#### NCERA-101 Station Report

Station: Alabama Agriculture Experiment Station (Auburn University) Preparer: Daniel E. Wells, Associate Professor, Department of Horticulture

#### 1. New Facilities and Equipment.

We increased our capacity to run trials in controlled environments by adding three new container farms from AmplifiedAg. Two of the farms will be used for production of leafy greens for on-campus consumption as part of our FoodU program which trains undergraduate students in a wide array of food production technologies, including CEA. A third farm will be used to run propagation trials and to model controlled environments for production of vegetable seedlings and rooted cuttings of ornamental plants. We are also planning to convert another shipping container farm, manufactured by Freight Farms, into a growth chamber to be used for plant trials.

# 2. Unique Plant Responses.

In trials using a split-root system, in which a standard hydroponic nutrient solution, in combination with various rates of added salinity, was provided to one half of a plant's root system and raw (fresh) water was supplied to the other half of the same plant's root system, red kale plants preferentially produced more root biomass on the freshwater side while 'Favorita' tomatoes preferentially produced more root biomass in the nutrient/salinity side. This phenomenon was observed in several trials under various salinity conditions. We suspect the difference can be explained by nutrient demand and differences in root physiology and are currently designing experiments to determine the reason for the differences.

We also observed some surprising results in plant trials in our poultryponics system in which we utilize novel water treatment strategies to convert poultry processing wastewater into a usable nutrient solution for plants. In previous years, we tested full supplementation of the wastewater to bring all macronutrients up to the level of the control hydroponic solution. This enabled growth in poultryponics treatments to keep pace with the hydroponic controls. Leaf tissue analysis suggested that only a few mineral nutrients were deficient in the wastewater including potassium, calcium, and magnesium. For the Year 3 trials, we therefore decided to only supplement these deficient nutrients up to the level found in the hydroponic solution. Shockingly, doing so actually suppressed plant growth below what was achieved on unsupplemented wastewater. This strange result kept occurring over multiple trials and leaf tissue analysis showed that supplementation vastly

improved potassium levels in the leaves (above levels in control) but further suppressed calcium and magnesium uptake. This led us to hypothesize that 1) targeted addition of Ca and Mg led to precipitation due to the unbalanced anions (high sulfate) in the wastewater and 2) the high potassium uptake was competing with divalent cation uptake. We have designed and are now implementing new experiments to test these hypotheses.

# 3. Accomplishments.

# a. Short-term outcomes:

- We continued to operate a poultryponics system and determined that wastewater from poultry processing plants can be successfully used to produce vegetables in a hydroponic-type system with minimal food safety risks. Results could allow for a novel waste management strategy for poultry producers and a novel water source for crop production, building efficiencies into both types of systems.
- We conducted several salinity tolerance trials on both leafy and fruiting vegetables. We utilize a split-root technique that allows for increased salinity tolerance. Yields of leafy vegetables were comparable to controls. For tomatoes, yields were lower as salinity increased, but consumer preference was high for fruits grown in split-root systems. These systems could be implemented to enhance produce quality.
- We developed a predictive model for assessing salt stress tolerance of Kale plants grown in aquaponics system via spatial and spectral predictive regression models. Using these techniques could help growers utilize higher salinity water without reducing yields.
- We determined that root removal from lettuce grown in a controlled environment did not reduced postharvest shelf-life when lettuce was stored properly. These findings will reduce food safety risks associated with consuming lettuce since roots can host common foodborne pathogens.
- We conducted tomato and lettuce trials in identical greenhouses using differing energy management techniques. In a low-energy greenhouse, lettuce yields and quality fluctuated greatly as the environmental conditions inside the greenhouse followed a seasonal pattern.

# b. Outputs:

Research and extension personnel at Auburn Univeristy published several peer-reviewed manuscripts and presented findings at regional, national, and international conferences. Topics of publications included aquaponics, poultryponics, split-root technique, postharvest quality and shelf-life, and food safety in CEA. Extension trainings on food safety in CEA were also conducted.

#### c. Activities:

- The poultryponics team, including PhD student Wellington Arthur, Brendan Higgins, and Daniel Wells, conducted studies on "poultryponics" to understand how biological treatment of the wastewater (using algae versus bacteria alone) impacts nutrient transformation and pathogen dynamics in the production of hydroponic lettuce.
- The aquaponics team, which includes several faculty and students at Auburn University, conducted multiple experiments aimed at optimizing performance and design of aquaponic systems including dietary probiotic additions to improve Tilapia growth and health in a biofloc system, leafy vegetable production using effluent from a biofloc system, split-root technique to improve salinity tolerance in kale and tomato, sensory evaluations of vegetables grown in aquaponics systems, and wastewater management techniques in a decoupled aquaponic system design.
- The GRACE project, led by Brendan Higgins, and including many faculty and students at AU, conducted trials on lettuce and tomato to better understand energy usage and yield potentials in low-tech greenhouses which are commonly-used in the southeast.
- Team members have actively collaborated outside of AU and have established collaborations with private industry partners and an economic development team from the state of Alabama.

# d. Milestones:

- We celebrated the second year of accomplishments of our NIFA SAS project by holding our second annual GRACE symposium at University of Florida on 3/27-3/28.
- Auburn University increased CEA research capacity by adding three new container farms which will allow for experimentation on propagation in controlled environments and the conversion of an existing container farm into a growth chamber to be used for plant growth trials.

# 4. Impact Statements.

• Plant growth trials at Auburn University utilizing a split-root technique have allowed for traditional CEA vegetable crops to be grown in higher salinity than is typical in hydroponics. Results from these trials will be key in integrating vegetable production

with brackish water aquaculture which promises to be more profitable than freshwater aquaculture. Resulting brackish water aquaponic systems will improve economics and provide a sustainable wastewater management strategy for brackish water aquaculture systems.

- The development of a food safety training program tailored for CEA producers is being led by research and extension personnel at Auburn University. This program will improve food safety protocols for CEA operations and will help educate growers and the general public about food safety risks and risk mitigation strategies.
- Postharvest quality studies conducted at Auburn University have quantified the shelf-life of CEA lettuce with and without roots. Removing roots from CEA lettuce prior to storage does not decrease shelf-life and may help reduce foodborne illnesses associated with lettuce.
- Auburn University's FoodU program trains undergraduate and graduate students by immersing them in the campus food system. Students are being prepared for careers in research or food production through hands-on exposure to state-of-the-art production techniques, including greenhouse hydroponics and aquaponics, container farms, market gardens, and a rooftop garden.
- As a result of the Auburn University Aquaponics Research Program, researchers at AU, including Daniel Wells, Timothy Bruce, David Cline, and Camila Rodrigues, were part of a team led by UC-Riverside who was awarded a grant, funded by USDA-NIFA, entitled "Mitigating antimicrobial resistance risk in aquaponic systems by selective capturing and electro-catalytic degradation of antibiotics in recirculating water". The subcontract for Auburn University is for \$300,000 and the term of the grant is from 07/01/2025 06/30/2027.

# 5. Published Written Works.

# Refereed Journal Articles

Oliveira, B. Padeniya, U., Bledsoe, J.W., Davis, D.A., Liles, M.R., Hussain, A.S., Wells, D.E., and T.J. Bruce. 2025. Evaluation of probiotic effects on the growth performance and microbiome of Nile tilapia (Oreochromis niloticus) in a high-density biofloc system. Aquaculture Nutrition. 2025, 5868806. <u>https://doi.org/10.1155./anu/5868806</u>

Mickos, V.P., Blanchard, C., Pizzo, J.S., Kitchens, S., Price, S., Wells, D., Rodrigues, C. Controlling *Salmonella enterica* in Water Recirculating Systems for Lettuce Production using a Bacteriophage Cocktail (Accepted in HortScience in May 2025). Rodrigues, C., Blanchard, C., Trandel-Hayse, M., Wells, D., Rehman, T. Post-harvest strategies to improve shelf-life of indoor-grown lettuce (accepted at Acta Horticulturae in November 2024).

Arthur, W., Z. Morgan, M. Reina, E. Drabold, D.E. Wells, D.V. Bourassa, Q. Wang, B.T. Higgins. 2024. Pilot-Scale Evaluation of Poultryponics: Insights into Nitrogen Utilization and Food Pathogen Dynamics. ACS ES&T Water. 4(9): 3964-3975. https://doi.org/10.1021/acsestwater.4c00262

Arthur, W., Z. Morgan, A.E. Inskeep, C. Browne, D.E. Wells, D.V. Bourassa, B.T. Higgins. 2024. Poultryponics: Cultivation of hydroponic lettuce using treated poultry processing wastewater for increased nitrogen neutrality. Bioresource Technology. 422: 132227. https://doi.org/10.1016/j.biortech.2025.132227

Arthur, W., C.K. Akplah, E.T. Drabold, S. Manjankattil, J. Smith, D.E. Wells, D.V. Bourassa, B.T. Higgins. 2024. Dosing *Salmonella* into Poultryponics: Fate of *Salmonella* during treatment of poultry processing wastewater and irrigation of hydroponic lettuce. Journal of Environmental Management. 377: 124559. <u>https://doi.org/10.1016/j.jenvman.2025.124559</u>

#### Extension Publications

Fogarty, S., Newbold, E., Dunn, L., Rodrigues, C., Calatena, R., Bihn, E., George, L., Machado, R., Sirsat, S., Callahan, C. Glossary of Aquaponic and Hydroponic Produce Safety Terms. National Food Safety Clearinghouse at the University of Vermont, 2025. Available at: <u>https://foodsafetyclearinghouse.org/resources/glossary-aquaponic-andhydroponic-produce-safety-terms</u>.

#### Poster and Oral Presentations

Blanchard, C., Trandel-Hayse, M., Rodrigues, C., Wells, D., and T. Rehman. 2024. Fresh weight of indoor-grown lettuce under different postharvest storage practices. HortScience 59(9):S379 – Presented at the 2024 ASHS Annual Conference. September 23-27, 2024. Honolulu, HI.

Trandel-Hayse, M., Wells, D., Rehman, T., Blanchard, C., Rodrigues, C., and MD Rahman. 2024. Nutritional quality and shelf-life of "living lettuce" through 28 days of cold storage. HortScience 59(9):S174 – Presented at the 2024 ASHS Annual Conference. September 23-27, 2024. Honolulu, HI.

Rodrigues, C. Mickos, V., Blanchard, C., and D. Wells. 2024. Bacteriophage as an alternative method to control Salmonella enterica in water-recirculated systems for lettuce production. HortScience 59(9):S127 – Presented at the 2024 ASHS Annual Conference. September 23-27, 2024. Honolulu, HI.

Padeniya, U., Lukwesa, D., Davis, D.A., Wells, D.E., and T.J. Bruce. 2024. Evaluating the influence of dietary immunostimulants on growth in Nile tilapia (Oreochromis niloticus) and romaine lettuce (Lactuca sativa) in a bioflocintegrated aquaponics system. Presented at Aquaculture America 2024. February 18-21, 2024. San Antonio, TX. https://wasblobstorage.blob.core.windows.net/meetingabstracts/AA2024AbstractBook.pdf

Lukwesa, D., Katende, A., Ayipio, E., and D.E. Wells. 2024. Evaluation of split-root system in shrimp effluent-based saline aquaponics for managing salinity stress in red kale ('KX-1') Brassica napus L. var Paularia. Presented at Aquaculture America 2024. February 18-21, 2024. San Antonio, TX. https://wasblobstorage.blob.core.windows.net/meetingabstracts/AA2024AbstractBook.pdf

Katende, A. Ayipio, E., Lukwesa, D., and D.E. Wells. 2024. Split-root hydroponics: investigating cherry tomato resilience to salinity stress. Presented at Aquaculture America 2024. February 18-21, 2024. San Antonio, TX. https://wasblobstorage.blob.core.windows.net/meetingabstracts/AA2024AbstractBook.pdf

Katende, A. Lukwesa, D. Bender, G., Smith, M.R., and D.E. Wells. 2024. Substrate combinations for reduced perlite reliance in tomato cultivation. HortScience 59(2) supplemental: SR63. Presented at the 2024 Annual Meeting of the Southern Region of ASHS. February 2-4, 2024.

Lukwesa, D., Lopez, J., Oyedele, R., Bartley, P., and D.E. Wells. 2024. Evaluating the combined effects of gypsum and split-root system on cherry tomatoes (Solanum lycopersicum var. cerasiforme) and red kale (Brassica napus L. var Pabularia 'KX-1') salinity tolerance threshold. HortScience 59(2) supplemental: SR63. Presented at the 2024 Annual Meeting of the Southern Region of ASHS. February 2-4, 2024.

Trandel-Hayse, M., Blanchard, C., Wells, D., Rehman, T., Rahman, Md, and C. Rodrigues. 2024. Postharvest quality and shelf-life of living lettuce: should growers keep or cut the roots? HortScience 59(2) supplemental: SR37. Presented at the 2024 Annual Meeting of the Southern Region of ASHS. February 2-4, 2024.

Katende, A., Lukwesa, D., Ayipio, E. and D.E. Wells. 2024. Split-root hydroponics: investigating cherry tomato resilience to salinity stress. HortScience 59(2) supplemental: SR12. Presented at the 2024 Annual Meeting of the Southern Region of ASHS. February 2-4, 2024.

Lukwesa, D., Katende, A., Wells, D.E., and E. Ayipio. 2024. Evaluating split-root

system in shrimp effluent-based saline aquaponics for managing salinity stress in red kale ('KX-1') (Brassica napus L. var. Pabularia). HortScience 59(2) supplemental: SR11. Presented at the 2024 Annual Meeting of the Southern Region of ASHS. February 2-4, 2024.

Pennington, M., Fain, G., Gamble, A., Pickens, J., and D. Wells. 2024. Effects of combining controlled release fertilizer and organic matter on nutrient retention in green roof media. HortScience 59(2) supplemental: SR8. Presented at the 2024 Annual Meeting of the Southern Region of ASHS. February 2-4, 2024.

Blanchard, C., Trandel, M., Rodrigues, C., Wells, D., Rahman, T. Fresh Weight of Indoor-Grown Lettuce under Different Postharvest Storage Practices. ASHS, Honolulu, Hawaii, 2024.

Sandoval, E.T., Blanchard, C., Trandel, M., da Silva, A.L.B.R., Rodrigues, C. Controlling *Salmonella enterica* in Roots of Indoor-Grown Lettuces. IAFP Annual Meeting, Long Beach, CA, 2024.

Rodrigues, C., Mickos, V., Blanchard, C., Wells, D. Controlling *Salmonella enterica* in Water Recirculating Systems for Lettuce Production using a Bacteriophage Cocktail. IAFP Annual Meeting, Long Beach, CA, 2024.

Rodrigues, C., Blanchard, C., Trandel-Hayse, M., Wells, D., Rehman, T. Post-harvest strategies to improve shelf-life of indoor-grown lettuce. V International Conference on Fresh-Cut Produce: Maintaining Quality and Safety, Foggia, Italy, 2024.

Volz, T., Rodrigues, C., Dunn, L.L., Jackson-Davis, A., Ferrarezi, R.S. Bridging the Gap: A Comprehensive Needs Assessment Survey to Identify Food Safety Knowledge Gaps Among Indoor Growers in the United States. Southern Region American Society of Horticultural Science, Atlanta, GA, 2024.

Rodrigues, C. Invited speaker "Food Safety for Indoor Agriculture" at the Alabama Fruit and Vegetable Growers Association Meeting, Gulf Shores, AL, 2024.

Rodrigues, C. Invited speaker "Mitigating Food Safety Hazards in Controlled Environment Agriculture" at the 22nd Southeast Regional Fruit and Vegetable Conference, Savannah, GA, 2024.

Rahman, M. H., & Rehman, T. U. (2024). Assessing Salt Stress Tolerance in Kale Plants Grown in an Aquaponics Environment Using a High-Throughput Phenotyping System. *American Society of Agricultural and Biological Engineers Annual International Meeting*, July 28-31, Marriott Anaheim – Anaheim, CA. Rahman, M. H., Rehman, T. U., Busby, S., Ru, S., & Sanz Saez de Jauregui, A. (2024). Drought Tolerance Assessment with Statistical and Deep Learning Models on Hyperspectral Images for High-throughput Plant Phenotyping. *International Conference on Precision Agriculture*, July 21-24, Manhattan, Kansas, USA.

Rahman, M. H., & Rehman, T. U. (2023). Assessing the salt stress tolerance of Kale plants grown in aquaponics system via spatial and spectral predictive regression models. *American Society of Agricultural and Biological Engineers Annual International Meeting*, July 9-12, Marriott Anaheim – Omaha, Nebraska.

#### <u>Training</u>

Rodrigues, C. Produce Safety Alliance Growers Training tailored to Hydroponic and Aquaponic Audience. Boston, MA, 2025.