

ISSR assays. Forty microsatellite primers (15- to 22-mer) from the University of British Columbia (Canada) were used in the study. A PCR reaction of 25 μ l contained similar ingredients as the above RAPD reactions except for primer and DNA polymerase, which were 0.2 μ M and 0.5 unit respectively. The PCR reaction was performed with a thermal program of 35 cycles of 94°C for 1 min, 50°C for 1min and 72°C for 2 min, with a final extension at 72°C for 7 min. Amplified products were separated in agarose electrophoresis as described in RAPD assays.

RESULTS

RAPD analysis

All primers tested successfully amplified the *Capsicum* DNA from all varieties. Twelve, two and three primers were able to detect DNA variations between parents of CMH#1, CMH#2, and CMH#3, respectively (Table II). Of the 86 amplified products generated from the 12 primers used for CMH#1, 48 were polymorphic and 38 were used as markers to differentiate the two parental genomes for CMH#1 purity test. There were five and six markers able to detect CMH#2 and CMH#3 hybridity, respectively (Table II). Interestingly, one RAPD primer AG14 detected hybridity of the three F₁ hybrids (Figure 1). Two primers AI02 and Y02 detected two F₁ hybridity. AI02 detected hybridity of CMH#1 and CMH#3, while Y02 detected that of CMH#1 and CMH#2.

ISSR analysis

Of the 40 UBC primers screened, 17 successfully amplified all the *Capsicum* DNA, of which only five detected polymorphisms between both parents of CMH#1, and three for CMH#3 (Table III). However, no primers were able to detect polymorphisms between parents for CMH#2. The five primers for CMH#1 namely UBC812, UBC842, UBC864,

UBC880 and UBC886 produced 26 bands in total, of which eight were markers detecting female parent 'Bangchang' and six were markers detecting male parent 'CM021'. The sizes of these 14 markers ranged from 300 to 1605 bp.

Markers for CMH#3 were generated from three primers including UBC867, UBC880 and UBC887 (Table III), which produced seven markers out of 25 bands in total (Table IV). UBC867 failed to detect a marker for male parent 'KKU-cluster', therefore only UBC880 and UBC887 were useful to test CMH#3 purity.

DISCUSSION

Among the three *Capsicum* hybrids, CMH#1 appeared to have the greatest number of polymorphic markers generated from both RAPD and ISSR analysis for hybrid purity test. This was most likely due to a greater genetic diversity of the parents than the other two hybrids. CMH#1 was derived from an interspecific cross between *C. annuum* X *C. chinense* parents, while CMH#2 and CMH#3 were from intraspecific crosses between *C. chinense* and *C. annuum*, respectively. When comparing polymorphisms between the parents that produced the intraspecific hybrids CMH#2 and CMH#3, the level of polymorphism appeared to be higher in CMH#3 than in CMH#2, which indicated that *C. annuum* '83-168' and 'KKU-cluster' were more diverse than 'CM021' and 'CM022' of *C. chinense*. The level of marker polymorphisms was also agreeable with their morphological attributes. Greater different characteristics between 'KKU-cluster' and '83-168' were identified than those between 'CM021' and 'CM022' (unpublished data).

In comparison of the two marker techniques for their ability to detect hybrid genetic purity, RAPD was able to detect all three F₁ hybridity, while ISSR failed to detect CMH#2 hybridity. Furthermore, RAPD produced a greater number of markers as well as average marker number per primer than ISSR.

Basically, RAPD and ISSR detect DNA variations at different parts of the genome. RAPD primers target randomly genome wide, while ISSR primers target regions between repetitive sequences or microsatellites. Of the six successful ISSR primers four were two-base repeats containing GA and CT, and two were 3- and 5-base repeats. Previous studies also reported that two-base repeats were effective in generating polymorphisms among different varieties in crop plants. CA and GA repeats were useful to generate markers in soybean (Wang *et al.*, 1998). Further example, two-base repeats were examined in Asiatic hybrid lily and reported that they successfully produced ISSR fingerprints with CT the most effective primers (Yamagishi *et al.*, 2002).

ACKNOWLEDGEMENT

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TABLE I

Capsicum hybrids and their respective parents

Hybrid	Female parent (P_1)	Male parent (P_2)
CMH#1	<i>C. annuum</i> 'Bangchang'	<i>C. chinense</i> 'CM021'
CMH#2	<i>C. chinense</i> 'CM021'	<i>C. chinense</i> 'CM022'
CMH#3	<i>C. annuum</i> '83-168'	<i>C. annuum</i> 'KKU-cluster'

TABLE II

Successful RAPD markers for genetic purity proof of the three Capsicum hybrids

Hybrid	Primer	Marker size (bp)	
		P ₁	P ₂
CMH#1	AE19	540	896
	AG14	1,159	804
		873	211
		694	
	AG16	845	791
	AI01	741	504
		348	
	AI02	903	488
		817	400
	AT07	393	367
	AU03	672	753
		480	
	AX01	831	662
		611	
	C1	1,397	672
		1,140	573
		947	
		400	
	Q05	1,146	479
	R12	364	520
		344	
	Y02	520	434
			300
CMH#2	AG14	350	749
			683
	Y02	863	1,030

TABLE II

Continued

CM11#3	AG14	200	969
	AI02	758	705
	AN19	640	705

TABLE III

Successful ISSR markers for genetic purity proof of the three Capsicum hybrids

Hybrid	Primer and sequence	Marker size (bp)	
		P ₁	P ₂
CMH#1	UBC812	1,144	300
	(GA) ₈ A		
	UBC842	947	857
	(GA) ₈ (CT)* G	701	
		501	
	UBC864	844	674
	(ATG) ₆		
	UBC880	964	657
	(GGAGA) ₃		
	UBC886	1,605	1,440
	(ACG)*(AGT)*(ACG)*(CT) ₇	500	571
CMH#2	-	-	-
CMH#3	UBC867	2,868	-
	(GGC) ₆		
	UBC880	1,022	912
	UBC887	1,022	882
	(AGT)*(ACG)*(AGT)*(CT) ₇		760
			500

* indicates either one of the three bases in the bracket were used.

TABLE IV

Successful markers generated from both RAPD and ISSR for genetic purity proof of the three Capsicum hybrids

Hybrid	Number of markers in P ₁			Number of markers in P ₂		
	RAPD	ISSR	Total	RAPD	ISSR	Total
CMH#1	22	8	30	16	6	22
CMH#2	2	0	2	3	0	3
CMH#3	3	3	6	3	4	7

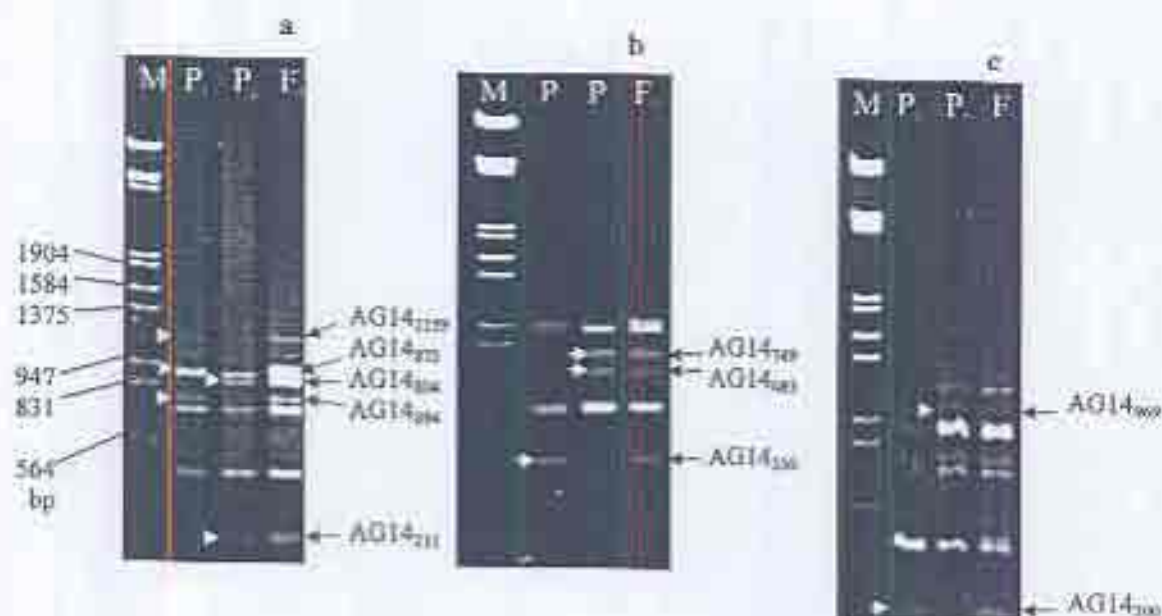


FIG. 1

RAPD analysis of the three *Capsicum* hybrids including a) CMH#1, b) CMH#2, and c) CMH#3 amplified by primer AG14; arrows indicate polymorphic markers presenting in both parents (P_1 and P_2) and their resulting F_1 . DNA fragment sizes were compared with molecular weight marker M (*Eco*R I + *Hind* III digested λ DNA).

Please quote No. 234/03

12 December 2003

Dr O Mongkolporn
Dept of Horticulture
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Nakhon Pathom 73140
Thailand

Dear Dr Mongkolporn,

The referee's comments on your paper are as follows

"Paper Ref. 234/03: Genetic purity test of F_1 chili hybrid using molecular analysis. By O. Mongkolporn, Y. Dokmaihom, C. Kanchana-udomkan and P. Pakdeevanaporn.

This is a generally well-presented paper describing a comparison of molecular techniques to detect hybridity in *Capsicum* (chili) F_1 hybrids. I think the genus *Capsicum* should appear in the title. I have indicated some clarification in Fig. 1, but they are fairly clear anyway. I think the results should be of value and that this paper is suitable for publication, subject to minor revision.

"1. Specific amendments to correct errors

Add *et al* to a couple of citations on p.3.

Correct '500' to '300' on p.6.

Insert reference to Table IV on p.6.

"2. Suggestions for the author to consider and accept or reject

Include the name *Capsicum* in the title. Use the form, ' F_1 hybrid chili' in the title.

Consider labelling of specific markers as indicated on Fig. 1.

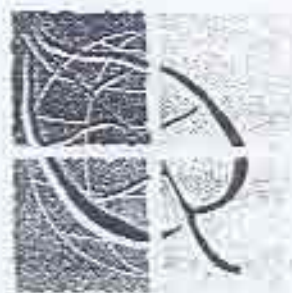
"3. General comments for improvement

Confirm that the marker size 350bp should appear in Table II both for the female parent of CMH#2 and the male parent of CMH#1. Label the female and male parents P_1 and P_2 respectively to facilitate the interpretation of Fig. 1. Clarify statement on p.7 concerning two-base repeats. Give sequence for primer UBC867."

With the above minor corrections and clarifications, the paper should be acceptable.

Yours sincerely,

Editor



Department of Horticulture, KU

24 December 2003

Dr A. R. Rees,
Editor Journal of Horticultural Science & Biotechnology
PO Box 3015, Littlehampton
West Sussex, BN16 3SX UK

Dear Dr Rees,

Refer to the manuscript number 234/03

Thank you for the acceptance of our manuscript titled 'Genetic purity test of F_1 chili hybrid using molecular analysis'. We would like to express a great appreciation to the reviewer's comments and suggestion. We have amended the manuscript following his comments. Details are as follows.

1. Correction of errors:

- *et al.* has been added to Koller... and Lu... on page 3.
- 300 bp has been replaced 500 bp on page 6.
- Reference for Table IV has been added on page 6.

2. Suggestions:

- *Capiscum* has been included in the title, as seen in the current form.
- Specific markers have been labelled.

3. General comments:

- The marker of 350 bp was generated from primer AG14 and was present in the parent 'CM021'. However, this marker was monomorphic between P_1 (Bangchang) and P_2 (CM021) of the hybrid CMH#1. Therefore, the marker appeared in Table II for only P_1 (CM021) of the CMH#2 (please see Fig 1 for details of the labelled markers). We insist the data as originally presented is correct.
- P_1 and P_2 have replaced in all Tables.
- Usefulness of the two-base repeats has been clarified on page 7.
- The sequence of the primer UBC867 has been supplied.

Enclosed please find a hard copy of the amended manuscript with a floppy diskette containing three files including text, tables and figure.

Yours sincerely,

Dr Orarat Mongkolporn
Lecturer

Development of a bioassay to study anthracnose infection of chili fruit caused by

Colletotrichum capsici

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Abstract. A fruit inoculation bioassay was developed for studying anthracnose infection of chili by *Colletotrichum capsici*. Four fruit maturity stages, immature green, mature green, color turning and ripe red, and three inoculation methods, drop, injection and wound/drop, were applied to an anthracnose susceptible variety 'PBC4714' of *Capiscum chinense* Jacq. Injection and wound/drop methods resulted in anthracnose symptoms developing at all fruit stages as early as 3-5 days after inoculation, while the drop method failed to cause any symptoms of anthracnose within 9 days after inoculation. The mature and immature green chili fruits appeared to be more susceptible to anthracnose than the more mature fruit stages. All the disease score parameters including lesion length, lesion width, lesion area and area under the disease progress curve were highly correlated indicating that any of the disease evaluation methods could be used to assess germplasm for resistance.

Additional keywords: *Capiscum chinense* Jacq., fruit maturity, inoculation method, disease scores

Introduction

Chili is an economically important vegetable crop of Thailand with anthracnose disease being a major constraint to pre- and post-harvested chili fruits. Anthracnose is caused by either *Colletotrichum capsici* (Syd.) E.J. Butler & Bisby or *C. gloeosporioides* (Penz.) Penz. & Sacc. in Penz. *C. capsici* has also been reported to infect a wide range of legume species (Pring *et al.*, 1995).

Fruit maturity stage has been shown to be important in the infection and colonisation of chili fruit by *C. gloeosporioides* with red fruit being more resistant than green fruit (Oh *et al.*, 1999a; Oh *et al.*, 1999b; Kim *et al.*, 2001; Kim *et al.*, 2002). However, Manandhar *et al.* (1995) reported that both ripe and green chili fruits reacted the same to either *C. capsici* or *C. gloeosporioides*.

Three laboratory inoculation methods (injection, drop and wound/drop) for studying anthracnose diseases of chili have been developed. The injection and drop methods were developed at the Asian Vegetable Research and Development Center (AVRDC), Taiwan (AVRDC Report 1997, unpublished data). The injection method involved the direct penetration of the chili fruit epidermis with a microinjector followed by inoculation with a fungal spore suspension. The drop method involved placing a fungal spore suspension on the surface of the chili fruit. The wound and drop method (Lin *et al.*, 2002) involved wounding the surface of the fruit by pricking with a pin then placing a drop of fungal spore suspension over the wounded tissue.

The injection method is considered to be harsher than the drop method as chili fruits are wounded before inoculation. The injection method has been used to detect anthracnose resistance in a few chili varieties (AVRDC Report 1998, unpublished data).

However, the injection method might be too severe for other chili varieties, which did not contain high resistance, and hence, may not be able to differentiate between moderate resistance and susceptible phenotypes in some parental lines. The wound/drop method was able to differentiate moderately resistant from susceptible varieties (Lin *et al.*, 2002).

This study aimed to investigate the effect of different chili fruit maturity stages and inoculation methods on anthracnose expression caused by *C. capsici*, and to develop an anthracnose disease scoring system for assessing chili host reaction to infection by *C. capsici*.

Methods

Fruit maturity stages and pathogen isolate

Capiscum chinense Jacq. 'PBC4714', an anthracnose susceptible line, was grown in a shade house between June and November 2002 at the Tropical Vegetable Research Center (TVRC), Kasetsart University, Kamphaengsaeu Campus, Thailand. Three fruits each at four different maturity stages, including immature green (30 days after flowering, DAF), mature green (35 DAF), color turning (40 DAF) and ripe red (45 DAF), were collected from the chili plants. Pedicels and calyces were removed from the harvested fruits. Fruits were surface sterilised with 1% (v/v) sodium hypochlorite for 5 min, and washed twice with distilled water. The fruits were wiped dry with paper towel and placed on a polystyrene tray that was then placed in a plastic box of 20 x 30 x 10 cm³, that contained 500 ml distilled water.

A highly aggressive single-spore isolate of *Colletotrichum capsici*, isolated from infected fruit collected from the field at Kasetsart University, was cultured on

potato dextrose agar at 27°C under continuous fluorescent light. Conidia from seven-day-old cultures were harvested by adding 5-10 ml of sterilised distilled water onto the culture, which was then gently swirled to dislodge the conidia. The conidia suspension was filtered through two layers of muslin cloth and the concentration was adjusted to 10^6 conidia/ml.

Inoculation method

The fruits were inoculated using the drop, injection and the wound/drop methods (Lin *et al.*, 2002). The drop method was executed by placing 10 µl of conidia suspension in the middle portion of each chili fruit. The injection method was accomplished by injecting 1 µl of spore suspension into a chili fruit wall at 1 mm depth using an injector Micro Syringe™ model 1705 TLL with dispenser PB600-1 (Hamilton, Switzerland). The wound/drop method was conducted by pin pricking the chili fruit wall to a 1 mm depth and then placing 6 µl of conidia suspension onto the pin-pricked wound. The inoculated fruits were incubated at 25°C, 100% RH and dark for 24 hrs and then 12-12 hrs light-dark cycle. Humidity was not controlled after symptoms started to appear.

Evaluation of anthracnose symptoms

Disease symptoms were evaluated by measuring lesion area, length and width, and area under the disease progress curve (AUC). The AUC was calculated from the relationship between lesion area and inoculation time. The symptoms were evaluated at 3, 5, 7 and 9 days after inoculation (DAI). Fruit sizes were also recorded.

Statistical analysis

A factorial analysis in complete randomised design was performed, considering two factors as follows: 1) inoculation methods including drop, injection and wound/drop; and 2) chili fruit maturity including immature green, mature green, color turning and ripe red. Statistical analysis of variance (ANOVA) and correlation analysis were performed using Statistical Analysis Software (SAS) for WindowTM release 6.12.

Results and Discussion

Development of anthracnose on different fruit maturity stages

The severe injection inoculation method was selected to compare the development of anthracnose symptoms at different fruit maturity stages. Lesion area (LA) measured at all chili fruit stages were not significantly ($P=0.05$) different from 3 to 5 DAI except for the mature green stage where the lesions were significantly larger than for the ripe red fruit stage at 5 DAI. At 7 and 9 DAI, mature green fruit showed the largest LA however, the LA was not significantly different from that of immature green fruits (Fig. 1).

These results suggested that mature green fruits were more susceptible to anthracnose than the more mature fruits. Similarly, in previous studies, red chili fruit were shown to be more resistant to infection by *C. gloeosporioides* than unripe mature green chili fruit (Oh *et al.*, 1999a; Oh *et al.*, 1999b; Kim *et al.*, 2001; Kim *et al.*, 2002). In contrast, Manandhar *et al.*, (1995) showed that chili fruit infected by *C. capsici* at both green and ripe fruit stages reacted the same and developed similar size symptoms.

Development of anthracnose by different inoculation methods

The three inoculation methods caused different severity levels of anthracnose on chili fruits. The injection and wound/drop methods showed similar trends of anthracnose development, where the lesion area of all fruit stages gradually increased from 3 to 5 DAI and dramatically increased after 5 DAI (Fig. 1). In contrast, the drop method failed to cause anthracnose symptoms at all chili fruit stages up to 9 DAI.

In comparing the injection and wound/drop method, there was no difference in the degree of anthracnose incidence on the chili fruit measuring by average lesion area at 9 DAI. However, the injection appeared to be a more severe method, because it could cause infection to all chili fruits, and the first visual symptoms were detected as early as 3 DAI. While no visual symptoms were detected at 3 DAI, when inoculated by the wound/drop method. The earliest appearance of the first symptom by wound/drop detected on mature green was at 5 DAI.

Wounding is a key factor to accelerate infection process. Cuticular wax layers of plants are one of the first barriers to fungal infection. The initial infection processes of *Colletotrichum* species included conidia germination, appressoria and infection hyphae formation, which were necessary for subsequent cuticular penetration through the host surface (Bailey *et al.*, 1992). Pring *et al.* (1995) reported that when host tissues of a range of legumes (*Cicer arctium*, *Lupinus angustifolius*, *Pisum sativum*, *Vigna radiata* and *V. unguiculata*), were wound inoculated with *C. capsici*, symptoms appeared approximately 4-6 days earlier than unwounded inoculation.

The lack of infection within the nine day post inoculation period using the drop method may have been due to the physical barrier of the cuticular wax layer on chili fruit at all maturity stages which prevented or delayed the direct infection by the

pathogen. If the inoculated fruit were incubated for longer, symptoms could have possibly developed however, detached chili fruits started to senesce after 9 days under laboratory conditions thus it was not practical to assess symptom development after 9 days.

Comparison of anthracnose evaluation parameters

Correlation analysis of four anthracnose evaluation parameters including, lesion length (LL), lesion width (LW), lesion area (LA), average of LL and LW (AL) and area under the disease progress curve (AUC) was performed using mature green fruits and the injection method. All anthracnose evaluation parameters were highly correlated and highly significant ($r^2 > 0.9$, $P < 0.01$), implying that any of these parameters could be used in a breeding program to evaluate resistance to anthracnose phenotype. The simplest and most convenient evaluation method was measuring lesion length.

In conclusion, only the injection and wound/drop methods caused anthracnose symptoms on chili fruit at all stages of fruit ripening, with the injection method being slightly more severe than the wound/drop method. Green chili fruits appeared to be more susceptible to anthracnose than the more mature fruits. All disease scoring parameters including lesion length, lesion width, lesion area, average of lesion length and lesion width, and area under curve, were highly correlated.

Acknowledgements

This research was partly funded by the Kasetsart University Research and Development Institute and the Thailand Research Fund. Field and laboratory experiments were facilitated by the Tropical Vegetable Research Center and the Asian Regional Center/Asian Vegetable Research and Development Center, respectively. The

authors would like to thank Associate Professor Dr Somsiri Sangchote, Department of Plant Pathology, Kasetsart University for the supply of *Colletotrichum capsici* isolate number 158ci, and the Asian Vegetable Research and Development Center for the chili germplasm used in this study.

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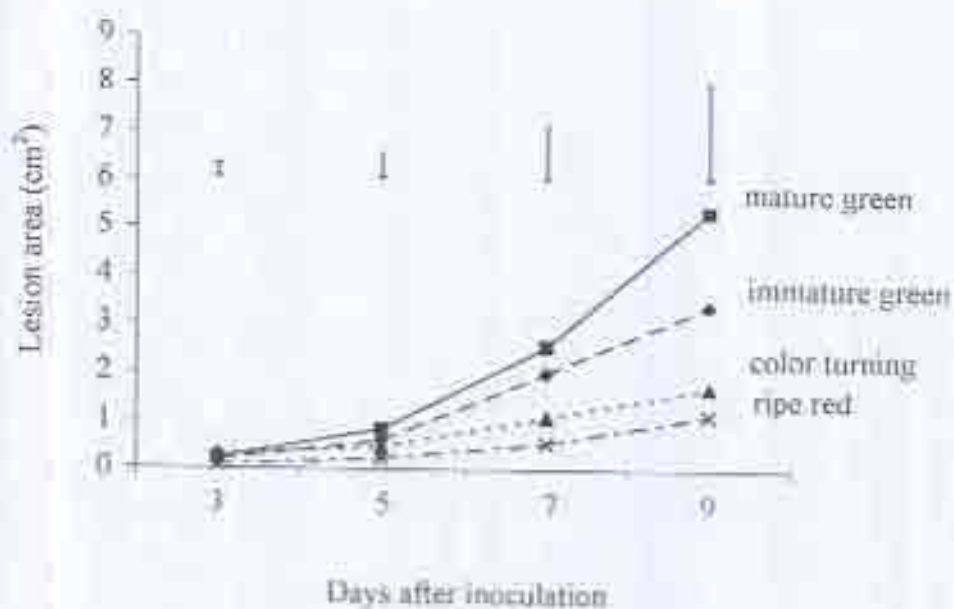


Fig. 1 Development of anthracnose symptoms on *Capsicum chinense* 'CM022' as measured by lesion area on four different chili fruit maturity at 3, 5, 7 and 9 days after inoculation inoculated by injection method. Bars at each time represent LSD at $p=0.05$

SUMMARY OF TPS 2002

The First International Conference on Tropical and
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A Laboratory Evaluation for Resistance to Anthracnose in Chili

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Abstract

A laboratory evaluation of resistance to chili anthracnose (*Colletotrichum capsici*) is being developed. The study was aimed to identify a suitable chili fruit maturity stage, a suitable inoculation method and a disease scoring system, to be employed in our chili breeding program for anthracnose resistance. The results showed that mature green chili fruits exhibited the most severe symptoms. Injection method accelerated the symptoms four days faster the drop method. Disease evaluation by lesion sizes and lesion area/fruit size ratio was highly correlated.

Introduction

Anthracnose (*Colletotrichum* spp.) is one of the major diseases of chili in Thailand. Two causal pathogen species, *capsici* and *gloeosporioides*, have been identified in Thailand, where *C. capsici* is found to be predominant (Sangchote, 1999). A breeding program to improve resistance to anthracnose in chili has been established. In a selection process for disease resistance, bioassay plays a key role in resistance evaluation, which could be done through either a field- or laboratory-based method. A field evaluation is disadvantageous because it is less likely to control environmental factors affecting the disease expression, while the laboratory method is opposite. To study genetic control of disease resistance, it is important that phenotype expression truly reflect their genotypes. Therefore, the more controlled laboratory evaluation has been developed to be used in our chili breeding program. The aims of this study were to develop a suitable inoculation method; to identify a suitable fruit maturity stage; and to develop disease scoring system for chili fruit anthracnose, for the evaluation of resistance to anthracnose.

Materials and Methods

Plant material Four varieties of chili including *Capsicum annuum* L. cv. 'Bangchang', *C. chinensis* 'CM021', *C. chinensis* 'CM022', F₁ ('CM021' x 'CM022') and F₁ ('Bangchang' x 'CM021') were used in the study. Five fruits of each four different maturity stages including immature green, mature green, colour turning and ripe red, were harvested from all chili varieties. The harvested fruits were surface sterilized with 1% sodium hypochlorite.

Inoculation methods A highly aggressive monoconidial isolate 158ci of *Colletotrichum capsici* was used as causal pathogen. Two inoculation methods were used: drop inoculation by dropping 10 µl of spore suspension at 5 x 10⁶ conidia/ml, and injection by inject 1 µl of spore suspension at 1 x 10⁶ conidia/ml into chili fruit epidermis at 1 mm depth. The inoculated fruits were incubated at 25°C, 100% RH and dark for 24 hrs and then 12-12 hrs light-dark (AVRDC, 1998).

Disease evaluation Disease symptoms were evaluated between 3-9 days after inoculation (DAI) by measuring lesion length, width and area at the inoculation sites. Fruit sizes were also recorded.

Results and Discussion

Chili fruits at mature green stage showed the most severe symptoms after inoculated with *C. capsici*. Disease symptoms by injection appeared at three DAI, which was four days faster than that by the drop inoculation. All of the parameters used to evaluate anthracnose were highly correlated (Table 1).

Table 1 Correlation between the parameters evaluating chili anthracnose at 8 DAI

Param. abbrev.	Lesion length (mm)	Lesion	Lesion width (mm)	Lesion width/fruit width	Lesion area (mm ²)
abbr.		length/lesion length			
Coefficient	0.9374	0.9120	0.9522	0.9302	0.9149
Probability	0.0010	0.0001	0.0001	0.0001	0.0003

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A Novel Source of Resistance to Anthracnose in Chilli

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Abstract

Anthracnose (*Colletotrichum capsici*) is one of the most severe diseases of tropical chilli (*Capiscum* spp.). The inheritance of resistance to anthracnose was investigated in F_1 , F_2 and BC_1 from a cross between a resistant *C. chinense* and a susceptible commercial cv. 'Bangchang'. The resistance appeared to be controlled by a single recessive gene.

Introduction

Anthracnose (*Colletotrichum* spp.) is an economically important disease of chilli production in Thailand. Under conditions favorable to the disease development, pre- and postharvest fruit losses of over 50% have been reported. Two significant causal pathogens found in Thailand are *C. capsici* (Syl.) Butler & Bisby and *C. gloeosporioides* Pertz (Sangchote, 1999). Up to date no high resistance have been found in *C. annuum* L. which is the only chilli species widely grown in Thailand and worldwide.

High resistance to anthracnose has been identified in an accession of *C. chinense* (AVRDC, 1998). Our preliminary study have exhibited this accession was resistant to both species of *Colletotrichum*. Characterisation of the resistance genes in this *C. chinense* genotype is important as they may be sources of a novel resistance, which may be transferred and incorporated to Thai elite chilli to develop anthracnose resistant genotypes.

This paper reports preliminary genetic study of anthracnose resistance that expressed in the interspecific cross of Thai susceptible *C. annuum* cv. 'Bangchang' and an anthracnose resistant *C. chinense*.

Materials and Methods

Plant materials: Five fruits at mature green stage were harvested from each plant of chilli populations including parents, F_1 , F_2 and BC_1 of the cross resistant *C. chinense* x cv. 'Bangchang'.

Bioassays and disease evaluation: A highly aggressive monoklonal isolate 158c of *C. capsici* was prepared at concentration of the 10^6 conidia/ml. The harvested fruits were surface sterilised with 10% sodium hypochlorite and then inoculated by injection method (AVRDC, 1998). The inoculated fruits were incubated at 25°C, 100% RH with darkness for 24 hrs, then 12 hrs light was added. Length and area of disease lesion were recorded at 3, 5, 7 and 9 days after inoculation (DAI).

Results and Discussion

All F_1 (Bangchang x *C. Chinense*) plants appeared to be susceptible indicating that the resistance was likely to be a recessive trait. However, maternal effect will be further investigated in the reciprocal F_1 (*C. chinense* x Bangchang). The observed phenotypic segregation in the F_2 and BC_1 well fitted with the expected ratio of 1:3 and 0:1 for the susceptibility:resistance, respectively (Table 1). These segregation ratios indicated that a single recessive gene conferred the resistance to anthracnose derived from the *C. chinense*.

Table 1 Segregation of phenotypic resistance and susceptibility to anthracnose caused by *Colletotrichum capsici* in F_1 , F_2 and backcross chilli populations of the cross *C. chinense* x Bangchang, as evaluated by contingency Chi-square values for goodness-of-fit to Mendelian inheritance ratios.

Population	No. of plants inoculated	Phenotypes		Expected ratio	χ^2	Probability
		Resistant	Susceptible			
<i>C. Chinense</i>	5	5	0	-	-	-
Bangchang	12	0	12	-	-	-
F_1 (Bangchang x <i>C. Chinense</i>)	12	0	12	-	-	-
F_2	30	9	21	1:3	0.4	0.91
BC_1 (F_1 x Bangchang)	16	0	16	0:1	not available	not available

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