

M-23

Synthetic design of soybean for improved aquaculture feed traits

*Edgar B. Cahoon**, Center for Plant Science Innovation & Department of Biochemistry, University of Nebraska-Lincoln, Nebraska, USA

Hae Jin Kim, Center for Plant Science Innovation & Department of Biochemistry, University of Nebraska-Lincoln, Nebraska, USA

Tam Nguyen, Center for Plant Science Innovation & Department of Biochemistry, University of Nebraska-Lincoln, Nebraska, USA

Hanh Nguyen, Center for Plant Science Innovation & Department of Agronomy and Horticulture, University of Nebraska-Lincoln, Nebraska, USA

Shirley Sato, Center for Plant Science Innovation & Department of Agronomy and Horticulture, University of Nebraska-Lincoln, Nebraska, USA

Lili Hou, Center for Plant Science Innovation & Department of Agronomy and Horticulture, University of Nebraska-Lincoln, Nebraska, USA

Thomas E. Clemente, Center for Plant Science Innovation & Department of Agronomy and Horticulture, University of Nebraska-Lincoln, Nebraska, USA

The use of soybean in aquaculture feed rations is particularly attractive as a sustainable substitute to the current practice of feeding meal and oil from low-value marine fish to high value farm-raised fish. Soybean is a viable feedstock for aquaculture production due in large part to the demonstrated quality of its meal as a protein source. The economic viability of soybean as a land-based source of aquaculture feed can be further enhanced by development of germplasm that produces fish oil-type fatty acid compositions and high value carotenoids, such as astaxanthin, for fish flesh pigmentation. Such traits would effectively complement the soybean meal fraction for a superior plant-derived aquaculture feed source. Using a conventional gene stacking approach, we recently demonstrated the ability to combine germplasm for two fatty acid traits and an astaxanthin trait to generate soybean seeds with up to 45 $\mu\text{g/g}$ astaxanthin and oil containing 3% to 5% of the fish-oil type fatty acid eicosapentaenoic acid (EPA). This was accomplished through a multi-year crossing effort that resulted in the stacking of seven transgenes. More recently, we have used synthetic biology approaches to increase the rate of gene stacking and to reduce the genetic complexity of an optimized aquaculture oil profile. Using gene synthesis and modular gene assembly, a single 25.5 kb T-DNA was generated that contained seed-specific expression cassettes for nine transgenes to produce EPA (five transgenes), astaxanthin (three transgenes), and vitamin E tocotrienol antioxidants (one transgene). This T-DNA was successfully introduced into soybean to generate lines with seeds containing up to 13% EPA, >100 $\mu\text{g/g}$ astaxanthin, and ~1,000 $\mu\text{g/g}$ tocotrienols. These lines are now undergoing field evaluation and seed bulking for aquaculture feeding trials.