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Analytical Testing Report

Antifungal Product Testing

05/21/2021

Job #: 21143

Product Tested: Mold Guard New Pharmaceutical Formula

Date Received: 04/02/2021

Date Analyzed/Tested: 04/02/2021-04/30/2021

Date Printed Report: 05/21/2021

Sample Type: Antifungal Solutions (Liquids)

Analysis: Standard Practice for Determining Resistance of Materials to Fungi

Client: Mold Guard Inc. - 3 Place Ville-Marie Suite 400 Montréal, PQ H3B 2E3 -
www.moldguardinc.com

Contact: Vladimir Podlipskiy



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Protocol:

The protocol consists of, selection of specimens, sterilization of specimens, treatment of specimens, inoculation of the specimens with suitable organisms, exposure of inoculated specimens under conditions favorable to growth, and examination and rating for visual growth. The Nutrient-Salts Agar Medium was prepared by dissolving following reagents in 1 L of distilled water:

Potassium dihydrogen orthophosphate (KH_2PO_4) 0.7 g

Magnesium sulfate ($\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$) 0.7 g

Ammonium nitrate (NH_4NO_3) 1.0 g

Sodium chloride (NaCl) 0.005 g

Ferrous sulfate ($\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$) 0.002 g

Zinc sulfate ($\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$) 0.002 g

Manganous sulfate ($\text{MnSO}_4 \cdot \text{H}_2\text{O}$) 0.001 g

Agar 15.0 g

Potassium monohydrogen orthophosphate (K_2HPO_4) 0.7 g

Test medium was sterilized by autoclaving at 121°C (250°F) for 20 min. pH of the medium was adjusted by the addition of 0.01 N NaOH solution so that after sterilization the pH is between 6.0 and 6.5.

Five fungi cultures were prepared by growing them on a 1.5% potato glucose agar plate (Fluka) at 25°C for 7 days. The cultures were visually inspected before the next step.

The fungal species tested included:

- a. *Penicillium chrysogenum* ATCC 10106
- b. *Trichoderma Virens*; T-1 ATCC 9645
- c. *Aureobasidium pullulans*; var. *Melanigenum*; QM 279c ATCC 15233
- d. *Telaromyces pinophilus* ATCC 9644
- e. *Chaetomium globosum*; QM 459 ATCC 6205

A spore suspension of each of the five fungi species was prepared in the following way. A sterile wooden toothpick was used for each species to gently scrape a square centimeter of the



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surface growth from each culture of the test organism. The material was then transferred from each toothpick into a 10 ml portion of basal salt solution.

All five fungal suspensions were further diluted. The final solutions consisted of the mold spore suspensions in a basal salt solution at a concentration of $\sim 1,000,000 \pm 200,000$ spores per ml.

Each sample was tested using Petri dishes (150 mm) that contained sterile nutrient salts agar (pH 6.5) and sterile drywall (2' diameter) treated with Mold Guard. The negative control was two pieces of sterilized dry wall not treated with Mold Guard.

The drywall material consisted of the following:

- a. Gypsum ($\text{CaSO}_4 > 85\%$)
- b. Layered Recycled paper ($<15\%$)
- c. Starch ($<3\%$)
- d. Crystalline silica ($<5\%$)

The 2-inch drywall pieces were embedded in the solidified basal salt agar and inoculated with 100 micro liters of the fungal suspension. The same treatment was used for the control sample. The five fungi strains were tested separately.

The inoculated test specimens were incubated at 28-30°C (82-86°F) with no less than 85% relative humidity. The length of the test was 28 days of incubation as recommended by the ASTM G21 Fungal Defacement Test. The control samples were terminated at 20 days for exhibiting an extensive growth rating of four. The samples were examined at 4, 12, 20, and 28 days after inoculation for visible effects of mold growth. The following rating system was used to score the test samples. See Table 1: Observed growth on Specimens.

Observed Growth on Specimens	Rating
None	0
Traces of growth ($>10\%$)	1
Light growth (10-30%)	2
Medium growth (30-60%)	3
Heavy growth (60-100%)	4

Table 1: Observed Growth on Specimens Rating



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Results:

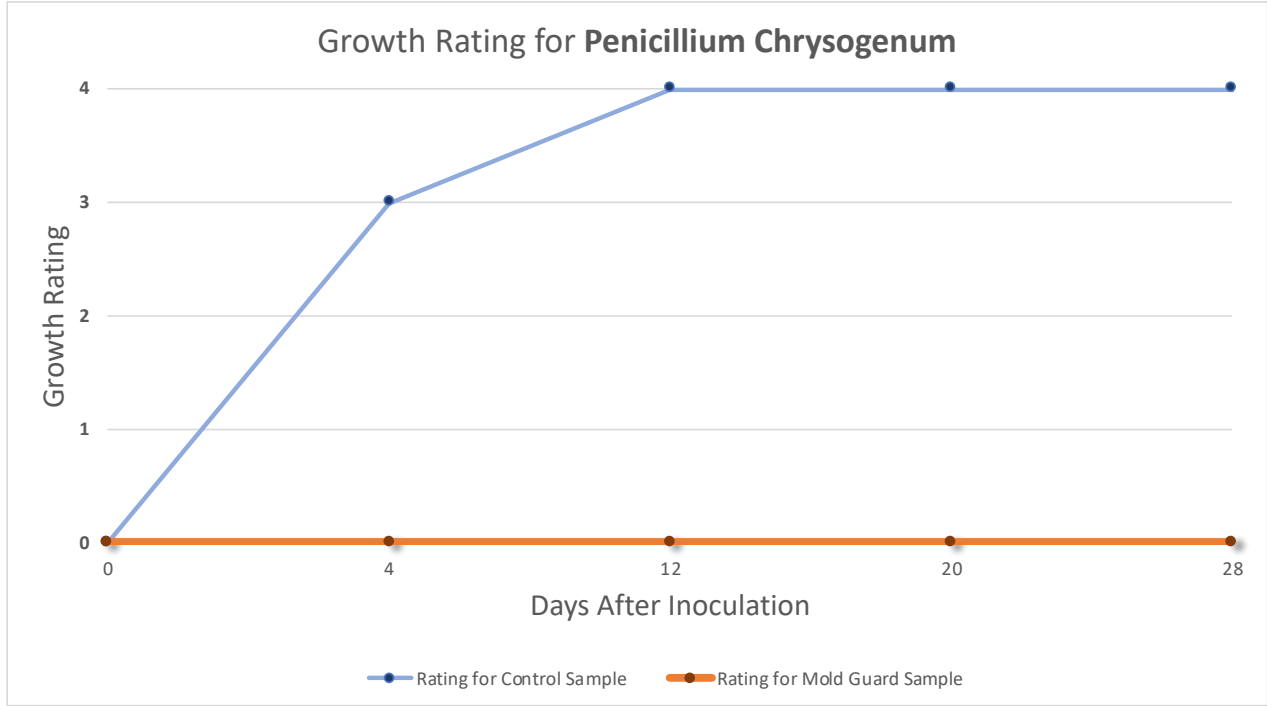


Chart 1: The control sample was overgrown by day 12. No mold growth occurred on the sample treated with Mold Guard.

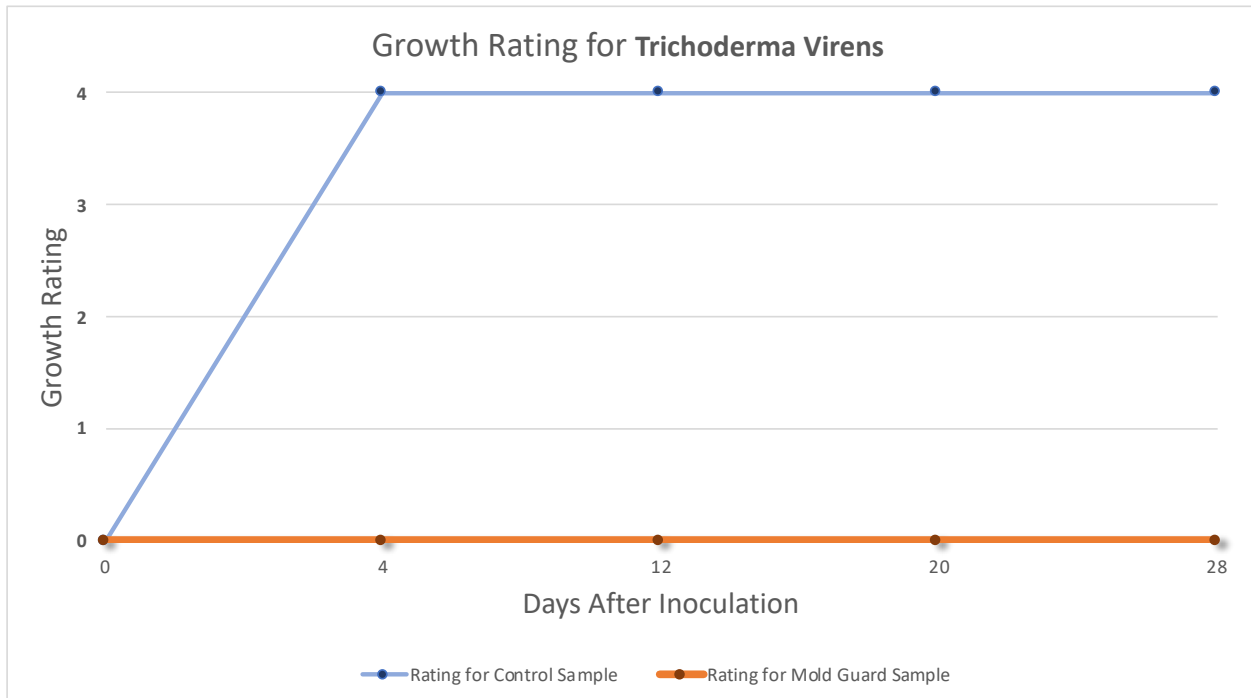


Chart 2: The control sample was overgrown by day 4. No mold growth occurred on the sample treated with Mold Guard.



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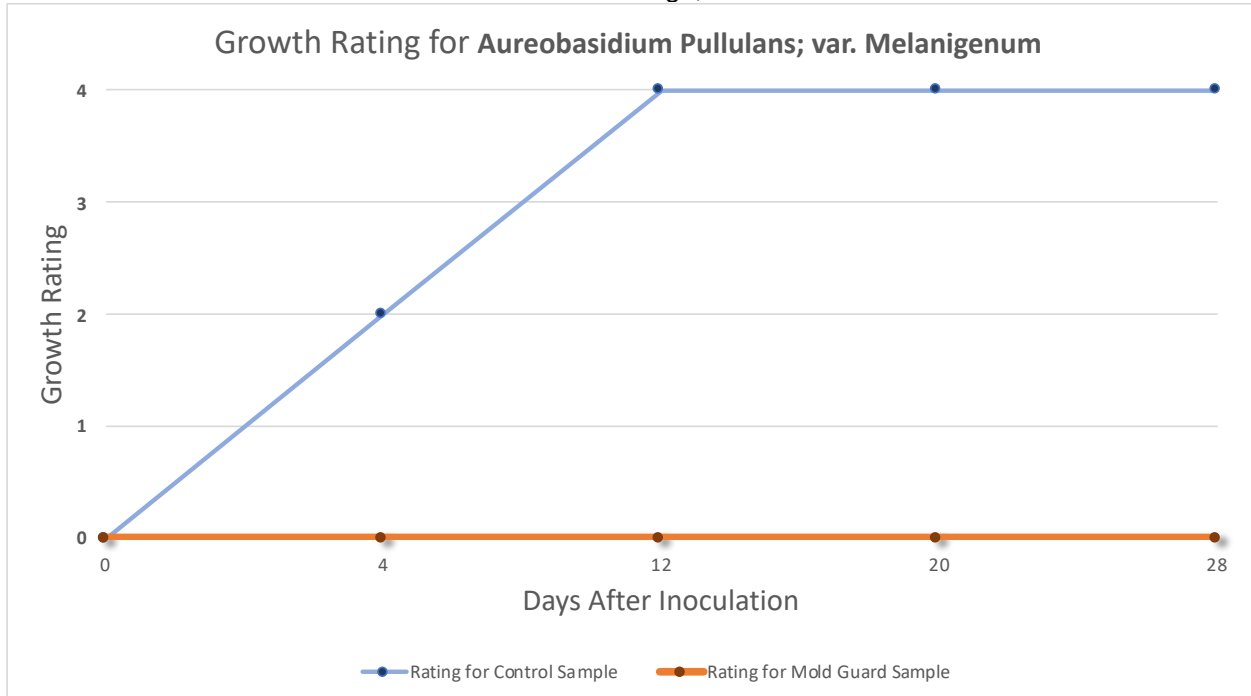


Chart 3: The control sample was overgrown by day 12. No mold growth occurred on the sample treated with Mold Guard.



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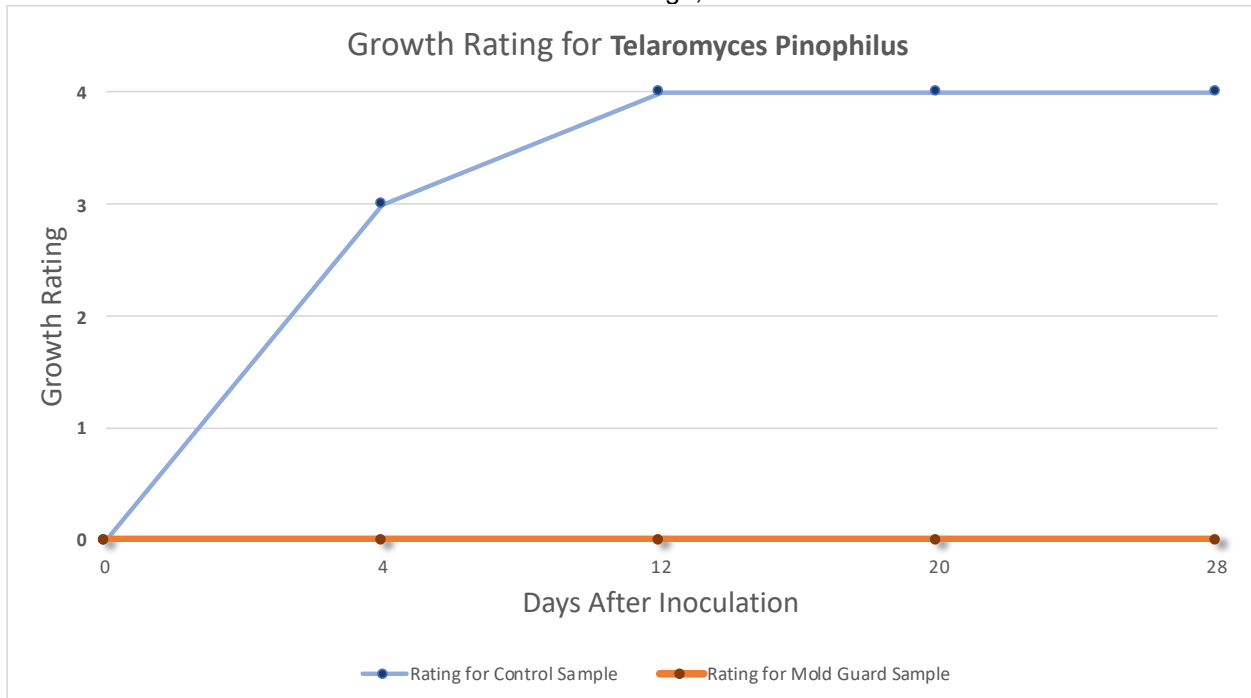


Chart 4: The control sample was overgrown by day 12. No mold growth occurred on the sample treated with Mold Guard.

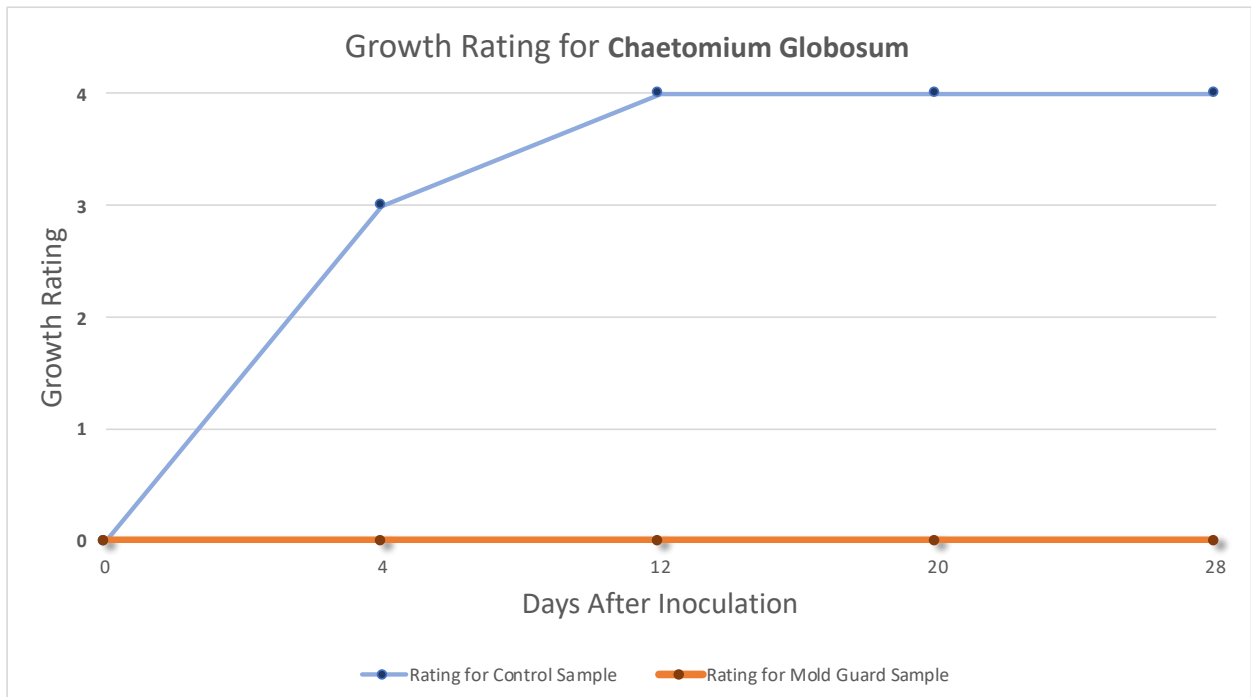


Chart 5: The control sample was overgrown by day 12. No mold growth occurred on the sample treated with Mold Guard.

Observations (see photographs in appendix):



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1. The dry wall treated with Mold Guard shows no signs of mold growth 28 days after inoculation.
2. The dry wall control samples not treated with Mold Guard and inoculated with penicillium chrysogenum, telaromyces pinophilus, and chaetomium globosum were under 60% covered in mold by day 4 and 100% covered by day 12.
3. The dry wall control samples not treated with Mold Guard and inoculated with trichoderma virens was 100% covered in mold by day 4.
4. The dry wall control sample not treated with Mold Guard and inoculated with aureobasidium pullulans; var. melanigenum was under 60% covered by day 4 and more than 90% covered by day 12.

Conclusion:

The drywall samples treated with Mold Guard prevented mold growth for 28 days (required by the ASTM G21 Fungal Defacement Test) at 28-30°C (82-86°F) with no less than 85 % relative humidity. Our data shows that Mold Guard has effective fungicidal activity and the ability to prevent fungi growth on a treated surface.

Test performed by Sergei Bibikov Ph.D.

Signed

Date:05-21-21

Scientist, Q.C. Officer Jim Polansky

Signed

Date: 05-21-21



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Appendix:

Sample ID	Rating				
	Strain 1 (<i>Penicillium chrysogenum</i>)	Strain 2 (<i>Trichoderma Virens</i> ; T-1)	Strain 3 (<i>Aureobasidium pullulans</i> ; var. <i>melanigenum</i> ; QM 279c)	Strain 4 (<i>Talaromyces pianophiles</i>)	Strain 5 (<i>Chaetomium globosum</i> ; QM 459)
Control Day 4	3	4	2	3	3
Mold Guard Day 4	0	0	0	0	0
Control Day 12	4	4	4	4	4
Mold Guard Day 12	0	0	0	0	0
Control Day 20	4	4	4	4	4
Mold Guard Day 20	0	0	0	0	0
Control Day 28	4	4	4	4	4
Mold Guard Day 28	0	0	0	0	0

Table 2: Observed growth ratings of samples evaluated at day 4, 12, 20 and 28 after inoculation.

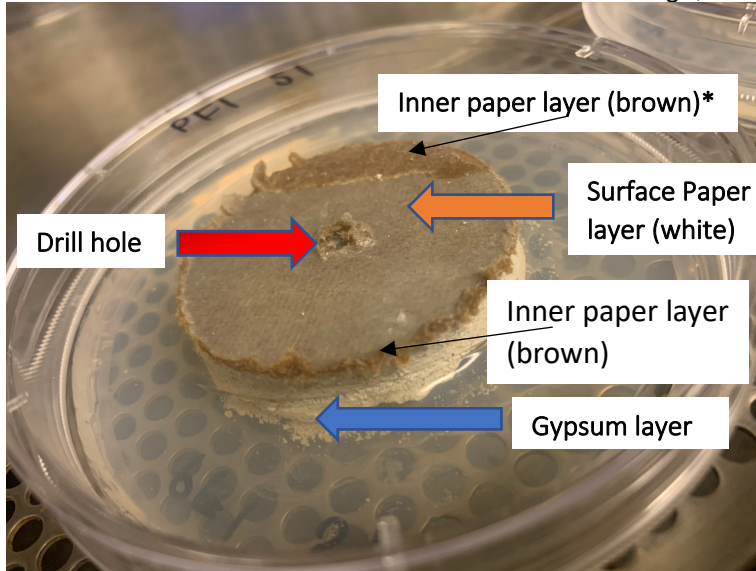


Image 1: Dry wall treated with Mold Guard on day 4, inoculated with **penicillium chrysogenum**.

The layers of the dry wall are labeled, starting from the bottom the gypsum layer, the brown inner paper layer and the white surface paper layer. During sterilization of the sample, the white surface paper layer was peeled back in some areas and exposed the brown inner paper layer, shown by the arrow labeled "inner paper layer (brown)*". The drywall sample was cut with a drill hole cutter which created the hole in the center of the sample. The hole works as an additional channel for the nutrition from the agar to penetrate the sample.

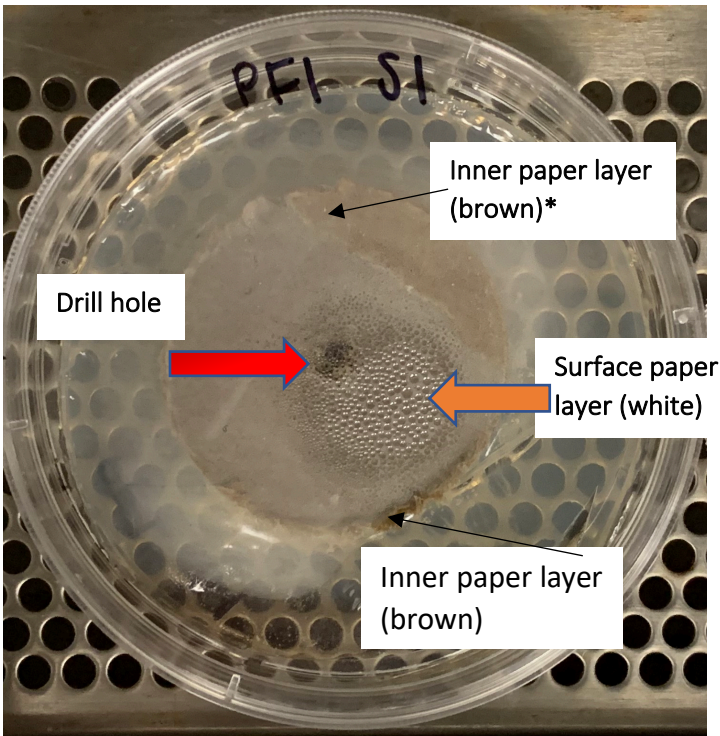


Image 2: Dry wall treated with Mold Guard on day 12, inoculated with **penicillium chrysogenum**.

Image 2 is the same sample shown in image 1. On day 12 moisture was observed, as seen in the photo. The moisture is a necessary component to maintain sufficient humidity outlined by the ASTM G 21 method "Standard Practice for Determining Resistance of Synthetic Polymeric Materials to Fungi".

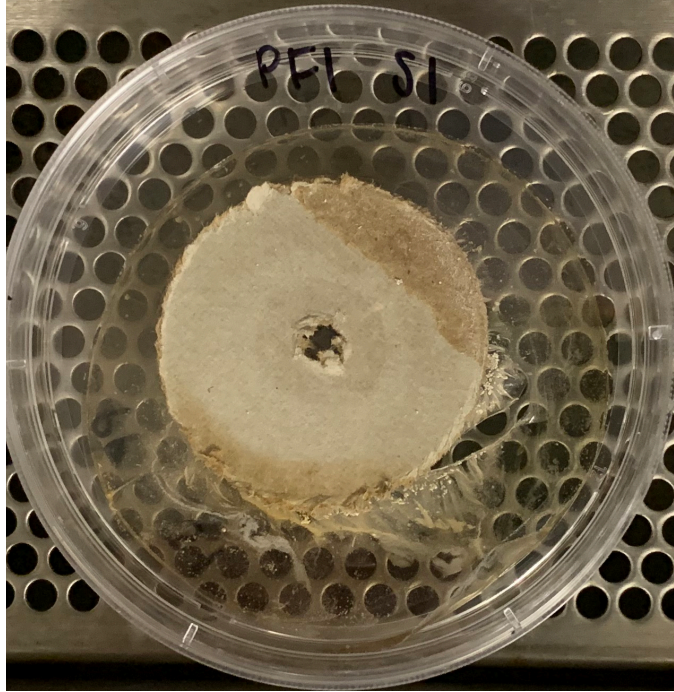


Image 3: Dry wall treated with Mold Guard on day 21, inoculated with **penicillium chrysogenum**.

Image 3 is the same sample shown in image 1 and image 2. By day 20 the moisture has decreased and no mold has grown.

Day 4	Day 12	Day 20	Day 28
Control with significant mold growth.	Control with heavy mold growth.	Control with heavy mold growth.	Control with heavy mold growth.

Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.
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Table 3: Dry wall pieces inoculated with *Penicillium chrysogenum*. The paper layer on the sample treated with Mold Guard was peeled back during the sterilization process.

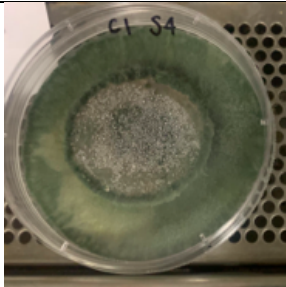
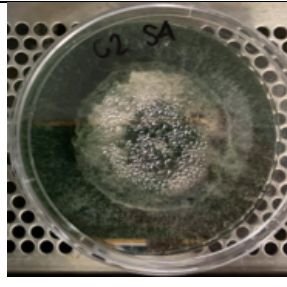
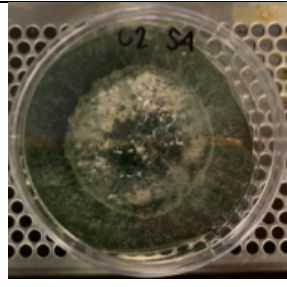
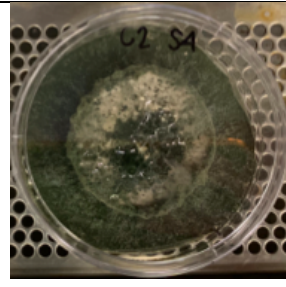
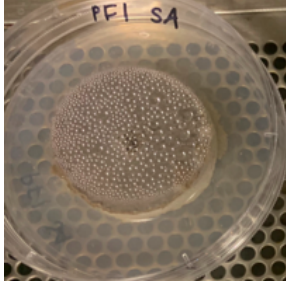
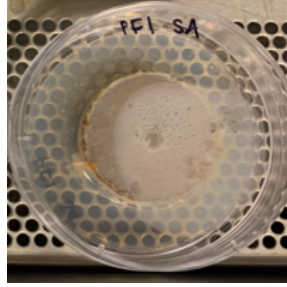
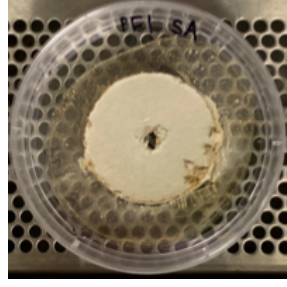
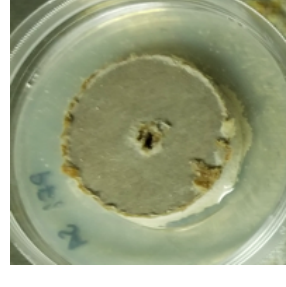
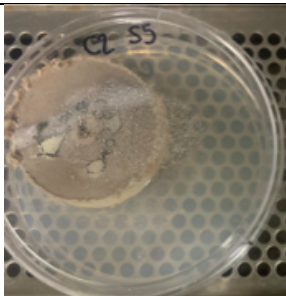
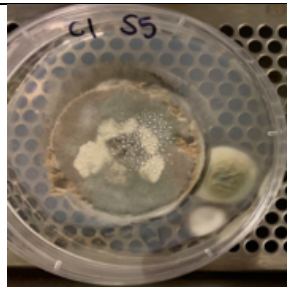
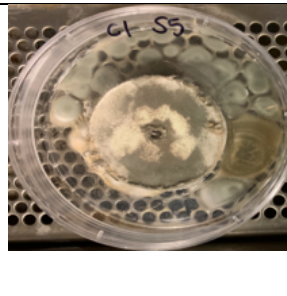
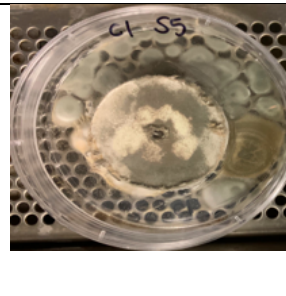
Day 4	Day 12	Day 20	Day 28
			
Control with heavy mold growth.	Control with heavy mold growth.	Control with heavy mold growth.	Control with heavy mold growth.
			
Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.

Table 4: Dry wall pieces inoculated with *Trichoderma Virens*; T-1.

Day 4	Day 12	Day 20	Day 28
			

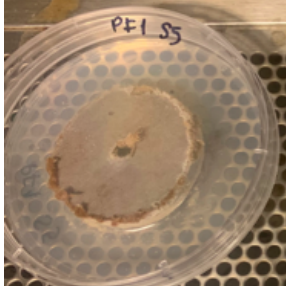
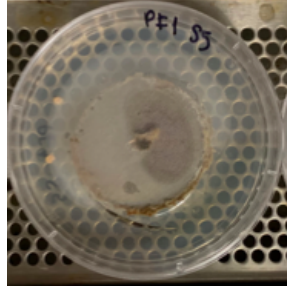
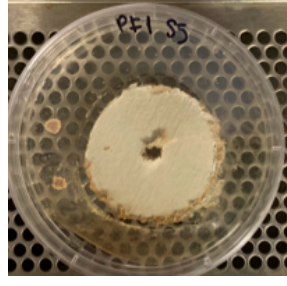
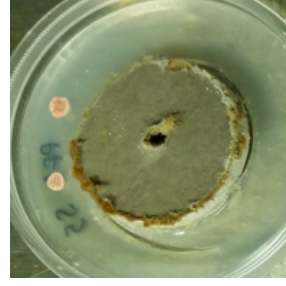
Control with significant mold growth.	Control with heavy mold growth.	Control with heavy mold growth.	Control with heavy mold growth.
			
Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.

Table 5: Dry wall pieces inoculated with *Aureobasidium pullulans*; var. *Melanigenum*; QM 279c.

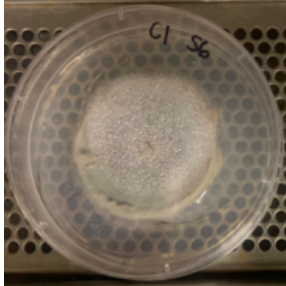
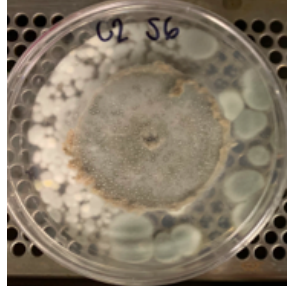
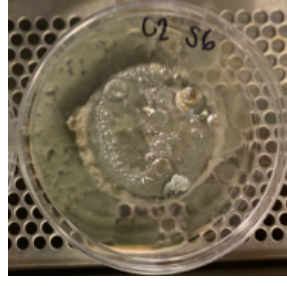
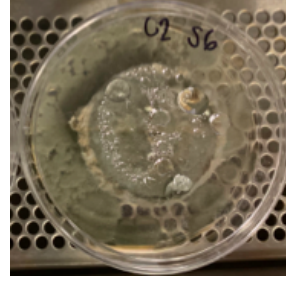
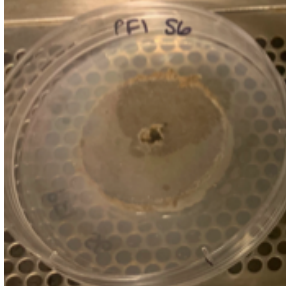
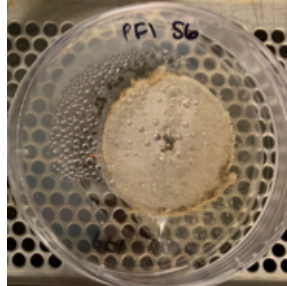
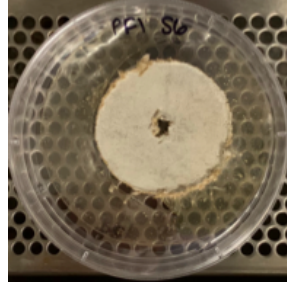
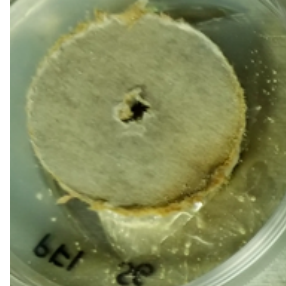
Day 4	Day 12	Day 20	Day 28
			
Control with heavy mold growth.	Control with heavy mold growth.	Control with heavy mold growth.	Control with heavy mold growth.
			
Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.

Table 6: Dry wall pieces inoculated with *Telaromyces pinophilus*.

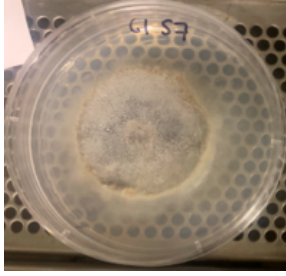
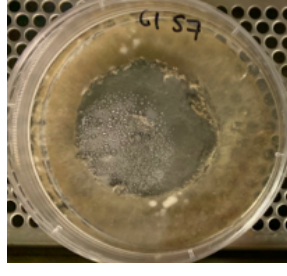
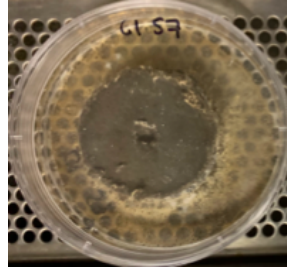
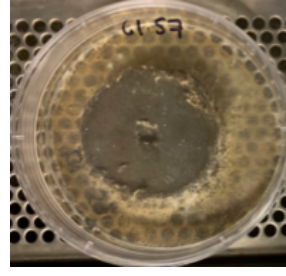
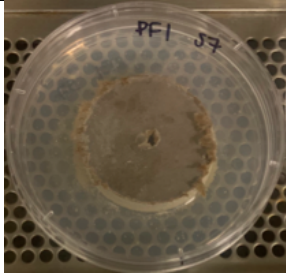
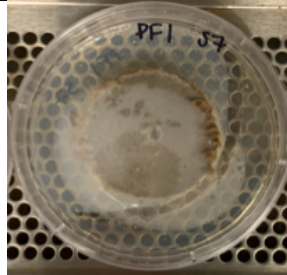
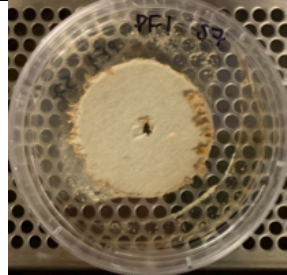
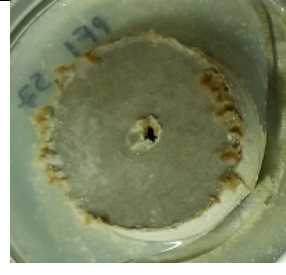
Day 4	Day 12	Day 20	Day 28
			
Control with heavy mold growth.	Control with heavy mold growth.	Control with heavy mold growth.	Control with heavy mold growth.
			
Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.	Treated with Mold Guard. No Mold Growth on dry wall.

Table 7: Dry wall pieces inoculated with *Chaetomium globosum*; QM 459.