



### Integrative DE analysis of snRNA and snATAC data:

- selected cell types: Myonuclei TI/TII of Old cohort vs Myonuclei TI/TII of Young cohort;
- differential snATACseq peak analysis (selected only promoter genomic regions) and DGE of snRNA;
- annotate our differential ATAC-seq regions to genes;
- select DE overlapped genes in both datasets ( $\text{Log}_2\text{FC} \geq 1$  &  $\text{Log}_2\text{FC} \leq -1$ ,  $\text{padj} < 0.05$ );
- perform correlation test of  $\text{Log}_2\text{FC}$ s (effect sizes) of both datasets;
- use gene information to test enrichment for GO sets.

To investigate whether the changes in open chromatin regions were correlated with the changes in gene expression levels, we performed an integrative analysis of the snATAC-seq and snRNA-seq data sets. A total of 401 overlapping genes were identified, including 213 upregulated and 188 downregulated genes in ATAC-seq, 230 upregulated and 171 downregulated genes in RNA-seq. We did a correlation analysis of the expression levels and chromatin openness of these 401 overlapping genes and observed a significant positive correlation between the differential gene expression and differential ATAC-seq signal ( $r^2 = 0.73$ ) consistent with the paper results.

Interestingly, some DE genes are not match with chromatine accesibility, indicating the regulation at other levels.