MSMB-Chapter2-Statistical Modelling

Aleeza Gerstein

2019-09-16

Chapter 2: Statistical Modeling

A simple example of statistical modelling

```
load(url("http://bios221.stanford.edu/data/e100.RData"))
e99 = e100[-which.max(e100)]
barplot(table(e99), space = 0.8, col = "chartreuse4")
```

```
library("vcd")
```

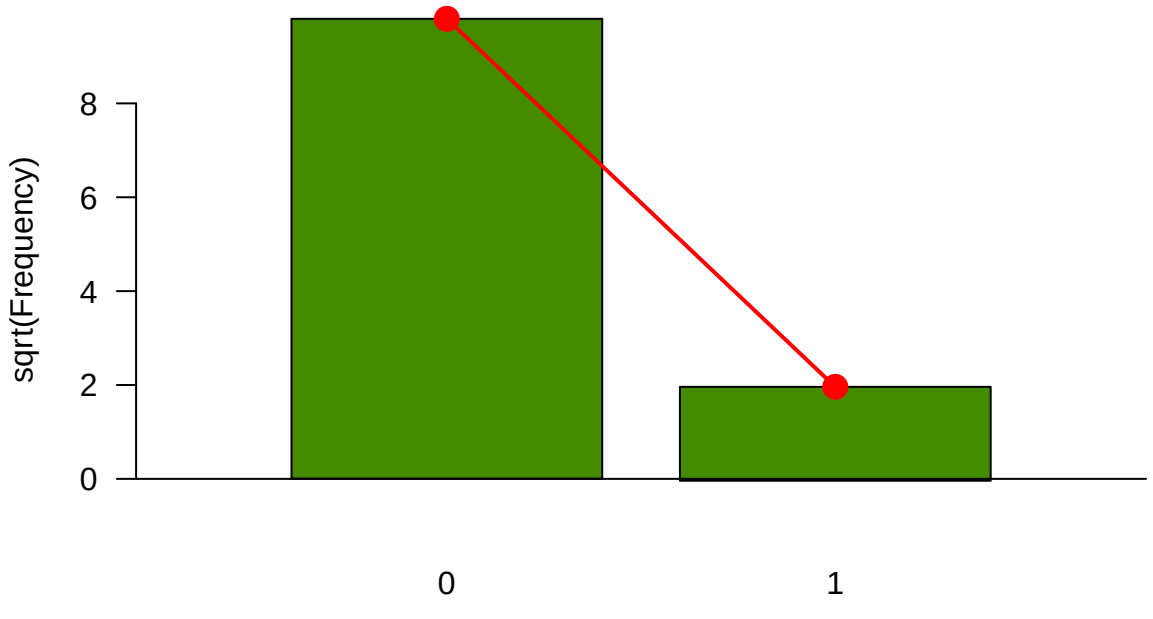
```
## Loading required package: grid
```

```
gf1 = goodfit(e99, "poisson")
rootogram(gf1, xlab = "", rect_gp = gpar(fill = "chartreuse4"))
```

We will learn later what this is?

Question 1: To calibrate what such a plot looks like a known poisson variable, use 'rpois' and $\lambda = 0.05$ to generate 100 Poisson distributed numbers and draw their rootogram

```
pv <- rpois(100, 0.05)
gf_pv = goodfit(pv, "poisson")
rootogram(gf_pv, xlab = "", rect_gp = gpar(fill = "chartreuse4"))
```



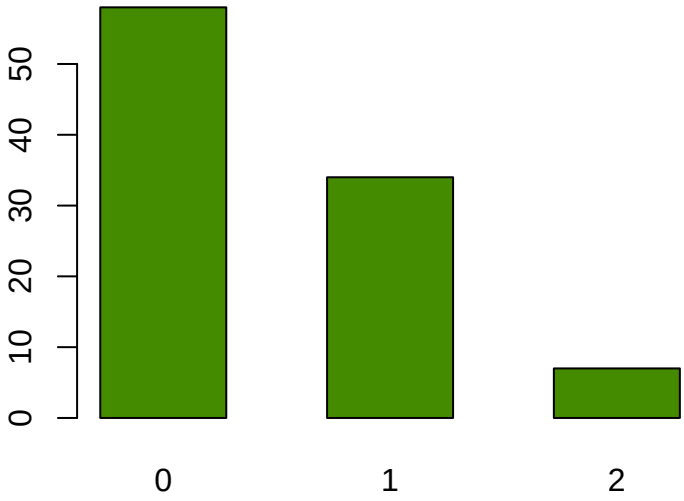


Figure 1: The observed distribution of the epitope data without the outlier.

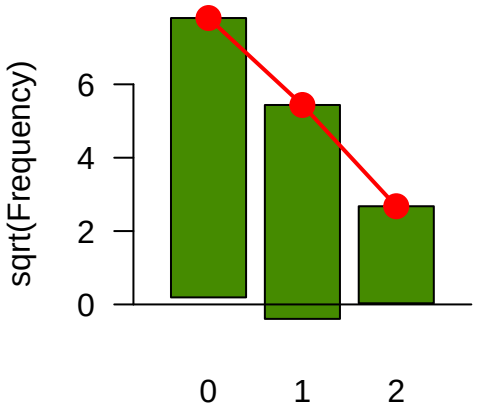


Figure 2: Rootogram showing the square root of the theoretical values as red dots and the square root of the observed frequencies as drop down rectangles. (We'll see a bit below how the `goodfit` function decided which λ to use.)

Question 2: Repeat the simulation with different values of λ. Can you find one that gives cout close to the observed count by trial and error?

```
table(e100)
```

```
## e100
##  0  1  2  7
## 58 34  7  1
```

```
pv0.5 <- rpois(100, 0.5)
table(pv0.5)
```

```
## pv0.5
##  0  1  2  3  4
## 61 32  5  1  1
```

```
loglikelihood  =  function(lambda, data = e100) {
  sum(log(dpois(data, lambda)))
}
```

```
lambdas = seq(0.05, 0.95, length = 100)
loglik = vapply(lambdas, loglikelihood, numeric(1))
#this is the same as
#loglik = sapply(lambdas, loglikelihood)
plot(lambdas, loglik, type = "l", col = "red", ylab = "", lwd = 2,
     xlab = expression(lambda))
m0 = mean(e100)
abline(v = m0, col = "blue", lwd = 2)
abline(h = loglikelihood(m0), col = "purple", lwd = 2)
```

```
m0
```

```
## [1] 0.55
```

```
gf  =  goodfit(e100, "poisson")
names(gf)
```

```
## [1] "observed" "count"    "fitted"   "type"     "method"   "df"
## [7] "par"
```

```
gf$par
```

```
## $lambda
## [1] 0.55
```

```
cb  =  c(rep(0, 110), rep(1, 10))
table(cb)
```

```
## cb
##   0   1
## 110  10
```

```
probs  =  seq(0, 0.3, by = 0.005)
likelihood = dbinom(sum(cb), prob = probs, size = length(cb))
plot(probs, likelihood, pch = 16, xlab = "probability of success",
     ylab = "likelihood", cex=0.6)
```

```
probs[which.max(likelihood)]
```

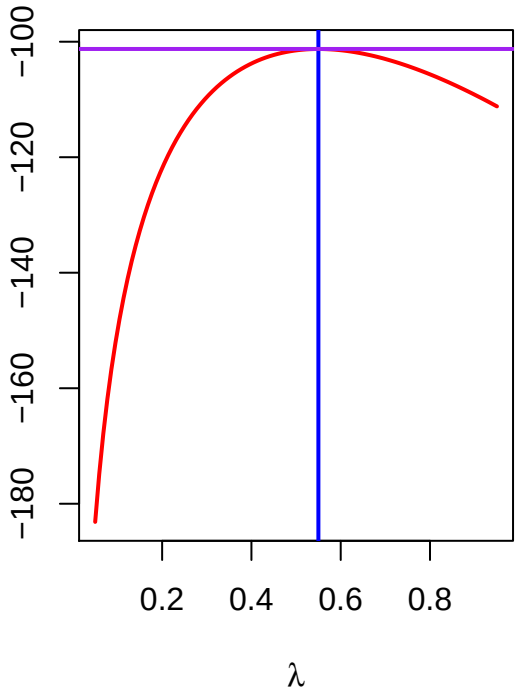


Figure 3: The red curve is the log-likelihood function. The vertical line shows the value of `m` (the mean) and the horizontal line the log-likelihood of `m`. It looks like `m` maximizes the likelihood.

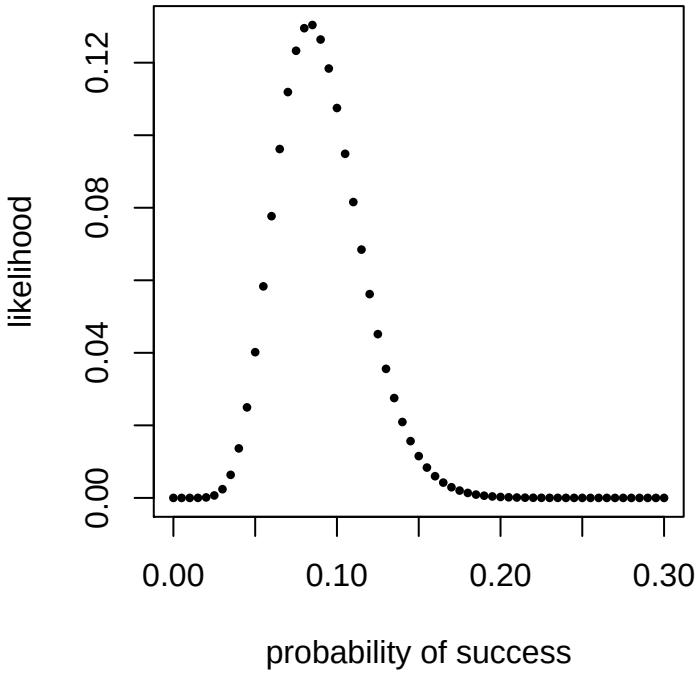


Figure 4: Plot of the likelihood as a function of the probabilities. The likelihood is a function on $[0, 1]$; here we have zoomed into the range of $[(ref : likely1 - 1), (ref : likely1 - 2)]$, as the likelihood is practically zero for larger values of p.

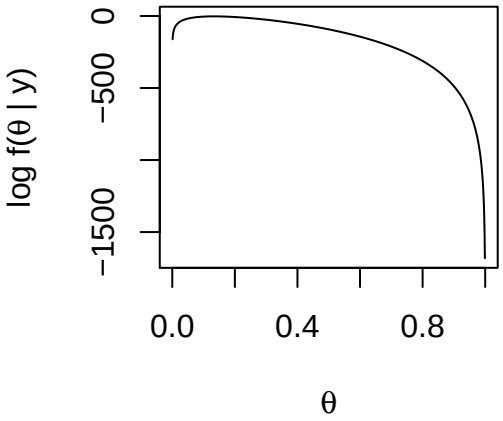


Figure 5: Plot of the log likelihood function for $n = 300$ and $y = 40$.

```
## [1] 0.085
```

```
stopifnot(abs(probs[which.max(likelihood)]-1/12) < diff(probs[1:2]))
```

```
loglikelihood = function(theta, n = 300, k = 40) {
  115 + k * log(theta) + (n - k) * log(1 - theta)
}
```

```
thetas = seq(0, 1, by = 0.001)
plot(thetas, loglikelihood(thetas), xlab = expression(theta),
     ylab = expression(paste("log f(", theta, " | y)")),type = "l")
```

```
library("Biostrings")
```

```
## Loading required package: BiocGenerics
```

```
## Loading required package: parallel
```

```
##
## Attaching package: 'BiocGenerics'
```

```
## The following objects are masked from 'package:parallel':
##
##     clusterApply, clusterApplyLB, clusterCall, clusterEvalQ,
##     clusterExport, clusterMap, parApply, parCapply, parLapply,
##     parLapplyLB, parRapply, parSapply, parSapplyLB
```

```
## The following objects are masked from 'package:stats':
##
##     IQR, mad, sd, var, xtabs
```

```
## The following objects are masked from 'package:base':
##
##     anyDuplicated, append, as.data.frame, basename, cbind,
##     colnames, dirname, do.call, duplicated, eval, evalq, Filter,
##     Find, get, grep, grepl, intersect, is.unsorted, lapply, Map,
##     mapply, match, mget, order, paste, pmax, pmax.int, pmin,
##     pmin.int, Position, rank, rbind, Reduce, rownames, sapply,
##     setdiff, sort, table, tapply, union, unique, unsplit, which,
##     which.max, which.min
```

```
## Loading required package: S4Vectors
```

```
## Loading required package: stats4
##
## Attaching package: 'S4Vectors'
## The following object is masked from 'package:base':
##
##     expand.grid
## Loading required package: IRanges
##
## Attaching package: 'IRanges'
## The following object is masked from 'package:vcd':
##
##     tile
## Loading required package: XVector
##
## Attaching package: 'Biostrings'
## The following object is masked from 'package:base':
##
##     strsplit
```

```
staph = readDNAStringSet("http://bios221.stanford.edu/data/staphsequence.ffn.txt", "fasta")
staph[1]
```

```
##   A DNAStringSet instance of length 1
##     width seq                                               names
## [1]  1362 ATGTCGGAAAAAGAAATTTGG...AAGAAATAAGAAATGTATAA lcl|NC_002952.2_c...
```

```
letterFrequency(staph[[1]], letters = "ACGT", OR = 0)
```

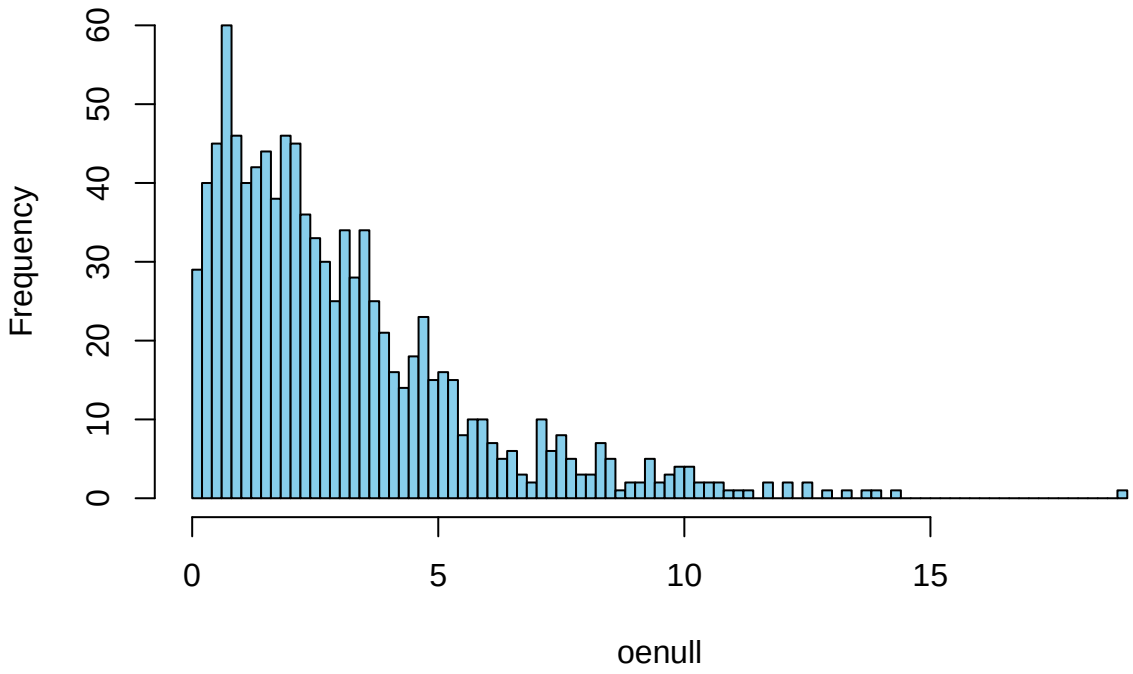
```
##   A   C   G   T
## 522 219 229 392
```

Question 2.9: Following a similar procedure to Exercise 1.8, test whether the nucleotides are equally distributed across the four possibilities for this first gene.

```
lf <- letterFrequency(staph[[1]], letters = "ACGT", OR = 0)
t(rmultinom(1, sum(lf), p = rep(1/4, 4)))
```

```
##      [,1] [,2] [,3] [,4]
## [1,]  334  327  379  322
```

```
oestat <- function(o, e) {
  sum((e-o)^2 / e)
}

B = 1000
n = sum(lf)
expected = rep(n / 4, 4)

oenull <- replicate(B,
  oestat(e = expected, o = rmultinom(1, n, p = rep(1/4, 4))))
hist(oenull, breaks = 100, col = "skyblue", main="")
```




```
oestat(e = expected, o = t(lf))
```

```
## [1] 184.4023
```

```
dmultinom(lf, prob = rep(0.25, 4))
```

```
## [1] 9.026662e-45
```

```
letterFrq = vapply(staph, letterFrequency, FUN.VALUE = numeric(4),
                   letters = "ACGT", OR = 0)
colnames(letterFrq) = paste0("gene", seq(along = staph))
tab10 = letterFrq[, 1:10]
computeProportions = function(x) { x/sum(x) }
prop10 = apply(tab10, 2, computeProportions)
round(prop10, digits = 2)
```

```
##   gene1 gene2 gene3 gene4 gene5 gene6 gene7 gene8 gene9 gene10
## A  0.38  0.36  0.35  0.37  0.35  0.33  0.33  0.34  0.38   0.27
## C  0.16  0.16  0.13  0.15  0.15  0.15  0.16  0.16  0.14   0.16
## G  0.17  0.17  0.23  0.19  0.22  0.22  0.20  0.21  0.20   0.20
## T  0.29  0.31  0.30  0.29  0.27  0.30  0.30  0.29  0.28   0.36
```

```
p0 = rowMeans(prop10)
p0
```

```
##         A         C         G         T
## 0.3470531 0.1518313 0.2011442 0.2999714
```

Outer probaqbility

```
cs = colSums(tab10)
cs
```

```
##  gene1  gene2  gene3  gene4  gene5  gene6  gene7  gene8  gene9 gene10
##   1362   1134    246   1113   1932   2661    831   1515   1287    696
```

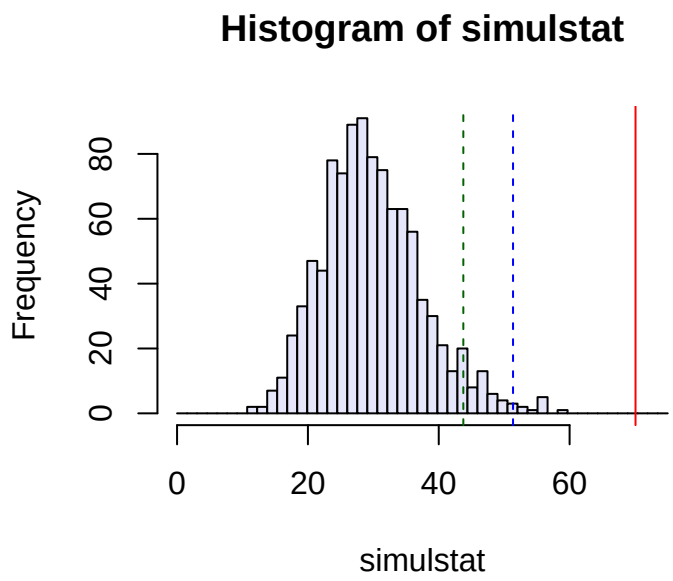


Figure 6: Histogram of `simulstat`. The value of `S1` is marked by the vertical red line, those of the 0.95 and 0.99 quantiles (see next section) by the dotted lines.

```
expectedtab10 = outer(p0, cs, FUN = "*")
round(expectedtab10)
```

```
##   gene1 gene2 gene3 gene4 gene5 gene6 gene7 gene8 gene9 gene10
## A   473   394    85   386   671   924   288   526   447    242
## C   207   172    37   169   293   404   126   230   195    106
## G   274   228    49   224   389   535   167   305   259    140
## T   409   340    74   334   580   798   249   454   386    209
```

```
randomtab10 = sapply(cs, function(s) { rmultinom(1, s, p0) } )
all(colSums(randomtab10) == cs)
```

```
## [1] TRUE
```

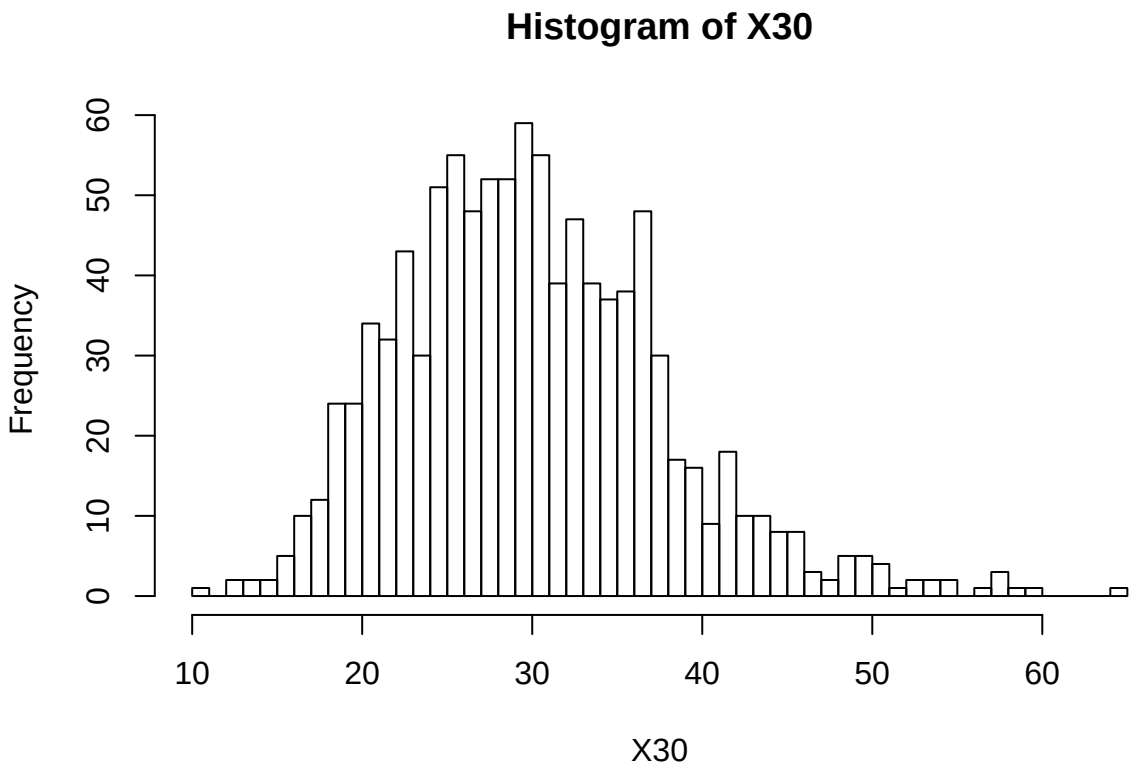
```
stat = function(obsvd, exptd = 20 * pvec) {
  sum((obsvd - exptd)^2 / exptd)
}
B = 1000

simulstat = replicate(B, {
  randomtab10 = sapply(cs, function(s) { rmultinom(1, s, p0) })
  stat(randomtab10, expectedtab10)
})

S1 = stat(tab10, expectedtab10)
sum(simulstat >= S1)
```

```
## [1] 0
```

```
hist(simulstat, col = "lavender", breaks = seq(0, 75, length.out=50))
abline(v = S1, col = "red")
abline(v = quantile(simulstat, probs = c(0.95, 0.99)),
       col = c("darkgreen", "blue"), lty = 2)
```

Question 2.10 a) Compare the 'simulstat' values and 1000 randomly generated χ^2_{30} random numbers by displaying them in histograms with 50 bins each.

```
X30 <- rchisq(1000, 30)
hist(X30, breaks =50)
```



b) Compute the quanties of the `simulstat` values and compare them to those of the χ^2_{30} distribution.}

```
qs = ppoints(100)
squant <- quantile(simulstat, qs)
quant <- quantile(qchisq(qs, df = 30), qs)
plot(squant, quant)
abline(0, 1)
```

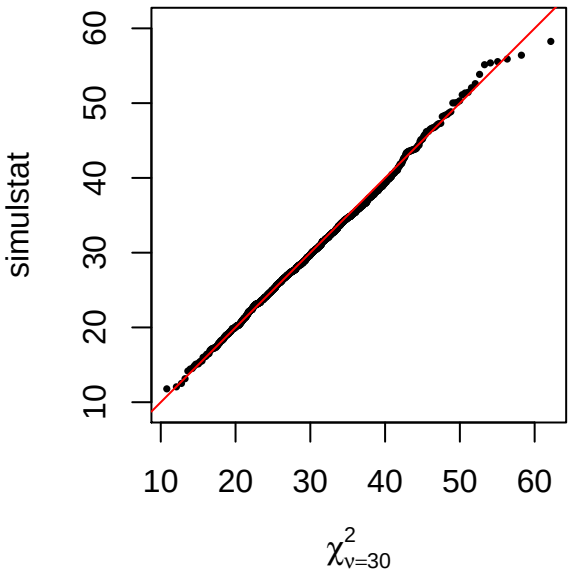
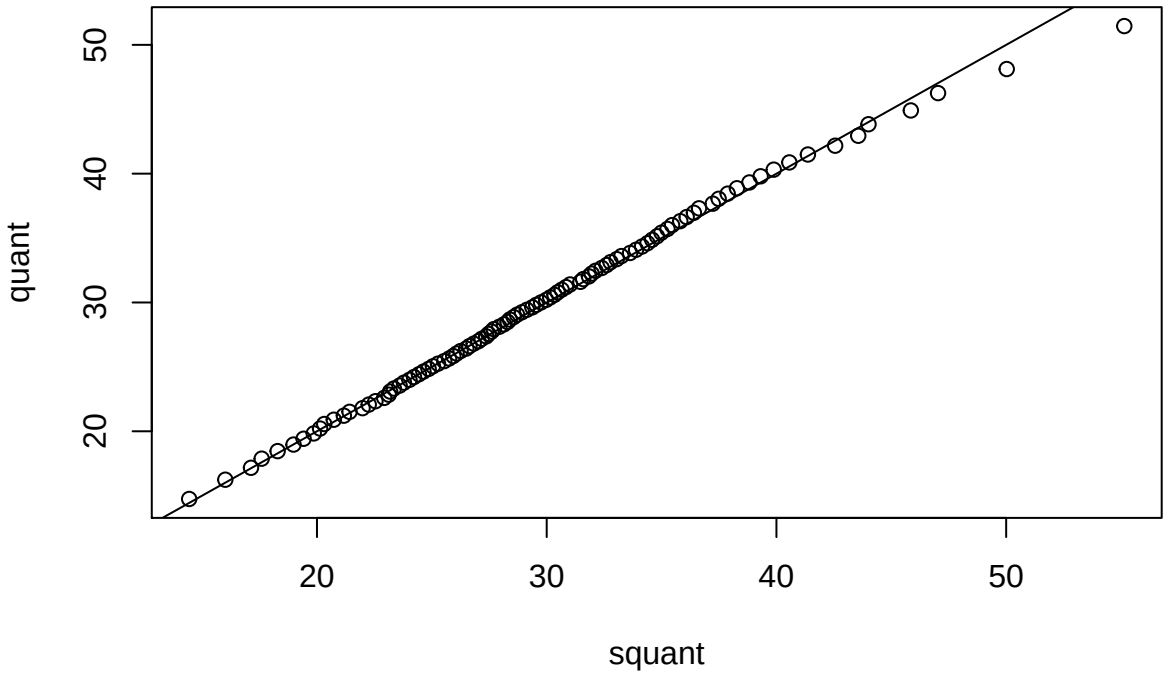


Figure 7: Our simulated statistic's distribution compared to χ^2_{30} using a QQ-plot, which shows the theoretical **quantiles** for the χ^2_{30} distribution on the horizontal axis and the sampled ones on the vertical axis.




```
qqplot(qchisq(ppoints(B), df = 30), simulstat, main = "",
       xlab = expression(chi[nu==30]^2), asp = 1, cex = 0.5, pch = 16)
abline(a = 0, b = 1, col = "red")
```

Compute the p-value that the counts are distributed with multinomial probability $p_A = 0.35$, $p_C = 0.15$, $p_G = 0.2$ $p_T = 0.3$ we observe a value as high as 70.1:

```
1 - pchisq(S1, df = 30)
```

```
## [1] 4.74342e-05
```

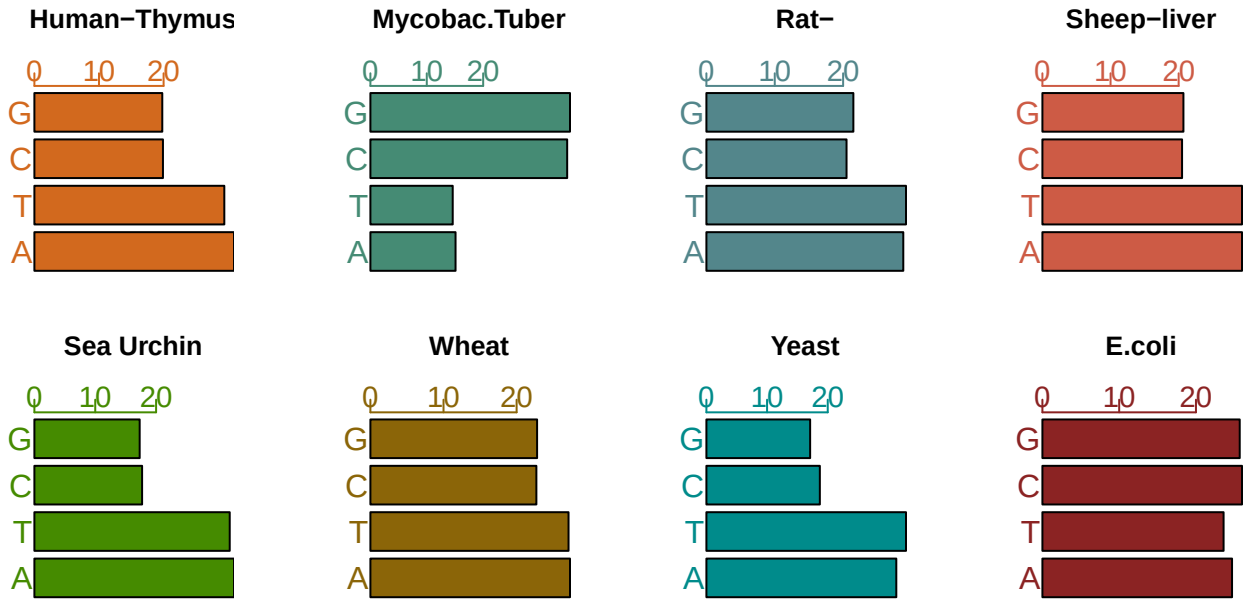


Figure 8: Barplots for the different rows in `ChargaffTable`. Can you spot the pattern?

2.7 Chagaff's Rule

Chagaff published percentages of the masses of different organisms for nucleotides:

```
load(url("http://bios221.stanford.edu/data/ChargaffTable.RData"))
ChargaffTable
```

```
##                   A    T    C    G
## Human-Thymus   30.9 29.4 19.9 19.8
## Mycobac.Tuber  15.1 14.6 34.9 35.4
## Rat-           28.8 29.2 20.5 21.5
## Sheep-liver    29.3 29.3 20.5 20.7
## Sea Urchin     32.8 32.1 17.7 17.3
## Wheat          27.3 27.1 22.7 22.8
## Yeast          31.3 32.9 18.7 17.1
## E.coli         24.7 23.6 26.0 25.7
```

Question 2.13: Do these data seem to come from equally likely multinomial categories? Can you suggest an alternative pattern? Can you do a quantitative analysis of the pattern?

No. G = C = 0.2; A = T= 0.3

Explain this why 'statChf'

```
statChf = function(x){
  sum((x[, "C"] - x[, "G"])^2 + (x[, "A"] - x[, "T"])^2)
}
chfstat = statChf(ChargaffTable)
permstat = replicate(100000, {
  permuted = t(apply(ChargaffTable, 1, sample))
  colnames(permuted) = colnames(ChargaffTable)
  statChf(permuted)
})
```

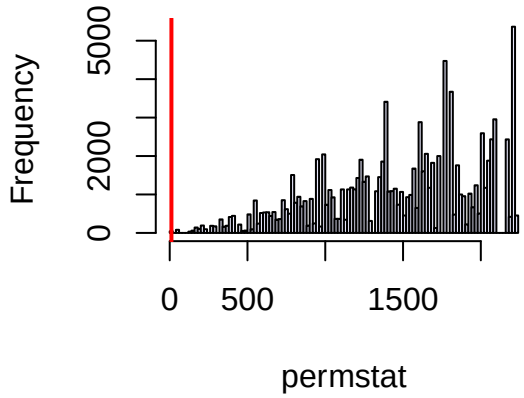


Figure 9: Histogram of our statistic `statChf` computed from simulations using per-row permutations of the columns. The value it yields for the observed data is shown by the red line.

```
pChf = mean(permstat <= chfstat)
pChf
```

```
## [1] 0.00015
```

```
hist(permstat, breaks = 100, main = "", col = "lavender")
abline(v = chfstat, lwd = 2, col = "red")
```

Question 2.14: When comparing 'pChf' we only looked at the values in the null distribution smaller than the observed value. Why did we do this in a one-sided way here?

2.7.1 Two categorical variables

```
load(url("http://bios221.stanford.edu/data/Deuteranopia.RData"))
Deuteranopia
```

```
##           Men Women
## Deute      19     2
## NonDeute 1981  1998
```

```
chisq.test(Deuteranopia)
```

```
##
##  Pearson's Chi-squared test with Yates' continuity correction
##
## data:  Deuteranopia
## X-squared = 12.255, df = 1, p-value = 0.0004641
```

2.7.2 A special multinomial: Hardy-Weinberg equilibrium

```
library("HardyWeinberg")
```

```
## Loading required package: mice
```

```
## Loading required package: lattice
```

```
##
## Attaching package: 'mice'
```

```
## The following objects are masked from 'package:IRanges':
##
##     cbind, rbind
```

```
## The following objects are masked from 'package:S4Vectors':
##
##     cbind, rbind
```

```
## The following objects are masked from 'package:BiocGenerics':
##
##     cbind, rbind
```

```
## The following objects are masked from 'package:base':
##
##     cbind, rbind
```

```
## Loading required package: Rsolnp
```

```
data("Mourant")
Mourant[214:216,]
```

```
##     Population    Country Total  MM  MN  NN
## 214    Oceania Micronesia   962 228 436 298
## 215    Oceania Micronesia   678  36 229 413
## 216    Oceania     Tahiti   580 188 296  96
```

```
nMM = Mourant$MM[216]
nMN = Mourant$MN[216]
nNN = Mourant$NN[216]
loglik = function(p, q = 1 - p) {
  2 * nMM * log(p) + nMN * log(2*p*q) + 2 * nNN * log(q)
}
xv = seq(0.01, 0.99, by = 0.01)
yv = loglik(xv)
plot(x = xv, y = yv, type = "l", lwd = 2,
     xlab = "p", ylab = "log-likelihood")
imax = which.max(yv)
abline(v = xv[imax], h = yv[imax], lwd = 1.5, col = "blue")
abline(h = yv[imax], lwd = 1.5, col = "purple")
```

```
phat  =  af(c(nMM, nMN, nNN))
phat
```

```
## [1] 0.5793103
```

```
pMM   =  phat^2
qhat  =  1 - phat
```

```
pHW = c(MM = phat^2, MN = 2*phat*qhat, NN = qhat^2)
sum(c(nMM, nMN, nNN)) * pHW
```

```
##       MM       MN       NN
## 194.6483 282.7034 102.6483
```

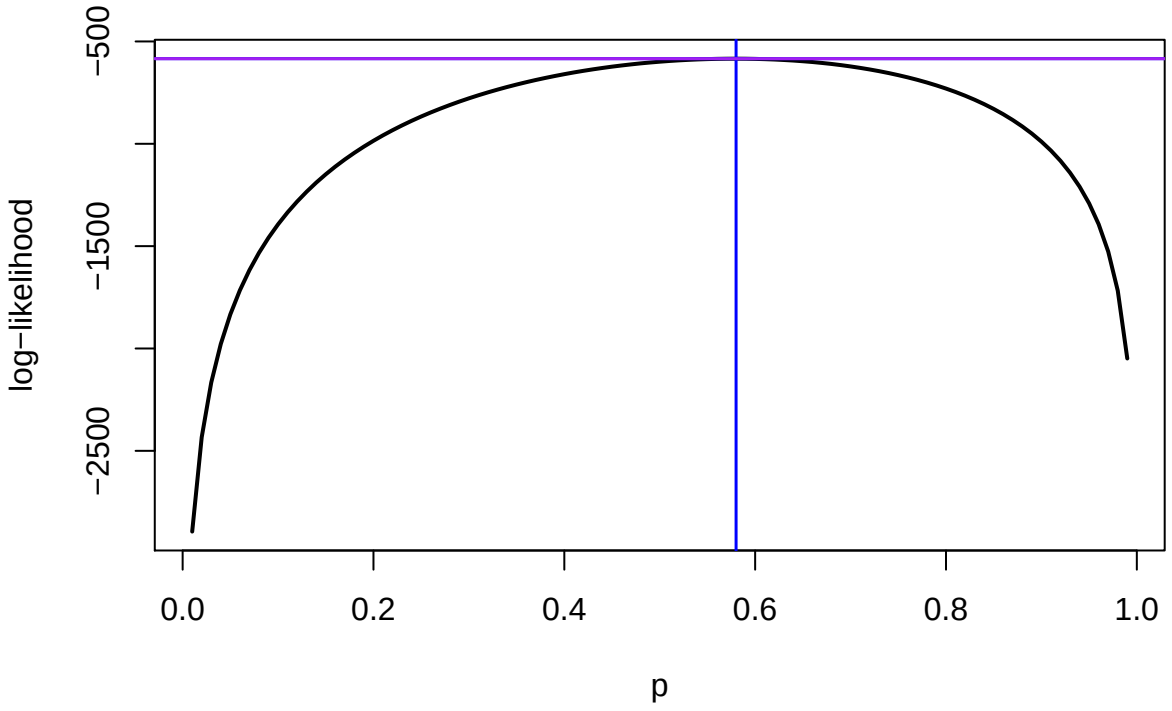


Figure 10: Plot of the log-likelihood for the (ref:chap2-r-HardyWeinberg-1-1) data.

```
pops = c(1, 69, 128, 148, 192)
genotypeFrequencies = as.matrix(Mourant[, c("MM", "MN", "NN")])
HWTernaryPlot(genotypeFrequencies[pops, ],
              markerlab = Mourant$Country[pops],
              alpha = 0.0001, curvecols = c("red", rep("purple", 4)),
              mcex = 0.75, vertex.cex = 1)
```

Question 2.16: Make the ternary plot as in the code above then add the other data points to it. What do you notice? You can back up your discussion with the 'HWChisq' function

```
HWTernaryPlot(genotypeFrequencies[-pops, ],
              markerlab = Mourant$Country[-pops],
              alpha = 0.0001, curvecols = c("red", rep("purple", 4)),
              mcex = 0.75, vertex.cex = 1)
```

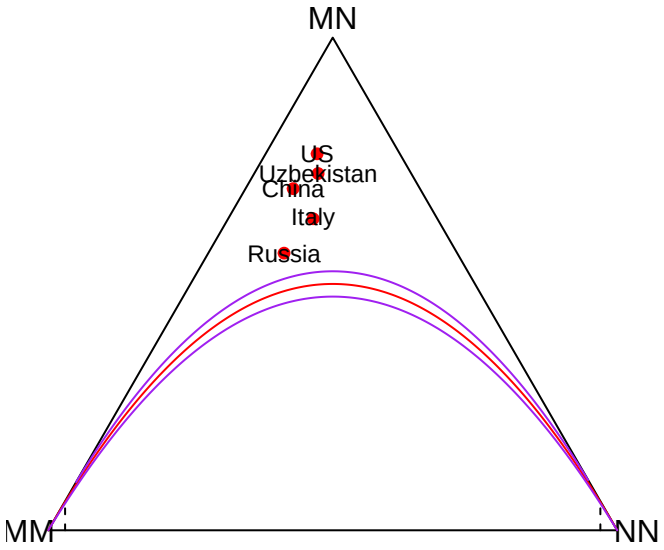
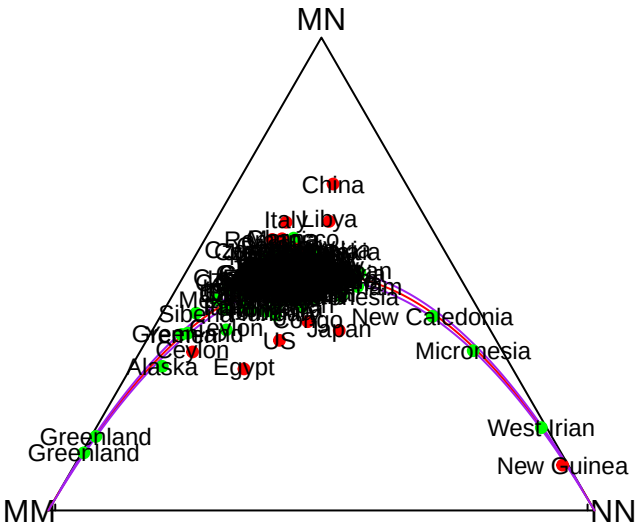


Figure 11: This **de Finetti plot** shows the points as barycenters of the three genotypes using the frequencies as weights on each of the corners of the triangle. The Hardy-Weinberg model is the red curve, the acceptance region is between the two purple lines. We see that the US is the furthest from being in HW equilibrium.




```
unique(Mourant$Population)
```

```
## [1] USSR    Eskimos Jews    Europe  Turkey  Asia    Africa  America Oceania
## Levels: Africa America Asia Eskimos Europe Jews Oceania Turkey USSR
```

```
t <- apply(genotypeFrequencies, 1, function(x) HWChisq(x)$pval)
```

```
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  144.1748 DF =  1 p-value =  3.25363e-33 D =  220.9169 f =  -0.1589235
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.235995 DF =  1 p-value =  0.07203652 D =  -7.715232 f =  0.07737265
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.3464892 DF =  1 p-value =  0.5561073 D =  -5.506968 f =  0.01392382
```

```
## Warning in HWChisq(x): Expected counts below 5: chi-square approximation
## may be incorrect
```

```
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.01751215 DF =  1 p-value =  0.8947205 D =  -0.693761 f =  0.01535081

## Warning in HWChisq(x): Expected counts below 5: chi-square approximation
## may be incorrect

## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.04787744 DF =  1 p-value =  0.8267987 D =  0.1177267 f =  -0.00262301
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.008482587 DF =  1 p-value =  0.9266178 D =  0.768 f =  -0.008326828
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  2.030071 DF =  1 p-value =  0.1542134 D =  16.0805 f =  -0.03262297
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  34.79255 DF =  1 p-value =  3.667752e-09 D =  -117.9812 f =  0.07307685
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  6.900338 DF =  1 p-value =  0.008617946 D =  35.78133 f =  -0.04846983
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  13.36419 DF =  1 p-value =  0.0002564751 D =  -90.71677 f =  0.03670936
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.0004297542 DF =  1 p-value =  0.9834606 D =  0.5661652 f =  -0.001165108
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5446964 DF =  1 p-value =  0.4604929 D =  10.58073 f =  -0.01374264
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.6087732 DF =  1 p-value =  0.4352501 D =  6.47025 f =  -0.02619219
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.355125 DF =  1 p-value =  0.06699611 D =  12.00037 f =  -0.07295075
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1464166 DF =  1 p-value =  0.7019836 D =  6.423477 f =  -0.006394214
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  5.153911 DF =  1 p-value =  0.02319407 D =  12.968 f =  -0.1045537
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1590772 DF =  1 p-value =  0.6900075 D =  3.65521 f =  -0.01315558
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  75.97485 DF =  1 p-value =  2.87301e-18 D =  153.7947 f =  -0.1222334
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.01592902 DF =  1 p-value =  0.8995655 D =  -1.781018 f =  0.003542334
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.0002437838 DF =  1 p-value =  0.9875427 D =  -0.6262961 f =  0.0004839528
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.003667017 DF =  1 p-value =  0.9517129 D =  -1.029919 f =  0.00211036
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.005589011 DF =  1 p-value =  0.9404059 D =  1.588852 f =  -0.001482261
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.3807199 DF =  1 p-value =  0.5372182 D =  4.707071 f =  -0.02249035
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.04590264 DF =  1 p-value =  0.8303529 D =  1.893122 f =  -0.008719768
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.871296 DF =  1 p-value =  0.1713267 D =  -8 f =  0.05925926
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1104575 DF =  1 p-value =  0.7396239 D =  -2.946429 f =  0.01176471
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.571377 DF =  1 p-value =  0.2100071 D =  11.33655 f =  -0.03535343
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.941419 DF =  1 p-value =  0.04711102 D =  -51.06031 f =  0.01934059
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
```

```
## Chi2 =  1.982894 DF =  1 p-value =  0.1590859 D =  11.401 f =  -0.04604623
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5305828 DF =  1 p-value =  0.4663619 D =  -6.252544 f =  0.02357204
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.01190939 DF =  1 p-value =  0.9130992 D =  -1.174676 f =  0.005228342
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.3854094 DF =  1 p-value =  0.5347223 D =  -5.138425 f =  0.02162287
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  6.180854 DF =  1 p-value =  0.01291399 D =  -20.439 f =  0.07700075
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.6971411 DF =  1 p-value =  0.403746 D =  -26.49561 f =  0.006695891
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.01093373 DF =  1 p-value =  0.9167214 D =  0.96 f =  -0.007326007
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.0007832292 DF =  1 p-value =  0.9776731 D =  -0.6210543 f =  0.001591953
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.0004957161 DF =  1 p-value =  0.9822368 D =  -0.0572123 f =  6.037873e-05
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  4.619097 DF =  1 p-value =  0.03161787 D =  37.45367 f =  -0.03111942
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.8322854 DF =  1 p-value =  0.3616125 D =  23.0096 f =  -0.009274361
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  5.551651 DF =  1 p-value =  0.01846327 D =  19.49032 f =  -0.07382427
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.78886 DF =  1 p-value =  0.05159477 D =  15.69025 f =  -0.06306123
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  2.028393 DF =  1 p-value =  0.1543837 D =  10.35281 f =  -0.05224809
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.4135673 DF =  1 p-value =  0.5201644 D =  -6.708544 f =  0.01710453
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.2455073 DF =  1 p-value =  0.6202564 D =  -5.8795 f =  0.01183285
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.707777 DF =  1 p-value =  0.05415947 D =  39.04728 f =  -0.02415615
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5879373 DF =  1 p-value =  0.4432179 D =  6.13385 f =  -0.02715701
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.000826124 DF =  1 p-value =  0.9770701 D =  0.1765782 f =  -0.000652005
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1451798 DF =  1 p-value =  0.7031849 D =  -3.932394 f =  0.01117372
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.07615999 DF =  1 p-value =  0.7825703 D =  -7.799917 f =  0.002646911
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.174822 DF =  1 p-value =  0.6758614 D =  -3.05279 f =  0.01849584
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.92292 DF =  1 p-value =  0.1655351 D =  -10.52933 f =  0.04807269
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  2.139793 DF =  1 p-value =  0.1435213 D =  -21.06248 f =  0.02606541
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.6048145 DF =  1 p-value =  0.4367469 D =  12.35813 f =  -0.0128244
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.471217 DF =  1 p-value =  0.2251537 D =  16.13448 f =  -0.02354142
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  8.692228 DF =  1 p-value =  0.003195697 D =  17.30815 f =  -0.1289806
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
```

```
## Chi2 =  0.70793 DF =  1 p-value =  0.4001319 D =  11.41815 f =  -0.01640346
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.9691839 DF =  1 p-value =  0.3248838 D =  11.28345 f =  -0.02267043
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.328076 DF =  1 p-value =  0.2491478 D =  34.83068 f =  -0.009553988
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.03908429 DF =  1 p-value =  0.8432818 D =  -6.318333 f =  0.001716324
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1897645 DF =  1 p-value =  0.6631127 D =  -3.034856 f =  0.01989615
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  11.89061 DF =  1 p-value =  0.0005641774 D =  -20.09702 f =  0.1493125
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.088042 DF =  1 p-value =  0.07886967 D =  -10.24705 f =  0.08084648
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.801502 DF =  1 p-value =  0.179531 D =  9.369813 f =  -0.05116476
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.711511 DF =  1 p-value =  0.05403845 D =  21.50579 f =  -0.0435345
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  7.355671 DF =  1 p-value =  0.006685156 D =  -19.34719 f =  0.09778854
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.002667343 DF =  1 p-value =  0.9588105 D =  -0.9848485 f =  0.001680672
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.9070016 DF =  1 p-value =  0.3409112 D =  7.372165 f =  -0.03224526
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  2.438673 DF =  1 p-value =  0.1183763 D =  9.881667 f =  -0.06626728
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  84.42257 DF =  1 p-value =  3.995607e-20 D =  78.37457 f =  -0.2706067
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.05827417 DF =  1 p-value =  0.8092448 D =  2.271593 f =  -0.008952853
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  48.05815 DF =  1 p-value =  4.137647e-12 D =  50.05882 f =  -0.2395833
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.8464939 DF =  1 p-value =  0.3575461 D =  7.53225 f =  -0.03062292
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  2.904765 DF =  1 p-value =  0.08831811 D =  13.704 f =  -0.05541537
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  8.079757 DF =  1 p-value =  0.004476244 D =  -21.359 f =  0.0915285
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.002767156 DF =  1 p-value =  0.9580476 D =  0.04435484 f =  -0.000241117
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.3425901 DF =  1 p-value =  0.5583379 D =  -3.392727 f =  0.0281805
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.080873 DF =  1 p-value =  0.079218 D =  10.26079 f =  -0.07730038
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  8.437928 DF =  1 p-value =  0.003674746 D =  -64.553 f =  0.03211507
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  12.05476 DF =  1 p-value =  0.0005166024 D =  27.20025 f =  -0.1113397
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1921472 DF =  1 p-value =  0.6611355 D =  -3.04 f =  0.02039721
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.134716 DF =  1 p-value =  0.2867717 D =  -28.86422 f =  0.01000193
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5114028 DF =  1 p-value =  0.4745322 D =  -12.77292 f =  0.01052765
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
```

```
## Chi2 =  0.1299846 DF =  1 p-value =  0.718448 D =  -2.573333 f =  0.01720449
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.01194549 DF =  1 p-value =  0.9129682 D =  1.839032 f =  -0.002464659
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.9405559 DF =  1 p-value =  0.3321349 D =  9.084203 f =  -0.02761863
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.4920306 DF =  1 p-value =  0.4830229 D =  30.39793 f =  -0.003998096
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.02429297 DF =  1 p-value =  0.8761418 D =  -1.62109 f =  0.006057408
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1323965 DF =  1 p-value =  0.7159601 D =  7.571257 f =  -0.004695561
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.72444 DF =  1 p-value =  0.0536216 D =  25.74437 f =  -0.03645192
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.072349 DF =  1 p-value =  0.3004155 D =  11.37037 f =  -0.02517632
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.000815 DF =  1 p-value =  0.3171134 D =  6.964183 f =  -0.04001581
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.0002798192 DF =  1 p-value =  0.9866538 D =  -0.4261472 f =  0.00163321
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.01918895 DF =  1 p-value =  0.8898261 D =  -1.228296 f =  0.007912835
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  5.257029 DF =  1 p-value =  0.02185831 D =  -18.24903 f =  0.07277808
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.9132209 DF =  1 p-value =  0.3392613 D =  7.061869 f =  -0.03585831
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  10.14062 DF =  1 p-value =  0.001450376 D =  21.04179 f =  -0.1230815
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  5.436096 DF =  1 p-value =  0.0197247 D =  14.68253 f =  -0.097353
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  2.880493 DF =  1 p-value =  0.08965854 D =  15.91541 f =  -0.04659288
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  9.159442 DF =  1 p-value =  0.00247438 D =  -18.75975 f =  0.1220069
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.684379 DF =  1 p-value =  0.1943436 D =  -7.7 f =  0.05982906
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  2.378741 DF =  1 p-value =  0.1229966 D =  -8.780271 f =  0.07361042
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  7.584971 DF =  1 p-value =  0.005885692 D =  -15.74953 f =  0.1228038
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  6.681023 DF =  1 p-value =  0.009744467 D =  17.8819 f =  -0.09685885
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  22.23425 DF =  1 p-value =  2.413303e-06 D =  29.9468 f =  -0.1842277
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  8.860607 DF =  1 p-value =  0.002913906 D =  28.94278 f =  -0.07779121
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  4.911152 DF =  1 p-value =  0.02668384 D =  -13.18271 f =  0.09861193
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.6260884 DF =  1 p-value =  0.4287938 D =  -6.979167 f =  0.02415111
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5364934 DF =  1 p-value =  0.4638896 D =  -11.65944 f =  0.01196872
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1351309 DF =  1 p-value =  0.7131703 D =  4.077072 f =  -0.009885658
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
```

```
## Chi2 =  0.9296296 DF =  1 p-value =  0.3349592 D =  10.30127 f =  -0.02408535
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.00109149 DF =  1 p-value =  0.9736445 D =  0.6410604 f =  -0.002016808
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.02948787 DF =  1 p-value =  0.8636574 D =  1.591934 f =  -0.007712555
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.9855184 DF =  1 p-value =  0.3208402 D =  9.449013 f =  -0.02795144
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.2436828 DF =  1 p-value =  0.6215588 D =  3.197664 f =  -0.02328922
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.9748977 DF =  1 p-value =  0.3234617 D =  -13.93652 f =  0.01812441
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  2.285431 DF =  1 p-value =  0.1305938 D =  14.42624 f =  -0.04109178
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.527046 DF =  1 p-value =  0.4678514 D =  14.56285 f =  -0.009475243
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.107158 DF =  1 p-value =  0.2927005 D =  -17.3787 f =  0.01654517
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.316154 DF =  1 p-value =  0.5739282 D =  -4.545556 f =  0.02042528
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.6995281 DF =  1 p-value =  0.4029423 D =  5.49805 f =  -0.03547084
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1034337 DF =  1 p-value =  0.7477473 D =  -2.062 f =  0.0176146
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  2.027621 DF =  1 p-value =  0.1544622 D =  -9.690086 f =  0.05595058
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.05657749 DF =  1 p-value =  0.8119895 D =  6.210429 f =  -0.002483884
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.003919973 DF =  1 p-value =  0.9500773 D =  0.7036329 f =  -0.005527517
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.6517352 DF =  1 p-value =  0.419493 D =  6.558255 f =  -0.02767877
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.321528 DF =  1 p-value =  0.06837824 D =  -11.94385 f =  0.07330043
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5794936 DF =  1 p-value =  0.4465109 D =  -5.153846 f =  0.03101852
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  257.1487 DF =  1 p-value =  7.178839e-58 D =  142.585 f =  -0.4520551
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.411656 DF =  1 p-value =  0.2347816 D =  5.647206 f =  -0.05712742
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.004548373 DF =  1 p-value =  0.9462301 D =  -0.9524092 f =  0.003067811
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.3865915 DF =  1 p-value =  0.5340965 D =  7.775519 f =  -0.01344723
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.34518 DF =  1 p-value =  0.06740212 D =  -15.0755 f =  0.05807753
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.580895 DF =  1 p-value =  0.2086318 D =  -9.68551 f =  0.0426326
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5194688 DF =  1 p-value =  0.4710684 D =  4.394135 f =  -0.03512333
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.035923 DF =  1 p-value =  0.08144021 D =  12.79703 f =  -0.05905331
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  19.62991 DF =  1 p-value =  9.398615e-06 D =  -28.53933 f =  0.1375129
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
```

```
## Chi2 =  10.8335 DF =  1 p-value =  0.0009967997 D =  -18.96212 f =  0.1308074
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  2.669591 DF =  1 p-value =  0.1022823 D =  -9.506616 f =  0.07397764
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.139921 DF =  1 p-value =  0.07639749 D =  11.23667 f =  -0.06619021
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.430547 DF =  1 p-value =  0.0640009 D =  15.5984 f =  -0.05663873
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.01769947 DF =  1 p-value =  0.8941623 D =  -1.413022 f =  0.005620323
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5206021 DF =  1 p-value =  0.4705849 D =  4.988433 f =  -0.03013948
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.000713165 DF =  1 p-value =  0.9786949 D =  -0.7998602 f =  0.0007146076
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  146.1379 DF =  1 p-value =  1.211144e-33 D =  95.96041 f =  -0.3830149
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.354865 DF =  1 p-value =  0.2444294 D =  8.493766 f =  -0.04300505
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1601897 DF =  1 p-value =  0.6889819 D =  3.000362 f =  -0.01739344
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.288761 DF =  1 p-value =  0.06975563 D =  24.70451 f =  -0.03371269
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  137.959 DF =  1 p-value =  7.439819e-32 D =  82.43134 f =  -0.414085
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.1390548 DF =  1 p-value =  0.7092225 D =  2.996328 f =  -0.01490683
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.2278697 DF =  1 p-value =  0.6331079 D =  3.000496 f =  -0.02429561
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.04569838 DF =  1 p-value =  0.830725 D =  2.423752 f =  -0.006558191
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.02150404 DF =  1 p-value =  0.8834142 D =  -1.72093 f =  0.005036069
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.002658075 DF =  1 p-value =  0.9588821 D =  0.1014235 f =  -0.0007301982
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  46.02595 DF =  1 p-value =  1.166967e-11 D =  -49.35077 f =  0.2357325
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.7054962 DF =  1 p-value =  0.400943 D =  6.95025 f =  -0.02807617
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.183676 DF =  1 p-value =  0.2766089 D =  12.408 f =  -0.02508734
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  4.44235 DF =  1 p-value =  0.03505795 D =  12.058 f =  -0.09728744
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.009584193 DF =  1 p-value =  0.9220126 D =  1.434472 f =  -0.002978768
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  6.225188 DF =  1 p-value =  0.01259455 D =  24.83833 f =  -0.06269747
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.2914313 DF =  1 p-value =  0.5893048 D =  -5.659687 f =  0.01461848
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.0012818 DF =  1 p-value =  0.97144 D =  0.1296004 f =  -0.0005459839
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.7388536 DF =  1 p-value =  0.3900284 D =  6.3196 f =  -0.03229552
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.6764863 DF =  1 p-value =  0.4107993 D =  9.200211 f =  -0.01970912
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
```

```
## Chi2 =  3.351774 DF =  1 p-value =  0.06713262 D =  -26.24257 f =  0.0326708
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  4.35653 DF =  1 p-value =  0.03686733 D =  14.57335 f =  -0.07859362
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.425034 DF =  1 p-value =  0.2325766 D =  -13.57976 f =  0.02726422
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  11.35796 DF =  1 p-value =  0.0007512515 D =  -21.68111 f =  0.1349326
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  12.87362 DF =  1 p-value =  0.0003332464 D =  -33.87901 f =  0.09479855
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.84693 DF =  1 p-value =  0.1741409 D =  -11.47977 f =  0.04236395
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.297344 DF =  1 p-value =  0.254699 D =  9.412698 f =  -0.03626024
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.9992242 DF =  1 p-value =  0.3174983 D =  6.027616 f =  -0.0469176
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  8.3686 DF =  1 p-value =  0.003817595 D =  33.50919 f =  -0.06352565
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5251952 DF =  1 p-value =  0.4686339 D =  -8.294584 f =  0.01703919
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5671085 DF =  1 p-value =  0.4514102 D =  -11.23508 f =  0.01349178
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.415354 DF =  1 p-value =  0.06459257 D =  13.25731 f =  -0.06506889
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.253838 DF =  1 p-value =  0.2628208 D =  -15.4113 f =  0.0213627
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  34.4094 DF =  1 p-value =  4.465612e-09 D =  39.14286 f =  -0.2238562
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  13.46418 DF =  1 p-value =  0.0002431617 D =  21.09789 f =  -0.1636737
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.03480967 DF =  1 p-value =  0.8519951 D =  -1.443139 f =  0.01085531
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  60.8135 DF =  1 p-value =  6.274629e-15 D =  -40.79528 f =  0.3492887
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.570515 DF =  1 p-value =  0.2101323 D =  -7.346899 f =  0.05814863
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.17896 DF =  1 p-value =  0.6722685 D =  -2.736554 f =  0.02185108
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  25.08128 DF =  1 p-value =  5.496383e-07 D =  36.18406 f =  -0.1732821
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.334906 DF =  1 p-value =  0.5627845 D =  -3.924959 f =  0.02583485
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.3479463 DF =  1 p-value =  0.555278 D =  5.036 f =  -0.0201469
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  19.3649 DF =  1 p-value =  1.079736e-05 D =  -24.93812 f =  0.1996038
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.331001 DF =  1 p-value =  0.0679855 D =  -16.08 f =  0.05394525
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.4230739 DF =  1 p-value =  0.5154072 D =  -5.885496 f =  0.02023303
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.4939089 DF =  1 p-value =  0.4821888 D =  -4.435289 f =  0.03311516
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  24.46085 DF =  1 p-value =  7.583506e-07 D =  85.09313 f =  -0.06561237
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
```

```
## Chi2 =  3.467243 DF =  1 p-value =  0.06259555 D =  -12.97927 f =  0.0684997
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  174.1815 DF =  1 p-value =  9.035356e-40 D =  82.21688 f =  -0.5328962
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.08108758 DF =  1 p-value =  0.7758289 D =  -2.497478 f =  0.01119958
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  28.15002 DF =  1 p-value =  1.122668e-07 D =  61.89341 f =  -0.114172
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.4623857 DF =  1 p-value =  0.4965111 D =  4.48375 f =  -0.03029228
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.000456616 DF =  1 p-value =  0.9829516 D =  -0.3466495 f =  0.002393217
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.3410853 DF =  1 p-value =  0.5592034 D =  8.579024 f =  -0.01068477
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.3169737 DF =  1 p-value =  0.5734321 D =  4.673218 f =  -0.01981632
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.05104855 DF =  1 p-value =  0.8212487 D =  1.6125 f =  -0.01312176
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  3.173079 DF =  1 p-value =  0.07486119 D =  16.8789 f =  -0.0484857
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  64.13849 DF =  1 p-value =  1.159733e-15 D =  -60.21745 f =  0.2632831
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.2555046 DF =  1 p-value =  0.6132255 D =  -3.493443 f =  0.02290881
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.5097411 DF =  1 p-value =  0.4752509 D =  -4.662069 f =  0.03222731
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.3770417 DF =  1 p-value =  0.5391907 D =  -3.8 f =  0.03044872
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.7512408 DF =  1 p-value =  0.3860837 D =  5.28932 f =  -0.04109465
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.9896148 DF =  1 p-value =  0.3198365 D =  5.1245 f =  -0.04817369
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  6.441667 DF =  1 p-value =  0.0111474 D =  14 f =  -0.1166667
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.008080521 DF =  1 p-value =  0.9283733 D =  1.323327 f =  -0.002738806
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.070909 DF =  1 p-value =  0.3007402 D =  -8.017699 f =  0.03611288
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.3002523 DF =  1 p-value =  0.5837243 D =  -3.422 f =  0.02750317
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.09548535 DF =  1 p-value =  0.7573162 D =  -1.91353 f =  0.01643735

## Warning in HWChisq(x): Expected counts below 5: chi-square approximation
## may be incorrect

## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  16.73636 DF =  1 p-value =  4.294986e-05 D =  -8.089721 f =  0.1282257

## Warning in HWChisq(x): Expected counts below 5: chi-square approximation
## may be incorrect

## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  0.0281032 DF =  1 p-value =  0.8668664 D =  -0.1361868 f =  0.003017241
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  7.308418 DF =  1 p-value =  0.006863233 D =  -21.22661 f =  0.08873014
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
```

```
## Chi2 =  0.2354712 DF =  1 p-value =  0.6274964 D =  -2.592552 f =  0.02214105
## Chi-square test with continuity correction for Hardy-Weinberg equilibrium (autosomal)
## Chi2 =  1.141699 DF =  1 p-value =  0.2852937 D =  6.648276 f =  -0.04703357
```

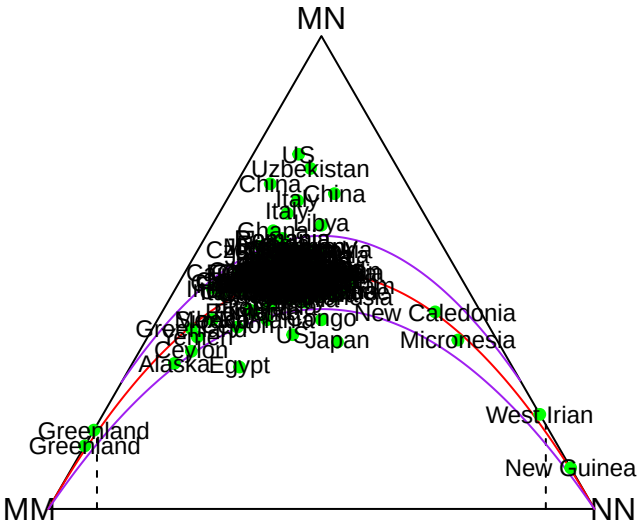
```
Mourant$p.val <- t
subset(Mourant, p.val < 0.05)
```

```
##     Population        Country Total   MM   MN   NN        p.val
## 1         USSR         Russia  5728 1743 3222  763 3.253630e-33
## 8       Europe        Austria  6557 2185 2993 1379 3.667752e-09
## 9       Europe        Austria  3000  914 1548  538 8.617946e-03
## 10      Europe        Austria 10000 3156 4761 2083 2.564751e-04
## 16      Europe Czechoslovakia   500  135  274   91 2.319407e-02
## 18      Europe Czechoslovakia  5110 1457 2824  829 2.873010e-18
## 28      Europe         France 10694 3356 5178 2160 4.711102e-02
## 33      Europe         France  1082  370  490  222 1.291399e-02
## 38      Europe        Germany  4867 1446 2482  939 3.161787e-02
## 40      Europe        Germany  1059  274  567  218 1.846327e-02
## 55      Europe        Germany   546  157  303   86 3.195697e-03
## 61      Europe        Hungary   554  209  229  116 5.641774e-04
## 65      Europe          Italy   800  263  357  180 6.685156e-03
## 69      Europe          Italy  1164  254  736  174 3.995607e-20
## 71      Europe          Italy   850  221  518  111 4.137647e-12
## 74      Europe          Italy  1000  417  424  159 4.476244e-03
## 78      Europe     Jugoslavia  8278 2895 3891 1492 3.674746e-03
## 79      Europe     Jugoslavia  1000  304  543  153 5.166024e-04
## 94      Europe        Romania  1035  376  465  194 2.185831e-02
## 96      Europe        Romania   694  197  384  113 1.450376e-03
## 97      Europe        Romania   604  147  331  126 1.972470e-02
## 99      Europe        Romania   641  250  270  121 2.474380e-03
## 102     Europe        Romania   528  196  225  107 5.885692e-03
## 103     Europe        Romania   743  198  405  140 9.744467e-03
## 104     Europe        Romania   672  204  385   83 2.413303e-06
## 105     Europe        Romania  1503  425  802  276 2.913906e-03
## 106     Europe          Spain   535  153  241  141 2.668384e-02
## 128       USSR     Uzbekistan  1265  207  916  142 7.178839e-58
## 136       Asia         Ceylon  1068  607  358  103 9.398615e-06
## 137       Asia         Ceylon   660  319  252   89 9.967997e-04
## 144       Asia          China  1004  134  693  177 1.211144e-33
## 148       Asia          China   812  181  563   68 7.439819e-32
## 154       Asia          Japan   841  233  320  288 1.166967e-11
## 157       Asia          Japan   500  137  272   91 3.505795e-02
## 159       Asia          Japan  1633  536  842  255 1.259455e-02
## 165       Asia          Japan   743  187  400  156 3.686733e-02
## 167       Asia          Japan   646  207  278  161 7.512515e-04
## 168       Asia          Japan  1465  523  647  295 3.332464e-04
## 172       Asia          Japan  2121  576 1122  423 3.817595e-03
## 177     Africa          Libya   700  126  428  146 4.465612e-09
## 178     Africa        Marocco   521  137  300   84 2.431617e-04
## 180     Africa          Egypt   508  250  152  106 6.274629e-15
## 183     Africa          Ghana   853  243  490  120 5.496383e-07
## 186     Africa          Congo   501  163  200  138 1.079736e-05
## 190    America         Canada  5734 2370 2764  600 7.583506e-07
## 192    America             US   619   90  473   56 9.035356e-40
## 194    America             US  2186  587 1208  391 1.122668e-07
```

```
## 201    America             US   937  372  337  228 1.159733e-15
## 207    America         Mexico   500  166  268   66 1.114740e-02
## 212    Oceania     New Guinea  1148   12  110 1026 4.294986e-05
## 214    Oceania     Micronesia   962  228  436  298 6.863233e-03
```

Question 2.17: Divide all total frequencies by 50, keeping the same proportions for each of the genotypes, and recreate the ternary plot. a) what happens to the points? b) what happens to the confience regions?

```
newgf = round(genotypeFrequencies / 50)
HWTernaryPlot(newgf,
              markerlab = Mourant$Country,
              alpha = 0.0001, curvecols = c("red", rep("purple", 4)),
              mcex = 0.75, vertex.cex = 1)
```



Confidence regions get bigger

2.7.3 Concatenating several multinomials: sequence motifs and logos

```
library("seqLogo")
load(url("http://bios221.stanford.edu/data/kozak.RData"))
kozak
```

```
##   [,1] [,2] [,3] [,4] [,5] [,6] [,7] [,8] [,9]
## A 0.33 0.25  0.4 0.15 0.20    1    0    0 0.05
## C 0.12 0.25  0.1 0.40 0.40    0    0    0 0.05
## G 0.33 0.25  0.4 0.20 0.25    0    0    1 0.90
## T 0.22 0.25  0.1 0.25 0.15    0    1    0 0.00
```

```
pwm = makePWM(kozak)
seqLogo(pwm, ic.scale = FALSE)
```

2.8 Modelling sequence dependencies: Markov chains

```
## Package:  markovchain
```

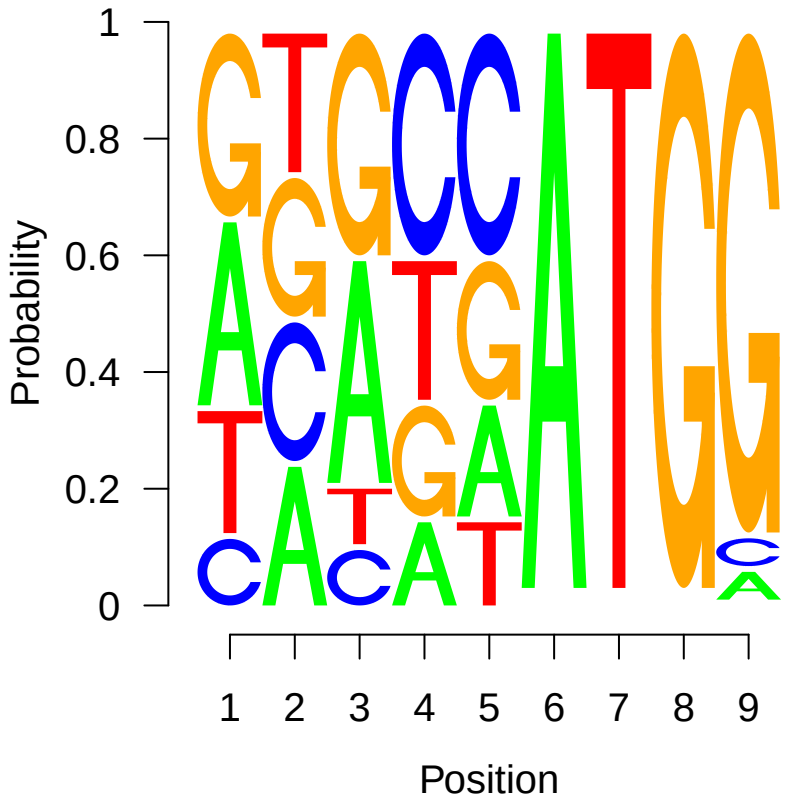


Figure 12: Here is a diagram called a sequence logo for the position dependent multinomial used to model the Kozak motif. It codifies the amount of variation in each of the positions on a log scale. The large letters represent positions where there is no uncertainty about which nucleotide occurs.

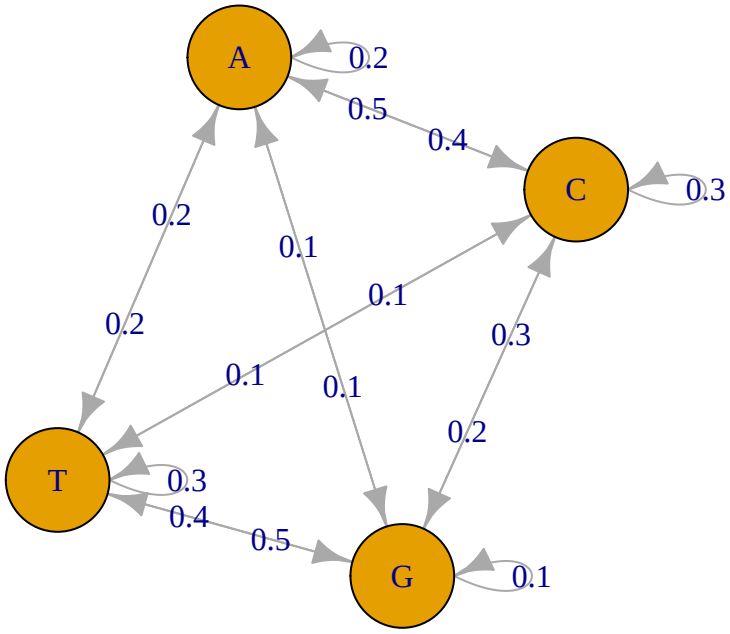


Figure 13: Visualisation of a 4-state Markov chain. The probability of each possible digram (e.,g., CA) is given by the weight of the edge between the corresponding nodes. So for instance, the probability of CA is given by the edge C→ A. We'll see in Chapter @ref(Chap:Images) how to use **R** packages to draw these type of network graphs.

```
## Version:  0.8.0
## Date:     2019-09-13
## BugReport: http://github.com/spedygiorgio/markovchain/issues
```

```
##
## Attaching package: 'igraph'
```

```
## The following object is masked from 'package:Biostrings':
##
##     union
```

```
## The following object is masked from 'package:IRanges':
##
##     union
```

```
## The following object is masked from 'package:S4Vectors':
##
##     union
```

```
## The following objects are masked from 'package:BiocGenerics':
##
##     normalize, path, union
```

```
## The following objects are masked from 'package:stats':
##
##     decompose, spectrum
```

```
## The following object is masked from 'package:base':
##
##     union
```

2.9 Baysian thinking

The *baysian paradigm* is a practical approach in which *prior* and *posterior* distributions are used as models of our knowledge **before** and **after** collecting some data and making an observation. We formalize our prior probability as $P(H)$. After seeing the data we have the posterior probability, $P(H|D)$.

2.9.1 Example: Haplotype frequencies

```
haplo6 <- read.table("../../data/haplotype6.txt",header = TRUE)
haplo6
```

```
##   Individual DYS19 DXYS156Y DYS389m DYS389n DYS389p
## 1         H1    14       12       4      12       3
## 2         H3    15       13       4      13       3
## 3         H4    15       11       5      11       3
## 4         H5    17       13       4      11       3
## 5         H7    13       12       5      12       3
## 6         H8    16       11       5      12       3
```

2.9.2 Simulation study of the Bayesian paradigm for the binomial

Instead of assuming our parameter Θ has one single value, the bayesian world view allows us to see it as a draw from a statistical distribution. When we are looking at a parameter that expresses a proportion or a probability, and that takes its values between 0 and 1, it is convenient to use the *beta distribution*

Why 50 & 350 for alpha and beta?

```
## Loading required package: reshape2
```

```
##
## Attaching package: 'reshape2'
```

```
## The following object is masked from 'package:tidyr':
##
##     smiths
```

The distribution of γ

What is the distribution of Y if Θ itself also varies according to some distribution. We call this the *marginal* distribution of Y.

```
rtheta = rbeta(100000, 50, 350)
y = vapply(rtheta, function(th) {
  rbinom(1, prob = th, size = 300)
}, numeric(1))
hist(y, breaks = 50, col = "orange", main = "", xlab = "")
```

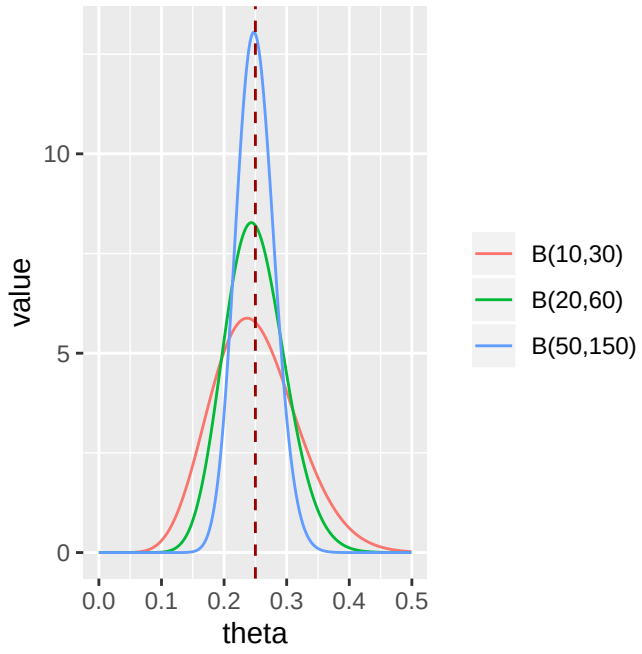
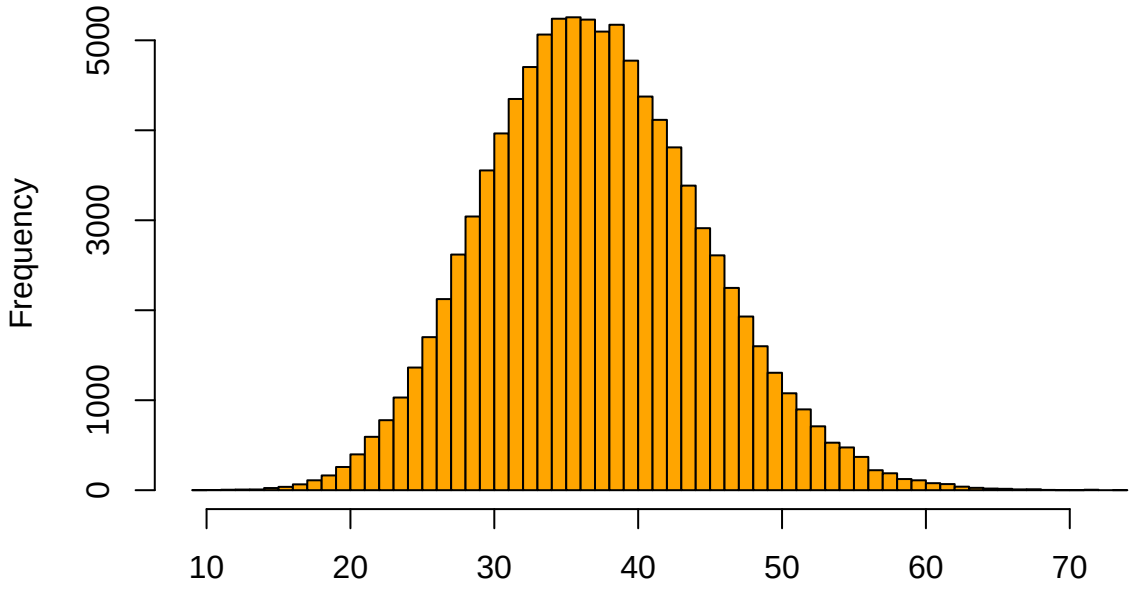


Figure 14: Beta distributions with $\alpha = 10, 20, 50$ and $\beta = 30, 60, 150$ used as a {prior} for a probability of success. These three distributions have the same mean ($\frac{\alpha}{\alpha+\beta}$), but different concentrations around the mean.



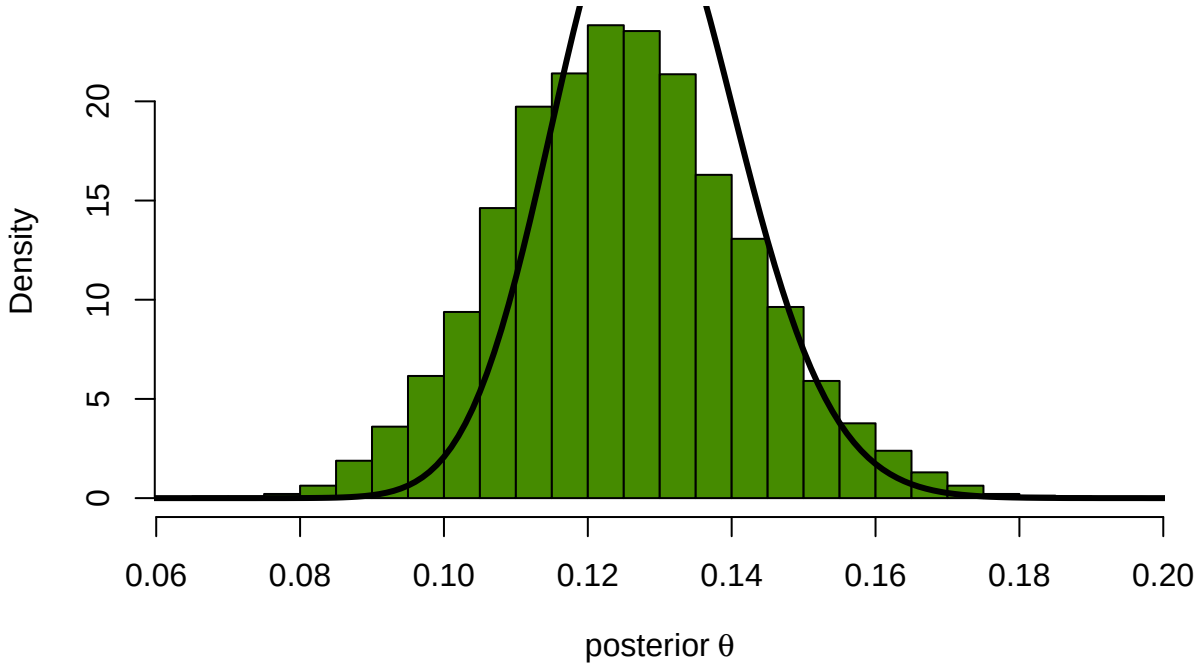

```
Mode <- function(x) {
  ux <- unique(x)
  ux[which.max(tabulate(match(x, ux)))]
}

Mode(y)
```

```
## [1] 36
```

Question 2.18: Verify that we can get the same result as in the above code chunk by using R's vectorization capabilities and writing 'rbinom(length(rtheta), rtheta, size = 300'.

Compute the posterior distribution of Θ byu conditioning on outcomes where Y is 40. Cmpare it to the theoretical posterior `densPostTheory`. HELP!

```
rtheta = rbeta(100000, 50, 350) #there are 10,000 observations, pulled from a beta distribution with sh
#y = vapply(rtheta, function(th) {rbinom(1, prob = th, size = 300)}, numeric(1))
#y is a vector of length rtheta where a single observation was drawn from the binomial disribution 300
# y = 40 is asking for the rtheta values that quasi-by-chance gave 40/300 successes from the binomial d

thetaPostEmp = rtheta[y == 40]  #~ 45% of all y values are equal to 40; 40 is a most common result
hist(thetaPostEmp, breaks = 40, col = "chartreuse4", main = "",
     probability = TRUE, xlab = expression("posterior"~theta))
densPostTheory  =  dbeta(thetas, 90, 610)
lines(thetas, densPostTheory, type="l", lwd = 3)
```



Check the means of both distributions computed above and see that they are close to four significant digits:

Calculate the integral under the curve

```
mean(thetaPostEmp)
```

```
## [1] 0.1250748
```

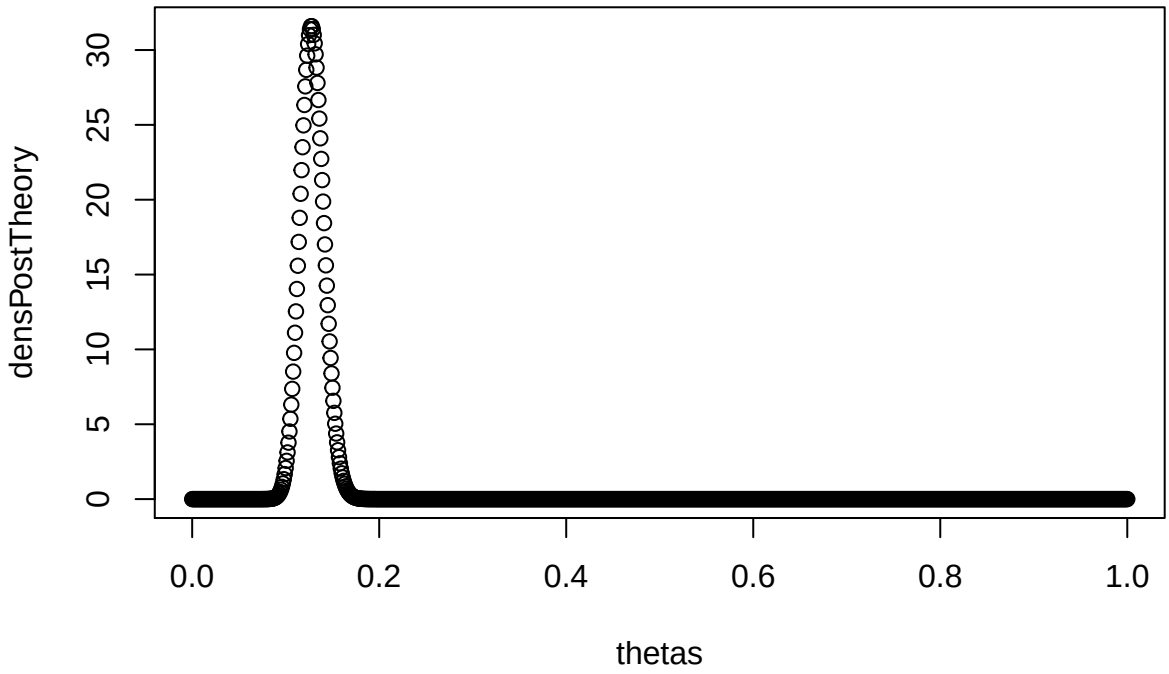
```
mean(rtheta)
```

```
## [1] 0.1249937
```

```
dtheta = thetas[2]-thetas[1]
sum(thetas * densPostTheory * dtheta)
```

```
## [1] 0.1285714
```

```
plot(thetas, densPostTheory)
```



To approximate the mean of the theoretical distribution `densPostTheory` we computed the integral $\int_0^1 \Theta f(\Theta) d\Theta$ using numerical integration, i.e., the sum over the integrand. Use *monte carlo integration* instead. Again, where do the shape numbers 90, 610 come from?? Why is this equivalent??

```
thetaPostMC = rbeta(n = 1e6, 90, 610)
mean(thetaPostMC)
```

```
## [1] 0.1285784
```

Check the concordance between Monte Carlo simulation sample `thetaPostMC` and our sample `thetaPostEmp` using a Q-Q plot

```
qqplot(thetaPostMC, thetaPostEmp, type = "l", asp = 1)
abline(a = 0, b = 1, col = "blue")
```

Question 2.19What is the difference between the simulation that results in 'thetaPostEmp' and the Monte Carlo simulation that leads to 'thetaPostMC'?

Posterior distribution is also a beta

The parameters $\alpha = 90$ and $\beta = 610$ were obtained by summing the prior parameters $\alpha = 50$ and $\beta = 350$ with the observed successes y = 40 and the observed failures n-y = 260, obtaining the posterior

$beta(90, 610) = beta(\alpha + y, \beta + (n - y))$

We can use this to give the best estimate we can for Θ with its uncertainty given by the posterior distribution. This is called the MAP estimate, $\frac{\alpha-1}{\alpha+\beta-2} = \frac{89}{698} = 0.1275$

Suppose we have a second series of data After seeing our previous data we now have a new prior, beta(90, 610). Now we collect a new data set with n = 150 observations and only y = 25 successes

The new posterior will be beta(90+25 = 115, 610+125 = 735); the mean is $\frac{115}{115+735} = 0.135$, thus one estimate of Θ is 0.135.

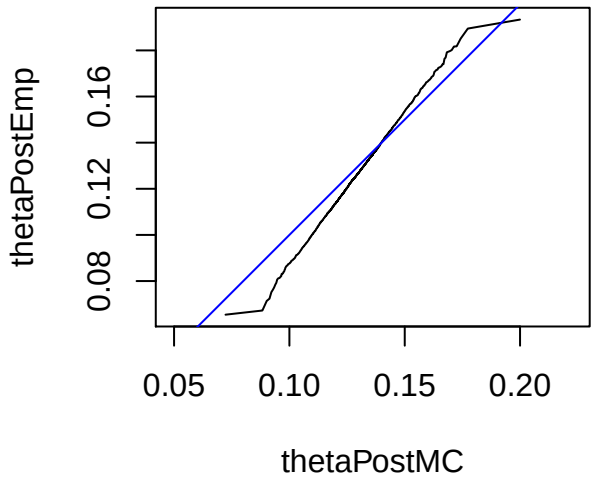


Figure 15: QQ-plot of our Monte Carlo sample `thetaPostMC` from the theoretical distribution and our simulation sample `thetaPostEmp`. We could also similarly compare either of these two distributions to the theoretical distribution function `pbeta(., 90, 610)`. If the curve lies on the line $y = x$ this indicates a good agreement. There are some random differences at the tails.

The theoretical *maximum a posteriori (MAP) estimate* would be the mode of beta(115, 735) = 0.134.

```
densPost2 = dbeta(thetas, 115, 735)
mcPost2 = rbeta(1e6, 115, 735)

sum(thetas*densPost2*dtheta)  #mean, by numeric integration
```

```
## [1] 0.1352941
```

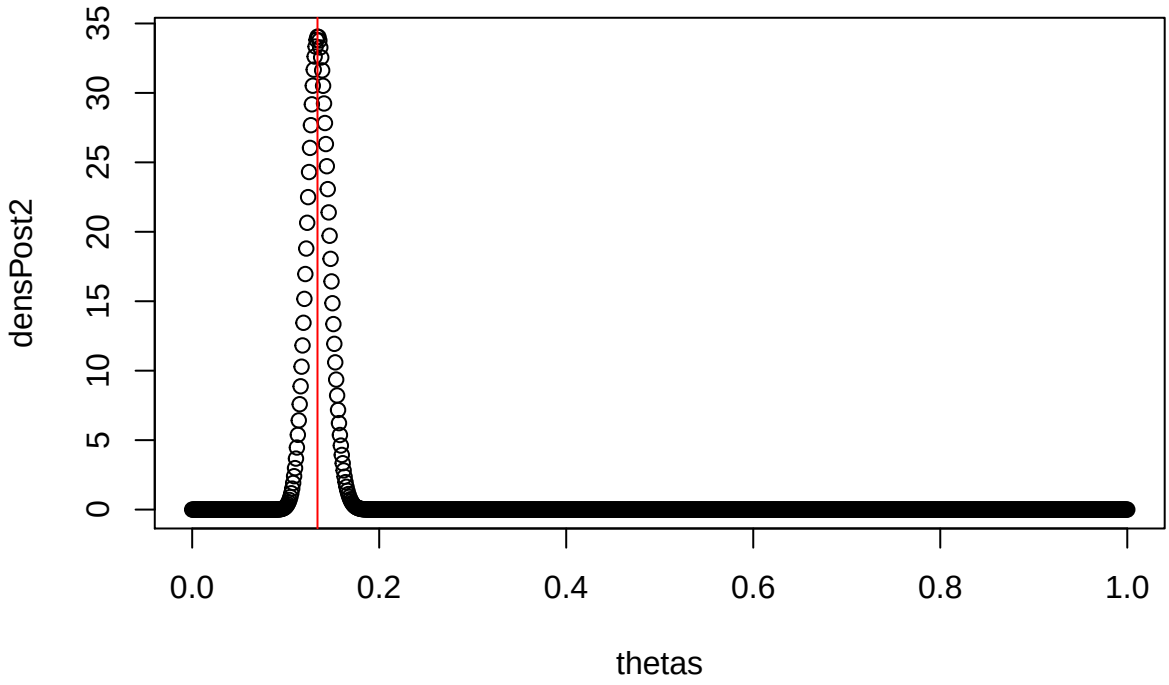
```
mean(mcPost2)  #mean, by MC
```

```
## [1] 0.1352928
```

```
thetas[which.max(densPost2)]
```

```
## [1] 0.134
```

```
plot(thetas, densPost2)
abline(v=thetas[which.max(densPost2)], col="red")
```



Question 2.20Redo all the computations replacing our original prior with a softer prior (less peaked), meaning that we use less prior information. How much does that change the final result?

```
rtheta = rbeta(100000, 50, 100) #there are 10,000 observations, pulled from a beta distribution with sh
densPostTheory  =  dbeta(thetas, 90, 610)
```

Confidence statements for the proportion parameter

Reach a conclusion about where the proportion actually lies given the data. The *posterior credibility interval* is a bayesian analog of a confidence interval. We can take the 2.5th and 97.5th percentiles of the posterior distribution $P(L \leq \Theta \leq U) = 0.95$ as

```
quantile(mcPost2, c(0.025, 0.975))
```

```
##      2.5%     97.5%
## 0.1131241 0.1590410
```

2.10 Example: occurence of a nucleotide pattern in a genome

```
library("Biostrings")
```

```
## GENETIC_CODE
## IUPAC_CODE_MAP
## vignette(package = "Biostrings")
## vignette("BiostringsQuickOverview", package = "Biostrings")
```

```
library("BSgenome")
```

```
## Loading required package: GenomeInfoDb
## Loading required package: GenomicRanges
## Loading required package: rtracklayer
```

```
##
## Attaching package: 'rtracklayer'
```

```
## The following object is masked from 'package:igraph':
##
##     blocks
```

```
ag = available.genomes()
length(ag)
```

```
## [1] 93
```

```
ag[1:2]
```

```
## [1] "BSgenome.Alyrata.JGI.v1"
## [2] "BSgenome.Amellifera.BeeBase.assembly4"
```

```
library("BSgenome.Ecoli.NCBI.20080805")
Ecoli
shineDalgarno <- "AGGAGGT"
ecoli <- Ecoli$NC_010473
```

```
window = 50000
starts = seq(1, length(ecoli) - window, by = window)
ends   = starts + window - 1
numMatches = vapply(seq_along(starts), function(i) {
  countPattern(shineDalgarno, ecoli[starts[i]:ends[i]],
               max.mismatch = 0)
}, numeric(1))
table(numMatches)
```

```
## numMatches
##  0  1  2  3  4
## 48 32  8  3  2
```

Question 2.22: What distribution might this fit? Poisson

What does the poisson plot below mean?

```
library("vcd")
gf = goodfit(numMatches, "poisson")
summary(gf)
```

```
##
##   Goodness-of-fit test for poisson distribution
##
##                       X^2 df  P(> X^2)
## Likelihood Ratio 4.134932  3 0.2472577
```

```
distplot(numMatches, type = "poisson")
```

Inspect the matches using the `matchPattern` function

```
sdMatches <- matchPattern(shineDalgarno, ecoli, max.mismatch =0)
sdMatches
```

```
##   Views on a 4686137-letter DNAString subject
```

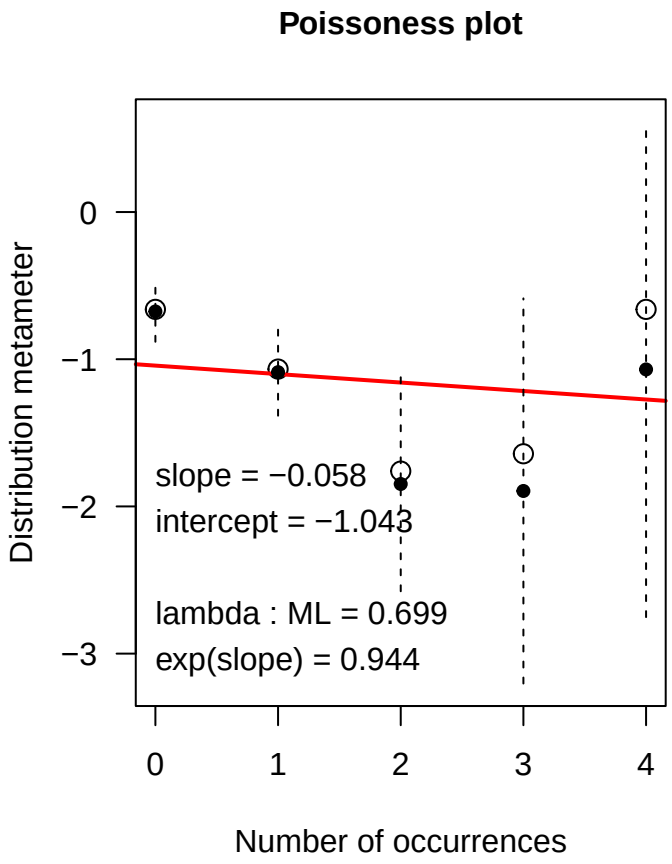


Figure 16: Evaluation of a Poisson model for motif counts along the sequence.

```
## subject: AGCTTTTCATTCTGACTGCAACGGGCAATATG...CAAATAAAAAACGCCTTAGTAAGTATTTTTC
## views:
##        start     end width
##  [1]   56593   56599     7 [AGGAGGT]
##  [2]  199644  199650     7 [AGGAGGT]
##  [3]  202176  202182     7 [AGGAGGT]
##  [4]  214433  214439     7 [AGGAGGT]
##  [5]  217429  217435     7 [AGGAGGT]
##  ...     ...     ...   ... ...
## [61] 4438786 4438792     7 [AGGAGGT]
## [62] 4498085 4498091     7 [AGGAGGT]
## [63] 4536658 4536664     7 [AGGAGGT]
## [64] 4546821 4546827     7 [AGGAGGT]
## [65] 4611626 4611632     7 [AGGAGGT]
```

What are the distances between them?

```
betweenmotifs <- gaps(sdMatches)
```

Find a model for the distribution of gap sizes betrween motifs. If they occur at random locations we expect them to follow an *exponential distribution*.

```
library("Renext")
```

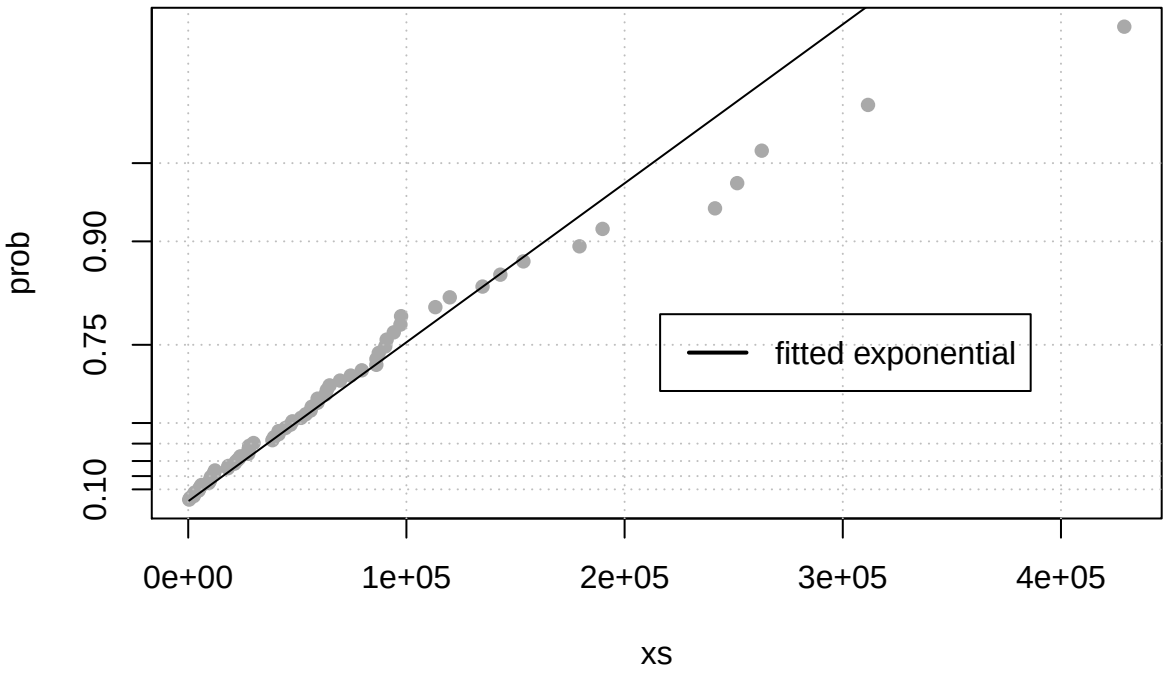
```
## Loading required package: evd
```

```
##
## Attaching package: 'evd'
```

```
## The following object is masked from 'package:igraph':
##
##     clusters
```

```
## The following object is masked from 'package:lattice':
##
##     qq
```

```
expplot(width(betweenmotifs), rate = 1/mean(width(betweenmotifs)),
        labels = "fitted exponential")
```



Question 2.23: There appears to be a slight deviation from the fitted line in Figure 2.23 at the right tail of the distribution, i.e., for the largest values. What could be the reason?

Bias in where genes are located?

2.10.1 Modeling in the case of dependencies

Dependency modelling using a **markov chain**. Discover differences between regions called CpG islands and the rest of the genome.

```
library("BSgenome.Hsapiens.UCSC.hg19")
chr8  =  Hsapiens$chr8
CpGtab = read.table("../../data/model-based-cpg-islands-hg19.txt",
                    header = TRUE)
nrow(CpGtab)
```

```
## [1] 65699
```

```
head(CpGtab)
```

```
##     chr  start    end length CpGcount GCcontent pctGC obsExp
## 1 chr10  93098  93818    721       32       403 0.559  0.572
## 2 chr10  94002  94165    164       12        97 0.591  0.841
## 3 chr10  94527  95302    776       65       538 0.693  0.702
## 4 chr10 119652 120193    542       53       369 0.681  0.866
## 5 chr10 122133 122621    489       51       339 0.693  0.880
## 6 chr10 180265 180720    456       32       256 0.561  0.893
```

```
irCpG = with(dplyr::filter(CpGtab, chr == "chr8"), #filter to only include chromosome 8
             IRanges(start = start, end = end))     #define Iranges object with start and end positions
```

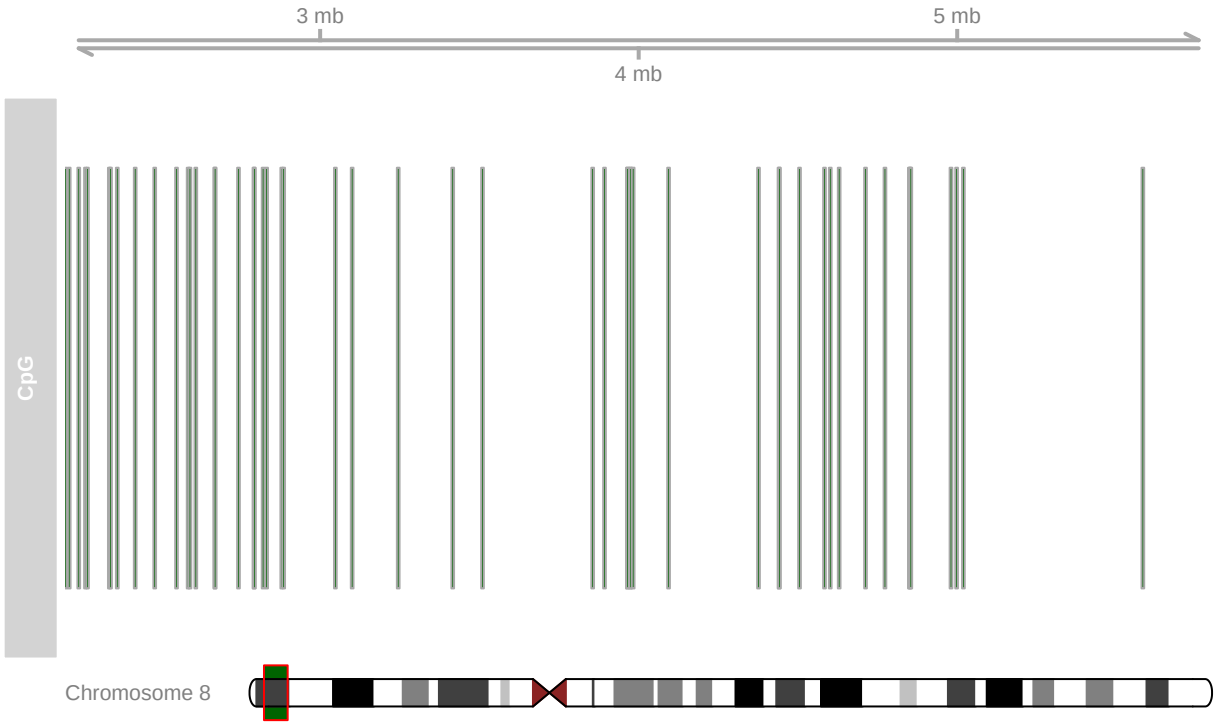


Figure 17: **Gviz** plot of CpG locations in a selected region of chromosome 8.

IRanges is a general "container" for mathematical intervals. **GRanges** is a container for the genomic locations and their associated annotations.

```
grCpG = GRanges(ranges = irCpG, seqnames = "chr8", strand = "+")
genome(grCpG) = "hg19"
```

```
library("Gviz")
ideo = IdeogramTrack(genome = "hg19", chromosome = "chr8")
plotTracks(
  list(GenomeAxisTrack(),
       AnnotationTrack(grCpG, name = "CpG"), ideo),
  from = 2200000, to = 5800000,
  shape = "box", fill = "#006400", stacking = "dense")
```

```
CGIview    = Views(unmasked(Hsapiens$chr8), irCpG)
NonCGIview = Views(unmasked(Hsapiens$chr8), gaps(irCpG))
```

Compute the transition counts in CpG islands and non-islands using the data:

```
seqCGI      = as(CGIview, "DNAStringSet") #there are 2855 different sequences
seqNonCGI   = as(NonCGIview, "DNAStringSet")
dinucCpG    = sapply(seqCGI, dinucleotideFrequency) #calculates all dinucleotides for a set of sequence
dinucNonCpG = sapply(seqNonCGI, dinucleotideFrequency)
dinucNonCpG[, 1]
```

```
##  AA  AC  AG  AT  CA  CC  CG  CT  GA  GC  GG  GT  TA  TC  TG  TT
## 389 351 400 436 498 560 112 603 359 336 403 336 330 527 519 485
```

```
NonICounts = rowSums(dinucNonCpG) #there are 16 different dinucleotides -> sum across all sequences
```

```
IslCounts  = rowSums(dinucCpG)
```

For a four state Markov chain we define the transition matrix as a matrix where the rows are the "From" state and the columns are the "to" state.

```
TI  = matrix( IslCounts, ncol = 4, byrow = TRUE)
TnI = matrix(NonICounts, ncol = 4, byrow = TRUE)
dimnames(TI) = dimnames(TnI) =
  list(c("A", "C", "G", "T"), c("A", "C", "G", "T"))
```

Use the counts of numbers of transitions of each type to compute frequencies and put them in matrices:

```
MI = TI /rowSums(TI)
MI
```

```
##            A         C         G         T
## A 0.20457773 0.2652333 0.3897678 0.1404212
## C 0.20128250 0.3442381 0.2371595 0.2173200
## G 0.18657245 0.3145299 0.3450223 0.1538754
## T 0.09802105 0.3352314 0.3598984 0.2068492
```

```
MN = TnI / rowSums(TnI)
MN
```

```
##           A         C          G         T
## A 0.3351380 0.1680007 0.23080886 0.2660524
## C 0.3641054 0.2464366 0.04177094 0.3476871
## G 0.2976696 0.2029017 0.24655406 0.2528746
## T 0.2265813 0.1972407 0.24117528 0.3350027
```

Question 2.24: Are the transitions different in the different rows? i.e., P(A|C) $\neq$ P(A|T) No.

Question 2.25: Are the relative frequencies of the different nucleotides different in CpG islands compared to elsewhere?

```
freqIsl = alphabetFrequency(seqCGI, baseOnly = TRUE, collapse = TRUE)[1:4]
freqIsl / sum(freqIsl)
```

```
##         A         C         G         T
## 0.1781693 0.3201109 0.3206298 0.1810901
```

```
freqNon = alphabetFrequency(seqNonCGI, baseOnly = TRUE, collapse = TRUE)[1:4]
freqNon / sum(freqNon)
```

```
##         A         C         G         T
## 0.3008292 0.1993832 0.1993737 0.3004139
```

Question 2.26: Use a χ^2 statistic to compare the frequencies between the observed and 'freqIsl' and 'freqNon' frequecies

```
t <- data.frame(freqIsl, freqNon)
chisq.test(t)
```

```
##
##  Pearson's Chi-squared test
##
## data:  t
## X-squared = 423898, df = 3, p-value < 2.2e-16
```

Given a sequence for which it's unknown if it's a CpG island or not, ask the probability if it is. Compute a score based on what is called the odds ratio ->use probabilities for each dinucleotide for islands and compare to probability for non-islands. Then take their ratio and see if it's larger or smaller than one.

Probbilities will be the products of many small terms and become small, so work around this by taking the logarithm. This is the *log-likelihood score*.

```
alpha = log((freqIsl/sum(freqIsl)) / (freqNon/sum(freqNon)))
beta  = log(MI / MN)
```

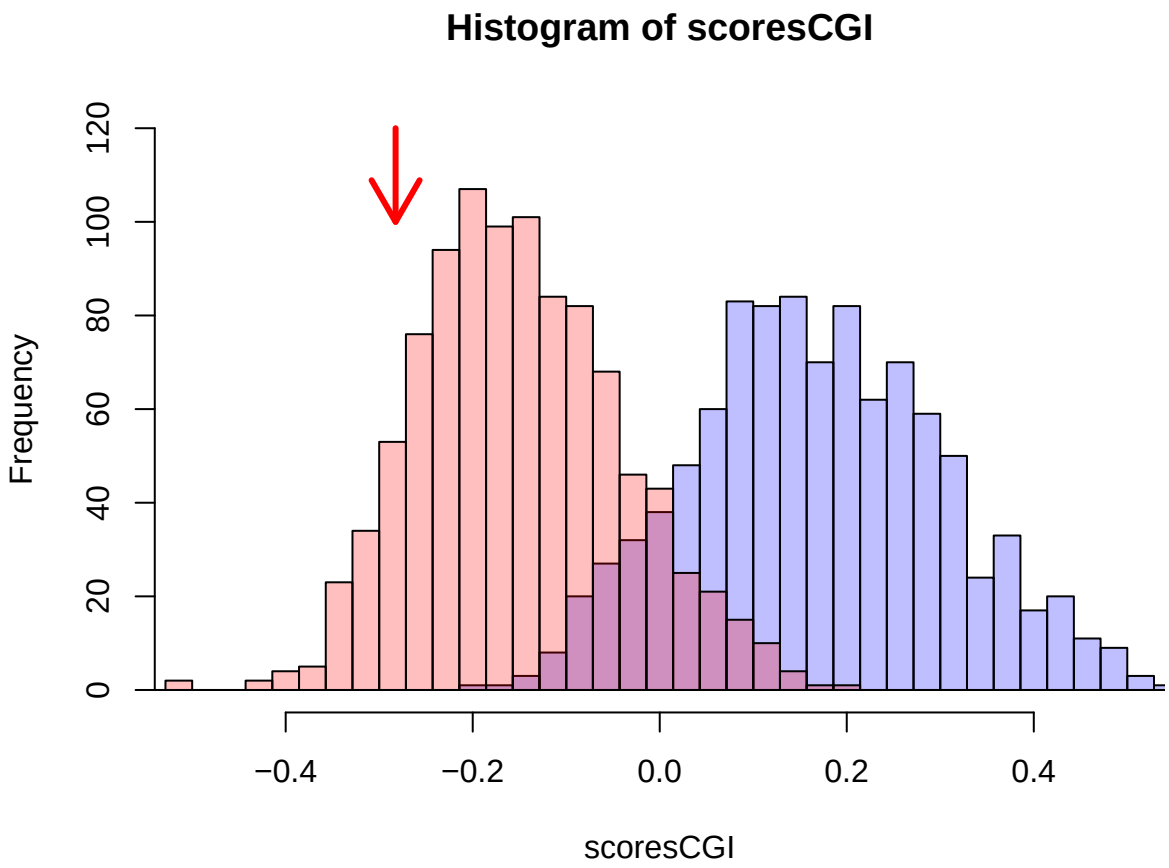
```
x <- "ACGTTATACTACG"
scorefun = function(x) {
  s <- unlist(strsplit(x, ""))
  score <- alpha[s[1]]
  if (length(s) >= 2)
    for (j in 2:length(s))
      score = score + beta[s[j-1], s[j]]
  score
}

testX <- scorefun(x)
```

Then pick sequences of length `len = 100` out of the 2855 sequences in the `seqCGI` object and then out of the 2854 `seqNonCGI` object. Drop sequences that contain any letter other than A, c, T, G (i.e. "."). Sample with probabilities proportional to their length minus `len` and pick subsequences of length `len` out of them.

```
generateRandomScores = function(s, len = 100, B = 1000) {
  alphFreq = alphabetFrequency(s)
  isGoodSeq = rowSums(alphFreq[, 5:ncol(alphFreq)]) == 0
  s = s[isGoodSeq]
  slen = sapply(s, length)
  prob = pmax(slen - len, 0)
  prob = prob / sum(prob)
  idx  = sample(length(s), B, replace = TRUE, prob = prob)
  ssmp = s[idx]
  start = sapply(ssmp, function(x) sample(length(x) - len, 1))
  scores = sapply(seq_len(B), function(i)
    scorefun(as.character(ssmp[[i]][start[i]+(1:len)]))
  )
  scores / len
}
scoresCGI    = generateRandomScores(seqCGI)
scoresNonCGI = generateRandomScores(seqNonCGI)
```

```
br = seq(-0.7, 0.7, length.out = 50) #need to change this to -0.7 or else the breaks did not span the r
h1 = hist(scoresCGI,    breaks = br, plot = FALSE)
h2 = hist(scoresNonCGI, breaks = br, plot = FALSE)
plot(h1, col = rgb(0, 0, 1, 1/4), xlim = c(-0.5, 0.5), ylim=c(0,120))
plot(h2, col = rgb(1, 0, 0, 1/4), add = TRUE)
```

```
arrows(testX, 120, testX, 100, lwd = 3, col="red")
```



We can consider this training data. Cool.

Exercises

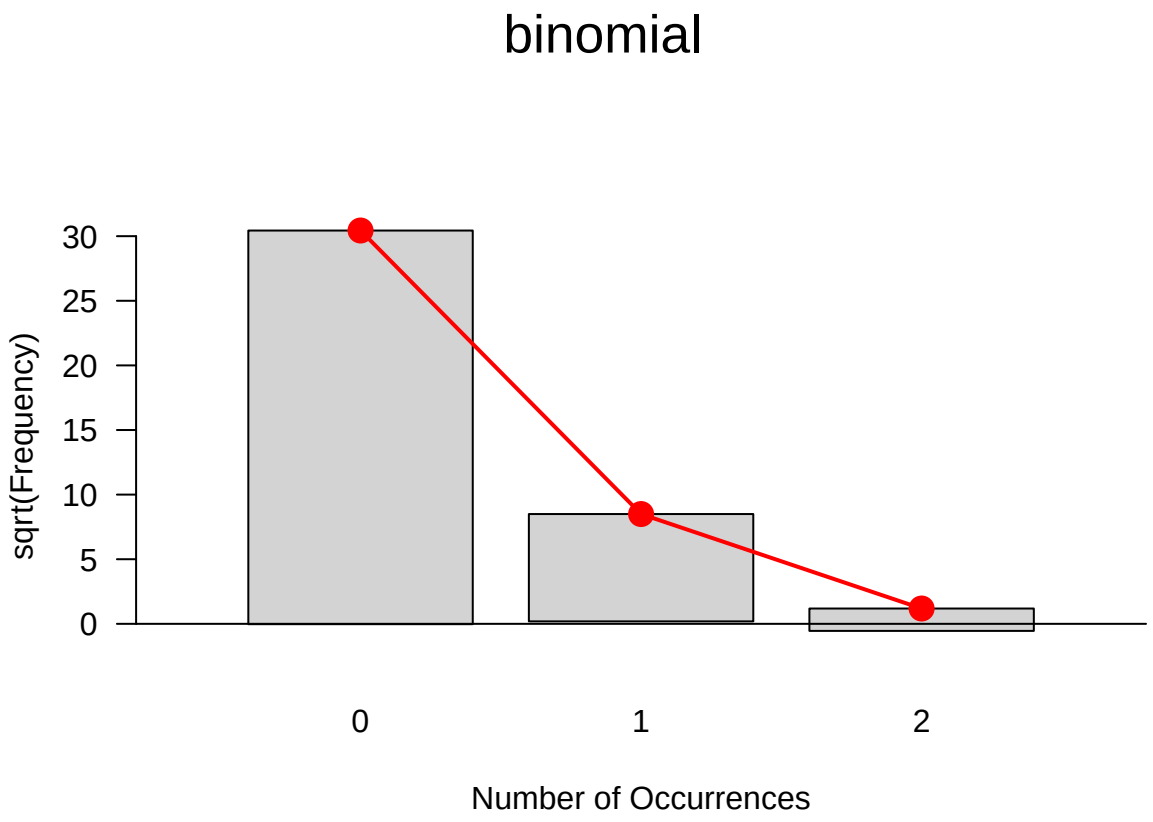
Exercise 2.1

Generate 1000 random 0/1 variables that model mutations occuring along a 1000-long gene sequence. These occur independently at a rate of 10^{-4}. Sum the 1000 positions to count how many mutaitons occur in sequences of length 1000. Find the correct distribution for these mutation sums using a goodness of fit test and make a plot to visualize the quality of the fit.

```
sums <- replicate(1000, {
                  sum(rbinom(1000, 1, 10^-4))
})

gf <- goodfit(sums, "binomial")
```

```
## Warning in goodfit(sums, "binomial"): size was not given, taken as maximum
## count
```

```
gf2 <- goodfit(sums, "poisson")
par(mfrow=c(1, 2))
rootogram(gf, main = "binomial")
```




```
rootogram(gf2, main = "poisson")
```

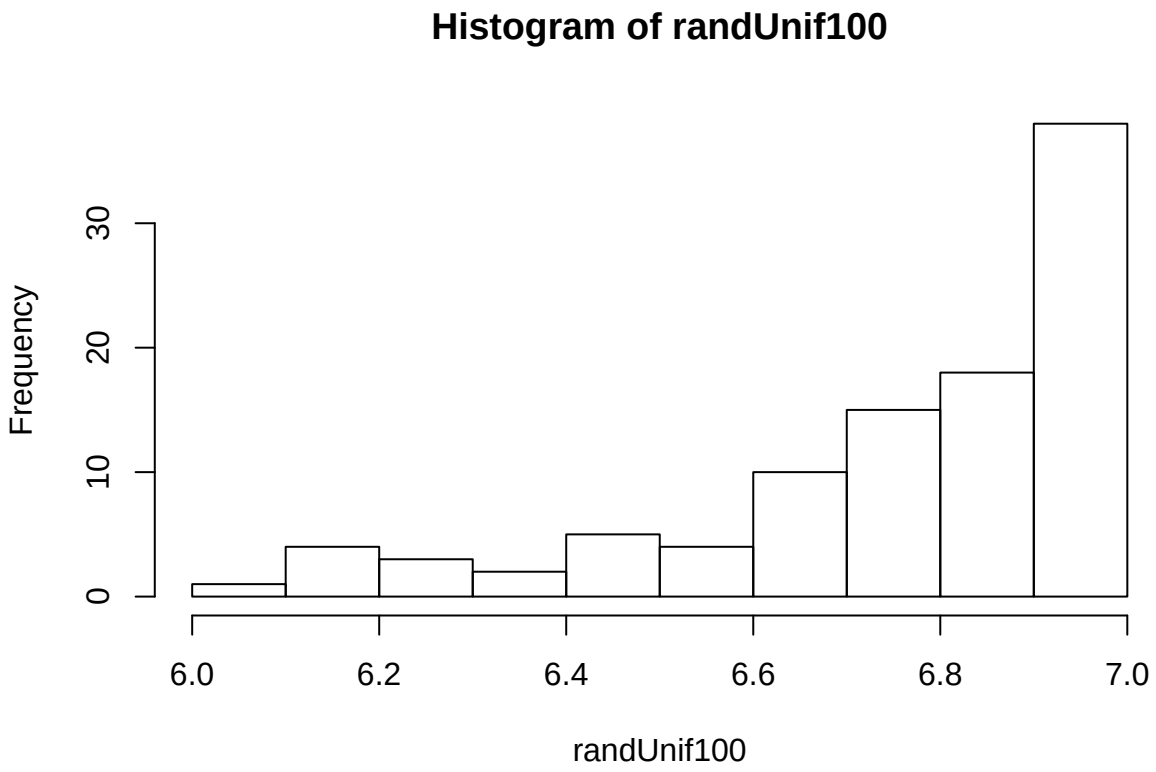
Exercise 2.2

Make a function that generates **n** random uniform numbers between 0 and 7 and returns their maximum. Execute the function for $\mathbf{n = 25}$. Repeat this procedure $B = 100$ times. Plot the distribution of these maxima. What is the maximum likelihood estimate of the maximum of a sample of size 25 (call it $\hat{\theta}$). Can you find a theoretical justification and the true maximum of θ?

```
randUnif <- function(n) max(runif(n, min = 0, max = 7))

randUnif(25)
```

```
## [1] 6.860117
```

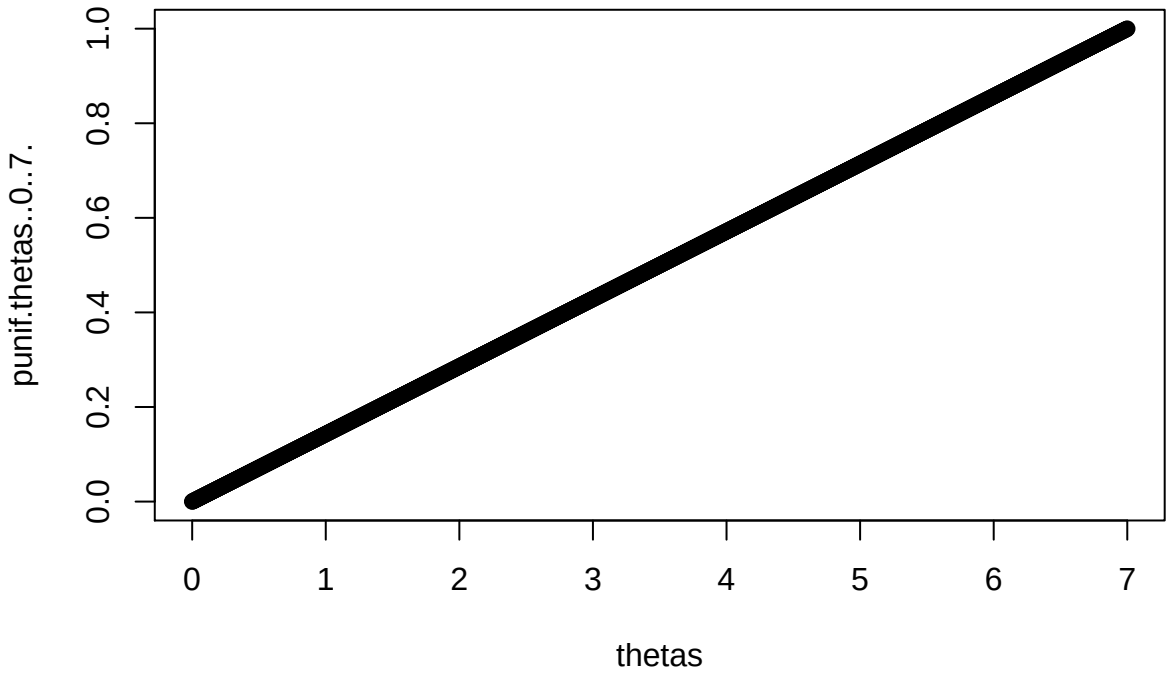
```
randUnif100 <- replicate(100, randUnif(25))
hist(randUnif100)
```




```
theta_hat <- mean(randUnif100)

thetas <- seq(0, 7, by = 0.005)
#likelihoodunif <- ?
```

Maximum should be 7?

```
plot(data.frame(thetas, punif(thetas, 0, 7)))
```



Exercise 2.3

A sequence of three nucleotides (a codon) taken in a coding region of a gene can be transcribed into one of 20 possible amino acids. There are 4^3 = 64 possible coding sequences but only 20 amino acids. We say the genetic code is redundant. The multiplicity (the number of codons that code fro the same amino acid) varies from two to six. The different codon spellings do not work with equal probabilities.

```
mtb = read.table("../../data/M_tuberculosis.txt", header = TRUE)
head(mtb, n = 4)
```

```
##   AmAcid Codon Number PerThous
## 1    Gly   GGG  25874    19.25
## 2    Gly   GGA  13306     9.90
## 3    Gly   GGT  25320    18.84
## 4    Gly   GGC  68310    50.82
```

The codons for profile are of the form CC* and they occur with the following frequencies:

```
pro  =  mtb[ mtb$AmAcid == "Pro", "Number"]
pro/sum(pro)
```

```
## [1] 0.54302025 0.10532985 0.05859765 0.29305225
```

Explore `mtb` using `table` to tabulate `AmAcid` and `Codon` varibles.

```
table(mtb$AmAcid)
```

```
##
## Ala Arg Asn Asp Cys End Gln Glu Gly His Ile Leu Lys Met Phe Pro Ser Thr
##   4   6   2   2   2   3   2   2   4   2   3   6   2   1   2   4   6   4
## Trp Tyr Val
##   1   2   4
```

```
table(mtb$Codon)
```

```
##
## AAA AAC AAG AAT ACA ACC ACG ACT AGA AGC AGG AGT ATA ATC ATG ATT CAA CAC
##   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1
## CAG CAT CCA CCC CCG CCT CGA CGC CGG CGT CTA CTC CTG CTT GAA GAC GAG GAT
##   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1
## GCA GCC GCG GCT GGA GGC GGG GGT GTA GTC GTG GTT TAA TAC TAG TAT TCA TCC
##   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1   1
## TCG TCT TGA TGC TGG TGT TTA TTC TTG TTT
##   1   1   1   1   1   1   1   1   1   1
```

How was the `PerThous` varible created?

```
mtb$PerThous
```

```
##  [1] 19.25  9.90 18.84 50.82 30.58 16.19 15.75 42.17 40.13  4.74  8.01
## [12] 32.66 48.65 12.85 10.91 59.84  3.19  1.29  3.54 14.48 15.10  5.27
## [23]  5.29 19.91 18.43  2.20  6.46 33.89 15.63  4.54  3.67 35.18 14.62
## [34]  1.62  2.22  6.63  0.88  0.47  6.08 14.62 17.93  1.62  6.17 23.35
## [45] 19.36  3.55  2.22 11.54 24.62  7.20  8.45 28.35 22.77  8.10  6.42
## [56] 15.83 50.45  4.74  5.44 17.27 31.56  6.12  3.41 17.03
```

```
length(mtb$PerThous)
```

```
## [1] 64
```

```
mtb$Number/sum(mtb$Number)*1000
```

```
##  [1] 19.2482944  9.8986552 18.8361604 50.8174611 30.5775158 16.1929977
##  [7] 15.7451554 42.1708303 40.1287584  4.7402849  8.0105756 32.6649670
## [13] 48.6541296 12.8460828 10.9066725 59.8449811  3.1884591  1.2944281
## [19]  3.5440548 14.4782525 15.1001731  5.2669832  5.2885570 19.9096430
## [25] 18.4299778  2.1960642  6.4617255 33.8864906 15.6261275  4.5431450
## [31]  3.6742416 35.1831504 14.6166224  1.6232426  2.2191258  6.6320841
## [37]  0.8822941  0.4686722  6.0771167 14.6173663 17.9337803  1.6202669
## [43]  6.1693633 23.3525241 19.3643465  3.5544698  2.2198698 11.5404959
## [49] 24.6164513  7.1996983  8.4494909 28.3509507 22.7685436  8.0961269
## [55]  6.4222975 15.8299627 50.4454990  4.7440045  5.4418054 17.2687121
## [61] 31.5602396  6.1217521  3.4056849 17.0321442
```

Write an R function that you can apply to the table to find which of the amino acids show sthe strongest *codon bias*, i.e., the strongest departure from uniform distribution among its possible spellings

```
numSpelling <- table(mtb$AmAcid)
expected <- 1/numSpelling


obsAA <- function(tab){
  deviation <- c()
  codon <- c()
  aa <- c()
  for(i in as.character(unique(tab$AmAcid))){
    sub <- subset(tab, AmAcid == i)
    deviation <- append(deviation, (sub$PerThous-sum(sub$PerThous)/nrow(sub))^2/sum(sub$PerThous)/nrow(s
    codon <- append(codon, as.character(sub$Codon))
    aa <- append(aa, as.character(sub$AmAcid))
  }
  data.frame(aa, codon, deviation)
}
```

```
t <- obsAA(mtb)
t[t$deviation == max(t$deviation),]
```

```
##     aa codon deviation
## 36 Ile   ATC  3.042324
```

Exercise 2.4

Display GC content in a running window along the sequence of **Staphylococcus aureus**. Read in a `fasta` file sequence form a file.

```
library("Biostrings")
staph = readDNAStringSet("http://bios221.stanford.edu/data/staphsequence.ffn.txt", "fasta")
```

Look at the complete staph object and then display the first three sequences in the set

```
staph
```

```
##   A DNAStringSet instance of length 2650
##         width seq                                               names
##     [1]  1362 ATGTCGGAAAAAGAAATTT...AGAAATAAGAAATGTATAA lcl|NC_002952.2_c...
##     [2]  1134 ATGATGGAATTCACTATTA...ACCAATCAGAACTTACTAA lcl|NC_002952.2_c...
##     [3]   246 GTGATTATTTTGGTTCAAG...TCATCAAGGTGAACAATGA lcl|NC_002952.2_c...
##     [4]  1113 ATGAAGTTAAATACACTCC...AGGTGAAATTATAAAGTAA lcl|NC_002952.2_c...
##     [5]  1932 GTGACTGCATTGTCAGATG...TGCAAACTTAGACTTCTAA lcl|NC_002952.2_c...
##     ...   ... ...
## [2646]   720 ATGACTGTAGAATGGTTAG...TCCTTTACTTGAAAAATAA lcl|NC_002952.2_c...
## [2647]  1878 GTGGTTCAAGAATATGATG...CCAAAGGGTGAGTGACTAA lcl|NC_002952.2_c...
## [2648]  1380 ATGGATTTAGATACAATTA...ATTCTGCTTAGGTAAATAG lcl|NC_002952.2_c...
## [2649]   348 TTGGAAAAAGCTTACCGAA...TAATAAAAAGATTAAGTAA lcl|NC_002952.2_c...
## [2650]   138 ATGGTAAAACGTACTTATC...TAAAGTTTTATCTGCATAA lcl|NC_002952.2_c...
```

```
staph[1:3,]
```

```
##   A DNAStringSet instance of length 3
##     width seq                                               names
## [1]  1362 ATGTCGGAAAAAGAAATTTGG...AAGAAATAAGAAATGTATAA lcl|NC_002952.2_c...
## [2]  1134 ATGATGGAATTCACTATTAAA...TACCAATCAGAACTTACTAA lcl|NC_002952.2_c...
## [3]   246 GTGATTATTTTGGTTCAAGAA...TTCATCAAGGTGAACAATGA lcl|NC_002952.2_c...
```

Find the GC content in sequence windows of width 100

```
window = 100
#starts = seq(1, length(staph$seq) - window, by = window)
#ends   = starts + window - 1
#GC_content = lapply(staph, function(x) {
#letterFrequency(x, letters = "GC", OR = 0)
#
#  sum(letterFrequency(staph[[1]][starts[i]:ends[i]], letters = "GC", OR = 0))

#  countPattern(shineDalgarno, ecoli[starts[i]:ends[i]],
#               max.mismatch = 0)
#}, numeric(1))
#table(numMatches)

#letterFrequency(staph[[1]], letters = "GC", OR = 0)
```