T.Pl.-ziekten 67 (1961): 562–565

IDENTITY OF RED CURRANT SPOON LEAF VIRUS[1]

Met een samenvatting: De identiteit van het lepelbladvirus van rode bes

BY

B. D. HARRISON

Rothamsted Experimental Station, Harpenden, Hertfordshire, England

INTRODUCTION

By inoculating a range of herbaceous species with sap from diseased red currant leaves, viruses causing ringspot symptoms were transmitted by HILDEBRAND (1942) in the United States, and by KLESSER (1951) and VAN DER MEER (1960) in Europe. HILDEBRAND identified his isolate as tomato ringspot virus but the possible identity of the European isolates with this or other viruses was not established. I have now obtained experimental evidence that VAN DER MEER's (1960) red currant spoon leaf virus is a strain of raspberry ringspot virus. Circumstantial evidence suggests that KLESSER's (1951) red currant ringspot virus is the same as red currant spoon leaf virus.

MATERIALS AND METHODS

The viruses used were red currant spoon leaf virus (kindly supplied by F. A. VAN DER MEER, Instituut voor Plantenziektenkundig Onderzoek, Wageningen); raspberry ringspot virus, "type" strain from raspberry in Scotland (HARRISON, 1958); KUNZE's (1958) virus from cherry affected by Pfeffingerkrankheit in Germany; and a virus obtained from cherry in The Netherlands by Miss H. J. PFAELTZER. The two last were provided by Dr. C. H. CADMAN, Scottish Horticultural Research Institute, Invergowrie, and are strains of raspberry ringspot virus (CADMAN, 1960 and unpublished results).

Antisera were obtained by injecting rabbits with partially purified virus preparations (HARRISON & NIXON, 1960) from *Petunia hybrida* Vilm. One intravenous injection was followed a month later by three intramuscular injections at fortnightly intervals. For intramuscular injections, 1 ml virus preparation was emulsified with 1.25 ml of FREUND's adjuvant, and 1 ml of the emulsion was injected into each hind leg. The rabbits were bled about a month after the last injection.

SYMPTOMATOLOGY

The symptoms caused by red currant spoon leaf virus (SLV) in *Nicotiana tabacum* L., *N. rustica* L., cucumber and *Chenopodium amaranticolor* Coste & Reyn. (VAN DER MEER, 1960) closely resemble those caused by raspberry ringspot virus (CADMAN, 1956; HARRISON, 1958). At Rothamsted, however, SLV occasionally caused enations on the undersides of systemically infected

[1] Accepted for publication 23 August, 1961.

leaves of cucumber var. 'Every-day'. The "type" culture of raspberry ringspot virus (RRV) did not do this, but the two isolates from cherry, which came from trees showing leaf enations, elicited more enations in cucumber than SLV. The enations in cucumber, however, were not produced as often by these viruses as by strains of tomato black ring virus, for which they were considered by SMITH (1957) to be diagnostic.

In *Chenopodium quinoa* Willd., SLV produced lesions in inoculated leaves and severe systemic symptoms. In bean (*Phaseolus vulgaris* L. var. 'The Prince') small chlorotic or necrotic lesions developed in inoculated leaves in winter, followed by systemic necrotic spotting. The reactions in these two species are typical of those caused by RRV. In *Petunia hybrida,* however, the symptoms developing in systemically infected leaves, bright yellow ringspot or mosaic patterns followed by almost complete bleaching of the shoots, were quite unlike those caused by the raspberry (HARRISON, 1958) and cherry isolates.

PROPERTIES *IN VITRO*

Sap from systemically infected leaves of *P. hybrida* was used as the source of virus for these tests, and was mixed with "Celite" abrasive before inoculation to *C. amaranticolor* assay plants. The results were:

Thermal inactivation point. Between 70 and 75 °C for 10 min.

Longevity in vitro. Between 3 and 4 weeks at 18 °C.

Dilution end-point. Between 1/1000 and 1/10000.

SLV thus has similar *in vitro* properties to RRV but seems slightly more stable to heating and to aging at room temperature. Electron micrographs, taken by H. L. NIXON and R. D. WOODS, of partially purified preparations of SLV showed numerous polyhedral particles indistinguishable from those of RRV (HARRISON & NIXON, 1960).

PLANT-PROTECTION AND SEROLOGICAL TESTS

P. hybrida plants systemically infected with RRV developed no additional symptoms after inoculation with SLV. Similarly, *P. hybrida* infected with SLV developed no additional symptoms when inoculated with RRV.

Precipitin tests in tubes, using antiserum to RRV or to SLV, and partially purified preparations of the two viruses, showed that each antiserum had the same precipitation end-point against both viruses (table 1). Cross-absorption tests showed that there were, at the most, only slight antigenic differences between the two viruses (table 1). Precipitin tests in agar gel supported this conclusion. Each virus formed a single line when allowed to react with its homologous or heterologous antiserum. When the two viruses were tested simultaneously against either one or other of the antisera, the respective precipitation lines joined with no suggestion of a spur.

DISCUSSION

The results described above show that SLV is a strain of raspberry ringspot virus, sharing most of its antigenic groups with the "type" strain, from which it can be distinguished by the symptoms produced in *P. hybrida.* Circumstan-

TABLE 1. Serological reactions of red currant spoon leaf (SLV) and raspberry ringspot (RRV) viruses.
Serologische reacties van lepelbladvirus van rode bes (SLV) en „ringspot"-virus van framboos (RRV).

Antiserum to *Antiserum tegen*	Antiserum absorbed with *Antiserum verzadigd met*	Antigen	
		RRV	SLV
RRV	–	1/1024*	1/1024
RRV	SLV	1/20	1/20
SLV	–	1/512	1/512
SLV	RRV	1/10	1/20

* Figures are the precipitation end-points of the antisera after 5 hr. incubation at 37 °C; the antigens were used at constant dilution.
De cijfers zijn de precipitatie-eindpunten van de antisera na 5 uur incuberen bij 37 °C; de antigenen werden in een constante verdunning gebruikt.

tial evidence strongly suggests that KLESSER's (1951) red currant ringspot virus is the same as SLV. Not only was KLESSER's virus isolated from plants originally obtained from The Netherlands, but its symptomatology in herbaceous plants and its properties *in vitro* resemble those of SLV; in particular it caused bright yellow ringspot symptoms and bleaching in young shoots of *P. hybrida* (Figure 2 in KLESSER's paper).

Tomato ringspot, the virus obtained from red currant in the United States, clearly differs from SLV. Both viruses cause ringspot symptoms in herbaceous plants, but they have different properties, and SLV did not precipitate with antiserum to the peach yellow bud mosaic strain of tomato ringspot virus (C. H. CADMAN, unpublished results). The natural mode of spread of the two viruses in red currant has not yet been established but other strains of the same viruses are soil-borne (WAGNON & BREECE, 1955; CADMAN & LISTER, 1961; CADMAN, 1956; HARRISON, 1956), and there is no reason to suspect that the red currant strains are exceptional in this respect.

SUMMARY

Red currant spoon leaf virus, isolated from red currant in The Netherlands, is a strain of raspberry ringspot virus; it shares most of its antigenic groups with the "type" strain from Scottish raspberry but causes distinctive symptoms in *Petunia hybrida*. It differs from tomato ringspot virus, obtained by HILDEBRAND (1942) from red currant in the United States. KLESSER's (1951) red currant ringspot virus is probably the same as red currant spoon leaf virus.

SAMENVATTING

Het hier beschreven onderzoek heeft uitgewezen, dat het in Nederland uit rode bes geïsoleerde lepelbladvirus (VAN DER MEER, 1960) een stam van het „ringspot"-virus van framboos is. Hoewel het lepelbladvirus de meeste antigenen gemeen heeft met de „type"-stam van het framboze-„ringspot"-virus, die uit een Schotse frambozeplant werd verkregen (tabel 1), kunnen beide worden onderscheiden op grond van de symptomen die zij op *Petunia hybrida*

teweegbrengen. Het lepelbladvirus verschilt van het „ringspot"-virus van tomaat, dat in de Verenigde Staten door HILDEBRAND (1942) uit rode bes werd geïsoleerd. Het door KLESSER (1951) bestudeerde „ringspot"-virus van rode bes is waarschijnlijk hetzelfde als het lepelbladvirus.

ACKNOWLEDGEMENTS

I am indebted to those mentioned above, for providing virus cultures and allowing me to quote unpublished results, and for technical assistance to Miss J. HILLIER.

REFERENCES

CADMAN, C. H., – 1956. Studies on the etiology and mode of spread of Scottish raspberry leaf curl disease. J. hort. Sci. 31: 111–118.

CADMAN, C. H., – 1960. Studies on the relationship between soil-borne viruses of the ringspot type occurring in Britain and Continental Europe. Virology 11: 653–664.

CADMAN, C. H. & R. M. LISTER, – 1961. Relationship between tomato ringspot and peach yellow bud mosaic viruses. Phytopathology 51: 29–31.

HARRISON, B. D., – 1956. Soil transmission of Scottish raspberry leaf-curl disease. Nature 178: 553.

HARRISON, B. D., – 1958. Further studies on raspberry ringspot and tomato black ring, soil-borne viruses that affect raspberry. Ann. appl. Biol. 46: 571–584.

HARRISON, B. D. & H. L. NIXON, – 1960. Purification and electron microscopy of three soil-borne plant viruses. Virology 12: 104–117.

HILDEBRAND, E. M., – 1942. Tomato ringspot on currant. Am. J. Bot. 29: 362–366.

KLESSER, P. J., – 1951. A virus disease of red currant (Ribes rubrum L.). Ann. appl. Biol. 38: 707–713.

KUNZE, L., – 1958. Ein Virus der Tabak-Ringflecken-Gruppe von Süsskirsche. Phytopath. Z. 31: 279–288.

MEER, F. A. VAN DER, – 1960. Onderzoekingen betreffende bessevirussen in Nederland I. Lepelblad van rode bes. T.Pl.-ziekten 66: 12–23.

SMITH, K. M., – 1957. A textbook of plant virus diseases. London.

WAGNON, H. K. & J. R. BREECE, – 1955. Evidence of retention of peach yellow bud mosaic virus in soil. Phytopathology 45: 696.