**Description of Supplementary Material Files**

**Supplementary Figures:**

**Supplementary Figure 1.** Distribution of the label-free data set after median sweep normalization.

**Supplementary Figure 2.** Graphic Summary of Label-Free pTyr Site Profiling on Major Myofilaments Proteins.

**Supplementary Figure 3.** Graphic Summary of Label-Free pTyr Site Profiling in titin.

**Supplementary Figure 4.** Canonical pathways upregulated and downregulated according to Ingenuity Pathway Analysis of the pTyr perturbation.

**Supplementary Figure 5.** Distribution of the protein expression intensities if the TMT-labeled MS/MS proteomics before and after median sweep normalization.

**Supplementary Figure 6.** Distribution of the TMT phosphoproteome intensities data set before and after median sweep normalization.

**Supplementary Table 1**: TMT proteomics myofilament subproteome signal and statistical analysis.

**Supplementary Table 2**: TMT proteomics myofilament subproteome phosphotyrosine signal and statistical analysis.

**Supplementary Table 3**: TMT proteomics myofilament subproteome phosphotyrosine signal normalized to total protein levels and statistical analysis.

**Supplementary Table 4**: Label-free proteomics, phosphoproteome quantification.

**Supplementary Table 5**: Label-free proteomics, data filtered and missing values imputation (k-means).

**Supplementary Table 6**: Label-free proteomics, TgErbB2 vs Ntg phosphotyrosine statistical analysis.

**Supplementary Table 7**: Label-free proteomics, R403Q-αMyHC vs Ntg phosphotyrosine statistical analysis.

**Supplementary Table 8**: IPA on TgErbB2 label-free proteomics data.

**Supplementary Table 9**: IPA on R403Q-αMyHC label-free proteomics data.

**Supplementary Table 10:** Kinase Substrate Enrichment Analysis & MoBaS Analysis.

**Supplementary Table 11:** Contractility and LV chamber dimensions by motion mode and tissue Doppler echocardiography.

**Supplementary Table 12**: IPA on TgErbB2 TMT proteomics data.