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EFFECTS OF REMOVAL OF LEAVES AND TOPS ON TRANSLOCATION OF POTATO VIRUS X AND POTATO VIRUS Y^N IN POTATO PLANTS[1]

Het effect van ontbladering en toppen op het transport van aardappel-X-virus en aardappel-Y^N-virus in aardappelplanten

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Removal of leaves from primarily infected plants does not stop translocation of potato virus X and potato virus Y^N from the stem to the tubers in potato plants. In some cases there is evidence that even more virus reaches the tubers. The removal of the top of a potato plant results clearly in a larger extent of infection of the tubers, as was demonstrated in experiments with both viruses. This effect proved to be greater according as the removed top was larger. Removal of leaves and tops apparently changes the physiological behaviour of potato plants in such a way that virus translocation is promoted. This means that in haulm killing, as is applied in seed potato growing, only perfect killing of the stems and leaves can result in stopping virus translocation to tubers; incomplete killing may have the opposite effect.

INTRODUCTION

In seed potato growing areas in the Netherlands it is common practice to destroy potato haulm mechanically or chemically towards the date seed potatoes have to be harvested. This enables the farmers to obtain the optimum profit of the growing period of the seed potatoes, since harvesting on or before a fixed date is necessary in order to qualify for a certain class of certification. As it is not possible to harvest all potatoes at that special date, applying some method of haulm killing is very useful. One of the problems connected with haulm killing is the fact that it is sometimes rather difficult to destroy the haulm completely. This depends, among other things, on the potato variety and on the stage of growth of the potato plant. Incomplete killing of the haulm may lead to renewed outgrowth of shoots which, as is known (BEEMSTER, 1961a), can lead to a rapid infection of the tubers by viruses. Whether a virus can be easily translocated downwards to the tubers from a defoliated stem was not known. As this, also, might play a role in the practice of haulm killing, some experiments on the subject were performed under glasshouse conditions. In these experiments translocation of potato virus X (PVX) and potato virus Y^N (PVY^N) was studied in defoliated and in topped plants, which were inoculated before defoliation or topping. Although in these experiments treatment of the potato plants was not quite the same as the one given normally in practice, it was thought that the experiments could at least give some idea of what might happen in the field.

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MATERIAL AND METHODS

The experiments, using the potato variety 'Bintje', were performed in a greenhouse at a temperature of 22–24 °C. The plants were grown in pots and were single-stemmed. The inoculations with PVX and PVY^N were carried out by sap using Carborundum as an abrasive. The inocula were obtained from stock cultures of the viruses from potato plants. The tubers of the plants were planted some time after harvesting without breaking dormancy. The plants growing from these tubers were tested for the presence of the viruses by using local lesion hosts. For PVX *Gomphrena globosa* leaves were used; for PVY^N A6 test leaves were used according to the method described by DE BOKX (1964). In some cases PVY^N could already be diagnosed visually and in these cases further testing was not performed. The tests showed that healthy-looking plants were often infected. To obtain a complete picture of the extent of tuber infection, each harvested tuber was cut into as many eye-cuttings as possible. In the tables the extent of infection is expressed in the numbers and percentages of infected eye-cuttings.

EXPERIMENTS AND RESULTS

a. *Experiment with PVY^N in 1961*

On 25 July 1961 75 'Bintje' tubers were planted and on 22 September (59 days after planting) one leaflet of the youngest full-grown leaf was inoculated with PVY^N. The plants were divided into 15 groups of 5 plants (A to O). The plants of the groups A, B, C, D and E were harvested 5, 7, 10, 12 and 14 days respectively after inoculation without previous defoliation. The plants of the groups F, G, H and I were completely defoliated (including the inoculated leaf) five days after inoculation and harvested 7, 10, 12 and 14 days after inoculation respectively. Plants of the groups J, K and L were defoliated seven days after inoculation and harvested 10, 12 and 14 days after inoculation respectively. Plants of the groups M and N were defoliated 10 days after inoculation and harvested 12 and 14 days after inoculation respectively. The plants of group O were defoliated 12 days after inoculation and harvested 14 days after inoculation.

In Table 1 the extent of infection of the eye-cuttings obtained after testing them is given. The amount of infection in general was rather low, having regard

TABLE 1. Infection with potato virus Y^N of eye-cuttings of potato tubers from plants defoliated at various dates after inoculation.
De besmetting met aardappel-YN-virus van oogstekken van aardappelknollen, afkomstig van de na inoculatie ontbladerde planten, op verschillende tijdstippen na de inoculatie.

Number of days between inoculation and harvest	Not defoliated	Number of days between inoculation and defoliation			
		5	7	10	12
5	A: 0/140[1] = 0%				
7	B: 2/96 = 2%	F: 0/119 = 0%			
10	C: 0/134 = 0%	G: 0/104 = 0%	J: 2/116 = 2%		
12	D: 9/93 = 10%	H: 0/110 = 0%	K: 3/129 = 2%	M: 3/125 = 2%	
14	E: 7/148 = 5%	I: 0/117 = 0%	L: 4/121 = 3%	N: 8/134 = 6%	O: 31/139 = 22%

[1] Numerator: number of infected eye-cuttings
Denominator: number of eye-cuttings tested

to the fact that the rate of PVY^N-translocation is normally rather high (BEEMSTER, 1961b, 1966). It is clear that at the fifth day after inoculation no virus had been translocated out of the inoculated leaves. Therefore, defoliation five days after inoculation did not lead to any infection of the tubers of the groups F to I. From the seventh day onward virus seems to have moved into the stem and sometimes into the tubers. The percentages of infection are more or less of the same order in defoliated and non-defoliated plants. Only in the plants defoliated 12 days after inoculation was a rather high percentage of infection found. The results given in Table 1 lead to the conclusion that removal of the leaves caused no significant differences in infection compared with non-defoliated plants.

b. *Experiment with PVX in 1963*

In 1963 the same type of experiment was performed with PVX. As the rate of translocation of PVX is generally somewhat lower than that of PVY^N the potato plants were inoculated in a younger stage of growth and harvested at longer intervals than was done in the experiment with PVY^N in 1961.

The 'Bintje' tubers were planted 11 July 1963. Three leaflets of the youngest full-grown leaf were inoculated on 22 August (42 days after planting). The single-stemmed plants were divided into 27 groups of five plants. Instead of giving the dates of defoliation and of harvesting in detail here, we refer to Table 2 in which the treatment of each of the groups is given, as in Table 1 for the previous experiment. The leaves, including the inoculated leaf, were removed 6, 9, 12, 15 and 18 days after inoculation. New outgrowths, which appeared later, were also removed as soon as possible. Table 2 gives the extent of infection of the eye-cuttings found after testing.

TABLE 2. Infection with potato virus X of eye-cuttings of potato tubers from plants defoliated at various dates after inoculation.

De besmetting met aardappel-X-virus van oogstekken van aardappelknollen, afkomstig van na de inoculatie ontbladerde planten, op verschillende tijdstippen na de inoculatie.

Number of days between inoculation and harvest	Not defoliated	Number of days between inoculation and defoliation				
		6	9	12	15	18
6	1/104[1] = 1%					
9	1/99 = 1%	6/106 = 6%				
12	11/122 = 9%	1/102 = 1%	5/103 = 5%			
15	3/133 = 2%	15/90 = 17%	9/108 = 8%	16/124 = 13%		
18	22/122 = 18%	26/96 = 27%	37/109 = 34%	17/97 = 14%	41/104 = 39%	
21	76/126 = 60%	45/99 = 46%	79/113 = 70%	63/126 = 50%	59/115 = 51%	37/118 = 31%
After ripening of the plants	88/232 = 38%	71/74 = 96%	90/100 = 90%	48/117 = 41%	82/119 = 69%	88/130 = 68%

[1] See legend to Table 1

The results show clearly that the extent of infection with PVX was much higher than in the experiment with PVY^N. The results for the group of plants which were not defoliated show that a very low percentage of infection was found six days after inoculation, the percentage increasing until 21 days after inoculation. In the group defoliated six days after inoculation, which at the time

of defoliation had only 1% infection of eye-cuttings, it appeared that after defoliation the translocation of virus towards the tubers continued, but with a tendency towards a higher infection of the tubers than in those from non-defoliated plants. The small amount of virus present in the stem at the time of removing the leaves probably multiplied somewhere in the stem at at least at the same rate as in the inoculated leaf. The stems of the defoliated plants looked much greener than the stems of the untreated plants. This suggests that in the defoliated stem photosynthetic processes became more active than they normally are in stems. It seems likely that this may have activated the processes of virus multiplication and of translocation which led finally to the infection expressed in Table 2. The groups of plants defoliated 9, 12, 15 and 18 days after inoculation gave more or less the same results as the group defoliated six days after inoculation.

In this experiment groups of plants were also harvested at the time of complete death of the green parts and results of this part of the experiment are also included in Table 2. The untreated plants reached this stage 49 days after inoculation, while the groups of plants defoliated 6, 9, 12, 15 and 18 days after inoculation did so 55, 41, 27, 27 and 27 days after inoculation respectively. It is curious that the group of non-defoliated plants shows the lowest percentage of infection and of the groups in which the plants were defoliated, the one defoliated only six days after inoculation shows the highest percentage of infection. The data on the time of ripening show that the group defoliated first had the longest life. This may explain the higher percentages of infection found in this group. Defoliation in a young stage of growth apparently offers the plant better possibilities of recovering than a later defoliation, even if the differences in time are relatively small, as was the case in this experiment.

c. *Experiment with PVX in 1964*

Besides the effect of defoliation on virus translocation, investigated in the previous experiments, it was of interest to study also the effect of removing a part of the plants. In the practice of haulm killing it may happen that only the upper parts of the plants are removed, the lower part, complete with some leaves, remaining intact. Translocation from these parts might also play a role, not only because virus already present could be translocated, but also because the leaves could be infected by aphids. Although infection of older leaves usually does not lead to much infection of tubers, the possibility of a change in behaviour of old leaves after removal of the top of the plants had to be studied. Accordingly the following experiments on this subject were made.

On 1 July 1964 'Bintje' tubers were planted and the single-stemmed plants were inoculated on 20 August with PVX. The inoculations were performed as follows.

Group A (13 plants): three leaflets of a leaf at a height of 100 cm were inoculated.

Group B (13 plants): as group A, but here the tops of the plants were cut off just above the inoculated leaves.

Group C (13 plants): three leaflets of a leaf at a height of 60 cm were inoculated.

Group D (13 plants): as group C, but here the tops of the plants were cut off just above the inoculated leaves.

All plants were harvested three weeks after inoculation. New outgrowths were

not removed. After testing each eye-cutting of all the harvested tubers the results given in Table 3 were obtained. In this table the mean yields in grams per plant of the tubers of each group are also given.

TABLE 3. Infection of tubers with potato virus X after inoculation and topping of potato plants.
De knolbesmetting met aardappel-X-virus na inoculatie en toppen van aardappelplanten.

	Tops of plants not removed		Tops of plants removed just above the inoculated leaf	
	Inoculated leaf at 100 cm	Inoculated leaf at 60 cm	Inoculated leaf at 100 cm	Inoculated leaf at 60 cm
Amount of infection	a:[1] 3/13= 23%	3/13= 23%	10/13= 77%	11/13= 85%
	b: 5/84= 6%	5/88= 6%	15/98= 15%	45/70= 64%
	c: 7/284= 2%	5/321= 2%	18/298= 6%	102/196= 52%
Mean weight of tubers per plant	179 g	170 g	197 g	117 g

1 a: Numerator: number of plants with infected tubers
Denominator: number of test plants
b: Numerator: number of infected tubers
Denominator: number of tubers tested
c: Numerator: number of infected eye-cuttings
Denominator: number of eye-cuttings tested

Table 3 shows that inoculation of a leaf at 100 cm gave exactly the same infection as inoculation at a height of 60 cm. In both cases only a small amount of virus (2% of the eye-cuttings became infected) was translocated to the tubers three weeks after inoculation. It may be concluded that the plants had already reached the stage at which mature plant resistance plays an important role. Topping of the plants proved to have an effect on virus translocation in both cases (groups B and D), which is especially evident in the number of plants showing infection of tubers. It is clear, however, that the effect on translocation is much more pronounced in the group topped and inoculated at 60 cm in which 52% of the eye-cuttings became infected. In this case topping forced the plants to a renewed growth of above ground parts (new shoots and leaves). During this process the plants apparently stopped further development of the tubers as the mean weight of tubers per plant at the time of harvest was considerably lower than those of the other three groups. This change in the physiology of the plants also led to a quickening of virus translocation which was certainly preceded by an activation of the process of virus multiplication. As regards virus translocation the plants behaved like young plants. The effect of topping at 100 cm did not have such a marked effect because in this case the mutilation of the plants was relatively small and consequently the recovery expressed in the formation of new shoots was hardly visible. The plants continued forming tubers and topping had only a very slight effect on the rate of virus translocation.

d. *Experiment with PVY^N in 1965*

On 6 August 1965 50 'Bintje' tubers were planted and grown as single-stemmed plants. The plants were divided into five groups of ten plants. On 27 September (52 days after planting) the plants were inoculated with PVY^N according to

the following scheme. In group A the fifteenth leaf (counted from the base of the plant) was inoculated; in groups B and C the tenth leaf and in groups D and E the fifth leaf. Immediately after inoculation the tops of the plants of the groups C and E were removed just above the inoculated leaves. Five plants from each group were harvested 14 and 20 days after inoculation. The presence of virus in the tubers was established in the usual way as described before. The results, which will be discussed together with those of the next experiment, are given in Table 4.

TABLE 4. Infection of tubers with potato virus Y^N after inoculation and topping of potato plants.
De knolbesmetting met aardappel-YN-virus na inoculatie en toppen van aardappelplanten.

	Tops of plants not removed			Tops of plants removed just above the inoculated leaf	
	Inoculated 15th leaf	Inoculated 10th leaf	Inoculated 5th leaf	Inoculated 10th leaf	Inoculated 5th leaf
Two weeks after inoculation	a:[1] 32/37 = 87%	29/40 = 73%	15/49 = 31%	31/40 = 78%	28/50 = 56%
	b: 88/109 = 81%	84/115 = 73%	35/122 = 29%	83/107 = 78%	67/108 = 62%
Four weeks after inoculation	a: 41/46 = 89%	38/46 = 83%	30/46 = 65%	37/42 = 88%	34/38 = 90%
	b: 132/143 = 92%	107/121 = 88%	93/130 = 72%	114/126 = 91%	99/104 = 95%

[1] a: Numerator: number of infected tubers
Denominator: number of tubers tested
b: Numerator: number of infected eye-cuttings
Denominator: number of eye-cuttings tested

e. *Experiment with PVX in 1965*

Fifty 'Bintje' tubers were planted and treated at the same date as those in the preceding experiment. In this experiment, however, the inoculation was carried out with PVX. The results of this experiment are given in Table 5.

TABLE 5. Infection of tubers with potato virus X after inoculation and topping of potato plants.
De knolbesmetting met aardappel-X-virus na inoculatie en toppen van aardappelplanten.

	Tops of plants not removed			Tops of plants removed just above the inoculated leaf	
	Inoculated 15th leaf	Inoculated 10th leaf	Inoculated 5th leaf	Inoculated 10th leaf	Inoculated 5th leaf
Two weeks after inoculation	a:[1] 5/46 = 11%	7/49 = 14%	3/37 = 8%	8/48 = 17%	32/46 = 70%
	b: 10/151 = 7%	10/166 = 6%	3/149 = 2%	9/141 = 6%	82/121 = 68%
Four weeks after inoculation	a: 21/62 = 34%	13/45 = 29%	26/55 = 47%	26/45 = 58%	51/44 = 84%
	b: 43/181 = 24%	40/159 = 25%	70/156 = 45%	89/166 = 54%	110/121 = 83%

[1] a: Numerator: number of infected tubers
Denominator: number of tubers tested
b: Numerator: number of infected eye-cuttings
Denominator: number of eye-cuttings tested

The results given in Table 4 for PVY^N lead to the conclusion that the older the inoculated leaf the fewer the tubers and eye-cuttings that become infected. This

was found to be true both two and four weeks after inoculation and is in agreement with earlier work on this subject (BEEMSTER, 1966). The infection with PVX (Table 5) was much less than that with PVY^N as could be expected (BEEMSTER, 1961b). In this experiment there was no significant decrease in infection when the inoculated leaf was older in the plants harvested two weeks after inoculation. The plants harvested four weeks after inoculation even showed the highest rate after inoculation of the fifth leaf. These results are not in agreement with earlier work (BEEMSTER, 1958) in which some experiments led to the conclusion that inoculation of old leaves with PVX leads to a smaller extent of tuber infection than inoculation of young leaves.

In experiments d and e topping of the plants was not performed above the 15th leaf, because it was thought to be of no influence, having in mind the results of experiment c. Topping of the plant above the fifth and tenth leaf led to a higher infection of tubers than was observed in the corresponding non-topped plants. This was found both in the experiment with PVY^N and the one with PVX and at both dates of harvesting.

As in experiment c the effect of topping was most striking when it was performed low. The infection with both PVY^N and PVX reached the highest value when topping was performed just above the fifth leaf and harvesting was four weeks after inoculation. Observations on the plants after topping showed that these plants produced new shoots. The plants topped just above the 10th leaf did likewise, but to a much smaller extent than those topped above the fifth leaf. This result was similar to that in experiment c. It is evident that this kind of mutilation of a potato plant generally favours those activities in the plant which are responsible for translocation of virus to tubers.

DISCUSSION

The results of the two experiments (a and b) on the effect of removal of the leaves on the rate of virus translocation in potato plants indicate that this removal does not result in a decreased rate of infection of the tubers produced by the defoliated plants. There is even a tendency towards an increased degree of tuber infection in some plants. The question arises whether the observed infection of tubers is due to translocation of virus already present at the time of removal of the leaves or to newly-formed virus. Earlier experiments (BEEMSTER, 1958, 1961a) have shown that the virus concentration in stems of primarily infected potato plants is relatively small. It has therefore to be assumed that most of the virus found in the tubers afterwards was newly formed in the stems. The formation of virus apparently starts very quickly after removal of the leaves. The effect found in the experiment most probably depends also on the stage of development of the plants. Possibly virus translocation would stop after removal of the leaves if rather old plants were treated. The stems of such plants would probably die and not, as occurred in our experiments, have the capacity to develop a very green colour. More work on this subject is needed. The results obtained here point to the fact that translocation of virus is indeed possible from bare stems, which fact has to be taken into account in the practice of seed potato growing.

The results of experiments c, d and e have demonstrated clearly that the removal of the tops of potato plants immediately after inoculation definitely promotes tuber infection with PVX or PVY^N. This means that the decapitated

plants have lost part of their mature plant resistance. The most plausible reason for this change in physiologic behaviour is to be found in the fact that the removal of a rather large part of the plant forces the plants to restore as far as possible their assimilatory apparatus (surface area of the leaves). Thus, the plants are brought back into a younger stage of development in which virus multiplication and translocation take place at a high rate. The fact that the removal of a part of the plant more or less stops tuber formation and promotes outgrowth of new shoots corresponds with the idea of a change in physiologic behaviour.

The experiments c, d and e also point to the fact that potato haulm killing practice has to be performed perfectly to attain its object. If part of the plant stays alive after the treatment the effect may well be the opposite of that at which the treatment aimed. There is less virus translocation in old intact plants than in plants which are incompletely destroyed.

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SAMENVATTING

In primair met aardappel-X- en aardappel-Y^N-virus besmette aardappelplanten werd het transport van virus naar de knollen voortgezet na ontbladering van de planten. In enkele gevallen bleek zelfs een zwaardere knolbesmetting op te treden. Het verwijderen van de top van aardappelplanten leidde in proeven met dezelfde virussen tot een sterkere knobesmetting dan in niet getopte planten. Dit gold des te meer naarmate de verwijderde top groter was.

Het verwijderen van bladeren en top brengt een zodanige verandering in het fysiologische gedrag van de aardappelplant teweeg dat er een verhoogde mate van virustransport, dus een zwaardere knolbesmetting optreedt. Voor de teelt van pootaardappelen betekenen deze resultaten dat de loofdoding alleen het beoogde doel zal bereiken als een volledige doding van de bovengrondse plantedelen wordt bewerkstelligd.

REFERENCES

BEEMSTER, A. B. R., – 1958. Transport van X-virus in de aardappel (Solanum tuberosum L.) bij primaire infectie. T. PlZiekt. 64: 165–262.

BEEMSTER, A. B. R., – 1961a. Translocation of leaf roll and potato virus Y in the potato. Proc. Conf. Potato Virus Diseases 4, Braunschweig 1960: 60–67.

BEEMSTER, A. B. R., – 1961b. Een vergelijking tussen het transport van X-virus en twee verschillende stammen van Y-virus in aardappelplanten. T. PlZiekt. 67: 278–279.

BEEMSTER, A. B. R., – 1966. The rate of infection of potato tubers with potato virus Y^N in relation to the position of the inoculated leaf. In: Viruses of plants. North Holland Publishing Company, Amsterdam: 44–47.

BOKX, J. A. DE, – 1964. Onderzoekingen over het aantonen van aardappelY^N-virus met behulp van toetsplanten. Diss., Wageningen.