Factor Analysis for Multiple Testing : an R package for large-scale significance testing under dependence

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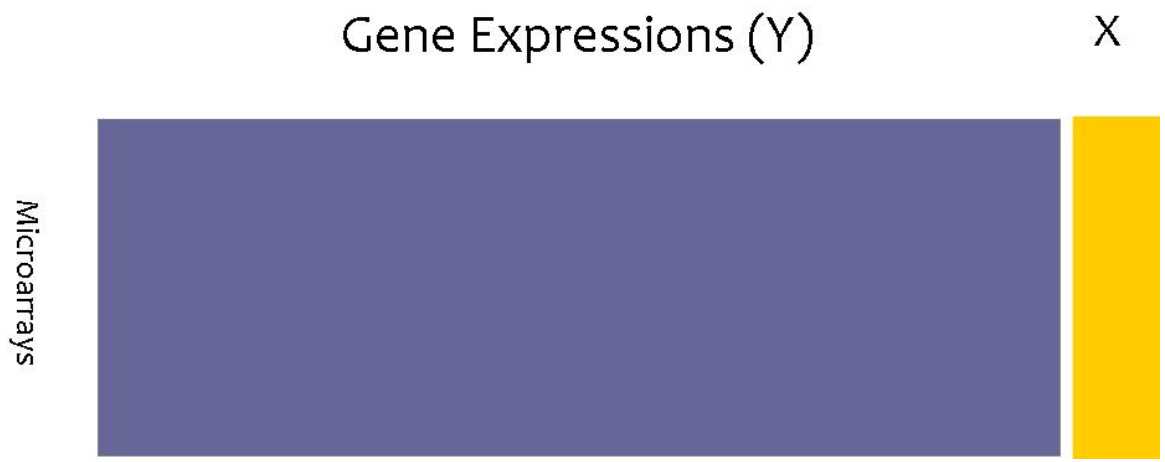
The UseR! Conference, July 2009
Agrocampus Ouest, France

Outline

Impact of dependence in multiple testing

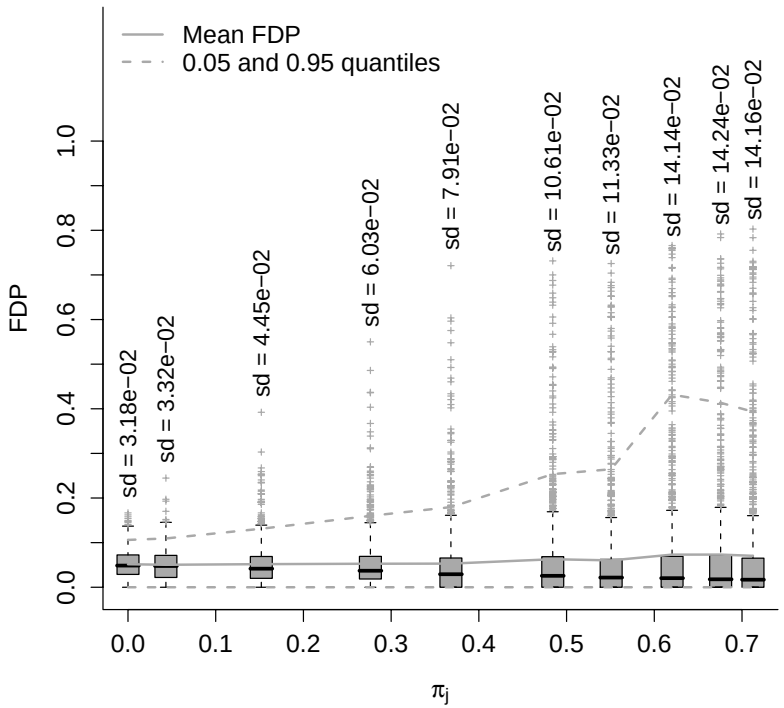
Multiple testing: to point out genes which expressions (Y) significantly depend on the experimental condition (X)
High dimension: a few microarrays and a huge number of gene expressions



A major concern: the biological links among genes and the high dimensional setting generates a large-scale correlation structure, which induces high instability in multiple testing procedures.

Distribution of error rates in multiple tests

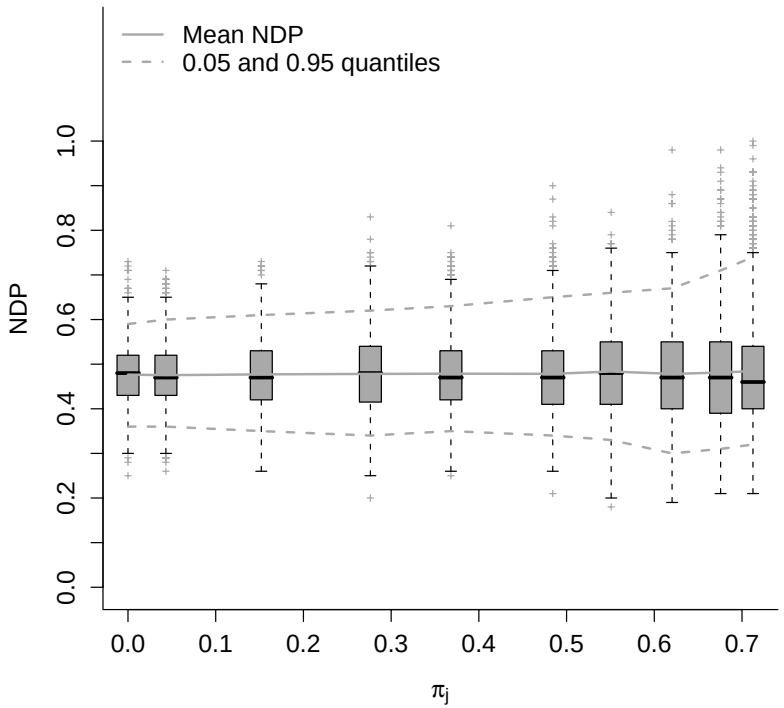
Distribution of False Discovery Proportion (V_t/R_t) on 1.000 simulated datasets/scenario (Friguet *et al.*, 2009, *JASA*)



	Declared H0	*Declared H1*	*Total*
True H0	$\mathbf{U_t}$	$\mathbf{V_t}$	*m0*
True H1	$\mathbf{T_t}$	$\mathbf{S_t}$	*m1*
	$m\text{-}R_t$	R_t	*m*

Distribution of error rates in multiple tests

Distribution of Non-Discovery Proportion ($T_t/m1$) on 1.000 simulated datasets/scenario (Friguet *et al.*, 2009, *JASA*)



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Factor Analysis for Multiple Testing

The common information shared by all the variables (m) is modeled by a factor analysis structure.
The common factors Z : small number ($q << m$) of latent variables (Friguet *et al.*, 2009, *JASA*)

Specific variability (uniqueness) Common variability

$$\Sigma = \Psi + BB'$$

$$Y^{(k)} = \beta_0^{(k)} + x'\beta^{(k)} + BZ + \varepsilon^{(k)}$$

Common factors

$$Z \sim N\left(0; I_q\right), V(\varepsilon) = \Psi$$

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Similar idea : Surrogate Variable Analysis method, Leek and Storey, 2007, 2008.

Factor-adjusted test statistics

The adjusted test statistics are conditionally centered and scaled version of usual test statistics

Conditional distribution of the usual test statistic $T^{(k)}$

$$\mathbb{E}(T^{(k)} \mid Z) = \tau_k + \frac{b_k'}{\sigma_k}\tau(Z), \quad \mathrm{Var}(T^{(k)} \mid Z) = \frac{\psi_k^2}{\sigma_k^2}.$$

Conditional centering and scaling

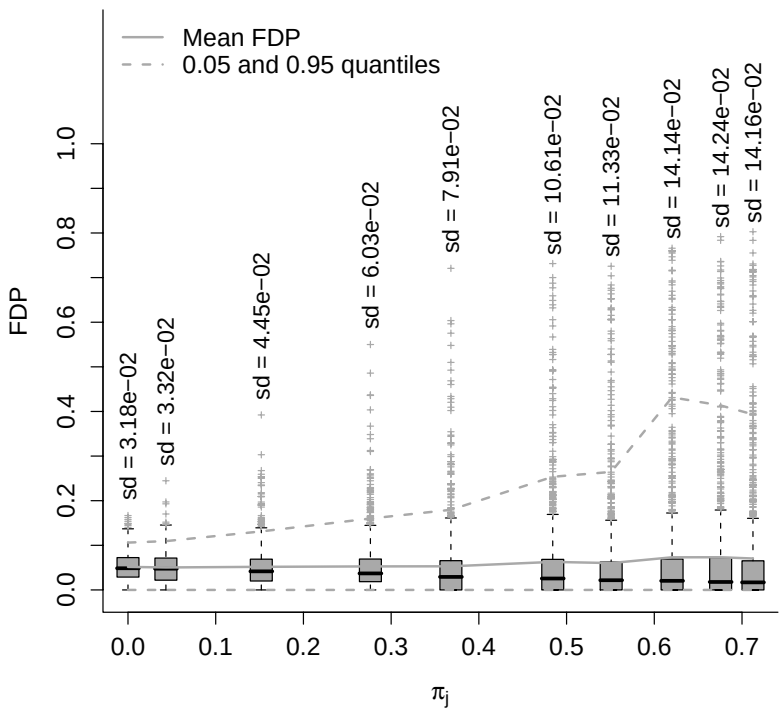
$$T_z^{(k)} = \frac{\sigma_k}{\psi_k}\left[T^{(k)} - \frac{b_k'}{\sigma_k}\tau(Z)\right].$$

with $\mathbb{E}(T_z^{(k)}) = \frac{\tau_k}{\sqrt{1-h_k^2}}$ and $\mathrm{Var}(T_z) = I_m$.

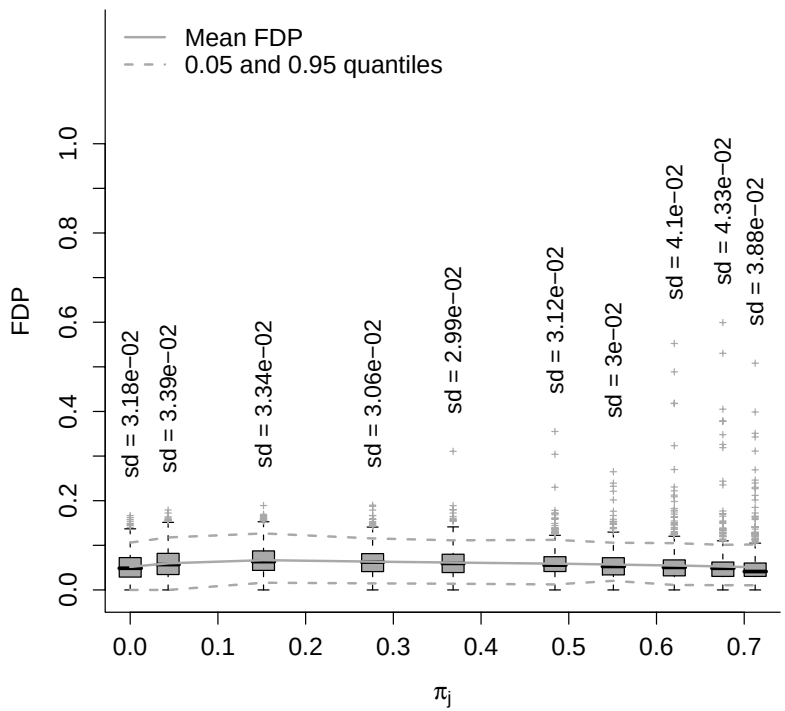
Distribution of error rates in multiple tests

Distribution of False Discovery Proportion on 1.000 simulated datasets/scenario (Friguet *et al.*, 2009, *JASA*)

Usual t-tests



Factor-adjusted t-tests



Distribution of error rates in multiple tests

Distribution of Non-Discovery Proportion on 1.000 simulated datasets/scenario (Friguet *et al.*, 2009, *JASA*)

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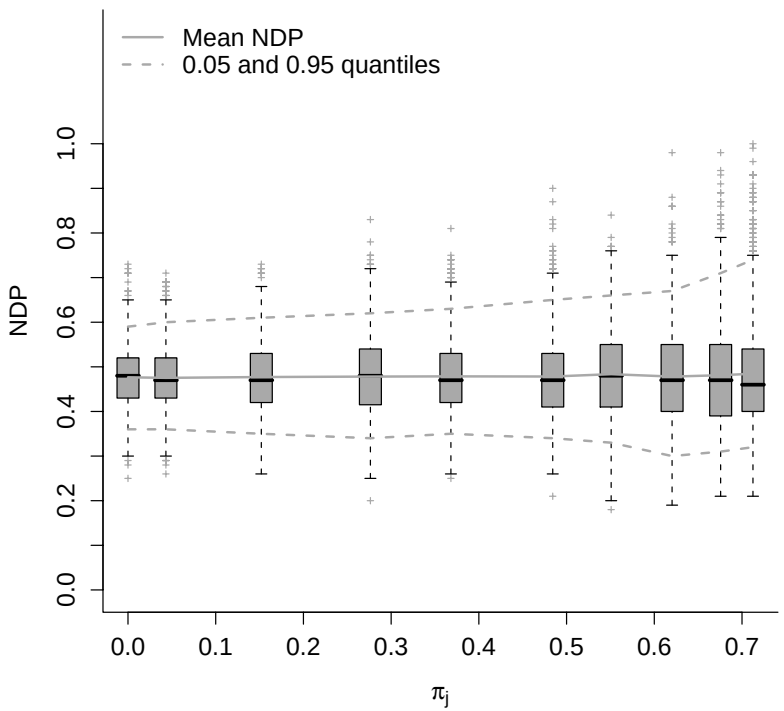

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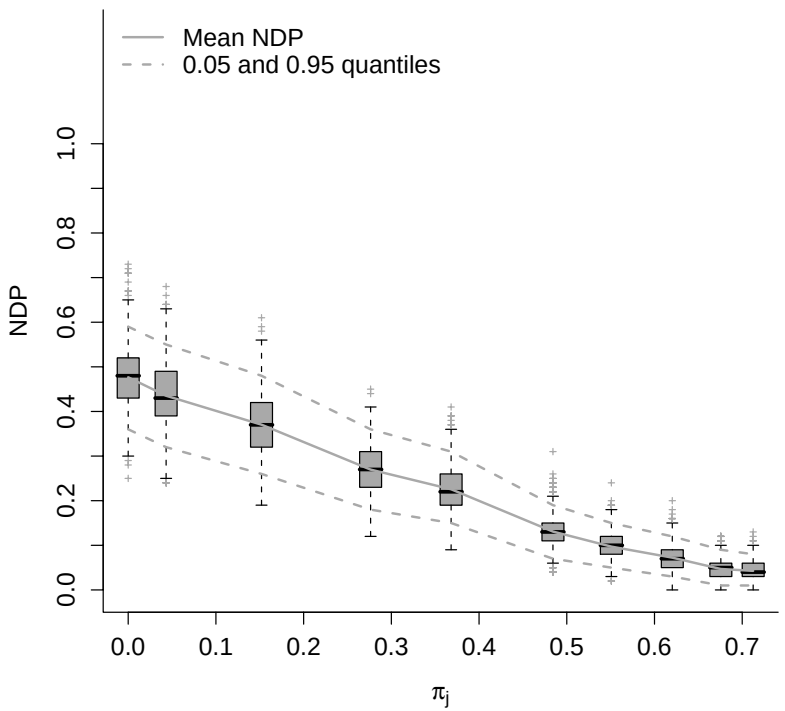

Outline

The FAMT package steps

1. Estimation of the number of factors
2. Factor Analysis model (using $\widehat{\mathcal{M}_0} = \{k,\ P_k \geq \alpha\}$)
3. Multiple testing : conditional statistics and p-values
   $\widehat{\mathcal{M}_0}$ updated, step 1 to 3 are done twice
4. Estimation of the proportion of null hypotheses
5. Benjamini and Hochberg's procedure to control the FDR

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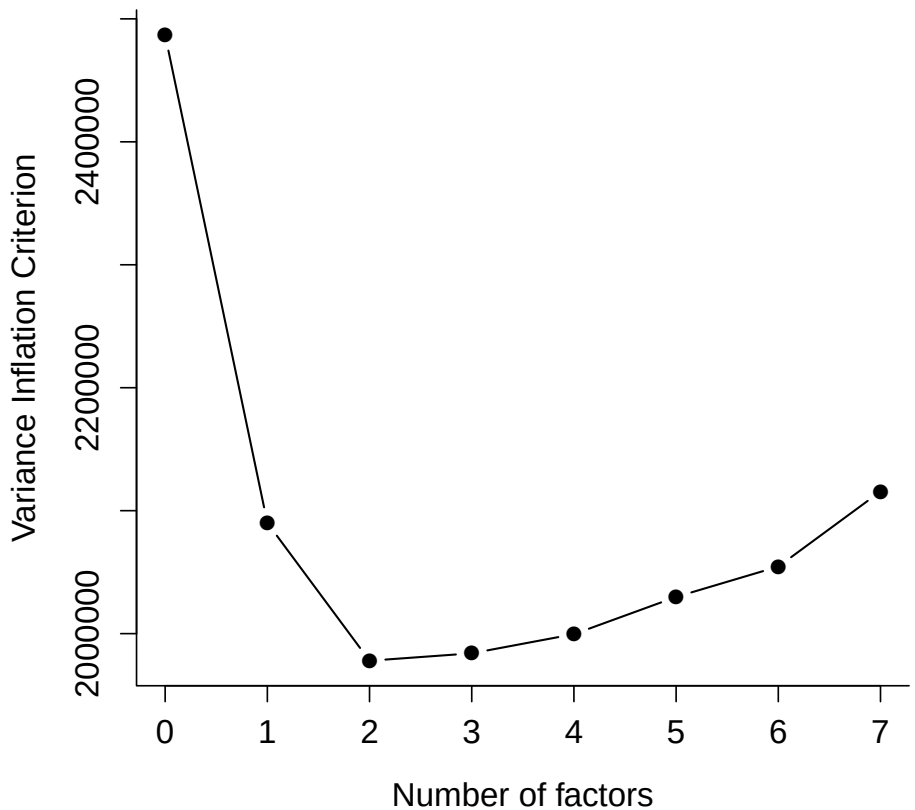
Illustration on the Lymphoma dataset (Alizadeh *et al.* 2000)

- 32 samples : 2 classes of B cell-like diffuse large cell lymphoma (DLCL) : germinal center B cell-like DLCL (18 samples) and active B cell-like DLCL (14 samples)
- Expression levels of 10295 genes

1/ Estimation of the number of factors

The number of factors is chosen to reduce the variance of the number of false positives in multiple tests.



2/ Factor Analysis model

To deal with high-dimension, the model parameters are estimated with an EM-algorithm (Rubin and Thayer, 1982) :

- E step : estimation of Z
- M step : estimation of B and Ψ

Specific variability (uniqueness) Common variability

$$\Sigma = \Psi + BB'$$

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Common factors

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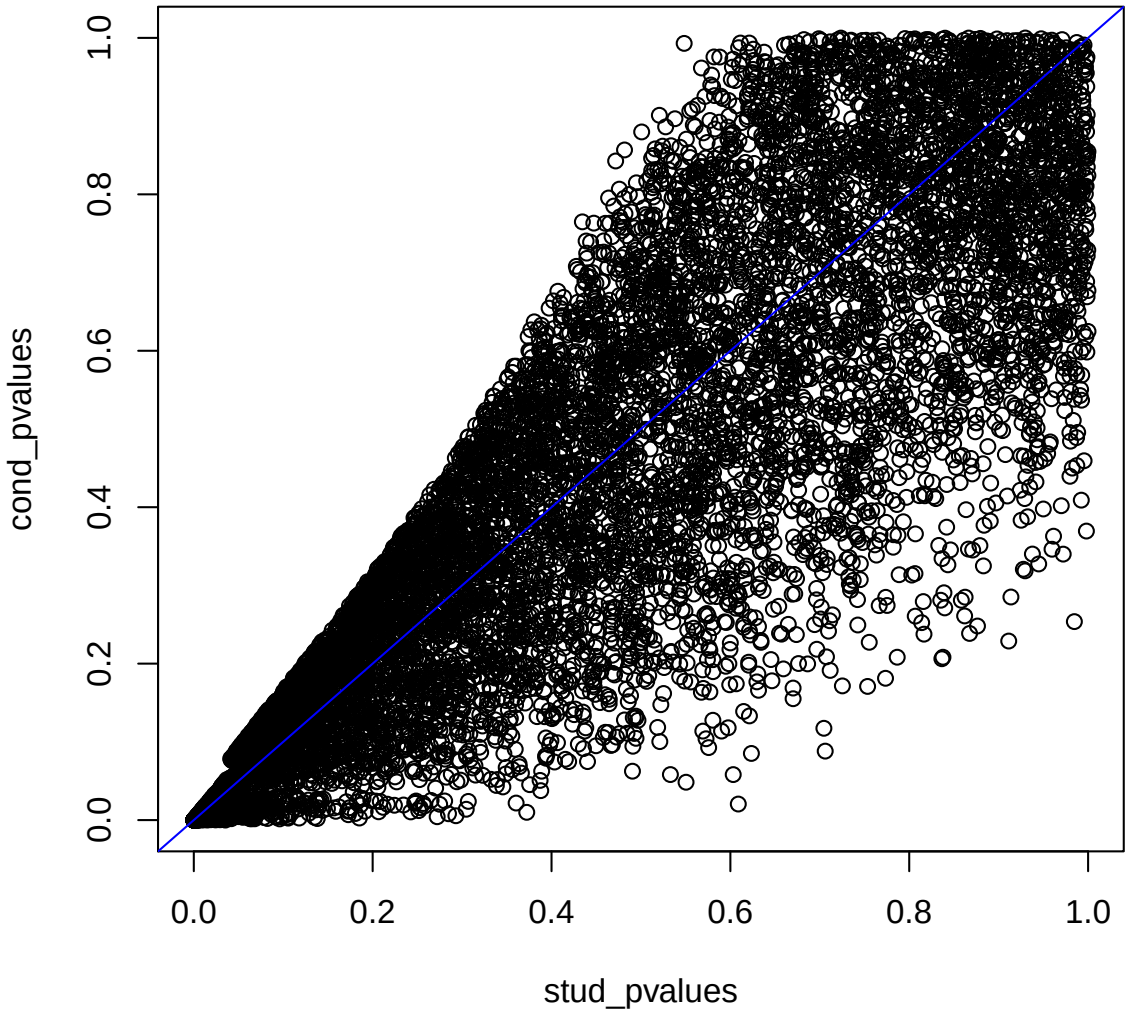
3/ Multiple testing (conditional p-values)



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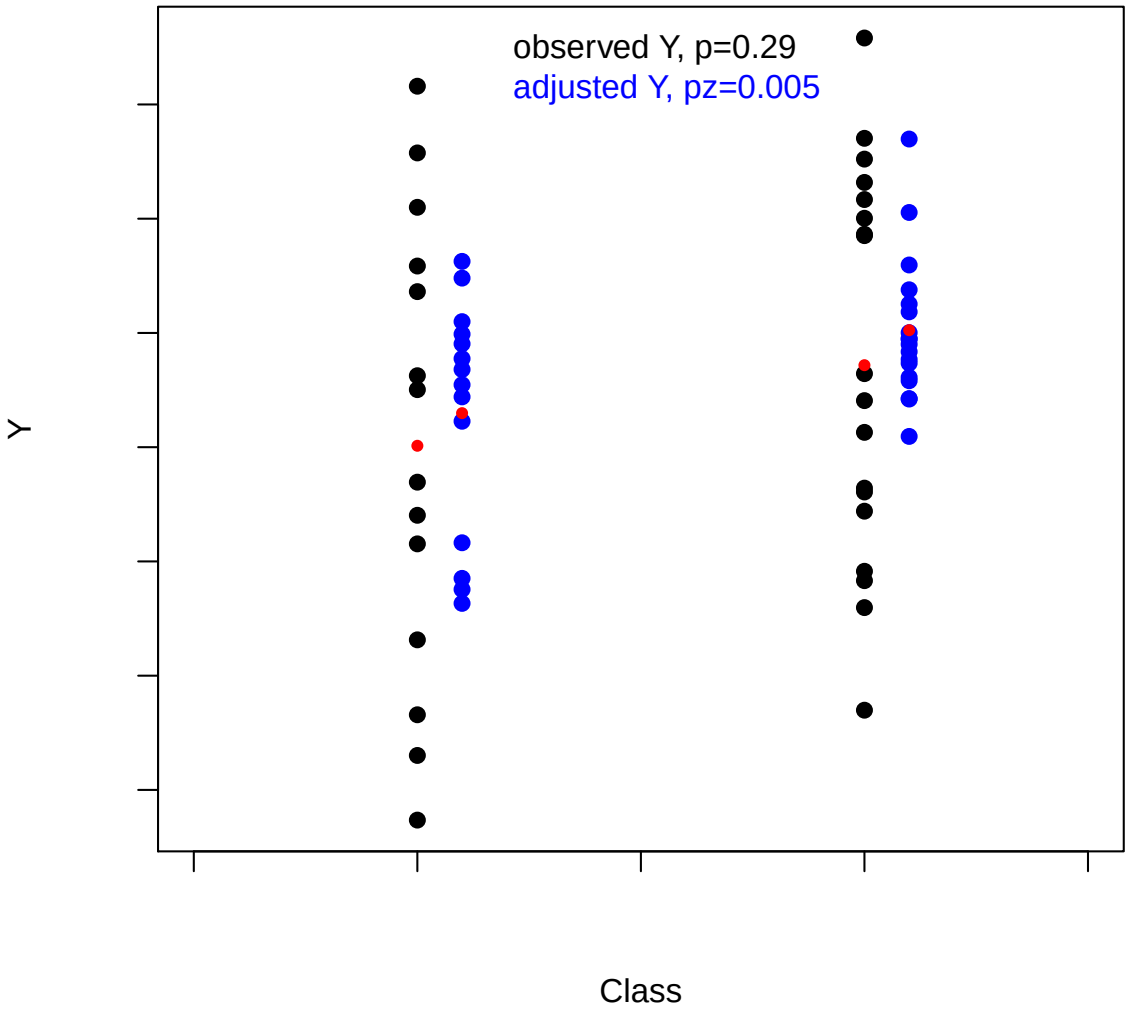
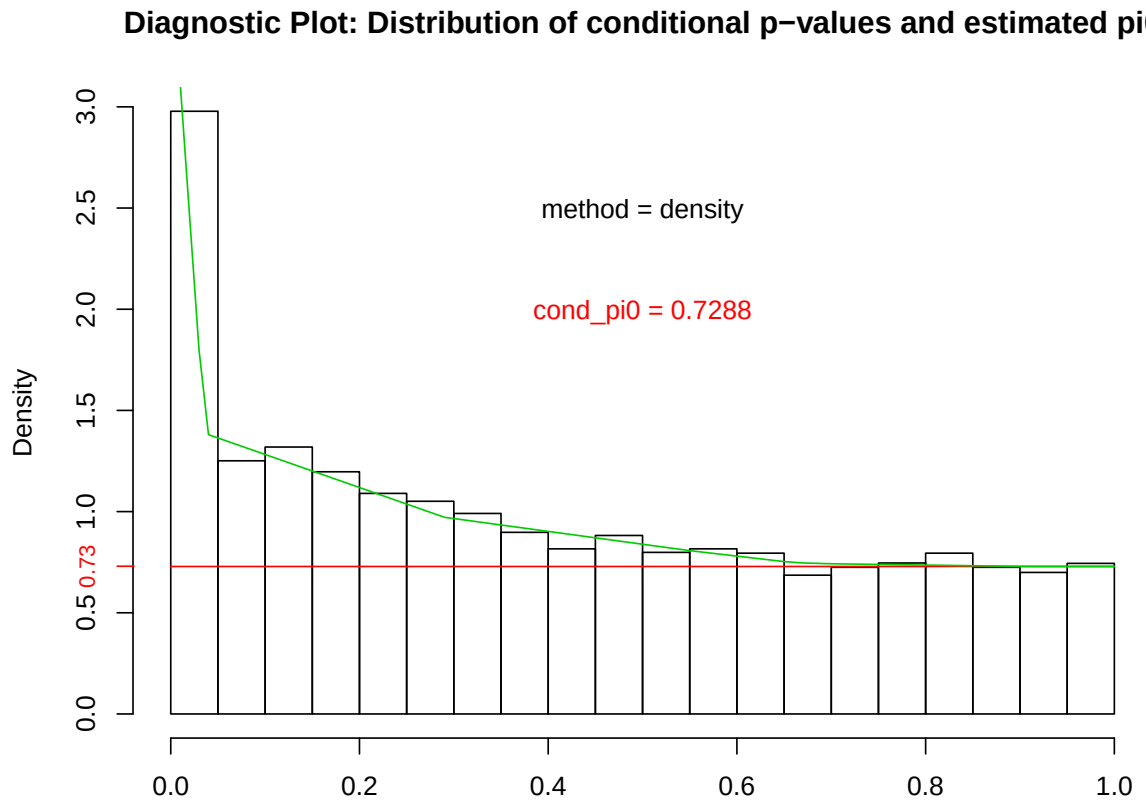


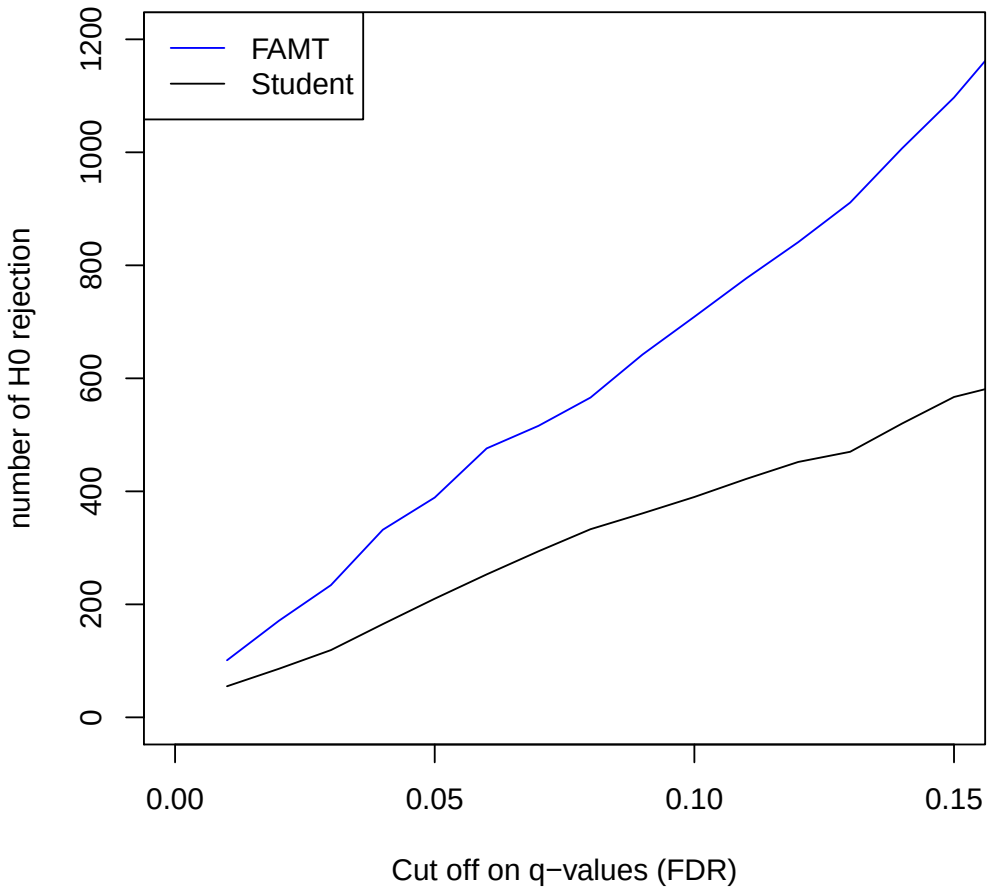
4/ Estimation of the proportion of null hypotheses

Key parameter to control the error rates.
FAMT provides 2 estimation algorithms :

- one based on the density of the conditional p-values
- the other uses a modified smoothing spline approach (based on Storey and Tibshirani, 2003).



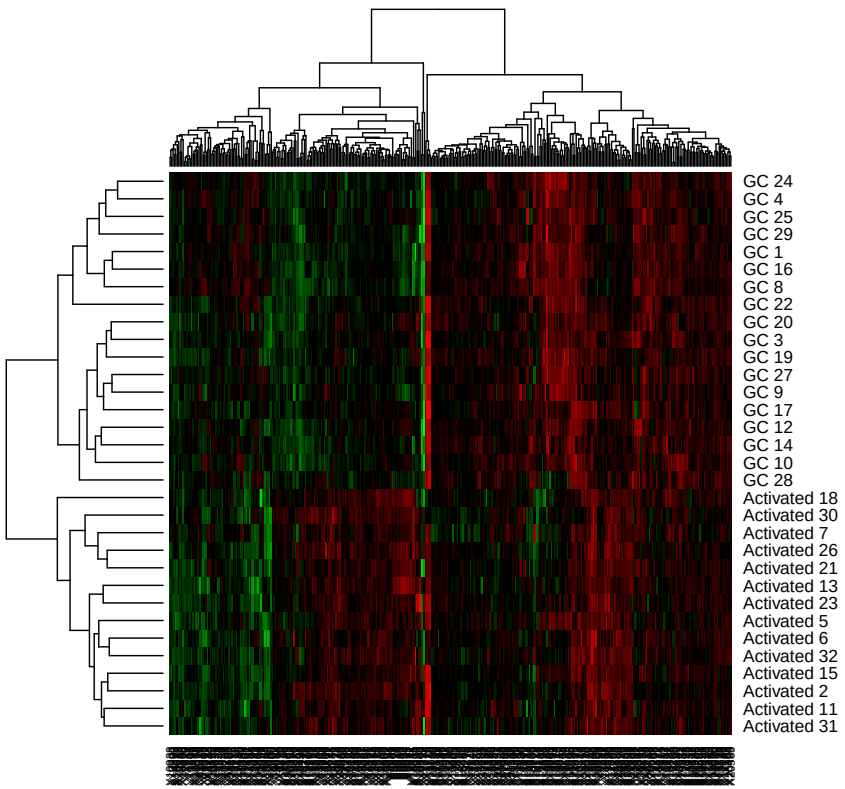
5/ Benjamini and Hochberg's procedure (q-values)



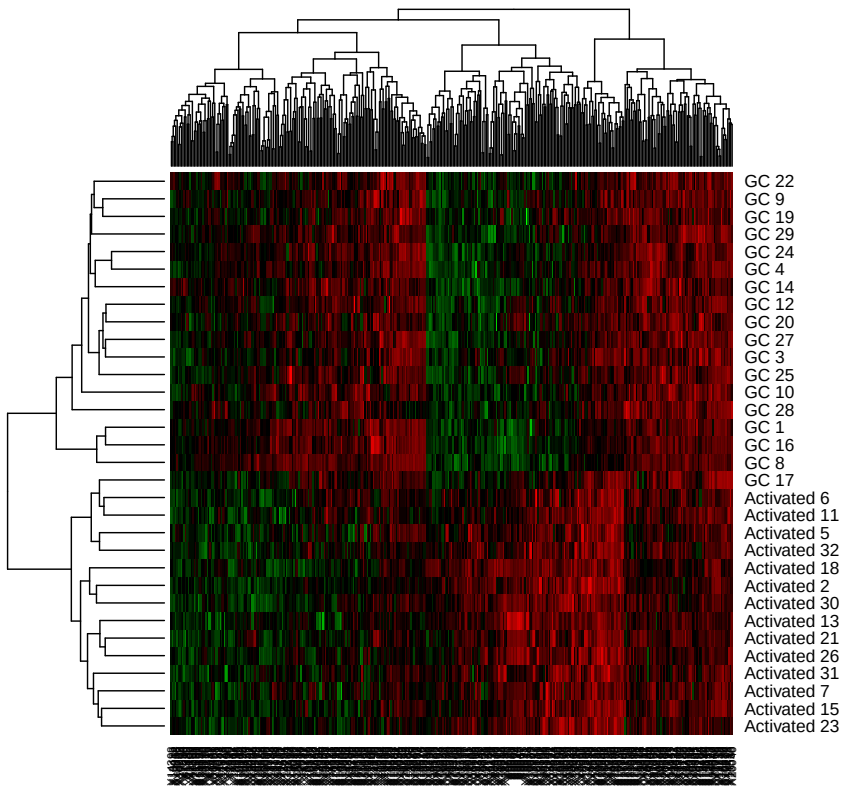
Heat maps

Cut off on the adjusted q-values : 5% FDR control level (389 genes)

Observed values



Factor-adjusted values



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Concluding comments

- FAMT procedure : large improvements in multiple testing procedures regarding the FDR control and the power (decreasing the non-discovery proportion)
- The interpretation of the factors can be useful for biologists
- The factor-adjustment of test statistics also decreases misclassification rates and improves stability of model selection in supervised classification
- FAMT  package available at *http://www.agrocampus-ouest.fr/math/FAMT*

Interpretation of the factors