CLASSICAL BREEDING IN THE DIATOM SPECIES Seminavis robusta FOR BIOTECHNOLOGICAL USE IN THE EUROPEAN AQUACULTURE INDUSTRY

Francesco Pisapia^{*1}, Peter Chaerle², Francesca Cucchi¹, Olga Chepurnova², Antera Martel Quintana¹, Wim Vyverman² and Juan Luis Gómez Pinchetti¹

¹ Spanish Bank of Algae (BEA), Canarian Science and Technology Park Foundation (FCPCT), University of Las Palmas de Gran Canaria (ULPGC), Muelle de Taliarte s/n, 35214 Telde, Spain. francesco.pisapia@fpct.ulpgc.es, francesca.cucchi.pagani@gmail.com, antera.martel@ulpgc.es juan.gomez@ulpgc.es,

² Laboratory for Protistology and Aquatic Ecology, Department of Biology, Ghent University, Krijgslaan 281-S8, 9000 Gent, Belgium.

peter.chaerle@ugent.be, olga.chepurnova@ugent.be, wim.vyverman@ugent.be

Abstract: Microalgae are a natural source of high-value fatty acids and could address the reduced availability of fish oil from pelagic fisheries and the increasing needs from the aquaculture sector and the ω -3 market (Chauton et al., 2015). Still, their exploitation on industrial scale is scarce, mainly due to the lack of highly performant strains.

Classical breeding constitutes a source of genetic diversification in microalgae, generating progeny strains with unique genetic heritage and bypassing the controversial issue of genetic modified organisms (GMOs). In the framework of the H2020 NewTechAqua project, our goal is to generate improved microalgae strains via classical breeding for application as aquafeed in European aquaculture. The heterothallic diatom species *Seminavis robusta* is particularly suitable for controlled breeding programs based on pedigree selection (Chepurnov et al., 2012).

Eight wild-type strains of *S. robusta* were purchased from the BCCM/DCG collection (PAE laboratory, UGENT, Belgium) and crossbred according to their sexual compatibility (De Decker et al., 2018). Single cells of 66 F1 progeny strains from eleven breeding pairs successfully developed into monoclonal cultures. All the 74 strains were cultured for twelve days under the same growth conditions in 6-well plates. Sixteen bright field and fluorescence CY5 images per well were taken every three days using the CytationTM 3 plate reader and imager from BioTek, and processed using Gen5 and ImageJ softwares. Cell density over time was estimated using the percentage of surface showing CY5 fluorescence over the total surface.

Sixteen F1 progeny strains aroused biotechnological interest as they showed faster growth compared to their wild-type parents. Further studies will address fatty acid composition and quantification for all the strains. The most performant F1 strains will undergo crossbreeding experiments in an iterative manner, using a pedigree-based approach. Our findings will ultimately contribute to achieve a more sustainable, resilient and cost-effective European aquaculture.

Key words: Classical breeding, Seminavis robusta, aquaculture, w-3 PUFAs, DHA, EPA

Acknowledgments: The authors acknowledge funding from the Horizon 2020 "NewTechAqua" project (PID: 862658) and staff mobility funding from the 8th call of the Assemble Plus TNA programme ("SEBREDIFACEA" project, PID: 13290).

References:

- Chauton, M. S., Reitan, K. I., Norsker, N. H., Tveterås, R., & Kleivdal, H. T. (2015). A techno-economic analysis of industrial production of marine microalgae as a source of EPA and DHA-rich raw material for aquafeed: Research challenges and possibilities. Aquaculture, 436, 95–103. https://doi.org/10.1016/j.aquaculture.2014.10.038
- Chepurnov, V. A., Chaerle, P., Vanhoutte, K., & Mann, D. G. (2012). How to Breed Diatoms: Examination of Two Species with Contrasting Reproductive Biology (pp. 323–340). https://doi.org/10.1007/978-94-007-5110-1_18
- De Decker, S., Vanormelingen, P., Pinseel, E., Sefbom, J., Audoor, S., Sabbe, K., & Vyverman, W. (2018). Incomplete Reproductive Isolation Between Genetically Distinct Sympatric Clades of the Pennate Model Diatom Seminavis robusta. Protist. https://doi.org/10.1016/j.protis.2018.05.003