Sociedad Argentina de Microbiología General

XIX CONGRESO DE LA SOCIEDAD ARGENTINA DE MICROBIOLOGÍA GENERAL

22 al 25 de octubre del 2024
Centro cultural y Pabellón Argentina de la Universidad Nacional de Córdoba, Córdoba, ARGENTINA.

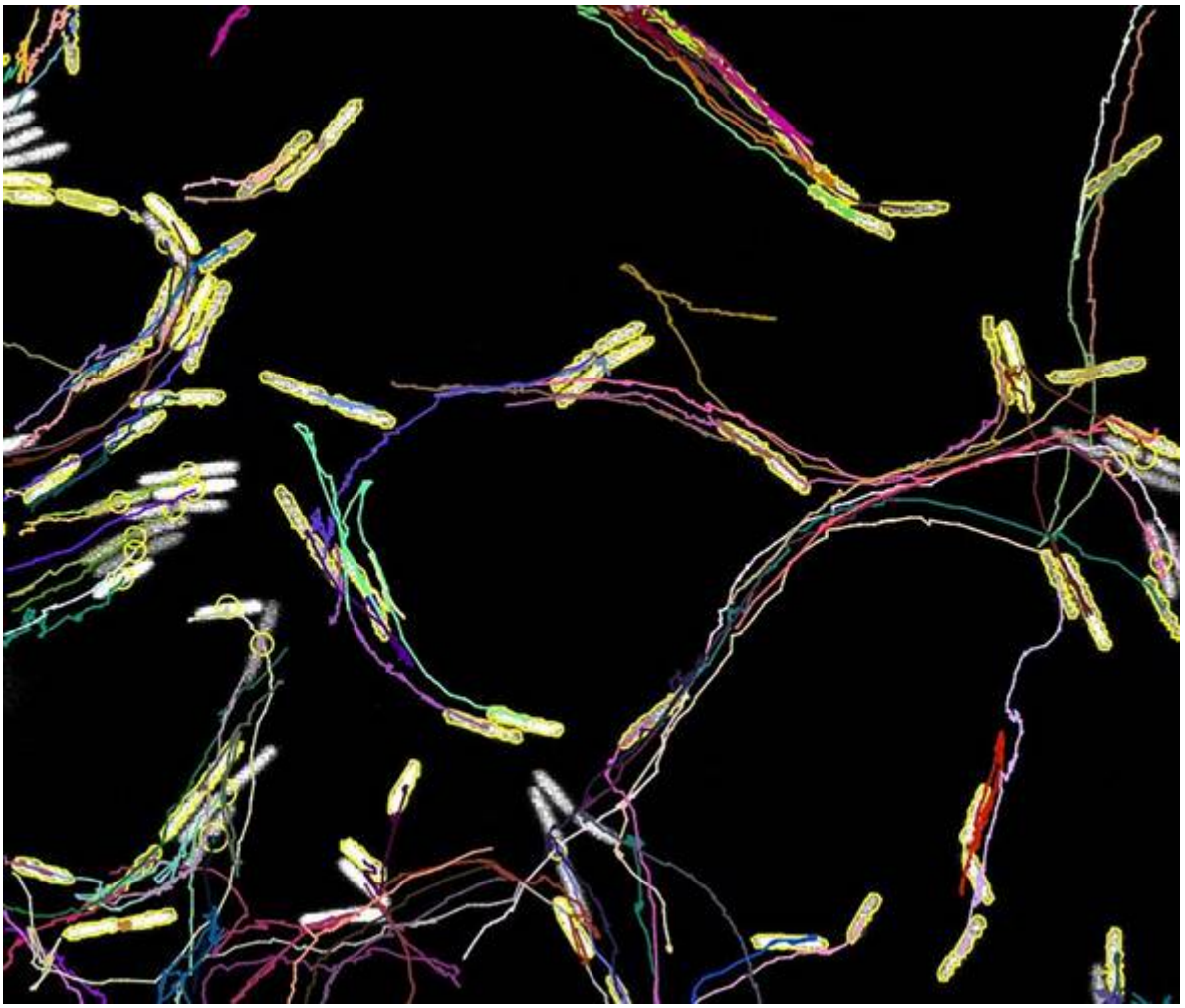

Foto: Se hace camino al andar. Celeste Dea. 1er puesto. Concurso fotográfico SAMIGE 20 años.

CHARACTERIZATION OF THE FUNCTIONALITY OF A SOLUBLE AND CHLOROPLASTIC DIACYLGLYCEROL ACYLTRANSFERASE (DGAT) THROUGH ITS EXPRESSION IN YEAST

Guaycochea, Santiago D. - Gómez Jousse, Micaela - Ferraro, Gisela - Bagnato, Carolina

Departamento de Eficiencia Energética y Biotecnología Ambiental (DEEBA) - Centro Atómico Bariloche - Comisión Nacional de Energía Atómica - San Carlos de Bariloche - Río Negro - Argentina. Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET) - CCT Patagonia Norte - San Carlos de Bariloche - Río Negro - Argentina.
Contacto: santyg45@gmail.com

Lipid metabolism in algae is complex and diverse. Microalgae are recognized as significant producers of triacylglycerols (TAGs), which are key precursors in various industries, such as biofuel production. This has driven a growing interest in identifying oleaginous algae, optimizing cultivation and lipid productivity, and studying the metabolic pathways leading to their synthesis. During a characterization study of the TAG synthesis pathway in *Chlamydomonas reinhardtii*, we identified a soluble, chloroplastic diacylglycerol acyltransferase (DGAT) that showed homology with the emerging DGAT3 family. However, this family has not yet been extensively characterized in terms of its activity and catalytic site. Moreover, its low homology with the DGAT1 and DGAT2 families makes it difficult to draw conclusions based on previous studies. To deepen our understanding of TAG synthesis pathways in algae, this study aimed to characterize the functionality of *C. reinhardtii* DGAT3. The yeast mutant strain H1246, which has a deficiency in neutral lipid synthesis, was used to characterize the activity in vivo in a eukaryotic system. In this way, any increase in the content of these lipids can be attributed exclusively to the expression of the enzyme under study. The DGAT3 sequence was cloned into the yeast expression vector pYES2. Yeast cells were transformed using the polyethylene glycol/lithium/acetate method, and induction was performed with galactose. Lipid accumulation and profile were evaluated by microscopy and thin-layer chromatography (TLC) in cultures of Saccharomyces cerevisiae wild-type, pYES2, and pYES2-DGAT3 H1246 strains. Activity was evaluated at different incubation times and with the addition of free fatty acids and different acyl acceptors. The expression of DGAT3 in yeast allowed us to observe a different pattern of lipid accumulation. In the wild-type strain, a growing accumulation of lipids was observed over time, while the strain transformed with DGAT3 showed accumulation at the end of the cultivation time. Lipid profile analysis by TLC showed that DGAT3 expression restored the synthesis of TAGs and waxes in the mutant yeast. This would indicate that *C. reinhardtii* DGAT3 can contribute to the accumulation of TAGs and waxes, and that this depends on the metabolic environment.

Palabras clave: TAGs - Lipids - Biodiesel - Microalgae