Pak. J. Bot., 15 (1): 19–24, 1983.

IDENTIFICATION OF SOME VIRUSES INFECTING SOLANACEOUS HOSTS IN SIND

G.R. SOLANGI, S.M. MOGHAL* and S.D. KHANZADA

Department of Plant Pathology, Sind Agricultural University, Tandojam

Abstract

Virus diseases identified on the basis of symptomatology, host range, serology and transmission were tobacco mosaic virus (TMV) in tomato, tobacco and chillies, X, Y and leaf roll virus (LRV) in potato and leaf curl virus (LCV) in tomato, tobacco and chillies. Leaf curl virus group transmitted by whitefly was predominantly present in Sind.

Introduction

Solanaceous hosts are known to be attacked by a number of virus diseases throughout the world. According to the descriptions of plant viruses (Anonymous, 1970-80) about 30 distinct viruses are reported to potentially infect solanaceous hosts of economic importance. Very little work appears to have been done on specific virus diseases in Pakistan. Kamal & Moghal (1968) reported some virus diseases on the basis of symptom expression but none were identified properly. In this paper, the incidence and identification of some viruses of solanaceous hosts in the Sind region are reported.

Materials and Methods

Samples of virus or virus-like diseases of tomato, potato, tobacco, chillies and eggplant were collected in polyethylene bags and stored at 4°C until processed.

Inoculum was prepared by grinding infected tissue in 0.02 M phosphate buffer, pH 7.2 (1 gm/ml) with a pestle and mortar and strained through two layers of cheese cloth. The leaves of test plants were lightly dusted with 400-mesh carborundum, inoculated mechanically with infective sap and then rinsed with tap water to remove excess inoculum. All plants were raised in an insect – free room maintained at 24 – 27°C with 14 h-day length provided with artificial fluorescent light. Symptoms were recorded after two weeks as suggested by Bos (1970). Viruses isolated from different hosts were propagated in tobacco.

*Plant Pathology Section, Argicultural Research Institute, Tandojam, presently at The Pakistan Agricultural Research Council, ISLAMABAD.

Virus transmission studies were conducted with seed, dodder and the insects according to the methods described by Noordam (1973). To provide virus-free conditions, all plants were raised in an isolation room. For vector experiments insect cages, 100 x 100 x 80 cm made of wooden frames with glass and fine netwire panels (20 mesh/cm) to accommodate 10 pots each containing 2-3 test plants were used. One month old plants were preferred for virus transmission.

Serological tests were carried out by the tube precipitin technique (Ball, 1974). Test antigen was prepared by extracting sap from infected tobacco leaves in 0.02 M phosphate buffer, pH 7.2, containing 0.15% thioglycollic acid. The extracts were emulsified with equal volumes of chloroform and centrifuged at 5000 rpm for 15 minutes and supernatants were saved. Twofold dilutions of antisera were prepared in 0.14 M NaCl and mixed with test antigen (1:1) in small glass tubes. Reactions were noted after incubating the tubes at 37°C for 2 h and then overnight at 4°C Control tests were run with sap from healthy tobacco leaves.

Table 1. Incidence and symptoms of viruses in solanaceous hosts under field conditions.

Host.	No. of places visited.	No. of fields examined	% infection		Conditions and symptoms
			Range	Average	
Tobacco	5	10	20-45	32	Vein clearing, mosaic and chlorotic areas, yellowing, curling and thickening of leaves; association of aphids and whiteflies.
Tomato	10	30	50-85	69	Mild to severe mosaic of leaves and fruits, curling and thickening of leaves, bushy growth, association of whiteflies.
Chillies	10	20	20-35	30	Mild mosaic, stunting of plants, yellowing, curling and thickening of leaves, association of whiteflies.
Potato	5	7	20-45	35	Mild to severe mosaic, rugosity, yellowing, thickening and rolling of leaves, association of aphids.
Eggplant	10	20	2-6	3.5	Mild mosaic, slight curling and yellowing of leaves, association of whiteflies.

Results

1. *Incidence and nature of infection:* Virus diseases are widely prevalent and distributed in various fields and localities surveyed (Table 1). Mean incidence in tomato, potato, tobacco, chillies and eggplant being 69, 35, 32, 30 and 3.5%, respectively. Maximum infection ranging between 50-85% was recorded in tomato. Plants of all ages exhibited a variety of symptoms which were complex in tomato and chillies. Whiteflies were found to be commonly and abundantly associated with all the hosts.

2. *Host range and symptomatology:* Host range studies were made over a protracted period and a large number of plant species were artificially inoculated with extracts from infected plants. Symptoms produced by the sap transmissible viruses are presented in Table 2.

Typical virus symptoms as local lesions, vein clearing, mosaic and mottle and stunting appeared in 12 indicator plants. *Nicotiana* spp. and *Chenopodium* spp. were commonly infected with sap of all the diseased plants. *Datura stramonium and Gomphrena*

Table 2: Reaction of indicator plants following mechanical inoculations with infective sap.

Plant species.	Tobacco	Tomato.	Potato.	Chillies.	eggplant
Chenopodium album L.	NLL	CLL	CLL	CLL	CLL
C. amaranticolor C & R	NLL	NLL	LL,VC	CLL	CLL
C. murale L.	ni	ni	NLL,VC	ni	ni
Nicotiana tabacum L.	LI,M, VC,VN	M.VC	VC,M, VN,	m	ni
N. glutinosa L.	NLL,VN M	LI	VC,M	m	ni
N. clevelandii A. Gray	LI	VC,M	VC,M	m	ni
Lycopersicon esculentum L.	M,CA	VC,M.S	M	m,CA	ni
Solanum tuberosum L.	ni	ni	LI,M.	ni	ni
S. melongena L.	LI,M	ni	ni	ni	VC,M,S
Datura stramonium L.	ni	ni	CLL,M	ni	ni
Gomphrena globosa L.	ni	ni	CLL,CA	ni	ni

CA = Chlorotic areas (locally)
CLL = Chlorotic local lesions
NLL = Necrotic local lesions
VC = Vein clearing
VN = Veinal necrosis
M = Severe mottle or mosaic
m = mild mottle
LI = Latent infection
S = Stunting
ni = no infection

globosa gave positive reaction with infected potato only. A limited number of indicator plants was infected with sap from diseased eggplant. Plant species not infected were: *Brassica* spp. *Cucumis* spp. *Cyamopsis psoroloides* DC, *Cajanus cajan* L, *Glycine max* (Merr.) L, *Lathyrus odoratus* L, *Medicago spp., Phaseolus mungo* L, *Pisum sativum* L, *Vicia faba* L, *Vigna unguiculata* (L.) Walp., *Spinacea oleracea* L, and *Zea mays* L.

3. *Transmission of diseases: Seed transmission:* Indicator plants were inoculated with sap extracted from seedling raised from seeds of diseased plants. Seeds collected from infected tobacco, chillies and eggplant produced healthy seedlings but 60% of those from tomato seeds were infected. Back inoculation from infected tomato seedlings on to tobacco plants confirmed the presence of virus.

Dodder transmission: Ten cm pieces of dodder vines (*Cuscuta reflexa*), collected from citrus plants, were placed for parasitization on tobacco plants showing severe curling. After 3 weeks it was trained on a set of 12 healthy tobacco plants. All plants under test became infected and exhibited typical curling symptoms after about 4 weeks.

Insect transmission: Suspect vectors i.e. whitefly (*Bemisia tabaci* Genn.) and aphids (unkown sp.) were attempted in summer and spring season, respectively. Before each test, insects were starved for 1-2 h. Whiteflies were first fed on leaf-curled tobacco plants for 24 h before being transferred to test plants in groups of 12-18 insects for 2-day inoculation feeding. Aphids were allowed an acquisition period for 2-5 minutes on mosaic tobacco plant and for 1 h on infected potato plant and allowed inoculation feeding on respective test plants for same length of period.

Transmission succeeded in both the cases. Leaf curling and stunting symptoms developed in tobacco after 25 days and continued upto maturity. The results indicate that *B. tabaci* is an efficient vector of leaf curl disease. In test with aphids, mosaic appeared in 20 out of 30 tobacco plants after 2 weeks which was confirmed by mechanical inoculation on healthy plants. Leaf rolling and yellowing symptoms developed in 6 out of 10 potato plants after 30 days. All these symptoms did not appear in mechanical inoculations.

4. *Serology:* The results of serological tests are given in Table 3. All the samples of different antisera were imported and showed no reaction with healthy plants antigens. Positive reactions were obtained with antisera to TMV, PVX and PVY. No reaction took place with antisera to cucumber mosaic virus, tobacco ring spot virus, potato virus S and potato virus A in any of the tests.

Table 3.Serological testing of viruses of solanaceous hosts.

Antisera	Tobacco	Tomato	Potato	Chillies	Eggplant
TMV (Tobacco Mosaic virus)	64	16	0	8	2
TMV	32	32	0	8	0
PVX (Potato virus X)	0	0	16	0	0
PVX	0	0	16	0	0
PVY (Potato virus Y)	0	0	32	0	0
PVY	0	0	32	0	0
PVY	0	0	16	0	0

The figures are the dilution end points of antisera reached. All the antigens were prepared from tobacco leaves after propagation of viruses isolated from different hosts. 0 indicates no reaction.

Discussion

Evidence is presented that disease symptoms recorded in solanaceous hosts (Table 1) are caused by the viruses. Symptomatology and reactions in differential hosts proved the presence of mechanically transmissible viruses (Table 2), and serological tests confirmed their identity (Table 3). On the basis of these tests, viruses identified are TMV in tobacco, tomato and chillies, and PVX and PVY in potato. It is desirable to further characterize these viruses biologically and physically. The virus in eggplant caused symptoms in eggplant and certain alternate hosts. i.e. *C. album, C. amaranticolor* and *N. tabacum,* but its identity was not confirmed specifically as the serological tests were inconclusive. TMV was found to be seed-borne and severe viral infections in tomato as encountered in the fields may be attributed to seed-borne nature of disease. This agrees well with Broadbent (1952). Aphids appeared to be actively involved in the transmission and spread of PVY and PLRV in potato. It was tentatively identified as *Myzus persicae,* but it needs to be confirmed. LCV in tobacco was transmitted by whitefly as well as by dodder. In India, leaf curl in tomato and chillies is also shown to be transmitted by whitefly (John, 1957). Abundance of whitefly population and presence of susceptible hosts throughout the year may account for wide spread occurrence of leaf curl viruses in solanaceous hosts. Similar observations have been made by Ahmad (1978, 79) in leguminous hosts. Thus, leaf curl viruses, transmitted by whitefly, are predominantly present in the region and it will be interesting to study virus-vector relationship for this complicated virus group and to find out effective control measures.

Acknowledgements

We are thankful to D.G. Robertson, Research Scientist, International Potato Centre at Islamabad, for providing seeds of indicator plants and antisera to different viruses and to Mr. A.S. Kazi, Entomology Section, Agricultural Research Institue, Tando Jam for the identification of whitefly.

References

Ahmad, M. 1978. Whitefly (*Bemisia tabaci*) transmission of a yellow mosaic disease of cowpea, *Vigna unguiculata*. *Pl. Dis.Reptr.*, 62:224-226.

Ahmad, M. 1979. Studies with a whitefly-transmitted yellow vein virus of *Digera alternifolius*. *Phytophath Z.*, 96:21-24.

Anonymous (1970-1980) *Descriptions of plant viruses* (1-215). Commonwealth Mycol. Inst. Kew, Surrey, England.

Ball, E.A. 1974. Serological tests for the identification of plant viruses. Amer. Phytopath. Soc. Pl. Virology Comm. Minnesota U.S.A. pp. 31.

Bos, L. 1970. *Symptoms of virus diseases in plants*. Inst. Phytopath. Res. Wageningen, The Netherland, pp. 206.

Broadbent, L.H. 1952. The epidemiology of tomato mosaic virus. XI. Seed transmission of tomato mosaic virus. *Ann. Appl. Biol.*, 56:177-205.

John, T. 1957. A review of plant viruses in India. *Jour. Madras Univ.*, 27:373-450.

Kamal, M. and S.M. Moghal. 1968. *Studies on plant diseases of south west Pakistan*. Govt. Pr. Karachi pp. 207.

Noordam, D. 1973. *Identification of plant viruses-Methods and Experiments*. Centre for Agric. Publ. and Doc. Wageningen pp. 207.