SUPPLEMENTAL MATERIAL

Table S1. The primers of qPCRs.

Etv5-F	AGAAAGAGGAAGTTTGTGGAC
Etv5-R	ATGAAGCACCAAGTTATCAGAC
Rbpms-F	CATTCAAGGGCTATGAAGGTTCTC
Rbpms-R	GCCTTAGCAAACTCTAGTCGT
Baz2b-F	CTTAGTTCTACAGCCAGCCC
Baz2b-R	CCATTTATACCTTTCTCAGGACCA
Calcoco1-F	GATAAGATCCTGAAGCTGAGTG
Calcoco1-R	TCCAGCAGTTCCTGTTTCTC
Pbxip1-F	TCTCACCTGCTTACTTTGGA
Pbxip1-R	TTCTTTCGTGACCTCTTCTTCAG
Cbfa2t2-F	AAACACCTTGACCATGCACTG
Cbfa2t2-R	GCTGAGAATCGTTGCTGAGAG
Zfp292-F	CTTACCTTGTCTGTCTTTGCAC
Zfp292-R	GAGTGAGTTCCCAAGCACAG
Zfp827-F	TTTAAAGTGAAGGAGGAGCCCA
Zfp827-R	TTGTAGCTTTCCGCTGAGAG
Zfp945-F	AAACTTCCTTAAGACTCAGGTG
Zfp945-R	CTTGAATGTTAACAGTTCCTGGG
Chd6-F	TAAACATGTGGAACGACCTG
Chd6-R	TCTGAAAGGAATGCGATGGA

Table S2. Gene expression matrix of individual modules (see Excel file)

Table S3. Enriched GO terms and Reactome and KEGG pathways in individual modules (see Excel file)

Tables S4. Enriched cardiac phenotype- and disease-associated gene sets in individual modules (see Excel file)

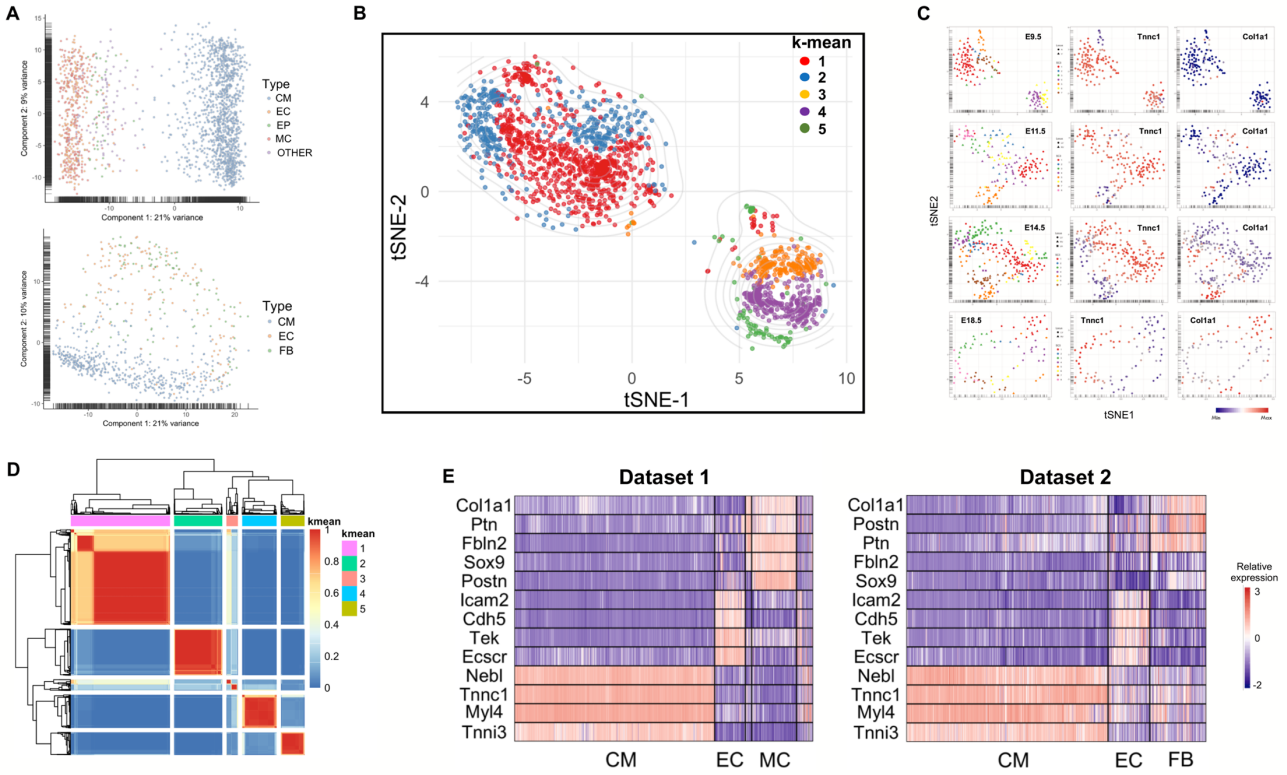
Table S5. Motif enrichment of 186 TFs (see Excel file)

Table S6. 186 TFs in the five selected modules and their predicted targets (see Excel file)

Table S7. Enrichments of TF targets in GO terms, pathways and cardiac phenotype- and disease-associated gene sets (see Excel file)

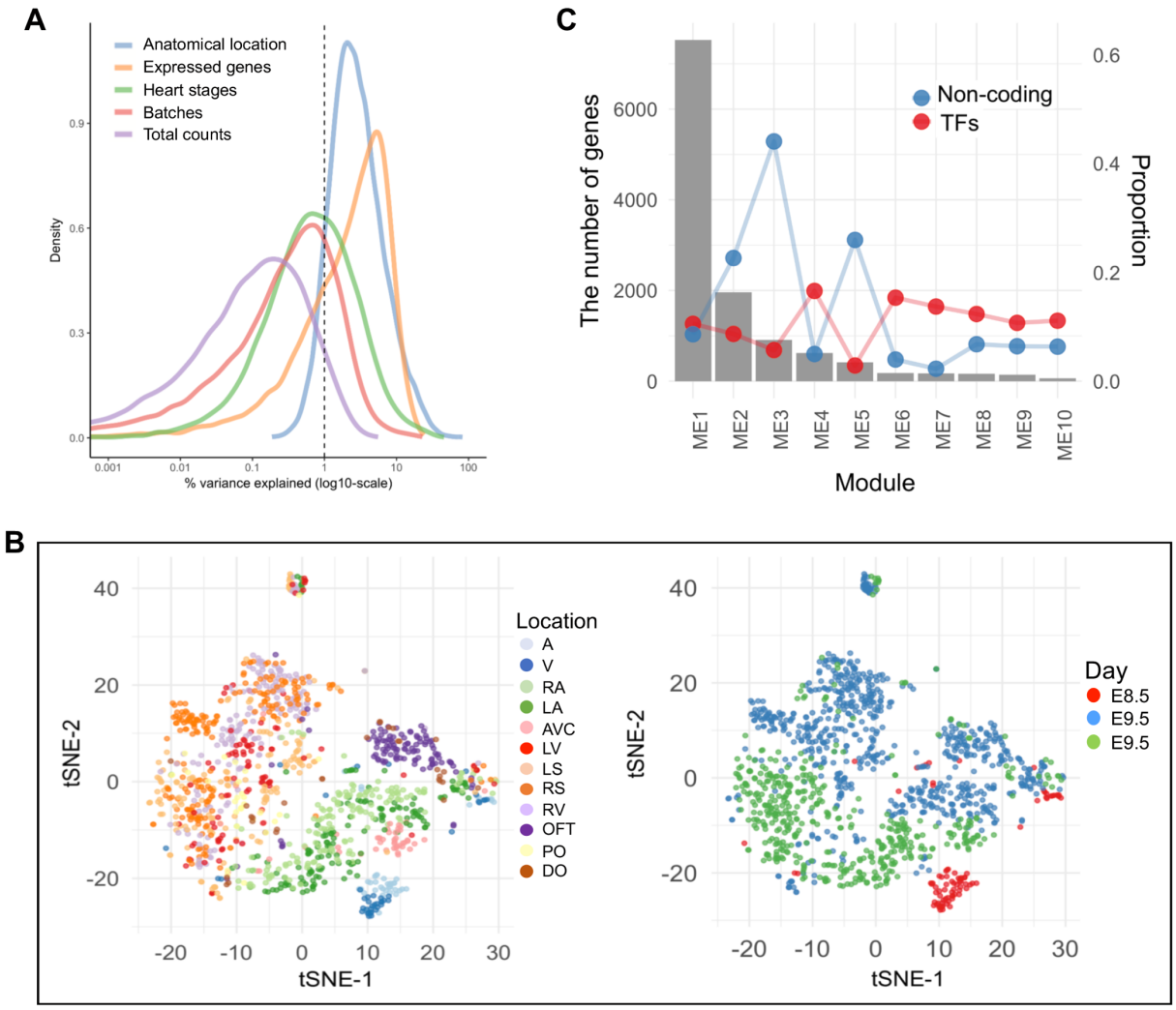
Table S8. DE genes of atrial, ventricular or OFT subgroups (see Excel file)

Figure S1. Clustering analysis of cardiac cell types in both datasets.



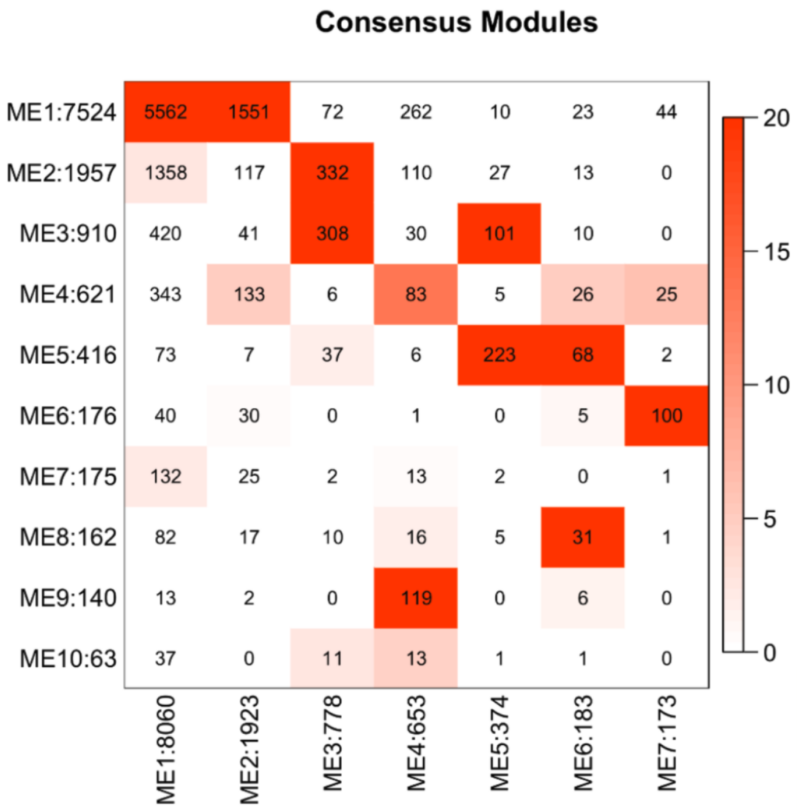
A. PCA (principal component analysis) of cells in dataset 1 and 2. **B,C**. Clustering of cardiac cells in dataset 1 and 2. B) shows t-SNE (t-distributed stochastic neighbor embedding) map of cardiac cells in dataset 1, while C) shows t-SNE maps with k-mean clustering of cardiac cells in dataset 2 at four developmental stages (E9.5 ~ 18.5), with expression pattern of selected marker genes. **D**. Transcriptional similarity of cells as revealed by k-mean clustering for dataset 1. **E**. Expression of marker genes used for identification of cardiomyocytes and other cell types for both datasets.

Figure S2. t-SNE map and module preservation of cardiomyocytes.



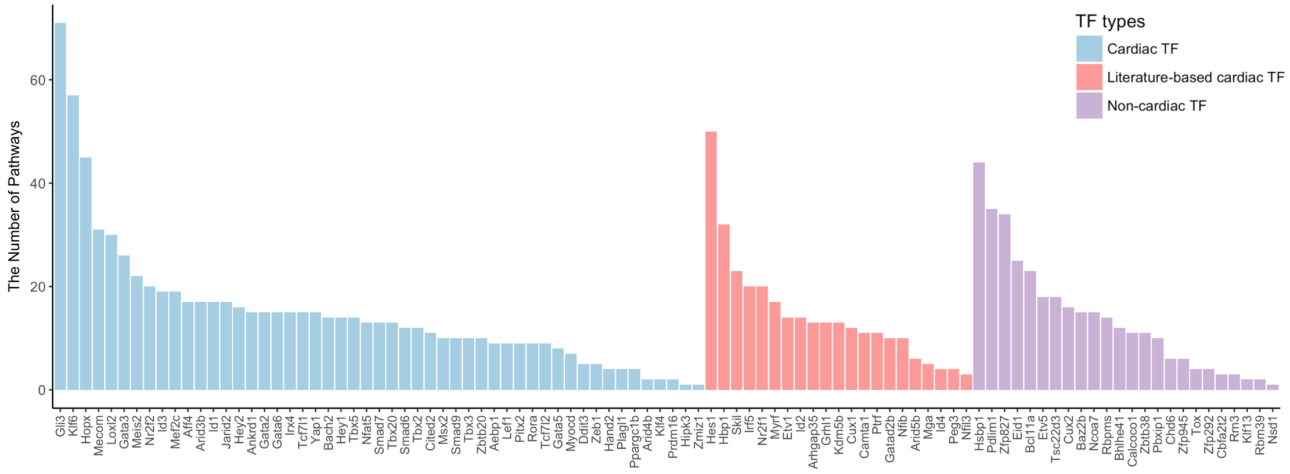
A. Gene expression variances in dataset 1 explained by different factors. **B**. The t-SNE (t-distributed stochastic neighbor embedding) map of CMs (cardiomyocytes) in dataset 1, with cells colored by anatomical locations (left panel) or stages (right panel). **C**. Gene numbers in individual modules. The left y-axis shows module sizes (ranging from 63 to 7,524), and the right y-axis indicates the proportions of TFs (transcription factors) and non-coding genes.

Figure S3. Module preservation.



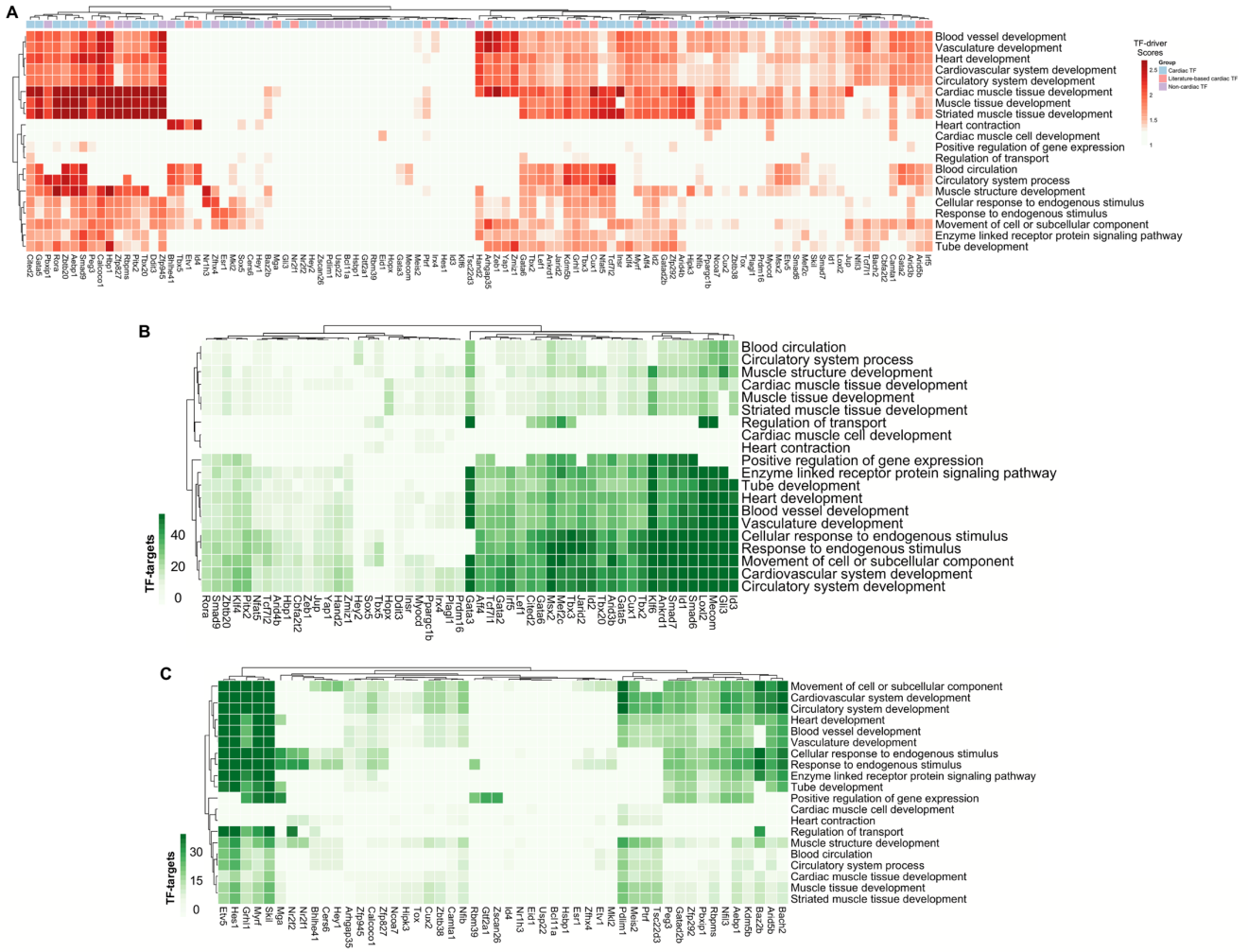
The y-axis shows module assignments in dataset 1, while x-axis shows consensus modules in dataset 1 and 2 combined, with each square indicates the numbers of overlapped genes. The color indicates the significance of overlaps.

Figure S4. Numbers of pathways regulated by candidate TFs.



The bar plot shows the numbers of enriched pathways (y-axis) under the regulations of cardiac, literature-based cardiac and non-cardiac TFs (transcription factors, the detailed version of Figure 6B).

Figure S5. TF-targets in enriched heart developmental pathways.



A. Heat-map displays the predicted regulations of the candidate TFs (transcription factors) in 20 representative heart terms enriched in the selected WGCNA (weighted gene co-expression network analysis) modules (the detailed version of Figure 6C). The colors in heat-map shows the TF-driver scores for the functional terms, representing the regulatory effects of individual TFs on the terms. **B**. Top enriched cardiac pathways regulated by cardiac TFs, with color indicating the number of TF targets. **C**. The numbers of genes in the (**B**) pathways that were predicted to be targets of literature-based cardiac or non-cardiac TFs.

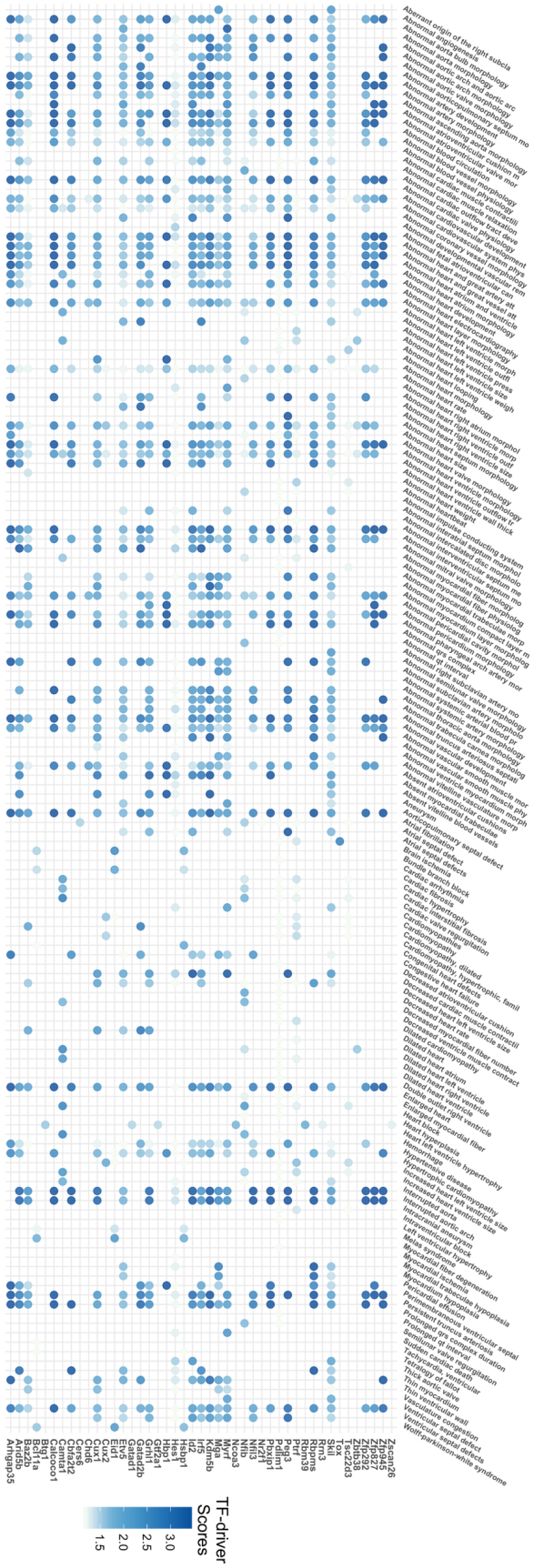
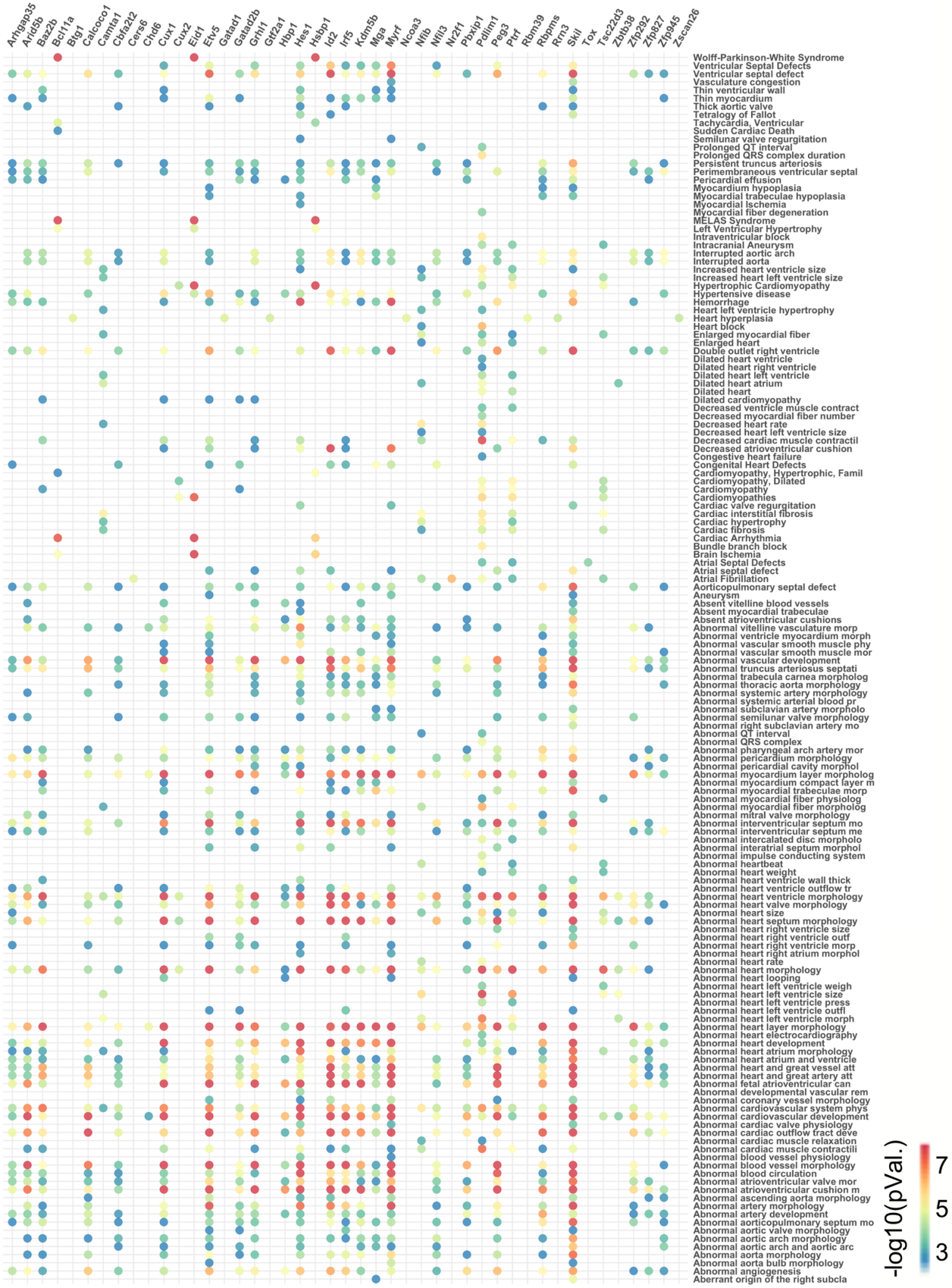


Figure S6. TF-targets in enriched cardiac disease-associated gene sets.

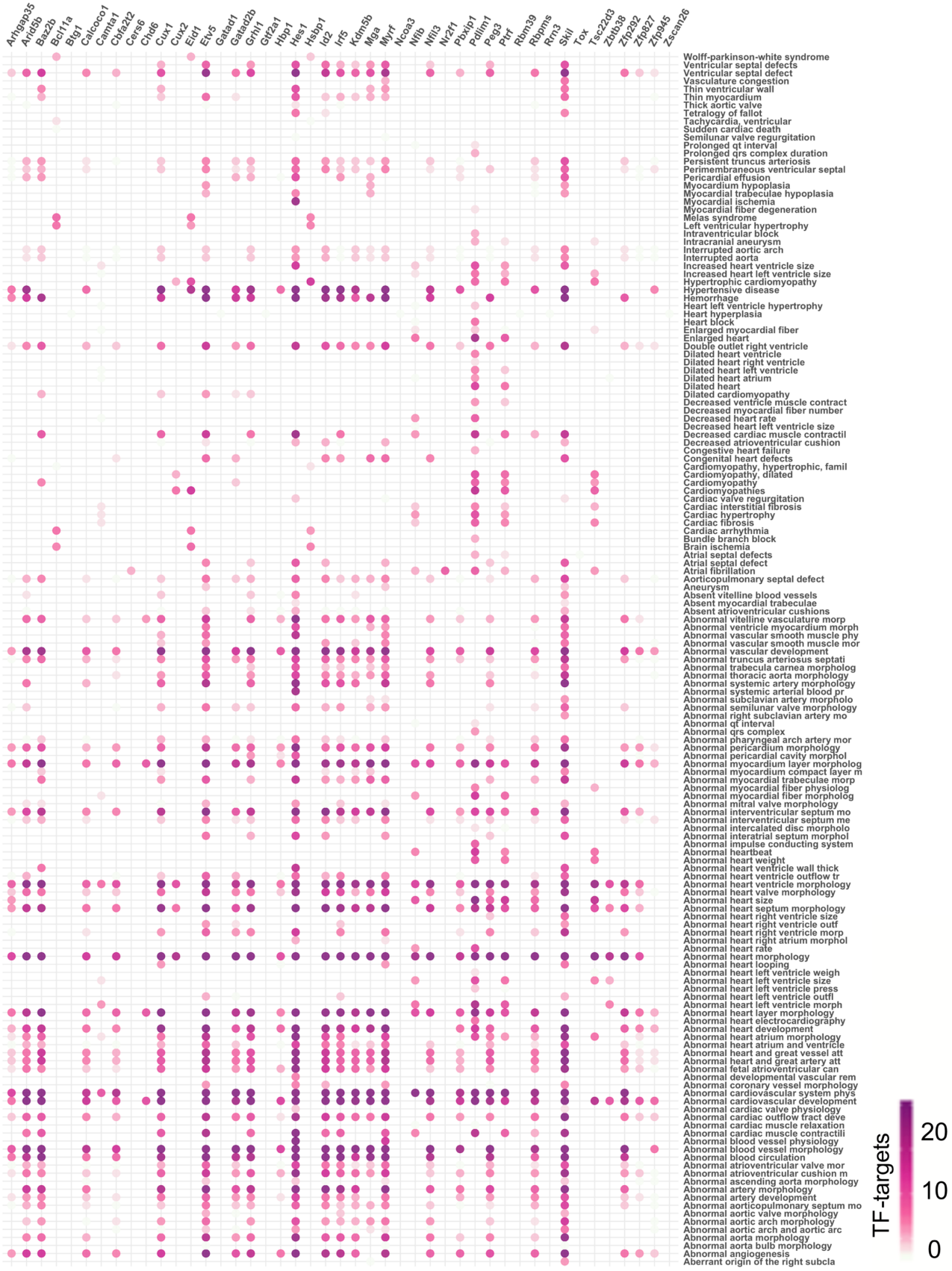
Heat-map shows the detailed version of Figure 6D, and displays the predicted roles of all the literature-based cardiac and non-cardiac TFs (transcription factors), measured as their TF-driver scores (colors) for different heart phenotype- or disease-associated gene sets.

Figure S7. Enrichment analysis of TF-targets in cardiac disease-associated gene sets.



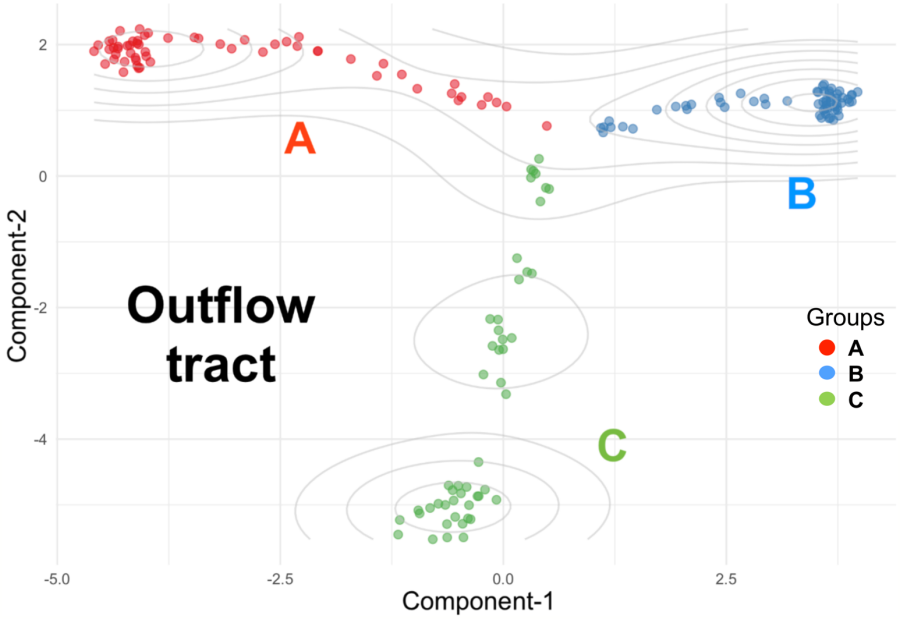
Heat-map shows the enrichment of the targets of literature-based cardiac or non-cardiac TFs (transcription factors) in different heart phenotype- or disease-associated gene sets, with color for enrichment significance.

Figure S8. The number of TF-targets in cardiac disease-associated gene sets.



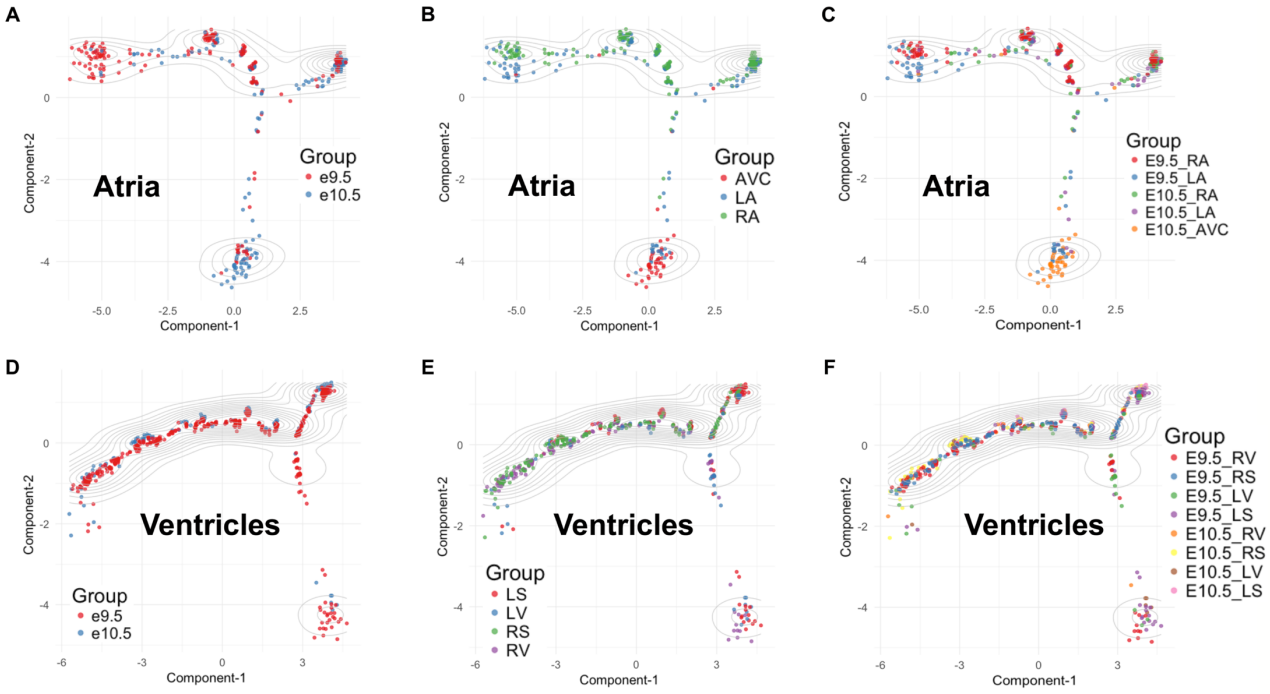
Heat-map shows the number of targets of literature-based cardiac or non-cardiac TFs (transcription factors) enriched in different heart phenotype- or disease-associated gene sets that are shown in Figure S7, with color for the number of targets.

Figure S9. A trajectory of CMs at outflow tract.



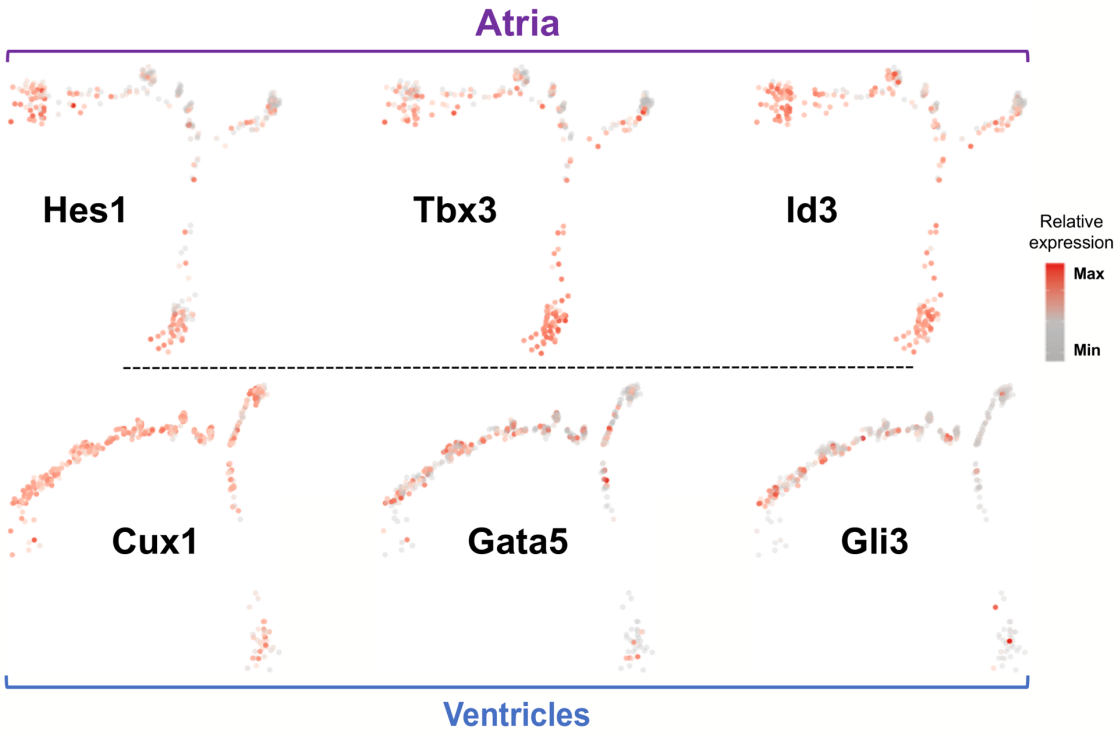
The trajectory indicates three groups (A, B and C) of CMs (cardiomyocytes) at outflow tract, with colors for different groups.

Figure S10. Trajectories of atrial and ventricular CMs (cardiomyocytes).



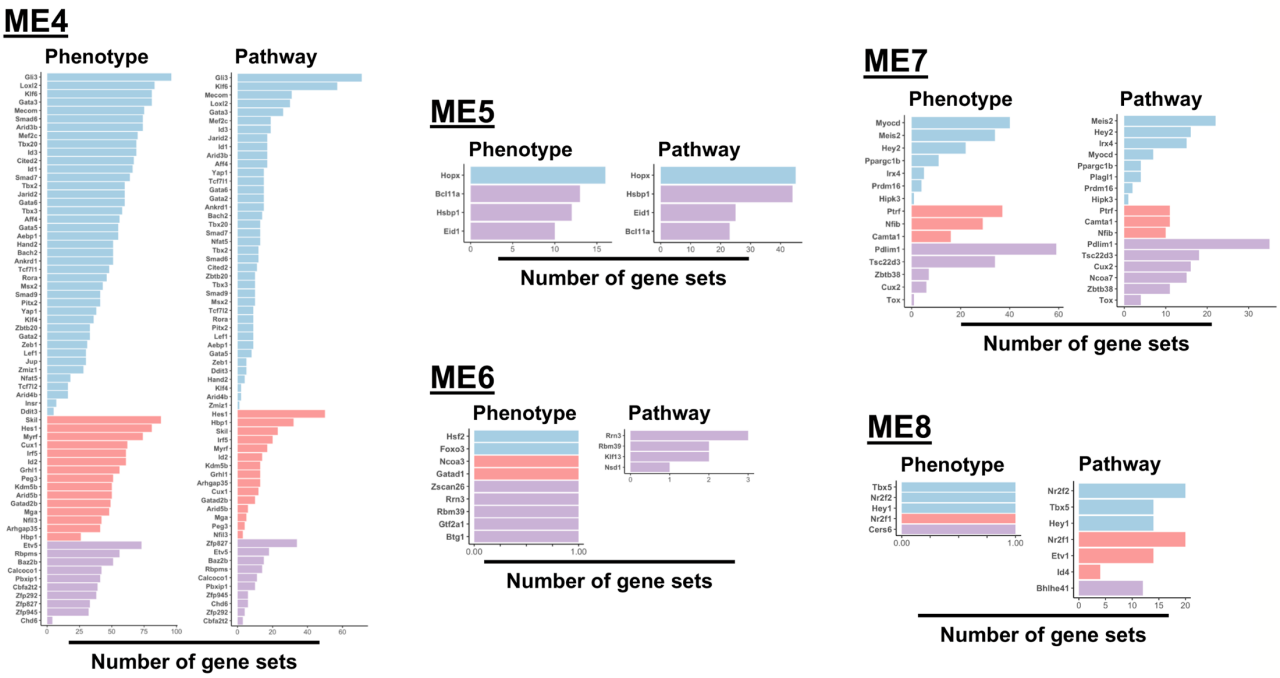
The atrial trajectory, with color indicating cells at different heart stages (**A**), at anatomical locations (**B**), or by spatiotemporal factors (**C**). The ventricular trajectory, with color by heart stages (**D**), anatomical locations (**E**), or spatiotemporal factors (**F**).

Figure S11. Expression patterns of representative atrial and ventricular TFs.



The plots display expression patterns of three representative DE (differentially expressed) TFs (transcription factors) of atrial subgroups or ventricular subgroups, according to Figure 8F.

Figure S12. Summary of the full list of the spatiotemporal TFs.



The bar-plot displays the number of the enriched gene sets under the regulation of the candidate TFs (transcription factors): cardiac, literature-based cardiac and non-cardiac, in the five selected modules (the detailed version of Figure 9).