Neth. J. Pl. Path. 81 (1975) 45–48 SHORT COMMUNICATION

Evidence for a soilborne nature of freesia leaf necrosis

H. J. M. VAN DORST

Glasshouse Crops Research and Experiment Station, Naaldwijk

Accepted 17 September 1974

This paper reports the results of experiments to determine whether or not freesia leaf necrosis (FLN) is soilborne. The agent which causes this disease has yet to be isolated. The vertical transmission of the disease to the new corms and cormels suggests that the causal agent has a viral etiology (Van Dorst, 1973). Attempts to transmit the agent by aphids or sap inoculation techniques were unsuccessful. As no method of transmission has been found, the relation between the disease and the soil in which the plants are cultivated, was studied.

Soil samples were collected at nine nurseries of which at seven the disease occurred, but each with a different history of soil treatment (Table 1). In addition, material was collected from decomposing freesia plant remains. These remains were mixed with sterilized sand and white peat to obtain normal growth.

In the first experiment 10 healthy cormels, 15 infected with freesia mosaic virus (FMV) and 10 infected with FLN were planted in trays each with one of the soil samples. The cormels with FLN were included in this experiment to provide the vector, supposedly occurring in the soil, with a virus source. Cormels with FMV were used in order to detect an infection with FLN more readily as a double infected plant will show severe leaf necrosis (Van Dorst, 1973). The plants were grown under conditions described previously (Van Dorst, 1973). At the end of the growing season the new corms and cormels were harvested and prepared for indexing in the next season after planting in soil treated with methylbromide (Table 1).

Infection with leaf necrosis, at an incidence of 100% in soil from nursery A, occurred in six of the seven soil samples collected at nurseries where freesias have been grown. No transmission was found in soil from nursery D, nor in soil from those (E and I) where no freesias had been cultivated. Transmission was also found in the tray with the mixture of decomposed remains, sand and white peat. Because plants infected with FLN were planted in each tray, it was not clear whether the vector acquired the causal agent from these plants, or contained it already. This was tested in a second experiment, in which only a small number of the soil samples used in experiment 1 were tested.

In each tray were planted 24 healthy and 24 FMV infected cormels of the cv. Rose Marie and 24 FMV infected cormels of cv. Marion. The results listed in Table 1 show that there was transmission of the causal agent of FLN from the soil indicating that the agent as well as the vector occurred in both the contaminated soil and the decomposing remains.

The infection rate appeared to be lower in the second experiment than in the first

Table 1. Transmission of freesia leaf necrosis in remains of freesia plants and in soil samples collected at nurseries with different soil treatments and at most of which, freesias have been grown.

Nursery	History of soil treatment	Percentage of freesia plants infected				
		1st experiment cv. Rose Marie		2nd experiment cv. Rose Marie		cv. Marion
		healthy	FMV[3]	healthy	FMV	FMV
A[1]	Freesias for 15 years; no soil sterilization	100	93	69	93	89
B[1]	Freesias for 7 years; no soil sterilization	75	73	0	7	5
C[1]	Last soil sterilization 2 years ago	63	70	33	42	40
D[1]	Last soil sterilization 1 year ago	0	0			
E[1]	No freesias grown	0	0			
F[1]	Last soil sterilization 2 years ago; soil collected around infected plants	91	83			
G[2]	No freesias grown for 12 years	10	0			
H[2]	Freesias for 5 years; no soil sterilization	20	7			
I[2]	No freesias grown	0	0			
J	Plant remains	70	71	5	10	9
Control	Sterilized potting soil	0	0	0	0	0

[1]At this nursery the freesias were grown in a glasshouse.
[2]At this nursery the freesias were grown in the open.
[3]FMV = freesia mosaic virus.

Tabel 1. Overdracht van bladnecrose bij knolfreesia in een monster van een freesia-afvalhoop en in zeven grondmonsters van freesia-bedrijven waar de grond op verschillende wijze was behandeld en twee grondmonsters van bedrijven, waar geen freesia's waren geteeld.

Table 2. Leaf necrosis infection in freesia plants grown at three water regimes and two temperatures.

Water regimes	Percentage of cv. Rose Marie plants infected			
	16°C		12°C	
Cormels used:	healthy	FMV[1]	healthy	FMV
wet	86	94	57	57
moist	72	84	41	28
dry	27	45	17	18
control (sterilised potting soil) moist	0	0	0	0

[1]FMV = freesia mosaic virus.

Tabel 2. De infectie van knolfreesia's, die in natte, normaal vochtige en droge grond stonden en bij twee verschillende temperaturen werden gekweekt.

one. This reduction in the ability to transmit or to infect might be explained by drying out of the soil during the course of the first experiment, which lasted 7 months.

All soil samples used were surveyed for nematodes. Only in soil from nursery I was one *Trichodorus pachydermis* specimen found. No *Xiphinema* nor *Longidorus* specimens were seen. *Olpidium brassicae* was found in freesia roots grown in soil samples from nurseries A, B and F (Table 1) and the soil used in Table 2. Tobacco necrosis virus could not be demonstrated in these soil samples using the test developed by Teakle (1962).

Finally, the relation between the infection rate and the moisture content of the soil was studied. The soil used for this experiment was collected at a nursery where freesias have been cultivated for several years. Three water regimes, viz. wet, moist (= normal condition) and dry (watered with 4:2:1 volumes respectively) were tested; a set of each was kept at 12 °C and 16 °C. Each treatment was made with 18 healthy and 30 FMV infected cormels with 3 per pot. Healthy and FMV infected cormels, planted in sterilized potting soil and watered as normal, served as controls. The harvested corms and cormels were prepared and indexed for infection (Table 2). It is clear that with an increase in moisture content of the soil the percentage of infected plants increased. The percentage of infected plants was higher at 16 °C than at 12 °C.

The results obtained show that an agent in the soil is necessary for infection of freesia with FLN. The mode of transmission is still unknown. Because a higher number of infected plants were obtained in moist and wet soils, water may be necessary for the movement and/or growth of the vector. Nematodes known to be vectors of plant viruses were not found in the soil samples studied.

It has been suggested that the causal agent of FLN has a viral etiology (Van Dorst, 1973). The survival of the infectious agent and also that of the vector in very different conditions, as in dried-out soil and decomposing plant material, points to a fungus-virus relationship. The agent infecting freesia resembles lettuce big vein virus in that it has not been isolated, neither observed in dip preparation nor transmitted by sap-inoculation (Grogan and Campbell, 1966), but has been transmitted through soil. Therefore one could expect the virus to survive in a resting stage of a fungus.

Acknowledgments

The author is greatly indebted to Dr D. Peters, Wageningen, for his help and advice; to Dr H. A. van Hoof, Wageningen, for the examination of the soil samples for nematodes and testing the soil for the occurrence of tobacco necrosis virus, and to Miss W. Th. Runia for her skilful assistance. Finally I thank Dr M. G. Schultz for his help in correcting the English text.

Samenvatting

Overdracht van freesiabladnecrose via grond

In een tweetal experimenten kon worden aangetoond dat de infectie van knolfreesia's plaatsvindt vanuit de grond (Tabel 1). Infectie treedt ook op als de knollen in verterende resten van freesiaplanten worden gepoot (Tabel 1). Een groter aantal zieke planten wordt gevonden wanneer de freesiaplanten worden geteeld in natte grond (Tabel 2). Temperatuur blijkt ook van invloed te zijn: bij 16 °C traden meer infecties op dan bij 12 °C. Omdat we het pathogeen nog steeds niet hebben kunnen isoleren en

mede gelet op de wijze van verspreiding, zou men mogen veronderstellen dat de verwekker van bladnecrose bij freesia veel gelijkenis vertoont met bobbelbladvirus bij sla.

References

Dorst, H. J. M. van, 1973. Two new disorders in freesias. Neth. J. Pl. Path. 79: 130–137.
Grogan, R. G. & Campbell, R. N., 1966. Fungi as vectors and hosts of viruses. A. Rev. Phytopath. 4: 29–52.
Teakle, D. S., 1962. Necrotic symptoms of tobacco necrosis virus in roots. Phytopathology 52: 1037–1040.

Address

Proefstation voor de Groenten- en Fruitteelt onder Glas, Zuidweg 38, Naaldwijk, the Netherlands.