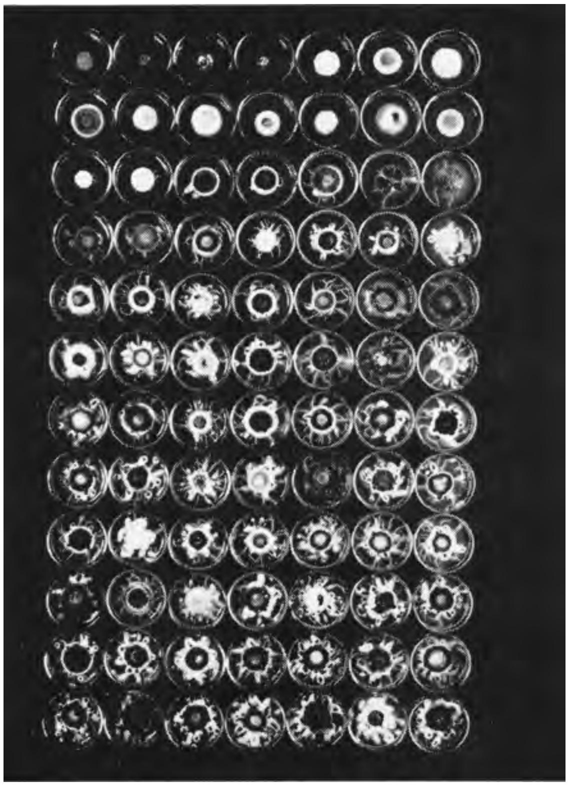
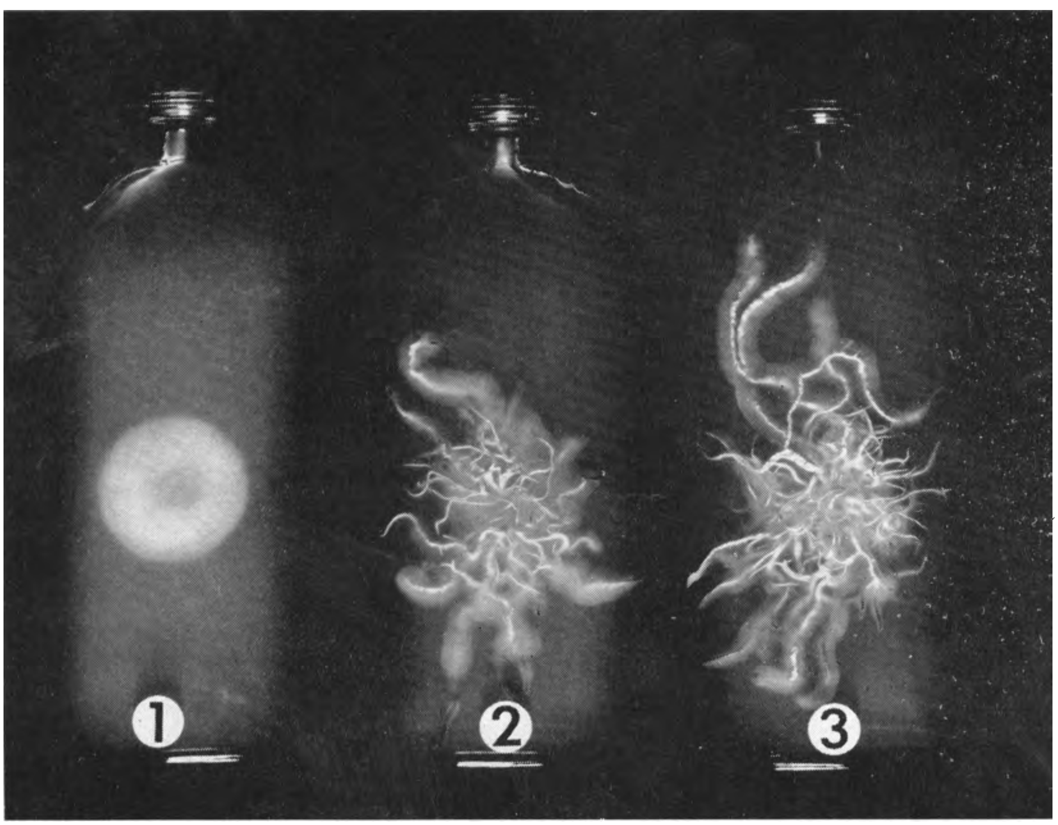


Variation in cultural characteristics of Armillaria mellea. Eighty-four isolates of the fungus from different hosts and from different parts of California were grown under similar conditions to show the variation which occurs naturally in this fungus.

Research on *Armillaria* . . . THE OAK ROOT

ROBERT D. RAABE · A. R. WEINHOLD · W. D. WILBUR

Influence of ethyl alcohol on rhizomorph production by Armillaria mellea: (1) basal medium; (2) basal medium plus 150 ppm ethyl alcohol; and (3) basal medium plus 1500 ppm ethyl alcohol.



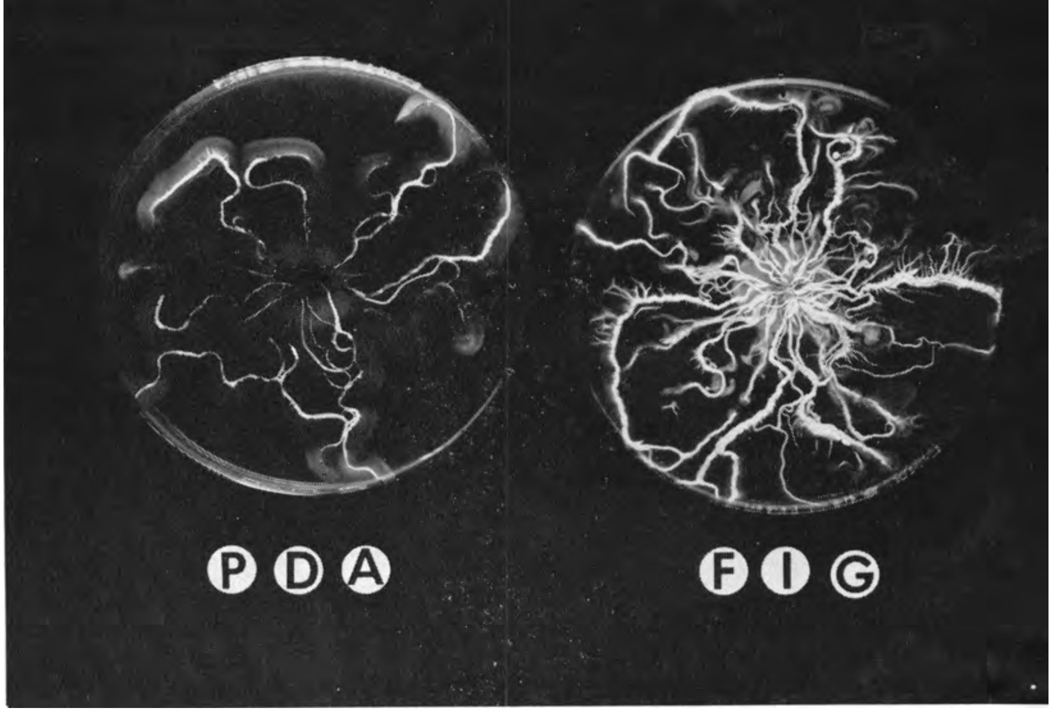
ARMILLARIA MELLEA, the oak root fungus, is an important disease-producing organism in California. The common name of the fungus is somewhat misleading for, though it is found on oaks, it also attacks and damages or kills many other plants including such crop plants as peaches, almonds, citrus, grapes, walnuts, apricots, plums, and many ornamentals.

Although it was apparent as early as 1881 that this disease was important to California agriculture, most of the early work consisted of listing the plants found susceptible to attack. One of the many observations was the association between the disease and susceptible crops planted on land cleared of oak trees—probably leading to the name "oak root fungus."

One of the earlier U. C. researchers, W. T. Horne of the Department of Plant Pathology, Berkeley, showed (in 1914) that cutting roots by ditching around infected plants slowed the progress of the fungus. That same year, he also suggested the use of carbon bisulfide as a control measure but apparently did not experiment with the material. There is evidence however, that at least one lemon grower in southern California began treating in-

mellea,

FUNGUS



Effect of wood extract on the growth of Armillaria mellea in culture. Fungus to left is growing on a standard laboratory culture medium. Fungus to right is growing on a medium extracted from fig wood.

fested areas and diseased trees with carbon bisulfide as early as 1916.

The first published recommendation for use of this chemical in controlling the disease appeared in 1926 in "Citrus Diseases and Their Control," by H. S. Fawcett of the Citrus Experiment Station at Riverside, who was aware of the field work carried on by the growers. In a later edition of the book he also reported on some of the experimental work of Donald E. Bliss, who joined the Citrus Experiment Station staff around 1933. Although Bliss experimented with carbon bisulfide, he did not report his findings until 1941, two years after Harold E. Thomas and a student, Lewis Lawyer, of the Department of Plant Pathology at Berkeley, published on the use of carbon bisulfide for controlling the disease.

One of the most significant of many articles by Thomas was in regard to the infection process in which he showed the importance of the rhizomorphs (black, rootlike strands making up the vegetative part of the fungal plant). He also started testing fruit trees and ornamental plants to find their resistance to the fungus. This project was stopped briefly following the war, reactivated in 1953, and was summarized recently in *California Agriculture* (March 1966). Thomas was also successful in making both greenhouse and field inoculations.

Meanwhile, at Riverside, Bliss also succeeded in making inoculations in the greenhouse. In his research he found that temperatures were important because plants grown at conditions unfavorable for their optimum growth were more susceptible to *A. mellea.* He also found that roots fumigated with carbon bisulfide were quickly overgrown by another soil-borne fungus, *Trichoderma viride.* He theorized that this fungus produced an antibiotic material which aided in the destruction of the oak root fungus following fumigation. This work is being continued. It has also been found that when *Armillaria* is weakened by heating, drying, or sub-lethal doses of carbon bisulfide, the fungus will survive in sterile, but not in natural soil. It appears that the treatments reduce the vigor of *Armillaria* so that it can be attacked and killed by *Trichoderma.* A testing program was also started to determine the possibility of resistance in plants adapted to warmer areas—particularly *Citrus* species. In such tests, it has been shown that although the commonly used rootstocks—sweet orange, lemon, mandarin, trifoliate, Troyer and lime—are susceptible, Brazilian sour is almost completely resistant to the fungus, and that Yuzu and Ichang hybrids have enough resistance to warrant continued testing. Unfortunately, the rootstocks showing resistance to the oak root fungus lack resistance to other important diseases so their use is not practicable at present. Field tests are being continued, however.

Researchers at Riverside are now conducting further experiments on control of the fungus by fumigation. It has been shown that carbon bisulfide or methyl bromide will control *Armillaria* in roots as deep as 10 ft. A concentration of 500 ppm of methyl bromide persisting seven days or longer will predispose *Armillaria* to attack by *Trichoderma.* This concentration in a sandy loam soil can be obtained with a surface application of 1 lb per 100 sq ft and then covering the soil with a 6-mil polyethylene tarp. Fumigant concentrations and flow rates are being studied on several soil types and at different moisture levels to enable workers to use this type of fumigation with precision. Work at Riverside also includes further study of an early practice by which *Armillaria* may be controlled through exposure to air of the crowns of infected plants.

Present work at Berkeley is being carried on in several general areas. One is in regard to the nutrition of the fungus with special reference to rhizomorph formation. In these studies it has been shown that ethyl alcohol and other closely related alcohols stimulate the production of rhizomorphs in culture media (see photo). Thus, many studies on the growth and development of the fungus can be made in a completely defined chemical medium.

In another phase of the project, an investigation is being made of the infection process with emphasis on mode of penetration, alterations in host tissue during and following invasion, and the role that the fungus plays in these changes. The information gained from these studies will result in better understanding of the formation and functions of the rhizomorphs so that methods of control may be developed which will interfere with their development and functioning.

Other current work is directed toward understanding the effect of environment upon the growth and development of the fungus, with particular reference to rhizomorph production. These studies have shown that the fungus is extremely variable in its growth characteristics on culture media and that this variation is inherited (see photo). Studies have also shown that different isolates of the fungus vary in the severity of their attacks upon a given type of plant. In connection with these studies, greenhouse inoculation techniques have been improved so that considerably less time is needed to infect plants. In addition, laboratory studies have shown that there are naturally occurring substances in some woody plants which greatly stimulate rhizomorph production (see photo). One of the objectives is to isolate and identify these stimulatory substances. The effects of temperature, light, and varying levels of oxygen and carbon dioxide upon the growth of the fungus are also being studied. It has been found that *Armillaria* is inhibited by light—particularly by the shorter, or blue, wavelengths. It also has been shown that the fungus grows well at reduced levels of oxygen and at very high levels of carbon dioxide. These characteristics help to explain its survival in roots considerably below the surface of the soil. The environmental factors influencing the production of the fruiting bodies (mushrooms) of the fungus are also being investigated; though fruiting bodies have been produced under laboratory conditions, as yet not all factors involved in fruiting have been determined.

Research on control of this important fungus and the disease it causes has been and is being approached from three directions: control through resistance; control by the use of chemical fumigation; and biological control. Although the fungus occurs worldwide throughout the temperate zones, much of our knowledge of how the fungus grows and how it is controlled has been developed at the University of California.

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Pre-emergence herbicides offer the advantage of reducing early competition in seedling alfalfa stands. The disadvantages of present pre-emergence herbicides is that they have to be incorporated, which adds extra expense, and they do not control all of the broad-leaved weeds. Benefin shows promise as a pre-emergence herbicide due to good selectivity in alfalfa and full-season grass control. Post-emergence herbicides have the advantage that treatment is not necessary until the problem exists. The dinitros and 2,4-DB (ester) will control weeds if they are small. Bromoxynil (not presently registered for use on alfalfa) kills a larger number of weeds and larger weeds than the other post-emergence herbicides. With the use of post-emergence herbicides, timing the application when the weeds are small and when most of them have emerged is essential. This report results from three years of trials using pre- and post-emergence applications of various herbicides. This information does not constitute a weed control recommendation by the University of California. For current weed control recommendations, local farm advisors should be consulted.

WEEDS ARE A MAJOR PROBLEM in the establishment of a new stand of alfalfa. In seedling alfalfa a severe weed problem can completely crowd out the stand. When the weeds are this abundant the young plants cannot compete for soil nutrients, sunlight, and moisture. If the weeds are less abundant, they may not crowd out the stand, but they weaken the young plants and retard growth, which delays the first cutting. The quality of hay is greatly reduced when weeds are present, resulting in a lower selling price.

Many herbicides have been tested in alfalfa field trials in recent years. Some of these show a high degree of selectivity, producing clean, weed-free alfalfa that sells for as much as $15 per ton more than weed-contaminated alfalfa.

Pre-emergent herbicides

In 1963 a large-scale field trial was established with EPTC (Eptam) to compare methods of application (granules versus liquid dripped in irrigation water), rates, and effectiveness for weed control. Because of the size of the plots (25′ by 1200′), there was only one replication of each treatment. Material was applied with equipment commonly available to alfalfa growers.

Treatments were all applied at planting time. The EPTC granules were incorporated with a disk and harrow. EPTC-6E was dripped in the irrigation water after seed was sown and worked into the soil.

All rates of granular EPTC gave good weed control. There were damage symptoms and some temporary stunting of the alfalfa at the 3 lb rate. The test demonstrated that weed control is not as effective in water-runs as is the granular application; a better stand was established by bringing the alfalfa up with natural rainfall. Weeds in the trial were groundsel, shepherds purse, fiddleneck, mustard, nettle-leaf goosefoot, and volunteer oats. EPTC gave satisfactory control

EPTC treatments	Weeds (in 4 sq ft)	Alfalfa plants (per sq ft)
Check (not irrigated)	13	33
EPTC-6E at 1.25 lbs per acre; water run	6	32
EPTC-6E at 2.35 lbs per acre; water run	3	13
Granular, 1 lb per acre (not irrigated)	2	58
Granular, 2 lbs per acre (not irrigated)	1	28
Granular, 3 lbs per acre (not irrigated)	2	27
Check (irrigated)	10	20

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