Using *crlmm* to genotype data from Illumina's Infinium BeadChips

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1 Getting started

In this user guide we read in and genotype data from 40 HapMap samples which have been analyzed using Illumina's 370k Duo BeadChips. This data is available in the *hapmap370k* package. Additional chip-specific model parameters and basic SNP annotation information used by CRLMM is stored in the *human370v1c* package. These can be downloaded from http://rafalab.jhsph.edu/software.html and must be installed for the following code to work.

2 Reading in data

The function `readIdatFiles` extracts the Red and Green intensities from the binary `idat` files output by Illumina's scanning device. The file `samples370k.csv` contains information about each sample.

```
> options(width = 50)
```

```
> library(Biobase)
> library(crlmm)
> library(hapmap370k)
> data.dir = system.file("idatFiles", package = "hapmap370k")
> samples = read.csv(file.path(data.dir,
+     "samples370k.csv"), as.is = TRUE)
> samples[1:5, ]
```

```
  HapMap.Name Gender         Plate Well
1     NA06991 Female WG1000442-DNA  E11
2     NA07000 Female WG1000442-DNA  D08
3     NA10859 Female WG1000453-DNA  B02
```

```
4     NA11882 Female WG1000453-DNA  D08
5     NA06993   Male WG1000447-DNA  D11
  SentrixPosition
1    4030186347_A
2    4030186263_B
3    4019585415_B
4    4031058127_B
5    4031058211_B
```

```
> RG = readIdatFiles(samples, path = data.dir,
+     arrayInfoColNames = list(barcode = NULL,
+         position = "SentrixPosition"),
+     saveDate = TRUE)
```

Reading in this data takes approximately 90 seconds and peak memory usage was 1.2 GB of RAM on our linux system. The `RG` object is an *NChannelSet* which stores the Red and Green intensities, the number of beads and standard errors for each bead-type. The scanning date of each array is stored in `protocolData`.

```
> class(RG)

[1] "NChannelSet"
attr(,"package")
[1] "Biobase"

> dim(RG)

Features  Samples
  381079       40

> slotNames(RG)

[1] "assayData"         "phenoData"
[3] "featureData"       "experimentData"
[5] "annotation"        "protocolData"
[7] ".__classVersion__"

> channelNames(RG)

[1] "G"   "Gnb" "Gse" "R"   "Rnb" "Rse"

> exprs(channel(RG, "R"))[1:5, 1:5]
```

```
      4030186347_A 4030186263_B 4019585415_B
10008          321          170         2961
10010         1738         3702         3105
10025           80          101          145
10026         5043         1856         6519
10039         4905         2464         9080
      4031058127_B 4031058211_B
10008         3468          262
10010         3425           70
10025           29           21
10026         8304         9872
10039         9788        10867
```

```
> exprs(channel(RG, "G"))[1:5, 1:5]
```

```
      4030186347_A 4030186263_B 4019585415_B
10008         4183         4484         3765
10010         2593           51         3824
10025         2768         2322         3435
10026          216         2840          211
10039          297         3016          345
      4031058127_B 4031058211_B
10008         3558         6502
10010         3528         6154
10025         3471         3608
10026          164          188
10039          361          380
```

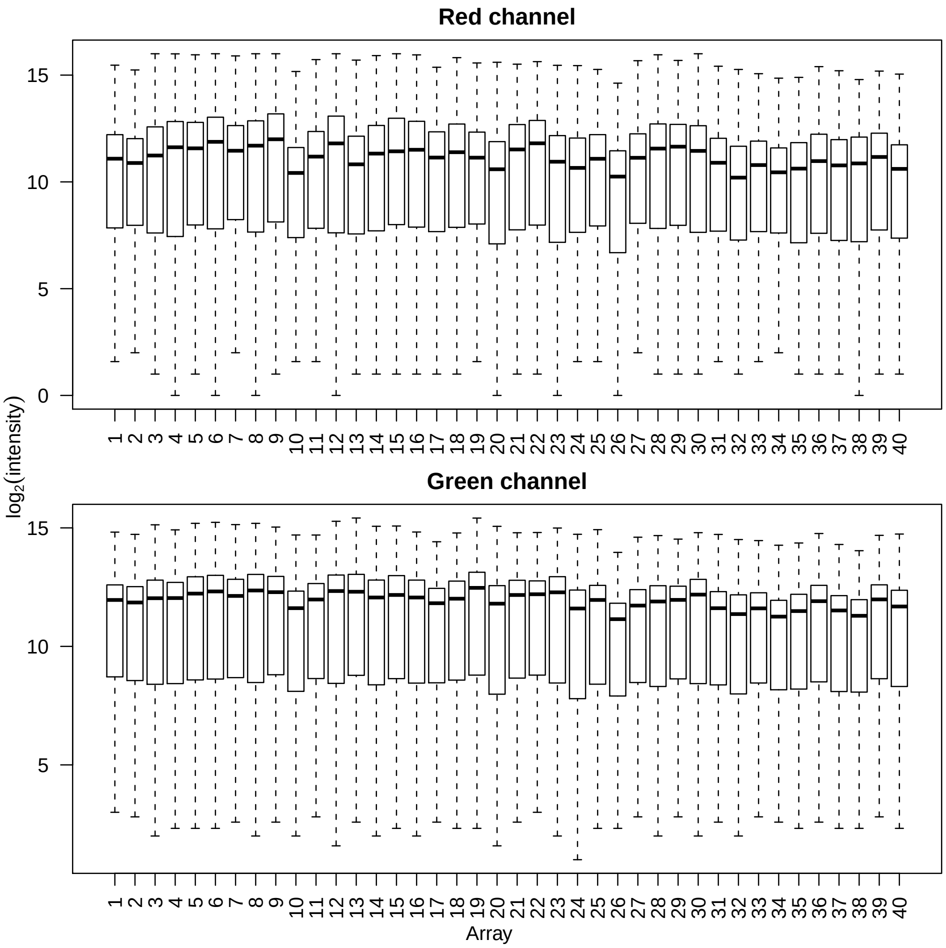
```
> pd = pData(RG)
> pd[1:5, ]
```

```
             HapMap.Name Gender         Plate
4030186347_A     NA06991 Female WG1000442-DNA
4030186263_B     NA07000 Female WG1000442-DNA
4019585415_B     NA10859 Female WG1000453-DNA
4031058127_B     NA11882 Female WG1000453-DNA
4031058211_B     NA06993   Male WG1000447-DNA
             Well SentrixPosition
4030186347_A  E11    4030186347_A
4030186263_B  D08    4030186263_B
4019585415_B  B02    4019585415_B
4031058127_B  D08    4031058127_B
4031058211_B  D11    4031058211_B
```

```
> scandatetime = strptime(protocolData(RG)[["ScanDate"]],
+     "%m/%d/%Y %H:%M:%S %p")
> datescanned = substr(scandatetime, 1,
+     10)
> scanbatch = factor(datescanned)
> levels(scanbatch) = 1:16
> scanbatch = as.numeric(scanbatch)
```

Plots of the summarised data can be easily generated to check for arrays with poor signal.

```
> par(mfrow = c(2, 1), mai = c(0.4, 0.4,
+     0.4, 0.1), oma = c(1, 1, 0, 0))
> boxplot(log2(exprs(channel(RG, "R"))),
+     xlab = "Array", ylab = "", names = 1:40,
+     main = "Red channel", outline = FALSE,
+     las = 2)
> boxplot(log2(exprs(channel(RG, "G"))),
+     xlab = "Array", ylab = "", names = 1:40,
+     main = "Green channel", outline = FALSE,
+     las = 2)
> mtext(expression(log[2](intensity)), side = 2,
+     outer = TRUE)
> mtext("Array", side = 1, outer = TRUE)
```



3 Genotyping

Next we use the function `crlmmIllumina` which performs preprocessing followed by genotyping using the CRLMM algorithm.

```
> crlmmResult = crlmmIllumina(RG = RG, cdfName = "human370v1c",
+     sns = pData(RG)$ID, returnParams = TRUE)
```

This analysis took 470 seconds to complete and peak memory usage was 3.3 GB on our system. The output stored in `crlmmResult` is a *SnpSet* object.

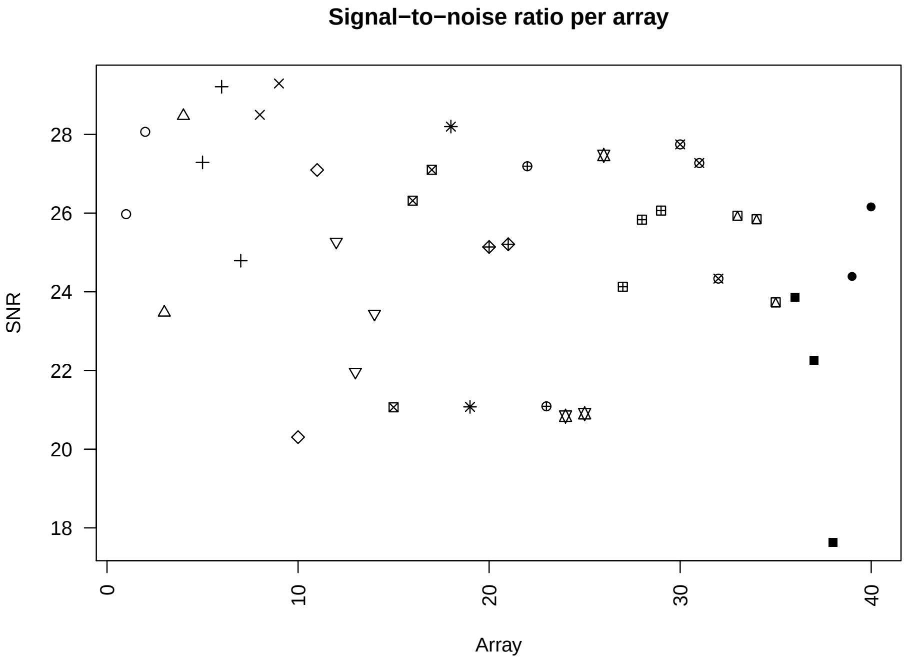
```
> class(crlmmResult)
```

```
[1] "SnpSet"
attr(,"package")
[1] "Biobase"
```

```
> dim(crlmmResult)

Features  Samples
  346451       40

> slotNames(crlmmResult)

[1] "assayData"         "phenoData"
[3] "featureData"       "experimentData"
[5] "annotation"        "protocolData"
[7] ".__classVersion__"

> calls(crlmmResult)[1:10, 1:5]

           1 2 3 4 5
rs12354060 3 3 3 3 3
rs6650104  1 1 1 1 1
rs12184279 3 1 3 3 3
rs12564807 1 1 1 1 1
rs3115860  2 1 1 2 2
rs3115850  2 2 2 2 2
rs7515489  3 3 3 1 1
rs12124819 1 2 2 1 1
rs17160939 1 1 1 1 1
rs12086311 3 3 3 3 3
```

Plotting the *SNR* reveals no obvious batch effects in this data set (different symbols are used for arrays scanned on different days).

```
> plot(crlmmResult[["SNR"]], pch = scanbatch,
+     xlab = "Array", ylab = "SNR", main = "Signal-to-noise ratio per array",
+     las = 2)
```



4 System information

This analysis was carried out on a linux machine with 32GB of RAM using the following packages:

```
> sessionInfo()

R version 2.10.0 RC (2009-10-23 r50188)
x86_64-unknown-linux-gnu

locale:
 [1] LC_CTYPE=en_US.iso885915
 [2] LC_NUMERIC=C
 [3] LC_TIME=en_US.iso885915
 [4] LC_COLLATE=en_US.iso885915
 [5] LC_MONETARY=C
 [6] LC_MESSAGES=en_US.iso885915
 [7] LC_PAPER=en_US.iso885915
 [8] LC_NAME=C
 [9] LC_ADDRESS=C
[10] LC_TELEPHONE=C
[11] LC_MEASUREMENT=en_US.iso885915
```

```
[12] LC_IDENTIFICATION=C

attached base packages:
[1] tools     stats     graphics  grDevices
[5] utils     datasets  methods   base

other attached packages:
[1] human370v1cCrlmm_1.0.0 hapmap370k_1.0.0
[3] crlmm_1.3.23           Biobase_2.5.8
[5] weaver_1.11.1          codetools_0.2-2
[7] digest_0.4.1

loaded via a namespace (and not attached):
 [1] affyio_1.13.5        annotate_1.23.4
 [3] AnnotationDbi_1.7.20 Biostrings_2.13.54
 [5] DBI_0.2-4            ellipse_0.3-5
 [7] genefilter_1.25.11   IRanges_1.3.97
 [9] mvtnorm_0.9-7        oligoClasses_1.7.16
[11] preprocessCore_1.7.9 RSQLite_0.7-3
[13] SNPchip_1.9.8        splines_2.10.0
[15] survival_2.35-7      xtable_1.5-5
```