Gene expression

gep2pep: a bioconductor package for the creation and analysis of pathway-based expression profiles

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Abstract

Summary: Pathway-based expression profiles allow for high-level interpretation of transcriptomic data and systematic comparison of dysregulated cellular programs. We have previously demonstrated the efficacy of pathway-based approaches with two different applications: the drug set enrichment analysis and the Gene2drug analysis. Here, we present a software tool that allows to easily convert gene-based profiles to pathway-based profiles and analyze them within the popular R framework. We also provide pre-computed profiles derived from the original Connectivity Map and its next generation release, i.e. the LINCS database.

Availability and implementation: The tool is implemented as the R/Bioconductor package gep2pep and can be freely downloaded from https://bioconductor.org/packages/gep2pep.

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Supplementary information: Supplementary data are available at Bioinformatics online.

1 Introduction

The use of genome-wide expression profiling technologies has transformed the way in which scientists approach the study of molecular mechanisms. Millions of assays have been performed and public databases have been developed to collect such immense amount of data, thus improving both reproducibility of the original studies and reusability for novel investigations (Rung and Brazma, 2013). Many different computational approaches have been developed to deal with the inherent complexity of transcriptomic data and to help mining biological knowledge out of them. The gene ontology (GO) (Ashburner et al., 2000) has been one of the most popular tools to provide systematic insights into the activity of transcriptional programs by factoring in the expression of multiple genes together through the gene set enrichment analysis (GSEA).

GSEA is commonly used to aid biological interpretation downstream of transcriptomic data analyses. However, we recently proposed a different approach, in which GSEA is rather part of the data preprocessing phase. In particular, we use GSEA to convert gene expression profiles (GEPs) to pathway expression profiles (PEPs). This allows to develop analytic approaches that use dysregulated gene sets (although we refer to them as pathways for simplicity) as their elementary variables, as opposed to single genes. We demonstrated the efficacy of the approach with two different tools: (i) the drug set enrichment analysis (DSEA) (Napolitano et al., 2016), which allows to identify pathways that are consistently dysregulated across a set of drugs, and (ii) the Gene2drug analysis (Napolitano et al., 2018), which allows to perform gene-drug prioritization based on the pathways that the molecular target of interest is involved in. Both tools have been previously released as closed source web applications.

Here, we present gep2pep, an R/Bioconductor package that implements the pathway-based expression profiles paradigm. It supports conversion of large collections of GEPs to PEPs and provides routines to perform DSEA-like and Gene2drug-like analyses. Together with the package, we provide two large collections of PEPs, respectively derived from the Connectivity Map 2.0 (Cmap) (Lamb et al., 2006) and its next generation released within the LINCS project (Subramanian et al., 2017). Finally, we present an update of the DSEA tool which takes advantage of these new data (see Supplementary Material).

2 Materials and methods

2.1 Implementation

The gep2pep R/Bioconductor package supports the management of large collections of heterogeneous profiles, exploiting the HDF5 format in combination with the Repo (Napolitano, 2017) package for objects management. It also supports import of gene set collections from the MSigDB database (Liberzon et al., 2011). Large datasets are handled through parallelization and partial results management support.
2.2 Converting GEPs to PEPs
PEPs are created from GEPs using GSEA. Given N GEPs related to a set of experimental conditions, and a database of M pathways such as those included in the GO, a GSEA is performed for each \((c, p)\) pair, where \(c\) is a condition and \(p\) is a pathway. Therefore, each \((c, p)\) pair is assigned an Enrichment Score (ES) and its corresponding \(P\)-value according to the Kolmogorov–Smirnov test (KST), giving rise to an \(M \times N\) matrix \(E\) of ESs and an \(M \times N\) matrix \(P\) of \(P\)-values. \textit{gep2pep} allows to perform further PEP-based analyses using any of \(E, P\) or their element-wise product, according to: \(-log(P)\). \textit{gep2pep} also implements merging of multiple PEPs into a single PEP, along the lines of Iorio \textit{et al.} (2010) for GEPs. Given a collection of PEPs \((E, P)\), the resulting merge is the row-wise average of \(E\), and the row-and-aggregation of \(P\) by the Fisher’s method.

2.3 Analyzing PEPs
The \textit{gep2pep} package supports two kinds of pathway-based analyses (see Fig. 1): condition set enrichment analysis (\textit{CondSEA}) and pathway set enrichment analysis (\textit{PathSEA}). Both are based on a GSEA-like procedure performed on PEPs as opposed to GEPs. CondSEA first ranks the PEPs row-wise. Then, given a set of conditions as input, it computes their KST within each row. Enriched rows correspond to pathways that are consistently dysregulated across the input conditions. When conditions are drug-induced GEPs, we call this approach \textit{DSEA} (Napolitano \textit{et al.}, 2016). Conversely, PathSEA first ranks the PEPs column-wise. Then, given a set of pathways as input, it computes their KST against each column. Enriched columns correspond to conditions in which most of the input pathways appear consistently dysregulated. When conditions are drug-induced GEPs and the input pathways are related to a pharmacological target, we call this approach \textit{Gene2drug} (Napolitano \textit{et al.}, 2018).

Overall, we computed \((1309 + 17 974) \times 14 645 \approx 350 \text{,}000 \text{,}000\) of ES—\(P\)-value pairs using a computer cluster. We have used these data to build a new version of the DSEA website (http://dsea.tigem.it/lincs), where the full dataset in the \textit{gep2pep} format can be obtained (see Supplementary Material).

3 Conclusion
We introduced \textit{gep2pep}, an R/Bioconductor package implementing a pathway-based approach to the analysis of GEPs. We also provided a large collection of pre-computed PEPs. We hope that an offline, structured and carefully documented tool for generic pathway-based approaches will allow more researchers to develop original applications under this new paradigm.

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References


