Effect of Water Stress in Cypress on the Development of Cankers Caused by *Diplodia pinea* f. sp. *cupressi* and *Seiridium cardinale*

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ABSTRACT

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When greenhouse-grown cypress seedlings were predisposed to extreme water deficit (ψ of −4.5 to −5.5 MPa) before or after inoculation, the expansion of Diplodia cankers on stems was enhanced. None of the water deficit regimes affected the development of Seiridium canker. Under summer field conditions, the development of Diplodia canker on stems and branches was enhanced in nonirrigated plants, compared with surface-irrigated ones. The expansion of Seiridium cankers on branches, however, was markedly decreased by the water stress. These results implicate drought as a factor contributing to outbreaks of Diplodia canker during the unseasonably dry years of 1984–1986.

In recent years, two important canker diseases of cypress (*Cupressus sempervirens* L.), caused by *Seiridium cardinale* (Wagener) Sutton & Gibson (*Coryneum cardinale* Wagener) and *Diplodia pinea* (Desm.) Kickx, Petrak, & Sydow f. sp. *cupressi*, were found in Israel (9,10). Both Seiridium and Diplodia cankers, characterized by gum oozing and bark discoloration on the stem and branches, lead to branch dieback and whole-tree mortality. During the years 1984–1986, which were characterized by low annual rainfall, both the incidence of Diplodia canker and the severity of infection were increased. Water stress during the summer months of these years was suspected to have favored the development of Diplodia canker, because enhanced invasion of the stems of water-stressed pine seedlings has been demonstrated (1). Seiridium cankers are most common on young, fast-growing trees (11); there are no indications to relate the epidemiology of Seiridium canker to any stress factor. Water stress in trees, either as a predisposition factor or during the incubation period, has been shown to affect the development of various canker-causing pathogens (4,6). The objective of the present work was to study the influence of water stress on the development of cypress cankers

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Table 1. Influence of preinoculation plant water potential on the development of Diplodia and Seiridium cankers on stems of potted cypress saplings at two temperatures in the greenhouse

Interval between last watering and inoculation (days)	22 C: Water potential at inoculation (MPa)	22 C: Canker length[y] (mm) Diplodia canker	22 C: Canker length[y] (mm) Seiridium canker	26 C: Water potential at inoculation (MPa)	26 C: Canker length[z] (mm) Diplodia canker	26 C: Canker length[z] (mm) Seiridium canker
2	—	—	—	−0.40	32.4 a	38.6 a
4	−1.20	27.3 a	35.7 a	−0.98	38.0 a	29.4 a
12	−4.52	64.7 b	42.7 a	−3.79	39.2 a	37.2 a
24	−5.50	117.9 c	40.2 a	−5.50	96.6 b	33.6 a

[y]Canker length was recorded 35 days after inoculation. The data are means of 10 replicates from different trees. Means in the same column followed by the same letter do not differ significantly ($P = 0.05$) according to Duncan's multiple range test.
[z]Canker length was recorded 28 days after inoculation. The data are means of 15 replicates from different trees. Means in the same column followed by the same letter do not differ significantly ($P = 0.05$) according to Duncan's multiple range test.

caused by the two different pathogens, under greenhouse and field conditions.

MATERIALS AND METHODS

Greenhouse and field experiments. In temperature-controlled greenhouse experiments, 4-yr-old cypress saplings (approximately 10 mm in diameter and 90–100 cm tall) planted in red sandy loam soil in 2-L polyethylene bags were used for inoculations. In field experiments, 6-yr-old cypress saplings (16–20 mm in diameter and 130–150 cm tall) grown in red sandy loam soil were inoculated. Various levels of water stress were obtained by withholding irrigation and allowing plants to deplete soil moisture over different lengths of time before or after inoculation.

Inoculation. Bark inoculations were made by removing the outer layers of the bark (5 × 3 mm) with a knife and placing a 5-mm disk of a culture of either *S. cardinale* or *D. p.* f. sp. *cupressi* over the exposed tissue. The inoculated area was covered with wet cotton and wrapped with plastic ribbons; both were removed after 5 days. In greenhouse experiments, each of 10–15 stems was inoculated with one pathogen at a point 40 cm below the apex. In field inoculations, 15 stems and 10 branches of different plants were inoculated with one pathogen 60 and 15 cm below the apex, respectively. At the end of the incubation period (28–40 days after inoculation) the periderm at the canker margin was removed, and the length of the bark discoloration was measured.

Measurement of water potential. The xylem water potential of plants was monitored with a pressure chamber (8) at the terminal 10- to 15-cm section of two twigs from each sapling for each reading. Pressure readings were taken between 5 and 8 a.m. The measurement of plant water potentials (ψ) lower than −5.5 MPa was discontinued because of safety regulations.

RESULTS

Greenhouse experiments. The predisposition of cypress saplings to various water potentials affected the development of Diplodia canker but not that of Seiridium canker (Table 1). At 22 C the expansion of Diplodia cankers was gradually enhanced as the duration of the water deficit and its magnitude increased prior to inoculation. At 26 C a similar trend was observed, but the cankers were significantly longer only when seedlings were subjected to extremely low ψ (Table 1). Postinoculation water stress significantly enhanced the expansion of Diplodia cankers only when the water deficit was maintained for 24 days, reaching ψ values of at least −5.5 MPa. Seiridium canker was not significantly affected by any water regime (Table 2).

Table 2. Influence of postinoculation plant water potential on the development of Diplodia and Seiridium cankers on stems of potted cypress seedlings at 26 C in the greenhouse

Interval from inoculation until first watering[y] (days)	Water potential before watering (MPa)	Canker length[z] (mm) Diplodia canker	Canker length[z] (mm) Seiridium canker
4	−0.98	47.6 a	36.7 a
12	−3.55	46.5 a	37.8 a
24	−5.50	136.0 b	45.5 a

[y]All plants were watered at inoculation and irrigated regularly after the first postinoculation watering.
[z]Canker length was recorded 35 days after inoculation. The data are means of 10 replicates from different trees. Means in the same column followed by the same letter do not differ significantly ($P = 0.05$) according to Duncan's multiple range test.

Field experiments. The development of Diplodia canker on stems was not influenced by various preinoculation surface irrigation regimes (Table 3). However, when plants that were not irrigated before inoculation were also deprived of water after inoculation, larger lesions developed (Table 3). Disease development on branches inoculated with either pathogen was studied only in extreme treatments (Table 3). Diplodia canker was enhanced by prolonged water stress, but the expansion of Seiridium cankers under such conditions was significantly decreased (Table 3). When the unwatered plants were grouped according to ranges of ψ at symptom recording, the elongation of Diplodia cankers was influenced by the degree of water stress (Table 4).

DISCUSSION

With potted saplings, neither predisposition, sensu Yarwood (12), nor postinoculation water stress affected the development of Seiridium canker, but both enhanced the expansion of Diplodia cankers under extremely negative values of ψ. Recently, Chou (2) found that stems of *Pinus radiata* D. Don became infected by *D. pinea* only when needle ψ dropped to −2.5 MPa. Schoeneweiss (5) found an increase in bark and wood colonization by *Botryosphaeria dothidea* (Fr.) Ces. & De Not. in stems of red-osier dogwood and sweetgum seedlings which were wilted with ψ of −0.6 to −0.3 MPa. Seedlings were not kept at constant ψ, as described by Schoeneweiss (5), but were exposed to a gradually increasing water stress until disease development was accelerated. Although water stress accelerated the expansion of Diplodia cankers, it was not a prerequisite for canker development, as was reported with some weak or unaggressive pathogens (6). By the scheme suggested by Schoeneweiss to express the relation between pathogen aggressiveness and the level of water deficit (7), the aggressiveness of *D. p.* f. sp. *cupressi* and

Table 3. Influence of plant water potential on the development of canker on stems and branches of cypress seedlings grown in field plots under different pre- and postinoculation irrigation regimes

Frequency of watering[x]		Water potential (MPa)			Canker length on branches[z] (mm)	
Pre-inoculation	Post-inoculation	On inoculation day	At symptom recording	Diplodia canker length on stems[y] (mm)	Diplodia canker	Seiridium canker
1-wk interval	1-wk interval	−1.18	−1.48	44.0 a	62.0 a	63.2 a
4-wk interval	1-wk interval	−2.96	−2.03	48.4 a	—	—
No watering	1-wk interval	−3.67	−2.27	44.8 a	—	—
No watering	No watering	−3.12	−3.97	91.6 b	120.8 b	28.0 b

[x]The last rain fell on 18 April 1987. Watering started on 18 June 1987 as specified. Plants were inoculated on 12 August, and canker length was recorded on 21 September 1987.

[y]The data are means of 15 replicates from different trees. Means followed by the same letter do not differ significantly ($P = 0.05$) according to Duncan's multiple range test.

[z]The data are means of 10 replicates from different trees. The means in each column differ significantly ($P = 0.05$) according to Duncan's multiple range test.

Table 4. Relation between plant water potential of field-grown cypress seedlings without irrigation and the development of Diplodia canker on stems

Water potential[y] (MPa)	Canker length[z] (mm)
−2.2 to −2.7	55.6 a
−3.2 to −3.6	85.2 ab
−4.3 to −4.7	85.6 ab
−5.5	126.0 b

[y]Plants were grouped according to ranges of water potential at symptom recording, 40 days after inoculation.

[z]The data are means of six to eight replicates. Means followed by the same letter do not differ significantly ($P = 0.05$) according to Duncan's multiple range test.

S. cardinale could be classified as medium and high, respectively. In our experiments, values of ψ such as −5.5 MPa were tolerated by the seedlings. These extreme ψ levels are very low compared with those reported for other conifers (8). *C. sempervirens* is a drought-resistant tree and succeeds in areas with a mean annual rainfall of 350 mm (3).

Results of the field experiment agree with greenhouse findings for *D. p.* f. sp. *cupressi*, in which the development of cankers was significantly increased only when plants were exposed to extreme stress conditions. In contrast, the expansion of Seiridium cankers under field conditions was markedly limited, whereas in the greenhouse it was not affected by any water stress. The inhibition of infection by *S. cardinale* during the summer has been observed (10). Results of the field experiment confirm this observation. The enhanced development of Diplodia canker under conditions of high water stress supports our hypothesis that the increased severity of Diplodia canker during 1984–1986 could have resulted from prolonged drought.

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