

**Genetic engineering of marine alga potential for biofuel applications and acclimation of strain in natural system**Shashi Kumar<sup>1</sup>, Prachi Nawkarkar<sup>1</sup>, and Amit K Singh<sup>2</sup><sup>1</sup>International Centre for Genetic Engineering and Biotechnology (ICGEB), India<sup>2</sup>Michigan State University, USA**Abstract**

During in vitro culture of wild algae acclimation to artificial environment pose a big challenge to thrive back to the natural system. Many algae when subjected to mutation or genetic transformation and maintained for long time in vitro environment face difficulty in becoming accustomed to a new climate or new environmental conditions. Photoautotrophic algae are important for producing sustainable biofuel, which is impeded due to low uptake of CO<sub>2</sub> from aquatic environments to saturate the activity of the carboxylating enzyme RuBisCO for maximal fixation of carbon dioxide. This greatly reduces the catalytic activity of RuBisCO in fixing CO<sub>2</sub> for producing more biofuel molecules and other important products. Our group has genetically engineered a commercially important oleaginous marine alga *Parachlorella kessleri*-I by overexpressing bicarbonate transporters from another green alga to enhance the supply of carbon dioxide to RuBisCO. This genetic modification approach has tremendously increased the CO<sub>2</sub> accumulation in GM *P. kessleri*-I around the pyrenoid. The higher supply of CO<sub>2</sub> has favored the carboxylation reaction in RuBisCO over photorespiration (oxygenation reaction) thereby enhancing the biomass productivity, which made the transgenic cells fat. The GM alga displayed about two-fold higher carbonic anhydrase activity while 3-4-fold higher neutral lipid accumulation when results were compared with the wildtype algae. Also, higher time course starch accumulation was observed around the pyrenoid in electron micrograph of transgenic alga. Further results were encouraging for life cycle assessment and scale up study, transgenic alga showed 4-fold more biomass productivity in presence of bicarbonate compared to wildtype cells. The enhanced lipid accumulation was complemented by elevated concentrations of saturated and mono-unsaturated fatty acids, which is advantageous for the production of high-quality biofuel using GM algae. prior to transfer the modified microalgae to natural light, we have carried out the comparative studies of different light intensities on a marine alga *P. Kessleri*-I. There were no significant differences observed in malondialdehyde (MDA) content during growth in high light (HL). The higher NPQ in HL grown alga than low light (LL) grown alga suggests that NPQ plays an important role in photoprotection during acclimation of transgenic alga when exposed to HL. Thus, optimizing light condition for acclimation in natural conditions, external supply of bicarbonate and intracellular transport of inorganic carbon has significantly improved the commercial viability of genetically modified *P. kessleri*-I for producing sustainable biofuels.

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## **Biography:**

Shashi Kumar is a Team Leader at ICGEB in New Delhi, India. He has graduated in Genetics from the University of Delhi, 1998, Postdoctoral research at University of Virginia, University of Central Florida, University of California Berkeley and Scientist at Yulex Inc., USA and USDA, Albany, USA. He has participated in establishing the “Centre for Advanced Bio-Energy Research” supported by the Department of Biotechnology, Government of India. His area of interests include metabolic engineering for drug biosynthesis, metabolic engineering of rubber plant for hypoallergenic latex, DNA barcoding, development of sustainable algal biofuel technology and genetic engineering of marine algae for higher lipid and biomass.