

Genetic Engineering Yeast to Produce Lipid Based Biofuel for the Future

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The commercial production of biofuel is not yet economically viable due to slow rates of production from microalgae. This problem could be alleviated by transitioning to *Saccharomyces cerevisiae* (yeast) for the production of biodiesel as *Saccharomyces cerevisiae* is used to produce 100% of bioethanol. Biodiesel includes triacylglycerol (TAG) lipids and diacylglycerol-N,N,N-trimethylhomoserine (DGTS) lipids. Through the BTA1cr gene from microalgae, those lipids synthesized can be used as a source of biodiesel. In E.P.Y part one, *Saccharomyces cerevisiae* was transformed with the pBTA1 plasmid allowing yeast with recombinant DNA (rDNA) to synthesize DGTS lipids at 8% lipid to biomass ratio, with the control untransformed cells being 0% lipid to biomass ratio. In E.P.Y part two, the transformed cell's lipid yields were increase using guided evolution to select the cells with higher lipid concentration making an oleaginous stain of yeast. Environmental stress includes starch limitation with ACCase inhibitors. The cells are displaced into a solution with desilt H2O and ACCase inhibitors. The ACCase inhibitor stops the process of lipid synthesis by targeting the enzyme acetyl-CoA carboxylase (ACCase) which is an enzyme crucial in the lipid synthesis cell process. The control of this experiment was regular *Saccharomyces cerevisiae*, as the average lipid to biomass ratio was found using procedure L. The results show that guided evolution works with a targeted stress environment to increase the lipid to biomass ratio of yeast to isolate oleaginous cells, allowing a new strain of yeast with a lipid to biomass ratio of 40.3%, which is a 506.5%-fold increase. This new stain of yeast can be used to produce both bioethanol and biodiesel in sufficient amount as a potential biofuel.

Awards Won:

Arizona State University: Arizona State University Intel ISEF Scholarship

University of Arizona: Tuition Scholarship Award