Tropical Plant Pathology (2018) 43:263–270
https://doi.org/10.1007/s40858-017-0206-9

SHORT COMMUNICATION

Survey on alternative hosts of *Corynespora cassiicola*, the cause of the leaf and calyx spot, in the surroundings of roselle fields in Mexico

Javier Hernández-Morales[1] · Daniel Leobardo Ochoa-Martínez[1] · Santo Ángel Ortega-Acosta[1] · Ricardo Vega-Muñoz[2]

Received: 27 July 2017 / Accepted: 12 December 2017 / Published online: 4 January 2018
© Sociedade Brasileira de Fitopatologia 2017

Abstract

Corynespora cassiicola causes the leaf and calyx spot of roselle (*Hibiscus sabdariffa*) in Mexico. A survey was conducted to identify potential inoculum sources of this pathogenic fungus on cultivated plants and weeds in the main producing areas of roselle in Mexico. Identification of the fungus species was based on morphology and sequences of partial Actin and EF-1α genes. The fungus was found infecting *Solanum lycopersicum*, *Chromolaena odorata*, *Senna alata*, *Hyptis suaveolens*, *Eugenia oerstediana*, *Passiflora viridiflora*, *Momordica charantia*, *Ricinus communis* and *Gossypium hirsutum*. To our knowledge, all these plant species, with the exception of *Solanum lycopersicum*, are new records as hosts for *C. cassiicola* in Mexico, and the species *Eugenia oerstediana* and *Passiflora viridiflora* as new hosts worldwide. Pathogenicity tests showed that all *C. cassiicola* isolates of different plant species were able to induce lesions in roselle leaves and calyces. Disease management should include eradication of these alternative hosts.

Keywords *Hibiscus sabdariffa* · Disease spotting · New hosts · Weeds

In Mexico, roselle (*Hibiscus sabdariffa* L.) is grown 20,061 ha yearly. Guerrero state produces >70% of this crop (SIAP 2016), where a leaf and calyx spot disease, caused by the ascomycete fungus *Corynespora cassiicola* (Berk. & M.*A. Curtis*) C.T. Wei, has been observed in recent years. The fungus affects the calyces that are commercialized. On leaves and calyces, up to 100% incidence has been reported (Ortega-Acosta et al. 2015). *Corynespora cassiicola* causes leaf spot in a large number of plants mainly distributed in tropical and subtropical areas (Dixon et al. 2009). In the United States and Brazil, *C. cassiicola* has caused serious problems in cotton (*Gossypium hirsutum* L.), soybean (*Glycine max* L.), and tomato (*Solanum lycopersicum* L.) (Galbieri et al. 2014; Sumabat et al. 2017). In Asia and Africa, the fungus is responsible for the main disease on *Hevea brasiliensis* (Willd. Ex A. Juss.) Müll. Arg. (Deon et al. 2012). In Mexico, *C. cassiicola* has been recorded affecting cucumber (*Cucumis sativus* L.), chili (*Capsicum annum* L.), papaya (*Carica papaya* L.), *Hibiscus sabdariffa* L., and *Solanum lycopersicum* L. (Castro 1979; Ale-Agha et al. 2008; Tun-Suárez et al. 2011; Ortega-Acosta et al. 2015; Farr and Rossman 2017). Several studies have shown that several weeds and cultivated species are hosts of *C. cassiicola* and they may be important sources of inoculum for seasonal epidemics (Onesirosan et al. 1974; Cutrim and Silva 2003). Knowledge of the host range for *C. cassiicola* is important for the regional management of disease (Smith et al. 2009). In Guerrero state of Mexico, roselle is cultivated along with maize (*Zea mays* L.), with few weeding labor that results in the presence of a large number of weeds growing inside and on the periphery of the parcels, some of them surviving all year-round. These plants may be reservoirs of *C. cassiicola*, the causal agent of spotting of leaves and calyces of roselle. The main objective of this research was to identify alternative host plants of

Section Editor: Adalberto Café-Filho

✉ Santo Ángel Ortega-Acosta
ortega.santo@colpos.mx

[1] Fitosanidad-Fitopatología, Colegio de Postgraduados, Carretera México-Texcoco Km. 36.5, Montecillo, CP. 56230 Texcoco, Estado de México, Mexico

[2] Botánica, Colegio de Postgraduados, Carretera México-Texcoco Km. 36.5, Montecillo, CP. 56230 Texcoco, Estado de México, Mexico

C. cassiicola in the roselle producing area in Guerrero, Mexico, and assess the pathogenicity of the strains isolated from those hosts on roselle.

From May to June 2015, plants exhibiting leaf spot symptoms were collected from within and border of roselle field crops. The collected plants were placed in polyethylene bags, which were labeled and sent to laboratory for processing. Leaf pieces (0.2 cm) were taken from the border of diseased tissues, surface-disinfested using 1% sodium hypochlorite for 2 min, rinsed three times with autoclaved distilled water, dried with sterile absorbent paper, and transferred to Petri dishes with PDA. The plates were incubated for one week at 28 °C (Onesirosan et al. 1974). Whenever a fungal-growth was observed, the colonies were transferred to Petri dishes with fresh PDA. The colonies were purified by monosporic culture technique. From the fungi obtained from different hosts, an isolate was selected for further morphological and molecular identification. The plants which were hosts of *Corynespora cassiicola* were taxonomically identified and deposited in the Herbario Hortorio CHAPA of the Colegio de Postgraduados.

For each isolate, spores were transferred to glass slides and 50 conidia were measured (length x width). The color, contour and shape of conidia were recorded. Images were acquired with an AmScope® MU 1000 camera attached to a microscope (Nikon Eclipse Ci). Measurements were made with a micrometric ruler in ImageTool® v3.0 image analyzer (Hernández-Livera et al. 2005). Color and shape of colonies were recorded and both morphological and cultural characteristics were compared with literature (Wei 1950; Ellis 1971). One isolate was selected to represent a single host and molecularly identified. Mycelia of *C. cassiicola* of 12-days old cultures grown in Petri dishes with PDA were scraped from the surface of the agar with a sterile metallic tweezer. Genomic DNA was extracted using AP protocol described by Sambrook and Russel (2001). PCR was performed for the amplification of a segment of the Actin and EF-1α genes using ACT-512F/ACT-783R and EF1-728F/EF1-986R primers, respectively (Carbone and Kohn 1999). The PCR mixture consisted of: 2 μL buffer (1 X), 0.6 μL $MgCl_2$ (2.0 μM), 0.2 μL dNTP's (0.2 mM), Primer (10 μM) forward 0.6 μL and reverse 0.6 μL, 0.1 μL Taq DNA polymerase (0.5 U) (Promega®) and 10 ng of the DNA extracted from the fungus. The final mixture was adjusted with sterile ultra-pure water to a 10 μL volume. The Eppendorf tubes were placed in a thermocycler (Techne-TC-512®) with the following amplification program for both sets of primers: initial denaturation at 94 °C for 2 min; 40 cycles at 94 °C for 1 min, 58 °C for 1 min and 72 °C for 1 min; with a final extension of 72 °C for 7 min (Shimomoto et al. 2011). Amplified PCR products were purified with Wizard® Genomic DNA Purification Kit (Promega®) and they were sequenced. The sequences were assembled and edited using the SeqMan Pro module from the DNASTAR LaserGene® program. The consensus sequences were compared and deposited at the GenBank.

Table 1 Host plants of *Corynespora cassiicola* in Guerrero, Mexico

[a]Code	Municipality	Locality	Geographic Coordinates	[b] m a.s.l.	Family	Common name	Scientific name	Symptoms observed
Ortega-Acosta et al.1	Ayutla	Cotzálzin	16°54′55.53″ N, 99°5′26.16″ O	342	Passifloraceae	–	*Passiflora viridiflora* Cav.	Leaf spots
Ortega-Acosta et al.2	Ayutla	Cotzálzin	16°54′55.53″ N, 99°5′26.16 O	342	Solanaceae	Wild tomato	*Solanum lycopersicum* L.	Leaf spots, and blight
Ortega-Acosta et al. 3	Ayutla	San José de la Hacienda	16°58′41.34″ N, 99°5′59.61″ O	392	Malvaceae	Cotton	*Gossypium hirsutum* L.	Spotting leaf and calyces
Ortega-Acosta et al.4	Ayutla	San Miguel	16°59′15.24″ N, 99°5′56.32″ O	385	Labiatae	Wild spikenard	*Hyptis suaveolens* (L.) Poit.	Leaf spots, and blight
Ortega-Acosta et al.5	Tecoanapa	Tecuantepec	16°59′42.96″ N, 99°14′50.59″ O	437	Fabaceae	Candlestick plant	*Senna alata* (L.) Roxb.	Leaf spots with center dark brown
Ortega-Acosta et al.6	Tecoanapa	Tecuantepec	16°59′47.96″ N, 99°14′55.96″ O	441	Myrtaceae	Capulin	*Eugenia oerstediana* O. Berg	Leaf spots with straw-colored center, black concentric rings, and purple halo
Ortega-Acosta et al.7	Tecoanapa	Tecuantepec	16°59′47.86″ N, 99°15′15.51″ O	439	Euphorbeaceae	Castor bean	*Ricinus communis* L.	Leaf spots with a straw-colored center, and blight
Ortega-Acosta et al.8	Tecoanapa	Xalpatláhuac	17°16.29″ N, 99°20′4.27″ O	690	Cucurbitaceae	Bitter melon	*Momordica charantia* L.	Leaf spot with straw-colored center, black concentric rings, and blight
Ortega-Acosta et al.9	Tecoanapa	Tecuantepec	16°59′51.10″ N, 99°14′54.03″ O	443	Asteraceae	–	*Chromolaena odorata* (L.) R.M. King & H. Rob.	Leaf spots, and blight

[a] Collection code botany deposited in Herbario-Hortorio CHAPA of Colegio de Postgraduados

[b] Metres above sea level

Table 2 Host plants of *Corynespora cassiicola* reported worldwide

Host	Codes for *C. cassiicola* isolates	Geographic location	New host for *C. cassiicola*	New report for Mexico	References
S. lycopersicum	JIT	Several reports, including Mexico	–	–	Farr and Rossman (2017)
C. odorata	M26	Palau	–	+	Dixon et al. (2009)
S. alata	GUG	Cuba	–	+	López et al. (2002); Wei (1950)
H. suaveolens	CHI	Guam	–	+	Smith (2008)
E. oerstediana	CAP	No reports for this species	+	+	Farr and Rossman (2017)
P. viridiflora	PAS	No reports for this species	+	+	Farr and Rossman (2017)
M. charantia	MAT	China, Florida, Japan, Korea, Northern Mariana Islands, Palau, Venezuela	–	+	Kwon et al. (2005); Li et al. (2010); Alfieri et al. (1984); Dixon et al. (2009); Shimomoto et al. (2011); Farr and Rossman (2017)
R. communis	HGA	India	–	+	Sarbhoy et al. 1971
G. hirsutum	ALG	China, India, United States	–	+	Sarbhoy et al. (1971); Conner et al. (2013); Wei et al. (2014); Butler et al. (2016)

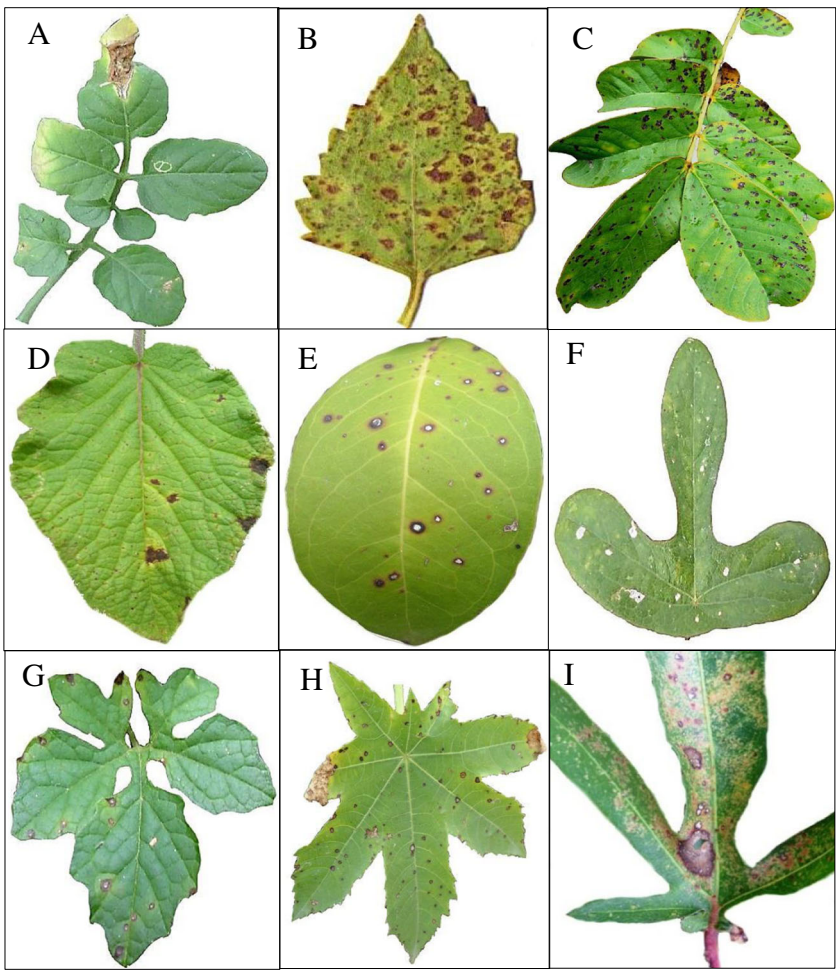

Fig. 1 Leaf spots in different plant species from which *C. cassiicola* were isolated. A = *S. lycopersicum*; B = *C. odorata*; C = *S. alata*; D = *H. suaveolens*; E = *E. oerstediana*; F = *P. viridiflora*; G = *M. charantia*; H = *R. communis*; I = *G. hirsutum*

Table 3 Comparison of *Corynespora cassiicola* sequences in GenBank

Hosts	Isolates	Species	[a]Actin	%	[b]EF-1α	%
S. lycopersicum	JIT	*C. cassiicola*	MF000832	100	MF000880	100
C. odorata	M26	*C. cassiicola*	MF000833	99	MF000881	100
S. alata	GUG	*C. cassiicola*	MF000834	100	MF000882	99
H. suaveolens	CHI	*C. cassiicola*	MF000835	100	MF000883	100
E. oerstediana	CAP	*C. cassiicola*	MF000836	100	MF000884	99
P. viridiflora	PAS	*C. cassiicola*	MF000837	100	MF000885	100
M. charantia	MAT	*C. cassiicola*	MF000838	100	MF000886	100
R. communis	HGA	*C. cassiicola*	MF000839	100	MF000887	99
G. hirsutum	ALG	*C. cassiicola*	MF000840	99	MF000888	99

[a] = Access numbers for the actin partial gene

[b] = Access numbers for the EF-1α partial gene. % = Percentage of similarity with *C. cassiicola* sequences available from GenBank

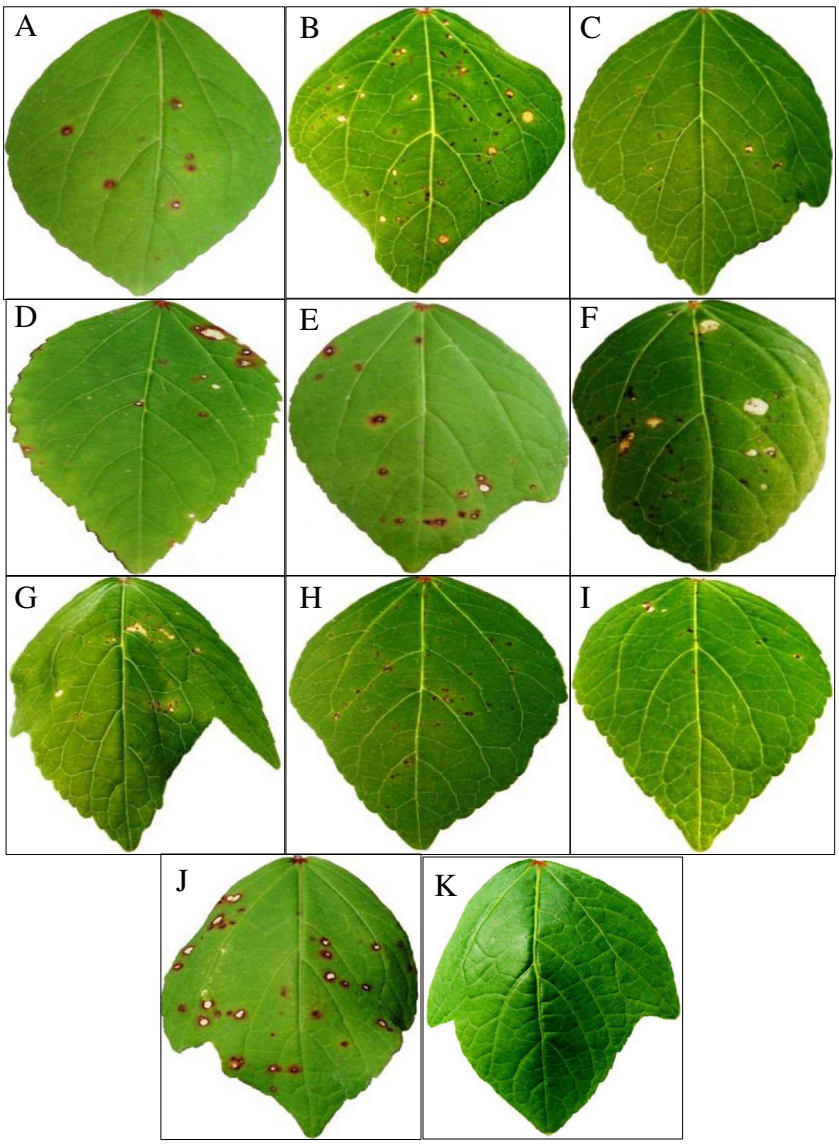

Fig. 2 Symptoms on leaves of *Hibiscus sabdariffa* caused by *C. cassiicola* isolated from different plant species. Twenty days after inoculation. A = JIT; B = M26; C = GUG; D = CHI; E = CAP; F=PAS; G = MAT; H=HGA; I = ALG; J = CCHFR (positive control); K = Autoclaved distilled water (negative control)

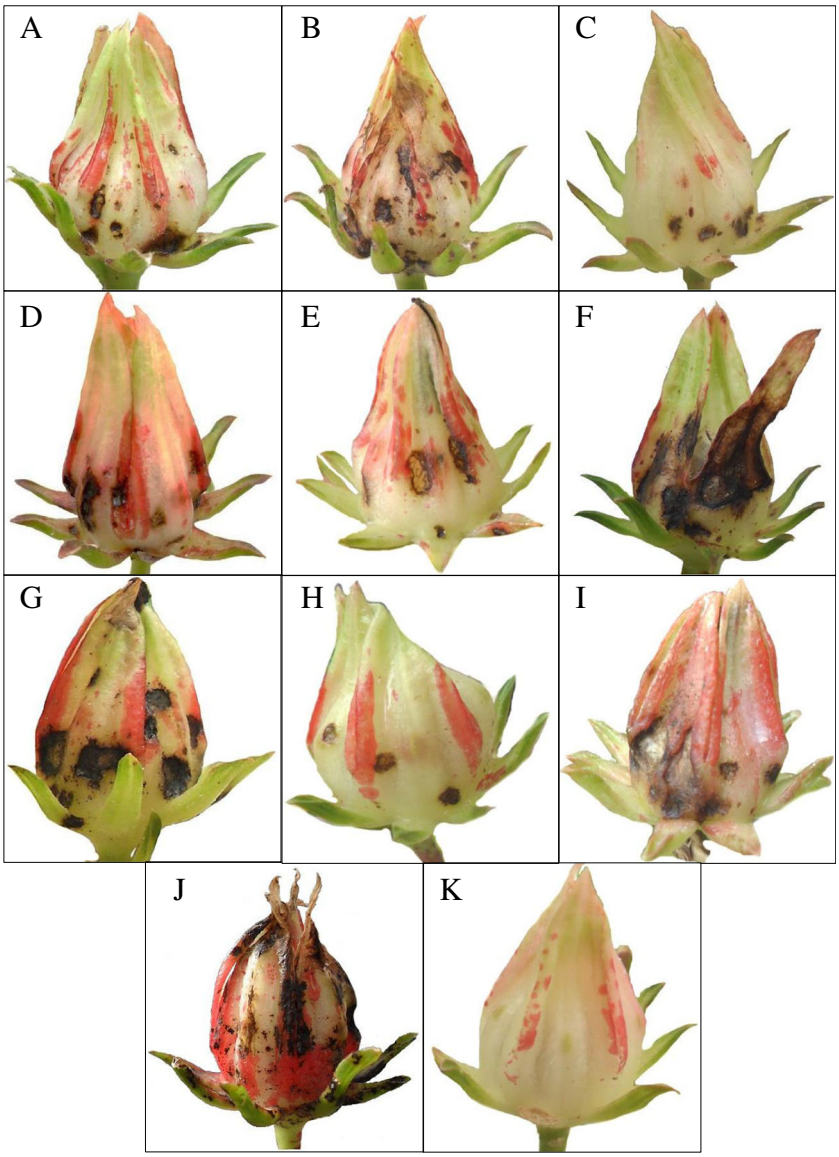

Fig. 3 Symptoms on calyces of *Hibiscus sabdariffa* caused by *C. cassiicola* isolated from different plant species. Twenty days after inoculation. A = JIT; B = M26; C = GUG; D = CHI; E = CAP; F=PAS; G = MAT; H=HGA; I = ALG; J = CCHFR (positive control); K = Autoclaved distilled water (negative control).

Isolates were grown on PDA at 28 °C during 7 days. Conidia were collected in a glass objects with autoclaved distilled water and adjusted to 2×10^5 spores/mL (Jin-Hyeuk and Chang-Seuk 2003; Ortega-Acosta et al. 2015). One *C. cassiicola* isolated, named CCHFR (Access No. KM207768) was utilized as positive control. Plants of roselle cultivar "Criollo de Guerrero" were grown in 15 cm diameter pots during 4.5 months. The inoculum suspension of each isolate considered as a treatment was sprayed with a hand atomizer on ten roselle plants (100 mL/plant) (Jin-Hyeuk and Chang-Seuk 2003). As a negative control, autoclaved distilled water was sprayed on five roselle plants. Isolates were not tested against their respective host of origin. The plants were covered with polyethylene bags during the first 48 h, and then kept under greenhouse conditions. Temperature and relative humidity were recorded with a digital hygrothermograph (Extech®-Rht10). To increase relative humidity an ultrasonic humidifier was placed on the center of the experiment. The average temperature and relative humidity recorded during the study period was 24 °C and 75%, respectively. Twenty days after inoculation the absence or presence of symptoms were registered. Ten fragments of plant symptom tissues from the leaves and calyx were collected from each inoculated isolate (treatment). Re-isolation was carried out using the methodology described above. Fragments of asymptomatic tissue from the control treatment were also inspected.

A total of fifty-four plant species were collected from fields cultivated with roselle and *Corynespora cassiicola* was isolated from nine of them: *S. lycopersicum* L., *Chromolaena odorata* (L.) R.M. King & H. Rob, *Senna alata* (L.) Roxb, *Hyptis suaveolens* (L.) Poit, *Eugenia oerstediana* O. Berg, *Passiflora viridiflora* Cav., *Momordica charantia* L., *Ricinus communis* L. and *Gossypium hirsutum* L. (Tables 1 and 2). Symptoms

Table 4 Pathogenicity tests on *Hibiscus sabdariffa* with ten isolates of *Corynespora cassiicola*

Hosts	Isolates	Inoculated plants/plants with symptoms	Symptoms	
			Leaves	Calyces
S. lycopersicun	JIT	10/10	Circular irregular spots with straw-colored centers, black borders, and evident purple rings	Circular to irregular sunken spots
C. odorata	M26	10/10	Circular irregular spots with straw-colored centers, and black borders	Circular to irregular sunken spots, and blight in the apex of the calyx
S. alata	GUG	10/10	Circular irregular spots with straw-colored centers, and black borders	Circular to irregular sunken spots
H. suaveolens	CHI	10/10	Circular irregular spots with straw-colored centers, black borders, and evident purple rings	Circular to irregular sunken spots
E. oerstediana	CAP	10/10	Circular irregular spots with straw-colored centers, black borders, and evident purple rings	Circular to irregular sunken spots
P. viridiflora	PAS	10/10	Circular irregular spots with straw-colored center, and black borders	Circular to irregular sunken spots, and blight in the apex of the calyx
M. charantia	MAT	10/10	Circular irregular spots with straw-colored centers, and black borders	Circular to irregular sunken spots, and blight in the apex of the calyx
R. communis	HGA	10/10	Circular irregular spots with straw-colored centers, and black borders	Circular to irregular sunken spots
G. hirsutum	ALG	10/10	Circular irregular spots with straw-colored centers, and black borders	Circular to irregular sunken spots
H. sabdariffa (Positive control)	CCHFR	10/10	Circular irregular spots with straw-colored centers, black borders, and evident purple rings	Circular to irregular sunken spots, and blight in the apex of the calyx
Negative control	Autoclaved distilled water	5/0	No showed symptoms	No showed symptoms

observed in these plants differed in leaf spot size and shape (Table 1, Fig. 1).

The fungus growth on PDA showed a wide range of morphological diversity according to the host from where they were isolated. The main characteristics of the colonies were round or in polygon, gray, green or cream color, with abundant aerial mycelium. The conidia were olivaceous to brown, with straight or slightly curved contours; oblong, cylindrical or oval, from 284.62–11.17 μm × 10.52–4.0 μm (length x width), and from 0 to 15 pseudosepta, typical characteristics of *C. cassiicola* (Wei 1950; Ellis 1971). Based on the GenBank search with the BLAST tool (Zhang et al. 2000; Morgulis et al. 2008), the identity of the sequences of the PCR-amplified fragments (Actin and EF-1α) of the different fungi isolates varied from 99 to 100% with *C. cassiicola* (Table 3).

Under greenhouse conditions, all *C. cassiicola* isolates (including the positive control) caused necrosis in leaves and calyces. The plants sprayed only with sterile distilled water (negative control) showed no symptoms (Figs. 2 and 3, Table 4). The re-isolated fungi of leaves and calyces of inoculated plants that showed symptoms were identified as *C. cassiicola* based on their cultural and morphological characteristics (Wei 1950; Ellis 1971). No mycelial growth was observed in plant tissues sprayed only with autoclaved distilled water (negative control).

In this study, we found a range of hosts of *Corynespora cassiicola*, which were able to cause the leaf and calyx spot in roselle. The differences in symptoms in the original host may be due to differences in plant age, infection timing or environmental conditions (Table 1, Fig. 1). In Mexico, *C. cassiicola* has been reported as a pathogen of cucumber (*Cucumis sativus* L.) (Castro 1979), chili (*Capsium annum* L.) (Tun-Suárez et al. 2011), roselle (*Hibiscus sabdariffa* L.) (Ortega-Acosta et al. 2015) and tomato (*Solanum lycopesicum* L.) (Farr and Rossman 2017). In our study, all but *S. lycopersicum* are new reports of hosts of *C. cassiicola* in Mexico (Table 2). The fungus has been reported affecting *Passiflora foetida* L. and *Passiflora* sp., in Guam and Florida, respectively; and on *Eugenia uniflora* L. in Guam (Alfieri et al. 1984; Dixon et al. 2009). No reports were found on *E. oerstediana* and *P. viridiflora* (Table 2). Therefore, these two species of plants are new hosts for *C. cassiicola*.

In pathogenicity tests, symptoms caused by the different isolates inoculated on roselle plants were similar to previously reported (Ortega-Acosta et al. 2015). Besides, slight variations of symptoms were observed across the isolates, which agrees with reports by Kingsland (1986) who cross-inoculated *C. cassiicola* isolates obtained from tomato and cucumber. The authors suggested that there is some degree of specificity

towards the host of origin. A similar pattern was observed in the present study. With the exception of cotton, all hosts are non-cultivated weeds in the roselle producing area, which grow on the edge or within the field. Cotton is generally grown in family orchards close to roselle fields. Conidia of *C. cassiicola* are able to disperse by the wind (Jayasinghe 2000; Smith et al. 2009), hence these hosts are potential sources of inoculum for epidemics in roselle crop. Weeds and cultivated plants that act as *C. cassiicola* reservoirs may favor the emergence of the disease (Onesirosan et al. 1974; Cutrim and Silva 2003). In Nigeria, Onesirosan et al. (1974) conducted studies with *C. cassiicola* isolates from *Aspilia africana* (P. Beauv. ex Pers.) C.D. Adams and *Lepistemos* sp. Weeds. When the pathogen was inoculated on cotton, it caused typical severe symptoms. In Brazil, Cutrim and Silva (2003) isolated *C. cassiicola* from tomato which was inoculated on two weeds, *Commelina benghalensis* L., and *Vernonia cinerea* (L.) Less., where it caused typical lesions. The authors suggested eradication of these weeds as a measure to combat the disease. Qi et al. (2011) inoculated *C. cassiicola* isolates from different hosts on *Hevea brasiliensis* and found results similar to our study. Further surveys of *C. cassiicola* on other weeds and cultivated plant species in the roselle producing areas of Guerrero may be important to expand knowledge of the potential hosts, which should not be grown or eliminated in the vicinities of roselle crops.

Acknowledgments Authors are thankful to the Fondo Sectorial SAGARPA-CONACYT (project "163972") for financial support.

References

Ale-Agha N, Jensen M, Brassmann M, Kautz S, Eilmus S, Ballhorn DJ (2008) Rare or remarkable microfungi from Oaxaca (south Mexico). Part II. Communications in Agricultural and Applied Biological Sciences 73:147–155

Alfieri JSA, Langdon KR, Wehlburg C, Kimbrough JW (1984) Index of plant diseases in Florida (revised). Florida Department of Agriculture and Consumer Services, Division of Plant Industry Bulletin 11:1–389

Butler S, Young-Kelly H, Raper T, Cochran A, Jordan J, Shrestha S, Lamour K, Mengistu G, Castro-Rocha A, Shelby P (2016) First report of target spot caused by *Corynespora cassiicola* on cotton in Tennessee. Plant Disease 100:535

Carbone I, Kohn LM (1999) A method for designing primer sets for speciation studies in filamentous ascomycetes. Mycologia 91:553–556

Castro MAS (1979) Leaf blight caused by *Corynespora*: a new disease on cucumber (*Cucumis sativus*) in the valley of Culiacan, Sinaloa, Mexico and its chemical control. Plant Disease Report 63:599–560

Conner KN, Hagan AK, Zhang L (2013) First report of *Corynespora cassiicola*-incited target spot on cotton in Alabama. Plant Disease 97:1379

Cutrim FA, Silva GS (2003) Patogenicidade de *Corynespora cassiicola* a diferentes espécies de plantas. Fitopatologia Brasileira 28:193–194

Deon M, Scomparin A, Tixier A, Mattos CRR, Leroy T, Seguin M, Roeckel-Drevet P, Pujade-Renaud V (2012) First characterization of endophytic *Corynespora cassiicola* isolates with variant cassiicolin genes recovered from rubber trees in Brazil. Fungal Diversity 54:87–99

Dixon LJ, Schlub RL, Pernezny K, Datnoff LE (2009) Host specialization and phylogenetic diversity of *Corynespora cassiicola*. Phytopathology 99:1015–1027

Ellis MB (1971) Dematiaceous Hyphomycetes. CMI, Kew, Surrey, England

Farr DF, Rossman AY (2017) Fungal databases, systematic mycology and microbiology laboratory, ARS, USDA. Accessed on June, 2017

Galbieri R, Araújo DCEB, Kobayasti L, Girotto L, Matos JN, Marangoni MS, Almeida WP, Mehta RY (2014) Corynespora leaf blight of cotton in Brazil and its management. American Journal of Plant Sciences 5:3805–3811

Hernández-Livera RA, Llanderal-Cazáres C, Castillo-Márquez LE, Valdez-Carrasco J, Nieto-Hernández R (2005) Identification of larval instars of *Comadia redtenbacheri* (Hamm) (Lepidoptera: cossidae). Agrociencia 39:539–544

Jayasinghe CK (2000) Corynespora leaf fall: the most challenging rubber disease in Asian and African continents. Bulletin of the Rubber Research Institute of Sri Lanka 42:56–64

Jin-Hyeuk K, Chang-Seuk P (2003) Leaf spot of cotton rose caused by *Corynespora cassiicola* in Korea. Mycobiology 31:57–59

Kingsland CG (1986) Pathogenicity and epidemiology of *Corynespora cassiicola* in the Republic of Seychelles. Tropical Pest Management 32:283–287

Kwon JH, Jee HJ, Park CS (2005) Corynespora leaf spot of balsam pear caused by *Corynespora cassiicola* in Korea. The Plant Pathology Journal 21:164–166

Li B-J, Zhao Y-J, Gao W, Shi Y-X, Xie X-W (2010) First report of target leaf spot caused by *Corynespora cassiicola* on balsam pear in China. Plant Disease 94:127

López MO, López D, Veitía M (2002) Nuevos registros de patógenos fúngicos de la guacamaya francesa (*Senna alata* (L.)) en Cuba. Fitosanidad 6:53–54

Morgulis A, Coulouris G, Raytselis Y, Madden TL, Agarwala R, Schäffer AA (2008) Database indexing for production mega BLAST searches. Bioinformatics 24:1757–1764

Onesirosan PT, Arny DC, Durbin RD (1974) Host specificity of Nigerian and North American isolates of *Corynespora cassiicola*. Phytopathology 64:1364–1367

Ortega-Acosta SÁ, Hernández-Morales J, Ochoa-Martínez DL, Ayala-Escobar V (2015) First report of *Corynespora cassiicola* causing leaf and calyx spot on roselle in Mexico. Plant Disease 99:1041

Qi Y-X, Zhang X, J-J P, Liu X-M, Lu Y, Zhang H, Hui-Qiang Z, Yan-Chao L, Yi-Xian X (2011) Morphological and molecular analysis of genetic variability within isolates of *Corynespora cassiicola* from different hosts. European Journal of Plant Pathology 130:83–95

Sambrook J, Russel DW (2001) Rapid isolation of yeast DNA. In: Sambrook J, Russell DW (eds) Molecular cloning: a laboratory manual. Cold Spring Harbor Laboratory, New York, pp 631–632

Sarbhoy AK, Lal G, Varshney JL (1971) Fungi of India (1967–71). Navyug Traders, New Delhi

Shimomoto Y, Sato T, Hojo H, Morita Y, Takeuchi S, Mizumoto H, Kiba A, Hikichi Y (2011) Pathogenic and genetic variation among isolates of *Corynespora cassiicola* in Japan. Plant Pathology 60:253–260

SIAP (2016) Servicio de información agroalimentaria y pesquera. Accessed 23 June 2017

Smith LJ (2008) Host range, phylogenetic and pathogenic diversity of *Corynespora cassiicola* (Berk. & Curt.) Wei. Ph.D. dissertations, University of Florida, Gainesville, Florida, USA

Smith LJ, Datnoff LE, Pernezny K, Schlub RL (2009) Phylogenetic and pathogenic characterization of *Corynespora cassiicola* isolates. Acta Horticulturae 808:51–56

Sumabat LG, Kemerait RC, Brewer MT (2017) Host-specialized populations of *Corynespora cassiicola* causing emerging target spot epidemics in the southeastern U.S. Phytopathology 107: S12

Tun-Suárez JM, Castillo-Peraza ME, Cristóbal-Alejo J, Latourneerie-Moreno L (2011) Etiología de la mancha foliar del chile dulce (*Capsicum annuum* L.) y su control *in vitro* en Yucatán, México. Fitosanidad 15:5–10

Wei CT (1950) Notes on *Corynespora*. Mycological Papers 34:1–10

Wei YX, Zhang H, Pu J-J, Liu X-M (2014) First report of target spot of cotton caused by *Corynespora cassiicola* in China. Plant Disease 98:1006

Zhang Z, Schwartz S, Wagner L, Miller W (2000) A greedy algorithm for aligning DNA sequences. Journal of Computational Biology 7:203–214