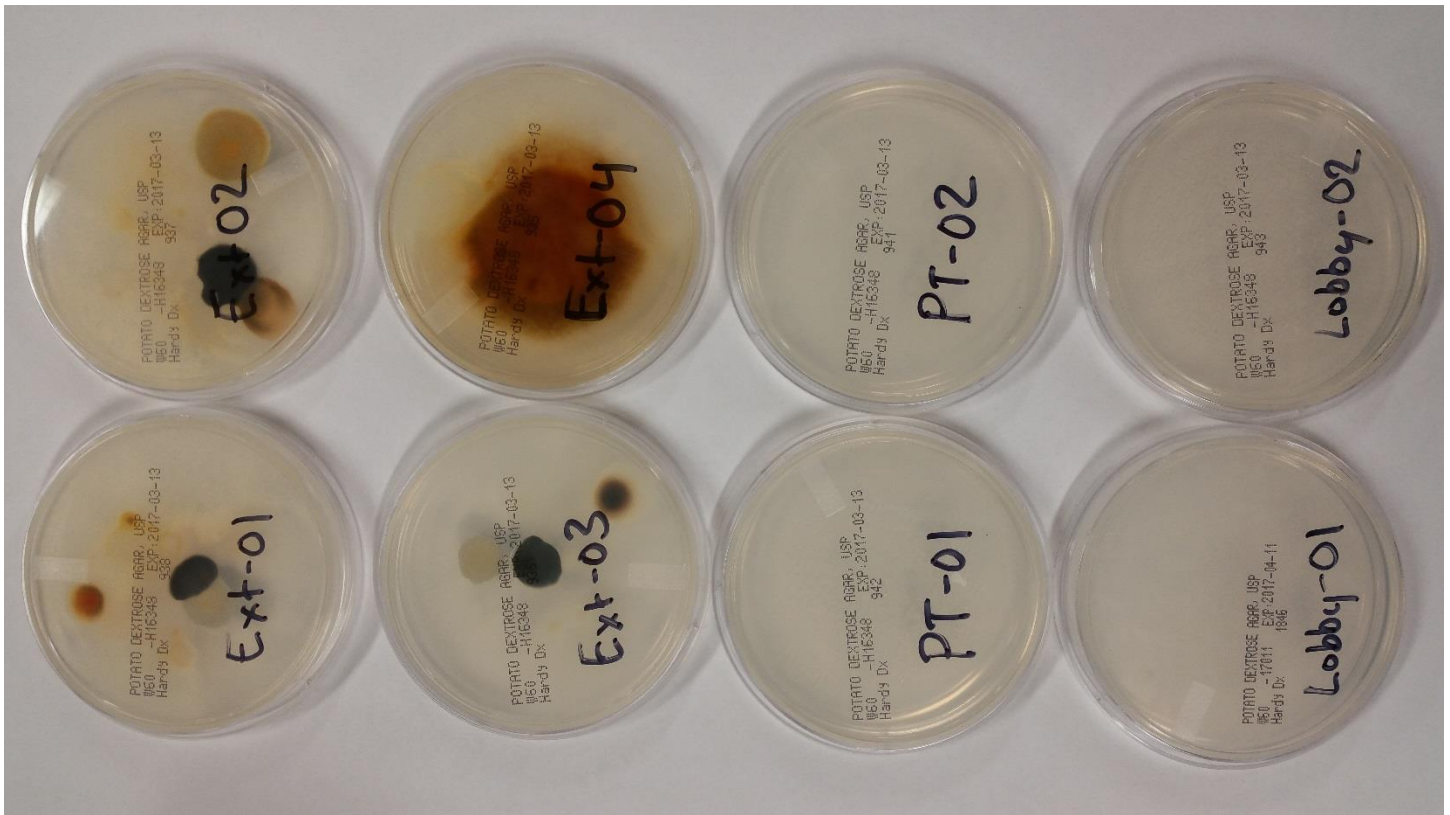


Table #1
In-Treatment - Inoculated Stainless Steel and Plastic 5mm Coupons, continued

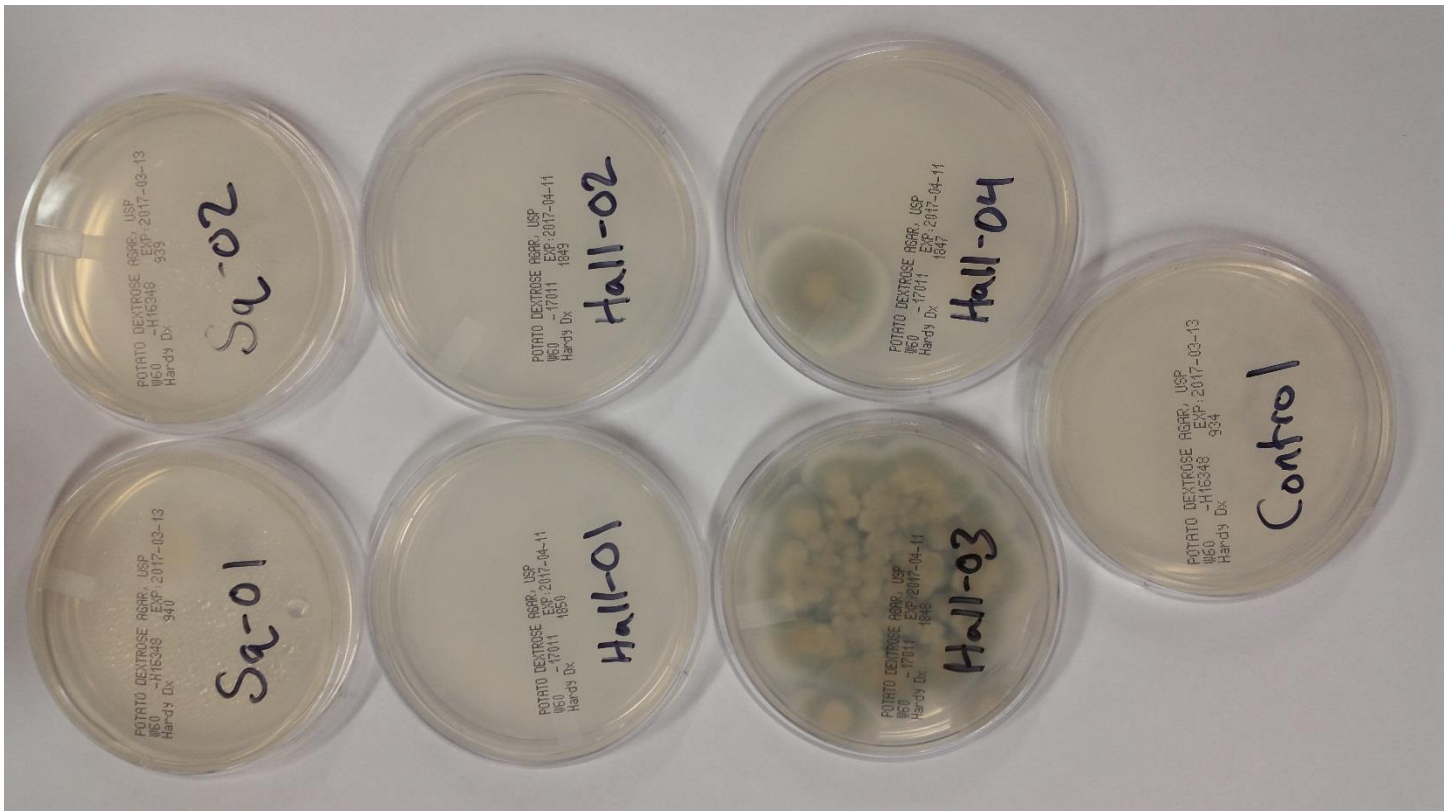
3	Stainless	A	856,000	5.93 ± 0.01		
	Plastic		845,500	5.93 ± 0.01		
	Stainless	B	889,000	5.95 ± 0.01		
	Plastic		879,000	5.94 ± 0.02		
	Stainless	C	881,000	5.94 ± 0.01	346,000,000	8.54 ± 0.02
	Plastic		882,000	5.95 ± 0.01	340,500,000	8.53 ± 0.02
	Stainless	D	888,900	5.95 ± 0.02		
	Plastic		875,000	5.94 ± 0.01		
	Stainless	E	895,500	5.95 ± 0.02		
	Plastic		880,000	5.94 ± 0.01		
4	Stainless	A	2,850,000	6.45 ± 0.02		
	Plastic		2,790,000	6.45 ± 0.01		
	Stainless	B	3,100,000	6.49 ± 0.02		
	Plastic		2,950,000	6.47 ± 0.01		
	Stainless	C	3,000,000	6.48 ± 0.02	343,900,000	8.54 ± 0.01
	Plastic		2,950,000	6.47 ± 0.02	342,250,000	8.53 ± 0.01
	Stainless	D	3,150,000	6.50 ± 0.02		
	Plastic		2,980,000	6.47 ± 0.01		
	Stainless	E	2,990,000	6.48 ± 0.01		
	Plastic		2,750,000	6.44 ± 0.01		
Positive Controls Not Exposed	Stainless		-	-	345,000,000	8.54 ± 0.02
	Plastic		-	-	355,500,000	8.55 ± 0.02
Positive Controls Sterile Water	Stainless		-	-	0	0
	Plastic		-	-	0	0

* Values are mean ± standard deviation, N=4 for each treatment location, N=4 for traveling control, initial population 8.6 ± 0.02 CFU/5g

In-treatment Air Samples

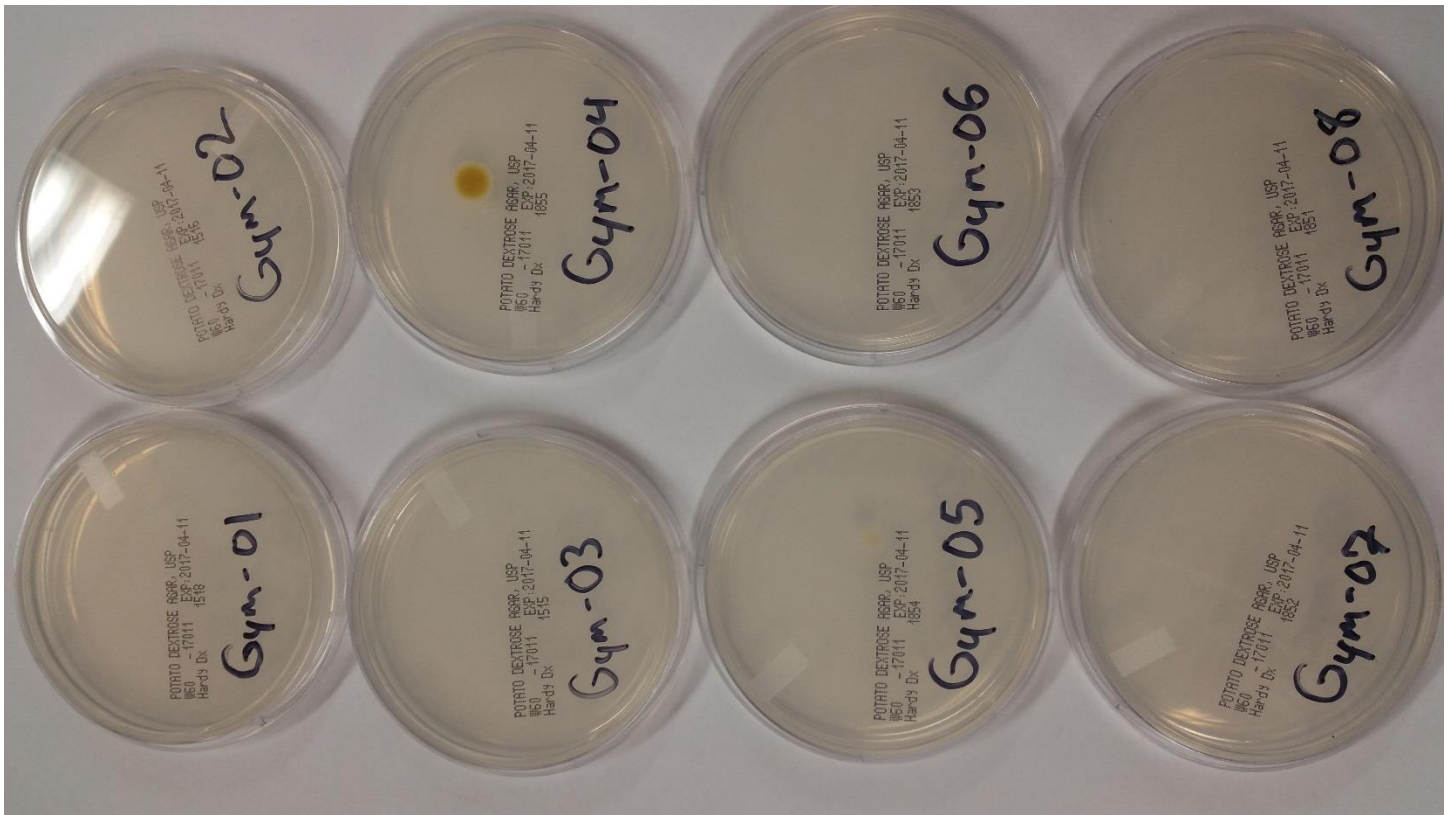


Outside air, PT and Lobby Air Samples

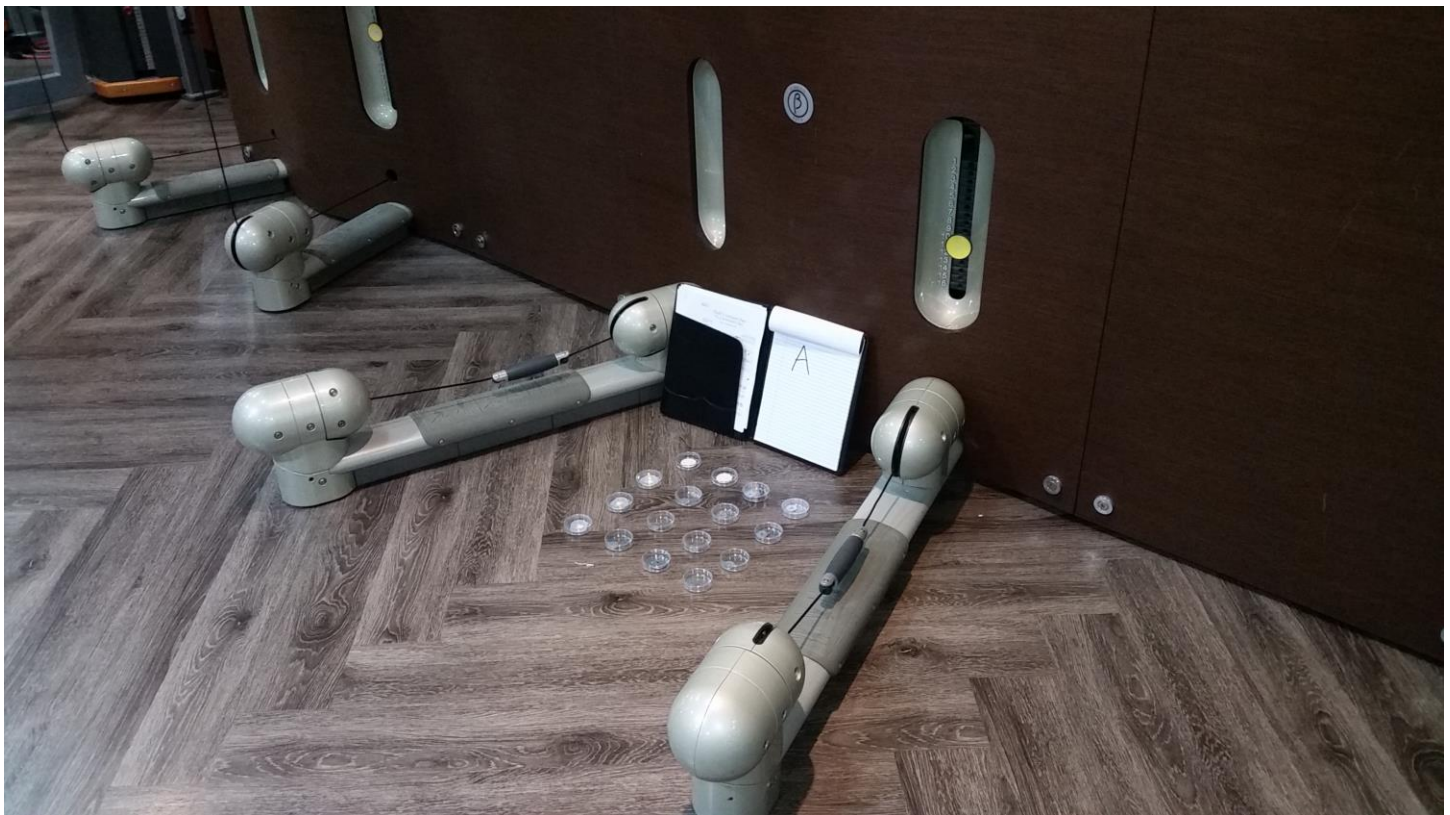


SA, Hall and Control

In-treatment Air Samples



Gym air samples



Stainless Steel and Plastic Coupon Location A

In-treatment Air Samples



Stainless Steel and Plastic Coupon Location B

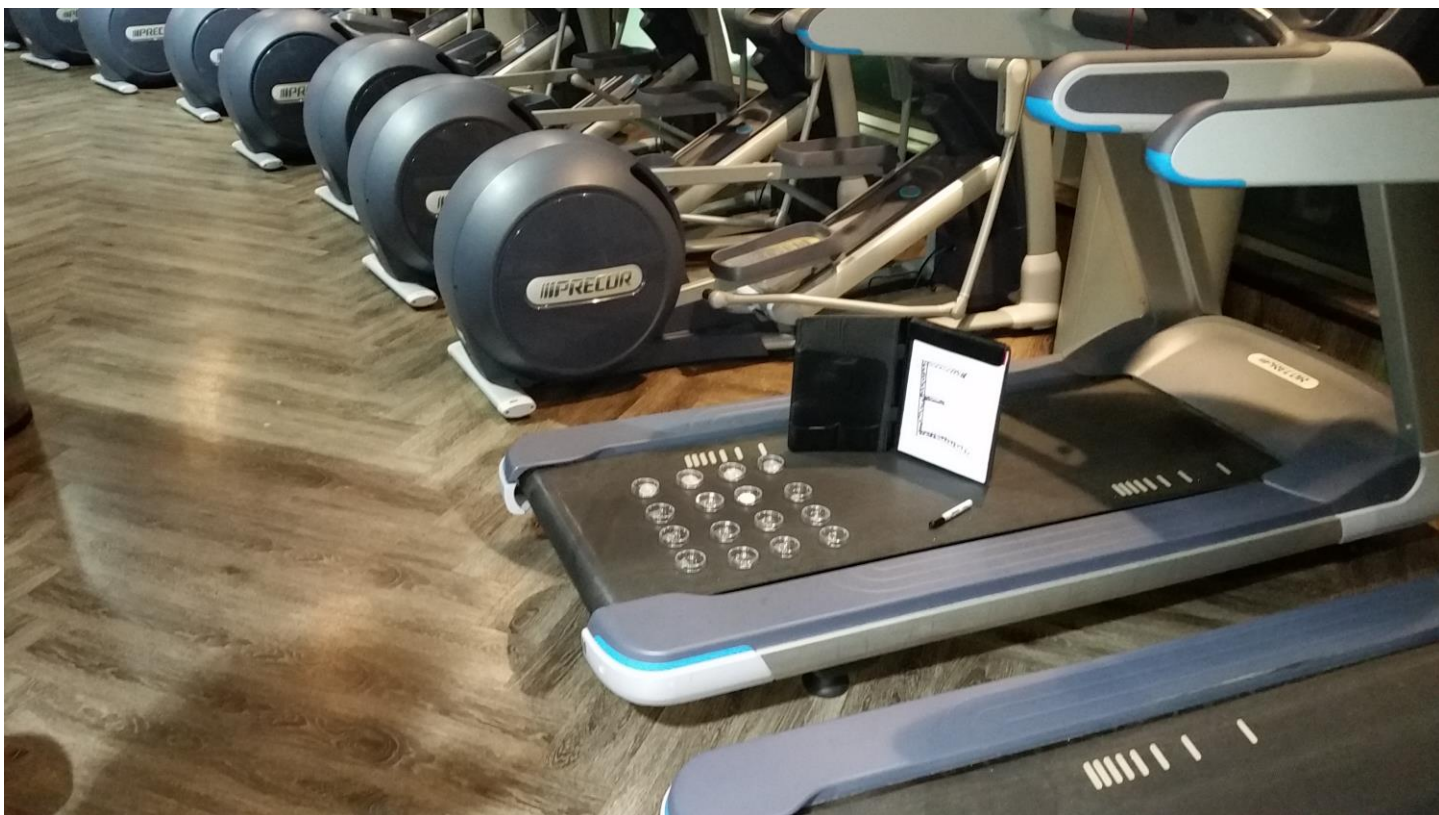


Stainless Steel and Plastic Coupon Location C

In-treatment Air Samples



Stainless Steel and Plastic Coupon Location D



Stainless Steel and Plastic Coupon Location E

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PhoenixAire
1100 North Glebe Road, Suite 600
Arlington VA 22201

February 17, 2017

In-Treatment Air and Surface Report – [REDACTED] Large Workout Area

A. Summary – Air Samples

Pre-treatment air samples were not taken and In-treatment 02/07/17 with results given below.

Sample Date	Treatment	Number of samples	Location	Average (cfu/m ³)	Range	Standard Deviation
02/07/17	In-Treatment	8	Gym	50	0/100	33.4
		2	PT	33	33/33	0
		2	Lobby	67	33/100	47.4
		2	SA	50	33/64	24.0
		4	Hall	33	0/67	23.6
		4	Exterior	2,917	2,567/3,367	295.8
		1	Control	0	0/0	-

Background

All air samples were taken via the MB-2 air sampler, 30 liters per sample throughout the various locations given above with results normalized to colony forming units per cubic meter of air (CFU/m³).

Given below is a review of the finding of the types of airborne organisms found in the above given locations and outside during this in-treatment sampling.

Species	Raw Count	Species	Raw Count
Aspergillus fumigatus	3,234	Penicillium brevicompactum	1,093
Penicillium, aspergillus types	2,134	Stachybotrys chartarum (atra)	705
Cladosporium sphaerospermum	2,056	Mortierella, spp	556
Penicillium purpurogenum	1,985	Basidiospores spp	327

Pre-treatment samples were not taken.

In-treatment bioburden in the aforementioned rooms average **47 cfu/m³** which is < 100 cfu/m³ and considered **clean and acceptable** per the Target Air Quality scale given below.

Observations

In-treatment exterior air samples ranged from **2,567** to **3,367 cfu/m³** and exhibits that a significant amount of the bioburden is coming from the outside air.

Target Air Quality

Air quality scale for workplaces, public buildings, schools, and homes are as follows:

- < 100 cfu/m³ is considered **clean and acceptable**.
- 100 to 300 cfu/m³ is **marginal**.
- > 300 cfu/m³ is **not acceptable** and needs corrective action.

In most cases, air quality < 100 cfu/m³ has shown a decrease in the overall bioburden and odors.

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B. Summary – Contact (Surface) Swabs

Pre-treatment surface swab samples were taken 11/09/16 and In-treatment samples taken 02/07/17 with results given below.

Sample Date	Treatment	Number of samples	Location	Average (cfu/cm ²)	Range	Standard Deviation	Percent Reduction
10/10/16	Pre-Treatment	9	Various Gym Locations	1,584	151/6,103	1,552.9	-
02/07/17	In-Treatment			51	11/105	27.3	96.8
10/10/16	Pre-Treatment	1	Negative control	0	0/0	-	-
02/07/17	In-Treatment			0	0/0	-	-

Pre-treatment average bioburden in the above rooms was **1,584 cfu/cm²** which is > 10 cfu/cm² which is **not acceptable** and needs corrective action.

In-treatment bioburden in the above mentioned rooms' average **51 cfu/cm²** and shows a 96.8% reduction from the pre-treatment samples, however, remains > 10 cfu/cm² which is **not acceptable** and needs corrective action.

Target Contact Surface Quality


Contact surface quality scale for workplaces, public buildings, schools, and homes are as follows:

- < 45 cfu total or < 1.67 -log, or < 5 cfu/cm² is considered **clean and acceptable**.
- 140 to 260 cfu total or 2.15 to 2.41-log, or 5 to 10 cfu/cm² is considered **marginal**.
- > 260 cfu total or > 2.41 -log, or > 10 cfu/cm² is considered **not acceptable** and needs corrective action.

In most cases, air quality < 45 cfu total or < 5 cfu/cm² has shown a decrease in the overall bioburden and odors.

Please contact me if there are questions or if further information is needed.

Respectfully submitted,


Rick Falkenberg, Ph.D., AFS
Senior Principal Scientist

- In-Treatment						
Room	Swab Lot No.	Plate Number	Surface Swab Sample Location	10x10x10 cm	Raw Count	CFU/cm2
Control 1	2017-01	1	Control Plate	0	0	0
N/A	2017-01	1	Treadmill	10x10x10	4,500	45
N/A	2017-01	2	Bike	10x10x10	5,550	56
N/A	2017-01	3	Row	10x10x10	2,250	23
N/A	2017-01	4	Stair	10x10x10	6,800	68
N/A	2017-01	5	Elliptical	10x10x10	3,300	33
N/A	2017-01	6	Front Desk	10x10x10	8,125	81
N/A	2017-01	7	Hoist Gym	10x10x10	4,500	45
N/A	2017-01	8	Medicine balls	10x10x10	7,127	71
N/A	2017-01	9	Bosus	10x10x10	4,800	48
N/A	2017-01	10	Technogym	10x10x10	10,500	105
N/A	2017-01	11	Massage tables	10x10x10	9,300	93
N/A	2017-01	12	Foam mats	10x10x10	5,200	52
N/A	2017-01	13	Lat pull down	10x10x10	5,800	58
N/A	2017-01	14	Barbells & wts	10x10x10	3,300	33
N/A	2017-01	15	Marpo rope pull	10x10x10	1,600	16
N/A	2017-01	16	Bosu knee lift	10x10x10	3,100	31
N/A	2017-01	17	Adj. bench	10x10x10	10,100	101
N/A	2017-01	18	Freeweights	10x10x10	3,300	33
N/A	2017-01	19	Cable station	10x10x10	1,100	11
N/A	2017-01	20	Leverage shoulder	10x10x10	2,100	21
Total Adjusted Raw Count					102,352	
Total CFU/cm2						1,024

Avg 51
 Low 11
 High 105
 SD 27.3

- Large Room - In-treatment						
Room	Plate Lot No.	Air Sample Location	Air Sample Location	Raw Count	Corrected Count	CFU/m3
Control 1	934	Control Plate	Unopened	0	0	0
B	1515	1	Gym main area	2	2	67
B	1516	2		3	2	67
B	1518	3		2	2	67
B	1855	4		1	1	33
B	1854	5		0	0	0
B	1853	6		0	0	0
B	1852	7		2	2	67
B	1851	8		3	3	100
B	1846	9	Lobby 1	1	1	33
B	943	10	Lobby 2	3	3	100
B	939	11	SA 1	2	2	67
B	940	12	SA 2	1	1	33
B	1848	13	Hall 1	1	1	33
B	1847	14	Hall 2	2	2	67
B	1849	15	Hall 3	0	0	0
B	1950	16	Hall 4	1	1	33
B	941	17	PT 1	1	1	33
B	942	18	PT2	1	1	33
B	938	19	Exterior 1	73	89	2,967
B	937	20	Exterior 2	69	83	2,767
B	936	21	Exterior 3	65	77	2,567
B	935	22	Exterior 4	81	101	3,367
Total Adjusted Raw Count					375	
Total CFU/cm2						12,367

Avg 50
 Low 0
 High 100
 SD 33.4

Avg 33
 Low 0
 High 67
 SD 23.6

Avg 2,917
 Low 2,567
 High 3,367
 SD 295.8

Practical Air Testing Solutions



November 20, 2016

PhoenixAire
1100 North Glebe Road, Suite 600
Arlington VA 22201

Pre-Treatment Air and Surface Report – [REDACTED]

Summary

Pre-treatment surface (swab) samples were taken 10/10/16 with results given below.

Sample Date	Treatment	Number of samples	Location	Average (cfu/cm ²)	Range	Standard Deviation
10/10/16	Pre-Treatment	20	Various Gym Locations	1,584	151/6,103	1,552.9
		1	Negative control	0	0/0	-

The pre-treatment bioburden in all of the above rooms are > 10 cfu/cm² which is **not acceptable** and needs corrective action.

Target Contact Surface Quality

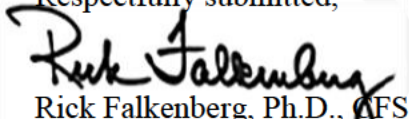
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- > 260 cfu total or > 2.41-log, or > 10 cfu/cm² is considered **not acceptable** and needs corrective action.

In most cases, air quality < 45 cfu total or < 5 cfu/cm² has shown a decrease in the overall bioburden and odors.

Please contact me if there are questions or if further information is needed.

Respectfully submitted,


Rick Falkenberg, Ph.D., CFS
Senior Principal Scientist

Room	Swab Lot No.	Plate Number	Surface Swab Sample Location	10x10x10 cm	Raw Count	CFU/cm2
Control 1	2017-01	1	Control Plate	0	0	0
N/A	2017-01	1	Treadmill	10x10x10	125,250	1,253
N/A	2017-01	2	Bike	10x10x10	205,125	2,051
N/A	2017-01	3	Row	10x10x10	185,252	1,853
N/A	2017-01	4	Stair	10x10x10	195,210	1,952
N/A	2017-01	5	Elliptical	10x10x10	181,250	1,813
N/A	2017-01	6	Front Desk	10x10x10	510,255	5,103
N/A	2017-01	7	Hoist Gym	10x10x10	75,258	753
N/A	2017-01	8	Medicine balls	10x10x10	301,450	3,015
N/A	2017-01	9	Bosus	10x10x10	210,252	2,103
N/A	2017-01	10	Technogym	10x10x10	105,200	1,052
N/A	2017-01	11	Massage tables	10x10x10	125,254	1,253
N/A	2017-01	12	Foam mats	10x10x10	610,255	6,103
N/A	2017-01	13	Lat pull down	10x10x10	78,500	785
N/A	2017-01	14	Barbells & wts	10x10x10	45,265	453
N/A	2017-01	15	Marpo rope pull	10x10x10	23,524	235
N/A	2017-01	16	Bosu knee lift	10x10x10	25,250	253
N/A	2017-01	17	Adj. bench	10x10x10	100,236	1,002
N/A	2017-01	18	Freeweights	10x10x10	25,500	255
N/A	2017-01	19	Cable station	10x10x10	25,120	251
N/A	2017-01	20	Leverage shoulder	10x10x10	15,100	151

Avg 1,584
 Low 151
 High 6,103
 SD 1,552.9

Total Adjusted Raw Count 3,168,506

Total CFU/cm2 31,685

© 2006 The Authors

10.10.16

[illegible]

