

Editorial

Solid-State Direct Fungal Bioconversion of Biomass to Fuels and Chemicals: The Next Frontier in Sustainable Biorefining

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In recent years, the term *sustainable biorefining* has evolved from being merely a technical jargon to a wide-ranging multidisciplinary scientific and engineering enterprise. Numerous publications, conferences, patents, discoveries, and innovations devoted on this topic have resulted since and the pipeline for the production of such outputs continues. By definition, biorefineries consist of integrated processing of biomass (forestry, agricultural crops, organic residues, municipal wastes, aquatic biomass, etc.) into a spectrum of marketable bio-based products (food, feeds, materials, chemicals) and bioenergy (fuels, power, energy) using a wide range of mechanical, thermal, chemical, and biological conversion processes [1]. Adding a sustainability component to biorefining implies that the endeavor seeks to improve the quality of human life while living within the carrying capacity of supporting ecosystems (i.e., the earth) with regards to environmental, social, and economic aspects [2]. Microbial fermentation involving bacteria, yeasts, algae, and fungi which utilize biochemical reactions occurring within microbial cells to convert carbon and nutrients into cellular growth, energy, and metabolic products have long been a key integral part of biorefining technologies. However, most of these processes employ submerged liquid fermentation methods, which necessitate pretreatment and hydrolysis of solid biomass substrates into soluble fermentable sugars that are more readily assimilable by the microorganisms in aqueous media using a variety of physicochemical and enzymatic means. These steps not only add to production and capital costs but in the case of chemical hydrolysis could generate processing waste streams and by-product compounds that could pose inhibitory and toxic effects towards microbial growth and metabolism; thus potentially requiring detoxification of the hydrolyzates prior to fermentation [3]. Therefore, the use of consolidated biological processes that involve direct microbial growth and utilization of solid biomass substrate for the production of value-added bioproducts could alleviate this problem and improve the economical and environmental sustainability of fermentation processes.

For a more efficient microbial fermentation process for the bioconversion of biomass to fuels, chemicals, and other bioproducts, the use of solid-state integrated hydrolysis and fermentation

employing mixed fungal cultures is a potential solution that deserves particular attention. Among the different kinds of microorganisms used in fermentation processes, filamentous fungi such as those belonging to the Zygomycetes, Ascomycetes, and Basidiomycetes stand out due to their expansive portfolio of products such as enzymes, organic acids, alcohols, single cell oils and proteins, biopolymers such as chitosan and glucans, and antibiotics and bioactive compounds with wide ranging applications [4-8]. What sets fungi apart from other microorganisms is their ability to synthesize and excrete a variety of hydrolytic enzymes such as amylases, pectinases, cellulases, xylanases, and ligninases that they use for breaking down polymeric carbohydrates in biomass to fermentable sugars for direct bioconversion to bioproducts with minimal to no physicochemical and/or enzymatic pretreatment. Furthermore, fungi in their natural habitats typically grow in symbiotic associations with other fungal strains on solid substrates such as soils or plant materials and use metabolic synergisms for the complete degradation of biomass to supply carbon and nutrients for growth and metabolism [9]. Mixed culture fermentations have long been the basis for most ancient fermentation processes especially in the area of food and beverage products. For biorefining applications, fungal strains with high lignocellulolytic activities could be combined with strains with biosynthetic capabilities for specific products (i.e., ethanol, organic acids, other metabolites) for direct bioconversion of lignocellulosic biomass to the desired bioproducts with minimal to no external pretreatment and hydrolysis steps. The integrated process could be performed in a single bioreactor or bioprocessing equipment and eliminates intermediate processing steps of biomass hydrolysis such as neutralization or detoxification prior to fermentation. Furthermore, the use of mixed cultures with synergistic metabolic activities could minimize fermentation inhibition due to buildup of biomass degradation intermediates since there would be a continuous flux of carbon towards the production of the metabolites.

However, the development of industrial bioprocesses utilizing consortia of compatible fungal strains that complement each other with regards to hydrolytic and biosynthetic activities for specific bioproducts, let alone large-scale solid-state fermentation cultures face significant technological and engineering challenges that will require focused and concerted research efforts from the scientific community. Challenges specific to solid-state fermentation and bioprocessing include difficulties in sensing, monitoring, and control of bioprocess variables; and fermentation kinetics modeling brought about by build-up of temperature, pH, O₂, substrate, and moisture gradients in static or intermittently agitated solid substrates [9]. Without reliable process monitoring techniques and mathematical models that could describe these complex systems precisely and accurately, the design of bioreactors with appropriate control

systems could prove to be difficult. A few investigators have already recognized these research needs [10-17]. Moreover, for fermentation cultures with multicomponent microbial systems, the complexity of the system further increases. For specific systems and target products and applications, comprehensive investigations must be conducted to determine the appropriate fungal mixed culture composition and culture phasing periods for stability and balance of the system (i.e. preventing culture domination by one strain component); optimize cultivation and downstream conditions; collect reliable bioprocess parameter and biochemical kinetic data for the development of precise and accurate mathematical models; and apply molecular methods to elucidate fungal consortia dynamics and metabolic gene expression mechanisms.

There is a high potential for improving the sustainability of microbial fermentation processes for the production of bio-based products using this approach. A few investigators have already tested this concept of solid-state fungal co-cultivation on improving cellulolytic enzyme activities with positive results [7]. However, its application for the direct production of specific biorefinery products is severely lacking. This could constitute a whole new biorefinery paradigm based on the application of live fungal biocatalysts for the direct bioconversion of biomass under solid-state culture conditions to fuels and chemicals. Unfortunately, the field of solid-state fermentation has not yet reached the level of sophisticated development and application compared with submerged fermentation technologies due to lack of interest and awareness in this field and the inherent difficulties of the challenges ahead. There is a great need for a unified approach on basic and fundamental research with regards to mass and heat transfer phenomenon, the potential of gradients as driving forces, and the molecular aspects of fungal bioactivity in mixed consortia and solid media. Overcoming these challenges could be considered as a new frontier in sustainable biorefining for many years to come.

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