



<u>Alejandro Ibanez Salazar</u>, Jocelin Itzel Ramírez Alonso, Luz María Teresita Paz Maldonado, Sergio Rosales Mendoza, Ruth Elena Soria Guerra; Universidad Autónoma de San Luis Potosí, Facultad de Ciencias Químicas, San Luis Potosí, S.L.P 78240; sithalex_@hotmail.com

Key words: microalgae, triacylglycerols, biofuels

Introduction. Chlamydomonas reinhardtii is a photosynthetic microalgae, which is now regarded as a new biotechnology platform (1).It has a multiple biotechnological advantages. In particular, it is a naturally fatty acid accumulator under stress (2). However, reduced biomass and lipids observed under these restrictive nutrient conditions (3). Since the fuel demand at the global level is increasing, the search of new fuel sources in a low-cost and environmentally friendly constitutes priority. These manner а characteristics are meeting by biofuels. Genetic engineering approaches to increase the accumulation of lipids in Chlamydomonas reinhardtii is proposed as an alternative in this field. Interestingly, several plant genes encoding for transcription factors have been associated with the increase of total oil content when they are over expressed. This is the case of the soybean Dof11 transcription factor (4).

In this study, we present a strategy to over express the *Dof11* transcription factor in *Chlamydomonas reinhardtii* in order to pursue the development of Chlamydomonas strains with improved capacity on fatty acid production.

Methods. Expression vector construction.

Genetic transformation by Agrobacterium tumefaciens.

Selection and propagation of candidate clones.

Molecular analysis of putative transformants. Sudan III staining

Results.

Expression vector construction

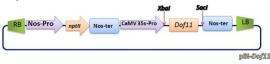


Fig.1 Expression vector pBI-Dof11. This vector is based in the pBI121 backbone, modified by replacing the GUS gene by the gene encoding the transcription factor Dof11.

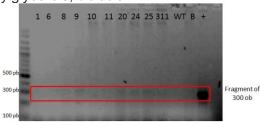


Fig.2 PCR to detect the *Dof11* gene from genomic DNA extracted from *Chlamydomonas reinhardtii* putative transformants. Lines: 1 to 311, putative transformants; s, WT, wild type, untransformed line; B, Blanco; +, Positive control, pBI-Dof11

Identified positive lines: 8, 9, 20, 24, 25, 311.

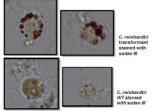


Fig. 3 Microscopy analys of Sudan III stained clones. Note stronger signals present in the transformants in comparison to the WT strain.

Conclusions. We have generated a number of candidate clones which are likely increased in lipid production. These clones are currently under bioreactor based propagation as well as transcriptional and lipid profile characterization. These strains represent an important tool in the development of new biofuel production platforms.

Acknowledgements. This work is under funding by CONACYT # 151480. Authors thank to Bioreactors Lab from the FCQ-UASLP.

References.

1. Rosales-Mendoza S, Paz-Maldonado LM, Soria-Guerra RE. (2012). Plant Cell Rep. 31(3):. 479-94.

2. Zi Teng Wang, Nico Ullrich, Sunjoo Joo, Sabine Wattenschmidt, Ursula Goodenough. (2009). Eukaryotic cell. 8(12):. 1856-68.

3. Loera-Quezada MM, J Olguin E. (2010). Latinoam Biotecnol Amb Algal. 1(1):. 91-116.

4. Hui-Wen Wang, Bo Zhang, Yu-Jun Hao, Jian Huang, Ai-GuoTian, Yong Liao, Jin-Song Zhang, Shou-Yi Chen. (2007). The plant Journal. 52:. 716-729.