What's new?

2016/07/29

<u>"Global overview " tab</u>

A "Global Overview" tab has been added to assess and visualize the precision across all replicates independent of either the condition, number of peptides or labeling time. The BoxPlot representation summarizes the variability across replicates and is identical to the one used in the Peptide Plot tab, except that the deuterium uptake values of all peptides are considered. As an example, the following results are generated with the test.csv file provided with the software. An excellent reproducibility is observed between each replicate for both conditions (Fig.1). This representation also highlights the differences of deuterium uptake measured in both conditions: more deuterium is incorporated in the presence of EDTA (blue boxplots) than in the presence of calcium (red boxplots).

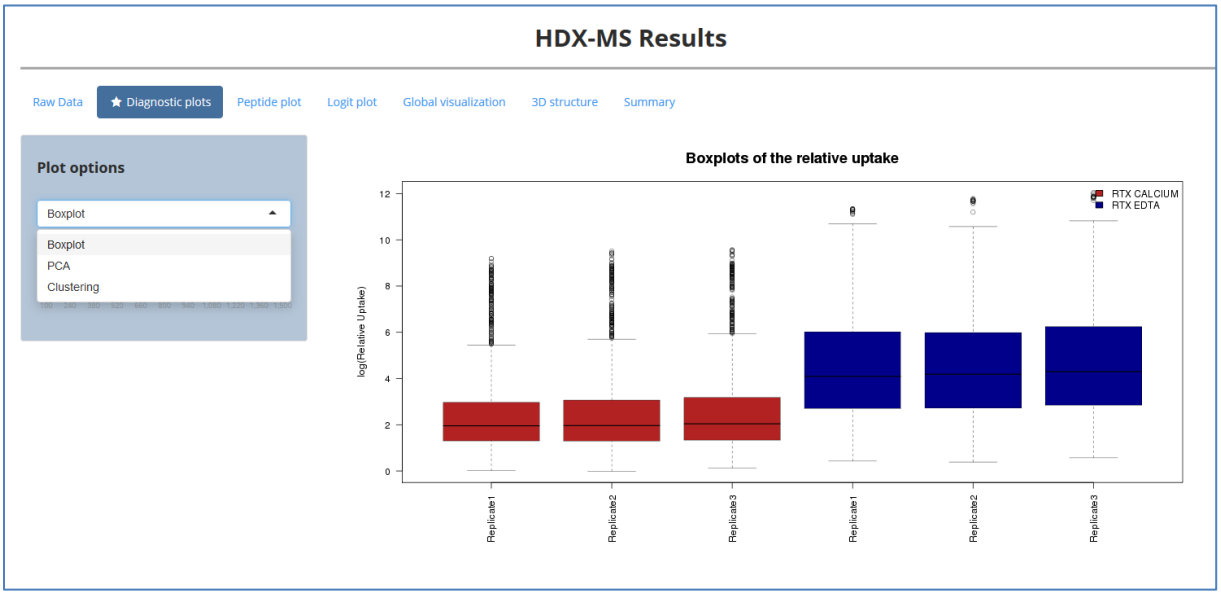

Figure 1. BoxPlots of the variability across replicates generated with the test.csv file.

The Principal Component Analysis (PCA) summarizes the effects of the variance on the entire HDX-MS datasets independent of either the conditions or labeling time (Fig. 2). In the example file, the condition represents the first principal component (PC1). Here, 99.76% (PC1) of the variability of the entire HDX-MS dataset can be explained by the condition *only*. PC2 and PC3 do not discriminate the data in this dataset.

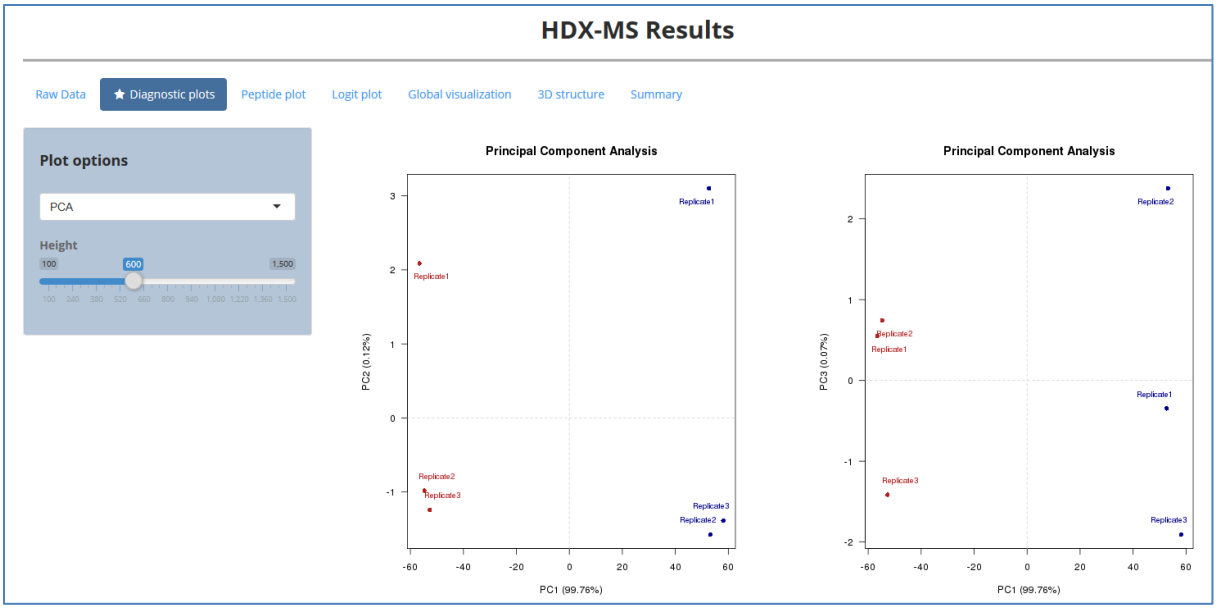

Figure 2. PCA plots generated with the test.csv file.

Finally, the dendrogram uses the euclidean distance between replicates to cluster them using the Ward criterion (Fig.3).

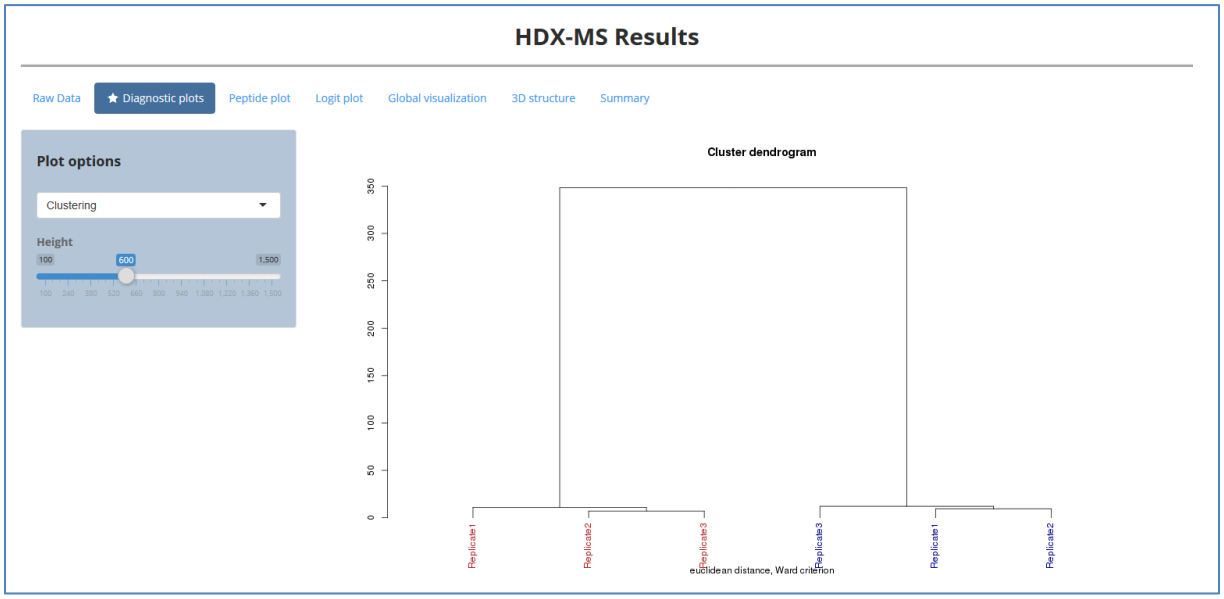

Figure 3. Cluster dendrogram generated with the test.csv file

“Biological threshold”

A “biological threshold” has been added in the “Global visualization” tab. This parameter allows the identification of statistically significant peptides displaying Fractional Uptake Difference values below the user-defined threshold (default value sets to 2%). In some cases, very small variations between conditions are considered as statistically significant by MEMHDX due to the very low variability measured between replicates. In such a scenario, the deuterium uptake curves of a peptide measured in two conditions show no difference by eyes (*i.e.*, both curves overlap) and would therefore not be considered as biologically relevant by the user.

By fixing a “biological threshold”, the user can easily identified such peptides and decide if the amplitude of variations observed between condition is biologically relevant or not.