Journal of Agricultural Technology 2015 Vol. 11(8): 2457-2461
Available online http://www.ijat-aatsea.com
ISSN 1686-9141

Biological Activity of metabolites from *Lepiota procera* against plant pathogen (*Colletotrichum capsici*)

Phadungpran, Phaophilat[*]; Pongnak, Wattanachai [1]and Soytong, Kasem[1]

Department of Plant Production Technology, Faculty of Agricultural Technology, King Mongkut's Institute of Technology Ladkrabang , Bangkok 10520, Thailand

Phadungpran P., Pongnak W. and Soytong K. (2015). Biological Activity of metabolites from Lepiota procera against plant pathogen (Colletotrichum capsici). Journal of Agricultural Technology. 11(8): 2457-2461.

Bioactivity tests of crude extracts against *Colletotrichum capsici* causing anthracnose of chilli were tested. Result showed that crude extracts of *Lepiota procera* at concentration 1,000 ppm. gave the highest percent inhibition of colony of *C. capsici* causing anthracnose of chilli which 13.40, 16.00 and 46.40 %, respectively with this, inhibition of spore production were 66.45, 57.09 and 80.63 percent respectively.

Key words: Metabolites, Crude extracts, *Lepiota procera*, *Colletotrichum capsici*

Introduction

Chilli (*Capsicum annum* L.) is an important cash crop grown under both tropical and subtropical conditions. India is the largest grower, consumer and exporter of chilli, currently exporting dry chilli and chilli products to over 90 countries around the world (Singal, 1999). *Colletotrichum* is one of the most important plant pathogens worldwide causing the economically important disease anthracnose in a wide range of hosts including cereals, legumes, vegetables, perennial crops and tree fruits (Bailey and Jeger, 1992). Among these hosts, chilli (Capsicum spp.),an important economic crop worldwide (Poulos, 1992).

Anthracnose, derived from a Greek word meaning 'coal', is the common name for plant diseases characterized by very dark, sunken lesions, containing spores (Isaac, 1992). Generally, anthracnose disease is caused by *Colletotrichum* species which belongs to the Kingdom Fungi; Phylum Ascomycota, Class Sordariomycetes; Order Phyllachorales; and Family Phyllachoraceae. The anamorphs are *Glomerella* species. Anthracnose of chilli was first reported from New Jersey, USA, by Halsted (1890) in 1890 who described the causal agents as *Gloeopsorium piperatum* and *Colletotrichum nigrum*. These taxa were then considered as synonyms of *C. gloeosporioides* by von Arx (1957). Anthracnose causes extensive pre- and postharvest damage to chilli fruits causing anthracnose lesions. Even small anthracnose lesions on chilli fruits reduce their marketable value (Manandhar *et al.*, 1995). Many post-harvest diseases of fruit exhibit the

phenomenon of quiescence in which symptoms do not develop until the fruit ripens. *Colletotrichum* species are the most important pathogens that cause latent infection (Jeffries et al., 1990).

The objective was to evaluate the metabolites from *Lepiota procera* to control *Colletotrichum capsici* causing anthracnose of chilli.

Materials and methods

Poisonous mushroom- *Lepiota procera* was used in this study.

Isolation of pathogen and pathogenicity test

Colletotrichum capsici causing anthracnose of chilli was isolated from leaf symptom by tissue transplanting techniques and performed pathogenicity test followed Koch's Postulate.

Extraction of biological active substances

The bioactive compounds were extracted of *Lepiota procera* were cultured in potato dextrose broth (PDB) at room temperature (28-30 C) for 45 days. Fungal biomass were collected by moving from PDB, filtered through cheesecloth and air-dried overnight. Fresh and dried fungal biomass was recorded. Dried fungal biomass were ground with electrical blender, extracted with 200 ml hexane (H) and shaken for 24 hour at room temperature. The filtrate from ground biomass was separated by filtration through Whatman No.4 filter paper. The filtrate was evaporated in vacuo to yield crude extract. The marc was further extracted with ethyl acetace (EtOAc) and methanol (MeOH) respectively using the same procedure as hexane. Each crude extract was weighted, and then kept in refrigerator at 4 C until use.

Biological activity against anthracnose of chilli caused by C. capsici

The crude extracts were tested for inhibition of the most aggressive isolate of C. *capsici*. The experiment was conducted by using 3 x 6 factorial in Completely Randomized Design (CRD) with four replications. Factor A represented crude extracts which consisted of crude hexane, crude ethyl acetate and crude methanol and factor B represented concentrations 0, 10, 50, 100 and/or 500, and 1,000 μg/ml. Each crude extract was dissolved in 2% dimethyl sulfoxide (DMSO), then mixed into potato dextrose agar (PDA) before autoclaving at 121C, 15 1bs/inch2 for 30 minutes. The tested pathogen were cultured on PDA and incubated at room temperature for 5 days, and then colony margin was cut by 3 mm diameter sterilized cork borer. The agar plug of pathogen was transferred to the middle of PDA plate

(5.0 cm diameter) in each concentration and incubated at room temperature (28-30C) for 5 days. Data were collected as colony diameter and computed the percentage of inhibition. Data were statistically computed analysis of variance. Treatment means were compared with DMRT at P=0.05 and P=0.01.

Results

Result showed that methanol crude extract from *Lepiota procera* gave significantly highest inhibition of 46.40 % for the colony growth of C. *capsici* at the concentration of 1,000 ppm when compared to the control. Crude methanol from *Lepiota procera* gave significantly highest inhibited the spore production of C. *capsici* as 80.63 % (Fig.1) and followed by crude hexane 66.45 % and crude ethyl acetate inhibited 57.09 % (Tables 1).

Table 1. Crude extracts of *Lepiota procera* testing for Colonydiameter and Spore production inhibition of *Colletotrichum capsici*

Crude extracts	Concentration (ppm)	Colonydiameter (cm)	Growth inhibition(%)	Number of spores (10^{x6})	Growth inhibition(%)
	0	5.00	0.00	3.16	0
	10	4.88	2.40	2.87	9.17
Crude Hexane	50	4.78	4.40	2.27	28.16
	100	4.45	5.00	2.09	33.86
	500	4.59	8.20	1.87	40.82
	1000	4.33	13.40	1.06	66.45
	0	5.00	0.00	3.17	0
	10	4.85	4.00	3.08	2.83
Crude EtOAc	50	4.75	5.00	2.37	25.23
	100	4.65	7.00	2.12	33.12
	500	4.50	10.00	1.87	41.09
	1000	4.20	16.00	1.36	57.09
	0	5.00	0.00	3.15	0
	10	4.78	4.40	2.64	16.19
Crude MeOH	50	4.53	9.40	2.33	26.03
	100	4.15	17.00	2.18	30.79
	500	3.80	24.00	1.66	47.30
	1000	2.68	46.40	0.61	80.63
C.V.%		3.96	11.53	3.96	93.78

/1 Average of four replications. Means followed by a common letter are not significantly differed by DMRT at P=0.01.

/2 Inhibition (%)=R1-R2/R1x100 where R1 was colony diameter of pathogen in control and R2 was colony diameter of pathogen in treated plates.

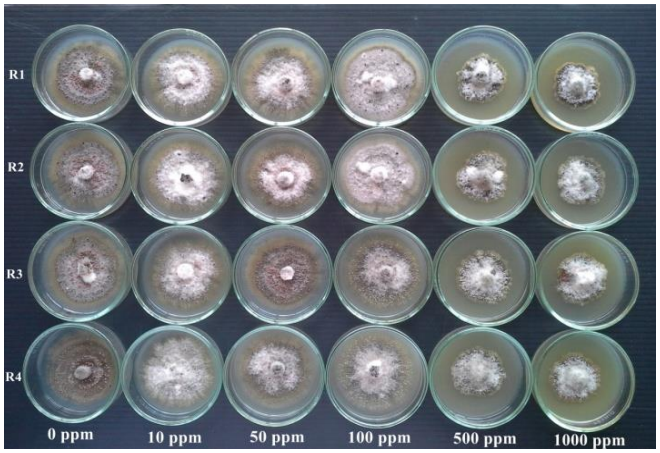

Fig 1. Biological activity against anthracnose of chilli caused by C. capsici on PDA with crude methanol (MeOH) from Lepiota procera

Discussion

Result showed that crude extracts from *Lepiota procera* at concentration 10, 50, 100, 500 and 1000 ppm. to control chilli anthracnose causing by *Colletotrichum capsici* at concentration of 1000 ppm gave the highest percent inhibition of colony of C. *capsici* causing chilli anthracnose which 46.40 percent with this, percent inhibition of spore production were 80.63 percent. Similar report from Nimmomgkol P.(2004) stated that the crude extracts against *Colletrichum dematium* causing anthracnose of chilli at concentration of 1000 ppm. Gave the highest percent inhibition of colony of *C. dematium* causing chilli anthracnose which 18.75 percent and inhibition of spore production were 30.56 percent.

Acknowledgements

The authors wish to acknowledge the support of Faculty of Agricultural Technology, KMITL, Bangkok, Thailand for funding this research.

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