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Encapsulation of *Trichoderma harzianum* with sodium alginate and evaluation of efficacy against fungal plant pathogen

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Abstract - Plant pathogenic fungi pose significant agricultural challenges, potentially as the cause of crop damage and reduced yields. This study evaluated the effectiveness of bioproducts formulated by encapsulating *Trichoderma harzianum* spores with sodium alginate for controlling 3 plant pathogenic fungi: *Fusarium* sp., *Curvularia* sp., and *Alternaria* sp. The assessment employed the dual culture methods on a potato dextrose agar (PDA) medium. The research revealed that this bioproduct's wet and dry forms, stored at different temperatures (4 °C and 28±2 °C), could maintain their spherical, uniform, and green appearances. Spore counts and spore survival for wet and dry forms stored at 4 °C were 2.53×10^6, 1.01×10^6 and 1.31×10^6, 1.10×10^5 spores/ml, respectively. Spore counts and spore survival for those stored at 28±2 °C were 3.18×10^6, 1.11×10^6 and 1.29×10^6, 1.00×10^5 spores/ml, respectively. Regarding the control of pathogenic fungi growth, both wet and dry forms stored at 4 °C exhibited inhibition percentages ranging from 64.10% to 82.35% and 64.74% to 81.25%, respectively. Similarly, those stored at 28±2 °C were shown to have inhibition

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percentages ranging from 65.38% to 84.31% and 66.66% to 86.27%, respectively. Statistically significant differences ($p<0.05$) were observed in the inhibition rates of all 3 pathogenic fungi between the two forms of bioproducts stored at different temperatures.

Keywords: Biocontrol, bioproduct, encapsulation, plant pathogen, *Trichoderma harzianum*

1. Introduction

Many farmers favor using chemicals to combat plant pathogenic fungi due to their quick results. However, this approach has led to significant adverse consequences, such as environmental pollution and harmful health effects on farmers and consumers. It also poses the risk of increasing the chemically resistant strains of fungi (Mbarga et al., 2012). Moreover, the persistent application of chemicals has resulted in cumulative effects within cultivated areas, resulting in soil pollution with substantial ecological damage. In addition, in today's world, there is a growing tendency to consume agriculturally produced goods that meet safety standards. In response to these aforementioned issues, there has been an increasing interest in employing biological control processes, either before or after harvest, by using microorganisms like fungi, yeast, and bacteria. This potential has primarily been driven by their environmentally friendly characteristics (Sellitto et al., 2021). Utilizing *Trichoderma* spp. fungi for controlling fungal plant pathogens emerge as a viable alternative due to their efficiency and widespread global acceptance (Asad, 2022). *Trichoderma* spp. are fungi that inhabit the soil and can produce many conidia and hyphae. These fungi are known for their antagonistic properties as being characterized by rapid growth and adaptability to various habitats (Siddiquee et al., 2009).

Trichoderma spp. belong to a group of fungi with antagonistic activity against various plant pathogenic fungi (Adnan et al., 2019). As reported by Abbas et al. (2022), *Trichoderma* spp. fungi have been utilized in biological control due to their several antagonistic mechanisms, including competition for nutrients, parasitism, production of antibiotics, and the release of enzymes to break down the cell walls of plant pathogenic fungi (Zafra & Cortés-Espinosa, 2015). Moreover, they could promote plant resistance to pathogens, benefiting plant health without harming humans (Zeilinger et al., 2016). Therefore, improvement of the stability and application methods of *Trichoderma* spp. products are essential. However, many commercial products have used live spores of *Trichoderma* spp. that require storage below 28°C to maintain efficacy, which could not be practically applied in real environmental settings. (Maruyama et al., 2020). Also, several factors could affect the efficiency of the product, such as storage, temperature, moisture, and pH levels. These factors resulted in the low stability of fungi due to either a low release of spores in the target area or the rapid decomposition of important substances. As a result, the efficacy of *Trichoderma* spp. for inhibition of plant pathogenic fungi is compromised (Fraceto et al., 2018). A report on the encapsulation of *Trichoderma harzianum* with sodium alginate has shown the potential to enhance the control of plant pathogenic fungi (Mohd Anuar et al.,

2020). Thus, encapsulation may represent a viable option for preserving the stability and efficacy against the growth of fungal plant pathogens.

Therefore, this research aimed to study the production of bioproducts that encapsulated the spores of *T. harzianum* and test for their efficacy against the growth of fungal pathogens, including *Fusarium* sp., *Curvularia* sp., and *Alternaria* sp.

2. Materials and methods

2.1 Fungi

The fungus *T. harzianum* was provided courtesy of the Biological Control Research Unit, Faculty of Technology, Mahasarakham University.

Plant pathogenic fungi include *Fusarium* sp., known for causing root rot, stem rot, and seed rot; *Curvularia* sp.; and *Alternaria* sp., known for causing leaf spots, leaf blight, and dirty panicle. These fungi were provided courtesy of the Biological Control Research Unit, Faculty of Technology, Mahasarakham University.

2.2 Encapsulation of *T. harzianum* spores with sodium alginate

The encapsulation of fungal spores of *T. harzianum* with sodium alginate was carried out through the process of ionic gelation, as described by Maruyama et al. (2020). This involved dripping a sodium alginate suspension into a solution of calcium chloride dihydrate ($CaCl_2.2H_2O$) using sodium alginate ($C_5H_7O_4COONa$) at a concentration of 2% (w/v). The sodium alginate was dissolved in sterile distilled water, stirred homogeneously for 30 min, and adjusted for a pH of 5.5 to 5.6. After that, 1 part of *T. harzianum* spore suspension was mixed with 1 part of the sodium alginate solution (v/v) to result in a concentration of 10^8 spore/ml. Then, the mixture was stirred homogeneously for 15 min. Subsequently, a peristaltic pump with a dropper tube diameter of 2 mm was used to drip the mixture into a calcium chloride dihydrate solution with a concentration of 0.1 M. While dripping, a magnetic bar was used for stirring to prevent the aggregation of microorganisms. The bioproducts soaked in calcium chloride dihydrate solution were stored at 4 °C for 24 h to ensure their stability. After that, the bioproducts were rinsed with sterile distilled water and dried in a sterile cabinet (laminar airflow) at 25±2 °C for 1 h, resulting in the production of a bioproduct that encapsulated the spores of *T. harzianum*. These bioproducts were then divided into 2 parts, wet and dry granules. The dry granules were further dried in a hot air oven at 40 °C for 12 h, as described by Locatelli et al. (2018). Subsequently, both wet and dry bioproducts were packed in opaque aluminum ziplock bags with a size of 8×10 cm (width × length) and a thickness of 0.16 mm. They were stored at 4 °C and 28±2 °C for two weeks.

2.3 Study of general characteristics of bioproducts

The size and weight of bioproducts encapsulating the spores of *T. harzianum* were examined, and 100 granules from each condition both wet and dry were measured for the width and length using a vernier caliper, while weight was measured using an Analytical Balance with 4 digits (Ohaus, pa323c, USA).

The pH test in this study is adapted from the methods of Kawicha et al. (2020). Each bioproduct (0.50 g) was dissolved in 2% sodium citrate ($NaC_6H_5O_7$) solution (49.50 ml). It took 1 h to dissolve the wet granules of the bioproduct and 8 h to dissolve the dry granules, and the solutions were then measured for pH using an Eutech model pH 700 instrument. The experiments were performed in triplicates and calculated for average.

For determination of water activity (aw), each bioproduct (1 g) was measured for water activity using an Aqualab series 3 (Decagon, Pullman, USA) with an accuracy of ±0.003. The experiments were repeated 3 times, and then calculated for the average.

The external surface characteristics of bioproducts were studied using a scanning electron microscope (SEM) at the Central Lab Center, Mahasarakham University.

2.4 Determination of spore quantity of bioproducts after encapsulation

Quantification of spores in bioproducts in this study was adapted from the methods of Maruyama et al. (2020). Each bioproduct (0.1 g) was dissolved in 2% sodium citrate solution (9.90 ml). The bioproduct in the form of wet granules took 1 h to dissolve completely, and the dry granules took 8 h. Then, 1 drop of 0.1% Tween 20 (Pubchem, USA) was added to the solution and followed by shaking with a vortex mixer for 1 min to disperse the fungal spores. The fungal spores were counted under a microscope with hemacytometer. The experiments were performed in triplicates and calculated for the average. After that, examine the number of the spores in 1 granule by using the average weight of wet and dry granule bioproducts stored in different temperatures as shown in Table 1.

2.5 Testing the survival spore of *T. harzianum* in bioproducts after encapsulation

This test was adapted from the method of Kawicha et al. (2020). It started with sampling wet and dry granule bioproducts under each storage condition, amounting to 0.1 g of each type dissolved in 2% sodium citrate and quantity 9.90 ml. Wet bioproducts took 1 h to dissolve, while dry bioproducts took 8 h to dissolve. Then, added 1 drop of Tween 20 (Pubchem, USA) at a concentration of 0.1 % and shook with a vortex mixer for 1 min to allow the fungal spores to dissolve. Then, it was diluted from 10^{-2} to 10^{-5} and each dilution level was grown on a PDA medium using a spread plate method with 100 µL on the surface of the PDA. The experiment was repeated with 3 dilutions and incubated at 28±2°C for 48 h. The number of colonies germinated on the PDA medium was then counted and calculated for the average. After that, the survival rate of the spores in 1 granule was examined by using the average weight of wet and dry granule bioproducts stored in different temperatures as shown in Table 1.

2.6 Testing the efficacy of bioproducts that encapsulating fungal spores of *T. harzianum* against the growth of *Fusarium* sp., *Curvularia* sp., and *Alternaria* sp.

This experiment was adapted from the methods described by Maruyama et al.

(2020). Bioproducts encapsulating *T. harzianum* spores, 1 wet granule and 1 dry granule encapsulating spores in their respective storage conditions, were placed on the potato dextrose agar (PDA) using the dual culture method. The bioproducts and plant pathogenic fungi were placed in an opposite direction at 5 cm apart with 2 cm distance from the edge of the petri dish. The tests were performed in triplicates. PDA plates were incubated at 28±2 °C. The radius of fungal colonies was measured 7 days later. The percent inhibition of radial growth (PIRG) was calculated according to the formula described by Gamliel et al. (1989) as follows: The percentage of control (PIRG) = $[(R_1 - R_2) / R_1 \times 100]$, where R_1 was the radius of the control fungal colony (mm), and R_2 was the radius of the fungal colony grown together with the bioproduct (mm).

2.7 Statistical analysis

Data were analyzed by descriptive statistics for percentage, mean and standard deviation, compared by factorial in complete randomized design (CRD), F-test and Duncan's multiple range test (DMRT) at 95% confidence interval ($p<0.05$).

3. Results and discussion

3.1 General characteristics of the bioproducts encapsulating *T. harzianum* spores

The size and weight of bioproducts stored at 4 °C and 28±2 °C were examined as follows: For wet granules stored at 4 °C and 28±2 °C, average widths were 3.32±0.51 and 3.23±0.42 mm, average lengths were 3.88±0.53 and 3.66±0.61 mm, average weights were 0.0234±0.0026 and 0.0239±0.0013 g, respectively.

Regarding dry granules stored at 4 °C and 28±2 °C, average widths were 0.94±0.13 and 0.92±0.15 mm, average lengths were 1.20±0.16 and 1.21±0.21 mm, average weights were 0.0015±0.0003 and 0.0015±0.0002 g, respectively as shown in Table 1.

The external characteristics of wet encapsulating granules stored at 4 °C and 28±2 °C were examined respectively. It was observed that these granules maintained a uniform green color and possessed round shapes with smooth surfaces. However, when compared to the control granules, they appeared to be dark, primarily due to the presence of spores of *T. harzianum*, as well as the dry granules stored at 4 °C and at 28±2 °C, they were found spherical in shape with a deep green color. When compared to the control granules, they appeared to be dark, primarily due to the presence of spores of the *T. harzianum*, as shown in Figure 1. This was consistent with the report of Maruyama et al. (2020), which reported that the bioproduct granules prepared without fungi control exhibited a generally spherical shape with slightly translucent white color, while the bioproduct granules with *T. harzianum* spores exhibited a spherical shape with green color. When studying the external surface characteristics using a scanning electron microscope (SEM), the wet granules that were stored at 4 °C and 28±2 °C were a uniform surface texture with minimal roughness, possibly due to the dispersion of *T. harzianum* spores. In contrast to the control granules, the surface of the dry granules that was

stored at 4 °C and at 28±2 °C were found to have a withered or cracked surface with some roughness and numerous pores due to moisture loss during the drying process, as shown in Figure 2. These observations were consistent with the study Maruyama et al. (2020), which reported that the surface texture of bioproduct granules could become rough due to drying process. This was also consistent with the findings of Qi et al. (2023), which reported that, after drying, the bioproduct granule's surface became smaller than with visible cracks and numerous pores, facilitating the release of spores from the bioproduct.

Table 1. Characteristics of bioproducts (alginate beads encapsulating with *T. harzianum* spores in the forms of wet and dry granules)

Bioproducts	Storage temperature	Formula	Characteristics				
			Width (mm)	Length (mm)	Weight (g)	pH value	AW value
wet granule	4 °C	with spore	3.32±0.51^{a}	3.88±0.53^{a}	0.0234±0.0026^{a}	6.80±0.04^{a}	0.987±0.01^{a}
		control	3.20±0.45^{b}	3.65±0.78^{b}	0.0219±0.0030^{b}	6.83±0.04^{a}	0.989±0.01^{a}
	28±2 °C	with spore	3.23±0.42^{ab}	3.66±0.61^{b}	0.0239±0.0013^{a}	6.82±0.02^{a}	0.994±0.01^{a}
		control	3.17±0.57^{b}	3.69±0.51^{b}	0.0205±0.0027^{b}	6.83±0.02^{a}	0.993±0.01^{a}
dry granule	4 °C	with spore	0.94±0.13^{c}	1.20±0.16^{c}	0.0015±0.0003^{c}	6.33±0.13^{b}	0.448±0.03^{b}
		control	0.93±0.15^{c}	1.19±0.19^{c}	0.0014±0.0003^{c}	6.26±0.04^{b}	0.443±0.02^{b}
	28±2 °C	with spore	0.92±0.15^{c}	1.21±0.21^{c}	0.0015±0.0002^{c}	6.31±0.06^{b}	0.446±0.00^{b}
		control	0.91±0.13^{c}	1.22±0.19^{c}	0.0013±0.0003^{c}	6.28±0.04^{b}	0.430±0.02^{b}

Note: control = no spore
Note: Mean ± SD.
Vertically different letters $^{a,b,c,\cdots}$ indicate a statistically significant difference ($p<0.05$)

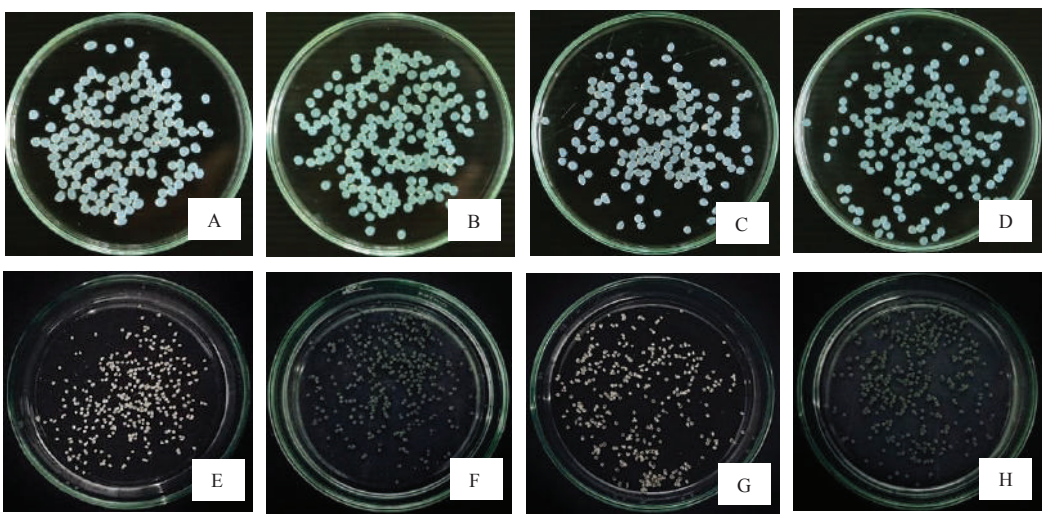


Figure 1. Bioproducts with encapsulated *T. harzianum* spores stored at 4 °C and 28±2 °C, wet granule 4 °C(control) (A), wet granule 4 °C (B), wet granule 28±2 °C (control) (C), wet granule 28±2 °C (D), dry granule 4 °C (control) (E), dry granule 4 °C (F), dry granule 28±2 °C (control) (G), dry granule 28±2 °C (H)

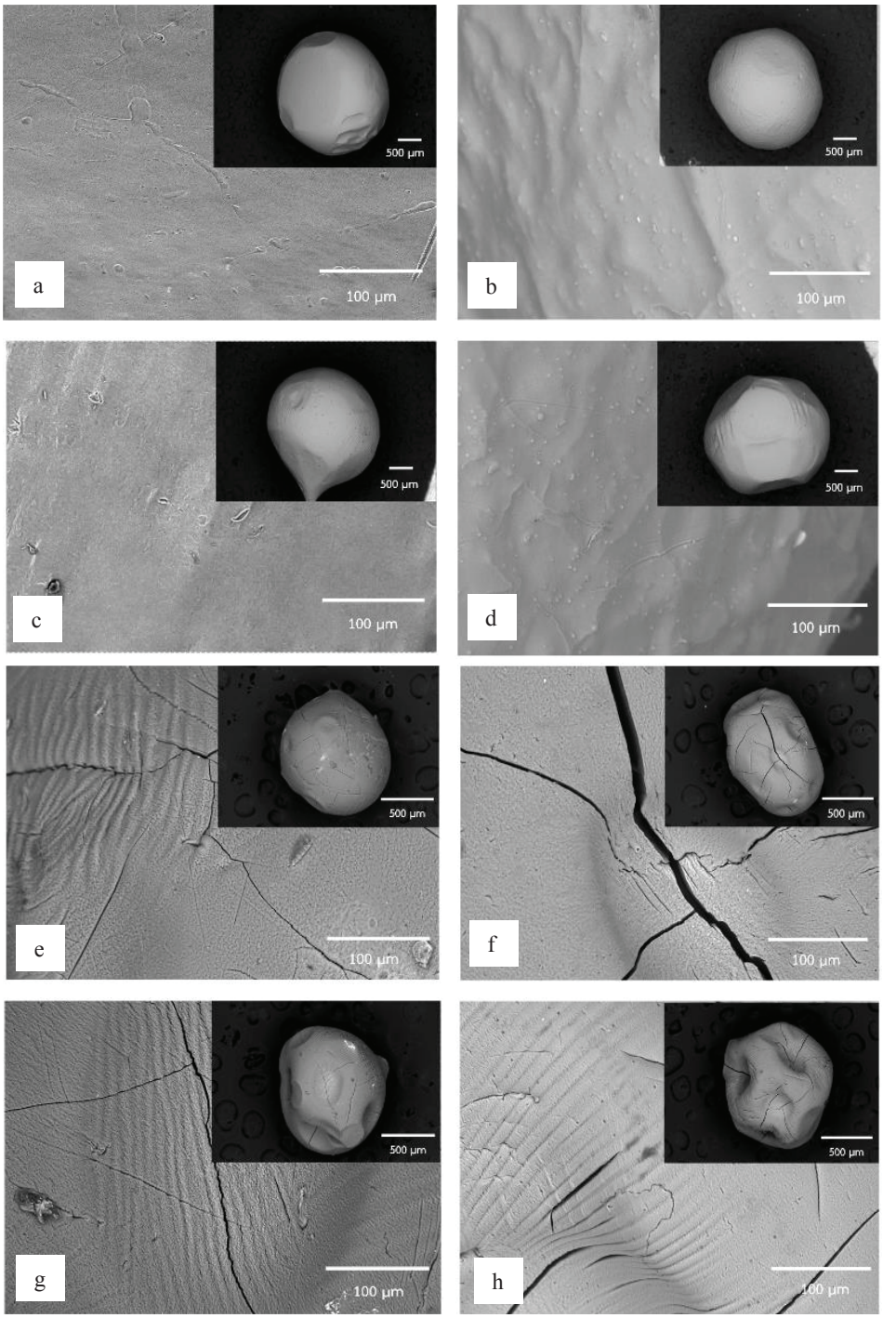


Figure 2. The surface of the bioproducts encapsulating with the *T. harzianum* spores was examined by a scanning electron microscope (SEM). The images were acquired at magnifications of 25 times (insert) and 300 times, wet granule 4 °C (control) (a), wet granule 4 °C (b), wet granule 28±2 °C (control) (c), wet granule 28±2 °C (d), dry granule 4 °C (control) (e), dry granule 4 °C (f), dry granule 28±2 °C (control) (g), dry granule 28±2 °C (h)

3.2 Spores quantity of bioproducts after encapsulation

The results of this study demonstrated that the wet granule showed a lower spore quantity than the dry granule. This difference was attributed to the higher weight of wet granule than the dry granule, affected the number of granules used in the test and impacted the spore quantity. However, when considering the number of spores in 1 granule, it was found that the average spore quantity of the wet granules was higher than that of the dry granules when the initial spore count was 10^8 spores/ml. The reason that the dry granule had a lower number of spores may be due to the drying process, according to research by Locatelli et al. (2018), which reported that the number of spores varied depending on the duration of drying. During drying, they lost moisture due to exposure to temperature and long drying conditions, resulting in a decrease in spore concentration.

Table 2. Quantity of *T. harzianum* spores in bioproducts under storage condition.

Bioproducts	Storage temperature	Spore volume (spore/ml)	
		bioproducts 0.1 g	bioproducts 1 granule
wet granule	4 °C	$1.08\times10^7\pm0.14^b$	$2.53\times10^6\pm0.03^a$
	28±2 °C	$1.33\times10^7\pm0.52^b$	$3.18\times10^6\pm0.12^a$
dry granule	4 °C	$8.83\times10^7\pm0.62^a$	$1.31\times10^6\pm0.01^b$
	28±2 °C	$8.67\times10^7\pm0.80^a$	$1.29\times10^6\pm0.01^b$

Note: Mean ± SD.
Vertically different letters $^{a,b,c,\cdots}$ indicate a statistically significant difference ($p<0.05$)

3.3 The survival spore of *T. harzianum* in bioproducts after encapsulation under each storage condition

The number of survival spores per granule of bioproducts revealed that the wet granule had a higher survival rate than the dry granule, as shown in Table 3. Moisture loss may have occurred because of the drying process due to prolonged exposure to temperature and drying conditions. Consequently, there is a reduction in the concentration of spores (Locatelli et al., 2018), affecting the survival of *T. harzianum* spores in the dry granule bioproducts. In addition, wet granule bioproducts may be more suitable for the growth and survival of spores during storage than dry granule bioproducts because the wet granule bioproducts stored at 4 °C and at 28±2 °C gave water activity values (aw) of 0.987 and 0.994, respectively, as shown in Table 1. This resulted on the growth of *T. harzianum*, which is consistent with the research of Santamarina and Rosello (2006) who reported that the *T. harzianum* grew well when the water activity value (aw) was between 0.950 and 0.995. The survival rate of wet granule bioproducts stored at 28±2 °C was higher than that at 4 °C, possibly due to the temperature conditions at 28±2 °C being suitable for the growth of *T. harzianum*, which is consistent with the research of Gupta and Sharma (2013) who reported that *T. harzianum* grew well at 25 °C to 30 °C, grew slowly at 37 °C, and no growth was observed at 45 °C. However, spore survival tests of *T. harzianum* after encapsulation showed that the ability of

sodium alginate to encapsulate fungal spores and the storage conditions of the two bioproducts showed different effects on *T. harzianum* survival.

Table 3. Survival rate of *T. harzianum* spores in bioproducts under storage condition

Bioproducts	Storage temperature	Quantity of surviving T. harzianum spores (spore/ml)	
		bioproducts 0.1 g	bioproducts 1 granule
wet granule	4 °C	$4.33\times10^{6}\pm0.58^{b}$	$1.01\times10^{6}\pm0.14^{a}$
	28±2 °C	$4.67\times10^{6}\pm0.58^{b}$	$1.11\times10^{6}\pm0.14^{a}$
dry granule	4 °C	$7.33\times10^{6}\pm1.15^{a}$	$1.10\times10^{5}\pm0.02^{b}$
	28±2 °C	$6.67\times10^{6}\pm0.58^{a}$	$1.00\times10^{5}\pm0.01^{b}$

Note: Mean ± SD.
Vertically different letters $^{a,b,c,\cdots}$ indicate a statistically significant difference ($p<0.05$)

3.4 Efficacy of bioproducts encapsulating the spores of *T. harzianum* against *Fusarium* sp., *Curvularia* sp., and *Alternaria* sp.

The inhibition percentages of wet granules stored at 4 °C against the growth of *Fusarium* sp., *Curvularia* sp., and *Alternaria* sp. were 78.94±0.00, 64.10±1.10, and 82.35±5.88, respectively. The inhibition percentages of wet granules stored at 28±2 °C were 83.33±1.80, 65.38±1.92, and 84.31±3.39, respectively. The dry granules stored at 4 °C showed the inhibition percentages at 81.25±0.00, 64.74±1.10, and 78.43±3.39, respectively, dry granules stored at 28±2 °C had the inhibition percentages at 85.41±7.21, 66.66±1.10, and 86.27±3.40, respectively, as shown in Table 4.

The results of this test demonstrate that *T. harzianum* encapsulated with sodium alginate exhibited statistically significant differences ($p<0.05$) in their ability to control the growth of all 3 tested phytopathogenic fungi. The inhibition percentages growth of *Fusarium* sp. and *Alternaria* sp. were relatively high, as shown in Tables 4 and 5. The inhibition percentage falls within the range of 64.10% to 86.27%. The different temperature storage preservation conditions and drying process for the bioproducts were found to have no statistically significant impact on their ability to control the growth of the tested fungal hyphae, this was according to the research conducted by Mancera-López et al. (2019), which reported that *T. harzianum* encapsulated with sodium alginate could control the growth of 4 different pathogenic fungi: *Penicillium citrinum, Aspergillus awamori, Rhizoctonia solani*, and *Aspergillus niger*. When encapsulated with sodium alginate, *T. harzianum* can grow on the PDA surface, completely covering the surface when tested with *P. citrinum*, and when tested with *A. awamori* and *R. solani*, it can also grow more than 75.00% on PDA surface. However, its growth and colonization on PDA surfaces with *A. niger* were less than 25.00%. This might be attributed to the rapid growth of *A. niger*. Furthermore, this was consistent with the research by Maruyama et al. (2020), which reported that *T. harzianum* encapsulated with sodium alginate, both in wet and dry forms and tested in combination with *Sclerotinia*

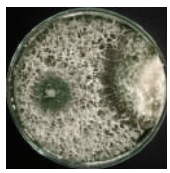
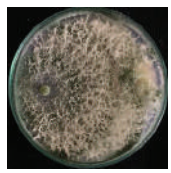
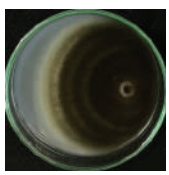
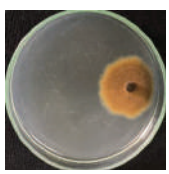
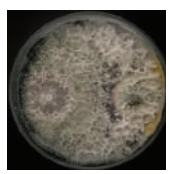
sclerotiorum, could effectively colonize 2 out of 3 portions of the potato dextrose agar (PDA). In this experiment, it was observed that *T. harzianum* encapsulated with sodium alginate could maintain the number of viable spores without adversely affecting its ability to control plant pathogenic fungi. Furthermore, this also promoted sporulation during storage, resulting in even spore distribution throughout the granules of the bioproduct. This aligned with the research by Mancera-López et al. (2019), which reported that when granule bioproducts were placed on the PDA surface, nutrients were absorbed into the granule bioproduct containing the spores of *T. harzianum* and promoting the distribution of spores in the granules. This, in turn, led to rapid and uniform germination of spores, which contributed to the effectiveness of controlling plant pathogenic fungi. However, the effectiveness of bioproducts for controlling plant pathogenic fungi may be influenced by other factors, such as the variability in strains of the tested plant pathogenic fungi. The results of this test showed that when comparing the efficacy for controlling plant pathogenic fungi based on the types of bioproducts and the storage conditions, there was no statistically significant difference. Nonetheless, when testing both types of bioproduct (wet and dry) in each storage condition along with the 3 types of plant pathogenic fungi, it was found that the drying process and storage temperature did not affect the efficacy of the bioproducts, but the differences clearly depend on the type of plant pathogen tested, with the inhibition percentage the fungi, *Curvularia* sp. being the lowest.

Table 4. Inhibition percentages of plant pathogenic fungi by each bioproduct after 7 days of storage at 4 °C or 28±2 °C

pathogenic fungi	% Inhibition of fungal growth			
	wet granule		dry granule	
	4 °C	28±2 °C	4 °C	28±2 °C
Fusarium sp.	78.94±0.00^{aA}	83.33±1.80^{aA}	81.25±0.00^{aA}	85.41±7.21^{aA}
Curvularia sp.	64.10±1.10^{bA}	65.38±1.92^{bA}	64.74±1.10^{bA}	66.66±1.10^{bA}
Alternaria sp.	82.35±5.88^{aA}	84.31±3.39^{aA}	78.43±3.39^{aA}	86.27±3.40^{aA}

Note: Mean ± SD.
Vertically different letters $^{a,b,c,\cdots}$ indicate a statistically significant difference ($p<0.05$)
Horizontally different letters $^{A,B,C,\cdots}$ indicate a statistically significant difference ($p<0.05$)

Table 5. The inhibition of pathogenic fungal growth by bioproducts encapsulating with *T. harzianum*

pathogenic fungi	control	Bioproducts and Storage temperature			
		wet granule		dry granule	
		4 °C	28±2 °C	4 °C	28±2 °C
Fusarium sp.					
Curvularia sp.					
Alternaria sp.					

Note: control = not containing bioproducts

4. Conclusion

Using *T. harzianum* fungus to control plant pathogenic fungi positively impacts the environment, agricultural production and consumers. However, to enhance the persistence and effectiveness of the fungus, this experiment showed that the bioproducts produced by encapsulating *T. harzianum* with sodium alginate using the ionic gelation process were a method for producing bioproducts. The efficiency in controlling plant pathogenic fungi, especially *Fusarium* sp., *Curvularia* sp., and *Alternaria* sp., was tested using a pair culture test on PDA media. In conclusion, they can highly inhibit plant pathogenic fungi up to 80% when wet and dry granules are stored at different temperatures. The drying process and storage temperature do not affect the bioproducts. The results highlight a positive impact and the potential future field application.

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