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Microbial Lipid Production

Methods and Protocols

Edited by

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Preface

Lipids derived from conventional sources such as oil seeds include building blocks of cellular membranes. They are utilized to make substances used as energy storage, insulation, a method of cellular communication, and protection. With population growth and increasingly limited farmland, there is an ever-growing demand to develop alternate sources of lipids to meet our food and energy needs. Oleaginous microorganisms such as bacteria, algae, yeast, fungi, and thraustochytrids are a promising, largely untapped, resource for lipid production. Microbial lipids offer some unique advantages, such as high content of unique polyunsaturated fatty acids that are widely used for dietary purposes and of chemically modified lipids that confer desirable physical properties. Lipid content in microorganisms depends upon genetic constituents and environmental conditions such as pH, temperature, exposure to natural light, and nitrogen content in the media. There are several advantages of producing lipids in microorganisms such as lower cultivation costs, ability to produce a diverse range of lipids using genetic manipulation, and the possibility of producing lipids year-round with limited space and infrastructure without the need for agricultural land.

The 24 chapters of this book provide comprehensive routinely used methods for isolation and characterization of lipids and for their production using various oleaginous organisms. The methods presented consider sugars derived from different substrates including chemically pretreated agricultural residues, industrial residues containing lignin, food wastes, and industrial waste water in an approachable format. The authors have also provided detailed applications using oleaginous organisms to transform substrates into a variety of products including bio-crude, high-value fatty acids, biofuels such as biodiesel, neutral lipids, volatile fatty acids, and surfactants. Protocols are presented in a basic standard outline format conducive to adaptation to suit specific application needs. This book is aimed at the novice, and therefore each technique is complete and assumes no prior knowledge. Novel screening protocols to identify oleaginous organisms with exceptional lipid yields, genetic engineering approaches to increase microbial lipid content, methods to judge fuel quality from microbial lipids, and life cycle analysis are some of the unique topics that are covered in this book.

Much of the success of the series is due to the “Notes” section, which describes where common procedural problems are identified, and solutions are discussed along with alternative procedures. It is in this section where important, practical details are presented that are rarely included in other published works. This is how the authors have passed along their practical experience to help mentor readers. Most of the chapters are focused on specific laboratory methods, but the first chapter provides a comprehensive review about lipids derived from plants and various microorganisms, an overview of available analytical techniques to characterize lipids, and their applications in various processes.

It has been inspiring to see the natural diversity of lipid molecules with varying properties and functions synthesized by microorganisms using complex genetic and enzymatic machinery. Also, it is equally inspiring to see the numerous analytical techniques and protocols now available for lipid researchers to use to analyze structure and function and to pursue diverse applications. Though it was challenging to organize such a range of chapter topics to fit together in a common theme, all the contributing authors are commended for their expertise, ensuring each chapter was on topic, and for writing in a clear and

direct style. I take this opportunity to thank both my wife and two daughters for sacrificing family time and motivating me to complete the book at times when my morale ebbed. I thank all my colleagues in the College of Technology and administrator at the University of Houston for their support and encouragement that made this book possible. Also, I thank Dr. Patricia Slinger and Dr. Bruce Dien from USDA ARS, Peoria, for providing the cover picture and also helping me to critically review several chapters in this book.

Houston, TX, USA

Venkatesh Balan

Cover Illustration Caption

Shown in the figure are cells of *Saitoella coloradoensis* strain NRRL YB-2330 containing numerous lipid granules. This strain was discovered to tolerate and produce abundant lipid when cultivated on ammonia fiber expansion (AFEX)-pretreated enzyme saccharified hydrolyzates of corn stover (Slininger et al. 2016; Dien et al. 2016). *Saitoella*, NRRL YB-2330, was isolated by L. J. Wickerham from insect frass collected in 1950 by staff of the U.S. Forest Service from an Engelmann spruce (*Picea engelmannii*) growing in the White River National Forest, Meeker, CO, USA. Taxonomically described by Kurtzman and Robnett (2012), NRRL YB-2330 is recognized as a member of the genus *Saitoella* and as a new species distinct from NRRL Y-17804, type strain of *S. complicata*, the only other *Saitoella* species in collection.

- Kurtzman CP and Robnett CJ (2012) *Saitoella coloradoensis* sp. nov., a new species of the Ascomycota, subphylum Taphrinomycotina. *Antonie van Leeuwenhoek* 101:795–802.
- Slininger PJ, Dien BS, Kurtzman CP, Moser BR, Bakota EL, Thompson SR, O’Bryan PJ, Cotta MA, Balan V, Jin M (2016) Comparative lipid production by oleaginous yeasts in hydrolyzates of lignocellulosic biomass and process strategy for high titers. *Biotechnology and Bioengineering* 113:1676–1690
- Dien BS, Slininger PJ, Kurtzman CP, Moser BR, O’Bryan PJ (2016a) Identification of superior lipid producing *Lipomyces* and *Myxozyma* yeasts. *AIMS Environmental Science*, 3:1–20.

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