

Presenting the Results of an Analysis - annotating data and creating useful output for one-color arrays

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Scenario

Bioconductor

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We assume that

- We are working with some sort of one-color arrays
 - Affymetrix
 - Nimblegen
 - Illumina
- Some of this is applicable to two-color arrays as well
- We want to map some identifier to gene information

Annotation Packages

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- Chip - level packages
 - hgu133plus2.db
 - illuminaHumanv3ProbelD
- Species - level packages
 - org.Hs.eg.db
 - org.Mm.eg.db
- biomaRt
- GO.db
- KEGG.db

Chip - Level Data

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What mappings can we do?

```
> ls("package:hgfocus.db")
```

```
[1] "hgfocus"  
[2] "hgfocusACCNUM"  
[3] "hgfocusALIAS2PROBE"  
[4] "hgfocusCHR"  
[5] "hgfocusCHRENGTHS"  
[6] "hgfocusCHRLOC"  
[7] "hgfocusENSEMBL"  
[8] "hgfocusENSEMBL2PROBE"  
[9] "hgfocusENTREZID"  
[10] "hgfocusENZYME"
```

Annotation Table Contents

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What is in a given table?

> *?hgfocusUNIGENE*

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What is in a given table?

```
> ?hgfocusUNIGENE
```

```
> head(toTable(hgfocusUNIGENE))
```

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What is in a given table?

```
> ?hgfocusUNIGENE
```

```
> head(toTable(hgfocusUNIGENE))
```

	probe_id	unigene_id
1	1007_s_at	Hs.631988
2	1053_at	Hs.647062
3	117_at	Hs.654614
4	121_at	Hs.469728
5	1255_g_at	Hs.92858
6	1294_at	Hs.16695

Gene Names

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Gene name for one illumina probe:

```
> get("1010243", illuminaHumanv3ProbeIDGENENAME)
```


Gene Names

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Gene name for one illumina probe:

```
> get("1010243", illuminaHumanv3ProbeIDGENENAME)
```

```
[1] "lipoprotein, Lp(a)-like 2"
```

Gene Names

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Gene name for one illumina probe:

```
> get("1010243", illuminaHumanv3ProbeIDGENENAME)
[1] "lipoprotein, Lp(a)-like 2"
```

And for multiple probes:

```
> illumina <- c("10008", "10010", "10017", "10019")
> mget(illumina, illuminaHumanv3ProbeIDGENENAME)
```

Gene Names

Bioconductor

Gene name for one illumina probe:

```
> get("1010243", illuminaHumanv3ProbeIDGENENAME)
```

```
[1] "lipoprotein, Lp(a)-like 2"
```

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And for multiple probes:

```
> illumina <- c("10008", "10010", "10017", "10019")
```

```
> mget(illumina, illuminaHumanv3ProbeIDGENENAME)
```

```
$`10008`
```

```
[1] "arginine-glutamic acid dipeptide (RE) repeats"
```

```
$`10010`
```

```
[1] NA
```

```
$`10017`
```

```
[1] NA
```

```
$`10019`
```

Reverse Mapping

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Say we want to know how many probesets
on the hgfocus chip map to the 'caffeine metabolism' pathway

```
> library(KEGG)  
> get("Caffeine metabolism", revmap(KEGGPATHID2NAME))
```

Reverse Mapping

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Say we want to know how many probesets
on the hgfocus chip map to the 'caffeine metabolism' pathway

```
> library(KEGG)
> get("Caffeine metabolism", revmap(KEGGPATHID2NAME))

[1] "00232"
```

Reverse Mapping

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Say we want to know how many probesets
on the hgfocus chip map to the 'caffeine metabolism' pathway

```
> library(KEGG)
> get("Caffeine metabolism", revmap(KEGGPATHID2NAME))
[1] "00232"
> get("00232", revmap(hgfocusPATH))
```

Reverse Mapping

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Say we want to know how many probesets
on the hgfocus chip map to the 'caffeine metabolism' pathway

```
> library(KEGG)
> get("Caffeine metabolism", revmap(KEGGPATHID2NAME))
[1] "00232"
> get("00232", revmap(hgfocusPATH))
[1] "214440_at"    "206797_at"    "207609_s_at"
[4] "1494_f_at"    "207718_x_at"  "208327_at"
[7] "210301_at"
```

Practice

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What is the gene symbol for Entrez Gene ID '1234'?

The gene name?

What Entrez Gene IDs map to the 'Caffeine metabolism'
pathway?

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What is the gene symbol for Entrez Gene ID '1234'?

```
> get("1234", org.Hs.egSYMBOL)
```

```
[1] "CCR5"
```

The gene name?

What Entrez Gene IDs map to the 'Caffeine metabolism' pathway?

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What is the gene symbol for Entrez Gene ID '1234'?

```
> get("1234", org.Hs.egSYMBOL)
```

```
[1] "CCR5"
```

The gene name?

```
> get("1234", org.Hs.egGENENAME)
```

```
[1] "chemokine (C-C motif) receptor 5"
```

What Entrez Gene IDs map to the 'Caffeine metabolism' pathway?

Practice

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What is the gene symbol for Entrez Gene ID '1234'?

```
> get("1234", org.Hs.egSYMBOL)
```

```
[1] "CCR5"
```

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The gene name?

```
> get("1234", org.Hs.egGENENAME)
```

```
[1] "chemokine (C-C motif) receptor 5"
```

What Entrez Gene IDs map to the 'Caffeine metabolism' pathway?

```
> caff <- get("Caffeine metabolism",  
+           revmap(KEGGPATHID2NAME))  
> get(caff, revmap(org.Hs.egPATH))
```

```
[1] "9"      "10"     "1544"  "1548"  "1549"  "1553"
```

```
[7] "7498"
```

What is biomaRt?

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- Package for annotating using Biomart
- Multiple annotation sources
- Use webservice (Rcurl) or database (RMySQL)
- Species level
- Some manufacturer IDs
- Much greater amount of information

Basic Usage

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Let's load the package and see what annotation choices we have

```
> library(biomaRt)
> listMarts()
```

Basic Usage

Bioconductor

Let's load the package and see what annotation choices we have

```
> library(biomaRt)
```

```
> listMarts()
```

```
[1] ENSEMBL 49 GENES (SANGER)
```

```
[2] ENSEMBL 49 HOMOLOGY (SANGER)
```

```
[3] ENSEMBL 49 PAIRWISE ALIGNMENTS (SANGER)
```

```
[4] ENSEMBL 49 MULTIPLE ALIGNMENTS (SANGER)
```

```
[5] ENSEMBL 49 VARIATION (SANGER)
```

```
[6] ENSEMBL 49 GENOMIC FEATURES (SANGER)
```

```
[7] VEGA 30 (SANGER)
```

```
[8] MSD PROTOTYPE (EBI)
```

```
[9] UNIPROT PROTOTYPE (EBI)
```

```
[10] HIGH THROUGHPUT GENE TARGETING AND TRAPPING (SANGER)
```

```
[11] GRAMENE (CSHL)
```

```
[12] REACTOME (CSHL)
```

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Ensembl

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Available Datasets

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We will use Ensembl annotations

```
> mart <- useMart("ensembl")
```

What datasets are available?

```
> listDatasets(mart)
```


Available Datasets

Bioconductor

We will use Ensembl annotations

```
> mart <- useMart("ensembl")
```

Introduction

What datasets are available?

Background

```
> listDatasets(mart)
```

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```
[1] "oanatinus_gene_ensembl"  
[2] "gaculeatus_gene_ensembl"  
[3] "cporcellus_gene_ensembl"  
[4] "lafricana_gene_ensembl"  
[5] "stridecemlineatus_gene_ensembl"  
[6] "scerevisiae_gene_ensembl"  
[7] "eeuropaeus_gene_ensembl"  
[8] "etelfairi_gene_ensembl"  
[9] "ptroglodytes_gene_ensembl"  
[10] "cintestinalis_gene_ensembl"  
[11] "ppygmaeus_gene_ensembl"  
[12] "ocuniculus_gene_ensembl"
```

Homo sapiens

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We use the Homo sapiens dataset

```
> mart <- useMart("ensembl", "hsapiens_gene_ensembl")
```

Checking attributes and filters ... ok

Now some terminology

- attribute - thing(s) we want to get
- filter - input data type
- value - input values

Available attributes

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```
> listAttributes(mart)
```

	name	description
1	affy_hcg110	AFFY HCG110
2	affy_hg_focus	AFFY HG FOCUS
3	affy_hg_u133_plus_2	AFFY HG U133-PLUS-2
4	affy_hg_u133a	AFFY HG U133A
5	affy_hg_u133a_v2	AFFY HG U133A 2
6	affy_hg_u133b	AFFY HG U133B

Available filters

Bioconductor

```
> listFilters(mart)
```

```
                name
1          affy_hc_g110
2          affy_hc_g110-2
3          affy_hg_focus
4          affy_hg_focus-2
5  affy_hg_u133_plus_2
6  affy_hg_u133_plus_2-2
```

```
                description
1      Affy hc g 110 ID(s)
2      Affy hc g 110 ID(s)
3      Affy hg focus ID(s)
4      Affy hg focus ID(s)
5  Affy hg u133 plus 2 ID(s)
6  Affy hg u133 plus 2 ID(s)
```

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Simple query

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We want the Entrez Gene ID for Affy ID 1007_s_at
from HG-U133A chip

```
> getBM(attributes = c("affy_hg_u133a", "entrezgene")
+       filters = "affy_hg_u133a",
+       values = "1007_s_at",
+       mart = mart)
```

Simple query

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We want the Entrez Gene ID for Affy ID 1007_s_at from HG-U133A chip

```
> getBM(attributes = c("affy_hg_u133a", "entrezgene")
+       filters = "affy_hg_u133a",
+       values = "1007_s_at",
+       mart = mart)
```

```
affy_hg_u133a  entrezgene
1      1007_s_at      780
```

Practice

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What is the gene name for Ensembl ID ENSG00000112715?
What chromosome is it located on?
What are the start and stop coordinates?

Practice

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What is the gene name for Ensembl ID ENSG00000112715?

```
> getBM(c("ensembl_gene_id", "description"),  
+       "ensembl_gene_id", "ENSG00000112715",  
+       mart)
```

```
ensembl_gene_id  
1 ENSG00000112715
```

1 Vascular endothelial growth factor A precursor (VEGF)

What chromosome is it located on?

What are the start and stop coordinates?

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What is the gene name for Ensembl ID ENSG00000112715?
What chromosome is it located on?

```
> getBM(c("ensembl_gene_id", "chromosome_name"),  
+       "ensembl_gene_id", "ENSG00000112715",  
+       mart)
```

```
ensembl_gene_id chromosome_name  
1 ENSG00000112715                6
```

What are the start and stop coordinates?

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What is the gene name for Ensembl ID ENSG00000112715?

What chromosome is it located on?

What are the start and stop coordinates?

```
> getBM(c("sequence_gene_chrom_start",  
+        "sequence_gene_chrom_end",  
+        "ensembl_gene_id"),  
+      "ensembl_gene_id", "ENSG00000112715",  
+      mart)
```

```
  gene_chrom_start gene_chrom_end  
1           43845926           43862202  
  ensembl_gene_id  
1 ENSG00000112715
```

Bulk Annotating

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Scenario

- Data have been analyzed
- We want to present results to layperson
- Content-rich
- Easy to use
- Choices:
 - HTML
 - Text

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- `annaffy`
- `biomaRt` and `annotate`
- `affycoretools`

annaffy

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- HTML tables
- text tables
- `aaf.handler()`

```
> aaf.handler()
[1] "Probe"
[2] "Symbol"
[3] "Description"
[4] "Chromosome"
[5] "Chromosome Location"
[6] "GenBank"
[7] "Gene"
[8] "Cytoband"
[9] "UniGene"
[10] "PubMed"
[11] "Gene Ontology"
[12] "Pathway"
```

affycoretools and annaffy

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First a bit of setup...

```
> library(affycoretools)
> load(paste(.path.package("affycoretools"),
+           "/doc/exprSet.Rdata", sep=""))
> prbs <- featureNames(eset)[500:550]
```

Create tables

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```
> probes2table(eset = eset, probids = prbs,  
+             lib = "hgfocus.db", text = TRUE,  
+             filename = "test")
```

affycoretools and biomaRt

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Useful when data to be annotated does not have a chip-level annotation package.

```
> probes2tableBM(eset = eset, probids = prbs[1:10],  
+               species = "hsapiens",  
+               ann.source = "affy_hg_focus",  
+               filename = "test2", affyid = TRUE,  
+               mysql = FALSE)
```