Identification and Incidence of Peanut Viruses in Georgia[1]

C. W. Kuhn*[2], J. W. Demski[3], D. V. R. Reddy[4], C. P. Benner[2], and Mandhana Bijaisoradat[2]

ABSTRACT

Surveys of peanuts in Georgia in 1983 detected peanut mottle virus (PMV), peanut stripe virus (PStV), and peanut stunt virus. The mild strain of PMV was by far the most prevalent virus in commercial peanuts; it occurred in every field and an average incidence of 15-20% was observed when the growing season was about two-thirds complete. The necrosis strain of PMV was noted in 39% of the fields, but the incidence was less than 0.1%. A new severe strain of PMV (chlorotic stunt) was identified in two fields. PStV was found at four locations; in each case the infected plants were near peanut germplasm lines from The People's Republic of China. Mixed infections of PMV and PStV occurred frequently. Peanut stunt virus was noted only in one research field in 1983. Numerous serological and sap inoculation tests did not detect tomato spotted wilt virus or cowpea chlorotic mottle virus.

Key Words: Peanut viruses, disease incidence, survey.

A systematic survey of peanut viruses was made in Georgia in 1973 (15). Peanut mottle virus (PMV) was found in every peanut (*Arachis hypogaea* L.) field surveyed, and peanut stunt virus occurred in less than 2% of the fields. The survey showed that 26% of the peanut plants were infected with PMV. When the disease incidence value was combined with the yield loss per diseased plant (at least 20%) (12, 15), the estimated yield loss caused by PMV was over 5% in 1973.

A new survey for peanut viruses was conducted in 1983 for two primary reasons. First, since 1973 several newly identified viruses have been found in peanuts, both in Georgia and in other parts of the world: cowpea mild mottle virus (1), groundnut crinkle virus (8), groundnut eyespot virus (7), a luteovirus from groundnut rosette diseased plants serologically related to beet western yellows virus (2), peanut chlorotic leaf streak virus (16), peanut clump virus (6, 19), a potyvirus causing peanut mild mottle (20), peanut green mosaic virus (17), and peanut stripe virus (PStV) (5). Cowpea chlorotic mottle virus (CCMV) has been isolated and identified from peanuts in experimental plots, and plants with symptoms similar to tomato spotted wilt (TSWV), a previously reported virus in the United States (10), have been noted in commercial peanut fields (J. W. Demski and C. W. Kuhn, *personal observations*). Second, there has been a change in the use of peanut cultivars. During the last 15 years, cultivar Florunner has become increasingly dominant, accounting for about 95% of the total peanut acreage in Georgia.

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[2]Department of Plant Pathology, University of Georgia, Athens, GA 30602.

[3]Department of Plant Pathology, Georgia Experiment Station, Experiment, GA 30212.

[4]International Crops Research Institute for the Semi-Arid Tropics, Hyderabad 500016, India.

Materials and Methods

Two surveys were conducted in the southwestern section of Georgia during 1983: (i) July 5 and 6, 6-9 weeks after planting and (ii) August 22 and 23, 12-15 weeks after planting. Thirty-nine fields in 13 counties were evaluated. Different sections of each field were inspected by three and four people on the first and second surveys, respectively. Plants were observed for symptoms typical of the mild strain (M) of PMV, and leaves from a few plants were collected. The incidence of PMV-M was estimated by counting the number of infected plants of 100 consecutive plants in a row (3-4 counts/field/person). A thorough search was made for plants with symptoms atypical of PMV-M, and leaves from all such plants were collected. Furthermore, in 23 of the 39 fields leaves were collected at random from 100 plants/field, irrespective of symptoms.

The leaf samples from plants with symptoms were processed in two ways. First, one leaflet from each plant was ground in 0.05 M potassium phosphate buffer, pH 7.5, containing 0.1% mercaptoethanol, 0.2% sodium diethyldithiocarbamate and 1% Celite. The resulting sap was used to inoculate Florunner peanut, *Phaseolus vulgaris* L. cv Topcrop, *Glycine max* (L.) Merr. cv Bragg, *Vigna unguiculata* Walp. cv California Blackeye, *Nicotiana glutinosa* L., *N. tabacum* L. cv Xanthi, and *Lycopersicon esculentum* Mill. The following specific disease reactions were considered useful in diagnosis: PMV-mild mottle on Florunner and Bragg and local necrotic lesions on Topcrop; peanut stunt virus-chlorosis and severe stunt on Florunner and local chlorosis on California Blackeye; TSWV-chlorosis, ringspots, and stunt on Florunner, bronzing on tomato, and local necrotic lesions on Xanthi; PStV-stripe on Florunner and no symptoms on Topcrop. Reactions other than these would suggest the need for additional studies for identification of other viruses. Second, another leaflet was ground in phosphate-buffered saline (0.02 M potassium phosphate, pH 7.3) containing 0.15 M NaCl, 0.003 M KCl, and 0.2% diethyldithiocarbamate for the enzyme-linked immunosorbent assay (ELISA). The ELISA procedure was described previously (3).

The leaf samples collected at random were evaluated by ELISA. Sap from leaflets of 10 plants was placed in each well of ELISA plates and tested with antibody-enzyme conjugates prepared against PMV-M, PStV, and CCMV.

Results

Peanut mottle virus. The visual identification of PMV-M was confirmed by positive ELISA determinations and typical symptoms produced on Florunner peanut (mild mottle) and Topcrop bean (necrotic local lesions) after mechanical inoculation. Twenty samples, each from a different field, were all positive. Additional corroboration of our ability to identify PMV-infected plants was made with the 2,300 plants collected at random and tested by ELISA (see below).

Similar to the 1973 survey, PMV-M was present in every peanut field observed, which we attribute to its seed transmission (11). Mottle disease incidence was only 3-4% at the first survey date, indicating limited secondary spread from a speculated 0.1 to 1.0% frequency of seed transmission in commercial peanut seed (12) (Table 1). Two fields, however, had as many as 25% of their plants infected with PMV-M at a plant growth stage before pods had set. At the time of the second survey (two-thirds of the growing season had been completed), an average of 15-20% of the plants were infected with PMV-M (Table 1). Nearly sixty percent of the fields had a disease incidence of 20% or higher; two

Table 1. Incidence of peanut mottle virus in peanuts in Georgia in 1983.

County	Survey no. 1		Survey no. 2	
	Fields observed (no.)	Average incidence of PMV (%)	Fields observed (no.)	Average incidence of PMV (%)
Calhoun	2	2	1	3
Colquit	2	3	2	25
Decatur	-[a]	-	1	30
Early	-	-	2	6
Miller	-	-	2	1
Mitchell	2	7	-	-
Schley	-	-	2	11
Sumter	3	6	4	37
Taylor	-	-	1	8
Terrell	3	2	1	20
Thomas	2	1	2	20
Tift	2	5	1	8
Worth	2	2	2	35

[a]Not observed.

Table 2. Single and mixed infections of peanut stripe virus (PStV) and peanut mottle virus (PMV) in experimental field plots in 1983.*

County	Plot site	Plants tested (no.)	Plants infected with		
			PStV	PMV	PStV+PMV
Oconee	Plant Sciences Farm	218	80	105	49
Spalding	Plant Introduction	16	16	6	6
Sumter	Plant Materials Center[b]	6	6	5	5
Sumter	Plant Materials Center[b]	13	3	--[d]	--
Tift	Research (breeding)[c]	10	6	--	--
Tift	Research (breeding)[c]	8	4	--	--
Tift	Research (breeding)[c]	226	13	--	--

[a]Individual plants were tested serologically (enzyme-linked immunosorbent assay) for PStV and PMV.

[b]Collections made on different dates: August 22 and September 30.

[c]Collections made on different dates: July 6, October 5, and November 16.

[d]No test for PMV.

of these fields had 50 and 80% incidence. However, 33% of the fields had less than 10% incidence.

Peanut leaves collected at random from 2,300 plants during the survey were tested by ELISA by combining sap from leaflets of 10 plants in each well. Each of the 230 ELISA samples contained PMV, indicating at least 10% incidence and probably considerably more.

On the first survey, the necrosis strain (N) of PMV was observed in seven fields (39%). The strain was identified by serology and unique necrosis symptoms on Florunner and Argentine peanuts. No plants with PMV-N were noted during the second survey. We speculate that the PMV-N infected plants were severely stunted and hidden by the abundant foliage of both healthy and PMV-M infected plants.

A new PMV variant with striking symptoms was isolated from plants in Taylor and Worth Counties. Four types of symptoms were noted: (i) distinct chlorosis (spots and rings), (ii) narrowing of leaflets, (iii) small leaves and severe stunting of plants, and (iv) strong leaf rolling. The symptoms were distinct from previously described strains of PMV (14). After serial passage through local lesions on Topcrop bean, the variant's relationship to PMV was established by serology, electron microscopy, and diagnostic hosts. Plants with similar striking symptoms were observed in nine of the 39 fields surveyed.

Peanut stripe virus. A new virus was isolated from experimental peanut plots in 1982 (5). It was purified, characterized, and named PStV. In 1983 the virus was found in experimental fields in four counties in Georgia (Table 2). The virus was identified on the basis of diagnostic hosts, serology (immunodiffusion and ELISA), electron microscopy, and physicochemical properties.

With one possible exception, PStV was not found in any commercial peanut fields in 1983. On the two survey trips, leaves were collected from plants with possible PStV symptoms and tested by ELISA. None of the 72 plants (in seven counties) collected on trip one or the 176 plants (in nine counties) from trip two had PStV. Many of the 248 leaf samples were also inoculated onto peanuts in the greenhouse. One of the inoculated peanuts (sample from Schley County) later reacted positively with PStV antiserum (ELISA). However, since there was no positive reaction with the leaf sample from the field, it is likely that the PStV-infected plant in the greenhouse could have resulted from a greenhouse contamination.

No special attempt was made to determine the incidence of PStV in experimental plots in 1983. However, the 218 plants tested by ELISA in the Oconee County plot (Table 2) represent about 45% of the plants in the plot. Therefore, the 37% incidence level is probably similar for the whole plot. Seeds used to plant this plot were obtained from plant breeders in five southeastern states. Their seed lots were believed to have been contaminated with PStV and PMV during the 1982 growing season.

Tests for other viruses. Peanut stunt virus was not observed in any peanut fields surveyed in 1983. However, the virus was detected by symptomatology in a research plot in Spalding County and then identified by diagnostic hosts.

Plants with symptoms typical of TSWV (10) have been observed previously in Georgia by the first two authors. Similar plants were observed again on the two survey trips in 1983. A few (less than 0.1%) such plants were found in five fields. TSWV could not be identified by diagnostic hosts or by a serological test (ELISA). Furthermore, two of the TSWV-suspected plants had PMV strain chlorotic stunt (described above) which causes stunting and chlorotic ringspot symptoms somewhat similar to TSWV symptoms. We believe it is probable that all of the plants with these types of symptoms were caused by PMV and not TSWV.

Peanut is a symptomless host of CCMV (500-800 μg

of virus/g of tissue), and it has been found in peanut research plots in Georgia (Kuhn, *personal observation*). The virus was not detected by ELISA in 185 individual peanut samples believed to be infected with PMV or PStV or in 2,300 plants (230 ELISA samples) collected at random during the second survey trip.

Mixed infections. Mixed infections with PStV and PMV were relatively frequent (22%) in the Oconee County plot (Table 2). Many plants in the plot were stunted and had ringspot symptoms; neither disease reaction is typical for either PStV or PMV infections. The severe disease reaction was not reproduced when peanuts were inoculated in the greenhouse with sap from leaf tissue infected with both viruses. Therefore, we are uncertain about the cause of the stunt and ringspot symptoms. Mixed infections also were found in experimental plots in other counties (Table 2).

Discussion

The incidence of PMV-M in Georgia in 1983 appeared to be lower (15-20%) than the incidence in 1973 (26%) (15), although the difference is probably not significant. General observations of peanuts over a period of 20 years indicate that incidence of PMV varies from year to year. However, we believe the two estimates of disease incidence (1973 and 1983) are realistic and representative of long-term effects of PMV. Furthermore, we speculate that PMV incidence is governed by environmental factors such as rainfall and temperature which affect aphid multiplication, their movement, and consequently their ability to cause secondary spread of the virus (4). Since neither commercial lines nor plant introductions are known to be resistant to PMV-M (13), it remains a serious threat to the peanut industry.

The mild strain of PMV continues to be the dominant one. The necrosis strain was observed more frequently in 1983 than 1973, but its incidence was too low to cause an economic problem. A new variant of PMV which causes severe stunting was identified in 1983. The variant has remained stable under greenhouse conditions for over six months, and we suggest that it should be named the chlorotic stunt (CS) strain of PMV. Peanut fields should be monitored routinely for these severe disease producing strains (N and CS) of PMV. They may cause disease losses comparable to groundnut rosette in Africa (18) and TSWV in India (9).

PStV was found at four locations in Georgia in 1983. In all cases the infected plants were grown at experiment stations where peanut germplasm lines from The People's Republic of China were growing nearby (5). Although extensive observations and serological tests were made, no PStV was found in commercial peanuts. Every effort possible should be made to attempt to keep the virus, which is seed-transmitted, from getting into peanut seed production which provides seeds for planting each year's crop.

We believe peanut stunt virus occurs infrequently in peanuts in the southern part of Georgia because white clover, the primary overwintering host, is not prevalent and the aphid vectors of the virus rarely move from plants outside the peanut fields to in-field plants during the hot summer months.

Other than PStV and PMV-CS, no new viruses were detected in 1983. Since all plants in the survey with symptoms atypical of PMV were evaluated, it appears unlikely that other viruses are a threat to peanuts at this time. Furthermore, the continuous culture of primarily one peanut cultivar (Florunner) has not increased the incidence of PMV, promoted new strains of existing viruses which have reached significant levels of incidence, or provided an opportunity for new viruses to become established.

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