Estimating permutation p-values using MatrixEQTL

In our pipeline we first reformat the data per gene and then for each preprocessed gene run step8_MatrixEQTL script which runs multiple bootstraps.

First, load the data for MatrixEQTL. Arguments here are in the same style as original script. They give information about the chromosome on which gene is located, number of subsamples to be used for estimation (no more then total number of samples recorded in specification file), random seed, window size and which model to be used.

Specification file still will be used, since it is required at earlier steps linking in this pipeline. It is not necessary if you choose a different way to provide the path to the data.

Once initial setup is done we read relevant (multigene) data

Load genotype data. Check that there is no snps with too small variance.

Load geneinfo and design matrix. Ensure that covariates don't have too small variance. Load gene expression (already quantile normalized format provided in the example).

```
genepos_infile_name = sprintf("%s/geneInfo_prepr_%s.txt", cnt.dir, model)
geneInfo = read.table(genepos_infile_name,
                      header = T, as.is = T)

genepos = geneInfo[geneInfo$chr==sprintf("chr%s", chr),1:4]
genepos[,2] = gsub("chr", "", genepos[,2])
for(col1 in 3:4)genepos[,col1] = as.numeric(genepos[,col1])
colnames(genepos) = c("geneid", "chr", "left", "right")

genepos_file_name = sprintf("%s/genepos_%s.dat", int.dir, suff0)
colnames(snpspos) = c("snpid", "chr", "pos")
write.table(genepos[blocki,], file=genepos_file_name,
            row.names=F, col.names=T, quote=F, sep="\t")

covariates_file_name = sprintf("%s/kmat_%s.csv", int.dir, model)
covar =  read.csv(covariates_file_name, as.is=T, header=F)
covar = as.matrix(covar)

converge = 1e-4
varl = apply(covar,2,var)

updvar = which(varl<converge)
for(i in updvar){
  if(length(varl[-updvar])>0){
    correct = sqrt(mean(varl[-updvar]))/sqrt(varl[i])
  }else{
    correct = 1/sqrt(varl[i])
  }
  xm = mean(covar[,i])
  covar[,i] = xm+(covar[,i]-xm)*correct
}
cvrt = SlicedData$new()
cvrt = cvrt$CreateFromMatrix(t(covar))


SNP_file_name = sprintf("%s/SNP_%s.txt", int.dir, suff0)

write.table(g.ini, SNP_file_name, row.names=T,
            col.names=T, quote=F, sep="\t")

exprj = read.table(expression_file_name)


pvOutputThreshold = 1;
errorCovariance = numeric();

genepos_file_name = sprintf("%s/genepos_%s.dat", int.dir, suff0)
colnames(snpspos) = c("snpid", "chr", "pos")
colnames(genepos) = c("geneid", "chr", "left", "right")
write.table(genepos[blocki,], file=genepos_file_name,
            row.names=F, col.names=T, quote=F, sep="\t")

rownames(exprj) = genepos$geneid[blocki]
```

Permutation estimate rewritten as a function.

If you ran eigenMT prior to running this pipeline, you can use that result to get a better guess of effective number of tests but it can be skipped. In such case 1\4 of simple number of SNPs can be used as a proxy for initial guess of effective number of snps procedure will try to adjust it if initial guess too inconsistent based on first 100 iterations

Lets illustrate calculation of permutation p-value estimate. We take the values generated in step4_runboot.R and fit glm predicting probability of observing more extreme result (then observed in bootstrap) by log10(minimum p-value). After fitting glm, predict permutation p-value based on log10(minimum p-value) Effective number of tests will be ratio of predicted permutation p-value and minimum p-value (trimmed between 1 and number of SNPs)

```
#boots = read.csv(sprintf("%s/short_boot_pval_9_1.csv", perm.dir), as.is=T)
#eigenMT = read.csv(sprintf("%s/upd_eigenMT_9_1.csv", out.dir), as.is=T)
nperm = 100
y = me$vals$perm>nperm
pvalb = me$vals$pvalb
kp3 = (y/nperm)>=0    & (y/nperm)<=0.3
kp3a = (y/nperm)>0    & (y/nperm)<=0.3

y1 = log10(y/nperm)
x1 = log10(pvalb)
glm13 = glm(cbind(y[kp3],nperm-y[kp3])~x1[kp3], family="binomial")
summary(glm13)
```

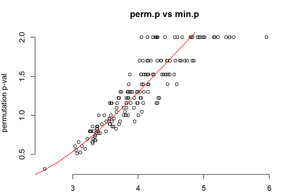
```
##
## Call:
## glm(formula = cbind(y[kp3], nperm - y[kp3]) ~ x1[kp3], family = "binomial")
##
## Deviance Residuals:
##     Min       1Q   Median       3Q      Max
## -2.4308  -0.8271  -0.1418   0.5668   2.4591
##
## Coefficients:
##             Estimate Std. Error z value Pr(>|z|)
## (Intercept)  5.33509    0.26368   20.23   <2e-16 ***
## x1[kp3]      2.07084    0.07162   28.90   <2e-16 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for binomial family taken to be 1)
##
##     Null deviance: 1394.65  on 194  degrees of freedom
## Residual deviance:  175.93  on 193  degrees of freedom
## AIC: 736.96
##
## Number of Fisher Scoring iterations: 5
```

```
xval = log10(eigenMT$p.value)
pred.perm = logit(glm13$coef[1]+glm13$coef[2]*xval)
c(xval, pred.perm)
```

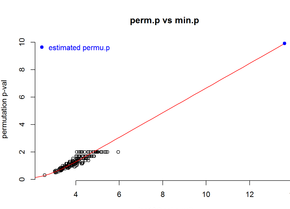
```
##              (Intercept)
## -1.360419e+01  1.221040e-10
```

```
xlim = range(-c(x1, xval))
ylim = range(-log10(y/nperm))
ylim[2] = -log10(pred.perm)

plot(-x1, -log10(y/nperm), xlab="-log10(boot p-val)",
     ylab="permutation p-val", bty="n", main="perm.p vs min.p")
#o = order(x1[kp3])
#xf = x1[kp3][o]
#yf = glm13$fitted.values[o]
#lines(-xf, -log10(yf), col="red")
fit = seq(0, min(c(xval,x1)), length.out=50)
pred.perm0 = logit(glm13$coef[1]+glm13$coef[2]*fit)
lines(-fit, -log10(pred.perm0), col="red")
```




```
fit = seq(0, xval, length.out=50)
pred.perm0 = logit(glm13$coef[1]+glm13$coef[2]*fit)
plot(-x1, -log10(y/nperm), xlab="-log10(boot p-val)",
     ylab="permutation p-val", bty="n", main="perm.p vs min.p",
     xlim=xlim,ylim=ylim)
lines(-fit, -log10(pred.perm0), col="red")
points(-xval, -log10(pred.perm), col="blue", cex=1, pch=19)
legend("topleft", "estimated permu.p", text.col="blue", pch=19,
       col="blue", bty="n")
```



Or using less extreme effect size: Lets illustrate calculation of permutation p-value estimate. We take the values generated in step4_runboot.R and fit glm predicting probability of observing more extreme result (then observed in bootstrap) by log10(minimum p-value). After fitting glm, predict permutation p-value based on log10(minimum p-value) Effective number of tests will be ratio of predicted permutation p-value and minimum p-value (trimmed between 1 and number of SNPs)

```
nperm = 100
eigenMT = me$$umm
y = me$$vals$perm>nperm
pvalb = me$$vals$pvalb
kp3 = (y/nperm)>=0    & (y/nperm)<=0.3
kp3a = (y/nperm)>0    & (y/nperm)<=0.3

y1 = log10(y/nperm)
x1 = log10(pvalb)
glm13 = glm(cbind(y[kp3],nperm-y[kp3])~x1[kp3], family="binomial")
summary(glm13)
```

```
##
## Call:
## glm(formula = cbind(y[kp3], nperm - y[kp3]) ~ x1[kp3], family = "binomial")
##
## Deviance Residuals:
##     Min       1Q   Median       3Q      Max
## -3.6001  -0.8029   0.0196   0.6533   2.6110
##
## Coefficients:
##             Estimate Std. Error z value Pr(>|z|)
## (Intercept)  5.61056    0.28387   19.77   <2e-16 ***
## x1[kp3]      2.15272    0.07981   27.25   <2e-16 ***
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
##
## (Dispersion parameter for binomial family taken to be 1)
##
##     Null deviance: 1281.24  on 159  degrees of freedom
## Residual deviance:  164.29  on 158  degrees of freedom
## AIC: 656.12
##
## Number of Fisher Scoring iterations: 5
```

```
xval = log10(eigenMT$p.value)
pred.perm = logit(glm13$coef[1]+glm13$coef[2]*xval)
c(xval, pred.perm)
```

```
##              (Intercept)
## -5.067591473  0.004073853
```

```
xlim = range(-c(x1, xval))
kp = y!=0
ylim = range(-log10(y/nperm)[kp])
ylim[2] = max(c(ylim[2], -log10(pred.perm)))

plot(-x1, -log10(y/nperm), xlab="-log10(boot p-val)",
     ylab="permutation p-val", bty="n", main="perm.p vs min.p", ylim=ylim)
o = order(x1[kp])
xf = x1[kp][o]
yf = glm13$fitted.values[o]
lines(-xf, -log10(yf), col="red")
points(-xval, -log10(pred.perm), col="blue", cex=1, pch=19)

fit = seq(0, min(c(xval,x1)), length.out=50)
pred.perm0 = logit(glm13$coef[1]+glm13$coef[2]*fit)
lines(-fit, -log10(pred.perm0), col="red")
```