STUDIES ON THE EPIDEMIOLOGY OF PECAN SCAB CAUSED BY *CLADOSPORIUM CARYIGENUM* IN OKLAHOMA

By

ALINA MARIA MARENCO

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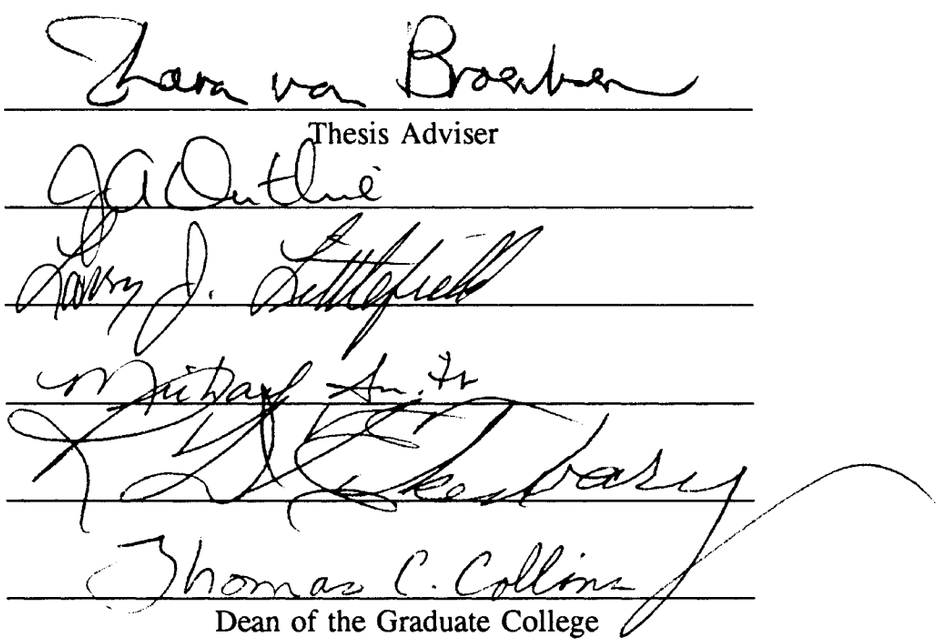

Thesis Adviser

Dean of the Graduate College

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CHAPTER I

INTRODUCTION

The pecan, *Carya illinoensis* (Wang.) K. Koch is a crop regarded native to the United States, especially to the riverbottom forests of the Mississippi drainage system (Brison, 1974). In Oklahoma there are about 48,432 acres of pecans under production, of which over about 37,384 acres are native pecans; the rest are improved cultivars (U. S. Department of Commerce, 1978).

The pecan scab disease caused by *Cladosporium caryigenum* (Ell. et Lang.) Gottwald, is the most important disease of pecans nationwide. Large amounts of fungicides are used to control this disease. At present, the pecan industry faces economic problems due to the increased costs of fungicides and other chemicals. Approximately 50% of variable production costs are invested in pest control, making it difficult for pecan growers to balance production costs and benefits.

To produce a profitable crop, prevent damage to the environment, and avoid development of resistance of *Cladosporium caryigenum* to the fungicides now available through excessive or exclusive use, fungicides should be applied only when needed. The current practice of applying fungicides according to a fixed spray schedule often results

in more applications than necessary. The obstacle yet to overcome is to identify what environmental factors are optimum for development of this disease in Oklahoma under field conditions.

Field research on how pecan scab develops under natural conditions and how this relates to weather factors such as relative humidity, temperature, and leaf wetness have been lacking over the years. Although some of these relationships have been investigated under laboratory conditions, it is necessary to determine if the field situation corresponds to such findings. In addition to field trials investigating the development of the disease, studies on the effect of temperature on the growth and sporulation of different Oklahoma isolates of *Cladosporium caryigenum*, and the inhibition by different fungicides is appropriate.

Determination of what factors play an important role in development of disease could be used to develop a threshold for the application of fungicides. Preliminary data from field experiments could serve as the foundation for further studies. The ultimate goal would be to produce a forecasting model that allows prediction of weather conditions favorable for development of scab. Such a system would reduce production costs, facilitate a low input type production, and help protect the environment.

Studies concerning inhibition of growth of *Cladosporium caryigenum* by fungicides could be important in revealing whether the scab fungus has developed some degree of

resistance to any of the fungicides tested, information which would be important when recommending treatment applications to growers. Finally, the growth and sporulation studies give us some insight into the nature of Oklahoma isolates of the fungus and how these factors may relate to disease development in the field.

CHAPTER II

TAXONOMY OF THE PECAN SCAB FUNGUS

The pecan scab fungus was first described in 1885 by G. Winter as *Fusicladium effusum* (Demaree,1928). In 1888, Ellis and Everhart announced the description of a new fungus *Fusicladium caryigenum* Ell. et Lang., on leaves of *Carya olivaeformis* Nutt. [= *C. illinoensis*] (Gottwald, 1982). However, Orton (1905) reported that *F. caryigenum* and *F. effusum* Wint. were the same.

Demaree (1928) proposed the reclassification of the fungus as a member of the form genus *Cladosporium* with the name *C. effusum*, comb. nov., based on the catenulate arrangement of the conidia, a character which excludes the fungus from the form genus *Fusicladium* (Demaree, 1928).

When Hughes (1953) divided Hyphomycetes into eight sections, he placed *Cladosporium* in section IA that contained "those genera with conidia usually developing in acropetal succession as blown out ends at the apex of simple or branched conidiophores which do not then increase in length". *Fusicladium* was placed in section II that contained "genera with conidia arising as blown-out ends of apex of simple or branched conidiophores and the ends of successively produced new growing points developing to one side of the previous conidium" (Hughes, 1953).

Lentz stated that the production of conidia in chains was not acceptable as the sole basis for excluding the pecan scab fungus from *Fusicladium* (Lentz, 1957). He believed that the catenulate arrangement of conidia had less significance than an indeterminate conidiophore (Gottwald 1982). Lentz concluded that the pecan scab fungus should be called *Fusicladium effusum* Wint.

Baron (1968) extended and discussed in detail the Hughesian system. He placed *Fusicladium* in the group Sympodulosporae whose members never produce conidia in chains, and *Cladosporium* in the series Blastosporae whose members produce conidia forming acropetal chains or occurring singly.

Gottwald (1982), in his examinations of numerous specimens from the National Fungus Collection, concluded that the pecan scab fungus could not be included in the Sympodulosporae because of its copious production of conidial chains as opposed to *Fusicladium*'s single-producing conidia characteristic. Also other characteristics, such as the presence of intercalary conidiogenous cells, ramoconidia, variability of conidium shape, formation of acropetal chains of blastoconidia, indicated closer natural relationships to members of *Cladosporium*-like fungi. Gottwald also used epidemiological considerations to further demonstrate the relationships between the pecan scab fungus and *Cladosporium*, and finally determined that the pecan scab fungus belongs to the genus *Cladosporium* as originally proposed by Demaree in 1928 (Gottwald, 1982). Nevertheless

it can not be called *C. effusum* as this name was used before Demaree, to describe a different fungus, at the present known to belong to *Cercospora* (Lentz, 1957). Therefore Gottwald proposed the new combination based on the synonym *F. caryigenum* as *Cladosporium caryigenum* (Ell. et Lang.) Gottwald comb. nov.

DISEASE CYCLE AND OVERWINTERING SITES

Demaree (1924) described the symptoms caused on pecan by the scab fungus *Cladosporium caryigenum*. He reported spots of irregular and undefined outline, olive brown in color, turning to almost black with age (as spores are washed away); mainly affecting young growing tissues of leaves, nuts and current year's twigs. Leaves become resistant with age. He reported pedicel infection as an important factor related to nut death, because girdling of pedicels disrupts the flow of water and nutrients to the nuts. Infected nuts are smaller in size, defective or made useless. Diseased tissue turns black, becomes hardened and has a cracked appearance. Twig dieback is observed only on current year's growth. Catkins and dormant buds are seldom infected.

Gottwald and Bertrand (1982) studied spore dispersal in the field. They determined that sporulating lesions are dark brown to black with a velvety appearance, and once they degenerate and become old, have a gray flat appearance. They found that periods of rapid decrease in vegetative wetness played an important role for spore dispersal of *C.*

caryigenum (= *F. effusum*) in the orchard. They concluded that it is not rainfall but reductions in vegetative wetness immediately after rain or dew that intensifies aerial conidia concentration of *C. caryigenum* (= *F. effusum*). Conidia are primarily dispersed by wind, but rainsplash is an important factor to restricted inoculum dispersal (Gottwald,1985).

Under laboratory conditions, Gottwald (1982) demonstrated that spore release of *C. caryigenum* decreases with RH above 40% but is stimulated when relative humidity is less than 40% and also by quick changes in RH.

Mode of infection and establishment of parasitism were studied by Latham and Rushing (1988), who observed that 83.2% of conidia had germinated 12 hr after inoculation. Some germinating conidia produced an appresorium adjacent to the conidium, while others produced an appresorium distally. They suggest this could be due to the occurrence of physiological races. Penetration of host cuticle was achieved by 62.2% of the germ tubes at 48 hr after inoculation. *C. caryigenum* grows subcuticularly. All hyphal growth is intercellular, restricted to the middle lamella of neighboring epidermal cells. Conidiation occurs 7-8 days after inoculation (Latham and Rushing,1988,1991).

Demaree (1928) cited as overwintering sites and sources of primary inoculum, scab lesions on twigs, nuts that have been destroyed by the disease, empty shucks and leaves, those lesions on the mid-veins of pinnae, rachises and petioles which produce the overwintering stromata.

WEATHER FACTORS AND THE DEVELOPMENT OF FUNGICIDE APPLICATION THRESHOLDS

Understanding how weather factors lead to production and dispersal of *C. caryigenum* conidia and subsequent disease development is of utmost importance to better timing of fungicide applications (Latham, 1982).

There have been a number of reports on the influence of weather on pecan scab. In 1956, Converse reported 25°C as the optimum temperature for germination in the laboratory, and also found that no germination occurred at or below 90.1% relative humidity.

Valli (1964) pointed out that cold, dry weather was unfavorable for the conidiation and infection process, as well as for the production of the young tissue needed for infection. He also reported, from other sources, that the optimum temperature for germination was 23.9°C (75°F), leading to infection after 6 hours of wet vegetation.

Gottwald (1984) reported the optimum temperature range for infection to be 20 to 30°C, even though notable infection still occurred in the 10 - 35°C range. He also found that a minimum of 2 hr of leaf wetness was required for infection, but maximum infection was achieved after 36 hr of continuous wetness. These studies were conducted in growth chambers under controlled environmental conditions.

Pecan scab lesions were first observed 7 - 9 days after inoculation, in greenhouse studies (Gottwald, 1985; Latham, 1982). Increase in length of leaf wetness periods leads to increase in lesion numbers. Gottwald (1985) reported

maximum lesion production at 20°C after a wetness period of 32 to 48 hours, and at 25°C after a wetness period of 48 hours with few lesions and little disease during wetness periods of less than 9 hours.

Frequency of fungicide application will depend on the scab pressure, which in turn is influenced by weather (Hargrove, Balsdon, Pickering, 1991). Valli (1964) provided a quick method of estimating probability of scab infection by multiplying the wetting periods in hours by the mean temperature in degrees centigrade during the wetting period. If the result is 140 or above, scab infection is likely to occur. The effectiveness of this method has not been evaluated.

Hunter (1978) declared that to develop an effective spray program based on weather data we must be able to predict the development of the disease in relation to weather conditions. He stated that under saturated atmosphere, the optimal temperature for spore production was 25°C (77°F) with noticeable numbers of spores being produced at 20°C (68°F). Based on correlation of the effectiveness of past spray schedules and weather data, Hunter postulated that 100 hours of leaf wetness was conducive to development of scab on the susceptible 'Schley' cultivar. This served as the foundation for the 100 hours leaf-wetness approach that was used as a guide in 1977 at Byron, Georgia, to start fungicide application.

McVay and Gazaway (1980) concluded that the monitoring of leaf wetness could be a positive asset for the pecan

grower. In their experiments, monitoring of leaf wetness periods and timing of fungicide applications only when indicated appeared to be superior to the fixed spray program for the control of pecan scab. They arbitrarily used 120 hours of leaf wetness as a threshold for each fungicide application.

The Texas Pecan Integrated Pest Management Manual (Cooper et al., 1983) recommended spray applications based on 100 accumulated hours of 90 % relative humidity used as a threshold level. They arrived at this figure based upon work done in Georgia, Alabama and Texas. However, they also said this monitoring system does not fit for areas where there is infrequent occurrence of 90% or greater relative humidity.

Cooper (1984) used a similar system as a pest management treatment where fungicides were applied if 100 hours of 90% or greater relative humidity had been accumulated in the 21.1 - 29.4°C range and two weeks had elapsed since the previous fungicide application.

In spite of the above, Latham (1982) does not agree with forecasting theories on setting 100 to 120 cumulative hours of leaf wetness as a threshold for fungicide applications. His data showed that scab developed on test trees 7 - 9 days after a rainy day when the leaf wetness periods were only 12 - 16 hours.

Hargrove et al. (1990) tested a 16-hour leaf wetness scab spray rule in eight different sites in Georgia. They proved this was more effective than fixed sprays in reducing

both the number of spray applications and excessive or unnecessary use of chemicals.

Clearly there are major differences in the thresholds proposed. This situation probably developed because these thresholds were obtained from data collected under controlled conditions that may not hold true in the field or by using the weather parameters recorded between sprays in a particular schedule that controlled scab. No data are available on the relationship between the various weather parameters with potential to be used in developing thresholds and actual disease evolution in the field.

DISEASE RATING METHODS FOR PECAN SCAB

Horsfall and Barratt (1945) developed a scoring system to record severity of plant diseases. They selected 50% as a midpoint, since "below 50%, the eye sees the amount of diseased tissue, while above 50%, it sees the amount of diseased free tissue". This system has been modified for use with pecan scab.

According to Bertrand and Gottwald (1986), Phillips et al. (1952) made a pictorial representation of five categories of pecan scab infection on nuts. Currently the two main systems used to evaluate pecan diseases are the Hunter and Roberts system, based on Phillips' illustration, and the modified Horsfall-Barratt system.

Both visual systems estimate percentage of diseased tissue by separation into different categories depending on the actual disease present on nuts and leaves. The

difference between them is that there are more infection level categories in the modified Horsfall-Barratt that may allow the user to register differences in amount of disease present during the later stages of scab disease.

In the field, leaves with more than 50% scabbing often defoliate, but scabbed nuts generally remain on trees. The Horsfall-Barrat system is especially useful for evaluating nut damage in high scab situations.

FUNGICIDES IN PECAN SCAB CONTROL

The amount of fungicides used annually in pecans places this crop as the third largest user of fungicides in the United States after peanuts and deciduous fruits (Littrell and Bertrand, 1981). Diverse studies on fungicide performance in controlling pecan scab have been carried out through the years. Cole (1964) reported that prior to 1940 lime and lime sulfur combinations were used to control scab. During the 1940's the use of carbamates began, and they were used until becoming ineffective. Along with these changes, spraying equipment also improved. By the 1980's a number of additional fungicides were available for management of pecan diseases including dodine, benomyl, and triphenyltin hydroxide. All of these have certain limitations (Littrell and Bertrand, 1981). Dodine is not particularly effective. Resistance to benomyl has forced abandonment of this fungicide in the southeast U.S. The tins may be lost because of environmental and worker safety concerns.

More recently a new class of fungicides, referred to as

sterol inhibitors has been discovered to be effective in controlling pecan scab (Littrell, 1983). One of these , propiconazole, has been registered for pecans. This fungicide can be applied up to 5 days after inoculation and still completely control scab (Latham, 1983). Another sterol inhibiting fungicide, fenbuconazole, is also expected to be registered for the 1995 growing season. In addition to their limited systemic activity and their low non-target toxicity, sterol inhibiting fungicides offer the possibility of post-treatment after key periods of weather favorable for infection.

However the potential for build up of resistance to sterol-inhibiting fungicides also exists, since it has been demonstrated under laboratory conditions that sterol-inhibiting-fungicide- resistant fungal mutants can be easily obtained (Littrell, 1983). In a crop which is as intensely sprayed with fungicides as pecans, the danger of fungicide resistance is always a threat. *Cladosporium caryigenum* has shown the ability to develop fungicide-resistant strains. This was first proved in Georgia, where benomyl-tolerant isolates were obtained; and in every case where tolerance was found, use of Benlate^{R} (benomyl) had been frequent or exclusive as the only control for scab in leaves (Littrell and Lindsey, 1976).

Littrell and Lindsey (1977) indicated that benomyl-tolerant strains survive in nature as well as the benomyl-sensitive and also maintained virulence. The effect of

benomyl-tolerant strains in treatments containing Benlate^{R} in the spray program was evident. In the case of Benlate^{R}, resistance occurred after less than three years of use. The product was registered for use in 1973, and in 1975 tolerant isolates were found in Georgia.

Benomyl resistance has not been documented in Oklahoma, and growers still rely heavily on this fungicide. Benomyl has not been used as intensively in Oklahoma and is used in rotation with other fungicides, especially tins. Barnes (1967) showed that benomyl is a valuable tool in Oklahoma to extend spray interval to 3-4 weeks.

Effective use of fungicides in disease management requires knowledge of the mechanisms of pathogen survival, the sources of primary inoculum, and how secondary spread occurs. With proper timing of fungicide applications by means of a forecasting system based on weather or anticipation of infection periods (Littrell and Bertrand, 1981), the number of fungicide applications might be reduced and the selection pressure thus decreased (Littrell, 1983).

HOST RESISTANCE AND PHYSIOLOGICAL SPECIALIZATION OF THE PECAN SCAB FUNGUS

Use of resistant cultivars is one of the methods used to control diseases. Pecans show a wide range of response to scab, varying from susceptible to moderately susceptible to moderately resistant to resistant (Cooper et al., 1983).

Wetzstein and Sparks (1983) made studies to correlate structural characteristics of pecan cultivars with

resistance. They found higher contents of phenolic compounds in mesophyl tissues and bundle sheath cells of immature leaves of resistant cultivars than those of susceptible cultivars. Low trichome density was correlated with resistance, as well as collapsed, desiccated trichomes and less size variation .

It has been shown in in vitro studies that juglone was fungitoxic to *C. caryigenum* at levels of 0.05 mg/g of liquid culture. The possible role of juglone in disease resistance was studied by Borasjani, Graves and Hedin (1985). They found juglone in twigs stayed consistently low throughout the season and that concentrations were higher in whole ground tissues such as leaflets and shucks. They proposed that the role of juglone in resistance depends on its presence and availability at or near infection sites.

Tannin and isoquercitrin also have been studied. They inhibit the in vitro growth of *C. caryigenum*. Laird, Graves and Hedin (1990) studied the influence of condensed tannin and isoquercitrin from pecan on growth of selected isolates of *C. caryigenum*. They observed different abilities of the isolates to tolerate inhibitory levels of both compounds which might affect isolate prevalence on a given host phenotype.

The factors involved in resistance toward scab disease are not totally elucidated or completely understood. Breeding for resistance in pecans faces many problems including: 1) the high degree of genetic diversity and adaptability of the scab fungus requires a wide range of

fungal isolates representative of the pecan belt be used in screening potentially resistant host plants, 2) the lack of correlation between resistance in leaf and nut tissues and differences in susceptibility between young and older tissues (Graves, 1986), and 3) the more resistant cultivars often lack one or more desirable horticultural characteristics (Hunter, 1977).

Demaree (1929) reported that the scab fungus attacked certain cultivars as 'San Saba', 'Georgia', 'Delmas', but not others such as 'Schley', 'Van Deman', 'Pabst', even when planted in the same locality or continuous rows. Furthermore, he observed that a particular cultivar that was susceptible in one district could be resistant in another. He attributed this to the existence of physiological forms of the pecan scab fungus. He carried out inoculation experiments and found that when inoculating to the original host, percentage of infection was higher than when inoculating to hosts of other cultivars, showing in this manner the variability in adaptability of forms of the pathogen to some cultivars and the probable existence of physiologic specialization.

In greenhouse studies in Oklahoma, Converse (1960) separated three physiological races of the scab fungus from the 'Squirrel', 'Western' and 'Sovereign' cultivars on the basis of pathogenicity to four pecan cultivars. He carried out tests with resistant and susceptible pecan leaf discs. He observed that diffusible substances inhibit conidial growth differentially, and he associated that with

resistance. Since then no more studies of this type have been done in Oklahoma.

Latham (1976) pointed out how the then recently introduced pecan cultivars, 'Cherokee' and 'Wichita', showed more severe symptoms than already established cultivars and pointed out the necessity for evaluating cultivars for resistance to scab before being released and distributed.

Hunter, et. al. (1986) observed how resistance to scab is overcome by the scab fungus, as happened with 'Stuart', widely planted in the early 1900's because it was considered resistant to scab. In 1956 'Stuart' was reported susceptible in Mississippi and now is proving so in most of the southeastern United States. In Oklahoma 'Stuart' still has some level of resistance.

CHAPTER III

STUDIES ON THE RELATIONSHIP BETWEEN WEATHER FACTORS AND DISEASE DEVELOPMENT IN THE FIELD

MATERIALS AND METHODS

A) Field Site and Material

The relationship between weather factors and development of pecan scab was studied at the Pecan Research Station, near Sparks, Lincoln County, Oklahoma during the growing seasons of 1993 and 1994.

The orchard consisted of about half the scab susceptible 'San Saba Improved' pecan cultivar, grafted to native rootstocks, and half native pecan trees. The 'San Saba Improved' trees were 43-years-old at the end of these studies. The age of the natives was unknown but varied. 'San Saba Improved' trees were arranged in rows running in a diagonal from southeast to northwest. Natives were randomly distributed within the orchard. The soil was a Port silt loam (fine-silty, mixed, thermic; Cumulic Haplustoil; mollisols). In the 'San Saba Improved', white clover (*Trifolium repens* L.) was the ground cover. This helped to maintain nitrogen availability as well as encourage beneficial insects. The ground cover in the native pecan trees was mowed grass sod. The site was selected for study

because of its central position in the Oklahoma pecan production area and because there was a history of recurring pecan scab in the orchard. The disease existing in this orchard occurred naturally; no artificial inoculations where involved. No fungicide treatments were applied. During 1994 trees were treated with chlorpyrifos (Lorsban^{R}) to control insects and with zinc to prevent deficiency. Those two chemicals were applied on May 12, 1994.

B) Rating Method and Ratings

Two terminal branches and two fruit clusters located on the southwest side of each tree were tagged with metal tags and plastic flagging tape on each of ten 'San Saba Improved' trees. The same number of samples was similarly tagged in the native trees.

The rating method used was the modified Horsfall and Barratt System (Horsfall and Barratt, 1945) that estimates percentage of diseased tissue by separation into eight categories depending on the actual diseased area present on fruit and leaves (Table I). Observations and ratings of fruit and leaves were taken weekly. Three compound leaves of each tagged branch were rated for disease symptoms. The first, second, and third leaves were rated during 1993, and the second, third and fourth during 1994. The reason for this change was that very often the first leaf is incompletely developed and may drop prematurely. Defoliation was recorded once leaflets started to drop, and

TABLE I

THE MODIFIED HORSFALL - BARRATT SYSTEM FOR RATING SCAB DISEASE ON PECAN FOLIAGE AND NUTS (HORSFALL, J. F. AND BARRATT, R. W. 1945. PHYTOPATHOLOGY 35:655)

Rating Category	% Diseased Area
1	0%
2	trace - 6 %
3	6 - 25 %
4	25 - 50 %
5	50 - 75 %
6	75 - 94 %
7	94 - 99 %
8	100 %

throughout the season, until the entire leaf was gone; however, the number of leaflets lost per week was not recorded.

The number of fruit present was recorded at the beginning of the sampling procedure. As the season progressed and fruit fell from the cluster, the number remaining in each cluster was recorded weekly. Disease of each individual fruit was rated according to the modified Horsfall and Barratt Method (Horsfall and Barratt, 1945). The fruit rating order was acropetal.

Disease ratings of tagged leaves and fruit were averaged for each observation day, and the results graphed for both 'San Saba Improved' and native pecans. Then, differences between average disease ratings on each of the sampling dates were calculated to show the change in disease severity over time. The change in disease severity over time was then graphed against key periods of independent weather variables (temperature, relative humidity, leaf wetness). The number of fruit dropped throughout the season was also averaged and the results graphed. The defoliation data was treated differently, since the exact amount of defoliation was not recorded, a scale was assigned as follows: 0 = entire leaf present, 1 = one or more leaflets absent, and 2 = entire leaf absent, which gave an index of defoliation.

The same sampling procedure was used during 1993 and 1994, except that fruits were not available during 1994 in the 'San Saba Improved' trees. Trees were rated during 1993

from May 28 to August 27 and during 1994 from May 16 to August 22.

C) Data Logger

A data logger (Campbell Scientific, Inc. Logan, UT), consisting of a CR10 Measurement and Control System configured with 29,900 data points of storage memory, powered by internal 12V DC batteries was used to collect and store the weather data. Three sensors were included in this system: (1) A temperature and relative humidity probe, model #207, placed in a 40 micrometer screen mesh housing. The probe was covered by a radiation shield and placed below a tree canopy about 1 m from the trunk and 1.3 m above ground. Both sensors were factory calibrated. (2) A rain gauge, model # TE525, adapted from the National Weather Service's "tipping bucket rain gauge", was placed about 1.8 m above ground. The unit measured each tipped cup occurring at each 0.01 inch (0.25 mm) of rain. Calibration was made by pouring 16 fl. oz (500 ml) of water into the tipping cup and getting 100 tips. (3) A leaf wetness sensor model #237, an electrical resistance measurement sensor with interlacing gold plated "fingers", was situated in the lower part of the tree canopy. This sensor measured the decrease of resistance between the "fingers" due to condensation.

The data logger was installed at a site at the center of the orchard. Temperature (°C), relative humidity (%), and leaf wetness (%) were measured every 60 seconds, and recorded as an average every hour. Rainfall was recorded as

the total rainfall collected whenever it occurred.

Data obtained from the data logger were transferred onto tapes using a cassette recorder. The data were converted into ascii and coma delineated files by means of a tape reader computer program. The coma delineated files were imported to the Lotus program, where data were arranged per day. The maximum, minimum and average daily temperatures were calculated, as well as the total number of hours of relative humidity > 90%, hours of leaf wetness > 25%, and the total rainfall per day. A day ran from the 0:00 A.M. hour through the 23:00 P.M. hour. In the secondary analysis, the average temperature, total hours of relative humidity > 90% and leaf wetness > 25% were calculated for the 7 day, 14 day and 14 to 7 day periods previous to each observation, and graphed against changes in disease ratings, in an attempt to identify key weather periods favorable to disease development.

RESULTS

A) Disease Ratings

During 1993, foliar disease appeared early in the season on the 'San Saba Improved' cultivar, and increased consistently throughout the season to a maximum 6.34 rating on August 27 (Fig. 1). In the native pecans very little disease was seen at the beginning of the season and continued steadily so throughout the season, with a maximum of 2.69 on August 27 (Fig. 1). Heavy defoliation was

observed in 'San Saba Improved', very early in the season, suggesting that this resulted from the scab disease. As foliar disease began to increase more rapidly on June 24 (Fig. 1) defoliation also increased (Fig. 2), especially during July 1 to July 9 where the defoliation index jumped from 0.4 to 1.1 during that period. No more major changes occurred after July 9.

Levels of disease for fruit reached higher levels than for leaves, in both 'San Saba Improved' and natives during 1993 (Fig. 3). Disease in 'San Saba Improved' fruit began to appear around mid June with a sharp increase starting July 9. The maximum rating for fruit in 'San Saba' was 8. For natives disease remained low with a maximum rating of 3.35 obtained on August 12 (Fig. 3). The number of fruit per cluster was recorded at the beginning of the trial; fruit started to drop very early in the season in both 'San Saba Improved' and natives, but in higher amounts in the 'San Saba Improved'. The number of fruit per cluster fell from 2.55 to less than 1.0 during 1993, for the 'San Saba Improved' (Fig. 4). The size of the fruit was very small and the crop was completely lost. Fruit of the native pecans attained fairly good sizes, although the number of fruit/cluster fell throughout the season from 3.4 to 1.0 at the end of the season (Fig 4).

During 1994, disease was already present by May 16. The disease appeared earlier in native pecans than in the 'San Saba Improved' cultivar, because budbreak in 'San Saba Improved' was delayed in 1994. On June 6 disease started to

increase more rapidly in the 'San Saba Improved'; a sharp increase occurred during the week beginning July 18 (Fig. 5). In the natives foliar disease remained at a consistently low level during the growing season. The maximum foliar rating was 2.13 by July 25, while 'San Saba Improved' reached a maximum of 3.74 by August 1. Nevertheless, the level of disease was not as high as in 1993. The defoliation index obtained this year was lower than the previous, with 1.2 the maximum obtained for the 'San Saba Improved', and approximately 0.5 for native pecans at the time the last observation was recorded (August 22) (Fig. 6).

For the 1994 season there was no flowering in 'San Saba Improved' and fruit set did not occur in these trees. In the native pecans disease in fruit started to increase sharply by July 18; the highest disease rating obtained was 3.21 by August 22 (last observation day) (Fig. 7). The size of the cluster fell from 3.4 fruit/cluster at the beginning of the season to 1.9 by August 22 (Fig. 8).

B) Weather

In 1993, average daily temperatures ranged from 15°C to 32.5°C during the rating period (Fig. 9). The maximum daily number of hours of relative humidity > 90% was 24 hr. (Fig. 10). The maximum number of daily hours of leaf wetness during the season was 17 hr (Fig.11). Total rainfall is shown in Fig. 12. For the 1994 season, the average daily temperatures ranged from 10°C to 30.9°C (Fig. 13). With

respect to percentage of relative humidity, there was some erroneous data due to sensor degradation. The maximum number of daily hours of leaf wetness was 18 (Fig. 14). Total rainfall is given in Fig. 15.

In general, weather factors seemed to be more conducive to disease during 1993 than during 1994.

C) Correlation Between Weather and Changes in Disease.

To evaluate how weather parameters affect disease development, only the data from the highly susceptible 'San Saba Improved' cultivar were considered in detail as the disease response increase there was sufficient to allow correlation of the effect of weather on disease. Although individual native pecan trees differ in their ability to resist scab, overall they were able to maintain disease at low levels, resulting in low disease ratings.

Average temperature, hours of relative humidity > 90% and hours of leaf wetness > 25% occurring 0 - 7, 7 - 14, and 0 - 14 days previous to each disease rating observation were graphed against the change in disease ratings for leaves and fruit. A curvilinear type of relationship was obtained for the weather variable temperature and disease change in leaves (Fig. 16) but not in fruit. The data did not show any relationships between relative humidity or leaf wetness and change in disease in leaves and fruit. During the 1994 growing season, the apparent relationship between temperature and disease change in leaves was not evident because of low levels of disease.

DISCUSSION

The susceptibility of the 'San Saba Improved' cultivar to scab was evident. Disease appeared early and increased evenly in these trees, most probably due to their genetic uniformity. In the natives, disease remained at a lower level in both leaves and fruit. Disease was not uniform from tree to tree due to the varied levels of resistance associated with a genetically diverse population. Resistance is an inherent characteristic in native pecans that makes them suitable for production with a low input type of management. These are important factors for growers to consider when planning their orchards and type of production.

fruit were also more affected in the 'San Saba Improved' cultivar than for natives even though fruit disease ratings were higher than foliar disease ratings in both 'San Saba Improved' and native pecans. This lack of correlation of resistance between leaves and fruit was pointed out by Hunter and Roberts (1977), and it represents a problem when breeding for scab resistance.

'San Saba Improved' fruit were affected at a very early stage of development, the reason why they remained small and never developed and why the crop was lost. In the native fruit, disease advanced slowly early in the season. This in addition to their inherent resistance allowed them to pass the critical young stage at which infection occurs, therefore attaining good sizes. Even though resistance is

an important characteristic to take into account for controlling scab disease, as demonstrated by the natives, some cultivars which are more susceptible are favored by the growers due to one or more desirable horticultural characteristics.

It is especially important to identify the threshold favorable to disease development in fruit and leaves at the early stages of development when there is much young, susceptible tissue to infect. As evident in the graphs (Figs. 1, 3, 5,), there is a critical time beyond which, if disease is not controlled, and under the right weather conditions, disease becomes uncontrollable in the susceptible cultivar. If this period could be identified, then chemical control measures could be aimed at this critical initial period favoring disease. In this matter the Oklahoma Mesonet project could be very helpful as a source of weather data throughout the state.

The fruit drop pattern observed in 1993 was similar to that observed by Hunter (1983). A steady fruit drop during May and June that, according to Hunter, has been referred to as the "first" and "May" drops, was followed by a period when there was little fruit drop during most of July. Fruit drop was resumed in late July, and continued until the end of the season. This last drop from July 29 onward (Fig. 4) was due to the scab disease. During the period July 9 to July 22, even when no fruit drop occurred, disease was increasing rapidly during the same interval until reaching a point where fruit were lost to disease (Fig. 3, 4). Hunter

(1983) showed that there is a direct correlation between grade of disease severity and fruit drop.

The high degree of defoliation observed in the 'San Saba Improved' and not in the natives where disease remained low suggests that this is a result of the scab disease. Early defoliation in 'San Saba Improved' prevented the trees from storing enough carbohydrate reserves, which affected flowering the following year, and no fruit were set during 1994 for this reason. Latham (1979) indicated the importance of leaf retention well into the fall to secure a good fruit set the following year. It is therefore important to control scab disease and avoid leaving orchards unmanaged once disease is present, to prevent early defoliation and subsequent loss of the following year's crop.

It is known that symptoms appear 7 to 9 days after infection (Latham, 1982; Gottwald, 1985). Since germination and ingress must occur before that, it was hypothesized that weather conditions occurring 14 to 7 days previous to the appearance of symptoms would be most important.

There seemed to be a relationship between temperature and change in disease in leaves (Fig. 16) which can be compared to the growth response of *Cladosporium caryigenum* to temperature under laboratory conditions. In the leaves the peak of this curve occurs around 27°C, which was also the optimum temperature for growth in the laboratory experiments (see Chapter IV). Frequent increases in disease

ratings occurred when the average temperature was between 23°C to 28°C. (Fig. 16). Nevertheless this relationship was not evident during 1994, which suggests that other factors were influencing change in disease during 1993 in the temperature range favorable for growth of the scab fungus, conditions that were not met during 1994, as demonstrated by the lower levels of disease (Fig. 5).

The data did not suggest any type of relationship between relative humidity or leaf wetness and disease development. By themselves none of these parameters was conducive to disease. Data in these graphs were randomly scattered, and there was no observable pattern that could give some information on how increase in hours of relative humidity > 90% or leaf wetness > 25% influence increase or change in disease.

Moisture is an important factor for spore germination. It is known from laboratory studies that *C. caryigenum* does not germinate at or below 90.1% relative humidity (Converse, 1956). This is the reason why the hours of relative humidity > 90% were chosen as a threshold for this study. It might be that relative humidity and temperature are important weather factors that interact to produce the right conditions conducive to development of pecan scab disease, an interaction that can not be adequately described at this preliminary stage of research.

There might have been several reasons to account for the apparent lack of correlation between weather factors and change in disease: (1) limitations in the method of

analysis, (2) the fact that the independent weather variables do not cause increase in disease by themselves but by the interaction among each other, and (3) errors introduced due to the sampling method, equipment and range of the parameters considered.

(1) Limitations in the method of analysis

The data have been analyzed in a linear fashion, but from the temperature graph vs disease change in leaves in 1993, it was observable that a curvilinear type of relationship might explain better the interaction occurring, even though the possibility exists that the observed effect might not be real. A better approach to take in the future would be to observe the data and evaluate the results using selected statistical models that might better describe the relationship occurring in the field. At this stage it is premature to do this. More locations as well as several years of observations would be needed to consolidate the data and choose the appropriate model to analyze the existing relationships in greater depth.

(2) Change in disease may be caused by the interaction of the dependent variables

Change in disease severity in this study has been evaluated and graphed against each independent weather variable separately (temperature, relative humidity, leaf wetness). These separate graphs limit the amount of information that can be extracted and applied to the field,

since in the field, change in disease is the result of interactions among the different weather factors influencing the pathogen and the host.

(3) Errors introduced due to methodology and equipment.

Although the most accurate disease estimations possible were made utilizing the Horsfall and Barratt rating system, this method is limited by the potential for human error in these estimations. Also this method lacks enough different categories in the early stages of disease to register changes in disease severity occurring at these stages. Other researchers have suggested that relative humidity > 90% might be important in the field for development of disease; therefore in the present study, as designed from its onset, only the hours of relative humidity > 90% were recorded. There was degradation of the sensitivity of the relative humidity sensor during the 1994 season, making data analysis difficult. Furthermore the figure of leaf wetness > 25% was chosen arbitrarily, and may not be an adequate threshold.

All these considerations should be taken into account for further studies.

CHAPTER IV

STUDIES ON THE EFFECT OF TEMPERATURE ON *C. CARYIGENUM* ISOLATES AND ON RESPONSE OF DETACHED LEAVES TO INOCULATION

1- The effect of temperature on the growth of *Cladosporium caryigenum* isolates

MATERIALS AND METHODS

Isolations were made from thirteen foliar samples collected during 1993 from several cultivars and from different locations in Oklahoma. The resulting isolates are given in Table II. Small pieces of tissue were placed on malt extract agar (MEA) to obtain the isolates. The MEA consisted of 15 g of agar and 10 g of malt extract dissolved in distilled water to make one liter. The isolates were grown in pure culture on MEA and then stored at room temperature in vials containing the same medium.

For the growth studies, isolates were cultured on MEA prior to the beginning of the experiment to have actively growing cultures. Disposable 100 x 15 mm petri dishes containing 20 ml of medium were marked with 2 perpendicular lines forming a cross. A number 2 cork borer was used to cut out pieces of agar from the outer edge of the colony of each isolate. This piece of agar was placed in the center of the petri dish containing MEA, at the junction of the

TABLE II

CULTIVARS AND PLACES OF ORIGIN
OF ISOLATES COLLECTED

Isolate Designation	Cultivar	Location Collected
CheO4	'Cheyenne'	Orsicana, TX
SSp1	'San Saba Improved'	Sparks, OK
SqRR1	'Squirrel's Delight'	Red River, OK
CCh1	'Colby'	Chetopa, KS
NRR	Native (I)	Red River, OK
NBF1	Native	Ada, OK
NV1	Native	Vinita, OK
PP2	'Peruche'	Ponca, OK
SAd1	Unknown	Adair, OK
WS1	'Western'	Sparks, OK
N2RR1	Native (II)	Red River, OK
BRR2	'Burkett'	Red River, OK
CwM1	'Cowley'	Miami, OK

cross. The temperatures used for these studies were 15°C, 18°C, 21°C, 24°C, 27°C, 30°C. Every isolate had triplicates (3 plates) at each incubation temperature. All plates were placed at the various temperatures in different incubators at the same time.

Plates were observed and the radial growth marked at four and seven days. Only in plates incubated at 15°C and 18°C were further observations made after twelve and fourteen days. Growth in plates was measured with a centimeter scaled rule. The distance from the center of the inoculum plug along the four pre-marked lines to the outer edge of the colony was measured for each isolate. The resulting four measurements of radial growth were averaged for each plate and the rate of radial growth per day was calculated for each plate. This experiment was conducted in both 1993 and 1994. The statistical analysis was done as a nested design with two treatment factors where temperature is crossed with year. The level of significance used was P = 0.05.

RESULTS

Growth occurred through the range of temperatures used in the experiment (Fig. 17). The lowest growth rate overall occurred at 15°C. As the temperature increased, growth rate increased, until reaching the apparent optimum of 27°C. Although growth was slower at 30°C, considerable growth still occurred. A similar trend was observed in the second experiment (Fig. 17), but this time growth at 24°C was

similar to growth at 30°C.

Average growth per day at 27°C was 0.52 cm/day in 1993, and 0.54 cm/day in 1994 (Fig. 17). The lowest growth rate was at the lowest temperature 15°C, with 0.24 cm/day and 0.26 cm/day in the 1993 and 1994 experiments, respectively. Growth at the highest temperature studied 30°C, was 0.50 cm/day and 0.49 cm/day, respectively.

Statistical analysis of the data for 1993 revealed that there was significant difference at P = 0.05 on the growth occurring at the different temperatures, except between growth at 27°C and 30°C. However, during 1994 there was a significant difference between the growth at 27°C and the growth at 30°C, although there was no significant difference between growth at 24°C and 30°C.

DISCUSSION

The isolates were collected from various geographic locations and different cultivars in an attempt to include as much genetic variation as possible. *Cladosporium caryigenum* grew at all the temperatures used (15°C, 18°C, 21°C, 24°C, 27°C and 30°C). The more limiting temperatures for growth were 15°C and 18°C. Nevertheless, in spite of the fact that there was no significant difference between growth at 27°C and 30°C in 1993, and between 24°C and 30°C in 1994, the temperature of 27°C may be regarded as the optimum temperature for growth from the point of view that it was the temperature at which the highest average growth was observed for both years.

Previously the range of temperatures between 20° - 30°C has been found to be optimum for other biological processes of *C. caryigenum*. Converse (1956) reported that the optimum temperature for germination was 25°C. Hunter (1978) observed that noticeable sporulation occurred in the range of 20° - 30°C, and that 25°C was the optimum for spore production. Gottwald (1984) reported the range of 20° - 30°C as optimum for infection by *C. caryigenum*, although he stated that considerable infection could occur at 10° - 35°C. All these experiments were carried out under controlled environmental conditions with no information about fluctuating temperatures, as would be experienced in the field. In the current experiments, the scab fungus was able to grow throughout the range tested. From the data above it could be said that the range of 20° - 30°C is a favorable range for *C. caryigenum* to carry out most of its physiological processes.

II- Effect of Temperature on Sporulation of Isolates of *C. caryigenum*.

MATERIALS AND METHODS

After two weeks of growth at six temperatures (15°C, 18°C, 21°C, 24°C, 27°C and 30°C), plates from the growth studies (see above) were examined under the microscope using a 4x objective, and were visually rated for the amount of sporulation according to the following scale: 0 = none, 1 = poor, 2 = low, 3 = medium and 4 = high. Each set of triplicate plates for each isolate and temperature was rated, averaged and the results graphed.

Single plates in each category (1 - 4) were randomly chosen from the different temperatures, regardless of the isolate, making a subset of 69 plates total. The spores on these plates were counted to determine the accuracy of the visual rating system. A SPotlite^{R} hemacytometer, with a 0.100 mm depth and a "V" load system (Baxter Healthcare Corporation) was used. For this purpose 5 ml of distilled water was poured onto each plate, the colony was scraped with a glass rod to dislodge spores, and aliquots of the resulting spore suspension were counted. The counts were compared with the visual ratings.

The experiments studying effect of temperature on sporulation of the scab fungus were carried out twice. The results were analyzed statistically using a nested design with two treatment factors where temperature is crossed with year. The level of significance used was P = 0.05. The

hemacytometer spore count data were not statistically analysed.

RESULTS

During the 1993 experiment, the average sporulation rating increased as temperature increased, reaching an optimum at 27°C, and was only slightly less at 30°C, the upper limit temperature used (Fig. 18). Statistical analysis revealed that 27°C was the optimum temperature for sporulation ($P = 0.05$). The results obtained in the second experiment were erratic and therefore discarded. For example, sporulation ratings were higher at the lowest temperature than at other temperatures that are more appropriate for growth and sporulation of *C. caryigenum*.

However, a more important problem was that the hemacytometer counts to test the reliability of the visual estimation of sporulation method showed that visual estimation was unreliable. There was substantial variation in the actual amount of spores/ml for plates within the same category (poor, low, medium or high) (Table III).

DISCUSSION

Although the 1993 sporulation experiment using the visual rating method appeared to show that good sporulation was obtained at 27°C (Fig. 18), the hemacytometer counts showed that visual estimation of sporulation was unreliable. One factor that might have prevented an accurate estimate was that sometimes conidial chains where growing very close

TABLE III

HEMACYTOMETER COUNTS OF SPORES IN PLATES PREVIOUSLY RATED WITH THE VISUAL ESTIMATION METHOD

Isolate	Temperature	#Spores x 10^4/ml
POOR RATING		
SQRR1	15	17.5
CCH1	15	8.5
WS	18	5.2
N2RR1	18	3.1
NRR	18	5.4
SAD	18	0.8
NBF	18	2.8
CWM1	18	3.5
PP2	30	24.1
Average		7.9
Range		0.8 - 24.1
LOW RATING		
WS	15	11.2
SSP1	15	12.0
BRR2	15	32.4
NV1	15	12.6
SQRR1	18	10.2
CHEO4	18	29.4
PP2	18	9.3
N2RR1	21	13.6
SAD	21	6.1
NRR	24	15.6
NBF	24	7.9
CWM1	24	9.3
Average		14.1
Range		6.1 - 32.4
MEDIUM RATING		
BRR2	18	33.8
CCH1	18	32.8
SQRR1	21	11.3
WS	21	21.4
SSP1	21	16.1
CHEO4	21	17.8
NV1	21	22.3
NRR	27	14.4
SAD	27	34.2
NBF	27	26.6
PP2	27	59.3

TABLE III (Continued)

Isolate	Temperature	#Spores x 10^4/ml
CWM1	30	11.7
Average		25.1
Range		11.3 - 59.3
	HIGH RATING	
NV1	24	26.5
CCH1	24	27.3
N2RR1	24	17.2
SQRR1	27	19.6
SSP1	27	56.7
BRR2	27	62.9
WS	30	17.3
CHEO4	30	55.3
Average		35.3
Range		17.3 - 62.9

together and in other instances chains were fairly well distributed on the plate. Assessment of sporulation should therefore be done by other methods rather than by visual estimation. A better method to follow in the future would be the counting of spores by means of the hemacytometer, even though this is very time consuming. Also better control of the relative humidity might help to produce more consistent results.

III- Differential Response of Detached Leaves of 2 Cultivars to Inoculation with Three Isolates of *C. caryigenum*

MATERIALS AND METHODS

To determine if detached leaves could be used to study if different isolates caused different levels of infection, a preliminary inoculation procedure using detached leaves was attempted. Three isolates were used: CheO4, NV1, CCh1, (see Table II) and two susceptible pecan cultivars 'Burkett' and 'Squirrel's Delight'. Spores suspensions were prepared as described above except 3 ml of sterile distilled water were added to each plate containing an isolate. Suspensions were adjusted to make a total volume of 10 ml of each isolate. The concentrations which resulted were as follows: CheO4 - 4.13 X 10^4 spores/ml, NV1 - 8.04 X 10^4 spores/ml, and CCh1 -7.2 X 10^4 spores/ml.

Detached leaflets of 'Burkett' and 'Squirrel's Delight' were placed in petri dishes with moistened paper towels for a total of six plates per cultivar, each containing two

leaflets. The spore suspensions were sprayed onto the leaves in the plates, the plates were incubated at 27°C, and the leaves were examined every four days for infection until twelve days after inoculation.

RESULTS AND DISCUSSION

The method was unreliable in producing consistent lesions. It was difficult to obtain leaves free from disease at the time the leaf samples were collected (in July, the time during which a second flush occurred in pecans). Since it was not possible to disinfect leaves secondary fungi sometimes gave lesions. Also browning occurred in the absence of infection on some leaves. This test was repeated with similarly unreliable results. The method therefore was not suitable for further use.

CHAPTER V

THE EFFECT OF FUNGICIDES ON THE GROWTH OF *C. CARYIGENUM* ISOLATES

MATERIALS AND METHODS

For this experiment the thirteen isolates (see Table II) were transferred to MEA plates so they could grow and be ready for use as inoculum once preparation of the agar plates amended with fungicides at different concentrations was complete.

Media with the following fungicides was prepared: Orbit^{R} (Propiconazole: 1-[[2-(2,4-dichlorophenyl)-4-propyl-1,3-dioxolan-2-yl]methyl]-1H-1,2,4-triazole) from CIBA Corporation; Supertin^{R} (triphenyltin hydroxide) from GRIFFIN Corporation; Benlate^{R} (Benomyl: Benomyl[methyl 1-(butylcarbamoyl)-2 benzimidazolecarbamate]) from E.I. DU PONT DE NEMOURS and Company; and RH 7592, an experimental product from the ROHM and HAAS company. The concentrations of total product (a.i. + inert materials) used were 0.01 ml/L (0.01 ul/ml), 0.1 ml/L (0.1 ul/ml), 1.0 ml/L (1 ul/ml) and 10 ml/L (10 ul/ml) for Orbit^{R}, Supertin^{R} and RH 7592, and 0.01 g/L (10 ug/ml), 0.1 g/L (100 ug/ml), 1.0 g/L (1000 ug/ml) and 10 g/L (10000 ug/ml) for Benlate^{R}. Refer to Table IV for actual concentrations of active ingredients.

TABLE IV

CONCENTRATION OF PRODUCT AND EQUIVALENT A.I. CONCENTRATION OF FUNGICIDES TESTED

Fungicide	Concentration of Product	Concentration of a.i.
propiconazole (Orbit^{R})	0.01 ml/L	4.18 ug/ml
	0.1	41.8
	1.0	418.0
	10.0	4180.0
triphenyltin hydroxyde (Supertin^{R})	0.01 ml/L	4.0 ug/ml
	0.1	40.0
	1.0	400.0
	10.0	4000.0
RH 7592 2F	0.01 ml/L	2.28 ug/ml
	0.1	22.8
	1.0	228.0
	10.0	2280.0
benomyl (Benlate^{R})	0.01 g/L	5.0 ug/ml
	0.1	50.0
	1.0	500.0
	10.0	5000.0

For each fungicide four liters of MEA were prepared and allowed to cool to 48°C before adding the respective fungicides. The same procedure was used for all fungicides and concentrations. Also MEA with no fungicide was prepared to be used as control. Twenty ml of medium was added to each plate. The plates had previously been marked as described above so that radial growth could be determined.

Because of the size, the experiment was completed in two sets, each of which had a control. Once media amended with the different fungicides and concentrations were ready, the 13 isolates were transferred to these plates and incubated at 27°C, the optimum temperature at which they grew better overall, according to the growth experiment (Chapter IV).

Plates were observed and marked for radial growth at four and at six days, at which time the fastest growing isolate in the control plates had reached the edge of the plate, and the experiment was terminated. Measurement and calculation of the average daily radial growth rate was as described above.

The fungicide experiment was carried out twice during 1994. The results were statistically analyzed using a nested design with two treatment factors where concentration is nested within test. The level of significance used was $P = 0.05$.

RESULTS

In the first experiment, only the lowest tested concentrations of either triphenyltin hydroxide or propiconazole allowed radial growth (Figs. 19 and 20). The average radial growth for all 13 isolates was 0.06 cm/day at the lowest concentration of triphenyltin hydroxide (4.0 ug/ml) and 0.19 cm/day at the lowest concentration of propiconazole (4.18 ug/ml). Average radial growth in control plates for these two fungicides was 0.54 cm/day. The statistical analysis showed that there were differences (P=0.05), between the lowest concentrations of propiconazole and triphenyltin hydroxide and the control plates.

With the experimental product RH 7592 2F, the pecan scab fungus exhibited limited growth in plates amended with all the concentrations (Fig. 21). The average growth/day was 0.21, 0.11, 0.14 and 0.10 cm/day from the lowest to the highest concentration.

The average radial growth/day in plates amended with benomyl was 0.47, 0.42 and 0.32 cm/day at the concentrations of 5, 50 and 500 ug/ml, respectively (Fig. 22). No growth occurred at the highest concentration of benomyl (5000 ug/ml). For control plates average radial growth/day was 0.51 cm/day. The lowest concentration of benomyl (5 ug/ml) was not significantly different from the control; however, growth at the other concentrations (50, 500 and 5000 ug/ml) was significantly different.

In the second experiment, where growth occurred in

plates amended with the different concentrations of the fungicides, it was significantly different from the control plates in all tests. These results support those of the previous experiment and are not given here.

DISCUSSION

Many years have passed since Barnes (1971) stated that benomyl was very effective for controlling *C. caryigenum*, and the scenario has changed. Littrell and Lindsey (1976) reported the existence of tolerant isolates at some counties in Georgia, after 3 years of repeated use of benomyl. The isolates in this study were considered tolerant if growth occurred in agar plates amended with 5 ug/ml of benomyl. From the point of view that tolerant isolates are those that grow on agar plates amended with the fungicide under study, all the thirteen isolates collected were found to be tolerant at concentrations even as high as 50 ug/ml and 500 ug/ml of benomyl. This agrees with Littrell (1980) whose isolates grew at concentrations of 640 ug/ml of benomyl. In the current experiment only the highest concentration (5000 ug/ml), a concentration illegal and unfeasible to apply, could stop growth of the fungus. Tolerance to benomyl had not been documented in Oklahoma until now. Growers will have to reconsider the way they are using the few fungicides available. At present some growers have already switched to triphenyltin hydroxide because they believe it gives better control than benomyl when they were rotating it with benomyl.

Propiconazole and triphenyltin hydroxide were effective in controlling growth of *C. caryigenum* at all concentrations used except for the lowest concentrations (4.18 ug/ml and 4.0 ug/ml, respectively). Although growth occurred it was very limited, especially with triphenyltin hydroxide, and statistically, growth even at the lowest concentration of both fungicides, was significantly different from the control. This reflects the effectiveness they have shown in the field and the sensitivity of *C.caryigenum* to these fungicides. However Littrell (1981) pointed out that in Greece the efficacy of triphenyltin hydroxide for control of *Cercospora* leaf spot in sugar beets had decreased. This is not the case at present with *Cladosporium*. However it is something to consider in the future and when making recommendations to pecan growers. Although very limited growth of *C. caryigenum* occurred at all concentrations of the experimental product RH 7592 2F, the statistical analysis showed that growth at all concentrations was reduced significantly compared to the control. The concentrations of active ingredient, which were chosen to include the recommended rates, were lower than for all the other concentrations of fungicides tested. This material works well in the field, so either this reduction in growth is effective or other mechanism must have came into play.

Few fungicides are available for the control of pecan scab, and use of some is limited due to phytotoxicity, ineffectiveness (dodine) or development of tolerant strains, as found in Georgia for benomyl (Littrell and Bertrand,

1981). Now in Oklahoma for the first time, the occurrence of tolerant isolates to benomyl has been documented. However, benomyl is useful to control other diseases of pecan, e.g. powdery mildew, liver and brown spots. Littrell and Lindsey (1976) showed that benomyl tolerant strains survive in nature as well as the sensitive ones and remain virulent, so tolerant strains are likely to remain present once selected.

No fungicide should be used as the sole basis for control; neither should chemical control be the only option employed. Management strategies should emphasize the appropriate use (rotation) of the effective fungicides still available, in conjunction with other non-chemical control measures to control pecan scab, and thus prevent further development of tolerance to the available fungicides. Consequently, the need for a practical forecasting system becomes more important, since a system that permits rapid communication to growers allows them to take timely action. Furthermore, contamination of the environment is also reduced. Reduced number of applications would also limit the exposure of pathogen to fungicides and thereby reduce the probability of the development of tolerance. The objective is not to eliminate fungicides in pecan scab control but to improve their usage.

CHAPTER VI

SUMMARY AND CONCLUSIONS

A. Field Studies

Studies were conducted at the Pecan Research Station to determine the relationships existing between weather factors such as temperature, relative humidity, leaf wetness and rainfall, and the development of the pecan scab disease caused by the fungus *Cladosporium caryigenum*. The site was located near Sparks, in Lincoln County, Oklahoma. The orchard consisted of the 'San Saba Improved' cultivar and native pecan trees. Disease occurred naturally in the orchard.

Terminal branches and fruit clusters were tagged in both the 'San Saba Improved' cultivar and native pecans. Weekly observations and disease ratings on fruit and leaves were done using the modified Horsfall and Barratt System (Horsfall and Barratt, 1945). The number of fruit/cluster were recorded throughout the season. Defoliation was also rated using a defoliation index.

A data logger, manufactured by Campbell Scientific, Inc., Logan, UT, was installed at the center of the orchard. The sensors used with the data logger consisted of a temperature and relative humidity probe, a rain gauge, and a

leaf wetness sensor.

During 1993 foliar disease appeared early in the 'San Saba Improved' cultivar and continued increasing throughout the season to a maximum of 6.34 (on a scale of 1 = no disease to 8 = 100% disease) on August 27. In the natives, disease remained low and only reached a maximum of 2.69 on August 27. The defoliation index was 1.1 for the 'San Saba Improved' and around 0.5 for the natives. Disease on fruit reached higher rating categories than leaves.

In 1994, disease started first in the native pecans, since budbreak in the susceptible cultivar 'San Saba Improved' was delayed that year. However, disease in native pecans remained low while in 'San Saba Improved' ratings kept increasing and reached higher levels than in native pecans. Nevertheless disease was lower than in 1993.

Weather factors apparently were more favorable for disease development during 1993 than during 1994. Average temperature, hours of relative humidity > 90% and leaf wetness > 25% occurring 0 - 7, 7 - 14 and 14 - 7 days previous to each observation were graphed against change in disease. It was hypothesized that the weather occurring 14 - 7 days previous to each observation would be more important to symptom expression since it has been reported that appearance of symptoms occurs 7 - 10 days after infection (Latham, 1982; Gotwald, 1985). For the 1993 data a curvilinear relationship was observed between temperature and increase in disease in leaves but not in fruit. The data did not show any primary relationship existing between

relative humidity or leaf wetness and increase in disease in leaves and fruit. During 1994, this type of relationship between temperature and leaves was not observed due to low disease levels. Hours of relative humidity > 90% and of leaf wetness gave the same results as in 1993.

The nature of the relationship between weather and disease development is not a simple one. Analysis of future data should be done by using statistical models that would allow better explanations of this relationship. More sites and additional years of study are necessary before thresholds can be established. The Oklahoma Mesonet System could be very helpful in providing quick access to weather information for use in disease forecasting once models are developed.

The genetic resistance of the native pecans apparently played an important role in maintaining low levels of disease. This important benefit needs to be taken into account when planning orchards, since levels of resistance in an orchard will determine the management strategies to be carried out. Growers, however, often favor cultivars that are more susceptible, therefore, determining the threshold at which disease develops so that well timed fungicide applications can be made would be very important.

Since fruit affected in the early stages of growth do not develop and may result in the entire crop being lost, it is critical to protect the fruit during this period. Early defoliation greatly influences negatively the setting of fruit the following year. Unmanaged or mismanaged orchards

can lead to losses not only of the current year crop but also the following year.

B. Laboratory Studies

Studies were carried out under laboratory conditions to observe the effect of temperature on *Cladosporium caryigenum* isolates. Thirteen isolates were obtained from different cultivars and from various locations in Oklahoma. Actively growing, pure cultures of the thirteen isolates were exposed to temperatures of 15°C, 18°C, 21°C, 24°, 27°C and 30°C. The average daily radial growth for all isolates was determined at each temperature. The optimum temperature for growth of *C. caryigenum* under the conditions of this experiment was found to be 27°C. However, growth occurred at all temperatures in the range.

As a second part of these studies, the sporulation of cultures grown at the same temperatures as above for twelve days was estimated using a visual rating scale. The optimum temperature for sporulation was found to be 27°C. However, the method was subsequently shown to be unreliable when actual counts of spores from plates rated visually were determined. The assessment of sporulation should be done by actual counting rather than visual estimation.

Although the optimum temperature for growth of the isolates of *C. caryigenum* collected was 27°C, the fungus also could grow very well at both 24°C and 30°C. Sporulation was also best in this range (24°C - 30°C). Under field conditions, knowledge of the optimum growth and

sporulation temperatures in conjunction with other weather factors may be important in the identification of a threshold for disease development.

A preliminary inoculation procedure was attempted to determine if different isolates produced different levels of infection. Spore suspensions from three isolates were sprayed onto field-collected, detached leaves of two highly susceptible cultivars and incubated under moist conditions at 27°C. Consistent production of lesions could not be obtained by using this method, and contamination by other fungi occurred. A major problem was to obtain leaves of highly susceptible cultivar from the field that were free from disease. The method was evaluated as not suitable for further use.

Information on the differences in pathogenicity of various isolates would be important for pecan breeders, which often can not test new cultivars against a wide range of naturally occurring strains. Improved cultivars selected after exposure to a wider range of strains could have a greater probability of displaying resistance for a longer time, which in turn would benefit the grower. More effort to develop an inoculation technique that would offer better results is certainly warranted.

In other studies, the effects of various fungicides on the growth of the thirteen *Cladosporium caryigenum* isolates was assessed. Propiconazole, triphenyltin hydroxide, benomyl and the experimental product RH 7592 2F were used to amend MEA plates onto which actively growing cultures of the

isolates were transferred.

Of the four fungicides tested, triphenyltin hydroxide completely stopped growth of the fungus at the concentration of 40.0 ug/ml a.i., and propiconazole stopped growth at 41.8 ug/ml a.i. For benomyl, even though higher concentrations were used, growth occurred in all concentrations but the highest (5000 ug/ml). The growth which occurred at all but that concentration was similar to control levels. The experimental product RH 7592 2F markedly limited growth but did not stop growth at any concentration. Results were consistent with the experiments carried out during 1994.

All the thirteen isolates of the scab fungus tested had developed tolerance to benomyl in Oklahoma. Growers must reconsider the type of fungicides and management strategies to use for control of pecan scab in the future, to prevent the development of tolerance to the few fungicides still available in the market.

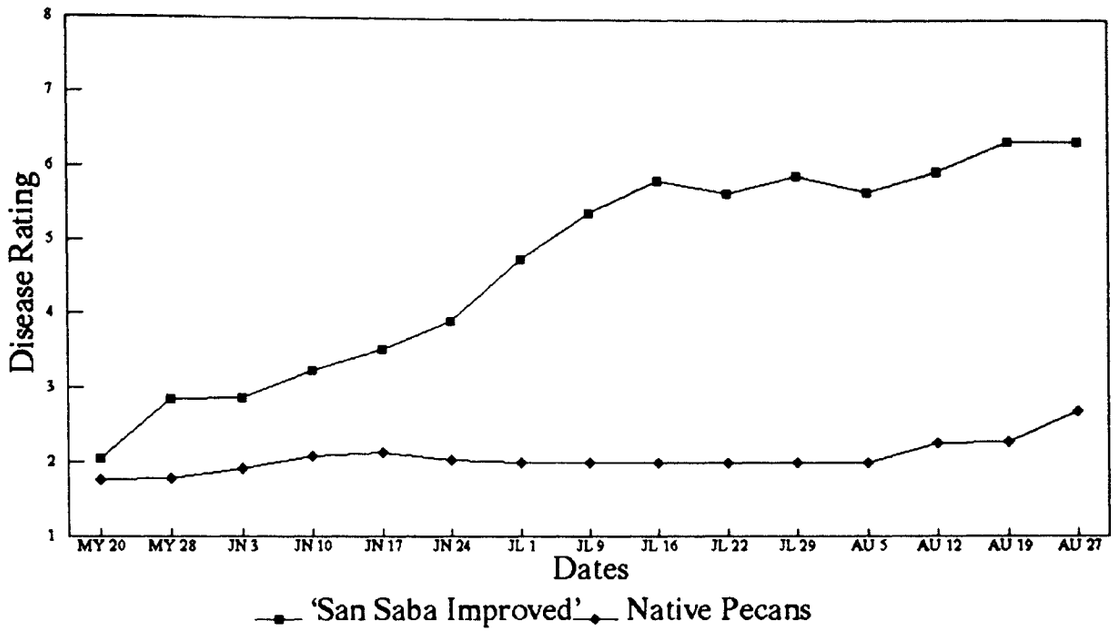

Figure 1. Leaf Disease Ratings for the 1993 Pecan Growing Season

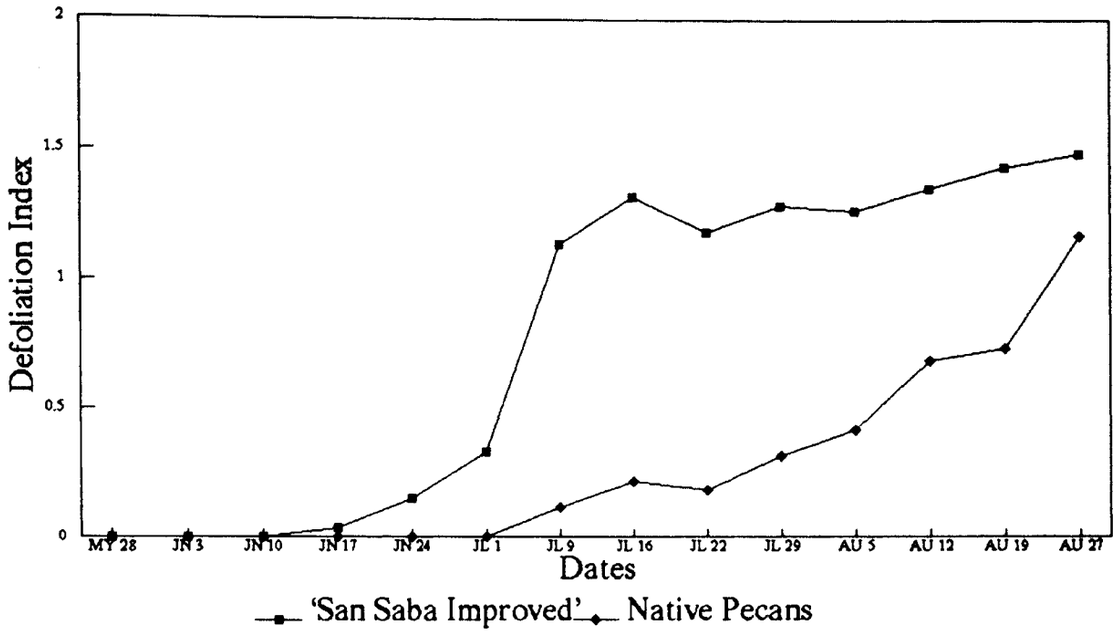

Figure 2. Defoliation During the 1993 Pecan Growing Season

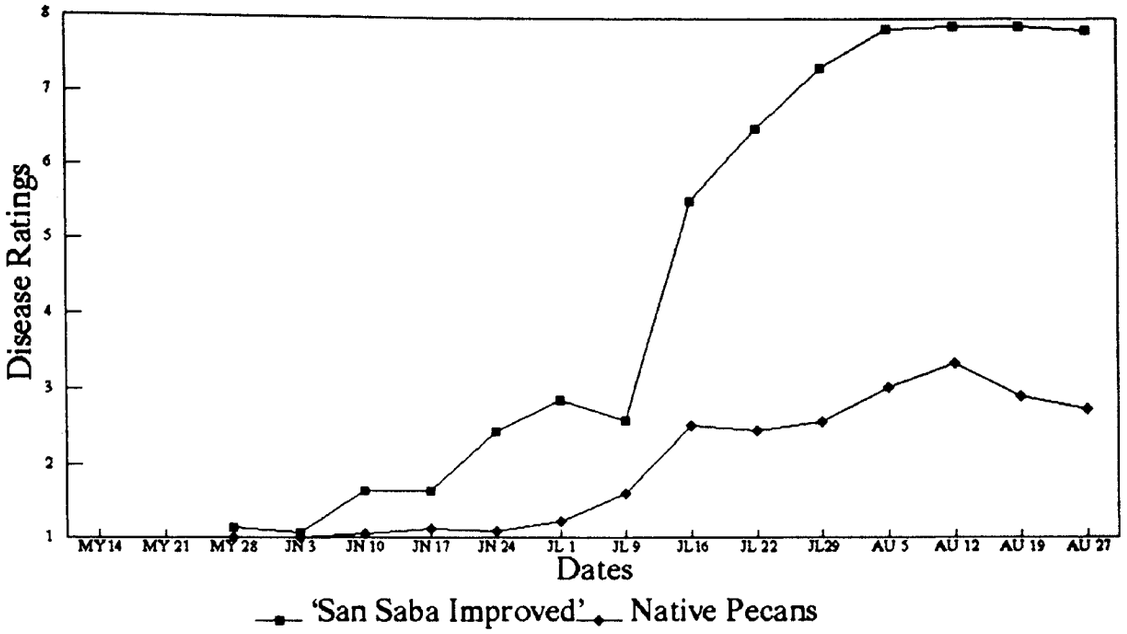

Figure 3. Fruit Disease Ratings for the 1993 Pecan Growing Season

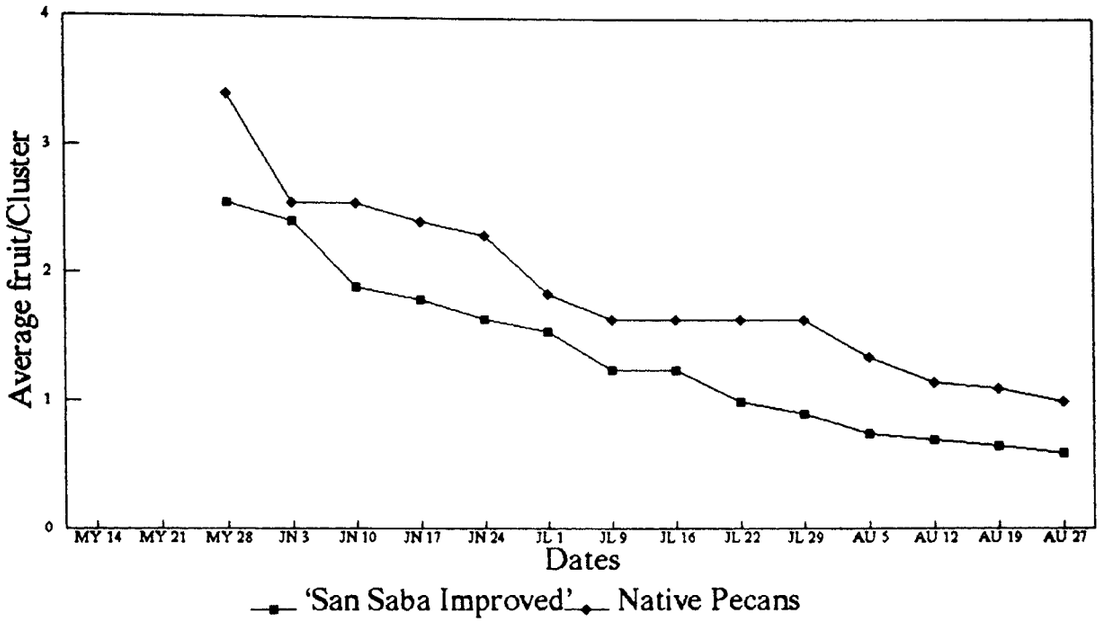

Figure 4. Fruit Retention During the Pecan Growing Season 1993

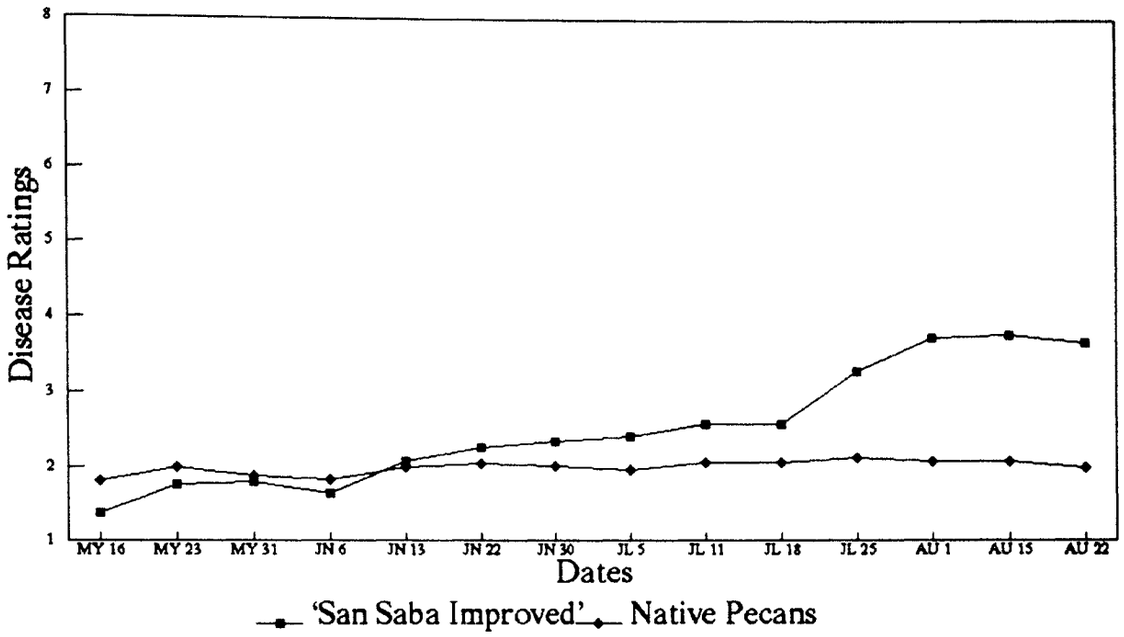

Figure 5. Leaf Disease Ratings for the 1994 Pecan Growing Season

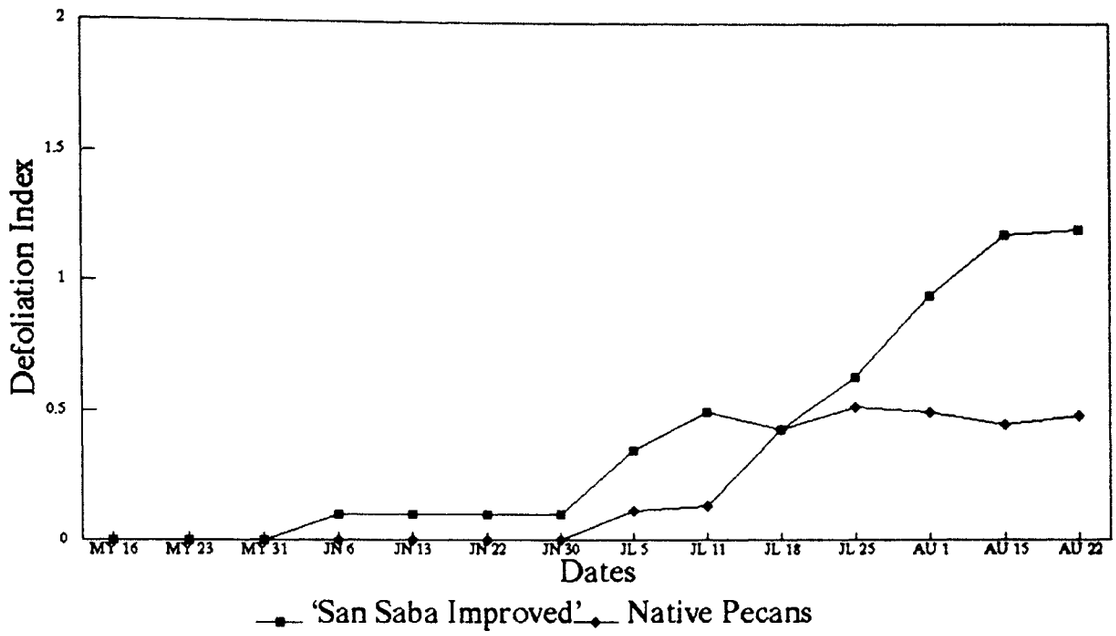

Figure 6. Defoliation During the 1994 Pecan Growing Season

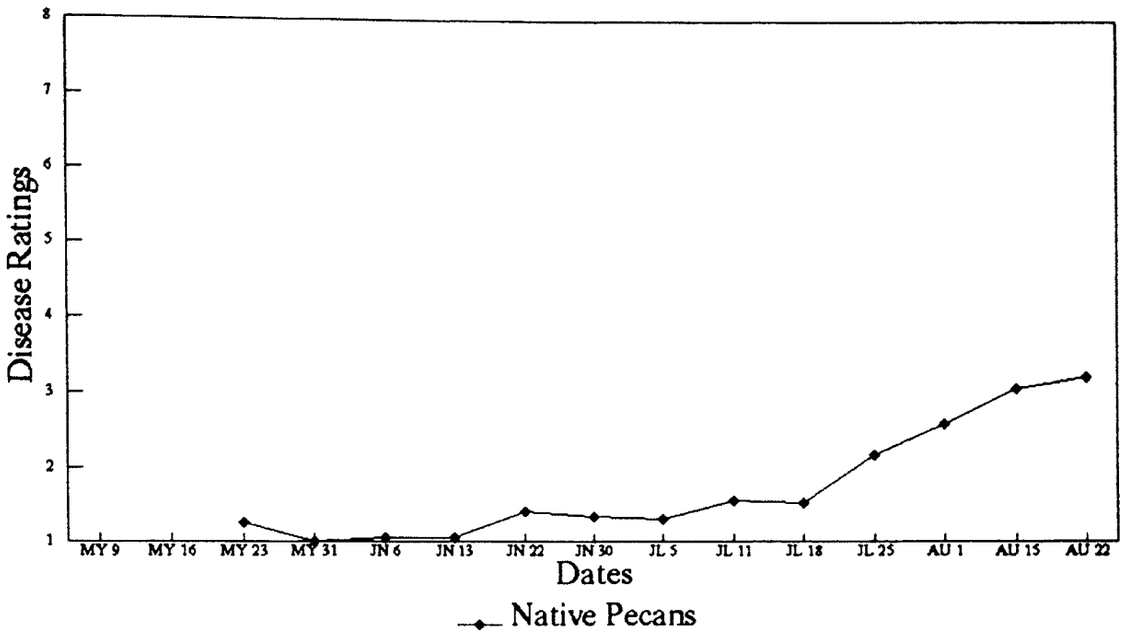

Figure 7. Fruit Disease Ratings for the 1994 Pecan Growing Season

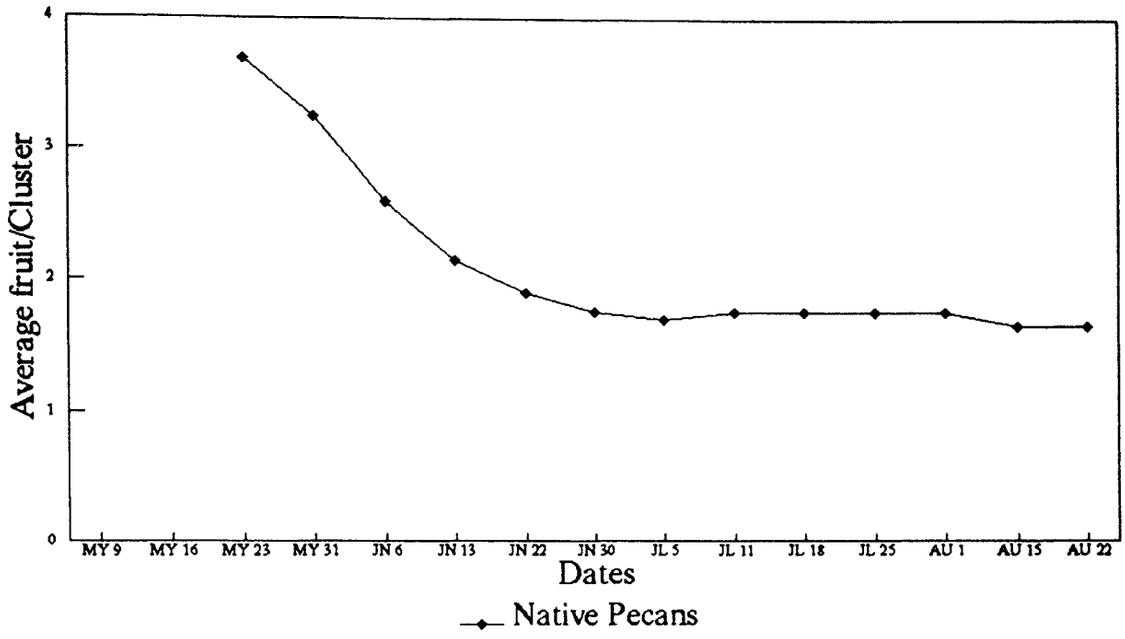

Figure 8. Fruit Retention During the 1994 Pecan Growing Season

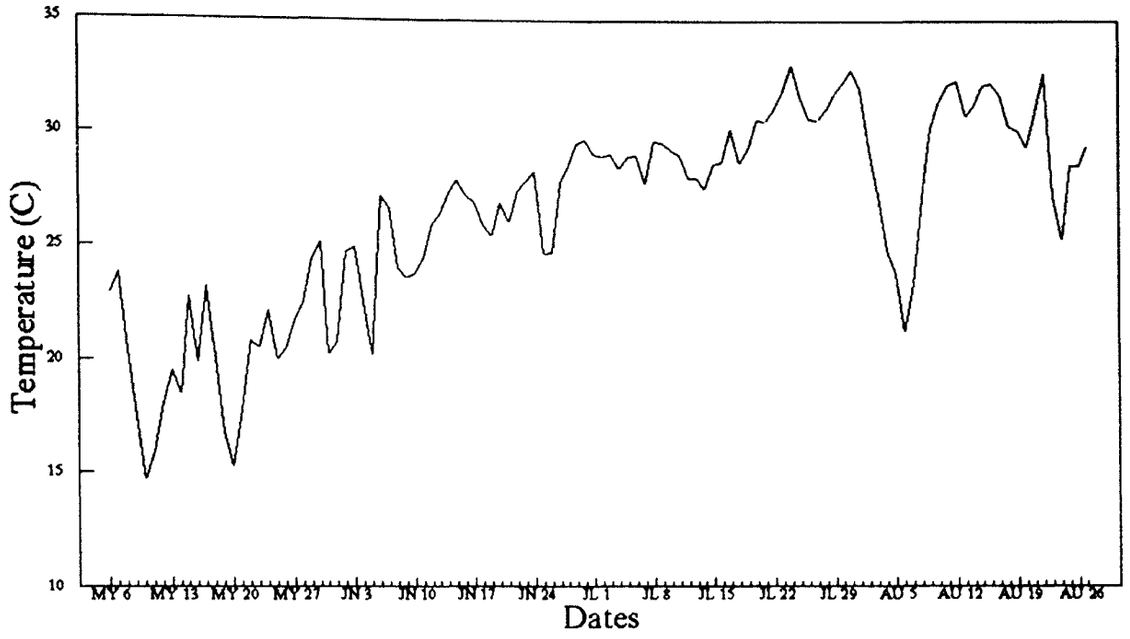

Figure 9. Average Daily Temperatures Occurring May through August 1993 at Sparks, OK

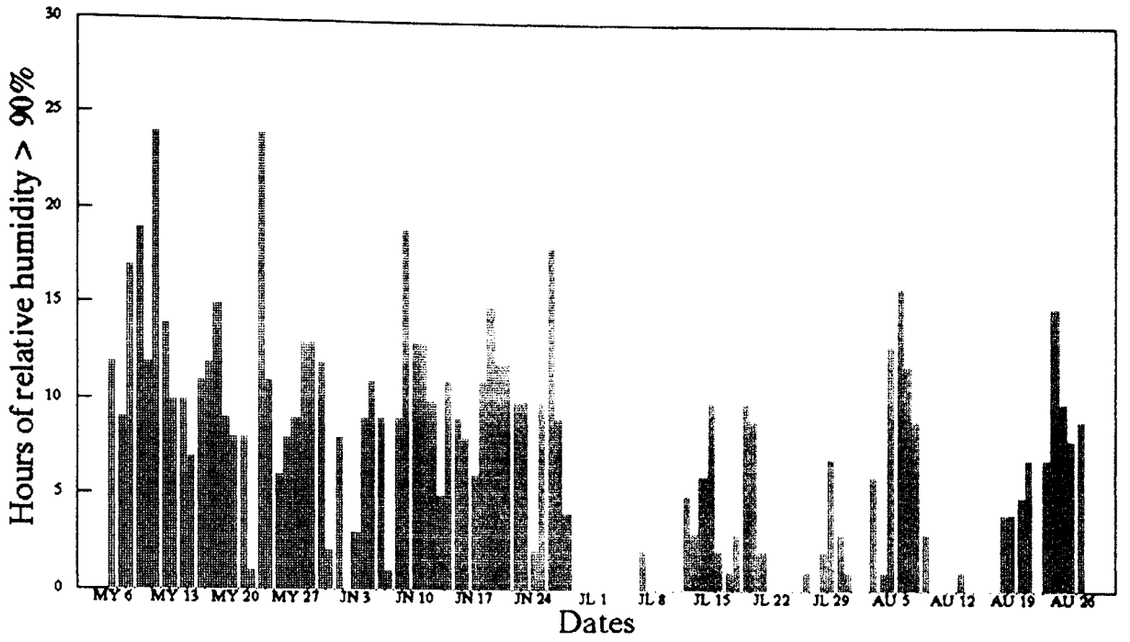

Figure 10. Hours of Relative Humidity > 90% Occurring May through August 1993 at Sparks, OK

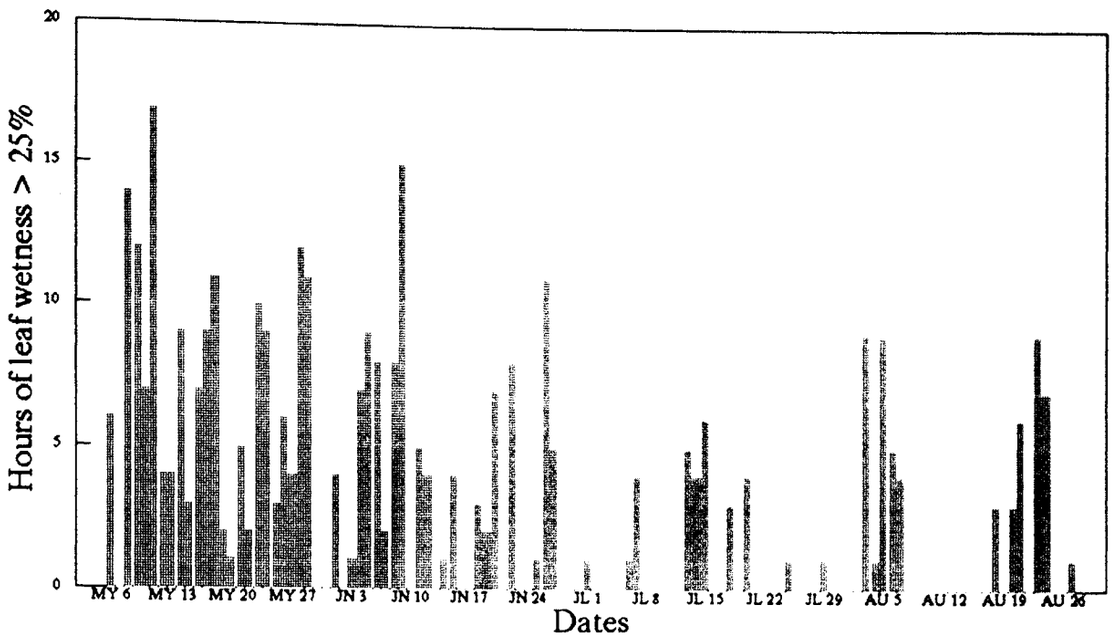

Figure 11. Hours of Leaf Wetness > 25% Occurring May through August 1993 at Sparks, OK

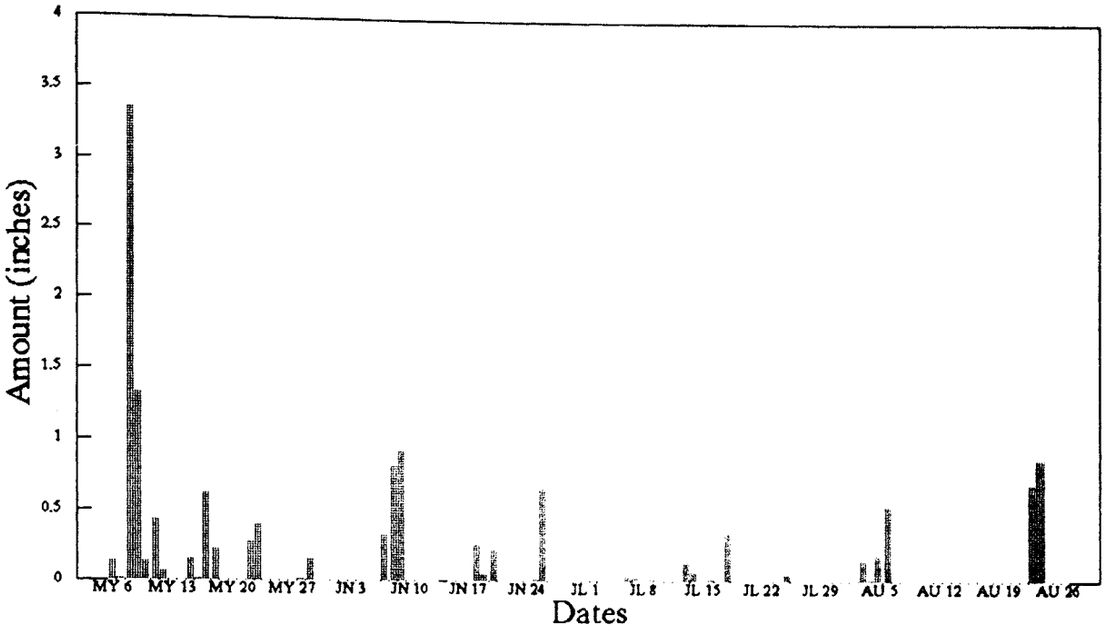

Figure 12. Total Rainfall Occurring May through August 1993 at Sparks, OK

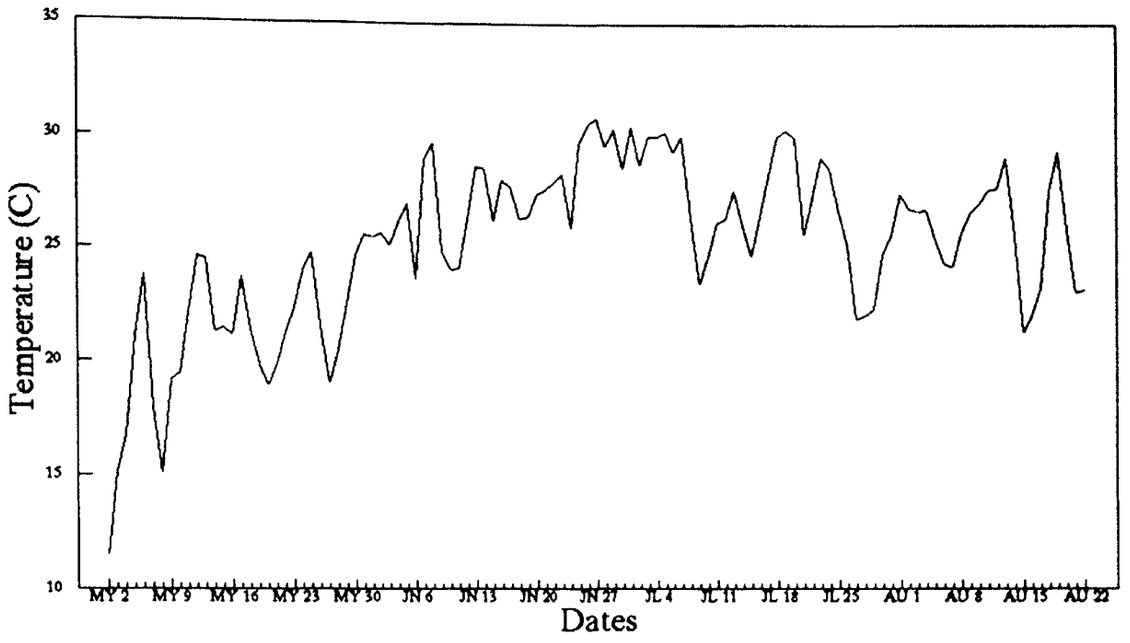

Figure 13. Average Daily Temperatures Occurring May through August 1994 at Sparks, Ok

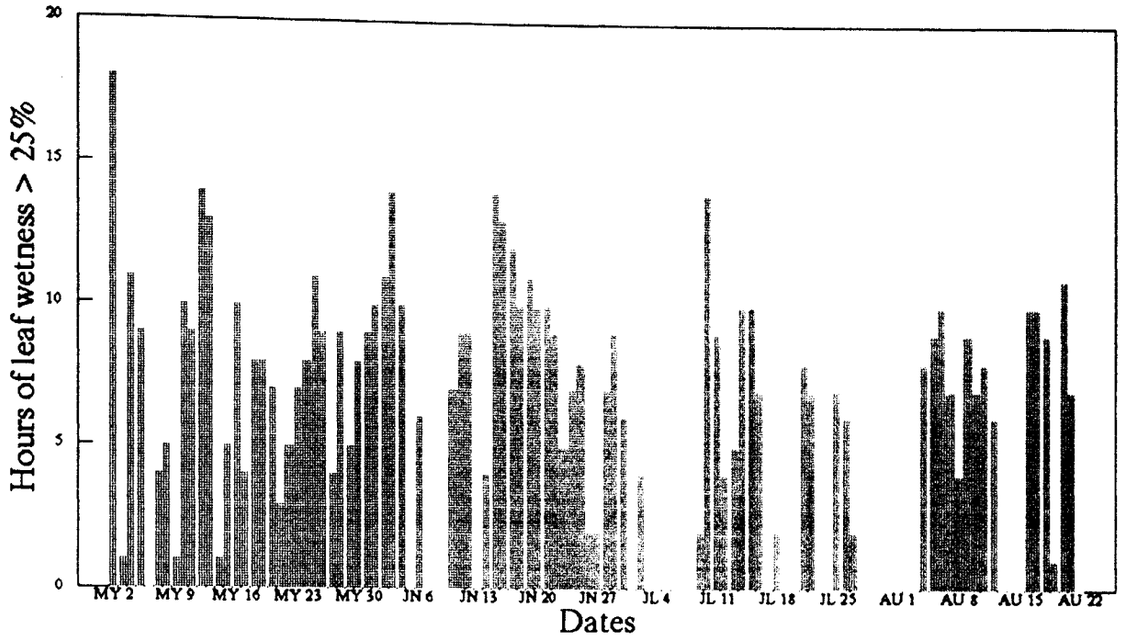

Figure 14. Hours of Leaf Wetness > 25% Occurring May through August 1994 at Sparks, OK

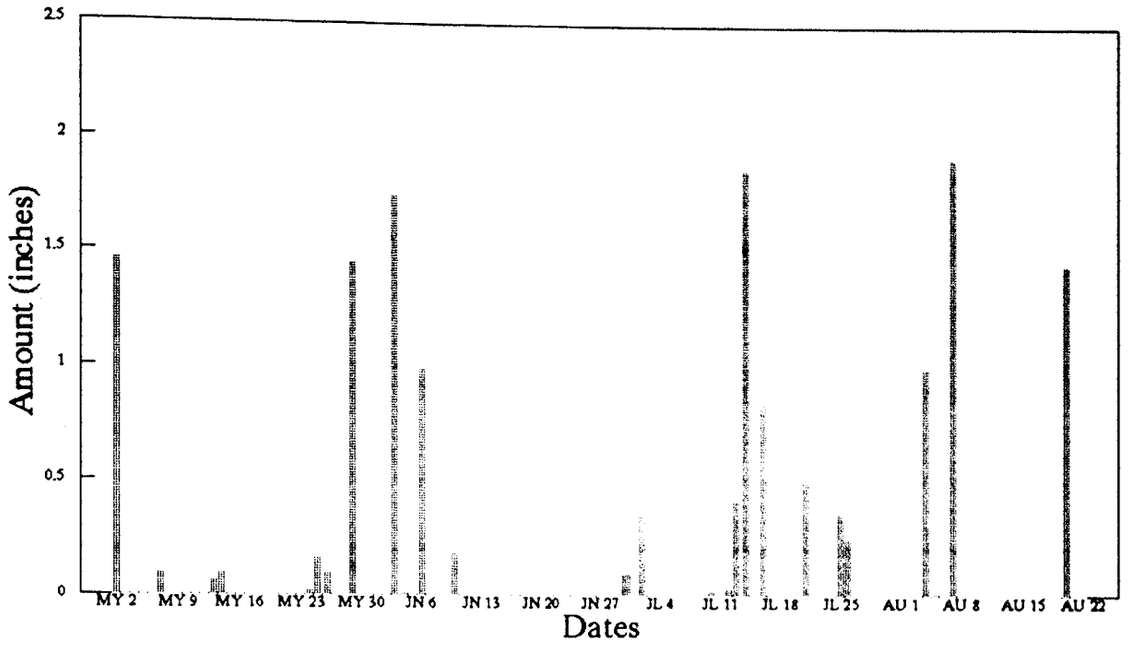

Figure 15. Total Rainfall Occurring May through August 1994 at Sparks, OK

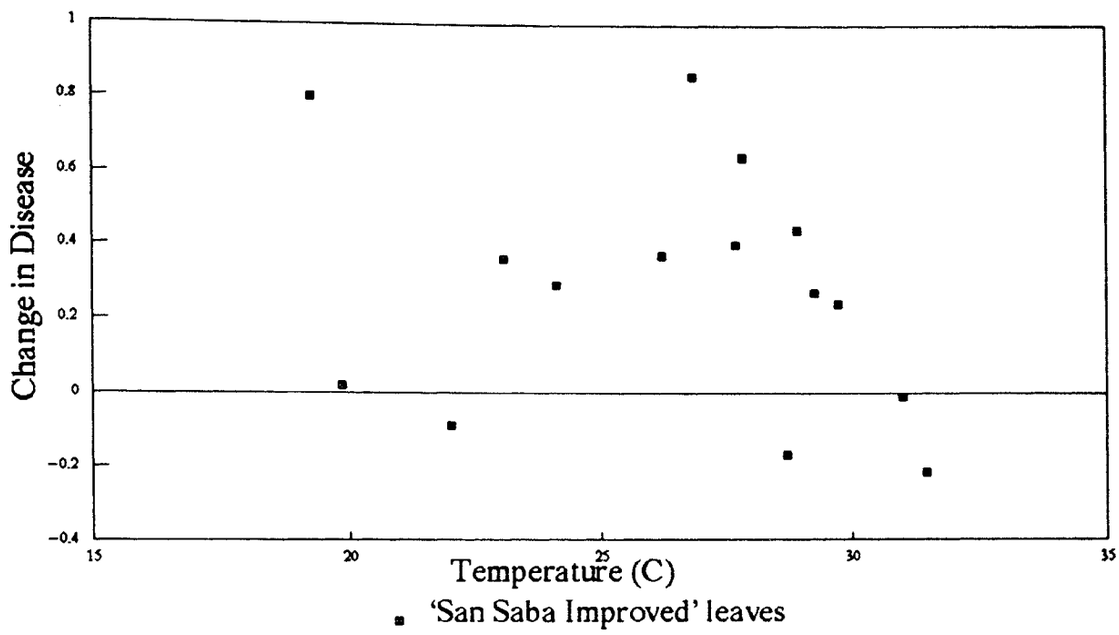

Figure 16. Temperature Occurring the Previous 14 - 7 Days to Each Observation vs Disease Change in 'San Saba Improved' Leaves During 1993, Pecan Research Station, Sparks, OK

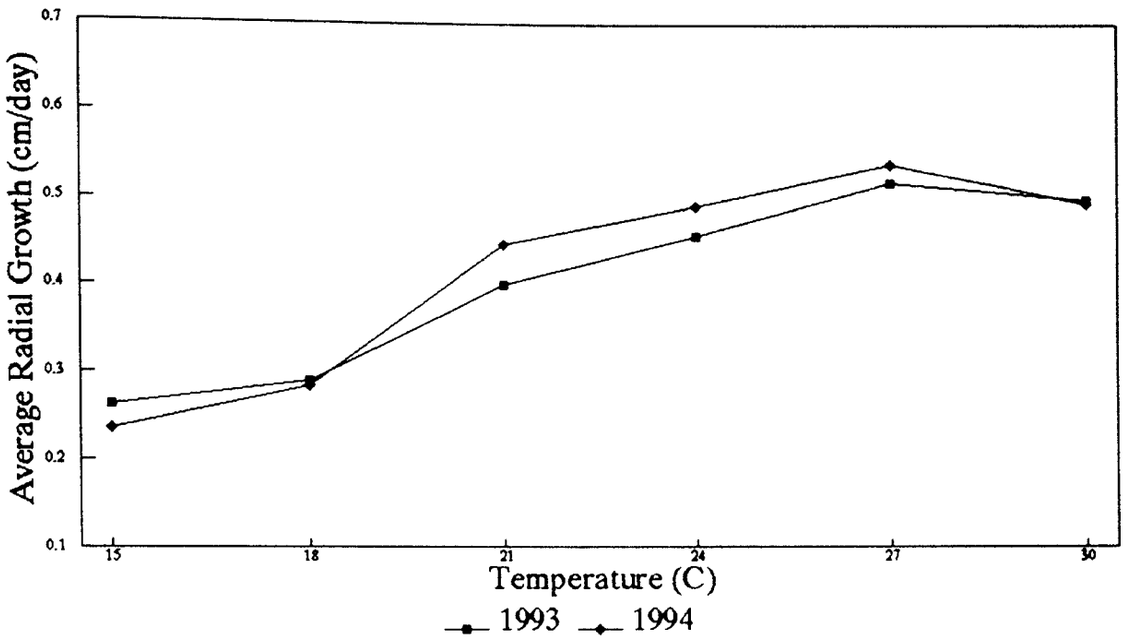

Figure 17. Effect of Temperature on Radial Growth of *C. caryigenum* isolates (1993 and 1994)

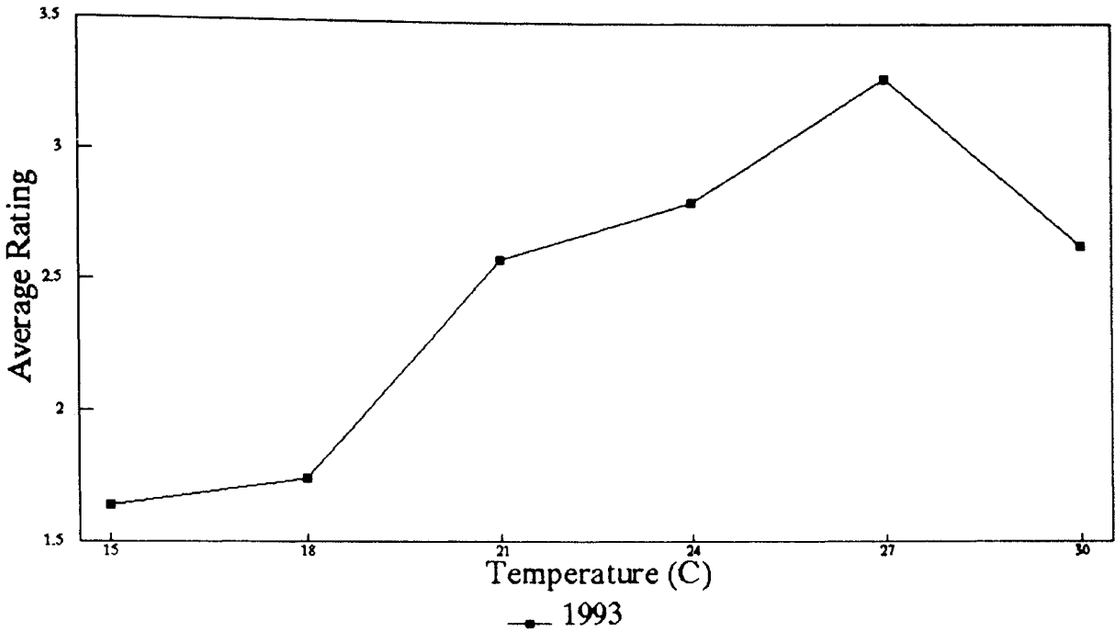

Figure 18. Effect of Temperature on Sporulation of *C. caryigenum* Isolates (1993)

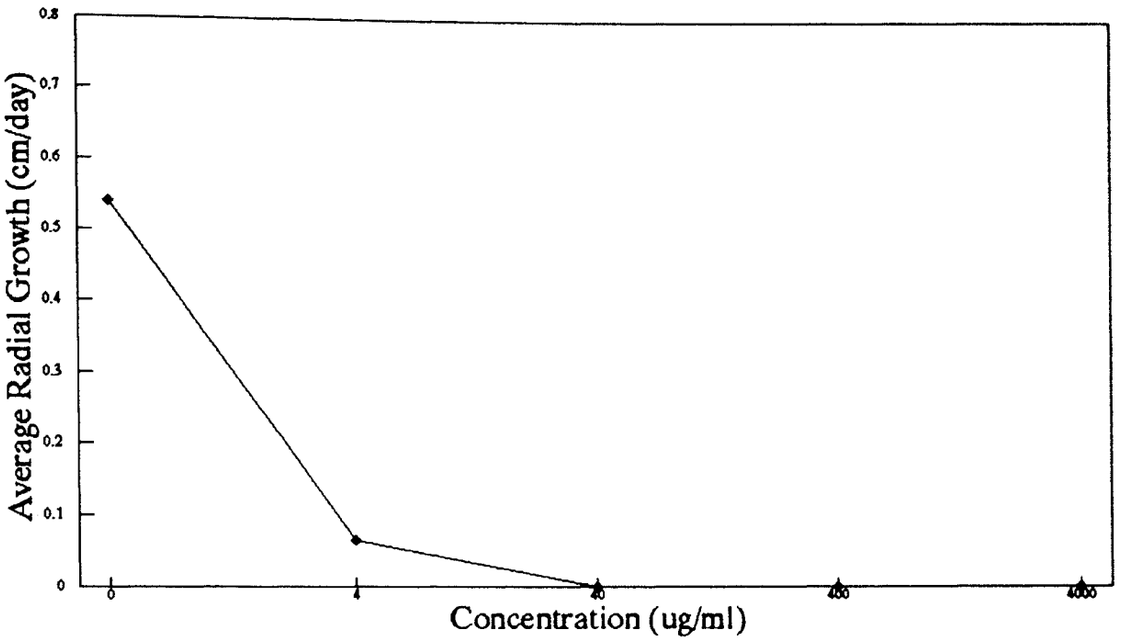

Figure 19. Radial Growth of Isolates of *C. caryigenum* in Agar Plates Amended with Different Concentrations of Triphenyltin Hydroxide

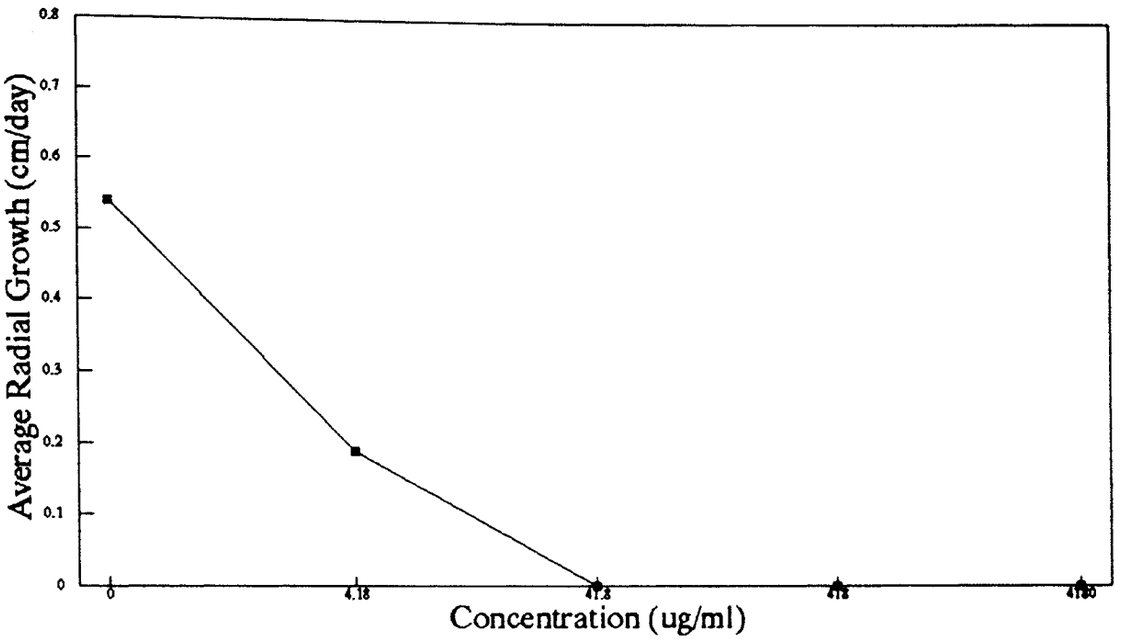

Figure 20. Radial Growth of Isolates of *C. caryigenum* in Agar Plates Amended with Different Concentrations of Propiconazole

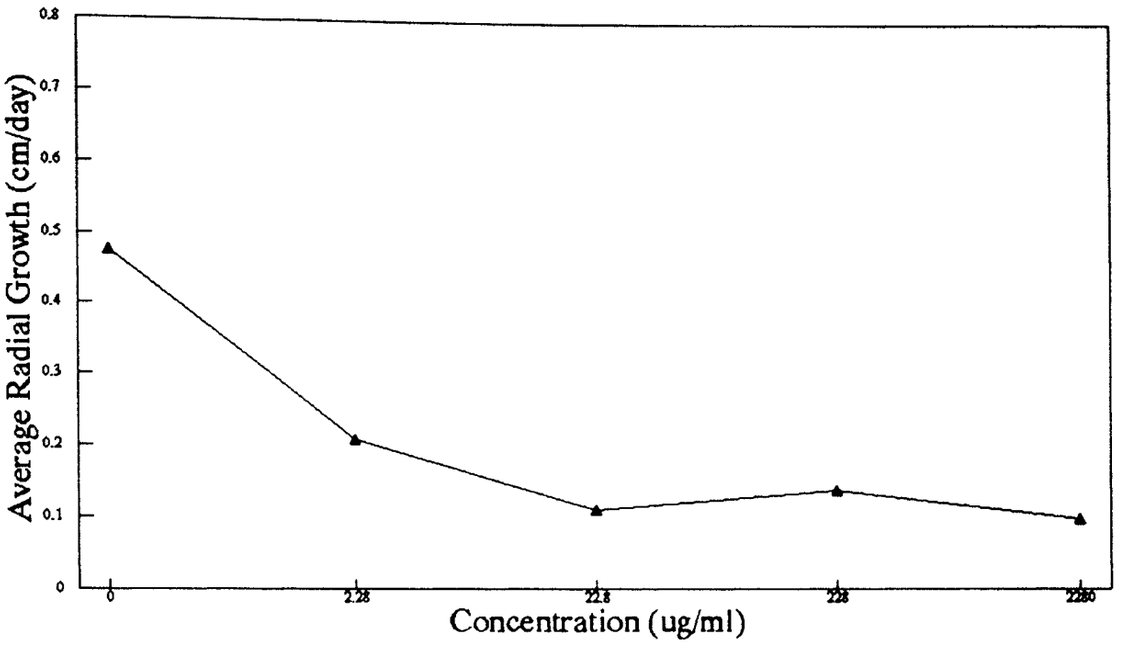

Figure 21. Radial Growth of Isolates of *C. caryigenum* in Agar Plates Amended with Different Concentrations of the Experimental Product RH 7592 2F

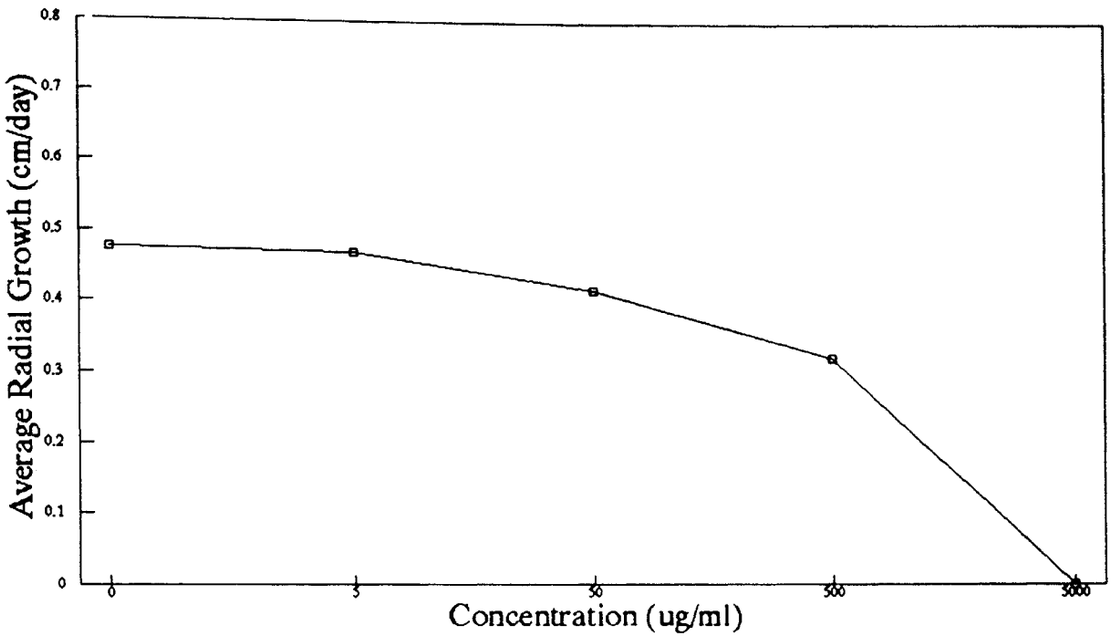

Figure 22. Radial Growth of Isolates of *C. caryigenum* in Agar Plates Amended with Different Concentrations of Benomyl

BIBLIOGRAPHY

Barnes, G. L. 1971. Effectiveness of extended interval applications of benomyl for control of pecan scab. Plant Disease Reporter 55(8):711-713.

Bertrand, P. F., and Gottwald, T. R. 1986. Evaluating fungicides for pecan disease control. In K. Hickey (ed.), Methods for evaluating pesticides for control of plant pathogens. APS Press. St. Paul, MN.

Bertrand, P. F., and Gottwald, T. R. 1984. The effect of deletion of late season fungicide sprays on pecan yield and quality. Proc. S. E. Pecan Growers Assoc. 77:51-54.

Borazjani, A., Graves, C. H. Jr., and Hedin, P. A. 1985. Occurrence of juglone in various tissues of pecan and related species. Phytopathology 75:1419-1421.

Brison, F. R. 1974. The pecan industry - past, present and future. Chap. 1. pp 1-26. In: Pecan culture. Capital Printing, Austin, TX.

Cole, J. R. 1964. Some methods for control of pecan scab, *Fusicladium effusum* (Wint.), from 1909 to the present. Proc. S. E. Pecan Growers Assoc. 57:88-89.

Converse, R. H. 1956. The production and germination of conidia of *Cladosporium effusum* (Wint.) Demaree in the laboratory. (Abstr.). Phytopathology 46:9

Converse, R. H. 1960. Physiologic specialization of *Fusicladium effusum* and its evaluation in vitro. Phytopathology 50:527-531.

Cooper, J. N. 1984. Growth and yield response of pecan to three levels of pest management in central Texas. M. S. thesis. Texas A & M University, College Station, TX. 125 pp.

Cooper, J. N., Johnson, J. D., McEachern, G. R., and McWhorter, G. M. 1983. Pecan Scab Monitoring, p. 3.9-3.14. In: Texas Pecan Integrated Pest Management Manual. TX Agr. Ext. Serv., Texas A & M University, College Station, TX.

Demaree, J. B. 1924. Pecan scab with special reference to

sources of the early spring infections. J. Agric. Res. 38:321-333.

Demaree, J. B. 1928. Morphology and Taxonomy of the pecan scab fungus, *Cladosporium effusum* Wint. comb. nov. J. Agric. Res. 37:181-187.

Demaree, J. B., and Cole, J. R. 1929. Behavior of *Cladosporium effusum* (Wint.) Demaree on some varieties of pecan. J. Agric. Res. 38:363-370.

French, W. J., and Ball, J. C. 1975. Pecan pest control options for small farmers. Proc. S. E. Pecan Growers Assoc. 68:63-66.

Gazaway, W. S., and McVay, J. R. 1981. Fungicide spray trials for scab control in Alabama. Proc. S. E. Pecan Growers Assoc. 74:179-185.

Gottwald, T. R. 1982. Taxonomy of the pecan scab fungus *Cladosporium caryigenum*. Mycologia 74(3):382-390.

Gottwald, T. R. 1985. Influence of temperature, leaf wetness period, leaf age, and spore concentration on infection of pecan leaves by conidia of *Cladosporium caryigenum*. Phytopathology 75:190-191.

Gottwald, T. R. 1982. Spore discharge by the pecan scab pathogen, *Cladosporium caryigenum*. Phytopathology 72:1193-1197.

Gottwald, T. R. 1984. Effect of temperature, post inoculation, leaf wetness, inoculum concentration and leaf age on infection of pecan by *Cladosporium caryigenum*. (Abstr.) Phytopathology 74:628.

Gottwald, T. R., and Bertrand, P. F. 1982. Patterns of diurnal and seasonal airborne spore concentration of *Fusicladium effusum* and its impact on a pecan scab epidemic. Phytopathology 72:330-335.

Gottwald, T. R., and Bertrand, P. F. 1983. Effect of time of inoculation with *Cladosporium caryigenum* on pecan scab development and nut quality. Phytopathology 73:714-718.

Gottwald, T. R., and Bertrand, P. F. 1988. Effects of an abbreviated pecan control program on pecan scab disease increase and crop yield. Plant Disease 72:27-32.

Gottwald, T. R., and Wood, B. W. 1984. The effects of scab and powdery mildew on photosynthesis and respiration of pecan fruit and foliage. Proc. S. E. Pecan Growers Assoc. 77:139-143.

Gottwald, T. R., and Wood, B. W. 1985. Decreased net photosynthetic and dark respiration rates of pecan fruit and foliage in response to infection by *Cladosporium caryigenum*. Plant disease 69:800-803.

Graves, C. H., and Hedin, P. A. 1986. Resistance in pecans. Proc. S. E. Pecan Growers Assoc. 79:45-49.

Graves, C. H., JR., Neel, W. W., and Coats R. E. 1984. Evaluation of new fungicides and strategies for control of pecan diseases in Mississippi. Proc. S. E. Pecan Growers Assoc. 77:55-60.

Graves, C. H., MacGown, M. W., Hedin, P. A., and Filer, T. H. 1986. Histochemical localization of juglone and related constituents of pecan. Phytopathology 76:205-208.

Graves, C. H., Neel, W. W., and Coats R. E. 1980. Fungicide performance on pecans in Mississippi: a five year summary. Proc. S. E. Pecan Growers Assoc. 73:141-156.

Hedin, P. A., Graves, C. H., and Langhans, V. E. 1978. Disease resistance: the most efficient way to cut production costs. Proc. S. E. Pecan Growers Assoc. 71:121-125.

Hargrove, W. W., Balsdon-wise, J., and Pickering, J. 1991. Developing a wetness-based fungicide schedule for the control of pecan scab, *Cladosporium caryigenum* (Ell. Et lang.) Gottwald at multiple sites using the remote automated intelligence network, RAIN. P.107-115. In: Wood, B. W. and Payne, J. A. (eds) Pecan Husbandry: Challenges and Opportunities. First National Pecan Workshop Proc. U. S. Dept. of Agri., Agri. Res. Serv., ARS-96

Horsfall, J. G., and Barratt, R. W. 1945. An improved grading system for measuring plant diseases. Phytopathology 35:655.

Hughes, S. J. 1953. Some folicolous hyphomycetes. Canad. J. Bot. 31:560-576.

Hughes, S. J. 1953. Conidiophores, conidia, and classification. Canad. J. Bot. 31:577-659.

Hunter, R. E. 1983. Influence of scab on late season nut drop of pecan. Plant Dis. 67:806-807.

Hunter, R. E., and Payne, J. A. 1980. Modified spray schedules for control of scab in Georgia and Texas. Proc. S. E. Pecan Growers Assoc. 73:61-66.

Hunter, R. E., and Roberts, D. D. 1977 Problems and

approaches related to screening for scab resistance in pecans. Proc. S. E. Pecan Growers Assoc. 70:69-75.

Hunter, R. E., Newton, J. E., and Kolb, M. C. 1978. Preliminary research on a fungicide spray schedule for pecan scab based on weather data. Proc. S. E. Pecan Growers Assoc. 71:171-177.

Hunter, D., Thompson, T., and Sanderlyn, R. 1986. Control of pecan diseases through genetic resistance. Proc. S. E. Pecan Growers Assoc. 79:51-54.

Jones, A. L. 1981. Fungicide resistance: past experience with benomyl and dodine and future concerns with sterol inhibitors. Plant Disease 65:990-992.

Laird, D. W., Graves, C. H., Jr., and Hedin, P. A. 1990. In vitro reactions of *Cladosporium caryigenum* with pecan condensed tannins and isoquercitin. Phytopathology 80:147-150.

Latham, A. J. 1976. Scab susceptibility of some newer pecan cultivars. Proc. S. E. Pecan Growers Assoc. 69:65-67.

Latham, A. J. 1979. Some factors influencing pecan scab, *Fusicladium effusum*, development. Auburn Agric. Exp. Stn. Highlights Agric. Res. 26:7.

Latham, A. J. 1982. Forecasting pecan scab according to hours of leaf wetness. Proc. S. E. Pecan Growers Assoc. 75:209-212.

Latham, A. J. 1982. Effects of some weather factors and *Fusicladium effusum* conidium dispersal on pecan scab occurrence. Phytopathology 72:1339-1345.

Latham, A. J. 1983. Control of *Cladosporium caryigenum* on pecan leaves and nut shucks with propiconazole (CGA-64250). Plant Dis. 67:1136-1139.

Latham, A. J., and Rushing, A. E. 1988. Development of *Cladosporium caryigenum* in pecan leaves. Phytopathology 78:1104-1108.

Lentz, P. L. 1957. Taxonomy of the pecan scab fungus. Mycologia 48:874-878.

Littrell, R. H. 1980. A review of the distribution of benomyl tolerant isolates of the pecan scab fungus. Proc. S. E. Pecan Growers Assoc. 73:47-50.

Littrell, R. H. 1983. New approaches for controlling pecan scab in the future. Proc. S. E. Pecan Growers Assoc. 76:49-51.

Littrell, R. H., and Lindsey J. B. 1976. Detection of benomyl-tolerant and sensitive pecan scab isolates in production groves. Proc. S. E. Pecan Growers Assoc. 69:77-79.

Littrell, R. H., and Lindsey J. B. 1977. Control of pecan scab in the presence of benomyl-tolerant and sensitive scab strains. Proc. S. E. Pecan Growers Assoc. 70:83-86.

Littrell, R. H., and Hunter, R. E. 1979. Laboratory and field studies for suppression of primary inoculum of the pecan scab fungus. Proc. S. E. Pecan Growers Assoc. 72:71-83.

Littrell, R. H., and Bertrand, P. F. 1981. Management of pecan of pecan fruit and foliar diseases with fungicides. Plant Disease 65(9):769-774.

McVay, J. R., and Gazaway, W. S. 1980. Measurement of leaf wetness as a means to determine fungicide applicatio timing for pecan scab control II. Proc. S. E. Pecan Growers Assoc. 73:121-125.

Ring, D. R., Payne, J. A., and Snow, J. W. 1986. A pest management program for pecans. Proc. S. E. Pecan Growers Assoc. 79:161-166.

Rushing, A. E., and Latham, A. J. 1991. Some ultrastructural observations of *Cladosporium caryigenum* growth in pecan leaves. Phytopathology 81:1102-1108.

Sanderlin, R. S., Broach, K. E. 1983. Fungicide control of pecan diseases, 1982 (scab, *Cladosporium caryigenum* and vein spot, *Gnomonia nerviseda* on *Carya illinoensis*). Am. Phytopathol. Soc. Fungic. Nematic. Test Results. 38:130-131.

United States Department of Commerce. 1978. Census of Agriculture. 1978. Oklahoma State and County Data. Vol 1. Pt. 36.

Valli, V. J. 1964. Weather conditions as related to pecan scab infection. Proc. S. E. Pecan Growers Assoc. 57:85-87.

Wetzstein, H. Y., and Sparks, D. 1983. Anatomical indices of cultivar and age-related scab resistance and susceptibility in pecan leaves. J. Amer. Soc. Hort. Sci. 108(2):210-218.

Wood, B. W., Gottwald, T. R., and Reilly, C. C. 1988. Pecan phylloplane chemicals influence germination of pecan scab conidia. J. Amer. Soc. Hort. Sci. 113(4):616-619.

VITA

Alina Maria Marenco

Candidate for the Degree of

Master of Science

Thesis: STUDIES ON THE EPIDEMIOLOGY OF PECAN SCAB CAUSED BY *CLADOSPORIUM CARYIGENUM* IN OKLAHOMA

Major Field: Plant Pathology

Biographical:

Personal Data: Born in Masaya, Nicaragua, on June 7, 1961, the daughter of Luis And Consuelo de Marenco.

Education: Graduated from Pureza de Maria School,Managua, Nicaragua, in December 1977; received Bachelor of Science degree in Biology from the National Autonomous University of Nicaragua, Leon, in November 1984. Completed the requirements for the Master of Science degree with a major in Plant Pathology at Oklahoma State University in December 1994.

Experience: Employed as Specialist in Microbiological Control of Medicines, SOLKA Laboratories, Managua, Nicaragua, January 1986 to December 1987; Quality Control Manager for Nabisco Cristal, Managua, Nicaragua, January 1988 to November 1989.

Professional Memberships: American Phytopathological Society.