Leptosphaeria korrae, a Cause of the Spring Dead Spot Disease of Bermudagrass in California

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ABSTRACT

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During the past 5 yr, spring dead spot has become the most destructive disease of bermudagrass (*Cynodon dactylon*) in California. Symptoms of the disease and signs of the fungus on roots, stolons, and culm bases are described. *Leptosphaeria korrae* was shown to be one cause of the disease. Three sterile, dematiaceous fungi resembling *Leptosphaeria* spp. caused similar symptoms. *L. korrae* caused greater damage to Tifgreen bermudagrass plants grown at 13 than at 24 C.

From 1978 to 1984, Tifgreen bermudagrass (*Cynodon dactylon* (L.) Pers.) home lawns in the San Joaquin Valley of California, from Fresno to Bakersfield, have been commonly and severely affected with a disease resembling spring dead spot (SDS). SDS is a destructive disease of bermudagrass of unknown etiology that has occurred in the southeastern and south central United States for the past 35 yr (1,4). A disease with the same name and symptoms has occurred in Australia since 1961, and in 1967, Smith (2,3) demonstrated that the disease was caused by two loculoascomycetous fungi, *Leptosphaeria narmari* and *L. korrae* (5). The cause of the disease in the United States has not yet been demonstrated, however. The California SDS disease appears in the spring, after the breaking of winter dormancy, as circular patches of dead and dying bermudagrass plants (Fig. 1). Affected areas vary from a few centimeters to 0.5 m or more in diameter. Diseased areas are depressed because the upper leaf blades, upper leaf sheaths, and upper culms of the diseased bermudagrass plants are dead, bleached, and desiccated, and the roots, stolons, and basal portions of culms are affected with a brown to black dry rot.

The disease usually does not appear until the bermudagrass plants are 2 yr or older, but in subsequent years, the disease frequently reappears in or near the same spots. By summer, dead patches usually have filled in with new growth spreading in from the sides and/or with new growth arising from surviving plants within the patches. In 1983, the California SDS disease appeared for the first time on golf greens and fairways in the southern part of the state. The fact that the disease has appeared on home lawns and golf courses not only in the hot inland San Joaquin Valley but also along the cool California coast suggests that the fungus may be destructive over a wide range of soil types and climates. The disease occurs most commonly on Tifgreen bermudagrass but also occurs on the cultivars Santa Ana and Common.

This paper describes the signs and symptoms of the SDS disease on bermudagrass in California and results of experiments that establish *L. korrae* as one cause of the SDS disease in California. In addition, three isolates of a fungus that were sterile, gray, and septate (resembling *Leptosphaeria* spp.) were also shown to cause the disease.

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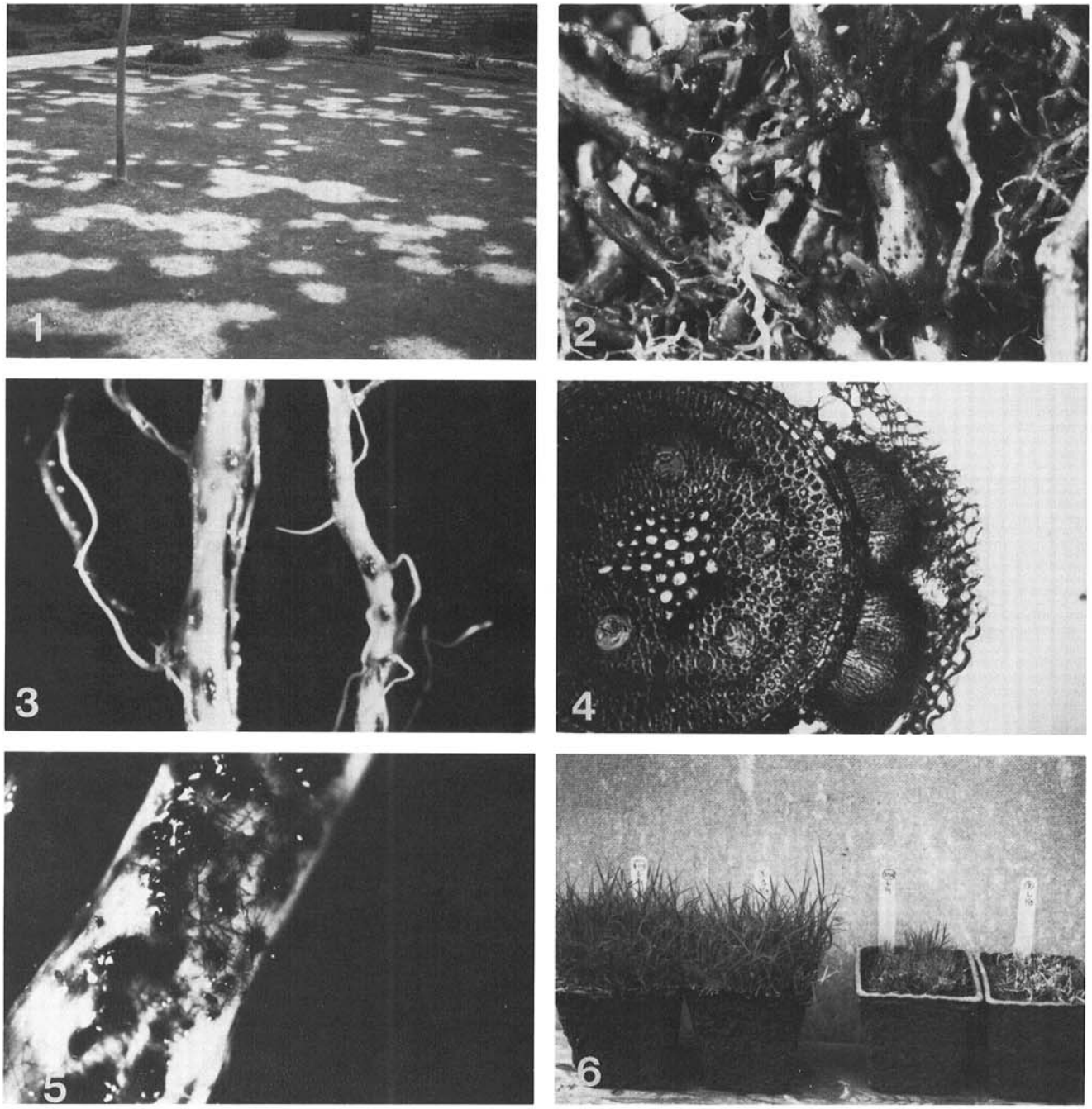
MATERIALS AND METHODS

Isolations. Tifgreen bermudagrass plants with symptoms of SDS were removed from a 3-yr-old home lawn in Visalia, CA, and brought to the laboratory for isolation. Stolons showing brown lesions were washed in running tap water for 2 hr, and incipient to moderately advanced lesions were removed with a sterile scalpel. The pieces were disinfested in 0.5% NaOCl for 1 min, rinsed in sterile water, and plated on acidified water agar (pH 4.9). Sclerotia that formed on the surfaces of the stems, stolons, and leaf sheaths were round, flat or cushion-shaped, and brown (Figs. 2 and 5), whereas sclerotia that formed within the cortical region of the roots were fusiform and brown to black (Fig. 3). Both types of sclerotia were surface-disinfected in 0.5% NaOCl for 1 min and submerged in acidified water agar. When septate hyphae emerged from the lesions and from the two types of sclerotia, hyphal-tip transfers were made to 9-cm petri plates containing potato-dextrose agar (PDA). These sterile fungi, which all appeared identical, were incubated in both light and darkness on a laboratory bench.

Inoculations. The bermudagrass hybrid Tifgreen (*C. dactylon* (L.) Pers. × *C. transvaalensis* (Burt-Davy)) was used in the inoculation trials because of its susceptibility to SDS and its widespread use. Because Tifgreen must be vegetatively propagated, the apical portion of the stems (sprigs) were removed from mother plants that were grown without being clipped. The sprigs were disinfested in 0.5% NaOCl for 10 min and transplanted into 4-in. plastic pots containing steam-treated U.C. soil mix and allowed to grow on a greenhouse bench for 3 mo. Before use, some plants were uprooted, soil was washed off the roots, and representative areas of the plants

were plated onto acidified PDA. Because these plants appeared healthy, vigorous, and free of fungi, the remainder were used as follows: Each 4-in. pot was filled one-third with steamed U.C. soil mix, then the plants (with half of their lower root system removed) were placed in the pot and covered with a 1:6 mixture (v/v) of cornmeal-sand inoculum of the fungus and steam-treated U.C. soil mix. The inoculum was prepared by pipetting 10-ml sterile mycelial fragments from a 1-mo-old PDA culture into sterile cornmeal sand (250 ml) contained in a 15-cm petri plate. Inoculum consisted of fungi originating from the stolon lesions (SDS1), from the stem sclerotia (SDS2), or from root sclerotia (SDS3). Control plants were treated as described previously, using sterile cornmeal-sand mixture. Because the disease appears in the field on plants emerging from winter dormancy, half of the plants were incubated on a bench in the glasshouse and half were incubated on a bench in a lathhouse from January to April 1983.

The effects of growth of plants at 13 and 24 C on the SDS disease was carried out using procedures identical to those used in the previous experiment, except two different fungal isolates were used. One isolate (SDS1) used in the previous experiment was recovered from a diseased stolon and another (SD54) was isolated from a single germinating ascospore obtained from a pseudothecium that formed on a spring dead spot-affected stolon collected in Bakersfield, CA. This fungus was identified by J. Walker, mycologist with the New South Wales Department of Agriculture, Australia, as *L. korrae* (5), one of the



Figs. 1–6. (1) Tifgreen bermudagrass lawn affected with spring dead spot (SDS). **(2)** Stolons and roots affected with dry brown rot typical of SDS. Note round sclerotia on the stolon surfaces. **(3)** A second type of sclerotia that is formed within the cortex of infected roots. Note fusiform shape. **(4)** Close-up of two sclerotia that were formed within the cortex of the infected root. Note hyphae occurring within the protoxylem and metaxylem vessels. **(5)** Close-up of the external type of sclerotia that are formed on stolon surfaces and lower stem bases. Note associated hyphae. **(6)** Effect of temperature on SDS development. The plant on the left in each pair was the healthy, uninoculated control and the plant on the right was inoculated. The pair of plants on the left was grown in the greenhouse and the pair on the right was grown in the lathhouse during winter 1982–1983.

causal agents of SDS established by Smith (2) in 1967. Immediately after inoculation, plants were placed for 2 mo in each of two controlled-temperature facilities, one regulated at 13 C and the other at 24 C. Each unit received 12 hr of light daily.

RESULTS

Symptoms and signs. Culm bases, buds, rhizomes, stolons, and roots may be mildly to severely affected with a black to brown dry rot (Fig. 2), which starts out on all organs as small, brown flecks. Secondary roots as well as primary roots are affected. Occasionally, secondary roots are infected and killed before they emerge from the primary roots. More frequently, the secondary roots are infected at the point where they are joined to the primary root. Gray, septate runner hyphae consisting of from one to as many as four hyphae are joined together at intervals and grow more or less parallel on the root surface. Hyphae commonly appeared in and between the cortical cells. Brown to black fusiform sclerotia consisting of parallel hyphae arising internally from the endodermal tissues (Figs. 3 and 4) appeared in the cortex of the infected primary roots (Fig. 4). The occurrence and numbers of sclerotia formed in the roots varied greatly. Microscopic examination of thin sections of living infected roots not only revealed the internal sclerotia but also hyphae and a brown occluding substance in the primary and secondary xylem (Fig. 4). Infection of the stele is relatively uncommon and occurs late in disease development.

In addition to the sclerotia formed internally in the roots, sclerotia are also produced externally on the basal leaf sheaths, on the basal portion of the culms beneath the leaf sheaths, and on the stolons (Fig. 2). These external sclerotia are brown, round, and cushion-shaped and have a network of gray, septate hyphae associated with them; when young, they tend to be flat (Fig. 5). The external sclerotia and associated hyphae are easily detached from the plant surface to which they adhere. Frequently, brown, elongate lesions occur beneath the sclerotia on the stolons and basal portion of the stems.

Inoculations. In April, 4 mo after inoculation, inoculated bermudagrass plants in the lathhouse partially or completely failed to recover from winter dormancy, whereas uninoculated control plants recovered in a normal manner (Fig. 6). In contrast, a comparable set of inoculated plants incubated for the same period in the greenhouse was slightly stunted (10%) and mildly chlorotic, with a 29% reduction in the number of stems (Fig. 6). All roots of inoculated plants were affected with typical SDS symptoms, but symptoms on stolons and lower stems were much milder in plants grown in the greenhouse than in those grown in the lathhouse. Symptoms of SDS were identical whether the inoculum arose from the fungus isolated from lesions on the stolons or from either of the two types of sclerotia. Control plants remained healthy. When isolations were made from diseased plants inoculated with the sterile fungus originating from stolon lesions or the two types of sclerotia, the same sterile, gray, septate fungus was always recovered. On PDA, the fungi were first white, and then gradually became gray to black.

Growth of foliage and roots of healthy bermudagrass was vigorous at 24 C but minimal at 13 C. Both isolates produced typical symptoms of the SDS disease, but symptoms were much more severe at 13 than at 24 C, and isolate SDS4 (from ascospore) was more pathogenic than isolate SDS1. Plants infected with either isolate at 13 C grew extremely slowly, and 2 mo later, all plants in 10 of 10 pots inoculated with isolate SDS4 were dead, whereas only plants in three of 10 pots inoculated with isolate SDS1 were dead. At 13 C, typical severe symptoms and signs of the fungus (root and stem sclerotia) were evident on the roots as well as large areas of the buds, stolons, and lower stem bases. In contrast, at 24 C, inoculated plants were only slightly affected, but roots were affected much more than buds, stolons, and culms. Isolate SDS4 was more pathogenic than isolate SDS1 since the former caused more root browning, less root formation, and a much higher incidence and severity of lesion formation on the buds, stolons, and lower stem bases. Although a few root sclerotia were evident at 24 C on an occasional infected root, stem sclerotia were not observed on any of the affected stolons or stem bases infected with either isolate. When isolations were made from diseased plants, the same sterile, gray, septate fungi were always recovered.

DISCUSSION

The SDS disease of bermudagrass in Australia was shown by Smith (2,3) to be caused by two fungi, *L. narmari* and *L. korrae*. According to Smith, *L. narmari* is the usual cause of SDS in Australia, whereas *L. korrae* has been isolated extremely rarely. Smith stated that both fungi were sterile on agar media and that the mycelia were at first white on PDA. Both gradually became a gray to black, but *L. narmari* differed from *L. korrae* in that it was first white, then buff, and finally gray to black.

The fungi causing the SDS disease that we have worked with in California were also sterile in culture and were first white, then gradually turned gray to black. This suggests, but does not prove, that our sterile isolates may also be *L. korrae* rather than *L. narmari*. Isolate SDS4, however, is definitely known to be *L. korrae* since it originated from a single ascospore that was morphologically identical to *L. korrae*. This means that we have shown that at least one cause of the SDS disease in California is *L. korrae*. Whether the remaining isolates (ie, SDS1, SDS2, and SDS3) that were sterile are *L. korrae* or *L. narmari* is not known because to date they have not produced pseudothecia and concomitant ascospores either in culture or in SDS-affected plants. The possibility that the unidentified isolates may be a third undescribed fungus is not probable since the symptoms of the disease, associated signs of the fungus, and growth characteristics of the fungus on PDA were identical to those described by Walker and Smith (5) for *L. narmari* and *L. korrae* in Australia.

The fact that the disease was very severe on inoculated, slow-growing bermudagrass plants incubated during the winter in the lathhouse or in an environmental growth chamber regulated at 13 C suggests that the fungus is more damaging under cool conditions. These conditions may favor development of the pathogen more than they favor growth of the host. In our experiments, the bermudagrass plants were not mowed. It is possible that had the plants been mowed weekly, they would have been weakened further and therefore might have incurred even greater damage by the fungus. SDS-affected plants grown at 13 C were not dormant; this indicates that the severe damage caused by the fungi in the field during the winter does not depend on the bermudagrass plants being dormant. Information is needed on whether dormant plants are attacked, whether they are more susceptible than actively growing plants, and the approximate minimum and maximum temperatures at which both species of *Leptosphaeria* cease to attack plants.

A disease of bermudagrass with the same field symptoms and the same name also occurs in the southeastern and south central United States. Although the disease appears to be transmissible (1), the causal organism of the disease has not yet been determined.

ACKNOWLEDGMENT

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LITERATURE CITED

1. Lucas, L. T. 1979. Spring dead spot of bermudagrass. Pages 183-187 in: Advances in Turfgrass Pathology. B. G. Joyner and P. O. Larsen, eds. Harcourt Brace Javanovich, Duluth, MN.
2. Smith, A. M. 1967. Spring dead spot of couch grass turf in New South Wales. M.Sc. thesis. University of Sydney, Sydney, Australia. 114 pp.
3. Smith, A. M. 1971. Spring dead spot of couch grass turf in New South Wales. J. Sports Turf Res. Inst. 47:54-59.
4. Wadsworth, D. F., and Young, H. C., Jr. 1960. Spring dead spot of bermudagrass. Plant Dis. Rep. 44:516-518.
5. Walker, J., and Smith, A. M. 1972. *Leptosphaeria namari* and *L. korrae* sp. nov., two long-spored pathogens of grasses in Australia. Trans. Br. Mycol. Soc. 58:459-466.