1970

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Recommended Citation

Brown, D.H., Silberborg, S.B. 1970. Effect of cycloheximide on Hypoxylon cankers on Aspen. Plant Disease Reporter 54(6).

EFFECT OF CYCLOHEXIMIDE ON HYPOXYLON CANKERS ON ASPEN

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Abstract

Direct applications of cycloheximide in fuel oil to Hypoxylon cankers on aspen arrested canker activity and enlargement for 5 years. Tree survival increased with increased concentrations of cycloheximide.

INTRODUCTION

The devastating effect of Hypoxylon pruinatum (Klotzsch) Cke. (H. mammatum (Wahl.) Miller) present in 18 million acres of aspen in the Lake States has been described by Anderson (2). Results of studies made in Michigan, Wisconsin, and Minnesota showed that the disease kills 1 to 2% of the standing aspen volume annually, which is equivalent to 31% of the net growth. An economical means of reducing these losses would be welcomed by public and private forest land managers, especially in regions having large acreages of aspen.

A small-scale chemical test was initiated in 1958 to determine the effectiveness of various concentrations of the antibiotic cycloheximide (Acti-dione BR)[2] in the control of Hypoxylon canker of aspen. The results of these tests are reported here. A brief report was made previously (8).

MATERIALS AND METHODS

A mixed even-aged stand of quaking aspen (Populus tremuloides) and bigtooth aspen (P. grandidentata) near La Fayette, New York was selected as the test area. Trees with active cankers on the bole, less than three-quarters around the circumference and 5 feet or less above the ground, were selected for treatment (Fig. 1). The trees were assigned to seven treatments consisting of: 1) no. 2 eastern fuel oil with incision and perforation, 2) cycloheximide (Acti-dione BR); 50 ppm in fuel oil with incision and perforation, 3) cycloheximide, 100 ppm in fuel oil with incision and perforation, 4) cycloheximide, 150 ppm in fuel oil with incision and perforation, 5) cycloheximide, 200 ppm in fuel oil with incision and perforation, 6) cycloheximide, 150 ppm in fuel oil with excision, and 7) cycloheximide, 200 ppm in fuel oil with excision.

In treatments 1 through 5, cankers in equal numbers were either incised with a knife or perforated with a special tool as part of a pre-spray treatment. In the first type of pre-spray treatment, vertical incisions into the xylem were spaced at 1- to 1 1/2-inch intervals across the face of the canker. The incisions extended approximately 1 inch beyond the vertical and lateral limits of bark discoloration. In the second type of pre-spray treatment, the entire canker, including about a 1-inch border beyond the edge of discoloration, was perforated with a special tool. The perforation tool, consisting of a wood block with numerous, evenly-spaced nails protruding from one side, was placed with the nail points against the bark and then tapped until the points penetrated the wood. The entire edge of discoloration in the bark was marked with a

FIGURE 1. Untreated Hypoxylon canker on aspen.

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[2]The material was supplied by the Upjohn Company, Kalamazoo, Michigan. Use of trade name does not imply endorsement by USDA, Forest Service.

wax pencil as a reference point for future measurements. A liberal amount of freshly mixed cycloheximide solution[3] was brushed on the incised and perforated cankers. Fuel oil controls were treated in a similar manner.

In treatments 6 and 7, a third type of pre-spray treatment involved the excision or complete removal of diseased bark. Bark was removed until internal mottling or discoloration was no longer visible in the cut edges along the perimeter of the excised area. A generous amount of fuel oil with cycloheximide was immediately applied to the exposed wood and surrounding bark.

All treatments were made in June 1958 and examined in September 1958 and March 1963. Canker growth measurements were made only once, in September 1958.

RESULTS AND DISCUSSION

The test trees were examined in September 1958, approximately 3 months after treatment. Canker growth or extension was measured from the wax pencil outline to the vertical and horizontal limits of visible bark discoloration. The average upward, downward and lateral growth for 18 cankers in treatments 2 through 5 was 4.1, 3.3, and 1.3 inches, respectively. Very little variation in growth rates existed among these treatments. The one canker in treatment 1 that could be measured had upward, downward, and lateral extensions of 5.5, 4.0, and 1.5 inches, respectively. The growth data for all cankers is comparable to that reported for artificially-induced Hypoxylon cankers (5, 11). Phytotoxicity was not evident in any of the treatments.

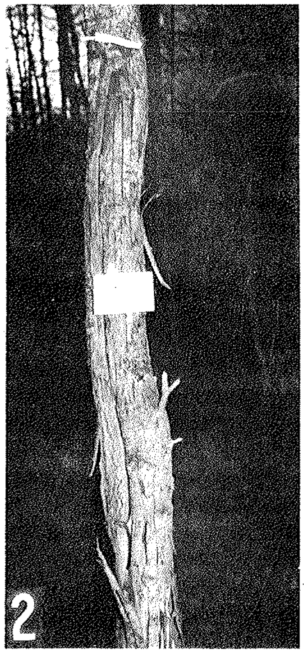
In the spring of 1963, five years after treatment, a significant change in canker activity was apparent. At least one canker in each treatment, with the exception of the fuel oil controls, had some callus (Table 1). Interference with canker activity increased with increased concentrations of the antibiotic. Each treatment, with two exceptions, had one canker that was ar-

Table 1. Effect of direct application with cycloheximide on Hypoxylon cankers on aspen 5 years after treatment.

Fuel oil treatment	Treated cankers (no.)	Cankers with callus (no.)	Arrested cankers with callus[a] (no.)	Dead trees no.	Dead trees %
1) Controls with incision and perforation	4	0	0	4	100
2) 50 ppm with incision and perforation	6	1	1	5	83
3) 100 ppm with incision and perforation	6	2	0	4	67
4) 150 ppm with incision and perforation	6	3	2	4	67
5) 200 ppm with incision and perforation	6	4	1	3	50
6) 150 ppm with excision	2	1	1	1	50
7) 200 ppm with excision	2	1	1	1	50
Total	32	12	6	22	

[a]Arrested cankers did not display any symptoms or signs of the disease and were completely contained by callous tissue.

[3]All solutions were checked for possible crystallization of the antibiotic on the bottom of the containers. No crystallization was found after the solutions had been stored at room temperature for several days.



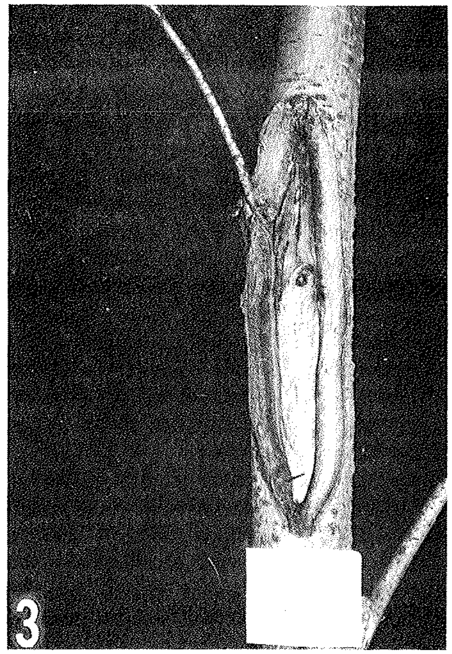


FIGURE 2. Hypoxylon canker treated with cycloheximide, 200 ppm in fuel oil following incision. Callus is evident around perimeter 5 years after treatment.

FIGURE 3. Hypoxylon canker treated with cycloheximide, 200 ppm in fuel oil following excision. Thick callus is present 5 years after treatment.

rested. All arrested cankers showed a steady, total containment of the disease with annual increases in callous tissue and no evidence of disease breakover in or beyond the callus. Callus associated with arrested cankers is different from that described for untreated cankers on native aspens (P. tremuloides and P. grandidentata and white poplar (P. alba)). In the latter situation the fungus frequently grows beyond the callous tissue, sometimes repeatedly, with the result that ridges or zones of callus are formed (4, 5). The callus on trees successfully treated with cycloheximide completely surrounded the perimeter of the treated area and appeared as a single enlarging ridge (Figs. 2, 3). A cross section of a treated canker indicated callous tissue began forming in 1959 and was closing over the wound. A study in Alberta (3) revealed strong callus formation on untreated cankers but no recovery of the tree from the disease.

The effect of treatment was apparent in the fate of trees as well as in canker activity. Trees treated with cycloheximide survived longer than the controls and survival increased with increasing concentrations of the antibiotic (Table 1). Untreated trees possessing natural or artificially-induced infections generally die within 2 to 5 years, depending on the age of the tree and the location of the cankers (1, 5, 11).

Treatments with cycloheximide that failed to inhibit Hypoxylon cankers on aspen in Colorado (9) differed from the treatments reported here in two respects. First, the basal 5-foot portion of the bole on each infected tree was sprayed. The sprayed area may not have contained a canker. Secondly, canker incisions of any type were not part of the treatment technique. The importance of canker incisions in suppressing blister rust cankers on eastern white pine has been demonstrated (10). Bark incisions apparently facilitate the penetration of the spray solution and in turn provide a more direct contact between the spray and the fungus.

Cycloheximide has been tested in culture against H. pruinatum. One test showed that cycloheximide at 100 ppm inhibited the mycelial growth of the fungus (6). A more extensive series of tests indicated that the antibiotic at 22 ppm completely inhibited fungal growth in culture (7).

Practical and economical means of direct control of Hypoxylon cankers on aspen is not known. Even though the promising results reported here are too limited to be conclusive, they do suggest that further testing is warranted. A successful chemical treatment would have potential application where cankers occur close to the ground (3) or on high value trees.

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