

Temperature Tolerance and Survival of *Ceratocystis fagacearum* in Texas

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ABSTRACT

Lewis, R., Jr. 1985. Temperature tolerance and survival of *Ceratocystis fagacearum* in Texas. Plant Disease 69:443-444.

Isolates of *Ceratocystis fagacearum* from Texas and North Carolina grew most rapidly at 22–26 C and survived without growth for 10 days at 34 C. Some isolates survived for 3 hr at 45 C, but all were dead after 4, 24, and 48 hr, respectively, at 45, 42, and 37 C. Sapwood temperatures of *Quercus virginiana*, healthy and affected by oak wilt, were compared with ambient temperature for 2 days in July in Kerrville, TX. During the hottest part of the day, sapwood temperatures were lower in healthy than in diseased trees and root collar temperatures were lower than those of the trunk at 1.4-m height or of ambient air. Sapwood temperatures in the root collar of a healthy tree remained at 24.5 C when ambient temperature surpassed 36 C. Air temperatures in Texas in summer permit survival of *C. fagacearum*, and sapwood temperatures in the lower trunk and roots of healthy trees permit its growth.

Ceratocystis fagacearum (Bretz) Hunt, which causes oak wilt, is inhibited by high temperatures (13,15) but is well established in central Texas, where maximum daytime temperatures in summer may exceed 40 C (1,9). The fungus was cultured from a red oak in Dallas in 1961 (4), but doubt about its ability to survive periods of high temperature in Texas was expressed in 1975 (16). Isolates of *C. fagacearum* from more northerly areas survive for only a short period and are difficult to culture from host tissues when temperatures are 30–35 C (2,5). Because of its sensitivity to high temperature, *C. fagacearum* was believed to be excluded from most of the South (13,15,16). Surveys showed little or no southward movement of oak wilt during the 1960s and 1970s (12,13); however, recent surveys confirmed epiphytotic oak wilt in central Texas (1). The purpose of this report is to explain how *C. fagacearum* is able to survive and grow in Texas when summertime temperatures exceed its presumed tolerance level.

MATERIALS AND METHODS

Six isolates of *C. fagacearum* from Texas and one from North Carolina (ATCC 27789) were used in growth and survival tests at different temperatures. One Texas isolate, collected by Dooling in 1961 (4), was maintained on potato-dextrose agar (PDA) in a refrigerator at

5–10 C during 1964–1980. The remaining Texas isolates were obtained from *Quercus virginiana* Mill. or *Q. shumardii* var. *texana* (Buckl.) Ashe during 1979–1981. All isolates were grown on PDA in petri dishes at 26 C for 10 days before experiments.

Growth of various isolates was compared at different temperatures. Agar disks (5 mm in diam.) with mycelium and conidia were removed from the outer 2 cm of colonies of *C. fagacearum*, placed on PDA in the centers of petri dishes, sealed in plastic bags, and incubated in darkness at 18, 22, 26, 30, and 34 C. Each dish was sealed with laboratory film. Nine dishes of each isolate were incubated at each temperature. After 10 days, colony diameters were measured.

Similar agar disks were used to test fungal survival at 37, 42, and 45 C for various periods of time. After exposure, the dishes were transferred to a chamber at 26 C and incubated in darkness for 10 days. The percentage of disks from which *C. fagacearum* grew at 26 C was recorded for each isolate. Five disks in each of nine petri dishes were evaluated for each isolate.

Xylem temperatures in one healthy and three diseased live oaks were measured at 0800–0900, 1330–1430, and 1630–1730

hours CDT on 18 and 19 July 1978. The three diseased trees were in the advanced phase of oak wilt (10), showing sparse foliage and dieback. Temperatures were measured in holes 50 mm deep × 5 mm in diameter drilled at 1.4 m above ground level and at the root collar on the north, east, south, and west sides of each tree. The holes were drilled through bark that was about 10 mm thick. The electronic probe of a portable Tele-thermometer was inserted in the base of each hole. The same holes were used each time temperatures were measured. Ambient temperatures were measured concurrently.

RESULTS

All seven isolates of *C. fagacearum* grew at 18, 22, 26, and 30 C but not at 34 C. There was variation in growth of different isolates and growth at different temperatures. Texas isolates grew fastest and equally well at 22 and 26 C (Table 1). Least growth occurred at 30 C.

When dishes were removed from the 34-degree chamber after 10 days and placed on a bench at 25–29 C, *C. fagacearum* grew from 57% of the agar disks containing Texas isolates and from none of the disks containing the North Carolina isolate. Survival beyond 10 days at 34 C was not tested.

Fungal survival was affected by both temperature and exposure time. None of the seven isolates grew after exposure to 45 C for 4 hr, 42 C for 24 hr, or 37 C for 48 hr. The North Carolina isolate was apparently killed during just 2 hr at 45 C, but none of the Texas isolates died during this time. Even after 3 hr at 45 C, three of the six Texas isolates were still alive in some of the agar disks. *C. fagacearum* was still alive in about one-third of the disks after either 4 hr at 42 C or 24 hr at 37 C (Table 2).

Xylem temperatures were lowest and most stable at the root collar of the healthy tree and highest and most variable at 1.4-m height in the trunks of

Table 1. Radial growth of Texas and North Carolina isolates of *Ceratocystis fagacearum* on potato-dextrose agar during 10 days at different temperatures

Temp. (C)	Colony diameter (mm) ^a							Mean
	DA ^b	KE	SA	BA	FR	LA	NC	
18	43.0	35.5	29.6	25.8	30.2	15.7	18.5	28.34
22	70.9	52.7	42.5	39.2	39.1	32.8	34.5	44.50
26	75.0	50.5	40.4	39.6	37.1	28.8	45.5	45.30
30	33.8	22.8	21.3	16.3	11.8	3.4	8.5	16.80

^a Average in nine petri dishes.

^b Texas isolates: DA = Dallas, KE = Kerrville, SA = San Antonio, BA = Bandera, FR = Fredericksburg, and LA = LaGrange. North Carolina isolate: NC = ATCC 27789.

Accepted for publication 1 December 1984.

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Table 2. Survival of Texas and North Carolina isolates of *Ceratocystis fagacearum* on potato-dextrose agar at different temperatures

Temp. (C)	Exposure time (hr)	Isolate survival (%) ^a							
		DA ^b	KE	SA	BA	FR	LA	NC	Mean
37	24	30.0	46.9	30.0	43.1	46.9	34.2	26.7	36.8
42	4	13.1	46.9	25.8	46.9	30.0	36.8	46.9	35.2
45	1	90.0	73.1	90.0	76.9	90.0	90.0	75.0	83.6
45	3	0.0	0.0	16.9	0.0	13.1	17.7	0.0	6.8

^a An arc sine transformation of the percent survival for *C. fagacearum* mycelium and conidia on 45 potato-dextrose agar disks.

^b Texas isolates: DA = Dallas, KE = Kerrville, SA = San Antonio, BA = Bandera, FR = Fredericksburg, and LA = LaGrange. North Carolina isolate: NC = ATCC 27789.

Table 3. Xylem temperatures at 50-mm depth in a healthy live oak and in three oaks affected by oak wilt in Kerrville, TX, during 18 and 19 July 1978

Measurement site	Tree condition	Temperature (C) ^a		
		0800-0900 hours	1330-1430 hours	1630-1730 hours
Root collar	Healthy	24.5	24.7	24.4
	Diseased	25.1	27.8	27.9
		25.9	28.3	29.4
		26.2	29.2	28.7
1.4-m Height	Healthy	26.1	27.3	27.7
	Diseased	29.3	32	33.3
		27.6	31.9	34.7
		28.1	31.8	33.6
Ambient air	25.8	35.5	36.1	

^a Average of measurements on the north, east, south, and west sides of the tree on two consecutive days.

diseased trees (Table 3). Root collar temperatures in the healthy tree remained near 24.5 C during each of the three daily periods when temperatures were measured. Temperatures were highest at 1630-1730 hours, when average maxima were 24.4, 27.7, 29.4, 34.7, and 36.1 C, respectively, in healthy root collar, healthy trunk, diseased root collar, diseased trunk, and ambient air. During the hottest part of the day, xylem temperatures at 1.4-m height in diseased trees were similar to those of ambient air. Root collar temperatures in these same trees were lower than ambient, however, and healthy tree temperatures were lower than those in diseased trees.

DISCUSSION

The optimum temperature for growth of Texas isolates of *C. fagacearum* was within the range reported for northern isolates. Northern isolates grow best at 22-25 C, with little or no growth at 30-32 C (11,15). The Texas isolates grew best at 22-26 C, with moderate to little growth at 30 C. Texas isolates survived without growth for 10 days at 34 C.

Texas isolates and northern isolates are similar in their tolerance to high temperatures. Northern isolates survived less than 3 hr in wood blocks at 49 C (6), less than 2 hr as conidia at 40 C (3), and

less than 3 days at 37 C (11). Some Texas isolates in this study survived for 3 hr at 45 C and 24 hr at 37 C. Air temperatures of 45 C for 3 hr or 37 C for 24 hr are not likely to occur in the South. Therefore, summertime temperatures are not high enough to eradicate *C. fagacearum* in Texas as previously suggested (16).

Temperatures suitable for growth and survival of *C. fagacearum* occur in the root collar and lower bole of infected oaks during the hottest periods of summer in Texas. Oak wilt infections are favored by moderate (24-28 C) temperatures during spring and autumn (7,8,10), but they occasionally develop during the summer in Texas when maximum daytime temperatures exceed 37 C. Xylem temperatures in the lower part of the bole of the healthy tree remained within the optimum range for both fungal growth and wilt development.

Sapwood temperatures were higher and less favorable for growth of *C. fagacearum* in the three diseased trees than in the healthy tree when the weather was hot. This was presumably due to a reduction in the flow of water through diseased sapwood and a correlated reduction in the cooling effects of the transpiration stream. Sapwood temperatures at the 1.4-m level were higher and less favorable for fungal growth than

those at the root collar in both healthy and diseased trees during the two hottest periods of the day. During summer in Texas, it is difficult to isolate *C. fagacearum* from twigs taken from trees with oak wilt (1,8,9). During this period, xylem temperatures in the upper crown might favor microorganisms other than *C. fagacearum*.

In the southern United States, a high-temperature boundary does not appear to exist for oak wilt. I believe the disease can develop at any place in the South where susceptible oaks grow. Tainter and Gubler (14) suggested that temperature alone does not limit the spread of oak wilt in Arkansas. Instead, competing microorganisms, especially *Hypoxylon* spp., may be responsible for slowing its spread.

ACKNOWLEDGMENT

This research was supported in part by the U.S. Department of Interior through the Lyndon B. Johnson National Historic Park, Johnson City, TX.

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