

# Class07Lab

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First, read the file that includes the data for the lab:

```
url <- "https://tinyurl.com/UK-foods"
x <- read.csv(url)
x
```

		X	England	Wales	Scotland	N.Ireland
1	Cheese		105	103	103	66
2	Carcass_meat		245	227	242	267
3	Other_meat		685	803	750	586
4	Fish		147	160	122	93
5	Fats_and_oils		193	235	184	209
6	Sugars		156	175	147	139
7	Fresh_potatoes		720	874	566	1033
8	Fresh_Veg		253	265	171	143
9	Other_Veg		488	570	418	355
10	Processed_potatoes		198	203	220	187
11	Processed_Veg		360	365	337	334
12	Fresh_fruit		1102	1137	957	674
13	Cereals		1472	1582	1462	1494
14	Beverages		57	73	53	47
15	Soft_drinks		1374	1256	1572	1506
16	Alcoholic_drinks		375	475	458	135
17	Confectionery		54	64	62	41

###Question 1: How many rows and columns are in your new data named x? What R functions could you use to answer this question?

```
dim(x)
```

```
[1] 17  5
```

##Checking your data: Use the `view()` function to see all data in a new tab. Or use `head()` or `tail()` to preview the first/last 6 rows.

#Preview the first 6 rows:

```
head(x)
```

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

#Fix the code using `rownames()` to rename the first column properly:

```
rownames(x) <- x[,1]  
x <- x[,-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

```
dim(x)
```

```
[1] 17  4
```

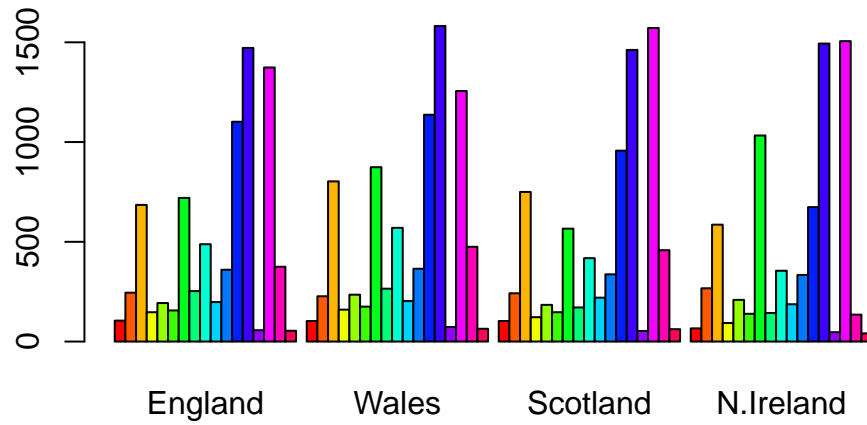
#Another approach: `x <- read.csv(url, row.names=1)` `head(x)`

###Question 2: Which approach to solving the ‘row-names problem’ mentioned above do you prefer and why? Is one approach more robust than another under certain circumstances?

I would prefer the second approach, re-reading the data file and setting the row names as the first column. Running the first approach code block (`x <- x[,-1]`) multiple times would remove additional columns from the dataset.

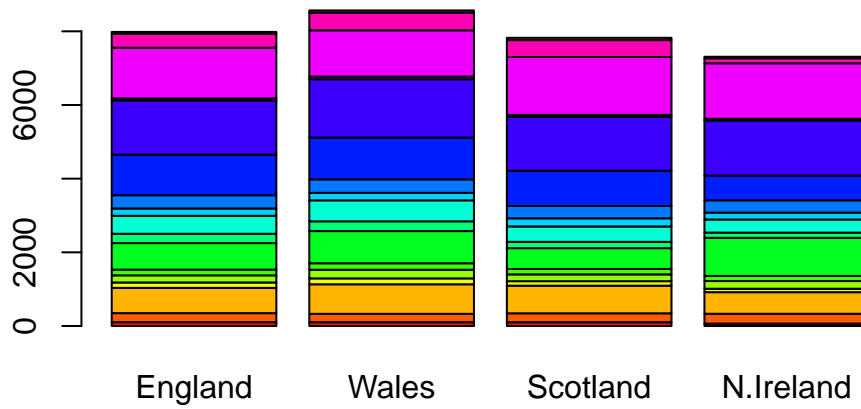
###Spotting major differences and trends:

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



###Question3: Changing what optional argument in the above barplot() function results in the following plot

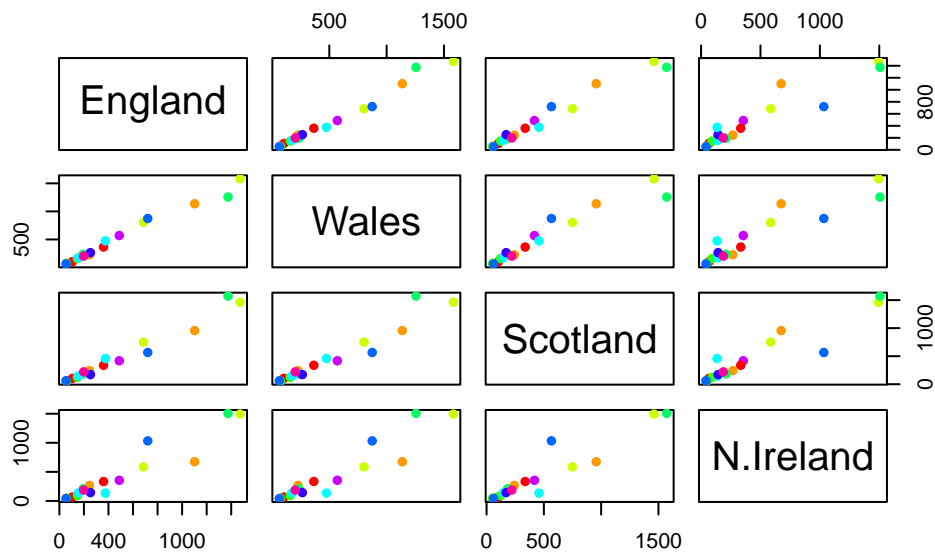
```
barplot(as.matrix(x), beside=F, col=rainbow(nrow(x)))
```



Changing the optional argument **beside** from TRUE to FALSE. False displays it so that the data is stacked instead of side-by-side.

###Question5: Generating all pairwise plots may help somewhat. Can you make sense of the following code and resulting figure? What does it mean if a given point lies on the diagonal for a given plot?

```
pairs(x, col=rainbow(10), pch=16)
```



The above is a scatter plot that represents the relationship between two pairs in 'x'. The diagonal represents the variables plotted against itself, so if there is a dot on the diagonal it means the y-value and x-value are the same.

###Question6: What is the main differences between N. Ireland and other countries of the UK in terms of this data-set?

The main difference between N. Ireland and other countries is that N. Ireland is more clustered near the 500 mark.

##PCA to the rescue: `prcomp()` function can be used for PCA innplementation.

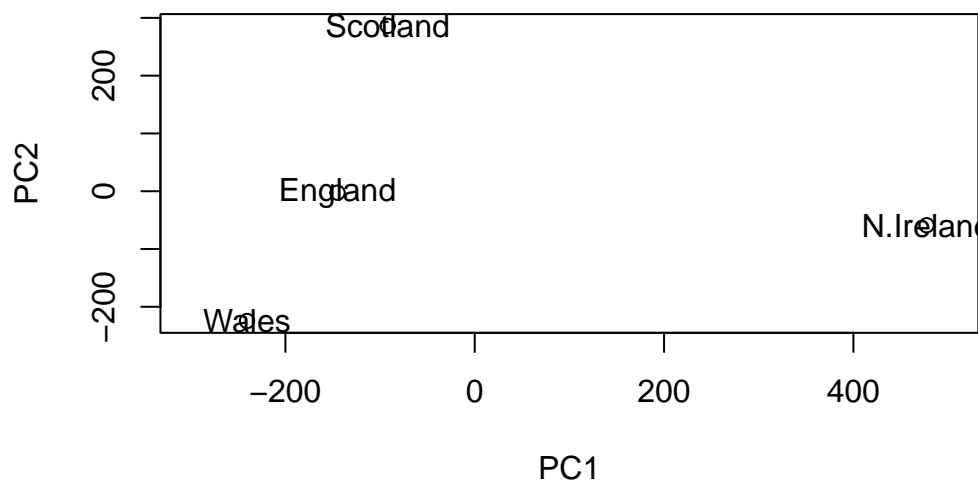
```
pca <- prcomp (t(x))
summary(pca)
```

Importance of components:

	PC1	PC2	PC3	PC4
Standard deviation	324.1502	212.7478	73.87622	3.176e-14
Proportion of Variance	0.6744	0.2905	0.03503	0.000e+00
Cumulative Proportion	0.6744	0.9650	1.00000	1.000e+00

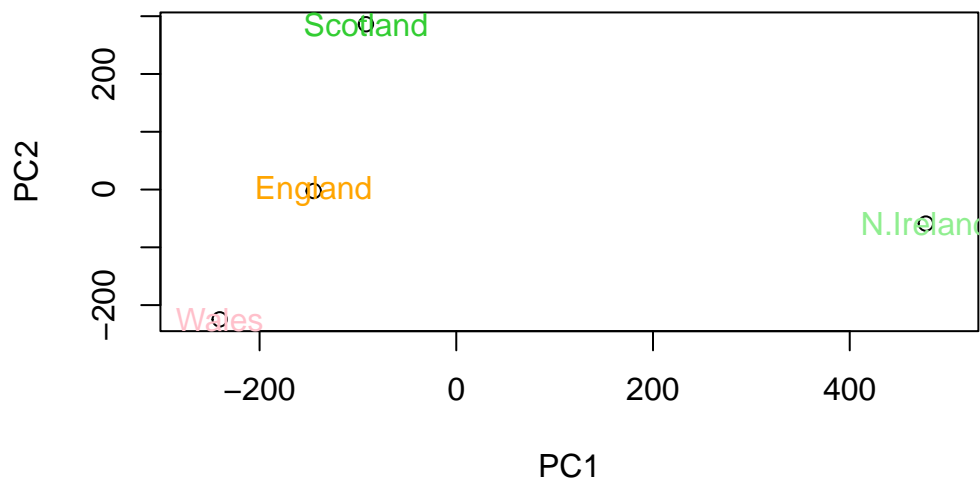
###Question7: Complete the code below to generate a plot of PC1 vs PC2. The second line adds text labels over the data points.

```
plot(pca$x[,1], pca$x[,2], xlab="PC1", ylab="PC2", xlim=c(-300,500))
text(pca$x[,1], pca$x[,2], colnames(x))
```



###Question8: Customize your plot so that the colors of the country names match the colors in our UK and Ireland map and table at start of this document.

```
country_colors <- c("orange", "pink", "lime green", "light green")
plot(pca$x[,1], pca$x[,2], xlab="PC1", ylab="PC2", xlim=c(-270,500))
text(pca$x[,1], pca$x[,2], colnames(x), col=country_colors)
```



## Calculating standard deviation:

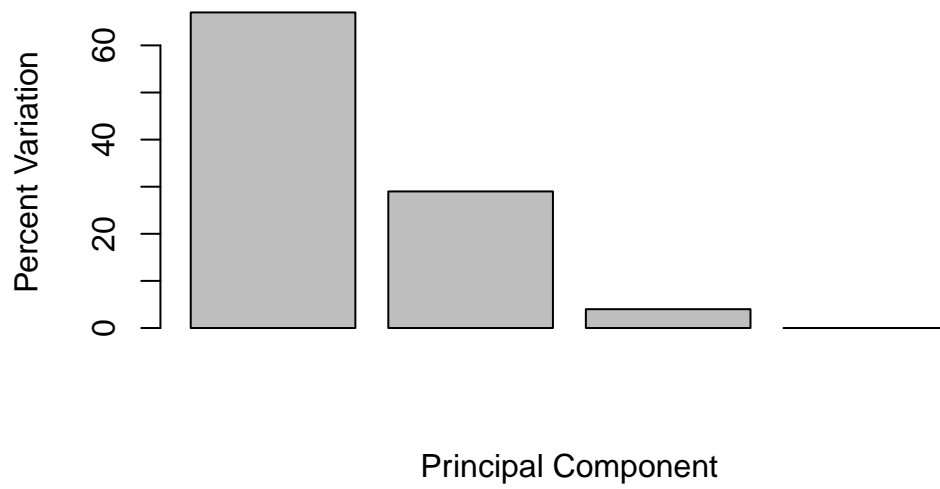
```
v <- round( pca$sdev^2/sum(pca$sdev^2)*100)
v
```

```
[1] 67 29 4 0
```

```
z <- summary(pca)
z$importance
```

	PC1	PC2	PC3	PC4
Standard deviation	324.15019	212.74780	73.87622	3.175833e-14
Proportion of Variance	0.67444	0.29052	0.03503	0.000000e+00
Cumulative Proportion	0.67444	0.96497	1.00000	1.000000e+00

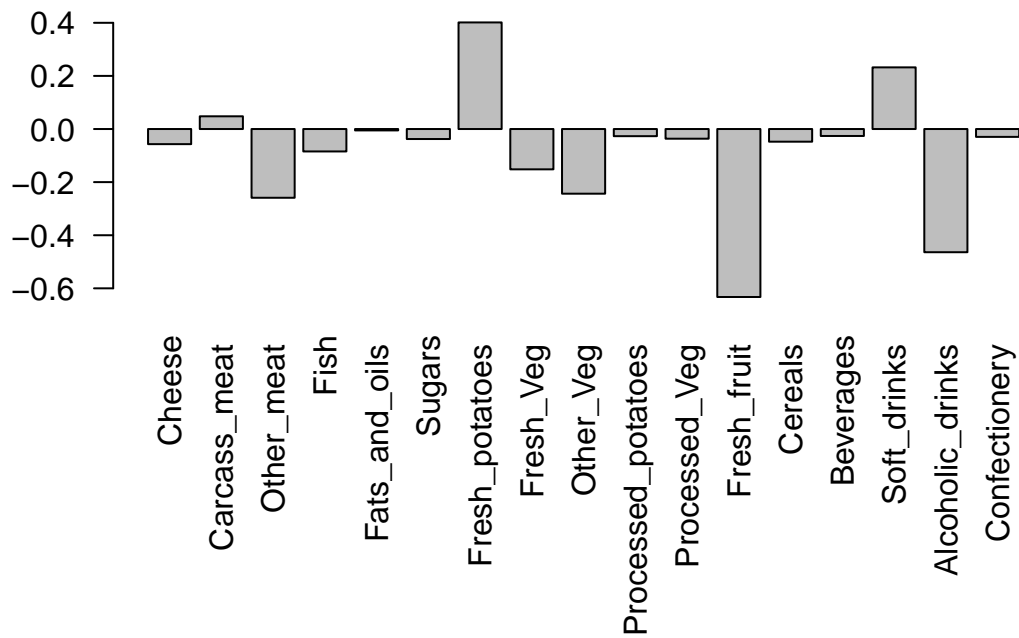
```
barplot(v, xlab="Principal Component", ylab="Percent Variation")
```



##Digging Deeper(variabele loadings):

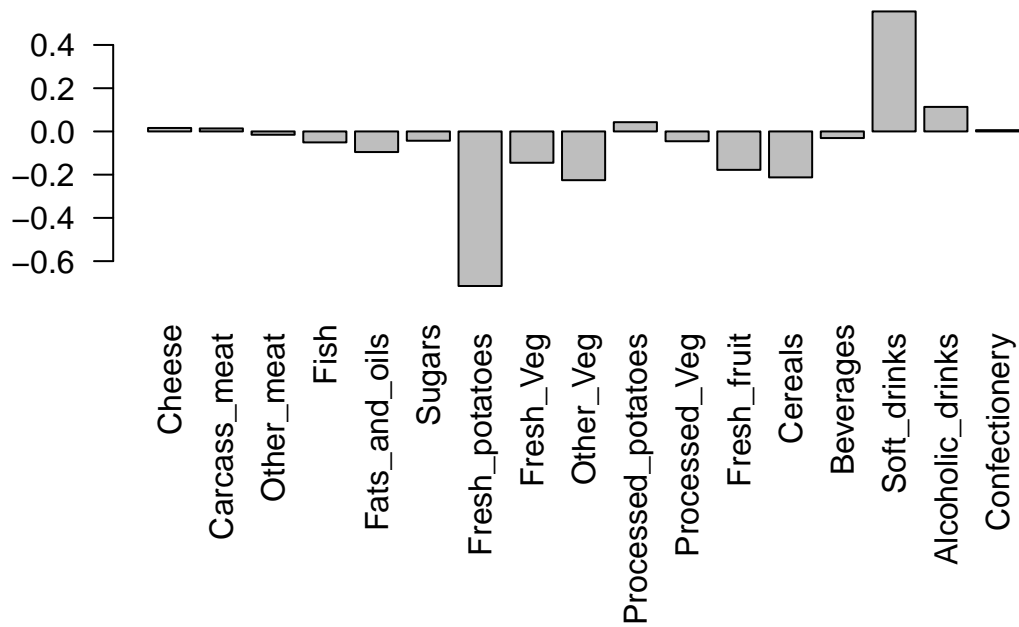
```
par(mar=c(10, 3, 0.35, 0))  
barplot( pca$rotation[,1], las=2 )
```





###Question9: Generate a similar ‘loadings plot’ for PC2. What two food groups feature prominently and what does PC2 mainly tell us about?

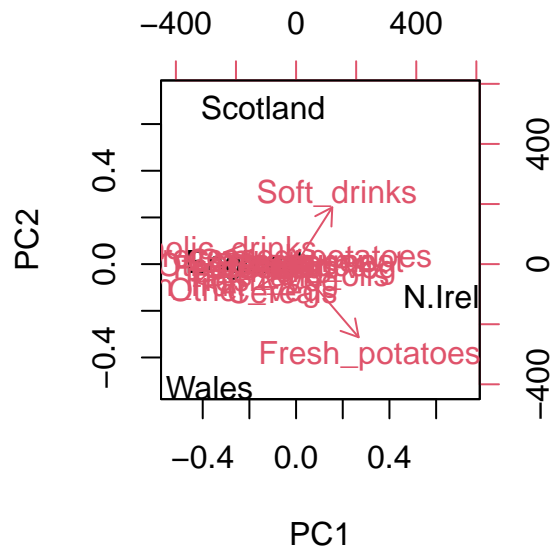
```
par(mar=c(10, 3, 0.35, 0))
barplot( pca$rotation[,2], las=2 )
```



The largest positive loading score is Soft Drinks. The largest negative score is Fresh potatoes. PC2 mainly tells us about what food groups are pushing other countries to the right or left side of the plot.

##Biplots:

```
biplot(pca)
```



##PCA of RNA-seq data: Read the data frame for a small RNA-seq count data set.

```
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

###Question10: How many genes and samples are in this data set? If the samples are columns and the genes are rows, there are 6 genes and 10 samples in this set.

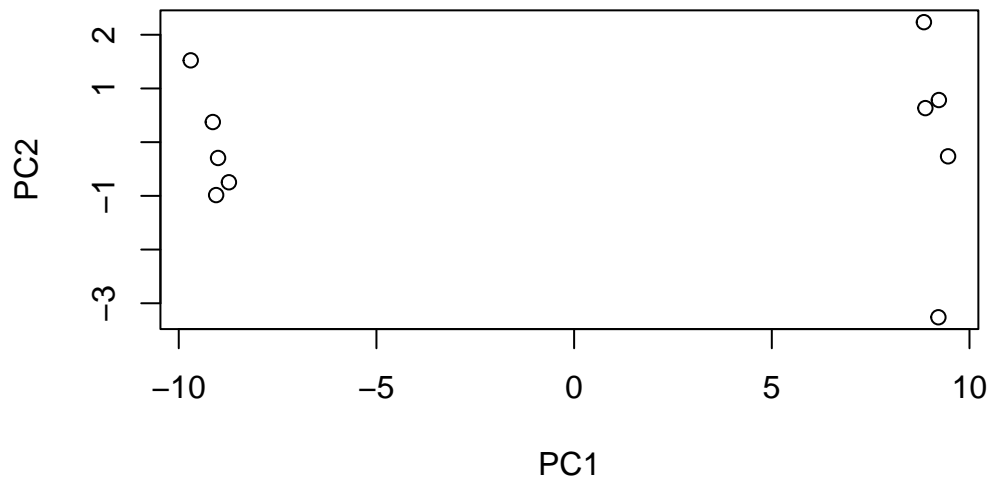
#Consider doing a PCA to better interpret the data:

```
#Begin by transposing the data:

pca <- prcomp(t(rna.data), scale=T)
```

```
#Plot
```

```
plot(pca$x[,1], pca$x[,2], xlab="PC1", ylab="PC2")
```



Examine the summary of the pca:

```
summary(pca)
```

Importance of components:

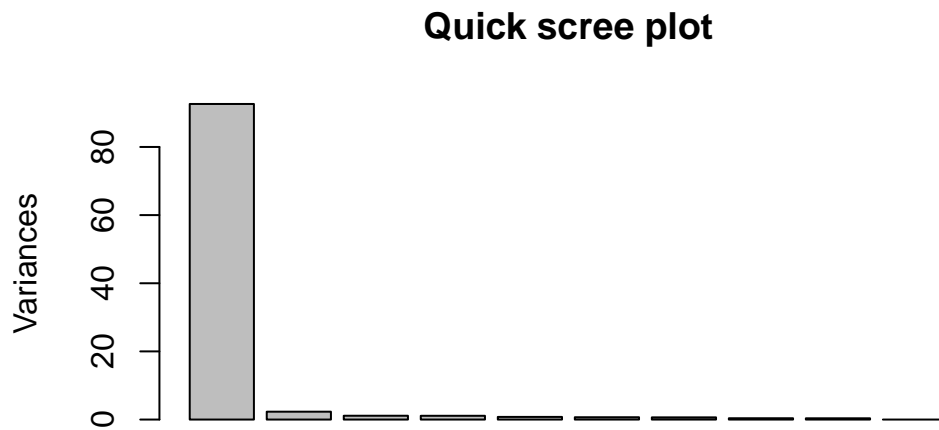
	PC1	PC2	PC3	PC4	PC5	PC6	PC7
Standard deviation	9.6237	1.5198	1.05787	1.05203	0.88062	0.82545	0.80111
Proportion of Variance	0.9262	0.0231	0.01119	0.01107	0.00775	0.00681	0.00642
Cumulative Proportion	0.9262	0.9493	0.96045	0.97152	0.97928	0.98609	0.99251

	PC8	PC9	PC10
Standard deviation	0.62065	0.60342	3.457e-15
Proportion of Variance	0.00385	0.00364	0.000e+00
Cumulative Proportion	0.99636	1.00000	1.000e+00

Obtain Bar plot:

```
plot(pca, main="Quick scree plot")
```



Use `pca$sdev` to calculate variation:

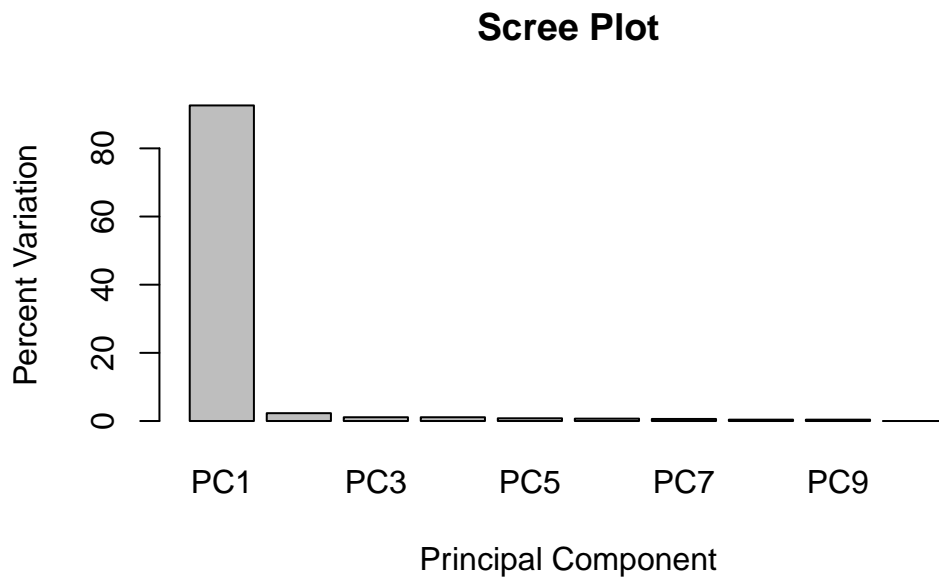
```
#Variation:
pca.var <- pca$sdev^2

#Percent variance:
pca.var.per <- round(pca.var/sum(pca.var)*100, 1)
pca.var.per

[1] 92.6  2.3  1.1  1.1  0.8  0.7  0.6  0.4  0.4  0.0
```

Generate Scree plot again using above data:

```
barplot(pca.var.per, main="Scree Plot",
        names.arg = paste0("PC", 1:10),
        xlab="Principal Component", ylab="Percent Variation")
```



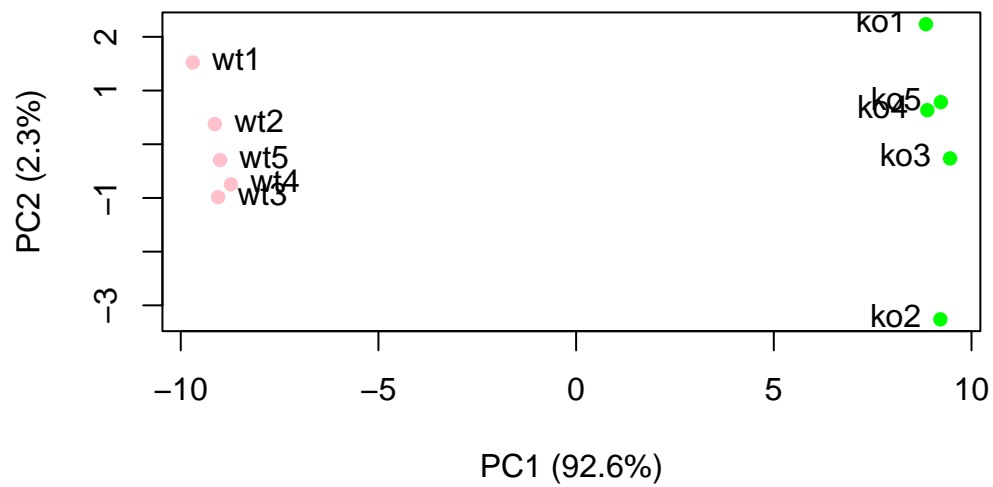
Now to make it colorful:

```
#Add vector for colors:

colvec <- colnames(rna.data)
colvec[grep("wt", colvec)] <- "pink"
colvec[grep("ko", colvec)] <- "green"

plot(pca$x[,1], pca$x[,2], col=colvec, pch=16,
     xlab=paste0("PC1 (", pca.var.per[1], "%)"),
     ylab=paste0("PC2 (", pca.var.per[2], "%)"))

text(pca$x[,1], pca$x[,2], labels = colnames(rna.data), pos=c(rep(4,5), rep(2,5)))
```

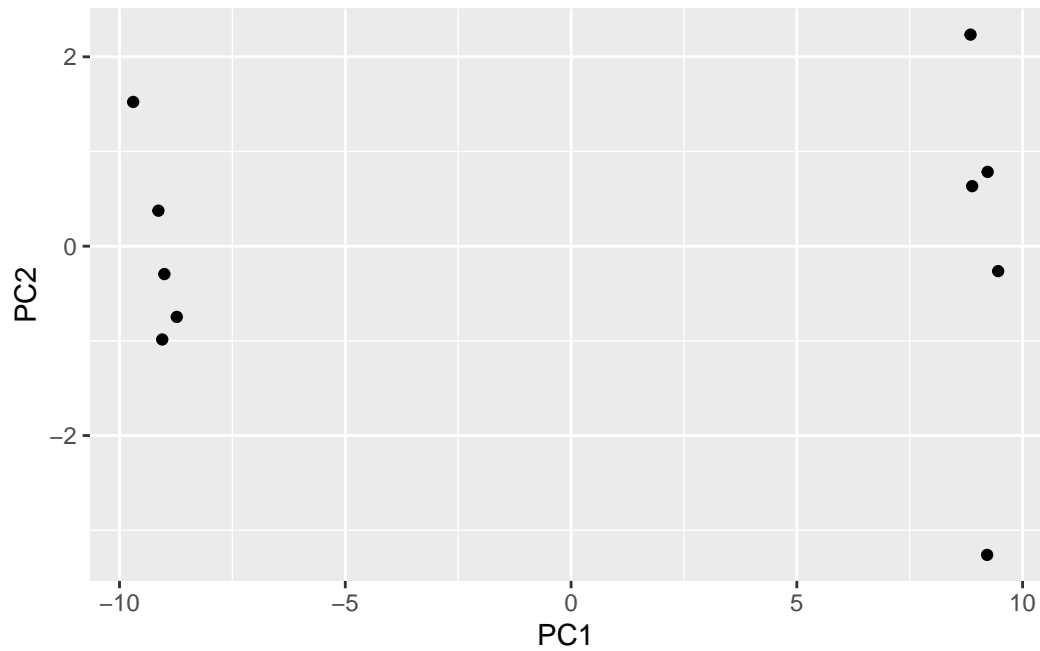


#Using ggplot:

create data frame for ggplot input with PCA results.

```
library(ggplot2)
df <- as.data.frame(pca$x)

ggplot(df) +
  aes(PC1, PC2) +
  geom_point()
```

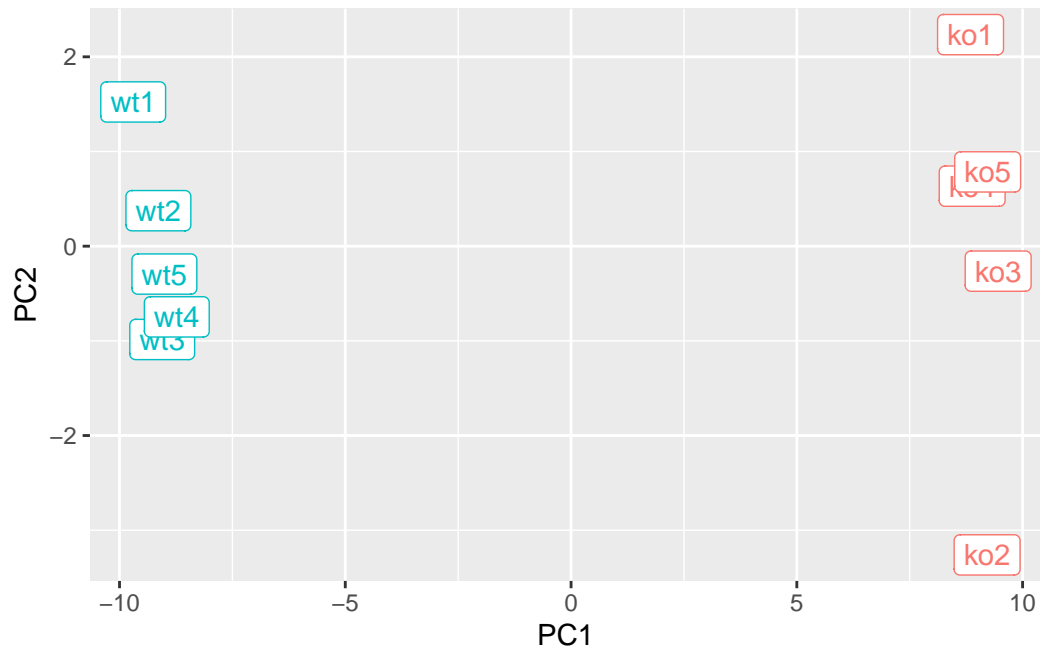


Add conditions:

```
df$samples <- colnames(rna.data)
df$condition <- substr(colnames(rna.data),1,2)

p <- ggplot(df) +
  aes(PC1, PC2, label=samples, col=condition) +
  geom_label(show.legend = FALSE)
p
```

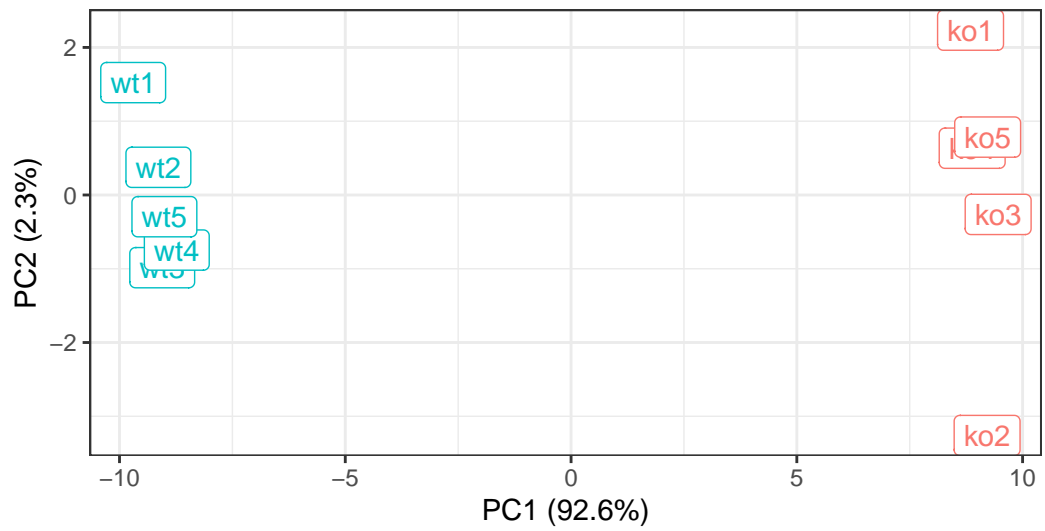




```
p + labs(title="PCA of RNA-Seq Data",
  subtitle = "PC1 separates wild-type from knock-out samples",
  x=paste0("PC1 (", pca.var.per[1], "%)"),
  y=paste0("PC2 (", pca.var.per[2], "%)"),
  caption="Class example data") +
  theme_bw()
```

## PCA of RNA-Seq Data

PC1 separates wild-type from knock-out samples



Class example data