oligene
PROVITRO - cells and more

oligoExpress – exploiting probe level information in Affymetrix GeneChip expression data

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useR! – The R User Conference 2006

Three ways to analyse GeneChip data

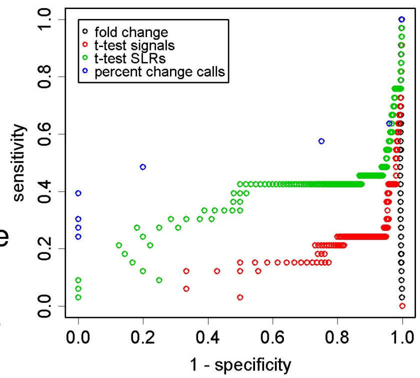
- **Absolute analysis (1 chip):**
  Signals, detection p-values, detection calls
- **Comparison analysis (1 chip vs 1 chip):**
  SLRs, change p-values, change calls
- **Group analysis (m chips vs n chips):**
  Statistics on absolute and comparison analysis results
  e.g.: t-test on signals, t-test on SLRs,
  percent of increase or decrease calls

Toy data: 6 hybridizations from the Affymetrix Latin Square Experiment (HG-U133A)

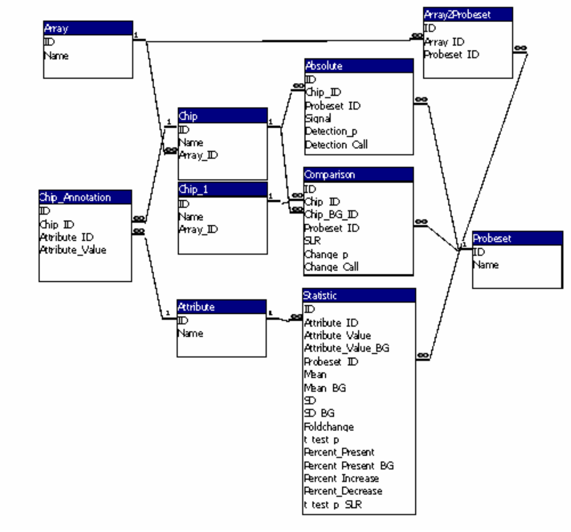
- Comparison of EXP1 (3 replicates) versus EXP2 (3 replicates)
- 30 spikes were mixed into the background RNA at 14 concentrations (0, 0.125, 0.25, …, 512 pmol)
- Concentrations of spikes differ between EXP1 and EXP2 (fold change = 2)
- SLRs and change calls were calculated between each pair of chips from EXP1 and EXP2 (9 comparisons)

Toy data: detection of the spiked transcripts

- Selection of candidates by thresholds on
  - fold change
  - t-statistics signals
  - t-statistics SLRs
  - percent of change calls indicating in- or decrease
- Count of the number of true and false positives



oligoExpress - database scheme



Data processing

- **Data sources:**
  - Expressions profiles: CEL files
  - Chip annotations: Excel sheet
- **Methods für absolute analysis:**
  - Available in library(affy) (Bioconductor project)
  - Functions mas5() and mas5calls() yield signals and detection p-values, respectively
- **Methods for comparison analysis:**
  - To my knowledge: not available from Bioconductor or other open source projects
  - cf. Affymetrix: *Statistical algorithms description document*
  - Own implementation (R code with integrated C functions)

Data annotation

- **Sample annotation:**
  - Entity-attribute-value (EAV) system
  - Sample names (CEL file names) → row names
  - Attributes names → column names
  - Matrix entries assign values of attributes to samples

- **Probe annotation:**
  - Mapping of probes to genes
  - Annotation of genes
    e.g. cytoband, function (gene ontology), pathway (KEGG), references (PubMed)

Data upload und retrieval

- ODBC is a generic interface to relational databases
- ODBC is supported by MS Access, PostgreSQL, MySQL, Oracle, …
- The library RODBC implements the ODBC database connectivity under R

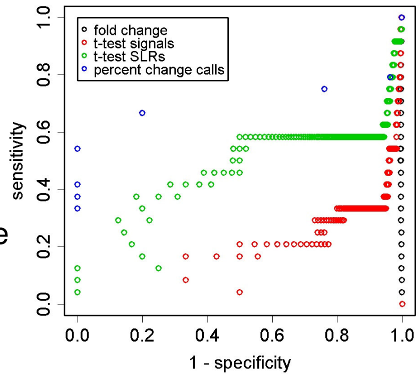
➢ **RODBC allows an easy and generic database management including definition of tables, data upload and data retrieval**

oligoExpress - conclusion

- **Concise mangement of all information from Affymetrix absolute and comparison analysis**
- **Flexible sample annotation by an EAV system, analysis of the corresponding biological groups**
- **Compatibility to all common database systems by usage of the RODBC interface**

Toy data: detection of the spiked transcripts **spikes with concentrations ≥ 1 pmol**

- Selection of candidates by thresholds on
  - fold change
  - t-statistics signals
  - t-statistics SLRs
  - percent of change calls indicating in- or decrease
- Count of the number of true and false positives


○ fold change
○ t-test signals
○ t-test SLRs
○ percent change calls
sensitivity
1 - specificity