**Intro:**

* Evaluation of normalization methods
* Normalization: important statistical method to adjust for systematic bias in data we cannot control for
* Make sample more comparable while preserving signal
* Adequate normalization is important
* Different methods work for different data sets
* Incorrect normalization can introduce new artificial patterns

**Data:**

* Peptides and Proteins (from MaxQuant)
* Protein = sum of peptides for given protein
* Peptide level about the same for given Protein
* Remove outliers (more than 1.5 IQR outside of 1st/3rd quantile) (**PP**)
* Create new Protein file (~5min)
* Meta data

**Filter:**

* Focus on ER/PR+, HER2 and control
* With and without Hydroxyurea treatment
* 3 replicates
* 2 batches
* PCA Protein: cell lines + treatment cluster (treatment/batch below)
* PCA Peptide: cluster by treatment/batch below; within cluster: cell lines

**Normalization:**

* Different methods work for different data sets
* Streamline evaluation
* Methods: log2, median, mean, VSN (\*), Quantile, Cyclic Loess (\*), Robust linear regression (\*), global intensity
* Different evaluation metrics: Total intensity, PCA, pooled coefficient of variance (PCV), pooled median absolute deviation (PMAD), pooled estimate of variance (PEV)
* PCV/PMAD/PEV: small good
* Cor: within group; high good
* Heatmap: Cluster by: Treatment groups, cell lines, batch/treatment
* Log2 ratio: distribution of log2 ratios of all treatment group combinations; centered around 0; VSN tighter but shifted
* NA: for imputation method selection (missing at random (MAR) or missing not at random (MNAR)); primary cluster by batch; secondary cluster by treatment groups

**DAtest:**

* Select normalization and imputation method
* LIMMA does not require imputation
* DAtest: comparison of different differential abundance/expression methods
* Automatically selects appropriate tests [**RUN**]
* Parameters and excluded tests
* Ranking of tests
* Log LIMMA best

**DAtest power:**

* Power evaluation over a range of effect sizes for a given test