DOI: 10.4103/ijmr.IJMR_1433_17

Haplotyping: Methods & protocols, I. Tiemann-Boege, A. Betancourt, editors (Humana Press, Springer Science + Business Media LLC, Switzerland) 2017. 326 pages. Price: Not mentioned.

ISBN 978-1-4939-6748-3

The era of large-scale sequencing has seen enormous growth in haplotype determination and its applications, such as accrediting low-frequency variants and describing the relationship between genetic variation and disease susceptibility. The recent technological advancements have generated enormous amounts of genotype data from genome-wide single-nucleotide polymorphism (SNP) microarrays and from whole-genome and whole-exome sequencing tools. The book has been written with an objective of describing various haplotyping methods based on the next-generation sequencing (NGS) data, and it also describes the use of traditional methods that can be adapted in any laboratory without the use of sophisticated equipment. This book is a part of the 1551th volume of the ongoing series, 'Methods in Molecular Biology', and is divided into seven sections comprising 15 chapters in all.

The opening section of the book 'Haplotyping with long-range PCR' comprises two chapters; the first chapter describes the use of allele-specific long-range PCR as a haplotyping protocol for the determination of the allelic phase of heterozygous sites on chromosomes. The authors mention that the method combines a proofreading polymerase with allele-specific primers that amplify matched templates. The methodology for primer designing and reaction protocol is explained in detail. They have pointed out this as a cost-effective method applicable for haplotyping SNPs separated by about 40kb with the use of real-time PCR. The second chapter describes the methods to measure the crossover frequency in plants at the hotspot scale. The authors have described three different methods in detail: (*i*) the titration methods for amplification, quantification and sequencing of single crossovers molecules; (*ii*) quantitative PCR methods

to measure crossover frequency; and (*iii*) application of high-throughput sequencing to stydy the crossover frequency in plants.

The second section of the book 'Selective sequencing of gene families (including the MHC)' includes four chapters; all the four chapters describe the use of recent technologies for sequencing of gene families. The first chapter describes the use of Pacific Biosciences (PacBio) single-molecule real-time platforms for haplotyping in gene families. The authors cite that PacBio offers two unique advantages to sequence gluten genes; the high throughput and the long reads. They further claim that although PacBio produces a high error rate, the errors are random, and with sufficient coverage, these errors can be cancelled out and high consensus accuracy can be reached. The book describes the applications of peptide nucleic acid probes for the enrichment of high molecular weight DNA in the next chapter. The authors state that this method provides unique advantages over conventional DNA-based enrichments as it increases the thermal stability, strand invasion and limited interferences with downstream PCR amplification. The next chapter outlines the cost-effective microarray-based sequence capture enrichment and NGS approach to characterize major histocompatibility complex (MHC) haplotypes. In this chapter, the use of NimbleGen sequence capture technology on solid surface is described and subsequently Roche/454 Life Sciences sequencing for the MHC genes. The fourth chapter enlightens the pedigree defined haplotypes and their applications to genetic studies with emphasis on MHC haplotypes and complex disease. The authors explain about the conserved extended haplotypes, ancestral haplotypes and genetic fixity, highlighting the MHC genes in this chapter.

The third and fourth parts of the book describe the haploid cell typing and haplotyping single chromosomes, respectively with two chapters in each section. Haplotypes have become useful tools for identifying mutations associated with human diseases and for developing personalized medicines. The first chapter of Part III describes the haplotyping of a non-meiotic diploid fungal pathogen using induced aneuploidies and SNP/comparative genomic hybridization microarray analysis. This chapter also describes the UAU1 transformation strategy to artificially induce whole chromosome trisomy of specific chromosomes. The next chapter about the whole-genome haplotyping of single sperm of *Daphnia pulex* provides experimental workflow for isolating single sperm cells using fluorescence-activated cell sorting and application of whole-genome amplification to a single sperm to generate enough DNA for library preparation for NGS.

Part IV describes a high-throughput and cost-effective experimental protocol to obtain high-resolution chromosomal haplotypes of each individual diploid genome by the single chromosome micro-dissection and sequencing approach. Details on the phased-genome sequencing through chromosome sorting are explained in the second chapter. The detailed protocol for a method capable of generating genomic sequences completely phased across the entire chromosome through Phase-Seq is provided here.

The next section of the book describes various genome-wide haplotyping techniques grouped under three chapters. The first chapter describes the long fragment read (LFR) technology, a DNA pre-processing method for genome-wide haplotyping by whole-genome sequencing. The second chapter describes a comprehensive and detailed protocol for an ultra-fast cost-effective haplotyping method, contiguity preserving transposition sequencing (CPT-Seq) for genome-wide haplotyping, assembly and single cell ATAC-Seq. The next chapter focuses on fosmid pool-based sequencing to haplotype whole genome; the method can be used in combination with or without whole-genome shotgun sequencing to resolve heterozygous SNPs.

In the field of haplotyping, one of the challenging aspects has been the characterization of rare haplotypes, since rare haplotypes need to be singled out from a large pool of wild types found in tissues, blood or other DNA sources. The sixth section describes the use of a high-throughput method to genotype hundreds of thousands of single molecules in parallel using bead-emulsion (BEH) haplotyping which will help in discovery of rare haplotypes by typing millions of single molecules.

The last section of the book is on the computational algorithms for haplotyping. This chapter is focussed more on the basic knowledge and step-by-step usage of a number of tools for haplotype inference on genotyping or NGS data. For each tool, the design philosophy and its targeted applications are also explained in detail.

Overall, this book is a valuable addition to the existing knowledge on haplotyping. It provides the up-to-date haplotyping methodologies with evidence for molecular genetic studies. All the topics have been

covered well and supplemented with flowcharts and tables wherever required. This book is recommended for scientists and students working in the field of molecular and population genetics.

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