Class 7: Machine Learning

Michelle Woo

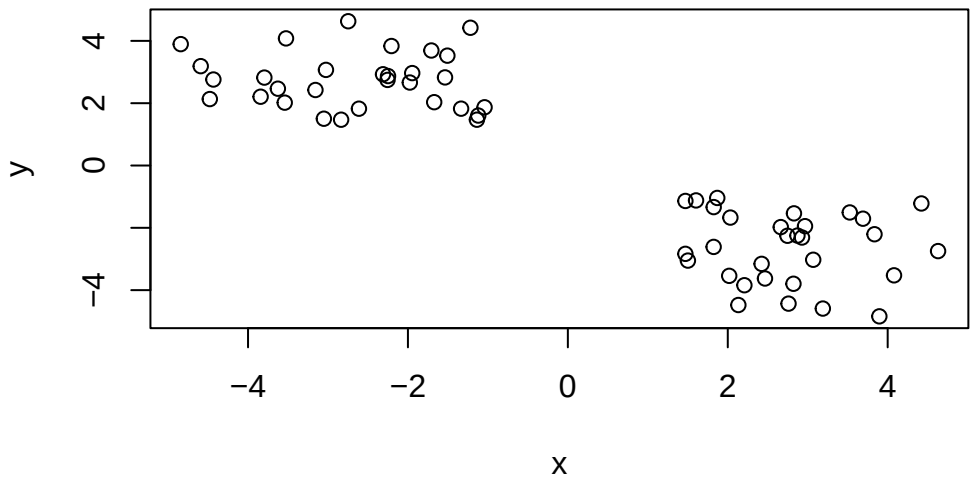
Example of K-means clustering

First step is to make up some data with a known structure, so we know what the answer should be

```
# generating random data
tmp <- c(rnorm(30, mean=-3), rnorm(30, mean=3))
tmp
```

```
 [1] -2.612790 -3.156766 -3.626662 -1.536235 -1.334884 -2.835436 -1.041552
 [8] -2.311686 -3.051507 -3.524817 -3.842329 -1.976277 -3.540879 -1.120426
[15] -4.433053 -2.746839 -4.591414 -2.206353 -4.477759 -3.025358 -1.946538
[22] -4.841679 -3.795691 -1.671556 -2.255078 -1.137502 -1.506960 -1.707061
[29] -1.218545 -2.248848  2.871401  4.421463  3.690979  3.526125  1.470185
[36]  2.747008  2.032718  2.821729  3.895937  2.966172  3.068899  2.132654
[43]  3.835145  3.188996  4.630303  2.759798  1.603296  2.018009  2.662816
[50]  2.207944  4.079429  1.501657  2.928335  1.868611  1.469840  1.824072
[57]  2.827889  2.465483  2.423512  1.823428
```

```
# visualizing in 3D
x <- cbind(x=tmp, y=rev(tmp))
plot(x)
```



Now we have some structured data in x. Let's see if k-means is able to identify the two groups

```
k <- kmeans(x, centers=2, nst=20)
k
```

```
K-means clustering with 2 clusters of sizes 30, 30

Cluster means:
          x         y
1 -2.644083  2.725461
2  2.725461 -2.644083

Clustering vector:
 [1] 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 2 2 2 2 2 2 2 2
[39] 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2

Within cluster sum of squares by cluster:
[1] 60.92531 60.92531
 (between_SS / total_SS =  87.7 %)

Available components:
```

```
[1] "cluster"      "centers"      "totss"        "withinss"     "tot.withinss"
[6] "betweenss"    "size"         "iter"         "ifault"
```

Let's explore and better understand k:

```
# how many elements are in each group?
k$size
```

```
[1] 30 30
```

```
k$centers
```

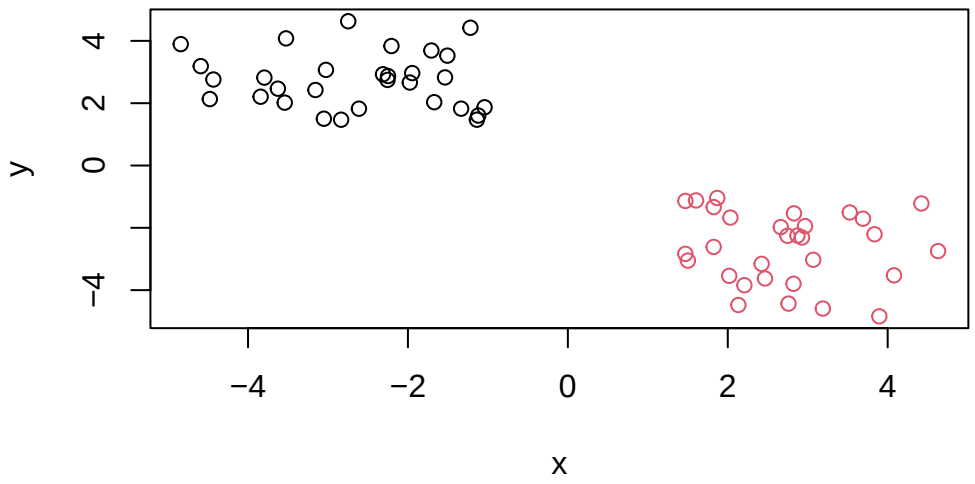
```
          x         y
1 -2.644083  2.725461
2  2.725461 -2.644083
```

```
# able to use this to color the plot
k$cluster
```

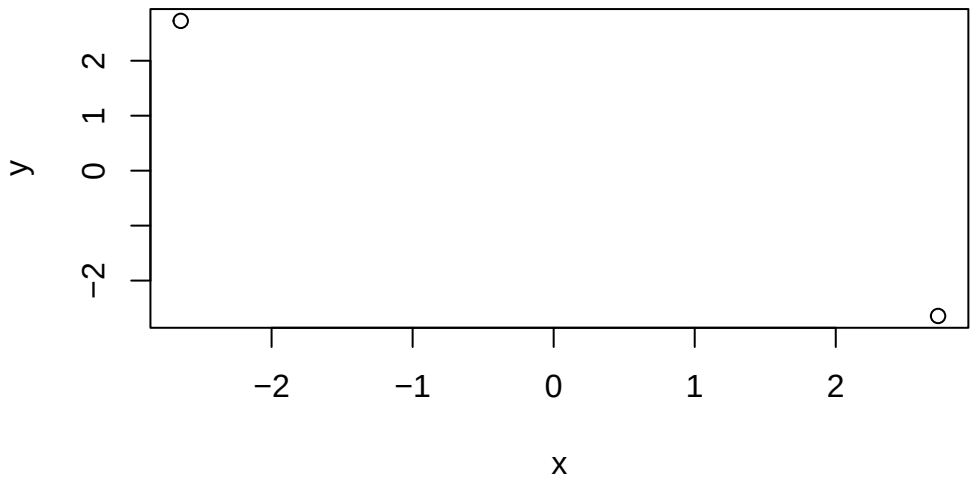
```
 [1] 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 2 2 2 2 2 2 2 2
[39] 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2
```

Refining the plot:

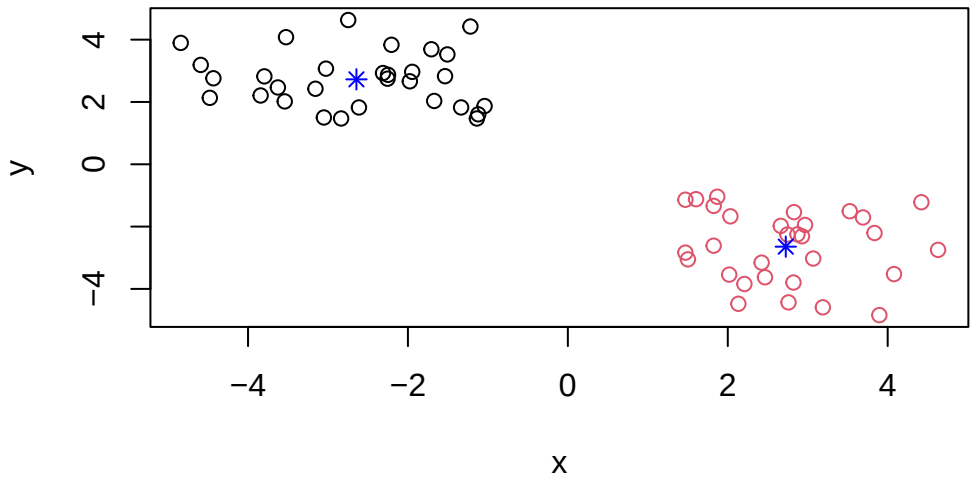
```
# coloring the different groups
plot(x, col=k$cluster)
```




```
# adding in cluster centers, plot(x, col=k$cluster)
plot(k$centers)
```



```r
# want to overlap the two above
plot(x, col=k$cluster)
points(k$centers, col = 'blue', pch = 8)
```



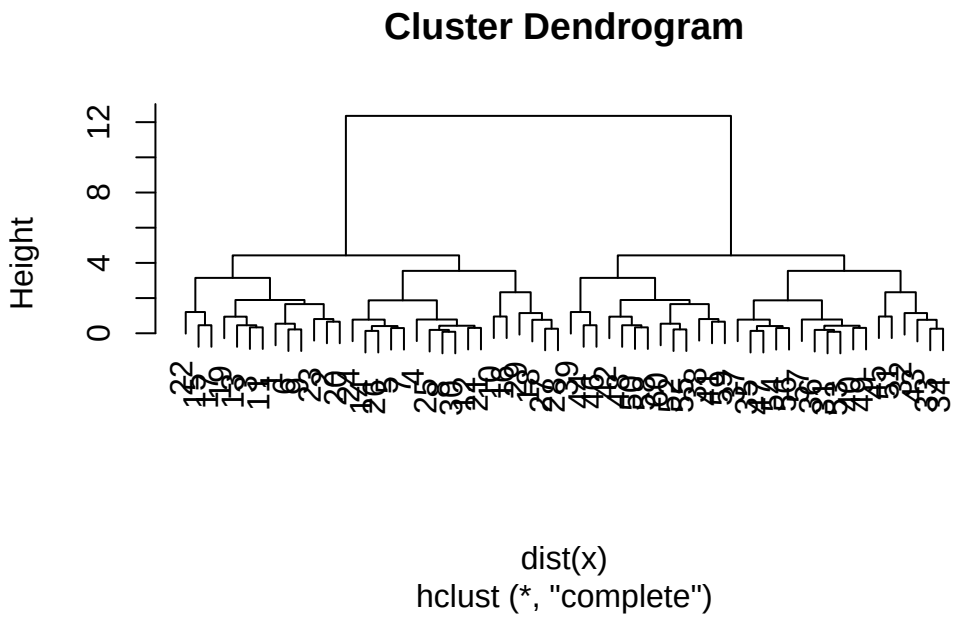
Example of Hierarchical Clustering

Let's use the same data as before, which we stored in `x`. We will use the `hclust()` function dist(x) calculates the distance between all the points, this is input required for clustering

```r
clustering <- hclust(dist(x))
clustering
```

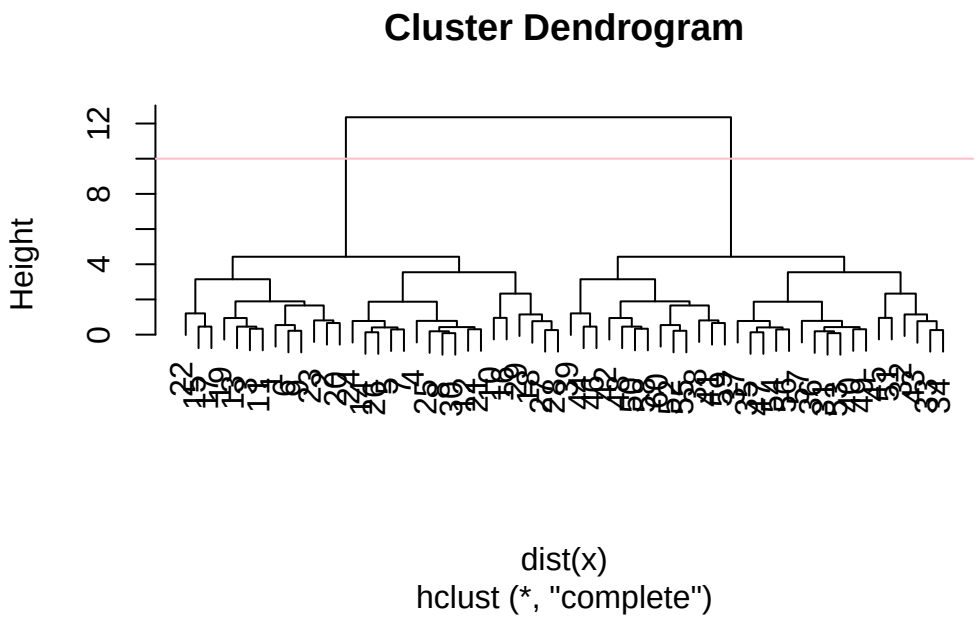
```
Call:
hclust(d = dist(x))

Cluster method   : complete
Distance         : euclidean
Number of objects: 60
```

```
# results in tree, plot function gives something different
plot(clustering)
```



Lets add a horizontal line

```
plot(clustering)
abline(h=10, col='pink') # results in 6 classifications
```



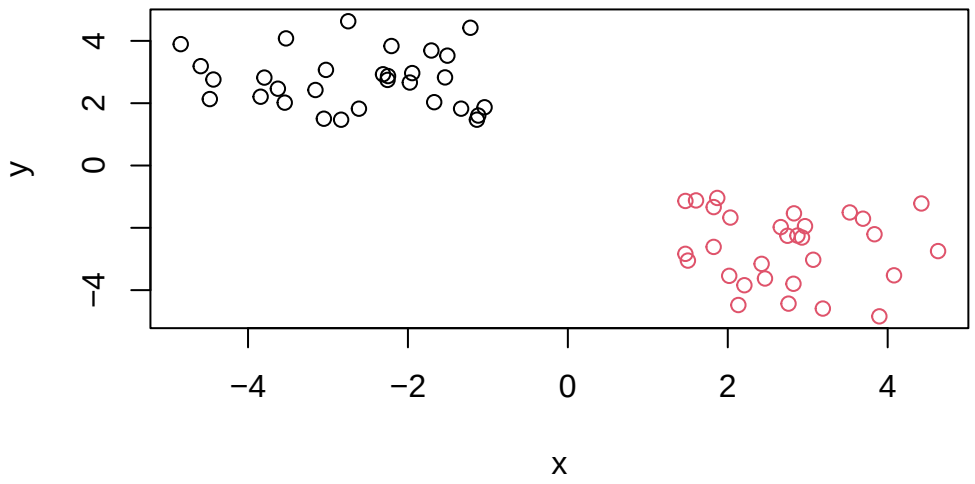
To get our results (i.e. membership vector) we need to 'cut' the tree at the chosen height. The function for doing that is `cutree()`

```
# able to get memebership clustering
subgroups <- cutree(clustering, h=10)
subgroups
```

```
 [1] 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 2 2 2 2 2 2 2 2
[39] 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2
```

Plotting this:

```
plot(x, col= subgroups)
```



You can cut your tree with the number of clusters you want:

```
cutree(clustering, k=2)
```

```
 [1] 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 2 2 2 2 2 2 2 2
[39] 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2 2
```

Principal Component Analysis (PCA)

PCA of UK food

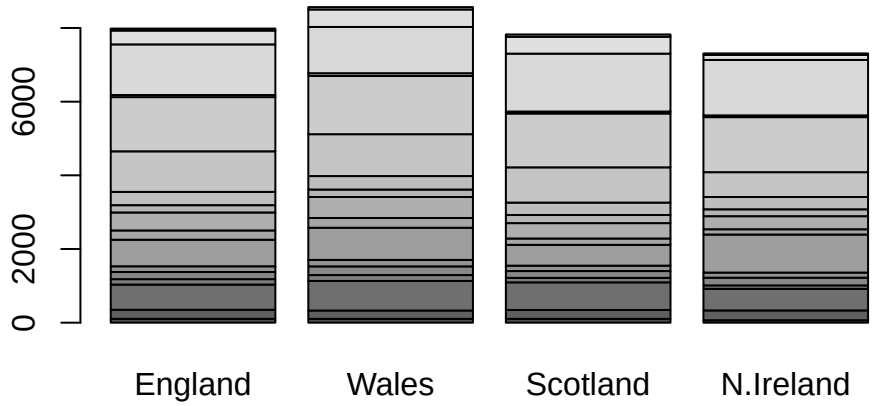
First, we need to read the data

```
url <- "https://tinyurl.com/UK-foods"

# making sure foods are the first column and for our rows
x <- read.csv(url, row.names=1)
head(x)
```

```
              England Wales Scotland N.Ireland
Cheese            105   103      103        66
Carcass_meat      245   227      242       267
Other_meat        685   803      750       586
Fish              147   160      122        93
Fats_and_oils     193   235      184       209
Sugars            156   175      147       139
```

Now we can generate some basic visualizations. We need to make x as a matrix to be able to plot it

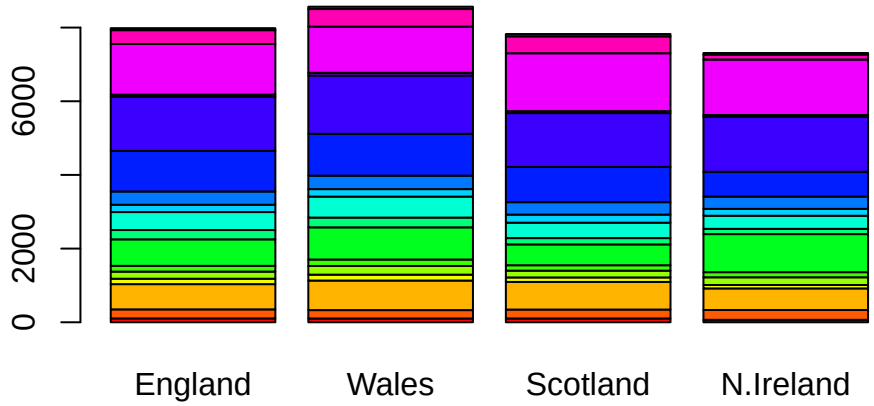
```
barplot(as.matrix(x))
```




```
rainbow(nrow(x))
```

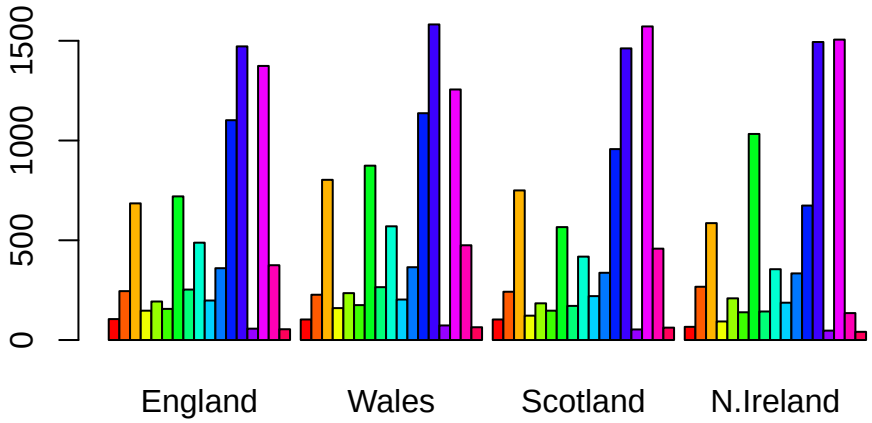
```
 [1] "#FF0000" "#FF5A00" "#FFB400" "#F0FF00" "#96FF00" "#3CFF00" "#00FF1E"
 [8] "#00FF78" "#00FFD2" "#00D2FF" "#0078FF" "#001EFF" "#3C00FF" "#9600FF"
[15] "#F000FF" "#FF00B4" "#FF005A"
```

```
# combining - giving color to the plot
barplot(as.matrix(x), col=rainbow(nrow(x)))
```



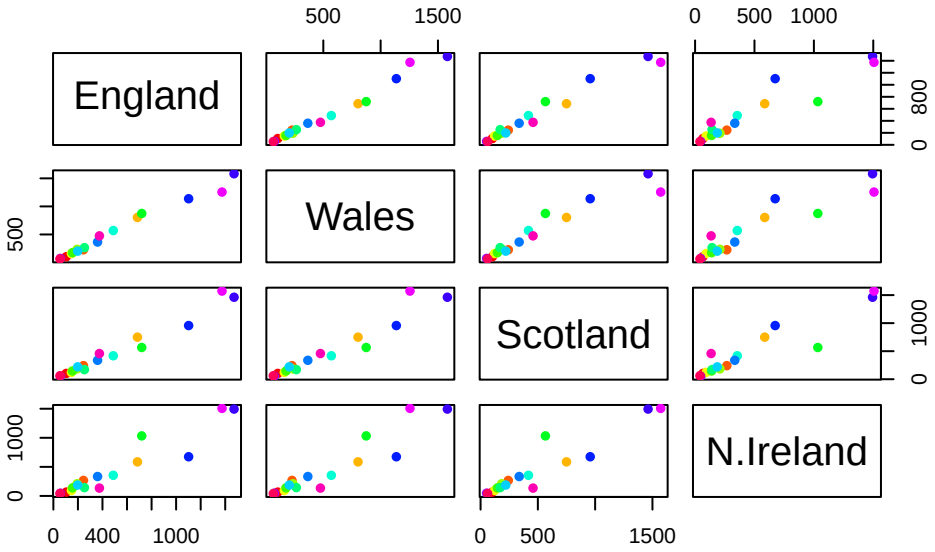
Lets refine our barplot

```
barplot(as.matrix(x), col=rainbow(nrow(x)), beside = T)
```



Other visualizations that can be useful:

```
pairs(x, col=rainbow(nrow(x)), pch=16)
```



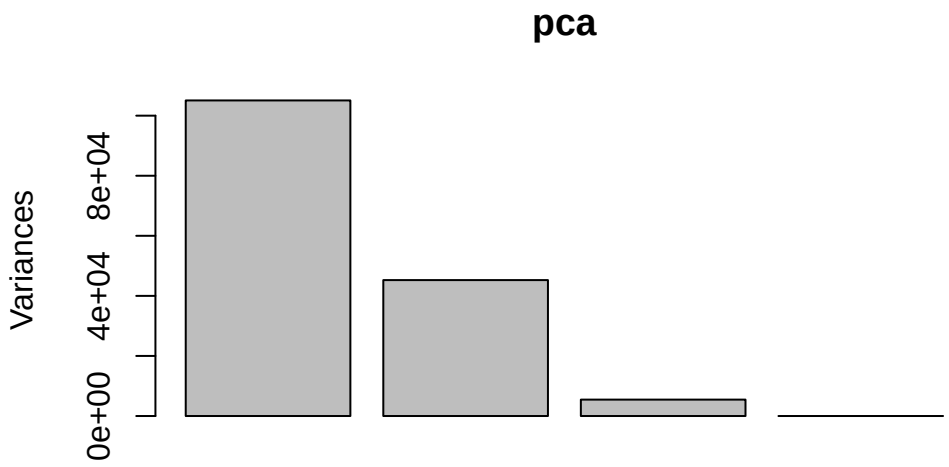
Lets apply PCA. For that, we need to use the command `prcomp()`. This function expects the transpose of our data

```
# t flips the rows and columns
# transpose_matrix <- t(x)
# pca <- prcomp(transpose_matrix)
pca <- prcomp(t(x))

summary(pca)
```

```
Importance of components:
                            PC1      PC2      PC3        PC4
Standard deviation     324.1502 212.7478 73.87622 4.189e-14
Proportion of Variance   0.6744   0.2905  0.03503 0.000e+00
Cumulative Proportion    0.6744   0.9650  1.00000 1.000e+00
```

Lets plot the PCA results:

```
plot(pca)
```



We need to access the results of the PCA analysis

```
attributes(pca)
```

```
$names
[1] "sdev"     "rotation" "center"   "scale"    "x"

$class
[1] "prcomp"
```

We can explore the `pca$x` dataframe:

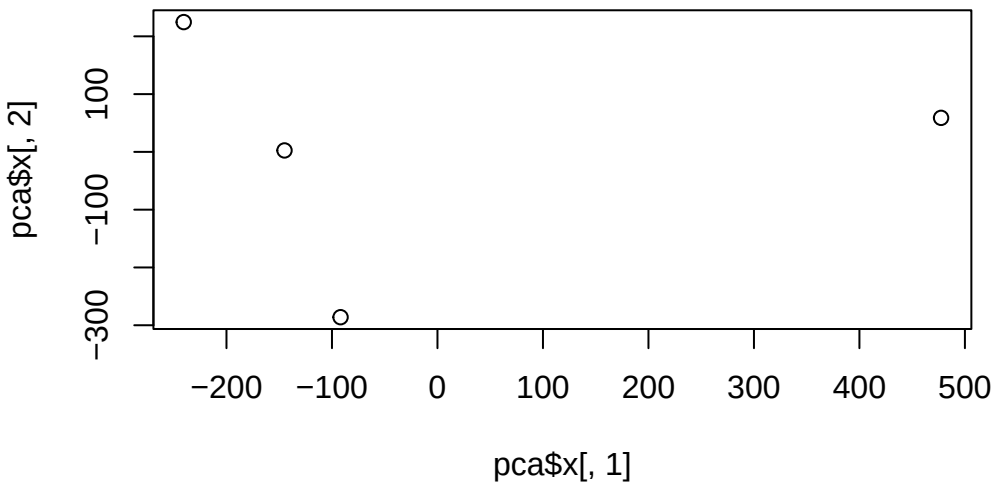
(all 4 components, we can now place 2 in x axis and 2 in y axis)

```
pca$x
```

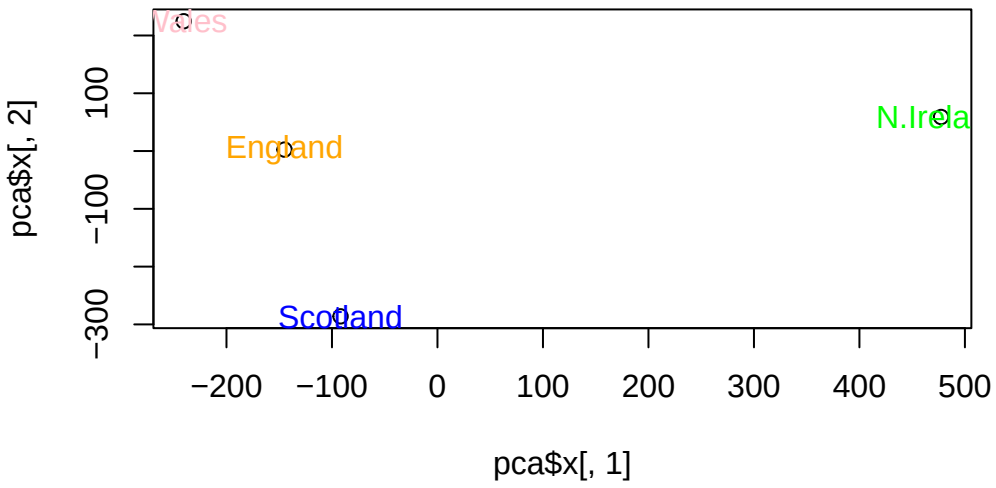
```
                PC1         PC2         PC3           PC4
England   -144.99315    2.532999 -105.768945  2.842865e-14
Wales     -240.52915  224.646925   56.475555  7.804382e-13
Scotland   -91.86934 -286.081786   44.415495 -9.614462e-13
N.Ireland  477.39164   58.901862    4.877895  1.448078e-13
```

Plotting:

```
plot(x=pca$x[,1], y=pca$x[,2])
```




```
# overlay country names and adding colors
plot(pca$x[,1], pca$x[,2] )
colors_countries <- c('orange', 'pink', 'blue', 'green')
text(x=pca$x[,1], y=pca$x[,2], colnames(x), col=colors_countries)
```



Q1. How many rows and columns are in your new data frame named `x`? What R functions could you use to answer this questions?

PCA of RNA-seq dataset

First step as always is to load the data:

```
url2 <- "https://tinyurl.com/expression-CSV"
rna.data <- read.csv(url2, row.names=1)
head(rna.data)
```

```
       wt1 wt2  wt3  wt4 wt5 ko1 ko2 ko3 ko4 ko5
gene1  439 458  408  429 420  90  88  86  90  93
gene2  219 200  204  210 187 427 423 434 433 426
gene3 1006 989 1030 1017 973 252 237 238 226 210
gene4  783 792  829  856 760 849 856 835 885 894
gene5  181 249  204  244 225 277 305 272 270 279
gene6  460 502  491  491 493 612 594 577 618 638
```

Q. How many genes and samples are in this data set?

```r
dim(rna.data)
```

```
[1] 100  10
```

There are 100 genes, and 10 samples.

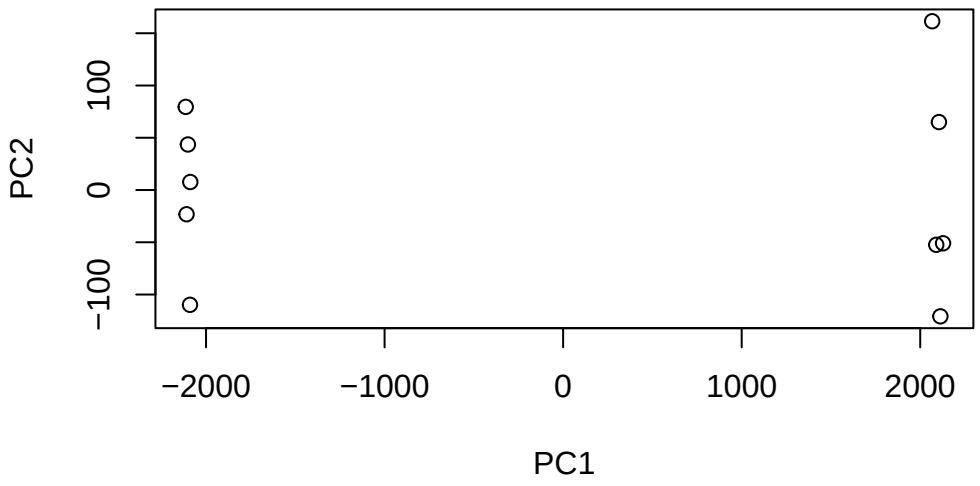
Now lets apply PCA:

```r
pca_rna <- prcomp(t(rna.data))
summary(pca_rna)
```

```
Importance of components:
                            PC1     PC2      PC3      PC4      PC5      PC6
Standard deviation     2214.2633 88.9209 84.33908 77.74094 69.66341 67.78516
Proportion of Variance    0.9917  0.0016  0.00144  0.00122  0.00098  0.00093
Cumulative Proportion     0.9917  0.9933  0.99471  0.99593  0.99691  0.99784
                           PC7      PC8      PC9      PC10
Standard deviation     65.29428 59.90981 53.20803 3.142e-13
Proportion of Variance  0.00086  0.00073  0.00057 0.000e+00
Cumulative Proportion   0.99870  0.99943  1.00000 1.000e+00
```

Lets plot the principal components 1 and 2

```r
plot(pca_rna$x[,1], pca_rna$x[,2], xlab='PC1', ylab='PC2')
```



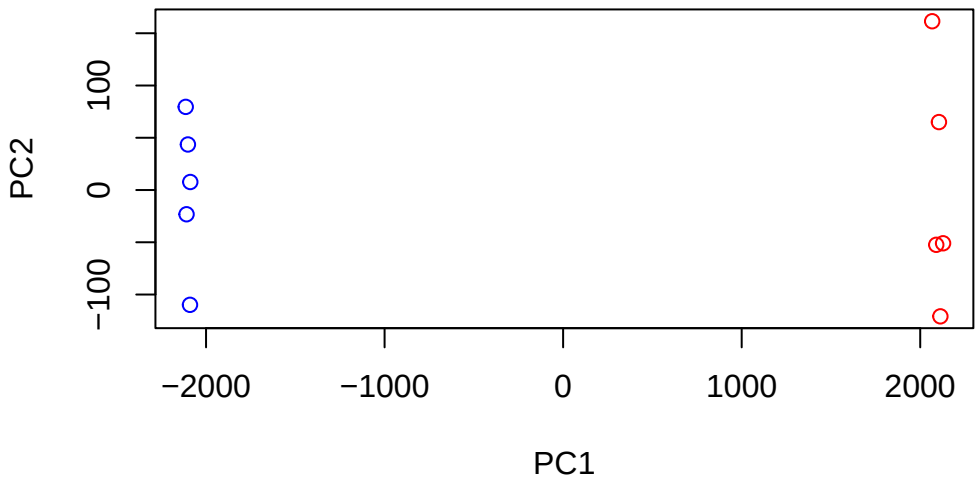

```
# checking the names
colnames(rna.data)
```

```
[1] "wt1" "wt2" "wt3" "wt4" "wt5" "ko1" "ko2" "ko3" "ko4" "ko5"
```

```
# generating a vector that will color the sample
cols_samples <- c(rep('blue', 5), rep('red', 5))
cols_samples
```

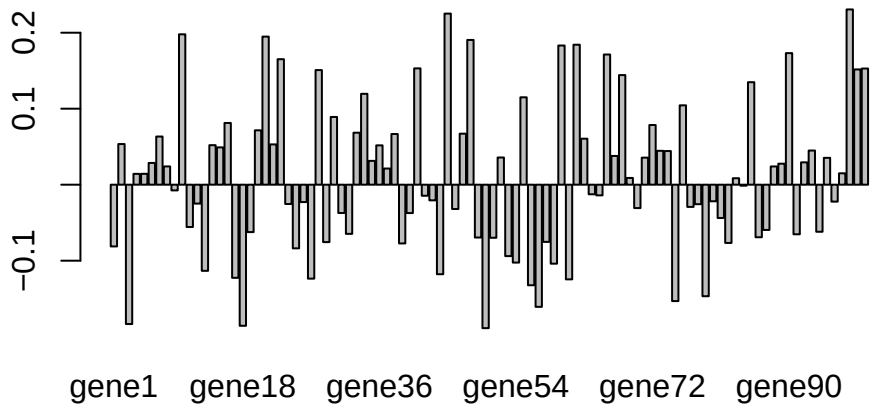
```
[1] "blue" "blue" "blue" "blue" "blue" "red"  "red"  "red"  "red"  "red"
```

```
# applying color to the plot
plot(pca_rna$x[,1], pca_rna$x[,2], xlab='PC1', ylab='PC2', col=cols_samples)
```



Identifying which gene is contributing the most

```
barplot(pca_rna$rotation[,1])
```




```
# identifying under- or overexpression
sort(pca_rna$rotation[,1])
```

```
      gene50       gene18        gene3       gene57       gene75       gene79
-0.188796985 -0.185668500 -0.183374164 -0.160771014 -0.153164404 -0.146803635
      gene56       gene61       gene27       gene17       gene44       gene13
-0.132330117 -0.124572881 -0.123615228 -0.122536548 -0.117808971 -0.113357525
      gene59       gene54       gene53       gene25        gene1       gene39
-0.103935563 -0.102503320 -0.093979884 -0.083761992 -0.081247810 -0.077306742
      gene82       gene29       gene58       gene51       gene49       gene86
-0.076658760 -0.075605635 -0.075274651 -0.069855142 -0.069530208 -0.069165267
      gene91       gene32       gene19       gene94       gene87       gene11
-0.065288752 -0.064721235 -0.062411218 -0.061938300 -0.059547317 -0.055698801
      gene81       gene40       gene31       gene46       gene70       gene77
-0.043780416 -0.037323670 -0.037219970 -0.031990529 -0.030784982 -0.029225446
      gene78       gene24       gene12       gene26       gene96       gene80
-0.025639741 -0.025407507 -0.024870802 -0.022868107 -0.022293151 -0.021824860
      gene43       gene42       gene65       gene64        gene9       gene84
-0.020617052 -0.014550791 -0.014052839 -0.012639567 -0.007495075 -0.001289937
      gene83       gene69        gene4        gene5       gene97       gene37
 0.008504287  0.008871890  0.014242602  0.014303808  0.014994546  0.021280555
      gene88        gene8       gene89        gene6       gene92       gene35
```

0.024015925	0.024026657	0.027652967	0.028634131	0.029394259	0.031349942
gene95	gene71	gene52	gene67	gene74	gene73
0.035342407	0.035589259	0.035802086	0.037840851	0.044286948	0.044581700
gene93	gene15	gene36	gene14	gene22	gene2
0.044940861	0.049090676	0.051765605	0.052004194	0.053013523	0.053465569
gene63	gene7	gene38	gene47	gene33	gene20
0.060529157	0.063389255	0.066665407	0.067141911	0.068437703	0.071571203
gene72	gene16	gene30	gene76	gene55	gene34
0.078551648	0.081254592	0.089150461	0.104435777	0.114988217	0.119604059
gene85	gene68	gene28	gene99	gene100	gene41
0.134907896	0.144227333	0.150812015	0.151678253	0.152877246	0.153077075
gene23	gene66	gene90	gene60	gene62	gene48
0.165155192	0.171311307	0.173156806	0.183139926	0.184203008	0.190495289
gene21	gene10	gene45	gene98		
0.194884023	0.197905454	0.225149201	0.230633225		