



UNIVERSITI PUTRA MALAYSIA

**DISTRIBUTION AND CHARACTERIZATION OF DISEASES OF
DRAGON FRUIT (*Hylocereus* SPP.) IN PENINSULAR MALAYSIA**

**MASANTO MASYAHIT
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By

MASANTO MASYAHIT

**Thesis Submitted to the School of Graduate Studies, University Putra Malaysia,
in Fulfilment of the Requirements for the Degree of Master of Science**

October 2010



Dedication

“I dedicated this thesis to my beloved families

As a written evidence which

Proved that I had been able to do the useful thing

Because of

Their struggle obstinacy

Which wishful me

Keep going to

Reach my dream “

Abstract of thesis presented to the Senate of University Putra Malaysia in fulfilment of the requirement for the degree of Master of Science

DISTRIBUTION AND CHARACTERIZATION OF DISEASES OF DRAGON FRUIT (*Hylocereus* SPP.) IN PENINSULAR MALAYSIA

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October 2010

Chair : Kamaruzaman Sijam, PhD

Faculty : Agriculture

Field surveys were conducted from December 2007 to August 2008 at 43 dragon fruit-orchards in Peninsular Malaysia with 50 posts of sampled plants per location. The results revealed that among recorded diseases, anthracnose, brown spot and necrotic spot were encountered in almost all sampled states with the range of frequency around 50 to 100%; whereas fruit brown rot was only found in Negeri Sembilan with frequency of 33.33%. Meanwhile, other diseases such as fruit fungal soft rot, stem end rot and bacterial soft rot occurred in 2, 5 and 6 surveyed states, respectively. Statistical analysis showed that the occurrence of anthracnose and necrotic spot diseases were maximum in Malacca with incidence and severity level around 58.00%; 21.20% and 72.00%; 30.00%, respectively. Meanwhile, the maximum occurrences of bacterial soft rot, brown spot and stem end rot were documented in Johore, Negeri Sembilan and Kelantan with incidence and severity levels of about 17.33%; 4.53%, 94.00%; 25.87%, and 14.00%; 4.20%, respectively. These diseases have strong relationships of incidence and severity with the range of



R^2 value around 0.8995 to 0.9978. Pearson correlation analysis resulted in the incidence of anthracnose which was significantly correlated with necrotic spot (r value of 0.852 at 0.05 level); whereas bacterial soft rot and brown spot were significantly interrelated with each other at the 0.01 level with r value of 0.515. Similarly, the Pearson coefficient correlation also revealed that bacterial soft rot was negatively correlated with temperature (r value of -0.478 at 0.01 level). The positive correlations were shown by bacterial soft rot and brown spot against altitude with r value of 0.508 and 0.523, respectively at 0.01 level. These diseases were categorized according to the characteristics of symptoms found on diseased plants and group of pathogenic agents. Although a number of bacteria and fungi species could be isolated and identified correspondingly based on their responses to biochemical sources within BiOLOG® Microplate as well as their cultural and morphological characteristics, the pathogenicity test showed that only some (e.g. *Enterobacter cloacae*, *Bipolaris* sp., *Botryosphaeria* sp., *Colletotrichum gloeosporioides*, and *Monilinia* sp.) positively resulted in similar symptoms with diseased plants in the field. In the mean time, the presence of viral infection was successfully detected on young stems with necrotic small mottle or spot symptoms. The spindle-shaped inclusion bodies of filamentous and rod-shaped *Cactus virus X* were observed at 4,000 and 20,000 X magnifications under transmission electronic microscopy (TEM). The findings derived from *in vitro* assays demonstrated that a temperature of 35°C could inhibit the colony diameter of the tested fungi up to around 7.36 mm for *Pestalotiopsis* sp. to 34.63 mm for *Fusarium* sp. during the final incubation period. Although pH 4 might restrict the colony diameter around 43.71 mm for *Botryosphaeria* sp. and 74.87 mm for *Colletotrichum gloeosporioides* on the last day of incubation, no significant effect of all pH levels against *Monilinia* sp. was found

after 4 days of incubation (DAI). For the effect of salinity, only the colony growth of *Bipolaris* sp. and *Botryosphaeria* sp. was affected by 100 ppm of salinity level up to 57.50 and 68.26 mm in diameter, respectively at 10 DAI; whereas other fungi could grow well under all salinity treatments. Meanwhile, the test carried out for antagonistic bacteria revealed that the ability of *Burkholderia cepacia*, *B. multivorans* and *Pseudomonas aeruginosa* in inhibiting the growth of *Bipolaris* sp., *C. gloeosporioides* and *Pestalotiopsis* sp. fungi was almost maximum at 8 DAI ranging from 61.80 to 85.11%. The effect of the employed bacteria was also found to be maximum against *Fusarium* sp. at 6 DAI with the range of inhibition percentage around 74.79 to 80.67% and this was maximum against *Botryosphaeria* sp. during the final incubation period with the range of 50.18 to 65.63%. The most maximum inhibition was shown by *B. cepacia* against *Monilinia* sp. since the first incubation period. In conclusion, anthracnose, brown spot and necrotic spot were three predominant diseases which have been nationwide found to occur on dragon fruit in Peninsular Malaysia. Statistically, temperature was found to negatively influence the occurrence of bacterial soft rot disease; whereas the altitude was positively correlated with the incidence of bacterial soft rot and brown spot diseases. Some fungal, bacterial and viral plant pathogenic have been proven as the causal agents of these diseases which mostly infected stem and fruit. Under *in vitro* condition, the colony growth of most fungi was affected by the temperature of 35°C, pH 4 and 100 ppm of NaCl concentration; whereas the maximum inhibition ability of antagonistic bacteria against the tested fungi commenced at 2 and 8 DAI. Good agricultural practices can decrease or even prevent some orchards from definite diseases. In addition, proper combination of environmental modification may be useful for the growth of this crop in the fields and its storage life at post harvest preservation. Nevertheless, further

study is required to investigate the probability of disease-resistant species among the dragon fruits species cultivated in Peninsular Malaysia which are not infected by certain diseases.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia
sebagai memenuhi keperluan untuk ijazah Master Sains

**TABURAN DAN PENCIRIAN PENYAKIT-PENYAKIT PADA BUAH MATA
NAGA (*Hylocereus SPP.*) DI SEMENANJUNG MALAYSIA**

Oleh

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Tinjauan-tinjauan lapangan dilakukan dari Disember 2007 sehingga Ogos 2008 di 43 ladang penanaman buah mata naga di Semenanjung Malaysia dengan sample yang mempunyai 50 tiang tanaman di setiap lokasi. Dapatan kajian menunjukkan bahawa antraknos, tompok coklat dan bintik nekrotik adalah di antara penyakit-penyakit yang tercatat dan dijumpai di hampir semua negeri pensampelan dengan rentang kekerapan sekitar 50 sehingga 100%; manakala penyakit reput coklat pada buah hanya ditemui di Negeri Sembilan dengan kekerapan 33.33%. Penyakit-penyakit lainnya seperti reput lembik coklat pada buah, reput hujung batang dan reput lembik bakteria masing-masingnya berlaku pada 2, 5 dan 6 negeri-negeri yang ditinjau. Analisis statistik menunjukkan bahawa kejadian penyakit antraknos dan bintik nekrotik adalah maksima di Melaka dengan taraf kejadian dan keterukan masing-masingnya sekitar 58.00%; 21.20% dan 72.00%; 30.00%. Sementara itu, kejadian maksima penyakit reput lembik bakteria, tompok coklat dan reput hujung batang dicatat di Johor, Negeri Sembilan dan Kelantan dengan taraf kejadian dan keterukan masing-masingnya sekitar 17.33%; 4.53%, 94.00%; 25.87%, dan 14.00%; 4.20%.

Penyakit-penyakit ini mempunyai hubungan yang kuat antara taraf kejadian dan keterukannya dengan nilai R^2 di antara 0.8995 hingga 0.9978. Sementara itu, analisis pertalian Pearson menunjukkan kejadian antraknos yang secara nyata saling berkaitan dengan bintik nekrotik (nilai r 0.852 pada taraf 0.05); manakala penyakit reput lembik bakteria dan tompok coklat didapati berhubung-kait secara terus dengan satu sama yang lain pada taraf 0.01 dengan nilai r 0.515. Koefisien pertalian Pearson juga menyatakan bahawa penyakit reput lembik bakteria berhubung-kait secara negatif dengan suhu (nilai r -0.478 pada taraf 0.01. Hubungan positif yang ditunjukkan oleh penyakit reput lembik bakteria dan tompok coklat terhadap altitud adalah dengan nilai r masing-masingnya 0.508 dan 0.523 pada taraf 0.01. Penyakit-penyakit ini dikategorikan mengikut ciri-ciri gejala pada tanaman yang sakit dan kelompok ejen-ejen patogenik. Walaupun sejumlah spesies bakteria dan kulat dapat diasingkan dan dikenalpasti secara bersesuaian berasaskan kepada sumber-sumber biokimia dalam Microplate BiOLOG® dan ciri-ciri kultural dan morfologinya, ujian patogenisiti menunjukkan bahawa hanya sesetengah daripadanya seperti *Enterobacter cloacae*, *Bipolaris* sp., *Botryosphaeria* sp., *Colletotrichum gloeosporioides*, dan *Monilinia* sp. menghasilkan gejala yang sama secara positif seperti tanaman-tanaman yang sakit di lapangan. Dalam pada itu, kehadiran jangkitan virus juga telah berjaya dikesan pada batang muda dengan pertanda bintik atau tompok kecil nekrotik. Tubuh-tubuh inklusi berbentuk gelendong daripada *Cactus virus X* yang berbentuk filamen dan batang panjang diamati pada pembesaran 4,000 dan 20,000X dibawah mikroskop transmisi elektronik (TEM). Penemuan-penemuan ujian *in vitro* menunjukkan bahawa suhu 35°C dapat menjejaskan garis tengah koloni kulat yang diuji sehingga berkisar pada 7.36 mm bagi *Pestalotiopsis* sp. dan sehingga 34.63 mm bagi *Fusarium* sp. pada jangkamasa pengeraman

terakhir. Walaupun pH 4 boleh menyekat garis tengah koloni pada kisaran 43.71 mm bagi *Botryosphaeria* sp. dan 74.87 mm bagi *Colletotrichum gloeosporioides* pada hari terakhir pengeraman, tiada kesan nyata daripada semua peringkat pH terhadap *Monilinia* sp. dijumpai daripada semua peringkat pH selepas 4 hari pengeraman (HSP). Untuk kesan paras kandungan garam, hanya pertumbuhan koloni daripada *Bipolaris* sp. dan *Botryosphaeria* sp. didapati telah dipengaruhi oleh paras kandungan garam 100 ppm sehingga masing-masing garis tengah koloninya 57.50 dan 68.26 mm pada 10 HSP; sedangkan kulat lainnya mampu tumbuh dengan baik dibawah semua perlakuan paras kandungan garam berkenaan. Ujian bakteria antagonistik menunjukkan bahawa kemampuan *Burkholderia cepacia*, *B. multivorans* dan *Pseudomonas aeruginosa* dalam menjejaskan pertumbuhan kulat *Bipolaris* sp., *C. gloeosporioides* dan *Pestalotiopsis* sp. yang kebanyakannya maksima pada 8 HSP dengan peratusan di antara 61.80 hingga 85.1%. Kesan bakteria-bakteria yang diuji tersebut juga maksima terhadap *Fusarium* sp. pada 6 HSP dengan peratusan penghambatan sekitar 74.79 hingga 80.67% dan ini juga didapati maksima terhadap *Botryosphaeria* sp. pada pengeraman terakhir dengan peratusan sekitar 50.18 hingga 65.63%. Penghambatan yang paling maksima ditunjukkan oleh *B. cepacia* terhadap *Monilinia* sp. sejak jangkamasa pengeraman pertama. Kesimpulannya, antraknos, tompok coklat dan bintik nekrotik ialah tiga penyakit utama yang terjadi pada buah mata naga secara merata di kesemua negeri di Semenanjung Malaysia. Secara statistik, suhu didapati mempengaruhi kejadian penyakit reput lembik bakteria secara negatif; manakala altitud berhubung-kait secara positif dengan kejadian penyakit reput lembik bakteria dan tompok coklat. Kajian juga membuktikan bahawa beberapa kulat, bakteria dan virus patogenik tumbuhan sebagai ejen-ejen penyebab penyakit-penyakit ini yang kebanyakannya

menjangkiti batang dan buah mata naga. Dibawah kondisi *in vitro*, pertumbuhan koloni kebanyakan kulat dipengaruhi oleh suhu 35°C, pH 4 dan paras kandungan garam 100 ppm; manakala kemampuan penghambatan yang maksima didapati daripada bakteria antagonistik terhadap kulat-kulat yang diuji bermula pada 6 HSP. Oleh itu, amalan-alaman pertanian yang baik boleh menurunkan dan bahkan mencegah penyakit tertentu daripada menyerang sesetengah ladang. Penggabungan pengubahsuaian persekitaran yang tepat juga dapat membantu di dalam pertumbuhan tanaman di lapangan dan jangkamasa penyimpanan buah semasa pengawetan yang dilakukan selepas penuaian. Walau bagaimanapun, kajian lanjut masih diperlukan untuk menyiasat kemungkinan species tahan penyakit atau tidak boleh dijangkiti penyakit-penyakit tertentu di antara semua jenis buah mata naga yang diusahakan di Semenanjung Malaysia.

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I certify that a Thesis Examination Committee has met on 14 October 2009 to conduct the final examination of Masanto Masyahit on his thesis entitled “Distribution and Characterization of Diseases of Dragon Fruit (*Hylocereus Spp.*) in Peninsular Malaysia” in accordance with Universities and University Colleges Act 1971 and the Constitution of Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The Committee recommends that the student be awarded the Master of Science.

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DECLARATION

I declare that the thesis is my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously, and is not concurrently, submitted for any other degree at Universiti Putra Malaysia or at any other institution.

MASANTO MASYAHIT

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