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Efficacy of Nano Particles from *Chaetomium cupreum* to Control *Phytophthora* spp. Causing Root Rot of Durian

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Durian is a tropical fruit crop which is highly prized culturally and economically in South-East Asia. *Phytophthora* spp. was found to be pathogenic isolate which causing root rot of durian and confirmed pathogenicity test of the virulent pathogen on detached leaves. The tested nano particles; nano-CCH, nano-CCE and nano-CCM derived from *Chaetomium cupreum* to inhibit mycelial growth and spore production of *Phytophthora* spp. showed the ED_{50} values of 3.49, 3.47 and 3.80 µg/ml, respectively. This research finding is firstly reported using nano particles derived from *Chaetomium cupreum* to inhibit *Phytophthora* spp. causing root rot of durian.

Keywords: *Chaetomium cupreum, Phytophthora* spp.

Introduction

Durian (*Durio zibethinus Murr*.) is king of tropical fruit refer to two facts of the fruit. Its superlative fresh, which is highly nutritional and its appearance, which resembles the thorny thrones of the Asian kings of old. Durian is one of the most famous fruit in South-East Asia. The fruit is very famous not only due to the taste richness but also the strong odour. Durian is an economically fruits in Thailand. The country is the world's largest producer and exporter of durian, followed by Malaysia and Indonesia (Somsri, 2014). In past, root rot has been reported to the serious rate of infection of durian because monoculture planting and high fertilizer applications could lead the increment of disease incidence caused by fungi such as *Phytophthora palmivora* and *Pythium* spp. Chemical compounds have been used to control plant diseases, but abuse in their employment has favored the development of pathogens resistant to fungicides. This research, the use of *Chaetomium cupreum* that antagonize plant pathogens

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is risk-free when it results in enhancement of resident antagonists. Moreover, biological control agents (BCAs) could reduce levels of fungicide.

Materials and methods

Morphological Studies

The fungal obtained from Chiang Mai University, were study based on the macroscopic and microscopic characteristics.

The macroscopic characteristics of colony appearance were determined including growth pattern and texture and growth rate onto PDA plates. For microscopic characteristics shapes of zoosporangia were observed by using a light microscope.

Pathogenicity Test

Pathogenicity test was done by the plug inoculation method. The healthy durian detached leaves were sterilized by 10% sodium hypochlorite. The surface detached leaves were made wounds by sterilized needle. The agar plug of pathogen inoculated to wound on detached leaves. The controls were processed similarly but transferred an agar plug without the pathogen.

In vitro test of nano particles from Chaetomium cupreum to inhibit Phytophthora spp.

The experimental to test for inhibition of mycelial growth and sporangium formation of *Phytophthora* spp. by using poison food method. The Experimental was conducted by using Completely Randomized Design (CRD) with four replications. The concentration of nano particles; nano-CCH, nano-CCE, nano-CCM as follow: 0, 3, 5, 10, 15 ppm. Each concentration was dissolved in 2% dimethyl sulfoxide (DMSO), then mixed into potato dextose agar(PDA) and added chitosan before autoclave at 121 ℃ for 20 minutes. The agar plug of pathogen was moved to PDA plates in each solvent and concentration. After incubation at 25 ℃ collected data as colony diameter, number of sporangia, inhibition percentage and Effective dose ED_{50} values of mycelial growth and sporangium formation. Data was statistically computed analysis of variance. Treatment means were compared with DMRT at P = 0.05

Results and discussion

Morphological Studies

The fungal growth rapidly and colonized the plate within 4 days on PDA. Colony morphology on PDA is a chrysanthemum pattern with aerial mycelium. Sporangia are globose and ovoid shape, which was papillate. Zoospores were directly released from sporangia when flooded in water.

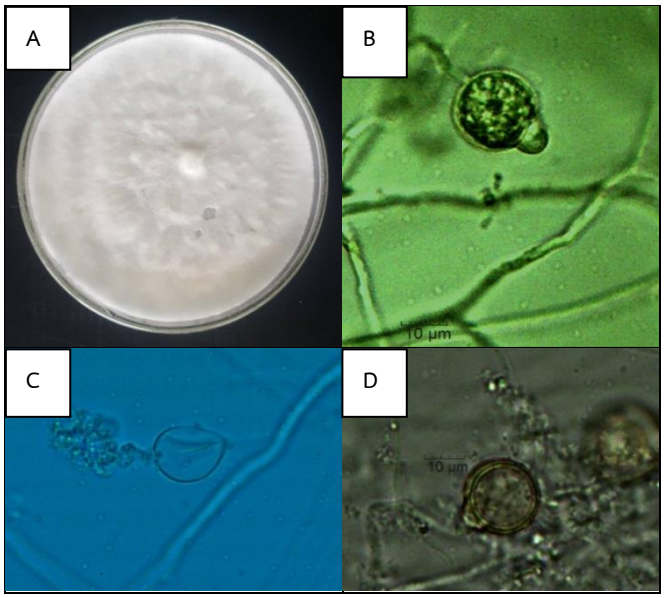


Fig. 1. Morphological characteristics of *Phytophthora* spp.)A(; Colony appearance on PDA)B(; Shape of sporangia)C(; Zoospore release from sporangia)D(; Oogonia

Pathogenicity test

Pathogenicity test on detached leaves after 3 days by the plug inoculation method. Leaves showed symptoms of brown hydrolysis expand around agar plug of pathogen. In control, Leaves remained healthy.

Fig. 2. Pathogenicity test of *Phytophthora* spp. on detached leaves.)A(; The inoculated pathogen)B(; The inoculated cont

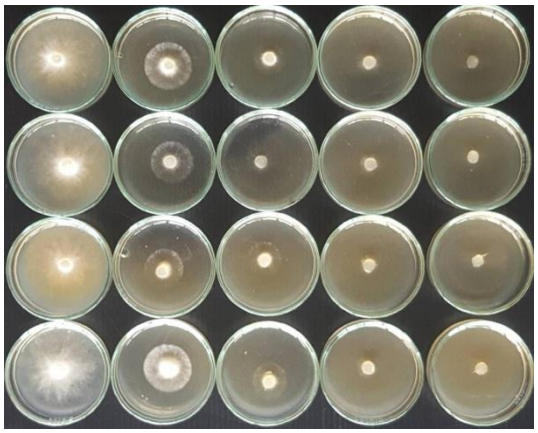
Nano particle test

Nano-CCH at concentrations of 3, 5, 10, 15 ppm were tested the colony growth inhibition of *Phytophthora* spp. which were 51.25, 61.75, 69.00 and 86.00 % respectively (Figure 2). Test inhibition of sporangia information of *Phytophthora* spp. which were 44.97, 64.39, 82.36 and 96.42 % respectively)Table 1(when compared to the control. Nano-CCE at concentrations of 3, 5, 10, 15 ppm were tested the colony growth inhibition of *Phytophthora* spp. which were 56.75, 60.50, 67.75 and 86.00 % respectively (Figure 2). Test inhibition of sporangia information of *Phytophthora* spp. which were 45.98, 64.50, 79.01 and 97.32 % respectively)Table 1(when compared to the control. Nano-CCM at concentrations of 3, 5, 10, 15 ppm were tested the colony growth inhibition of *Phytophthora* spp. which were 44.75, 61.75, 74.75 and 86.00 % respectively (Figure 2). Test inhibition of sporangia information of *Phytophthora* spp. which were 32.14, 73.21, 89.73 and 99.55 % respectively)Table 1(when compared to the control. Meanwhile ED_{50} values of nano-CCH, nano-CCE, nano-CCM were 3.49, 3.47 and 3.81 µg/ml respectively) Table 1(. Similar reports were confirmed by Soytong (2001), Kean (2010) that biological product of *Ch. cupreum* significantly inhibited *P. palmivora* of durian. The crude extracts of *Ch. cupreum* against mycelial growth of *P. nicotianae* causing root rot in citrus (Hung *et al*., 2015b) and pomelo (Hung *et al*., 2015a).

Table 1. Effect of nano particles from *Ch. cupreum* to inhibit *Phytophthora* spp.

Nano particles	Concentration)ppm(	Colony diameter)cm(	Inhibition of colony growth)%(	Number of sporangia $)\times10^6($	Inhibition of sporangia)%(	ED_{50}
Nano CCH	0	5.00[a]	-	56.00[a]	-	3.49
	3	2.43[c]	51.25[f]	30.81[c]	44.97[f]	
	5	1.91[e]	61.75[d]	19.93[d]	64.39[e]	
	10	1.55[f]	69.00[c]	9.87[f]	82.36[c]	
	15	0.70[h]	86.00[a]	2.00[h]	96.42[a]	
Nano CCE	0	5.00[a]	-	56.00[a]	-	3.47
	3	2.16[d]	56.75[e]	30.25[c]	45.98[f]	
	5	1.97[e]	60.50[d]	19.87[d]	64.50[e]	
	10	1.61[f]	67.75[c]	11.75[f]	79.01[c]	
	15	0.70[h]	86.00[a]	1.50[h]	97.32[a]	
Nano CCM	0	5.00[a]	-	56.00[a]	-	3.81
	3	2.76[b]	44.75[g]	38.00[b]	32.14[g]	
	5	1.91[e]	61.75[d]	15.00[e]	73.21[d]	
	10	1.26[g]	74.75[b]	5.75[g]	89.73[b]	
	15	0.70[h]	86.00[a]	0.25[h]	99.55[a]	
C.V. %)		4.79		6.68		

Average of four replications. Means followed by a common letter are not significantly different by DMRT at P = 0.05



Nano CCH

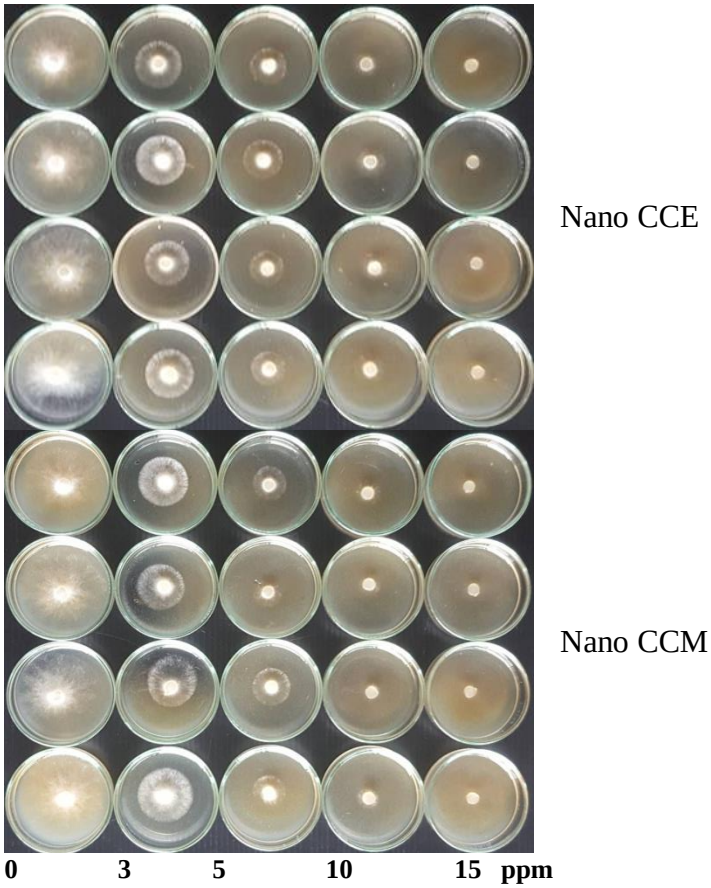


Figure 3. Testing nano-CC from *Ch. cupreum* against *Phytophthora* spp.

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