

ANTIFUNGAL COMPOUNDS  
PRODUCED IN  
ANTAGONISTIC COMPETITION  
BETWEEN  
MARINE FUNGI

By: Muhammad Burhan Tariq



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Dr. Marc J. Orbach  
School of Plant Sciences

# ANTIFUNGAL COMPOUNDS PRODUCED IN ANTAGONISTIC COMPETITION BETWEEN MARINE FUNGI

Muhammad Tariq

## ABSTRACT

Competition among fungi has been characteristic of antibiotic relationships between microbes, leading to the discovery of novel antimicrobial compounds. This study observed such antagonistic relationships between marine fungi isolated from coral off the coast of Woods Hole, Massachusetts. Competition assays were conducted on these fungal isolates against two chosen competing species on PDA plates. Three fungal isolates were observed to release antifungal compounds inhibiting the growth of the competing species. Methanol extracts were taken from each of the three fungal isolates and  $^1\text{H}$  and  $^{13}\text{C}$  NMR spectra obtained. The three fungi were shown to produce antifungal compounds, not observed in previous studies. 14 fractions were obtained from subjecting the methanol extracts from each of the three fungi to chromatography. The final step remains to test these fractions for antifungal activity leading to the isolation and identification of the antifungal compounds.

## INTRODUCTION

Fungi have long been known for their antagonistic interactions with other microbes leading to the discovery of novel chemical agents such as penicillin. However, one of the main types of these interactions is among different species of fungi themselves. Antagonism between species of fungi plays a central role in determining the composition, organization, and pattern of succession within fungal ecosystems (3). This antagonism is a result of antibiotic competition for occupation of niche and resources vital for survival and proliferation. Therefore, the antibiotic chemical agents produced should be dependent upon the competing species and the particular environmental niche. The goal of this project was to study fungal antibiotic competition in understudied marine fungi isolated from a distinctive environment,

leading to the discovery of new antifungal compounds.

Fungal isolates from the coral *Astrangia* off the shore of Woods Hole, Massachusetts were obtained by Dr. M. A. Mandel, O. Yarden, and M. J. Orbach during the summer of 2009 when they conducted research at the Marine Biological Laboratory in Woods Hole. These isolates were ideal subjects for this study due to the fact that fungi from this environment are understudied and arise in an ecological niche very distinct from that of previously studied terrestrial fungal competition (9). These isolates were assayed for competition against two chosen competing species and changes in growth rate were measured. Furthermore, those fungal isolates exhibiting signs of antibiotic competition over a distance were used to attempt to isolate and identify the antifungal compounds (1).

## METHODS AND MATERIALS

### *Identification of Fungal Isolates*

The fungal isolates used in this study had their rDNA ITS regions as well as part of the 28S rDNA sequenced. Preliminary assignment of species was done based on the top ten BLAST hits for each isolate (5).

### *Competition Bioassays*

Eleven fungal isolates were grown on Potato Dextrose Agar (PDA) for one week. The fungal isolates were 7D2, 13B4, 24C4, 1B5, 29A6, 23C2, 21B3, 21C2, 9D5, 25A8, and 10B1. From these cultures, 1 mm plugs were used to inoculate the competition assay plates. The competition assays were performed on PDA plates with two media plugs taken out 2 cm from the edge of the plate and 4.5 cm from each other. One of these spaces was inoculated with one of the coral fungal isolate plugs and the other was inoculated with a plug of the competing species. Two competing species were used in this study, *Colletotrichum trifolii* (CT) and *Cladosporium cladosporioides* (CC). These fungi grew at significantly different rates. They were chosen based on data that these species or related species were found in the coral environment and the different growth rates factored out rate of growth as a variable. Two repetitions were conducted for each competition for a total of four competition assays per fungal isolate. Control plates were set up in the same manner, except with no competing species. All inoculations were performed in a laminar flow hood to avoid contamination of the plates.

Selected isolates that showed signs of inhibition were re-assayed with three additional replicates with controls on 30 ml PDA plates.

Growth measurements were made in three parameters of length and width (Figure 4.1). The first parameter labeled A was a measurement of the height of the fungus, a vertical length at the initial inoculation site from the top edge to the bottom edge of the fungus. The second parameter labeled B was a measurement of the inside width of the fungus, a horizontal length from the initial inoculation site to the edge of the fungal colony growing towards the center of the plate. The third parameter labeled C was a measurement of the outside width of the fungus, a horizontal length from the initial inoculation site to the edge of the colony growing towards the outer edge of the petri dish. These measurements were taken for each fungal isolate and each competing species.

### *Methanol Extraction*

Fungal isolates found to exhibit antibiotic activity were extracted with methanol for isolation of the antifungal compounds (2). The extraction was performed following growth of the fungal isolates on PDA plates for one week, with three repetitions for each isolate. To each plate, 10 ml of 100% methanol was added and incubated for five minutes. Next, the agar was cut into small cubes, approximately 0.5 cm in length, using a scalpel. These cubes and the methanol were transferred to a 250 ml Erlenmeyer flask. To each flask, 45 ml of additional 100% methanol was added. The flasks were shaken for 1.5

hours and the methanol extract was collected using vacuum filtration with Whatman No. 1 filter paper to remove the agar and fungal materials. The filtrate was washed with 10 ml of additional methanol.

#### *Chemical Assays<sup>1</sup>*

The methanol extracts were used to obtain whole fraction <sup>1</sup>H NMRs by evaporating the methanol and resolating in MeOD. Furthermore, <sup>13</sup>C NMRs were obtained as well (2). Then about half the sample was taken and subjected to silica gel chromatography with select fractions used for obtaining NMR data. Finally, selected compounds are to be identified and characterized with melting point, spectroscopy, and their antifungal activity is to be tested.

#### *Analysis of Antagonistic Components*

Fractions from the chromatographed extracts are to be used to measure antifungal activity by placing select amounts on Whatman No. 1 filter paper and introducing this to plates containing the competing species; inhibition of compounds will be assessed by observing and measuring the growth rate of the fungus around the filter paper in comparison to a control. Finally, the minimum inhibitory concentration (MIC) is to be determined (1).

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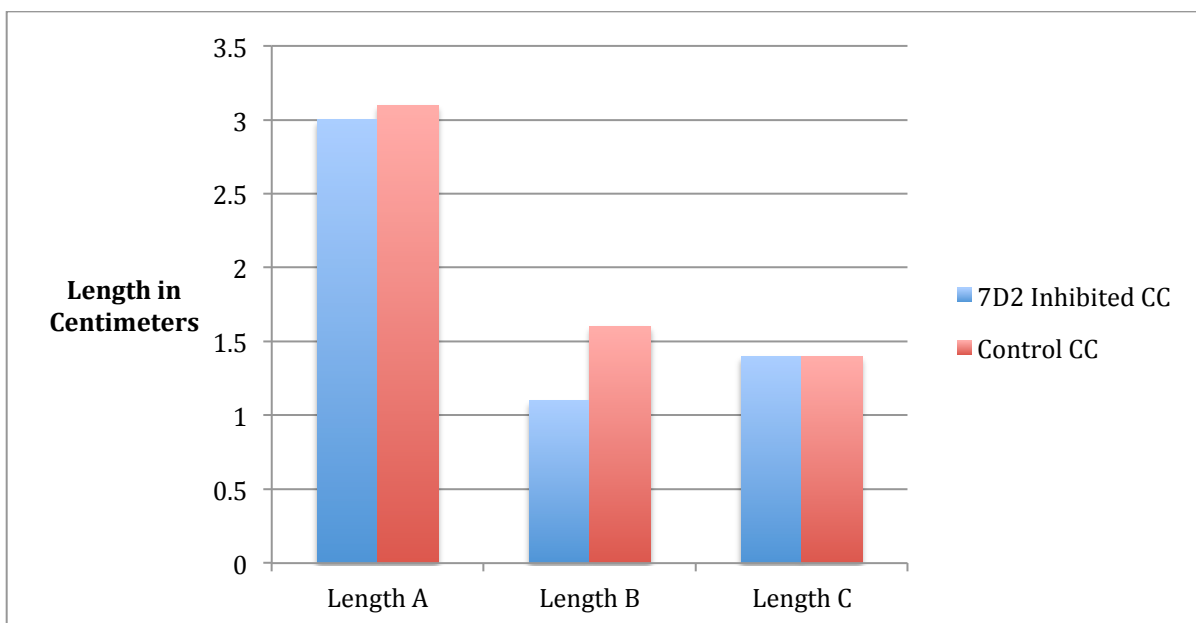
<sup>1</sup> Conducted by Dr. Robert Bates from the Department of Chemistry and Biochemistry, University of Arizona

## RESULTS

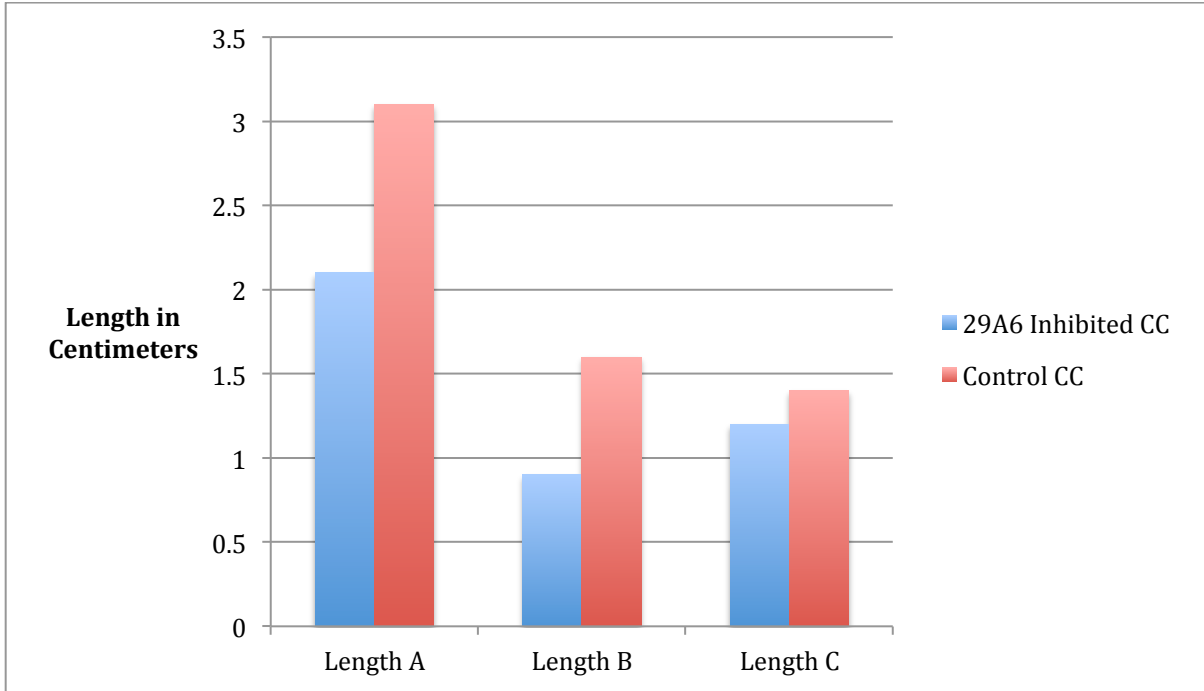
Three fungal isolates (7D2, 29A6, and 10B1) exhibited signs of antibiotic interactions (Figure 5.1), defined as a decrease in growth by at least 25% in at least one dimension of the competing species as compared to the controls. The top BLAST hits for these fungi were *Nectria haematococca* (7D2), *Penicillium dipodomyicola* (29A6), and *Penicillium atrovenetum* (10B1). The fungal isolate 7D2 released an antifungal compound into the media, which caused hyphal damage. The competing species *C. trifolii* was observed to show a blackening of the interior-facing circumference of the fungus. The fungal

isolate 29A6 had a cloud of halo, decreasing the clarity of the media around its circumference that projected outwards. This was visible after the second day of growth (Figure 5.2). The fungal isolate 10B1 resulted in a scarlet pigmentation of the media in competition with the competing species *C. cladosporioides*. The NMR of 29A6 indicated high levels of citrate (doublets at around 2.8 and 2.9 ppm), while the NMR of the other two isolates indicate high levels of glucose in the extracts. The raw data for the growth measurements, NMR spectra, and chromatography data are presented in Appendix A and B respectively.

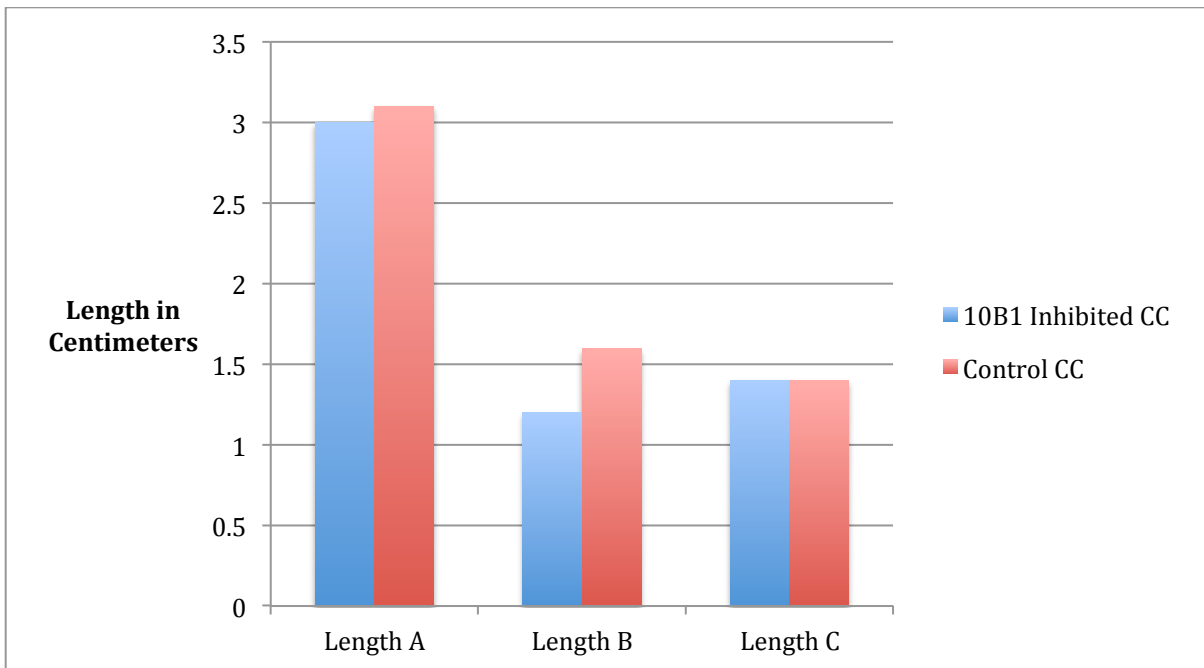
**Figure 1.1:** Comparison of control and 7D2 inhibited growth dimensions of *C. cladosporioides* (CC) at 7 days.



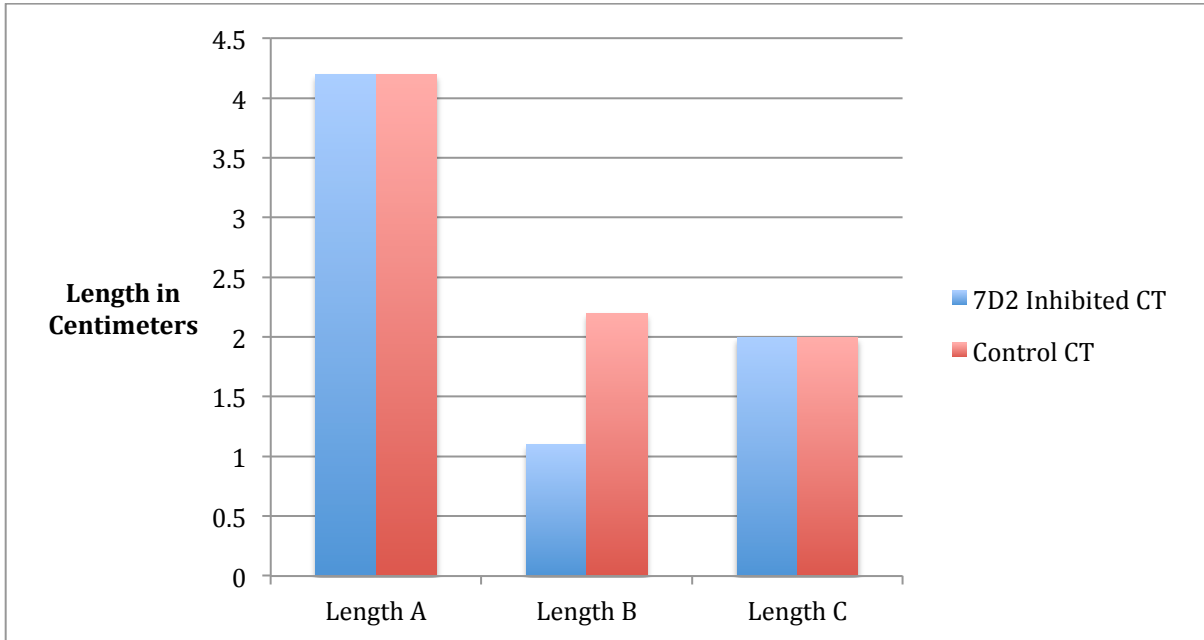
**Figure 1.2:** Comparison of control and 29A6 inhibited growth dimensions of *C. cladosporioides* (CC) at 7 days.



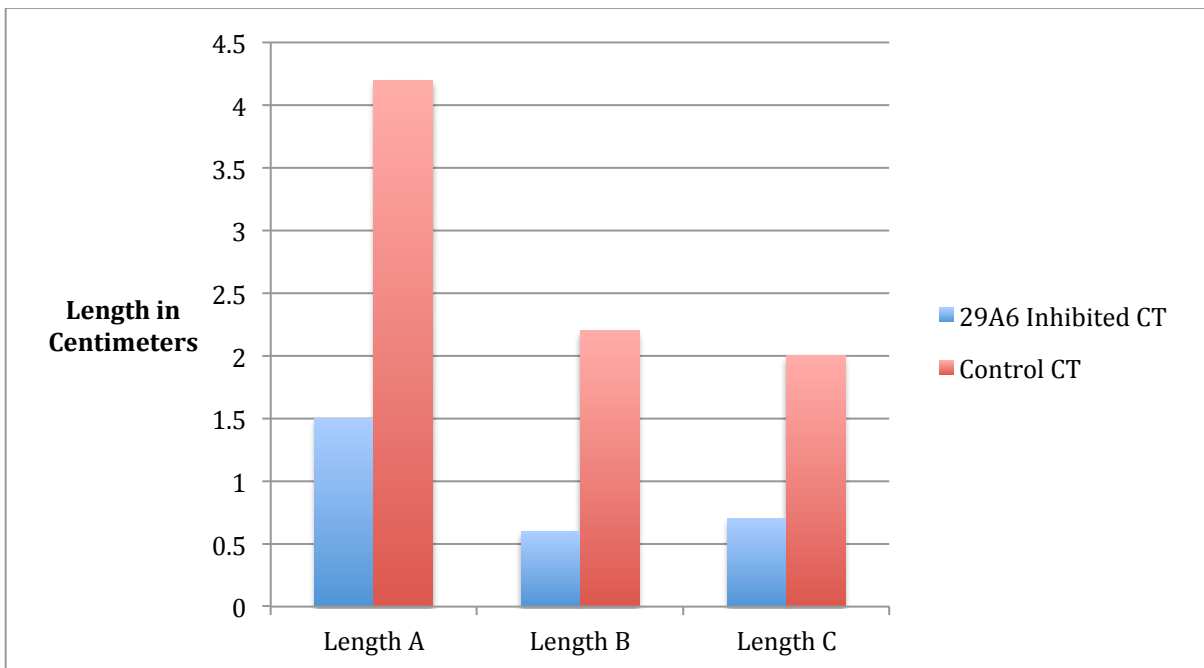
**Figure 1.3:** Comparison of control and 10B1 inhibited growth dimensions of *C. cladosporioides* (CC) at 7 days.



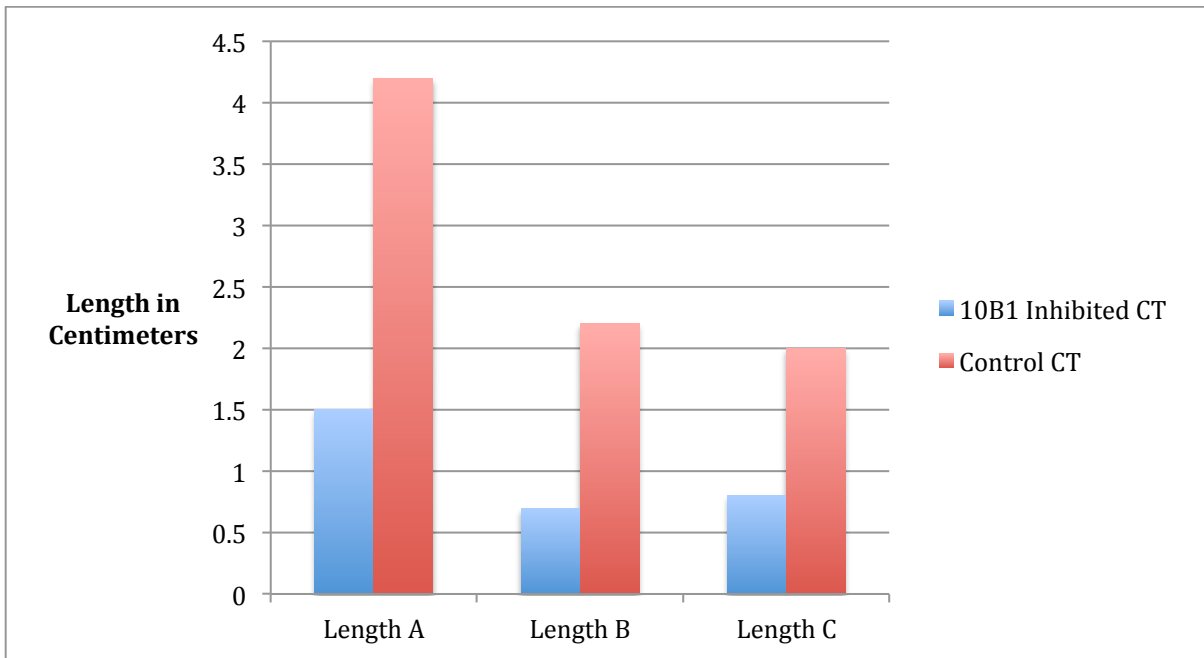
**Figure 2.1:** Comparison of control and 7D2 inhibited growth dimensions of *C. trifolii* (CT) at 7 days.



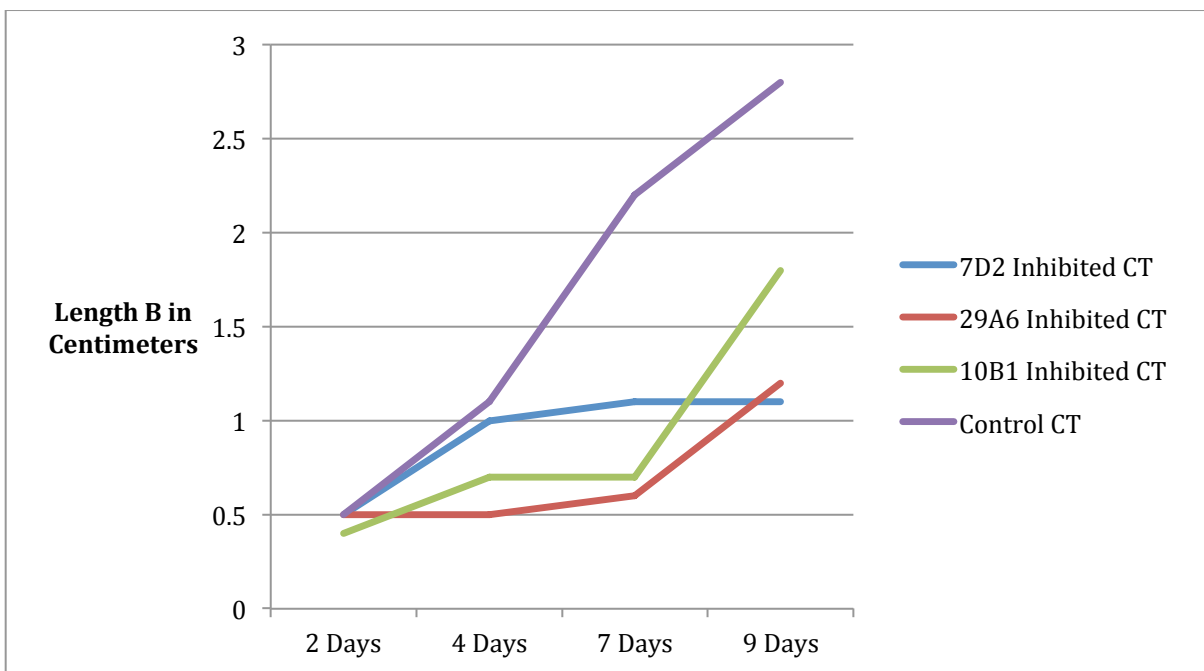
**Figure 2.2:** Comparison of control and 29A6 inhibited growth dimensions of *C. trifolii* (CT) at 7 days.



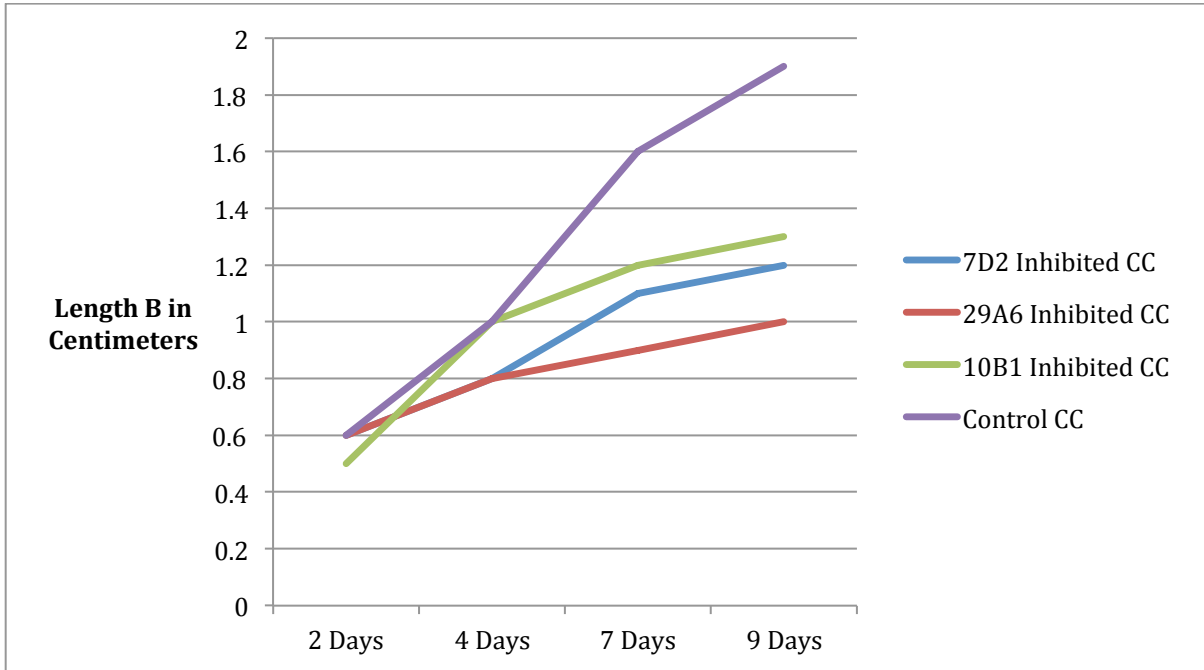
**Figure 2.3:** Comparison of control and 10B1 inhibited growth dimensions of *C. trifolii* (CT) at 7 days.



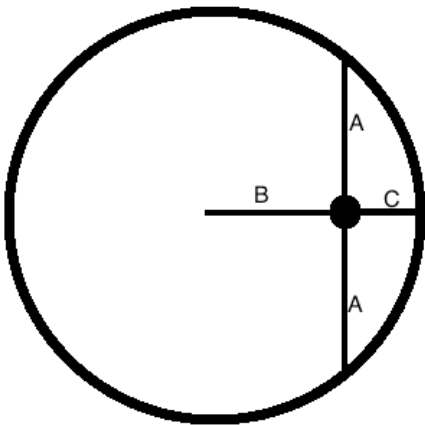
**Figure 3.1:** Comparison of control and fungal isolate inhibited growth of *C. trifolii* (CT) over the competition time period.



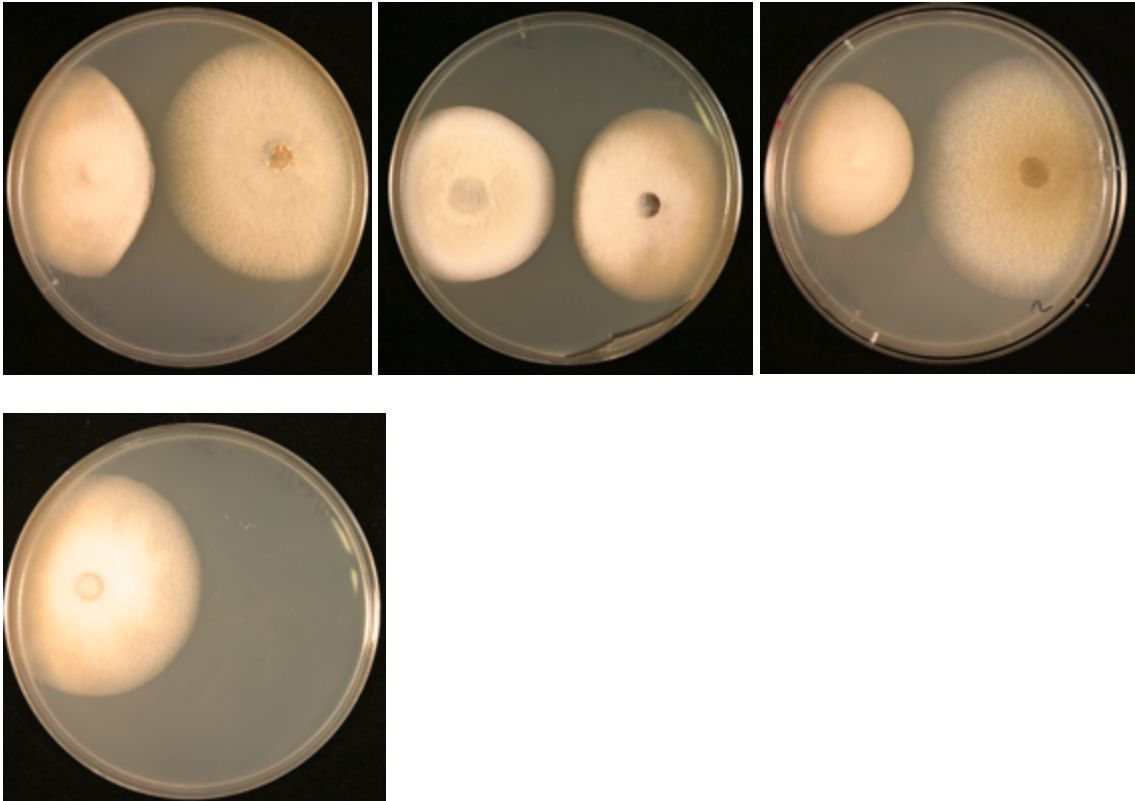
**Figure 3.2:** Comparison of control and fungal isolate inhibited growth of *C. cladosporioides* (CC) over the competition time period.



**Figure 4.1:** A schematic showing the three length and width dimensions of fungal growth measurement (A, B, and C).



**Figure 5.1:** Photographs of the competition plates at 7 days. From left to right, 7D2, 10B1, and 29A6 are seen competing against *C. trifolii*. The competing species, *C. trifolii*, is seen on the left in the picture and the fungal isolate is on the right. The last picture in the second row is the control of *C. trifolii* (CT).



**Figure 5.2:** Photograph of 29A6 at 2 days showing the cloud of halo.



## DISCUSSION

Three (7D2, 10B1, 29A6) of the eleven fungal isolates (7D2, 13B4, 24C4, 1B5, 29A6, 23C2, 21B3, 21C2, 9D5, 25A8, 10B1) exhibited signs of antibiosis over a distance against the two competing species. All tested isolates either showed antibiosis by growth inhibition in both competing species or did not affect either species. No tested isolates showed antibiotic relationships selective for just one of the competing species. The study by Suzuki *et al* discovered the antifungal compound Haematocin from a terrestrial isolate of the fungus *Nectria haematococca* similar to the coral isolate 7D2. The study by Dombrink-Kurtzmann *et al*, described the release of patulin from the fungus *Penicillium dipodomyicola* (similar to strain 29A6) and the study by Porter *et al*, showed that *Penicillium atrovenetum* (similar to strain 10B1) released 3-nitropropionate. However, our study found none of these compounds present in the NMR spectra taken of the methanol extracts from the marine isolates 7D2, 29A6, and 10B1. This indicates that the antifungal compounds released by the three isolates are novel compounds as compared to the compounds released by those three terrestrial species.

As figures 3.1 and 3.2 indicate, the growths of the competing species diverge and are most significantly inhibited starting from the fourth day of competition. Furthermore, figures 1.1-2.3 show that 7D2 inhibits growth primarily in length B, while 29A6 and 10B1 inhibit growth in all three measured lengths. This indicates that

7D2 releases its antifungal compound later than the other two isolates, because the other two dimensions (A and C) are occupied by the fungus early in growth and the edge of the plate is reached. Furthermore, this can be explained by differing diffusional and release patterns of the antifungal compounds in the three isolates. Although the majority of inhibition is apparent after the fourth day, figures 3.1 and 3.2 show that some inhibition is present very early in the 29A6 and 10B1 competition assays. This is most apparent in figure 3.1, in which it is indicated that 29A6 releases its antifungal compounds even before 10B1.

The competition assay for 29A6 had a cloud of disfiguration in the clarity of the media, most likely due to the release of the antifungal compound, or precipitation of something in the media. This was further supported by the observation that inhibition of the competing species was most prominent when this cloud of haze made contact with the competing fungus. Furthermore, the scarlet pigmentation present in the 10B1 competition assay against *C. cladosporioides* is most likely due to the combined effect of compounds released by both fungi. This is supported by the observation that the pigmentation is not present in the competition assay against *C. trifolii* or in any of the controls.

The NMR spectrum of 29A6 indicates a citrate product. However citrate is a normal product of fungal metabolism and is not likely to be the antifungal compound. However, a derivative of citrate could be showing the NMR

results and be the antifungal compound. In the NMR spectra of the other two isolates, high levels of glucose were observed. This might be expected because glucose (dextrose is a term used to describe glucose) is a major component of the growth media. The low concentrations of other compounds may suggest that the antifungal compounds produced are very potent by causing such a strong inhibition of growth in the competing species.

A limitation to this study was the small number of fungal isolates used to represent a diverse marine ecosystem. This restricts the observation of a fraction of fungal isolates, which exhibit antibiotic relationships. Furthermore, the use of a limited number of competing species restricts the number of specific

antagonistic relationships that could be observed. Additionally, the lack of a complete species identification on the three fungal isolates exhibiting antifungal activity is a limitation to the study.

The future goals of this study are the testing of the isolated fractions found in Appendix B to the antifungal compound produced by each fungal isolate. After identification, the isolated antifungal compounds will be analyzed to determine their potential utility as anti-fungal, or anti-microbial drugs.

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## APPENDIX A

*C- Contact with other fungus*

*CE- Contact with edge of plate*

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
7D2	1.4	0.7	0.6	0.9	0.5	0.4	1.2	0.5	0.6	2
	1.4	0.5	0.4	0.9	0.5	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
13B4	1.1	0.5	0.4	1	0.7	0.5	1.2	0.5	0.6	2
	1.2	0.5	0.8	1.1	0.6	0.6				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
24C4	1.3	0.7	0.8	1.1	0.5	0.5	1.2	0.6	0.8	2
	1.2	0.7	0.7	1	0.5	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
1B5	3	1.5	1.2	0.8	0.4	0.4	2.5	1.5	1	2
	2.8	1.8	1.2	0.8	0.4	0.4				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
29A6	2	1	0.5	1	0.5	0.4	1.5	0.8	0.6	2
	2	0.5	1.1	0.9	0.5	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
23C2	1	0.6	0.5	1	0.4	0.5	1	0.5	0.5	2
	1.1	0.5	0.5	0.9	0.4	0.4				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21B3	4	1.7	2.3	0.9	0.5	0.4	3.8	2	1.8	2
	3.5	2	1.6	1.1	0.5	0.3				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21C2	3	1.5	1.6	0.9	0.5	0.4	3	1.7	1.5	2
	2.9	1.5	1.7	0.9	0.6	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
9D5	1.5	0.8	0.7	0.9	0.4	0.5	1.5	0.7	0.8	2
	1.4	0.7	0.7	0.8	0.5	0.4				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
25A8	1.1	0.5	0.6	1	0.4	0.6	1.1	0.5	0.4	2
	1	0.5	0.5	1.2	0.5	0.4				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
10B1	1.2	0.5	0.8	0.9	0.4	0.5	1	0.5	0.5	2
	0.9	0.4	0.6	0.9	0.4	0.5				

Competing Species Control	Length A (cm)	Length B (cm)	Length C (cm)	Age of Culture in Days
CT	0.9	0.5	0.5	2

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
7D2	2.8	1.8	1.3	2.2	1	1	2.5	1.2	1.1	4
	2.9	1.5	1.4	2.5	1	1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
13B4	2.1	1.1	1	2.4	1.1	1	2.1	1.1	0.9	4
	2	1	0.7	2.3	1.4	1.2				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
24C4	2.4	1.3	1.1	2.1	1.2	1	2.7	1.5	1.2	4
	2.5	1.2	1.3	2.1	1.2	1.1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
1B5	7/CE	4.8	2/CE	2	1	0.8	7/CE	5	2/CE	4
	7/CE	5.2	2/CE	1.8	0.7	1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
29A6	3	1.5	1.4	2	1.1	1.1	3	2	1.4	4
	3	1.3	1	1.3	0.5	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
23C2	2.4	1.1	1.1	2.1	1.4	1.1	2.4	1	1	4
	2.1	1	0.9	2.2	1.1	1.1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21B3	7/CE	6.5	2/CE	1.8	0.6	0.9	7/CE	6.5	2/CE	4
	7/CE	6.5	2/CE	1.9	0.6	1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21C2	7/CE	5	2/CE	2	1	1	7/CE	5.3	2/CE	4
	7/CE	5.2	2/CE	2.1	0.7	1.1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
9D5	2.9	1.5	1.5	2.1	1.2	1.1	3	1.5	1.5	4
	3	1.5	1.5	2.2	1.2	1.1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
25A8	1.8	0.8	0.8	2.3	1.2	1.3	1.8	0.8	0.9	4
	1.8	0.7	0.9	2.4	1.2	1.1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
10B1	2	1	1	1.8	0.8	1	1.9	0.8	1	4
	2	0.8	11.6	1.6	0.7	0.7				

Competing Species Control	Length A (cm)	Length B (cm)	Length C (cm)	Age of Culture in Days
CT	2.1	1.1	1	4

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
7D2	5.1	2.6	2/CE	4.4	1.1	2/CE	5	2.5	2/CE	7
	5.2	2.7	2/CE	4.4	1.1	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
13B4	3.3	1.7	1.7	4.4	2	2/CE	3.5	1.8	1.8	7
	3.3	1.7	1.5	4.9	2.2	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
24C4	4.2	2	2/CE	4.1	2.1	2/CE	4.4	2.3	2/CE	7
	4.2	2	2/CE	4.2	2.3	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
1B5	7/CE	6.5	2/CE	2.4	2.1	2/CE	7/CE	6.5	2/CE	7
	7/CE	6.5	2/CE	2	2	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
29A6	3	2.5	2/CE	2	1.3	1.5	3.5	2	2/CE	7
	2.5	2	1.5	1.5	0.6	0.7				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
23C2	3.5	2	1.8	4	2.1	2	4	1.8	1.9	7
	3	2.2	2/CE	4.5	2	2				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21B3	7/CE	6.5	2/CE	1.7	C	1.5	7/CE	6.5	2/CE	7
	7/CE	6.5	2/CE	1.7	C	C				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21C2	7/CE	6.5	2/CE	2	C	1	7/CE	6.5	2/CE	7
	7/CE	6.5	2/CE	2	C	C				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
9D5	5.1	2	2/CE	4.3	2.1	2/CE	5.3	2.6	2/CE	7
	5.2	2.2	2/CE	4.3	2	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
25A8	2.7	1.8	1.9	4.4	1.8	1.8	2.6	1.8	1.9	7
	2.4	1.8	1.9	4.4	1.8	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
10B1	2.5	1.4	1.3	2	1	1	2.5	1.5	1.5	7
	2.3	1	2	1.5	0.7	0.8				

Competing Species Control	Length A (cm)	Length B (cm)	Length C (cm)	Age of Culture in Days
CT	4.2	2.2	2/CE	7

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
7D2	6.5	3.4	2/CE	4.9	1.1	2/CE	6.5	3.4	2/CE	9
	6.6	3.3	2/CE	4.8	1.1	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
13B4	4	2	1.9	5.5	2.2	2/CE	4.3	2.2	1.9	9
	4	1.8	1.7	5.5	2.2	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
24C4	5.3	2.1	2/CE	5.1	2.1	2/CE	5.1	2.7	2/CE	9
	5.2	2.1	2/CE	5.2	2.3	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
1B5	7/CE	6.5/CE	2/CE	2.4	C	1.2	7/CE	6.5/CE	2/CE	9
	7/CE	6.5/CE	2/CE	1.7	C	1.1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
29A6	7/CE	4.4	2/CE	3.9	1.2	2/CE	7/CE	4.3	2/CE	9
	7/CE	4.2	2/CE	3.8	1.2	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
23C2	5.4	2.3	2/CE	6	2.3	2/CE	5.3	2.5	2/CE	9
	5.2	2.1	2/CE	6	2.2	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21B3	7/CE	6.5/CE	2/CE	1.7	C	1	7/CE	6.5/CE	2/CE	9
	7/CE	6.5/CE	2/CE	1.8	C	0.9				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21C2	7/CE	6.5/CE	2/CE	1.9	C	1.2	7/CE	6.5/CE	2/CE	9
	7/CE	6.5/CE	2/CE	2	C	1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
9D5	5.8	2.2	2/CE	5.3	2	2/CE	6.1	3	2/CE	9
	6	2.1	2/CE	5.3	2	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
25A8	3.4	1.5	1.6	5.5	2	2/CE	3.2	1.6	1.6	9
	3.3	1.4	1.7	5.4	2.1	2/CE				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CT Length A (cm)	CT Length B (cm)	CT Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
10B1	3.7	2.2	2/CE	5.5	1.7	2/CE	5.1	2.6	2/CE	9
	4.6	2.3	2/CE	5.6	1.8	2/CE				

Competing Species Control	Length A (cm)	Length B (cm)	Length C (cm)	Age of Culture in Days
CT	5.4	2.8	2/CE	9

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
7D2	1.4	0.6	0.7	1.2	0.6	0.6	1.2	0.5	0.5	2
	1.6	0.7	0.8	1.1	0.6	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
13B4	1.3	0.7	0.7	1.1	0.6	0.5	1.2	0.5	0.6	2
	1.4	0.7	0.7	1	0.6	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
24C4	1.5	0.8	0.7	1.1	0.7	0.5	1.2	0.6	0.8	2
	1.7	0.8	0.9	1.1	0.5	0.7				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
1B5	3.4	2	1.8	1	0.6	0.4	2.5	1.5	1	2
	3.5	1.7	2/CE	1	0.6	0.4				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
29A6	2.1	1.1	1.1	1.1	0.6	0.5	1.5	0.8	0.5	2
	2.1	1	1	1	0.5	0.4				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
23C2	1.5	0.7	0.7	1.1	0.6	0.5	1	0.5	0.5	2
	1.5	0.7	0.8	1.1	0.6	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21B3	5.5	2.6	2/CE	1.1	0.6	0.5	3.8	2	1.8	2
	5.5	2.7	2	1.1	0.6	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21C2	5	2.5	2/CE	1.1	0.6	0.5	3	1.7	1.5	2
	5	2.7	2/CE	1.1	0.6	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
9D5	1.8	0.8	0.9	0.9	0.5	0.4	1.5	0.7	0.8	2
	1.7	0.9	0.8	1.1	0.6	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
25A8	1.2	0.6	0.7	1.1	0.5	0.4	1.1	0.4	0.5	2
	1	0.6	0.7	1.1	0.6	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
10B1	1.4	0.6	0.9	1.1	0.6	0.5	1	0.5	0.5	2
	1.4	0.8	0.7	1.1	0.5	0.4				

Competing Species Control	Length A (cm)	Length B (cm)	Length C (cm)	Age of Culture in Days
CC	1.1	0.6	0.5	2

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
7D2	3.1	1.6	1.6	1.7	0.8	0.9	2.5	1.1	1.2	4
	3	1.4	1.7	1.9	1	0.7				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
13B4	2.3	1.1	1	1.7	1	0.7	2.1	1	0.9	
	2.2	1.1	1.2	1.8	1	0.9				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
24C4	3	1.5	1.7	1.7	0.9	1	2.7	1.5	1.2	4
	2.7	1.5	1.4	2	1.2	1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
1B5	5.4	2	2/CE	1.6	0.9	0.8	7/CE	3	2	4
	5.4	2	2/CE	1.6	0.7	0.8				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
29A6	4.2	2	2/CE	1.7	0.8	0.8	3	2	1.5	4
	4.2	2.2	2/CE	1.8	0.9	0.7				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
23C2	2.6	1.5	1.5	1.6	0.7	0.8	2.4	1	1	4
	2.5	1.4	1.4	2	1	0.9				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21B3	7/CE	6.5/CE	2/CE	1.1	0.6	0.5	7/CE	6.5/CE	2/CE	4
	7/CE	6.5/CE	2/CE	1.2	0.6	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21C2	7/CE	6.5/CE	2/CE	1.3	0.7	0.7	7/CE	6.5/CE	2/CE	4
	7/CE	6.5/CE	2/CE	1.2	0.8	0.6				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
9D5	3.1	1.7	1.6	1.6	0.9	0.8	3	1.5	1.5	4
	3.3	1.7	1.6	1.6	1	0.6				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
25A8	1.8	0.8	1	1.8	1	0.7	1.8	0.8	0.9	4
	2	1.1	1	1.8	0.9	0.7				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
10B1	2.2	1	1.2	1.8	1	0.8	1.9	0.8	1	4
	2.5	1	1.4	1.8	1	0.9				

Competing Species Control	Length A (cm)	Length B (cm)	Length C (cm)	Age of Culture in Days
CC	2	1	0.8	4

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
7D2	5.5	2.5	2/CE	3	1.2	1.5	5.1	2.5	2/CE	7
	5.6	2.8	2/CE	3	1.1	1.4				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
13B4	3.5	1.7	1.7	3	1.5	1.5	3.5	1.8	1.8	7
	3.6	1.8	1.7	2.8	1.5	1.3				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
24C4	4.5	2.2	2/CE	3	1.8	1.5	4.4	2.3	2/CE	7
	4.7	2.3	2/CE	2.9	1.5	1.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
1B5	7/CE	6.5/CE	2/CE	1.5	0.5	0.9	7/CE	6.5/CE	2/CE	7
	7/CE	6.5/CE	2/CE	1.6	0.7	1.1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
29A6	7/CE	3.5	2/CE	2	1	1.4	3.5	2	2/CE	7
	7/CE	3.5	2/CE	2.1	0.9	1.2				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
23C2	4.3	2.2	2/CE	3	1.4	1.5	4	1.8	1.9	7
	4.1	2.1	2/CE	3	1.5	1.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21B3	7/CE	6.5/CE	2/CE	1.1	C	0.5	7/CE	6.5/CE	2/CE	7
	7/CE	6.5/CE	2/CE	1.1	C	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21C2	7/CE	6.5/CE	2/CE	1.1	C	0.5	7/CE	6.5/CE	2/CE	7
	7/CE	6.5/CE	2/CE	1.1	C	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
9D5	5.5	2.8	2/CE	2.7	1.5	1.1	5.3	2.6	2/CE	7
	5.4	2.8	2/CE	2.9	1.5	1.4				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
25A8	3	1.5	1.4	2.9	1.5	1.5	2.6	1.9	1.8	7
	2.9	1.3	1.5	3	1.4	1.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
10B1	3.4	1.4	1.8	3	1.2	1.4	2.5	1.5	1.5	7
	3	1.3	2/CE	3.1	1.4	1.3				

Competing Species Control	Length A (cm)	Length B (cm)	Length C (cm)	Age of Culture in Days
CC	3.1	1.6	1.4	7

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
7D2	7/CE	3.6	2/CE	3.5	1.2	1.6	6.5	3.3	2/CE	9
	7/CE	3.4	2/CE	3.6	1.2	1.6				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
13B4	4.5	2.3	2/CE	3.6	1.7	1.7	4.3	2.2	1.9	9
	4.5	2.4	2/CE	3.7	1.8	1.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
24C4	5.4	2.5	2/CE	3.1	1.7	1.8	5.1	2.7	2/CE	9
	5.1	2.4	2/CE	3.7	1.9	1.9				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
1B5	7/CE	6.5/CE	2/CE	1.5	C	0.9	7/CE	6.5/CE	2/CE	9
	7/CE	6.5/CE	2/CE	1.6	C	1.1				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
29A6	7/CE	4.2	2/CE	2.2	C	1.4	7/CE	4.2	2/CE	9
	7/CE	4.1	2/CE	2.1	C	1.3				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
23C2	5.1	2.4	2/CE	4	1.4	1.6	5.3	2.5	2/CE	9
	5.2	2.5	2/CE	4	1.7	1.6				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21B3	7/CE	6.5/CE	2/CE	1.1	C	0.5	7/CE	6.5/CE	2/CE	9
	7/CE	6.5/CE	2/CE	1.1	C	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
21C2	7/CE	6.5/CE	2/CE	1.1	C	0.5	7/CE	6.5/CE	2/CE	9
	7/CE	6.5/CE	2/CE	1.1	C	0.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
9D5	6.4	3	2/CE	3.6	C	1.6	6.1	3	2/CE	9
	6.3	3.1	2/CE	3.6	C	1.5				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
25A8	3.2	1.4	1.6	3	1.8	1.9	3.2	1.5	1.6	9
	3.4	1.8	1.9	3.2	1.8	1.9				

Fungal Isolate	Length A (cm)	Length B (cm)	Length C (cm)	CC Length A (cm)	CC Length B (cm)	CC Length C (cm)	Control Length A (cm)	Control Length B (cm)	Control Length C (cm)	Age of Culture in Days
10B1	3.5	1.7	2/CE	3.8	1.5	1.7	4.1	2.6	2/CE	9
	3.7	1.6	2/CE	3.7	1.4	2/CE				

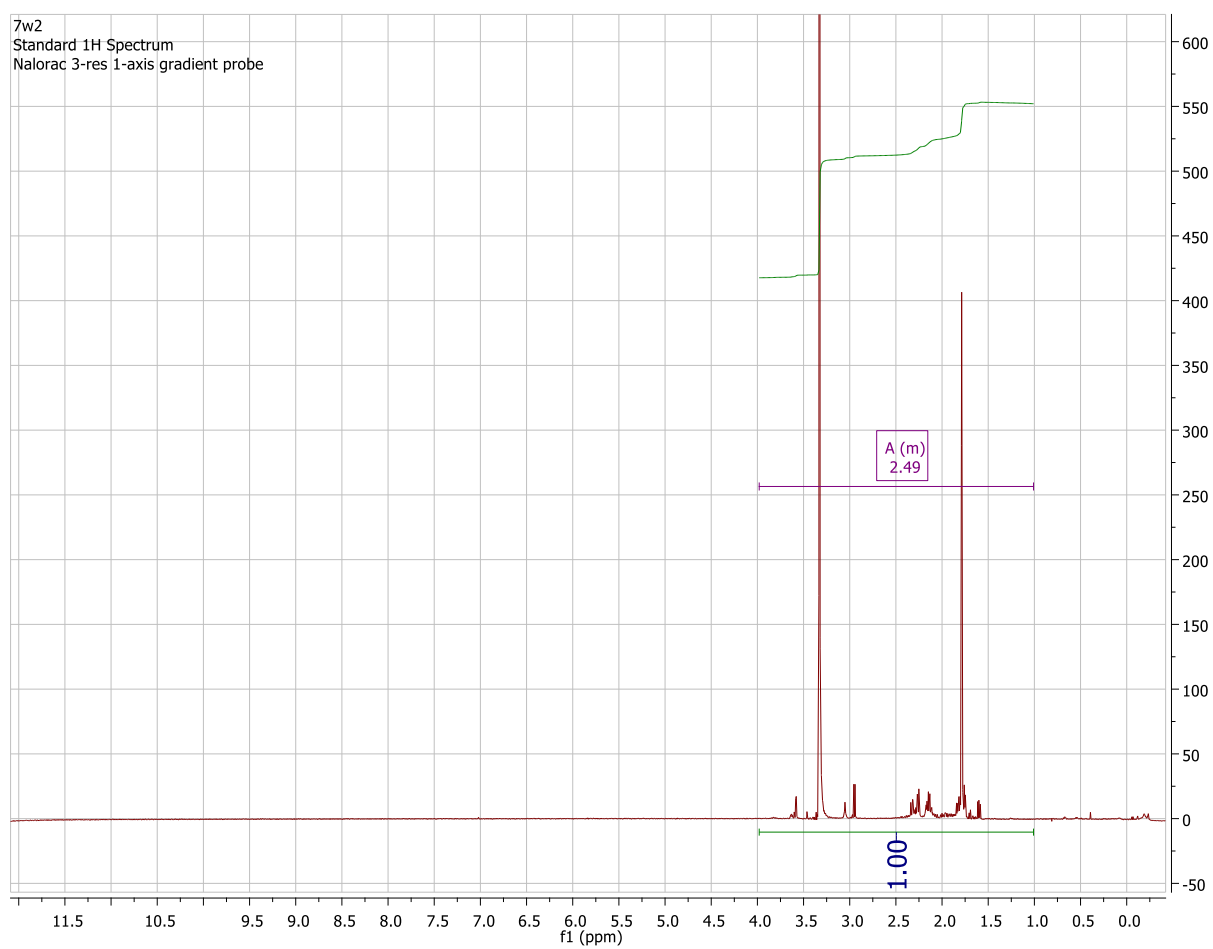
Competing Species Control	Length A (cm)	Length B (cm)	Length C (cm)	Age of Culture in Days
CC	3.8	1.9	2/CE	9

- **Data for 11 and 14 days not shown.**
- **Data for repeated assays on 7D2, 29A6, and 10B1 with Daily measurements not shown.**

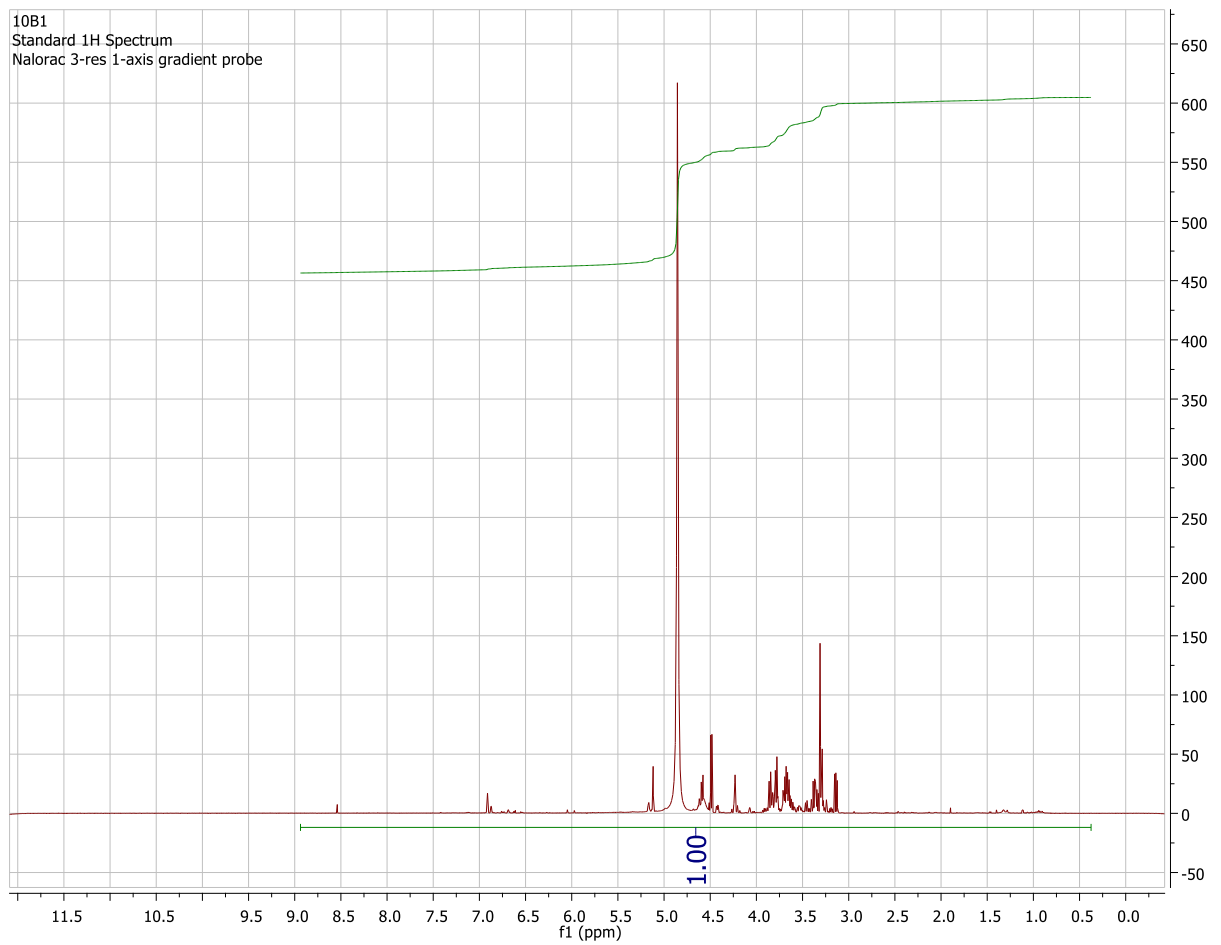
## APPENDIX B

$^1\text{H}$  NMR Spectra: obtained from Dr. Robert Bates from the Department of Chemistry and Biochemistry, University of Arizona

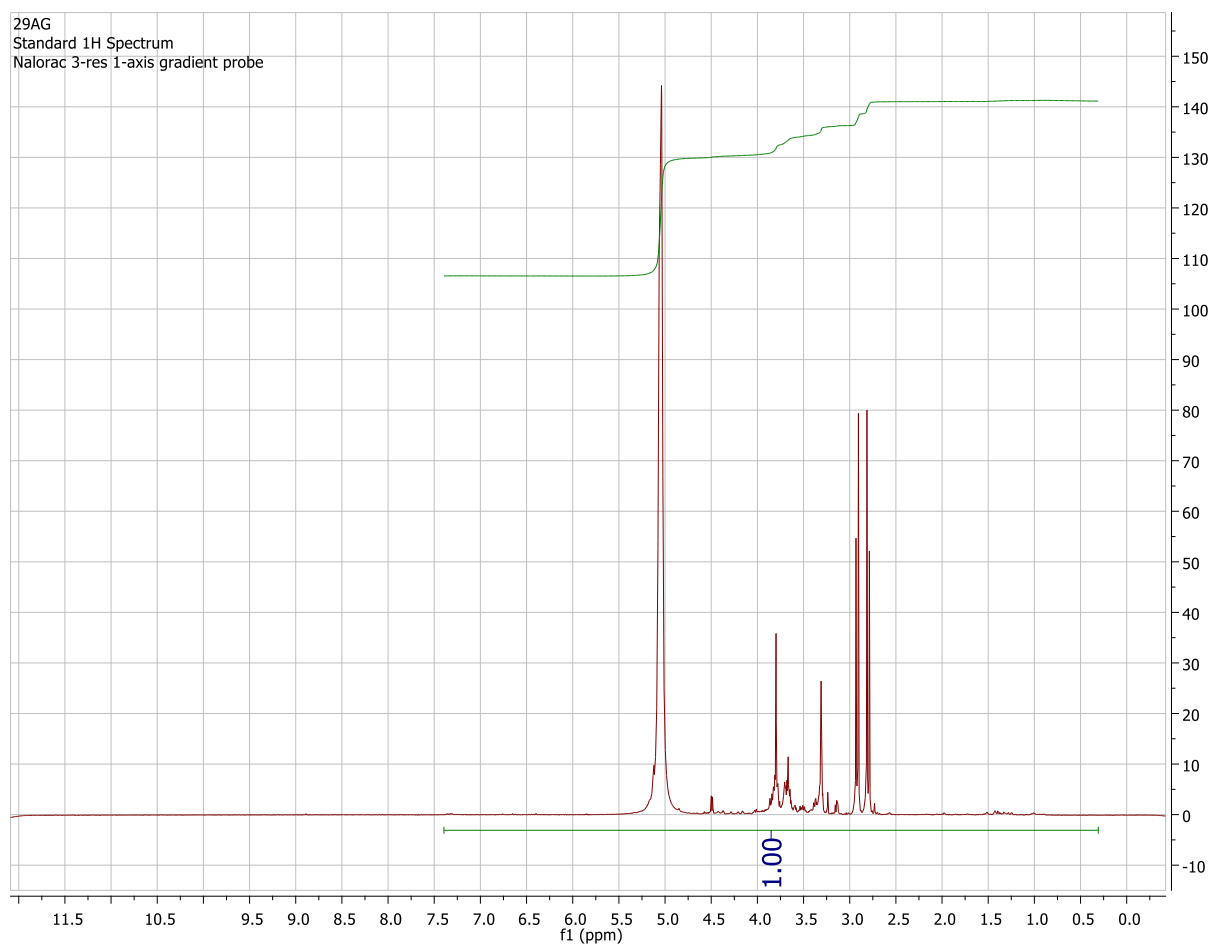
### FUNGAL ISOLATE 7D2 (*NECTRIA HAEMATOCOCCA*)



# FUNGAL ISOLATE 10B1 (*PENICILLIUM ATROVENETUM*)



# FUNGAL ISOLATE 29A6 (*PENICILLIUM DIPODOMYICOLA*)



Chromatography Data- obtained from Dr. Robert Bates from the Department of Chemistry and Biochemistry, University of Arizona

Samples from silica gel column chromatography 11/2012

All <sup>1</sup>H NMR spectra run in MeOH-d<sub>4</sub> except 7D2-8 and 10B1-10, which were run in D<sub>2</sub>O

**29A6** -Half the total sample after evaporation of MeOH (260 mg) put on column with ~10% MeOH/EtOAc); most fractions 80 mL, last fraction 300 mL. Overall from NMR: ~50% citrate, 40% glucoses, rest mostly another sugar not yet identified.

<u>fraction</u>	<u>eluent</u>	<u>~mg</u>	<u>NMR results</u>
29A6-1-2	EtOAc	44	Mostly fats; some citrate ion and aromatics
29A6-3-4	15% MeOH	77	About equal amounts of citrate and glucose (α&β)
29A6-5-6	50% MeOH	64	Mostly glucoses and citrate; another sugar
29A6-7	MeOH	77	Like above plus fatty acids

**7D2** -Attempted to put 88 mg on column with MeOH, but only 25 mg dissolved; 80 mL fractions except first was 30 mL

<u>fraction</u>	<u>eluent</u>	<u>~mg</u>	<u>NMR results</u>
7D2-1	50% MeOH	3	Mostly dioctyl phthalate (DOP, a common plasticizer impurity; 50% EtOAc not worth testing unless other fractions are inactive)
7D2-2	50% MeOH	15	Mostly glycosides; smaller amounts of glycerides
7D2-3-4	50% MeOH	5	Mostly glycerides; small amounts of glycosides
7D2-5-7	MeOH	5	Some glycosides and glycerides
7D2-8	insol MeOH	64	Complex mixture of sugar derivatives

**10B1** -Attempted to put 75 mg on column with MeOH, but only 60 mg dissolved; 80 mL fractions.

<u>fraction</u>	<u>eluent</u>	<u>~mg</u>	<u>NMR results</u>
10B1-1	MeOH	5	Mostly fatty acid derivatives; a little DOP; not worth testing unless other fractions are inactive
10B1-2	MeOH	41	Mostly sugar derivatives; some fatty acids and aromatics
10B1-3-4	MeOH	6	Mostly fatty acids and sugars; traces of aromatics
10B1-5-9	MeOH	8	Fatty acids and sugars
10B1-10	insol MeOH	6	Brown crystals removed from top of column; very broad peaks of sugar derivative or perhaps Fe complex.