

Parsley latent virus, a new and prevalent seed-transmitted, but possibly harmless virus of *Petroselinum crispum*

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Abstract

Parsley latent virus, a hitherto undescribed virus, was isolated from 38 out of 54 samples of seed of parsley (*Petroselinum crispum*) of 17 out of 24 cultivars and from all five European countries tested, but not from some samples from the USA. It could easily be detected in seedlings and also in seeds germinated on moist filter paper, but not in dry seeds or in seeds soaked in water. Strawberry latent ringspot virus was detected in five samples. The parsley virus is symptomless in parsley and caused latent systemic infection in *Gomphrena globosa*, three cultivars of *Spinacia oleracea* and weak and often transient systemic symptoms in *Chenopodium amaranticolor*, *C. giganteum*, *C. glaucum* and *C. quinoa*, but did not infect any other species out of all 32 species of seven plant families tested in total.

The virus could easily be transmitted mechanically but not by seven aphid species in the non-persistent manner. Dilution end-point was between 100 and 1000, thermal inactivation between 55 and 60°C and ageing in vitro between 7 and 10 days.

Purification yielded a single infectious component. The particles were spherical, ca. 27 nm in diameter, with a sedimentation coefficient of 127.5 S, a buoyant density of 1.449 g/ml, an RNA content of 36% and one type of protein with a relative molecular mass of 22×10^3 . Purification without Triton and urea resulted in preparations with aggregates each consisting of 12 particles in icosahedral array.

The virus differs from all viruses described so far and did not show clear serological affinity with antisera to any of 34 widely differing viruses tested. It does not seem of direct practical importance and may be easily overlooked.

Additional keywords: seed transmission, strawberry latent ringspot virus.

Introduction

When studying host range and symptoms of celery mosaic virus and other viruses of Umbelliferae we soon detected that plants of parsley (*Petroselinum crispum*) grown as test plants from seed in a virus glasshouse contained an apparently undescribed virus. It did not produce symptoms in parsley but caused systemic symptoms in *Chenopodium quinoa* not expected for celery mosaic and differing from those of the seed-transmitted celery latent virus (Bos et al., 1978). Soon thereafter the virus was found to be prevalent in seed lots of various parsley cultivars and origins. We have therefore studied it for its potential importance and for its identity.

Materials and methods

Virus isolates. After initial detection in 1975, the virus was soon isolated from various seed samples. Since no differences on test plants were observed between isolates, almost all research was on isolate Pe6 from seed of cv. 'Extra fijne krul'

propagated in France. Seed samples were obtained via the General Netherlands Inspection Service for Vegetable and Flower Seeds (NAK-G) and directly from seed companies.

Maintenance and propagation was in *Chenopodium quinoa* which started to show systemic symptoms after 10 to 11 days. Long-term storage was in leaf material of *C. quinoa*, dried and stored over CaCl_2 at 4°C.

Biological studies were in the conventional ways using *C. quinoa* as an assay and indicator host for its systemic reaction. Plants were grown and kept in an insect-proof glasshouse at 18-22°C. Additional light from Philips SON/T lamps was given during short days to lengthen the light period to 12 h a day.

Host-range tests were with two to eight plants or more per species depending on plant size. Back inoculations were made to one but usually two plants of *C. quinoa*, from inoculated and non-inoculated leaves, at least 14 and 21 days after inoculation, respectively.

Insect-transmission tests were with virus-free aphid cultures provided by the IPO Department of Entomology. Aphids were first starved for 1 to 2 h and then given acquisition access on infected *C. quinoa* for 10 to 15 min. They were then left for 10 to 30 min on virus-free *C. quinoa* plants (5 plants per aphid species with 10 aphids per plant), thereafter left overnight for another inoculation access period on a second series of test plants, and then killed with an aphicide.

Virus detection in seeds was investigated with commercial seed lot Pe33 of which most of the seeds were infected. Testing was of dry seeds, seeds soaked in water for 24 h, and seeds germinated on moist filter paper at room temperature for 11 days and of seedlings or young plants raised in flats. Seeds or seedlings were tested individually by grinding whole seeds or leaf samples in a mortar with some water and inoculation onto two plants of *C. quinoa*. Serial testing of seed samples of unknown state of health was always by sowing seeds in flats and testing five groups of ten seedlings by sap inoculation onto two to three plants each of *C. quinoa*, *C. amaranticolor* and cucumber. The latter species was added to more readily detect contamination by a nepovirus. In addition, of such samples often ten plants were tested individually.

Virus purification was from leaves of systemically infected *C. quinoa* plants with clear symptoms harvested 12 to 15 days after inoculation. Portions of 100 g were homogenized with a Waring blender in 500 ml of 0.18 M phosphate-citric acid buffer pH 7 containing 0.1% thioglycolic acid. The homogenate was filtered through two layers of cheesecloth, and 2.5% Triton X-100 and 1M urea were added. The mixture was stirred overnight and then centrifuged for 10 min at 6 000 g (all g values will be given at R_{\max}). The supernatant was centrifuged for 2.5 h at 115 000 g and the resulting pellets were resuspended in 60 ml of 0.1 M tris-HCl pH 9. The suspension was stirred for 2 h at 4°C and then centrifuged for 10 min at 8000 g. The supernatant was spun for 2.5 h at 86 000 g and the resulting pellets were resuspended in 2 ml of 0.1 M tris-HCl pH 9. After stirring overnight at 4°C the suspension was centrifuged for 10 min at 8000 g and 1-ml portions loaded onto 10-40% sucrose gradients in 0.1 M tris-HCl pH 9. The gradients were spun for 2 h at 24 000 rpm in a Beckman SW27

rotor. The tube contents were fractionated using an ISCO density-gradient fractionator and the virus fractions, diluted 1:1 with 10 mM glycine-NaOH pH 9, were centrifuged for 2.5 h at 86 000 g. The pellets were resuspended in 1 ml 10 mM glycine-NaOH pH 9. Thereafter 0.5 ml of this virus suspension was mixed with 3 ml of CsCl solution (0.7010 g/ml) in a Beckman SW41 tube, overlaid with paraffin oil and centrifuged for ca 19 h at 30 000 rpm. The virus band was recovered by puncturing the bottom of the tube and the CsCl was removed by dialyzing against 10 mM glycine-NaOH pH 9.

Biophysical measurements. The sedimentation coefficient at infinite dilution was determined by the graphical method of Markham (1960) using a Spinco Model E ultracentrifuge with schlieren optics, and the buoyant density was measured as described by Maat et al. (1978). The degradation of the virus and polyacrylamide gel electrophoresis to determine the nature of the nucleic acid and the relative molecular mass of the coat protein subunit were done as described before (Huttinga and Mosch, 1976).

Serology. For antiserum production a rabbit was injected with fresh highly purified Pe6-virus preparations. For each injection the virus obtained from 300 g of leaf material was used. Two intravenous injections with an interval of three days were followed two weeks later by an intramuscular injection with an emulsion of virus and Freund's incomplete adjuvant. After another two weeks, bleeding was started.

Serological testing was by double diffusion in agar using purified virus preparations. Antisera to tomato bushy stunt virus and *Pelargonium* leaf curl virus were kindly provided by Dr M. Hollings, Littlehampton, England, to raspberry bushy dwarf virus by Dr A.F. Murrant, Invergowrie, Scotland, to Myrobalan latent ringspot virus by Dr J. Dunez, Bordeaux, France, to elm mottle virus by Dr A.T. Jones, Invergowrie, Scotland, to a mosaic from broad bean (MF) by Dr J.C. Devergne, Antibes, France, to a pear pollen virus by Dr J.A. Tomlinson, Wellesbourne, England, and to sowbane mosaic virus by Dr G. Morvan, Montfavet, France. Most other antisera were from own stock. Of each antiserum four dilutions were tested, ranging from undiluted to 1:256, the dilutions used depending on the homologous antiserum titre.

Electron microscopy in crude sap was after staining with 2% PTA pH 6.5 or with ammonium molybdate. Purified samples were negatively stained with 1% PTA pH 6 in water or with 1% uranylacetate in water. Preparations were examined in a Philips EM 300. Particle size measurements were made with TMV as an internal standard.

Results

Host range and symptoms. The virus has been isolated several times from seed of a number of parsley cultivars (see below). Plants grown from seedlings that proved to be infected at testing have been kept under observation for extended periods of time, but were never found to differ from virus-free plants simultaneously studied. Out of 32 plant species of seven families tested only four *Chenopodium* species, *Gomphrena globosa* (high concentration) and *Spinacia oleracea* 'Califlay', 'Dorema,

and 'Géant d'Hiver', contracted systemic infection. *C. amaranticolor*, *C. giganteum*, *C. glaucum* and *C. quinoa* were the only species producing (systemic) symptoms. Latent local infection was obtained in *C. ficifolium*, *C. foetidum*, *C. murale* and *C. vulvaria*. No infection could be detected in *C. ambrosioides*, *C. capitatum* and *C. foliosum*. Other plant species tested without becoming infected were Cucurbitaceae: *Cucumis sativus* 'Gele Tros'; Leguminosae: *Phaseolus vulgaris* 'Bataaf', *Pisum sativum* 'Koroza' and 'Rondo', and *Vicia faba* 'Kompakta'; Solanaceae: *Datura stramonium*, *Nicotiana clevelandii*, *N. glutinosa*, *N. hybrida* 'Christie', *N. megalosiphon*, *N. rustica*, *N. tabacum* 'White Burley', *Petunia hybrida* and *Physalis peruviana*, and the Umbelliferae: *Ammi majus*, *Apium graveolens* var. *dulce*, *Coriandrum sativum* and *Daucus carota* 'Amsterdamse Bak'. In one experiment a latent local infection was obtained in *Beta vulgaris* and a latent systemic infection in *Lactuca sativa*, but another experiment with these species was negative.

Chenopodium quinoa reacted with systemic symptoms only. They ranged from weak vein clearing to fine and diffuse chlorotic spotting usually associated with the venation. Such symptoms mostly started 10 to 11 days after inoculation in the lower part of the first non-inoculated leaves and were thereafter more general in younger leaves that often became slightly deformed (Fig. 1). There was hardly any effect on plant growth and habit, and plants often completely recovered. In several instances symptoms were weak and could easily be overlooked. In *C. amaranticolor* symptoms were similar (Fig. 2) but hardly noticeable and mostly absent. In *C. giganteum* and *C. glaucum*, the two only other species reacting, symptoms resembled those of *C. quinoa*, although slightly more malforming in *C. glaucum*, and plants also readily recovered. No local-lesion host was detected.

Persistence of infectivity. Dilution end-point was between 100 and 1000, thermal-inactivation point was between 55 and 60°C, and ageing in vitro between 7 and 10 days. Infectivity persisted in leaf material dried and stored over CaCl₂ for at least 2½ years.

Insect-transmission tests. The virus was not transmitted in the non-persistent manner by any of the following aphid species: *Acyrtosiphon pisum*, *Aphis fabae*, *Aulacorthum solani*, *Macrosiphum euphorbiae*, *Myzus ascalonicus*, *M. persicae* (tested three times) and *Rhopalosiphum padi*.

Seed-transmission tests. Seed transmission in inoculated plants was only tested in *C. quinoa*. Seed harvested from such plants inoculated with isolates Pe 26, Pe29 and Pe33 was sown and all 50 seedlings examined with each isolate were visually normal. After testing these in two groups of 25 onto *C. quinoa* no reaction was obtained.

The virus could neither be detected in 10 dry parsley seeds with Pe33 and in 20 seeds soaked for 24 h all tested individually, nor in 4 groups of 10 soaked seeds. After germination of seed from the same lot on moist filter paper for 11 days, however, the virus was detected in 8 out of 10. Testing of seedlings grown in flats in the glasshouse resulted in positive reactions with all 4 groups of 10 seedlings and with all 10 seedlings tested individually.

In the serial tests of several commercial seed samples 38 out of 54 samples tested were found to contain the virus and infection percentages varied from some to

Fig. 1. Systemic symptoms of parsley latent virus in *Chenopodium quinoa*.

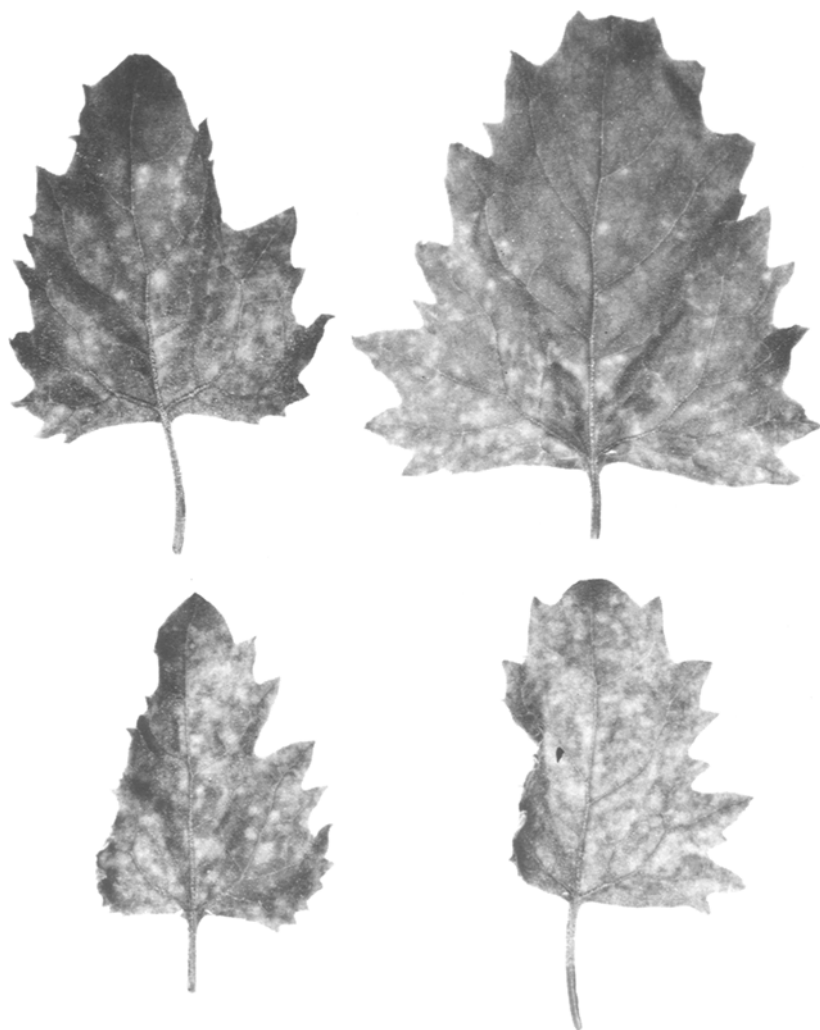


Fig. 1. Systemische symptomen van het latente peterselievirus in *Chenopodium quinoa*.

practically 100, as with Pe33. Infection rates usually were high. Infection was detected in 17 out of 24 cultivars tested in seed from all six European countries from which samples had been obtained but not in samples of five cultivars from the USA. Symptoms were never observed in seedlings and plants grown from infected seed.

Five samples also contained strawberry latent ringspot virus (SLRV), although apparently in low concentrations. This virus could be easily distinguished from the parsley virus by its severe systemic reaction on *C. quinoa* and *C. amaranticolor* and by systemic symptoms on cucumber seedlings. To detect the contamination with SLRV, addition of the latter two indicators hosts was therefore considered essential. Identification was confirmed serologically. In case of infection or contamination

Fig. 2. Weak and rarely occurring systemic symptoms of parsley latent virus in *Chenopodium amaranticolor*.



Fig. 2. Zwakke, zelden voorkomende systemische symptomen van het latente peterselievirus in *Chenopodium amaranticolor*.

with SLRV, the parsley virus escaped detection because of its much weaker symptoms, absence of a differential host and, at the time of testing, lack of an antiserum.

Virus purification and some biophysical properties. During first attempts to purify the virus it clearly tended to aggregate. However, this could be greatly overcome by the described purification method and high amounts of infectious virus were obtained.

In sucrose-gradient centrifugation the virus sedimented in one zone and in the electron microscope uniform spherical particles were found (Fig. 3). The sedimenta-

Fig. 3. Electron micrographs of parsley latent virus: A) in crude sap with ammonium molybdate; B) in PTA after purification via a sucrose gradient. Bars represents 100 nm.

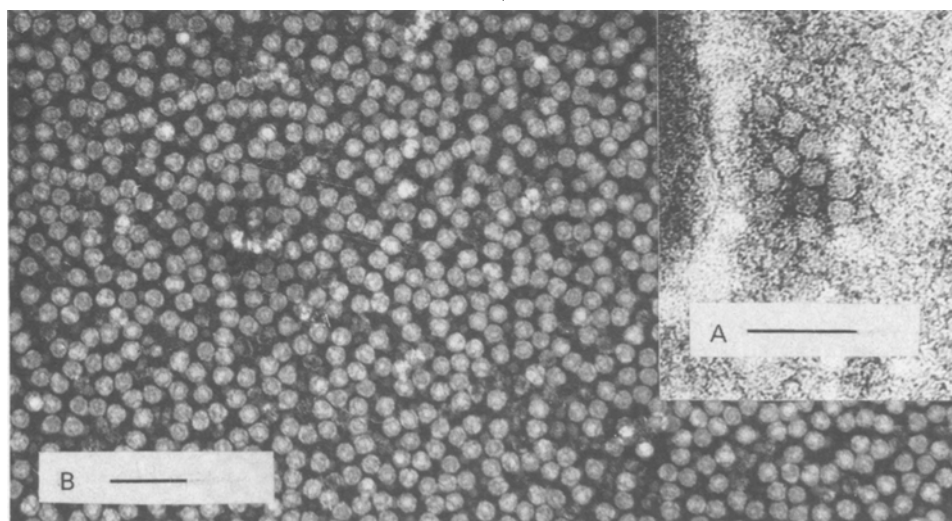


Fig. 3. Elektronenfoto's van het latente peterselievirus: A) in ruw sap met ammoniummolybdaat; B) in PTA na zuivering via een suikergradiënt. Vergrotingsstaaf geeft 100 nm weer.

tion coefficient at infinite dilution in 10 mM glycine-NaOH pH 9 at 20°C was 127.5 S. In density-gradient centrifugation in CsCl the virus reached equilibrium in a single zone with a buoyant density of 1.449 g/ml. From this value a nucleic-acid content of 36% was calculated according to Sehgal et al. (1970).

The degraded virus showed one protein band and one nucleic-acid band in polyacrylamide gel electrophoresis. The protein had a relative molecular mass of 22×10^3 . The nucleic acid was sensitive to RNase.

When purification was done in 0.18 M phosphate-citric acid pH 7 only, an extra zone was found in sucrose-gradient centrifugation. This zone contained clusters of 12 particles (Fig. 4).

Fig. 4. Electron micrographs of aggregating particles of parsley latent virus after purification in phosphate-citric acid buffer and sucrose-gradient centrifugation, fixed in glutaraldehyde and stained in uranylacetate; B, C, D are enlargements of aggregates in 2-fold (B), 3-fold (C) and 5-fold (D) symmetry. Bars represents 100 nm.

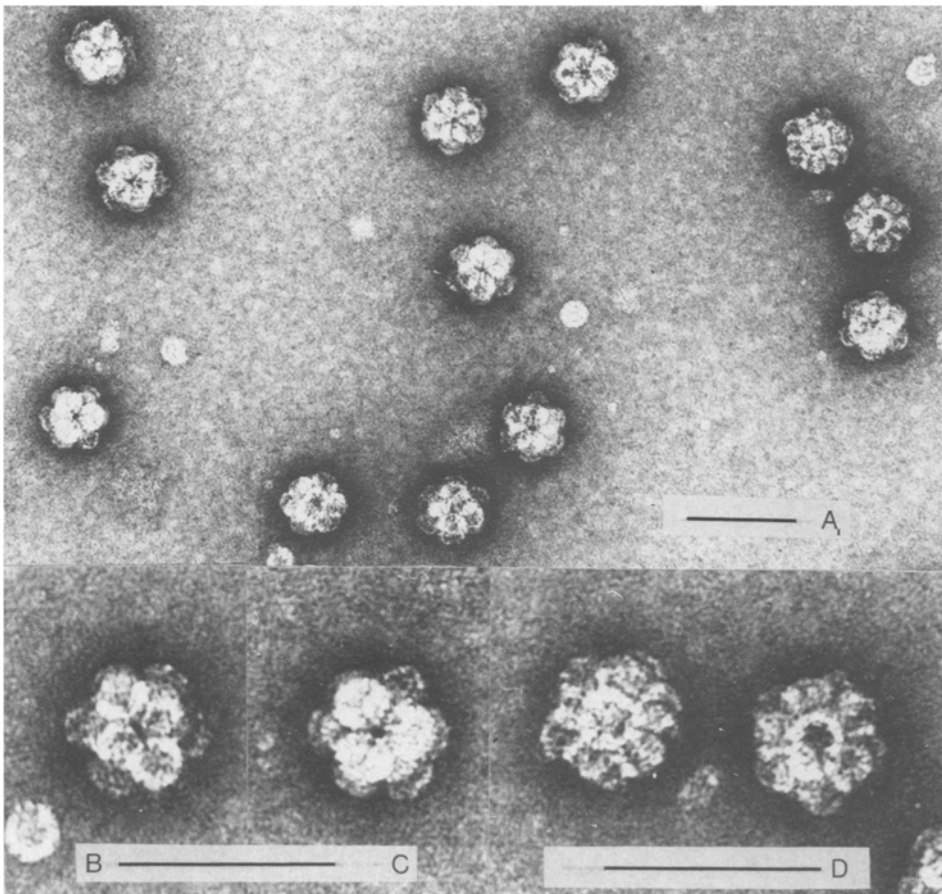


Fig. 4. Elektronenfoto's van geaggregeerde deeltjes van het latente peterselievirus na zuivering in fosfaatci-troenzuurbuffer en suikergradiëntcentrifugering, gefixeerd in glutaraaldehyde en gekleurd in uranylacetaat; B, C, D zijn vergrote opnamen van aggregaten in 2-voudige (B), 3-voudige (C) en 5-voudige (D) symmetrie. Vergrotingsstaven geven 100 nm weer.

Table 1. Results of Ouchterlony double-diffusion tests, performed to identify Pe6 virus. The figures represent the highest antiserum dilutions still reacting, or the titres obtained.

Viruses or antisera tested	Reactions of homologous antisera	Reactions of Pe6 antiserum	Reactions of antisera with Pe6 antigen
Alfalfa mosaic virus	64	—	—
Apple mosaic virus	8	—	—
Arabid mosaic virus	256	—	—
Bean pod mottle virus			—
Bean southern mosaic virus			—
Broad bean wilt virus	256	—	—
Carnation mottle virus			—
Carnation ringspot virus	256	—	—
Cherry leafroll virus	256	—	—
Cocksfoot mild mosaic virus	256	—	—
Cowpea mosaic virus-yellow strain			—
Cowpea mosaic virus-severe strain			—
Cucumber mosaic virus -B32	64	—	—
Cucumber mosaic virus -yellow strain	64	—	—
Cucumber mosaic virus -red currant	8		—
Cucumber mosaic virus - <i>Nerine</i>		—	
Elm mottle virus			—
Ge 36 virus	64	—	—
Lilac ring mottle virus	256	—	—
Lonicera virus (unknown)		—	—
MF-Devergne			—
Myrobalan latent ringspot virus			—
Pear pollen virus			—
Pelargonium leaf curl virus			—
Pelargonium virus L128	64	—	—
Prunus necrotic ringspot virus	32	—	—
Raspberry bushy dwarf virus			—
Raspberry ringspot virus	256	—	—
Red clover mottle virus			—
Sowbane mosaic virus			—
Strawberry latent ringspot virus	256	—	—
Tobacco necrosis virus- <i>Lonicera</i>	256	—	—
Tobacco necrosis virus-B4	256	—	—
Tobacco ringspot virus	256	—	—
Tobacco streak virus-Sb3	64	—	—
Tomato aspermy virus-1	256*		4
Tomato aspermy virus-2	256*	—*	—
Tomato aspermy virus-3	256*	—	—
Tomato black ring virus	256	—	—
Tomato bushy stunt virus			—
Tomato ringspot virus	256	—	—
Turnip yellow mosaic virus			—
Pe6	1024	1024	1024

—No reaction observed; * Reaction with TAV-2 antigen.

Tabel 1. Resultaten van de Ouchterlony-dubbele-diffusieproeven ter identificatie van het Pe6-virus. De getallen geven de hoogste antiserumverduunningen weer waarbij nog reactie optrad, of de verkregen titers.

Serology. Results of serological experiments are summarized in Table 1. Besides the homologous antiserum (titre 1024), only one antiserum to a chrysanthemum isolate of tomato aspermy virus (TAV) reacted very weakly with Pe6. Two antisera to other TAV isolates did not react with Pe6, nor did the Pe6 antiserum react with a TAV isolate. The results indicate that Pe6 differs clearly serologically from any of the viruses included in the serological experiments. With the agar double-diffusion test, Pe6 could be detected in crude sap of *C. quinoa*, but not in crude extracts from infected parsley.

Electron microscopy. In crude sap of *C. quinoa* no particles have ever been observed with PTA. In ammonium molybdate some spherical particles were found on one occasion (Fig. 3A). In purified preparations (Fig. 3B) particles were spherical, uniform and ca. 27 nm in diameter. The particles obtained after purification using phosphate-citric acid buffer pH 7 partly occurred in characteristic aggregates of 12 particles with icosahedral symmetry (5-3-2) (Fig. 4).

Discussion

In recent years virus research of umbelliferous crops has gained increasing interest (e.g. Wolf and Schmelzer, 1972). Prior to 1970 only a few reports have been published on viruses isolated from naturally infected parsley plants, viz. on celery mosaic virus (Severin and Freitag, 1938), cucumber mosaic virus (Wellman, 1935; Kovachevsky, 1965) and an unidentified virus resembling celery mosaic virus or parsnip mosaic virus (Férel et al., 1969).

More systematic surveys on parsley viruses have thereafter been reported for East Germany (DDR) by Wolf (1970) and Wolf and Schmelzer (1972) (in the following list denoted as W and W&S, respectively), and for Britain by Frowd and Tomlinson (1972: F&T). The viruses they detected were alfalfa mosaic virus (W, later also isolated in the USA by Campbell and Melugin, 1971), carrot mottle virus (W; F&T), celery mosaic virus (W; F&T; simultaneously also isolated in the USA by Sutabutra and Campbell, 1971), cucumber mosaic virus (W; F&T), broad bean wilt virus (W; F&T), tomato black ring virus (W&S), an unidentified 500 nm virus, possibly belonging to the potexvirus group (F&T), and another new unnamed and undescribed virus (W&S). In the DDR celery mosaic virus and cucumber mosaic virus were the most prevalent ones, whereas in Britain carrot mottle virus was the most frequently isolated virus. There, the latter virus was considered the principal cause of severe stunting of parsley with leaf chlorosis and reddening. Upon back inoculation to parsley, the other four viruses isolated in Britain induced slight symptoms or none. Cucumber mosaic virus and especially tomato black ring virus might pass via parsley seed because of their known seed transmission in other hosts. Our detection of strawberry latent ringspot virus in some of the seed samples tested adds another virus to the list of viruses naturally infecting parsley. It has earlier been reported to be seed transmitted up to 100% in celery (*Apium graveolens* var. *dulce*), where seedlings grown from infected seed also were symptomless (Walkey and Whittingham-Jones, 1970).

The latent virus we have detected to occur in seed of several cultivars and from various origins, and now characterized, differs from all of the above-listed viruses in

host range and symptoms and from most of them in particle morphology and size. It resembles nepoviruses for its latency in most hosts and its high rates of seed transmission in parsley. However, its physical properties make it impossible to classify the virus in any of the known plant virus groups. The virus could not be transmitted by seven aphid species in the non-persistent manner and persistent transmission is most unlikely because of its seed transmission.

The virus differs from nepoviruses (Harrison and Murrant, 1977) in much smaller coat protein (although in molecular mass resembling the two proteins of cherry rasp-leaf virus), in having only one RNA-containing particle type instead of two and no empty top component. Its particles sediment at 127.5 S fitting for nepovirus bottom components. Serological experiments have also shown that the virus clearly differs from 33 spherical viruses, including nepoviruses as well as cucumovirus, tombus and tymoviruses, and from alfalfa mosaic virus. Unfortunately, our experiments did not prove whether the tomato aspermy virus preparation (TAV) to which the TAV-antiserum that reacted with Pe6 (Table 1) had been prepared was contaminated with Pe6 or a Pe6-related virus, or that TAV and Pe6 are related viruses. *Nicotiana rustica* in which TAV had been propagated is a non-host of Pe6 and this suggests contamination of TAV with Pe6 to be unlikely. Moreover, only one TAV antiserum out of three reacted with Pe6, and Pe6 antiserum did not react with the TAV isolate tested. Also, biophysically Pe6 differs from cucumoviruses, and no non-persistent aphid transmission could be obtained with seven aphid species tested. Persistent transmission was not tested since this is unlikely for a seed-borne virus.

The parsley virus resembles beet cryptic virus in particle morphology, in absence of symptoms in its natural hosts and in high rates of seed transmission. However, the latter virus is not mechanically transmissible, is slightly bigger (30 nm), may have more than one particle component, and the relative molecular mass of its protein (36×10^3) is higher than that of our virus (Kassanis et al., 1977, 1978). Our virus is readily transmissible mechanically and did not systemically infect the cultivar of *Beta vulgaris* tested.

The icosahedral aggregates of particles of the parsley virus observed in an extra zone after purification in phosphate-citric acid buffer are similar to those reported for tobacco necrosis virus satellite (Kassanis and Woods, 1968), a kale strain of radish mosaic virus (Kassanis et al., 1973) and carnation ringspot virus (Tremaine et al., 1976). However, these viruses clearly differ biologically and biophysically¹. Since the virus could not be identified with any known virus we have now described it in detail as a new virus and propose the name *parsley latent virus*.

As yet the virus seems of academic interest only since its host range is very limited and none of the crop species tested reacted with symptoms. Potentially, however, the virus is of importance since it theoretically may cause disease in plant species not yet tested, although a vector is not yet known, and since it may aggravate symptoms of other viruses and other pathogens if occurring in complex. The virus could not be detected in dry or soaked seeds but was easily isolated from seedlings infected from seed. In several seed origins incidence was high, but no local lesion hosts were found and symptoms in *Chenopodium* spp. were weak and often transient. Thus, the virus

¹ Note added in proof: Purified parsley latent virus did not react with an antiserum to beet cryptic virus kindly sent by Mr R.F. White, Rothamsted Experimental Station, Harpenden, England, whereas it clearly reacted with the homologous antiserum.

may easily escape attention and may have done so in earlier surveys of parsley for viruses.

Samenvatting

Latent peterselievirus, een nieuw, maar mogelijkwerwijs onschadelijk, met zaad overgaand virus van Petroselinum crispum

In zaailingen van peterselie (*Petroselinum crispum*) werd een nog niet eerder beschreven virus aangetroffen. Het virus kon niet worden aangetoond door toetsing van droge of in water geweekte zaden op *Chenopodium quinoa* maar wel in op filtreerpapier gekiemde zaden en vooral in zaailingen. Het werd aangetroffen in 38 van de 54 getoetste herkomsten, in 17 van de 24 getoetste rassen en in zaad vermeerderd in alle zes hierop onderzochte Europese landen maar niet in enkele zaadmonsters uit de USA. In sommige monsters bevatten nagenoeg alle zaden het virus. In vijf herkomsten werd eveneens het nog niet eerder in peterselie gerapporteerde latente aardbeikringvlekkenvirus geconstateerd. Dit virus kan bij toetsing gemakkelijk worden herkend door systemische symptomen in *C. amaranticolor* en komkommer.

In geïnfecteerde peterselieplanten zijn geen afwijkingen waargenomen. Het virus kon niet op non-persistente wijze worden overgebracht met zeven bladluisoorten maar wel gemakkelijk met sap. Van 32 getoetste plantesoorten van zeven families, waaronder vier schermbloemigen, kon het virus slechts worden overgebracht op vier *Chenopodium*-soorten, *Gomphrena globosa* en alle drie getoetste spinazierassen. Alleen *C. quinoa* (Fig. 1), *C. giganteum*, *C. glaucum* en soms ook *C. amaranticolor* (Fig. 2) reageerden met vaak voorbijgaande systemische symptomen. Een lokale-lesietoetsplant werd niet gevonden. Zaadovergang bij *C. quinoa* kon niet worden aangetoond.

Voor de houdbaarheid van het infectievermogen werden de volgende waarden gevonden: verdunningseindpunt 100-1000, thermaal inactiveringspunt 55-60°C en houdbaarheid in vitro 7-10 dagen.

Zuivering door homogenisatie in fosfaatcitroenzuurbuffer, behandeling met Triton X-100 en ureum en differentiële en daarna dichtheidsgradiëntultracentrifugering leverde preparaten op met uniforme deeltjes van ca. 27 nm diameter (Fig. 3B), een sedimentatiecoëfficiënt van 127,5 S, een zweefdichtheid van 1,449 g/ml, een RNA-gehalte van 36% en een relatieve moleculaire massa van de eiwitonder-eenheid van 22×10^3 . Bij zuivering zonder toepassing van Triton en ureum werd een extra zone verkregen met aggregaten van 12 deeltjes in icosaeëdrische rangschikking (Fig. 4). In ruw plantesap waren slechts met grote moeite enkele deeltjes met behulp van de elektronenmicroscop te vinden.

Het virus reageerde niet met antisera tegen 33 bolvormige virussen en luzerne-mozaïekvirus (Tabel 1). Of de zwakke reactie verkregen met één antiserum tegen het tomate-aspermievirus een verre serologische verwantschap inhoudt, dan wel het gevolg is van een verontreiniging, werd niet vastgesteld.

Het virus wordt beschouwd als een geheel nieuw virus waarvoor de naam *latent peterselievirus* wordt voorgesteld. Het lijkt door zijn beperkte waardplantenreeks en symptoomloosheid in de vatbaar bevonden soorten, behalve in enkele als toetsplant te gebruiken *Chenopodium*-soorten, nauwelijks van praktische betekenis.

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