Electronic Supplementary Material (ESI) for Molecular Omics.

Supporting Information

Rbm20 ablation is associated with changes in the expression of titin-interacting and metabolic proteins

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Table S1. Number of protein groups identified across eight biological replicates of WT rat hearts. A total of 2425 protein group identifications were shared across all eight biological replicates.

Replicate	Protein Groups
WT 1	2644
WT 2	2654
WT 3	2672
WT 4	2645
WT 5	2685
WT 6	2627
WT 7	2670
WT 8	2635
Total Shared	2425

Table S2. Number of protein groups identified across eight biological replicates of RBM20 KO rat hearts. A total of 2379 protein group identifications were shared across all eight biological replicates.

Replicate	Protein Groups
KO 1	2583
KO 2	2647
KO 3	2588
KO 4	2633
KO 5	2655
KO 6	2656
KO 7	2657
KO 8	2619
Total Shared	2379

Table S3. Differentially expressed genes in the Rbm20 KO rat myocardium with expression quantified at both the protein and transcript levels.[1]

				RNAseq		
Gene name	**UniProt ID**	**Protein names**	**Proteomics (d21)**	**d1**	**d20**	**d49**
Ankrd1	Q8R560	Ankyrin repeat domain-containing protein 1	↑	-	-	↑
Casq1	P19633	Calsequestrin-1	↓	-	↑	↑
Ech1	Q62651	Delta(3,5)-Delta(2,4)-dienoyl-CoA isomerase, mitochondrial	↓	↓	-	-
Fhl1	Q9WUH4	Four and a half LIM domains protein 1	↑	↑	↑	↑
Gamt	P10868	Guanidinoacetate N-methyltransferase	↓	-	-	↓
Hmgcs2	P22791	Hydroxymethylglutaryl-CoA synthase, mitochondrial	↓	↓	↓	-
Igsf1	Q925N6	Immunoglobulin superfamily member 1	↓	-	↓	-
Maoa	P21396	Amine oxidase [flavin-containing] A	↑	-	-	↑
Msrb2	Q4FZX5	Methionine-R-sulfoxide reductase B2, mitochondrial	↑	-	↑	↑
Myh7	P02564	Myosin-7	↓	↑	↑	↑
Penk	P04094	Proenkephalin-A	↑	-	↑	↑
Tnni1	P13413	Troponin I, slow skeletal muscle	↓	-	↑	-

Up and down arrows denote proteins with significantly up- and down-regulated expression, respectively, while "-" denotes that expression was not changed significantly between WT and KO.

Table S4. Differentially expressed mitochondrial (GO: 0005739) genes in Rbm20 KO rat myocardium as determined by proteomics and previously by RNAseq.[1]

				RNAseq		
Gene name	**UniProt ID**	**Protein names**	**Proteomics (d21)**	**d1**	**d20**	**d49**
Acadsb	P70584	Short/branched chain specific acyl-CoA dehydrogenase, mitochondrial	↑	N/A	N/A	N/A
Akr1b7	Q5RJP0	Aldose reductase-related protein 1	↓	N/A	N/A	N/A
Ccnb1	P30277	G2/mitotic-specific cyclin-B1	N/A	-	↑	-
Dguok	A0A0G2K478	Deoxyguanosine kinase	N/A	↓	↑	-
Fundc1	Q5BJS4	FUN14 domain-containing protein 1	↓	N/A	N/A	N/A
Hmgcs2	P22791	Hydroxymethylglutaryl-CoA synthase, mitochondrial	↓	↓	↓	-
Nif3l1	Q4V7D6	NIF3-like protein 1	↓	N/A	N/A	N/A
Nit2	Q497B0	Omega-amidase NIT2	↓	N/A	N/A	N/A
Oxct1	B2GV06	Succinyl-CoA:3-ketoacid coenzyme A transferase 1, mitochondrial	↑	N/A	N/A	N/A
Pdk4	O54937	[Pyruvate dehydrogenase (acetyl-transferring)] kinase isozyme 4, mitochondrial	↓	N/A	N/A	N/A
Tomm20	Q62760	Mitochondrial import receptor subunit TOM20 homolog	↑	N/A	N/A	N/A
Trub1	Q5M934	Pseudouridylate synthase TRUB1	N/A	-	↑	-

Up and down arrows denote proteins with significantly up- and down-regulated expression, respectively, while "-" denotes that expression was not changed significantly between WT and KO. N/A indicates that the protein/transcript was not detected.

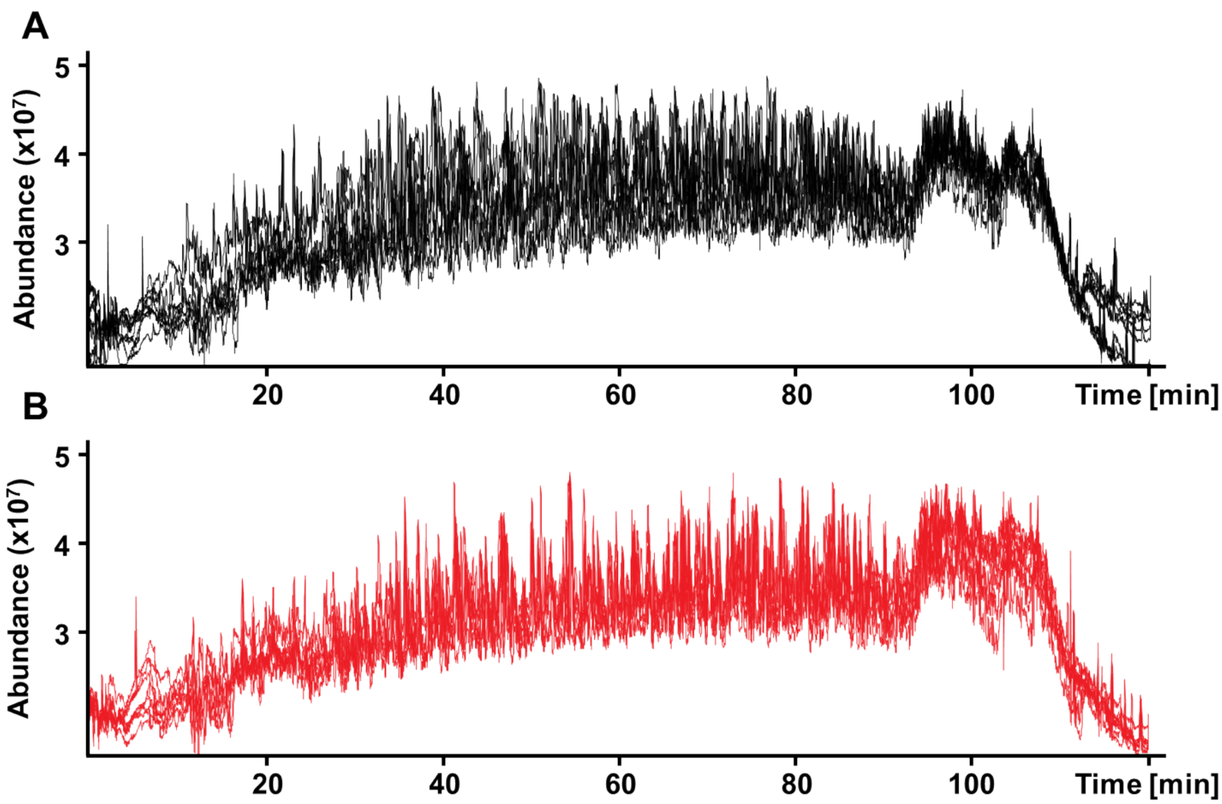

Figure S1. Overlay of total ion chromatograms from eight WT (A) and eight RBM20 KO (B) biological replicates. Excellent reproducibility between biological replicates was observed.

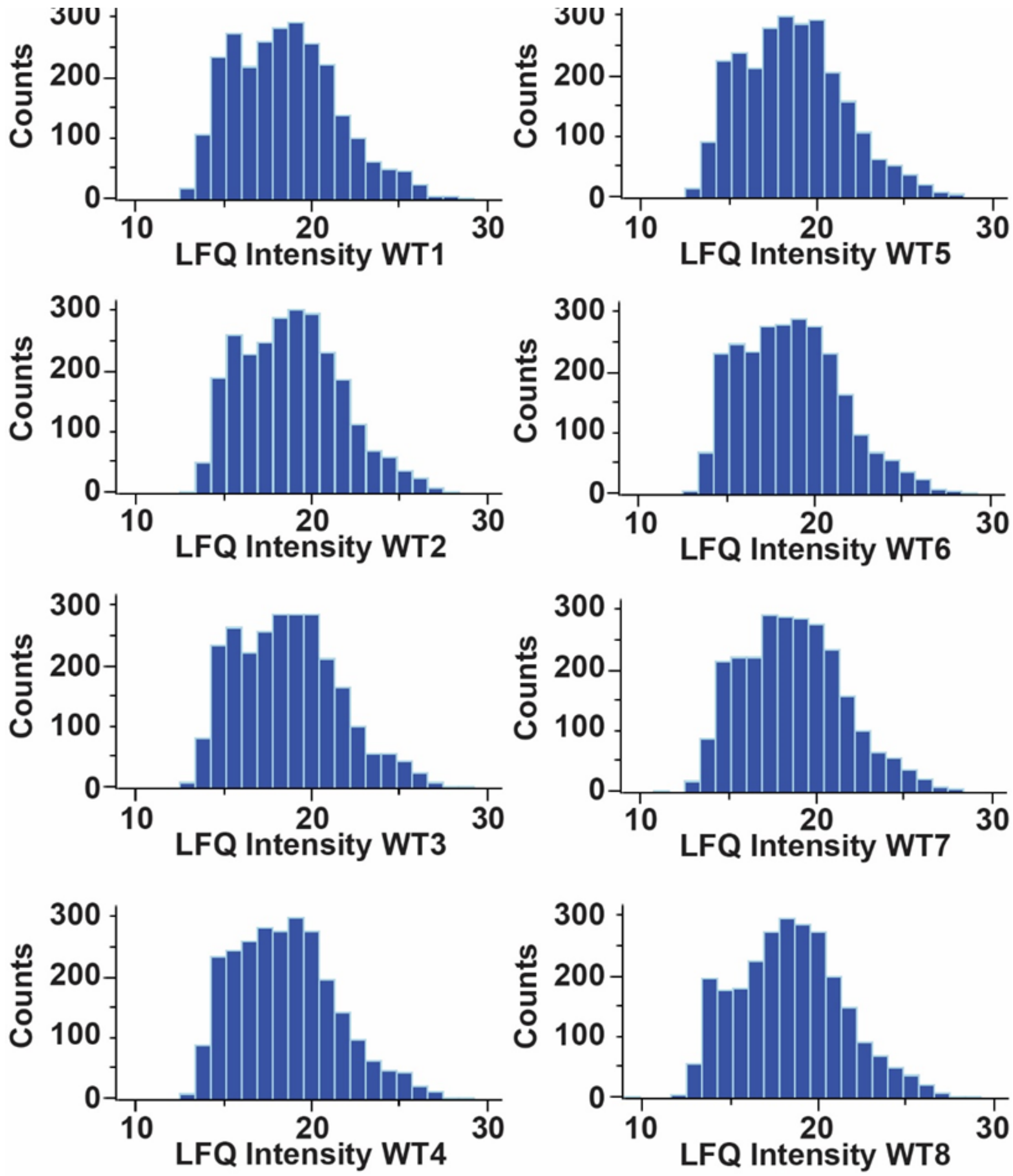

Figure S2. Histograms showing $\log_2$ transformed peptide LFQ intensities for identified proteins binned by intensity range for the eight WT rat heart biological replicates.

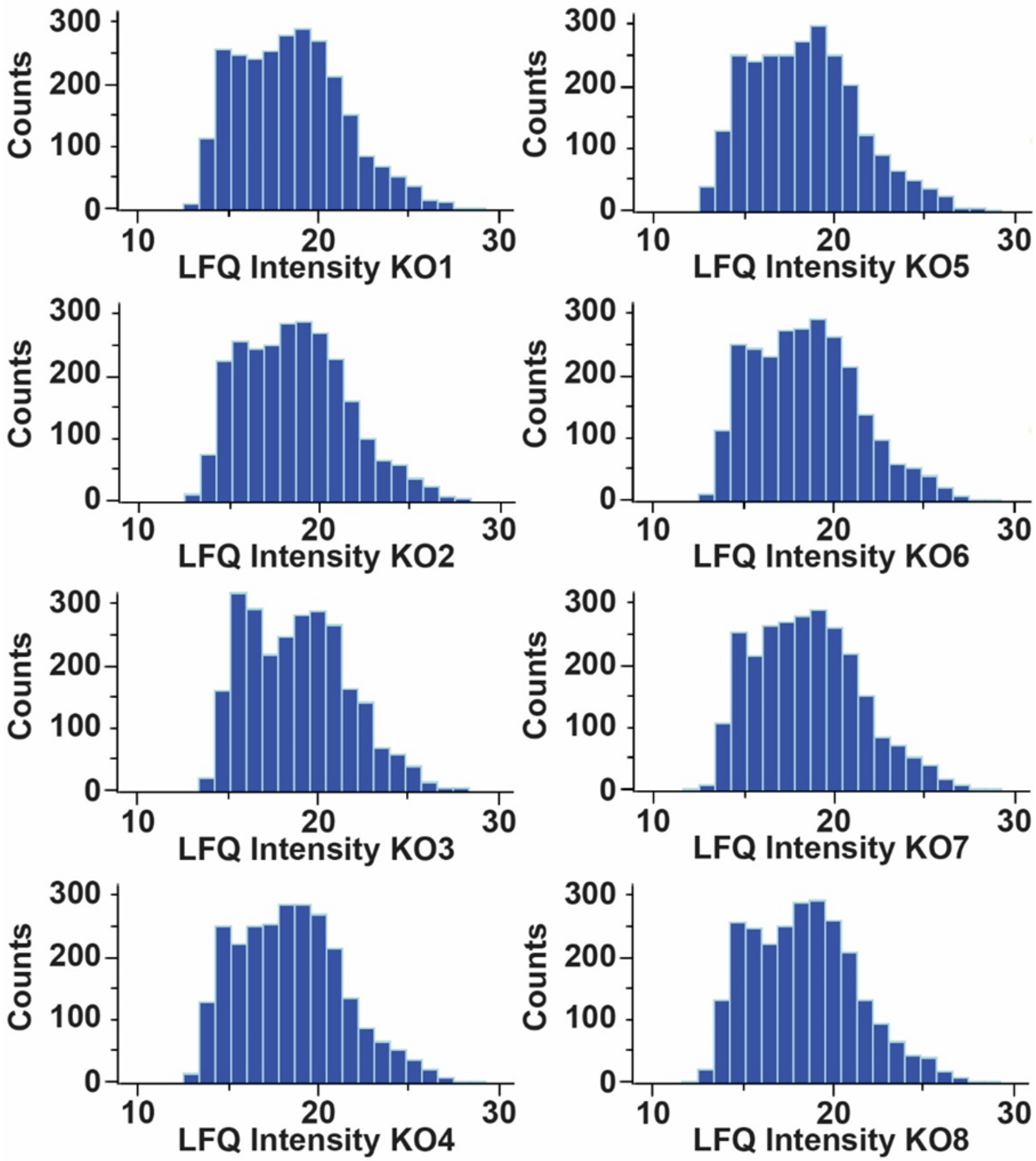

Figure S3. Histograms showing $\log_2$ transformed peptide LFQ intensities for identified proteins binned by intensity range for the eight RBM20 KO rat heart biological replicates.

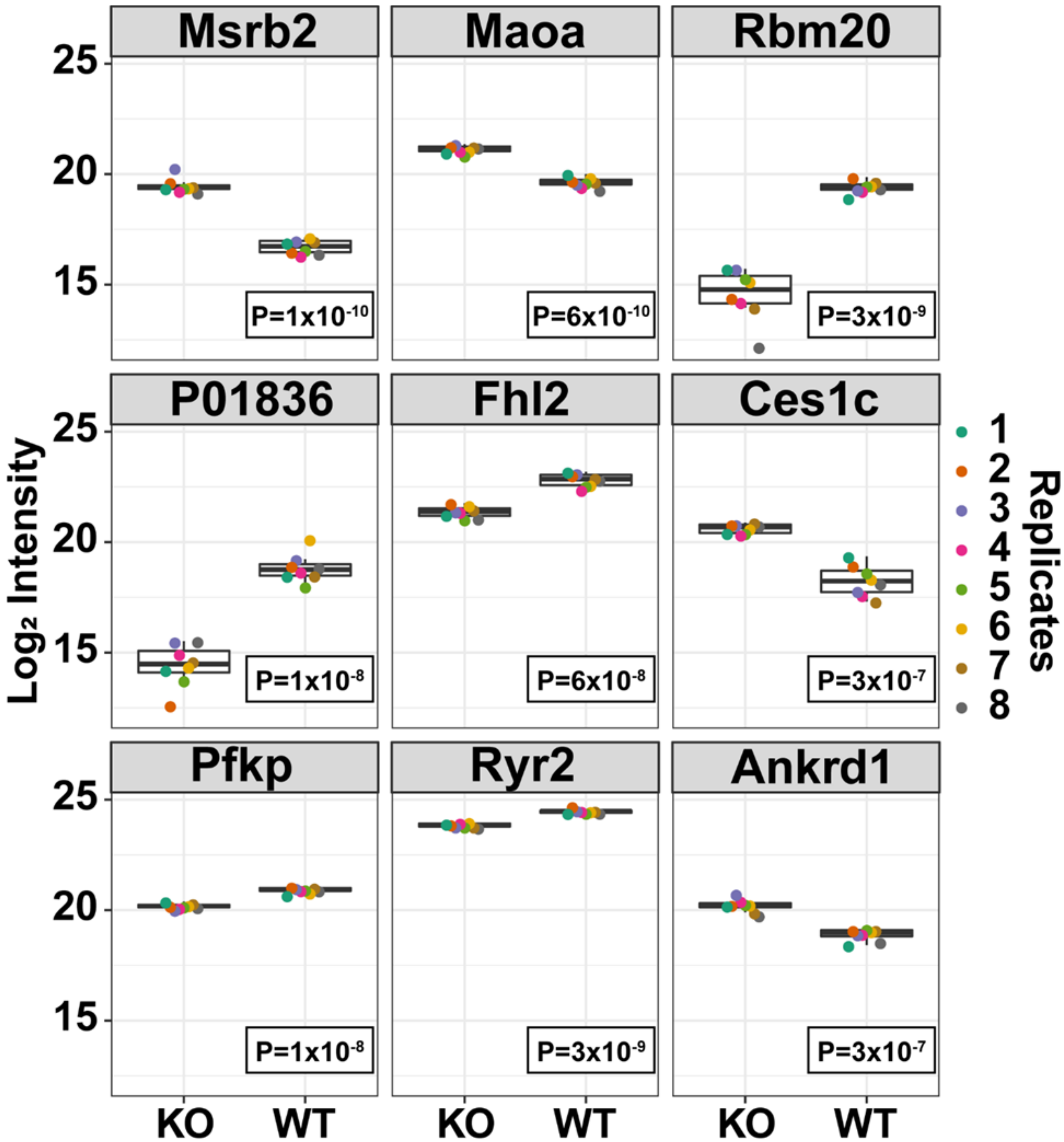

Figure S4. Boxplots of the WT and KO expression levels, shown as LFQ intensities, of differentially expressed proteins with the nine highest $-\log_{10}$ P values. Expression of proteins not detected in a given sample group, such as RBM20, which was not detected in any KO samples, were generated by imputation using Perseus.

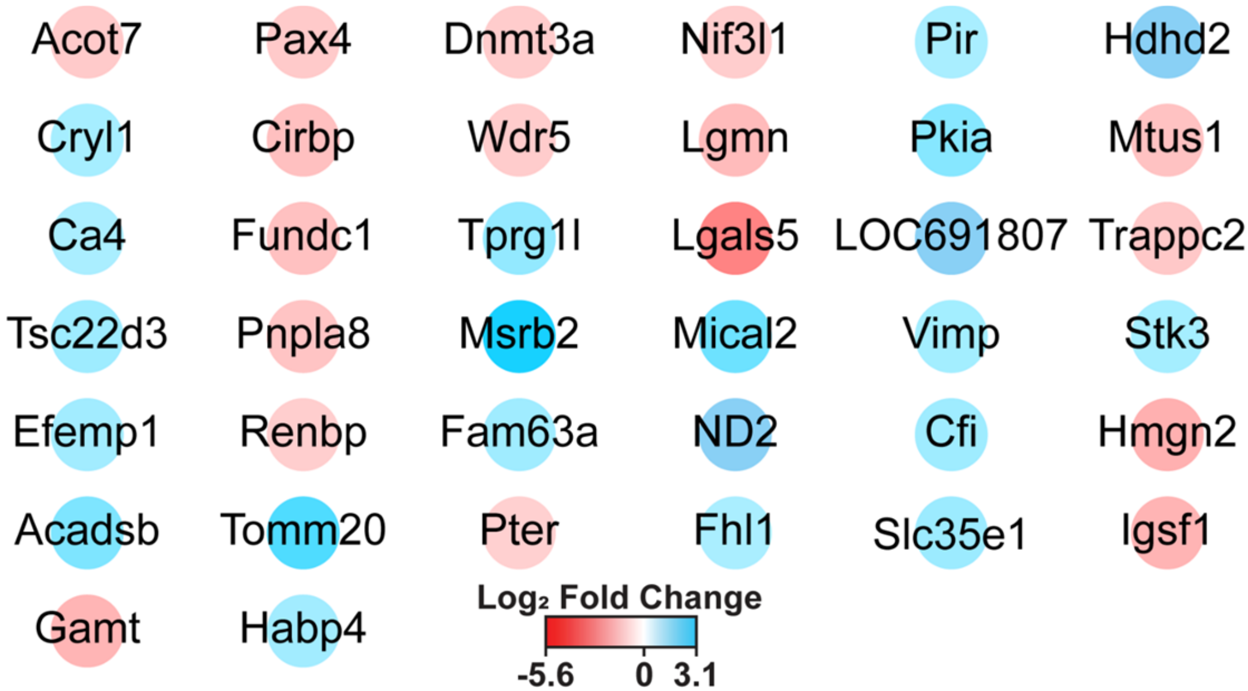

Figure S5. Identified proteins expressed significantly differently in RBM20 KO rats with no known interactors in the 103 DEPs, color coded by Log_2 fold-change in expression.