Update of Activities at the Georgia Envirotрон
The University of Georgia, Griffin Campus

ENVIROTÓN EQUIPMENT UPGRADES

CONVIRON GROWTH CHAMBERS

Dehumidification systems were installed in six CG72 growth chambers. Dehumidifiers are CargoCaire 150 units comprising a SiGel dessicant wheel. The dehumidifiers are installed on top of the chambers with inlet/outlet ducting on either side of the CG72 evaporative coils. The units are separately controlled with a wall-mounted humidistat in the CG-72 machine room. The humidification option in the CG-72 chambers has been deactivated to prevent conflicting control between the dehumidification and humidification units.

A separate temperature monitoring system has been installed in the nine growth chambers. Each chamber has been equipped with a type T thermocouple sensor, and all thermocouples are connected to a single Campbell Scientific CR21-X datalogger equipped with a reference temperature sensor. The datalogger is connected to the Envirotрон LAN through an Campbell Scientific MD9 coaxial interface which in turn is connected to an NL-100 network link interface.

GREENHOUSES

Six of the eight greenhouses have been retrofitted with Wadsworth Step50 greenhouse controllers, along with redesigns of the actuation of the pad louvers and retrofitting of Wadsworth rack and pinion arms to open and close the roof vents.

GROWTH CHAMBER RESEARCH

Morphological Characterizations of Clover for Determination of Genetic Redundancy

Brad Morris, Plant Genetics Resources Conservation Unit.

Genetic redundancy is of prime concern for curation of crop species. The USDA, ARS, PGRCU *Trifolium subterraneum* collection is suspect of having genetic redundant accessions. Our goal was to determine whether or not genetic redundant accessions do in fact exist within the U.S. subterraneum clover collection. Subclover seed were planted in potting soil within each of five 4” plastic pots. A sub-sample of 90 subclover accessions were tested. Soon after seed germination, each pot utilized in the test were moved to growth chambers at the Georgia Envirotрон. Plants were grown in a 16 hour photoperiod regime with a 27 °C / 17 °C day/night temperature setting. Successful morphological characterizations were recorded for leaf marking, flower color, and stipule color.

Combined Effects of Elevated Carbon Dioxide Levels and Temperature on the Biology of the Mealybug *Phenacoccus madeirensis* Green (Homoptera: Pseudococcidae)

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The combined effects of elevated CO₂ levels (400 and 700 µL/L) and temperatures (20, 25 and 30 °C) on the development, survival and reproduction of two generations of the mealybug *Phenacoccus madeirensis* were investigated. Mealybugs were reared on chrysanthemums grown in growth chambers set at a specific CO₂ level and temperature. The duration to egg hatching and to adulthood of the mealybugs was recorded by examining the mealybug cohorts daily. Hatching rates of eggs and survival rate to adulthood were determined by recording the number of individuals that successfully molted into the next developmental stage. The proportion of females in the population was determined by fractioning the number of females over the total number of adults at the end of the experiment. Adult females were isolated in leaf cages and their eggs were collected daily to determine fecundity. The nutritional status (carbon concentration, nitrogen concentration, and the relative water content of leaves) of chrysanthemum were also studied to interpret the performance of mealybugs at elevated CO₂ level and temperature. The development of mealybug is temperature-dependent. Duration of development did not differ among different CO₂ level treatments and generations. A female completed its development in about 20 days at 30 °C, 28 days at 25 °C, and 47 days at 20 °C. Males have longer duration of development than females. Survival rates, proportion of females, fecundity, duration of reproduction, and the parameters of host plant nutritional status did not differ significantly among temperature and CO₂ level treatments and between generations.
Fate of *Escherichia coli* O157:H7 in Manure Compost Applied to Soil and Vegetables

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Animal waste in the form of raw manure or composted manure is routinely applied to the land as a crop fertilizer and/or soil amendment. A potential risk arising from the disposal of animal waste of fecal origin is the spread of enteric pathogens. Many outbreaks or cases of *E. coli* O157:H7 infection have been associated with water or food directly or indirectly contaminated with animal manure. Cross-contamination of produce from manure or improperly composted manure used on the farm can be a source of pathogen contamination during preharvest. Although competition with soil microorganisms and adverse environmental conditions can reduce pathogens, there is little information regarding the ability of *E. coli* O157:H7 to survive in manure-amended soils. In this study, our objective was to determine the fate of *E. coli* O157:H7 in soil and on vegetables in a controlled and contained plant growth chamber environment.

A five-strain mixture of green fluorescent protein (GFP)-expressing *E. coli* O157:H7 was prepared and inoculated at 10⁷ CFU/g into the compost. The inoculated compost was mixed with Tifton clay soil at a ratio of 1:100. Twenty horticultural pots for each of baby carrot and green onion plants were filled with inoculated and fertilized soil (ca. 5000 g). Three healthy transplants of each plant were planted into each pot 100 mm apart from each other, and then irrigated with city tap water. The pots were placed in the Envirotron with control of light, temperature, and CO₂ levels. Special air filters was installed to prevent pathogens from spreading to the environment. Plants were irrigated every other day, and fertilized with soluble fertilizer (Sam's Choice Deep Feeding All purpose Food) every two weeks. Soil samples from around the plant (Soil), plant leaves and stem samples (Plant), and soil samples just under the roots (S/p) in triplicate were analyzed for *E. coli* O157:H7 at approximately weekly intervals for the first four weeks, and every 2 weeks for the rest of plant growth cycle (up to 3 months). Soil moisture content and pH were also determined. Over a period of 64 days in onion, the population of GFP-expressing *E. coli* O157:H7 in soil and soil under roots samples was steadily reduced by 3 log, whereas in plant samples was reduced by 2. With carrot, it took 84 days to achieve a reduction of 2.3 log in soil. Seventy days were needed to get a reduction of 1.7 log in carrot plant.

Image analysis for non-destructive and non-invasive quantification of root growth and soil water content in rhizotrons.

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Studies aiming at quantification of roots growing in soil are often constrained by the lack of suitable methods for continuous, nondestructive measurements. A system is presented in which maize (*Zea mays* L.) seedlings were grown in acrylic containers – rhizotrons – in a soil layer 6-mm thick. These thin-layer soil rhizotrons facilitate homogeneous soil preparation and nondestructive observation of root growth. Rhizotrons with plants were placed in an Envirotron CG72 growth chamber, on a rack slanted to a 45° angle to promote growth of roots along the transparent acrylic sheet. At 2- to 3-day intervals, rhizotrons were placed on a flatbed scanner to collect digital images from which root length and root diameters were measured using RMS software. Images taken during the course of the experiment were also analyzed with QUACOS software that measures average pixel color values. Color readings obtained were converted to soil water content using images of reference soils of known soil water contents.

To verify that roots observed at the surface of the rhizotrons were representative of the total root system in the rhizotrons, they were compared with destructive samples of roots that were carefully washed from soil and analyzed for total root length and root diameter. A significant positive relation was found between visible and washed out roots. However, the influence of soil water content and soil bulk density was reflected on seminal roots rather than first order laterals that are responsible for more than 80% of the total root length. Changes in soil water content during plant growth could be quantified in the range of 0.04 to 0.26 cm³ cm⁻³ if image areas of 500 x 500 pixel were analyzed and averaged. With spatial resolution of 12 x 12 pixel, however, soil water contents could only be discriminated below 0.09 cm³ cm⁻³ due to the spatial variation of color readings.

Results show that this thin-layer soil rhizotron system allows researchers to observe and quantify simultaneously the time courses of seedling root development and soil water content without disturbance to the soil or roots.