Occurrence of Wheat Spindle Streak Mosaic Virus on Winter Wheat in Georgia

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ABSTRACT

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A virus disease not previously reported in Georgia was observed in winter wheat during the spring of 1984. The cause was identified as wheat spindle streak mosaic virus (WSSMV) based on symptomatology, mode of transmission, enzyme-linked immunosorbent assay, and serum-specific electron microscopy. Wheat spindle streak mosaic (WSSM) was observed in a number of locations in Georgia, primarily in an area near Plains, and also near Hartsville, SC. It was found on two wheat cultivars, Coker 797 and Florida 301, and on rye. A significant yield difference was found between healthy and diseased plants (22 and 35%) of Florida 301 at two of three locations. Yield reduction resulted mainly from a decrease in tillering. Other yield components that were reduced significantly were kernel weight, straw weight, and total biomass. This is the first report of WSSMV in the southeastern United States.

Wheat spindle streak mosaic (WSSM) was first described on winter wheat in southeastern Canada in 1960 (14). The disease is characterized by chlorotic to necrotic spindle-shaped streaks in the leaves, slight stunting, reduced tillering, and reduced seed yield. WSSM is found throughout the eastern and midwestern wheat-growing areas of the United States (2,7,23,24) and in other countries (1,11,18). The disease has been reported most often in the Great Lakes area of the United States and Canada.

The causal agent, wheat spindle streak mosaic virus (WSSMV) is soilborne with flexuous, filamentous particles 200-3,000 nm long and 15-18 nm wide (6,8,20). In the host, the particles are scattered throughout the epidermal and paranchyma cells and are difficult to detect in thin section (1,21). Infected cells also contain pinwheel inclusions (8).

Symptoms of the disease are produced only during periods of cool temperatures (8-12 C), and disease development ceases at temperatures higher than 18 C (15,16). The virus is thought to be vectored by *Polymyxa graminis* Led., a soilborne plasmodiophoraceous fungus (10,18,19). The virus can survive for years in the soil. Mechanical transmission of the virus

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through sap is difficult. The most efficient transmission is accomplished in contaminated soil (15,17). Grain yield has been reduced from 14 to 44% in susceptible wheat cultivars through the reduction of tillering (3-37%) (23). In Canada, yields were reduced from 7 to 60% according to data from individual plant samples or small plots (3,15).

The objectives of this study were to determine the incidence of WSSMV in Georgia's winter wheat crop and the effect of the virus on Florida 301, a widely grown cultivar.

MATERIALS AND METHODS

Field survey. During the spring of 1984, wheat fields chosen at random throughout central and southern Georgia were surveyed for visible symptoms of WSSM, with particular attention given to low or wet areas. Plants showing typical WSSM symptoms were collected and taken to the laboratory for further testing.

Serology. Antiserum to WSSMV was obtained from K. Z. Haufler (5). All field samples were tested with direct enzymelinked immunosorbent assay (ELISA). Selected samples were tested with serum-specific electron microscopy (SSEM) to confirm the reliability of the ELISA. The direct ELISA procedure employed was similar to that described by Lister (9) and Rejeshwari et al (12), with the exception that antigen buffer contained a high (0.4) molar phosphate (4) and 0.1 M EDTA. Gamma globulins from the antiserum to WSSMV were used at 10 mg/ml. The

enzyme conjugate was used at a 1/500 dilution. All ELISA reactions were measured by reading absorbence at 410 nm in a Dynatech ELISA reader. A minimum of 20 samples per field from plants with and without symptoms were tested by ELISA. Reactions having a 1.5-fold increase or higher over healthy controls were considered positive.

For SSEM, 300-mesh grids were carbon-coated and covered with a film of Parlodion. These grids were then floated on a 20-µl drop of a 1/100 dilution of antiserum in 0.1 M Na₂HPO₄-NaH₂PO₄ buffer at pH 7.0. Grids were incubated at room temperature for 30 min, then rinsed with phosphate buffer and blotted dry. Small leaf pieces (about 1 g) from infected wheat leaves were triturated with a mortar and pestle. One to 2 ml of phosphate buffer was added to the homogenized tissue. Grids, previously coated with the WSSMV antiserum, were floated on a 20-µl drop of extract and incubated at room temperature for 1 hr, then rinsed again with phosphate buffer and blotted dry. The grids were negatively stained with 2% ammonium molybdate and viewed under a Philips 201C electron microscope at ×20,000.

Transmission and symptomatology. Five winter wheat cultivars were planted in WSSMV-contaminated soil collected from Sumter County, GA, near Plains. The cultivars were Holley, Florida 301, Florida 302, Coker 797, and an advanced breeding line, 781014. The plants were grown in a growth chamber at 10–12 C and observed over a 4-mo period for symptom expression. Each cultivar was tested by ELISA to determine the presence of WSSMV.

Yield study. Three WSSMV-infected wheat fields were chosen for yield loss assessment. The fields, all planted with Florida 301, were located in Plains, 5 mi. north of Plains, and 8 mi. north of Oglethorpe, GA. A 2-m row section of obviously infected plants was harvested along with a corresponding section of uninfected plants. Twelve pairs of rows were harvested from each of the three fields. The number of infected and total tillers was determined for each harvested section. Several yield components were

also determined, including grain yield, straw weight, total biomass, harvest index (grain yield divided by total weight of grain and straw), weight of 1,000 kernels, weight of seed from 10 heads, and number of seed from 10 heads.

RESULTS

Symptomatology. WSSMV produced the typical spindle-shaped chlorotic lesions on the leaves and stems of both Florida 301 and Coker 797 and an unknown rye cultivar. Symptoms were generally found on the lower leaves and included greenish yellow to white spots parallel to the leaf veins. The spots were elongated with tapered ends, which gave them the characteristic spindle shape. Later in the season, as the weather moderated, new leaves had faint or no symptoms.

Diseased plants were generally restricted to low or wet areas of fields, but in some instances, they were more uniformly distributed throughout the field. Areas of fields with chlorotic, stunted plants were obvious in early March, when plants were still in the rosette stage. As the disease progressed, areas of yellowed and stunted plants became easily recognizable. Heading of infected plants was delayed 7–10 days.

Disease distribution. The disease was found at 18 locations in Georgia (Table 1. Fig. 1) and at Hartsville, SC. Fields with moderate to severe damage (10-20\% of plants showing WSSM symptoms) were found in Sumter County in Plains and near Americus, in Taylor County near Butler, and in Macon County near Oglethorpe (Table 1). Other fields with lesser amounts of the disease were found in Macon County near Marshallville, in Peach County near Fort Valley, in Spalding County near Griffin, in Washington County near Davisboro, and in Schley County near Ellaville. The Ellaville field was unusual in that the virus was found infecting wheat and volunteer rye, which was scattered throughout the field. Small grains have been grown at the Hartsville, SC, site for many years, and mosaiclike virus symptoms are often observed. Losses to soilborne viruses were especially high in 1984 among susceptible cultivars. WSSM was not found in the following Georgia counties: Laurens (three locations), Johnson (two), Burke (one), Colquit (one), and Tift (one). Additionally, one wheat field was inspected near Ouincy. FL, that was negative for WSSM. In all the Georgia locations, the disease was found only on Coker 797 or Florida 301, except for the one rye infection. When plants with WSSM symptoms from each field were assayed by ELISA, all gave a positive reaction.

Identification. The virus was successfully transmitted from field soil to the five wheat cultivars tested (Holley, Florida 301, Florida 302, Coker 797, and 781014).

After 15 wk at 10-12 C, leaves of each of these cultivars had the chlorotic spindleshaped lesions characteristic of WSSM. The identity of the virus was determined by ELISA in all plants that showed symptoms. Samples taken from the field (Table 1) and from the five cultivars inoculated in the growth chamber had a positive reaction to WSSMV antisera with this technique. Values, as measured by a Dynatech reader, that had at least a 1.5-fold increase above the healthy controls were considered positive. Identification of the long, filamentous particles of the virus was confirmed by SSEM. The observation of virus particles correlated with positive reactions by ELISA (Table 1).

The resting spores of *P. graminis* were found associated with the roots of wheat plants collected from the field and those grown in contaminated soil in the growth chamber.

Yield loss assessment. WSSM reduced yield 22 and 35% at two locations in Georgia (Table 2). The third location (5 mi. north of Plains) showed a reduction, but it was not significant (P = 0.05). Straw weight and total biomass was significantly reduced in all cases. Harvest index, weight of seed from 10 heads, and number of seed from 10 heads did not differ significantly at any of the locations. The total number of tillers differed significantly at the two locations that had significant yield losses. The 1,000-kernel weight was significantly different (P = 0.05) at the two Plains locations.

DISCUSSION

WSSM has been observed for the first time in the winter wheat crop in the Southeast. It was found in 18 fields throughout Georgia and was particularly severe in four fields near Plains. We also identified the disease near Hartsville, SC. The virus was only found infecting wheat cultivars Coker 797 and Florida 301 in the field. Other cultivars observed in the field were not diseased as determined by

visual observation and ELISA. It is not known if other cultivars have field resistance or if the virus was not present where they were grown. Most of the wheat acreage in the Coastal Plain area of southern Georgia was planted with susceptible cultivars Coker 797 and Florida 301 in 1984. The virus was also found infecting rye, which has not been previously reported as a host for the virus in this country (18). The virus has been reported on rye in the German Democratic Republic (11).

WSSMV reduced the grain yield of Florida 301 significantly at two locations. The yield reduction in these two fields was quite high, 22 and 35%. This is in the range of previously reported yield losses of 7–60%, depending on the cultivar (3,15,22). Losses of this magnitude could have a devastating effect on the wheat crop if a large enough area became infected.

Winter conditions in Georgia and much of the Southeast are favorable for

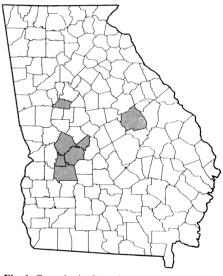


Fig. 1. Counties in Georgia with winter wheat naturally infected with wheat spindle streak mosaic virus.

Table 1. Survey for wheat spindle streak mosaic on winter wheat in Georgia and verification of wheat spindle streak mosaic virus by ELISA and serum-specific electron microscopy

County	Location	No. of fields surveyed	No. of fields infected ^a	ELISA reacton ^b	SSEM°
Macon	Marshallville		3	2.4	+
	Oglethorpe	6	1	1.8	NT
Peach	Fort Valley	5	3	1.7	NT
Schley	Ellaville ^d	3	1	1.4	NT
Spalding	Griffin		1	1.7	NT
	Experiment Station	6	Ī	1.6	NT
Sumter	Americus		3	1.9	NT
	North of Plains		1	1.8	+
	Plains	8	1	2.4	+
Taylor	Butler	3	ī	2.9	+
Washington	Davisboro	4	i	1.5	NT
	Hartsville, SC	1	1	2.2	NT

^aBased on visual observation of symptoms that were confirmed by ELISA.

^bThe values are the multifold increase over healthy controls. Although a minimum of 20 samples were taken from each field, data represent the average reading from the first plant tested (that had symptoms of wheat spindle streak mosaic) in the representative field.

 $^{^{}c}$ + = Long, filamentous rods were observed; NT = not tested.

^dVirus sample found in rye.

Table 2. Yield components of Florida 301 infected with wheat spindle streak mosaic virus at three locations in Georgia

Yield component	Treatment	North of Plains	Plains	Oglethorpe
Harvest index ^y	Healthy	0.45 a	0.43 b	0.40 a
	Diseased	0.48 a	0.45 a	0.45 a
Straw weight (g)	Healthy	151.3 a	187.6 a	198.3 a
311411 11-18-11 (8)	Diseased	122.6 b	112.7 b	127.1 b
1,000 Kernel	Healthy	40.0 a	41.1 a	35.6 a
weight (g)	Diseased	36.0 b	34.2 b	33.8 a
Seed from 10	Healthy	233.9 a	240.2 a	287.5 a
heads (g)	Diseased	240.0 a	253.1 a	255.7 a
Total biomass	Healthy	271.2 a	328.5 a	329.8 a
(grain + straw) (g)	Diseased	234.0 b	204.4 b	229.5 b
Total no. of tillers	Healthy	179.8 a	192.9 a	184.7 a
	Diseased	150.3 a	136.3 b	151.8 b
No. of diseased tillers	Healthy	2.9 b	2.8 b	4.0 b
no. or discussed times	Diseased	85.7 a	95.1 a	117.7 a
Diseased	Healthy	1.7 b	1.5 b	2.0 b
tillers/row (%)	Diseased	57.0 a	69.8 a	77.5 a
Yield (g/plot)	Healthy	119.9 a ^z	140.8 a	131.5 a
rield (g/ prot)	Diseased	111.4 a	91.6 b	102.4 b
Yield (kg/ha)	Healthy	3,363 a	3,948 a	3,693 a
11010 (116)	Diseased	3,128 a	2,569 b	2,872 b
Yield loss (%)		7.1	34.9	22.1

^yNumbers are mean of 12 replicates (each plot was one row 2 m long).

P. graminis and WSSM development. In central Georgia, the mean monthly temperatures are 7.3, 6.8, and 6.9 C and mean monthly rainfall is 11.4, 11.8, and 11.9 cm for December, January, and February, respectively. These temperatures are favorable for P. graminis growth and the infection of wheat (16). Rainfall of more than 11 cm/mo ensures sufficient soil moisture for the dissemination of zoospores under near-soilsaturation conditions. Indeed, WSSM was observed in most wheat fields in low areas where soil saturation occurred during rainy periods and in fields that had continuous wheat production. A similar situation has been documented with takeall on wheat, which has become a widespread problem in the Southeast as a result of continuous wheat culture (13).

The overall incidence of the virus in Georgia must be determined to assess its potential impact. The survey in the spring of 1984 showed that incidence of WSSMV varies in different fields from high (10-20% infected) to no plants infected. This varied incidence is to be expected because environmental factors affect both the fungus and WSSMV development. Furthermore, the soil types are variable in much of the Southeast,

varying from heavy clay to sandy loam and from relatively level fields on the Coastal Plain to rolling hills on the Piedmont. Therefore, if susceptible cultivars are grown more extensively, as continuous wheat production on land with poor drainage, a much greater yield loss may be observed.

The susceptibility of the wheat cultivars grown in the Southeast to WSSMV needs to be determined. If the virus does become more prevalent and causes a greater economic loss, a good source of resistance will be important.

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Paired means with the same letter are not significantly different (P = 0.05) as determined by the paired t test.