EDITORIAL

User guides for biologists to learn computational methods

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System-wide studies of a given molecular type are referred to as “omics.” These include genomics, proteomics, and metabolomics, among others. Recent biotechnological advances allow for high-throughput measurement of cellular components, and thus it becomes possible to take a snapshot of all molecules inside cells, a form of omics study. Advances in computational modeling methods also make it possible to predict cellular mechanisms from the snapshots. These technologies have opened an era of computation-based biology. Component snapshots allow the discovery of gene-phenotype relationships in diseases, microorganisms in the human body, etc. Computational models allow us to predict new outcomes, which are useful in strain design in metabolic engineering and drug discovery from protein-ligand interactions. However, as the quantity of data increases or the model becomes complicated, the process becomes less accessible to biologists. In this special issue, six protocol articles are presented as user guides in the field of computational biology.

Keywords: computational biology, machine learning, microbiome, Ribo-seq, drug discovery

Introduction

Computational methods to gain insight into biological systems, including the evolution of genes and proteins, were first introduced in the 1960s (Ouzounis and Valencia, 2003). Since then, many computational methods have been developed to analyze protein/nucleotide sequences and to predict protein structures. In the era of omics, molecules inside cells (nucleotide sequences, transcripts, proteins) can be captured as a snapshot by high-throughput technologies. Based on the snapshots, computational models can be developed to fill the gaps between snapshots and to predict new outcomes.

Rapid advances in fields associated with computational bio-

logy, such as bioinformatics and systems biology, have allowed deeper insight into the mechanisms of cells, the relationship between cell components and diseases, etc. As the methods have become more sophisticated, they also have become less accessible to biologists. This special issue presents six different articles describing protocols that can be used readily by biologists and bioengineers. The aim of this issue is to introduce step-by-step instructions for scientists who are not familiar with computational approaches.

Microbiome analysis

The human body is colonized by at least 10 times more microorganisms than human cells that comprise it (NIH Human Microbiome Project [HMP] Working Group *et al.*, 2009). This community of microorganisms (known as a microbiome) has been an attractive source of new information in understanding human diseases and the physiology of environmental microbes (Gilbert *et al.*, 2018). Clear differences between the influence of organisms in the microbiome and the influence of genetic variation can be shown in cases such as obesity, autism, and allergies (Gilbert *et al.*, 2018). Recent research has led to novel treatments such as, for example, fecal translation in the case of *Clostridium difficile* infection.

Traditional approaches to analyze microorganism communities include isolating bacterial species by cultivating them on media under controlled conditions. This approach is not always successful in quantifying microorganisms and may overlook many species because of differences in media requirements between species. Recently, next-generation sequencing (NGS) of 16S rRNA has allowed analysis of microorganism communities in the human body or the surrounding environment. To analyze the 16S rRNA sequences, they must be pre-processed, mapped to known microorganism species, and analyzed taxonomically, all of which can be achieved using computer systems. Three articles are included in this special issue: microbiome analysis from 16S rRNA sequences (Kim *et al.*, 2020), analysis of environmental microbiomes (Jo *et al.*, 2020), and machine-learning methods for microbiome analysis (Namkung, 2020). These guides will not only assist in analysis, but also expand understanding of microbiomes.

Jo *et al.* (2020) introduce a step-by-step protocol for the analysis of a microbial community obtained from shotgun metagenomic sequencing by using *in silico* tools. The community may be sampled from soil, deep water, the human body, or other microbiomes of interest. The article includes

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pre-processing of metagenomic datasets, taxonomy profiling, and functional annotation of sequenced genes to infer the function and physiology of the community.

Kim *et al.* (2020) introduce a protocol for NGS-based microbiome studies by analyzing the 16S rRNA sequences of a microbial community. The article includes pre-process sequencing of raw data, diversity analysis, and taxonomic classification. This protocol article uses a QIIME2 core installed on a virtual machine, which is relatively easy to follow for those who are not bioinformaticians.

Namkung (2020) introduces fundamental knowledge on machine-learning algorithms and clustering/dimensionality reduction methods for the development of risk-prediction models and for analysis of microbiome profiles, respectively. Python is used to build a code to cluster microbiome profiles or to predict risk probability from the profiles. Specifically, the sklearn library is used for clustering, principle component analysis, and support vector machine.

Ribo-Seq analysis

NGS has influenced methods of conducting experiments and understanding underlying cellular mechanisms. NGS has been utilized to discover genomic sequences, to analyze the relationship between genetic variation and disease phenotype, to reconstruct metabolic pathways displaying physiological behaviors, to identify microorganisms living in the gut, etc.

In general, RNA-seq has been used widely to measure the abundance of transcripts in a cell (known as a transcriptome) (Nagalakshmi *et al.*, 2008). A transcriptome provides more information than genomic data because transcript abundance is correlated with the activity of working genes. With a transcriptome, we can find which genes are working actively and determine how active they are. Despite this usefulness, transcript abundance may not be correlated directly with protein levels. Proteins perform a wide range of functions and define the status of cells. Therefore, in order to understand cellular systems and behaviors accurately, the entire complement of proteins in a cell (known as a proteome) should be measured. At present, it is still difficult to measure a whole proteome.

Ribo-seq has been developed to measure the abundance of transcripts that are translated actively (known as a translatome), which is correlated directly with protein levels, by footprinting ribosome-bound transcripts (Ingolia *et al.*, 2009). In the early days, Ribo-Seq was used to study translation initiation, codon adaptation, translation elongation, translational control, etc. These days, Ribo-Seq is used to assess the levels of proteins.

Choe *et al.* (2020) introduce a step-by-step guide for Ribo-Seq analysis for those who are not bioinformaticians using a Simple Translatome Analysis Tool for Ribo-Seq (STATR). The article discusses how to conduct Ribo-Seq experiments and process raw data, as well as genome-wide differential expression analysis. The authors analyze the actively transcribing genes of the multi-drug resistance pathogen, *Staphylococcus aureus*, and find that the accessory gene regulator (Agr), a major regulator of virulence and pathogenesis, is translated in response to linezolid.

Metabolic engineering

Engineering microorganisms to construct microbial cell factories has gained attention as a sustainable technology alternative to current chemical factories. According to recent reports, refinery chemicals, energies and fuels, plastics, cosmetics, and even drugs are produced from microorganisms (Lee *et al.*, 2012). However, metabolic networks and their genetic regulatory systems are wired in a highly complicated way and therefore it is difficult to intuitively predict the outcomes of genetic engineering of microorganisms. To understand cellular networks, omics data have been measured (genome, transcriptome, metabolome, proteome, etc.) and analyzed to depict a big picture of living organisms. Recently, computational methods have been developed to predict the outcome of gene modifications (Kabimoldayev *et al.*, 2018; Gu *et al.*, 2019).

Jeon *et al.* (2020) introduce a method to simulate a specific growth rate and theoretical production yield of succinic acid as an example by using COBRapy. Simulation and prediction of metabolic systems in microorganisms would stimulate the construction of microbial cell factories efficiently.

Drug discovery

Classical drug discovery strategy for a single drug requires 10–20 years to develop and the process costs approximately \$1.8 billion (Paul *et al.*, 2010). In the drug discovery process, hit discovery, lead optimization, and preclinical tests require approximately 5–10 years (Marx *et al.*, 2016). In preclinical tests, many drug candidates are withdrawn due to unexpected cardio- or hepato-toxicity. Fortunately, recent rapid advances in computer science allow for the development of prediction models for virtual screening. In addition, computational models of protein-drug interactions and machine-learning models for absorption, distribution, metabolism, excretion, and toxicity (ADME-Tox) have been developed using deep learning algorithms.

Shaker *et al.* (2020) introduce a user guide for *in silico* drug discovery that includes protein structure prediction, active site prediction, protein-drug interaction (docking), and ADME-Tox prediction, a virtual strategy for drug discovery. This might encourage biologists, even those who are not familiar with computer science, to carry out research on drug discovery.

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