

A brief overview on pitahaya (*Hylocereus* spp.) diseases

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Abstract

The objective of this contribution is to present some recent advances in pitahaya (*Hylocereus* spp.) diseases research conducted in various parts of the world, thereby providing some information for promoting further studies on plant protection of the pitahaya species. Pitahaya is a cactus fruit crop of high demand at national and international level. It is cultivated in more than 10 countries worldwide. Prevalent form of cultivation until little more than 20 years was in small-scale family production. However, since its cultivation in commercial plantations and further domestication started in earnest, some symptoms of rotting and spots in stems and fruits were being observed. In some cases, commercial production plots had to be abandoned due to the diseases. In the decade of 1990s in Mexico, some studies on stem soft rot disease were initiated. It was found that at least two Enterobacteria were involved. With regard to the causing agent of the spots on stems, it was identified as fungus specie, *Botryosphaeria dothidea* Ces. & de Not, anamorph *Fusicoccum* sp. Morphological and physiological characterization were carried out in Mexico, to develop integrated management strategies. Anthracnose is yet another fungal disease that could become aggressive as it affects fruits and stems. Etiological studies in USA and Japan revealed the causing agent to be a fungus *Colletotrichum gloesporioides* Penz. Meanwhile, researchers in Taiwan described some morphological and genomic characteristics of a virus, *Cactus Virus X*. This virus causes a kind of mosaic on the stem, a mixture of light and dark green areas. Perspective of pitahaya as a commercial crop is promising. In terms of crop protection, however, it is necessary to determine genetic diversity of plant pathogen species as well as that of the causing agents and to develop integrated control measures. Moreover, few studies were made on the diseases in post-harvest. The viral disease is not yet to be reported in Mexico, but some personal observations appear to indicate that it is already present in commercial plantations in the country. It is thus imperative to initiate sampling, detection, and identification activities using traditional and molecular techniques. The final objective of all aforementioned aspects is to conserve and successfully manage pitahaya resource.

Keywords: *Hylocereus*, bacterial diseases, etiological studies, fungal diseases.

Resumen

El objetivo de esta contribución es presentar algunos avances recientes en la investigación de las enfermedades de la pitahaya (*Hylocereus* spp.) conducidos en varias partes del mundo, por lo tanto se proveerá de información para promover estudios posteriores sobre la fitoprotección de especies de pitahaya. La pitahaya es un cactus frutal cultivado de alta demanda a nivel nacional e internacional. Es cultivado en más de 10 países alrededor del mundo. La forma prevalente de producción y cultivo 20 años previos fue familiar a pequeña escala. Sin embargo, a partir de su cultivo en plantaciones comerciales y posterior domesticación, algunos síntomas de pudrición y manchas en tallos y frutos se empezaron a observar. En algunos casos, la totalidad de las plantaciones tuvieron que abandonarse por efecto de las enfermedades. En el decenio de 1990 en México, algunos estudios sobre la pudrición blanda del tallo se iniciaron. Evidencias sobre que al menos dos Enterobacterias estaban involucradas fueron bien soportadas. Respecto al agente causal de manchas en los tallos, se identificó a *Botryosphaeria dothidea* Ces. & de Not, anamorfo *Fusicoccum* sp. La caracterización morfológica y fisiológica se realizó en México, así como algunas estrategias de manejo integrado. La antracnosis

es otra de las enfermedades fungosas que podría llegar a ser agresiva por su efecto en los frutos y en los tallos. Los estudios etiológicos en USA y Japón revelaron que el agente causal es el hongo *Colletotrichum gloesporioides* Penz. Mientras tanto, investigadores en Taiwán describieron algunas características morfológicas y genómicas de un virus, el *Cactus Virus X*. Este virus causa un tipo de mosaico en el tallo, una mezcla de áreas verde claro y oscuras. La perspectiva de la pitahaya como cultivo comercial es promisorio. En términos de protección del cultivo, sin embargo resulta necesario determinar la diversidad de los aislamientos de hongos y bacterias que continúan disminuyendo la producción y la calidad de los frutos así como desarrollar medidas de control integrado. Sin embargo, pocos estudios se han realizado en enfermedades post-cosecha. La enfermedad viral aún no ha sido reportada en México, pero algunas observaciones personales parecen indicar que está presente en plantaciones comerciales en el país. Por lo tanto es imperativo iniciar actividades de muestreo, detección e identificación con el uso de técnicas tradicionales y moleculares. El objetivo final de todos los mencionados es conservar y manejar exitosamente al recurso de la pitahaya.

Palabras clave: *Hylocereus*, enfermedades bacterianas, estudios etiológicos, enfermedades fungosas.

Introduction

Pitahaya, *Hylocereus undatus* (Haworth) Britton & Rose, is an epiphytic cactus of anthropocentric importance for its fruits and stems at national and international level. In addition, some parts of the plant have uses in herbal medicine and in agroindustry. This species could be the most used and important among epiphytic and hemiephytic cacti (see Nobel and de la Barrera, 2004). This species is cultivated in at least 10 different countries worldwide and presents a promising future production increase. On the other hand, yield of this crop can be diminished due to microorganisms such as bacteria, fungi, and viruses as well as some pest insects (a yet to be identified stem borer and a species of true bug). They can induce economic losses up to 44 % (Valencia-Botín *et al.*, 2004a). For these reasons, studies on the crop protection were initiated in 1990s in Colombia, Nicaragua, and Mexico.

Other plant diseases of minor importance are: Stem Mildew (*Helminthosporium* sp.), Stem Soft Rot (*Fusarium* sp.), and a green mold in fruits caused by a fugal complex of *Penicillium* sp., *Volutella* sp., and *Corynespora* sp. These diseases have been reported only in Nicaragua.

The objective of this paper is to describe recent advances in pitahaya disease research to bring forward further inquiry into the subject. Emphasis was made on research conducted in Mexico.

Bacterial diseases

Stem Soft Rot. During the 1990s, some etiological studies on stem soft rot diseases were initiated in Mexico. It has been found that at least two species of Enterobacteria were involved (Valencia *et al.*, 2003a). The disease causes the stem to rot in soft state, occasionally delimited by chlorotic haloes (Figure 1). Experimental trials showed that the rotting appeared 15 days after artificial inoculation.

Two pitahaya species (*Hylocereus undatus* and *H. purpusii*) were found to be susceptible to these bacteria, the first species being more susceptible than the latter. Plants deficient in calcium and nitrogen could develop more severe symptoms of the disease (Valencia *et al.*, 2003a).

A particular soft rot disease affecting stem and fruit in Malaysia was studied by Masyahit *et al.* (2009). They concluded that causal agent of the yellowish and brownish soft rot was *Enterobacter cloacae*. This bacteria has been reported as an opportunistic plant pathogen of papaya (*Carica papaya* L.) fruits in Hawaii causing internal yellowing (Nishijima *et al.*, 1987), internal bulb decay on onion (*Allium cepa* L.) in California and Colorado, USA (Bishop and Davis, 1990), leaf rot disease on odontioda orchids (*Odontioda* sp.) in Japan (Takahashi *et al.*, 1997), rhizome rot of edible ginger (*Zingiber officinale* Roscoe) in Hawaii (Nishijima *et al.*, 2004) and gray kernel disease on macadamia (*Macadamia integrifolia*) in Hawaii (Nishijima *et al.*, 2007). Other report of the bacterial disease include Malaysia causing soft rot (Masyahit *et al.*, 2009).



Figure 1. Symptoms of soft rot of stem caused by *Enterobacteria* in Maxcanú, Yucatán, Mexico (A) and in Malaysia (B) (Photos: Alberto J. Valencia-Botín (A) and Les Thorogood (B)).

Fungal diseases

Fish eye. With respect to spots on the stem, it was found that the causing agent was *Botryosphaeria dothidea* Ces. & De Not, anamorph *Fusicoccum* sp. (Valencia-Botín *et al.*, 2003b; Valencia-Botín *et al.*, 2004b). This disease can be characterized by early symptoms of chlorotic points in the stem, later developing into a form similar to fish eyes (Figure 2).



Figure 2. Chlorotic lesions and spots on the stem (fish eye) caused by *Botryosphaeria dothidea* Ces. & de Not, anamorph *Fusicoccum* sp.
Photo: Alberto J. Valencia-Botín.

There is a possible association between this disease and a species of true bug, *Leptoglossus* spp. (Homoptera, Sub-Order Heteroptera: Coreidae). The bug causes some lesions on the stem surface, from which the chlorotic halos may develop.

The fungus produced pycnidia in potato-dextrose-agar (PDA) medium 8 days after incubation under continuous fluorescent light at 22-25 °C. The pycnidium had a size of 180×150 µm. Its conidium observed in fruiting bodies was a hyaline unicell and had ellipsoid to fusoid form with distinctively truncated base. Its dimension was 12.15 to 19.44 × 4.86 to 8.51 µm, the mean being 16.49 × 5.90 µm (Valencia *et al.*, 2003b).

Histological studies demonstrated that the symptom of the disease was a result of epidermal cellular necrosis and of chlorenchyma demarcated by one or two layers of sclereids. Within these lesions, it was possible to find some pycnidia which are a sexual state of the fungus.

Morphological as well as physiological characterization of *Fusicoccum* isolates were made *in vitro* in Mexico for the purpose of incorporating into integrated management strategies against the disease. It was found that the mycelium of *Fusicoccum* grew better in PDA and EPA (Extract of Pitahaya-Agar) into 8.5 cm radial growth at 22 to 26 °C under 12 and 24 photoperiods (Valencia-Botín *et al.*, 2004a).

Anthracnose. This is also a fungal disease that affects not only fruits but also the stems. Etiological studies have been conducted in USA, Japan, Malaysia and Nicaragua (Kim *et al.*, 2000; Palmateer and Ploetz, 2007; Masyahit *et al.*, 2009). It was found that the causing agent was *Colletotrichum gloesporioides* Penz.

One characteristic of the disease is the presence of reddish brown lesions coalesced with conspicuous chlorotic haloes (Figure 3). Its colonies produce abundant hyaline conidia that are elongated to cylindrical unicells, measuring on the average $14.7 \times 5.0 \mu\text{m}$ at the ranges of 12.5 to 17.5×3.8 to $7.5 \mu\text{m}$.



Figure 3. Lesions of anthracnose caused by *Colletotrichum gloesporioides* Penz in pitahaya.
Photo: Aaron J. Palmateer, University of Florida.

Palmateer and Ploetz (2007) stated that the fungi isolated from a diseased pitahaya plant corresponded to *C. gloesporioides* in their morphological characteristics, except for the appressorium and hyphopodium structures. These isolates had a hyphopodium more spherical and lobulated measuring 10.9 (8.5 to 12.7) \times 9.1 (7.1 to 10.3) μm .

Determination of causing agent was performed under the basis of comparisons of Internal Transcribed Spacer (ITS) DNA sequences (Palmateer and Ploetz, 2007). Additionally, *C. gloesporioides* has been found causing reddish-orange spots with chlorotic halos on stems of *H. megalanthus* in Brazil (Takahashi *et al.*, 2008). No records or symptoms of this disease has been found during our visits to many fields in Mexico.

Recently, a particular disease causing small, circular, light brown, depressed lesions which may expand to form large areas of rot on flowers and fruits (Figure 4) was reported in South Florida (Tarnowski, *et al.*, 2010). The causal agent was identified as *Bipolaris cactivora* (Petra) (= *Drechslera cactivora*).

The same pathogen causes stem rot of the Cactaceae in Europe and the United States and a fruit rot on pitahaya in Japan (Durbin *et al.*, 1955; Taba *et al.*, 2007).

Other genera of fungi. Vazquez *et al.* (2007) found species of the genera *Fusarium*, *Alternaria*, *Cladosporium*, *Aspergillus* and *Rhizopus* to cause fruit decay in pitahaya. This is the first report of those pathogens in postharvest.



Figure 4. *Bipolaris* fruit rot lesions on pitahaya. Naturally developing lesions begin as A, circular, tan, and sunken lesion develop into rotten area with conspicuous dark spot of sporulation (B).

Photo: Aaron J. Palmateer, University of Florida.

Recently a disease in red-fleshed dragon fruit (*Hylocereus polyrhizus* [Weber] Britton & Rose) in Malaysia was reported. It causes small, circular, faint pink to beige lesions that generally coalesce as the symptoms progress. The pathogen associated with those symptoms was identified as *Curvularia lunata* (Wakker) Beodijn. This disease has been reported affecting pitahaya fruits only in Malaysia but not in other countries (Masratul *et al.*, 2009).

Viral disease

Cactus Virus X. Researchers at the National Taiwan University described morphological and genomic characteristics of a virus, Cactus virus X (CVX), which caused light and dark green mosaic on the stem. Based on the experimental trials, they found that the virus was transmitted mechanically and caused local necrosis in *Chenopodium amaranticolor* and chlorotic lesions in *C. quinoa*. These two plant species are classified under Amaranthaceae in APG II (Angiosperm Phylogeny Group II) system, while in others under Chenopodiaceae. The virus also caused necrotic lesions and chlorotic haloes in *Gomphrena globosa*. In *Celosia argentea*, small chlorotic stains may develop, followed by systemic infection of the disease. Both of these plant species belong to Amaranthaceae (Liou *et al.*, 2001).

Studies with electron microscope revealed that the virus had a form of flexible rod of 480 to 520 nm in size. Purified viral particles contained a principal protein of approximately 26 KDa. Analysis using immunodiffusion method indicated a positive reaction to the antiserum for CVX, but they did not react to antiserum for Bamboo Mosaic or Papaya Mosaic Virus. Genome sequencing of this virus has also been conducted (Liou *et al.*, 2004). CVX is considered as a pitahaya pathogen, but its presence in Mexico is not yet to confirmed.

Conclusions

Botryosphaeria dothidea, *Colletotrichum gloesporioides* and *Bipolaris cactivora* are fungi that affect stems and fruits of pitahaya. Only *B. dothidea* have been reported in Mexico. Etiological studies of bacterial diseases are still in progress in our country. Perspective of pitahaya crop protection must encompass in confirming the presence of causing agents by means of integration and complimentary use of molecular tools, such as ribosome and nuclear gene sequencing and RT-PCR, to develop quarantine or eradication measures and to promote integrated management of the diseases.

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