

Supplemental materials

Figure S1. Age-related changes of DNA methylation.

(A) Volcano plot demonstrating changes in methylation sites. The color keys indicate age-downregulated DMPs (blue) and age-upregulated DMPs (red).

(B) Heatmap showing the results of deconvolution of blood cells with the houseman method. Rows represent the cell and columns represent the individuals. The shade of color indicates the proportion of cells. CA: chronological age.

(C) Bar plot showing the enriched pathways of genes related to age-increased (top, red) and age-decreased (bottom, blue) differential DNA methylation sites. Key DMP-related genes enriched in the indicated terms are listed below. The different shade of colors indicates the $-\log_{10}P$ value from low to high.

(D) Bar plots showing the pathway enrichment results of the changing features at 35 years old and key DMP-related genes enriched in the indicated terms are listed below.

(E) Bar plots showing the enriched pathways of age-decreased genes potentially regulated by NRF1. The shade of color indicates the $-\log_{10}P$ value from low to high.

(F) Heatmap showing the most shared sites between different clocks and the methylation changes with aging. The different shades of color on the top represent different ages. Bins are colored from blue to red to indicate low to high methylation levels. CA: chronological age.

(G) Transcription factors whose transcriptional expression levels trend in the opposite direction to the methylation level of the motif binding site. Left, scatter plots showing the correlation between the mean methylation values of the motif and the expression of TF binding to the motif. Right, bar plots showing the enriched pathways of age-decreased genes potentially regulated by TFs. The shade of color indicates the $-\log_{10}P$ value from low to high.

(H) Bar plot showing the number of probes specific to each clock, with up-regulated methylation sites colored in red and down-regulated methylation sites colored in blue (top). Heatmap showing the enriched pathways of probe-related genes (bottom). The different shade of colors indicates the $-\log_{10}P$ value from low to high.

Figure S2. Characterization of the age-related DNA methylation changes in female.

(A) Dot plot showing the PCA distribution of methylation patterns. The different shades of color represent different ages.

(B) Scatter plot showing the correlation of mean methylation levels with chronological age. Correlation coefficient and P-value (Pearson's correlation analysis) are shown.

(C) Scatter plot showing the correlation of Shannon entropy with chronological age. Correlation coefficient and P-value (Pearson's correlation analysis) are shown.

(D) Venn plot showing the overlap between the changed features at two ages (left, age 35; right, age 55).

(E) Changes of age-related differential methylation positions with age (linear correlation, $|\Delta\text{Beta}/\text{year}| > 0.002$ and Benjamini-Hochberg adjust P value < 0.01 , corrected with BMI, gender and proportion of blood cells). Each row of the heatmap represents a methylation site, and each column represents an individual. Bins are

colored from blue to red to indicate low to high methylation levels. CA: chronological age.

(F) Sliding window analysis demonstrating the number of differentially methylated sites that change at different ages. Line plot showing the number of DMPs at different ages.

(G) Heatmaps showing the scale values of features changing at 35 years old (top) and 55 years old (bottom). The color of the bins indicates the scaled abundance of beta value from low (blue) to high (red).

(H) Bar plots showing the pathway enrichment results of the changing features at 35 and 55 years old and key DMP-related genes enriched in the indicated terms are listed below.

(I) Histogram showing the proportion of chromosomes in regions with high age-related methylation variation. The colors of the pots indicate different chromosomes.

(J) Density plot showing the distribution of significantly changing methylation sites on chromosomes with age, with up-regulated methylation sites colored in red and down-regulated methylation sites colored in blue (top). LOESS fitting plots showing the average methylation change in the distribution region of differentially methylated hotspots, methylation sites that change linearly with age are colored and the remaining sites are grey (linear correlation, $|\Delta\text{Beta}/\text{year}| > 0.002$ and Benjamini-Hochberg adjust P-value < 0.01 , corrected with BMI and proportion of blood cells), and the dashed line showing the average methylation beta value (bottom).

Figure S3. Characterization of the age-related DNA methylation changes in male.

(A) Dot plot showing the PCA distribution of methylation patterns. The different shades of color represent different ages.

(B) Scatter plot showing the correlation of mean methylation levels with chronological age. Correlation coefficient and P-value (Pearson's correlation analysis) are shown.

(C) Scatter plot showing the correlation of Shannon entropy with chronological age. Correlation coefficient and P-value (Pearson's correlation analysis) are shown.

(D) Venn plot showing the overlap between the changed features at two ages (left, age 35; right, age 80).

(E) Changes of age-related differential methylation positions with age (linear correlation, $|\Delta\text{Beta}/\text{year}| > 0.002$ and Benjamini-Hochberg adjust P value < 0.01 , corrected with BMI and proportion of blood cells). Each row of the heatmap represents a methylation site, and each column represents an individual. Bins are colored from blue to red to indicate low to high methylation levels. CA: chronological age.

(F) Sliding window analysis demonstrating the number of differentially methylated sites that change at different ages. Line plot showing the number of DMPs at different ages.

(G) Heatmaps showing the scale values of features changing at 35 years old (top) and 55 years old (bottom). The color of the bins indicates the scaled abundance of beta value from low (blue) to high (red).

(H) Histogram showing the proportion of chromosomes in regions with high age-related methylation variation. The colors of the pots indicate different chromosomes.

(I) Density plot showing the distribution of significantly changing methylation sites on chromosomes with age, with up-regulated methylation sites colored in red and down-regulated methylation sites colored in blue (top). LOESS fitting plots showing the average methylation change in the distribution region of differentially methylated hotspots, methylation sites that change linearly with age are colored and the remaining sites are grey (linear correlation, $|\Delta\text{Beta}/\text{year}| > 0.002$ and Benjamini-Hochberg adjust P-value < 0.01 , corrected with BMI, gender and proportion of blood cells), and the dashed line showing the average methylation beta value (bottom).

Figure S4. GSVA of genes in the age-related probe-gene pairs.

(A) GSVA of age-upregulated genes in probe-gene pairs. Box plots show the GSVA scores, with points representing the individual, and color keys indicate the age-accelerators (red) and age-decelerators (blue).

(B) GSVA of age-downregulated genes in probe-gene pairs. Box plots show the GSVA scores, with points representing the individual, and color keys indicate the age-accelerators (red) and age-decelerators (blue).

Figure S5. Age pace of DNA methylation-based multi-model aging clocks.

(A) Relationship between age pace predicted by DNAm-based multi-model age clock and multi-model age/chronological age. Scatter plots showing the correlation (Pearson's correlation analysis) between age pace and predicted age/ chronological age. Density plots of age pace, and the color keys indicate the age-accelerators (red) and age-decelerators (blue).

Figure S6. Associations between age pace of DNA methylation-based multi-model aging clocks and multi-omics features.

(A) Dot plots showing the multi-omics changed features in DNAm-based multi-modal age accelerators. Colors indicate the type of DNAm-based multi-modal age. DA: DNAmAge.

(B) Dot plots showing the number of features associated with age accelerated predicted by different clocks, and the color keys indicate the clock types. Heatmap showing differential genes, metabolites, and proteins, with rows representing the multi-model ages, columns representing the features, and the color keys indicate the differences and the class of features. The shade of color in the heatmap indicates the scaled abundance of features measurement.

(C) Heatmap shows differential phenotypes (T-test, P-value < 0.05), with rows corresponding to DNAm-based multi-model ages, columns corresponding to features, and the classification of the measurement phenotypes is annotated as bar plots above. The shade of color indicates the Log2foldchange value from low to high.

Supplementary Table Legends

Table S1. The distribution of age and gender in cohorts.

Table S2. Age-related differential methylation positions.

Table S3. Sliding window analysis results.

Table S4. Age-related probe-gene pairs.

Table S5. Gene set variance analysis differences between young and old for the DNAm-related aging-DEGs.

Table S6. Motif enrichment of probe-gene pairs.

Table S7. The final models to build the DNAm-based aging clocks.

Table S8. The predicted age of previous DNAm clocks.

Table S9. The predicted age of DNAm-based multi-model age

clocks. Table S10. The predicted age of 5 CpGs clock.

Table S11. DNAm-based multi model age pace related metabolites (age-accelerators vs age-decelerators).

Table S12. DNAm-based multi model age pace related proteins (age-accelerators vs age-decelerators).

Table S13. DNAm-based multi model age pace related genes (age-accelerators vs age-decelerators).

Table S14. DNAm-based multi model age pace related phenotypes and life factors (age-accelerators vs age-decelerators).

Fig S1

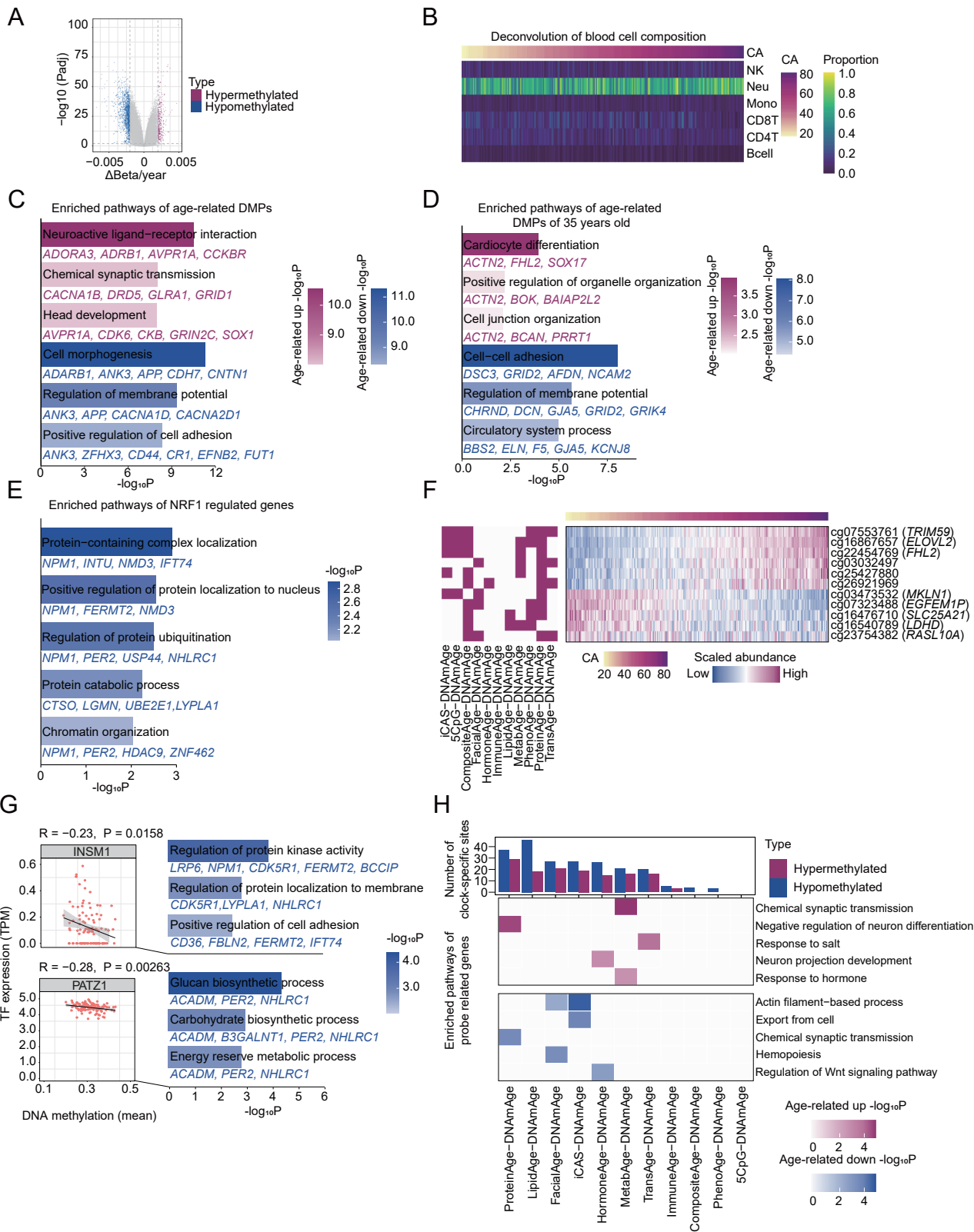


Fig S2

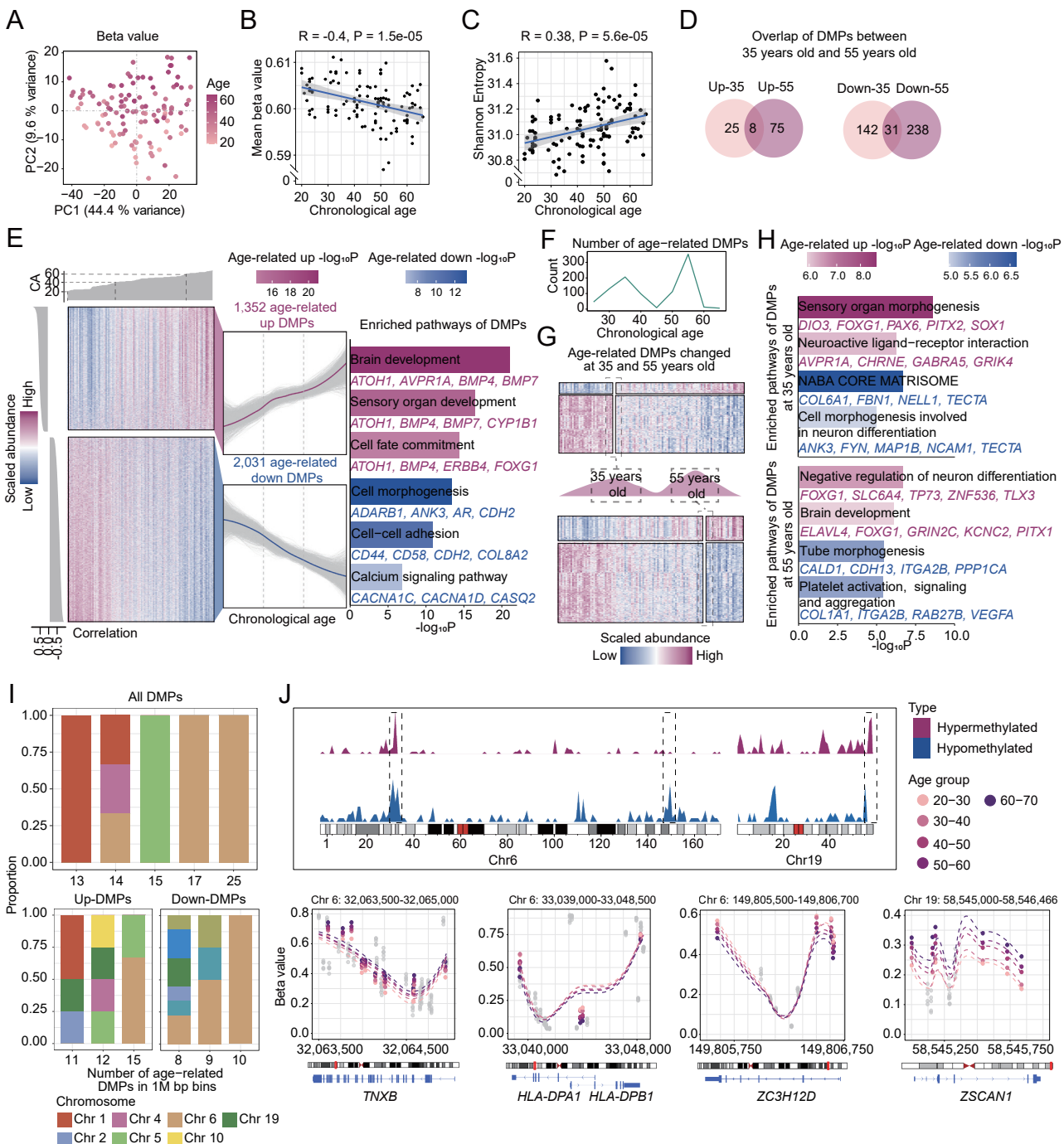


Fig S3

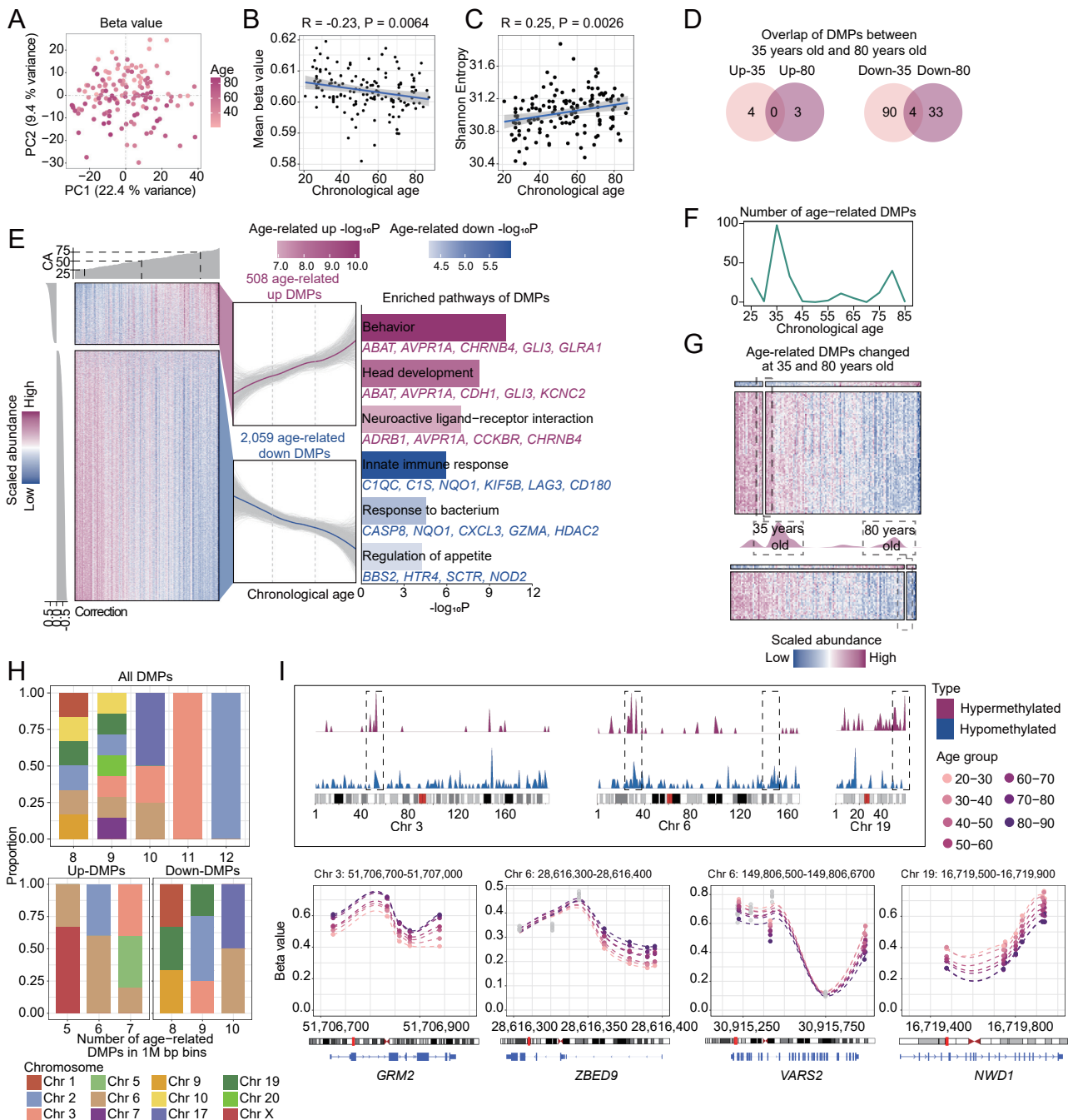
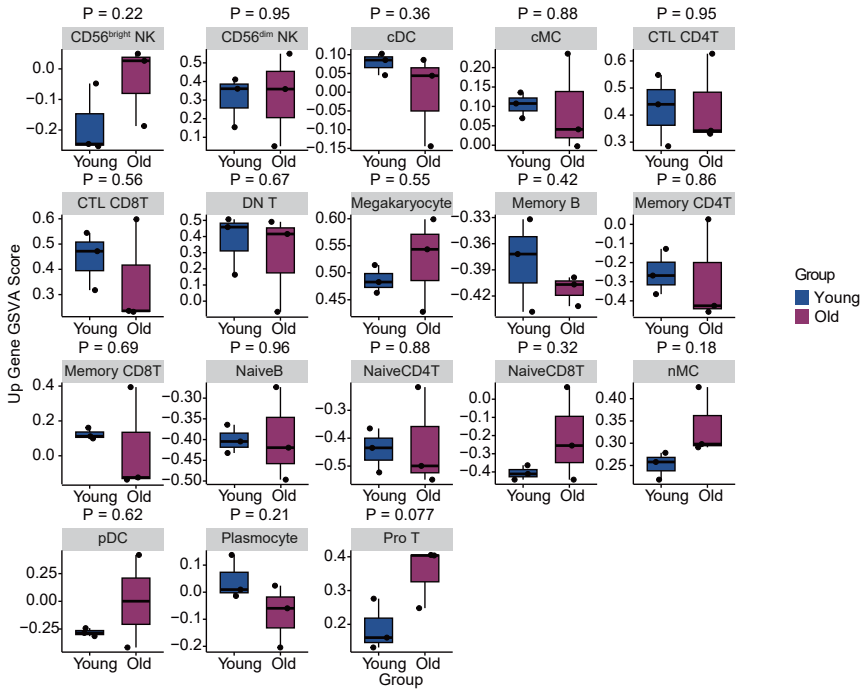
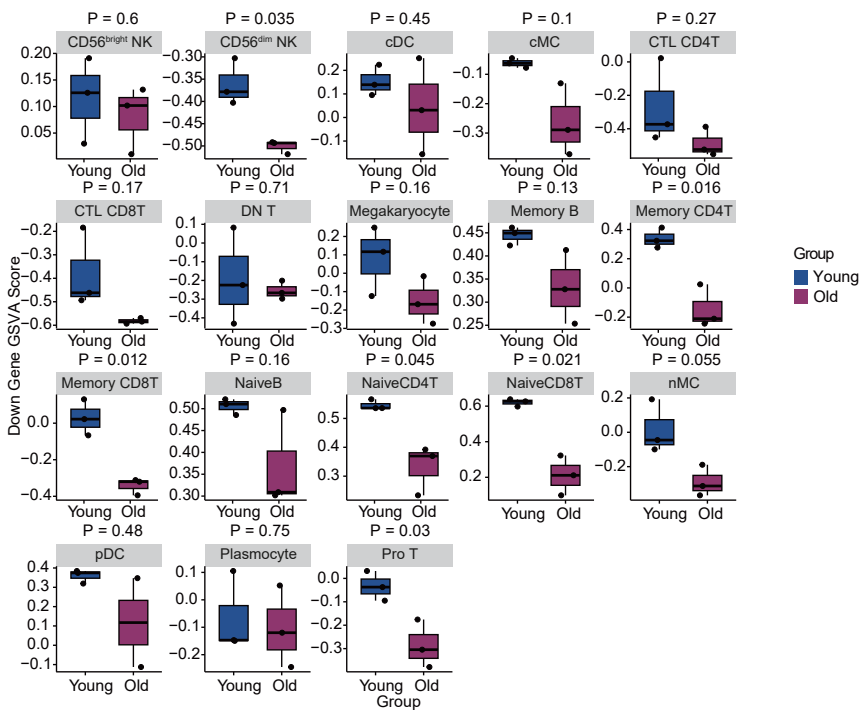


Fig S4

A



B



A

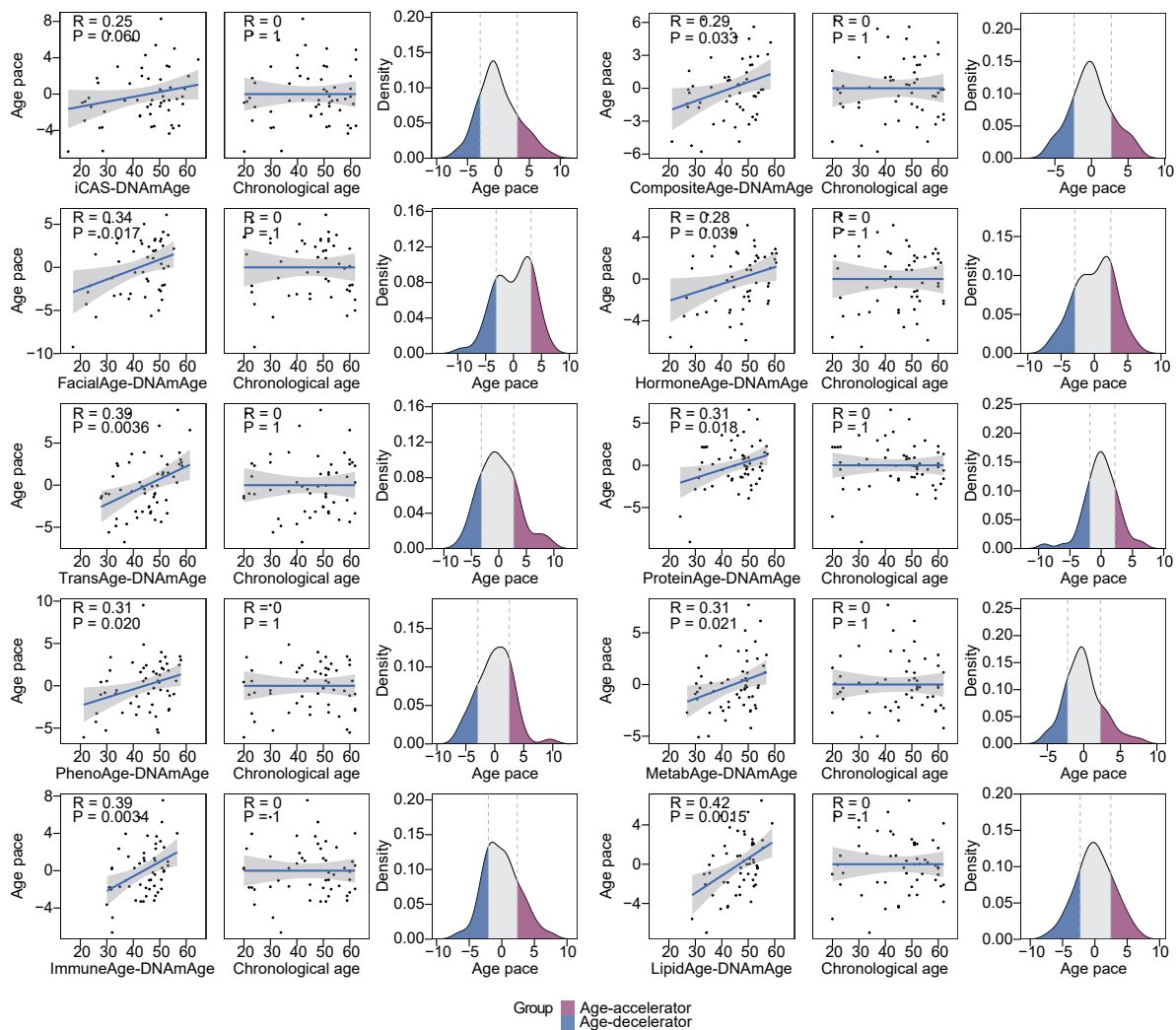
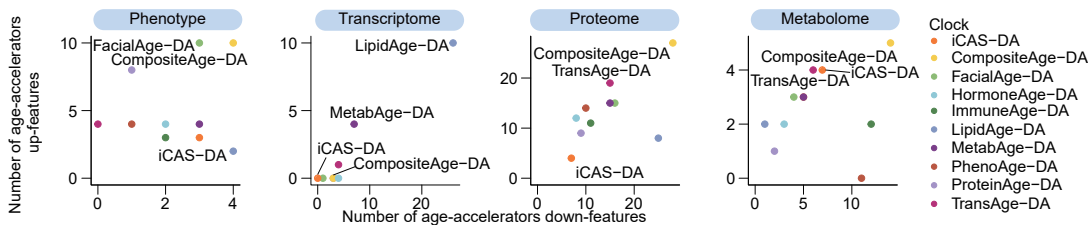
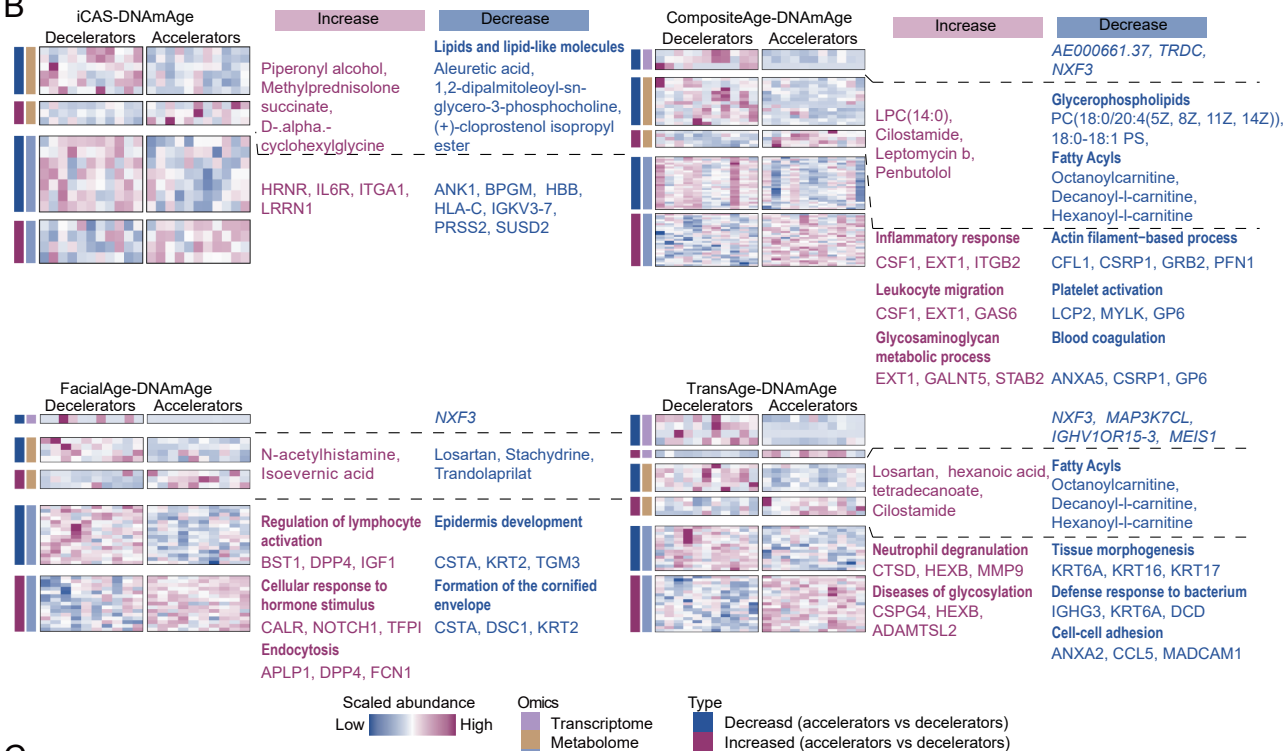


Fig S6

A



B



C

