http://www.pjbs.org

PJBS

ISSN 1028-8880

Pakistan Journal of Biological Sciences

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Pakistan Journal of Biological Sciences 9 (4): 759-765, 2006
ISSN 1028-8880

Influence of Aphids on the Epidemiology of Potato Virus Diseases (PVY, PVS and PLRV) in the High Altitude Areas of Turkey

[1]Hidayet Bostan, [1]Coskun Guclu, [2]Erdogan Ozturk, [3]Işil Ozdemir and [4]Havva Ilbagi
[1]Department of Plant Protection, [2]Department of Field Crops, Faculty of Agriculture,
Atatürk University, 25240 Erzurum, Turkey
[3]Ministry of Agricultural and Rural Affairs, Plant Protection Central Research Institute,
06172 Ankara-Turkey
[4]Department of Plant Protection, Tekirdag Faculty of Agriculture,
Trakya University, 59030 Tekirdağ-Turkey

Abstract: Potato plants belonging to cv. Morfona were found infected with *Potato leaf roll virus* (PLRV), *Potato virus Y* (PVY) and *Potato virus S* (PVS) with the rates of 3.40, 6.47 and 5.30% in field conditions during the 2003 season. In order to determine the variations in the incidence rates of those potato viruses, harvested tubers from this field used for seed potato for the following seasons for the years of 2004 and 2005. Infection rates for 2004 and 2005 were determined as 7.66 and 22.33% for PLRV; 27.0 and 91.0% for PVY; 21.0 and 77.33% for PVS, respectively. While infection rates of those viruses were relatively low until 2003, the number of infected plants increased in following years. The results obtained from this study revealed that spreading rates of PVY and PVS were higher than PLRV as PVY higher than PVS. Winged aphid counts in the yellow-pan traps during the potato-growing seasons of 2004 and 2005 indicated that *Myzus (Nectarosiphon) persicae* (Sulzer, 1776), *Therioaphis trifolii* (Monell, 1882) and *Aphis fabae* (Scopoli, 1763) were the most abundant aphid species in Central District of Erzurum Province where field trails were established. Nevertheless there was not significant fluctuation of those aphid populations between 2004 and 2005. In addition to those aphids, *Anoecia corni* (Fabricius, *Brachycaudus (Acaudus) cardui* (Linnaeus), *Cryptomyzus ribis* (Linnaeus), *Eulachnus rileyi* (Williams, 1911), *Hyperomyzus lactucae* (Linnaeus, 1758) and *Pterochloroides persicae* (Cholodkovsky) species were collected from the pans but their number were very low.

Key words: Potato viruses, epidemiology, aphid species

INTRODUCTION

Potato (*Solanum tuberosum* L.) is one of the most important crops in Turkey, with an annual production of nearly 5 millions tones from 200.000 ha of arable land. In order to keep this amount of yield, Turkey needs at least 125.000 tones of seed potato per year. There is no responsible state owned company that produced diseases-free seed potato in Turkey. To meet this demand of farmers, seed potatoes have been imported from European countries, multiplied and then have been distributed to the producers by some private companies. In such a way seed potato production is slow and time consuming so virus diseases tend to increase with each multiplication year and resulting in decreased yield and quality of propagation material. Although there are more than 30 viral pathogens that can impact potato worldwide (Salazar, 1996); a survey study dealing with potato virus diseases from seed potato tubers showed that *Potato leafroll virus* (PLRV); *Potato virus Y* (PVY) and *Potato virus S* (PVS) were identified as the most common viruses in potato crops individually or in combination with each other in Turkey (Bostan and Haliloglu, 2004). Among those viruses; PLRV is a non-propagative virus transmitted circulatively. PVY and PVS are transmitted non-persistent manner. They are transmitted by several aphid species of which *Myzus persicae* (Sulzer) is generally considered the most efficient and cosmopolitan vector of PLRV, PVY and PVS (Peters and Jones, 1981; Singh and Boiteau, 1984; Singh *et al*., 1997; Ragsdale *et al*., 2001). Furthermore, the green peach aphid is exceptional in its ability to transmit most of the aphid-transmitted viruses and can carry more than one virus simultaneously (MacKinnon, 1960). Acquisition and transmission of PLRV by aphids is a prolonged process, requiring stylet penetration to the

Corresponding Author: Dr. Hidayet Bostan, Department of Plant Protection, Faculty of Agriculture, Atatürk University, 25240 Erzurum-Turkey Tel: 90 442 231 2010 Fax: +90 442 231 1469

phloem, followed by an incubation period of a day or so and then another phloem penetration to transmit the virus (Bagnall, 1988). Because of PLRV persists throughout the aphid's life, it can be spread over longer distances by winged aphids and occurs in low titers in plant tissues and its vectors (Peters and Jones, 1981; Gildow, 1993). On the other hand, unlike PLRV, stylet-borne PVY and PVS viruses are acquired by aphid vectors in seconds from infected plants, transmitted immediately to healthy plants by short duration probes and the vector usually loses its infectivity within less than one hour (Raccah and Loebenstein, 1982; Sigvald, 1987). Therefore, any aphid visiting infected potato plants is a potential vector of non-persistent viruses so this type of virus transmission can not be controlled by the use of insecticide sprays. None of the insecticides could kill the aphids immediately within a few seconds so they acquire and inoculate the virus until they killed. Furthermore, other cultivated and non-cultivated solanaceous species can act as PLRV, PVS or PVY reservoirs (Sauza-Dias *et al*., 1993). Transmission efficiency of potato viruses transmitted by aphids differ greatly based on aphid species, aphid biotypes, abundance of aphids, transmission manner and characteristics of viruses, inoculum sources and host resistance to virus and vectors (Harrington and Gibson 1989; de Bokx and Piron 1990; Wilson and Jones, 1993; Novy *et al*., 2002). A prerequisite in efforts to reduce extensive yield losses due to viral diseases or at least to maintain the same level in potato production in subsequent growing seasons is the usage of certified virus-free tuber as seed for planting and to minimize the virus inoculum level. On the other hand, potato seed crops should be grown in areas not only isolated from commercial potato fields, but also in areas where the aphid vectors are absent, or occur only in relatively low numbers.

The aims of this study were: 1) to determine increments at the infection rate of PLRV PVY and PVS during three years period (2003, 2004 and 2005); 2) to identify the aphid species visiting to potato field; 3) to determine places having high altitude showing highland properties where fruits and vegetable crops have never been cultivated during potato growing season and weather conditions are suitable for seed potato multiplication and production.

MATERIALS AND METHODS

Screening of plant material for PLRV, PVY and PVS: This study was carried out under Erzurum High Land conditions during years of 2004 and 2005. Erzurum Province is located in the Eastern Anatolian Region of Turkey at the average altitude of 1,850 m from sea level. The climate is characterized with very cold and snowy winters and warm and relatively dry summer seasons. On the other hand, due to short vegetation period and adverse climatic conditions, vegetable and fruit crops are not grown in Erzurum. Therefore, potato, sugar beet, alfalfa, sainfoin, wheat and barley are the main crops.

In order to determine the increments at the infection rates of PLRV, PVY and PVS under field condition, potato cv. Morfona obtained from Agricultural Research Institute of Eastern Anatolia, Erzurum, Turkey was used as plant material during period of this study. For this purpose, first survey study was performed from dormant potato tubers harvested from experimental field plots. After tested, tubers randomly selected from this field were used as seed stock for next year. The second and third tests were performed to potato plant leaf samples collected from the same experimental field in the autumn of 2004 and 2005. For each test, randomly selected 300 tubers or leaf samples were tested individually for each virus by RT-PCR and presence of PLRV, PVY and PVS viruses were expressed as percent for per year.

Collection of winged aphids from the field: Winged aphids regardless of species were collected from experimental potato field plots from the beginning of August to the middle of October during 2-year period. For this purpose, three bright yellow pan traps consist of plastic dishpans (30×15 cm) filled with water (containing 0.002% detergent) were placed in the centers of potato field plots kept in throughout the potato-growing season. Aphids were collected from yellow-pans in every 3-4 days and stored in 70% ethanol in tagged bottles separately according to dates, months and years until to be counted and identified.

RNA extraction from dormant tubers and leaves for RT-PCR: For the extraction of nucleic acids from dormant potato tubers and leaves, the method Singh *et al*. (2002) was used. Briefly, an extraction solution (300 μL of 0.1 M Tris HCl, pH 7.4, 2.5 mM $MgCl_2$, 0.65% Na_2SO_3) and 10 U of RNase-free DNase I (Roshe Molecular Bochemicals) was added to a 1.5 mL micro-centrifuge tube, in which was collected 6 drops (150-180 μL) of tuber or leaf sap, obtained by Tuber Slicer (Electrowerk, Behcke and Co., Hannover, Germany). The mixture was vortexed for 10-15 sec and then incubated at 37°C for 10 min. Nucleic acids were extracted with phenol:chloroform:isoamyl alcohol (24:24:1) using equal volumes of sap and then precipitated with 1 vol. of isopropanol and 1/10 volume of 0.3 M sodium acetate and then incubated overnight at -20°C. The precipitate was collected by centrifugation

Table 1: List of primer pairs used for PLRV, PVY and PVS in RT-PCR

Virus	Sequence	Polarity	Fragment (bp)	Reference
PLRV	5'-CGC GCT AAC AGA GTT CAG CC-3'	Sense	336	Singh *et al.* (1995)
	5'-GCA ATG GGG GTC CAA CTC AT-3'	Antisense		
PVS	5'-TGG CGA ACA CCG AGC AAA TG-3'	Sense	187	Matousek *et al.* (2000)
	5'-ATG ATC GAG TCC AAG GGC ACTG-3'	Antisense		
PVY	5'-AAG CTT CCA TAC TCA CCC GC-3'	Sense	856	Nie and Singh (2002)
	5'-CAT TTG TGC CCA ATT GCC-3'	Antisense		

(12000 g, 15 min, 4°C), washed with 70% ethanol, dried under vacuum and dissolved in 1000 μL (leaves) 100 μL (tubers) of distilled water.

Reverse transcription polymerase chain reaction (RT-PCR): For the reverse transcription, 2.5 μL of nucleic acids were added to a 7.5 μL reaction mixture containing [50 mM Tris-HCl pH 8.3, 75 mM KCl, 10 mM DDT, 2.5 mM $MgCl_2$, 2.5 mM of each dNTPs (Promega), 0.2 μM of antisense primer, 20 U RNasin (Promega, Madison, WI) and 200 U Moleney Murine Leukemia virus-reverse transcriptase (MMLV-RT) (Invitrogen)]. Samples were incubated for 1 h at 42°C for reverse transcription and incubated at 95°C for 3 min to terminate the reaction.

The antisense and sense primer pairs and their fragment size are given in Table 1. For PCR, 2 μL of reverse transcription reaction were transferred to tubes containing 23 μL of the PCR mixture. The final conditions for PCR assay were as fellows: 10 mM Tris-HCl pH 8.3, 50 mM KCl, 1.5 mM $MgCl_2$, 0.2 mM of each dNTPs, 0.2 μM of each of antisense and sense primers and 0.625 U of *Taq* polymerase (Sigma). Samples were amplified in 35 cycles with a denaturation step 94°C, primer annealing 60°C, primer extension 72°C (each step of 1 min duration) and final extension at 72°C (10 min). Ten microliter of amplification products were electrophoresed on a 1.5% agarose gel containing 0.2 μg mL^{-1} ethidium-bromide and photographed under UV illumination. In order to determine of the size of amplified products in the gel, a low mass ladder (Invitrogen) was used.

RESULTS

Determination of the increasing rate of PLRV, PVS and PVY: In 2001-2002, a survey study was carried out and seed potato tubers were obtained from the main potato production Provinces of Bolu, Erzurum, Izmir, Niğde and Nevşehir in Turkey. Test results revealed that average rate of incidence of PLRV, PVY and PVS viruses were determined in those tubers as 13.28, 16.8 and 6.4%, respectively. On the other hand, these rates for PLRV, PVY and PVS in Erzurum were obtained as 5.90, 12.0 and 4.50%. Tubers belonging to Morfona, however, were found infected with PLRV, PVY and PVS at the rate of

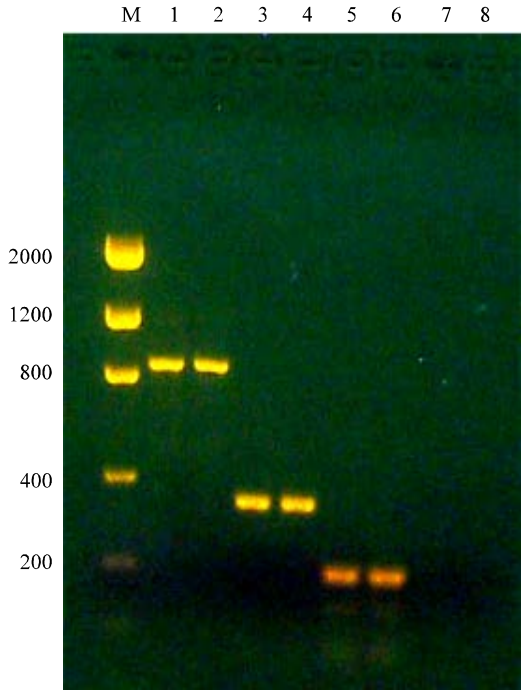

Fig. 1: Agorose gel electrophoresis showing PVY, PLRV and PVS specific PCR products. M, molecular size markers; Lane 1 and 2, PVY (856 bp); Lane 3 and 4, PLRV (336 bp); Lane 5 and 6, PVS (187 bp); Lane 7, negative control (from healthy sample); Lane 8, PCR control

3.40, 6.47 and 5.30% in 2003. Harvested tubers from the same field were used for planting material for the years of 2004 and 2005 in order to determine the variation in the incidence rate of viruses. Infection rates of viruses for 2004 and 2005 were determined as 7.66 and 22.33% for PLRV; 27.0 and 91.0% for PVY; 21.0 and 77.33% for PVS, respectively as exhibited in Table 2.

While infection rates of these viruses have been relatively low until 2003, the number of infected plants increased following years. The results obtained from this study revealed that the spread of PVY and PVS were higher than PLRV; as PVY was higher than PVS. On the other hand, it was shown that primer pairs were capable for the detection of PLRV, PVY and, PVS isolates in Turkey (Fig. 1).

Table 2: Incidence rates of *Potato leaf roll virus* (PLRV), *Potato virus Y* (PVY) and *Potato virus S* (PVS) according to years

		Infection rate of viruses (%)						
Years	No. of samples	PLRV	PVY	PVS	PLRV+PVY	PLRV+PVS	PVY+PVS	PLRV+Y+S
2003	170	4.1	6.4	5.3	0.0	0.0	0.0	0.0
2004	300	15.0	27.0	21.0	1.0	0.0	2.0	0.0
2005	300	36.0	91.0	59.0	22.0	19.0	46.0	16.0

Table 3: The number of winged-aphid species collected by yellow pans during 2004 and 2005

	2004					2005				
Aphid species	August	September	October	Total No.	Rate of species (%)	August	September	October	Total No.	Rate of species (%)
Aphis fabae	20	7	0	27	5.81	37	24	0	61	13.9
Aulocorthum solani	6	0	0	6	1.29	12	0	0	12	2.75
Hyalopterus pruni	14	0	0	14	3.01	0	8	0	8	1.83
Macrosiphum euphorbiae	4	0	0	4	0.86	11	0	0	11	2.52
Myzus persicae	100	55	178	333	71.76	92	79	103	275	63.07
Rhopalosiphum padi	23	0	0	23	4.95	9	0	0	9	2.06
Therioaphis trifolii	48	0	0	48	10.34	53	0	0	53	12.15
Uroleucon jaceae	0	0	9	9	1.93	0	2	5	7	1.60
Total	215	62	187	464		214	113	108	436	

Identification of aphid species collected from field: Collected winged aphid numbers by yellow-pan traps during the potato-growing seasons of 2004 and 2005 revealed that *Myzus (Nectarosiphon) persicae* (Sulzer, 1776), *Therioaphis trifolii* (Monell, 1882) and Aphis fabae (*Scopoli*, 1763) were the most abundant aphid species in Central District of Erzurum Province, as exhibited in Table 3. But, there was no significant fluctuation of these aphid populations between the years of 2004 and 2005. The percentage of *Aphis fabae*, *Aulocorthum solani*, *Hyalopterus pruni*, *Macrosiphum euphorbiae*, *Myzus* (*Nectarosiphon*) *persicae*, *Rhopalosiphum maidis*, *Rhopalosiphum padi*, *Therioaphis trifolii* and *Uroleucon* (*Uromelan*) *jaceae* species were calculated as 5.72, 1.2, 2.96, 0.84, 70.55, 4.87, 1.69, 10.16 and 1.90%, respectively in 2004. In 2005, the percentage of *Aphis fabae*, *Aulocorthum solani*, *Hyalopterus pruni*, *Macrosiphum euphorbiae*, *Myzus* (*Nectarosiphon*) *persicae*, *Rhopalosiphum maidis*, *Rhopalosiphum padi*, *Therioaphis trifolii* and *Uroleucon* (*Uromelan*) *jaceae* were calculated as 13.95, 2.74, 0.42, 2.51, 62.92 , 2.05, 1.60, 12.12 and 1.60%, respectively. In addition to these aphid species, *Anoecia corni* (Fabricius, *Brachycaudus* (*Acaudus*) *cardui* (Linnaeus), *Cryptomyzus ribis* (Linnaeus), *Eulachnus rileyi* (Williams, 1911), *Hyperomyzus lactucae* (Linnaeus, 1758) and *Pterochloroides persicae* (Cholodkovsky) were also collected from pans, but the number of these aphid species were very low.

When we look at the incidence rate of *M. persicae* in potato field, it started to be seen at the beginning of August in the years of 2004 and 2005. The greatest numbers of *M. persicae* was caught in the first half of October while *Aphis fabae* was appeared during all season. *Therioaphis trifolii* was only caught in August of 2004 and 2005 but not the other periods. Although other aphid species were occurred individually or relatively small numbers and their distributions were irregular.

DISCUSSION

The infection rates of PLRV, PVY and PVS were close to each other as indicated in Table 2 and PVY and PVS however were higher than PLRV. The reason might be PLRV having different transmission characteristics than the other viruses and the presence of other non-colonizing aphid species in this area. As a matter of fact, it was reported that any aphid visiting the potato plants is a potential vector of non-persistent viruses (Harrewijn *et al*., 1981) and more than 50 species of aphids have proven capable of transmitting PVY, including many species that can not colonize potato (Boiteau *et al*., 1988). Similarly, Peters and Jones (1981) suggested that short hovering flights performed by the green peach aphid summer migrants might be responsible for a considerable amount of PVY transmission. PLRV is transmitted by aphids colonizing on potato crops and not by all colonizing aphid species (Harrewijn *et al*., 1981). Non-persistent viruses, such as PVY can be transmitted by other aphids, such as *Rhopalosiphum maydis*, *Brachycaudus helichrysi* and *Acyrhosiphon pisum* which do not have potatoes as their host (Hoof, 1980). Furthermore, PLRV has a latent period of 8 to 72 h, but does not replicate in aphids (Eskandari *et al*., 1979) and PLRV persists throughout the aphid's life, it can be spread over longer distances than PVY and PVS, by wind-borne winged aphids (Peters and Jones, 1981).

Within-field spread of PLRV is often by apterae walking from plant to plant (Hanafi *et al*., 1989; Flanders *et al*., 1991; Hodgson, 1991). Apterous *M. persicae* tents to be more efficient vector of PLRV than winged morphs and nymphs are more efficient than adults. But, apterae do not appear to play a significant role in within field spread of PVY (Ragsdale *et al*., 1994). Besides, *M. persicae*, it was reported that PLRV can be transmitted by other potato-colonizing aphids, including *Aphis nasturtii* (buckthorn), *Aulacothum solani* (foxglove), *Macrosiphum euphorbiae* (potato) and *Myzus ascolonicus* (shallot) (MacGillivray, 1981, 1996; Singh *et al*., 1996) and *Capitophorus elaeagni*, *Rhopalosiphum maydis*, *Rhopalosiphum padi* (Halbert *et al*., 2003).

In this study, beside *Myzus (Nectarosiphon) persicae*, *Aulocorthum solani*, *Macrosiphum euphorbiae*, *Rhopalosiphum maidis*, *Rhopalosiphum padi*, other aphid species were not detected in this area. Furthermore, *Aulocorthum solani, Macrosiphum euphorbiae*, *Rhopalosiphum maydis* and *Rhopalosiphum padi* were also present, but the numbers of these aphid species were very low. Although *M. persicae* was the most abundant aphid than all the other aphid species but it can not be considered its population density was high.

The reason of various aphid species appear on potato fields from the second half of August of the year can be explained with the absence of any green plants species left except potato and sugar beet in this area. On the other hand, it is the fact that members of solanacae family crops are not grown in Erzurum except potato and sugar beet in contras to other potato production area in Turkey which could be source of inoculum for these viruses in the neighborhood was taken into account. It can be said that only infected plants in experimental field could be source of inoculum. Typically, virus tolerances for seed lots to be increased following year from the range of 0 to 1%, to the 1 to 5% infected tubers (Woodford *et al*., 1995).

Although there were no places where hard seeded fruit plant can grow except wild *Rosa* spp., how can this *M. persicae* go through winter? In this case, it could be explained by either gone through winter in some *Rosa* spp. or transmitted by wind from long distances, or there might be some unknown hosts for over wintering.

Even if most aphid species are restricted in their host range to a single plant genus or even particular species (Eastop, 1972), about 10% of aphid species, including all that colonize potato show annual alternation of host plants (Eastop, 1986). The primary over wintering host of heteroecious species is often a tree or woody shrub; the summer (secondary) hosts are commonly herbaceous and generally include a much broader range of host species. Only rarely is the primary host closely related to any of the summer hosts. *M. persicae* may be the most polyphagous of all aphid species: it is known to have more than 875 secondary hosts (Leanard *et al*., 1970; Tamaki, 1981).

However, the transmission of diseases agents from a host to another is the most important key in their biology (Mathews, 1992) and the diseases cycle of insect-vectored pathogens represent a level of complexity beyond that of the classic disease trial of susceptible host, viable inoculum and favorable environment (Bagnall, 1988; Ragsdale *et al*., 1994). But, unfortunately, in Turkey, research on aphid biology and ecology has been neglected in the aspect of virus epidemiology and management.

As a result, when we evaluated the population densities and diversity of aphid species collected from traps in the experimental field and take into account climatic condition, this type of agro-ecological regions could be used for seed potato multiplication and production.

REFERENCES

Bagnall, R.H., 1988. Epidemics of potato leafroll in North American and Europe linked to drought and sun spot cycles. Can. J. Plant Pathol., 10:193-202.

Boiteau, G., R.P. Singh, R.H. Parry and Y. Pelletier, 1988. The spread of potato virus Y° in New Brunswick potato fields: Timing and vectors . Am. Potato J., 65: 639-649.

Bostan, H. and K. Haliloglu, 2004. Distribution of PLRV, PVS, PVX and PVY (PVY^{N}, PVY° and PVY^{c}) in the seed potato tubers in Turkey. Pak. J. Biol. Sci., 7: 1140-1143.

De Bokx, J.A. and P.G.M. Piron, 1990. Relative efficiency of a number of aphid species in the transmission of potato virus Y^{N} in the Netherlands. Net. J. Plant Pathol., 96:237-246.

Eastop, V.F., 1972. Deduction from the present day host plants of aphids and related insects. In: Insect-Plant Relationships. Symp. R. Entomol. Soc. Lond., 6: 157-178.

Eastop, V.F., 1986. Aphid Plant Associations. In: Stone, A.R. and D.L. Hawksworht (Eds.). Coevolution and Systematic. Clarendon Pres, Oxford, pp: 35-54.

Eskandari, F., E.S. Sylvester and J. Richardaon, 1979. Evidence for lack of propagation of potato leafroll virus in its aphid vector, *Myzus persicae*. Phytopathology, 69: 45-47.

Flanders, K.L., E.B. Radcliffe and D.W. Ragsdale, 1991. Potato leafroll virus spread in relation to densities of green peach aphid (Homoptera: Aphididae): Implications for management thresholds for Minnesota seed potatoes. J. Econ. Entomol., 84: 1028-1036.

Gildow, F.E., 1993. Evidence for receptor-mediated endocytosis regulating luteovirus acquisition by aphids. Phytopathology, 83: 270-277.

Halbert, S.E., D.L. Corsini and M.A. Wiebe, 2003. Potato virus Y transmission efficiency for some common aphids in Idaho. Am. J. Potato Res., 80:87-91.

Hanafi, A., E.B. Radcliffe and D.W. Ragsdale, 1989. Spread and control of potato leafroll virus in Minnesota. J. Econ. Entomol., 82: 1201-1206.

Harrewijn, P., H.A. van Hoof and J.P.W. Noordink, 1981. Flight behavior of the aphid *Myzus persicae* during its maiden flight. Neth. J. Plant Pathol., 87: 111-117.

Harrington, R. and R.W. Gibson, 1989. Transmission of potato virus Y by aphids trapped in potato crops in southern England. Potato Res., 32: 167-174.

Hodgson, C., 1991. Dispersal of apterous aphids (Homoptera:Aphididae) from their host plant and its significance. Bull. Entomol. Res., 81: 417-427.

Hoof, H.A., 1980. Aphid vector of potato virus Y. Net. J. Plant Pathol., 86: 159-162.

Leanard, M.D., H.G. Walker and L. Enari, 1970. Host plants of *Myzus persicae* at the Los Angeles State and Country Arboretum, Arcadia, California. Proc. Entomol Soc. Wash., 72: 294-312.

MacGillivray, M.E., 1981. In: Compendium of Potato Diseases. The American Psychopathological Society, St. Paul, MN, pp: 101-103.

MacKinnon, J.P., 1960. Combined transmission by single aphids of two viruses that persist in the vector. Virology, 11: 425-433.

Mathews, R.E.F., 1992. Fundamentals of Plant Virology. Academic Pres, NY.

Matousek, J., J. Schubert, P. Didic and J. Ptaek, 2000. A broad variability of potato S carlavirus (PVS) revealed by analysis of RT-PCR-amplified virus sequences. Can. J. Plant Pathol., 22: 29-37.

Nie, X. and R.P. Singh, 2002. A new approach for the simultaneous differentiation of biological and geographical strains of potato virus Y by uniplex and multiplex RT-PCR. J. Virol. Meth., 104: 41-54.

Novy, R.G., Nasrudin, A., D.W. Ragsdale and E.B. Tadcliffe, 2002. Genetic resistance to potato Leafroll virus, potato virus Y and green peach aphid in progeny of *Solanum tuberosum*. Am. J. Potato Res., 79: 9-18.

Peters, D. and R.A.C. Jones, 1981. Potato Leafroll Virus. In: Hooker, W.J. (Ed.), Compendium of Potato Diseases. The American Psychopathological Society, St. Paul, MN, pp: 68-70.

Raccah, R.C.B and G. Loebenstein, 1982. Detection of potato virus Y in the aphid *M. persicae* by enzyme-linked imminosorbent assay (ELISA). Ann. Applied Biol., 101: 511-516.

Ragsdale, D.W., E.B. Radcliffe, C.D. DiFonzo and M.S. Connelly, 1994. Action Thresholds for an Aphid Vector of Potato Leafroll Virus. In: Zahnder, G.W., M.L. Powelson, R.K. Jansson and K.V. Raman (Eds.). Advances in Potato Pest Biology and Management. Am. Phytopathol. Soc., pp: 99-110.

Ragsdale, D.W., E.B. Radcliffe and C.D. DiFonzo, 2001. Epidemiology and Field Control of PVY and PLRV. In: Loebenstein, G., P. Berger, A.A. Brunt and R. Lawson (Eds.). Virus and Virus-like Diseases of Potatoes and Production of Seed-Potatoes. Kluwer Academic Publishers, The Netherlands, pp: 237-270.

Salazar, L.F., 1996. Potato Viruses and Their Control. CIP, Lima, pp: 214

Sauza-Dias, J.A.C., de, A.S. Costa and A.M. Nardin, 1993. Potato leafroll virus in solanaceous weeds in Brazil explains severe outbreaks of the disease in absence of known potato donor sources. Summa Phytopathologica, pp: 19-80.

Sigvald, R., 1987. Aphid migration and the importance of some aphid species as vectors of potato virus Y° (PVY°) in Sweden. Potato Res., 30: 267-283.

Singh, R.P. and G. Boiteau, 1984. Necrotic lesion host for potato virus Y useful in field epidemiological studies. Plant Dis., 68: 779-781.

Singh, R.P., J. Kurz, G. Boiteau and G. Bernard, 1995. Detection of potato leafroll virus in single aphids by reverse transcription polymerase chain reaction and its potential epidemiological application. J. Virol. Methods, 55: 133-143.

Singh, R.P., J. Kurz and G. Boiteau, 1996. Detection of stylet-borne and circulative potato virus in aphids by duplex reverse transcription polymerase chain reaction and its potential epidemiological application. J. Virol. Methods, 59: 189-196.

Singh, R.P., J. Kurz, G. Boiteau and L.M. Moore, 1997. Potato leafroll virus detection by RT-PCR in field-collected aphids. Am. Potato J., 74: 305-313.

Singh, R.P., X. Nie, M.R. Singh, R. Coffin and P. Duplessis, 2002. Sodium Sulphite inhibition in nucleic acid extraction for virus detection by RT-PCR. J. Virol. Methods, 99: 123-131.

Tamaki, G., 1981. Exploiting the ecological interaction of the green peach aphid on peach trees. United States Department of Agriculture, Technical Bulletin No. 1640, pp: 1-6.

Wilson, C.R. and R.A.C. Jones, 1993. Resistance to potato leafroll virus infections and accumulation in potato cultivars and the effects of previous with other viruses on expression of resistance. Aust. J. Agric. Res., 44: 1891-1904.

Woodford, J.A.T., C.A. Jolly and C.S. Aveyard, 1995. Biological factors influencing the transmission of potato leafroll virus by different aphid species. Potato Res., 38: 133-141.