

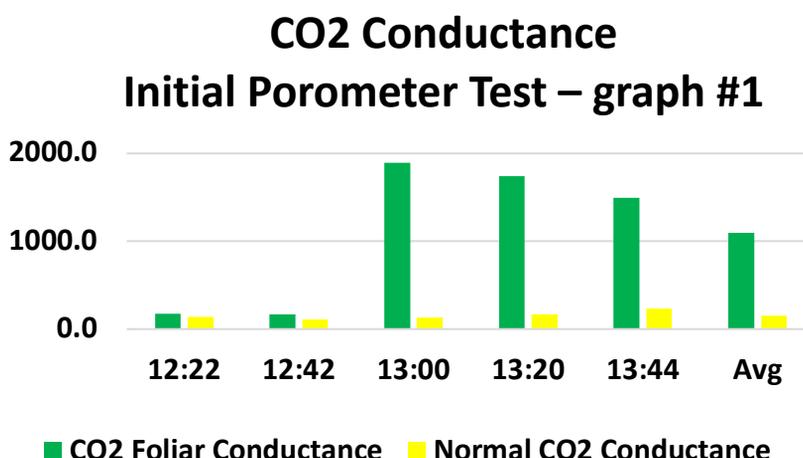
# Research Report #2

## St. Cloud State University

St. Cloud State University is conducting a series of four experiments to document the impacts of CO<sub>2</sub> delivered in supersaturated water to stomata via foliar misting. The first of these experiments has been initiated with seeding plants being prepared for foliar spray exposure or other control/ null treatments. Ongoing data from this experiment will be reported in the next reports.

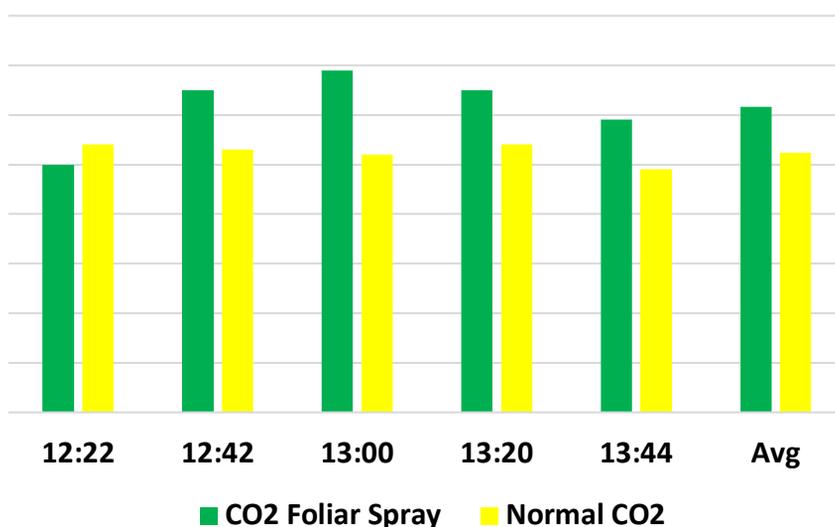
In our previous report (report #1), data for instantaneous physiological responses in plants treated with CO<sub>2</sub> enriched foliar spray were reported. This data largely focused on chlorophyll A changes in the plant in response to exposure. Instrumentation was acquired to provide additional data to clarify if the chlorophyll A responses were related to CO<sub>2</sub> enrichment.

Further instrumentation was acquired for these experiments, specifically, a SC-1 Leaf Porometer (ICT International) was acquired to measure conductance. Conductance is an estimate of the rate of CO<sub>2</sub> entering the leaf and/or water vapor exiting the plant leaf stomata.



This conductance metric is likely most directly related to CO<sub>2</sub> GRO Inc.'s technological approach to enhancing plant growth, which is to increase CO<sub>2</sub> availability to plants via super saturated water deposited on top of the leaf or under the leaf near the stomata. The exact approach is described further in the report. The average conductance increased by greater than eight fold.

### Chlorophyll A Measurements Initial Porometer Test - graph #2



The chlorophyll A content, as seen in the previous graph consistently shows a 15-20% increase in plants exposed to CO<sub>2</sub> enriched foliar compared to plants receiving no foliar spray; measured as a function of area.

Three experiments were run with the porometer. In all experiments chlorophyll A concentration was measured (Apogee MC-100) with CO<sub>2</sub> conductance (ICT International SC-1). In the first, both metrics were quantified every 20 minutes for 100 minutes. Two treatments were considered: 1) CO<sub>2</sub> enriched foliar spray and 2) no spray. Data for each metric was compared between treatments using a tTest for equal means. Both chlorophyll A (p=0.0077) and CO<sub>2</sub> conductance (p=0.0131) showed significant increases in the CO<sub>2</sub> exposed treatments, graph #1 and graph #2, respectively.

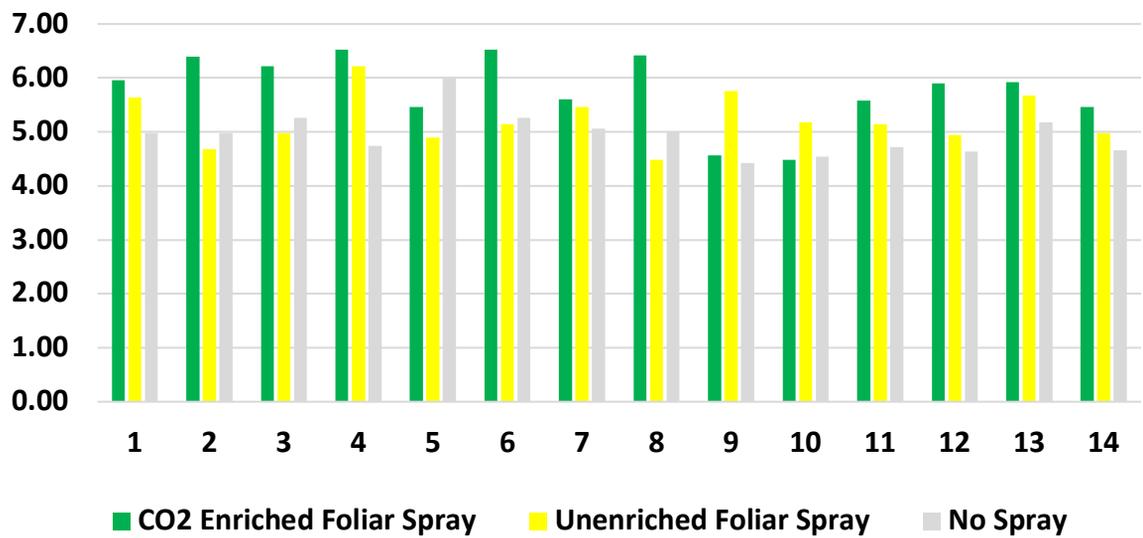
The second and third experiment were identical in treatments and metrics quantified. They only differed in duration of the experiment. This is a result of the time it takes to acquire a stomatal conductance estimate in comparison to chlorophyll A. In the second experiment the duration was 2 hours and 20 minutes and the third lasted 4 hours. Treatments for these experiments included: 1) CO<sub>2</sub> enriched foliar spray, 2) unenriched foliar spray, and 3) no spray. The unenriched foliar spray treatment was added to potentially reject the hypothesis that water vapor alone could explain the results from prior experiments. Results are seen in graphs #3 and #4.

In both experiments chlorophyll A was measured for 5 randomly selected leaves every 10 minutes immediately following treatments which were also applied every 10 minutes. Stomatal conductance was measured each hour for each treatment. Both experiments were consistent in showing higher chlorophyll A content and higher stomatal conductance in CO<sub>2</sub> exposed treatments.

ANOVA was used to compare chlorophyll A data in both experiments and stomatal conductance in the third experiment (only two estimates existed for experiment 2 making statistical comparison impossible). Significant differences existed between CO<sub>2</sub> exposed treatments for chlorophyll A ( $p=0.00057$ , exp2 and  $p=0.0000005.5$ , exp3) and stomatal conductance ( $p=0.00000074$ ). Notable is that NO significant difference existed between unenriched spray and no spray treatments, strongly suggesting that CO<sub>2</sub> availability was the factor increasing both chlorophyll A and stomatal conductance.

Important is that after 5 rounds of instantaneous experimentation, chlorophyll A content consistently shows a 10-20% increase in plants exposed to CO<sub>2</sub> enriched foliar compared to plants receiving no foliar spray or non-augmented foliar spray. Measurements were based on area per m<sup>2</sup> measurements.

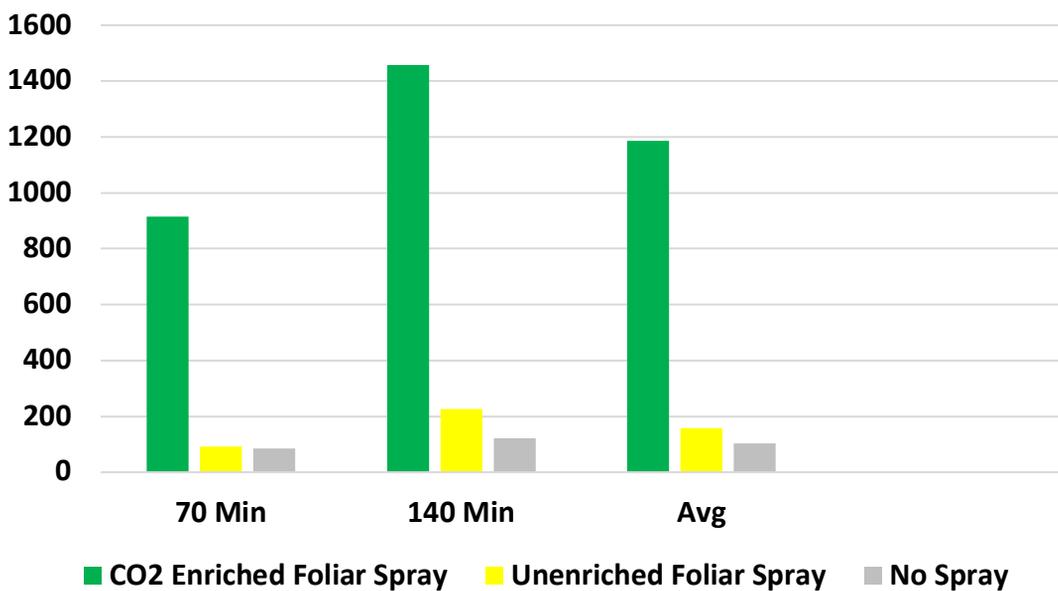
### Chlorophyll A – graph #3



In this experiment, treatments were applied every 10 minutes for 2 hours and 20 minutes .

Chlorophyll A measurements were taken from five random leaves from plants under each treatment every 10 minutes.

### CO2 Conductance Data On Leaf Surface graph #4



In this experiment, treatments were applied every 10 minutes for 2 hours and 20 minutes .

CO2 conductance measurements were taken from five random leaves at 70 and 140 minutes.

Further, 3 rounds of experimentation show a consistent increase in stomatal conductance of greater than 8 times in plants exposed to CO<sub>2</sub> enriched foliar spray compared to plants receiving no foliar spray or non-augmented foliar spray. This data continues to be encouraging and consistent with the hypothesis of significant growth enhancement with CO<sub>2</sub> delivery via foliar spray.