Statistical Analysis of Microarray Data

Visualization

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FORMER ADDRESS (1999-2011)
Université Libre de Bruxelles, Belgique
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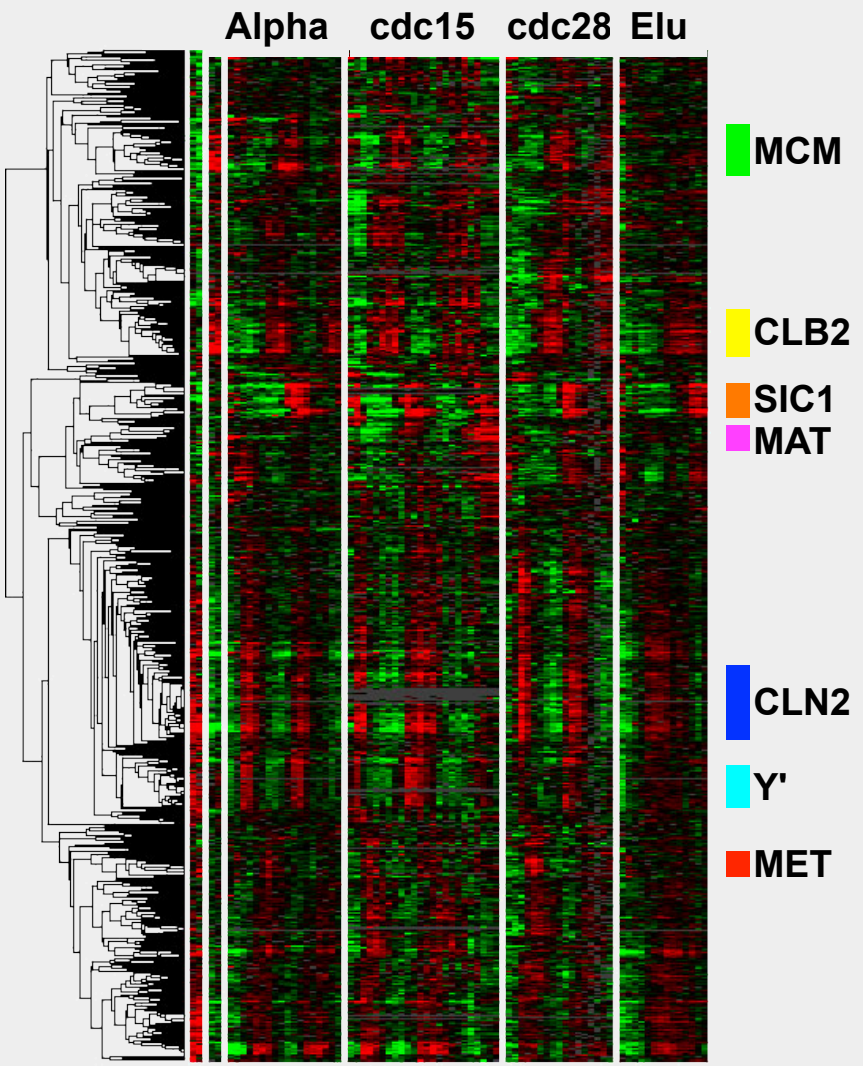
http://www.bigre.ulb.ac.be/

Heat maps



Spellman et al. (1998).
Mol Biol Cell **9**(12), 3273-97.

- Eisen (1998) introduced a visualization tool which allows to display the expression profiles of many genes.
  - Each row represents one gene, each column one chip.
  - Gene profiles can be aligned along the dendrogram resulting from hierarchical clustering.
- This visualization mode combines clustering and expression profiles.
- Problem of isomorphism:
  - The two outgoing branches from each intermediate node can be swapped arbitrarily.
  - The distance between two genes is represented on the horizontal axis (depth of the first parent node)
  - The vertical distance between two genes does not reflect the calculated distance. Some genes are direct neighbours on the vertical axis whereas they are very distant.

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Reduction in data dimension

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Why to reduce dimensionality ?

- A series of microarrays can be represented as a N x p matrix, where
  - each one of the p columns contains information about an experiment (different conditions, treatments, tissues)
  - each one of the N rows contains information about a spot (gene)
- Object dimensions
  - Each gene can be considered as a p-dimensional object (one dimension per experiment).
  - Each experiment can be considered as a N-dimensional object (one dimension per gene).
- Visualization
  - Visualization devices are restricted to 2 (printer) or at best 3 (space explorer) dimensions.
  - One would thus like to display objects in 2D or 3D, whilst retaining the maximum of information.
  - After reduction of dimensions, some clusters may already appear in the data set.
- Analysis
  - Some analysis methods loose their accuracy when there are too many vriables (over-fitting).
  - Reducing the data to a subset of dimensions will allow a trade-of between the loss of information and the gain in accuracy. In this case, the appropriate number of dimensions may be higher than 3, its choice depends on the data itself (e.g. number of objects per training group).

How to reduce dimensionality ?

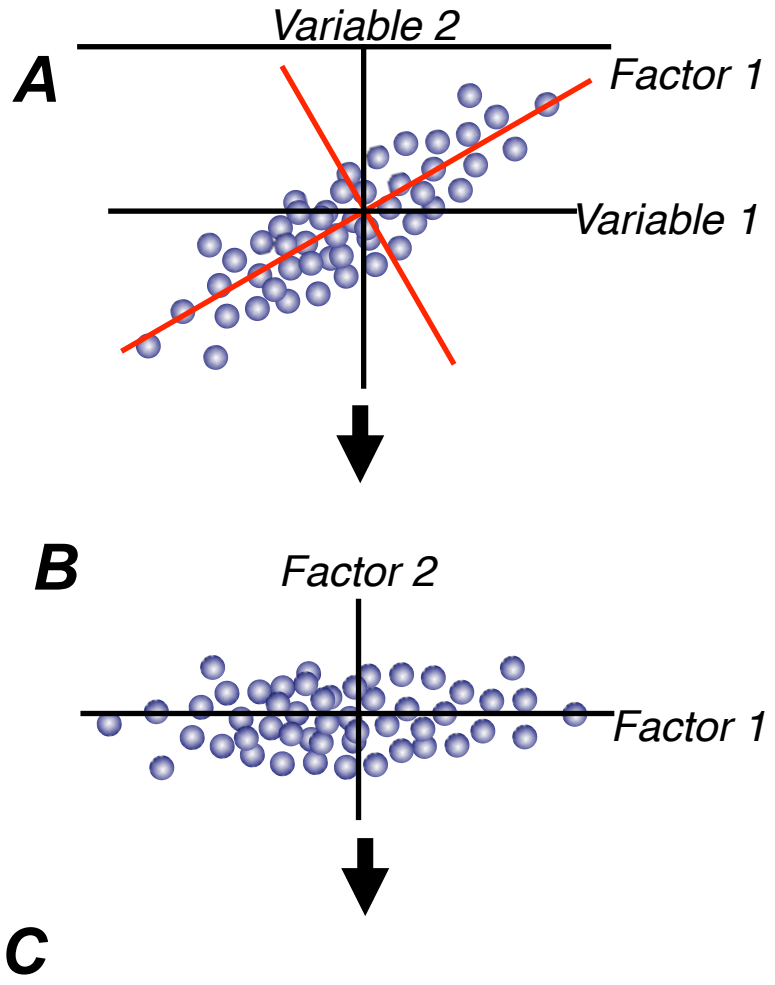
- Several methods are available for reducing the number of dimensions of a data set
  - Principal Component Analysis
  - Singular Value Decomposition
  - Spring embedding

Principal component analysis – Principle of the method

A. Multidimensional data
   - n objects, p variables (in this case p=2)

B. Principal components
   - n objects, p factors
   - Each factor is a linear combination of variables

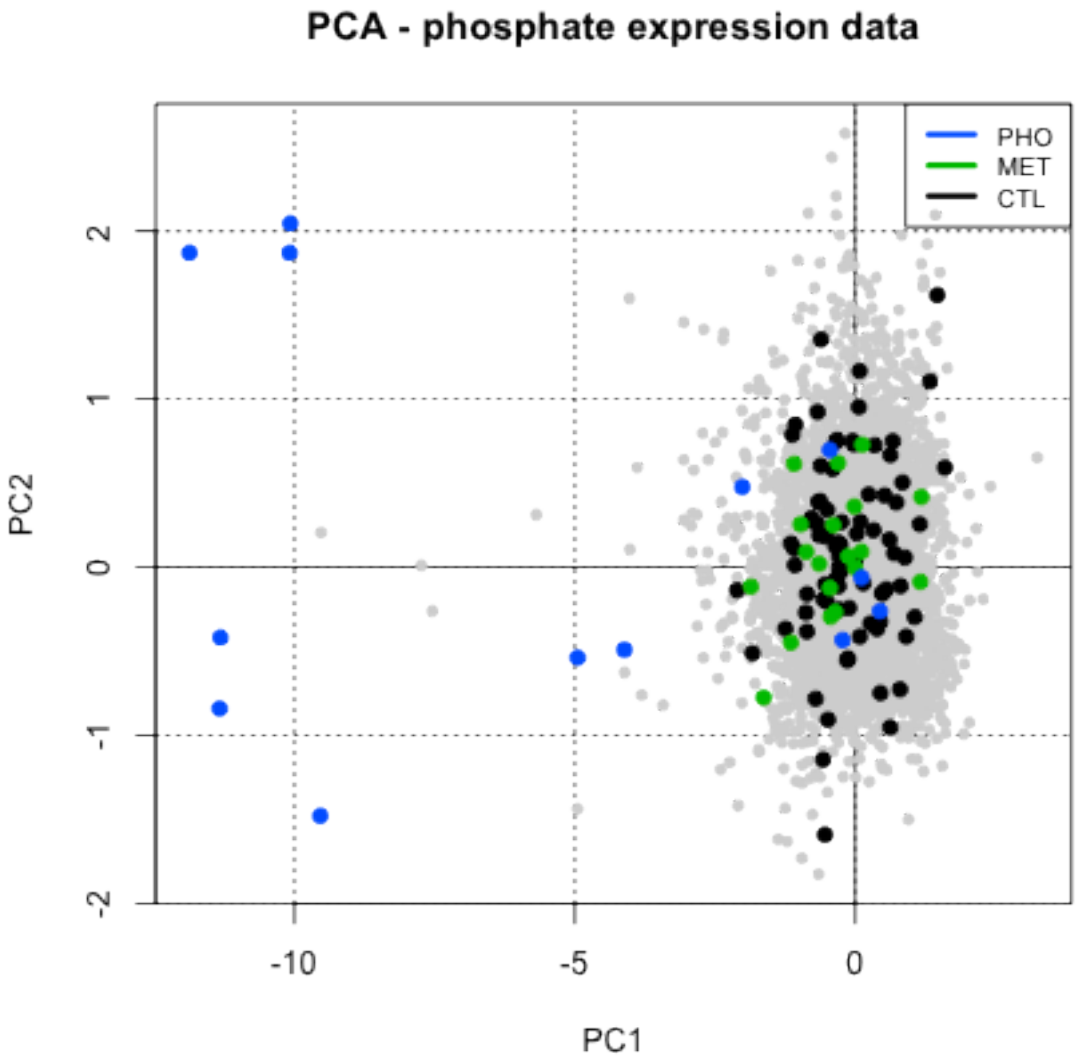
C. Reduction in dimensions
   - Selection of a subset of principal components
   - q factors, with q < p (in this case, q=1)



- Gilbert, D., Schroeder, M. & van Helden, J. (2000). Trends in Biotechnology 18) 487-495.

PCA example: phosphate data (Ogawa 2000)

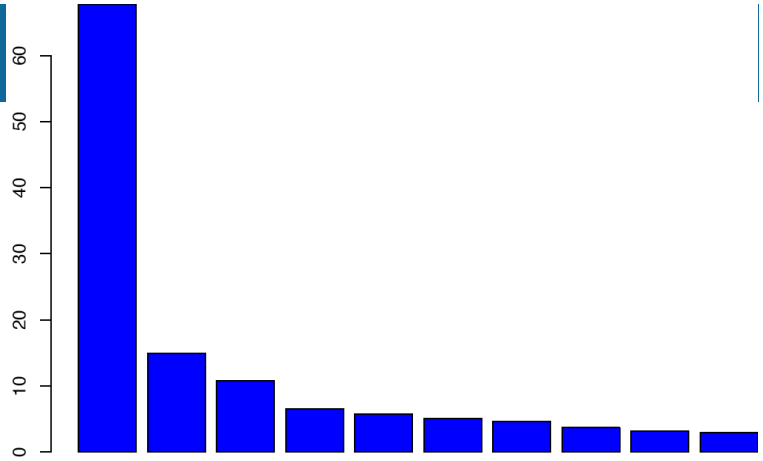
- The original data set contains 5783 objects (genes) characterized by 8 variables (samples).
- The plot shows the first (PC1 and second (PC2) principal components.
- Colors highlight some genes known to be regulated by phosphate (blue), methionine (green) or none of them(black).
- The first component visibly separates several of the phosphate-responding genes from the other genes.

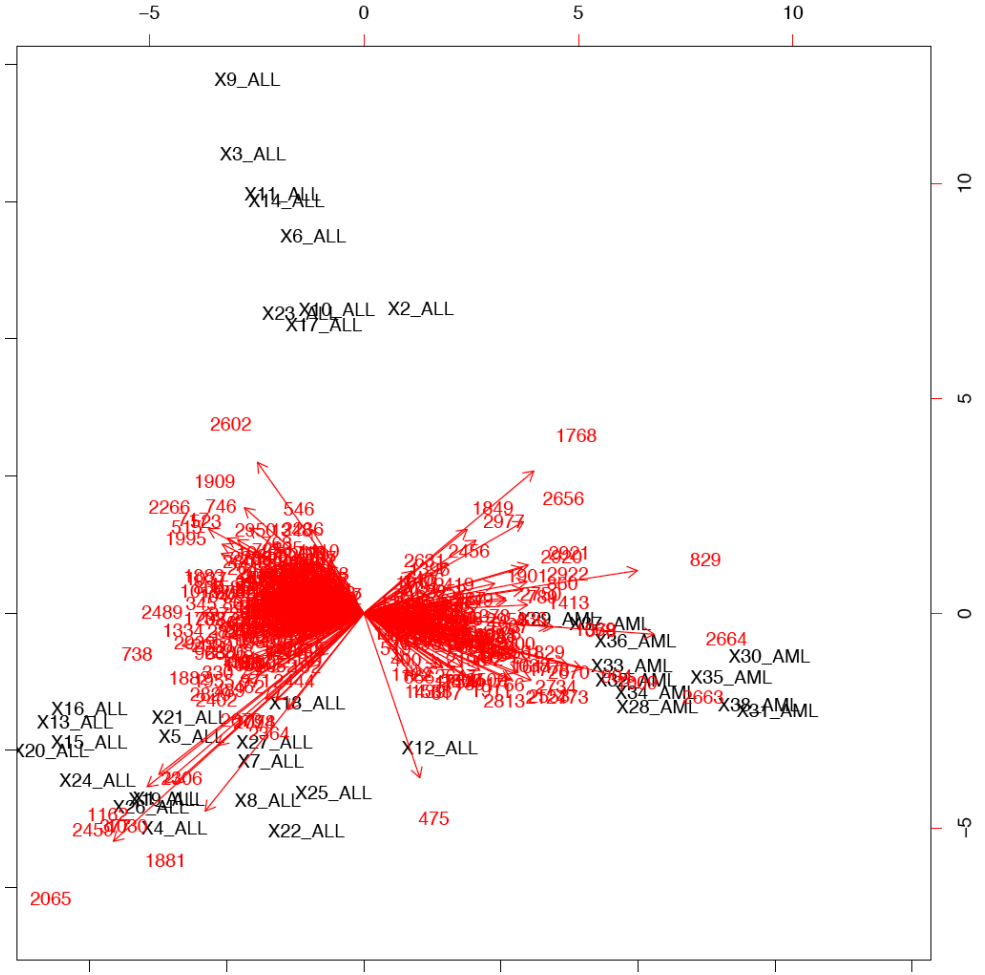


- Data from Ogawa et al. New components of a system for phosphate accumulation and polyphosphate metabolism in Saccharomyces cerevisiae revealed by genomic expression analysis. Mol Biol Cell (2000) vol. 11 (12) pp. 4309-21.
- Script: in statistics for bioniformatics, multivariate_analysis.R

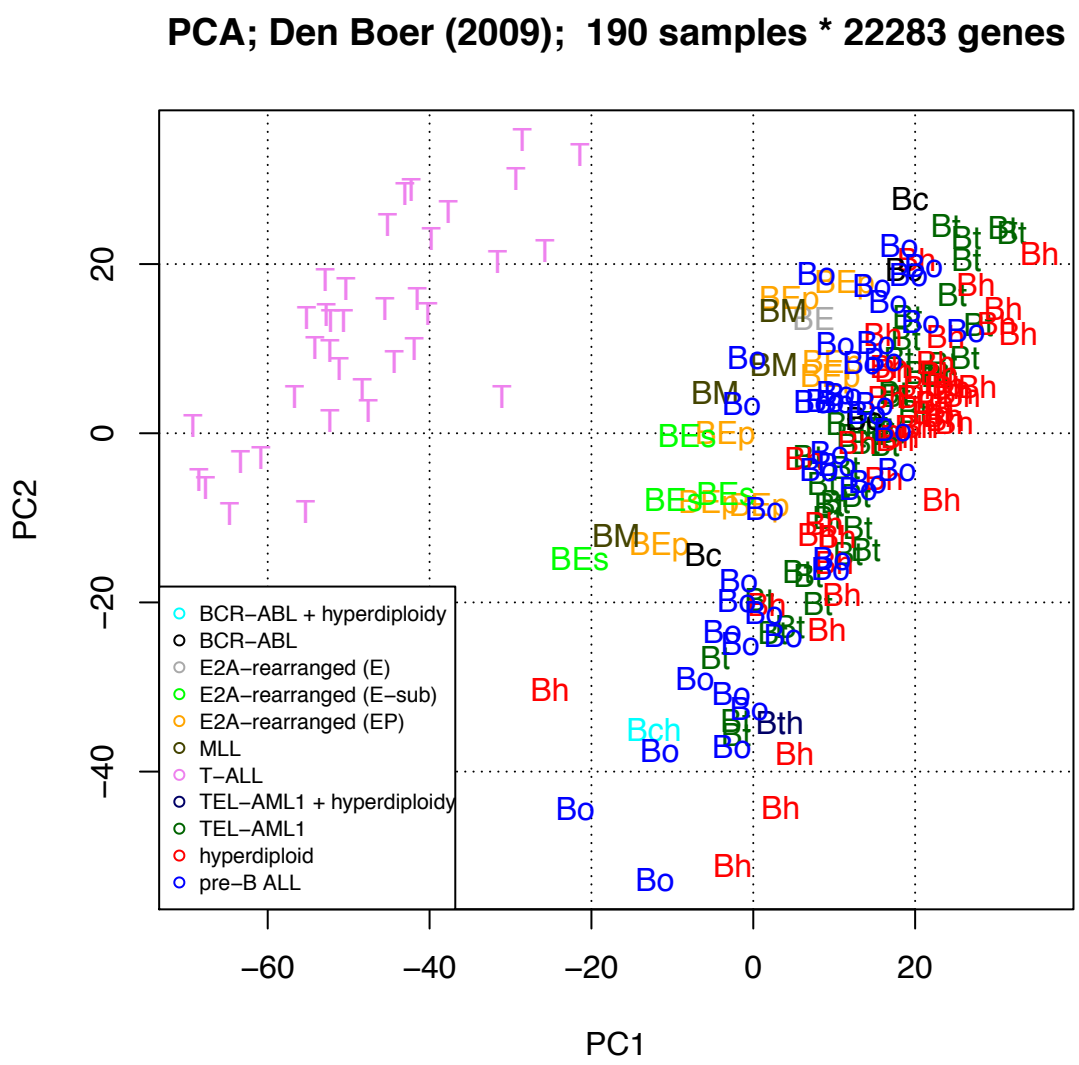
PCA on Golub data set

- PCA applied on a selection of 367 genes showing a significant difference in expression between cancer types (AML vs ALL).
- The first component
  - Explains 70% of the variance !
  - Perfectly separates the two main patient types (AML and ALL)
- The second component reveals two sub-groups among patients of the the ALL type.

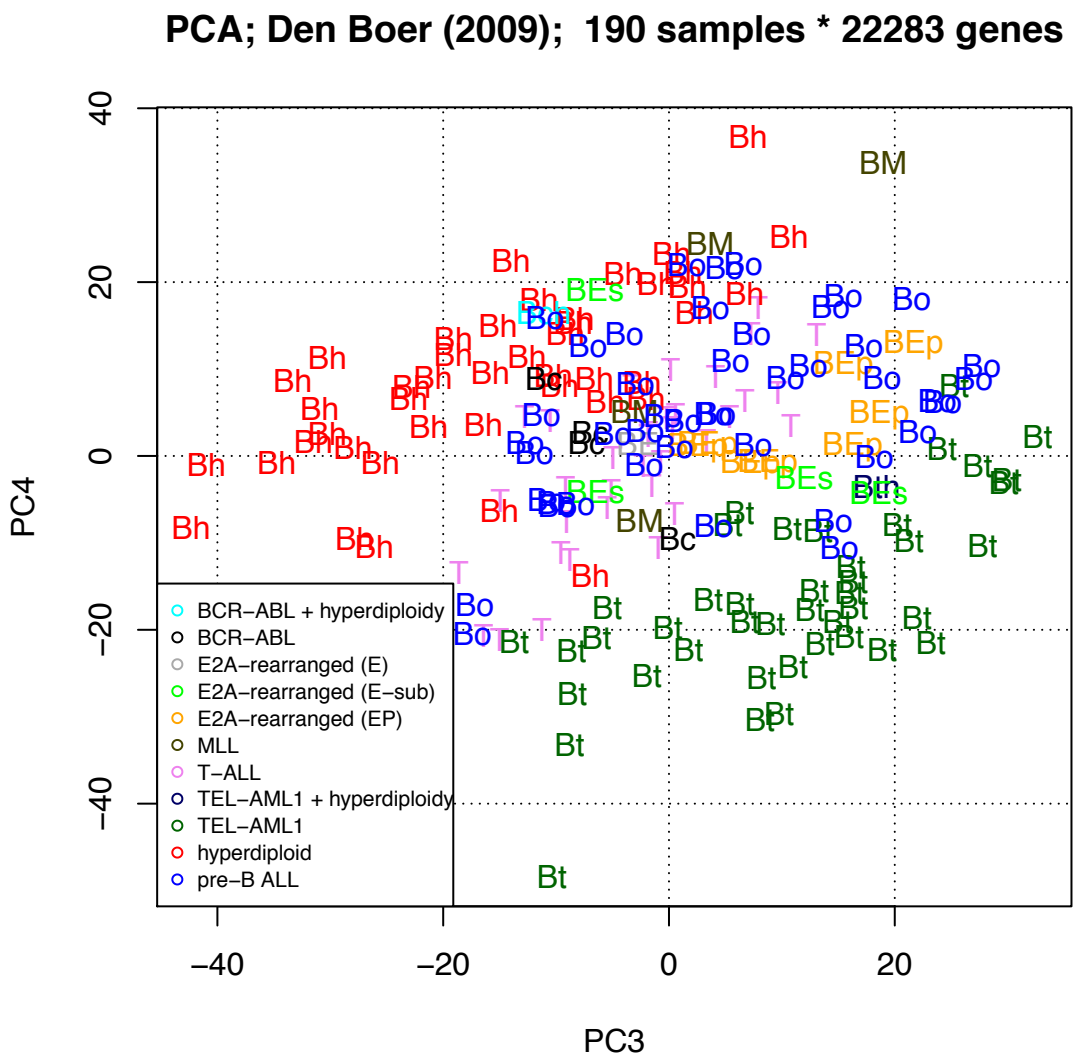




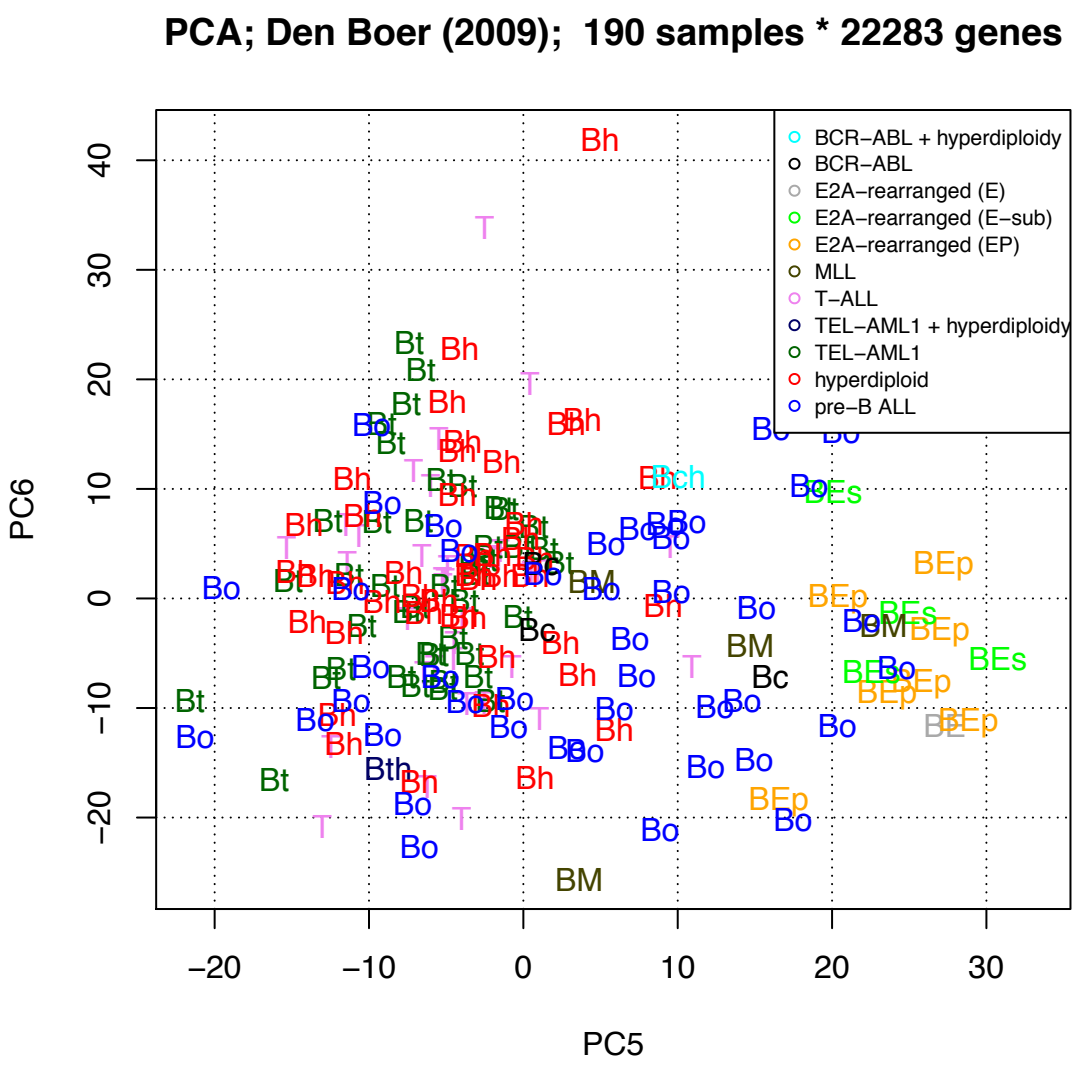
PCA – Den Boer (2009) – PC1 versus PC2



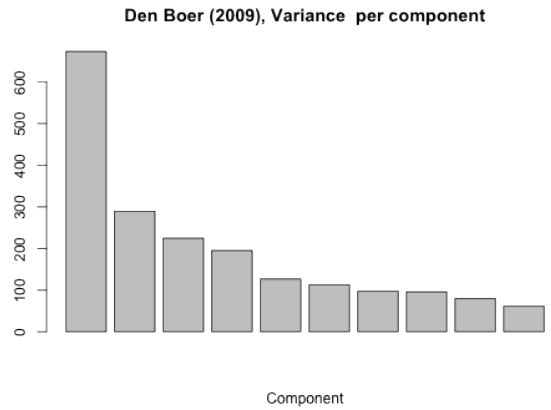
PCA – Den Boer (2009) – PC3 versus PC4

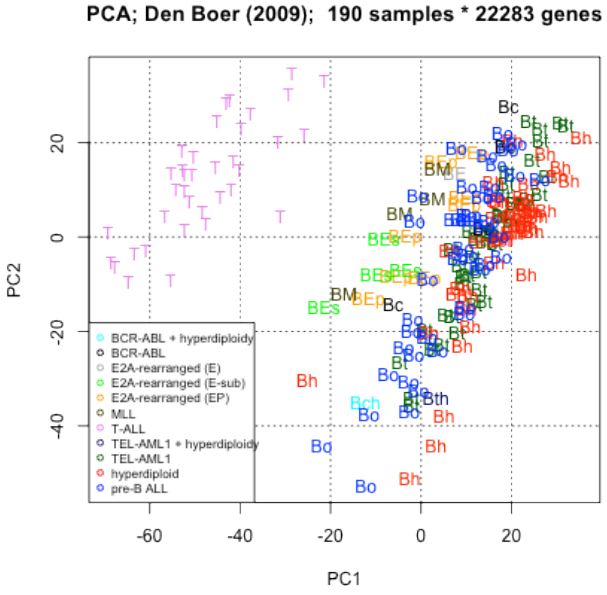


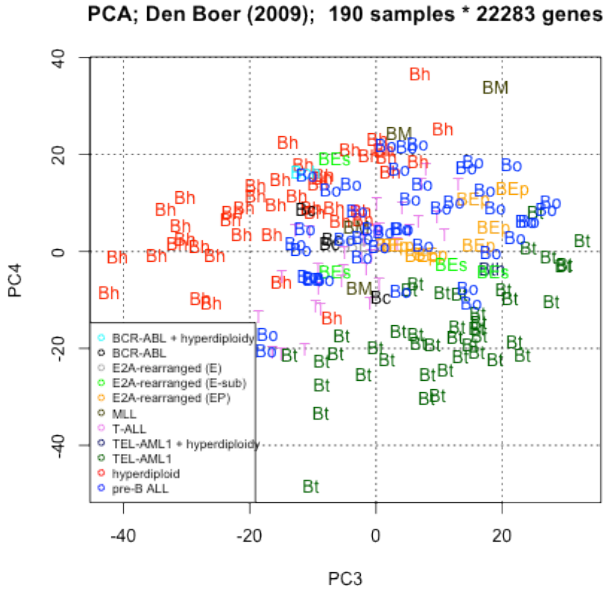
PCA – Den Boer (2009) – PC5 versus PC6

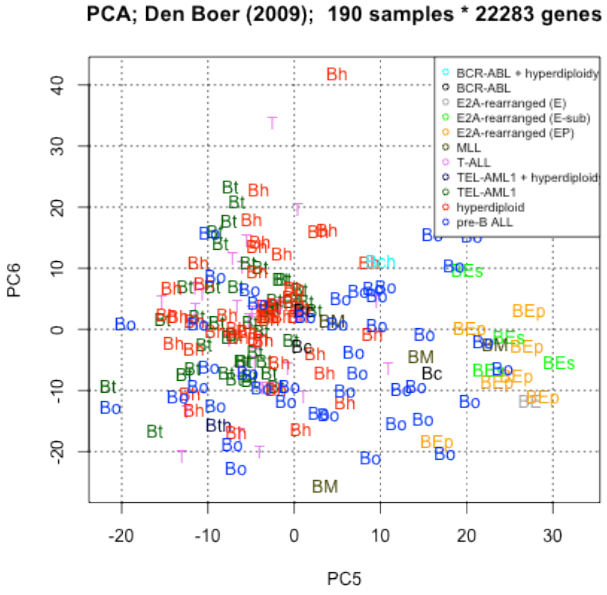


PCA – Den Boer (2009)

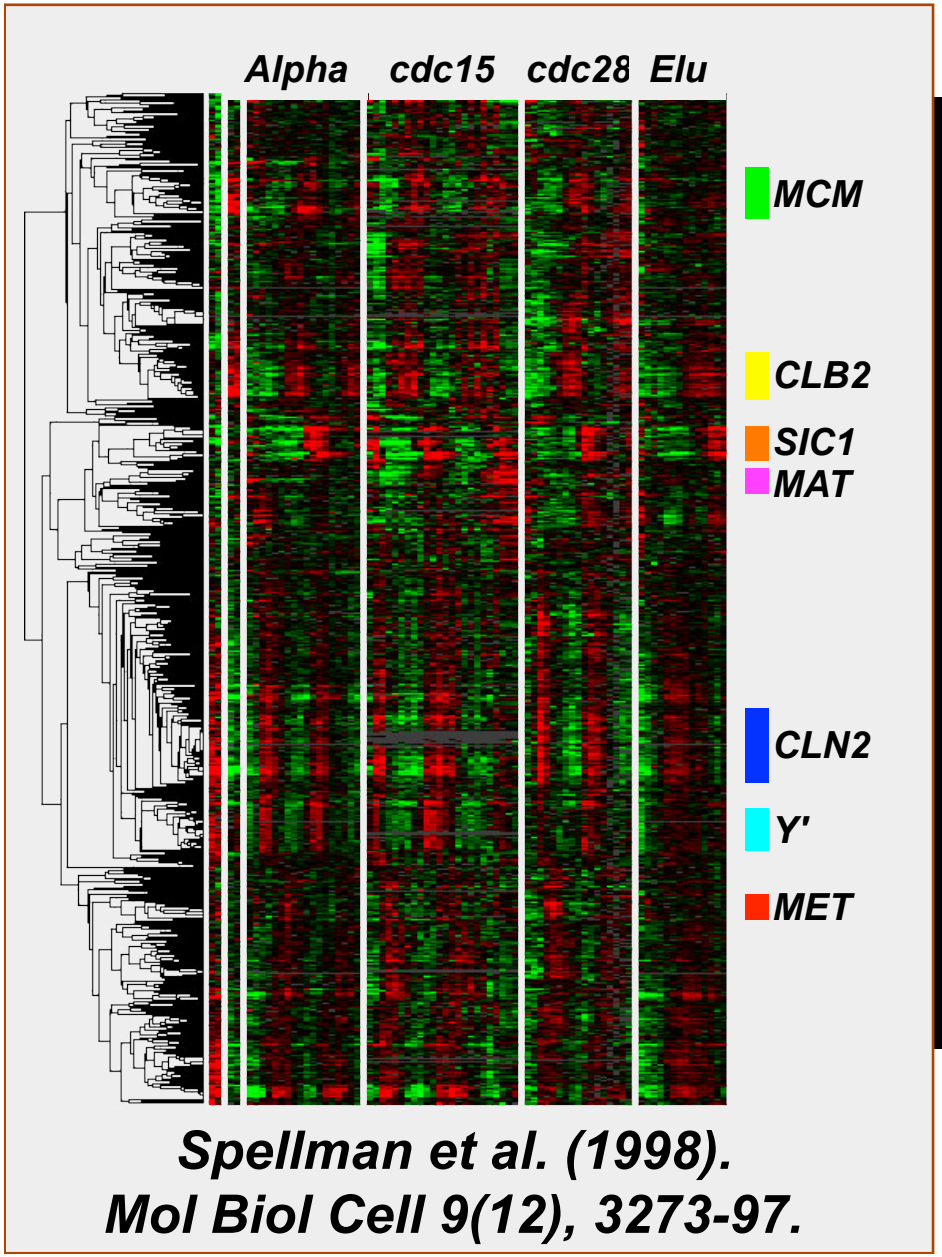








Singular value decomposition



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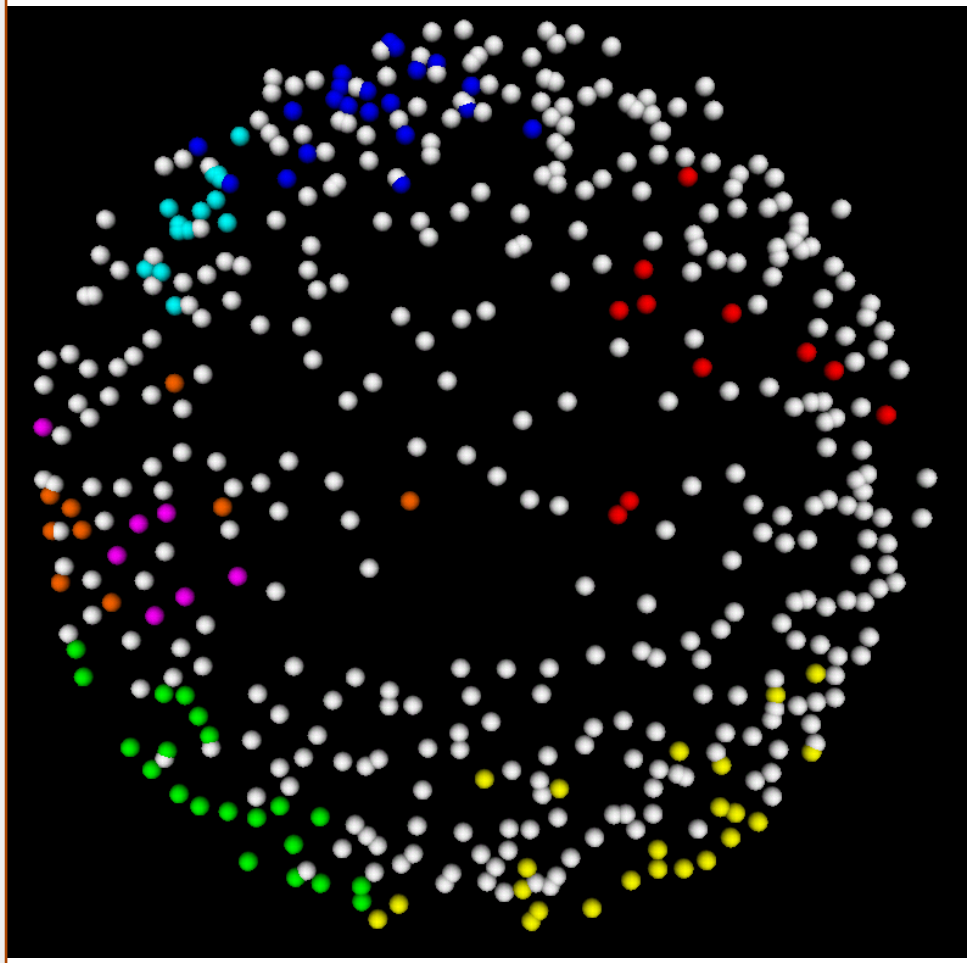
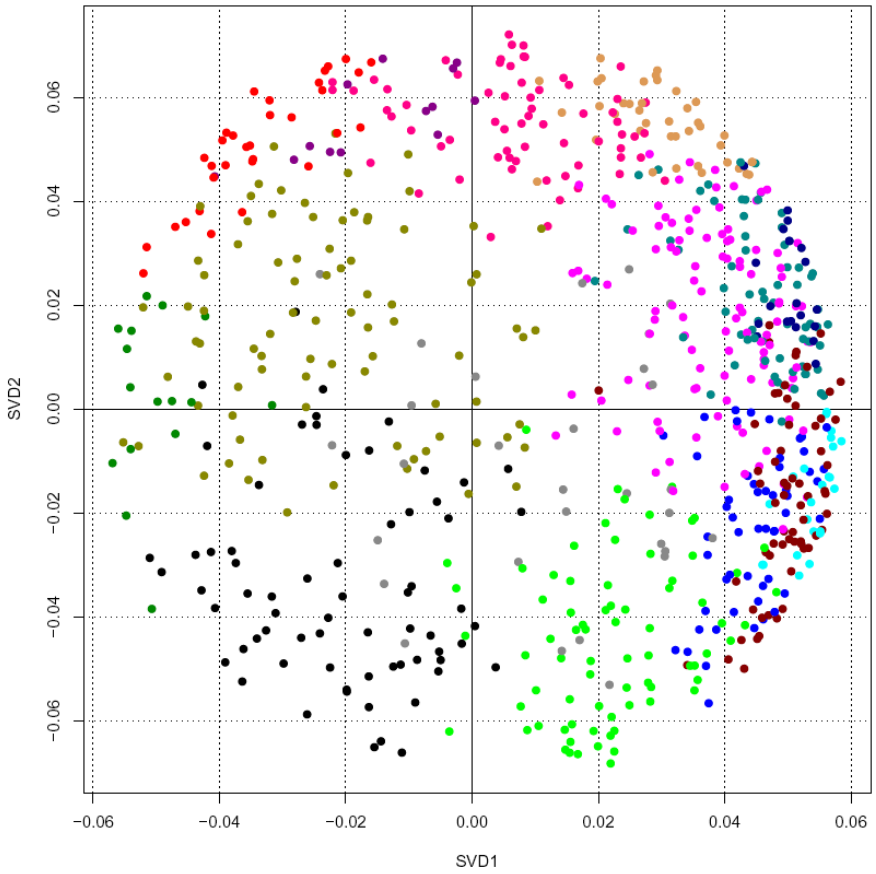


Gilbert et al. (2000).
Trends Biotech. 18(Dec), 487-495.

Singular value decomposition - Cell cycle

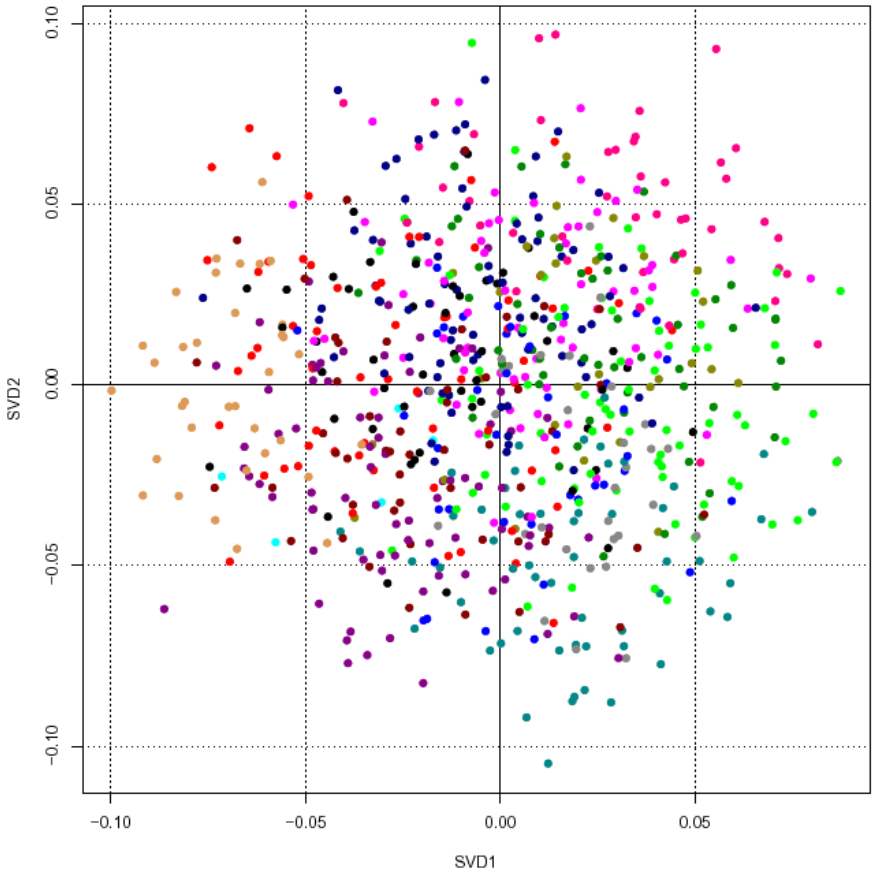
Cell cycle data

Spellman 98 elu



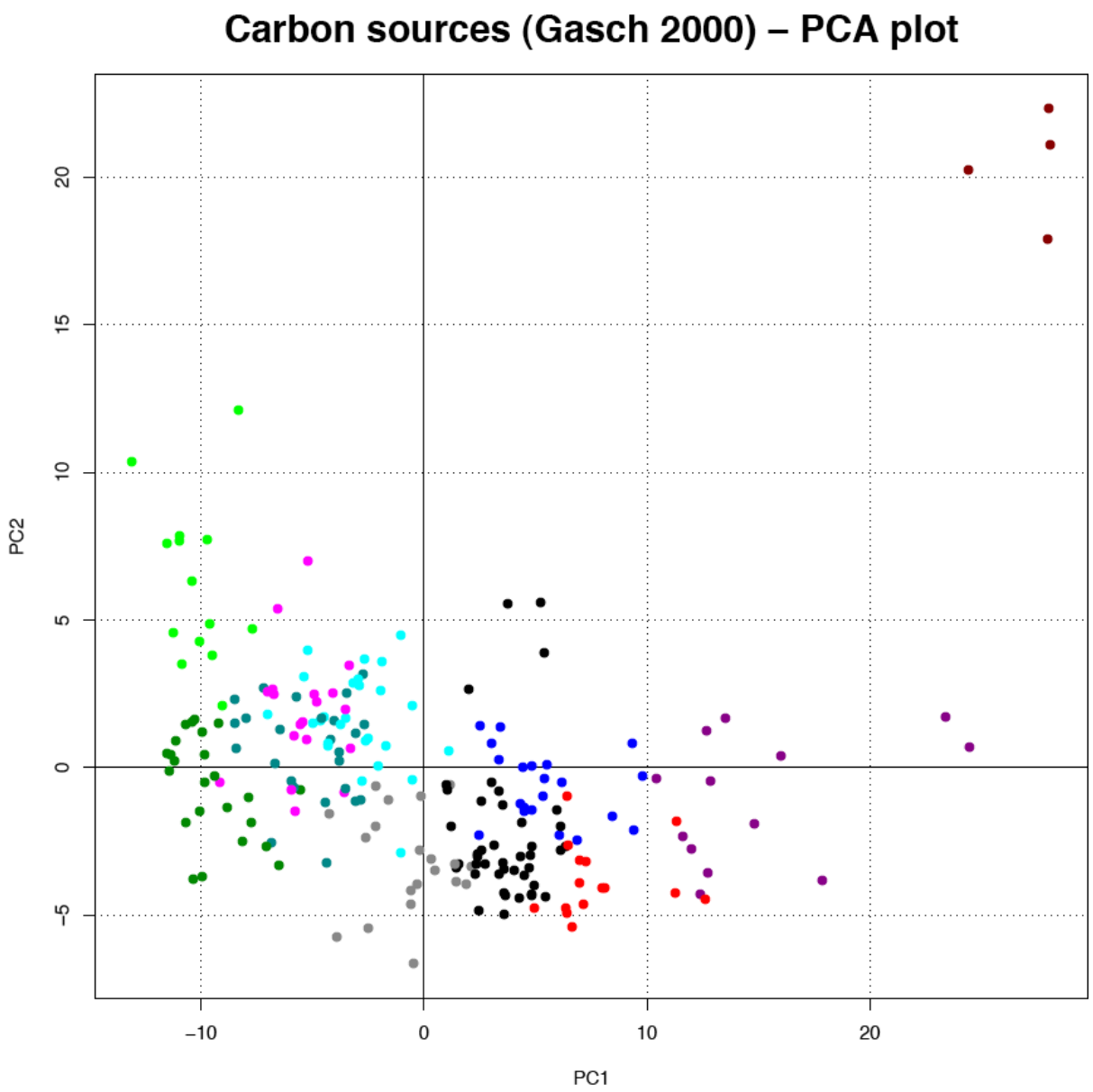
Randomized data

Spellman 98 elu permuted



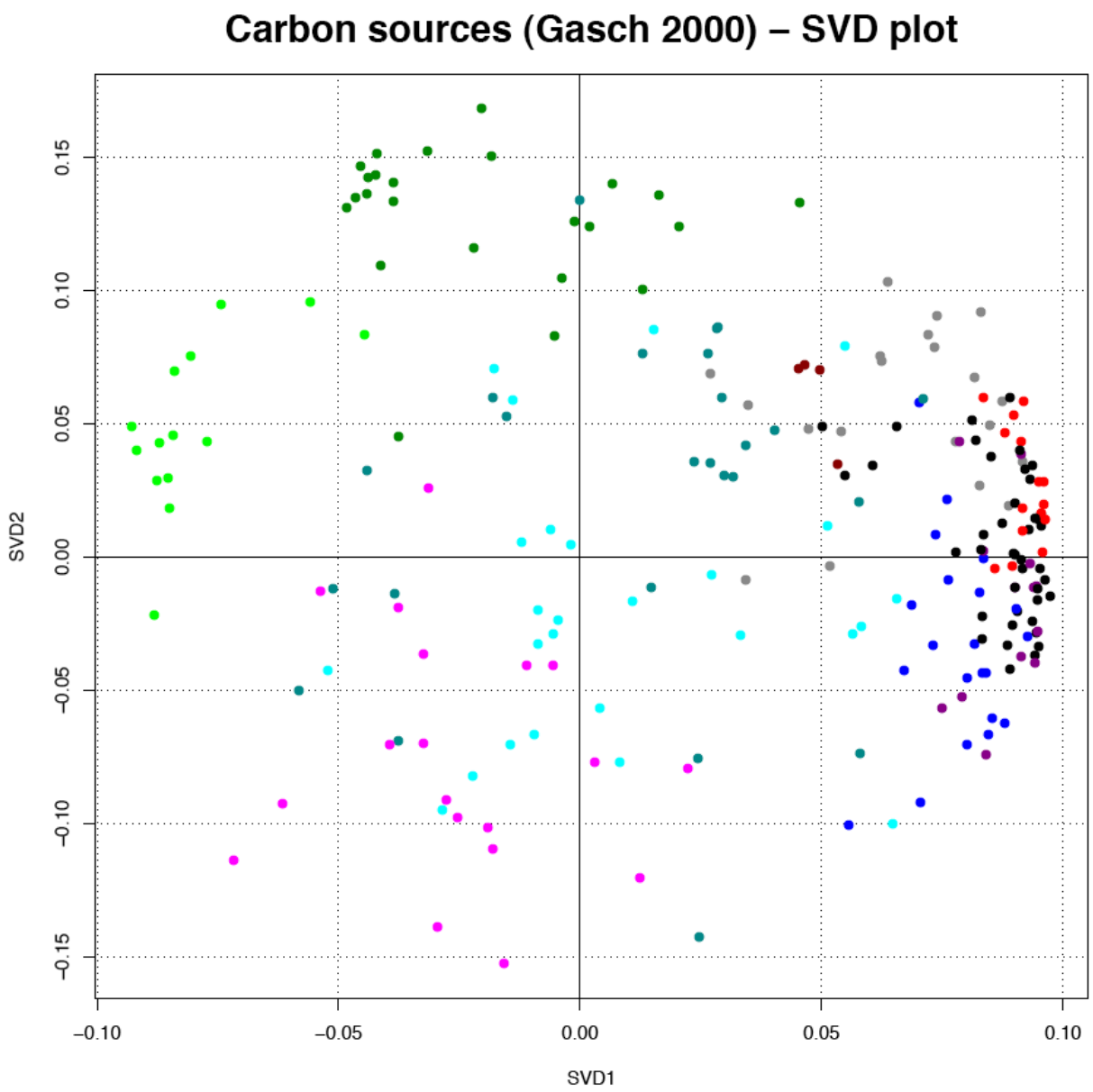
- Calculate a distance matrix between objects
  - in this case Pearson's coefficient of correlation
- Assign 2D-coordinates which reflect at best the distances

Data reduction with principal components



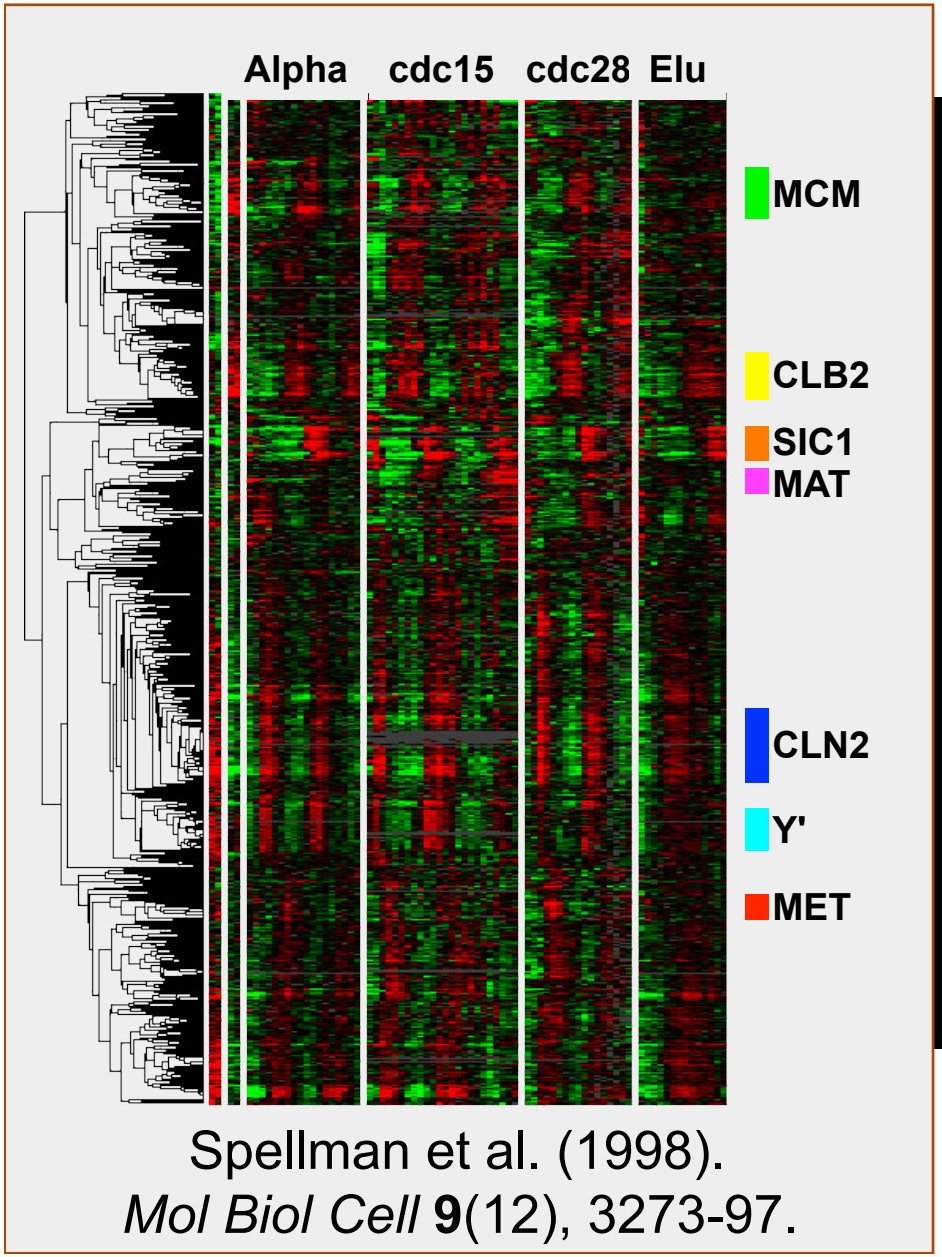
- Data from Gasch (2000). Growth on alternate carbon sources (13 chips).
- Selection of 224 genes which are significantly regulated in at least one chip.
- The plot represents the two first components after PCA transformation.
- Colours represent 15 clusters obtained with K-means clustering.

Singular value decomposition - Carbon sources



- Data from Gasch (2000). Growth on alternate carbon sources (13 chips).
- Subset of 224 genes significantly regulated in at least one chip.
- Singular value decomposition (SVD) on correlation matrix.
- The clusters are better separated than with PCA.
- The proximity between two dots reflects their correlation (within the constraints of the 2D space)

Singular value decomposition



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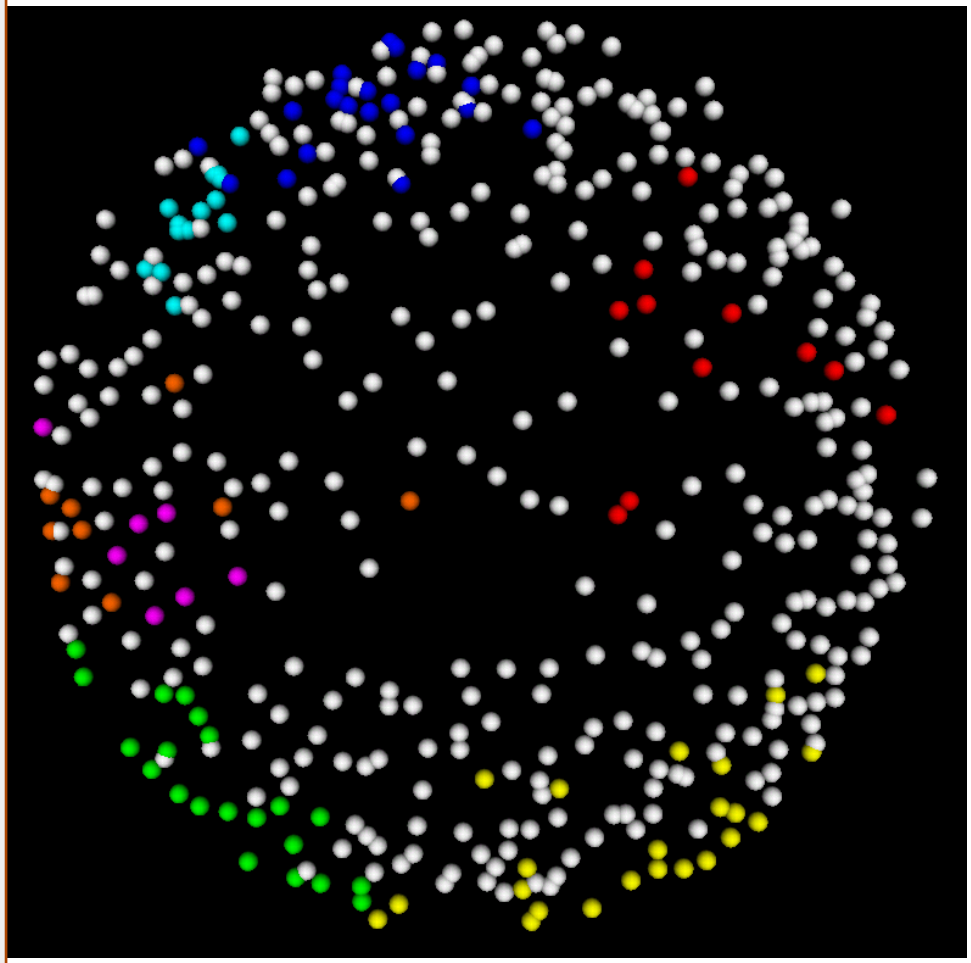

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Singular value decomposition - Cell cycle

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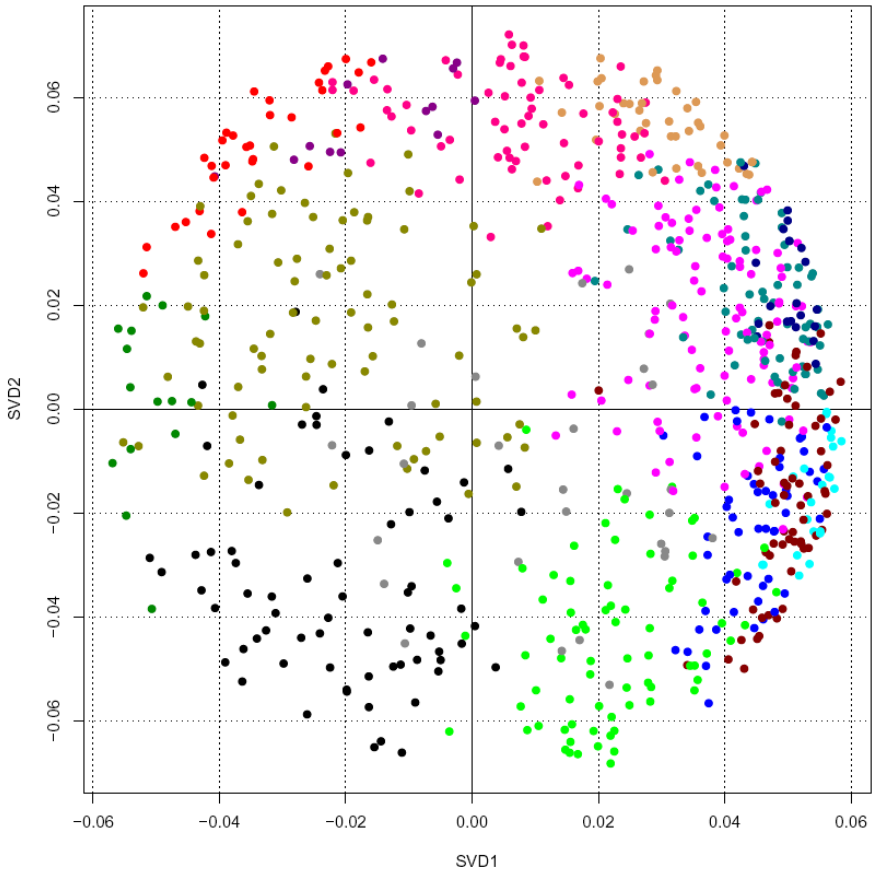
Spellman 98 elu



Randomized data

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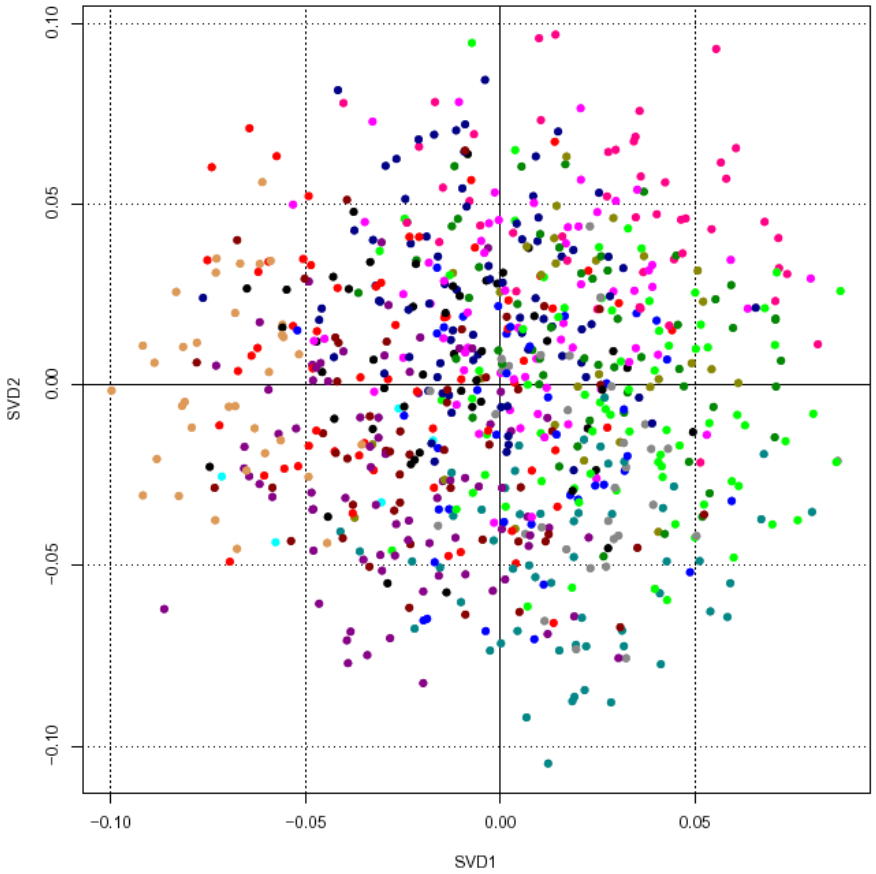

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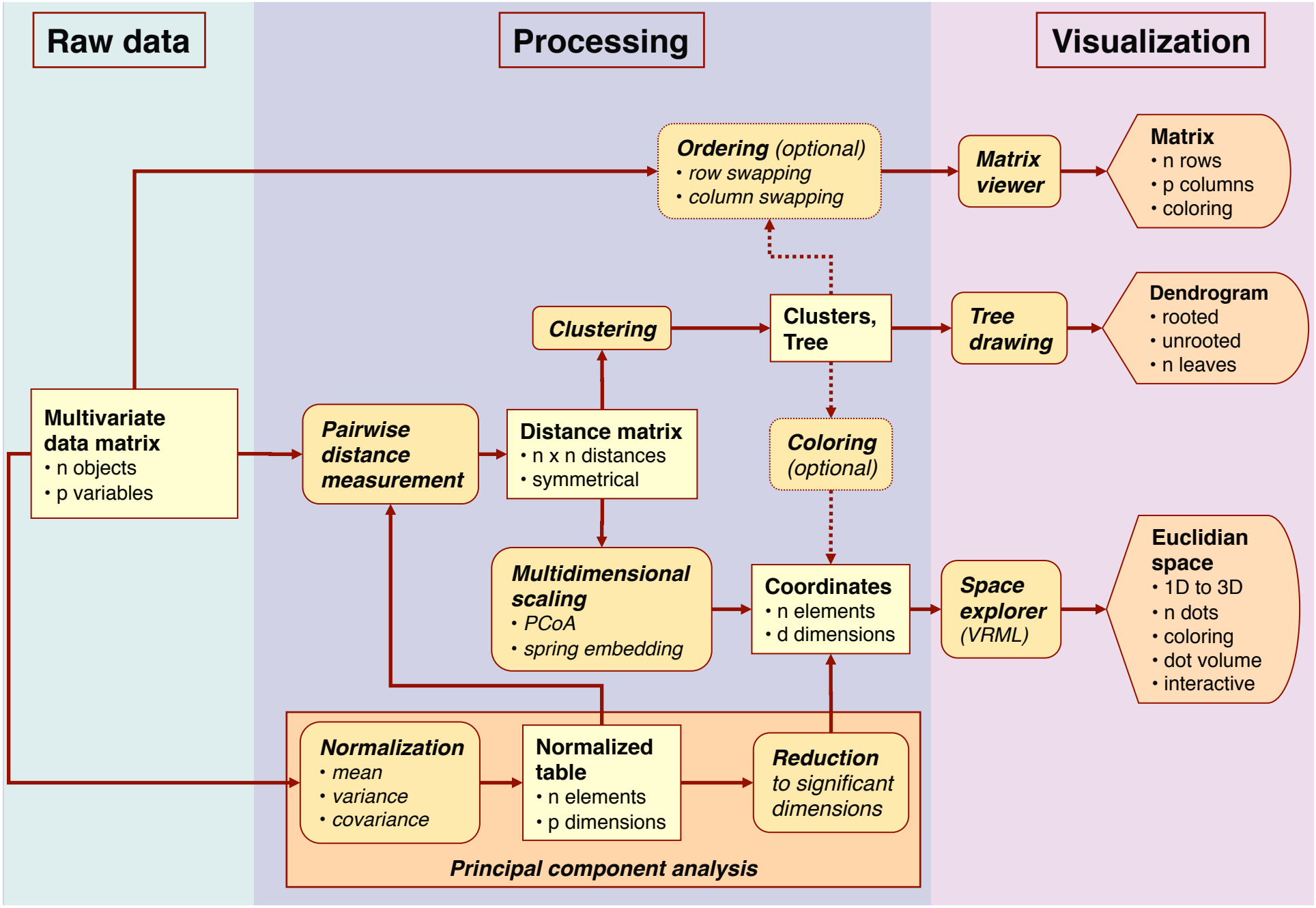

Adapted from Gilbert et al. (2000). *Trends Biotech.* **18**(Dec), 487-495.