Bioconductor's aCGH package

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1 Overview

This document presents an overview of the aCGH package, which provides wide basic functions for reading, analyzing and plotting array Comparative Genomic Hybridization data (Snijders et al. (2001)). Specific example for reading data in is using output of the custom freely available programs, SPOT and SPROC (Jain et al. (2002)). These programs provide image quantification and pre-processing. Outputs of all the other image processing software need to be combined into a single file containing observed values for each clone and samples and then read in as a matrix.

2 Data

The data used in the example was generated in in lab of Dr. Fred Waldman at UCSF Comprehensive Cancer Center (Nakao et al. (2004)). Array CGH has been done on 125 colorectal fresh-frozen primary tumors and the associations with various phenotypes were analyzed. To reduce running time, only 40 samples are used in the examples.

3 Examples

3.1 Creating aCGH object from log2.ratios and clone info files

Each array CGH object has to contain the log2ratios representing relative copy number along with the mapping information including but not limited to clone name, chromosome and kb relative to the chromosome. Optionally there may be phenotypes associated with each sample.

Note that when working with your own data, you will need to specify absolute path to those files of the path relative to your working folder. For instance, if you are working in the folder Project1 your data files are placed in the subfolder Project1/Data, then datadir = "Data" if you are using relative path.

3.2 Filtering and imputation for objects of class aCGH

Here we remove unmapped clones and clones mapping to Y chromosome, screen out clones missing in more than 25

```
> ex.acgh <-
+ aCGH.process(ex.acgh, chrom.remove.threshold = 23, prop.missing = .25, sample.quality.t</pre>
```

Here we impute missing observations using lowess approach. Note that occasionally, majority of the observations on chromosome Y may be missing causing imputing function to fail. Therefore, by default, the largest chromosome to be imputed is indexed as maxChrom=23 (X). Here we specify imputation for all chromosomes; however, in this example there are no data on chromosome Y.

```
> log2.ratios.imputed(ex.acgh) <- impute.lowess(ex.acgh, maxChrom=24)</pre>
Processing chromosome 1
Processing chromosome
Processing chromosome
Processing chromosome 4
Processing chromosome 5
Processing chromosome 6
Processing chromosome 7
Processing chromosome 8
Processing chromosome
                      9
Processing chromosome
                      10
Processing chromosome
                      11
Processing chromosome
                      12
Processing chromosome
                       13
Processing chromosome
                       14
Processing chromosome
                      15
Processing chromosome
                       16
Processing chromosome
                       17
Processing chromosome
                      18
Processing chromosome
                      19
Processing chromosome
                      20
Processing chromosome
                      21
Processing chromosome
                      22
                      23
Processing chromosome
3.3
     Printing, summary and basic plotting (fig. 1) for objects of class aCGH
> data(colorectal)
> colorectal
aCGH object
Call: aCGH.read.Sprocs(sproclist[1:40], "human.clones.info.Jul03.csv",
    chrom.remove.threshold = 23)
Number of Arrays 40
Number of Clones 2031
> summary(colorectal)
aCGH object
Call: aCGH.read.Sprocs(sproclist[1:40], "human.clones.info.Jul03.csv",
    chrom.remove.threshold = 23)
Number of Arrays 40
Number of Clones 2031
Imputed data exist
HMM states assigned
```

samples standard deviations are computed genomic events are assigned phenotype exists

> plot(colorectal)

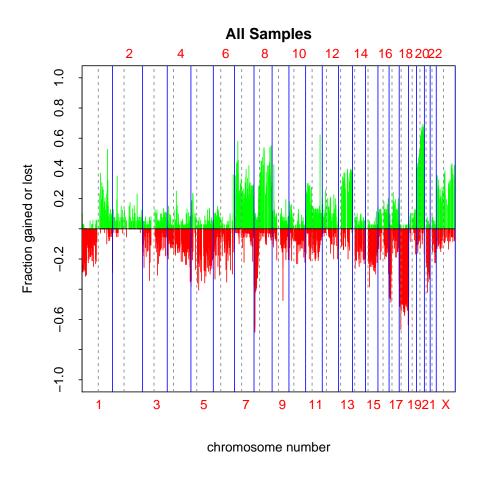


Figure 1: Basic Frequency Plot

```
> sample.names(colorectal)
 [1] "sprocCR31.txt" "sprocCR40.txt" "sprocCR43.txt" "sprocCR59.txt"
                                        "sprocCR75.txt" "sprocCR77.txt"
 [5] "sprocCR63.txt" "sprocCR73.txt"
 [9] "sprocCR96.txt" "sprocCR98.txt" "sprocCR100.txt" "sprocCR106.txt"
[13] "sprocCR112.txt" "sprocCR122.txt" "sprocCR124.txt" "sprocCR131.txt"
[17] "sprocCR135.txt" "sprocCR137.txt" "sprocCR146.txt" "sprocCR148.txt"
[21] "sprocCR150.txt" "sprocCR154.txt" "sprocCR159.txt" "sprocCR163.txt"
[25] "sprocCR169.txt" "sprocCR178.txt" "sprocCR180.txt" "sprocCR186.txt"
[29] "sprocCR193.txt" "sprocCR200.txt" "sprocCR204.txt" "sprocCR210.txt"
[33] "sprocCR212.txt" "sprocCR217.txt" "sprocCR219.txt" "sprocCR227.txt"
[37] "sprocCR232.txt" "sprocCR244.txt" "sprocCR246.txt" "sprocCR248.txt"
> phenotype(colorectal)[1:4,]
  id age sex stage loc
                                  hist diff gstm1 gstt1 nqo K12 K13 MTHFR ERCC1
1 31
      70
                                                                   2
                     O Adenocarcinoma
2 40
     71
                                                               2
                                                                   2
                                                                         2
                                                                               2
                     1 Adenocarcinoma
                                          1
                                                1
3 43
     59
           1
                 1
                     0 Adenocarcinoma
                                         NA
                                                1
                                                      1
                                                          1
                                                               2
                                                                   2
                                                                         2
                                                                               1
4 59 72
                 2
                                                               2
                                                                   2
           0
                     1 Adenocarcinoma
                                          1
                                                1
                                                      1
                                                           1
                                                                         1
                                                                              NA
  bat26 bat25 D5S346 D17S250 D2S123
                                                             mi2
                                                                       LOH k12
            0
1
      0
                   0
                                               0/1 unstable loci negative
2
            0
                                   1 >2 loci unstable, (NCI def) negative
3
      0
                           0
                                               0/1 unstable loci negative
                           0
                                   0
                                               0/1 unstable loci negative
  K12AA k13 K13AA M677 M1298 p16 p14 mlh1 BAT26 mlh1c
                                                                       mi misum
          0
                     1
                           0
                                1
                                   0
                                               0
                                                     0 0/1 unstable loci
                                                                              0
                                         1
2
          0
                     1
                           0
                                0
                                    0
                                         0
                                               0
                                                     0 >2 loci unstable
                                                                              3
3
          0
                                2
                                                     0 0/1 unstable loci
                     1
                           0
                                    0
                                         0
                                               0
                                                                              0
```

1

0

1 Complete

CGHSTAT

- 2 Complete
- 3 Complete
- 4 Not Done

Reading Sproc files

0

Here we demonstrate reading of the sproc files and combining them into one array CGH object. Sproc file format is specific to the custom SPROC processing software at UCSF Cancer Center.

```
> datadir <- system.file("examples", package = "aCGH")</pre>
> latest.mapping.file <-</pre>
          file.path(datadir, "human.clones.info.Jul03.txt")
> ex.acgh <-
          aCGH.read.Sprocs(dir(path = datadir,pattern = "sproc",
                           full.names = TRUE), latest.mapping.file,
                           chrom.remove.threshold = 23)
```

1

0

1

0

0

0 0/1 unstable loci

Trying to read E:/biocbld/bbs-3.1-bioc/tmpdir/RtmpI7gZj0/Rinst21bc3c4da9/aCGH/examples/sproc Trying to read E:/biocbld/bbs-3.1-bioc/tmpdir/RtmpI7gZj0/Rinst21bc3c4da9/aCGH/examples/sproc

```
Averaging duplicated clones
CTB-102E19
                   692 693
CTB-112F7
                  1692 1693
CTB-142024
                   1640 1641
CTB-339E12
                   1633 1634
CTB-36F16
                  1220 1221
DMPC-HFF#1-61H8
                        1662 1663
GS1-20208
                  662 663
RP1-97B16
                  256 257
RP11-119J20
                    409 410
RP11-13C20
                   153 154
RP11-149G12
                    815 816
                   825 826
RP11-172D2
RP11-175H20
                    821 822
                    183 184
RP11-176L22
RP11-188C10
                    817 818
RP11-1L22
                  147 148
RP11-204M16
                    785 786
RP11-238H10
                    850 851
RP11-23G2
                  176 177
RP11-247E23
                    178 179
RP11-268N2
                  813 814
RP11-30M1
                  166 167
RP11-39A8
                  158 159
RP11-47E6
                  170 171
RP11-72C6
                  1006 1007
RP11-83014
                   819 820
RP11-94M13
                   873 874
> ex.acgh
aCGH object
Call: aCGH.read.Sprocs(dir(path = datadir, pattern = "sproc", full.names = TRUE),
    latest.mapping.file, chrom.remove.threshold = 23)
Number of Arrays 2
Number of Clones 1952
```

3.5 Basic plot for batch of aCGH Sproc files. (fig. 2)

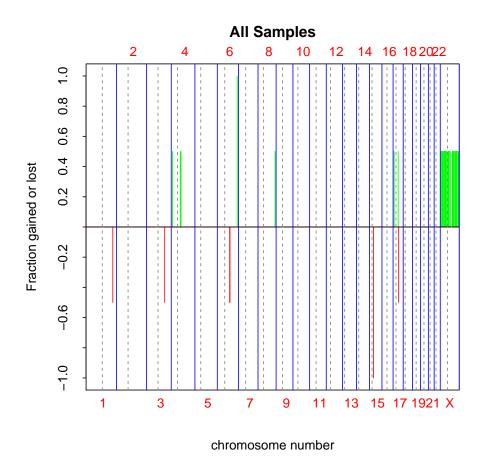


Figure 2: Basic plot for batch of aCGH Sproc files

3.6 Subsetting example

> cr <- colorectal[,1:3]</pre>

3.7 Basic plot for the ordered log2 ratios along the genome

The relative copy number is plotted along the genome with clones placed in the genomic order. We are plotting sample 2 here. (fig. 3). Chromosome Y is explicitly excluded.

> plotGenome(ex.acgh, samples=2, Y = FALSE)

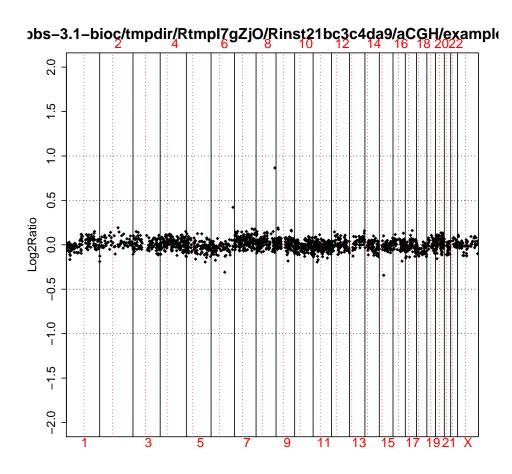


Figure 3: Basic plot for the ordered log2 ratios along the genome

3.8 Computing and plotting hmm states

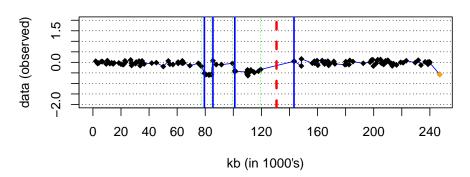
Unsupervised hidden markov model is repeatedly fitted to each chromosome for varying number of states (2 , ..., 5). The number of states is determined after all fits are done using model selection criterion such as AIC, BIC or delta-BIC. The model with minimal penalized negative log-likelihood is chosen for each selection criterion. Note, that some of the model fits are going to fail and are not going to be used in the final selection. Meanwhile , error message warning of the model fit failing will be printed during hmm runs. The user shoulld ignore those particular messages and related warnings.

For a given sample, each chromosome is plotted on a separate page along with its smoothed values(fig. 4). The genomic events such as transitions, focal aberrations and amplifications are indicated. The outliers are also marked.

```
> ## Determining hmm states of the clones. In the interest of time,
> ##we have commented this step out and used pre-computed results.
> ##hmm(ex.acgh) <- find.hmm.states(ex.acgh)</pre>
> hmm(ex.acgh) <- ex.acgh.hmm</pre>
> ## Merging hmm states
> hmm.merged(ex.acgh) <-</pre>
     mergeHmmStates(ex.acgh, model.use = 1, minDiff = .25)
> ## Calculating the standard deviations for each array. Standard error is
> ##calculated for each region and then averaged across regions. The final
> ##SDs for each samples are contained in sd.samples(exa.acgh)$madGenome.
>
> sd.samples(ex.acgh) <- computeSD.Samples(ex.acgh)
> ## Finding the genomic events associated with each sample using
> ##results of the partitioning into the states.
> genomic.events(ex.acgh) <- find.genomic.events(ex.acgh)
Finding outliers
Finding focal low level aberrations
Finding transitions
Finding focal amplifications
Processing chromosome
Processing chromosome
Processing chromosome
Processing chromosome
Processing chromosome
                       5
Processing chromosome
                       7
Processing chromosome
Processing chromosome
                       8
Processing chromosome
Processing chromosome
                       10
Processing chromosome
                       11
Processing chromosome
                       12
```

```
Processing chromosome 13
Processing chromosome 14
Processing chromosome
                      15
Processing chromosome
                      16
Processing chromosome 17
Processing chromosome 18
Processing chromosome 19
Processing chromosome 20
Processing chromosome 21
Processing chromosome 22
Processing chromosome 23
> ## Plotting and printing the hmm states either to the screen or into the
> ##postscript file. Each chromosome for each sample is plotted on a separate
> ##page
> ##postscript("hmm.states.temp.ps");plotHmmStates(ex.acgh, sample.ind=1);dev.off()
```

Sample 1 sprocCR31.txt - Chr 1 Number of states 2



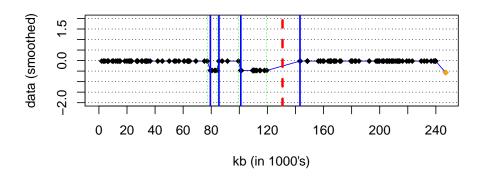


Figure 4: Plotting the hmm states found for colorectal data set.

3.9 Plotting summary of the tumor profiles

Here the distribution of various genomic events as well as their frequency by location is displayed. Run the function plotSummaryProfile(colorectal) which produces multi-page figure. Necessary to write out as ps or pdf files.

3.10 Overall frequency plot (fig. 5)

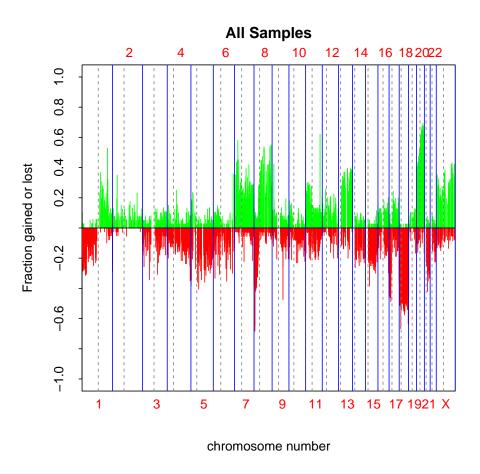


Figure 5: Overall frequency plot of the tumor profiles

summarize.clones() function is the text equivalent of plotFreqStat() - it summarizes the frequencies of changes for each clone across tumors and includes results of statistical comparisons for each clone when available.

> summarize.clones(colorectal)[1:10 ,]

	Clone	Target	${\tt Chrom}$	kb	NumPresent.All	NumGain.All
2	RP11-82D16	HumArray2H11_C9	1	2009	39	4
3	RP11-62M23	HumArray2H10_N30	1	3368	35	1
4	RP11-11105	HumArray2H10_B18	1	4262	38	1
5	RP11-51B4	HumArray2H10_Q30	1	6069	35	0
6	RP11-60J11	HumArray2H10_T30	1	6817	36	1
7	RP11-813J5	<pre>HumArray2H10_B19</pre>	1	9498	30	0
8	RP11-19901	HumArray2H10_W30	1	10284	39	1
9	RP11-188F7	HumArray2H9_C14	1	12042	36	1

```
10 RP11-178M15 HumArray2H9_F14
                                                             35
                                       1 13349
                                                                           1
11 RP11-219F4 HumArray2H9_I14
                                       1 14391
                                                             39
                                                                           1
   NumLost.All PropPresent.All PropGain.All PropLost.All
2
              7
                            0.98
                                          0.10
                                                         0.18
3
              7
                            0.88
                                          0.03
                                                         0.20
              9
                                                         0.24
4
                            0.95
                                          0.03
5
             10
                            0.88
                                          0.00
                                                         0.29
6
              7
                            0.90
                                          0.03
                                                         0.19
7
              8
                            0.75
                                          0.00
                                                         0.27
8
              5
                            0.98
                                          0.03
                                                         0.13
9
              4
                                                         0.11
                            0.90
                                          0.03
              4
10
                            0.88
                                          0.03
                                                         0.11
              7
                            0.98
                                          0.03
                                                         0.18
11
```

threshold.func() function gives the clone by sample matrix of gains and losses. "1" indicates gain and "-1" indicates loss.

```
> factor <- 3
> tbl <- threshold.func(log2.ratios(colorectal),
               posThres=factor*(sd.samples(colorectal)$madGenome))
> rownames(tbl) <- clone.names(colorectal)</pre>
> colnames(tbl) <- sample.names(colorectal)</pre>
> tbl[1:5,1:5]
            sprocCR31.txt sprocCR40.txt sprocCR43.txt sprocCR59.txt
RP11-82D16
                         0
RP11-62M23
                         0
                                        0
                                                       0
                                                                     -1
RP11-11105
                         0
                                        0
                                                       0
                                                                     -1
RP11-51B4
                         0
                                       NA
                                                       0
                                                                     -1
RP11-60J11
                         0
                                        0
                                                       0
                                                                     -1
            sprocCR63.txt
RP11-82D16
                         1
                         0
RP11-62M23
RP11-11105
                         1
RP11-51B4
                         0
RP11-60J11
                         0
```

fga.func() function gives the fraction of genome altered for each sample.

```
> col.fga <- fga.func(colorectal, factor=3,chrominfo=human.chrom.info.Jul03)
> cbind(gainP=col.fga$gainP,lossP=col.fga$lossP)[1:5,]
```

```
gainP lossP
[1,] 0.220098155 0.184029096
[2,] 0.025559893 0.004990002
[3,] 0.006184865 0.002350805
[4,] 0.107402285 0.148058176
[5,] 0.143115647 0.137430523
```

3.11 Testing association of clones with categorical, censored or continuous outcomes.

Use mt.maxT function from multtest package to test differences in group means for each clone grouped by sex. Plot the result along the genome displaying the frequencies of gains and losses as well well as height of the statistic corresponding to each clone(figs. 6 and 7.). The p-value can be adjusted and the horizontal lines indicate chosen level of significance.

> colnames(phenotype(colorectal))

```
[1] "id"
                "age"
                          "sex"
                                     "stage"
                                                "loc"
                                                           "hist"
                                                                     "diff"
                                     "K12"
[8] "gstm1"
                "gstt1"
                          "nqo"
                                                "K13"
                                                           "MTHFR"
                                                                     "ERCC1"
[15] "bat26"
               "bat25"
                          "D5S346"
                                     "D17S250" "D2S123"
                                                           "mi2"
                                                                     "LOH"
[22] "k12"
               "K12AA"
                          "k13"
                                     "K13AA"
                                                "M677"
                                                           "M1298"
                                                                     "p16"
[29] "p14"
               "mlh1"
                          "BAT26"
                                     "mlh1c"
                                                "mi"
                                                           "misum"
                                                                     "CGHSTAT"
```

- > sex <- phenotype(colorectal)\$sex
- > sex.na <- !is.na(sex)</pre>
- > index.clones.use <- which(clones.info(colorectal)\$Chrom < 23)</pre>
- > colorectal.na <- colorectal[index.clones.use,sex.na , keep=TRUE]</pre>
- > dat <- log2.ratios.imputed(colorectal.na)</pre>
- > resT.sex <- mt.maxT(dat, sex[sex.na], test = "t.equalvar", B = 1000)

b=10	b=20	b=30	b=40	b=50	b=60	b=70	b=80
b=110	b=120	b=130	b=140	b=150	b=160	b=170	b=
b=210	b=220	b=230	b=240	b=250	b=260	b=270	b=
b=310	b=320	b=330	b=340	b=350	b=360	b=370	b=
b=410	b=420	b=430	b=440	b=450	b=460	b=470	b=
b=510	b=520	b=530	b=540	b=550	b=560	b=570	b=
b=610	b=620	b=630	b=640	b=650	b=660	b=670	b=
b=710	b=720	b=730	b=740	b=750	b=760	b=770	b=
b=810	b=820	b=830	b=840	b=850	b=860	b=870	b=
b=910	b=920	b=930	b=940	b=950	b=960	b=970	b=

> plotFreqStat(colorectal.na, resT.sex, sex[sex.na], factor=3, titles =
+ c("Female", "Male"), X = FALSE, Y = FALSE)

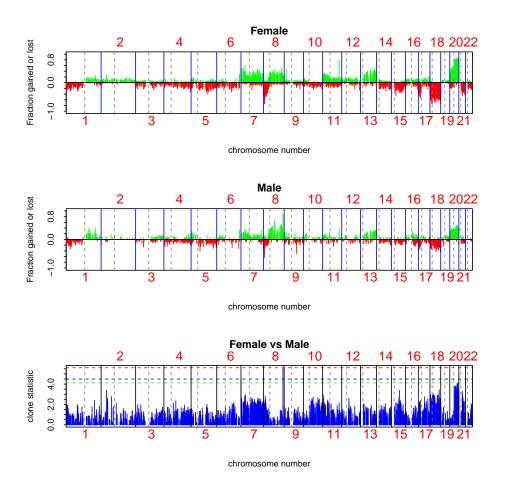


Figure 6: Frequency plots of the samples with respect to the sex groups

```
> plotSummaryProfile(colorectal, response = sex,
                           titles = c("Female", "Male"),
                           X = FALSE, Y = FALSE, maxChrom = 22)
                     Number of Transitions 0.0950684
                                                        Number of Chrom containing Transitions 0.174915
                                      0
               25
                                                        10
               15
                                                        2
               2
                                                        0
                       Female
                                     Male
                                                                Female
                                                                              Male
                     Number of Aberrations 0.841381
                                                         Number of Whole Chrom Changes 0.0174111
                                                        15
                                                        10
               100
                                                        2
               20
                                                        0
                       Female
                                     Male
                                                                Female
                                                                              Male
```

Figure 7: Plotting summary of the tumor profiles

Testing association of clones with categorical outcome for autosomal clones that are gained or lost in at least 10% of the samples. Note that the same dataset should be provided for creating resT object and for plotting. Pay attention that HMM-related objects including sample variability do not get subsetted at the moment. Note that currently two-stage subsetting does not work for HMM slots, i.e. two conditions (change and autosomal) need to be done in one iteration.

```
> factor <- 3
```

> resT.sex <- mt.maxT(dat, sex[sex.na],test = "t.equalvar", B = 1000)</pre>

b=10	b=20	b=30	b=40	b=50	b=60	b=70	b=80
b=110	b=120	b=130	b=140	b=150	b=160	b=170	b=
b=210	b=220	b=230	b=240	b=250	b=260	b=270	b=
b=310	b=320	b=330	b=340	b=350	b=360	b=370	b=
b=410	b=420	b=430	b=440	b=450	b=460	b=470	b=
b=510	b=520	b=530	b=540	b=550	b=560	b=570	b=
b=610	b=620	b=630	b=640	b=650	b=660	b=670	b=
b=710	b=720	b=730	b=740	b=750	b=760	b=770	b=
b=810	b=820	b=830	b=840	b=850	b=860	b=870	b=
b=910	b=920	b=930	b=940	b=950	b=960	b=970	b=

>

> minChanged <- 0.1

> gainloss <- gainLoss(log2.ratios(colorectal)[,sex.na], cols=1:length(which(sex.na)), thres=

> ind.clones.use <- which(gainloss\$gainP >= minChanged | gainloss\$lossP>= minChanged & clones

> colorectal.na <- colorectal[ind.clones.use,sex.na, keep=TRUE]

> dat <- log2.ratios.imputed(colorectal.na)</pre>

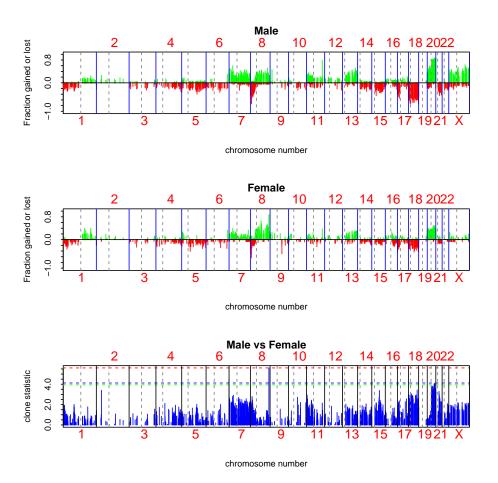


Figure 8: Frequency plots of the samples with respect to the sex groups for clones gained or lost in at least 10% of the samples

Testing association of clones with censored outcomes. Since there was no survival data available, we simulate data for a simple example to demonstrate creation and usage of basic survival object. We create an object equivalent to resT object that was created earlier. In the figure the samples are seprated into dead and alive/censored groups for ease of visualization. Nevertheless, statistic is computed and assessed for significance using proper survival object.

```
> time <- rexp(ncol(colorectal), rate = 1 / 12)</pre>
> events <- rbinom(ncol(colorectal), size = 1, prob = .5)
> surv.obj <- Surv(time,
                                 events)
> surv.obj
 [1] 3.9247017+
                5.1988040+ 4.8551209
                                          8.1572195+ 22.5084423+
                                                                  0.8149976
 [7] 32.6864907+ 5.6129255+ 7.0006015 21.5549388+ 7.1802027+
                                                                  0.2784021
[13] 26.1092680+
                 2.5408442+ 39.8711780
                                          1.3283006+ 2.5438853 15.7391636+
[19] 10.3774459
                  1.8108430
                              8.5394715
                                        18.1832044
                                                      9.3282889
                                                                 16.8230054+
[25] 12.7421092+ 15.5932252+ 22.5914185
                                          8.4289535
                                                      8.9287899
                                                                 14.0813353+
     4.3354278+
                 9.7139247+ 10.0814262 12.4875596 17.2799807+ 16.3092133+
[37] 75.2490877+ 42.2173193 10.6717370+ 8.4001867+
> stat.coxph <-
    aCGH.test(colorectal, surv.obj, test = "coxph",
          p.adjust.method = "fdr")
> stat.coxph[1:10 ,]
     index teststat
                            rawp
                                      adjp
1028
     1028 -3.240148 0.001194679 0.5497703
640
       640 -3.153093 0.001615501 0.5497703
1058
     1058 -3.051434 0.002277509 0.5497703
1029
     1029 -2.982017 0.002863562 0.5497703
144
       144 2.968494 0.002992634 0.5497703
543
       543 -2.934097 0.003345193 0.5497703
1270
     1270 -2.927760 0.003414138 0.5497703
150
            2.921159 0.003487313 0.5497703
1040
     1040 -2.914399 0.003563740 0.5497703
141
       141 2.891998 0.003828008 0.5497703
```

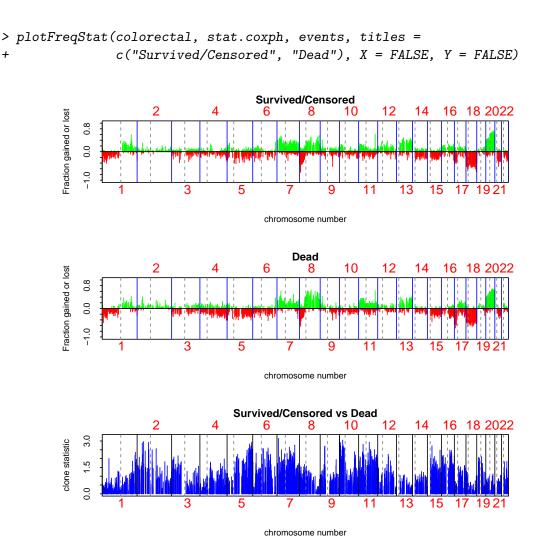


Figure 9: Frequency plots of the samples with respect to survival.

Deriving statistics and p-values for testing the linear association of age with the log2 ratios of each clone along the tumors. Here we repeat above two examples but using significance of linear regression coeffecient as a mesuare of association between genomic variable and continious outcome.

```
> age <- phenotype(colorectal)$age
> age.na <- which(!is.na(age))
> age <- age[age.na]
> colorectal.na <- colorectal[, age.na]
> stat.age <-
+ aCGH.test(colorectal.na, age, test = "linear.regression",
+ p.adjust.method = "fdr")
> stat.age[1:10 ,]
```

```
index
           teststat
                                       adjp
                             rawp
            3.259187 0.002399741 0.9952687
1735
      1735
1739
      1739
            3.184326 0.002941084 0.9952687
685
       685 -3.158061 0.003157117 0.9952687
            3.144471 0.003274723 0.9952687
1251
      1251
      1718
            3.118281 0.003513183 0.9952687
1718
1714
      1714
            3.112281 0.003570080 0.9952687
642
       642 -3.082287 0.003867826 0.9952687
639
       639 -3.012157 0.004658116 0.9952687
       643 -2.937882 0.005659632 0.9952687
643
           2.881404 0.006552898 0.9952687
1744
      1744
```

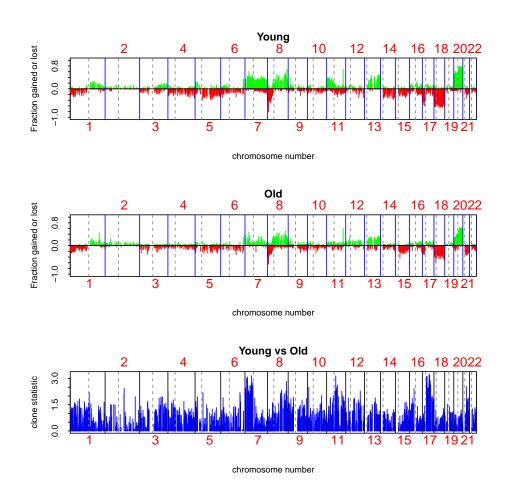


Figure 10: Frequency plots of the samples with respect to age.

Here we show example of how to create a table of results which can be later exported into other programs via *write.table*. First, Males vs Females:

- > sex <- phenotype(colorectal)\$sex</pre>
- > sex.na <- !is.na(sex)</pre>
- > index.clones.use <- which(clones.info(colorectal.na)\$Chrom < 23)</pre>
- > colorectal.na <- colorectal[index.clones.use,sex.na , keep=TRUE]</pre>
- > dat <- log2.ratios.imputed(colorectal.na)</pre>
- > resT.sex <- mt.maxT(dat, sex[sex.na], test = "t.equalvar", B = 1000)</pre>

b=10	b=20	b=30	b=40	b=50	b=60	b=70	b=80
b=110	b=120	b=130	b=140	b=150	b=160	b=170	b=
b=210	b=220	b=230	b=240	b=250	b=260	b=270	b=
b=310	b=320	b=330	b=340	b=350	b=360	b=370	b=
b=410	b=420	b=430	b=440	b=450	b=460	b=470	b=
b=510	b=520	b=530	b=540	b=550	b=560	b=570	b=
b=610	b=620	b=630	b=640	b=650	b=660	b=670	b=
b=710	b=720	b=730	b=740	b=750	b=760	b=770	b=
b=810	b=820	b=830	b=840	b=850	b=860	b=870	b=
b=910	b=920	b=930	b=940	b=950	b=960	b=970	b=

> sex.tbl <- summarize.clones(colorectal.na, resT.sex, sex[sex.na], titles = c("Male", "Femal > sex.tbl[1:5,]

	Clone	Targe	et C	hrom	kb	NumPresen	t.All	Num	Gain.All	NumLo	st.All
2	RP11-82D16	HumArray2H11_C	.9	1	2009		38		4		7
3	RP11-62M23	HumArray2H10_N3	30	1	3368		34		1		7
4	RP11-11105	HumArray2H10_B1	.8	1	4262		37		1		9
5	RP11-51B4	HumArray2H10_Q3	30	1	6069		34		0		10
6	RP11-60J11	HumArray2H10_T3	30	1	6817		35		1		7
	PropPresent	t.All PropGain.A	11	Propl	Lost.	All NumPre	esent.	Male	NumGain.	Male	
2		0.97 0.	11		0.	18		23		1	
3		0.87 0.	03		0.	21		20		1	
4		0.95 0.	03		0.	. 24		23		0	
5		0.87 0.	00		0.	.29		19		0	
6		0.90 0.	03		0.	. 20		20		0	
	NumLost.Mal	le PropPresent.M	ſale	Prop	Gain.	Male Prop	Lost.	Male	NumPrese	nt.Fe	nale
2		5 1	.00	1		0.04	(0.22			15
3		5 0	.87	•		0.05	(0.25			14
4		7 1	.00			0.00	(0.30			14
5		7	.83			0.00	(0.37			15
6		4 0	.87	•		0.00	(0.20			15
	NumGain.Fer	male NumLost.Fem	nale	Prop	Prese	ent.Female	Prop(Gain	.Female		
2		3	2			0.94	Į		0.20		
3		0	2			0.88	3		0.00		
4		1	2			0.88	3		0.07		
5		0	3			0.94	Į		0.00		
6		1	3			0.94	Ŀ		0.07		
	PropLost.Fe	emale stat	ra	wp ac	djp						
2		0.13 1.3456684	0.1	85	1						

3	0.14	1.2966513	0.214	1
4	0.14	0.7545065	0.445	1
5	0.20	1.9207531	0.066	1
6	0.20	0.5052960	0.640	1

3.12 Clustering samples

Here we cluster samples while displaying phenotypes as well as within phenotypes using chromosomes 4, 8 and 9 and display the phenotype labels, in this case, sex. We also indicate high level amplifications and 2-copy deletions with yellow and blue colors. (fig. 11).

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```
> par(mfrow=c(2,1))
> clusterGenome(colorectal.na, response = sex[sex.na],
                  titles = c("Female", "Male"),
                  byclass = FALSE, showaber = TRUE, vecchrom = c(4,8,9),
                  dendPlot = FALSE, imp = FALSE)
> clusterGenome(colorectal.na, response = sex[sex.na],
                  titles = c("Female", "Male"),
                  byclass = TRUE, showaber = TRUE, vecchrom = c(4,8,9),
                  dendPlot = FALSE, imp = FALSE)
                                     Female
                                       Male
                                                           9
                                       clone
                                      Female
                                       Male
                          4
```

Figure 11: Clustering of the samples by sex

clone