

# Maize Bushy Stunt and Corn Stunt: A Comparison of Disease Symptoms, Pathogen Host Ranges, and Vectors

L. R. Nault

Professor, Department of Entomology, Ohio Agricultural Research and Development Center (OARDC), Wooster 44691.

The technical assistance of William Styer, Jane Buth Todd, and Marian Coffey and the supply of seed for host range studies by H. H. Iltis,

University of Wisconsin and S. Pandey, CIMMYT, Mexico City, Mexico, are sincerely appreciated. Suggestions for improvement of

the manuscript by R. E. Davis, D. T. Gordon, and L. E. Williams are gratefully acknowledged. Approved for publication by Associate

Director, OARDC, as Journal Series Article 167-79.

Accepted for publication 10 December 1979.

## ABSTRACT

NAULT, L. R. 1980. Maize bushy stunt and corn stunt: A comparison of disease symptoms, pathogen host ranges, and vectors. *Phytopathology* 70:659-662.

The corn stunt spiroplasma (CSS) and maize bushy stunt mycoplasma (MBSM) are transmitted by *Dalbulus maidis*, *D. elimatus*, and *Graminella nigrifrons*; however, CSS is, but MBSM is not, transmitted by *Exitianus exitiosus* and *Stirellus bicolor*. The mean latent period for MBSM in *D. maidis* is  $24.2 \pm 1.9$  days and for CSS  $19.0 \pm 2.0$  days both at  $27 \pm 2$  C.

Teosinte (*Zea mays mexicana*) is a host for both CSS and MBSM, but *Z. perennis* and *Z. diploperennis* are hosts only for CSS. The mean latent period for MBSM in sweet corn is one-third to one-half the latent period

for CSS. Maize bushy stunt mycoplasma causes more severe stunting and proliferation of axillary and basal shoots than does CSS. At high temperatures, CSS (but not MBSM) causes chlorotic spots and stripes on leaves. At low temperatures chlorotic spots and stripes may not develop, and CSS may produce symptoms resembling those caused by MBSM. Evidence suggests that MBSM is similar, if not identical, to the "Mesa Central" and "Louisiana" strains of corn stunt.

Two distinct mycoplasmas are associated with corn stunting diseases. One of the mycoplasmas, the Rio Grande strain of corn stunt (13), forms motile helical filaments and has been called a spiroplasma (3,8,9). The corn stunt spiroplasma (CSS) has been cultured in vitro and its causal relationship to Rio Grande corn stunt has been demonstrated (5,19). The second mycoplasma is nonhelical (1,2,4) and produces symptoms in maize (*Zea mays* L.) similar to those reported for the "Mesa Central" (13) and "Louisiana" (12) strains of corn stunt. This latter mycoplasma has been designated the maize bushy stunt mycoplasma (MBSM) (4,15). The vernacular, achaparramiento-M, has recently been used for this pathogen in Mexico (1). Tetracycline antibiotics delay symptom development in plants and interfere with leafhopper transmission (1,2). Proof of its pathogenicity and causal relationship to Mesa Central and Louisiana corn stunts awaits culture of the pathogen.

In this paper I compare the symptomatology, host plant range, and leafhopper transmission of CSS and MBSM.

## MATERIALS AND METHODS

Plants infected with CSS were obtained from R. E. Davis (USDA-SEA-AR, Beltsville, MD) in 1972. This isolate has been cultured (5) and deposited in the American Type Culture Collection (ATCC 29051). The principal isolate of MBSM used in this study was obtained from southern Texas in 1975 (4). Other isolates observed for symptom expression were obtained from Peru in 1978 (16) and again from Texas in 1979 (Fig. 1A). *Dalbulus maidis* (DeLong & Wolcott) was obtained from H. N. Pitre (Mississippi State Univ., MS), *D. elimatus* (Ball), *Exitianus exitiosus* (Uhler) and *Stirellus bicolor* (Van Duzee) from R. F. Whitcomb (USDA-SEA-AR, Beltsville, MD), and *Graminella nigrifrons* (Forbes) and *Macrostes fascifrons* (Stol) were collected from grasses near Wooster, OH. Leafhoppers were reared in organically-covered aluminum framed cages  $19 \times 38 \times 38$  cm. A colony was started by infesting host plants in 10-cm-diameter pots with 100-200 adults per plant. The host plant for *D. elimatus* and *D. maidis* was maize. *Graminella nigrifrons*, *E. exitiosus*, and *M. fascifrons* were reared on oats (*Avena sativa* L.) and maize and *S. bicolor* was reared on rye (*Secale cereale* L.). Adults were removed from plants after an egg laying period of 4-7 days. This procedure

produced leafhoppers of nearly uniform age. Temperature of the rearing room was 24-29 C.

For transmission studies, leafhoppers were given a 4-day acquisition access period (AAP) on infected plants followed by a 17-day incubation period (IP) on healthy plants. Insects were then caged 10 or three per test plant for a 7-day inoculation access period. To determine the latent period of mycoplasmas in *D. maidis*, leafhoppers were exposed to corn infected with MBSM and CSS for a 48-hr AAP. Leafhoppers were then transferred to healthy corn plants for 10-day IP. Then 25 surviving leafhoppers from each group were selected and placed singly on Oh43 corn test plants in the two-leaf stage. Each leafhopper was transferred daily to a healthy test plant for 20 transfers or until death. Leafhoppers were held at  $27 \pm 2$  C throughout this experiment. Test plants were placed in a greenhouse and observed regularly for up to 6 wk after exposure to leafhoppers.

To determine latent periods and symptoms of mycoplasmas in maize, sweet corn test plants (cultivar Aristogold Bantam Evergreen) were exposed to inoculative leafhoppers for 48 hr. These plants exposed while in the two-leaf stage were then transplanted to 20- or 25-cm-diameter clay pots. These were then placed in a walk-in plant growth chamber set for 16 hr of light per 24-hr day with a temperature of 27 C (day) and 18 C (night) or 31 and 25 C and 65% RH. The light intensity in the chamber was 37,660 lux (3,500 ft-c). Plants were observed daily until well-formed ears had developed or the plants had died. Symptoms in the sweet corn Golden Cross Bantam and the inbred field corns Oh28 and Oh43 also were observed.

To determine host ranges of the mycoplasmas, six to 10 plants of each test species were exposed to 10 inoculative *D. maidis* for a 7-day inoculation access period. Test plants were observed in a greenhouse for 8 wk. Plants with disease symptoms were assayed for presence of CSS or MBSM by *D. maidis* transmission to sweet corn.

## RESULTS

**Leafhopper transmission.** *Dalbulus maidis*, *D. elimatus*, and *G. nigrifrons* transmitted MBSM but *E. exitiosus*, *S. bicolor*, and *M. fascifrons* did not (Table 1). *Dalbulus maidis*, *D. elimatus*, *E. exitiosus*, *G. nigrifrons*, and *S. bicolor* transmitted CSS but *M. fascifrons* did not. This is the first report of *E. exitiosus* and *S. bicolor* as vectors of CSS and confirms the status of the *Dalbulus* spp. and *G. nigrifrons* as CSS and MBSM vectors (14).

The mean latent period for MBSM in *D. maidis* was  $24.2 \pm 1.9$  days with a range of 22–28 days for nine individuals. The mean latent period for CSS in *D. maidis* was  $19.0 \pm 2.0$  days with a range of 17–23 days for 20 individuals.

**Latent period and symptomatology of mycoplasmas in corn.** The mean latent period for MBSM in Aristogold Bantam Evergreen sweet corn varied from 9.5 to 18.9 days, depending on temperature. First symptoms were a chlorosis of margins of whorl leaves followed by reddening of the tips of older leaves. Whorl leaves and all subsequently developing leaves showed varying degrees of marginal chlorosis, reddening, tearing, twisting, and shortening. Numerous tillers developed at leaf axils and plant bases (Fig. 1B, Table 2). Infected plants were stunted and developed numerous small ears (Table 2). Root systems of diseased plants were less extensive than were those of healthy plants. At high temperatures, plants died before maturity. Death of plants may not have been due solely to presence of MBSM, but also to soilborne pathogens that attack weakened plants.

In the sweet corn Golden Cross Bantam, and the inbreds Oh28 and Oh43 red coloration was absent and the number of basal and axillary tillers was fewer than that for Aristogold Bantam Evergreen. Symptoms observed for the Texas isolates of MBSM, collected in 1975 and 1979, were very similar to those observed for two Peruvian isolates (16).

The mean latent period for CSS in corn was shorter in plants held at higher temperatures (Table 2). At the high temperature regime, chlorosis of leaf margins was the first symptom observed and that was followed by reddening of older leaves. The red coloration was never as extensive as that observed in plants infected with MBSM. First symptoms were followed 2–4 days later by small chlorotic spots that developed at the bases of newly developing leaves. In successive leaves above those bearing first symptoms, the chlorotic spots coalesced to form chlorotic stripes that extended toward the leaf tips until entire leaves were affected (Fig. 1C). Plants were stunted and numerous ear shoots developed (Table 2). At the low temperature regime, symptoms were expressed by a yellowing and reddening at the bases of upper leaves (Fig. 1D). Diagnostic chlorotic spots and stripes did not always develop.

**Host range of mycoplasmas.** In addition to maize, three races of

TABLE 1. Leafhoppers tested as vectors and plants tested as hosts for maize bushy stunt mycoplasma (MBSM) and corn stunt spiroplasma (CSS)

Leafhopper species and plant hosts tested	MBSM	CSS
<b>Leafhoppers<sup>a</sup></b>		
<i>Dalbulus maidis</i>	24/27 <sup>b</sup>	31/31
<i>D. elimatus</i>	10/17	16/20
<i>Exitianus exitiosus</i>	0/38	22/28
<i>Graminella nigrifrons</i>	4/22	4/20
<i>Stirellus bicolor</i>	0/27	11/18
<i>Macrostelus fascifrons</i>	0/47	0/30
<b>Plant species<sup>c</sup></b>		
<i>Zea mays</i>	10/10	10/10
<i>Z. mays mexicana</i>	15/15	15/15
<i>Z. perennis</i>	0/10	2/10
<i>Z. diploperennis</i>	0/10	5/10
<i>Tripsacum lanceolatum</i>	0/6	0/6
<i>Coix lachryma-jobi</i>	0/6	0/6
<i>Sorghum halepense</i>	0/6	0/6
<i>S. vulgare</i>	0/6	0/6
<i>Vicia faba</i>	0/10	0/10
<i>Vinca rosea</i>	0/10	0/10

<sup>a</sup>Leafhoppers were given a 4-day acquisition access period followed by a 17-day incubation period. Insects were then caged 10 per test plant (except for *S. bicolor* and *E. exitiosus* at three per plant) for a 7-day inoculation access period. Controls were leafhoppers from stock colonies placed on sweet corn test plants to check for presence of pathogens. None transmitted CSS or MBSM during these investigations.

<sup>b</sup>Numerator is number plants infected; denominator is number plants exposed.

<sup>c</sup>Test species were each exposed to 10 inoculative *D. maidis* for a 7-day inoculation access period.

Mexican teosinte, *Z. mays* L. *mexicana* (Schrad.) Iltis, were susceptible to MBSM (Table 1). Maize bushy stunt symptoms in two collections of Central Plateau teosinte were stunting, chlorosis, splitting of leaves, and some tillering, but no leaf reddening. Symptoms in two collections of Chalco teosinte were: profuse tillering; extensive chlorosis; no reddening; and (in a Balsas teosinte) considerable tillering, chlorosis, and extensive reddening of leaves.

In addition to maize and the three races of teosinte, *Zea perennis* (Hitchcock) Reeves & Mangelsdorf and *Z. diploperennis* Iltis, Doebley & Guzman were susceptible to CSS. Diagnostic spotting and striping were seen in *Z. mays mexicana* and *Z. perennis*, but not in *Z. diploperennis*. Symptoms in the later species were stunting and reddening of leaves. Maize bushy stunt mycoplasma and CSS were recovered by *D. maidis* from all test species showing the respective symptoms of each pathogen.

## DISCUSSION

Maize bushy stunt mycoplasma and CSS produce distinctly different diseases in maize. Symptoms of MBSM are very similar to those previously reported for the Mesa Central (13) and Louisiana (12) strains of corn stunt. A discussion of these similarities was presented previously (15). These findings also parallel those recently published for Mesa Central corn stunt in Mexico (1,2).

The effects of temperature on latent periods and symptoms for MBSM and CSS offer some insight into reports on the distribution of corn stunt in Mexico. Maramorosch (12) noted that Mesa Central corn stunt occurred principally in the central plateau region, whereas Rio Grande corn stunt was found only at low elevations. In surveys for corn stunt in Mexico, Davis (6,7) observed maize with Rio Grande corn stunt symptoms at low and middle altitudes but not at high elevations. Microscopic examination of plants with these symptoms consistently revealed the presence of spiroplasmas. At middle and high elevations plants with Mesa Central corn stunt symptoms were observed; a few of these contained spiroplasmas. This anomaly may be due to the effects of lower temperatures at high altitudes on CSS symptom development. At low temperatures the latent period for CSS is greatly prolonged and symptoms are a diffuse chlorosis and reddening at the bases of upper leaves (Fig. 1D), symptoms which could mistakenly be attributed to Mesa Central corn stunt. In addition, disease development due to MBSM may be favored at high altitude over that caused by CSS in doubly infected plants.

Both CSS and MBSM have restricted host plant ranges within the Maydeae infecting only members of the genus *Zea*, but not *Tripsacum* and *Coix* or other Gramineae. The perennial teosintes, *Zea perennis* and *Z. diploperennis* were the only differential hosts found, both being susceptible to CSS but not MBSM. I did not confirm the earlier finding that two dicots, *Vinca rosea* and *Vicia faba* are susceptible to CSS (14). However, Markham et al (14) used *Euscelidius variegatus* as a vector, a species which unlike *D. maidis*, readily feeds on dicots.

*Stirellus bicolor* and *E. exitiosus* are differential vectors that

TABLE 2. Latent periods of maize bushy stunt mycoplasma (MBSM) and corn stunt spiroplasma (CSS) and their effects on corn growth<sup>a</sup>

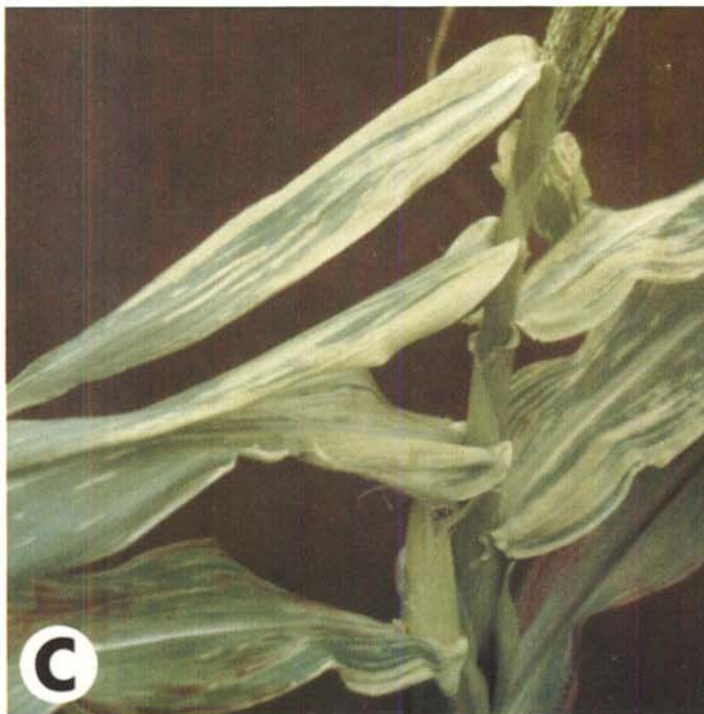
Temperature <sup>b</sup> (day/night)	Treatment	Latent period (days)	Plant ht. <sup>c</sup> (cm)	Number axillary <sup>c</sup> and basal shoots
27/18	MBSM	$18.9 \pm 1.3$	$50.8 \pm 6.5$	$7.3 \pm 1.1$
	CSS	$43.3 \pm 4.1$	$141.6 \pm 12.6$	$3.6 \pm 1.0$
	Control		$212.0 \pm 13.0$	$1.6 \pm 0.7$
31/25	MBSM	$9.5 \pm 0.9$	... <sup>d</sup>	... <sup>d</sup>
	CSS	$16.3 \pm 1.1$	$104.5 \pm 10.9$	$6.1 \pm 1.7$
	Control		$180.4 \pm 17.4$	$2.5 \pm 1.0$

<sup>a</sup>Values are averages and standard deviations from 8–10 plants for each treatment.

<sup>b</sup>Temperatures of walk-in growth chamber during 16-hr photophase/8-hr scotophase.

<sup>c</sup>Plant height and shoot number measured at plant maturity.

<sup>d</sup>Plants died before maturity.



**Fig. 1.** Symptoms of maize bushy stunt mycoplasma (MBSM) and corn stunt spiroplasma (CSS). **A**, MBSM-infected field corn, McAllen, TX, May 1979. **B**, MBSM-infected sweet corn grown in environmental chamber (see low-temperature regime, Table 2). **C**, CSS-infected sweet corn (see high-temperature regime, Table 2). **D**, CSS-infected sweet corn (see low-temperature regime, Table 2).

transmit CSS but not MBSM. The use of these leafhoppers as vectors or the perennial teosintes as hosts should prove useful in differentiating CSS and MBSM in doubly infected field-collected plants. Unfortunately, I found no selective vector or host that could be used to definitively identify MBSM from CSS in such mixed infections.

The potential for CSS and MBSM spread from the southern USA to the Corn Belt is limited by the narrow plant host ranges of these pathogens and probably does not depend heavily upon distribution and abundance of vectors. While the *Dalbulus* spp. are neotropical and limited in their distributions in this country (10) *S. bicolor*, *E. exitiosa*, and particularly *G. nigrifrons*, occur throughout the Corn Belt and are abundant (17,18). Should either CSS or MBSM adapt to a commonly occurring perennial host such as johnsongrass, *Sorghum halepense* L., these mycoplasmas could become a serious threat to northern corn-growing regions. Johnsongrass serves as the overwintering host for maize chlorotic dwarf virus and five of six strains of maize dwarf mosaic virus, causal agents of the two most serious viral diseases of maize in the USA (11).

#### LITERATURE CITED

1. BASCOPE, B. 1977. Agente causal de la llamada "raza mesa central" del achaparramiento del maiz. Maestro en Ciencias Thesis, Escuela Nacional de Agricultura, Chapingo, Mexico. 55 pp.
2. BASCOPE, B. and J. GALINDO. 1978. Mycoplasmic nature of the "Mesa Central" corn stunt. Page 78 in: Third Int. Cong. Plant Pathol., Munich, W. Germany. 16-23 August 1978. (Abstr.) Deutsch. Phytomed. Gesellschaft.
3. BOVE, J. M., and C. SAILLARD. 1979. Cell biology of spiroplasmas. Pages 83-153 in: R. F. Whitcomb and J. G. Tully, eds. The Mycoplasmas III, Plant and Insect Mycoplasmas. Academic Press, New York, NY. 351 pp.
4. BRADFUTE, O. E., L. R. NAULT, D. C. ROBERTSON, and R. W., TOLER. 1977. Maize bushy stunt—a disease associated with a non-helical mycoplasma-like organism. (Abstr.) Proc. Am. Phytopathol. Soc. 4:171.
5. CHEN, T. A., and C. H. LIAO. 1975. Corn stunt spiroplasma: isolation, cultivation and proof of pathogenicity. Science 188:1015-1017.
6. DAVIS, R. E. 1974. Occurrence of spiroplasma in corn stunt-infected plants in Mexico. Plant Dis. Rep. 57:333-337.
7. DAVIS, R. E. 1977. Spiroplasma: Role in the diagnosis of corn stunt disease. Pages 92-98 in: Proc. Maize Virus Dis. Coloq. and Workshop, 16-19 August 1976. Ohio Agric. Res. and Dev. Center, Wooster. 145 pp.
8. DAVIS, R. E. 1979. Spiroplasmas: Newly recognized arthropod-borne pathogens. Pages 451-484 in: K. Maramorosch and K. Harris eds. Leafhopper Vectors and Plant Disease Agents. Academic Press, New York, NY. 654 pp.
9. DAVIS, R. E., and J. F. WORLEY. 1973. Spiroplasma: motile, helical microorganism associated with corn stunt disease. Phytopathology 63:403-408.
10. DeLONG, D. W. 1950. The genera *Balbulus* and *Dalbulus* in North America including Mexico (Homoptera: Cicadellidae) Bull. Brooklyn Entomol. Soc. 45:105-113.
11. GORDON, D. T., and L. R. NAULT. 1977. Involvement of maize chlorotic dwarf virus and other agents in stunting diseases of *Zea mays* in the United States. Phytopathology 67:27-36.
12. GRANADOS, R. R., K. MARAMOROSCH, T. EVERETT, and T. P. PIRONE. 1966. Transmission of corn stunt virus by a new leafhopper, *Graminella nigrifrons* (Forbes). Contrib. Boyce Thompson Inst. 23:275-280.
13. MARAMOROSCH, K. 1955. The occurrence of two distinct types of corn stunt in Mexico. Plant Dis. Rep. 39:896-898.
14. MARKHAM, P. G., R. TOWNSEND, K. PLASKITT, and P. SAGLIO. 1977. Transmission of corn stunt to dicotyledonous plants. Plant Dis. Rep. 61:342-345.
15. NAULT, L. R., and O. E. BRADFUTE. 1979. Corn stunt: Involvement of a complex of leafhopper-borne pathogens. Pages 561-586 in: K. Maramorosch and K. Harris, eds. Leafhopper Vectors and Plant Disease Agents. Academic Press, New York, NY. 654 pp.
16. NAULT, L. R., D. T. GORDON, R. E. GINGERY, O. E. BRADFUTE, and J. CASTILLO LOAYZA. 1979. Identification of maize viruses and mollicutes and their potential insect vectors in Peru. Phytopathology 69:824-828.
17. PITRE, H. N., and L. W. HEPNER. 1967. Seasonal incidence of indigenous leafhoppers (Homoptera: Cicadellidae) on corn and several winter crops in Mississippi. Ann. Entomol. Soc. Am. 60:1044-1055.
18. STONER, W. N., and R. D. GUSTIN. 1967. Biology of *Graminella nigrifrons* (Homoptera: Cicadellidae), a vector of corn (maize) stunt virus. Ann. Entomol. Soc. Am. 60:495-505.
19. WILLIAMSON, D. L., and R. F. WHITCOMB. 1975. Plant mycoplasmas: a cultivable spiroplasma causes corn stunt disease. Science 188:1018-1020.