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"Phycosomal dynamics in xenic cultures of the alkalitolerant green microalga *Chlorella sp. SLA-04*"

ABSTRACT

The production of microalgal biomass and biofuel is an important component of the transition away from a petroleum-based economy. Industrial scale microalgal cultures are often xenic, meaning they are comprised of microalgae as well as a phycosome (*i.e.*, microbiome). The microalgal field has begun to appreciate the ubiquity and potential influence of the phycosome, but there remains a critical need for comprehensive research to unravel the intricate metabolic and ecological relationships between microalgae and the respective phycosome that can be comprised of mainly bacteria but also other microorganisms (*i.e*., archaea, fungi, protists, viruses). Phycosome research is essential for potentially using these interactions to enhance the stability, productivity, and cost-efficiency of industrial microalgal cultivation. *Chlorella* sp. SLA-04 is an oleaginous, alkalitolerant microalga isolated from the alkaline Soap Lake (Washington, USA). Under alkaline conditions, SLA-04 can be grown to high biomass levels without reliance on the delivery of concentrated CO_2, an improvement in producing competitively priced biomass and biofuel. The high pH, high alkalinity systems are able to capture CO_2 directly from the air in open systems (*e.g*., raceway ponds) but the open systems can be dynamic in terms of stability and productivity. Despite growing knowledge of the importance of phycosomes in open production systems, little is known about how alterations to cultivation conditions can be used to maintain a xenic system with controllable outputs, especially under high pH, high alkalinity conditions. The work outlined in this dissertation employed long term temporal community studies, open outdoor raceway experiments, diel-cycle-resolved temporal sampling coupled with activity-based probing (bioorthogonal non-canonical amino acid tagging (BONCAT)), and quantitative measures of algal physiology to better understand the relationship between microalgal phenotype and the respective phycosomes. SLA-04 phycosome composition and culture physiology were consistent over time when maintained in xenic cultures under low and high alkalinity. When xenic cultures were used in successive open, outdoor raceway experiments, compositional community changes coincided with seasonal temperature and light shifts, providing evidence that abiotic and biological environmental stresses impact directly and indirectly SLA-04 productivity and phycosome composition. By employing temporally resolved sampling and probing the relationship between diel-cycle-dependent metabolism and the phycosome, we identified active bacterial populations that may play a role in culture productivity. Expanding beyond augmenting SLA-04 productivity, aggregation of xenic cultures was assessed as a quantifiable phenotype, uncovering a relationship between aggregation, taxonomic composition and algal growth conditions (*i.e.,* alkalinity level). All together, these results represent an initial description of the ecology (*e.g*., composition, succession, activity) of alkaline microalgae cultures and provide methodology and perspective for future phycosome studies.

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