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New challenges in microalgae biotechnology

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Abstract

Photosynthetic protists, also called microalgae, have been systematically studied for more than a century. However, only recently broad biotechnological applications have fostered a novel wave of research on their potentialities as sustainable resources of renewable energy as well as valuable industrial and agro-food products. At the recent VII European Congress of Protistology held in Seville, three outstanding examples of different research strategies on microalgae with biotechnological implications were presented, which suggested that integrative approaches will produce very significant advances in this field in the next future. In any case, intense research and the application of systems biology and genetic engineering techniques are absolutely essential to reach the full potential of microalgae as cell-factories of bio-based products and, therefore, could contribute significantly to solve the problems of biosustainability and energy shortage.

Keywords: Algae biotechnology; Biofuels; Genetic engineering; Microalgae; Photosynthetic protists; Systems biology

Introduction

Energy and its sustainable production is one of the most important resources for mankind. Sunlight is by far the most important input of energy to Earth, and photosynthesis is the main biological process channeling solar energy into the biosphere. Eukaryotic microalgae are a taxonomically broad and heterogeneous group of phototrophic protists of increasing biotechnological interest due to their higher photosynthetic efficiencies relative to land plants (microalgae contribute up to 25% of global photosynthetic productivity), elevated growth rates and vast metabolic capabilities (Raven and Falkowski 1999). In particular, the green microalgae (*Chlorophyta*) share the same photosynthetic machinery as the higher plants, according to their close phylogenetic relationships.

Microalgae such as *Chlorophyceae* and *Bacillariophyceae* use sunlight energy and a simple set of abundant, cheap resources (carbon dioxide, water and minerals) to generate a potential large number of valuable products of technological interest. These products can be applied, either directly or after transformation, in industrial, pharmaceutical and agro-food processes; examples are carotenoids, oils, polysaccharides, pigments, bioethanol, hydrogen, microalgal biomass (Cadoret et al. 2012; Finazzi et al. 2010; León et al. 2008). Indeed, some microalgal species accumulate important amounts of these compounds under specific environmental conditions, so this biotechnologically relevant

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phenotypic feature is amenable to optimization by genetic engineering approaches (Cadoret et al. 2012; León et al. 2008).

This review summarizes the contributions presented by three microalgal biotechnologists at the symposium *New Challenges in Microalgae Biotechnology* held during the VII European Congress of Protistology, which was organized for the first time as a joint meeting in partnership with the International Society of Protistologists (VII ECOP – ISOP Joint Meeting) in Seville, Spain, 5–10 September 2015. The use of microalgae as sustainable oil sources for biofuels will be evaluated and discussed in the first section, given that some of these phototrophic protists intrinsically accumulate high oil levels (up to more than 80% of the dry weight). Significant recent advances in the development of genetic manipulation tools, aimed to improve biotechnological features of microalgae as sources of renewable resources, are presented in the second contribution. Finally, the application of computational modeling as a systems biology strategy to better understand microalgal metabolic and cell signaling networks will doubtless contribute to discover novel properties with relevant biotechnological implications, as is presented in the third contribution. In any case, it is clear that intense research and the application of genetic engineering are absolutely essential to reach the full potential of microalgae as cell factories and, therefore, will significantly contribute to solve the problems of biosustainability and energy shortage.

Biofuel from microalgae?

Microalgae are a polyphyletic group and a huge pool of biological diversity. Properties typical of higher plants are combined in microalgae with biotechnologically amenable attributes of microbial cells. These and other properties of microalgae (such as their metabolic plasticity, tolerance to extreme environmental conditions and amenability to genetic engineering), are valuable for bioindustry. These photosynthetic microorganisms are a source of compounds with commercial value, such as carotenoids, phycobiliproteins, polyunsaturated fatty acids, polysaccharides and an array of bioactive compounds for agriculture and food, feed, pharmaceutical, cosmetic, and chemical industries. Microalgae can also be of use in the recovery of wastewater and in abatement of carbon dioxide.

Microalgae have been proposed as an alternative source for renewable biofuel, capable of meeting the global demand for transport fuels. Although the “microalgae to biofuel” concept was first suggested in the 1940s, it has recently received new attraction and support. A seminal article by Yusuf Chisti published in 2007 (Chisti 2007) has been particularly effective in drawing the attention of researchers and investors. In this way, many research groups were attracted to the field, together with commercial ventures established thereafter. According to data available in Thomson Reuters’ Web of Science™, the number of published items per year on “biofuel from microalgae” has grown exponentially, from less than 5 before 2007 to over 390 in 2014.

Attention on microalgae for biotechnological reasons has also benefited from the fact that mass production of liquid biofuels from plant biomass is being increasingly questioned. The “food *versus* fuel” dilemma and the limitations in available fertile land for a world’s growing population are reasons to reconsider the biofuel production from crop plants (Searchinger et al. 2015). Microalgae represent an alternative to land plants, since cultures could be developed in non-arable land, employing brackish, saline or even waste water, as well as carbon dioxide from flue gases as carbon source. Values for expected fuel productivity around 20,000 L per hectare and year seem reasonable for outdoor culture of microalgae (Moody et al. 2014), although some substantially higher projections are frequently argued in the literature. However, most of the projected values originate from gross extrapolations, both in area and time, from short-term trials in small size facilities, if not directly from laboratory experiments. Analogous considerations apply to published life cycle assessments and to production prices appraisals for either biomass or biofuel from microalgae. The escalation of these processes offers a very challenging subject for applied research.

Up to now, scarcity of scientific and technical knowledge, as well as limited practical experience, determines a high price for microalgal biomass and the biofuel thereof. The lowest production cost in commercial algae production seems to be about US$ 4–5 per kg algal biomass. Significant R&D efforts are currently being addressed to the development of viable processes able to massively generate microalgal biofuels at prices that can compete with those of established fuels (Sing et al. 2013). The production step has to be considerably improved, but also harvesting, biomass drying and extraction of biofuel precursor and its conversion into the final product still need substantial optimization.

Selection of the most appropriate microalgal strains is a key issue (Fig. 1). Not just the content of the biofuel precursor (either fermentable sugars or fatty acids) should be considered, but rather the production capacity, looking for the optimal combination of product level and biomass productivity. The continuous culture approach is the most appropriate methodology for the screening of microalgae for the purpose of biofuel production, as it allows the determination of real productivity for a particular biofuel precursor (Del Río et al. 2015). Also crucial in the selection of the strain is the ability to develop outdoors as a monoalgal culture throughout the year. Many expectations are placed on the potential of genetic engineering for the generation of strains with superior productivity of either fatty acids or fermentable carbohydrates, but further development of novel techniques for efficient manipulation of microalgae is still needed.

Production of biofuels is largely policy-driven and its profitability has been questioned, even at oil prices above US$ 100/barrel (bbl). Current average price for crude oil is around