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Accessions of Australian Nicotiana species suitable as indicator hosts in the diagnosis of plant virus diseases

P. VAN DIJK, F.A. VAN DER MEER[1] and P.G.M. PIRON

Research Institute for Plant Protection (IPO), P.O. Box 9060, 6700 GW Wageningen, the Netherlands

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Abstract

When screening the genus *Nicotiana* for sensitive and differential hosts for a group of mechanically transmissible plant viruses with narrow host ranges, development of systemic symptoms was almost exclusively observed in species of three closely related sections of the subgenus *Petunioides*. These species were *N. miersii* (section *Acuminatae*), *N. bigelovii* and *N. clevelandii* (*Bigelovianae*) and *N. benthamiana*, *N. cavicola*, *N. ingulba*, *N. occidentalis*, *N. rosulata* and *N. rotundifolia* (*Suaveolentes*). Except for *N. benthamiana* and *N. clevelandii*, which are already known for their large virus ranges, they are new experimental hosts that appeared very useful for detection of viruses and for differentiation of viruses that closely resemble each other in host range. Accessions of the same species often varied largely in local and systemic viral response. Especially *N. benthamiana*-9, *N. miersii*-33 and *N. occidentalis*-37B (code numbers given by Tobacco Research Laboratory, Oxford, N.C., USA) are recommended for routine inoculation tests. The sensitive *Suaveolentes* species mentioned are native to the arid parts of Australia. Collections of these species deserve attention in studies on virus diseases of unknown etiology where experimental hosts are lacking.

Additional keywords: apple chlorotic leaf spot virus, apple stem pitting, broad spectrum test plants, carrot mottle virus, parsnip mottle virus, parsnip yellow fleck virus, potato virus M, potato virus S.

Introduction

Test plants are indispensable for detection, differentiation, identification and propagation of plant viruses and their strains. Various species of the genus *Nicotiana*, especially *N. clevelandii*, *N. glutinosa*, *N. rustica* and *N. tabacum* proved very useful for these purposes (Hollings, 1966; Fulton, 1979). More recently, *N. benthamiana* was recommended because of its large virus range (Quacquarelli and Avgelis, 1975; Christie and Crawford, 1978). The latter species was found to be a prerequisite for the detection of the *Anthriscus* strain of parsnip yellow fleck virus in Umbelliferae with viral dieback, enabling the belated diagnosis of the disease (Van Dijk and Bos, 1985). The lack of sensitive and differential hosts can seriously hamper progress in research on plant virus diseases.

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[1] To whom correspondence should be addressed.

The *Nicotiana* species commonly used in plant virology (Fulton, 1979) represent only a small part of the 66 presently recognized species of the genus (Smith, 1979). Therefore, we explored the genus *Nicotiana* in order to find good experimental hosts for a group of viruses with relatively narrow host ranges. During this study the tendency appeared that certain sections of the genus contain more species with a large 'virus range' (Christie and Crawford, 1978) than other sections do. The prospects of this tendency in the search for sensitive experimental hosts are the subject of this paper. Details of reactions of individual viruses in new hosts will be published elsewhere.

Materials and methods

Accessions of *Nicotiana* species tested, were kindly supplied by Dr V.A. Sisson, Tobacco Research Laboratory (TRL), Oxford, N.C., USA. Other accessions tested were seed stocks normally used at the Research Institute for Plant Protection (IPO). The accessions are indicated by their TRL code number or by the suffix IPO (Table 1). Three additional accessions of *N. occidentalis* that we compared with TRL accessions, were provided by Dr C.E. Fribourg, Universidad Nacional Agraria, Lima, Peru (accession Lima) and by the Zentralinstitut für Genetik und Kulturpflanzenforschung, Gatersleben, DDR (accessions G436 and G472). The collection of *Nicotiana* species tested (Table 1) represents 63 of the 66 species recognized by Smith (1979). *N. noctiflora* and *N. ameghinoi* (section *Noctiflorae*) and *N. longibracteata* (*Acuminatae*) are not represented. Additional species of the collection are: *N. angustifolia* (*Acuminatae*) (Goodspeed, 1954); *N. palmeri* (*Trigonophyllae*), now considered to be equal to *N. trigonophylla*; *N. sanderae* (*Alatae*), originating as a hybrid between *N. forgetiana* and *N. alata* (Smith, 1979); *N. eastii* (*Suaveolentes*), believed to be a hybrid between *N. maritima* and *N. suaveolens* (Horton, 1981).

In Table 1 *Nicotiana* species are arranged according to genetical relationships (Smith, 1979; V.A. Sisson, personal communication). Further classification of accessions of species into varieties or subspecies (Goodspeed, 1954; Burbidge, 1960; Horton, 1981) was not always known and is therefore not considered.

When necessary to achieve germination, seeds of *Nicotiana* species were soaked for one day in gibberellic acid (GA_3, 10^{-4} % w/v) before sowing.

Test plants were grown in glasshouses at 18-22 °C with additional illumination (SON-T) in winter to extend the daylength to 16 h.

Viruses, strains and number of isolates are mentioned in Table 1. Inoculum source plants were: infected *N. occidentalis*-37B for A2817, infected *Chenopodium quinoa* for ACLSV, infected *N. benthamiana*-IPO for the three viruses of umbelliferous plants and infected potatoes for PVM and PVS.

Systemic infection in accessions of *Nicotiana* species was detected by observation of symptoms in comparison with healthy control plants, by back-inoculation to suitable assay hosts (*N. occidentalis*-37B for A2817, *Phaseolus vulgaris* cv. Bataaf for CMotV and *C. quinoa* for PMotV and PYFV), or by ELISA (ACLSV, PVM and PVS).

Table 1. Systemic response[1] of a collection of *Nicotiana* species after inoculation with viruses of little aggressiveness to the genus *Nicotiana*.

Subgenus Section Species-accession	virus							
	A2817[2]	ACLSV[3]	CMotV[4]	PMotV[5]	PYFV-A[6]	PYFV-P[7]	PVM[8]	PVS[9]
Subgenus *Rustica*								
Section *Paniculatae*								
N. glauca-23[10]	—^{x}	.	.	.	.	.	—	—
N. glauca-23A	—^{x}	.	.	.	.	.	—	—
N. glauca-23B	—^{x}	.	—^{x}	.	—^{x}	.	—	—
N. glauca-23C	—^{x}	.	.	.	.	.	—	—
N. paniculata-40	—^{x}	.	.	.	.	.	.	.
N. paniculata-40A	—^{x}	.	.	.	.	.	.	.
N. paniculata-40B	—^{x}	.	.	.	.	.	.	.
N. paniculata-40C	—^{x}	.	.	.	.	.	.	.
N. knightiana-27	—	—	.	.	—	.	.	.
N. solanifolia-52	—	—	.	.	—	.	.	.
N. benavidesii-8	—^{x}	—	—^{x}	.	—^{x}	.	—	—
N. cordifolia-15	—^{x}	.	—^{x}	.	—^{x}	.	—	—
N. cordifolia-15A	—^{x}	.	—^{x}	.	—^{x}	.	—	—
N. raimondii-45	—	—	.	.	—	.	.	.
Section *Thyrsiflorae*								
N. thyrsiflora-57	—^{x}	.	.	.	—^{x}	.	.	.
Section *Rusticae*								
N. rustica-IPO[11]	—	—	.	—	—^{x}	—	—	—
Subgenus *Tabacum*								
Section *Tomentosae*								
N. tomentosa-58	—^{x}	.	.	.	.	.	.	.
N. tomentosa-58A	—^{x}	.	.	.	.	.	.	.
N. tomentosiformis-59	—^{x}	—	.	.	—	.	.	.
N. otophora-38	—^{x}	.	.	.	.	.	.	.
N. otophora-38B	—^{x}	.	.	.	.	.	.	.
N. setchellii-51	—^{x}	.	—^{x}	.	.	.	.	.
N. glutinosa-24	—^{x}	—	—	.	—	—	—	—
N. glutinosa-24A	—^{x}	—	S	.	—	—	(s)	—
N. glutinosa-24B	—^{x}	—	—	.	—	—	—	—
N. glutinosa-IPO	—	—	—^{x}	—	—^{x}	—^{x}	—	—
N. kawakamii-72	—^{x}	.	—^{x}	—^{x}	—^{x}	—^{x}	.	.
Section *Genuinae*								
N. tabacum cv. White Burley-IPO	—	.	—^{x}	—	—	—	—	—
Subgenus *Petunioides*								
Section *Undulatae*								
N. undulata-61A	.	.	—^{x}	.	—^{x}	.	.	.
N. arentsii-6	—^{x}	.	—^{x}	.	—^{x}	.	—	—
N. wigandioides-63	—^{x}	.	.	.	.	.	.	.
Section *Trigonophyllae*								
N. trigonophylla-60	s	—	s	—^{x}	—	—	(s)	—
N. palmeri-39	—^{x}	.	.	.	.	.	.	.
Section *Alatae*								
N. sylvestris-56A	.	.	—^{x}	.	—^{x}	.	.	.
N. langsdorffii-28A	—^{x}	.	.	.	.	.	.	.

Table 1. Continued.

Subgenus Section Species-accession	virus							
	A2817	ACLSV	CMotV	PMotV	PYFV-A	PYFV-P	PVM	PVS
Subgenus *Petunioides* (continued)								
Section *Alatae* (continued)								
N. alata-3	—^{x}	—	—^{x}	.	—^{x}	.	(s)	(s)
N. alata-4	—^{x}	.	.	.	.	.	—	—
N. sanderae-50B	—^{x}	.	.	.	.	.	.	.
N. forgetiana-21A	—^{x}	.	—^{x}	.	—^{x}	.	—	—
N. forgetiana-21B	—^{x}	.	.	.	.	.	—	—
N. bonariensis-11	—^{x}	.	—^{x}	.	—^{x}	.	—	(s)
N. longiflora-30	—^{x}	.	.	.	.	.	.	.
N. longiflora-30B	—^{x}	.	.	.	.	.	.	.
N. longiflora-30C	—	.	.	.	.	.	.	.
N. plumbaginifolia-43A	s	s	.	.	—	.	.	.
N. plumbaginifolia-43B	s	s	.	.	.	.	.	.
N. plumbaginifolia-43C	s	s	.	.	.	.	.	.
Section *Repandae*								
N. repanda-46	—	.	S	—^{x}	—^{x}	—^{x}	.	.
N. stocktonii-54	—	—	.	.	—	.	.	.
N. nesophila-34A	—^{x}	.	.	.	.	.	.	.
Section *Noctiflorae*								
N. petunioides-42	—^{x}	.	.	.	.	.	.	.
N. acaulis-1	—^{x}	.	.	.	.	.	.	.
Section *Acuminatae*								
N. angustifolia-5	—^{x}	—	—^{x}	.	—^{x}	.	—	—
N. acuminata-2	—^{x}	—	—^{x}	.	—^{x}	.	—	—
N. acuminata-2A	—^{x}	.	.	.	.	.	—	—
N. pauciflora-41	—^{x}	.	.	.	.	.	.	.
N. attenuata-7	—^{x}	.	S	.	—^{x}	.	—	—
N. miersii-33	s	(s)	s	S	S	S	S	(s)
N. corymbosa-16	—^{x}	—	—	—^{x}	—^{x}	s	—	—
N. linearis-29	—	—	s	—^{x}	—	s	s	(s)
N. spegazzinii-70	—^{x}	.	.	.	.	.	.	.
Section *Bigelovianae*								
N. bigelovii-10	—	—	S	—	s	(S)	—	—
N. bigelovii-12	—	—	S	s	(S)	(S)	—	—
N. bigelovii-13	—	—	.	s	S	(S)	—	—
N. clevelandii-14	—^{x}	—	S	.	—^{x}	.	—	—
N. clevelandii-IPO	—	—	S	(S)	(s)	S	—	—
Section *Nudicaules*								
N. nudicaulis-36	—^{x}	.	—^{x}	—^{x}	—^{x}	—^{x}	.	.
Section *Suaveolentes*								
N. benthamiana-9	—^{x}	—	S	S	S	S	—	—
N. benthamiana-9A	—^{x}	—	S	S	S	S	—	—
N. benthamiana-IPO	—	—	S	S	S	S	—	—
N. umbratica-69	—	—	S	—	(S)	—^{x}	—	—
N. cavicola-68	—	—	S	S	S	S	—	—
N. debneyi-17	—	—	—^{x}	—^{x}	—^{x}	—^{x}	(s)	(s)
N. debneyi-IPO	—	—	—	—^{x}	—^{x}	s	—	(s)
N. gossei-26	.	—	.	—^{x}	—^{x}	—^{x}	—	—
N. amplexicaulis-65	—	—	.	—^{x}	—^{x}	—^{x}	—	—
N. amplexicaulis-65A	.	.	.	—^{x}	—	—	—	—

Table 1. Continued.

Subgenus Section Species-accession	virus							
	A2817	ACLSV	CMotV	PMotV	PYFV-A	PYFV-P	PVM	PVS
Subgenus *Petunioides* (continued)								
Section *Suaveolentes* (continued)								
N. maritima-31	—	.	s	—^{x}	—^{x}	—^{x}	.	.
N. velutina-62	—	—	—	—^{x}	—^{x}	—^{x}	—	—
N. velutina-62A	.	—	S	—^{x}	—^{x}	—	(s)	(s)
N. velutina-62B	.	—	—	—^{x}	—	—	(s)	(s)
N. hesperis-67	—	—	—	—	—^{x}	—	—	—
N. hesperis-67A	—	—	—	(S)	—	S	(S)	—
N. occidentalis-37	—	—	—	—^{x}	—	s	(s)	—
N. occidentalis-37A	s	—	—	—^{x}	—	s	s	(s)
N. occidentalis-37B	S	S	S	(S)	(S)	S	s	(s)
N. occidentalis-G436[12]	—	—	.	.	.	.	.	.
N. occidentalis-G472	S	S	S	.	—^{x}	S	.	.
N. occidentalis-Lima[13]	—	—	.	.	.	.	.	.
N. simulans-66	—	—	.	—^{x}	—^{x}	—^{x}	—	—
N. megalosiphon-32	—	.	.	.	—	(S)	.	.
N. megalosiphon-IPO	—	—	(S)	—	—^{x}	(S)	(s)	—
N. rotundifolia-47	—	—	—	(S)	S	S	(s)	(s)
N. rotundifolia-47A	s	s	—	—^{x}	S	S	(s)	(s)
N. excelsior-19	—	—	—^{x}	—^{x}	—^{x}	—^{x}	(s)	(s)
N. excelsior-19A	—^{x}	—	(S)	—^{x}	—	—	s	(s)
N. eastii-18	—^{x}	—	—^{x}	—^{x}	—^{x}	s	—	—
N. suaveolens-55	—	—	.	—^{x}	—^{x}	—^{x}	—	—
N. suaveolens-55A	.	—	—	—^{x}	—^{x}	s	—	—
N. suaveolens-55B	.	—	—	—^{x}	—	s	—	—
N. suaveolens-55C	—	—	—^{x}	—^{x}	—^{x}	—	—	—
N. ingulba-64	—	—	S	(S)	S	S	—	—
N. exigua-20	—	—	—^{x}	—^{x}	—^{x}	—^{x}	(s)	—
N. exigua-20A	—^{x}	—	s	—^{x}	S	s	—	—
N. goodspeedii-25	—	—	.	—^{x}	—^{x}	—^{x}	—	—
N. rosulata-53	—	—	S	—^{x}	S	S	(S)	(s)
N. rosulata-53A	—^{x}	—	S	S	S	S	(S)	(s)
N. fragrans-22	—	.	—	—^{x}	—^{x}	—	—	—
N. africana-71	—^{x}	.	—^{x}	—^{x}	—^{x}	—^{x}	.	.

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[1] Symbols: . = not tested; — = systemic immunity; —^{x} = no systemic symptoms but not tested for systemic infection; s = symptomless systemic infection; S = systemic symptoms; ( ) = symbols between parentheses: not all isolates induced the reaction indicated.

[2] Unidentified virus isolate associated with apple stem pitting (Van der Meer, 1986).

[3] Isolate of apple chlorotic leaf spot virus (Lister, 1970).

[4] Three isolates of carrot mottle virus (Murant, 1974b).

[5] Three isolates of parsnip mottle virus (Watson et al., 1964).

[6] Three isolates of parsnip yellow fleck virus-*Anthriscus* strain (Murant, 1974a).

[7] Three isolates of parsnip yellow fleck virus-parsnip strain (Murant, 1974a).

[8] Six isolates of potato virus M (Wetter, 1972).

[9] Six isolates of potato virus S (Wetter, 1971).

[10] Code numbers from Tobacco Research Laboratory.

[11] IPO: Seed stock from Research Institute for Plant Protection.

[12] Code numbers preceded by 'G' from Zentralinstitut für Genetik und Kulturpflanzenforschung.

[13] Seed stock from Universidad Nacional Agraria.

Results

The results given in Table 1 and summarized in Table 2 show that of the accession-virus combinations tested, the percentage of combinations resulting in systemic symptoms, as well as the number of viruses involved, were much higher in the closely related sections *Acuminatae*, *Bigelovianae* and *Suaveolentes* of the subgenus *Petunioides* (Goodspeed, 1954) than in the remaining sections of the genus *Nicotiana*. The same, although to much lesser extent, is true when the latent systemic invasion of the viruses is included.

Different accessions of the same species often reacted differently to inoculation with one or more of the viruses applied (Table 1). The most extreme differences were detected between the accessions 37 and 37B of *N. occidentalis* (section *Suaveolentes*). While the latter appeared susceptible to all eight viruses and strains tested and developed symptoms with six of them, the former appeared immune to at least five of the viruses and strains and did not produce symtoms with the other three (Fig. 1). Because of retarded symptom expression (not mentioned in Table 1) *N. benthamiana*-9A (*Suaveolentes*) appeared a less convenient indicator than

Fig. 1. Different susceptibility of the morphologically rather similar *Nicotiana occidentalis* accessions 37 (left) and 37B (middle and right). Accession 37B is very susceptible and sensitive to apple chlorotic leaf spot virus (right) whereas 37 is immune. Healthy 37B plant in the middle.

Table 2. Cumulated systemic response of accessions of different sections and species of the genus *Nicotiana* after inoculation with viruses mentioned in Table 1.

Section(s) or species[1]	Number of accessions	Accession/virus combinations tested (%)	Systemic infection[2]		Systemic symptoms	
			s+S[3] (%)	number of viruses	S (%)	number of viruses
Acuminatae, *Bigelovianae* and *Suaveolentes*	54	85	37	8	18	7
Remaining sections	53	66	15	5	1	1
Acuminatae	9	64	42	8	11	5
Bigelovianae	5	93	41	4	30	4
Suaveolentes	40	89	36	8	18	7
Species from arid parts of Australia[4]	23	92	46	8	28	7
Remaining species of *Suaveolentes*	17	85	20	5	3	3

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[1] Data summarized per section or species from Table 1. *N. benthamiana*-IPO and *N. occidentalis*-Lima are not included since they proved to be identical to the accessions *N. benthamiana*-9 and *N. occidentalis*-37, respectively.

[2] Of the combinations not developing systemic symptoms, the number of combinations resulting in latent infection was estimated from the proportion of combinations that proved to be infected when tested.

[3] s = Symptomless systemic infection; S = systemic symptoms.

[4] The following species of the *Suaveolentes* section that are restricted to central and western-Australia (Burbidge, 1960): *N. benthamiana*, *N. umbratica*, *N. cavicola*, *N. gossei*, *N. velutina*, *N. hesperis*, *N. occidentalis*, *N. simulans*, *N. rotundifolia*, *N. excelsior*, *N. ingulba* and *N. rosulata*.

N. benthamiana-9 and -IPO, the latter two being identical in viral response as well as in morphology. Within most species differences in reactions to viruses between accessions, were associated with clear morphological differences. Within some species, however, accessions differing in viral response showed little or no morphological differences (Figs 1 and 2).

From the data in Table 1 it appears that accessions of *N. miersii* (*Acuminatae*), *N. bigelovii* and *N. clevelandii* (*Bigelovianae*) and *N. benthamiana*, *N. cavicola*, *N. occidentalis*, *N. rotundifolia*, *N. ingulba* and *N. rosulata* (*Suaveolentes*) were susceptible to at least 4 out of 8 viruses and strains applied. *N. clevelandii* and *N. benthamiana* are known broad spectrum test plants. Of the remaining species listed, only *N. occidentalis* has been sporadically mentioned before in literature (Paulsen and Fulton, 1968; Fernandez-Northcote, 1983; Horváth, 1983). However, these authors evidently did not use the sensitive accession 37B. The seeds of *N. occidentalis* which we received from C.E. Fribourg (accession Lima), were originally obtained from R.W. Fulton (Paulsen and Fulton, 1968) and the same seed lot was used by E.N. Fernandez-Northcote (personal communication). Plants which we obtained from these seeds, were morphologically similar to the TRL accession 37 and shared immunity to both apple viruses with this accession (Table 1). J. Horváth (personal communication) obtained his seeds from the Zentralinstitut für Genetik und Kulturpflanzenforschung, Gatersleben, DDR. Plants grown from both seed lots that we obtained from that institute, clearly differed in appearance from all three TRL accessions. One of the accessions (G472) was sensitive to both apple viruses whereas the other (G436) appeared immune to these viruses (Table 1).

With most accessions reacting with systemic symptoms, each virus could be recognized by its own typical symptoms. The two apple viruses for instance, which closely resembled each other in host range (Table 1), could easily be distinguished because A2817 induced a typical vein clearing and vein mosaic in *N. occidentalis*-37B, whereas ACLSV caused a severe mosaic and leaf distortion in the same host. *N. miersii*-33 turned out to be a differential host for the related carlaviruses PVM and PVS (Koenig, 1982), since all PVM isolates induced systemic stunting, necrosis and leaf malformation in this accession, while none of the PVS isolates caused any symptoms in it (Fig. 3). PYFV-A could be distinguished from the related PYFV-P because only the latter induced prominent systemic symptoms in *N. clevelandii*-IPO.

The local lesion response of some accessions, not mentioned in Table 1 for reasons of surveyability, was also useful for differentation of some viruses. For example, PMotV could be distinguished from PYFV because only the latter caused local lesions in *N. benthamiana*-9 or -IPO. Most isolates of PVM caused local lesions in *N. hesperis*-67A, *N. rosulata*-53 (Fig. 2) and *N. rotundifolia*-47A, while PVS isolates did not. *N. hesperis*-67A developed local lesions when inoculated with A2817 virus whereas ACLSV did not infect this accession, thus offering the possibility to separate A2817 virus from mixtures of A2817 virus and ACLSV.

With all viruses represented by more than one isolate, several accessions were infected by some, but not by all isolates. In the same way accessions often reacted with symptoms to some, but not to all isolates of a given virus. On the one hand this behaviour is an advantage because it enables to distinguish between isolates of a virus and offers possibilities to separate them from mixtures. On the other hand, however, it means that the accessions concerned are not suitable as indicators in routine testing.

Fig. 2. Different effects of a potato virus M variant on inoculated leaves (bottom row) and systemically infected leaves (top row) of the morphologically very similar *Nicotiana rosulata* accessions 53 (left) and 53A (right). Accession 53 reacts with clear local and systemic symptoms whereas 53A shows only very faint systemic symptoms.

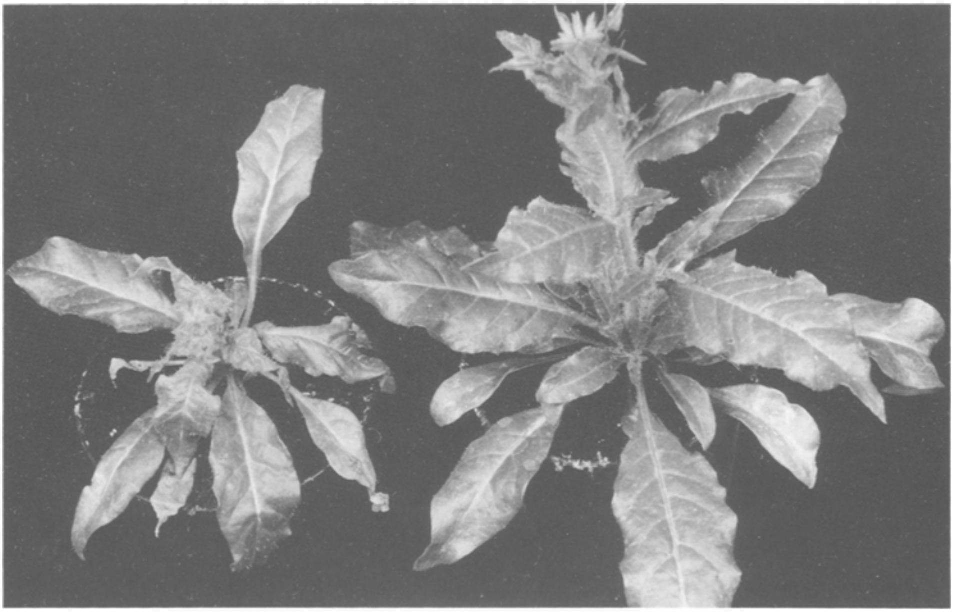

Fig. 3. *Nicotiana miersii* accession 33, a differential indicator for potato virus M (PVM) and potato virus S (PVS). Left: plant inoculated with PVM and showing stunting, necrosis and leaf malformations. Right: plant inoculated with PVS and systemically infected but not showing symptoms.

Seeds of the sensitive accessions generally germinated well and plants were easy to grow. Especially the sensitive *N. occidentalis* accessions 37B and G472 are of a suitable type with large smooth leaves that can easily be inoculated. However, slow growth and narrow leaves are unfavourable characteristics of *N. miersii*-33. Accessions of *N. bigelovii*, *N. cavicola* and *N. ingulba* are less useful because of aspecific symptoms.

Especially *N. benthamiana*-9, *N. miersii*-33 and *N. occidentalis*-37B seem to be suitable for routine inoculation tests because of their large virus ranges and sensitivity to different viruses.

Discussion and conclusions

Although during the course of our research most attention became directed to the section *Suaveolentes*, and thus no equal numbers of species of other sections were tested, our results indicate that susceptibility and sensitivity to viruses is more common in the closely related sections *Acuminatae*, *Bigelovianae* and *Suaveolentes*, than in the remaining part of the genus *Nicotiana*. In this respect our findings are in line with the results of Sievert (1972) who investigated largely the same collection of *Nicotiana* species and detected only one potato virus Y-resistant accession in those three sections, whereas 13 resistant accessions were found in the remaining sections of the genus.

Especially the rather big *Suaveolentes* section appears to contain some species of which particular accessions may be expected to be sensitive to a broad spectrum of viruses. This section comprises all 19 native Australian species (Burbidge, 1960) as well as *N. fragrans* and *N. africana* that are restricted to some South Pacific islands and Africa, respectively (Goodspeed, 1954; Smith, 1979; V.A. Sisson, personal communication). Surprisingly, the six species that we found especially sensitive to our viruses, are all from the arid western and inland areas of the continent (Burbidge, 1960). The division of the *Suaveolentes* section into one part consisting of species that are restricted to arid parts of Australia, and a second part with the remaining species, reveals that sensitivity to our viruses almost exclusively occurs in the former part of the section (Table 2).

One might speculate that there is a connection between the virus sensitivity of the western and inland species and their evolutionary history. These species are believed to have developed from *N. debneyi*, *N. fragrans* and *N. suaveolentes* as a result of hybridization and introgression, followed by geographical isolation when inland areas in the latest geological period gradually became desiccated (Goodspeed, 1954). In their isolated territories they may not have been challenged sufficiently by viruses to develop resistance. In this respect it is interesting that until recently there have been discussions as to whether or not plant viruses were present in Australia before the European settlement (White, 1973; Gibbs and Guy, 1979; Guy and Gibbs, 1985). It would be interesting to investigate Australian species of other cosmopolitan genera like *Solanum* and *Chenopodium* for the phenomenon discovered in *Nicotiana.*

Test plants that are susceptible and sensitive to a large range of viruses may be very useful when studying and comparing viruses with a restricted host range, and for studies on virus diseases of unknown etiology such as those of woody hosts. For the causal viruses of the latter, good herbaceous hosts are a prerequisite for propagation, identification and purification and may also be convenient indicator plants in routine indexing of selected stock material (Fulton, 1966; Van der Meer, 1982). Fulton (1966) stated: 'There seems to be no way to predict which herbaceous species will be susceptible to a virus infecting a woody host'. However, the results of our study focus the attention on a number of *Nicotiana* species indigenous to the arid parts of Australia. With regards to the large intraspecific differences in sensitivity that we observed, it seems most promising to concentrate on collections of those species when new virus indicators are needed.

Accurate identification and separate propagation of sensitive lines are of great importance. This has earlier been mentioned by Van der Want et al. (1975) who detected important differences in sensitivity between sources of *N. glutinosa*, *N. rustica* and *N. tabacum* cv. White Burley, used in different laboratories.

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Samenvatting

Collectienummers van Australische Nicotiana-soorten geschikt als toetsplant bij de diagnostiek van virusziekten van planten

In het genus *Nicotiana* werden vatbare, gevoelige en differentiële toetsplanten opgespoord door toetsing van vooral de collectie van het Tobacco Research Laboratory (Oxford, N.C., VS) met een aantal virussen waarvoor weinig of geen bruikbare toetsplanten bekend waren. Systemische reacties werden bijna uitsluitend in soorten van drie nauw verwante secties van het subgenus *Petunioides* waargenomen. Deze soorten waren *N. miersii* (sectie *Acuminatae*), *N. bigelovii* en *N. clevelandii* (*Bigelovianae*) en *N. benthamiana*, *N. cavicola*, *N. ingulba*, *N. occidentalis*, *N. rosulata* en *N. rotundifolia* (*Suaveolentes*). Behalve voor virusvermeerdering bleken deze soorten ook zeer geschikt te zijn voor differentiatie van virussen of virusstammen met een bijna gelijke waardplantenreeks.

Collectienummers van één en dezelfde soort konden sterk in lokale en systemische reactie verschillen. Vooral *N. benthamiana*-9, *N. miersii*-33 en *N. occidentalis*-37B kunnen voor routine-inoculaties worden aanbevolen. Met uitzondering van *N. benthamiana* en *N. clevelandii* zijn de genoemde soorten of genotypen nieuwe experimentele waardplanten voor de virologie. De soorten die van de sectie *Suaveolentes* worden genoemd, zijn inheems in de woestijnachtige gebieden van Australië. Verzamelingen daarvan verdienen nadere aandacht bij de diagnostiek van virusziekten waarvoor nog geen experimentele waarplanten voorhanden zijn.

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