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IMPROVING ROS RESISTANCE IN *CHLORELLA VULGARIS* TO INCREASE BIOMASS PRODUCTION

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The utilization of biomass for biofuel production is one of the key elements for the development of a sustainable energy supply. In particular, biomass from microalgae has gained increasing attention in recent years due to the many advantages over terrestrial crops. However, the economic feasibility of growing algae at an industrial scale is yet to be realized, in part due to biological constraints limiting biomass yield.

A key issue is the inefficient use of light at high irradiances typical of photobioreactors' massive culture. Due to mixing, cells experience peaks of high light, which result in an imbalance on rates of light absorption and CO2 fixation, and lead to massive release of ROS within the chloroplast. In particular singlet oxygen ($^1O_2$) is an unavoidable by-product of oxygenic photosynthesis, causing progressive oxidative damage, photoinhibition and ultimately cell death. Thus, the successful implementation of biomass and biofuel production facilities requires the development of strains with increased oxidative stress responses and enhancement of detoxification networks.

To this aim, we applied random mutagenesis and phenotypic selection to the fast-growing microalga *Chlorella vulgaris*. The organic dye Rose Bengal (RB), which generates singlet oxygen when illuminated, was added to the growth medium for the selection of 1O2-resistant mutants. Three putative interesting strains were found, showing a higher resistance to $^1O_2$, being able to survive to RB dose lethal for the parental strain.

Cultivation of two of these $^1O_2$-resistant strains in laboratory-scale showed higher productivity than the parental genotype, with a ~30% higher biomass yield in both dense cell suspensions and high irradiances, typical of industrial bioreactors. These results suggest that generation of mutants with higher resistance to ROS is a promising strategy in the development of domesticated microalgal strains for mass culture.