EFFECTS OF SULFUR DIOXIDE AND NITROGEN DIOXIDE ON VEGETATION

Six Month Progress Report for Contract No. EE-77-S-02-4368

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MASTER

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II. Progress (Six-month period beginning April 1 and ending September 30, 1979)

Effects of NO_2 on the Phytotoxicity of SO_2

During the spring, work on the measurement of SO_2 deposition velocities in short-term exposures was continued. The system used for these investigations is being expanded so that the rates of transpiration and CO_2 exchange during exposure to an air pollutant or combination of pollutants can also be continuously monitored.

But major emphasis of our work shifted to the preparation of new field facilities for the 1979 growing season. Twelve new open-top chambers were constructed and electrical power was installed at the field site.

The results of the 1978 field experiments indicated that for one of the two cultivars tested (Hark) all treatments of SO_2 with or without NO_2 reduced yield (bean dry weight). Therefore, in order to determine the lower limit for yield reduction in Hark soybean we chose the following treatments (which are 50% lower than the concentrations used in 1978): 0.1 or 0.3 ppm SO_2 with or without 0.1 ppm NO_2 , 0.1 ppm NO_2 alone, charcoal filtered air (chamber control), and ambient plots (no chambers).

Soybeans were planted on the same date as in 1978, June 5. Since the quality of the ambient air in Ithaca is better than in Yonkers, the open-top chambers were not placed over the plants until August 7. In 1978, the exclusion and charcoal filtration of ambient air in the open-top chambers was begun on July 19.

The exposures were made during the pod-filling stage from August 16 until September 13. During this period, the soybeans were exposed on 18 days from approximately 1030 to 1430 hours.

In all, there were seven different treatments in duplicate, giving a total of fourteen separate soybean plots. The plots were planted so that each soybean plant had six nearest neighbors 10 inches away as was used in the 1978 field experiments.

In mid-September, the plots were ranked according to the degree of senescence, i.e., the amount of yellowing of the older foliage. In order of decreasing senescence they were (1) high SO₂ + NO₂; (2) low SO₂ + NO₂; (3) high SO₂ alone, NO₂ alone, low SO₂ alone (all equal); (4) charcoal filtered air (chamber control); and (5) ambient (no chamber).

The plants were harvested on October 1. The pods were taken from each of 37 plants per plot and the following variables are being determined: number of pods, number of seeds, and dry weight of seeds.

Interactions of Air Pollutants, Plants, and Plant Pathogens

During the past six months, two studies have been initiated in the general area of SO₂-plant-plant pathogen interactions. One of these, the effects of SO₂ on Scleroderris canker of red pine, is a two-year field study and results are not yet available; however, a brief summary of what has been done follows. In late June, one half of each of 16 plots of red pine were inoculated with Gremmeniella abietina (syn. Scleroderris lagerbergii). In July and August, some of these plots were exposed to several sub-acute doses of SO₂ by means of a linear gradient exposure apparatus. Next summer, symptoms will be evaluated and measurements of the effects of SO₂ on the disease and host will be made. Additional plots will be exposed to SO₂ and some plots exposed to SO₂ this summer will be re-exposed. This design should provide information on effects of SO₂.

on infection and disease development.

Studies were also conducted to evaluate the effects of SO₂ on bacterial pustule of soybean (caused by Xanthomonas phaseoli var. sojensis). These studies utilized greenhouse-grown seedlings and were conducted in growth chambers. The experimental design involved exposing plants to 0.1 ppm SO₂ before, after, or both before and after inoculation with the pathogen. The results indicate that SO₂ exposure significantly inhibited infection and lesion development by the pathogen over time. We also found that in several cases, the effects of SO₂ exposure on lesion development were non-additive, i.e., the effects of exposure both before and after inoculation were greater than the sum of the effects of two individual exposures. Rates of lesion development, as measured by the slope of a regression of lesion size on time after inoculation, were consistently different, although statistical significance could not be demonstrated.

There were no statistically significant interactions of SO₂ and the pathogen on the plant as measured by sulfur uptake, leaf area, dry mass, or dry mass to leaf area ratio. There were significant reductions in leaf area and dry mass due to SO₂ alone. Additional investigations are planned to determine the effects of exposure regimes characteristic of point source fumigations on disease development.

Effects of SO₂ on Plant-Insect Interactions

Investigations of the effects of SO₂ on the use of pinto beans by the Mexican bean beetle were completed during this period. Adult females showed a slight but statistically significant preference for feeding on fumigated foliage. Although the pre-ovi-

position period of females fed fumigated tissue did not differ from that of the control females, females on fumigated foliage required a slightly longer time to mature each batch of eggs (1.7 vs. 1.4 days); this was offset by an increase in the number of eggs laid/female/day over a ten day period did not differ significantly (37.0 vs. 37.9 for experimental and control, respectively).

No differences could be detected in the rate of development or mortality of the immature stages. Although the weights of first and second instar larvae fed fumigated foliage were slightly greater than those of the control larvae, this difference was lost by the last instar and the growth curves were virtually identical. These results are presently being prepared for publication.

A reproducing colony of Mexican bean beetles has been established on soybeans and tests using the Mexican bean beetle/soybean system were started in late September. Also, most of the techniques needed to rear the imported cabbageworm continuously and to study the effects of SO₂ on the use of cabbage by this insect have been developed during this period.

Fumigation of balsam fir trees with SO₂ appears to result in some changes in the volatiles emanating from their needles. When trees were treated with 2 ppm SO₂ for 5 hours on 3 consecutive days, the total volatiles collected during two 4-hour periods after fumigation were considerably greater than from control trees. In addition, the quantity of β -pinene was disproportionately higher from fumigated trees. β -Pinene has previously been shown to be attractive to ovipositing spruce budworms. Bioassays of oviposition preference between fumigated and control trees were inconclusive due to a high degree of variability and bioassay procedures will

be refined to allow for detection of smaller differences.

Mode of Action and Metabolism of SO₂ in Plants

Exposure of many plants to low levels of SO₂ results in a decreased rate of photosynthesis as measured by net CO₂ uptake, and by CO₂ fixation. Removal of the plant from the SO₂ atmosphere results in a restoration of rates to previous levels. An increasingly large body of evidence indicates that the rate at which the carboxylase enzyme itself operates in vivo is in turn controlled by several factors. These are other enzymatic reactions, pH and light. Data obtained by Dr. Amundson in our group over the past year have demonstrated a marked difference in sensitivity to SO₂ between two soybean cultivars, Hark and Beeson, as measured by differences in yield of beans in field experiments. Whether differences in the response to SO₂ of the factors which control carbon fixation can be correlated with the field observations of Dr. Amundson will be determined.

Two enzymes in particular are thought to be pivotal in the control of the rate of CO₂ fixation in the chloroplast. These are fructose 1,6 biphosphatase and sedoheptulose 1,7 biphosphatase. These enzymes have been shown to be light-activated via a mechanism involving photosynthetic electron transport. Since SO₂ (in solution as sulfite) is known to be detoxified and converted to sulfate at that same site, i.e., at the green membranes of the chloroplast, in a series of reactions which are linked to electron transport, it became of interest to determine if SO₂ exerts its inhibitory effect on photosynthesis through an interference with this activation process. Soybean plants, both Hark and Beeson, have been exposed

to SO_2 (0.5 - 1 ppm) after being in continuous darkness for about 13 hours in the hopes that the chloroplast phosphatases would be completely inactive at the onset of fumigation and illumination. Chloroplasts were isolated from the leaves of soybeans after 1.5 hours of fumigation with SO_2 in the light, they were then lysed, and the stromal extracts obtained were assayed for fructose 1,6 biphosphatase activity. Enzyme activity in Hark was lower in the chloroplasts which had been isolated from fumigated leaves than in the corresponding controls, whereas the presence of SO_2 during illumination of Beeson resulted in elevated levels of enzyme activity. There are several reports in the literature of stimulation of photosynthesis by low levels of sulfite and perhaps the differing sensitivities of these two cultivars to SO_2 is reflected in the apparent stimulation of this key regulatory enzyme of carbon fixation in the SO_2 -resistant cultivar.

In addition, preliminary data indicate that transitory changes in pH alone can trigger increases in activity of chloroplast fructose 1,6 biphosphatase. It is possible that SO_2 in solution within the chloroplast acts as an uncoupler, preventing the alkalization of the stroma which normally occurs as electron transport is initiated. The chloroplast sugar phosphatases have an alkaline pH optimum which is the same as the one attained by the stroma on illumination. The presence of sulfite in the chloroplast may have the consequence that this transition does not take place and consequently the regulatory phosphatases remain inactive, thus effectively stopping the flow of intermediates through the reductive pentose phosphate cycle. Further experiments are planned in which the possible role of sulfite as an uncoupler will be examined.

III. Status

Effect of NO₂ on the Phytotoxicity of SO₂

We are on schedule with regard to the effect of NO₂ on the phytotoxicity of SO₂. The 1979 field season was just completed. Over the next several months, the yield data will be analyzed. In addition, laboratory studies will continue to determine the uptake rates of both SO₂ and NO₂ alone and in combination.

Interactions of Air Pollutants, Plants and Plant Pathogens

At the present time, we are on schedule with regard to planned studies of pollutant-pathogen interactions. During the next six months, work will proceed on studies of the effects of SO₂ on Scleroderris canker of red pine. In addition, experiments are planned to evaluate the effects of single and repeated short term peak exposures on bacterial pustule of soybean.

Effects of SO₂ on Plant-Insect Interactions

Studies of SO₂ effects on interactions between the spruce budworm and conifers are back on schedule while those involving the Mexican bean beetle and its hosts are very nearly on schedule. In the next period, assays will be conducted to determine the effects of the SO₂-induced changes in the release of host volatiles on oviposition by the spruce budworm. Studies on the Mexican bean beetle/soybean system (development times, mortality, growth curves, fecundity, and adult feeding preference) will be completed soon and work on characterizing factors responsible for changes in plant use will be initiated. If no such changes are found,

the groundwork has been completed to examine a crop plant/insect pest system.

Mode of Action and Metabolism of SO₂ in Plants

A possible effect of SO₂ has been detected on fructose-1,6, bis phosphatase, one of the regulatory enzymes of photosynthesis. Two soybean cultivars, Hark and Beeson, which have been established to differ with regard to their respective sensitivities to SO₂ appear also to differ with respect to the light-mediated activation of this regulatory enzyme. The direct role of pH in the activation of fructose 1,6,bisphosphatase was also examined.

These investigations will be expanded during the six month period, October 1979 - March 1980, to include determinations of effects of SO₂ on other regulatory enzymes of photosynthesis in soybean.

IV. Completed Outputs

Amundson, R. A. and L. H. Weinstein. Reduction of yield to two cultivars of soybean by exposure to SO₂ or SO₂ and NO₂ during the pod filling stage. In preparation.

Amundson, R. A. and L. H. Weinstein. Effects of short-term exposures of SO₂ and NO₂ alone or in combination of leaf diffusive resistance and leaf injury in several cultivars of soybean. In preparation.

Laurence, J. A. 1978. Effects of air pollutants on plant pathogen interactions. Paper presented at the annual meeting of the Air Pollution Control Association and the Third International Congress of Plant Pathology. Submitted to J. Air Poll. Contrl. Assoc.

Laurence, J. A. 1979. Response of maize and wheat to sulfur dioxide. Plant Dis. Reporter 63(6): 468-471. 1979.

Laurence, J. A., L. H. Weinstein, D. C. McCune, and A. L. Aluisio. 1979. Effects of SO₂ on southern corn leaf blight of maize and stem rust of wheat. Accepted for publication. Plant Disease Reporter.

Laurence, J. A., L. H. Weinstein, A. L. Aluisio, and D. C. McCune. 1979. Effects of SO₂ on three plant-virus combinations. Paper presented at the Fourth International Congress of Plant Protection.

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Dated October 19, 1979

Boyce Thompson Institute
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