

# hw2

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## Data Import

```
anno = read.table("/Users/Hana/Desktop/STA/sta46/hw2/SampleAnnotation.txt", as.is=TRUE, sep="\t", quote="\"",  
                  row.names=1, header=TRUE)  
x = read.table("/Users/Hana/Desktop/STA/sta46/hw2/expressiondata.txt", as.is=TRUE, sep="\t", quote="\"",  
               x = as.matrix(x)
```

## Define samples and colors and phenotype

```
samples = rownames(anno)  
colors = rainbow(nrow(anno))  
isNorm = anno$TissueType == "norm"  
isSick = anno$TissueType == "sick"  
isAcute = anno$TissueType == "acute"
```

## Data Transformation

It seems to be too dispersed when we try boxplot on the original data. So we transfer data into log-form to do the further analysis.

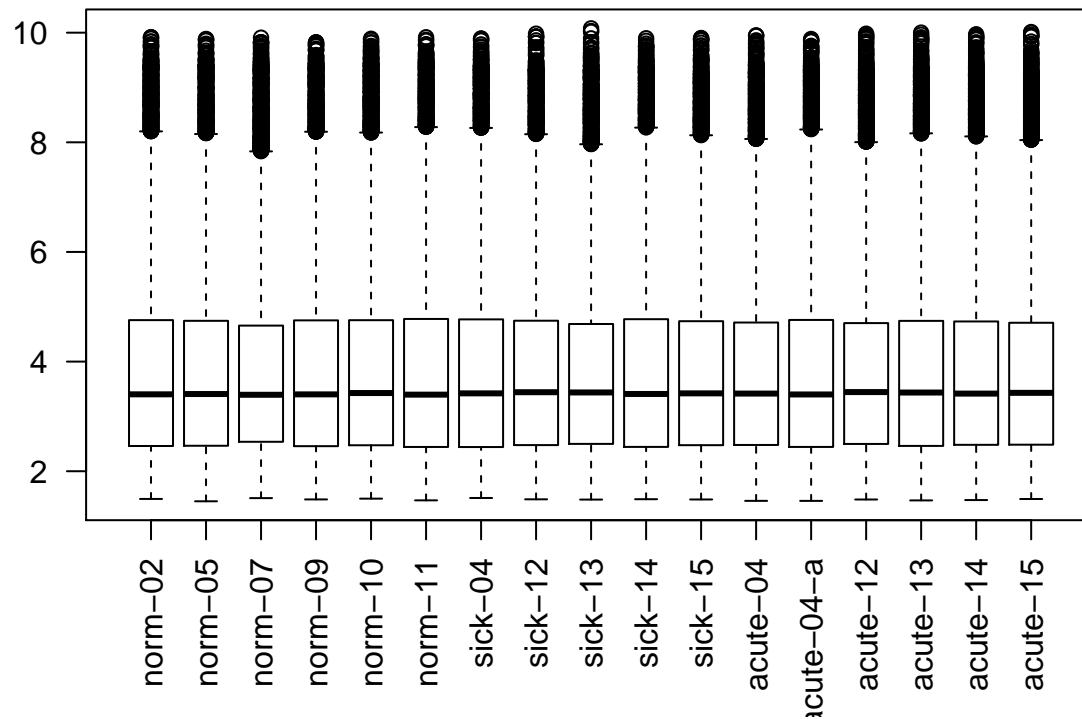
```
data <- log(x)
```

## Distributions

### Boxplot

```
boxplot(data, las=2, main="Boxplot")
```

## Boxplot



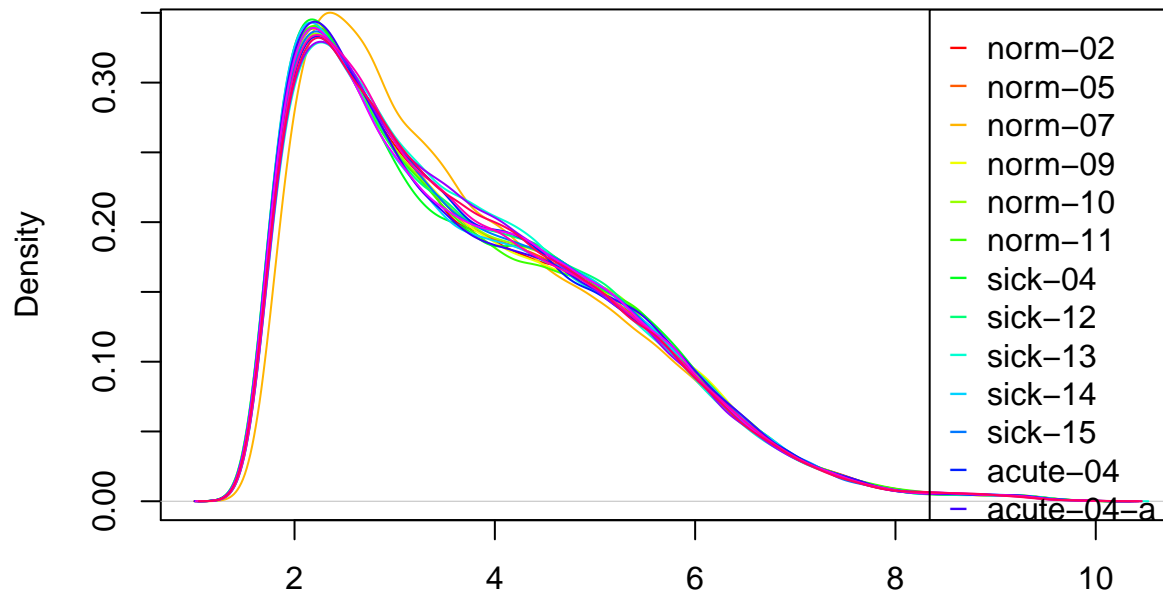
```
outliers = data[data[,1]>6|data[,2]>6|data[,3]>6|data[,4]>6|data[,5]>6|data[,6]>6|data[,7]>6|data[,8]>6
```

The plot shows us that there are some outliers of each sample, which refer to highly expressed genes.

## Density

```
plot(density(data[,1]),col=colors[1],main="Density Distribution")
for(i in 2:ncol(data))
{
  lines(density(data[,i]),col=colors[i])
}
legend("topright",col=colors,legend=samples,pch="-")
```

## Density Distribution

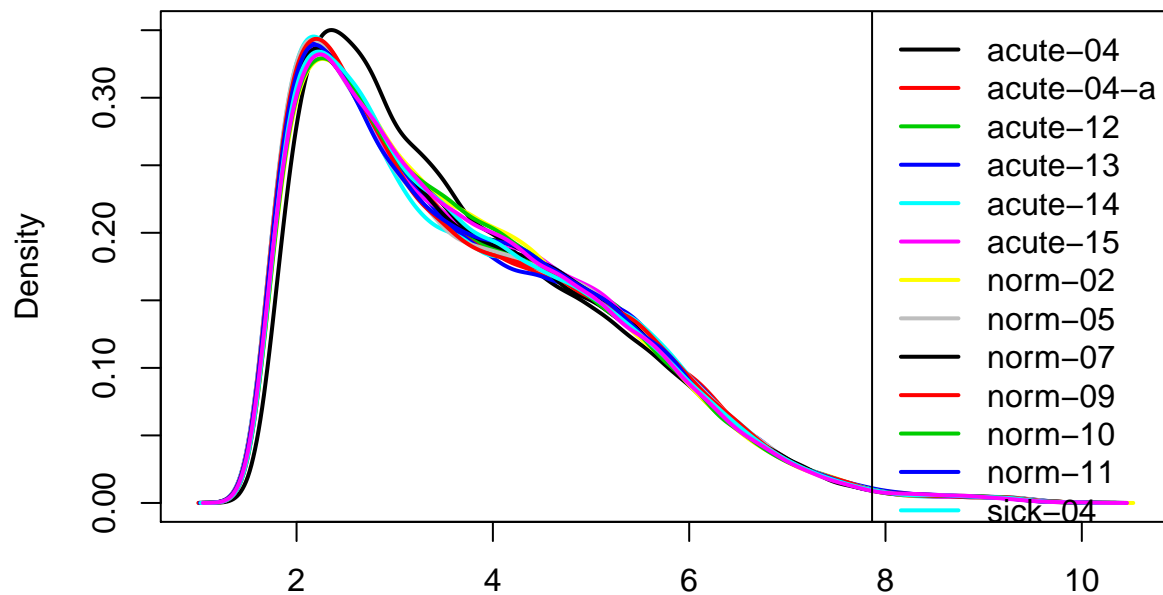


N = 54675 Bandwidth = 0.1523

limma::plotDensities

```
library(limma)
plotDensities(data, main="limma::plotDensities", legend="topright")
```

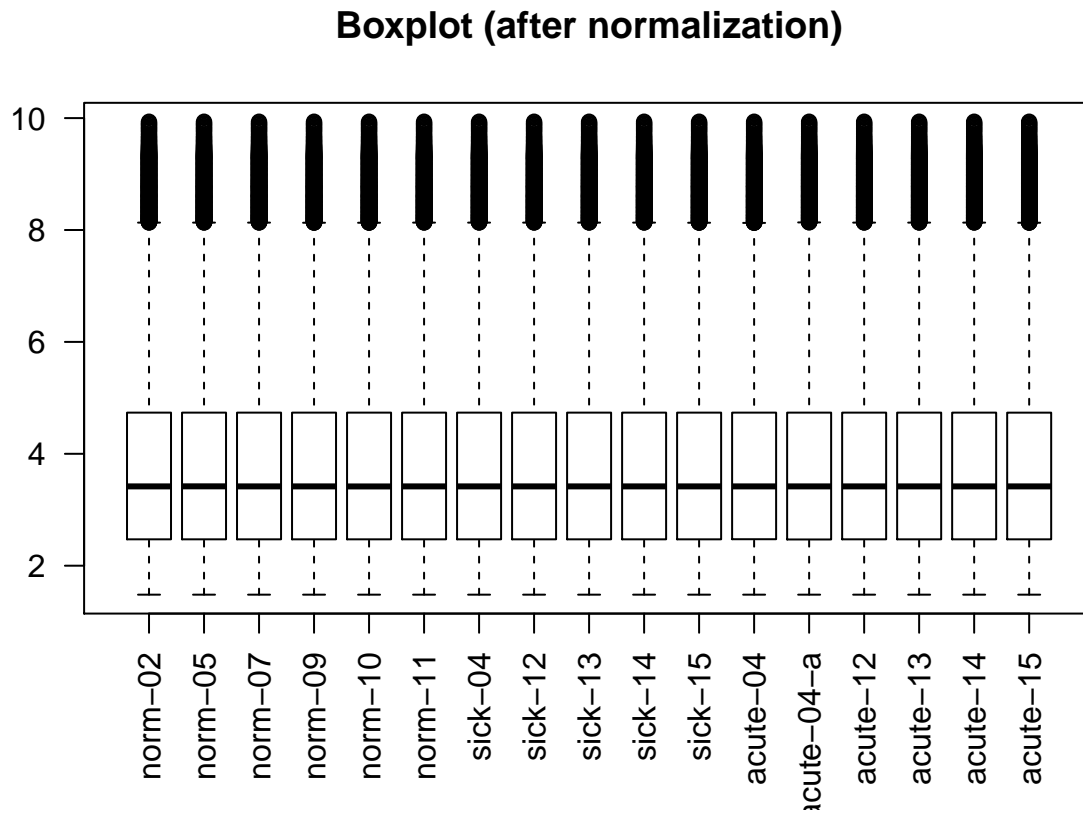
## limma::plotDensities



Intensity

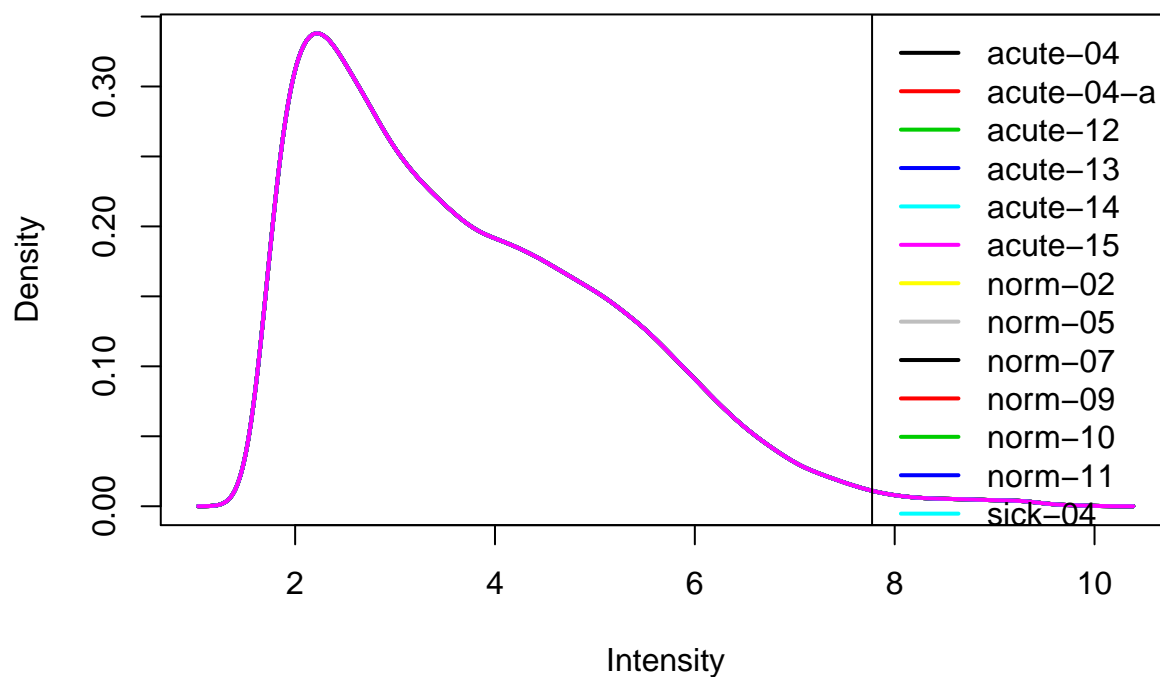
## Normalization

```
norm_data<-normalizeQuantiles(data,ties=TRUE)  
boxplot(norm_data,las=2,main="Boxplot (after normalization)")
```



```
plotDensities(norm_data,main="limma::plotDensities (after normalization)",legend="topright")
```

## limma::plotDensities (after normalization)

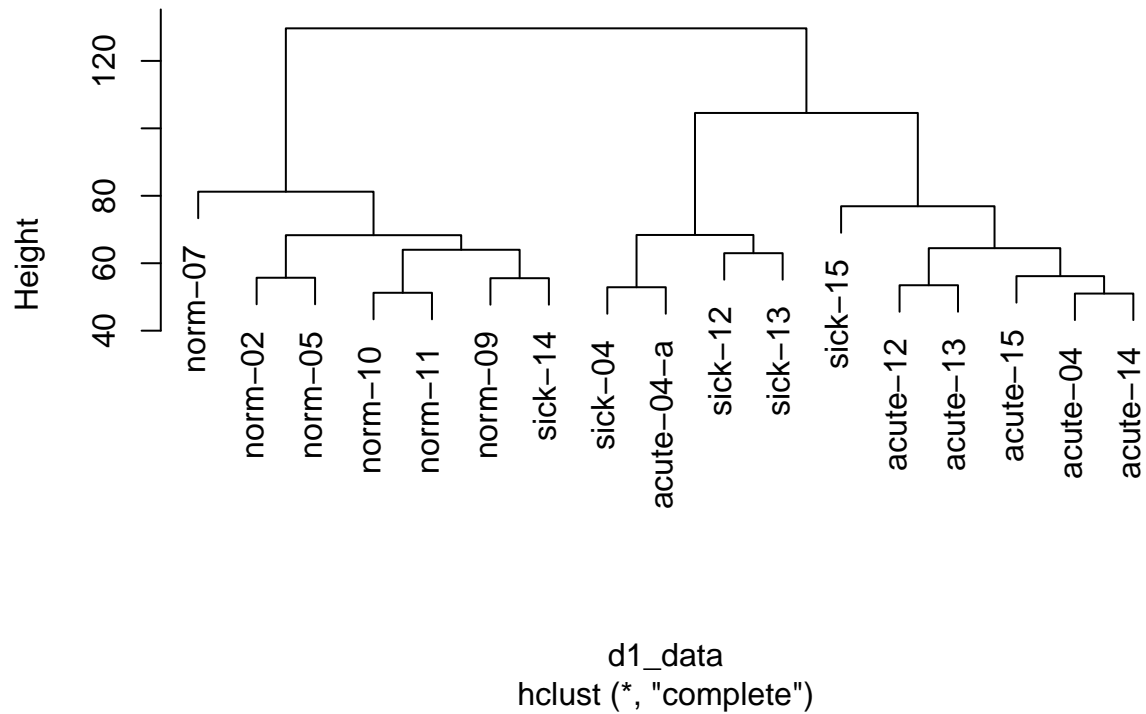


### Clustering

For patients:

```
d1_data<-dist(t(norm_data))  
clusters_p<-hclust(d1_data)  
plot(clusters_p,main="Clustering (for patients)")
```

## Clustering (for patients)



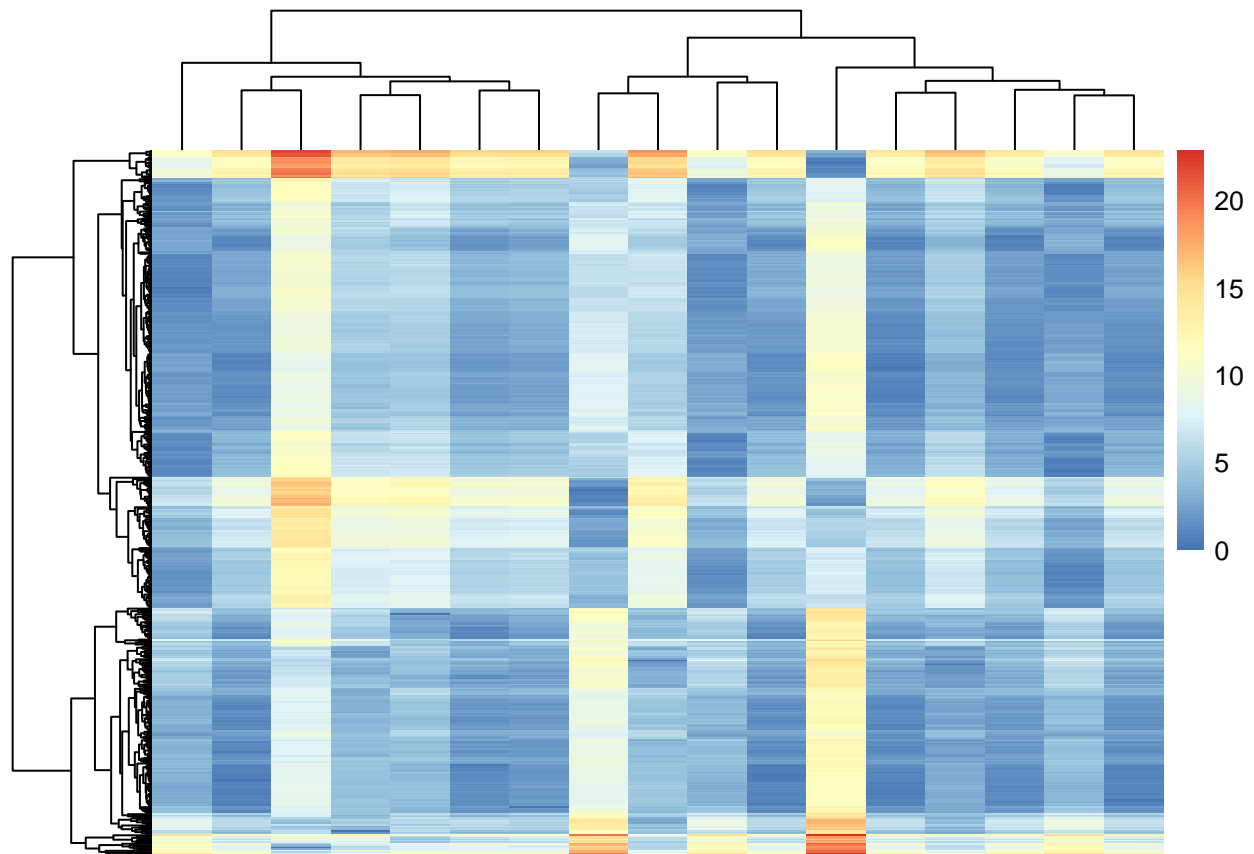
### For genes:

The whole dataset is too large to perform clustering among all genes. Instead, we select those genes which are outliers (log-value larger than 6 according to boxplot above) in at least one sample.

```
norm_outliers<-normalizeQuantiles(outliers,ties=TRUE)
d2_data <-dist(norm_outliers)
clusters_g<-hclust(d2_data)
plot(clusters_g,main="Clustering (for outliers)")
```

Heatmap visualization showing the relationship between `d2_data` (x-axis) and `hclust (*, "complete")` (y-axis). The color scale represents the height of the hierarchical clustering, ranging from 0 (white) to 1 (black). The diagonal is black, indicating a height of 1. The off-diagonal elements show the height of the clusters formed by the data points. The label "Height" is on the left side of the heatmap, and the number "35" is in the top right corner.

```
s_outliers=outliers[sample(nrow(outliers),500),]  
norm_s_outliers<-normalizeQuantiles(s_outliers,ties=TRUE)  
ds_data <-dist(norm_s_outliers)  
library(pheatmap)  
clusters_g2<-hclust(ds_data)  
pheatmap(ds_data,cluster_rows =clusters_g2,cluster_cols=clusters_p )
```

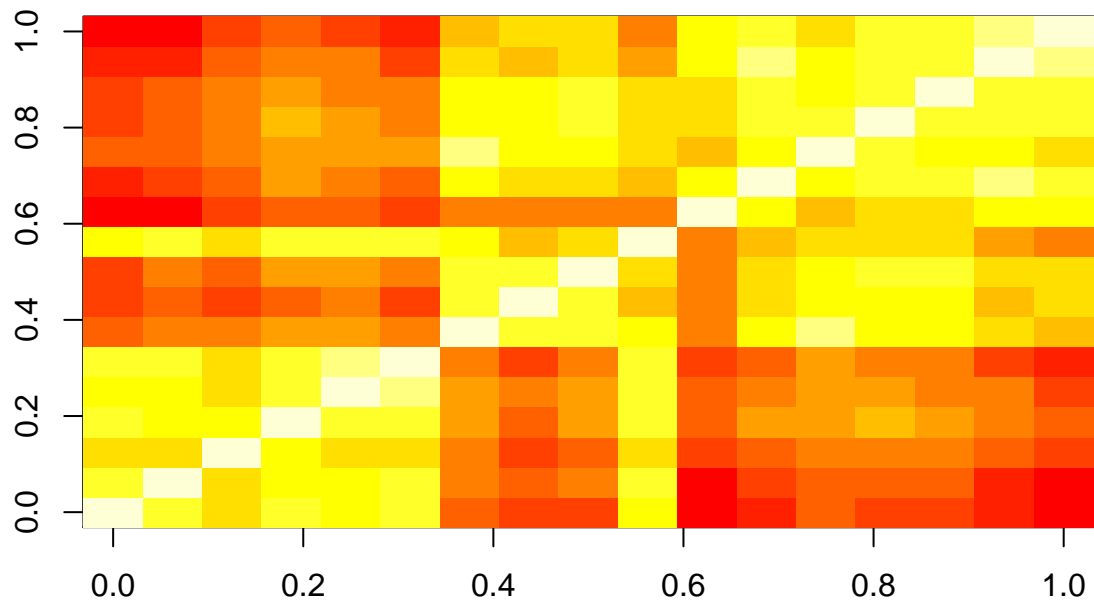


Correlation matrix

```
corr <- cor(norm_data)
image(corr,main="Correlation plot")
```



## Correlation plot

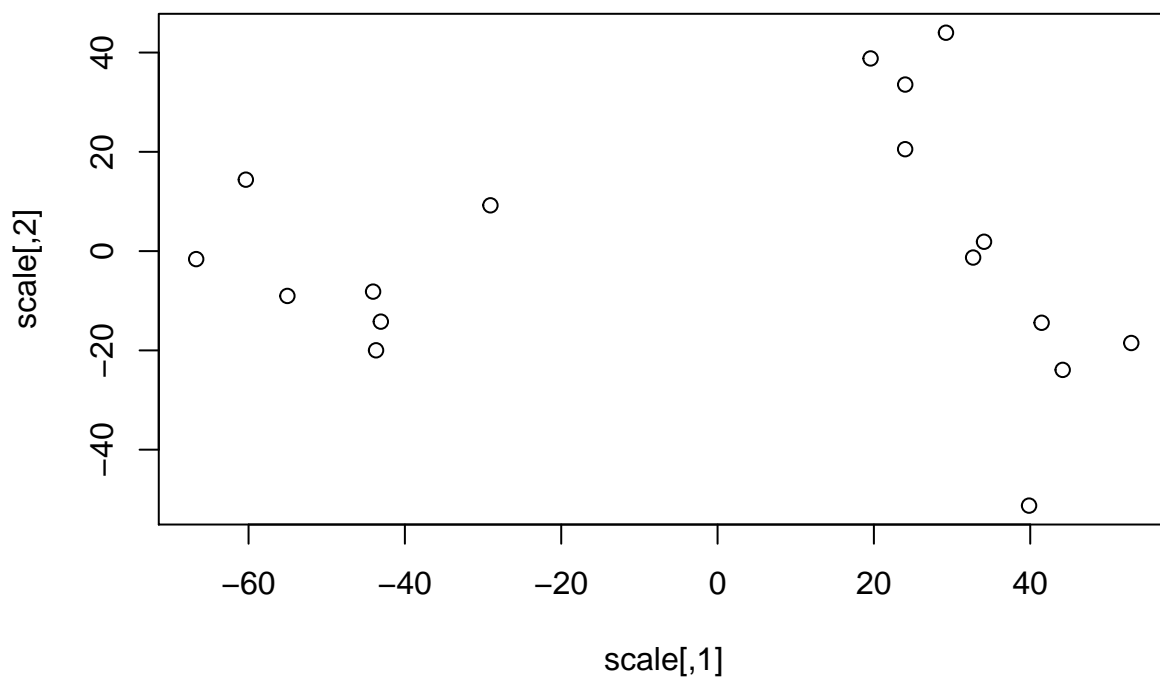


## Reduced dimensionality representation

### Classical Multidimensional Scaling

```
scale<-cmdscale(d1_data,k=2)
plot(scale,main="Reduced dimensionality representation(cmdscale)")
```

## Reduced dimensionality representation(cmdscale)



##### Principal Components Analysis

```
pca<-prcomp(norm_data)
plot(pca,main="Reduced dimensionality representation(pca)")
```

