Symptom expression and accumulation of potato virus Y (PVY°) and potato leaf roll virus in thirteen potato cultivars

JARI VALKONEN and EERIK MÄKÄRÄINEN


Necrotic local lesions developed in cvs. Matilda, Ostara, Record, Saturna, Stina, Hankkija’s (Hjan) Tanu and Hjan Timo and local ring spots in Olympia and Sieglinde (Siikli) following sap inoculation with the ordinary strain of potato virus Y (PVY°). Secondly infected cvs. Ostara, Pito, Siikli and Hjan Timo developed leaf drop. No infected progeny was produced by Matilda, Saturna and Hjan Tanu. In contrast, Bintje, Puikula and Sabina developed neither local lesions nor systemic necrosis, but showed mosaic symptoms following primary and secondary infection by PVY°. The ELISA absorbance values for potato leaf roll virus (PLRV) in Ostara, Pito and Saturna were less than 10% of those in the PLRV-infected Siikli. The ELISA values for PLRV in Olympia, Stina, Hjan Tanu and Hjan Timo were not significantly different from those of Siikli. The severity of the symptoms did not correlate with the concentration of PLRV in the potatoes.

Key words: hypersensitivity, virus accumulation, virus resistance, potato

**Introduction**

Potato virus Y (PVY) and potato leaf roll virus (PLRV) are the economically most important viruses of potato (*Solanum tuberosum* L.) in Europe, causing yield losses of up to 80% (De Bokx and van der Want 1987). In Scandinavia, PVY is more important than PLRV (Umaerus et al. 1979, Kurppa 1983). In Finland, the tobacco vein necrosis strain of PVY (PVYN) seems to be dominating, whereas the ordinary strain of PVY (PVY°) is the one most commonly encountered in other parts of Europe (Kurppa 1983, De Bokx and van der Want 1987).

Resistance in potato to viruses is important in the control of viral diseases. The types of resistance to PVY are hypersensitivity and extreme resistance (Ross 1986). Localized hypersensitivity is expressed as necrotic local lesions in the sap-inoculated leaves. Systemic hypersensitivity results in necrosis in the top or the top leaves of the PVY-infected plant. Ross (1986) defined extreme resistance as intensified local hypersensitivity. We define extreme resistance as the type where the virus concentration remains extremely low in an infected plant. The latter definition not only includes the type of resistance in which the multiplication of PVY is reduced (Barker and Harrison 1984), but also the possibility that the low virus concentration in plant tissue is due to the inhibited cell-to-cell spread of the virus (Valkonen et al. 1991).

The three components of resistance to PLRV in potato are restriction of virus multiplication, resistance to infection and inhibition of virus movement.
from foliage to tubers (Barker and Harrison 1985, 1986, Barker 1987). The first component was considered to be the most important by Barker and Solomon (1990), who detected restriction of PLRV accumulation in some cultivated potato genotypes. Furthermore, extreme resistance to PLRV has been found in a few wild potato species (Jones 1979, Brown et al. 1984, Brown 1991, Valkonen et al. 1992).

The local potato production in Finland is mainly based on Finnish, Swedish, German and Dutch cultivars. However, the information about the type and level of resistance to PVY and PLRV in the potato cultivars commonly grown in Finland is limited and data has been obtained mostly from the field and not from experiments performed under controlled conditions (Kurppa 1983, Stegemann and Schnick 1985, Kurppa and Hassi 1989). Determination of the type and level of virus resistance in the above potato cultivars was considered important for the strategic planning of potato breeding programme for virus resistance initiated recently as a joint project of the Department of Plant Production, University of Helsinki, and the Institute of Plant Breeding, Agricultural Research Centre of Finland. The present study was undertaken to test the symptom expression and accumulation of PVYO and PLRV in 13 potato cultivars grown in Finland as PVYO and PLRV are the most important viruses of potato worldwide, PVYO usually causes more severe symptoms in potato than does PVYN (De Boix and van der Want 1987), and PLRV may become more important also in Scandinavia as a consequence of the forecasted warming up of the climate (Carter 1992).

Material and methods

Viruses

One isolate each of PLRV and PVYO was obtained from fieldgrown potatoes in Sudan and England, respectively (El-Amin et al. 1990, Gibson et al. 1990). These virus isolates were used, because the above isolate of PVYO is well-characterized (Gibson et al. 1990, Valkonen et al. 1991, 1992) and resembles the Finnish PVYO isolate YSF11 of Kurppa (1983) in terms of biological properties, and because PLRV occurs only sporadically in Finnish potato fields and no local isolates of the virus were available. PVYO was maintained in Nicotiana tabacum cv. Samsun and PLRV in potato cv. Sieglinde.

Antisera

The antibodies and alkaline phosphatase conjugated antibodies to PVY and PLRV were obtained from Böhringer.

Plants and growing conditions

Virus-tested seed potatoes of cvs. Bintje, Ostara, Record and Saturna (Dutch), Olympia and Sieglinde (Siikli) (German), Matilda, Sabina and Stina (Swedish), and Pito, Puikula, Hankkija’s (Hjian) Tanu and Hjian Timo (Finnish) (Stegemann and Schnick 1985) were obtained from the Finnish Seed Testing Institute, Helsinki. The tubers were planted into pots 21 cm in diameter and filled with a mixture of steam-sterilized peat and washed sand (10:1 v/v) in the beginning of July 1991, and the pots were then sunk by halfway into a sand bed in an aphid-proof nethouse. The plants were watered daily and fertilized weekly with NPK fertiliser. Late blight (Phytophthora infestans (Mont.) de Bary) was controlled by applications of methalaxy, mancozeb and manebe. The average photoperiod was 16h and the mean temperatures at day and night were 26°C and 16°C, respectively. The tubers were harvested at the end of September and dried at room temperature for two days. After storage of four months at 4°C, five randomly taken progeny tubers per plant were planted into pots and grown in glasshouse as described above. The photoperiod was extended to 18 h by illumination with fluorescent lamps. The mean temperatures during day and night were as above.
Virus inoculation

PVY° was sap-inoculated by grinding leaves of infected tobacco at 1g/5ml of distilled water with a pestle and mortar, and rubbing the extract onto carborundum-dusted leaves of the potato plants. Two plants per potato cultivar were inoculated with PVY°. The two oldest leaves of four shoots per plant were inoculated at the emergence of the fifth leaf of the shoot (stage of development no. 305) (JEFFERIES and LAWSON 1991), whereas all other shoots were cut off.

PLRV was graft-inoculated, which is the most sensitive method of testing the resistance to PLRV (SWIEZYNSKI et al. 1989). Two plants per cultivar were inoculated at the emergence of the seventh leaf of the shoot (stage of development no. 307) (JEFFERIES and LAWSON 1991). Four shoots per plant were left growing; all other shoots were cut off. Two of the four shoots were then graft-inoculated with PLRV by removing a leaf from the upper part of the stem with its axillary bud, cutting the stem lengthways, sidegrafting an infected scion of cv. Siikli in place of the removed bud and binding it with Parafilm. The growth of the infected scion was taken as an indication of a successful graft union and the scions were removed three weeks after grafting.

Virus detection

The local and systemic symptoms were visually observed 14 and 28 days after sap inoculation of PVY°. The secondary symptoms were visually observed and the titres of PVY° and PLRV were determined by a double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) (CLARK 1981) in the progeny plants at the appearance of flower buds (stage of development no. 410) (JEFFERIES and LAWSON 1991). For DAS-ELISA, the uppermost fully expanded leaves of the plants
Table 1. Symptoms due to primary infection (sap-inoculated PVY\textsuperscript{O}), and symptoms and ELISA absorbance values (A\textsubscript{405}) ± standard deviation due to secondary infection by PVY\textsuperscript{O}, as determined at the time of flower initiation.

<table>
<thead>
<tr>
<th>Primary infection symptoms</th>
<th>Secondary infection</th>
<th>no. of tubers infected</th>
<th>symptoms</th>
<th>A\textsubscript{405} values</th>
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<tbody>
<tr>
<td></td>
<td>local</td>
<td>systemic</td>
<td></td>
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</tr>
<tr>
<td>Bintje</td>
<td>O</td>
<td>M</td>
<td>5/5</td>
<td>M, S, R</td>
</tr>
<tr>
<td>Matilda</td>
<td>NRS, VN</td>
<td>VN, IVN</td>
<td>0/5</td>
<td>O</td>
</tr>
<tr>
<td>Olympia</td>
<td>RS</td>
<td>VN, CS</td>
<td>4/5</td>
<td>M</td>
</tr>
<tr>
<td>Ostara</td>
<td>NS</td>
<td>O</td>
<td>2/5</td>
<td>M, LD</td>
</tr>
<tr>
<td>Pito</td>
<td>O</td>
<td>VN, CS</td>
<td>5/5</td>
<td>M, LD</td>
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<tr>
<td>Puikula</td>
<td>O</td>
<td>M</td>
<td>4/5</td>
<td>M, R</td>
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<tr>
<td>Record</td>
<td>NS</td>
<td>O</td>
<td>3/5</td>
<td>M</td>
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<tr>
<td>Sabina</td>
<td>O</td>
<td>M</td>
<td>5/5</td>
<td>M</td>
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<tr>
<td>Saturna</td>
<td>NRS, VN</td>
<td>VN</td>
<td>0/5</td>
<td>O</td>
</tr>
<tr>
<td>Siikli</td>
<td>RS</td>
<td>NRS, VN</td>
<td>2/5</td>
<td>M, LD</td>
</tr>
<tr>
<td>Stina</td>
<td>NRS, VN</td>
<td>VN, IVN</td>
<td>1/5</td>
<td>M, LD</td>
</tr>
<tr>
<td>Hjan Tanu</td>
<td>NRS</td>
<td>O</td>
<td>0/5</td>
<td>O</td>
</tr>
<tr>
<td>Hjan Timo</td>
<td>NRS</td>
<td>O</td>
<td>4/5</td>
<td>M, LD</td>
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<td>non-inoculated cultivars</td>
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O = no symptoms  
CS = chlorotic spots  
IVN = interveinal necrosis  
LD = leaf-drop  
M = mosaic  
NRS = necrotic ringspots  
NS = necrotic spots  
R = rugosity  
S = stunting  
VN = vein necrosis  
RS = ringspots

were sampled in duplicate, extracted at 1g/3ml of buffer and pipetted onto ELISA plates (Greiner Labortechnik) in aliquots of 200 µl. Absorbances were recorded at 405 nm (A\textsubscript{405}) after developing the colour reaction for 1 h using p-nitrophenyl as substrate.

**Results**

**PVY\textsuperscript{O}**

Necrotic local lesions were observed in cvs. Matilda, Ostara, Record, Saturna, Stina, Hjan Tanu and Hjan Timo following sap inoculation with PVY\textsuperscript{O} (Table 1), whereas Olympia and Siikli developed ring spots that remained green while the leaves turned yellow (Fig. 1). Bintje, Pito, Puikula and Sabina showed no symptoms in the sap-inoculated leaves.

Systemic vein necrosis following primary infection was observed in Matilda, Olympia, Pito, Sa-
turna, Siikli and Stina (Table 1). Systemic mosaic symptoms without necrosis were observed in Bintje, Puikula and Sabina, while Ostara, Record, Hjan Tanu and Hjan Timo were free of any.

Systemic mosaic symptoms were observed in the progeny plants of ten of the 13 cultivars following secondary infection by PVY\textsuperscript{O} (Table 1). Furthermore, Ostara, Pito, Siikli and Hjan Timo developed leaf drop, Bintje was heavily stunted and the growth of Puikula was poor. In contrast, the progeny of Matilda, Saturna, Stina and Hjan Tanu were free of symptoms and PVY\textsuperscript{O} according to ELISA.

**PLRV**

All the tested progeny tubers of each cultivar graft-inoculated with PLRV were infected with PLRV according to ELISA. However, the cultivars fell into three groups according to the PLRV titres (P=0.01) (Fig. 2). Olympia, Siikli, Stina, Hjan Tanu and Hjan Timo yielded high PLRV titres ($A_{405}$ > 1.33), Matilda, Puikula and Record yielded moderate PLRV titres (0.90 < $A_{405}$ < 0.96), and Ostara, Pito and Saturna yielded low PLRV titres ($A_{405}$ < 0.66). The PLRV titres of Bintje ranged between low and moderate and those of Sabina between moderate and high (LSD\textsubscript{0.01}= 0.34 for the $A_{405}$ values).

PLRV caused yellowing of the leaves and leaf roll in Matilda, Ostara, Siikli and Hjan Timo, yellowing of the leaves without leaf roll in Olympia, Puikula, Saturna and Hjan Tanu, and yellowing of the leaf margins in Pito. Symptomless infection by PLRV was detected in Bintje, Record, Sabina and Stina.

**Discussion**

Hypersensitivity reactions were observed in 10 of the 13 cultivars following infection with PVY\textsuperscript{O}. Expression of hypersensitivity is useful, as such a response induced by a virus or virus strain in a plant
can reduce the systemic spread of the virus (FRITIG et al. 1987). Indeed, no infected progeny was detected in Matilda, Saturna and Hján Tanu and few infected progeny tubers were produced by Stina. These cultivars reacted by local necrosis to primary infection of PVY\textsuperscript{O}. Similar results were obtained in Matilda by KURPPA and HASSI (1989).

There are few studies on the genetic control of hypersensitivity to PVY in potato. COCKERHAM (1970) identified four genes which control the hypersensitivity to all strains of PVY in a number of Solanum spp., of which the genes in S. chacoense Bitt. and S. demissum Lindl. acted also against potato virus A (PVA). JONES (1990) suggested that the strain group specific hypersensitivity to PVY\textsuperscript{O} in certain potato genotypes was controlled by a single dominant gene, \textit{N}_{\text{Ybr}}, which possibly originated in Katahdin and a Scottish potato clone 11-79 (DAVIDSON 1980), and by another dominant gene of unknown origin. Our results do not allow further comparisons of the genes for hypersensitivity to PVY between the cultivars of our study and those reported by COCKERHAM (1970) and JONES (1990), as those comparisons would require inoculations with other strains of PVY and PVA. However, the hypersensitivity to PVY\textsuperscript{O} in Hjan Timo, a cultivar derived from the cross Frühnudel \texttimes Katahdin (VARIS 1975), is presumably controlled by the gene \textit{N}_{\text{Ybr}} from Katahdin.

The progenies of Bintje, Puikula and Sabina were almost 100\% infected with PVY\textsuperscript{O}. The plants generated high PVY titres and showed severe mosaic symptoms without necrosis. The susceptibility of these cultivars to PVY hampers their cultivation in Southern Scandinavia. Therefore, Puikula and Sabina are mainly grown in the northernmost areas, where the spread of PVY is reduced due to the low populations of the PVY transmitting species of aphids (UMAERUS et al. 1979, SIGVALD 1984, KURPPA and RAJALA 1986).

The incidence of PLRV-infected progeny tubers was similarly high in all of the cultivars. However, there were variations in the PLRV concentrations between the progenies of cultivars. The concentration of PLRV in cultivars such as Ostara, Pito and Saturna was less than 10\% of that in Siikli. Restricted accumulation of PLRV is useful in potato as it potentially reduces the spread of PLRV by aphids (BARKER and HARRISON 1986).

The severity of the symptoms and the concentrations of PLRV did not strictly correlate in the potato genotypes tested. For example, apparent leaf roll and yellowing symptoms were observed in Ostara which exhibited the lowest PLRV concentration of all the cultivars. In contrast, no symptoms were observed in Stina which generated high concentrations of PLRV. The severity of symptoms observed in the PLRV-infected progeny of potato correlates rather with the physiological stage of the mother plants at the time of inoculation than with the PLRV concentrations in the progeny plants (BARKER and HARRISON 1986, BARKER and WOODFORD 1987).

The valuable types of virus resistance, i.e. hypersensitivity to PVY\textsuperscript{O} and restricted accumulation of PLRV, were incorporated in e.g. Ostara and Saturna. However, no cultivar exhibited extreme resistance to PVY and PLRV. Genes for extreme resistance are the obvious choice to be incorporated in new cultivars (JONES 1990), because extreme resistance acts against most, if not all strains of a virus and efficiently reduces the virus transmission by aphids (JONES 1979, 1990, GIBSON et al. 1990). Furthermore, virus strains cabable of overcoming extreme resistance are not common in the field (JONES 1985, ROSS 1986). Extreme resistance to PVY, PLRV and PVA is incorporated in the wild potato species of the Etuberosa group, e.g. S. brevivids Phil. (VALKONEN et al. 1992), and it has recently been transferred to some cultivated potato genotypes (PEHU et al. 1990, WILLIAMS et al. 1990, XU et al., unpublished results). The results suggest higher acceptability and productivity for most cultivars of the present study in Southern Scandinavia and other parts of Europe, where PLRV and PVY\textsuperscript{O} are the most disastrous viruses in potato, if genes for extreme resistance to PLRV and PVY could be incorporated into them.

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References


PLRV:saastuttamista mukuloista kasvaneiden Ostaran, Pidon, Siiklin ja Hjan Timon taimen alalehdet kuolivat ja jättivät roikkumaan varresta. PVYO:lla saastuttettujen Matildan, Saturnan ja Hjan Tunan mukuloista yksikään ei tuotannut virussaastunutta taimia, kun taas muiden PVY:lla saastuttettujen lajikkeiden mukuloista kasvaneissa taimissa ilmeni viherkirjoja, ja taimissa havaittiin korkeita PVY-pitoisuksia.