Bioinformatics Solutions Using R and Bioconductor

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•C and Perl were the languages of choice for the first generation of bioinformaticians: massive amounts of sequence data

•R is well suited to scientific challenges in **functional genomics** which employs technologies to measure abundances of biomolecules under different conditions

•Since 2001 ***Bioconductor*** project

Packages for analyzing high-dimensional (time series) data

obtained from high-throughput **functional genomics** assays:

e.g. expression microarrays, metabolic profiling:

- •Identifying Interesting Genes with **siggenes**
- •R package **limma**: Linear Models for Microarray Data
- •Reverse Engineering Genetic Networks using **GeneNet**
- •Package **GAGE**: Generally Applicable Gene Set Enrichment

Identifying Interesting Genes with **siggenes**

•Common task in microarray experiments: identification of genes whose expression values differ substantially between groups or conditions - **differentially expressed genes** (**degs**)
• **multiple testing** problems in which thousands of hypotheses are tested simultaneously
•Testing statistic and corresponding p-value are computed for each gene
•Raw p-values are adjusted for multiple testing:
➢**Significance Analysis of Microarrays** (**SAM**)
observed test statistics are plotted against expected under null hypotheses; points that differ from diagonal correspond to degs
➢**Empirical Bayes Analysis of Microarrays** (**EBAM**)
Models the distribution of observed test statistics as mixture of two components: one for degs and the other for not

R package **limma**: Linear Models for Microarray Data

➢A number of summary statistics are computed
for each gene and each contrast:
moderated t-statistic: ratio of log2-fold change
to its standard error
Standard errors have been moderated across genes
p-value:
obtained from *moderated t-statistic* after adjustment
for multiple testing using Benjamini and Hochberg's method
to control **False Discovery Rate** (**FDR**)

B-statistic: log-odds that the gene is differentially expressed
B=0 corresponds to 50/50 chance
➢ *moderated F-statistic*
for each gene combines the *t-statistics* for all contrasts
tests whether any of the contrasts are non-zero

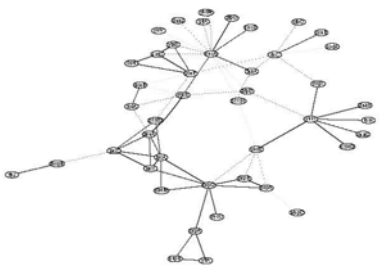
R Package **GAGE**:

Generally Applicable Gene Set Enrichment

•**Gene Set Analysis** (**GSA**) focuses on sets of related genes
•Incorporates prior knowledge of biological pathways in form of gene sets
•Small coordinated gene expression changes in a pathway can have major biological effect even if these changes are not statistically significant for any individual gene
•Uses all data instead of prefiltering for a short list of strongly differentially expressed genes
•**GAGE** can handle multiple microarray datasets with different sample sizes, exprimental designs, profiling techniques

Reverse Engineering Genetic Networks using **GeneNet**

•output - graph where each gene corresponds to a node and edges depict dependencies between nodes
• based on **Graphical Gaussian Model** (**GGM**) which represent multivariate dependencies by means of *partial correlation*
•model selection - assigning statistical significance to edges using local False Discovery Rate
•exploratory approach that may help to identify „**hubs**“ or **clusters of genes** that are functionally related or co-regulated



Bioconductor is more than just a repository of biology-related packages!!!

The project has driven a number of technological innovations:

•Management of package dependence hierarchies

•Interfaces between R and other software systems

•Biological metadata (e.g. genome annotations) packages

•More structured data formats than basic data types of R

•Common data structures that allow efficient exchange of data and computational results between different packages (Example is *ExpressionSet* - a class for storage data and info on a microarray experiment)