Barley Yellow Dwarf Viruses in Small Grains of Pennsylvania: Isolate Identification, Distribution, and Vector Efficiency

F. E. GILDOW and J. FRANK, Department of Plant Pathology, The Pennsylvania State University, University Park 16802, and D. BINGAMAN and C. POWELL, Bureau of Plant Industry, Pennsylvania Department of Agriculture, Harrisburg 17110

ABSTRACT

Gildow, F. E., Frank, J., Bingaman, D., and Powell, C. 1987. Barley yellow dwarf virus in small grains of Pennsylvania: Isolate identification, distribution, and vector efficiency. Plant Disease 71:922-926.

Barley yellow dwarf virus (BYDV) was identified in small grains collected from 1984 to 1986 from eight counties in Pennsylvania. Isolates of BYDV recovered from commercial fields in three environmentally distinct cereal management areas were compared with the four characterized New York isolates (RPV, RMV, MAV, and PAV) by a combination of enzyme-linked immunosorbent assay and aphid transmission specificity. BYDV was recovered from 300 of 376 plants selected for testing on the basis of symptom expression. Sixteen percent of the BYDV-positive plants were infected by more than one isolate of BYDV. Data combined from single and mixed infections indicate that the percentages of plants infected with isolates resembling RPV, RMV, MAV, and PAV were 19, 4, 9, and 82%, respectively. Isolates similar to SGV were not detected in this survey. Comparisons of two *Rhopalosiphum padi* aphid clones and five *Sitobion avenae* aphid clones collected in Pennsylvania with previously characterized clones of New York aphids indicated no differences in vector specificity for the four BYDV isolate types. Of the 329 *R. padi* and *S. avenae* collected from symptomless oat plants from fields in three counties, 15 were viruliferous for PAV, 1 for RPV, and 1 for MAV. Results suggest reservoirs for a variety of BYDV isolates occur throughout Pennsylvania and that future epiphytotics could occur under the appropriate environmental conditions.

Barley yellow dwarf (BYD) disease of small grains occurs worldwide and is induced by a range of viruses belonging to the luteovirus group (16). Five characterized New York barley yellow dwarf virus (BYDV) isolates were first identified by their aphid-vector transmission patterns (9). Recently, these five isolates were divided into two groups, group 1 (MAV, PAV, and SGV) and group 2 (RPV and RMV), on the basis of serological and nucleic acid properties and cytopathology of infected hosts (16). These isolates have been used as type isolates for describing a range of luteoviruses isolated from cereals from different locations (1,4,6,10,15,17,18). These studies indicated that the predominant isolate type or strain causing BYD varies with crop species and location and may be associated with aphid species distribution and population levels.

In Pennsylvania, BYD occurs annually in spring- and fall-planted small grains. Because of its low incidence, the disease has not been of economic concern in recent years. Previous work, however, indicated that in some years BYD can occur in epiphytotics, severely reducing the yield. For example, surveys by the Pennsylvania Department of Agriculture in the fall of 1975 indicated nearly 100% infection of some winter barley fields, resulting in considerable yield reductions (L. Forer, *personal communication*). Disease incidence also can vary from county to county. Although BYD occurred only sporadically throughout most Pennsylvania fields surveyed in the spring of 1986 and was of little concern throughout the state, samples were received from extension agents in Tioga County from uniformly yellowed and

Accepted for publication 19 February 1987.

© 1987 The American Phytopathological Society

stunted oat fields. Yield after harvest was estimated to be 70% of previous harvests. Serological tests indicated these oats were infected with an MAV-like isolate of BYDV. Recent field studies in Pennsylvania indicated that yields of BYDV-infected Nobel oats inoculated at growth stages 3 and 4 were significantly less than those of uninoculated oats (5). In addition, evidence suggested that BYDV infection of fall-planted cereals may severely reduce the probability of winter survival (3).

Because of the importance of small grains to Pennsylvania agriculture and the location of small-grain-breeding plots throughout the state, a survey was initiated to identify the components of BYD in Pennsylvania and to form a basis for future studies involving epidemiology and disease resistance. The purposes of this study were to identify isolates of BYDV in small grains, using a combination of aphid transmission and serological methods, to determine the geographical distribution of these isolates and to test aphids infesting small grains in Pennsylvania for their ability to transmit luteoviruses associated with BYD.

MATERIALS AND METHODS

Collections of barley (*Hordeum vulgare* L.), oats (*Avena sativa* L.), and wheat (*Triticum aestivum* L.) were made in eight counties of Pennsylvania, representing three environmentally distinct cereal-growing regions: southeastern (Lancaster County), central (Centre, Columbia, Lycoming, Montour, and Union counties), and western (Somerset and Westmoreland counties). Collections were of spring-planted (April) cereals, except for one collection each of fall-planted (September) barley and oats from Lancaster County and one collection each of fall-planted wheat in Columbia and Lycoming counties. Samples were collected from at least three fields randomly selected in each county. Plants selected for testing on the basis of symptoms resembling those induced by BYDV were kept wrapped in moist paper towels on ice and brought to the laboratory within 6 hr of collection. Leaf and stem pieces from individual plants were washed under tap water, blotted dry, and either divided equally among four dishes for aphid recovery tests or finely chopped with a razor blade and stored for 2–6 wk at −20 C until tested by enzyme-immunosorbent assay (EIA).

EIA was done on samples (2–4 g) of finely chopped frozen tissue, which was first powdered in liquid nitrogen, homogenized in 4 ml of phosphate-buffered saline (0.02 M potassium phosphate and 0.15 M sodium chloride, pH 7.0) containing 0.05% Tween 20, 2% polyvinyl pyrrolidone (mol wt 10,000), 0.2% bovine serum albumin, and 0.01% sodium azide, then clarified in 4 ml of chloroform with low-speed centrifugation. The polyclonal rabbit immunoglobulins specific for the New York RPV, RMV, MAV, and PAV isolates were supplied by W. F. Rochow (Cornell University). Methods for EIA were similar to those previously described for the double-antibody sandwich technique (11). All antisera were cross-absorbed with healthy oat concentrates as previously described (13). Absorbance values at 405 nm for healthy oat and barley samples ranged from 0.00 (same as buffer controls) to 0.03, with a mean of 0.01. Absorbance values for BYDV-infected tissues typically ranged from 0.2 to 1.9 for homologous reactions. Individual polystyrene plate wells were coated with 1 μg of globulin in 100 μl of coating buffer. Plant samples were tested using 200 μl of clarified homogenate per well. All incubations of plates were done for 24 hr at 4 C. Plates were evaluated on a Dynatech Minireader II (Dynatech Laboratories Inc., Chantilly, VA). Homogenates from field-collected plants or from plants infected in aphid transmission tests were compared in EIA with homogenates from healthy oats (*A. byzantina* C. Koch 'California Red' or 'Coast Black') or oats infected by one of four characterized BYDV isolates (13). These isolates are RPV-NY, transmitted specifically by *Rhopalosiphum padi* (L.); RMV-NY, transmitted by *R. maidis* (Fitch); MAV-NY, transmitted by *Sitobion avenae* (Fabricius); and PAV-NY, transmitted efficiently by *R. padi* and *S. avenae* and occasionally by *Schizaphis graminum* (Rondani).

Virus-free colonies of all aphid species used were grown on caged 15-cm pots of Barsoy barley maintained under constant fluorescent light at 18–20 C. All colonies were initiated with first-instar nymphs parthenogenetically produced over a 24-hr period by a single apterous adult of each species on detached, healthy barley leaves. The characterized (9) New York clones of *R. padi*, *R. maidis*, *Sitobion avenae*, and *Schizaphis graminum* were used for all virus recovery tests from field plants, for subsequent index bioassy tests, and for vector comparison studies. Colonies of *R. padi* and *S. avenae* from Pennsylvania were initiated as described above from single apterous adults collected from oats in Lancaster, Lycoming, and Somerset counties.

Aphid virus-recovery tests were done by allowing aphids of each species a 48-hr acquisition feeding on leaf tissue from each field-collected plant to be tested. Aphids were then given a 5-day inoculation access on 7-day-old seedlings of California Red oats. Ten aphid nymphs (first to third instars) were placed on each of three seedlings for each treatment. After this inoculation access feeding period, the plants were fumigated and observed over a 4-wk period for BYD symptoms. Plants that became infected then were tested for vector-specific transmission patterns by the four aphid species (index bioassay), by EIA, or by both methods to identify the BYDV isolates recovered from the original field plant.

To compare the New York and Pennsylvania clones of *R. padi* and *S. avenae* for vector competence and transmission efficiency, aphids were allowed a 48-hr acquisition period on detached leaves from healthy oats or oats infected with one of the four BYDV isolates tested. Second and third-instar nymphs then were given a 5-day inoculation access period on 7-day-old California Red oats. To estimate the percentage of viruliferous aphids moving into and within the field and to identify the BYDV isolates they carried, adult aphids were collected individually from symptomless oat plants in three fields from each of three counties (Lancaster, Somerset, and Union). About 70% of the aphids were alate forms. Aphids were not collected from the relatively few infected plants showing symptoms in these fields, because these plants were tested directly for BYDV and all aphids from those plants would be carrying the identified isolate. Aphids were stored in screw-top plastic vials on ice (4–8 hr) until placed singly on 7-day-old oat seedlings for a 5-day inoculation feeding. These plants then were fumigated and observed over a 4-wk period. Infected plants were tested by EIA to identify the transmitted virus isolate.

RESULTS

BYDV isolates occurring in Pennsylvania small grains were identified by three types of tests: direct double-antibody sandwich EIA of field samples, aphid recovery and transmission of BYDV from field plants, and BYDV transmission to healthy seedlings by field-collected aphids.

When field samples were tested directly by EIA and the results compared with EIA responses of the four characterized New York isolates, eight patterns of infection were identified (Table 1). Four patterns were typical of plants infected with each of the characterized NY isolates, and four patterns indicated plants were doubly infected by a combination of two distinct isolates. Mixed infections also were detected in aphid recovery tests, which were verified by EIA (Table 2). As noted previously (9), the MAV and PAV antisera cross-reacted in a heterologous manner with MAV- and PAV-like isolates. The EIA comparison indicated no major serological differences between New York and Pennsylvania isolates with the polyclonal antisera used. To simplify discussion, the Pennsylvania isolates of BYDV will be referred to as RPV, RMV, MAV, and PAV isolates with the understanding that they are similar but not identical to the

New York BYDV isolates. Of 282 field plants sampled for BYDV over a 3-yr period by EIA, 225 (80%) tested positive for one or more BYDV isolates (Table 3). Double infections were found in 16% of the infected plants. Data combined from single and mixed infections indicate that the percentages of plants infected with RPV, RMV, MAV, and PAV isolates were 23, 3, 11, and 79%, respectively. No major differences were observed in isolate distribution, although Westmoreland County did have a larger percentage of RPV isolates and only PAV isolates were detected in spring oats in Lancaster County. In general, however, PAV isolates predominated, with RPV, RMV, and MAV occurring sporadically. The RMV isolates tended to occur more frequently in mixed infections with PAV and MAV isolates.

To determine whether Pennsylvania virus isolates reacting to specific BYDV antisera were also vector-specific, isolates recovered from field plants were subjected to a series of aphid transmission tests followed by EIA. Transmission patterns and EIA responses for four isolates designated RPV-PA, RMV-PA, MAV-PA, and PAV-PA (Table 4) were similar to responses expected for the characterized New York RPV, RMV, MAV, and PAV isolates (14). No obvious differences in symptom severity or vector efficiency were detected between similar New York and Pennsylvania isolates transmitted by New York aphids to California Red oats.

To recover Pennsylvania isolates for study of vector-specificity, some field-collected plants were tested by aphid recovery and transmission to indicator oat seedlings. Other objectives of the aphid-recovery test were to identify isolates with unusual transmission characteristics and serologically distinct isolates not responding to EIA tests and to identify SGV-like isolates that were not specifically tested for in the EIA. When the four aphid species were used for virus recovery from field plants, BYDV was recovered from 74 of 94 plants tested (Table 2). The percentages of plants infected with RPV, RMV, MAV, and PAV isolates, in single and mixed infections, were 10, 5, 3, and 90%, respectively. RMV isolates were detected only in mixed infections. In all cases, aphid transmission tests agreed with subsequent EIA. No isolates with unusual vector patterns were recovered, and *Schizaphis graminum*-specific transmission, characteristic of the SGV isolate (7), did not occur.

When *R. padi* and *Sitobion avenae* were collected from symptomless plants in nine oat fields in three counties and tested for their ability to transmit BYDV, only 18 of 329 aphids (5%) were viruliferous. The numbers of aphids transmitting RPV, MAV, and PAV isolates were 1, 1, and 15, respectively. One *R. padi* transmitted both RPV and PAV simultaneously. *R. padi* and *S. avenae* were the only aphid species observed on cereals in the three fields examined in each of three counties in May and June of 1986.

Comparison of New York and Pennsylvania *R. padi* and *S. avenae* indicated no differences in vector-specific transmission patterns among clones of aphids from either area (Table 5). All *R. padi* efficiently transmitted only RPV and PAV. A single RMV transmission indicated that the specificity mechanism was not absolute. All five clones of *S. avenae* transmitted only MAV and PAV. In all cases, MAV was transmitted more efficiently than PAV.

Table 1. Comparison of enzyme-immunosorbent assays (EIA) of barley yellow dwarf virus-infected oats collected in 1986 from four counties in Pennsylvania with oats infected with the RPV, RMV, MAV, and PAV type isolates from New York

Isolate identification[a]	No. plants tested	Absorbance at 405 nm[b]			
		RPV	RMV	MAV	PAV
Healthy oats	4	0.00	0.01	0.01	0.00
RPV-NY	4	*0.57*	0.02	0.01	0.01
RMV-NY	4	0.01	*0.34*	0.01	0.01
MAV-NY	4	0.01	0.01	*0.54*	0.04
PAV-NY	4	0.01	0.03	0.23	*1.08*
RPV-PA	5	*1.94*	0.02	0.02	0.01
RMV-PA	1	0.00	*0.11*	0.01	0.00
MAV-PA	7	0.00	0.00	*0.87*	0.35
PAV-PA	98	0.01	0.01	0.19	*0.74*
PAV + RPV-PA	8	*0.97*	0.07	0.34	*0.95*
PAV + RMV-PA	1	0.05	*0.10*	0.41	*1.21*
MAV + RPV-PA	1	*0.17*	0.05	*0.15*	0.06
MAV + RMV-PA	1	0.05	*0.23*	*0.38*	0.07

[a]Mixed infections were verified by aphid transmission patterns.
[b]Values are means of absorbance for the number of plants tested with each of the antisera indicated. Each plant was tested individually against the four antisera. The threshold for determining a positive value was 10 times the absorbance of the mean value obtained for healthy controls (0.01 ± 0.01). Positive values for the homologous virus-antibody reactions are in italics. Positive values for the heterologous MAV-PAV and RPV-RMV interactions are not in italics.

Table 2. Identification and distribution of barley yellow dwarf virus isolates infecting barley, oats, or wheat collected in Pennsylvania from 1984 to 1986 by aphid-recovery vector-specific bioassay[a]

County	Date collected	Host plant	Number tested	Number of plants infected with isolate type shown				
				RPV	MAV	PAV	PAV + RPV	PAV + RMV
Centre	Jun. 1984	Oats	17	–	–	11	–	–
Lancaster	Nov. 1984	Barley	17	3	–	5	–	–
Columbia	Jun. 1985	Wheat	5	–	–	2	–	3
Lycoming	Jun. 1985	Wheat	10	–	–	5	–	–
Montour	Jun. 1985	Barley	5	1	–	3	–	1
Centre	Jun. 1986	Oats	10	–	–	10	–	–
Lancaster	Jun. 1986	Oats	10	–	–	10	–	–
Somerset	Jun. 1986	Oats	10	1	2	6	1	–
Union	Jun. 1986	Oats	10	–	–	9	1	–
Total			94	5	2	61	2	4
Percentage of 74	infected plants			7	3	82	3	5

[a]Recovery tests were initiated by allowing virus-free *Rhopalosiphum padi*, *R. maidis*, *Sitobion avenae*, and *Schizaphis graminum* a 48-hr acquisition access period on detached leaves from field collected plants, then a 5-day inoculation access period on 7-day-old California Red oat seedlings. Each seedling was infested with 10 aphids. Seedlings that became infected were then tested by enzyme-immunosorbent assay, as described in the text, or by a second aphid-recovery test.

Table 3. Results of enzyme-immunosorbent assay (EIA) identification of barley yellow dwarf virus (BYDV) isolates infecting barley, oats, and wheat collected in Pennsylvania from 1984 to 1986, showing the distribution of isolates among counties and crop species[a]

County	Date collected	Host plant	Number tested	Number of plants infected with isolate type shown							
				RPV	RMV	MAV	PAV	PAV + RPV	PAV + RMV	MAV + RPV	MAV + RMV
Lancaster	Nov. 1984	Barley	11	–	–	–	4	–	1	–	–
Lancaster	Nov. 1984	Oats	5	2	–	–	2	–	1	–	–
Westmoreland	May 1984	Barley	15	7	1	3	4	–	–	–	–
Westmoreland	Jul. 1984	Oats	30	4	–	1	4	–	–	–	–
Lancaster	Jun. 1984	Oats	5	–	–	–	3	–	–	–	–
Centre	Jun. 1985	Oats	10	1	–	–	2	3	–	–	–
Columbia	Jun. 1985	Oats	20	–	–	–	7	5	–	2	–
Lycoming	Jun. 1985	Oats	14	–	–	2	8	–	–	–	1
Lycoming	Jun. 1985	Wheat	5	–	–	–	3	–	–	–	–
Somerset	Jun. 1985	Oats	36	1	–	4	16	9	–	3	–
Centre	Jun. 1986	Oats	32	2	–	4	21	3	–	1	1
Lancaster	Jun. 1986	Oats	30	–	–	–	22	–	–	–	–
Somerset	Jun. 1986	Oats	39	2	–	3	30	2	–	–	–
Union	Jun. 1986	Oats	30	1	1	–	24	3	1	–	–
Total			282	20	2	17	150	25	3	6	2
Percentage of the 225 infected plants				9	1	8	67	11	1	3	1

[a] Leaf and stem tissue of individual field plants were finely chopped and stored in plastic bags at −20 C until tested. Greenhouse-grown healthy California Red oats or oats infected with the type isolates of RPV, RMV, MAV, or PAV-NY were used as controls. Two-gram tissue samples were powdered in liquid nitrogen and homogenized in 4 ml of 0.01 M phosphate-buffered saline containing 0.05% Tween 20, 2% polyvinyl pyrrolidone (mol wt 10,000), 0.2% bovine serum albumin, and 0.01% sodium azide, then clarified in 4 ml of chloroform.

DISCUSSION

Pennsylvania isolates of BYDV were similar to characterized New York isolates in vector transmission patterns and in response to virus-specific antisera in EIA tests. Combined results of both EIA tests and aphid recovery tests indicated that the percentages of 300 plants infected with RPV, RMV, MAV, or PAV isolates in single or mixed infections were 19, 4, 9, and 82%, respectively. Isolates similar to RPV, RMV, MAV, and PAV were recovered from two of three grain management areas of the state. In the southeastern region (Lancaster County), MAV was not detected and only PAV occurred in spring oats. RMV isolates were rare and usually occurred in mixed infections with PAV or MAV. This is consistent with the fact that *R. maidis* was not observed on spring cereals during this study. This aphid, however, is a major component of the aphid population on fall-planted wheat and barley, suggesting that RMV could occur at a higher incidence in these crops. SGV-like isolates were not detected in any of 94 plants tested by *Schizaphis graminum* transmission bioassays or by heterologous reactions to MAV and PAV antisera in any of 282 EIA-tested plants. Although *S. graminum* was previously reported (2) to occur in peak population levels in June, we did not observe this species during our aphid collections. The most common aphid observed was *Sitobion avenae*, followed by *R. padi.*

In June 1986, aphid populations were less than one aphid per 0.3 m of row in all fields checked. Adult aphids collected for study occurred singly on plants scattered randomly throughout the fields. As might be expected, incidence of BYDV, based on symptoms, also was very low. In most fields, infected plants were widely spaced and randomly distributed. These plants were severely stunted and yellowed, suggesting infection during an early growth stage. Significant secondary spread from these plants to adjacent plants was not apparent. These observations suggested early infection of young seedlings by migrant aphids that were unable to establish a significant population level. These observations also suggest that the adult aphids collected in June for testing were more likely to be migrant aphids moving into the crop from other areas. In Lancaster and Union counties, difficulties in collecting sufficient numbers of aphids for testing coincided with observations of many parasitized aphid mummies and active coccinellid predators.

The origin of the virus isolates in the 16% of infected plants from which more than one virus was identified is unknown but could be epidemiologically important in understanding BYDV survival and spread between crop species. Mixed infections of PAV + RMV, MAV + RMV, and MAV + RPV were identified. Transmission of each of these isolate combinations required either inoculation by two aphid species, each viruliferous for the vector-specific isolate it transmits,

Table 4. Characterization of vector-specific transmission patterns and enzyme-immunosorbent assay (EIA) responses of barley yellow dwarf virus (BYDV) isolates recovered from single oat plants collected in Pennsylvania

Isolate designation	No. plants (of 3) becoming infected after aphid inoculation feeding[a]								Mean A_{405nm} in EIA with antiserum indicated[b]			
	Recovery test				Index bioassay test							
	RP	RM	SA	SG	RP	RM	SA	SG	RPV	RMV	MAV	PAV
RPV-PA	1	0	0	0	3	0	0	0	*0.71*	0.04	0.01	0.01
RMV-PA	0	3	0	0	0	3	0	0	0.03	*0.36*	0.01	0.02
MAV-PA	0	0	3	1	0	0	3	0	0.01	0.01	*2.03*	0.24
PAV-PA	3	0	1	2	3	0	3	2	0.01	0.01	0.64	*2.50*

[a] BYDV recovery tests from field-collected oats and index tests were initiated by allowing *Rhopalosiphum padi* (RP), *R. maidis* (RM), *Sitobion avenae* (SA), and *Schizaphis graminum* (SG) a 48-hr acquisition access period on detached leaves from each plant, then a 5-day inoculation access period on 7-day-old California Red oats. Each seedling was infested with 10 aphids. Aphids of each species, fed first on healthy oats (as controls), did not transmit virus to any of six plants during a 5-day inoculation access period. The RMV-PA isolate was recovered initially from a plant also infected with PAV-PA; all other isolates were from oats infected with a single isolate type.

[b] Mean absorbance values for four healthy oats when tested with the four antisera were 0.01 ± 0.01. The threshold value for considering a reaction positive was, set arbitrarily and conservatively, at an A_{405nm} of 0.1. Homologous positive antiserum-virus reactions are in italics. The MAV-PAV heterologous reactions were also positive but easily differentiated.

Table 5. Comparison of New York and Pennsylvania clones of *Rhopalosiphum padi* and *Sitobion avenae* for vector-specific transmission of four New York isolates of barley yellow dwarf virus (BYDV)

Aphid species	Aphid source[a]	No. of aphids (of 20) that transmitted the BYDV isolate indicated[b]			
		RPV	RMV	MAV	PAV
R. padi	NY	18	1	0	16
	PA-LA	17	0	0	17
	PA-S	19	0	0	20
S. avenae	NY	0	0	18	10
	PA-LA-G	0	0	13	6
	PA-LA-R	0	0	9	4
	PA-LY	0	0	15	4
	PA-S	0	0	18	6

[a] Virus-free colonies were started with 24-hr-old nymphs produced on healthy barley leaves by one apterous adult aphid collected at each location. Colonies were maintained on caged Barsoy barley at 15 C under constant light. New York aphids (NY) were originally obtained from W. F. Rochow, USDA-ARS, Cornell University. Pennsylvania aphids (PA) were collected from Lancaster (LA), Lycoming (LY), and Somerset (S) counties. Two color forms of *S. avenae* were obtained from Lancaster County, a typical green form (G) and an atypical reddish form (R).

[b] Virus-free nymphs were given a 48-hr acquisition feeding on detached leaves from infected oats or on healthy oats as a control. Single aphids then were given a 5-day inoculation feeding on 7-day-old California Red oat seedlings. None of 120 aphids of each clone, fed on healthy oats, transmitted virus to any of 12 plants. *R. maidis* fed on the RMV source transmitted virus to eight of eight plants but did not transmit virus to any of eight plants when allowed an acquisition access only on healthy oats as healthy controls.

or dependent transmission (12) by a single aphid species that had previously fed on a plant infected by both isolates. The occurrence of MAV, PAV, and RPV is not unexpected because *S. avenae* and *R. padi* were commonly observed in the fields. Although *R. maidis* was not observed in the field, 20% of the mixed infections included RMV, which is usually transmitted specifically by *R. maidis* from single infections. One explanation for this observation is that plants infected with RMV and PAV or MAV were inoculated by RMV-viruliferous *R. maidis* that were unable to survive on the small grains and by *S. avenae* or *R. padi* that transmitted MAV or PAV. Another explanation is that migrant *S. avenae* or *R. padi* may have fed on plants doubly infected with more than one isolate, acquired transcapsidated virions negating the vector-specificity mechanism, and subsequently inoculated both isolates to the plant tested. The fact that one of 18 viruliferous field-collected aphids simultaneously transmitted two isolates supports this possibility. The role of mixed infections of fall-planted cereals serving as virus and aphid reservoirs needs to be examined to understand more clearly the role of dependent transmission in BYDV epidemiology.

Ideally, breeding programs should use a variety of local BYDV-isolates when selecting for BYDV resistance or tolerance (8). The identification and characterization of isolates and their geographical distribution, therefore, is important. Future control of BYDV also will require continued research on aphid migration patterns relative to environmental parameters, identifying epidemiologically significant overwintering reservoirs of BYDV and aphid vectors, and determining the effects of local virus isolates on available cultivars.

LITERATURE CITED

1. Clement, D. L., Lister, R. M., and Foster, J. E. 1986. ELISA-based studies on the ecology and epidemiology of barley yellow dwarf virus in Indiana. Phytopathology 76:86-92.
2. Coons, B. F. 1968. Aphid trapping with black-light lamps. J. Econ. Entomol. 61:309-310.
3. Delserone, L. M., and Frank, J. A. 1985. The effect of *Pyrenophora teres* and two isolates of barley yellow dwarf virus (BYDV) on the freezing resistance of winter barley. (Abstr.) Phytopathology 75:624.
4. Forester, R. L., and Rochow, W. F. 1983. Barley yellow dwarf viruses in Idaho wheat and corn. (Abstr.) Phytopathology 73:788.
5. Frank, J. A., and Gildow, F. E. 1986. Effects of the PAV-NY isolate of barley yellow dwarf virus on yield components of oats. (Abstr.) Phytopathology 76:563.
6. Gildow, F. E., and Rochow, W. F. 1983. Barley yellow dwarf in California: Vector competence and luteovirus identification. Plant Dis. 67:140-143.
7. Johnson, R. A., and Rochow, W. F. 1972. An isolate of barley yellow dwarf virus transmitted specifically by *Schizaphis graminum*. Phytopathology 62:921-925.
8. Jones, A. T., and Catherall, P. L. 1970. The effect of different virus isolates on the expression of tolerance to barley yellow dwarf virus in barley. Ann. Appl. Biol. 65:147-152.
9. Rochow, W. F. 1969. Biological properties of four isolates of barley yellow dwarf virus. Phytopathology 59:1580-1589.
10. Rochow, W. F. 1979. Field variants of barley yellow dwarf virus: Detection and fluctuation during twenty years. Phytopathology 69:309-310.
11. Rochow, W. F. 1982. Identification of barley yellow dwarf viruses: Comparison of biological and serological methods. Plant Dis. 66:381-384.
12. Rochow, W. F. 1982. Dependent transmission by aphids of barley yellow dwarf luteoviruses from mixed infections. Phytopathology 72:302-305.
13. Rochow, W. F., and Carmichael, L. E. 1979. Specificity among barley yellow dwarf viruses in enzyme immunosorbent assays. Virology 95:415-420.
14. Rochow, W. F., Muller, I., Tufford, L. A., and Smith, D. M. 1986. Identification of luteoviruses of small grains from 1981 through 1984 by two methods. Plant Dis. 70:461-464.
15. Seybert, L. J., and Wyatt, S. D. 1981. Identification of barley yellow dwarf virus strains present in eastern Washington. (Abstr.) Phytopathology 71:108.
16. Waterhouse, P. M., Gildow, F. E., and Johnstone, G. R. 1987. The Luteovirus Group. Description of Plant Viruses. Commonw. Mycol. Inst./Assoc. Appl. Biol., Kew, Surrey, England. In press.
17. Yount, D. J., and Carroll, T. W. 1983. Barley yellow dwarf luteoviruses in Montana cereals. Plant Dis. 67:1217-1222.
18. Zhou, G.-H., Cheng, Z.-M., Quan, Y.-T., Zhang, X.-C., and Rochow, W. F. 1984. Serological identification of luteoviruses of small grains in China. Plant Dis. 68:710-713.