Metagenomics is a key technology to the DNA-based exploration of the genomic potential from not-yet-cultivated microbes for ecology and biotechnology. Since the term “metagenome” was coined almost two decades ago, metagenomics has dramatically changed our view on many research areas such as microbial ecology, community biology, and microbiome research, and it has resulted in the rapid identification of many novel biomolecules with potential value to bio-based industrial processes. Function-driven metagenomics has been the focus of many laboratories around the world to quickly encounter novel functional genes encoding enzymes with new and/or improved traits. In this way, the diversity of biocatalysts and other valuable biomolecules useful for downstream applications increased significantly. Industries demand enzymes that can be directly applied in biotechnological processes and catalyze a wide variety of different reactions. Ideally, these biocatalysts/bioactive molecules should be highly active with a broad range of substrates under harsh reaction conditions and, at the same time, should possess a predictable substrate specificity and enantioselectivity. Today, only a limited number of truly well-suited enzymes fulfill these requirements. To identify novel biomolecules different strategies are employed: While the sequence-based detection of novel enzymes and other biomolecules certainly provides rapid access to novel genes and enzymes, it suffers from the fact that only sequences with putative function and similarities to already known genes are recovered. Function-based screens overcome this bottleneck but are limited by low hit rate due to the often poor capabilities of the employed host to express foreign genes and to produce active recombinant proteins. Thus, function-driven detection of novel biocatalysts or other valuable biomolecules is still a very time-consuming process that slows down development times for novel products. However, it has the huge advantage that functional biocatalysts and bioactive compounds are recovered.

In recent years, various novel technologies have been developed to access the metagenomes of microbial communities using high-throughput technologies often in combination with next-generation sequencing approaches. Within the second edition of this book, we provide up-to-date technologies on various function-based technologies currently used in metagenomics. Our goal is that this book serves as a manual for researchers who are interested in establishing metagenomics in their laboratories. All working steps involved are presented in the chapters: Starting from the DNA isolation from soils and marine samples followed by the construction and screening of the libraries for diverse enzymes and biomolecules. The book provides a comprehensive overview of current methods used to isolate DNA and construct large-insert and small-insert libraries from terrestrial and marine habitats, including plant and fungal microbiomes. It further summarizes methods for establishing metagenome libraries in non-*E. coli* hosts such as *Streptomyces*, and it highlights novel molecular tools ready to use for function-driven mining of metagenomic DNA. Lastly, several chapters provide detailed insights into screening protocols for a wide array of different genes encoding enzymes with relevance to biotechnology and ecology. Protocols are offered for the screening of lipases/esterases, cellulases, hydrogenases, ligninolytic enzymes,
glycosyl transferases, and quorum-quenching enzymes involved in the destruction of N-acyl homoserine lactone-based cell–cell communication signals. Furthermore, the book provides detailed screening protocols for phosphatases, poly-hydroxyalkanoate metabolism-related enzymes, stereoselective hydrolases, and microbial signals for the discovery of secondary metabolites. Finally, detailed insights into the pipeline necessary for the reconstruction of metabolic pathways are given.

In our view, this book provides a comprehensive collection of up-to-date protocols for metagenomics and tools for the recovery of many major types of biocatalysts and allows an easy setup of these screens in any microbiology laboratory.

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