PLANT VIRUSES AFFECTING PEPPERS IN JORDAN

BY

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I hereby recommend that this thesis prepared under my direction by Sami Fahed Batarseh entilted:

PLANT VIRUSES AFFECTING

PEPPERS IN JORDAN

be accepted as fulfilling the thesis requirement for the degree of MASTER OF SCIENCE.

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As members of the final Examination Committee, we certify that we have read this thesis and agree that it may be presented for final defense.

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INTRODUCTION

Peppers (Capsicum annuum L. and Capsicum frutescens L.) are among the important solanaceous crops in Jordan. approximately 7,984 donums were grown in Jordan, of which about 6,334 donums were planted in the Jordan Valley (1982). Most of the production was exported to the neighboring countries, which brought an annual income of about 650,781 Jordanian Dinars (11).

Virus diseases cause significant production problems for many vegetables and many viruses have been reported to occur in peppers and to cause yield reduction (7, 12, 13, 19, 23, 30, 43, 49).

Potato virus Y (PVY) and cucumber mosaic virus (CMV) were reported to occur in Jordan (28), while many viruses have been reported to infect peppers in several regions of the world (1, 2, 8, 9, 12, 13, 20, 22, 23, 26, 28, 34, 37, 41, 46, 49).

This study extends knowledge on virus diseases of peppers (Fig. 1), their indentification and assessment of actual prevalence.

LITERATURE REVIEW

Peppers are found naturally infected by more than twenty viruses. In addition there are about fourty two viruses which can infect peppers through mechanical inoculation (1, 6, 8, 13, 15, 19, 30, 32, 33, 45, 49).

Potato virus Y (PVY), cucumber mosaic virus (CMV), pepper veinal mottle virus (PVMV), pepper mottle virus (PMV) and tobacco etch virus (TEV) are commonly isolated from diseased peppers (7, 8, 14, 15, 20, 22, 24, 30, 33, 35, 39, 44, 45, 49).

Potato virus Y infects at least sixty plant species in the Solanaceae, Chenopodiaceae and Leguminosae. The disease resulted in a 44.33 percent reduction in pepper yields (9, 20, 21, 40). Infected pepper plants were severely mottled and stunted, local lesions followed by systemic necrosis causes death of a hypersensitive selection from Anahiem chili, mild to to severe mottle in Capsicum frutescens C V. Tabasco, and Datura stramonium L. is immune to infection by all tested strains (15, 21, 29, 37, 40, 44).

The virus is sap and aphids transmissible to pepper and number of other plants. No seed transmission has been observed (15, 21, 37). Many solanaceous weeds can be a major reservoir for the virus (40). Regarding viability of potato virus Y, in

crude extract it can withstand heating up to 55 C for 10 min. but not 57 C. The dilution end point is 10^{-2} - 10^{-3} and it loses it's infectivity between 2 - 3 days at 20 - 22C (15, 21, 37).

Cucumber mosaic virus has a very wide host range and a world-wide distribution. It is known to infect 775 species of plants representing 85 families and causes yield reduction in several food plants (19, 30).

Incidence of CMV in pepper (Capsicum annuum L.) may reach 80 - 100%, depending on environmental conditions, and is more likely to previal in the temperate regions, high temperatures apparently cause masking of symptoms (19, 46).

Besides being readily transmitted by mechanical inoculation, the virus is transmissible in the non-persistant manner by more than 60 species of aphids, particularly Myzus persicae and Aphis gossypii with the first of these the most efficient vector (5, 42, 46). Cucumber mosaic virus is seed-borne in seeds of at least four weed species and particularly those of Stellaria media (4, 5, 42).

In sap of diseased tobacco plants, cucumber mosaic virus has thermal inactivation point of about 70 C for 10 min., dilution end point of 10^{-4}, and longevity in vitro of 3-6 days (27, 31).

Pepper veinal mottle virus causes severe leaf chlorosis in Petunia hybrida and leaf mottling, severe leaf distortion and considerable loss of yield in naturally infected Capsicum annuum and Capsicum frutescens. The virus is sap and aphid transmissible to Solanaceae and other families. Myzus persicae and Aphis gossypii transmitted the virus in a non-persistant manner. This virus is not transmitted through the seeds of infected Solanum melongena, Datura metel, Capsicum annuum and Nicotiana clevelandii (14, 23, 24, 33, 35, 36, 43).

In Capsicum annuum sap, the thermal inactivation point is 60 - 65 C for 10 min., dilution end point 10^{-3}-10^{-4} and a longevity in vitro at 25 C for 7-8 days (14, 24, 33, 35, 45).

Pepper mottle virus infects many species of Solananceae, particularly of Capsicum and Nicotiana and is known to have several strains. Some isolates produce local lesions in Chenopodium amaranticolor the virus causes mottle diseases of Capsicum annuum and Capsicum frutescens with fruit distortion by some strains, one strain doesn't produce local lesions in Capsicum frutescens C V. Tabasco. It has a thermal inactivation point between 50 - 55 C for 10 min., dilution end point 10^{-2}- 10^{-3} and longevity invitro of 3 days at 28 - 36 C (7,47). Pepper mottle virus is transmitted in a non-persistant manner by several species of aphids, particularly Myzus persicae, Aphis gossypii, and Aphis craccivora, with the first of these the most efficient vector. It is sap transmissible

but not through seeds (7, 47, 49).

The severity of symptoms in cultivated plants depends greatly on the host species and cultivar and on the virus strain. Various physiological and chemical changes occur in virus-infected plants, depending on the stage of infection. Rapid spread in the field occurs mainly when aphid populations, especially of the green peach aphid (Myzus persicae), are high (4, 5, 18, 47, 49).

Tobacco etch virus in peppers poses a special problem due to severe wilt and death syndrome associated with Capsicum frutescens C V. Tabasco infection. TEV causes necrotic and/or Chlorotic mottle diseases of tobacco, pepper, tomato and other solanaceous plants, Sicklepod (Cassia obtusifolia) is a secondary host for TEV, the infected branches of this host are stunted and usually exhibit chlorotic mottle (9, 10, 16, 30).

More than ten species of aphids, and especially Myzus persicae, transmit the virus in a non-persistent manner and it is readily transmissible by inoculation of sap. The presence of naturally infected weed plants increases the propagation of this virus disease (5, 17, 18).

In tobacco sap, the thermal inactivation point of TEV is about 55 c° for 10 min., dilution and point about 10^{-4} and longevity in sap for 5 - 10 days at 20 C (10, 16, 30).

Tomato aspermy virus has a wide host range, infecting over 75 species in the families Chenopodiaceae, Compositae, and Solanaceae. Systemic symptoms in most species are much more severe in winter than in summer (38.25). The virus prevented seed formation in tomato but Cucumer mosaic virus did not.

The virus infects Capsicum frutescens L., Nicotiana tabacum L. conn., Datura stramonium L., petunia hybrida vilm; Lycopersicon esculentum Mill, Chenopodium quinoa and Chenopodium amaranticolor. In tobacco sap, the thermal inactivation point 55-60 C for 10 min, dilution end point 10^{-4}- 10^{-5} and longevity in vitro 2-6 days at 20 C (38).

The virus is sap transmitted and 10 species of aphids especially Myzus persicae and Aphis gossypii (38,12).

An isolate from Campanula rapunculoides was seed-borne in Stellaria media but not seed-borne in Nicotianaglutinosa, Nicotiana tabacum and lycopersicon esculentum (38).

MATERIALS AND METHODS

Sample collection and Indexing:-

Leaf samples from 203 plants showing virus-like symptoms were collected from different locations in the Jordan Valley. All samples were collected in plastic bags and stored in the refrigerator over night (App. 3). Samples were then tested for virus presence by mechanical inoculation, since pepper viruses were reported to infect many hosts, the following indicator plants were used to differentiate them (13, 21, 30, 37, 45, 45, 49). Chenopodium quinoa Willd, Chenopodium amaranticolor Coste & Reyn., Capsicum annuum L. C. V. Fancy, Capsicum frutescens L. C. V. Tabasco, Capsicum frutescens L. C. V. Anaheim Chili, Cucumis sativus L. C. V. Floramore, Cucumis melo L. C. V. Flexuous, Cucumis melo L. C. V. Black Seed, Cucurbita pepo L. C. V. Ara-non, Nicotiana glutinosa L., Nicotiana tabacum L. Conn. C.V. Havana 423, Amarathus caudatus L., Gomphrena globosa L., Phaseolus vulgaris L. C. V. Tender Green, Datura stramonium L., Datura metel L., Lycopersicon esculentum Mill. C.V. Raf, Petunia hybrida Vilm., and Nicandra physaloides (L.) Gaertn. (Tab. 1).

Inocula were prepared by macerating leaf samples in 0.1 M neutral phosphate buffer, containing 0.01 M sodium diethyl diethiocarbamate (Na-DIECA) and 0.0 1m cystein hydrochloride,

using a sterilized mortar and pestle (33, 34). The resulting extract of each sample was rubbed on leaves of virus indicator plants that had been dusted with carborundum particles of 6000 mesh. All inoculated plants were grown in methyl bromide-fumigated soil and maintained under green house conditions.

Incidence Study:-

Surveys on the incidence of virus symptoms in commercial pepper fields started in the Jordan Valley during mid-August 1983 and continued through mid-May 1984. This would cover the general pattern of pepper production which includes a fall period when transplants are moved to the field from mid-to-late August and a spring production period when transplants are moved to the field from mid-October untill mid November. Harvest in the spring continuous until mid-May.

The incidence study of mosaic diseases was determined in six representative pepper fields selected along the Jordan Valley, which include representative regions of pepper production in the Jordan Valley (App. 1, 2). For the fall pepper production season extending from mid-August 1983 to the end of December 1983, the survey covered fields in the following areas:-

Al-Kafrain (Capsicum annuum L.); El-Karameh (Capsicum frutescens L.); Deir-alla (Capsicum annuum L.); Kreiymeh (Capsicum frutescens L.); Wadi el-yabis (Capsicum frutescens

L.); and El-Mashare (Capsicum annuum L.).

Sites for the spring pepper production season extending from mid-October 1983 to mid-May 1984 included:-

El-Sakneh (Capsicum annuum L. inter planted with tomato and cucumber); El-Sakneh (Capsicum frutescens L.); El-Karameh (Capsicum frutescens L. inter planted with squash); Muthallath el-Masri (Capsicum annuum L. inter planted with tomato); Deir-alla (Capsicum annuum L.); North-Shuneh (Capsicum annuum L.); and North-Addasieh (Capsicum annuum L.).

Number of plants showing mosaic symptoms was counted at weekly intervals, each time out of 100-pepper plants in four rows taken at random in each pepper field. The percentage of plants with virus symptoms was calculated for each area on each date of collection.

Virus isolation:-

Out of a total 203 samples collected, 177 virus isolates were recovered on the basis of diagnostic reactions developed on the indicator plants. From these isolates two virus isolates were selected to represent the remaining isolates depending on the diagnostic reactions. 171 virus isolates represented by 83-907.isolate, while other six isolates represented by 83-950.

The selected isolates were passed through single lesion transfers in Chenopodium quinoa (for 83 - 950) and by serial

dilution on Capsicum frutescens (for 83 - 907), 0.1 ml of the extract was subjected to serial 10-fold dilutions, then tested for infectivity by carborundum mechanical inoculation into Capsicum frutescens. Plants that developed symptoms when challenged with the final diluted inoculum were considered as the source of the virus. The two isolates were maintained on Nicotiana glutinosa or Nicotiana tabacum H. 423.

Host range:-

All tested indicator plants were developed to the cotyledon or the first true leaf stage. At least 10-plants of every species to be tested were mechanically inoculated with the two isolates.

The inocula were prepared by macerating young infected leaves of source plants in 0.1 M neutral phosphate buffer containing Na-DIECA and cystein hydrochloride (0.01 M), using sterilized mortars and pestles.

Mechanical inoculations were made by rubbing a fore-finger on to leaves previously dusted with 6000 mesh carborundum. Plants were observed for symptoms for 3-4 weeks or longer after inoculation. Inoculated and tip leaves of all plants were then tested by mechanical indexing to Chenopodium quinoa (for 83 - 950) or Capsicum frutescens (for 83 - 907).

All plants used in these tests were kept throughout the

experiment in growth chambers (6000 Lux for 16 hr., 25-28 C).

Aphid transmission:-

The green peach aphid (Myzus persicae) was used for all transmission tests. For each virus isolate, non-viruliferous apterous aphids reared in caged virus-free eggplant (Solanum melongena L.) were transferred with a camel hair brush to plastic dishes, left to starve for one hour and then moved to young virus-infected source plants. About 400 aphids were kept on these infected plants for 5, 10, 15, 30, or 60 minutes and then five aphids per plant were transfered to 10 healthy plants. The inoculation feeding period was the same length as the acquisition feeding period. The aphids were then killed by insecticide spray.

Properties in crude sap:-

Infected leaves of Nicotiana glutinosa were macerated in 0.1 M neutral phosphate buffer containing 0.01 M Na-DIECA and Cystein hydrochloride 0.01 M (1gm/1 ml buffer) and extract was filtered through two layers of cheesecloth.

The thermal inactivation point (TIP) was determined by heating 0.5 ml of extract in eight test tubes at different temperatures (50, 55, 60, 65, 70, 75, 80, 85 C) for 10 min. then the heated extracts were stored in an ice bath until all aliquots received the predetermined heat treatment.

For dilution end point (DEP) another 0.1 ml of the extract was subjected to serial 10-fold dilutions. For the longevity in vitro (LIV) test 3 ml of the extract was kept at room temperature, then tested daily for infectivity by mechanical inoculation in to susceptible host plants. Treatments were assayed by mechanical inoculation of Capsicum annuum (for isolate 83 - 907) or Chenopodium quinoa (for isolate 83 - 950) Assay plants.

Seed transmission:-

To determine whether or not virus isolates (83-907 and 83 - 950) were seed transmitted, seeds of infected Nicotiana glutinosa (for 83-907), Capsicum annuum and Nicandra physaloides (for 83 - 950) were collected. About 200 seeds were sown in sterilized peatmoss in plastic pots, then transplanted into separate pots cotaining methyl bromide-fumigated-soil and kept in growth chambers. Plants were observed for about 6 - 8 weeks after transplanting, back-indexing mechanical inoculation tests were done on Chenopodium quinoa or Capsicum annuum (for 83-950), and on Capsicum frutescens (for 83-907).

Serology:-

Serological tests consisted of an agglutination precipitin test (for isolate 83 - 907) and an agar-double diffusion test (for isolate 83 - 950). Antisera of potato virus Y (obtained from Dr. G. I. Mink, Washington State University) was determined using an agglutination precipitin test method. Healthy

leaves of Nicotiana glutinosa were macerated in 0.1 M neutral phosphate buffer (1 gm/1ml buffer) and infected leaves of Nicotiana glutinosa (for isolate 83 - 907) were macerated in the same buffer, then two drops of each extract were placed in a Petri dish and mixed with one drop of potato virus Y antisera and subsequently observed for agglutination reactions.

Agar-double diffusion tests were conducted in Petri dishes of 0.8% ion agar dissolved in boiling distilled water to which both 0.85% NaCl and 0.25% sodium azide had been added. Crude sap extracted from infected Nicotiana glutinosa (for isolate 83 - 950) was used as antigen. Antiserum to tomato aspermy virus was kindly supplied by Dr. A.A. Brunt (Glass house crops research institute little Hampton. England) while cucumber mosaic virus antiserum was provided by Dr. G. I. Mink (Washington state University) Gel patterns developed as diffusion reaction between centeral well (5 mm in diameter) and peripheral wells (5 mm in diameter) 5 mm from the centeral well Antiserum of tomato aspermy virus and of cucumber mosaic virus was placed in the center and respective antigens perpared from 1:1 W/V tissue extracts in 0.03 M neutral phosphate buffer were placed in peripheral wells. Dishes were kept at laboratory temperature and examined daily for 3 days.

Electron microscopy:-

Samples of leaves from infected Nicotiana glutinosa (for

isolate 83 - 907) showing mottled symptoms were ground in 0.1 M neutral phosphate buffer. Drops of extract were mounted on EM grids with formvar film, then negatively stained with 2% potasium phosphotungstate (PTA, PH 6.5).

RESULTS

Out of a total of 203 samples collected, 177 virus isolates were recovered. Among the virus isolates 171 reacted in a similar pattern in differential host range tests and were identified later as potato virus Y. The remaining 6 isolates had a similar host range that were identified as tomato aspermy virus (Table. 1). Thus the study showed that PVY and TAV were recovered in 85% of the total samples tested in the fall of 1983 and the spring of 1984. PVY was recovered from sweet pepper and hot pepper fields in spring and fall seasons (App. 3). Of all PVY isolates recovered in the spring 72.19% and 27.81% were isolated from hot pepper and sweet pepper fields, respectively. In the fall season 75% and 25% of all PVY isolates were recovered from hot pepper and sweet pepper fields respectively.

TAV was recovered only in the fall season. Of all TAV 33.33 were recovered from sweet pepper fields, while 66.67% were recovered from hot pepper fields.

Of all isolations 97% were identified as PVY and 3% as TAV (Table. 2). PVY was the predominant virus and was more prevalent in the spring. TAV was only recovered from two locations (EL-Kafrain and EL-Mashare') (App. 3).

Table 1. Symptomology of two virus isolates from pepper on selected test plants.

	Virus isolates and symptoms [1] on test plants	
	Virus isolate (83-907)	Virus isolate (83-950)
Capsicum annuum	SM	SM
Capsicum frutescens	SM	SM
Datura stramonium	-Ve	SM
Nicandra physaloides	-Ve	SM & SS
Petunia hybrida	-Ve	SMe.
Gomphrina globosa	-Ve	SCR & NBL
Cucurbita pepo	-Ve	SM
Nicotiana glutinosa	SM	SM
Lycopersicon esculentum	SM	SM
Cucumis melo CV. flexuous	-Ve	SM & E

1. SM = Systemic mottle, -Ve = No symptoms, SS = Shoestring, SMe.= Systemic mosaic, E: Enation

Incidence study:-

The incidence of mosaic diseases affecting hot peppers was studied in three fields. The infection was first observed in late October or early November and reached maximum in late December. The maximum incidence ranged between 26 - 35% in two hot pepper fields and in a third field the incidence reached 6% by the end of November (Fig. 2). However, study of this last field was discontinued because the farmer ploughed the field. In sweet peppers, no visible symptoms could be detected in plants selected randomly for the purpose of incidence studies in three fields located in EL-Karameh, Kreiymeh, and Wadiel-yabis. However, tissue collected from sweet pepper fields not included in the incidence studies showed mosaic symptoms from which PVY and TAV were isolated (Table. 1).

In the spring season virus symptoms were observed in early and late February in hot pepper fields. The incidence of diseased pepper plants increased slowly to the end of March in all surveyed fields. By early April the incidence of mosaic diseases build up quickly and spread fast reaching 75 - 95% in three fields. However, the incidence in the fourth and fifth fields (EL-Sakneh and Muthel-Masri), was consistently low and the spread of the disease(s) was slow. In these fields the incidence reached 5 - 12% by mid-May (Fig. 3).

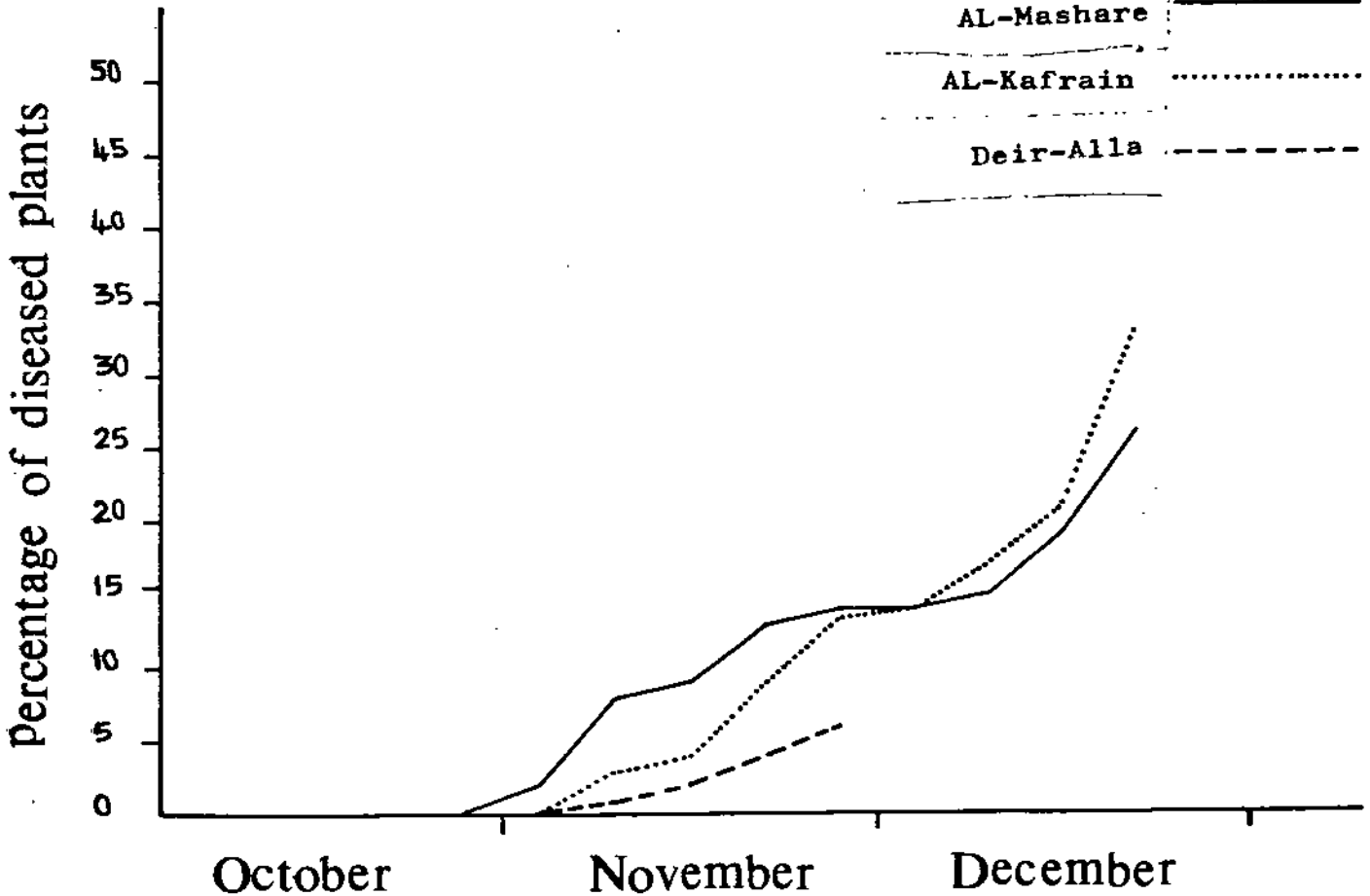

Fig. 2. Incidence of mosaic diseases in hot pepper fields (C. annuum) during the fall-planted growing period 1983 in the Jordan - Valley.

Fig. 3. Incidence of mosaic diseases in hot pepper fields (C. annuum) during the spring-planted growing period (1983 - 1984) in the Jordan - Valley.

Table 2. Identification of viruses isolated from pepper fields in fall and spring during the period 1983 - 1984.

Date/Plant species	Virus isolate recovered	PVY identification (83-907)			TAV identification (83-950)		
		H.R	H.R & phy. prop.	H.R. Phy. pro. E.M. and Serology	H.R	H.R.& Phy. Prop.	H.R,Phy. prop. and serology
Fall 1983	26						
C. annuum	17	15	1	1	2	0	0
C. frutescens	9	5	0	0	4	2	1
Spring 1984	151						
C. annuum	109	109	2	1	0	0	0
C. frutescens	42	42	0	0	0	0	0
Total	177	171	3	2	6	2	1

H.R. : Host range, Phy. Prop.: Physical properties, E.M.: Electron Microscopy.

In sweet peppers during the spring season diseased plants were first observed in late February and early March. The spread of the disease(s) was/were fast in one field (EL-Sakneh) reaching 100% by the end of April whereas it was very slow in the other field (EL-Karameh) with a final incidence of 12% by the end of the season (Fig. 4).

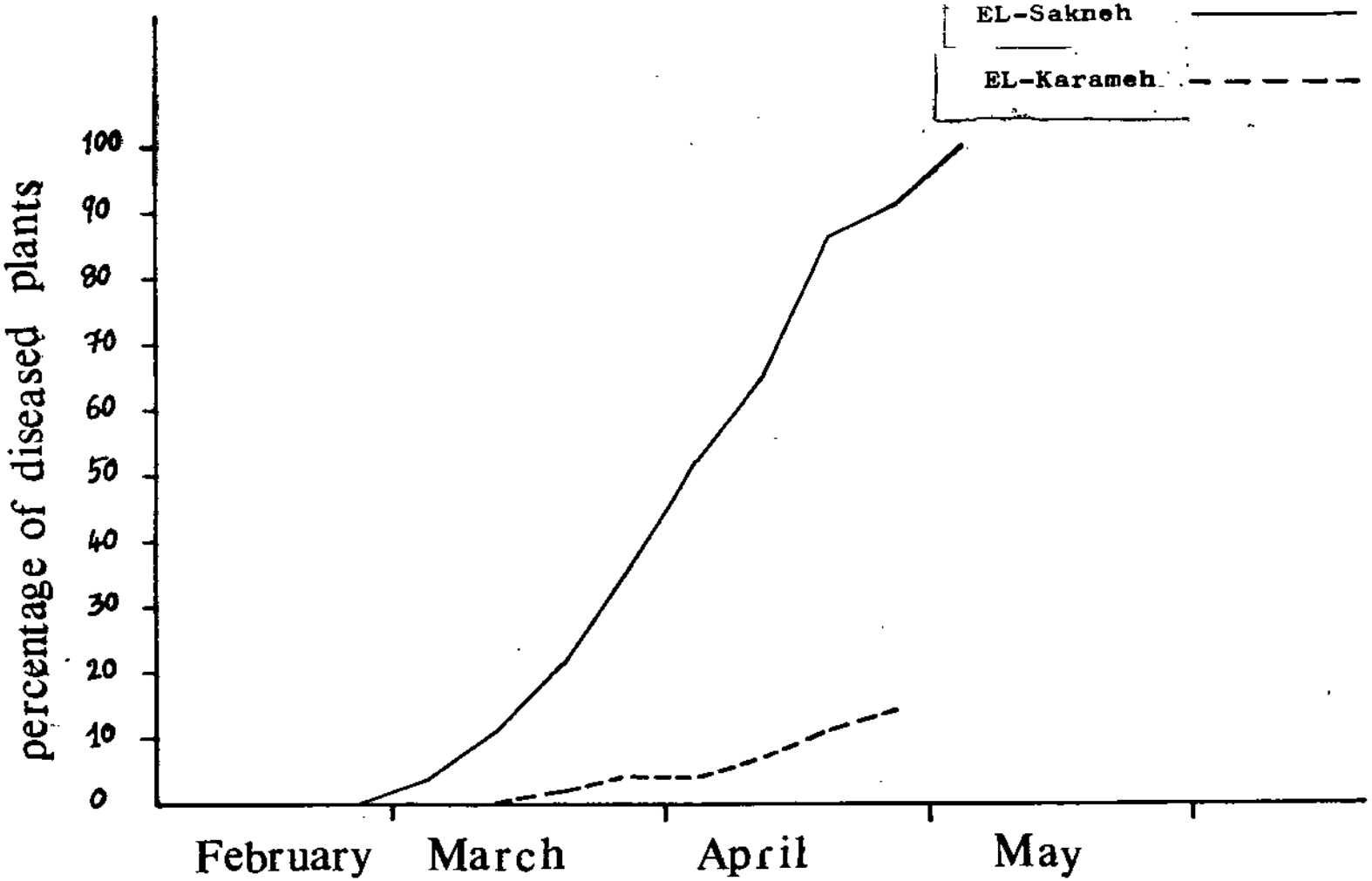

Fig. 4. Incidence of mosaic diseases in sweet pepper fields (C. Frutescens) during the spring-Planted growing period (1983 - 1984) in the Jordan Valley.

Host range and symptomology:-

Isolate 83 - 907 produced symptoms on pepper 18 days after inoculation. Mild to severe mottle appeared in the first true leaves (Fig. 5). Plants belonging to 23 species in 8 genera from 6 families were mechanically inoculated with crude extracts from Nicotiana glutinosa infected from field-collected peppers, Infection with this isolate was restricted to Solanaceae and was not transmissible to the plants of any other family tested.

The virus produced systemic mottle on Capsicum annuum, Capsicum frutescens, Nicotiana tabacum H. 423, Nicotiana glutinosa and Lycopersicon esculentum (Table. 3). None of the follwoing plants showed symptoms or proved to be symptomless carriers after back indexing through 30 days post inoculation:- Amaranthus caudatus, Datura stramonium, Vicia faba, Chenopodium quinoa, Chenopodium amaranticolor, Chenopodium album, Medicago sativa, Phaseolus vulgaris, Vigna ungiculata, Nicandra Physaloides, Cucumis melo, Cucumis sativus, Cucurbita pepo, Gomphrena globosa, Petunia hybrida, and Physalis angulata.

The remaining isolates represented by 83 - 950 produced systemic mottle coupled with filiform leaves and distorted fruits of pepper 21 days after inoculation (Fig. 6). Plants

Table 3. Host range and symptoms of PVY isolates (represented by virus isolate 83 - 907)

Host range	Symptoms[1]	Back indexing[2]	
		inoculated Leaves	Systemic Leaves
Capsicum annuum	SM	+ Ve[3]	+ Ve
Capsicum frutescens	SM	+ Ve	+ Ve
Nicotiana tabacum H. 423	SM	+ Ve	+ Ve
Nicotiana glutinosa	SM	+ Ve	+ Ve
Lycopersicon esculentum	SM	+ Ve	+ Ve

1. SM = Systemic mottle.
2. Back indexing on Capsicum frutescens CV. Anahiem Chili.
3. + Ve = Symptoms expressed.

belonging to 30 species in 24 genera from 6 families were mechanically inoculated with crude sap extracts from Nicotiana glutinosa infected from field collected peppers. The virus produced systemic mottle in Capsicum annuum, Capsicum frutescens, Nicotiana glutinosa, Nicotiana tabacum H. 423, Datura stramonium, Lycopersicon esculentum, and Cucurbita pepo. Systemic mosaic occurred in Petunia hybrida while systemic mottle followed by shoestring was seen in Nicandra physaloides. Systemic mottle and enation was seen in Cucumis melo CV. flexuous. Chlorotic or necrotic local lesions were produced on Chenopodum quinoa, Chenopodium album and Chenopodium amaranticolor if the source of inoculum was not peppers. Systemic yellowing converted to necrotic brown rings, in Gompherena globosa (Tab. 4).

None of the following plants showed systemic or were shown to be symptomless carriers after back indexing:-

Amaranthus caudatus, Medicago sativa, Phaseolus vulgaris CV. Tender Green, Phaseolus vulgaris CV. Gold Crop, Phaseolus vulgaris CV. Top crop, Vicia faba, Cucumis sativus CV. Zeas, Cucumis sativus CV. Floramore, Cucmis sativus CV. Baita Alapha, Cucumis mole CV. Black Seed, Cucumis melo CV. Charleston, Cucumis melo CV. Chilean black, Vigna ungiculata CV. California Black Eye and Brassica oleraceae.

Table 4. Host range and symptom reactions of TAV isolates (represented by virus isolate 83-950)

Host range	Symptoms[1]	Back indexing[2]	
		Inoculated leaves	Systemic leaves
Capsicum annuum	SM	+ Ve[3]	+ Ve[3]
Capsicum frutescens	SM	+ Ve[3]	+ Ve[2]
Nicotiana tabacum H.423	SM	+ Ve	+ Ve
Nicotiana glutinosa	SM	+ Ve	+ Ve
Datura stramonium	SM	+ Ve	+ Ve
Petunia hybrida	SMs.	+ Ve	+ Ve
Chenopodium quinoa	C.L.L.[4]	+ Ve	- Ve
Chenopodium amaranticolor	C.L.L.[4]	+ Ve	- Ve
Nicandra physaloides	SM & SS	+ Ve	+ Ve
Gomphrena globosa	SCR & NBL	+ Ve	+ Ve
Lycopersicon esculentum	SM	+ Ve	+ Ve
Cucurbita pepo	SM	+ Ve	+ Ve
Cucumis melo CV. flexuous	SM & E	+ Ve	+ Ve
Solanum melongena	LM	+ Ve	- Ve
Physalis angulata	Sch.	+ Ve	+ Ve
Datura metel	SM	+ Ve	+ Ve

1. SM = Systemic mottle, SMs = Systemic mosaic, CLL = Chlorotic Local lesions, SS = Shoe string, SCR & NBL = Systemic Chlorotic rings turned to necrotic brown lesions, E: Enation LM = local masked infection, Sch = Systemic chlorosis.
2. Back indexing on Chenopodium quinoa.
3. Back indexing on Capsicum annuum.
4. Source of inoculum not pepper.

Properies in Crude sape:-

The general properies of 83 - 907 were determined. The virus in crude sap extract of Nicotina glutinosa could withstand heating up to 50 C for 10 min. but not 55 C. The dilution and point of the virus was found to be 10^{-4} and it lost its infectivity between 5 - 7 days at 20 - 25 C.

The general properties of 83 - 950 in crude sap extract of Nicotiana glutinosa had thermal inactivation point of 60C. Dilution end point of 10^{-3} and longevity in vitro of 3 - days.

Transmission by aphids:-

Both isolates (83 - 907 and 83 - 950) were readily transmitted by Myzus persicae in non-persistant manner (Table 5,6). The percentage of infection decreased when aphids were allowed to remain on test plants for extended periods of feeding. This is expected with stylet-born viruses (14, 19).

Seed transmission:-

All plants grown from seeds collected from plants infected with the two isolates (83 - 907 and 83 - 950) did not show any symptoms even after back indexing by mechanical inoculation.

Serology tests:-

The agglutination precipitin test of healthy leaf extract did not show any reaction but infected leaf extract isolate

Table 5. Transmission of (isolate 83 - 907) by Myzus persicae.

Acquisition feeding period/ inoculation feeding period in minutes.	% of infected plants
5/5	86.7
10/10	60
15/15	69.6
30/30	64.6
60/60	48.1

Table 6. Transmission of (isolate 83 - 950) by *Myzus persicae*.

Acquisition feeding period/ inoculation feeding period in minutes.	% of infected plants
5/5	72.7
10/10	38.9
15/15	27.3
30/30	41.9
60/60	4.0

83 - 907) showed clearly positive agglutination precipitin reaction, consisting of distinctly clumped precipitate.

Agar double diffusion tests with tomato aspermy virus antiserum gave strong reactions with antigens of infected Nicotiana glutinosa (isolate83 - 950) in a white clear percipitin lines. There was no such reaction lines with extracts of healthy Nicotiana glutinosa (Fig. 7). Cucumber mosaic virus antiserum also gave weak reactions with antigens of infected Nicotiana glutinosa with less clear light precipitin lines.

Electron Microscopy:-

Flexuous, filamentous particles were consistently found in crude extracts from Nicotiana glutinosa leaves infected with PVY (isolate 83 - 907) and were never seen in healthy leaf extracts. Particles measured were between 690 - 750 nm long (Fig. 8).

DISCUSSION

On the basis of host range, symptomatology, mode of transmission, and general properties, the two isolates were identified as potato virus Y (for isolate 83 - 907) and tomato aspermy virus (for isolate 83 - 950).

The identification was further substantiated by serology for both isolates and electron microscope (for the PVY isolate only). Unlike pepper mottle virus, pepper veinal mottle virus, and tobacco etch virus, potato virus Y doesnot infect Datura stramonium which is immune to all PVY strains (15, 29, 40).

Tomato aspermy virus doesnot infect all cucurbits or cowpea as cucumber mosaic virus, but light precipitin lines in serology tests were observed between cucumber mosaic virus antiserum and tomato aspermy virus antigens (isolate 83 - 950). Tomato aspermy virus antigens produced clear precipitin lines with tomato aspermy virus antiserum. On this evidence isolate 83 - 950 was identified as tomato aspermy virus which is serologically related to cucumber mosaic virus (13, 25, 38).

Chenopodium quinoa, Chenopodium album and Chenopodium amaranticolor showed no symptoms and from which virus was not recovered by back indexing if they were inoculated with crude pepper sap prepared from 83 - 907 or 83 - 950 isolates. However, local lesions were observed if several other plants

infected with isolate 83 - 950 were used as source of inoculum on the three Chenopodium hosts. This observation may due to virus inhibitors present in pepper extracts (37, 44).

Incidence studies of diseased pepper plants showed hot pepper infection only of relatively low level in the fall and high total levels of infection in both hot and sweet peppers in the spring. These differences between fall and spring may be due to behavioral differences in winged aphid populations or seasonal sources of inoculum could be high in spring and low in fall (19, 27, 31). It is noticed that low incidence of potato virus Y in the spring season in two fields (El-Sakaeh and Muthalleth EL-Masri) of hot pepper and one field (El-Karameh) of sweet pepper was associated with inter cropping system in these fields. Intercropped plants were tomatoes and cucumber or squash or tomatoes, respectively. Such intercropping may result in greater attraction of hosts other than pepper to winged aphids or to partial shading of the pepper plants (6,18).

This study shows that during the 1983 and 1984 seasons potato virus Y and tomato aspermy virus were the viruses affecting peppers in the Jordan Valley. Of these two the former one was the most important since it was the predominant isolate (from 203 samples collected, 171 were PVY) and reached field incidence of 100%. Tomato aspermy virus was only found in six samples from two locations out of 203 samples

tested from 9 locations (App. 3), and field incidence was too low to calculate. From this evidence we can conclude that any attempt to control pepper viruses in the Jordan Valley should concentrate on potato virus Y.

SUMMARY IN ENGLISH

A survey was conducted to identify the most important viruses in pepper fields, and study their prevalence during the period 1983-1984, in the Jordan Valley.

177 virus isolates were recovered from a total of 203 samples collected through the study period. Of all virus isolates 96.61% were identified as potato virus Y and 3.39% as tomato aspermy virus.

Incidence of PVY was high and its spread was faster in spring as compared to that in the fall growing season.

TAV had a negligable role in the virus disease picture in peppers during the fall. However, the virus was recovered only in the fall growing season.

It is noticed that the low incidence of mosaic diseases were seen in pepper fields intercropped with tomatoes or cucurbits.

SUMMARY IN ARABIC

ملخـــص بالعربيـــة

" دراسة الأمراض الفيروسيه التي تصيب محصول الفلفل في الأردن "

تمت دراسة لأهم الأمراض الفيروسيه التي تصيب محصول الفلفل في الأردن خلال الفتـــــرة ١٩٨٣ ـ ١٩٨٤م. ولدى فحص العينات على النباتات المشخصه للأمراض الفيروسيه وعزلها قسّمـــت العينات الى مجموعتين، الأولى تحوى " ١٧١ " عينه مرضيه والثانيه تحوى " ٦ " عينات مرضيـــه. وبعد اكتمال الفحوصات على المدى العوائلي، الخواص الفيزيائيه، انتقال بواسطة الحشرات، انتقال بواسطة البذور، الفحص بواسطة الميكروسكوب الالكتروني ثم الفحص السيرولوجي لسلالتين مستنبتيـــن للمجموعتين، وجد أن محصول الفلفل يصاب بمرض فيروس البطاطا واى ومرض فيروس البندوره الأسبيرمي وان الأول هو السائد والأكثر انتشارا.

ولدى دراسة شدة المرض ووقت ظهوره وجد أن الاصابه في العروة الخريفيه تبدأ مع نهاية شهر تشرين الأول وبداية تشرين الثاني لتصل أشدها في نهاية كانون أول بنسبة تتراوح بين ٢٥ ـ ٣٥٪.

أما في العروة الربيعيه فان أعراض المرض تبدأ بالظهور خلال شهر شباط لتصل أشدها في نهاية شهر نيسان وبداية شهر أيار بنسبة تتراوح بين ٧٥ ـ ١٠٠٪. ولقد لوحظ ان شدة الاصابه فـــي الحقول المتداخله الزراعه قليله في كافة مواقع الدراسة لتصل أشدها بنسبة حوالي ١٥٪.

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Appendix 1. Region, Pepper species, and time of planting of the 6 - pepper fields that were assigned for the incidence study of the (1983) fall growing season.

Region	Pepper species	Time of planting
Kufrain	C. annuum	20-7-83
El-Karameh	C. frutescens	13-9-83
Deir-Alla	C. annuum	20-8-83
Kreiymeh	C. frutescens	23--8-83
Wadi El-Yabis	C. frutescens	25-8-83
El-Mashare	C. annuum	18-9-83

Appendix 2. Region, pepper species, and time of planting of the 7 - pepper fields that were assigned for the incidence study of the (1984) spring growing season.

Region	Pepper species	Time of planting
El-Sakneh	C. annuum	28-11-83
El-Sakneh	C. frutescens	28-11-83
El-Karameh	C. frutescens	20-10-83
Muth. El-Masri	C. annuum	25-10-83
Deir-Alla	C. annuum	15-10-83
North-Shuneh	C. annuum	25-10-83
North-Addasieh	C. annuum	15-10-83

Appendix 3. Viruses recovered from leaf samples collected from Various pepper fields, in the Jordan Valley at different areas. (1983 - 1984).

Collection Date	Area	Sample No.	Pepper spp.	Virus(s) recovered
4-6-83	N.Addasieh	83-877	C. annuum	PVY
20-7-83	N.Addasieh	83-907	C. annuum	"
25-7-83	N.Addasieh	83-909	C. annuum	"
9-8-83	El-Kafrain	83-910	C. annuum	"
9-8-83	El-Kafrain	83-911	C. annuum	"
9-8-83	"	83-912	"	"
9-8-83	"	83-913	"	"
9-8-83	"	83-914	"	"
9-8-83	"	83-915	"	"
9-8-83	"	83-916	"	"
9-8-83	"	83-917	"	-
13-8-83	El-Mashare	83-925	"	-
13-8-83	"	83-930	C.frutescens	-
20-8-83	El-Kafrain	83-939	C. annuum	"
20-8-83	"	83-940	"	"
20-8-83	"	83-941	"	"
20-8-83	"	83-942	"	"
20-8-83	"	83-943	"	"
20-8-83	"	83-944	"	-

Collection Date.	Area	Sample No.	Pepper spp.	Virus(s) recovered
20-8-83	El-Kafrain	83-945	C. annuum	PVY
20-8-83	"	83-946	C. frutescens	"
20-8-83	"	83-947	"	TAV
20-8-83	El-Kafrain	83-948	"	TAV
20-8-83	"	83-949	"	PVY
20-8-83	"	83-950	"	TAV
29-8-83	El-Moshare	83-954	C. annuum	PVY
29-8-83	"	83-955	"	TAV
29-8-83	"	83-956	C. frutescens	TAV
8-9-83	El-Kafrain	83-958	C. annuum	PVY
24-10-83	"	83-959	"	"
24-10-83	Wadi El-Yabis	83-960	C. frutescens	"
7-11-83	El-Kafrain	83-961	C. annuum	TAV
7-11-83	Deir-Alla	83-962	"	PVY
7-11-83	El-Mashare	83-963	"	"
7-11-83	"	83-964	"	"
7-11-83	"	83-965	"	"
21-11-83	El-Kafrain	83-966	"	"
21-11-83	"	83-967	"	"
21-11-83	Deir-Alla	83-978	"	"
21-11-83	El-Mashare	83-969	"	"
21-2-84	N.Shuneh	84-971	C. frutescens	"

Collection Date	Area	Sample No.	Pepper spp.	Virus(s) recovered
21-2-84	N.Shuneh	84-972	C. annuum	PVY
21-2-84	N.Addasieh	84-973	C. annuum	PVY
21-2-84	N.Shuneh	84-974	C. frutescens	"
21-2-84	"	84-975	"	"
21-2-84	N.Addasieh	84-976	C. annuum	"
21-2-84	"	84-977	C. frutescens	-
21-2-84	N.Shuneh	84-978	"	PVY
28-2-84	El-Sakneh	84-980	C. annuum	-
28-2-84	N.Addasieh	84-981	"	-
28-2-84	El-Sakneh	84-982	"	-
28-2-84	N.Shuneh	84-983	"	PVY
28-2-84	N.Addasieh	84-984	"	"
28-2-84	El-Sakneh	84-985	"	"
6-3-84	N.Shuneh	84-986	"	"
6-3-84	N.Addasieh	84-987	"	-
6-3-84	Deir-Alla	84-988	"	PVY
6-3-84	El-Sakneh	84-989	C.frutescens	PVY
10-4-84	N.Addasieh	84-1057	C.annuum	"
10-4-84	N.Shuneh	84-1058	"	"
10-4-84	Muth. El-Masri	84-1059	"	"
10-4-84	N.Addasieh	84-1060	"	"
10-4-84	Deir-Alla	84-1061	"	"

Collection Date	Area	Sample No.	Pepper spp.	Virus(s) recovered
10-4-84	N.Addasieh	84-1063	C. annuum	PVY
10-4-84	Muth.El-Masri	84-1064	"	-
10-4-84	"	84-1065	"	PVY
10-4-84	N.Addasieh	84-1066	"	PVY
10-4-84	El-Karameh	84-1067	C. frutescens	"
10-4-84	El-Sakneh	84-1068	"	-
10-4-84	"	84-1069	"	PVY
10-4-84	N.Shuneh	84-1070	C. annuum	PVY
10-4-84	El-Sakneh	84-1071	"	-
10-4-84	Deir-Alla	84-1072	"	PVY
10-4-84	"	84-1073	"	-
10-4-84	Muth-El-Masri	84-1074	"	-
10-4-84	N.Shuneh	84-1075	"	PVY
10-4-84	N.Addasieh	84-1076	"	"
10-4-84	El-Sakneh	84-1077	C. frutescens	"
10-4-84	N.Addasieh	84-1078	C. annuum	-
10-4-84	El-Sakneh	84-1079	C. frutescens	PVY
10-4-84	"	84-1080	"	"
10-4-84	Muth-El-Masri	84-1081	C. annuum	"
17-4-84	El-Sakneh	84-1082	C. frutescens	"
17-4-84	N.Addasieh	84-1083	C. annuum	"

Collection Date	Area	Sample No.	Pepper spp.	Virus (s) recovered
17-4-84	N.Addasieh	84-1084	C. annuum	PVY
17-4-84	El-Sakneh	84-1085	"	"
17-4-84	"	84-1086	"	-
17-4-84	"	84-1087	C. frutescens	PVY
17-4-84	El-Karameh	84-1088	"	-
17-4-84	"	84-1089	"	PVY
17-4-84	El-Sakneh	84-1090	"	"
17-4-84	N.Shuneh	84-1091	C. annuum	"
17-4-84	Deir-Alla	84-1092	"	"
17-4-84	El-Sakneh	84-1093	"	"
17-4-84	Deir-Alla	84-1094	"	"
17-4-84	El-Sakneh	84-1095	C. frutescens	"
17-4-84	"	84-1096	"	"
17-4-84	"	84-1097	"	"
17-4-84	Deir-Alla	84-1098	C. annuum	"
17-4-84	El-Karameh	84-1099	C. frutescens	"
17-4-84	El-Sakneh	84-1100	C. annuum	"
17-4-84	El-Sakneh	84-1101	C. frutescens	"
17-4-84	"	84-1102	"	"
17-4-84	N.Addasieh	84-1103	C. annuum	"
17-4-84	Deir-Alla	84-1104	"	"

Collection Date		Sample No.	Pepper spp.	Virus(s) recovered
	r-Alla	84-1105	C. annuum	PVY
	.Shuneh	84-1106	"	"
17-4-84	"	84-1107	"	"
17-	El-Sakneh	84-1108	"	"
	Deir-Alla	84-1109	"	"
	El-Karameh	84-1110	C. frutescens	"
-84	Deir-Alla	84-1111	C. annuum	"
25-4-84	"	84-1112	"	"
25-4-84	N.Shuneh	84-1113	"	"
25-4-84	Deir-Alla	84-1114	"	"
25-4-84	El-Sakneh	84-1115	C. frutescens	"
25-4-84	"	84-1116	"	"
25-4-84	N. Shuneh	84-1117	C. annuum	"
25-4-84	"	84-1118	"	"
25-4-84	N. Addasieh	84-1119	"	-
25-4-84	N. Addasieh	84-1120	C. annuum	PVY
25-4-84	N. Shuneh	84-1121	"	"
25-4-84	El-Sakneh	84-1122	C. frutescens	"
25-4-84	N. Shuneh	84-1123	C. annuum	"
25-4-84	Deir-Alla	84-1124	C. annuum	"
25-4-84	N. Shuneh	84-1125	"	"

Collection Date	Area	Sample No.	Pepper spp.	Virus(s) Recovered
25-4-84	N. Shuneh	84-1126	C. annuum	PVY
25-4-84	Deir-Alla	84-1127	"	"
25-4-84	"	84-1128	"	"
25-4-84	El-Sakneh	84-1129	C. frutescens	"
25-4-84	El-Karameh	84-1130	"	"
25-4-84	N. Addasieh	84-1131	C. annuum	"
25-4-84	"	84-1132	"	"
25-4-84	N. Shuneh	84-1133	"	"
25-4-84	"	84-1134	"	"
25-4-84	El-Karameh	84-1135	C. frutescens	-
25-4-84	El-Sakneh	84-1136	C. annuum	PVY
25-4-84	"	84-1137	C. frutescens	"
25-4-84	"	84-1138	"	"
25-4-84	El-Sakneh	84-1139	C. annuum	"
25-4-84	"	84-1140	"	"
25-4-84	"	84-1141	"	-
25-4-84	N. Shuneh	84-1142	"	PVY
25-4-84	El-Sakneh	84-1144	C. frutescens	"
25-4-84	N. Addasieh	84-1145	C. annuum	"
25-4-84	El-Karameh	84-1146	C. frutescens	-
25-4-84	Deir-Alla	84-1147	C. annuum	PVY

Collection Date.	Area	Sample No.	Pepper spp.	Virus(s) recovered
25-4-84	Deir-Alla	84-1149	C. annuum	PVY
25-4-84	El-Sakneh	84-1150	C. frutescens	"
25-4-84	N. Shuneh	84-1151	C. annuum	"
25-4-84	Deir-Alla	84-1152	"	"
25-4-84	El-Sakneh	84-1153	C. frutescens	"
25-4-84	"	84-1155	"	"
25-4-84	N. Addasieh	84-1156	C. annuum	"
25-4-84	Deir-Alla	84-1157	"	"
5-5-84	El-Sakneh	84-1203	"	"
5-5-84	N. Addasieh	84-1204	"	"
5-5-84	El-Sakneh	84-1205	C. frutescens	"
5-5-84	"	84-1206	"	"
5-5-84	N.Shuneh	84-1207	C. annuum	"
5-5-84	N. Addasieh	84-1208	"	"
5-5-84	N. Shuneh	84-1209	"	"
5-5-84	Deir-Alla	84-1210	"	"
5-5-84	"	84-1211	"	"
5-5-84	N. Addasieh	84-1212	"	"
5-5-84	Deir-Alla	84-1213	"	"
5-5-84	N. Shuneh	84-1214	"	"
5-5-84	El-Karameh	84-1215	C. frutescens	"

Collection Date.	Area	Sample No.	Pepper spp.	Virus(s) recovered
5-5-84	El-Karameh	84-1216	C. frutescens	PVY
5-5-84	El-Sakneh	84-1217	"	"
5-5-84	"	84-1218	"	"
5-5-84	Deir-Alla	84-1219	C. annuum	"
5-5-84	N. Shuneh	84-1220	"	"
5-5-84	Muth.El-Masri	84-1221	"	"
5-5-84	El-Karameh	84-1222	C. frutescens	"
5-5-84	El-Sakneh	84-1223	C. annuum	"
5-5-84	"	84-1224	"	"
5-5-84	"	84-1225	C. frutescens	"
5-5-84	El-Karameh	84-1226	"	"
5-5-84	N. Addasieh	84-1227	C. annuum	"
5-5-84	Deir-Alla	84-1228	"	"
8-5-84	"	84-1272	"	"
8-5-84	El-Sakneh	84-1273	C. frutescens	"
8-5-84	N. Addasieh	84-1274	C. annuum	"
8-5-84	El-Sakneh	84-1275	"	"
8-5-84	Deir-Alla	84-1276	"	"
8-5-84	N. Addasieh	84-1277	"	-
8-5-84	Deir-Alla	84-1278	"	PVY
8-5-84	El-Sakneh	84-1279	"	-

Collection Date.	Area	Sample No.	Pepper spp.	Virus(s) recovered
8-5-84	El-Sakneh	84-1280	C. frutescens	PVY
8-5-84	"	84-1282	"	"
8-5-84	N. Addasieh	84-1283	C. annuum	"
8-5-84	El-Sakneh	84-1284	"	"
8-5-84	Deir-Alla	84-1285	"	"
8-5-84	N. Shuneh	84-1286	"	"
8-5-84	"	84-1287	"	"
8-5-84	"	84-1288	"	"
8-5-84	El-Sakneh	84-1289	C. frutescens	"
8-5-84	N. Addasieh	84-1290	C. annuum	"
8-5-84	N. Addasieh	84-1291	"	"
8-5-84	Deir-Alla	84-1292	"	"
8-5-84	N. Shuneh	84-1293	"	"
8-5-84	"	84-1295	"	"