

GS01 0163

Analysis of Microarray Data

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Lecture 10: Exploring BioConductor

- How do we load CEL files into an AffyBatch? how can we merge batches? how can we partition batches?
- How do we check that it worked?
- How do we supply the associated phenoData?
- Given an AffyBatch, how do we look at it? boxplot, hist, ma-plots, ratio plots, PLM
- Given an AffyBatch, how do we fit it? expresso, justRMA
- Given an eset, what can we say about its contents?
- How can we get the probe level values for a probeset?

- How can we figure out what probeset corresponds to a given gene?
- How can we get the probe sequences for a probeset?

Loading CEL files into an AffyBatch

One theme for today is TMTOWTDI.

We're going to try to extend this from Perl over to BioConductor, and to the analysis of Affy data.

To begin with, let's say that we've got a set of CEL files. How do we pull them in?

There are several options. How do I survey them?

Loading CEL files into an AffyBatch

One theme for today is TMTOWTDI.

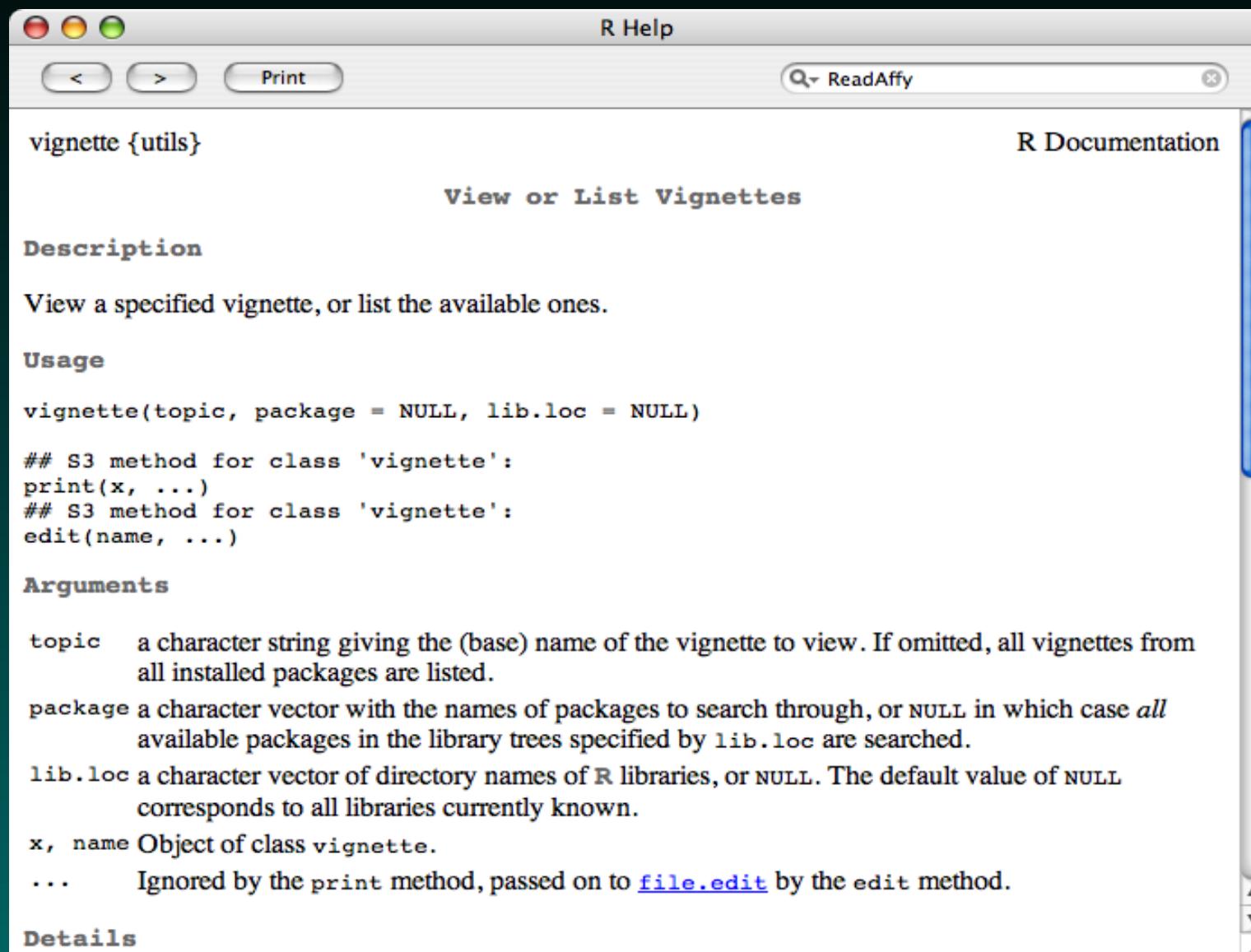
We're going to try to extend this from Perl over to BioConductor, and to the analysis of Affy data.

To begin with, let's say that we've got a set of CEL files. How do we pull them in?

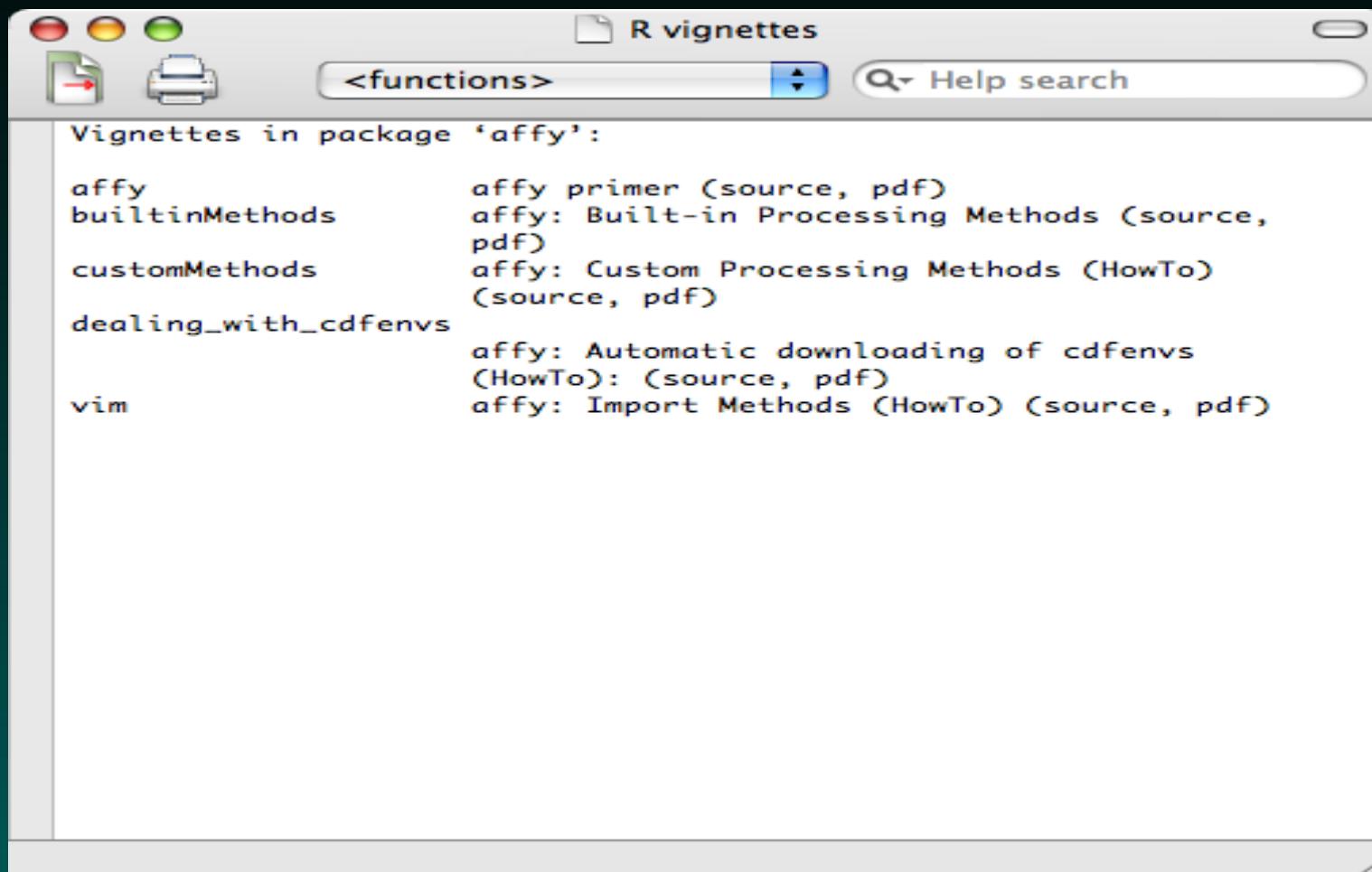
There are several options. How do I survey them?

```
> library(affy)  
> vignette("affy")
```

Reading the Fine Manual: Vignettes

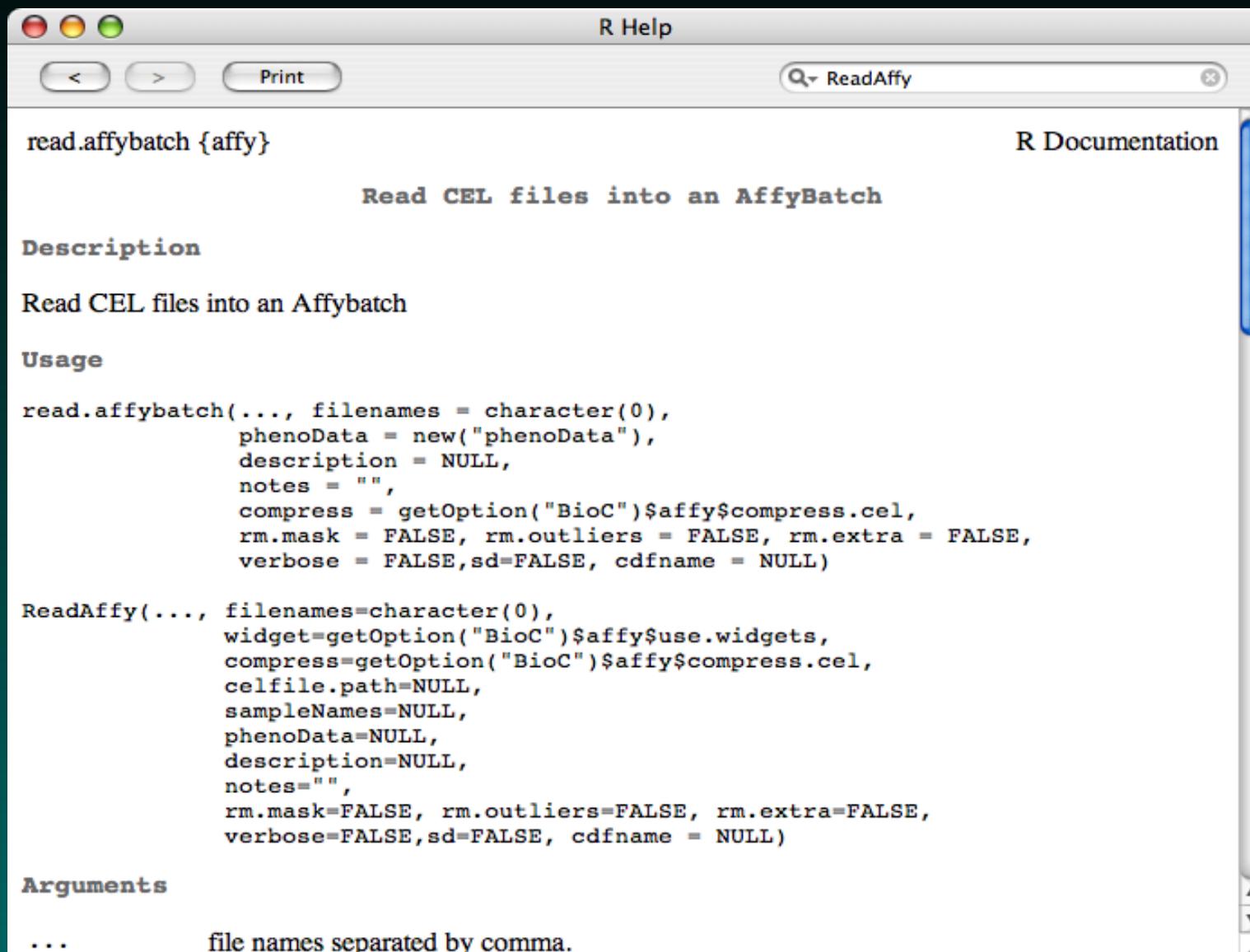


Listing Vignettes



```
> vignette(package = "affy");
```

ReadAffy: Help from Top



ReadAffy: ... to Bottom

The screenshot shows the R Help window for the 'ReadAffy' function. The title bar says 'R Help' and the search bar contains 'ReadAffy'. The main content area displays the documentation for 'ReadAffy'.

Description: obtain a MIAME instance. If left NULL but widget=TRUE then widgets are used. If left NULL and widget=FALSE then an empty instance of MIAME is created..

Value:
An AffyBatch object.

Author(s):
Ben Bolstad bmb@bmbolstad.com (read.affybatch), Laurent Gautier, and Rafael A. Irizarry (ReadAffy)

See Also:
[AffyBatch](#)

Examples:

```
if(require(affydata)){
  celpath <- paste(.path.package("affydata"), "celfiles", sep="/")
  fns <- list.celfiles(path=celpath, full.names=TRUE)

  cat("Reading files:\n", paste(fns, collapse="\n"), "\n")
  ##read a binary celfile
  abatch <- ReadAffy(filenames=fns[1])
  ##read a text celfile
  abatch <- ReadAffy(filenames=fns[2])
  ##read all files in that dir
  abatch <- ReadAffy(celfile.path=celpath)
}
```

[Package *affy* version 1.10.0 [Index](#)]

The Affy Index

R Help

Print Help Search

Methods for Affymetrix Oligonucleotide Arrays

Documentation for package `affy' version 1.10.0

User Guides and Package Vignettes

Read [overview](#) or browse [directory](#).

Help Pages

[A](#) [B](#) [C](#) [D](#) [E](#) [F](#) [G](#) [H](#) [I](#) [J](#) [L](#) [M](#) [N](#) [O](#) [P](#) [Q](#) [R](#) [S](#) [T](#) [U](#) [W](#) [X](#) [misc](#)

[S.AffyBatch](#) Class AffyBatch

--- A ---

<u>affy-options</u>	Options for the affy package
<u>affy.scalevalue.exprSet</u>	Scale normalization for exprSets
<u>AffyBatch</u>	Class AffyBatch
<u>AffyBatch-class</u>	Class AffyBatch
<u>affybatch.example</u>	AffyBatch instance affybatch.example
<u>affybatch.example2</u>	AffyBatch instance affybatch.example

Reading a list of files

The file subcels.txt contains 20 lines like this:

```
G:/Public/Singh-Prostate-Affymetrix/CelFiles/N01__normal.cel  
G:/Public/Singh-Prostate-Affymetrix/CelFiles/N58__normal.cel  
G:/Public/Singh-Prostate-Affymetrix/CelFiles/N59__normal.cel  
G:/Public/Singh-Prostate-Affymetrix/CelFiles/N61__normal.cel  
G:/Public/Singh-Prostate-Affymetrix/CelFiles/N62__normal.cel  
...
```

Reading a list of files

```
> basedir <- file.path("G:", "Public", "Singh-Prostate-Affym  
> celList <- read.table(file.path(basedir, "subcels.txt"))  
> celList[1:6, ]
```

```
[1] G:/Public/Singh-Prostate-Affymetrix/CelFiles/N01__normal.  
[2] G:/Public/Singh-Prostate-Affymetrix/CelFiles/N58__normal.  
[3] G:/Public/Singh-Prostate-Affymetrix/CelFiles/N59__normal.  
[4] G:/Public/Singh-Prostate-Affymetrix/CelFiles/N61__normal.  
[5] G:/Public/Singh-Prostate-Affymetrix/CelFiles/N62__normal.  
[6] G:/Public/Singh-Prostate-Affymetrix/CelFiles/N02__normal.  
20 Levels: G:/Public/Singh-Prostate-Affymetrix/CelFiles/N01__:
```

Reading the CEL files

```
> ABatch <- ReadAffy(cellList)
```

Error : file names must be specified using a
charactervector, not a 'list'

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oops...

The Evolution...

```
> cellList <- as.character(cellList$V1)
```

```
> ABatch <- ReadAffy(cellList)
```

Error : file names must be specified using a
charactervector, not a 'list'

The Evolution...

```
> cellList <- as.character(cellList$V1)
```

```
> ABatch <- ReadAffy(cellList)
```

Error : file names must be specified using a
charactervector, not a 'list'

oops...

```
> ABatch <- ReadAffy(filenames = cellList)
```

Ta Da!

Checking the Contents

```
> slotNames(ABatch)
```

```
[1] "cdfName"           "nrow"  
[3] "ncol"              "assayData"  
[5] "phenoData"         "featureData"  
[7] "experimentData"    "annotation"  
[9] ".__classVersion__"
```

```
> phenoData(ABatch)
```

```
rowNames: N01_normal.cel, N58_normal.cel, ..., T49_tumor  
varLabels and varMetadata:  
sample: arbitrary numbering
```

Looking at phenoData

```
> class(phenoData(ABatch))
```

```
[1] "AnnotatedDataFrame"
```

```
attr(,"package")
```

```
[1] "Biobase"
```

```
> slotNames(phenoData(ABatch))
```

```
[1] "varMetadata"      "data"
```

```
[3] "dimLabels"        ".__classVersion__"
```

```
> pd <- phenoData(ABatch)
```

```
> pd@data
```

	sample
N01_normal.cel	1
N58_normal.cel	2
N59_normal.cel	3
N61_normal.cel	4
N62_normal.cel	5
N02_normal.cel	6
N11_normal.cel	7
N18_normal.cel	8
N21_normal.cel	9
N34_normal.cel	10
T36_tumor.cel	11
T40_tumor.cel	12
T43_tumor.cel	13

```
T58__tumor.cel      14
T59__tumor.cel      15
T06__tumor.cel      16
T20__tumor.cel      17
T24__tumor.cel      18
T26__tumor.cel      19
T49__tumor.cel      20
```

```
> pd@varMetadata
```

```
labelDescription
sample arbitrary numbering
```

```
> pd@dimLabels
```

```
[1] "rowNames"     "columnNames"
```

Assigning phenoData

subsamples.txt:

Array name	Sample name	Status	Batch	Cluster
N01__normal	N01A	Normal	B2	A
N58__normal	N58A	Normal	B4	A
N59__normal	N59A	Normal	B3	A
N61__normal	N61A	Normal	B3	A
N62__normal	N62A	Normal	B3	A
N02__normal	N02B	Normal	B2	B
N11__normal	N11B	Normal	B2	B
N18__normal	N18B	Normal	B2	B
...				

Assigning phenoData

```
> p1 <- read.phenoData(file.path(basedir, "subsamples.txt"))
```

Error in scan(file, what, nmax, sep, dec, quote,
skip, nlines, na.strings, : line 2 did not have 7
elements

In addition: Warning message:
read.phenoData is deprecated, use read.AnnotatedDataFrame
instead

Assigning phenoData, pt 2

```
> p1 <- read.AnnotatedDataFrame(file.path(basedir,  
+ "subsamples.txt"), sep = "\t")  
> p1
```

rowNames: 1, 2, ..., 21 (21 total)

varLabels and varMetadata:

V1: read from file

V2: read from file

...: ...

V5: read from file

(5 total)

Not quite what we want.

Assigning phenoData, pt 3

```
> p1 <- read.AnnotatedDataFrame(file.path(basedir,  
+ "subsamples.txt"), sep = "\t", header = TRUE)  
> p1
```

rowNames: 1, 2, ..., 20 (20 total)

varLabels and varMetadata:

Array.name: read from file

Sample.name: read from file

...: ...

Cluster: read from file

(5 total)

```
> phenoData(ABatch) <- p1
```

Other ways of Reading Data

Are they all in one directory?

What is the list of filenames?

`read.affybatch` vs `ReadAffy`

GUI?

Other ways of Reading Data 1

```
kabagg$ ls ../../DataSets/SinghSmall  
N60__normal.CEL N61__normal.CEL N62__normal.CEL  
  
> ABSmall <- ReadAffy(celfile.path=  
"../../DataSets/SinghSmall"); # works
```

Other ways of Reading Data 2

```
kabagg$ ls ../../DataSets/SinghSmall2  
N60__normal.CEL.gz N61__normal.CEL.gz  
N62__normal.CEL.gz
```

```
> ABSmall <- ReadAffy(celfile.path=  
  "../../DataSets/SinghSmall2",  
  compress=TRUE); # works
```

This takes only about 1/3 the space...

Other ways of Reading Data 3

```
kabagg$ ls ../../DataSets/SinghSmall3  
N60.gz N61.gz N62.gz
```

```
> ABSmall <- ReadAffy(celfile.path=  
  "../../DataSets/SinghSmall3",  
  compress=TRUE); # fails  
  
> ABSmall <- ReadAffy(filenames=  
  "../../DataSets/SinghSmall3/N60.gz",  
  compress=TRUE); # works
```

This still takes only about 1/3 the space...

Quantification

```
> t0 <- date()  
> eset0 <- expresso(ABatch, bgcorrect.method = "rma",  
+   normalize.method = "quantiles", pmcorrect.method = "pm"  
+   summary.method = "medianpolish")
```

background correction: rma

normalization: quantiles

PM/MM correction : pmonly

expression values: medianpolish

background correcting...done.

normalizing...done.

12625 ids to be processed

| |
| #####|

```
> t1 <- date()
```

```
> t0
```

```
[1] "Tue Oct 02 12:54:29 2007"
```

```
> t1
```

```
[1] "Tue Oct 02 13:01:57 2007"
```

Quantification

```
> eset1 <- justRMA(filenames = cellList)
```

Error : the following are not valid files:

```
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P  
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P
```

```
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P:  
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P:  
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P:  
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P:  
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P:  
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P:  
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P:  
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P:  
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P:  
m:/Private/ArrayCourse/Lectures07/Lecture10/G:/Public/Singh-P:
```

```
> eset1 <- justRMA(filenames = cellList, celfile.path = "")
```

Error : the following are not valid files:

```
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/N01__normal.cel  
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/N58__normal.cel  
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/N59__normal.cel
```

/G:/Public/Singh-Prostate-Affymetrix/CelFiles/N61__normal.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/N62__normal.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/N02__normal.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/N11__normal.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/N18__normal.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/N21__normal.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/N34__normal.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/T36__tumor.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/T40__tumor.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/T43__tumor.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/T58__tumor.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/T59__tumor.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/T06__tumor.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/T20__tumor.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/T24__tumor.cel
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/T26__tumor.cel

```
/G:/Public/Singh-Prostate-Affymetrix/CelFiles/T49_tumor.cel
```

```
> t2 = date()  
> eset1 <- justRMA(filenames = cellList, celfile.path = NULL)
```

Background correcting

Normalizing

Calculating Expression

```
> t3 = date()  
> t2
```

```
[1] "Tue Oct 02 13:01:57 2007"
```

```
> t3
```

```
[1] "Tue Oct 02 13:02:34 2007"
```

The following method will also work:

```
> celpath <- file.path(basedir, "CelFiles")
> cels <- sub(celpath, "", cellList)
> eset1 <- justRMA(filenames = cels, celfile.path = celpath)
> t2 <- date()
```

The customized routines are better if they do what you want to do...
(also note that justRMA didn't build an AffyBatch.)

Just Because I'm Curious

```
> exprs(eset1)[1, 1:3]
```

N01__normal.cel	N58__normal.cel	N59__normal.cel
6.732579	6.893270	7.068655

Can we reconstruct this?

```
> ABatch.BG <- bg.correct.rma(ABatch)
> ABatch.BG.norm <- normalize.AffyBatch.quantiles(ABatch.BG)
```

These steps produce AffyBatch objects, with altered exprs.

What is the First Gene?

(well, ok, probeset)

```
> gn1 <- geneNames(ABatch.BG.norm) [1]  
> gn1
```

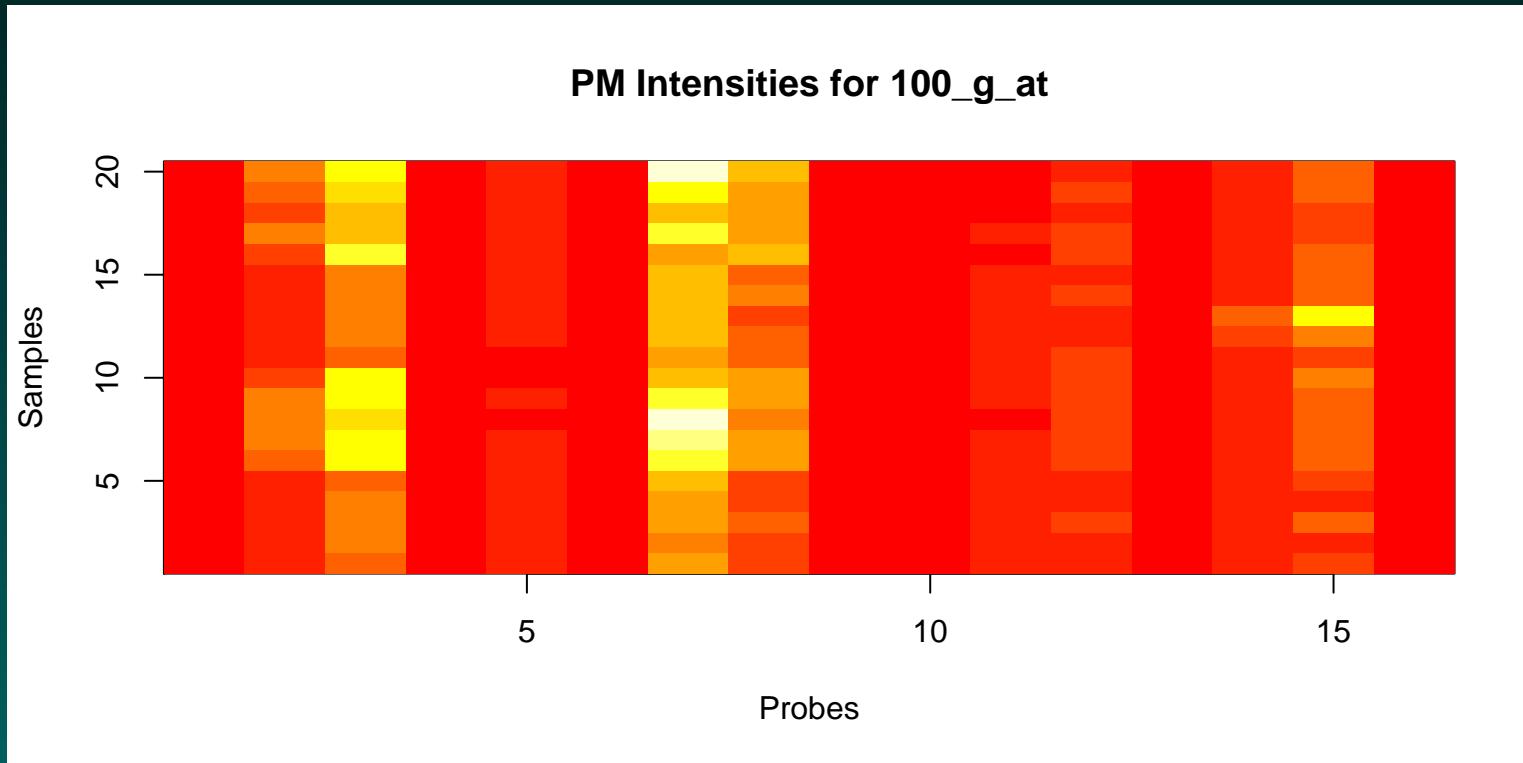
```
[1] "100_g_at"
```

Ok, now what are the values?

```
> pr1 <- pm(ABatch.BG.norm, gn1)
```

Looking at it, Take 1

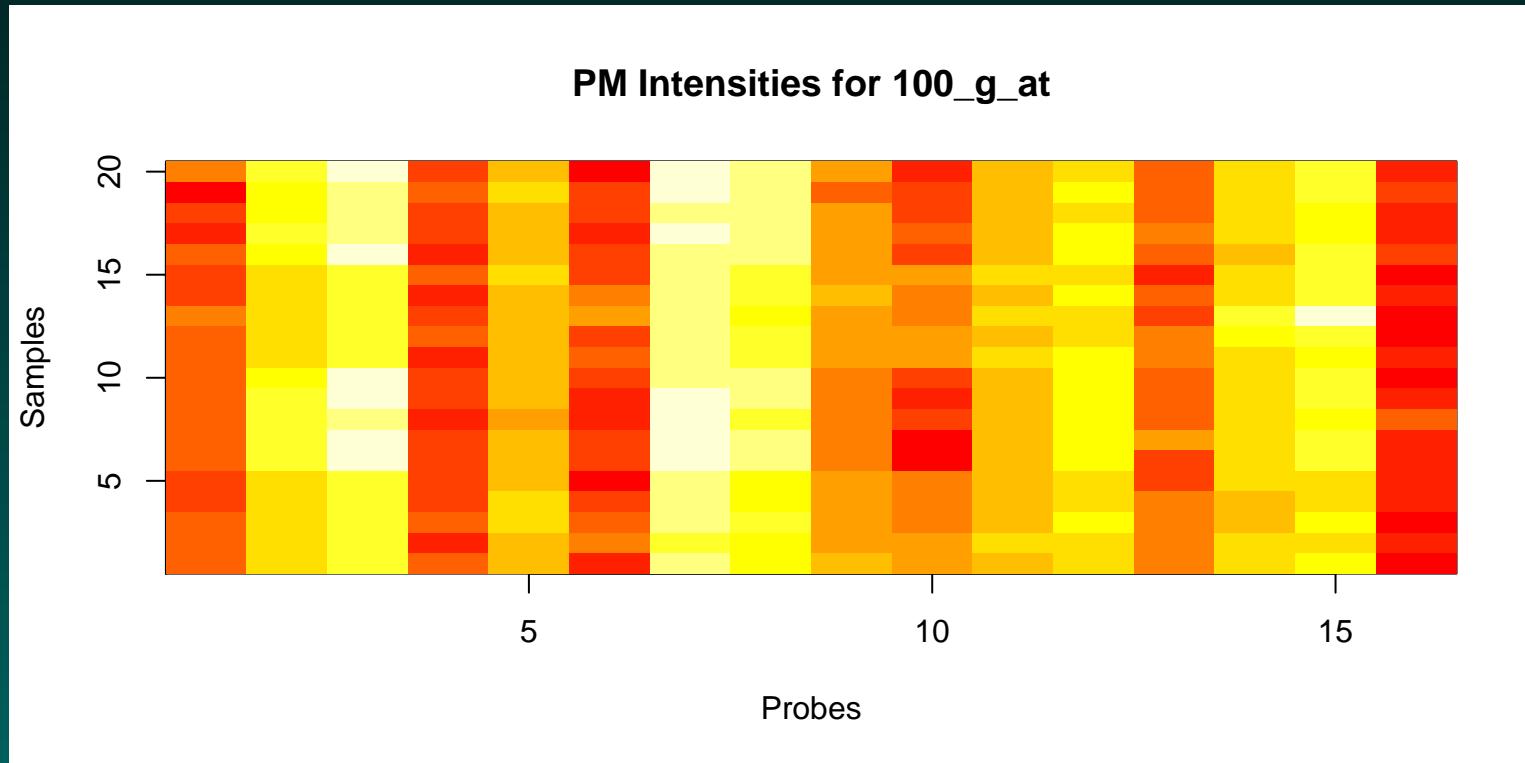
```
> image(1:nrow(pr1), 1:ncol(pr1), pr1, xlab = "Probes",
+       ylab = "Samples", main = paste("PM Intensities for",
+                                         gn1))
```



Some parallelism, but we may be missing something...

Looking at it, Take 2

```
> image(1:nrow(pr1), 1:ncol(pr1), log2(pr1), xlab = "Probes"  
+       ylab = "Samples", main = paste("PM Intensities for",  
+                                         gn1))
```



Logs!

Fitting the Probes

```
> pr1Fit <- medpolish(log2(pr1))
```

1 : 121.5456

Final: 120.8225

```
> names(pr1Fit)
```

```
[1] "overall"     "row"           "col"           "residuals"
```

```
[5] "name"
```

```
> (pr1Fit$overall + pr1Fit$col)[1:3]
```

1 2 3

6.732579 6.893270 7.068655

This is what we found before!

We can Check the Code

```
> medpolish
```

```
function (x, eps = 0.01, maxiter = 10, trace.iter = TRUE, na.rm = FALSE) {
  z <- as.matrix(x)
  nr <- nrow(z)
  nc <- ncol(z)
  t <- 0
  r <- numeric(nr)
  c <- numeric(nc)
  oldsum <- 0
  for (iter in 1:maxiter) {
    rdelta <- apply(z, 1, median, na.rm = na.rm)
    z <- z - matrix(rdelta, nr = nr, nc = nc)
    if (trace.iter) print(paste("Iteration", iter))
    if (abs(oldsum - sum(z)) <= eps * sum(z)) break
    oldsum <- sum(z)
  }
}
```

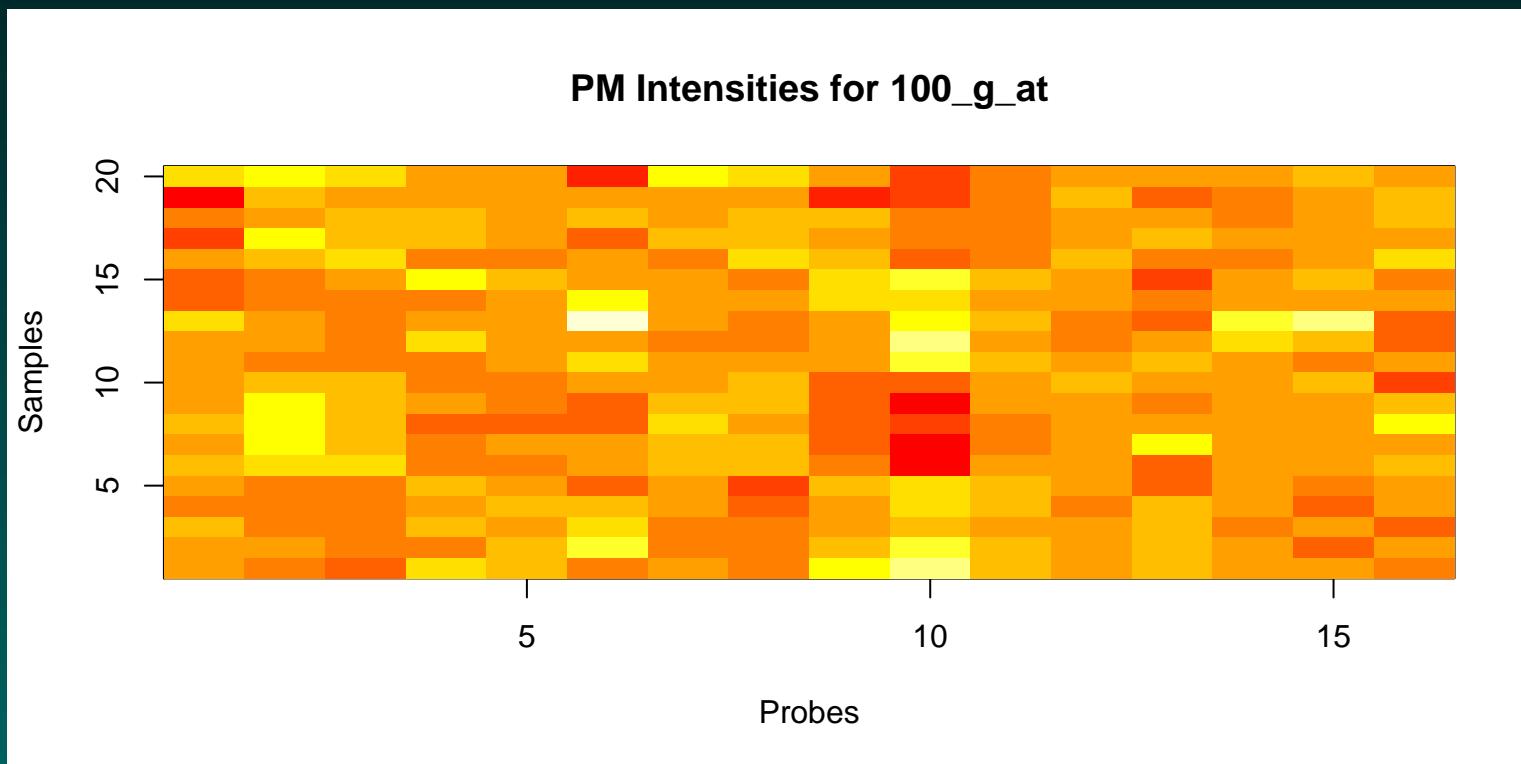
```
r <- r + rdelta
delta <- median(c, na.rm = na.rm)
c <- c - delta
t <- t + delta
cdelta <- apply(z, 2, median, na.rm = na.rm)
z <- z - matrix(cdelta, nr = nr, nc = nc, byrow = TRUE)
c <- c + cdelta
delta <- median(r, na.rm = na.rm)
r <- r - delta
t <- t + delta
newsum <- sum(abs(z), na.rm = na.rm)
converged <- newsum == 0 || abs(newsum - oldsum) < epsilon
newsum
if (converged)
  break
oldsum <- newsum
```

```
    if (trace.iter)
        cat(iterator, ":", newsum, "\n")
    }
    if (converged) {
        if (trace.iter)
            cat("Final:", newsum, "\n")
    }
    else warning(gettextf("medpolish() did not converge in %d
maxiter), domain = NA)
    names(r) <- rownames(z)
    names(c) <- colnames(z)
    ans <- list(overall = t, row = r, col = c, residuals = z,
                name = deparse(substitute(x)))
    class(ans) <- "medpolish"
    ans
}
```

<environment: namespace:stats>

and Check the Residuals

```
> image(1:nrow(pr1), 1:ncol(pr1), pr1Fit$residuals,  
+       xlab = "Probes", ylab = "Samples", main = paste("PM In  
+ gn1))
```



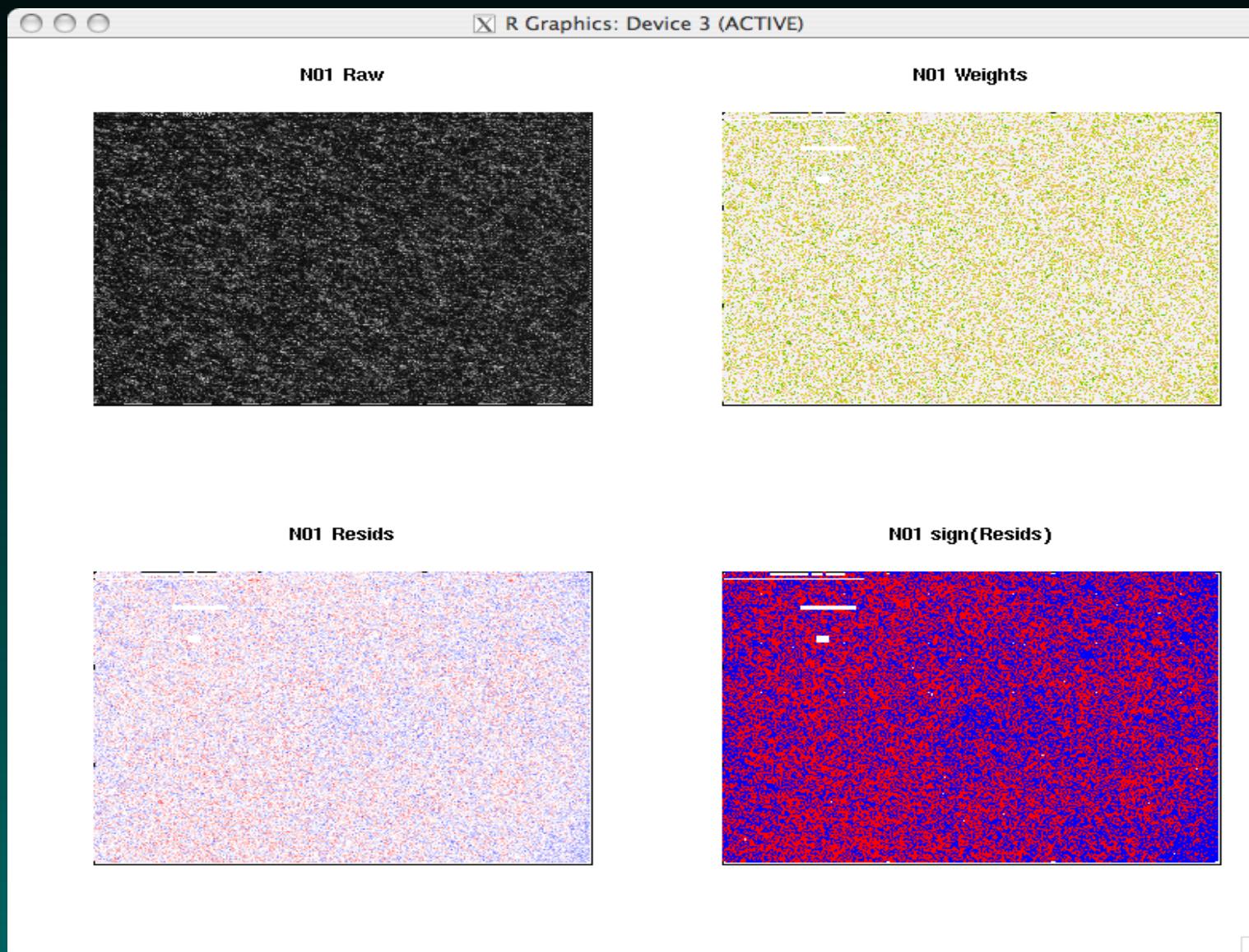
One other Fitting Approach: PLM

PLM = “Probe Level Model”

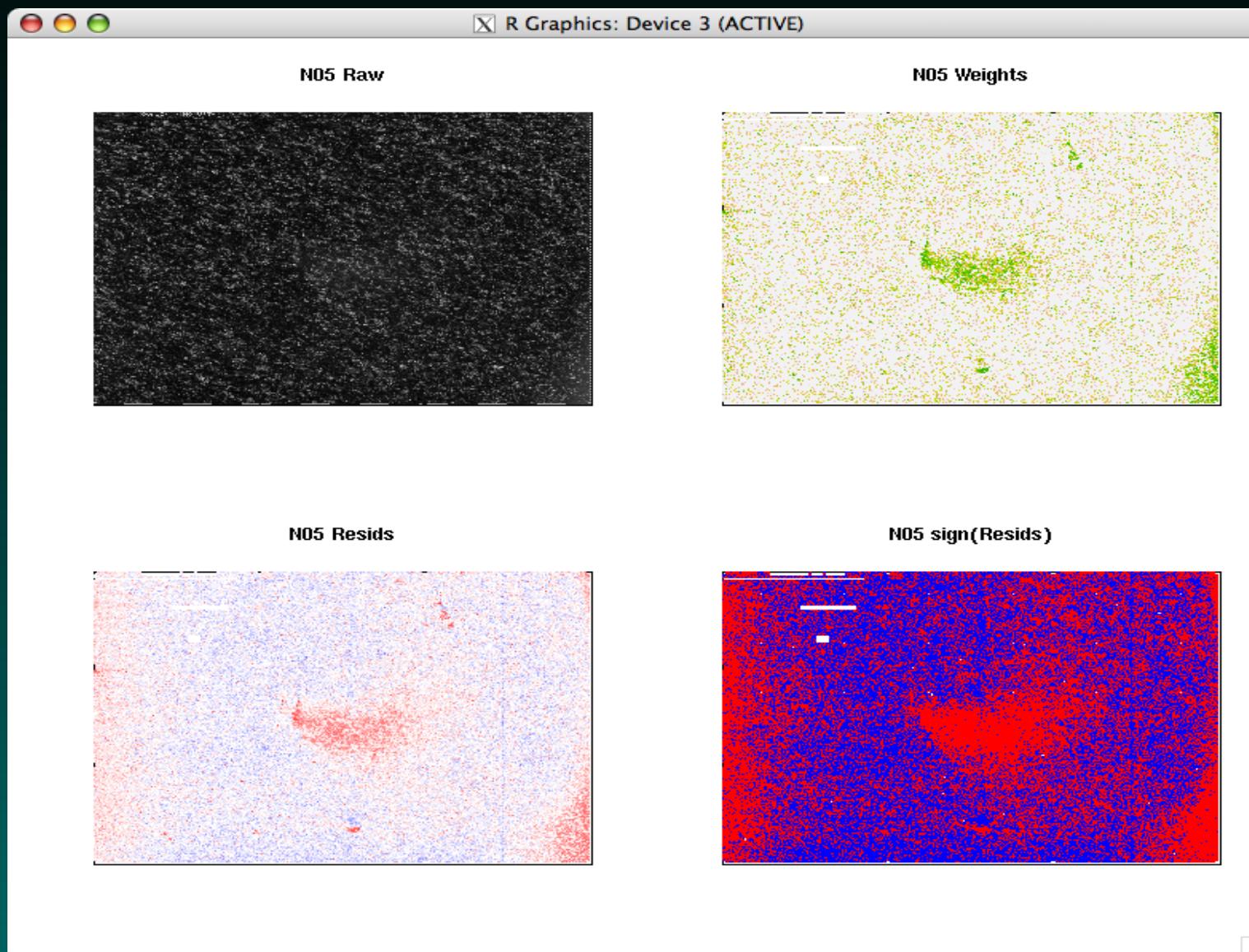
```
> library(affyPLM)
> plm1 <- fitPLM(ABatch)

> opar <- par(mfrow = c(2, 2))
> image(ABatch[, 1], main = "N01 Raw")
> image(plm1, type = "weights", which = 1, main = "N01 Weigh"
> image(plm1, type = "resids", which = 1, main = "N01 Resids"
> image(plm1, type = "sign.resids", which = 1, main = "N01 s
> par(opar)
```

Looking at N01



Looking at N05



Whence the Gene Name Info?

The screenshot shows the R Help interface for the 'hgu95av2' package. The title bar reads 'R Help' and the search bar contains 'hgu95av2'. The main content area is titled 'Affymetrix Human Genome U95 Set Annotation Data (hgu95av2)' and features a large blue 'R' logo. Below the title, there are two circular icons. The text 'Documentation for package `hgu95av2' version 1.12.0' is followed by a 'Help Pages' link. A vertical scroll bar is on the right side of the help window.

<u>hgu95av2</u>	Bioconductor annotation data package
<u>hgu95av2ACCNUM</u>	Map Manufacturer ids to GenBank Accession Numbers
<u>hgu95av2CHR</u>	Map Manufacturer IDs to Chromosomes
<u>hgu95av2CHRENGTHS</u>	A named vector for the length of each of the chromosomes
<u>hgu95av2CHRLOC</u>	Map Manufacturer IDs to Chromosomal Location
<u>hgu95av2ENZYME</u>	Map Between Manufacturer IDs and Enzyme Commission (EC) Numbers
<u>hgu95av2ENZYME2PROBE</u>	Map Between Enzyme Commission Numbers and Manufacturer Identifiers
<u>hgu95av2GENENAME</u>	Map Between Manufacturer IDs and Genes
<u>hgu95av2GO</u>	Map between Manufacturer IDs and Gene Ontology (GO)
<u>hgu95av2GO2ALLPROBES</u>	Map Between Gene Ontology (GO) Identifiers and all Manufacturer Identifiers in the subtree

```
> library("hgu95av2")
```

What Does This Package Contain?

```
> hgu95av2()
```

Quality control information for hgu95av2

Date built: Created: Mon Apr 23 12:21:36 2007

Number of probes: 12625

Probe number mismatch: None

Probe missmatch: None

Mappings found for probe based rda files:

hgu95av2ACCTNUM found 12625 of 12625

hgu95av2CHR found 12149 of 12625

hgu95av2CHRLOC found 11730 of 12625

hgu95av2ENZYME found 1861 of 12625

hgu95av2ENTREZID found 12225 of 12625

hgu95av2GENENAME found 12161 of 12625
hgu95av2GO found 11421 of 12625
hgu95av2MAP found 12121 of 12625
hgu95av2MIM found 10157 of 12625
hgu95av2PATH found 4322 of 12625
hgu95av2PFAM found 12046 of 12625
hgu95av2PMID found 12120 of 12625
hgu95av2PROSITE found 12046 of 12625
hgu95av2REFSEQ found 12004 of 12625
hgu95av2SYMBOL found 12161 of 12625
hgu95av2UNIGENE found 11973 of 12625

Mappings found for non-probe based rda files:

hgu95av2CHRENGTHS found 25
hgu95av2ENZYME2PROBE found 677
hgu95av2GO2ALLPROBES found 7501
hgu95av2GO2PROBE found 5339

```
hgu95av2PATH2PROBE found 189
```

```
hgu95av2PMID2PROBE found 127350
```

(we can also see this using `ls("package:hgu95av2")`.)

What Does This Package Contain?

```
> hgu95av2GENENAME
```

```
<environment: 0x05599244>
```

Almost everything in this package is an “environment”, which is the fancy name R uses for a hash table. We can access things by name.

```
> hgu95av2GENENAME$"1000_at"
```

```
[1] "mitogen-activated protein kinase 3"
```

We can access a lot of annotation!

What was Needed for Quantification?

```
> library("hgu95av2cdf")
> hgu95av2cdf$"1000_at"
```

	pm	mm
[1,]	358160	358800
[2,]	118945	119585
[3,]	323731	324371
[4,]	223978	224618
[5,]	313420	314060
[6,]	349209	349849
[7,]	199525	200165
[8,]	213669	214309
[9,]	236739	237379
[10,]	298099	298739

```
[11,] 282744 283384  
[12,] 281443 282083  
[13,] 349198 349838  
[14,] 297953 298593  
[15,] 317054 317694  
[16,] 404069 404709
```

These give the indices of the probes within the 409600-long vector of expression intensities.

What if We Want to Go in Reverse?

Given a probeset, I can find a gene name. What if I have a gene name, and I want something else?

Can we find “BAD”?

This is a gene symbol, so we probably want to work with the `hgu95av2SYMBOL` environment.

The key function for extracting items from an environment without the key is “contents”.

```
> tempSYM <- contents(hgu95av2SYMBOL)
> tempSYM[1]
```

```
$`1114_at`
[1] "BMP4"
```

Finding BAD in the Contents

```
> tempSYM[tempSYM == "BAD"]
```

```
$`1861_at`  
[1] "BAD"
```

```
> names(tempSYM[tempSYM == "BAD"])
```

```
[1] "1861_at"
```

This gives us the key!

Some of these queries are simplified if we invoke

```
> library("annotate")  
> getLL("1861_at", "hgu95av2")
```

1861_at

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One More Thing...

sequences?

```
> library("hgu95av2probe")
> data(hgu95av2probe)
> as.data.frame(hgu95av2probe[1, ])
```

	sequence	x	y	Probe.Set.Name
1	TGGCTCCTGCTGAGGTCCCCTTCC	395	301	1138_at
	Probe.Interrogation.Position	Target.Position		Target.Strandedness
1		2631		Antisense

So, What is BAD?

```
> as.data.frame(hgu95av2probe[hgu95av2probe$Probe.Set.Name =
+     "1861_at", ])
```

	sequence	x	y	Probe.Set.Name
14006	CAACCTCTGGCAGCACAGCGCTAT	403	485	1861_at
14007	AACCTCTGGCAGCACAGCGCTATG	402	485	1861_at
14008	CCTCTGGCAGCACAGCGCTATGGC	207	491	1861_at
14009	TGGGCAGCACAGCGCTATGCCCGCG	436	421	1861_at
14010	GCAGCACAGCGCTATGCCCGCGAGC	285	599	1861_at
14011	AGCACAGCGCTATGCCCGCGAGCTC	207	601	1861_at
14012	ACAGCGCTATGCCCGCGAGCTCCGG	632	337	1861_at
14013	CTATGCCCGCGAGCTCCGGAGGATG	196	609	1861_at
14014	TATGCCCGCGAGCTCCGGAGGATGA	624	167	1861_at
14015	GCTCCGGAGGATGAGTGACGAGTTT	440	447	1861_at

14016	GATGAGTGACGAGTTGTGGACTCC	291	467	1861_at
14017	ATGAGTGACGAGTTGTGGACTCCT	292	467	1861_at
14018	TGACGAGTTGTGGACTCCTTAAG	595	539	1861_at
14019	CGAGTTGTGGACTCCTTAAGAAG	488	515	1861_at
14020	GAGTTGTGGACTCCTTAAGAAGG	478	495	1861_at
14021	TGTGGACTCCTTAAGAAGGGACTT	502	207	1861_at

Probe.Interrogation.Position Target.Strandedness

14006	384	Antisense
14007	385	Antisense
14008	387	Antisense
14009	391	Antisense
14010	394	Antisense
14011	396	Antisense
14012	399	Antisense
14013	405	Antisense
14014	406	Antisense

14015	417	Antisense
14016	426	Antisense
14017	427	Antisense
14018	432	Antisense
14019	435	Antisense
14020	436	Antisense
14021	441	Antisense