Applied Biological Research 18 (3): 270-276; (2016)
DOI: 10.5958/0974-4517.2016.00042.2

POTENTIAL RESERVIOURS OF WHITEFLY TRANSMITTED BEGOMO-VIRUS CAUSING LEAF CURL DISEASE IN CHILLI

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(Received 1 September, 2015; accepted 8 September, 2016)

ABSTRACT

A total of 19 weeds *viz*., *Abutulon theopharasti, Achyranthus aspara, Ageratum conizoides, Cyperus esculentus, Malvestrum coromandelianum, Amaranthus viridis, Boerhavia diffusa, Calotropis gigantean, Cannabis sativa, Commelina benghalensis, Digera arvensis, Eclipta alba, Euphorbia hirta, Tinospora cordifolia, Ipomoea pestigridis, Parthenium hysterophorus, Phylanthus niruri, Physalis minima* and *Tribulus terrestris*, belonging to 12 families, were collected from chilli (*Capsicum annum*) fields in Punjab (India) with the aim to find out alternate hosts of Begomoviruses affecting drastically chilli crop. Of these, *Eclipta alba, Ageratum conizoides, Amaranthus viridis, Ipomoea pestigridis, Abutulon theopharasti, Cannabis sativa* and *Commelina benghalensis* showed typical leaf curl symptoms with vein clearing as predominant symptom exhibited by the above symptomatic weed species. Rest weeds did not possess any symptom. All the weed species were subjected to PCR and only *Commelina benghalensis* was found positive for Begomovirus with expected fragment size of ~575 bp of core protein region. Further, rolling circle amplification (RCA) technique was adopted to develop concatemer of circular DNA of virus. The RCA product when used as template in PCR showed amplification (~575 bp) for all the symptomatic and asymptomatic weed species, except *Calotropis gigantean*. The phylogenitic analysis revealed the presence of chilli leaf curl India virus, tomato leaf curl Jodepur virus and papaya leaf crumple virus in weed samples. Perusal of literature revealed that *Commelina benghalensis aspara, Boerhavia diffuda, Cyperus esculentus, Tinospora cordifolia* and *Ipomoea pestigridis* have not so far been reported as hosts of Begomoviruses, so this is the first report in this direction.

Key words: Begomovirus, chilli, leaf curl, whitefly, weed hosts

INTRODUCTION

During the last three decades viruses have emerged as devastating pathogens, particularly in tropics and subtropics, causing huge economic losses and threatening vegetable crop production. The family *Geminiviridae* is one of the largest plant virus families. Its members have a circular, single-stranded DNA (ssDNA) genome of approximately 2.7-5.2 kb encapsidated within twinned (geminate) icosahedral virions and are transmitted by whitefly (*Bemisia tabaci*) in a persistent

circulative manner and infect dicotyledonous plants (Van Regenmortel *et al*., 2000). Till date, begomoviruses have been reported from at least 16 different crop in India (Borah and Dasgupta, 2012). Weeds are world-wide distributed and have high environmental adaptability. The first studies on whitefly transmitted viruses were made in Latin America which date back 1930 and concern several weed species belonging to the families *Malvaceae* and *Euphorbiaceae* in Puerto Rico and Brazil (Cosata and Bennet, 1950). Weeds serve as reservoir or alternative hosts for begomovirus survival and spread in India and abroad (Gilbertson *et al.,* 1991). In India, weed hosts such as *Acanthosperomum hispidum*, *Ageratum conyzoides*, *Datura stramonium*, *Euphorbea geniculate, Gynandropisis pentaphylla, Parthenium hysterophorus, Sida* sp. and *Tribulus terrestris* have been reported as sources of inoculum for Begomoviruses (Shastry, 1984; Reddy *et al.,* 2005; Sivalingam*et al.,* 2007; Gupta *et al.,* 2010). Kang *et al.* (2004) reported the presence of cotton leaf curl virus in symptomless plants of chilli and China rose in Punjab. Gaikwad *et al.* (2010) reported brinjal, *Petunia hybrida*, calendula and marigold as a natural host for tomato leaf curl virus in Punjab. Pumpkin (*Cucurbita maxima*) and snap melon (*Cucumis melo*) have been found as potential reservoirs of Begomoviruses since they remain in field for a long time (Sharma and Kang, 2009).

Frequent mixed infections with Begomoviruses from different lineages make these weed species melting pots for the generation of new viruses through recombination, a phenomenon that is crucial for speciation and evolution in the family *Geminiviridae* (Seal *et al.,* 2006). The characterization of viral population infecting weeds offer significant evidence about ecological and evolutionary characteristics of these viruses, and convey clues as to whether they can infect and establish in plants. The aim of present study was to characterize the Begomoviruses infecting economically important weeds in Punjab (India) and to assess their diversity.

MATERIALS AND METHODS

Collection of samples

The symptomatic as well as asymptomatic prevalent weeds harbouring adult population of whiteflies growing within and around leaf curl affected chilli fields of PAU, Ludhiana (India) were collected in July 2013. A total of 19 weed samples belonging to different species were collected in labeled zip lock bags and brought to the laboratory. The identification of collected samples was authenticated by the Head, Department of Agronomy, PAU, Ludhiana, India. The type of symptoms exhibited by these weed species were recorded and catalogued.

Begomovirus detection

The genomic DNA of weed samples was extracted following CTAB method (Lodhi *et al.,* 1994) with suitable modifications and later on quantified using Thermo Scientific NanoDrop™ 1000. The sample DNA used as template was subjected to virus detection using PCR. The weed samples which were not amplified by universal degenerated primers (AV494/AC1048) as suggested by Wyat and Brown (1996) were subjected to RCA (rolling circle amplification) followed by PCR. The core CP region amplified by AV494/AC1048 primers with PCR or RCA + PCR was used for cloning and sequencing. The T/A cloning with InsTA clone kit (Fermentas) was performed, followed by bidirectional sequencing of clones of each weed species. The samples were out-sourced for sequencing to Xcelris Lab Ltd. (Ahmedabad) and sequencing was done using ABI 3730Xl DNA analyzer. The sequences of these were analyzed using bioinformatics tools except the construction of phylogenetic tree. For construction of phylogenetic tree, an online programme Phylogeny.fr was used (*http://www.phylogeny.fr/version2_cgi/index.cgi*) [Derepper *et al.,* 2008].

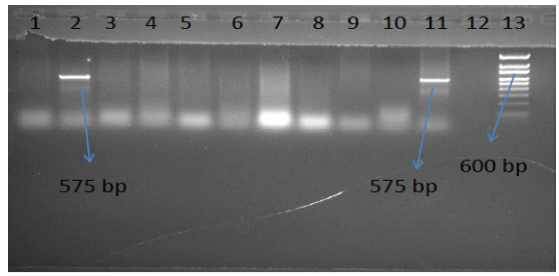
RESULTS AND DISCUSSION

A total of 19 weeds were collected at random from chilli growing field conditions to assess whether these serve as alternate hosts of Begomoviruses causing leaf curl disease. Of these *Eclipta alba, Ageratum conizoides,, Malvestrum coromandelianum, Ipomoea pestigridis, Abutilon theopharasti* and *Canabis sativa* showed typical symptoms of Begomovirus infection, with yellowing of veins as a predominant symptom. Rest thirteen weeds didn't possess any symptom. DNA extracted with CTAB method was quantified on Nano Drop and found good both in quantity and quality with 260/280 ratio ranging between 1.89-1.96. Among these 19 weed species subjected to regular PCR reaction with universal Begomovirus specific primers, only *Commelinia benghaliansis* was positive for Begomovirus with expected fragment size of ~575 bp. These results could be attributed to low titre of virus below the level of detection by regular PCR or the presence of sufficient differences in the nucleotide sequence between viral DNA and the probe to prevent hybridization at low stringency (Marcia *et al.,* 1997). To overcome low titre of virus, rolling circle amplification (RCA) technique blended with PCR was adopted. The RCA product of the DNA of 18 weed samples which were previously negative with regular PCR assay were subjected to PCR. The results showed amplification of ~575 bp fragment for all the symptomatic and asymptomatic weed species, except *Calatropis*. The results are in conformity with earlier workers (Radhakrishnan *et al.,* 2004, Sharma and Rishi, 2007, Sivalingam *et al.,* 2007, Fiallo *et al.,* 2010). A perusal of literature revealed that five weeds *viz., Commelina benghalensis, Cyperus esculentus, Boerhavia diffusa, Tinospora cordifolia* and *Ipomoea pestigridis* have not so for been reported as host of Begomoviruses, thus this forms the first report.

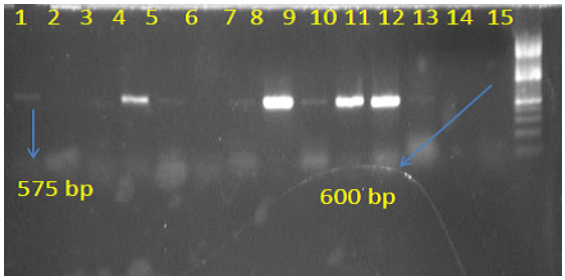
For partial characterization of Begomovirus, seven weed samples *viz., Abutulon theophrasti, Ageratum conyzoides, Amaranthus viridis, Cannabis sativa, Eclipta alba, Ipomoea pestigridis* and *Malvestrum coromandelianum* were subjected to partial genome characterization to know the virus associated with these samples. The RCA-PCR product of ~575 bp size amplified from core CP region of Begomovirus associated with weeds were cloned. The clones *viz.,* Abu. 1 (*Abutulon*

Table 1: Weeds identified as potential hosts of Begomoviruses

S. No.	Botanical name	English name	Family	Symptoms observed	Presence (+) or absence (-) of virus	
					PCR	RCA + PCR
1.	*Abutulon theophrasti*	Velvet leaf	Malvaceae	Vein yellowing & curling	-	+
2.	*Achyranthus aspara*	Chaff flower	Amaranthaceae	Symptomless	-	+
3.	*Ageratum conyzoides*	Billygoat weed	Asteraceae	Vein yellowing & curling	-	+
4.	*Amaranthus viridis*	Slender amaranth	Amaranthaceae	Symptomless	-	+
5.	*Cyperus esculentus*	Nut sedge	Amaranthaceae	Symptomless	-	+
6.	*Boerhavia diffusa*	Spreading hogweed	Nyctaginaceae	Symptomless	-	+
7.	*Calotropis gigantean*	Crown flower	Asclepiadaceae	Symptomless	-	-
8.	*Cannabis sativa*	Hemp	Cannabaceae	Yellowing of veins	-	+
9.	*Commelina benghalensis*	Dayflower	Commelinaceae	Symptomless	+	+
10.	*Digera arvensis*	False amaranth	Amarantahaceae	Symptomless	-	+
11.	*Eclipta alba*	False daisy	Asteraceae	Yellowing of veins	-	+
12.	*Euphorbia hirta*	Asthma-plant	Euphorbiaceae	Symptomless	-	+
13.	*Tinospora cordifolia*	Gulbel	Menispermaceae	Symptomless	-	+
14.	*Ipomoea pestigridis*	Morning glory	Convolvulaceae	Yellowing of veins	-	+
15.	*Malvastrum coromandelianum*	Clock plant	Malvaceae	Yellowing of veins	-	+
16.	*Parthenium hysterophorus*	Congress grass	Asteraceae	Symptomless	-	-
17.	*Phylanthus niruri*	Stonebreaker	Euphobiaceae	Symptomless	-	+
18.	*Physalis minima*	Native gooseberry	Solanaceae	Symptomless	-	+
19.	*Tribulus terrestris*	Puncturevine	Zygophyllaceae	Symptomless	-	+



(a)



(b)

Fig. 1: Agarose gel image showing amplification of virus using AV494/AC 1048 primers in weed samples; 1-10. 1-13.weed samples; 11. Positive control; 12. Negative control; 14, 15 100 bp ladder (G-Biosciences)

theophrasti), Age 1 (*Ageratum conyzoides*) Amran1 (*Amaranthus viridis*), Cana 1 (*Cannabis sativa*), Ecli1 (*Eclipta alba*), Ipo 1 (*Ipomoea pestigridis*) and Mal1 (*Malvestrum coromandelianum*) were sequenced and analyzed. The core CP gene sequence has been illustrated to be very useful in classification of Begomoviruses (Wyat and Brown, 1996; Brown *et al.*, 2012).

Phylogenetic analysis, based on partial genomic DNA sequences of 7 weed samples, revealed formation of 5 different sub-clades by weeds analyzed. Clone Cana 1 of *Cannabis* shared a close ancestor with duranta leaf curl Laxmangarh virus (Acc No. JN998444.1) and papaya leaf crumple virus (Acc No. HM140368. 1). Clones Abu 1 (*Abutulon theophrasti*) and Age 1 (*Ageratum conyzoides*) forming a sub-clade with common ancestor of tomato leaf curl Joydebpur virus that is distantly related to other Begomoviruses. Chilli leaf curl virus India isolate was found close ancestor of clones Ipo 1 (*Ipomoea pestigridis*) and Amran 1 (*Amaranthus viridis*). Two clones Ecli 1 from *Ecliptaalba* and Mal 1 from *Malvastrum coromandelianum* were closely related and formed a separate clade from the Begomovirues used for analysis (Fig 3). Similar observations were made with distance were noticed with distance matrix analysis where

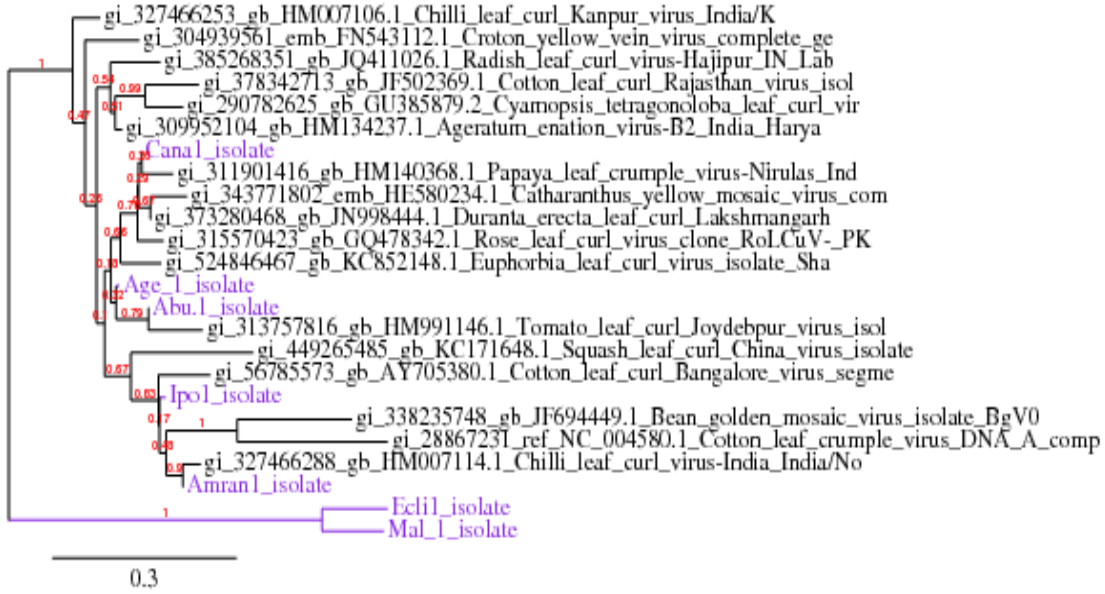


Fig. 3: Phylogenetic analysis of partial sequences of weed samples constructed using BioNJ method under Kimura 2 parameter model for nucleotide, employing interior branch test of 1000 replicas. The tree was constructed for 24 Begomovirus sequences in which clones *viz.*, Abu.1 (*Abutulon theophrasti),* Age 1 *(Ageratum conyzoides)* Amran1 (*Amaranthus viridis*), Cana1 *(Cannabis sativa),* Ecli1 *(Eclipta alba),* Ipo1 *(Ipomoea pestigridis)* and Mal1 *(Malvastrum* spp*)* are from weed samples collected in and around chilli fields of Ludhiana

Percent Identity

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23		
1		73.0	88.4	76.0	83.2	68.3	76.8	77.5	78.7	85.1	75.6	78.9	77.1	83.1	74.8	81.5	80.1	81.5	78.6	7.0	77.1	6.0	54.3	1	JF502369
2	36.5		72.2	74.3	73.7	65.8	71.9	73.8	72.7	80.0	72.1	73.5	74.6	77.8	71.9	76.7	74.8	76.0	73.1	6.4	72.5	7.1	52.8	2	KC171648
3	13.2	38.0		77.9	81.4	69.6	77.0	80.0	79.8	90.4	78.5	83.0	77.8	86.3	76.2	85.2	83.6	83.3	81.6	5.8	78.5	6.5	53.2	3	GU385879
4	31.6	34.0	28.6		78.2	77.2	85.0	80.0	81.2	84.4	80.8	80.8	78.2	83.7	79.4	83.3	81.1	97.7	78.2	6.0	87.0	6.7	55.3	4	HM007114
5	19.7	35.4	22.4	28.2		68.1	75.4	77.9	78.4	81.6	75.7	78.7	76.9	82.5	74.3	82.7	81.5	89.6	77.2	6.0	81.6	6.7	53.7	5	AY705380
6	38.3	41.0	36.7	24.3	39.3		75.8	71.6	73.0	88.8	70.4	71.0	71.9	92.0	69.9	93.1	89.7	82.8	86.1	6.0	80.2	6.9	56.3	6	HM991146
7	30.2	37.1	29.5	19.9	33.3	25.9		80.0	80.8	89.0	86.7	79.1	76.5	91.5	85.3	91.2	88.6	83.3	85.5	3.1	80.2	8.2	55.7	7	HM007106
8	29.0	33.2	26.1	25.9	28.2	30.7	25.4		85.5	89.5	79.6	82.3	87.9	94.7	82.6	89.4	86.8	82.0	88.3	5.6	78.9	6.5	55.0	8	GQ478342
9	26.9	33.6	25.3	24.4	27.0	29.3	23.9	16.8		86.6	81.4	83.1	81.5	91.7	80.0	89.0	86.4	82.3	86.1	2.9	78.1	6.5	54.8	9	KC852148
10	16.6	23.5	10.4	17.6	21.1	12.2	11.6	11.4	14.9		90.8	90.5	89.1	87.5	89.6	86.7	85.4	78.1	83.0	4.9	77.9	6.7	60.6	10	HM134237
11	29.7	35.0	27.5	24.6	31.1	33.6	14.7	25.7	22.6	9.9		80.4	76.0	84.8	85.7	86.2	84.8	79.9	80.8	4.9	80.0	7.1	54.1	11	FN543112
12	27.0	35.0	21.7	24.9	27.8	33.2	25.5	20.9	20.0	9.6	23.2		81.4	86.7	80.1	85.6	84.4	84.1	82.4	11.1	78.5	6.2	54.3	12	JQ411026
13	30.4	33.4	29.3	28.5	29.8	32.6	28.0	12.0	21.9	11.5	29.5	21.6		94.3	82.8	90.6	88.2	81.8	88.5	5.3	78.9	6.5	54.6	13	HM140368
14	19.4	26.7	15.2	18.3	20.0	8.5	9.1	5.5	8.9	13.9	17.2	14.5	5.9		95.1	89.6	86.8	81.5	90.7	5.3	78.5	6.0	71.5	14	JN998444
15	34.0	36.8	32.5	27.1	34.4	35.6	18.1	21.2	24.9	10.9	18.1	26.1	20.0	5.1		89.0	86.2	81.5	88.5	5.8	78.3	6.7	53.9	15	HE580234
16	16.9	23.5	12.1	14.4	14.9	2.9	5.0	7.1	7.6	10.3	10.9	11.1	5.7	6.9	7.6		92.5	81.0	85.2	6.0	83.1	6.9	70.6	16	Abu
17	16.0	23.6	11.5	14.9	14.1	4.2	5.4	7.6	8.4	9.6	10.3	10.5	5.9	7.6	8.4	0.5		81.0	84.0	5.6	81.9	6.5	69.4	17	Age
18	20.8	25.6	18.8	1.9	10.8	19.0	18.4	19.8	19.4	22.3	19.8	17.7	20.1	20.1	20.8	15.4	15.7		76.6	7.6	86.5	8.1	74.0	18	Amran1
19	16.0	23.4	12.0	16.5	17.6	6.5	7.0	3.6	6.5	9.7	12.9	10.7	3.4	0.9	3.4	4.1	4.4	17.8		5.6	76.1	6.5	66.3	19	Cana1
20	125.5	137.7	129.9	118.8	127.1	119.8	113.6	109.3	112.0	122.1	129.2	125.4	117.5	115.9	112.6	121.8	122.9	122.5	112.5		6.4	84.5	2.9	20	Ecli1
21	15.4	22.0	13.5	3.0	8.9	11.0	11.3	12.7	14.3	14.6	11.9	13.2	12.6	13.2	13.5	6.2	7.0	3.0	10.0	111.9		7.1	67.2	21	Ipo1
22	117.8	134.8	118.4	112.4	120.9	109.2	107.1	101.1	107.1	115.4	119.8	114.8	107.1	107.7	105.1	110.3	111.8	125.0	105.5	13.7	106.5		4.3	22	Mal1
23	58.5	60.7	60.6	56.5	58.5	59.8	55.6	56.1	55.9	40.4	56.7	58.0	57.4	35.9	57.6	32.8	31.7	31.4	34.8	141.6	30.0	136.3		23	NC_004580
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23		

Divergence

Fig. 4: Distance matix of per cent nucleotide identities based on core CP sequences of weed clones and selected Begomoviruses

Mal 1 and Ecli 1 shared 84.5% homology with each other but shares maximum of 11.1% nucleotide with radish leaf curl virus (Fig. 4). Hence it proved that these two weeds could harbour Begomovirus species. The weed *I. pestigridis* and *A. viridis* are potential source of chilli leaf curl India virus, whereas Abutilon ad Ageratum are potential reservoir of tomato leaf curl Joydebpur virus. Cannabis is potential reservoir of papaya leaf crumple virus. These viruses are found to be infecting chilli as well as weeds in Punjab and causing chilli leaf curl disease and a serious threat to crop.

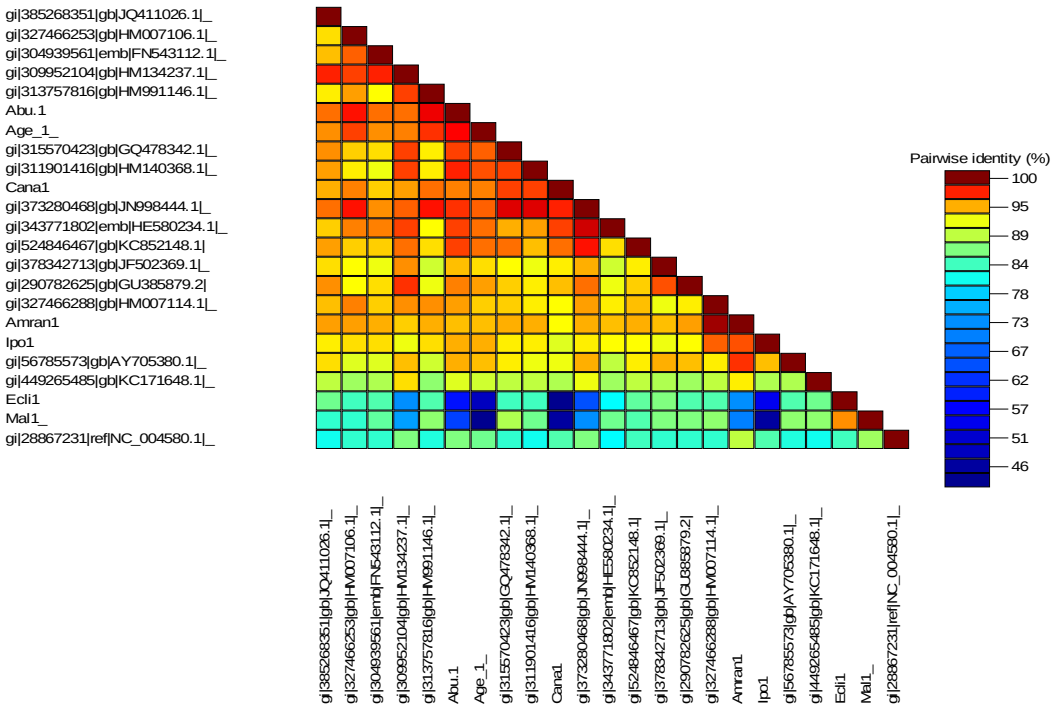


Fig. 5: Closely related sequences of Begomoviruses with weed clones based on similarity scores using pairwise sequence identity tool (SDTv1.0)

Pair-wise distance analysis from SDT V 1.0 indicated that the two clones Mal 1 and Ecli 1 were closely related to each other but distantly related to Begomoviruses reported (Fig. 5). As per the ICTV classification on the basis of core coat protein analysis these two clones from weeds are treated as new (Brown *et al.*, 2012). The association of tomato leaf curl Joydebpur virus has been reported from Punjab by Shih *et al.* (2007), but association of papaya leaf crumple virus with chilli and weeds is a new finding. Therefore, sanitation in and around the chilli crop could be an effective way of management of this disease.

The current study proved that weeds are potential sources of Begomoviruses. Since, these weeds persist throughout the year, so may serve as permanent reservoirs of Begomoviruses and facilitate their carry over to the crops of economic importance. The study demonstrated that not only symptomatic hosts but asymptomatic (symptomless carriers) as well carry Begomoviruses and farmers usually neglect these (asymptomatic ones), resulting in carry over and build-up of disease. Therefore, need is to go for clean cultivation, not only in one's field but in the vicinity as well.

REFERENCES

Borah, B.K. and Dasgupta, I. 2012. Begomovirus research in India a critical appraisal and the way ahead. *Journal Bioscan*, **37:** 791-806.

Brown, J.K., Fauquent, C.M., Briddon, R.W., Zerbini, M., Moriones, E. and Castilo, N.J. 2012. Family *Geminiviridae*. pp. 351-373. **In**: *Virus Taxonomy: Classification and Nomenclature of Viruses – 9th Report of the International Committee on Taxonomy of Viruses* (eds. A.M.Q. King, M.J. Adams, E.B. Carstens and E.J. Lefkowitz). Elsevier Academic Press, USA.

Chattopadhyay, B., Singh, A.K., Yadav, T., Fauquet, C.M., Sarin, N.B. and Chakraborty, S. 2008. Infectivity of the cloned components of a Begomovirus: DNA beta complex causing chilli leaf curl disease in India. *Archives of Virology*, **153:** 533-539.

Cosata, A.S. and Bennet, C.W. 1950. Whitefly transmitted mosaic of *Euphorbia prunifolia*. *Phytopathology,* **40:** 266-283.

Dereeper, A., Guignon, V., Blanc, G., Audic, S., Buffet, S., Chevenet, F., Dufayard, J.F., Guindon, S., Lefort, V., Lescot, M., Claverie, J.M. and Gascuel, O. 2008. Phylogeny.fr: robust phylogenetic analysis for the non-specialist. *Nucleic Acids Research*, **36:** 465-469.

Fiallo, O.E., Navas, C.J. and Martinez, Z.Y. 2010. Two novel Begomoviruses belonging to different lineages infecting *Rhinchosia minima*. *Archives of Virology,* **155:** 2053-2058.

Gaikwad, A.K., Sharma, A., Cheema, D.S. 2010. Tomato leaf curl Palampur virus infecting calendula and marigold in Punjab. *Indian journal of Virology,* **21:** A47.

Gilbertson, R.L., Hidayat, S.H., Martine, R.T., Leong, S.A., Faria, J.C., Morales, F. and Maxwell, D.P. 1991. Differentiation of bean infecting geminiviruses by nucleic acid hybridization probes and aspects of bean golden mosaic in Brazil. *Plant Disease*, **75:** 336-342.

Gregorio, J., Bernal, A., Banuelos, H.B., Alpuchi, A.G. and Hernandez, C. 2010. Analysis of a new strain of *Euphorbia mosaic virus* with distinct replication specificity unveils a lineage of Begomoviruses with short Rep sequences in the DNA-B intergenic region. *Virology*, **7:** 275.

Gupta, V.K., Sharma, R., Singh, S., Jindal, J. and Dilawari, V.K. 2010. Efficiency of *Bemisia tabaci* (Gennadius) populations from plant-hosts for acquisition and transmission of cotton leaf curl virus. *Indian Journal of Biotechnology,* **9:** 271-275.

Haider, M.S., Tahir, M., Saeed, A.H., Shah, A.H., Rashid, N., Javed, M.A. and Iqbal, J. 2007. *Vinca minor*: another host of a tomato infecting Begomovirus in Pakistan. *African Crop Science Conference Proceedings*, **8:** 905-907.

Kang, S.S., Athar, M. and Cheema, S.S. 2004. Additional hosts for cotton leaf curl Geminivirus. *Journal of Research PAU,* **41:** 85-88.

Li, J. and Zhou, X.P. 2010. Molecular characterization and experimental host-range of two Begomoviruses infecting *Clerodendrum cyrtophyllum* in China. *Virus Genes*, **41:** 250-259.

Lodhi, M.A., Ye, G.N., Weeden, N.F. and Reisch, B.A. 1994. Simple and efficient method for DNA extraction from grapevine cultivars and Vitis species. *Plant Molecular Biology Reporter*, **12:** 6-13.

Marcia, E.R. and Wayne, A.M. 1997. Genetic diversity among geminiviruses associated with weed species *Sida* spp, *Myacroptilium lathyroides* and *Wissadula amplissima* from Jamaica. *Plant Disease*, **81:** 1251-1258.

Morales, F.J. 2010. Distribution and dissemination of Begomoviruses in Latin America and the Caribbean. *Archives of Virology*, **146:** 415-441.

Radhakrishnan, G., Malathi, V.G. and Verma, A. 2004. Detection of DNA A and DNA β associated with cotton leaf curl and some other plant diseases caused by whitefly transmitted geminiviruses. *Indian Phytopathology*, **57:** 53-60.

Reddy, R.V.C., Colvin, J., Muniyappa, V. and Seal, S. 2005. Diversity and distribution of Begomoviruses infecting tomato in India. *Archives of Virology*, **150:** 845-867.

Sastry, K.S.M. 1984. Strains of tomato leaf curl virus and its perpetuation under field conditions. *Journal of Turkish Phytopathology*, **13:** 87-90.

Seal, S.E., VanderBosch, F. and Jeger, M.J. 2006. Factors influencing Begomovirus evolution and their increasing global significance: Implications for sustainable control.*Critical Reviews in Plant Science*, **25:** 23-46.

Sharma, A. and Kang, S.S. 2009. Cucurbits: A new host for Begomovirus in Punjab. *Plant Disease Research*, **24:** 51-53.

Sharma, P. and Rishi, N. 2007. Cotton leaf curl disease, an emerging whitefly transmissible Begomovirus comlex. *Plant Viruses*, **1:** 127-134.

Shih, S.L., Tsai, W.S., Green, S.K. and Singh, D. 2007. First report of tomato leaf curl Joydebpur virus infecting chilli in India. *Plant Pathology*, **56:** 341.

Singh, J.A.S., Sahimann, H.S. and Kapur, S.P. 1994. Studies on whitefly *Bemisia tabaci* transmitted cotton leaf curl virus disease in Punjab. *Journal of Insect Science*, **7:** 194-198.

Sivalingam, P.N., Padmalatha, K.V., Mandal, B., Monga, D., Ajemra, B.D. and Malathi, V.G. 2007. Detection of Begomoviruses by PCR in weeds and crop plants in and around cotton field infected with cotton leaf curl disease. *Indian Phytopathology*, **60:** 360-361.

Sivalingam, P.N., Sumiya, K.V. and Malathi, V.G. 2011. Carrot as a new host for a Begomovirus: Yellow mosaic disease of carrot reported in India. *New Disease Reporter*, **23:** 34.

Van Regenmortel, M.H.V., Fauquet, C.M., Bishop, D.H.L., Carstens, E.B., Estes, M.K., Lemon, S.M., Maniloff, J., Mayo, M.A., McGeoch, D.J., Pringle, C.R. and Wickner, R.B. 2000. *Virus Taxonomy: The Classification and Nomenclature of Viruses. The 7th Report of the International Committee on Taxonomy of Viruses.* San Diego, Academic Press, USA.

Wyatt, S.D. and Brown, J.K. 1996. Detection of geminiviruses in aqueous extracts by polymerase chain reaction. *Phytopathology*, **86:** 1288-1293