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A Study of Dicotyledonous Weed Species as Hosts of *Potato Yellow Mosaic Trinidad Virus* (PYMTV)

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Abstract: Begomoviruses represent one of the major groups of phytopathogens of many economically important crops worldwide. In Trinidad, *Potato yellow mosaic trinidad virus* (PYMTV) has been implicated as the causal agent of disease epidemics in tomato since the late 1980's. Surveys of dicotyledonous weeds found in commercial tomato cultivations in addition to weeds found in non-tomato-growing areas were conducted to determine possible PYMTV inoculum sources other than tomato. Detection of viruses was confirmed through dot blot hybridization and immunocapture-PCR (IC-PCR) amplification. None of the weeds tested in the study were hosts of PYMTV. However, two weed species were found to be naturally infected with two distinct begomoviruses as confirmed through partial nucleotide sequence comparisons. There was also no evidence of mixed begomovirus infections.

Key words: Alternate hosts, diagnosis, whitefly-transmitted geminiviruses

INTRODUCTION

PYMTV (Family: *Geminiviridae*; genus: *Begomovirus*) is ubiquitous in tomato (*Lycopersicon esculentum* Mill.) in Trinidad[1]. Several factors may contribute to the epidemic status of begomovirus disease including the extent and distribution of cultivated and non-cultivated host species[2]. Apart from their epidemiological importance, weeds that harbor dual or multiple begomovirus infections may facilitate recombination between the constituent begomoviruses, resulting in the emergence of recombinant viruses[3]. The specific objectives of this study were (I) to detect PYMTV in dicotyledonous weed species by dot blot hybridization and IC-PCR amplification and (ii) to identify other begomoviruses that may be present in dicotyledonous weeds.

MATERIALS AND METHODS

Two surveys were conducted. The first was an intensive one that targeted six commercial tomato-growing areas between March-April, 2000. The fields were heavily infected (80 to 100% infection) with PYMTV and in close proximity to each other thereby maintaining a high level of primary inoculum in the tomato-growing areas. The second survey was conducted between April and September, 2000 in non-tomato-growing areas.

Detection of PYMTV by dot blot hybridization and IC-PCR amplification: Dot blot hybridization was carried out to detect the presence of PYMTV with a PYMTV DNA-A specific probe using a DIG (digoxigenin-dUTP) DNA labeling and detection kit (Roche Diagnostics/Boehringer-Mannheim Inc., Germany) based on the method of Rampersad and Umaharan[1]. Immunocapture-PCR (IC-PCR) amplification of infected leaf extracts for subsequent sequencing was carried out using degenerate primers as described by Rampersad and Umaharan[4]. Extracts of PYMTV-infected tomato leaves and healthy leaf extracts of weeds were used as positive and negative controls, respectively.

RESULTS

Dot blot hybridization and IC-PCR assays: Samples collected from tomato growing areas (681 samples: 81 dicotyledonous weed species belonging to 30 families) were all negative for PYMTV detection in both dot blot hybridization and IC-PCR assays (Table 1). This indicated that the virus was not present within or outside the tomato growing areas. IC-PCR detected two distinct begomoviruses in only two weed species, *Sida rhombifolia* (L.) and *Rhynchosia minima* (L.).

Nucleotide sequence comparisons: Target sequences for alignment (using DNASTAR and CLUSTALW and

Table 1: Locations visited, weed species collected and tested for PYMTV infection

Family	Species	Location[a]	Dot- blot +/-	PCR +/-/NT[b]	Symptoms[c] and Number tested[d]
Amaranthaceae	*Amaranthus* spp.	A to Q	-	NT	NS (0/34)
Asteraceae	*Bidens pilosa* L. var. Alba (L.) O.E. Shultz	A to Q	-	NT	NS (0/15)
Asteraceae	*Eclipta prostrata* L.	A to Q	-	NT	NS (0/29)
Portulacaceae	*Portulaca oleracea* L.	A to Q	-	NT	NS (0/45)
Asteraceae	*Parthenuim hysterophora* L.	A to Q	-	NT	NS (0/41)
Acanthaceae	*Blechum pyridiatum* (Lam.) Urb.	A to Q	-	NT	NS (0/21)
Euphorbiaceae	*Acalypha arvensis* L.	A to Q	-	NT	NS (0/4)
Compositae	*Emilia fosberii/sonchifolia* L. (D.C.)	A to Q	-	NT	NS (0/26)
Mimosaceae	*Mimosa pudica* L.	A to Q	-	-	S - Chl (B:1/15)
Cucurbitaceae	*Momordica charantia* L.	A to E	-	-	S - Chl (A, B: 5/15)
Piperaceae	*Peperomia pellucida* L. Kunth	A to Q	-	NT	NS (0/40)
Euphorbiaceae	*Phyllanthus amarus* L.	A to Q	-	-	S - Chl (A, B: 7/26)
Lythraceae	*Ammannia latifolia* L.	A, B	-	-	S - Chl (B: 1/5)
Euphorbiaceae	*Euphorbia hirta* L.	A to Q	-	NT	NS (0/30)
Euphorbiaceae	*Euphorbia heterophylla* L.	A to Q	-	NT	NS (0/12)
Euphorbiaceae	*Euphorbia hypericifolia* L.	A to Q	-	NT	NS (0/6)
Euphorbiaceae	*Euphorbia hyssopifolia* L.	A to E	-	-	S - Chl (E:1/5)
Fabaceae	*Crotalaria retusa* L.	A, B	-	NT	NS (0/7)
Capparidacea	*Cleome rutidosperma* L.	A	-	NT	NS (0/3)
Rubiaceae	*Spermacoce verticillata* L.	A to E	-	NT	NS (0/22)
Compositae	*Syndrella nodiflora* L. Gaertner	A, B, D, E, R	-	NT	NS (0/27)
Rubiaceae	*Spermacoce assurgens* L.	L	-	-	S - Chl (E:1/5))
Amaranthaceae	*Alternanthera tenella* L.(D.C.)	E	-	-	S - Chl (B:1/2)
Leguminosae	*Pueraria phaseoloides* D.C.	R,C	-	NT	NS (0/16)
Leguminosae	*Rhynchosia minima* L.	X,W	-	-	S –YMc (E: 5/5)
Leguminosae	*Calopogonium mucunoides* Desv.	X	-	-	S -Chl (E: 5/5)
Malvaceae	*Malachra fasciata* L.	A, D	-	-	S- Y/Pink Mc(A, L: 8/8)
Malvaceae	*Abelmoschus moschatus* L.	K	-	-	S Chl (L:1/1)
Malvaceae	*Lablab purpureus* L. Sweet	B	-	NT	NS (0/10)
Onagraceae	*Ludwigia* sp.	C	-	NT	NS (0/1)
Convolvulaceae	*Merremia aegyptia* (L.) Urb., (L.) Vahl.	A, B, T	-	-	S - Chl (B:15/17)
Verbenaceae	*Stactytarpheta cayennensis*	E	-	NT	NS (0/1)
Malvaceae	*Hibiscus sabdariffa* L.	E	-	-	S -YMc (E:2/2)
Malvaceae	*Sida acuta* J. Burm	A to O, S	-	-	S -YMc (J, K, V: 10/15)
Malvaceae	*Sida rhombifolia* L.	A to O, S	-	-	S -YMc (J, K, V: 5/15))
Compositae	*Struchium sparganophora*	A	-	NT	NS (0/1)
Leguminosae	*Teramnus volubilis* P. Browne	A	-	NT	NS (0/1)
Convolvulaceae	*Ipomoea squamosa* L.	A	-	NT	NS (0/1)
Compositae	*Ageratum conyzoides* L.	A	-	NT	NS (0/3)
Campanulaceae	*Sphenoclea zeylanica* Gaertner	A	-	NT	NS (0/2)
Compositae	*Ambrosia cumanensis* L. (D.C.)	A	-	NT	NS (0/1)
Maranthaceae	*Alternanthera philoxeroides*	A	-	NT	NS (0/3)
Boraginaceae	*Heliotropuim indicum* L.	A	-	NT	NS (0/1)
Sterculiaceae	*Melochia marducata* L.	A	-	NT	NS (0/1)
Boraginaceae	*Heliotropium procumbens* Mill.	A	-	NT	NS (0/1)
Verbenaceae	*Phyla betulifolia*	A	-	NT	NS (0/2)
Labiatae	*Ocimum sanctum* L.	A	-	NT	NS (0/1)
Polygonaceae ?	*Polygonum* sp. ?	A	-	NT	NS (0/1)
Euphorbiaceae	*Caperonia palustris* A. St. Hil.	A	-	NT	NS (0/3)
Capparidaceae	*Cleome serrata* L.	A	-	NT	NS (0/1)
Phytolaccaceae	*Microtea debulis* Sw.	A	-	NT	NS (0/2)
Compositae	*Pluctea cardinensis* Cass.	A	-	NT	NS (0/3)
Apocynaceae	*Catharanthes roseus* (L.) G. Donf.	A	-	NT	NS (0/1)
Solanaceae	*Physalis angulata* L.	A, D	-	NT	NS (0/2)
Leguminosae	*Flemingia strobilifera* Roxb. ex Aiton f.	A,B	-	NT	NS (0/5)
Vitaceae	*Cissus verticillata* L.	A	-	NT	NS (0/1)
Compositae	*Vernonia cenerea* (L.) Less.	A	-	NT	NS (0/10)
Euphorbiaceae	*Dalechampia scandens* L.	B	-	NT	NS (0/1)
Convolvulaceae	*Merremia quinquanfolia* L.	S,X	-	NT	NS (0/2)
Malvaceae	*Malachra fasciata.*	A, D	-	NT	NS (0/10)
Urticaceae	*Laportea aestuans* (L.) Chew (Benth.)	A	-	NT	NS (0/6)
Fabaceae	*Macroptelium lathyroides*	A	-	NT	NS (0/5)
Loganiaceae	*Spigelia anthelmia* L.	D	-	NT	NS (0/4)
Amacardiaceae	*Spondias mombin* L.	D	-	NT	NS (0/1)
Verbenaceae	*Stachytarpheta jamaicensis* (L.) Vahl.	B, D, I, R	-	-	S - Chl (B:1/13)

Table 1: Continue

Family	Species	Location[a]	Dot- blot +/-	PCR +/-/NT[b]	Symptoms[c] and Number tested[d]
Rubiaceae	*Hamelia erecta* Jacq.	D	-	-	S - Chl (D:1/1)
Onagraceae	*Ludurgia octovaits*	D	-	NT	NS (0/1)
Araceae	*Colocasia esculenta* Schott. (Jacq.)	D	-	NT	NS (0/1)
Apocynaceae	*Prestonia quinquangulares*	D	-	NT	NS (0/1)
Convolvulaceae	*Ipomea batatas* L.	C	-	NT	NS (0/2)
Euphorbiaceae	*Croton lobtus* L.	C	-	NT	NS (0/1)
Solanaceae	*Solanum bicolor* L.	C	-	NT	NS (0/1)
Leguminosae	*Vigna unguilata* L. Walp.	B	-	NT	NS (0/10)
Fabaceae	*Centrosema pubescens* (D.C.) Benth	B	-	NT	NS (0/1)
Scrophulariaceae	*Scoparia dulcis* L.	B	-	NT	NS (0/1)
Verbenaceae	*Lantara camara* L.	R	-	NT	NS (0/5)
Malvaceae	*Malachra alceifolia* Jacq.	D, L	-	-	S-YMc (D, L:3/3)
Solanaceae	*Lycopersicon esculentum* Mill.	A to E	+	+	S- YMc, Crk, (A-E)

Key: [a]Location - Tomato-growing areas: Aranguez (North and South) = A; Pasea/Macoya = B; Cunupia = C; Tabaquite = D; Chaguaramas = E. Non-tomato-growing areas: Valencia = F; Sangre grande = G; Brazil = H; San Rafael = I; Talparo = J; Guaico Tamana = K; Todd's Road = L; Princes Town = M; New Grant = N; Rio Claro = O; Mayaro = P; Manzanilla = Q; Maracas Bay area/ Blanchisseusse = R; Santa Cruz = S; San Juan = T; Maraval = U; Toco Main Road = V; Montserrat Hills = W; Mt Hope = X. [b]NT- Not tested with PCR. [c] Symptoms: NS – No symptoms; S – symptomatic: Chl – chlorosis of leaves; YMc – Yellow mosaic on leaves; Y/pink Mc – yellow or pink mosaic on leaves; [d]Number tested: () – numerator indicates the number of symptomatic plants, denominator indicates the total number of plants sampled.

BLASTN-http://www.ncbi.nlm.nih.gov/blast/) were selected from the major representative phylogenetic clusters of New World begomoviruses. The core cp region (AV1 ORF) was comparatively analyzed because of high percentage of similarity shared among geminiviruses, the availability of these sequences in GenBank[5,6]. Comparisons of the core cp region show that the Trinidad isolate (AY217344) is highly homologous to *Rhynchosia mosaic virus* (RhMV) that infects pigeon pea in Puerto Rico (AY062025) at 88.0%. The virus infecting *S. rhombifolia* (AY217345) seems to be closely related to *Sida golden mosaic virus* (SiGMV) (AF070923) at 89.2%. Based on sequence data and the absence of polymorphic bands in IC-PCR, the results also indicated that there were no mixed infections of PYMTV with other begomoviruses.

DISCUSSION

RhMV and SiGMV have been identified to infect *R. minima* and *S. rhombifolia* in Trinidad. RhMV has recently been implicated as a disease pathogen in tobacco in Mexico and in pigeon pea in Puerto Rico[7,8]. While these findings may not yet be critical to disease epidemics in tomato, they serve to highlight the importance of certain virus reservoirs in weed species to potential disease epidemics in other commodity crops. *Sida* spp. is a common roadside weed in the region and it is not surprising that SiGMV has also been found in Central America[9].

According to the results of this study, the absence of PYMTV in any of the dicotyledonous weed species sampled, even in the presence of high primary inoculum, suggests that PYMTV may have a limited naturally-infected host range. These findings are congruent with other host range studies conducted for *Tomato mottle virus* (ToMoV) in Florida[10] and *Bean golden mosaic virus* (BGMV) in the wider Caribbean[9]. PYMTV is an emergent virus that exclusively infects tomato which, in turn, is the only known inoculum source. Its absence in dicotyledonous weeds indicates rapid diversification due to high selection pressure if the virus is indigenous or introduction of the virus from an external source.

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REFERENCES

1. Rampersad, S.N. and P. Umaharan, 2003. Identification of resistance to *Potato yellow mosaic virus-trinidad isolate* (PYMTV) among Lycopersicon species. Plant Dis., 87: 686-691.
2. Salati, R., M.K. Nahkla, M.R. Rojas, P. Guzman, J. Jaquez, D. Maxwell and R.L. Gilbertson, 2002. Tomato yellow leaf curl virus in the Dominican Republic: Characterization of an infectious clone, virus monitoring in whiteflies and identification of reservoir hosts. Phytopathology, 92: 487-496.
3. Méndez-Lozano, J., I. Torres-Pacheco, C.M. Fauquet and R.F. Rivera-Bustamante, 2002. Interactions between geminiviruses in a naturally occurring mixture: *Pepper huasteco virus* and *Pepper golden mosaic virus*. Phytopathology, 93: 270-277.

4. Rampersad, S.N. and P. Umaharan, 2003. Detection of begomoviruses in clarified plant extracts: A comparison of standard, direct-binding and immunocapture PCR techniques. Phytopathology, 93: 1153-1157.
5. Brown, J.K., A.M. Idris, I. Torres-Jerez, G.K. Banks and S.D. Wyatt, 2001. The core region of the coat protein gene is highly useful for establishing the provisional identification and classification of begomoviruses. Arch. Virol., 146: 1581-98.
6. Mayo, M.A. and C.R. Pringle, 1998. Virus taxonomy. J. Gen. Virol., 79: 649-57.
7. Ascencio-Ibáñez, J.T., G.R. Argüello-Astorga, J. Méndez-Lozano and R.F. Rivera-Bustamante, 2002. First report of *Rhynchosia golden mosaic virus* (RhGMV) infecting tobacco in Chiapas, Mexico. Plant Dis., 86: 692.
8. Idris, A.M., J. Bird, D.M. Rogan and J.K. Brown, 2002. Molecular characterization of *Rhynchosia mosaic virus*-Puerto Rico associated with symptomatic Rhynchosia Minima and *Cajanus cajan* in Puerto Rico. Plant Dis., 86: 558.
9. Gilbertson, R.L., S.H. Hidayat, R.T. Martinez, S.A. Leong, J.C. Faria, F. Morales and D.P. Maxwell, 1991. Differentiation of bean-infecting geminiviruses by nucleic acid hybridization probes and aspects of Bean golden mosaic in Brazil. Plant Dis., 75: 336-342.
10. Polston, J.E., E. Hiebert, R.J. McGovern, P.A. Stansly and D.J. Schuster, 1993. Host range of *Tomato mottle virus*, a new geminivirus infecting tomato in Florida. Plant Dis., 77: 1181-1184.