



FINAL REPORT ON THE GREENGUARD PILOT STUDY OF MICROBIAL RESISTANCE (Phases 1 and 2)

The Method and Current Participants

Mold growth on construction materials and furnishings is an unfortunate byproduct of elevated moisture and soil indoors. Some construction materials and indoor furnishings may support microbial growth and, thus, become indoor sources of biocontaminants or lead to product deterioration. Therefore, the potential for mold growth on building and interior furnishing materials is an important issue for the design, specification, and construction communities. The test method, based on EPA's research guide, published as ASTM D 6329-98 (2003), "Standard Guide for Developing Methodology for Evaluating the Ability of Indoor Materials to Support Microbial Growth Using Static Environmental Chambers" is designed to provide a quantitative measurement of microbial susceptibility (resistance) on building materials with reliability and reproducibility across a wide variety of building materials. A nine month laboratory study (Phase 1), followed by a year long pilot study of products submitted for testing (Phase 2), were performed to confirm and validate the choice of mold, consistency of results from control samples, the time course of mold growth, and that different materials respond differently.

Greenguard has completed the both phases of the pilot study undertaken to evaluate the method efficacy and the ability to rank products across building material product lines. The first phase of the study was focused on developing a baseline across newly manufactured products and establishing the ranking system applicability. In the second phase, a call for participants was made, and answered, to allow manufacturers to be stakeholders in the process, gain valuable information about their newly manufactured products' ability to grow mold, and see how products vary across types and constituency. Ten manufacturers participated in the study. Tested products included insulation, adhesive, flooring, and wallcovering. Those manufacturers who participated in the study and had products that were found to be mold resistant are listed along with their products in Attachment 1. Only those manufacturers who agreed to have their products listed are acknowledged.

Details of the Method Based on the Results of Phase 1

Overview

In the test, samples and control coupons (Whatman Filter paper) are inoculated with measured amounts of spores of a mold (*Penicillium brevicompactum*) that commonly occurs in buildings with water or moisture damage. One set of test and control coupons are harvested immediately (within one hour) as the time 0 incubation for use as the reference, or baseline, value. The other set of samples and controls are then incubated under controlled conditions at 95% RH (relative humidity) for three (3) weeks. The amount of mold recovered from the coupons is quantified at time 0 and at the end of three weeks of incubation to determine the material's degree of



resistance to mold growth. This is done by plating spores washed from the coupons and counting the colonies after incubation. Mold growth on the test material leads to increased amounts of mold recovered from the coupons. Results are reported on a scale of one (1) to five (5), with 1 being least resistant (highly susceptible to mold growth) and 5 being most resistant to mold growth, based on a quantitative count of mold colonies.

The test predicts a materials performance in terms of its ability to resist microbial colonization under the stated conditions. The test determines microbial resistance of new materials as manufactured and not as installed, soiled, or weathered. As such, the test is not a guarantee of product performance, since installation, modification of materials, aging, etc. all may impact the microbial resistance of materials.

The test was designed to provide reliable, quantitative, reproducible results across a wide variety of building materials within a reasonable amount of time under realistic and reproducible conditions favorable for mold growth.

Frequently Asked Question

Why a single mold? This may be better stated as “why not use a mixture of several molds”. The reason for a single mold rather than a mixture is related to the goal of having an objectively quantifiable test result and to having a test applicable and comparable across a wide variety of materials. When a sample is inoculated with multiple molds, there is competition among those molds not only on the sample, but also on the media used to harvest the mold. Each mold would need to be counted individually. However, the dominant mold(s) will vary across different product types and mold growth varies depending on the species. Additionally, competition by molds on the harvesting media may result in mold on the harvesting media present in different ratios as on the sample media.

Why *Penicillium brevicompactum*? There are several reasons for the choice of *Penicillium brevicompactum*.

- 1) *Penicillium brevicompactum* is naturally found growing in damp conditions on a wide variety of materials and, thus, makes the test applicable across a wide variety of materials under the test conditions used. It is representative of the most commonly encountered genus, *Penicillium*, found in problematic indoor environments. It is widespread in nature, in soil, and decaying vegetation. It is common on damp walls and building materials, and it is also present in dust from floors, bedding, and upholstered furniture.
- 2) *Penicillium brevicompactum* is relatively easy to handle and keep viable, helping to ensure reliable, consistent, reproducible results. This has been validated by the reproducibility of internal process control samples (Fig.1).
- 3) *Penicillium brevicompactum* grows in isolated colonies that do not overgrow another and, thus, makes them easy to count and quantify (Fig.2).

- 4) *Penicillium brevicompactum* requires only 4 days of growth to have countable colonies.
- 5) *Penicillium brevicompactum* shows a different response to different materials, in the expected manner. From the validation study, materials expected to be more resistant to mold growth show less growth of *Penicillium brevicompactum* than materials expected to be less resistant (Fig.3).

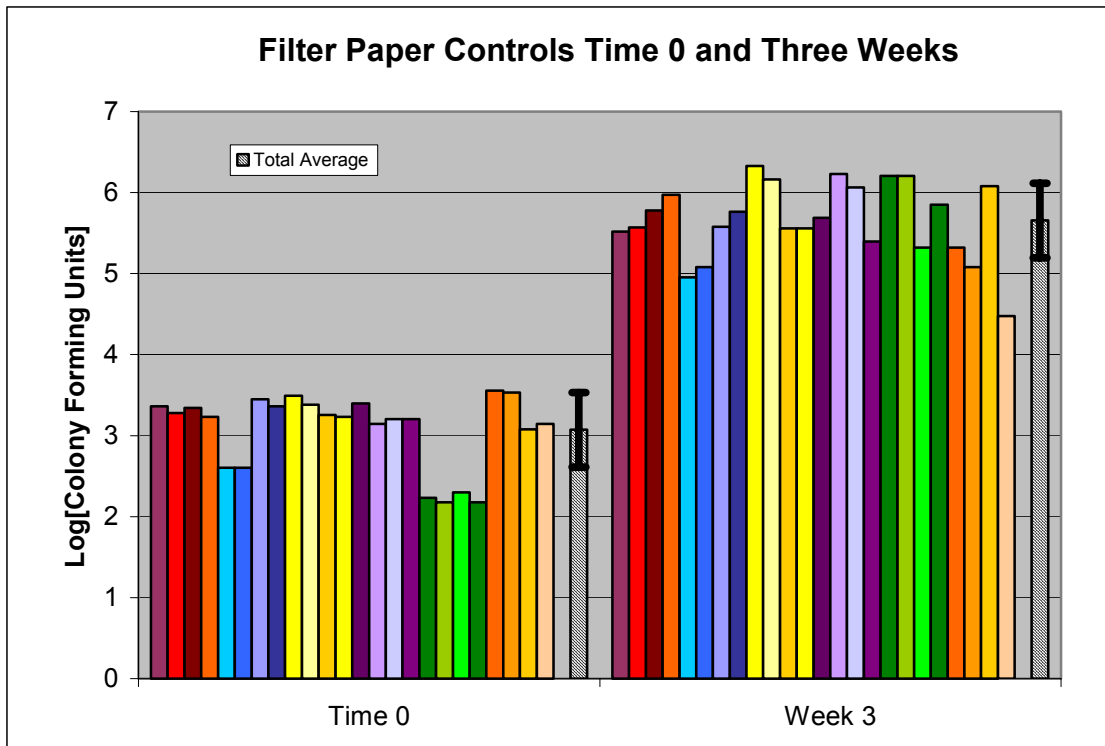


Figure 1 *Penicillium brevicompactum* growth on filter paper control samples at time 0 and after 3 weeks (n = 24 separate runs)



Figure 2 *Penicillium brevicompactum* colonies growing on media

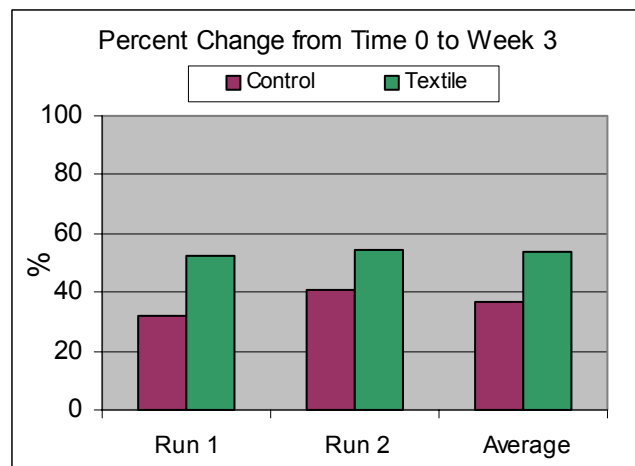
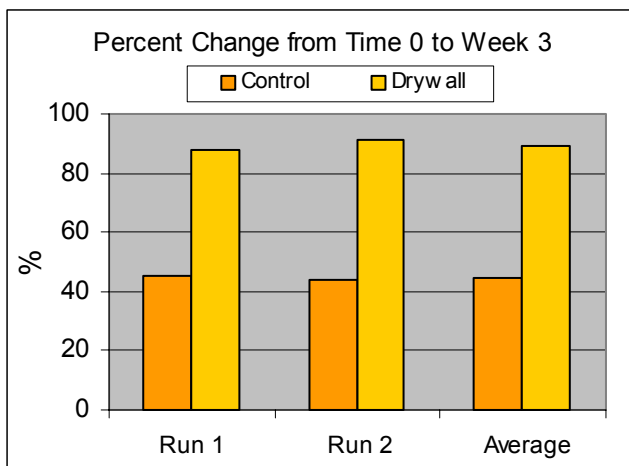


Figure 3 *Penicillium brevicompactum* growth on drywall and textile

Why 3 weeks at 95% RH? Very simply, these conditions enable a balance between rigorous testing and delivering fast results.

Why 95% RH? This creates a condition favorable to mold growth within a reasonable time frame. It mimics near condensation conditions, which occur in the case of water damage or poor humidity control, conditions which favor mold growth.

Why 3 weeks? Three weeks gives molds sufficient time to grow on a spectrum of materials from resistant to non-resistant while enabling a fast turn around time for the customer. Results from the validation study show a reproducible time course for the growth of *Penicillium brevicompactum* across a wide variety of materials with a plateau in the response at three weeks (Fig. 4). Again, this allows for consistent results across a wide variety of materials.

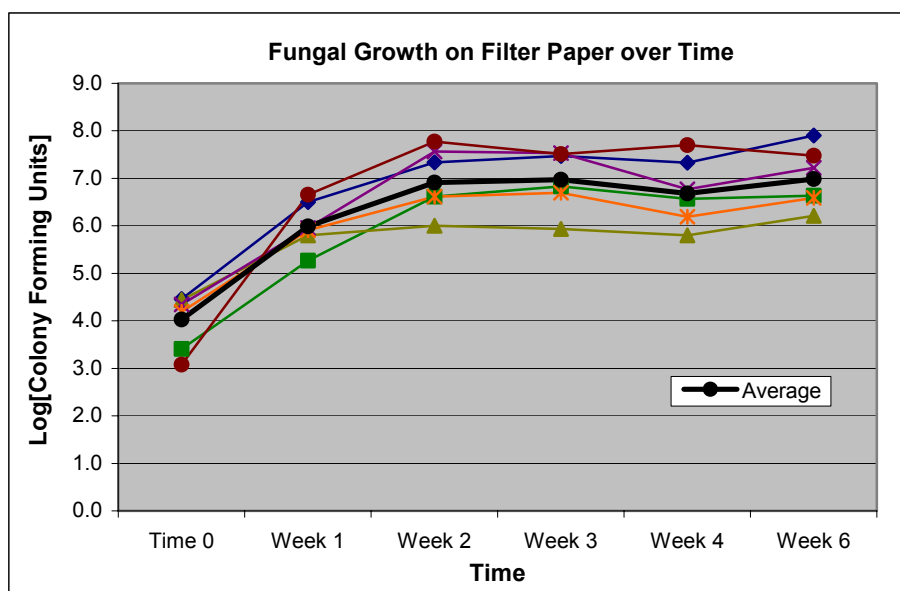


Figure 4 *Penicillium brevicompactum* growth time curves on filter paper

What Is the Expected Ranking System?

The results will be reported by product category and subcategories (if applicable). The initial results will be represented for each product category on a proposed scale of 1 to 5, with 1 being least resistant (or highly susceptible to mold growth) and 5 being most resistant (or least susceptible to mold growth). It will be based on a log scale of colony forming units, where the increase, or reduction, of colonies determines the ranking. This scale has been proposed using data obtained from recognized susceptible materials for each category, such as paper based wall materials, economy grade ceiling tiles, insulation, textiles, and engineered wood.



Ranking	Name	Definition
1	Not Resistant	Growth comparable to highly susceptible materials. 5 log increase in mold growth at 3 weeks.
2	Slightly Resistant	Growth comparable to slightly resistant materials. 3-4 log increase in growth at 3 weeks.
3	Moderately Resistant	Growth comparable with moderately resistant materials. Approximately 2 log increase in mold growth at 3 weeks.
4	Resistant	Growth comparable to resistant materials. Up to a 1 log increase at 3 weeks.
5	Highly Resistant	Growth comparable with highly resistant materials. No significant increase in growth (<20%) at 3 weeks.

Information to Date Based on the Validation Study (Phase One)

Initial method development (Phase 1) indicated that repeatable and consistent results can be obtained with this new method and that the proposed scale on a category basis is applicable. This second report presents the actual product data and an update on the proposed scale and ranking system.

Results from Phase 2

The combined results from Phases 1 and 2 based on the originally proposed ranking system are presented in Figure 5. However, review of the data indicated that the number of microbial colony forming units present after three weeks of incubation was a more reliable, and more relevant, measure since the concern is mold growth at 3 weeks, not a change in mold counts from time 0 to 3 weeks later. Thus, a revised ranking system based on the three-week results is proposed.

Ranking	Name	Definition
1	Highly Susceptible to Mold Growth	Growth comparable to highly susceptible materials. Log(CFU) > 7.5 at 3 weeks.
2	Susceptible to Mold Growth	Growth comparable to susceptible materials. Log(CFU) ≤ 7.5 and > 5.5 at 3 weeks.
3	Resistant to Mold Growth	Growth comparable to resistant materials. Log(CFU) ≤ 5.5 and > 2.5 at 3 weeks.
4	Highly Resistant to Mold Growth	Growth comparable with highly resistant materials. Log(CFU) ≤ 2.5 at 3 weeks.



FIGURE 5

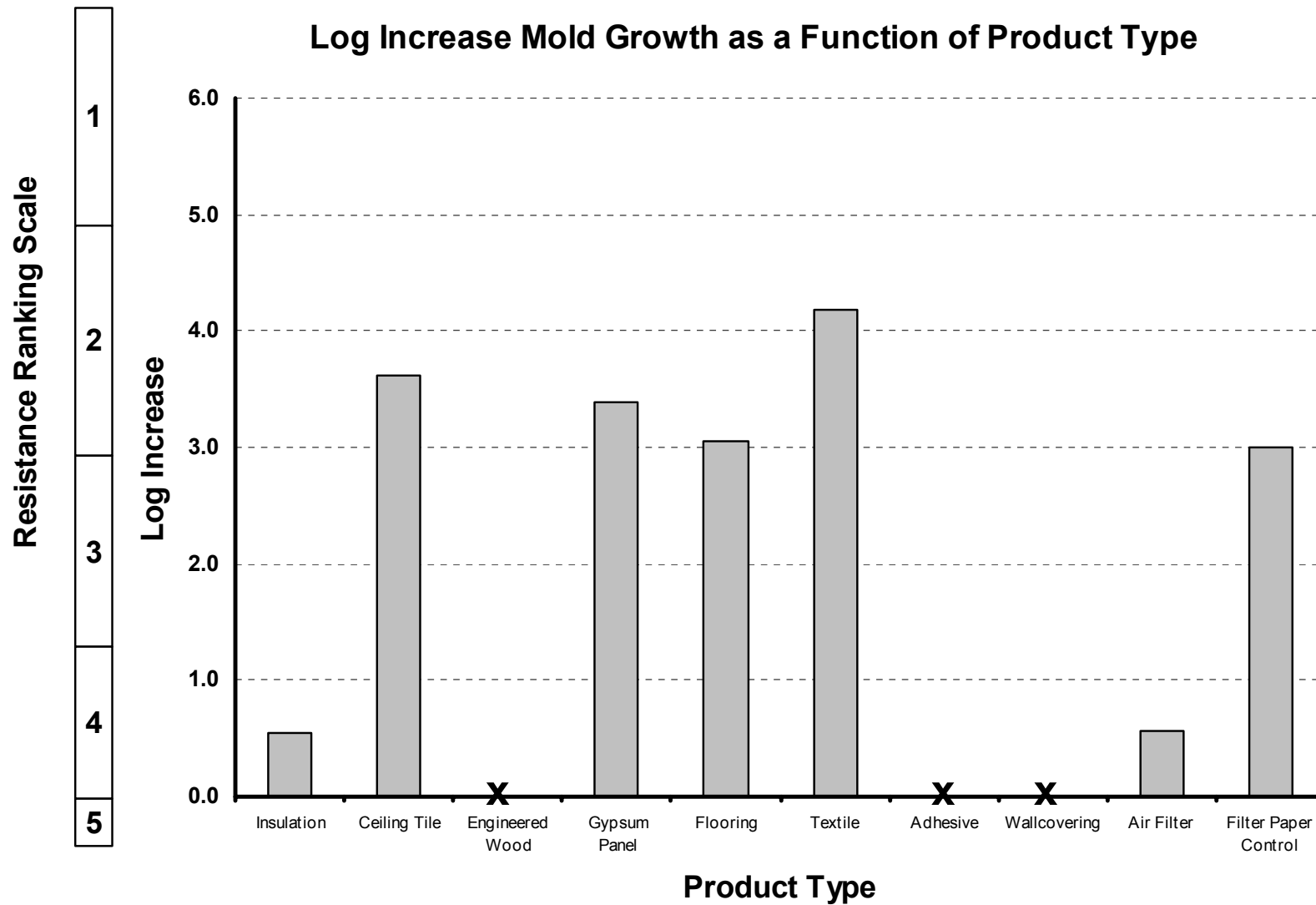




FIGURE 6

3 Week Mold Counts as a Function of Product Type

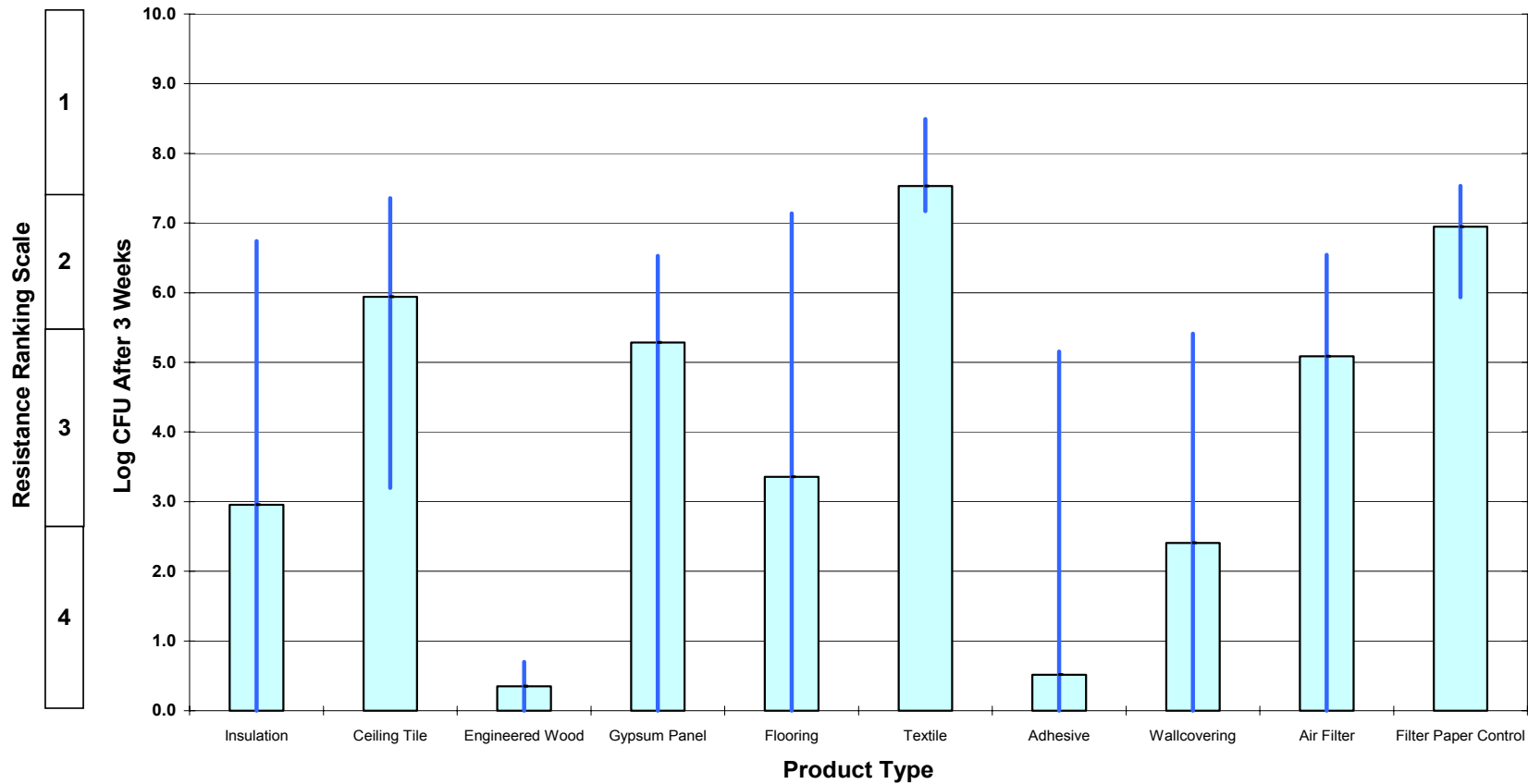


Figure 6 shows the 3-week average CFU data as a function of product of type, along with the updated Resistance Ranking Scale. Also included on the graph are bars indicating the range of values measured for each product type.

Summary Results

The results of the Pilot Study can be summarized as follows:

1. Throughout the two phases of the Pilot Study, the filter paper control 3 week log (CFU) results were consistent at 6.95 ± 0.49 (n=17). This consistency was maintained over the two-plus year period during which the Pilot Study was conducted and the tests run by a number of different analysts. Thus, the filter paper is useful as an internal control and reference and demonstrates the reproducibility of the method.
2. The method demonstrates variation in the measured CFU's at 3 weeks, both between and within product types, consistent with expectations based on product composition. This demonstrates that the method can differentiate between materials that are resistant and those that are susceptible to mold growth. That said, while *P. brevi* is a useful mold indicator for the product types shown, we are investigating the use of other molds more appropriate for specialized product types (e.g. masonry products).
3. The revised ranking system of 4 is appropriate for the range of products tested in this study and provides sufficient differentiation among the product's abilities to support mold growth.



Attachment 1 Summary of Microbial Resistant Products

Manufacturer	Product Name
Armacell, LLC	Tubolit – Polyethylene Pipe Insulation AP/Armaflex – Elastomeric Sheet Insulation AC/Accoflex – Elastomeric Pipe Insulation
Lonseal	LonFloor Vista SRG Greenmedic LonFloor #3902AB LonFloor Vista Greenmedic
DesignTex, Inc.	Wallcovering – Flip Wallcovering – Sumatra Wallcovering – Fern Wallcovering – Benchmark
Freudenberg Nonwovens	Air Filter T60 Air Filter Mini 95 Air Filter MV95
Mapei, Inc.	Ultra/Bond ECO 220 Ultra/Bond ECO 575 Ultra/Bond ECO 290
W. F. Taylor	2087 – EVR White Sheetgoods Adhesive 2055 – Performance Plus Fast Tack Carpet Adhesive 2091 – Tuff-Grip TPS Luxury Vinyl Tile and Plank Adhesive 2071 – Tuff Lok X-Linking Wood Flooring Adhesive
Thermafiber	Sound Attenuation 2.5# @ 3” Curtainwall Insulation Faced CW-90 2”

See AQS final report on Microbial Resistance Testing presented to the Greenguard Environmental Institute.