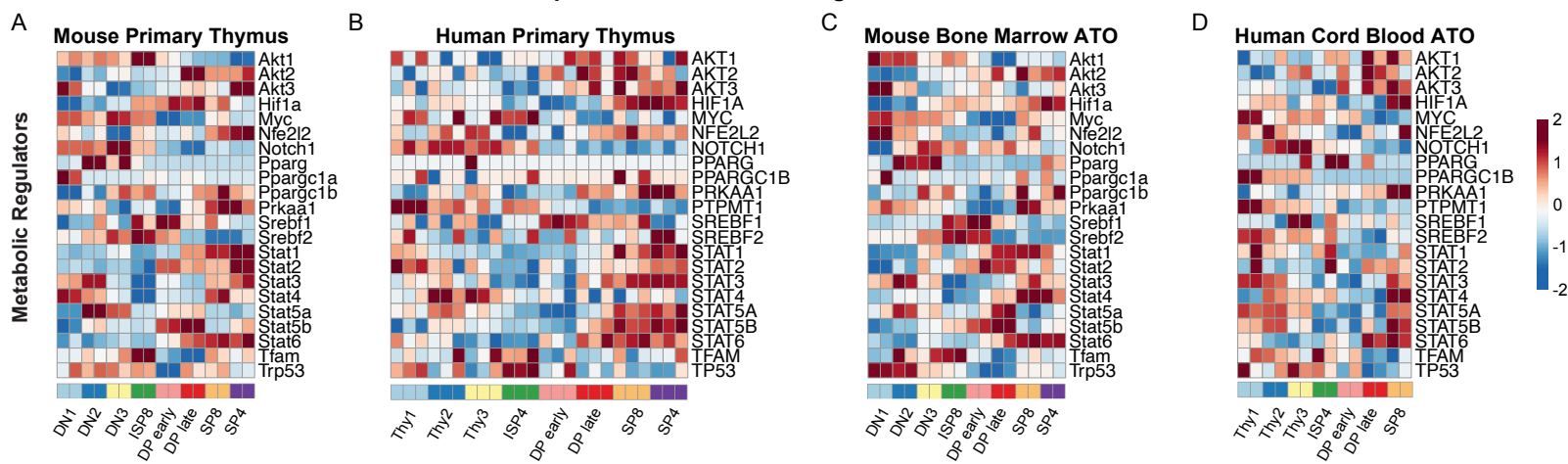
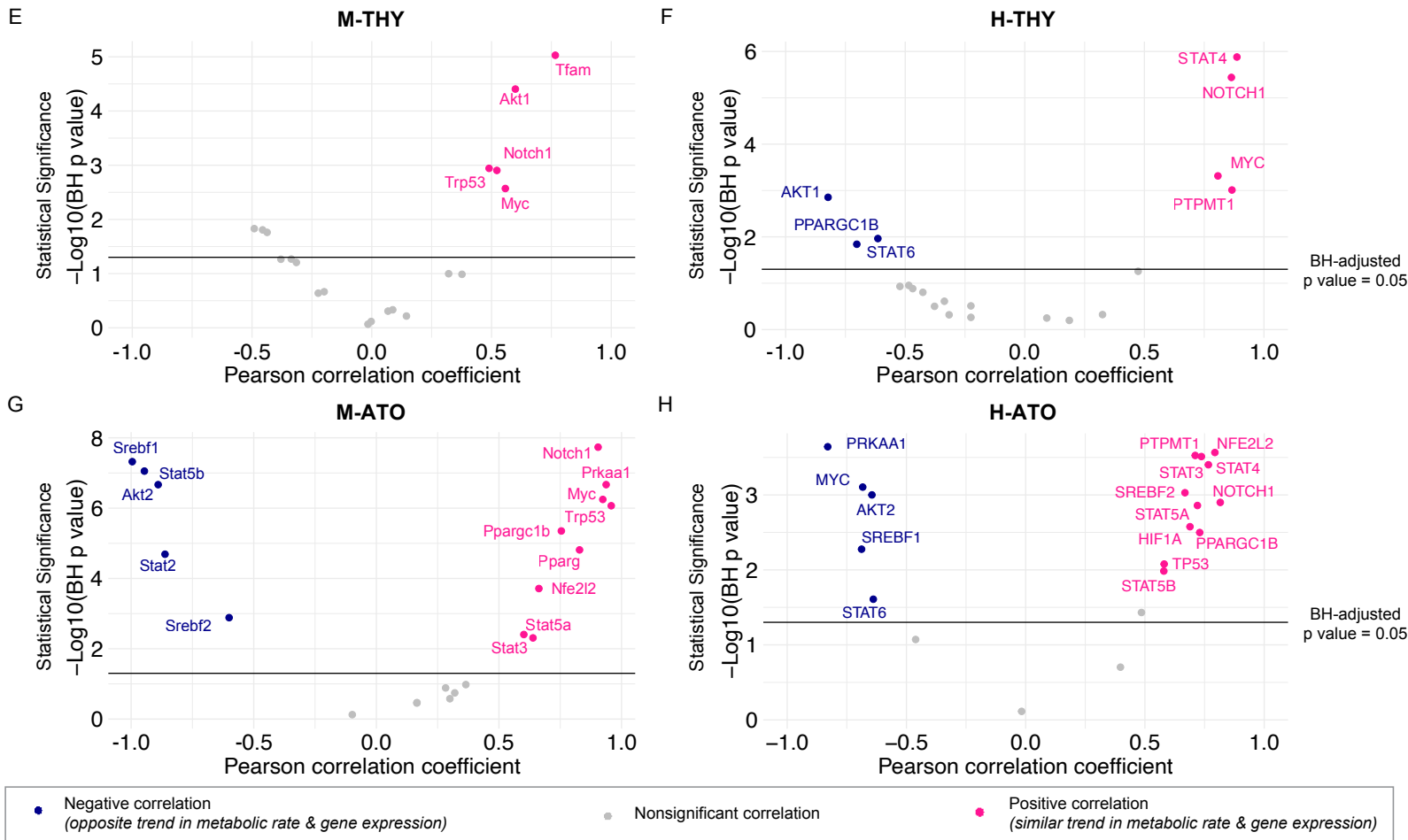


## Supplementary Figure 5

### Expression of Metabolic Regulator Genes



### Correlation of basal OCR and metabolic regulator genes



### Supplementary Figure 5 (related to Figure 6):

(A-D) Heatmaps and summary line plots of genes encoding key signaling molecules and transcription factors that regulate metabolic programs in (A) M-THY, (B) H-THY, (C) M-ATO, and (D) H-ATO. The x axis indicates cell populations isolated for analysis. Each individual heatmap represents z-scores of normalized variance-stabilized gene expression data. (n=2 independent replicates per population in M-THY; n=3 in H-THY; n=2 in M-ATO; n=2 in H-ATO).

(E-H) Scatter plots show correlation between gene expression of metabolic regulators and basal respiration (OCR) in (E) M-THY, (F) H-THY, (G) M-ATO, and (H) H-ATO. The x axis represents the Pearson correlation coefficient R values calculated for individual genes within the specified metabolic pathway for all thymocyte populations for which respective extracellular flux data was collected. The y axis represents the statistical significance of each R value using  $-\log_{10}$  of Benjamini-Hochberg adjusted p-values. Solid black line indicates an adjusted p-value of 0.05; all R values above the line are statistically significant. Genes with R values  $>0.5$  (pink) are positively correlated with metabolic flux data, while genes with R values  $<-0.5$  (blue) are negatively correlated with metabolic flux data. Nonsignificant or weak correlation R values  $>-.05$  or  $<0.5$  are indicated in grey. (M-THY: n=6 flux analysis values for 5 populations, n=2 RNA-seq replicates; H-THY: n=3 flux analysis values for 5 populations [except for ISP4: n=2], n=3 RNA-seq replicates; M-ATO: n=6 flux analysis values for 3 populations, n=2 RNA-seq replicates; H-ATO: n=6 flux analysis values for 3 populations [except for ISP4: n=5], n=2 RNA-seq replicates)