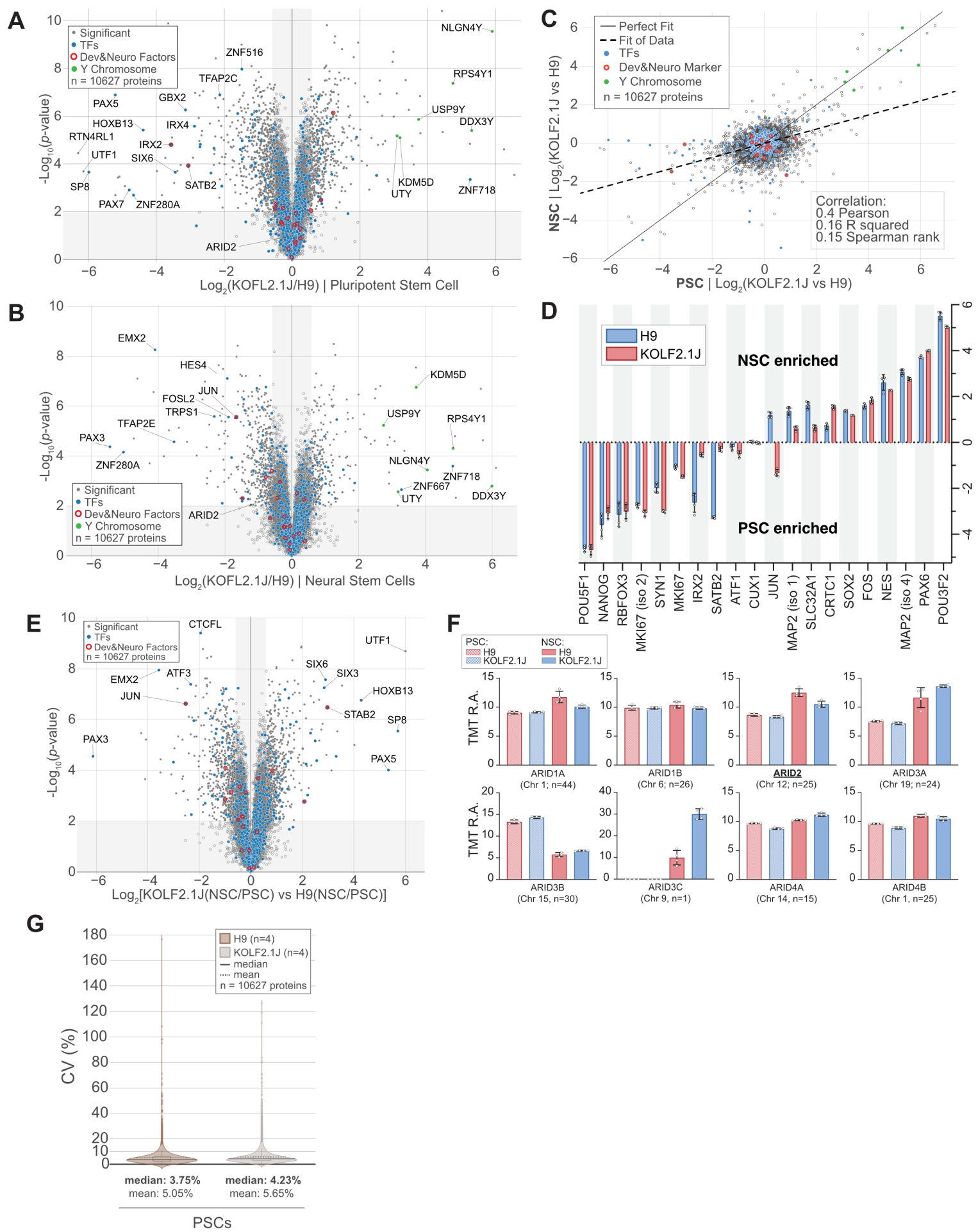


**Supporting Information:**  
**Nam et al (2022)**

**Figure S1**



**Figure S1. Quantitative comparison of H9 and KOLF2.1 proteome remodeling upon Neural Stem Cell differentiation.** Related to Figure 1

(A) Volcano plot shows the pluripotent stem cells  $-\log_{10}$ -transformed  $p$ -value versus the  $\log_2$ -transformed ratio of KOLF2.1J/H9 for quantification.  $p$ -values were calculated by two-sided Welch's t-test (adjusted for multiple comparisons); for parameters, individual  $p$ -values and  $q$ -values, see **Supplementary Table 1**. Individual proteins are shown in gray open circles, significantly changed protein: filled gray circle, transcription factors: filled blue circles, and developmental & neuronal markers: red open circles; 10627 proteins were quantified.

(B) Volcano plot shows the neural stem cells  $-\log_{10}$ -transformed  $p$ -value versus the  $\log_2$ -transformed ratio of KOLF2.1J/H9 for quantification.  $p$ -values were calculated by two-sided Welch's t-test (adjusted for multiple comparisons); for parameters, individual  $p$ -values and  $q$ -values, see **Supplementary Table 1**. Individual proteins are shown in gray open circles, significantly changed protein: filled gray circle, transcription factors: filled blue circles, and developmental & neuronal markers: red open circles; 10627 proteins were quantified.

(C) Proteome level correlation plot of  $\log_2(\text{KOLF2.1J}/\text{H9})$  from neural stem cell (y axis) or pluripotent stem cell (x axis) upon differentiation. The calculated correlation factors (Pearson, R squared, and Spearman rank) are indicated.

(D) Plot of relative abundance with marker proteins enriched in pluripotent stem cell (PSC) or neural stem cell (NSC) of either cell lines.

(E) Volcano plot shows the  $-\log_{10}$ -transformed  $p$ -value versus the  $\log_2$ -transformed ratio of  $\text{KOLF2.1J}(\text{NSC}/\text{PSC})/\text{H9}(\text{NSC}/\text{PSC})$  for quantification.  $p$ -values were calculated by two-sided Welch's t-test (adjusted for multiple comparisons); for parameters, individual  $p$ -values and  $q$ -values, see **Supplementary Table 1**. Individual proteins are shown in gray open circles, significantly changed protein: filled gray circle, transcription factors: filled blue circles, and developmental & neuronal markers: red open circles; 10627 proteins were quantified.

(F) Individual relative abundance of the ARID family of DNA binding proteins. Individual quantified proteins are shown in a bar graph representing their TMT relative abundance (R.A.) in the two cell lines (H9 and KOLF2.1J) and both cell types (PSC and NSC). Chromosome location (Chr) and number of quantified peptides ( $n$ ) is indicated for each protein.

(G) Violin plots representing the coefficient of variation across biological replicates for both cell lines: center line, median; center dotted line, mean; box limits correspond to the first and third quartiles; whiskers, 1.5x interquartile range; outliers shown as empty circles. Computed CVs showed 5% or less variation across samples.



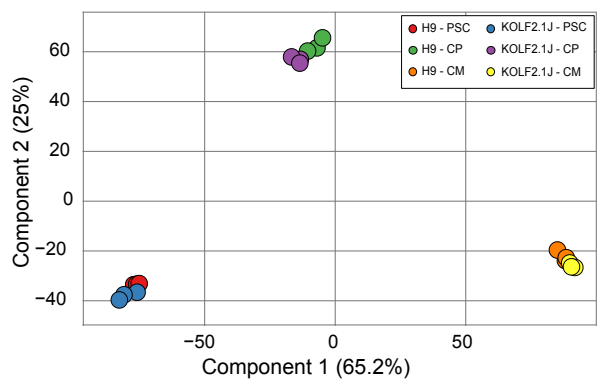
**Figure S2. Quantitative phosphoproteome analysis of H9 and KOLF2.1 upon Neural Stem Cell differentiation.** Related to Figure 2

(A) MeanRank score from kinase enrichment analysis for H9 down-phosphorylated proteins in NSC to PSC comparison ( $\text{Log}_2 < -1$ ,  $p$ -value  $< 0.01$ ).

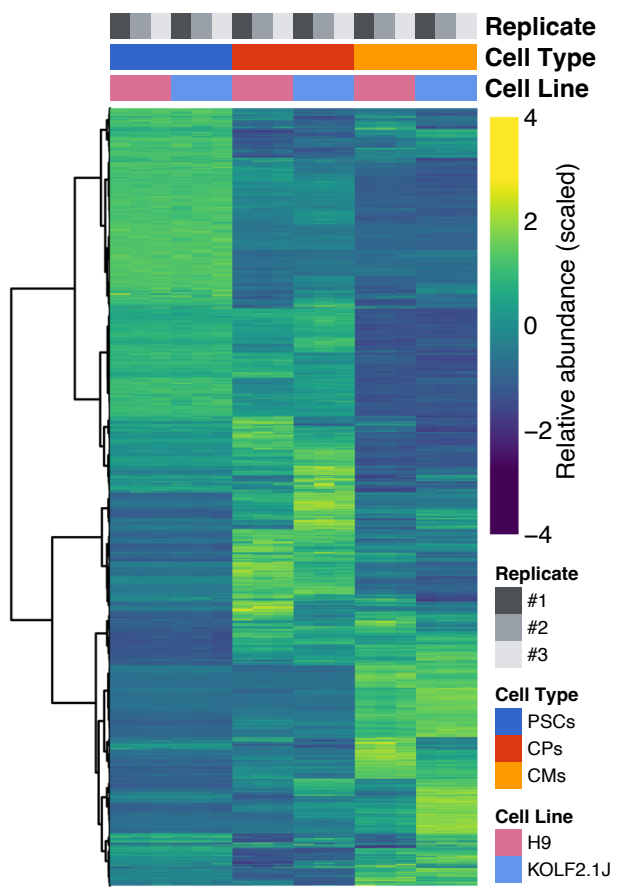
(B) MeanRank score from kinase enrichment analysis for KOLF2.1J down-phosphorylated proteins in NSC to PSC comparison ( $\text{Log}_2 < -1$ ,  $p$ -value  $< 0.01$ ).

**Figure S3**

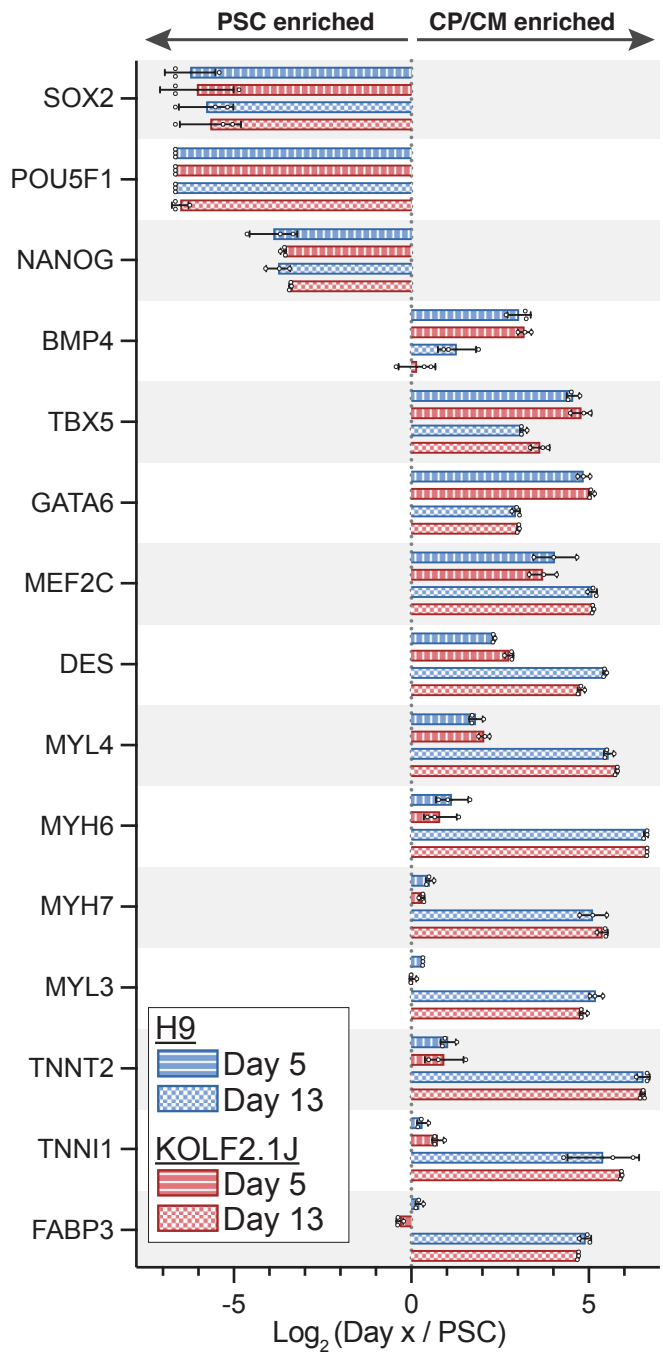
**A**



**B**



**C**



**Figure S3. Quantitative proteome remodeling analysis upon cardiomyogenesis.** Related to Figure 3.

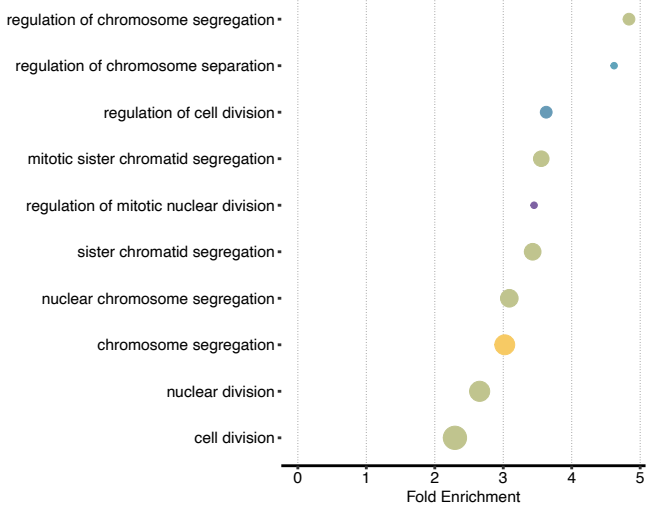
(A) Principal component analysis (PCA) of proteomics data obtained from 18plex TMTpro based analysis. The inset indicates color coding for individual cells (PSC, Pluripotent stem cell; CP, Cardiac progenitors; CM, Cardiomyocytes).

(B) Heatmap of protein's relative abundance during the cardiac differentiation. Hierarchical clustering heatmap and associated dendrogram of the three time points was computed using Euclidean distance and Ward linkage on z-transformed scaled TMT abundance values.

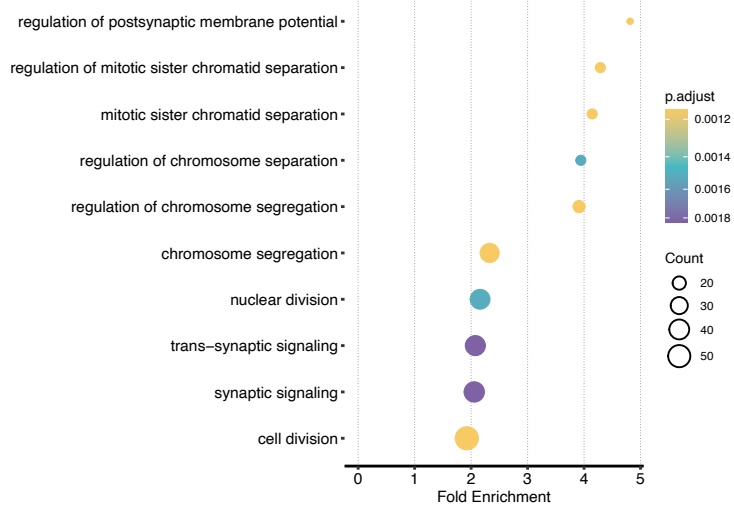
(C) Plot of relative abundance for both cell line, with protein markers enriched in day 5 cardiac progenitor (CP) state or day 13 cardiomyocytes (CM) state, compared to day 0 pluripotent stem cell (PSC) state.

**Figure S4**

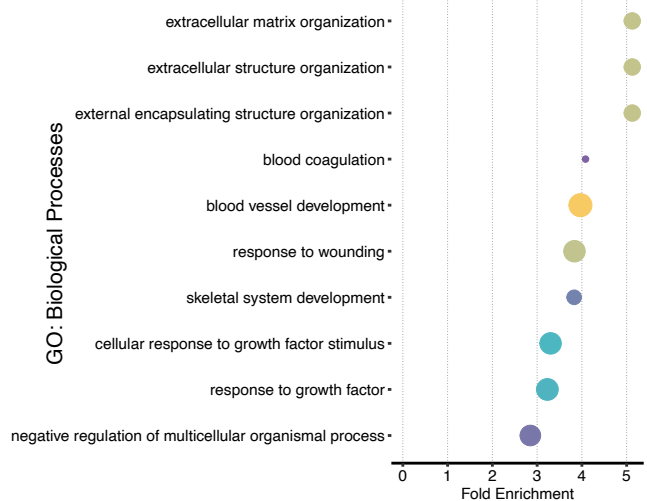
**A** H9 - Cluster 8



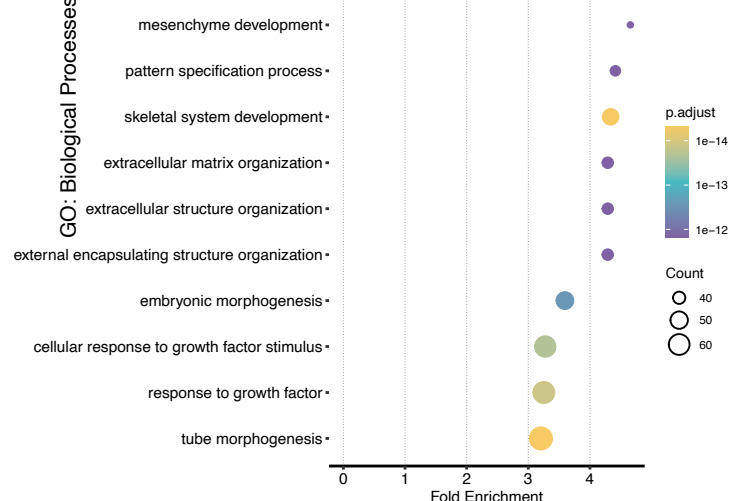
**B** KOLF2.1J - Cluster 8



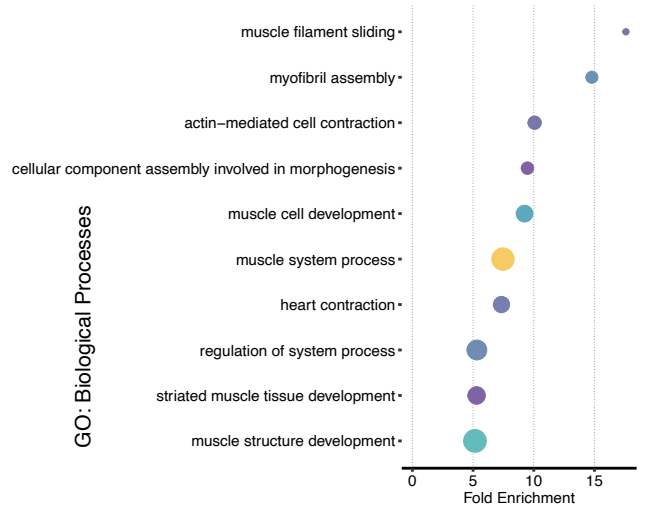
**C** H9 - Cluster 6



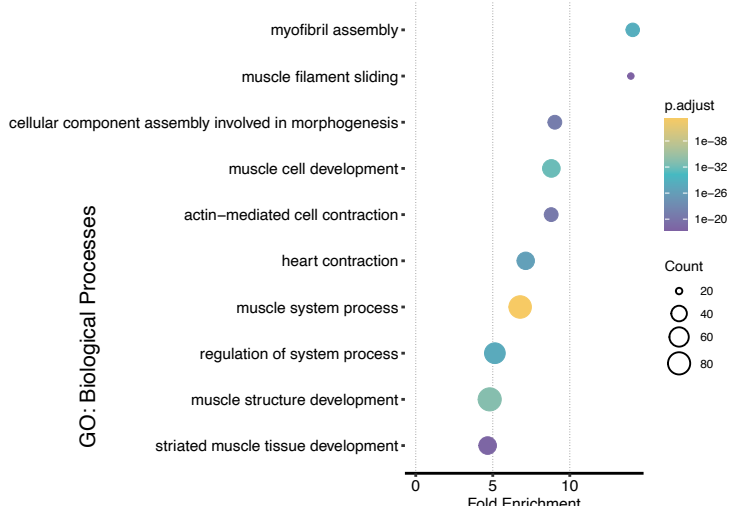
**D** KOLF2.1J - Cluster 6



**E** H9 - Cluster 1



**F** KOLF2.1J - Cluster 1





**Figure S4. Gene ontology enrichment analysis of temporal clusters upon cardiomyogenesis.**

Related to Figure 4.

- (A) The enriched gene ontology (GO) terms for proteins in cluster 1 in H9 pluripotent stem cells.
- (B) Similar to (A) but for KOLF2.1J dataset.
- (C) The enriched gene ontology (GO) terms for proteins in cluster 6 in H9 pluripotent stem cells.
- (D) Similar to (C) but for KOLF2.1J dataset.
- (E) The enriched gene ontology (GO) terms for proteins in cluster 8 in H9 pluripotent stem cells.
- (F) Similar to (E) but for KOLF2.1J dataset.