# Epidemiology of European Apple Canker in California

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ABSTRACT

Factors affecting the incidence of European apple canker on Red Delicious, Golden Delicious, and Rome Beauty apple in California were studied. The disease is widely distributed in Sonoma County, and is present in other coastal counties. Isolates of *Nectria galligena* from paper birch, yellow birch, and bigtooth aspen did not infect apple trees when inoculated at leaf scars, but an isolate from apple was pathogenic on pear. European canker was found on pear for the first time in California. The major period of infection of apples was in November, with some occurring in December and none in the spring. Analysis of factors affecting development of the disease in 1970-71 showed that only leaf fall was limiting. However, in 1971-72 rain and temperature also were limiting factors. Multiple regression equations that were generated accounted for 83-94% of the variation in infection. Conidia were the major infective propagules and ascospores played an insignificant role in infection. Ascospores were water-borne or wind-borne; always in low numbers. Ascospore discharge showed no diurnal periodicity, however, it was influenced by duration of both rain and temperature.

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*Additional key words:* infection, dispersal, inoculum.

*Nectria galligena* Bres. causes cankers on many species of hardwood trees throughout the world (2, 31). The only major study on European canker of pome fruit trees in western United States was done by Zeller (31) in Oregon. Severe damage was observed on pear trees and lesser infection on apple trees. Many workers have contributed to an understanding of this disease on a worldwide basis, especially in England (1, 3, 4, 5, 10, 12, 16, 18, 25, 26, 29, 31). Nevertheless, effective control in some regions has not been obtained. Recent studies have shown that control measures effective in some areas are ineffective in others due to different epidemiological conditions (25, 26, 27).

Although European apple canker disease was reported in California in 1909 (28) no serious outbreaks were observed until the spring of 1955 when severe damage, especially to young Red Delicious and Gravenstein apple trees, was reported in Sonoma County (22). The disease became widespread in Sonoma County during the 1960's, culminating in a severe epidemic in the fall of 1970.

Wilson (26) noted several differences between the canker disease in California and in other parts of the world. Ascocarps generally did not mature under California conditions, nor were conidia present during the summer months. He found that fresh stem wounds of apple trees could be infected as late as February. Inoculations made in autumn often did not produce symptoms until after bud break in the spring. In contrast, Lortie (15) in Canada, and Swinburne (25) in Northern Ireland, state that ascospores were the most important propagule and that the major infections occur in the spring.

Studies reported herein deal with distribution of the disease in California, hosts and pathogenicity of *N. galligena*, effects of climatological and biological factors on infection, factors affecting development and dispersal of ascospores, and ascosporic infection.

MATERIALS AND METHODS.—*Distribution of the disease in California.*—A survey of three major apple-growing regions for European apple canker was made between November 1969 and September 1970. Six representative orchards in Sonoma, five in Santa Cruz, three in El Dorado, and three in Mendocino counties were inspected. Transects crisscrossing the orchard blocks were walked and all trees observed for canker presence.

*Hosts and pathogenicity.*—Inocula for the pathogenicity tests were prepared by culturing different isolates of *N. galligena* on Matsushima's (19) medium modified by adjustment to pH 5.2 with 1.0 M KOH and omission of vitamins. Cultures were grown 8-10 days at 20 C under Sylvania Gro-Lux fluorescent lights (approximately 1,076 lux); spores were suspended in sterile distilled water, and concentration adjusted with a hemacytometer. Isolates from paper birch (*Betula papyrifera* Marsh.), yellow birch (*B. alleghaniensis* Britt.), bigtooth aspen (*Populus grandidentata* Michx.), and Red Delicious apple were used. Senescing leaves were removed, and 20 random leaf scar inoculations per isolate ($2 \times 10^3$ spores/scar) were made with conidia on a total of eight 2-year-old potted Gravenstein and Golden Delicious trees. The trees were bagged with wetted polyethylene for 72 hours and kept in darkness for 1 month at 13 C. Thereafter they were chilled at 5 C for 1 month, and then held at 15.5 C for 1 month with 12 hours per day light (approximately 5,380 lux). They were moved outdoors in February 1972, and canker production was observed until after bud break in April.

*Effects of climatological and biological factors on infection.*—Determination of time and quantity of infection on the three cultivars was done by numbering consecutively all leaves on specific twigs of the current year's growth with a waterproof marking pen. The twigs

were marked transversely into 5-cm sections starting at the base, so that when a leaf scar appeared it could be related to the leaf that had fallen. Leaf fall was recorded at weekly intervals. In 1970-71, 10 to 12 randomly selected twigs per tree were marked in this manner on three trees per cultivar. In 1971-72, five twigs per tree were used on five trees per cultivar. The trees were randomly selected in a small mixed plot of Red Delicious, Golden Delicious, Rome Beauty, and Jonathan cultivars 8-9 years old. The Red Delicious trees were severely infected with the disease and provided the best opportunity to study infection in the Sebastopol (Sonoma County) area. After leaf fall, observations were made twice weekly or weekly for the appearance of leaf-scar cankers from December to April in both years. The incidence of cankers at leaf scars was compiled on a weekly basis. Since, in this region, most leaf scars on Red Delicious, Golden Delicious, and Rome Beauty become immune to infection within 1 week, the time of infection was accurately determined (8).

A climate recorder (23) was used to note ambient temperature, wind velocity, and wind direction in 1970-71. Rain in 1970-71 was recorded using a recording rain gauge, but in 1971-72 data from the official U.S. Weather Station, Graton, California, were used. This station was located approximately 3.2 km from the experimental plot. Temperature and relative humidity were recorded with a hygrothermograph in a standard weather shelter in both years.

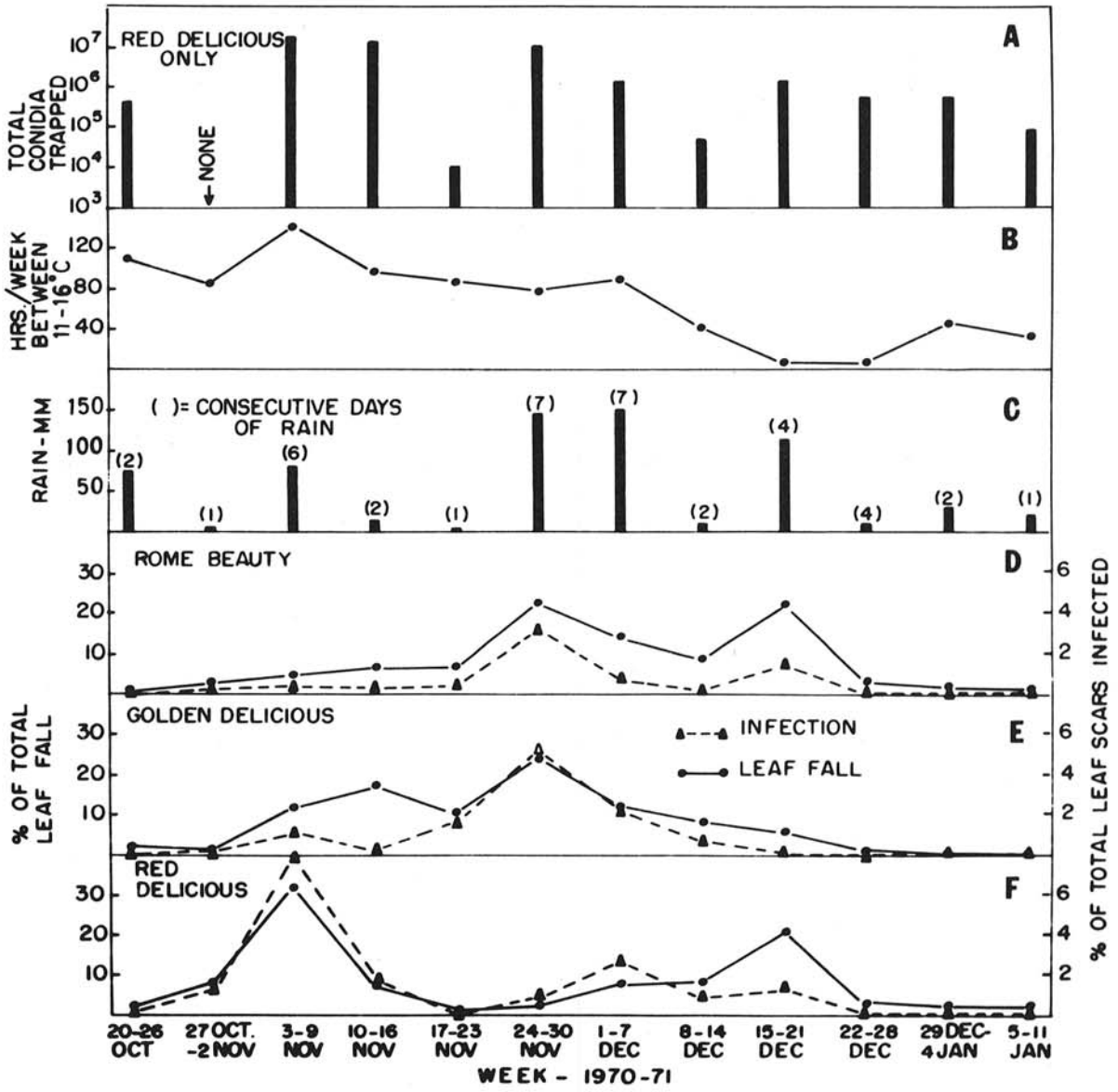


Fig. 1. Biological and weather parameters that influenced *Nectria galligena* infection of three apple cultivars in Sonoma County, California, between October 1970 and January 1971. **A)** Total conidia trapped per week in a limb funnel trap on Red Delicious only. **B)** Number of hours per week between 11-16 C. **C)** Millimeters of rain per week and consecutive days of rain. **D, E, F)** Percent of total leaf fall and of total leaf scars infected per week for Rome Beauty, Golden Delicious, and Red Delicious, respectively.

Dispersal of conidia was noted in two ways. First, a 10-cm diameter funnel was placed on the underside of a cankered 6.3-cm diameter scaffold branch and connected to a plastic jug with flexible plastic tubing. The funnel caught almost all of the water running down the limb. The water was collected twice weekly or weekly and conidial counts were made with a hemacytometer. In 1970-71, conidial estimates of this type were done only on Red Delicious. In 1971-72, this method was used to determine weekly levels on the three cultivars. In the second method, a 10-cm diameter funnel filled with 25 pieces of actively sporulating, 5-month-old Red Delicious twigs was also used to check the relative conidial levels in 1970-71. The funnel was mounted on a 1.2-m steel stake and connected to a plastic jug. Conidia were counted as before. Only data from the limb trap are reported since both trapping methods yielded similar results.

*Factors affecting development and dispersal of ascospores and ascosporic infection.*—The following studies were made during the period from October 1970 to May 1971 to determine the population of ascospores in two heavily infected Sonoma County orchards with active perithecial production. Visual observations were made of perithecial development on cankered branches with abundant fruiting bodies.

Production of ascospores was studied using two funnel traps as described in the conidial study. One trap was the same as that used to observe conidial production except that ascospore production was noted when the new perithecia were produced on the 1-year-old cankers. The second funnel contained 40 pieces of Red Delicious bark with perithecia in all states of development. Ascospores caught in rainwater were counted twice weekly and the data were combined either on a weekly or monthly basis.

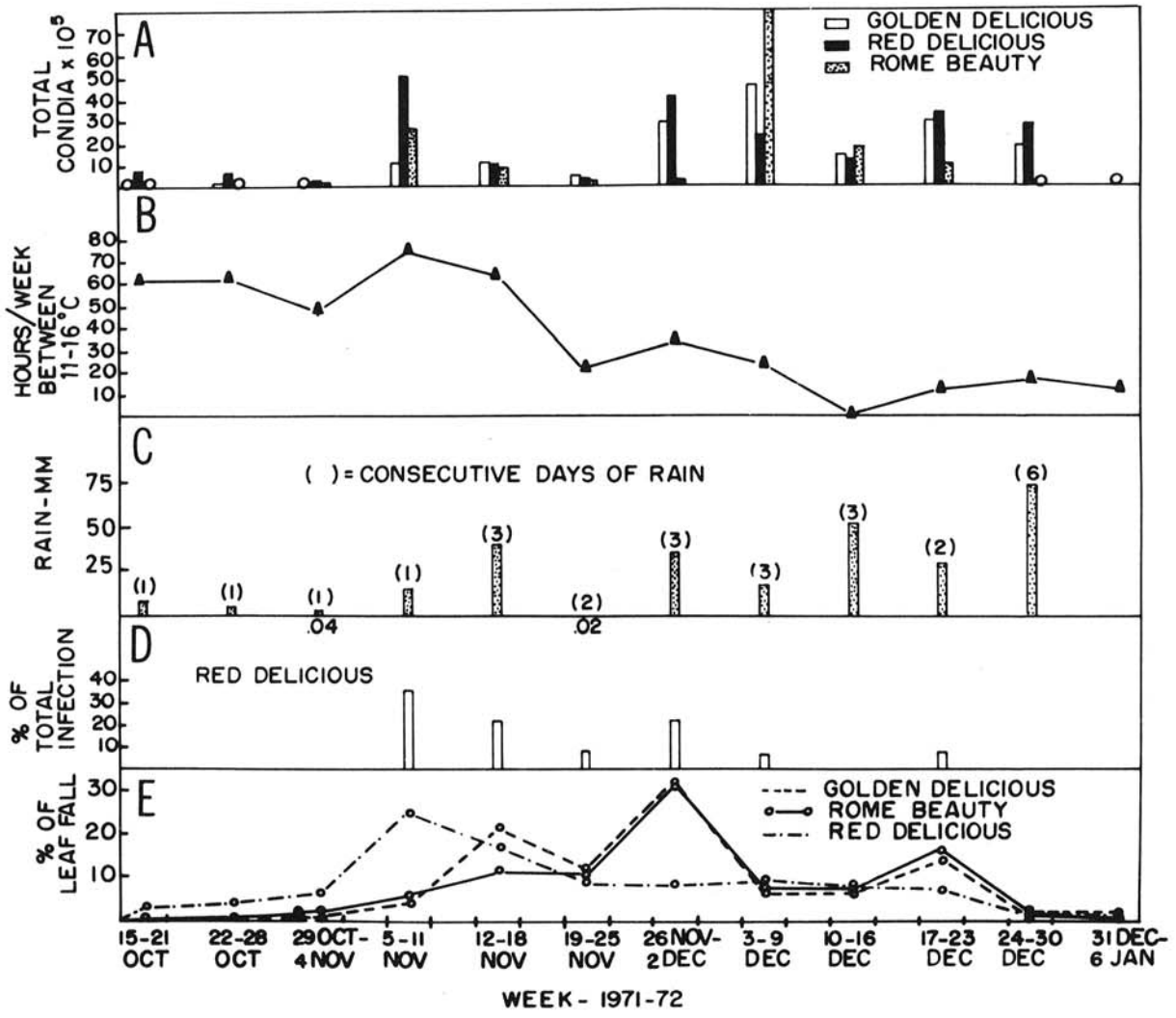


Fig. 2. Biological and weather parameters that influenced infection of Red Delicious apple by *Nectria galligena* in Sonoma County, California between October 1971 and January 1972. **A)** Total conidia per week trapped in limb traps on three apple cultivars. 0 = no spores trapped that week. **B)** Number of hours per week between 11-16 C. **C)** Millimeters of rain per week and numbers of consecutive days of rain. **D)** Percent of total leaf scar infection per week, for Red Delicious. **E)** Percent of total leaf fall per week for Golden Delicious, Rome Beauty, and Red Delicious.

A Burkard 7-day recording volumetric spore trap (Burkard Manufacturing Co., Ltd., Rickmansworth, Herts, England) was placed at ground level and ringed with 50 cankered Red Delicious branches containing numerous perithecia to try to correlate ascospore discharge and dispersal with weather parameters. Techniques used for trap adjustment, spore mounting, and adhesive preparation were as described in the instructions for the Burkard sampler. The meterological equipment was the same as for the infection studies.

Daily airborne ascospore concentration under normal conditions was determined with a Burkard spore trap placed at foliage height.

RESULTS.—*Distribution of the disease in California.*—Surveys of apple orchards in major apple regions indicated that the causal organism was present in Sonoma and Santa Cruz counties. The fungus was not found in the important apple-growing counties of El Dorado or Mendocino. However, it has been reported to occur in Mendocino County (W. O. Mc Cartney, *personal communication*). No reports exist from Butte County. *N. galligena* was reported in 1963 (24), in one orchard in Santa Cruz County and the orchard was subsequently destroyed. Nevertheless, an intensive survey of small orchards in the Santa Cruz mountains in September 1970 showed active cankers on McIntosh, Red Delicious, and Jonathan cultivars. The orchardist indicated that this type of canker had been observed for more than 20 years in that district. Wilson and Nichols (28) reported the presence of *N. galligena* in Del Norte and Humboldt counties, but apples are not extensively grown in these areas.

Rainfall above 100 cm per year, foggy weather, and moderate temperatures appear to be unifying factors in the occurrence of the causal organism in California. Watsonville, a large coastal apple area in Santa Cruz County, appears to escape the disease because it receives only approximately 58 cm of rain a year. One important coastal area of several hundred acres in Mendocino County has almost identical climate and apple culture to the heavily infected areas of Sonoma County. However, examination of orchards in this area showed no evidence of the disease. At this time escape seems to be the only explanation.

Sierra foothill apple districts differ from the coastal regions in that summer fogs are absent, otherwise climate and apple culture are similar. It was thought that summer fog might be an important factor in influencing fall inoculum level and ultimately infection, but this was not substantiated in a study in Sebastopol during the summer of 1971. Thus, the reason for the absence of European canker in the Sierra foothill region remains unclear.

*Hosts and pathogenicity.*—Native California hardwood species surrounding heavily infected apple orchards showed no evidence of infection or cankers caused by *N. galligena* during 1970-72; however, since it is a major pathogen of hardwoods in the Northeast, pathogenicity of several hardwood isolates on apple was tested.

Sixty percent of the inoculations with Red Delicious isolates were positive, whereas all other inoculations were negative. An apple isolate, however, was capable of infecting pear trees. Six inoculations of mature Bartlett pear trees with $2 \times 10^3$ conidia per leaf scar were all

TABLE 1. The correlation of infection by *Nectria galligena* in Red Delicious apple in 1970-71 and 1971-72 with environmental and biological parameters, presented as partial correlation coefficients and coefficients of determination

| | Partial correlation coefficients[a] | | | Partial coefficients of determination for individual variables | |
|---|---|---|---|---|---|
| Variable | 1970-71 | 1971-72 | | 1970-71 | 1971-72 |
| Leaf fall | 0.900** | 0.834** | | 0.560 | 0.793 |
| Temperature | 0.640+ | 0.256 | | 0.346 | 0.002 |
| Rain | 0.231 | 0.051 | | 0.004 | 0.010 |
| Conidia | 0.471 | 0.648+ | | 0.020 | 0.080 |
| Wind | −0.356 | ... | | 0.004 | ... |
| | | | Total $R^2$ = | 0.933 | 0.890 |
| | | | Total R = | 0.966** | 0.943** |

[a]** = $P = 0.01$; + = $P = 0.10$.

TABLE 2. Simple linear correlation coefficients among variables used in multiple regression analyses of *Nectria galligena* infection of apple for 1970-71[a] and 1971-72[a]

| | Leaf fall | Temperature | Rain | Conidia | Wind | Infection |
|---|---|---|---|---|---|---|
| Leaf fall | | 0.2609 | 0.3007 | 0.6025*[b] | 0.4159 | 0.8755** |
| Temperature | 0.5006+[b] | | 0.1793 | 0.5907* | 0.0619 | 0.5884* |
| Rain | −0.0163 | −0.3446 | | 0.3069 | 0.5998* | 0.3261 |
| Conidia | 0.4763 | 0.0978 | 0.3665 | | 0.3034 | 0.7833** |
| Wind | ... | .... | ... | ... | | ... |
| Infection | 0.8929**[b] | 0.4863 | 0.0614 | 0.6772* | ... | |

[a]Data above diagonal line are for 1970-71, those below are for 1971-72.
[b]** = $P = 0.01$; * = $P = 0.05$; + = $P = 0.10$.

positive. In most cases the twig died. The pear cankers were produced 10 days earlier than those resulting from concurrent inoculations on Red Delicious twigs. Only one pear tree in an orchard, cultivar Bartlett, was observed with European canker, and symptoms were similar to those on Red Delicious. This is the first record of this disease on pear in California. An apple isolate was capable of causing infection of quaking aspen (*P. tremuloides* Michx.) if wounds were made into the sapwood. Inoculum used was mycelium grown on sterile rye grains (T. Hinds, *personal communication*). In inoculations where cankers appeared *N. galligena* was reisolated.

*Effect of climatological and biological factors on infection.*—A positive correlation between leaf fall and infection on the three cultivars was found in 1970-71 (Fig. 1-D to F). It is obvious that a combination of factors must coincide for infection to occur. These include leaf fall at the appropriate time, moisture, conidial production, and a favorable temperature. A period of very high infection occurred on Red Delicious in early November 1970 coincident with 6 days of rain, maximum conidial production, and maximum number of hours per week between 11-16 C (Fig. 1-A, B, C, F). It was evident that when one or more of the critical factors was not present, infection generally decreased markedly (Fig. 1-A to F). For example, although a second peak leaf fall of Red Delicious occurred in December, temperatures were too low to provide a large increase in infection. Also, on 8-14 December, infection of Red Delicious decreased due to low rain and concomitant low spore production. A somewhat similar situation was observed with Golden Delicious on 10-16 November when little infection occurred, presumably due to low rainfall. Why an increase in infection occurred the next week is not clear, but rain did fall on 23 November and continued for 14 more days.

Similar data were obtained in 1971-72 when overall infection was extremely low (Fig. 2). This year, neither Golden Delicious nor Rome Beauty had any observable infection, and Red Delicious had only 14 infections in 747 marked leaf scars. The previous year, 136 out of 801 scars were infected. Two factors may have been responsible for this; low rainfall in November, with few consecutive days of rain, and comparatively fewer hours per week between 11-16 C than were observed in 1970-71. Data presented on conidia are not comparable from year-to-year, but Fig. 2-A and 2-E do show that peaks in conidia produced on Golden Delicious and Rome Beauty did not coincide with peak leaf fall of these cultivars. Although rainfall data on 5-11 November (Fig. 2-C) seem to contradict the need for several days of consecutive moisture, rain occurred on 11 November and continued for 3 additional days, thus giving the leaf scars exposed on 11 November, 4 days of moisture; enough for some infection.

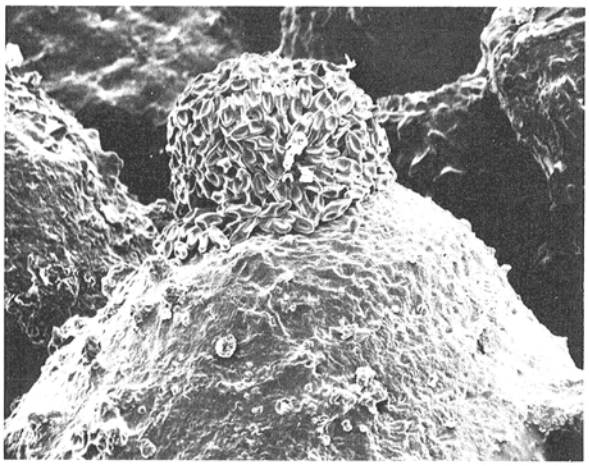

**Fig. 3.** Scanning electron micrograph of the upper portion of a perithecium of *Nectria galligena* and ascospores accumulated at the ostiole. (approximately × 250). Photo courtesy of E. Butler and L. Petersen.

The epidemiological data were subjected to multiple regression analysis (6, 7). The correlations presented are in two forms, simple correlation coefficients and partial correlation coefficients. In neither case can a cause and effect relationship be inferred. Partial correlations are quite valuable since the common influence of one or more variables is removed, and only the residual is correlated with the dependent variable of particular interest. By this means, a more specific correlation can be obtained. In both years leaf fall contributed most significantly to the variation in infection, as shown by the partial correlation coefficient and partial coefficients of determination (Table 1). In both years, if all factors except leaf fall are deleted from the regression equation generated, no statistically different multiple correlation coefficient is obtained between the two equations. Thus, on a statistical basis, to predict infection using the factors studied, only the use of leaf fall data are warranted. In 1970-71, temperature (hours between 11 and 16 C) was influential ($P \leq 0.10$) in predicting infection but not in 1971-72, probably because both infection and the number of hours in this temperature range were low. The same relationship held for rainfall in that year. The highest infection occurred when the greatest number of hours in the 11-16 C range occurred in November (Fig. 2-B, D). The nonsignificant partial correlation coefficients for rain and conidia in 1970-71 are best explained by the fact that neither was a limiting factor for infection in that year (Fig. 1-A, C). Wind probably plays little role in dispersing water-borne conidia within the canopy, and would not be expected to have a significant effect on infection. Equations generated in both years account for highly significant amounts of the variation observed in infection: 93% in 1970-71 and 89% in 1971-72 (Table 1). Various significant simple linear correlations were found in 1970-71 (Table 2). Infection was highly correlated with leaf fall, temperature, and conidia. Both leaf fall and temperature showed significant relations with conidia. In 1971-72, only leaf fall and conidia were significantly related to infection.

Multiple regression analysis of Golden Delicious and Rome Beauty data for 1970-71 yielded very similar results; that is, the strongest correlation was between leaf fall and infection. With both cultivars the equations generated accounted for 83-84% of the variation observed.

Data obtained in 1970-71 indicated the presence of relatively high concentrations of conidia during the

period from the start of infection until the end of April 1971, when rains decreased greatly. Nevertheless, the peak periods of conidial production on Red Delicious were in November and December in both years (Fig. 1-A, 2-A).

Incubation periods on the three cultivars in 1970-71 were slightly different. Symptoms were expressed in Red Delicious in approximately 2-3 months, whereas the two other cultivars required 3-4 months. In 1971-72, infections occurred only on Red Delicious and these incubated 4-5 months, possibly due to temperature differences and the dry fall, winter, and spring.

The phenomenon of delayed symptom expression, in some cases until after bud break, as reported by Crowdy (5) and Wilson (26) was observed on all three cultivars but mostly on Rome Beauty and Golden Delicious.

*Factors affecting development and dispersal of ascospores and ascosporic infection.*—The following study was done to ascertain if bursting buds were susceptible to infection in spring. Inoculation of opening buds of Red Delicious trees with a droplet of sterile distilled water containing $2 \times 10^3$ conidia resulted in only two cankers out of 60 inoculations. The inoculated twigs were wetted previous to inoculation, bagged with moistened polyethylene bags for 1 week, and kept at 13 C. Although some infection did occur, it appears that even under apparently optimum conditions our most susceptible cultivar is not very amenable to this type of infection. It is possible, however, that ascosporic infection would differ from that caused by conidia.

After initial rains in October 1970 fully mature perithecia were observed, many with spores accumulated at the ostioles (Fig. 3). This phenomenon was quite common, suggesting that ascospores, like conidia, are readily water-borne. On 10 December 1970, protoperithecia were observed on twigs in the funnel trap used for the conidial study, and their subsequent development was followed. Ascospore production started during the week of 22-28 December 1970 reaching a peak in April 1971 (Fig. 4). The fact that ascospore production peaked in April is evidence that perithecia of *N. galligena* function as do those of most other Pyrenomycetes. Very little precipitation occurred after 1 May and no more water-borne ascospores were caught. Nevertheless, collections of immature and mature perithecia and sporodochia during summer months indicated the presence of normal, viable spores of both stages. Although there usually are no rains from June to mid-September, frequent night and morning fogs seem to be sufficient to keep both stages of the fungus viable. Immature ascoscarps in April probably matured in October-November with fall rains.

The relation of rainfall to the number of ascospores caught in rainwater and in the air around a spore trap ringed with sporulating cankers is shown in Fig. 5. Airborne ascospore dispersal was not directly correlated with rainfall intensity, but ascospores caught in water were. Although monthly data do not show this clearly, daily counts of airborne ascospores indicated that with heavy rainfall, spores were rapidly washed from the air, whereas the number of spores caught in water was, in general, more directly related to amount of rainfall during the same period. It is probable that the peak load of airborne ascospores caught in December 1970 (Fig. 5-C)

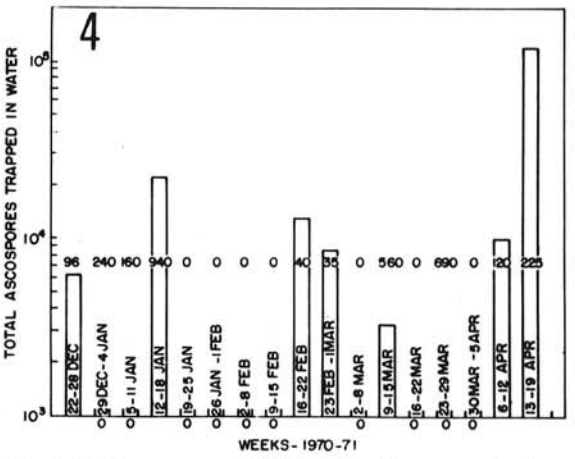


**Fig. 4.** Total ascospores of *Nectria galligena* caught in a water-funnel trap from 22 December 1970 to 19 April 1971. The funnel was filled with cankered 1-year-old twigs that contained no perithecia, but which started producing protoperithecia by early December 1970. Numbers written above the bars indicate milliliters of water caught in the trap during that week.

was due principally to the perithecia produced the previous spring with some contribution coming from the new perithecia produced in December (Fig. 4). This is probably also true for the windborne stage, but is unverified. Most ascospores caught in late winter to spring presumably were due to new perithecia initiated sometime in December and thereafter. Ascospore production was low in April-May 1971, in comparison with that in December-January (Fig. 5-B, C). However, April and May had the two highest daily ascospore counts, but due to extremely low rainfall, on a monthly basis, they were lower in total ascospore discharge.

Climatological data were correlated with daily airborne ascospore counts from November 1970 to May 1971 by multiple regression analysis. The predictors used were: (i) hours of rainfall; (ii) hours between 5 and 10 C; (iii) hours between 11 and 16 C; (iv) hours between 17 and 21 C; (v) hours of 100% RH; (vi) cm of rain; and (vii) wind velocity. All variables were tabulated on a daily basis. The dependent variable was either the number of spores caught per day or the logarithm of the number. Partial correlation coefficients were 0.3524 and 0.3698 for daily hours of rain and number of hours between 5 and 10 C, respectively. These were significant, $P = 0.05$. The partial correlation coefficient for number of hours between 11 and 16 C was 0.3222 ($P \leqslant 0.10$). All other variables were unimportant in predicting ascospore dispersal as measured by a Burkard spore sampler. Results using either the logarithm of the spore catch or arithmetic values were not significantly different. The coefficient of determination ($R^2$) showed that the multiple regression equation produced, accounted for only 38% of the variation in spore catch. Maturation of ascocarps and varying efficiency of the Burkard trap under differing climatic conditions probably accounted for having a lower $R^2$ than anticipated. Analysis of daily ascospore counts in a heavily infected orchard with abundant perithecia showed only low numbers of wind-borne ascospores. Any role that ascospores might play in

infection would almost entirely have to be as water-borne propagules, as mentioned earlier. Since *natural* conidial levels (Fig. 1-A) are greater than 100 times the ascospore levels obtained by placing 40 pieces of bark with abundant perithecia in funnel traps, it is extremely doubtful that ascospores play an important role in infection in California. Long-range spread of the disease, under these conditions, would more likely be due to planting infected stock than to wind-borne ascospores.

Fig. 6 indicates that the daily ascospore discharge of *N. galligena* is not controlled by a diurnal periodicity mechanism. As noted previously, rain is needed for discharge, but amount of discharge is not directly related to rainfall intensity. Mean ambient temperatures were within the 5-10 C range which has been found to correlate significantly with ascospore discharge.

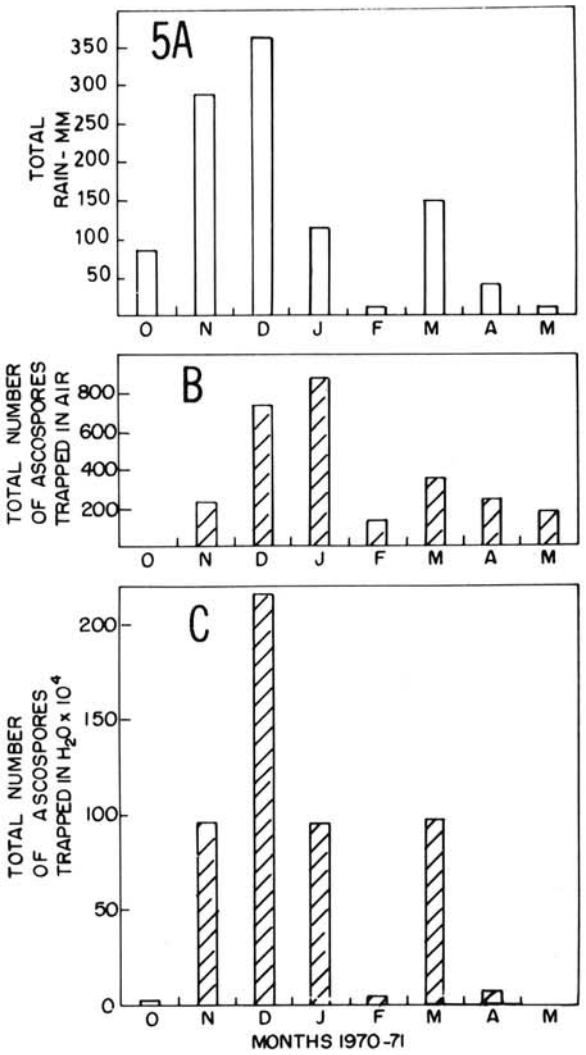


**Fig. 5-(A to C)** Monthly rainfall and ascospore production of *Nectria galligena* during the period from October 1970 to May 1971. **A)** Rainfall data from the official U.S. Weather Station, Graton, California. **B)** Number of airborne ascospores caught in a Burkard trap (10 liters of air per minute) ringed with heavily cankered limbs. Counts were made every 2 hours on a weekly basis, and combined for each month. **C)** Number of ascospores caught in water by use of a funnel trap. Twice weekly data were totaled on a monthly basis.

Observations from November 1971 to April 1972 showed very little development of perithecia in the same orchards where the 1970-71 studies were made. This paucity of perithecia may be correlated with the extremely hot summer and relatively dry October-November in 1971.

DISCUSSION.—Nichols and Wilson (22) noted that Red Delicious was the most susceptible cultivar to canker under California conditions and that Gravenstein was next in susceptibility. Golden Delicious and Rome Beauty were more resistant. Our observations over a 3-year period support this conclusion. Cankers on scaffold branches of Rome Beauty, Gravenstein, and Golden Delicious generally callus over, whereas infected Red Delicious branches are almost invariably killed.

Zagaja et al. (30) noted that Starking Red Delicious and McIntosh cultivars were very susceptible to canker in Poland. The few plantings of McIntosh that exist in Sonoma County, California, also are extremely susceptible to *N. galligena*. Golden Delicious, in France, was one of the less susceptible cultivars whereas, in contrast to our observations, Starking Delicious was resistant (3). Crowdy (5) felt that field resistance observed in Bramley's Seedling was due to formation of an effective morphological barrier against the spread of incipient leaf scar infection. Under field conditions in California, Red Delicious is more susceptible than Rome Beauty or Golden Delicious. This was substantiated by inoculations (8).

In Oregon, much greater disease severity was noted on pear than on apple (31). Anjou, Bosc, Howell, Surprise, and Old Home were the most susceptible cultivars, whereas Bartlett and Winter Nelis were generally free from canker. Only one pear tree (cultivar Bartlett) was observed with canker in California.

*N. galligena* is quite polyphagus, thus, infected hardwood stands were suspected as the source of inoculum for adjacent apple orchards. This suspicion, however, was not substantiated. On the Pacific Slope, Zeller (32) reported *N. ditissima* var. *major* Wollenweber on *Alnus* sp. *N. ditissima* is presently considered synonymous with *N. galligena* (13). Few reports on cross pathogenicity exist. Apple isolates from California will infect pear, but various hardwood isolates did not infect Gravenstein or Golden Delicious cultivars when inoculated at leaf scars without wounding. Manion and French (17) showed that red maple (*Acer rubrum* L.) isolates could canker paper birch and balsam poplar (*Populus tacamahaca* Mill.), but the paper birch isolate didn't infect balsam poplar. They used spores and mycelium in wound inoculations. Infection in California is predominantly through leaf scars, although at times pruning-wound infections have been observed. Lortie (14) was unable to infect yellow birch and sugar maple (*A. saccharum* Marsh.) through leaf scars using ascosporic or conidial inoculum. He did get good infection when various types of wounds were made. The isolate used on yellow birch was obtained from that species, but the sugar maple was inoculated with a red maple isolate. Information concerning pathogenic specialization of *N. galligena* is inadequate and needs further study.

Wiltshire (29), in England, was one of the first to observe leaf scars as the major infection court of pome trees. He reported that most infection was autumnal, but on certain cultivars spring infection was predominant. He also observed that trees which dropped their leaves later seemed to have less infection. Kennel (12), in Germany, found that artificial inoculations were most successful when done in September and October. In England, Marsh (18) showed that inoculations in October and April produced cankers, but not in November or January. Zeller (31) reported that autumn and spring infection of pears could occur. Wilson (26) was able to infect apple trees from December through February in California, but he made fresh transverse cuts through the leaf scar region to facilitate entrance of the fungus. Data presented herein indicate that November is the critical month for infection and by mid-December there is almost no new infection (Fig. 1-D, E, F, and 2-D). Spray trials in California achieved 90-99% control of European canker in the epidemic year 1970-71 with one or two fall applications of Difolatan or Bordeaux mixture (9). This tends to indicate essentially no spring infection. Some of the infection attributed by some authors to ascospores or conidia at bud burst may actually be due to "confined" lesions (latent fall infections) as described by Crowdy (5) and Wilson (26). We observed this phenomenon under both controlled and field conditions. Indeed, if the fungus becomes active after bud break, the ensuing canker would obviously occur at the base of the twig if one or more of the apical leaf scars had been infected in the autumn. The canker would be at the union of the bud and the shoot if the leaf scar subtending an intercalary bud had been infected. Therefore, it would be impossible to distinguish between spring and fall infection under field conditions, if the fungus had been latent since autumn.

Periods of peak conidial discharge may vary depending upon the geographical region. For example, Lortie and Kuntz (16) in Canada, trapped conidia only from May through August with peaks in May and June, whereas Kennel (12) in Germany, reported peak conidial production on new cankers between July and September. Bulit (3) observed the peak conidial period in the summer in France. Munson (21) and Swinburne (25), in the British Isles, indicated a period of conidial dispersal similar to that of California. Swinburne (25) showed a more lengthy peak conidial production period than in California, but the maximum production was in November also. It can be seen that conidial release does not always coincide with leaf fall, a necessary occurrence for major leaf scar infection. Summer dispersal, however, could be of great importance for infection of the fruit calyx. Under California conditions this is rarely important (20).

Lack of infection in 1971-72, as noted, may have been due to low rain and low temperatures during the infection period. Critical studies on the effect of temperature on infection are lacking; however, in California infection in the field occurred only when temperatures were between 5 and 16 C, but principally between 10 and 16 C. Peak conidial and ascosporic production also occurred at the latter temperatures. These results are in general agreement with those of Swinburne (25).

There is little information on the effect of duration of wetting of twigs on infection, but it is probable that the low infection observed in 1971-72 was due to both the low

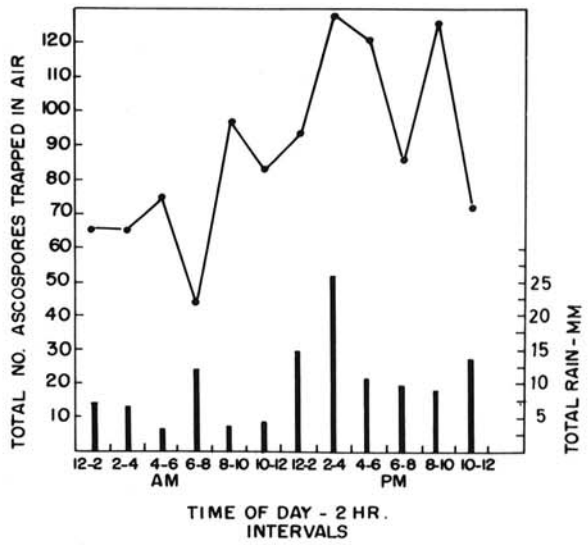


**Fig. 6.** Effect of time of day and rain on ascospore discharge of *Nectria galligena*. Ascospore values are totals for a representative 20-day period during December 1970 and January 1971.

intensity of the rains and their short duration. Wilson (26) showed that a minimum of 12 hours of moisture was needed to obtain low levels of infection on Red Delicious trees, but he wounded the leaf-scar area before inoculation. Gravenstein leaf-scar inoculations (8) resulted in 10% infection after 6 hours of wetness, but otherwise these results agree with those of Wilson, who found that several days of continuous wetness were needed for high levels of infection. Two other factors of importance are inoculum concentration and the ability of conidia to withstand desiccation. Results indicated that at least 50 conidia are needed to infect a Red Delicious leaf scar, and 500-5,000 conidia per leaf scar are needed to obtain reasonably high amounts of infection (8). This is probably due to the statistical improbability of a spore penetrating an open vessel element, since direct penetration does not occur (1). *N. galligena* conidia are sensitive to desiccation. For example, viability of conidia dropped to one-third of normal after 3 hours at 88% RH and 11 C (8). Although exact observations are lacking on rain duration in 1971-72, it is probable that days with low rainfall had periods of desiccation which adversely affected the survival of conidia and their ability to infect leaf scars. More critical studies are needed to elucidate the infection process, effect of inoculum potential, and the requirement for 6 hours of moisture. Crowdy (5) reported that conidia are sucked into the open vessel segment for up to 1 hour after leaf abscission. Thus it is difficult to explain the need for long periods of wetness to achieve infection.

Perithecial development in California would probably be similar to that in Northern Ireland (25) and Germany (12) if sufficient spring and summer rains with moderate temperatures were present.

Any infective role played by ascospores would likely be as water-borne propagules due to the common occurrence of agglutinated spore masses at the ostioles. Zeller (31) observed this phenomenon and Lortie (14)

reported it under controlled conditions with 90-100% RH. Both Lortie (15) and Swinburne (25) postulated that ascospores play a dominant role in infection, but their trapping method may have precluded obtaining a valid estimate of conidia, as both used petroleum-jelly-covered slides affixed approximately 1-2 cm from perennial cankers. The older cankers with abundant perithecia usually produce less conidia than new cankers on 1-year-old twigs. Additionally conidia, being water-borne, would not stick readily to these slides, except when raindrops containing spores dried on the slides. This method also would not accurately measure water-borne ascospores. It is possible that some of the "spring infection" in Northern Ireland (25) may be due to latent or "confined" lesions. In the spring, since only very minute areas, viz. bud scale scars or cracking leaf scars, are susceptible and since there is a high dilution factor with airborne inoculum, it would seem that extremely high levels of ascospores would be needed for infection. Under California conditions, as shown by water traps or a Burkhard volumetric sampler, few ascospores are caught at any time of the year even when baiting of the traps is done. These results make it very doubtful that ascospores play an important role in infection in either spring or fall.

Lortie (14) emphasized that perithecia need to be wetted and then permitted to begin drying before ascospores are discharged. Our field studies indicate that airborne ascospores were readily caught during rain as long as a strong "scrubbing effect" (11) did not occur. These results substantiate those of Swinburne (25) who observed that the duration, but not the intensity of rainfall is strongly correlated with ascosporic discharge.

## LITERATURE CITED


1. ASHCROFT, J. M. 1934. European canker of black walnut and other trees. West Va Agric. Exp. Stn. Bull. 261. 52 p.
2. BRANDT, R. W. 1964. Nectria canker of hardwoods. U.S. Dep. Agric., For. Ser., Forest Pest Leaflet 84. 7 p.
3. BULIT, J. 1957. Contribution a l'étude biologique du Nectria galligena Bres. agent du chancre du Pommier. Ann. Inst. Natl. Rech. Agron., Ser. C. Ann. Epiphytol. 8:67-89 (Rev. Appl. Mycol. 36:767-768. 1957).
4. CROWDY, S. H. 1949. Observations on apple canker. III. The anatomy of the stem canker. Ann. Appl. Biol. 36:483-495.
5. CROWDY, S. H. 1952. Observations on apple canker. IV. The infection of leaf scars. Ann. Appl. Biol. 39:569-580.
6. DIXON, W. J. 1970. Regression analysis. Pages 218-257d *in* W. J. Dixon, BMD; biomedical computer programs. University of California Press, Berkeley, Calif.
7. DRAPER, N. R., and H. SMTIH. 1966. Selecting the "best" regression equation. Pages 163-178 *in* Draper, N. R., and H. Smith, Applied regression analysis. John Wiley and Sons, New York.
8. DUBIN, H. J. 1972. Epidemiology and factors affecting fungicidal control of European apple canker. Ph.D. Thesis, University of California, Davis. 82 p.
9. ENGLISH, H., H. J. DUBIN, F. J. SCHICK, and K. O. ROBERTS. 1971. Chemical control of leaf scar infection by Nectria galligena in apple. Phytopathology 61:1320-1321 (Abstr.).
10. GRUBB, N. Y. 1921. Test of fungicides on apple trees. J. Pomol. 2:93-114.
11. HIRST, J. M., and O. J. STEDMAN. 1963. Dry liberation of fungus spores by raindrops. J. Gen. Microbiol. 33:335-344.
12. KENNEL, W. 1963. Zur Pathogenese des Obstbaumkrebses (Nectria galligena Bres.) am Apfel. Gartenbauwissenschaft 28:29-64.
13. LOHMAN, M. L., and A. J. WATSON. 1943. Identity and host relations of Nectria species associated with diseases of hardwoods in the United States. Lloydia 6:77-108.
14. LORTIE, M. 1962. The Nectria canker and its incitant. Ph.D. Thesis, University of Wisconsin, Madison. 118 p.
15. LORTIE, M. 1964. Pathogenesis in cankers caused by Nectria galligena. Phytopathology 54:261-263.
16. LORTIE, M., and J. KUNTZ. 1963. Ascospore discharge and conidial release by Nectria galligena Bres. under field and laboratory conditions. Can. J. Bot. 41:1203-1210.
17. MANION, P. D., and D. W. FRENCH. 1967. Nectria galligena and Ceratocystis fimbriata cankers of aspen in Minnesota. For. Sci. 13:23-28.
18. MARSH, R. W. 1939. Observations on apple cankers. II. Experiments on the incidence and control of shoot infection. Ann. Appl. Biol. 26:458-469.
19. MATSUSHIMA, T. 1961. A new medium for identification of hyphomycetes. Trans. Mycol. Soc. Jap. 2:118-120.
20. MC CARTNEY, W. O. 1967. An unusual occurrence of eye rot of apple in California due to Nectria galligena. Plant Dis. Rep. 51:279-281.
21. MUNSON, R. G. 1939. Observations on apple canker. I. The discharge and germination of spores of Nectria galligena Bres. Ann. Appl. Biol. 26:440-457.
22. NICHOLS, C. W., and E. E. WILSON. 1956. An outbreak of European canker in California. Plant Dis. Rep. 40:952-953.
23. SCHULTZ, H. B., and F. A. BROOKS. 1956. A spot climate recorder. Bull. Am. Meteorol. Soc. 37:160-165.
24. STOUT, G. L. 1963. Bureau of Plant Pathology. Pages 109-117 *in* Forty-third Annual Rep. Calif. Dep. Agric. Bull. 52.
25. SWINBURNE, T. R. 1971. The seasonal release of spores of Nectria galligena from apple cankers in Northern Ireland. Ann. Appl. Biol. 69:97-104.
26. WILSON, E. E. 1966. Development of European canker in a California apple district. Plant Dis. Rep. 50:182-186.
27. WILSON, E. E. 1968. Control of European canker of apple by eradicative and protective fungicides. Plant Dis. Rep. 52:227-231.
28. WILSON, E. E., and C. W. NICHOLS. 1964. European canker of apple. Calif. Dep. Agric. Bull. 53:151-155.
29. WILTSHIRE, S. P. 1922. Studies on the apple canker fungus. I. Leaf scar infection. Ann. Appl. Biol. 8:182-192.
30. ZAGAJA, S. W., D. F. MILLIKAN, W. KAMINSKI, and T. MYSZKA. 1971. Field resistance to Nectria canker in apple. Plant Dis. Rep. 55:445-447.
31. ZELLER, S. M. 1926. European canker of pomaceous fruit trees. Oregon Agric. Exp. Stn. Bull. 222. 52 p.
32. ZELLER, S. M. 1935. Some miscellaneous fungi of the Pacific Northwest. Mycologia 27:449-466.