

UW OSHKOSH BETTERAIR ENVIRONMENTAL SAMPLING STUDY



Care Partners Assisted Living Facility

Little Chute Wisconsin

December 1, 2016 – May 3, 2017

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Study Design

The study was designed to assess the influence of the BetterAir environmental probiotic system on levels of microbial contamination at Care Partners Assisted Living community in Little Chute, Wisconsin. Environmental sampling took place in four double-occupancy units (two each in two separate wings of the facility). All four rooms were immediately adjacent to a facility HVAC system, which separately controlled air flow to the respective wing of the facility. As part of the study, two of the rooms at the far end of one wing of the facility (room 3 and room 4) would be treated with the BetterAir environmental probiotic, while two other rooms at the far end of the opposite wing of the facility (room 7 and room 8) would serve as the untreated control rooms. Rooms 3 and 7 are identical in layout and are at the distal end of each wing. Rooms 4 and 8 are identical in layout and are adjacent to the neighboring room with an HVAC unit situated between them. The two wings of the facility are separated by a common area which contains the kitchen, dining area, and lounge room.

Environmental sampling was conducted using a moistened swab method to lift microbial cells from surfaces of an area of approximately 100 cm² from each surface sampled. This was done for three different types of surfaces. 1) HVAC duct surfaces located approximately 50 cm from the left and right of the vent opening, 2) wood surfaces including locations on window stools directly above the vent openings and the upper surface of the door leading into the room, and 3) carpeted surfaces located near the entrance, middle, and far end of the room. These locations were selected during a pilot study which determined these areas to be reliable for recovering microbial contaminants. During the duration of the study, both HVAC units were set to run the fan in continuous operation such that airflow through the HVAC system was constant to all rooms of the facility. To test microbial distribution through the HVAC system, the stream of air from vents was also sampled by placing inverted Petri plates directly on the vent grill.

To assess the presence of different types of microbial contamination, four different forms of microbial growth media were employed. 1) Sabouraud Dextrose Agar (SDA) designed to promote the growth of fungal spores, 2) Mannitol Salt agar (MSA) designed to exclude the growth of Gram negative bacteria and allow the growth of Gram positive bacteria with differential identification of presumptive *Staphylococcus aureus* cells, 3) MacConkey agar (MAC) designed to exclude the growth of Gram positive bacteria and allow the growth of Gram negative bacteria with differential identification of presumptive fecal indicator bacteria (FIB) that belong to the family *Enterobacteracea*, and 4) Eosin Methylene Blue agar (EMB) that functions similarly to MAC, but with somewhat relaxed stringency of inhibition. To increase the stringency of EMB to select against Gram positive bacteria, plates were amended with 1.5 ug/ml ampicillin to eliminate all growth of the environmental probiotic strains.

Sampling Methods

Surfaces were sampled using rayon-tipped transport swabs (Puritan Medical, ME, USA) moistened with a phosphate-buffered saline solution (pH 7.2) to maximize recovery from surfaces while preventing growth-in-transit artifacts compared to using growth-promoting broth as a hydrating solution. During an initial pilot study it was predetermined that using a single swab per type of growth medium plated provided sufficient recovery on petri plates from carpet samples and so each swab sample of this type of surface was transferred to a single petri plate of media using a three-step coverage approach to completely expose the plate while removing microbes from the surfaces of the swab material. Thus, three faces of the swab were extracted through three passes over the plate in separate directions for each swab sample taken. It was also predetermined that samples of non-carpet surfaces (i.e. vent ducts, window stools, and door surfaces) contained far too many microbial cells for direct transfer onto plates. Swab samples from these surfaces were diluted into 1 ml of phosphate buffer identical in formulation to the solution used to moisten all swabs for sampling. Just prior to plating the samples on growth media, the swabs were vortexed three times at high speed for the duration of 15 seconds each to release microbial cells from swab fibers. Sub-samples (100 microliters each) were plated on all four types of growth media using standard spread-plate techniques. A ten-fold dilution factor was applied to petri plate inoculated in this way. HVAC air samples were taken using SDA, MSA, MAC, and EMB plates directly exposed to air flow over the outflow vent (plates were positioned in a downward facing direction on the vent surface) for a period of 60 minutes while the HVAC system was operating. After petri plates were exposed to samples (swab or air flow), the plates were incubated at 31°C for three days to promote microbial colony formation.

After microbial growth, colonies were counted using a counting pen and qualitative assessments of colony identity were made on each type of growth media. The following qualities were used to categorize the environmental contaminants:

Mold – growth on SDA with colonies showing presence of aerial mycelium.

Gram positive bacteria – growth on MSA

Staphylococcus aureus – growth on MSA with presence of yellow halo in the agar around colonies and with golden colony pigmentation and diameter of 1 – 2 mm.

Gram negative bacteria – growth on MAC or EMB

Fecal indicator bacteria – growth of enteric bacteria such as *Escherichia coli* on MAC or EMB with opaque colonies pigmented from pink to light purple (MAC) or pink to blue (EMB).

For the numerous plates containing high levels of fungal contamination, plates were placed on a light box to help visualize the spaces between colonies and two portions of the plates were counted, averaged, and multiplied by their fractional surface area of the plate. For the most contaminated plates the sample was re-plated with further dilution from a duplicate unprocessed sample swab. The list of samples from each unit during sampling dates included:

4 swabs from window stools directly above vent ducts (2 plated each on 4 types of growth media)

4 swabs from vent ducts at arm's length (approx.. 50 cm) interior (2 plated each on 4 types of growth media)

12 swabs from carpet sites (12 plated from 3 different locations on 4 types of growth media)

4 vent outflow samples consisting of one for each type of media exposed for 60 minutes for each unit (all were incubated and counted)

Study Duration

A period of base line sampling took place between December 1, 2016 and January 19, 2017. On February 21, 2017 the BetterAir HVAC delivery system was activated to distribute probiotic into room 3 and 4. At the same time, in-room BetterAir devices were also installed and activated in room 3 and 4 to ensure that the probiotic was delivered adequately to the two test room. After a period of approximately two weeks to allow for probiotic accumulation, sampling of the test and control rooms resumed on March 3, 2017. The study concluded on May 3, 2017 after three successive sampling visits were conducted during the period of active probiotic dispersal.

Results

Overall, the results of the study demonstrated sufficient quantities of ambient microbial populations in each of the units sampled to assess the contribution of environmental decontamination methods. However, vent air flow samples from all four rooms demonstrated very low numbers of microbial cells being distributed through the HVAC system even during the deployment of the BetterAir environmental probiotic system; microbial loads were never observed above 4 colonies over the duration of an hour during any sampling visit or for any type of medium employed (Table A1). While the result was unexpected, it is possible that the sampling method for air flow was inadequate for detecting airborne microbial cells. While not a substantial source of detectable environmental microbes through the air flow, the ductwork provided among the highest levels of ambient microbial populations. Here, fungal cells accounted for all or nearly all of the recovered cells for many of the samples taken at this location (Fig. 1). A notable exception was immediately following deployment of the BetterAir environmental probiotic system in mid-February. After a two week period of time to allow for

probiotic accumulation, the rise in levels of Gram positive bacteria in all four rooms was clearly observed (Fig. 2).

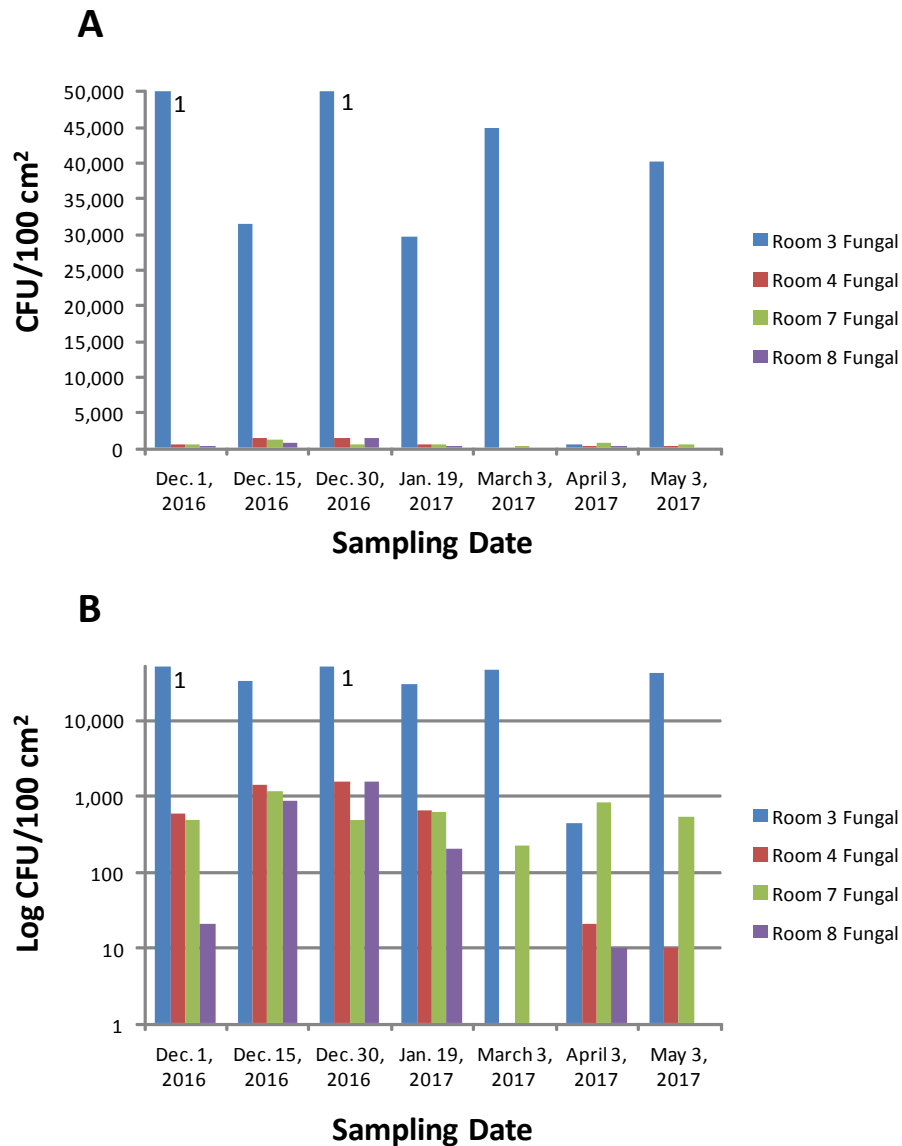


Figure 1. Vent interior mold. Samples were taken from vent duct interior surfaces approximately 50 cm distance from vent opening. Approximately 100 cm² surface was swabbed and diluted in 1 ml saline with 0.1 ml subsamples plated on SDA medium. The ten fold dilution factor has been applied to CFU values to account for the dilution. The limit of detection for this assay was 50,000 CFU/100 cm². Sample marked 1 are above the detection limit. Extreme levels of mold contamination in room 3 obscures underlying trends in rooms 4-8 (panel A) which can be observed by semi-log transformation of the data (panel B).

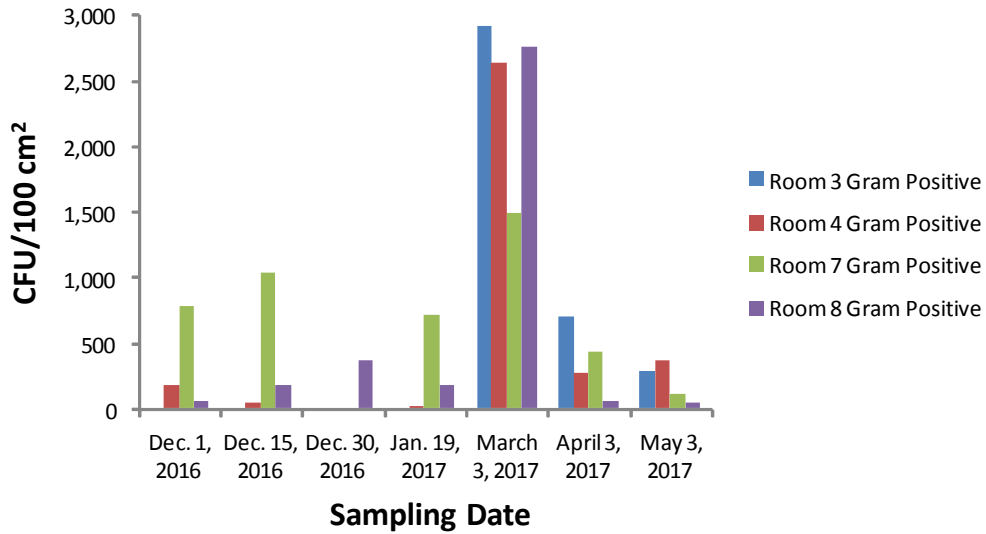


Figure 2. Vent interior Gram positive bacteria. Samples were taken from vent interior surfaces approximately 50 cm distance from vent opening. Approximately 100 cm² surface was swabbed and diluted in 1 ml saline with 0.1 ml subsamples plated on MSA medium. The ten fold dilution factor has been applied to CFU values to account for the dilution.

It is unclear at this time why all of the rooms would have experienced similar increases in Gram positive bacterial loads, but one explanation would be that the facility, on the whole, experienced a common circulation of and therefore distribution of the BetterAir probiotic. The colony morphologies on MSA plates were consistent with those observed in laboratory investigations of the environmental probiotic strains prior to initiating the study (Fig. 3). There were too few presumptive *Staphylococcus aureus* cells on vent surfaces to draw any conclusions about the influence of probiotics on this contaminant (Table A2).

The increase in Gram positive bacteria correlated with the total elimination of fungal contamination from vent duct interior surfaces on March 3, 2017 in room 4 where levels fell from nearly 1,000 cells per 100 cm² to undetectable levels. To rule out any error during plating, duplicate samples for this date were plated for room 4 with the same result. Initially, this response was not observed in room 3; however, one month later (April 3, 2017) a two-log (> 99%) reduction in fungal cells on vent duct interior surfaces was observed (Fig. 1A and 1B). Mold contamination levels began to rebound in room 3 by May 3rd and in room 4 by April 3rd. Interestingly, the high initial levels of mold in rooms 3 and 4 formed colonies that were black in color. As mold levels rebounded, samples were dominated by colonies that were white in color, possibly indicating a shift in fungal species took place during a period of overall lower contamination. Mold contamination also declined in one of the control rooms (room 8) but not in room 7. It is worth noting that Gram positive bacterial loads were much lower in room 7

than in room 8 (Fig. 2). If a critical threshold of probiotic cell density was not achieved on duct surfaces in room 7, it might account for the discrepancy between these two rooms.



Figure 3. Mannitol salt plate containing Gram positive bacteria. The colony morphology of Gram positive bacteria found in rooms after deployment of the BetterAir device is consistent with the strains found in the probiotic liquid.

A general decrease in Gram negative bacterial contamination on vent duct interior surfaces was also observed following activation of the BetterAir probiotic delivery systems. Here, the results were more variable, however, in the three consecutive sampling periods following delivery of the probiotic strains, all four rooms demonstrated low or undetectable levels of Gram negative bacteria, whereas prior measurements varied widely but were often in the hundreds of colonies per sample (Fig. 4A). The same trend was observed with presumptive fecal indicator bacteria (FIB) in the three consecutive sampling periods following delivery of probiotic strains (Fig. 4B). It must be noted here that the high variability in Gram negative and especially FIB detection (including some periods where they were not detected in duct surface samples prior to activation of the BetterAir devices); however, the decrease in Gram negative bacterial loads are also striking following the activation of the BetterAir probiotic system.

Like duct surfaces, the solid surfaces also experienced a rise in Gram positive bacterial loads associated with the activation of the BetterAir probiotic system (Fig. 5) as expected for delivery of the environmental probiotic strains. And like the duct surfaces, this occurred in all four rooms that were sampled. Here again, it is difficult to account for the larger Gram positive bacterial load in room 7 and room 8 which should have served as the untreated control room other than that air flow through the facility may have also delivered the probiotic strains to these control rooms. The solid surface samples demonstrated a marked decrease in fungal contamination following activation of the BetterAir devices (Fig. 6). Fungal levels remained low on these surfaces in room 3 for the three consecutive sampling dates following delivery of the

environmental probiotic. The average fungal contamination prior to activation amounted to approx. 3,600 CFU/100 cm² and after averaged only 326 CFU/100 cm², indicating a sustained >10-fold suppression of mold counts. Room 4 was also on track to demonstrate a similar suppression of mold numbers, however, the average mold levels prior to delivery of the environmental probiotic were much lower (723 CFU/100 cm²) in room 4 than in room 3 and they rebounded on the May 3, 2017 sampling date.

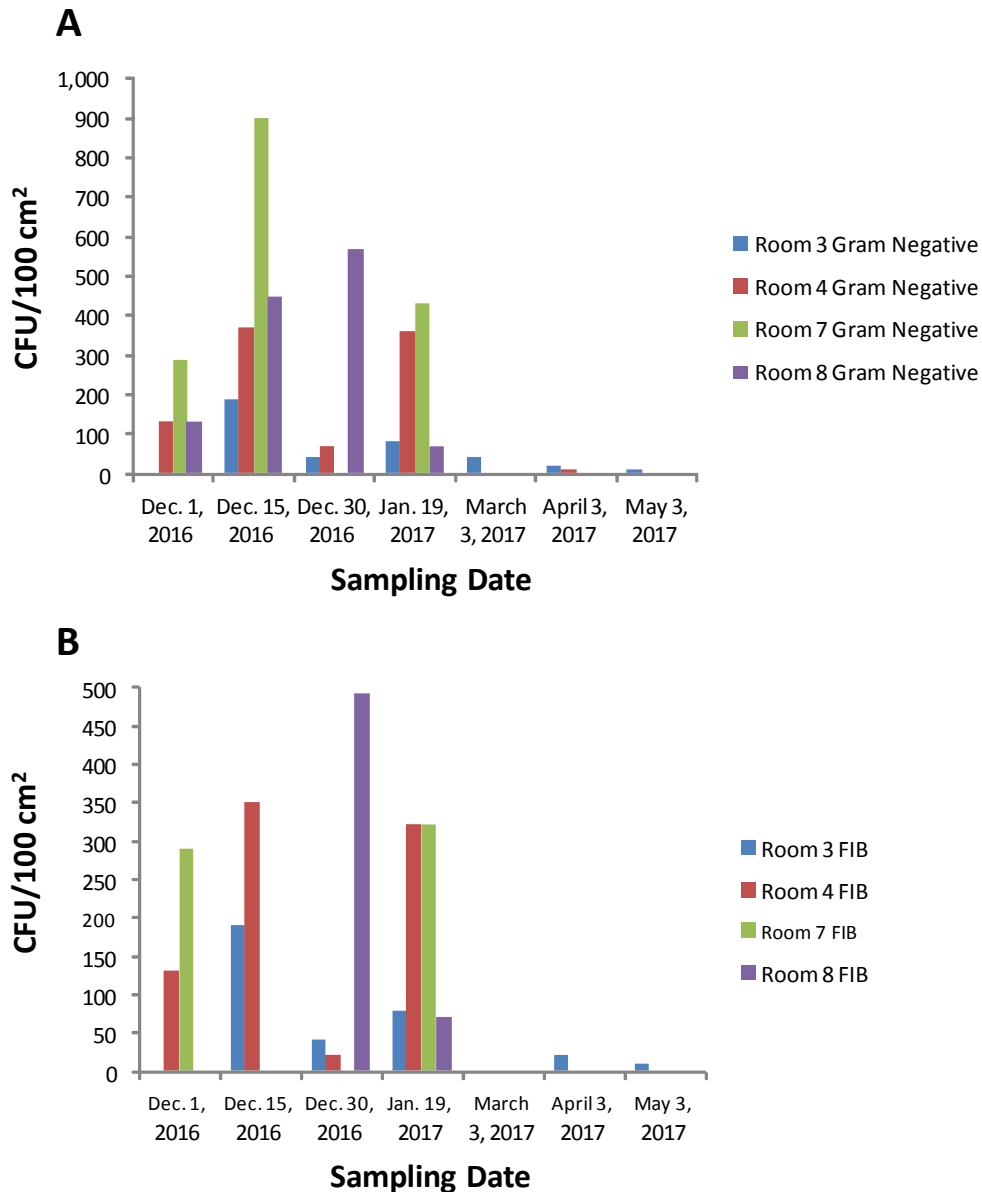


Figure 4. Vent interior Gram negative bacteria. Samples were taken from vent interior surfaces approximately 50 cm distance from vent opening. Approximately 100 cm² surface was swabbed and diluted in 1 ml saline with 0.1 ml subsamples plated on MAC and EMB media. The ten fold dilution factor has been applied to CFU values to account for the dilution. Total Gram negative bacteria (panel A) and presumptive fecal indicator bacteria (FIB) (panel B) were enumerated using the two forms of selective and differential growth media.

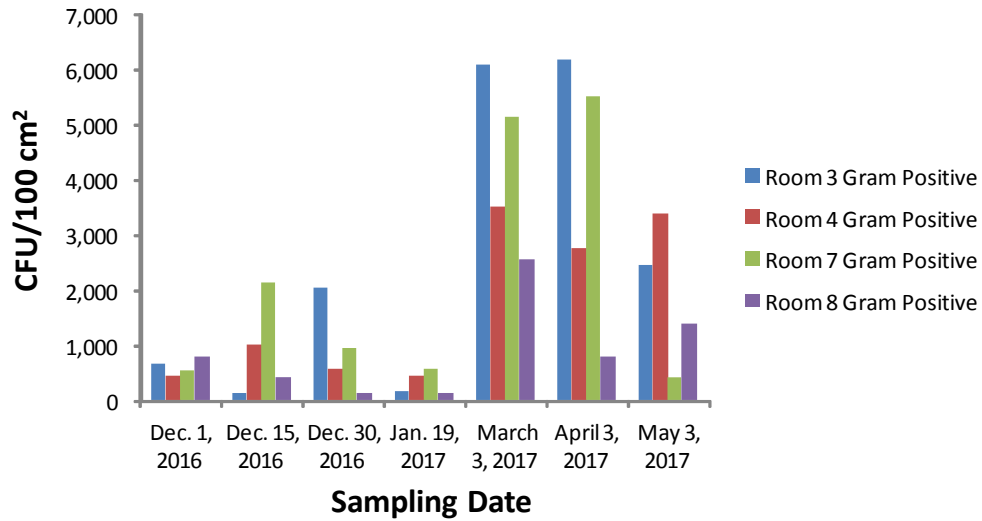


Figure 5. Solid Surface Gram positive bacteria. Samples were taken on window stools (lowermost interior horizontal surface) directly above vents and on the upper surface of the door to the room. Approximately 100 cm² surface was swabbed and diluted in 1 ml saline with 0.1 ml subsamples plated on MSA. The ten fold dilution factor has been applied to CFU values to account for the dilution.

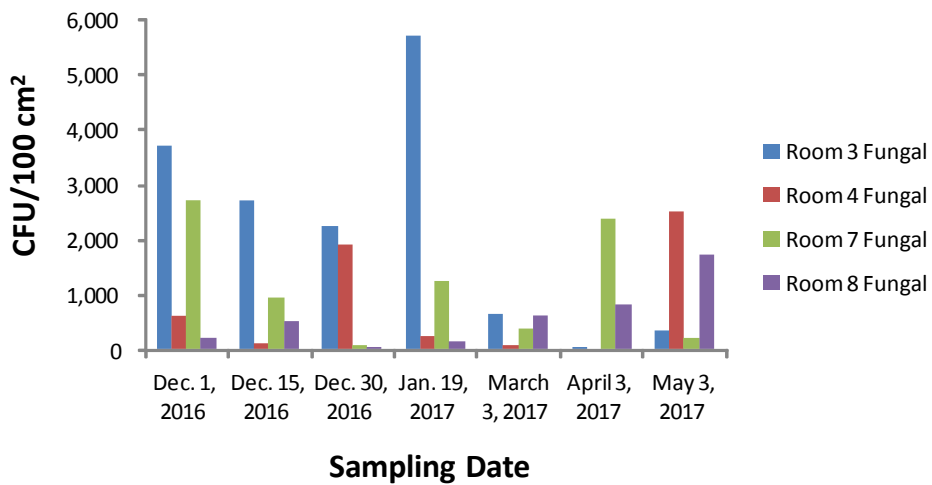


Figure 6. Solid Surface mold. Samples were taken on window stools (lowermost interior horizontal surface) directly above vents and on the upper surface of the door to the room. Approximately 100 cm² surface was swabbed and diluted in 1 ml saline with 0.1 ml subsamples plated on SDA. The ten fold dilution factor has been applied to CFU values to account for the dilution.

At times, high numbers of presumptive *S. aureus* cells were detected on solid surfaces. While no distinct, downward trend was noted, averages after treatment were lower than prior to treatment in rooms 3, 4, and 8 (Table A3). High numbers of Gram negative and fecal indicator bacteria were also detected in samples of the solid surfaces of all rooms. However, like presumptive *S. aureus* cells, these bacteria did not appear to follow a discernible downward trend in correlation with the dispersal of the environmental probiotic (Table A4) on these surfaces.

Carpeted surface, regardless of the sample location (entryway, middle, or far end of room), were often lower and much more variable than either the duct interior or solid surface samples. Unlike the duct and surface samples, a large increase in overall Gram positive bacterial counts was not observed following activation of the BetterAir devices (Fig 7A). This may be an indication that regular cleaning, such as vacuuming or carpet scrubbing, the residential units was sufficient to disrupt the accumulation of the environmental probiotic. Levels of mold and Gram negative bacteria were often low and only detected in high numbers in certain samples (Table B2). These results are consistent with cleaning followed by point source contamination events that would happen to be detected in some samples periodically. As such, the values from these samples are too variable to draw conclusions as to the correlation between the delivery of environmental probiotic and mold or Gram negative bacterial contamination levels. However, the incidence of colonies consistent with *S. aureus* appeared to diminish following the activation of the BetterAir devices (Fig. 7B). Prior to activation in room 3, presumptive *S. aureus* colonies were detected on each sampling day ranging from 18-492 CFU/100 cm² (average 242 CFU/100 cm² overall) and after activation such colonies were observed in only 4 instances (essentially undetectable). Likewise, in room 4, levels ranged from 7-132 CFU/100 cm² (average 56 CFU/100 cm² overall) prior to activation and only a single colony consistent with *S. aureus* was found after activation, again essentially undetectable.

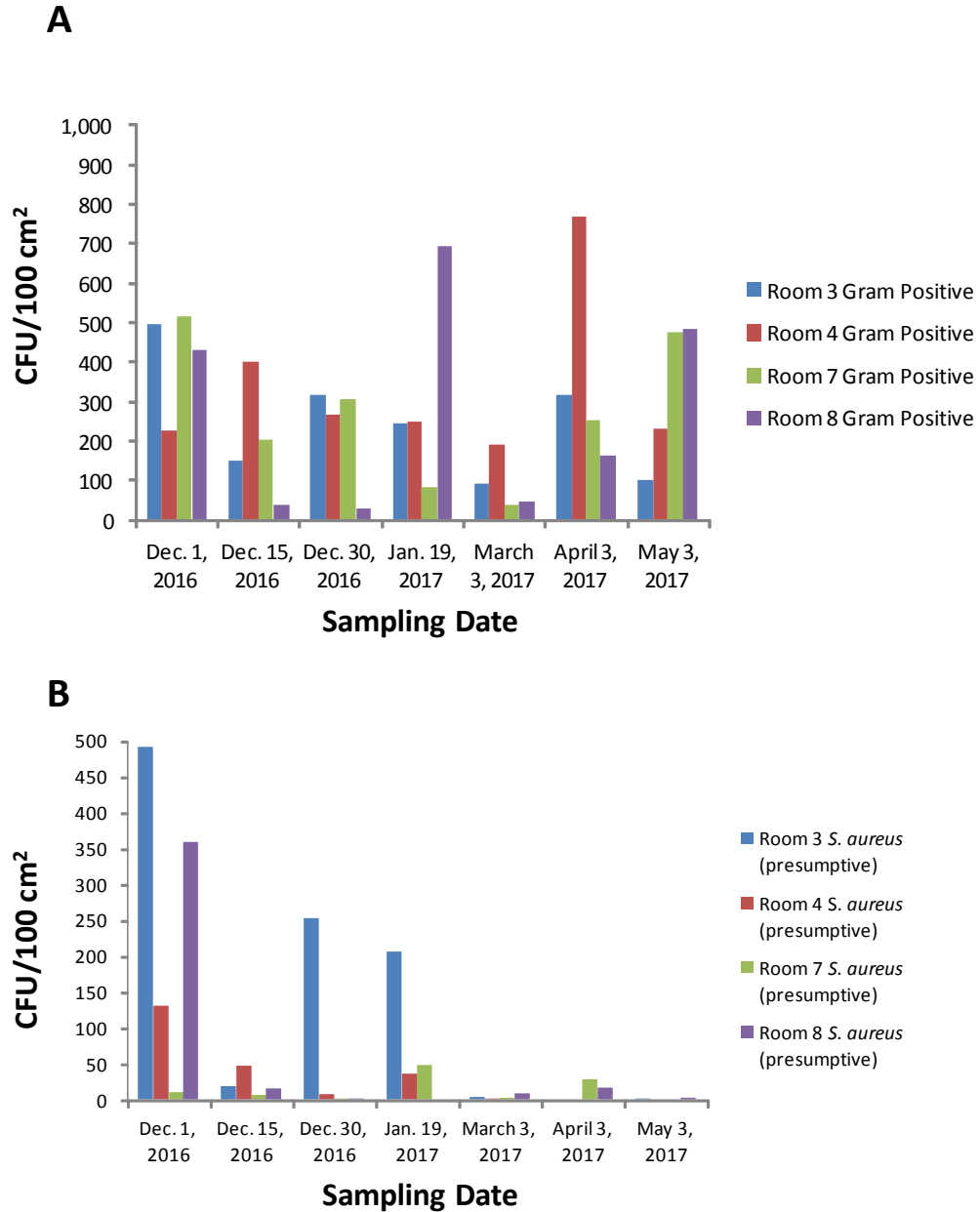


Figure 7. Carpeted surface Gram positive bacteria. Samples were taken from 100 cm² surfaces in three locations within each room. Each general location was swabbed separately four times and directly transferred to growth media without dilution. Colonies produced on agar plates were counted and were regarded as a direct representation of the surface area sampled. Total Gram positive bacteria (panel A) and presumptive *Staphylococcus aureus* colonies (panel B) were determined based on the selective and differential qualities of MSA plates.

Conclusions

Although many of the samples were highly variable both from room to room and on different sampling dates, such variability in the data is to be expected in a facility where residents, animals, and support staff have regular interactions within and between rooms, windows and doors are opened or shut at different times and for different durations, and cleaning schedules and thoroughness are potentially different among rooms. As the goal of the study was to ascertain what affect the BetterAir environmental probiotic delivery systems would have on an actively utilized living environment, all the data must be viewed in consideration of such numerous uncontrolled (and uncontrollable) variables. Moreover, especially as it pertains to carpet samples, although the specific time of day during which sampling took place and the intervals between samples were controlled, the schedule of room cleaning was not a controlled variable and while active carpet cleaning was never observed during sampling dates, it is certainly possible the some of the rooms may have been cleaned just prior to sampling while on other dates, a good deal of time may have passed between cleanings.

In considering such variables, there were some clear trends that correlate implementation of the BetterAir environmental probiotic with reduced levels of contamination:

- 1) Levels of Gram positive bacteria consistent with environmental probiotic increased after activation of the BetterAir delivery systems indicating that deployment onto duct and solid surfaces was effective.
- 2) Levels of Gram negative bacteria (including fecal indicator bacteria) decreased on vent duct interior surfaces, essentially dropping to undetectable levels in all four rooms after activation of the BetterAir delivery systems.
- 3) Mold levels decreased on vent duct interior surfaces and solid surfaces following activation of the BetterAir delivery system and, when levels rebounded, it appeared to be a different species of fungus that gave rise to mold colonies (i.e. white rather than black pigmented mold).

Additional considerations

The study was designed to separate controls (room 7 and 8) at the far end of the wing of the facility not treated with environmental probiotic from treated rooms (room 3 and 4) and the far end of the wing treated with environmental probiotic through the HVAC system and through in-room direct delivery. In practice, however, such a distinction may not be feasible because the dispersal of the probiotic through the HVAC system in one wing may result in circulating levels in all parts of the facility. Movements of occupants and staff along with recirculation via cold air return and redistribution through the ventilation system could feasibly contribute to sufficient air mixing (especially over the long periods of time covered in the study) so as to

render the control rooms essentially indistinguishable from those directly treated with the environmental probiotic. It is unclear why the methods employed should have failed to detect cells being dispersed through the ventilation system. However, because direct airflow samples were insufficient to detect the microbial cells (probiotic or otherwise), the contribution of airflow to the dispersal of cells remains unknown. As both an in-room delivery system and a BetterAir HVAC delivery system were used in treated rooms, it is not possible to determine the source of surface associated environmental probiotic other than to assume that probiotic cells measured on the interior surface of the duct almost certainly must have come from the HVAC delivery system as outward air flow through the vents was continuous throughout the entire duration of the study.

APPENDIX I

TABLE A1. Vent outflow samples at an exposure time of 1 hour with continuous air flow.

	Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 3							
Fungal	0	0	1	0	0	0	0
Gram Positive	0	0	1	0	1	0	1
<i>S. aureus</i> (presumptive)	0	0	0	0	0	0	0
Gram Negative	0	0	1	0	0	0	0
FIB	0	0	0	0	0	0	0
Room 4							
Fungal	0	1	0	0	1	1	1
Gram Positive	0	0	1	0	0	0	0
<i>S. aureus</i> (presumptive)	0	0	0	0	0	0	0
Gram Negative	0	2	1	0	1	0	0
FIB	0	0	0	0	0	0	0
Room 7							
Fungal	0	0	3	0	0	0	0
Gram Positive	0	0	0	0	0	2	0
<i>S. aureus</i> (presumptive)	0	0	0	0	0	0	0
Gram Negative	0	0	2	0	0	0	0
FIB	0	0	0	0	0	0	0
Room 8							
Fungal	0	0	1	0	0	0	0
Gram Positive	4	0	1	0	1	0	0
<i>S. aureus</i> (presumptive)	2	0	0	0	0	0	0
Gram Negative	0	0	1	0	0	0	0
FIB	0	0	0	0	0	0	0

Values are indicated in total CFU formed after 72 hours of incubation.

Shaded region indicates rooms and time period sampled during direct probiotic application.

TABLE A2. Presumptive *Staphylococcus aureus* colonies on vent interior surfaces.

	Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 3	0	0	0	0	0	0	0
Room 4	0	0	0	0	20	0	0
Room 7	70	0	0	0	0	0	0
Room 8	10	0	0	70	0	0	0

Values are indicated in total CFU formed after 72 hours of incubation.

Shaded region indicates rooms and time period sampled during direct probiotic application.

TABLE A3. Presumptive *Staphylococcus aureus* on solid surfaces.

	Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 3	0	30	170	20	7	10	10
Room 4	280	700	10	20	17	0	50
Room 7	140	10	10	20	0	120	0
Room 8	370	120	0	80	0	10	0

Values are indicated in total CFU formed after 72 hours of incubation.

Shaded region indicates rooms and time period sampled during direct probiotic application.

NOTE: Averages: Room 3 (before) = 55 CFU per day, (after) = 9. Room 4 (before) = 253 CFU per day, (after) = 22 CFU per day. Room 7 (before) = 45 CFU per day, (after) = 40 CFU per day. Room 8 (before) = 173 CFU per day, (after) = 3 CFU per day.

TABLE A4. Gram negative bacteria on solid surfaces.

	Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 3	120	1,680	3,320	1,600	5,770	980	790
Room 4	180	50	180	30	1,860	80	885
Room 7	580	570	110	10,080	685	380	5,620
Room 8	200	460	890	30	360	200	550

Values are indicated in total CFU formed after 72 hours of incubation.

Shaded region indicates rooms and time period sampled during direct probiotic application.

NOTE: While the numbers are high, there appears to be little in the way of distinguishable trends here. Numbers initially go up in 3 and 4.

TABLE A5. Mold on carpeted surfaces.

	Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 3	98	0	6	0	0	0	0
Room 4	46	1	1	66	0	0	0
Room 7	16	2	4	12	1	2	0
Room 8	78	0	0	0	0	0	0

Values are indicated in total CFU formed after 72 hours of incubation.

Shaded region indicates rooms and time period sampled during direct probiotic application.

APPENDIX II

Table B1. Averaged data from vent duct samples

		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 3	Fungal	50,000	31,350	50,000	29,520	44,790	440	40,010
Room 3	Gram Positive	0	0	0	0	2,920	700	280
Room 3	S. aureus (presumptive)	0	0	0	0	0	0	0
Room 3	Gram Negative	0	190	40	80	40	20	10
Room 3	FIB	0	190	40	80	0	20	10
		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 4	Fungal	570	1,380	1,490	630	0	20	10
Room 4	Gram Positive	180	40	0	10	2,640	270	370
Room 4	S. aureus (presumptive)	0	0	0	0	20	0	0
Room 4	Gram Negative	130	370	70	360	0	10	0
Room 4	FIB	130	350	20	320	0	0	0
		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 7	Fungal	470	1,150	480	600	220	820	530
Room 7	Gram Positive	780	1,040	0	720	1,490	430	110
Room 7	S. aureus (presumptive)	70	0	0	0	0	0	0
Room 7	Gram Negative	290	900	0	430	0	0	0
Room 7	FIB	290	0	0	320	0	0	0
		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 8	Fungal	20	850	1,490	200	0	10	0
Room 8	Gram Positive	60	180	370	180	2,760	60	40
Room 8	S. aureus (presumptive)	10	0	0	70	0	0	0
Room 8	Gram Negative	130	450	570	70	0	0	0
Room 8	FIB	0	0	490	70	0	0	0

NOTE: Limit of Detection Set at 50,000 CFU/100 cm²

Table B2. Averaged data from floor samples

		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 3	Fungal	98	0	6	0	0	0	0
Room 3	Gram Positive	492	149	313	243	89	316	98
Room 3	S. aureus (presumptive)	492	18	253	206	3	0	1
Room 3	Gram Negative	179	75	48	78	5	39	9
Room 3	FIB	0	73	46	0	4	25	8
		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 4	Fungal	46	1	1	66	0	0	0
Room 4	Gram Positive	226	400	264	248	188	766	230
Room 4	S. aureus (presumptive)	132	47	7	36	1	0	0
Room 4	Gram Negative	85	95	19	13	33	46	0
Room 4	FIB	0	33	6	0	31	19	0
		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 7	Fungal	16	2	4	12	1	2	0
Room 7	Gram Positive	514	204	306	83	36	252	475
Room 7	S. aureus (presumptive)	10	6	1	48	2	28	0
Room 7	Gram Negative	97	20	33	31	2,469	19	3
Room 7	FIB	0	16	18	2	2	5	2
		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 8	Fungal	78	0	0	0	0	0	0
Room 8	Gram Positive	431	38	27	694	44	162	486
Room 8	S. aureus (presumptive)	359	15	1	0	8	16	2
Room 8	Gram Negative	44	12	137	23	3	188	57
Room 8	FIB	43	9	6	0	2	44	27

Table B3. Averaged data from surface samples

		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 3	Fungal	3,710	2,710	2,250	5,690	630	10	340
Room 3	Gram Positive	680	150	2,040	180	6,087	6,190	2,440
Room 3	S. aureus (presumptive)	0	30	170	20	7	10	10
Room 3	Gram Negative	120	1,680	3,320	1,600	5,770	980	790
Room 3	FIB	20	40	1,720	730	20	340	780
		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 4	Fungal	620	110	1,910	250	90	0	2,500
Room 4	Gram Positive	460	1,010	570	460	3,520	2,770	3,400
Room 4	S. aureus (presumptive)	280	700	10	20	17	0	50
Room 4	Gram Negative	180	50	180	30	1,860	80	885
Room 4	FIB	20	10	10	20	200	30	580
		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 7	Fungal	2,700	940	70	1,240	370	2,380	210
Room 7	Gram Positive	550	2,130	960	570	5,140	5,530	430
Room 7	S. aureus (presumptive)	140	10	10	20	0	120	0
Room 7	Gram Negative	580	570	110	10,080	685	380	5,620
Room 7	FIB	0	330	50	10	685	340	1,240
		Dec. 1, 2016	Dec. 15, 2016	Dec. 30, 2016	Jan. 19, 2017	March 3, 2017	April 3, 2017	May 3, 2017
Room 8	Fungal	210	520	30	150	620	800	1,720
Room 8	Gram Positive	780	420	130	140	2,560	780	1,390
Room 8	S. aureus (presumptive)	370	120	0	80	0	10	0
Room 8	Gram Negative	200	460	890	30	360	200	550
Room 8	FIB	60	30	700	10	10	150	110