TITLE: Potential for Physiological Management of Symptom Expression by Turfgrasses Infected by Bipolaris sorokiniana

INVESTIGATORS:

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1991 Funding: $20,000

CLIMATIC REGION: Cool Humid
USGA REGION: Great Lakes

IOWA STATE UNIVERSITY

Potential for Physiological Management of Symptom Expression by Turfgrasses Infected by *Bipolaris sorokiniana*

1991 Research Grant: $65,00
(First year of support)

Dr. Clinton F. Hodges
Principal Investigator

Ethylene has been found to contribute substantially to the loss of chlorophyll in leaves of *Poa pratensis* infected by *Bipolaris sorokiniana*. The physiology of the elevated endogenous ethylene levels is unknown, but the increase is known to cause some of the yellowing often associated *B. sorokiniana* leaf spot. This research project was initiated to determine if the endogenous ethylene, or its mode action, could be manipulated to prevent the loss of chlorophyll in infected leaves and prevent yellowing. The ultimate objective is to develop a means of controlling disease symptom expression. Infection would not be prevented, but yellowing would not occur and normal mowing procedures would remove infected leaves. The use of fungicides for diseases of this type could be substantially reduced or eliminated.

Initial studies with aminooxyacetic acid (AOA) have provided some positive results. AOA blocks the enzymatic conversion of S-adenosyl-L-methionine (SAM) to 1-aminocyclopropane-1-carboxylic acid (ACC) in the biosynthetic pathway of ethylene in higher plants. This action ultimately reduces the total ethylene produced. Leaves of inoculated plants showed elevated levels of endogenous ethylene at 24, 48, 72, and 96 hr after inoculation; peak endogenous ethylene production occurred at 48 hr (1000 μl l^{-1}). Plants inoculated and treated with AOA (10^{-3}M) produced less endogenous ethylene at all sampling times than untreated inoculated plants. At 48 hr, the leaves of inoculated plants treated with AOA produced 642 μl l^{-1} of endogenous ethylene compared to 1000 μl l^{-1} produced by untreated inoculated plants.

Chlorophyll loss was initiated after peak endogenous ethylene production (48 hr) and became progressively more severe with time. Chlorophyll content of leaves of untreated inoculated plants was 57% of that in healthy control leaves at 96 hr after inoculation; chlorophyll content of leaves of AOA treated and inoculated plants was 81%. The decrease in endogenous ethylene in infected leaves of AOA treated plants clearly decreased the chlorophyll loss associated with the disease. No phytotoxic effects were observed to be associated with AOA.

These initial observations are encouraging and suggest that manipulation of symptom expression (yellowing) by leaves infected with *B. sorokiniana* may be feasible. Several additional substances that interfere with ethylene biosynthesis, or with the mode of action of ethylene, will be examined during the remainder of this year and into next year.

A. PREFACE

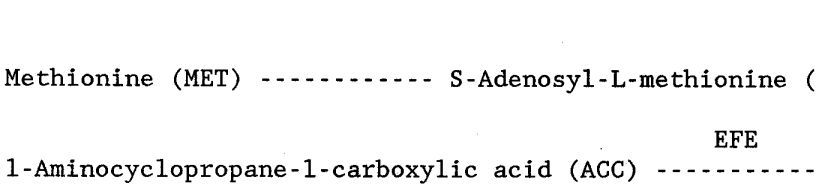
Please note that the reporting period for this year is from June 1, 1991, through October 15, 1991. Funding for the project was not received until May of 1991, and research was initiated in June. Therefore, the report covers slightly less than four months.

B. OBJECTIVES

The primary objective of this project for 1991 was to evaluate substances known to prevent the biosynthesis and/or mode of action of the endogenous ethylene generated during the infection of Poa pratensis by Bipolaris sorokiniana. The endogenous ethylene generated is a major physiological factor in the yellowing of infected leaf tissue. Control of this process could feasibly reduce or eliminate the use of fungicides for the control of this disease (and possibly others).

C. OBSERVATIONS

The primary source of endogenous ethylene during infection is from the biosynthetic pathway in the host plant. Some ethylene also is produced by the pathogen from a second pathway, but the amount is believed to be negligible in most cases. Therefore, prevention of biosynthesis and/or mode of action of endogenous ethylene produced by the host was the target of the 1991 studies. The higher plant biosynthetic pathway for ethylene is as follows:



Studies conducted in the first 4 months of this year have concentrated on the use of aminooxyacetic acid (AOA) to prevent the conversion of SAM to ACC and thereby decrease the ethylene generated during infection. Ten milliliters of AOA (10^{-3}M) were applied to the soil in pots containing P. pratensis each of 3 days preceding inoculation, and then each of 4 days during pathogenesis which included the assay days for endogenous ethylene evolution by the infected leaves.

Inoculated plants were assayed for ethylene generation at 24, 48, 72, and 96 hours after inoculation. Ethylene elevated sharply in response to infection at 24 and 48 hr after inoculation (Fig. 1). The ethylene then progressively declined at 72 and 96 hr. Chlorosis of infected tissue started after the ethylene peak at 48 hr. AOA did not prevent the rise in ethylene associated with infection (Fig. 1), but it did keep the level below that of untreated plants. It was especially effective at preventing the 48 hr peak associated with untreated inoculated plants

(Fig. 1). Endogenous ethylene in untreated inoculated plants peaked at 48 hr at 1000 $\mu l\ l^{-1}$ compared to 642 $\mu l\ l^{-1}$ in inoculated plants treated with AOA (Fig. 1). Levels of ethylene in untreated and AOA treated control plants were very similar and no signs of phytotoxicity were observed in response to the AOA. Please note that the data collected has not been analyzed statistically to date.

To determine the yellowing effect, or loss of chlorophyll, chloroplasts were extracted from leaf tissue from plants assayed for endogenous ethylene at 96 hr after inoculation. Leaves of inoculated plants contained 57% of the chlorophyll in noninoculated control leaves (Fig. 2). Inoculated plants treated with AOA contained 81% of the chlorophyll in noninoculated control leaves. AOA did not have any negative effect on the chlorophyll content of treated control plants.

These preliminary results are encouraging. We have had time to test only one substance (AOA) of several that attack the biosynthesis of endogenous ethylene, and chlorophyll loss was substantially reduced by the AOA. Other substances under study for the remainder of this year and into next year will attack ethylene biosynthesis at several stages; some also will interfere with the mode of action of ethylene. AOA may not be the most efficient ethylene inhibitor we will find, but in future studies its action combined with hormonal substances that retard senescence may prove very effective in preventing the chlorosis associated with leaf spot.

Principal Investigator

10/24/91
Date

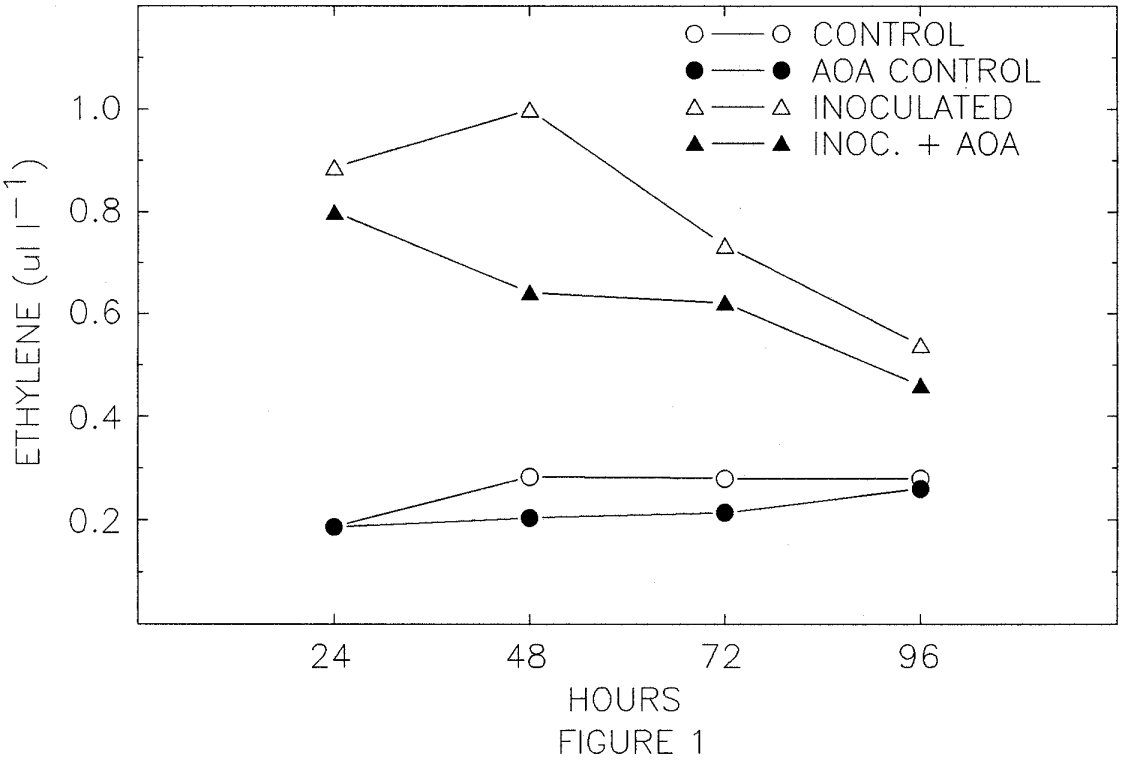


FIGURE 1

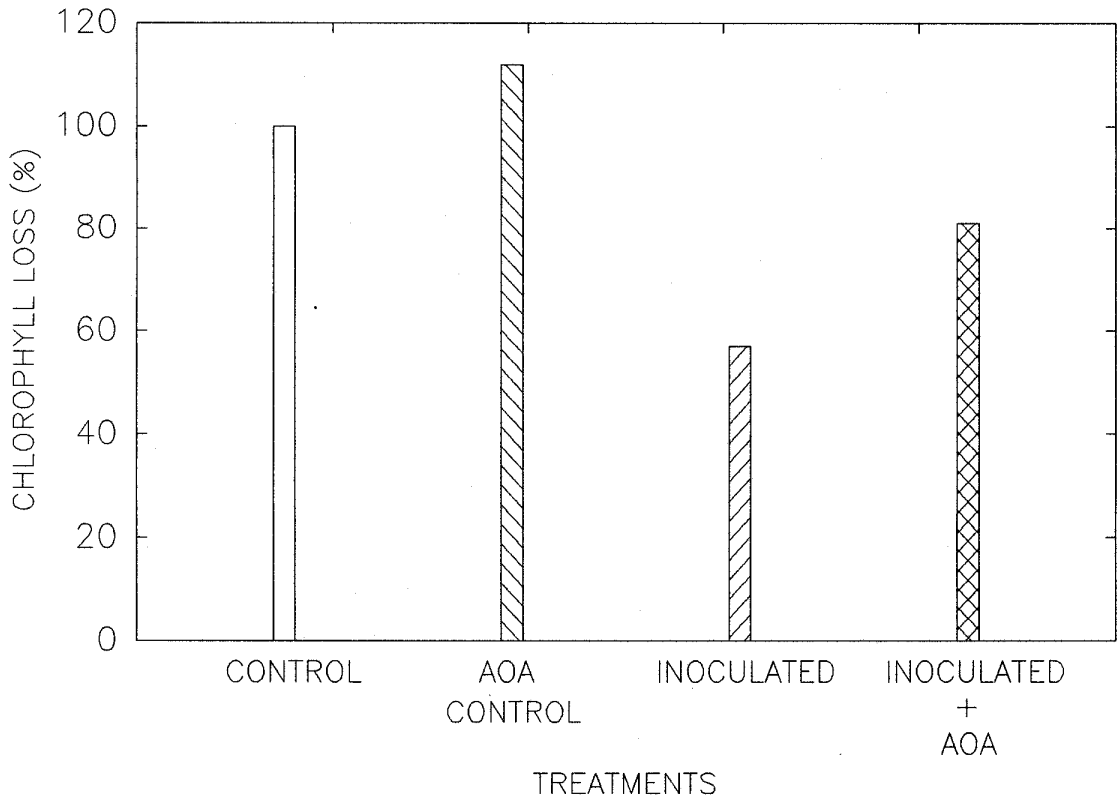

120
100
80
60
40
20
0
CHLOROPHYLL LOSS (%)
CONTROL
AOA CONTROL
INOCULATED
INOCULATED + AOA
TREATMENTS

FIGURE 2