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POTATO VIRUS M TRANSPORT IN INFECTED PLANTS

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A study of virus transport in plants yields a lot of data enriching our knowledge on the viruses themselves as well as on their interaction with diseased plants [6, 7]. Besides it yields numerous data which allow to understand and to solve some practical problems in agriculture. It is known, for example, that not all potato tubers from a diseased plant have to be infected, that the number of infected tubers decreases when the time of infection is delayed and that the virus is not transported to the tubers when plant infection is sufficiently late [1]. The aim of this study was to obtain the data on the direction and rate of potato virus M translocation in plants and on tuber infection on plants which had been inoculated in different periods.

MATERIALS AND METHODS

One of the severe potato virus M (PVM) isolates (M 24) described by Kowalska [5] was used throughout this study. Potato plants cv. Uran infected with this virus isolate obtained from the Department of Genetics of the Potato Research Institute at Młochów served as a virus source.

Tomato cv. Najwcześniejszy plants were used in the experiments on the direction and rate of PVM translocation. These plants were inoculated in a 4 leaf stage by rubbing inoculum onto 3rd and 4th leaves from the base of plant. They were grown in a greenhouse in which the temperature was maintained at 18-28^{o}C level and 18 hours day was secured by mercury lamps giving the light of minimum 6 000 lx. The presence of PVM in leaves of these plants was tested by se-

rological microprecipitin test with commercial antiserum produced by Serological Laboratory of the Potato Research Institute at Gdańsk.

The aim of the first experiment was to check how fast PVM escapes from inoculated leaves. Twenty five tomato plants were inoculated and divided in 5 groups - 5 plants in each group. Inoculated leaves were detached off the plants of succesive groups in 4, 8, 12, 16 or 20 days following inoculation. All plants were individually tested for PVM 6 weeks after the leaves had been detached off the last group of plants. The sap for testing was obtained from the top, fully expanded leaves. Twenty five of uninoculated tomato plants served as controls.

The aim a second experiment was to obtain the data on the direction and rate of PVM translocation in tomato plants. Twenty five plants were inoculated and divided in 5 groups - 5 plants in each group. In a week sfter inoculation the first leaf samples were tested for PVM. Each sample contained 5 leaves (1 leaf from each of 5 plants of first group) from the same node. The succesive groups of plants were tested by the same manner in 2, 3, 4 and 5 weeks following inoculation.

The experiments on potato tuber infection with PVM were conducted on virus-free cv. Irys plants obtained from the Department of Genetics and Potato Breeding of the Potato Research Institute. At the end of May 75 tubers were planted in the experimental field in 3 rows 70 cm apart. Thirty three days later 25 plants in a border row were sap-inoculated with PVM by rubbing the inoculum onto 10 leaflets in a upper part of each plant. The plants in a second border row were inoculated 54 days after tuber planting. The plants in the middle row were left as controls. The tubers were harvested in 90 days after planting. Three tubers from each plant were selected for PVM testing - one from the middle part of the stem, second from the upper part, and the third from the lower part of the same stem. Unfortunately, the stolons were destroyed in many plants at the harvesting time and the localization of tubers was possible only for 10 plants inoculated in 33 days after planting, 5 plants inoculated in 54 days after planting and 10 control plants. The tubers from these plants were tested for PVM after 3 months of storage. The equivalent weight of flesh from each tuber was ground in a mortar and used for bioassay of bean cv, Red Kidney plants [4].

After taking these samples for bioassay the tubers were planted in a greenhouse and the plants were bioassayed for PVM on Red Kidney plants. A 3 g leaf sample from each plant was taken for this test from the upper part of the plant.

All inoculations were made by rubbing the sap obtained by grindind the leaves or tuber parts in a mortar in the presence of and equal amount (w/v) of 0.067 M phosphate buffer pH = 7.5. Inoculated leaves had been previously dusted with carborundum 600 mesh and were rinsed with tap water after inoculation.

RESULTS

Antiserum against PVM did not react with the sap from control tomato cv. Najwcześniejszy plants nor the control antiserum from rabbit reacted with the sap from PVM inoculated plants. On the other hand, antiserum against PVM reacted with the sap from all PVM inoculated plants, even from those which the leaves were detached off at the 4th day following inoculation (Table 1). It means that all inoculated plants became infected with PVM and that 4 days period was sufficient for PVM escaping from inoculated leaves. The intensity of serological reactions indicated that the amount of PVM was the highest in the plants which the inoculated leaves were detached off in 8th and 12th day following inoculation. The least amount of a virus was present probably in the plants which the leaves were detached off in 4th day following inoculation (Table 1).

PVM was not detected in leaves of any of 6 or 7 nodes of tomato cv. Najwcześniejszy plants tested in 1 or 2 weeks after inoculation. The virus was not detected even in inoculated leaves from 3rd and 4th nodes (Table 2). The tests made in 3 weeks after inoculation shown that PVM was present in inoculated leaves and in leaves from 2 succesive nodes above the inoculated leaves. In 4 and 5 weeks following inoculation PVM was detected in leaves from all nodes except two youngest, top leaves (Table 2).

PVM was not detected in any of tubers from control potato cv. Irys plants nor in any of plants inoculated 54 days after planting. These results were confirmed by testing the plants grown from these tubers.

Table 1

PVM detection in tomato cv. Najwcześniejszy plants which inoculated leaves were detached off in different time following inoculation

No. of days after inoculation in which inoculated leaves were detached off	No. of plant	Result of serological testing		
		PVM-a + CP	C-a + IP	PVM-a + IP
5	1	-	-	+
	2	-	-	+
	3	-	-	+
	4	-	-	+
	5	-	-	+
8	6	-	-	++
	7	-	-	++
	8	-	-	++
	9	-	-	++
	10	-	-	++
12	11	-	-	++
	12	-	-	++
	13	-	-	++
	14	-	-	++
	15	-	-	++
16	16	-	-	+
	17	-	-	+
	18	-	-	+
	19	-	-	+
	20	-	-	+
20	21	-	-	+
	22	-	-	+
	23	-	-	+
	24	-	-	+
	25	-	-	+

PVM-a - antiserum against PVM, C-a - control antiserum, CP - control plants, IP - inoculated plants.

- No precipitate.

+ Visible precipitate,

++ Heavy precipitate.

Table 2

Serological PVM detection in leaves
from different nodes of tomato cv. Najwcześniejszy plants
in succesive weeks following inoculation

Node from the base of plant	PVM detection in successive weeks				
	1	2	3	4	5
1	-	-	-	+	+
2	-	-	-	+	+
3*	-	-	+	+	+
4*	-	-	+	+	+
5	-	-	+	+	+
6	-	-	+	+	+
7	nt	-	-	+	+
8	nt	nt	-	+	+
9	nt	nt	-	+	+
10	nt	nt	nt	-	+
11	nt	nt	nt	-	-
12	nt	nt	nt	nt	-

- PVM not detected, + PVM detected, nt not tested (no such node on a plant), * inoculated leaves.

On the other hand, PVM was detected in almost all, except three, tubers from the plants inoculated 33 days after planting (Table 3). The number of lesions on Red Kidney leaves indicated that PVM was present in a highest concentration in tubers from middle part of the stem and in the lowest concentration in tubers from the upper part of the stem (Table 3). These results were fully confirmed by testing the plants grown from these tubers (Table 4).

Table 3

The number of local lesions on Red Kidney bean leaves inoculated with sap from potato cv. Irys tubers obtained from plants inoculated with PVM 33 days after planting (average from 3 Red Kidney leaves)

Localization of the tuber on plant stem	Lesion number for plant no.:										Average lesion number
	1	2	3	4	5	6	7	8	9	10	
Upper	1	44	2	12	19	10	0	40	15	0	14
Middle	24	81	12	41	120	39	31	52	41	4	44
Bottom	14	54	4	23	78	13	12	53	13	0	26

Table 4

The number of local lesions on Red Kidney bean leaves inoculated with sap from potato cv. Irys plants grown from tubers harvested from plants inoculated with PVM 33 days after planting (average from 3 Red Kidney leaves)

Localization of the tuber on plant stem	Lesion number for plant no.:										Average lesion number
	1	2	3	4	5	6	7	8	9	10	
Upper	1	120	7	30	48	27	0	88	36	0	36
Middle	66	227	30	101	309	104	74	115	98	10	113
Bottom	36	153	10	57	199	34	29	118	32	0	67

DISCUSSION

The data obtained in an experiment in which inoculated leaves were detached off the plants in different times following inoculation indicate that PVM could escape from inoculated leaves first 4 days following inoculation. This result is in agreement with the results obtained for other plant viruses [6, 7]. It seems that a small amount of a virus is translocated to the plant in such a short time because PVM concentration in plants on which inoculated leaves were left for 8 or 12 days was probably higher which was indicated by the heavier precipitation with antiserum.

On the other hand, PVM concentration in plants on which inoculated leaves were left for 16 or 20 days seemed again lower at the moment of testing. Dziewońska et al. [3] have proved, that when the temperature is high enough (above 20°C) the concentration of PVM in plants is quickly increasing at the beginning of infection process, it reaches some maximum, and then starts to decrease again. Then it is conceivable that in our experiments which were conducted in high temperature conditions PVM concentration reached its peak quite shead of testing time in plants which inoculated leaves were left on for 16 or 20 days. Such interpretation is possible when we accept that inoculated leaves are significant virus source for the whole plants in at least 20 days following inoculation.

The data which were obtained in a second experiments are in favor of such interpretation. PVM was not detected in inoculated and in two leaves just above the inoculated leaves till the 3rd week after inoculation and in the rest of leaves, except two youngest, it was detected at 4th week after inoculation.

The tubers of potato cv. Irys plants which had been inoculated 54 days after planting were not infected with PVM. Similar results were obtained by Beemster [2] for potato virus Y and they are in accordance with a common opinion that potato tubers may escape infection if plant infection is sufficiently late.

On the other hand the plants which had been inoculated 33 days after planting yielded the tubers which were almost wholly infected. The highest PVM concentration was found in tubers from the middle part of the stem. These tubers are first to initiate and they are initiated when the plants are still young and the virus replication

and translocation are still active [3] so they may obtain a heavy virus load. The tubers which were initiated later on the higher and lower parts of the stem could obtain the lower virus load since in the time of their initiation the plants were already older and the virus replication and transport were gradually inhibited [1]. Then it is possible that the highest virus concentration in the biggest tubers which was found by Beemster [2] was connected not with a tuber size but rather with their initiation time.

Finally, the correlation between the results of bioassaying of tubers and of plants which were grown from these tubers (Tables 3 and 4) indicates that, at least for young plants, PVM concentration in plants was influenced by the concentration of this virus in tubers which these plants were grown from.

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TRANSPORT WIRUSA M ZIEMNIAKA W PORAŻONYCH ROŚLINACH

S t r e s z c z e n i e

Testem serologicznym wykryto wirus M ziemniaka (PVM) nawet w roślinach pomidora odmiany Najwcześniejszy, z których inokulowane liście usunięto już w 4 dni po inokulacji, choć ilość wirusa w roślinach, z których inokulowane liście usuwano nieco później była większa. Testując serologicznie liście pomidora odmiany Najwcześniejszy z różnych pięter roślin w kolejnych tygodniach po inokulacji PVM wykryto wirus - pierwszy raz po 3 tygodniach od inokulacji w liściach inokulowanych i położonych bezpośrednio nad nimi. W późniejszych terminach testowania wykrywano PVM we wszystkich liściach z wyjątkiem liści najmłodszych. Nie wykryto PVM w bulwach odmiany Irys zebranych z roślin inokulowanych wirusem po 54 dniach od posadzenia, ani w roślinach wyrosłych z tych bulw. Liczba plamek lokalnych na liściach pierwotnych fasoli Red Kidney świadczy, że najwięcej PVM zawierały bulwy ze środkowej części pędów roślin odmiany Irys, inokulowanych wirusem po 33 dniach od posadzenia. Ilość wirusa w bulwach zawiązujących się później była mniejsza. Wyniki te potwierdziło badanie roślin wyrosłych z tych bulw.

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ТРАНСПОРТ ВИРУСА М КАРТОФЕЛЯ В ПОРАЖЁННЫХ РАСТЕНИЯХ

Р е з ю м е

Путём серологической пробы обнаружен был вирус М картофеля (PVM) даже у растений помидора Раннеспелый, из которых инокулированные листья удалялись уже через 4 дня после инокуляции, хотя число вируса в растениях, инокулированные листья у которых удалялись немного позже, было выше. Исследуя серологическим путём листья помидора Раннеспелый из разных ярусов растения, на очередной неделе после инокулировки PVM, обнаружен был вирус уже через 3 недели после инокули-

ровки в инокулированных листьях и в листьях расположенных непосредственно над ними. В более поздние сроки исследований PVM обнаружен был во всех листьях за исключением наиболее молодых листьев.

Не обнаружен был PVM ни в клубнях картофеля сорта Irys, выращенного из растений инокулированных вирусом в 54 дня после посадки, ни в растениях выросших из этих клубней. Число энденемических пятнышек полученных на первичных листьях фасоли Red Kidney свидетельствует, что больше всего PVM содержали клубни средней части побегов растений картофеля сорта Irys, инокулированных вирусом в 33 дня после посадки. Число вируса в клубнях завязывающихся позже, было меньше. Результаты эти были подтверждены исследованиями растений, выросших из этих клубней.