Host Reactions of Mechanically Transmissible Legume Viruses of the Northern Temperate Zone

R. Hampton, L. Beczner, D. Hagedorn, L. Bos,
T. Inouye, O. Barnett, M. Musil, and J. Meiners

Authors are members of the International Working Group on Legume Viruses: respectively, Research Plant Pathologist, Agricultural Research Service, U.S. Department of Agriculture, Oregon State University, Corvallis, OR 97331; Plant Virologist, Research Institute of Plant Protection, Budapest, Hungary; Professor, Department of Plant Pathology, University of Wisconsin, Madison, WI 53706; Virus Research Officer, Institute of Phytopathological Research, Wageningen, The Netherlands; Plant Virologist, College of Agriculture, University of Osaka Prefecture, Sakai, Osaka, 591, Japan; Associate Professor, Department of Plant Pathology and Physiology, Clemson University, Clemson, SC 29631; Plant Virologist, Institute of Virology, Slovak Academy of Sciences, Bratislava 9, Czechoslovakia; and Chief, Applied Plant Pathology Laboratory, Agricultural Research Service, U.S. Department of Agriculture, Beltsville, MD 29795.

Contribution of The Agricultural Research Service, U.S. Department of Agriculture, in cooperation with the Agricultural Experiment Station, Oregon State University, Corvallis. Technical Paper 4594 of the latter.

Accepted for publication 30 December 1977.

ABSTRACT

HAMPTON, R., L. BECZNER, D. HAGEDORN, L. BOS, T. INOUYE, O. BARNETT, M. MUSIL, and J. MEINERS. 1978. Host reactions of mechanically transmissible legume viruses of the northern temperate zone. Phytopathology 68: 989-997.

Reactions of 23 plant hosts to inoculation with each of 38 legume viruses, and a key to aid in the diagnosis of these viruses, are presented. The virus isolates, investigated at locations where they are indigenous, were either type cultures or had been equivalently characterized. Each of the hosts was derived from standardized seedstocks. Therefore, each investigator tested specific viruses against identical host germplasm. The key is based on some 1,700 symtomatological data and virus host ranges obtained from selected plant hosts. The final key formulation was derived from evaluations of all possible combinations and hierarchies of plant hosts. Assuming that each virus will infect and induce characteristic symptoms when inoculated into defined cultivars under suitable conditions, the key is an aid to identification for each of the 38 viruses tested. Evaluations of two to four separate isolates of certain viruses supported the diagnostic value of the key.

The diverse plant family, Leguminosae, includes at least 35 species cultivated for protein-rich food, feed and forage, and green manure. Crop losses in *Phaseolus* beans due to virus infection were reported as early as 1917 (8), and the destructiveness of virus-induced legume diseases has become increasingly apparent since that time. Weiss (11) pioneered in classifying viruses infectious to legumes by host range and symptomatology, and later (12) compiled host relations for viruses described primarily on leguminous crops. Bos and van der Want (3) proposed research on legume viruses collectively. Bos et al. (2) suggested standardized procedures for identifying these viruses, and later Bos (1) published an updated compendium of viruses naturally infectious to leguminous plants.

Over the past 25 yr, an era of increasing emphasis on controlling virus-induced diseases of *Phaseolus* and *Pisum* crops in the USA , Europe, and Japan, new technology facilitating virus purification and characterization has dramatically enhanced plant virus identification. Although there has been a strong emphasis recently on intrinsic viral properties, it now is being recognized increasingly that viral genetics are significantly expressed as observable virus interactions with specific hosts. During this same era, several workers independently have developed sets of host differentials for diagnosing certain indigenous legume viruses. Diagnostic information, however, usually has been published only for individual viruses, sometimes communicated comprehensively only among colleagues, and with a few exceptions (2, 4, 5) has lacked standardization or international scope.

The International Working Group on Legume Viruses was established in 1962 to promote international comcumication and cooperation among plant pathologists working with legume viruses and to standardize identification criteria and nomenclature for these viruses. This working group recently published a revised list of host plants proven valuable for definitive work with legume viruses (9). We recognize, however, that significant differences in virus sensitivity and growth habit exist among selections of virus indicator plants (10), and that widely applicable information on virus-host interactions is dependent upon standardized test-plant seedstocks. A repository of reference seedstocks (6), including some 93 plant types of the Plant Virus Indicator Collection (7), now has been established at Fort Collins, CO, USA (see Materials and Methods, *Plant hosts*).

The present study was undertaken to standardize host ranges and reactions for type cultures of important

00032-949X/78/000 173$03.00/0

legume viruses of the northern temperate zone on uniform plant hosts, and to formulate a key to aid in the diagnosis of these viruses. We recognize that current diagnostic procedures typically consist of the development of preliminary host-reaction information, determination of virus particle morphology, and serological identification. When ambiguities or new viruses are encountered during such information development, however, new or reinforcing lines of evidence are required. Host reactions developed by this investigation are intended as preliminary guidelines for workers with limited laboratory facilities or as supplementary information for well-equipped workers. We expect users of our key to depend, for final virus identification, upon confirmatory ultrastructural, serological, and, when necessary, physico-chemical properties of virus isolates.

MATERIALS AND METHODS

Plant hosts.—The 23 plant hosts tested in this study are described in Table 1. They comprise 19 species, 17 genera,

TABLE 1. Uniform plant hosts that were tested against 38 legume virus isolates.

Host species	Cultivar/Selection	NSSL Accession no.[a]
Antirrhinum majus L.	Mixed Colors (Tetra Giant Ruffled Mixed)	92506
Chenopodium amaranticolor Coste & Reyn.	Corvallis strain	93611
Cucumis sativus L.	Chicago Pickling	92507
Datura stramonium L.	R. Fulton strain	93613
Glycine max (L.) Merr. I	Bragg	90508
Glycine max II	Davis	90509
Gomphrena globosa L.	A. F. Ross strain	93612
Lycopersicon esculentum Mill.	Marglobe	92508
Medicago sativa L.	DuPuits	92509
Nicotiana glutinosa L.	Corvallis strain	91533
Nicotiana tabacum L.	Samsun NN	90496
Petunia hybrida Hort. Vilm.-Andr.	King Henry	96919
Phaseolus vulgaris L. I	Bountiful	89342
Phaseolus vulgaris II	Black Turtle Soup	92518
Phaseolus vulgaris III	Pinto III	89343
Phlox drummondii Hook.	Tall Mixed Colors	92510
Pisum sativum L. I	Perfected Wales	92511
Pisum sativum II	Dark Skin Perfection	92512
Spinacea oleracea L.	Bloomsdale Long Standing	92513
Trifolium pratense L.	Kenland	92514
Trifolium repens L.	New Zealand	92515
Vicia faba L.	(minor) Bell bean	92516
Vigna unguiculata (L.) Walp.	California Cowpea #5 (Early Ramshorn, California Blackeye #5)	92517

[a] Seedlot accession numbers for reference samples kept at the National Seed Storage Laboratory, Fort Collins, Colorado 80521, U.S.A.

and seven plant families, and were chosen for documented value in detecting or distinguishing certain viruses. In a few cases (*Antirrhinum, Petunia, Phlox*) they were selected merely on the basis of commercial availability of seed representing standardized host phenotypes. Seedlots of the plant hosts were gathered at one location, subdivided into eight equal quantities for distribution to the eight investigators and into a reserve supply. Thus, each investigator tested documented isolates of viruses on plant host germplasm identical to

TABLE 2. Legume virus isolates tested against 23 uniform plant hosts.

Virus	Selected synonym	Isolate/Cooperator
Alfalfa Mosaic	Lucerne mosaic virus	Necrotic strain, Beczner / (2,5,8)[a]
Adzuki Bean Mosaic		AB-S / (6)
Bean Common Mosaic	*Phaseolus* virus 1	Westlandia strain (= type strain)/ (3)
Bean Pod Mottle		Beltsville strain / (7)
Bean Southern Mosaic	Bean mosaic virus 4	Beltsville strain / (7)
Bean Yellow Mosaic	*Phaseolus* virus 2	B-25 strain, Bos / (3,5,8)
Bean Yellow Severe Mosaic		Gil-6 strain, Hampton / (5)
Broadbean Wilt	P.O. pea streak virus	Beczner isolate / (2)
Clover Blotch		CBV 1 isolate / (8)
Clover Yellow Mosaic	Pea mottle virus	Casper strain / (2)
Clover Yellow Vein	Pea necrosis virus	ATCC PV-123 / (1)
Clover (Red) Mottle		TpM 25 isolate / (8)
Clover (Red) Necrotic Mosaic		TpM 34 isolate / (8)
Clover (Red) Vein Mosaic	Wisconsin pea stunt virus	RK 31, Hagedorn / (3,4,8)
Clover (White) Mosaic	*Trifolium* virus 1	Wageningen strain, Bos / (2,5,8)
Cowpea Aphidborne Mosaic	Cowpea common mosaic virus	Florida strain, Purcifull & Zettler / (1)
Cowpea Chlorotic Mottle		Type culture, Kuhn / (1)
Cowpea Mosaic	Cowpea yellow mosaic virus	Yellow strain, Agrawal / (1)
Cucumber Mosaic	Cucumber virus 1	P 146 / (6)
Desmodium Yellow Mottle		Type culture, Scott / (1)
Lettuce Mosaic	*Lactuca* virus 1	69-053 / (6)
Pea Dwarf Mosaic	(Related to broadbean wilt)	P 108 / (6)
Pea Early Browning	Vroege-verbruiningsvirus van erwt	E 116, Bos / (3)
Pea Enation Mosaic	*Pisum* virus 1	Madison strain, Hagedorn / (4)
Pea Seedborne Mosaic	Pea leafroll mosaic virus	C 4-26 / (5,4)
Pea Streak	Sweet clover virus	Wisconsin strain, Hagedorn / (3,4,5)
Peanut Mottle	Groundnut mottle virus	M 2, Kuhn / (1)
Peanut Stunt	Groundnut stunt virus	Raleigh strain, Hebert / (1)
Soybean Mild Mosaic		SV-115, Takahashi / (6)
Soybean Mosaic	Soybean virus 1	Madison strain, Hagedorn / (4)
Soybean Stunt		SSV-A, Takahashi / (6)
Tobacco Ringspot	*Nicotiana* virus 12	Demski isolate / (2)
Tobacco Streak	*Nicotiana* virus 8	Red node strain, Beltsville / (7)
Tomato Black Ring	Lettuce ringspot virus	Schmelzer isolate / (2)
Tomato Spotted Wilt	*Lycopersicum* virus 3	Corvallis strain / (5)
Turnip Mosaic	Cabbage black ring virus	PN-9 / (6)
Watermelon Mosaic	Muskmelon mosaic virus	Molnár isolate / (2)
Wisteria Vein Mosaic		Ws 2B, Bos / (3)

[a] Numerical reference to cooperators testing viruses for this study. Strain designated was selected as representative; results presented in Table 1 were contributed by first designated cooperator. Cooperator reference numbers are as follows: Barnett (1), Beczner (2), Bos (3), Hagedorn (4), Hampton (5), Inouye (6), Meiners (7), and Musil (8). Results presented in Table 3.

Table 3. Symptoms induced in 23 plant hosts by 38 legume virus isolates

LEGEND:

Local Symptoms
Systemic Sympt.

HOST PLANT	Alfalfa Mosaic	Azuki Bean Mosaic	Bean Common Mosaic	Bean Pod Mottle	Bean Southern Mosaic	Bean Yellow Mosaic	Bean Yellow Severe Mosaic	Broadbean Wilt	Clover Blotch	Clover Yellow Mosaic	Clover Yellow Vein	Clover (Red) Mottle	Clover (Red) Necrotic Mosaic
Antirr. majus	ℓ[a] Mo,Ma	ℓ -*	- -*	- -	- -	- -	- -	0 0	- -	ℓ Mo	ℓ -	- -	- -
Chen. amarant.	LL Mo,Ma	-* -*	- -*	- -	- -	NS Mo,LC,N	LL$_n$ -	LL$_c$ Mo,Ma,N	Chl VChl	LL$_n$ Mo	LL$_n$ (VB)	- -	LL$_n$ -
Cuc. sativus	LL$_c$ VC,Mo	- -	- -*	- -	- -	- -*	- -	- -	Chl Chl	ℓ s	- -	- -	LL$_c$ -
Dat. stramon.	LL VC,Mo Ma	-* -*	- -*	- -	- -	- -*	- -	LL,RS Mo	- -	ℓ Mo	- -	- -	? -
Glyc. max I	LL Mo	- -	ℓ -	-* Mo	LL$_c$ Mo	? VB,VC Mo	LL$_n$ Mo,NS	LL$_c$ Mo	- Chl	ℓ Mo,VC	- -	LL$_n$,RS$_n$ VChl,Chl	LL$_n$ Chl
Glyc. max II	LL W,N	- -	? -	-* Mo,Stu	LL$_c$ Mo	? VN,LC,Mo,Stu	- -	LL$_c$ Mo	- Chl	ℓ Mo,VC	- -	- VChl,Ma	LL$_n$ Chl
Gomph. glob.	RS$_n$ Mo	- -*	ℓ -	- -	- -	? -*	- -	LL$_n$ LC,Mo	(LL$_n$) -	LL$_n$ s	- -	- -	LL$_n$ -
Lycop. escul.	LL Mo,NS	-* -*	- -*	- -	- -	- -*	- -	ℓ s	- (VC)	ℓ -	- -	- -	- -
Medic. sativa	ℓ s	-* -*	- -	- -	- -	- -	- -	- -	- Mo	LL$_n$ Mo	ℓ Mo,RSc	- Mo	- -
Nicot. glut.	LL Mo	- -*	0 0	- -	- -	0 0	- -	LL$_c$ Mo,RS	RS$_n$ Mo	ℓ -	- -	- -	RS$_n$ (s)
Nicot. tab.	- Mo,Ma	- -*	- -*	- -	- -	ℓ -	LL$_n$ -*	LL$_n$,RS Mo	RSn Mo	- -	RSc,NS -	- -	RS$_n$ (s)
Petunia hyb.	LL VC,Mo	-* -*	0 0	- -	- -	0 0	- -	LL$_c$ VC,Mo	(RS$_n$) Mo	- -	- -	- ?	RS$_n$ (s)
Phas. vulg. I	LL -	LL -	LL$_c$ Mo,Ma	LL$_n$,VN Mo	LL$_c$ Mo	LL$_c$ Mo,Ma,Stu	LL$_n$ Ep,N	LL$_n$ -	ChlS Mo,Ma	ℓ s	LL$_c$ ChlS	LL$_n$,Chl -	LL$_n$,VN N
Phas. vulg. II	LL -	- -	LL$_c$ Mo	RS$_n$,LC s	- Mo	VN VN,Mo,Ma,Stu	LL$_n$ Ep,N	LL$_n$, -	ChlS Mo,Ma	ℓ Mo	LL$_c$ Mo,Stu	LL$_n$,VN -	LL$_n$,VN N
Phas. vulg. III	LL -	LL -	- -	LL$_n$,LC,VN s	LL$_n$,VN s	ℓ ChlS Mo,Stu	LL$_n$ Ep,N	LL$_n$ -	VN,VB Mo,Ma	ℓ s	ℓ ChlS	LL$_n$,VN -	Chl,VN N
Phlox drum.	RS,LL Mo,Str	0 0	- -*	- -	- -	- -	- -	0 0	- -	ℓ -	- -	- -	- -
Pisum sat. I	LL Mo	- -	- -	- -	- s	- -*	- -	ℓ Mo,Stu	- Mo,N	LL$_c$ VC,Mo	LL$_c$ Chl	LL$_n$ Chl,Stu	LL$_n$ Str
Pisum sat. II	NS VC,Mo	- -	- -	- s	- -	- -*	- -	LL$_n$ Mo,Stu	- Mo,N	LL$_c$ VC,Mo	- -	- Stu,N	LL$_n$,VN Str
Spin. oler.	LL -	-* -*	- -*	- -	- -	ℓ -	LL$_n$ -*	ℓ Mo,Stu	ℓ(ChlS) -	LL$_c$ Mo,LR,Stu	- -	- -	ChlS -
Trifol. prat.	ℓ Mo,Ma	-* -*	- -	- -	- -	ℓ s	LL$_n$,N -	- -	- Mo	LL$_c$ Mo	- -	- Mo	ℓ Mo,Ma
Trifol. rep.	ℓ Mo,Ma	-* -*	- -	- -	- -	- -	ℓ s	- -	- s	ℓ Mo	ℓ Mo	- ?	ℓ Mo
Vicia faba	LL Str,N	- -*	- -	- -	- s	ℓ VC,Mo	LL$_n$,N NS,N	ℓ Mo	- Mo	LL$_n$ VC,Mo (NS)	LL$_n$ Mo, Stu, RS$_n$	NS N	(NS) Chl
Vigna unguic.	LL -	LL Mo	ℓ Mo	- s	- s	ℓ s	- -	ℓ VC,Mo	ChlS,LL$_n$ Mo,Ma	ℓ -	- -	RSn -	LL$_n$,VN Chl

TABLE 3, Contd.

Local Symptoms Systemic Sympt. HOST PLANT	Clover (Red) Vein Mosaic	Clover (White) Mosaic	Cowpea Aphid-borne Mosaic	Cowpea Chlorotic Mottle	Cowpea Mosaic	Cucumber Mosaic	Desmodium Yellow Mottle	Lettuce Mosaic	Pea Dwarf Mosaic	Pea Early Browning	Pea Enation Mosaic	Pea Seed-borne Mosaic	Pea Streak
Antirr. majus	ℓ -*	(ℓ) -	ℓ -	ℓ -	ℓ -	-* Mo	ℓ s	-* -*	LL -	ℓ s	- -	- -	ℓ -
Chen. amarant.	- -*	- -	LL_c -	- -	LL_n -	LL -*	- -	LL VC,NS, ChlS	LL VC,ChlS Stu,Ma	LL_n -	LL_c -	LL_n -	- -*
Cuc. sativus	- -	LL_c -	- -	- -	- -	LL ChlS VC,Mo	ℓ s	-* -*	-* -	LL_c -	- -	- -	- -*
Dat. stramon.	- -	- -	- -	ℓ -	ℓ -	-* Mo	- -	-* -*	LL -	ℓ -	- -	- -	ℓ -
Glyc. max I	- -	ℓ Mo	ℓ -	LL_n -	LL_c VB	- -	LL_n Mo	- -	-* -	ℓ Mo	0 0	- -	- (s)
Glyc. max II	- -	ℓ Mo	ℓ s	LL_c Mo	LL_c Mo	- -	LL_n VC	- -	-* -	ℓ -	0 0	- -	- -
Gomph. glob.	- -*	- -	ℓ s	LL_c, RS_c s	ℓ s	-* Mo	ℓ s	LL NS	-* Mo	LL_n -	LL_c Mo	- -	LL_n -
Lycop. escul.	- -	ℓ -	- -	LL_c -	- -	-* Mo,Ma, Stu	- -	-* -*	ℓ -	- -	- -	- -	- -*
Medic. sativa	- -	LL_c s	ℓ Mo	- -	- -	-* -	- -	-* -	-* -	- -	- -	- -	-* s
Nicot. glut.	0 0	- -	ℓ -	ℓ s	ℓ -	-* VC,Mo, Ma,Stu	ℓ s	-* -*	LL RS_c	0 0	- -	- -	0 0
Nicot. tab.	-* -*	- -	- -	ℓ -	ℓ -	-* VC,Mo	- -	-* -*	ℓ -	RS Str	- -	- -	- -
Petunia hyb.	0 0	- -	- -	ℓ s	- -	-* VC,Mo	- -	- -	0 0	0 0	- -	- -	0 0
Phas. vulg. I	ℓ -	LL_c Mo	LL_c ChlS	- -	LL_c s	LL VC,LR Mo,Stu	LL_n -	-* -	LL -*	LL_n NS or RS_c	- -	- -	- -
Phas. vulg. II	LL_c -	LL_c Mo	LL_c Chl	ℓ Mo	LL_c Mo	LL VC,Mo, Stu	LL_n Mo	- -	-* -	LL_n RS_c,LC	0 0	- -	ℓ -
Phas. vulg. III	- -	LL_c Mo	LL_c,VN -	RS_c,VN Mo,Str	ℓ s	- VC,Mo, Stu	LL_c Mo	- -	- -	LL_n RS,Mo, LC	0 0	- -	- -
Phlox drum.	- -	- -	- -	ℓ s	ℓ VC,ChlS	-* Mo	ℓ s	-* -*	- Mo	RS_n s	- -	- -	- ?
Pisum sat. I	- VB,Ma, Stu,Str	LL_c Mo,LR	- -	- -	- -	-* VC,VB, Mo	ℓ -	- VC,Mo	- VC,Mo, Ma,Stu	LL_n LR,VC	Chl Mo,Ma	- LR,Stu	W VB,Ma, N
Pisum sat. II	N VB,Ma, Stu,Str	LL_c Mo,LR	ℓ ChlS	- -	ℓ Mo	-* VC,VB, Mo	ℓ s	- VC,Mo	- VC,Mo, Ma,Stu	LL_n LR,VB	Chl Mo,Ma	- LR,Stu	W VB,Ma, N
Spin. oler.	- -	- -	ℓ Chl	ℓ -	LL_c Mo	LL VC,Mo	- -	-* -*	- VC,Mo, Ma,Stu, N	- -	- -	- -	- s
Trifol. prat.	- (VC,Mo)	ℓ Mo	- -	ℓ -	- -	-* -	- -	-* -	-* -	ℓ -	- -	- -	ℓ s
Trifol. rep.	- s,Str	ℓ Mo	- -	- -	- -	-* -	ℓ -	-* -	-* -	- s	- -	- -	(ℓ) -
Vicia faba	LL_n VC,Str	LL_n Mo	LL_n Mo	ℓ -	ℓ s	LL NS or Mo	- -	-* -	- VC,Mo Stu	ℓ s	- -	- LR,Mo, Ma	LL_n Mo,Str
Vigna unguic.	- -*	LL_c VC,Mo	LL_c VB,Stu, Chl	LL_n Mo	LL_c Mo	LL VC,Mo Stu	ℓ s	- -	- -	LL_n -	- -	- -	- -*

TABLE 3, Contd

LEGEND:

Local Symptoms
Systemic Sympt.

HOST PLANT	Peanut Mottle	Peanut Stunt	Soybean Mild Mosaic	Soybean Mosaic	Soybean Stunt	Tobacco Ringspot	Tobacco Streak	Tomato Black Ring	Tomato Spotted Wilt	Turnip Mosaic	Watermelon Mosaic	Wisteria Vein Mosaic
Antirr. majus	- -	LL_c Mo	- Mo	ℓ -	0 0	LL_n, (RS) Mo,NS	- -	ℓ s	- -	ℓ -	- -	ℓ -
Chen. amarant.	- -	LL_c VB	LL ChlS, VC,Mo	LL_c -	LL -*	LL_n Mo,Ma	- -	- -	-* -*	LL VC,ChlS	LL_n -	LL_c -
Cuc. sativus	ℓ Chl	LL_c Mo,RS_c	LL -	- -	-* VC,Mo	LL_c Mo	- Mo	LL_c Mo	- -	-* -	ℓ VC,Mo	- -*
Dat. stramon.	- -	ℓ Mo	- -	- -	LL -	LL_c Mo	- -	ℓ Mo	LL Mo,Ma	-* -	- -	- -*
Glyc. max I	LL_c VB,RS_c	- -	LL VC,ChlS	VN,Mo VN,Mo	LL VC,Mo	LL_n NS,Mo, Stu	- Mo	ℓ Mo	- -	- -	LL_n -	- -
Glyc. max II	- -	ℓ s	LL VC,ChlS Mo,Ma	VN,Chl Mo,Ma	LL VC,Mo, Stu	LL_c NS,Mo Stu	- Mo	ℓ Mo	- -	-* -*	ℓ -	- -
Gomph. glob.	- -	LL_c s	LL ChlS	ℓ -	LL -	ℓ s	- -	LL_n s	- (Ma,W)	LL -	ℓ -	ℓ -
Lycop. escul.	- -	ℓ Mo	- s	- -	-* -	LL_n -	- -	LL_n,RS Mo,NS	LL_c Ma,RS_n	-* -	- -	- -*
Medic. sativa	- -	- -	-* -	0 0	-* -	- -	- -	- -	- -	-* -*	- -	- -
Nicot. glut.	- -	ℓ Mo	LL VC,ChlS	- -	- VC,Mo, ChlS	ℓ s	- -	LL_c s	LL_n NS,N	LL VC,Mo, NS,Stu	- -	0 0
Nicot. tab.	- -	ℓ Mo	LL VC,VB, Mo	ℓ -	- VC,RS_c ChlS	LL_n RS	- -	LL_n,RS VC,Mo, NS	LL_n NS,N	LL -	ℓ -	ℓ -
Petunia hyb.	- -	ℓ VC,Mo	-* VC,Mo	ℓ VC	- VC,Mo, ChlS	LL_c Mo	- -	LL_n Mo	LL -*	LL VC,Mo, NS,Stu	- -	0 0
Phas. vulg. I	LL_c,VN s	LL_c,Ep Mo,Ma	- -	LL_c -	- -	LL NS,N	VN,Ep Str	LL_c NS,Mo, Ma	- -	-* -	ℓ -	LL_n -
Phas. vulg. II	LL_n,VN Chl,Str, N	LL_c,Ep Mo,Ma	LL -	LL_n, VN -	- -	LL_n LR,Str, NS,Stu	VN,Ep Str	LL_n Mo	- -	-* -	- -	- -
Phas. vulg. III	LL_c Chl	Ep Mo,Ma	LL -	LL_c -	- -	LL_n LR,Str, NS,Stu	VN,Ep Str	LL_c NS,Mo	- -	-* -	ℓ -	- -
Phlox drum.	- -	LL_c, Str,Ma Str,VN, Mo	LL -	- -	ℓ -	LL_n -	- -	ℓ NS,Mo	- -	- -	- -	- -
Pisum sat. I	ℓ VB,Mo	ℓ Ma,Mo	ℓ Mo,Stu	LL_n VC	-* -	LL_c,LR Mo	- -	- -	- -	-* -	- -	- -
Pisum sat. II	ℓ VB,Mo	ℓ Ma,Mo	- VC,Mo, Stu	LL_c s	-* -	LL_c Mo	- -	ℓ Mo	- -	-* -	- -	- -
Spin. oler.	- -	LL_c ChlS, Mo,Ma	LL VC,N ChlS	ℓ -*	- s	LL_c VC,Mo, LR,Stu	- -	- -	- -	LL ChlS, Stu,N	LL_c -	ℓ -
Trifol. prat.	ℓ -	ℓ s	-* -	- -	-* -	ℓ -	- -	- -	- -	-* -*	- -	- -
Trifol. rep.	- -	ℓ Mo	-* -	- -	-* -	ℓ -	- -	- -	- -	- -	- -	- -
Vicia faba	LL_n Mo	N Mo,RS_c	ℓ -	Chl,LL_c s	-* -	LL_n Mo	- -	- -	- -	LL s	ℓ Mo	LL_c ChlS
Vigna unguic.	ℓ Mo	LL_c VB,Mo	LL VC,NS, Stu	LL_c Mo	LL -	LL_n,RS Str,N	- Mo	LL_c LR,VC, Stu	- -	- -	ℓ -	- -

that employed by the other investigators. The reserve supply of each plant host seedlot was deposited in the Seedbank for Plant Virus Indicators (see Table 1 for accession numbers) at the National Seed Storage Laboratory, Fort Collins, CO 80521 USA, under the care of L. N. Bass.

To the extent possible, 10 plants of each of the 23 hosts were inoculated simultaneously with a given virus. Simultaneous inoculation of the 23 plant hosts was facilitated by coordinated plantings of hosts according to the time required from seed planting to the development of a usable plant (varied from 2 to 5 wk). Rapidly developing species usually were inoculated on fully expanded primary leaves; more slowly developing species were inoculated on secondary leaves on the fourth to eighth nodes. Several vigorous, healthy plants of each host were maintained as buffer-abraded, noninoculated controls during each test period. Soil mixtures, fertilizers, and cultural practices which promoted optimal growth and vigor of plant hosts were employed at each location.

Virus isolates.—Thirty-eight virus isolates, described in Table 2, were investigated. Each investigator tested isolates that were indigenous to his area and were recognized as typically representative of a known virus. Three separate isolates of five viruses (alfalfa mosaic virus, bean yellow mosaic virus, red clover vein mosaic virus, white clover mosaic virus, and pea streak virus) were investigated. The other 33 viruses were represented in this study by single isolates. In most cases, isolates tested had been documented in publications, and many are regarded as type cultures. Reference sources of these isolates, in the form of desiccated, infected plant tissue, were prepared by the investigators. Inoculum was prepared from plants previously determined to be satisfactory propagation hosts.

Inoculation procedure.—Host plants were inoculated by mechanical abrasion of the upper leaf surface. Mechanical inoculation was facilitated by dusting silicon carbide powder onto the leaf surfaces before inoculum application. Specific buffers, tissue extractions, and inoculum application methods were left to the discretion of each investigator. Inoculum infectivity, but not virus concentration, was monitored by investigators during this study.

Symptomatology.—Host plants were observed for 3 to 7 wk after inoculation for the development of localized and/or systemic symptoms. Virus-induced symptoms were categorized and recorded under 24 descriptive symbols previously agreed upon by the investigators. Leaves of noninoculated control plants were observed to distinguish between virus-induced symptoms and aging or other anomalous effects. Inoculated plants were scrutinized for localized and systemic symptoms at least three times, the last time after casual observation had indicated a steady-state host response to inoculation (Table 3). Anomalous symptoms which were observed in noninoculated control plants of *Petunia,* soybeans, and tomato at some locations, when tested as possibly having been induced by seedborne viruses, were found not to be pathogen-induced, and were discounted from the symptomatology of inoculated plants.

Unless otherwise designated, instances of systemic latent infection were determined and recorded. For this purpose, plants that had remained free of symptoms approximately 7 wk after inoculation with a given virus were assayed (back-inoculated) on another host of proven sensitivity to that virus. Most investigators did not test for localized latent infection, and a negative reaction for inoculated leaves was used therefore to designate absence of symptoms rather than absence of virus.

For the purpose of formulating a key from host range and symptomatological information supplied by the eight investigators, data were entered into punched cards under two virus-host interaction categories: definitive symptoms induced (localized or systemic or both: "positive") or symptoms not induced ("negative"). Missing or inconclusive data were entered into the cards as color codes. Various selections and arrangements (hierarchies) of hosts and formulations of trial keys were evaluated by card sorting. The key finally accepted was formulated on three basic premises: (i) the numbers of plant host hierarchies should provide adequate host germplasm by which to test and differentiate viruses of distinctive pathogenic capabilities, (ii) strategic hosts to be arranged in the hierarchical system should be selected which both divide the 38 viruses into approximately equal groups and respond to inoculation by these viruses by producing distinctive and reliable symptoms, (iii) to the extent possible, within the limits of missing or inconclusive data, hosts should be selected which have been acknowledged, historical differential hosts for well-known viruses.

Greenhouse test environment.—Environmental conditions varied significantly among the eight locations at which cooperative tests were undertaken, although the northern latitude of test locations varied only from 34° 40′ at Clemson, SC, USA to 51° 58′ at Wageningen, The Netherlands. Five of the locations were dominated by island or coastal climates, whereas three locations were dominated by less-temperate continental climates. At all locations, however, winter greenhouse environments were somewhat comparable in temperature (16 to 22 C), photoperiod (10 to 13 hr), and solar radiation intensity (80 to 180 langleys). Tests undertaken at one location in May and June were exposed to temperatures of almost 40 C and a 14-hr photoperiod. The effects of adverse

←

---

[a] Symbols used for virus-induced symptoms, Table 3:

O = Not tested; - = No symptoms; ? = Abnormal plant appearance, nondiscript; (ℓ) = Variable, symptom not always expressed; * = No back inoculation to test for latent infection; Chl = Chlorosis; ChlS = Chlorotic spots; Ep = Epinasty; ℓ = Latent, localized; LC = Leaf curl (upward); LL = Local lesions, unspecified, LL_c (chlorotic), LL_n (necrotic); LR = Leaf roll (downward); Ma = Malformation (distortion, crinkle, savoying, strap leaf, fern leaf); Mo = Mottle or Mosaic; N = Necrosis, general; NS = Necrotic spots (restricted systemic necrosis, zones, spots, flecks); RS = Ring spot, unspecified, RS_c (chlorotic), RS_n (necrotic); s = Latent, systemic; Str = Streak (stem necrosis, progressive leaf necrosis); Stu = Stunt; VB = Vein banding (chlorotic band on each side of vein); VC = Vein clearing (loss of normal color, chlorosis of vein); VChl = Vein chlorosis; VN = Vein necrosis; W = Wilting.

TABLE 4. Diagnostic key for 38 mechanically transmissible legume viruses.

SECTION I - Viruses inducing symptoms on *Vigna unguiculata*

- *V. unguiculata*[a] -- positive
  - *S. oleracea* -- positive
    - *L. esculentum* -- positive
      - *G. max* (Bragg) -- positive
        - *G. globosa* -- positive
          - *P. vulgaris* (Bountiful) -- local lesions ALFALFA MOSAIC (-, 20 x 60)[b]
          - *P. vulgaris* (Bountiful) -- chlorotic spots, systemic mosaic CLOVER BLOTCH[c] (o, 28)
        - *G. globosa* -- negative TOBACCO RINGSPOT (o, 28)
      - *G. max* (Bragg) -- negative
        - *G. globosa* -- local lesions PEANUT STUNT (o, 30)
        - *G. globosa* -- systemic symptoms CUCUMBER MOSAIC (LEGUME STRAIN) (o, 30)
    - *L. esculentum* -- negative
      - *N. glutinosa* -- positive
        - *C. sativus* -- positive
          - *P. vulgaris* (Bountiful) -- positive CLOVER (RED) NECROTIC MOSAIC (o, 30)
          - *P. vulgaris* (Bountiful) -- negative SOYBEAN MILD MOSAIC (o, 25)
        - *C. sativus* -- negative BROAD BEAN WILT (o, 25)
      - *N. glutinosa* -- negative
        - *P. vulgaris* (Pinto 111) -- positive COWPEA APHID-BORNE MOSAIC (–, 750)
        - *P. vulgaris* (Pinto 111) -- negative COWPEA MOSAIC (o, 28)
  - *S. oleracea* -- negative
    - *V. faba* -- positive
      - *P. hybrida* -- positive SOYBEAN MOSAIC (–, 750)
      - *P. hybrida* -- negative
        - *G. max* (Davis) -- positive
          - *C. sativus* -- positive CLOVER (WHITE) MOSAIC (–, 480)
          - *C. sativus* -- negative CLOVER (RED) MOTTLE (o, 30)
        - *G. max* (Davis) -- negative PEANUT MOTTLE (–, 750)
    - *V. faba* -- negative
      - *C. sativus* -- positive
        - *P. sativum* (DS Perfection) -- positive
          - *G. max* (Davis) -- positive TOMATO BLACK RING (o, 30)
          - *G. max* (Davis) -- negative PEA EARLY BROWNING (=, 105, 215)
        - *P. sativum* (DS Perfection) -- negative TOBACCO STREAK (RED NODE) (o, 28)
      - *C. sativus* -- negative
        - *P. vulgaris* (Bountiful) -- positive AZUKI BEAN MOSAIC (-, 750)
        - *P. vulgaris* (Bountiful) -- negative COWPEA CHLOROTIC MOTTLE[d] (o, 25)

SECTION II - Viruses inducing no symptoms on *Vigna unguiculata*

- *V. unguiculata* -- negative
  - *P. vulgaris* (Bountiful) -- positive
    - *V. faba* -- positive
      - *G. max* (Bragg) -- positive
        - *N. tabacum* -- positive BEAN YELLOW SEVERE MOSAIC (–, 750)
        - *N. tabacum* -- negative BEAN YELLOW MOSAIC (–, 750)
      - *G. max* (Bragg) -- negative
        - *P. sativum* (DS Perfection) -- positive PEA DWARF MOSAIC (o, 25)
        - *P. sativum* (DS Perfection) -- negative
          - *M. sativa* -- positive CLOVER YELLOW VEIN (–, 750)
          - *M. sativa* -- negative WISTERIA VEIN MOSAIC (–, 750)
    - *V. faba* -- negative
      - *G. max* (Davis) -- positive
        - *P. vulgaris* (Pinto 111) -- localized symptoms
          - *P. vulgaris* (B Turtle) -- localized symptoms BEAN POD MOTTLE (o, 30)
          - *P. vulgaris* (B Turtle) -- systemic symptoms BEAN SOUTHERN MOSAIC (o, 28)
        - *P. vulgaris* (Pinto 111) -- localized and systemic symptoms DESMODIUM YELLOW MOTTLE (o, 30)
      - *G. max* (Davis) -- negative BEAN COMMON MOSAIC[d,e] (–, 750)
  - *P. vulgaris* (Bountiful) -- negative
    - *P. sativum* (DS Perfection) -- positive
      - *P. vulgaris* (B Turtle) -- positive
        - *S. oleracea* -- positive CLOVER YELLOW MOSAIC (–, 520)
        - *S. oleracea* -- negative CLOVER (RED) VEIN MOSAIC (–, 645)
      - *P. vulgaris* (B Turtle) -- negative
        - *V. faba* -- positive
          - *C. amaranticolor* -- positive PEA SEEDBORNE MOSAIC (–, 750)
          - *C. amaranticolor* -- negative PEA STREAK[f] (–, 620)
        - *V. faba* -- negative
          - *C. amaranticolor* -- local lesions PEA ENATION MOSAIC (o, 30)
          - *C. amaranticolor* -- localized and systemic symptoms LETTUCE MOSAIC (–, 750)
    - *P. sativum* (DS Perfection) -- negative
      - *C. sativus* -- positive
        - *N. glutinosa* -- positive SOYBEAN STUNT (o, 30)
        - *N. glutinosa* -- negative WATERMELON MOSAIC (–, 750)
      - *C. sativus* -- negative
        - *D. stramonium* -- positive TOMATO SPOTTED WILT (0, 70-90)
        - *D. stramonium* -- negative TURNIP MOSAIC[d] (–, 720)

---

[a] Diagnostic plant-host, elaborated in Table 1.

[b] Particle shape (o = sphere, – = elongated particle), size in nm.

[c] Clover blotch virus induces variable chlorotic spots in *S. oleracea* and obscure vein clearing in *L. esculentum;* these symptoms may not be dependable under certain greenhouse conditions.

[d] The user of the diagnostic key is encouraged to seek reactive hosts from Table 1, for confirmation of viruses whose classification is based heavily upon negative host reactions.

[e] Although the Westlandia strain of bean common mosaic induced symptoms on *V. unguiculata*, it is assumed that the majority of BCMV isolates lack this capability.

[f] One of four cooperators indicated that pea streak virus induced symptoms on *C. amaranticolor*. Placement of this virus in the key is based upon non-induction of symptoms in *C. amaranticolor*

greenhouse environments on virus-host interactions were not accounted for in this study.

RESULTS AND DISCUSSION

The localized and systemic reactions induced by 38 virus isolates on 23 plant hosts, excluding 40 missing reactions, are presented in Table 3. Among these virus-host interactions there were 186 instances of localized latent infections and 51 instances of systemic latent infections. Twenty-seven virus-host interactions resulted in both localized and systemic latent infection. *Antirrhinum majus* L., *Trifolium pratense* L., and *T. repens* L. sustained latent infections more frequently than the other plant hosts. Desmodium yellow mottle virus uniquely induced both localized and systemic latent infections in seven of 23 plant hosts inoculated.

Variability in symptoms and host range due to virus isolate differences was appraised by comparing host reactions induced by three isolates for each of five viruses. Agreement of results obtained for isolates by the investigators was substantial. Minor disagreements were noted and hosts emphasizing isolate differences were avoided in formulating the key. Significant deviations in isolate character are footnoted in Table 4.

From these data, a key (Table 4) was constructed after successively evaluating various arrangements of selected plant hosts and hierarchies of these hosts. In accord with the premises previously discussed, a key was formulated which placed major emphasis on the responses to 38 viruses by *Vigna unguiculata, Spinacea oleracea, Phaseolus vulgaris, Lycopersicon esculentum, Vicia faba,* and *Pisum sativum*. Fourth- through seventh-order subdivisions in the keys were facilitated by the reactions to viruses by 12 additional plant hosts.

Dichotomous choices, preferred for toxonomic keys, were constructed in all possible cases, based upon whether (positive) or not (negative) viruses induced obvious symptoms on key hosts. When this was not possible because of identical host ranges among viruses, differentiation was achieved by qualitative symptomatology.

Coincident with our final choice of plant hosts and hierarchical arrangement of these hosts for the key, several viruses tended to be grouped together according to natural host relationships. For instance, four of the "clover viruses" were virulent to *V. unguiculata,* whereas the five "bean viruses" lacked virulence to this host. Similarly, three "pea viruses" were virulent to *P. sativum* and *V. faba,* but lacked virulence to *V. unguiculata* and *P. vulgaris*.

Although we have pursued the ideal of developing a definitive key to aid in diagnosing legume viruses, we have recognized several substantive limitations. Several qualifications are therefore appropriate. First, because negative ractions were employed as a virus-host reaction category in the key, and because reliance upon negative reactions accumulates as the user progresses through the key, confirmations of certain virus identities are dependent upon virus-host reactions from Table 3 which are supplemental to the key. Second, we recognize that numerous viruses will be discovered which are distinct from those included in this study; this, indeed, is one of the purposes of the key. We, therefore, do not imply that the use of this key shall necessarily result in the firm identification of an unknown virus. Third, different strains of the 38 viruses tested in this study are certain to induce different symptoms on some of the 23 plant hosts, thus posing identification inaccuracies. Fourth, although we attempted to select for use in the key virus-host interactions resulting in the most conspicuous symptoms, symptom anomalies due particularly to plant nutrition or test environment can be expected. Finally, the user should recognize that, since the key is based indirectly upon the interactions between viral and plant host nucleic acids and is indifferent to other virus properties, very dissimilar viruses with similar virulence on the selected hosts will be located proximally in the key. Because of this inevitable feature of the key, a "near miss" in applying the key to an unknown virus provides little meaningful information about that virus. Thus, a virus tentatively diagnosed by use of the key should be viewed by electron microscopy to determine whether or not its particle morphology matches that of the virus indicated by the key. Should particle morphology match the virus in the key, final identification should be confirmed by serology, and, where possible, by other properties of the virus particles.

LITERATURE CITED

1. BOS, L. 1964. Tentative list of viruses reported from naturally infected leguminous plants. Neth. J. Plant Pathol. 70:161-174.
2. BOS, L., D. J. HAGEDORN, and L. QUANTZ. 1960. Suggested procedures for international identification of legume viruses. Tidschr. Plantenziekten 66:328-343.
3. BOS, L., and J. P. H. VAN DER WANT. 1958. Virus diseases of leguminous plants. Landbouwvoorlichting 15:550-558, 573-587.
4. GIBBS, A. J., B. D. HARRISON, and A. F. MURANT (eds.). 1970-1976. Descriptions of plant viruses, Nos. 1-170. Commonwealth Mycological Institute and the Association of Applied Biologists, Kew, Surrey, England.
5. HAGEDORN, D. J. 1974. Diagnosis of pea viruses. Pages 15-19 *in* Virus diseases of pea, Pisum sativum. Phytopathol. Monogr. 9, Am. Phytopathol. Soc., St. Paul, Minnesota. 47 p.
6. JAMES, E. 1972. The organization of the National Seed Storage Laboratory. Pages 397-404 (Appendix No. 1) *in* Viability of Seed. Chapman and Hall, Ltd., London. 410 p.
7. SEEDBANK SUBCOMMITTEE, COMMITTEE ON VIROLOGY, AMERICAN PHYTOPATHOLOGICAL SOCIETY. 1977. Virus indicator species available. Phytopathol. News 11:26.
8. STEWART, V. B., and D. REDDICK. 1917. Bean mosaic. Phytopathology 7:61 (Abstr.).
9. VANDERVEKEN, J. J. F. 1976. International Working Group on Legume Viruses Report of the 'host range' committee. Parasitica 32:58-62.
10. VAN DER WANT, J. P. H., M. L. BOERJIAN, and D. PETERS. 1975. Variability of some plant species from different origins and their suitability for virus work. Neth. J. Plant Pathol. 81:205-216.
11. WEISS, F. 1939. A key to the typical viruses of leguminous crops. Plant Dis. Rep. 23:352-361.
12. WEISS, F. 1945. Viruses described primarily on leguminous vegetable and forage crops. Plant Dis. Rep. Suppl. 154:32-80.