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Supplemental Information

Rbf Activates the Myogenic Transcriptional Program to Promote Skeletal Muscle Differentiation

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SUPPLEMENTAL INFORMATION

SUPPLEMENTAL FIGURES:

Figure S1

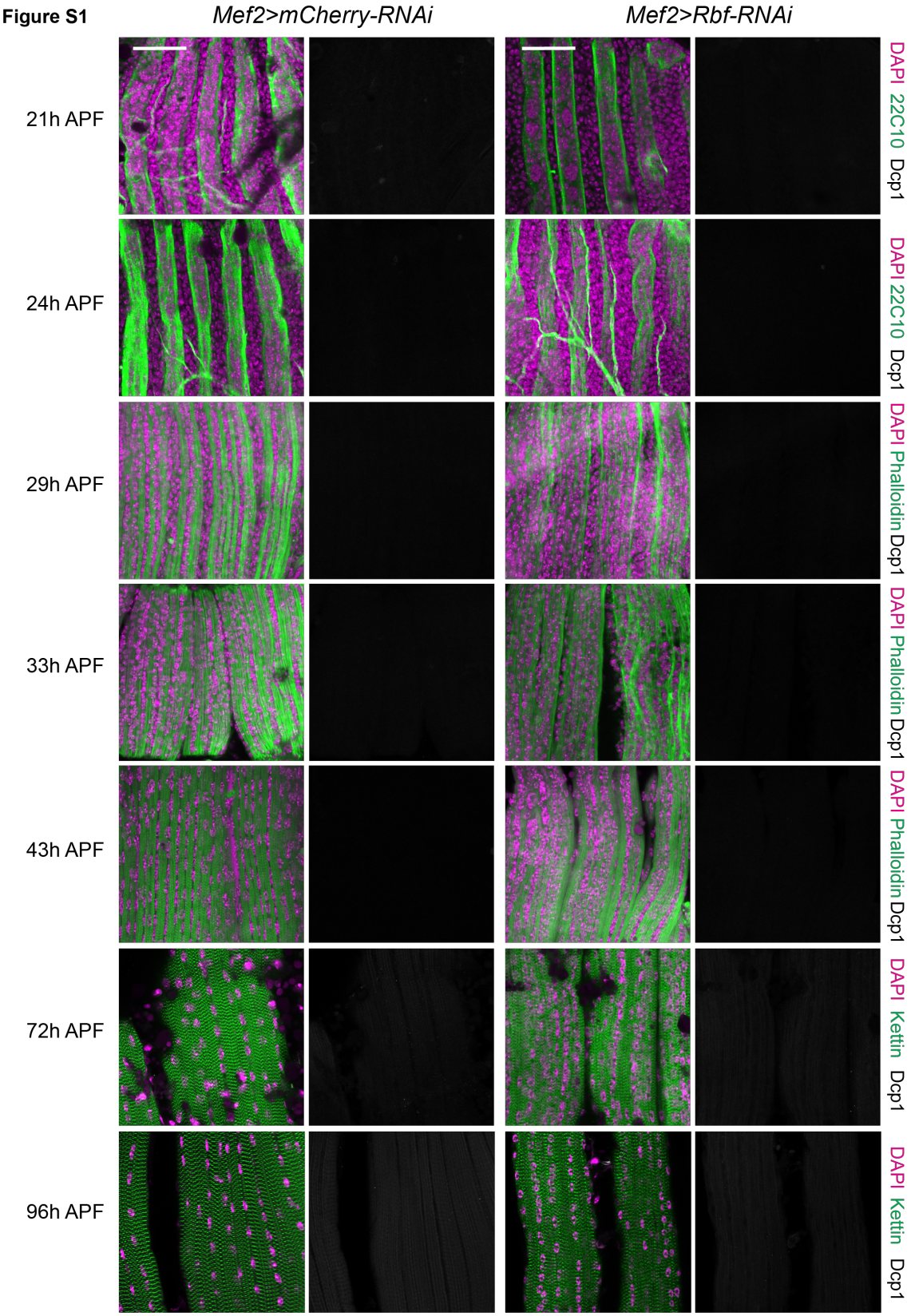

Figure S1: The loss of Rbf does not induce apoptosis mediated by dcp1 in developing indirect flight muscles. Related to Figure 2.

Developing DLMs stained over time from 21 to 96 h APF. Apoptotic cells were marked using anti-dcp1 antibody (right panel). Overall structure was analyzed using DAPI and Phalloidin or 22c10 to mark developing IFM or Kettin to stain sarcomere depending on the developmental stages (left panel). Single confocal sections are shown. Scale 40 μm. Full genotypes are *w-, UAS-Dicer2; +; Mef2-GAL4/UAS-mCherry-RNAi* (left panel) and *w-, UAS-Dicer2; +; Mef2-GAL4/UAS-Rbf-RNAi* (right panel)

Figure S2

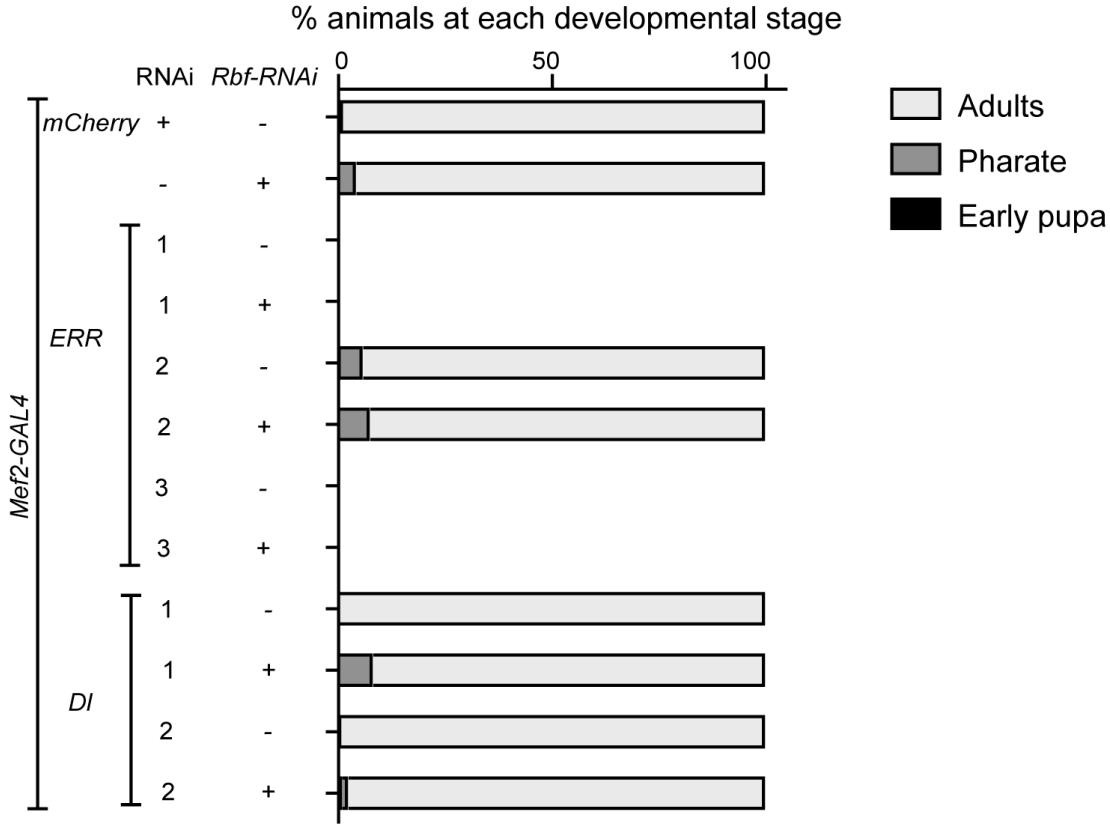

Figure S2: Rbf interacts with other transcription factors. Related to Figure 7.

Genetic interactions between Rbf and candidates were analyzed by knocking down the expression of each gene with or without *Rbf-RNAi* in muscles using *Mef2-GAL4* driver. The candidates tested are estrogen-related receptor (ERR) and Delta (Dl). Lethality during pupa development was analyzed as % animals scored at each developmental stage. Adults: light grey, Pharate: grey, Early pupa: dark grey. N= 94 - 319 flies per genotype. Two independent experiments. Genotypes are *Mef2-GAL4/UAS-mCherry-RNAi*, *UAS-Rbf-RNAi/Mef2-GAL4,* three RNAi lines were used for ERR (1: *UAS-ERR-RNAi*^{HMJ23520}, 2: *UAS-ERR-RNAi*^{JF02431} and 3: *UAS-ERR-RNAi*^{HMC03087}), and two for Dl (1:*UAS-Dl-RNAi*^{GD1238} and 2: *UAS-Dl-RNAi*^{JF02825}).

SUPPLEMENTAL TABLES:

Table S7: Primer sequence. Related to STAR Methods

Gene	Forward primer sequence	Reverse primer sequence	Application
twi	AAGCTCAGCAAGATCCAGACC	GAGCTGGCCGATCCATACG	RT-qPCR
Mef2	CGGATATCATGAGCCTCAACAC	CGTGGAACCATTGCTATTCTGC	RT-qPCR
htl	AACGCATCGAAACCGTTCAC	TGGTGCGCTGTTTCTGTATC	RT-qPCR
stumps	AACAAGGTGGTTGCTCTGCT	ACTGCAGGGTGTAGGGATTG	RT-qPCR
Act88F	ATGGTGGGTATGGGTCAGAA	CTTCTCCATGTCGTCCCAGT	RT-qPCR
Mhc	AAGAACGACCTCGAGAACCAG	TCGGCCTTCTTCTTCTGCTG	RT-qPCR
fln	GGCAAAGAGGGACAAACAAC	ACTACGAGTGCTCATCCGTTC	RT-qPCR
RpL32	TACAGGCCCAAGATCGTGAAG	GACGCACTCTGTTGTCGATACC	RT-qPCR
RpL30	GCAAATACTGCCTGGGCTAC	ACTTCAGTCTTGGCCAGCAT	RT-qPCR
Lmpt-transcript K	AGTGGCTGCCCTAAAACAAC	ATTGCTCCTCTGCTGCGATAG	RT-qPCR
sals	GCAAGCCATGAAGAAGAACCAG	TCGTCTTCGTCTAGCTTCATGG	RT-qPCR
Tm1-transcript H	ATAAGCAGCCGCAGCAAAAG	TTTCTGCTGCCTCTGTTTCG	RT-qPCR
Tm2	TCCAAGATCATGGAGCTGGAG	TTCATCTCGCGCTTGAACTC	RT-qPCR
Mlc2	TTCTCTGTGTTCTCCCAGAAGC	TTGTCGGCATCCATGAGTTG	RT-qPCR
how	ATCTGTCCGATGACCTGCATG	TTAGCTCATCTTCGCCTTCGG	RT-qPCR
salm	ACTACAGGAGCCACACCAAAG	ATGTGTTGCTTCAGGTTGCC	RT-qPCR
Rbf	AACAACGTGAAGCAGCTGAG	ACTTGGCGAGGAATTTCTGC	RT-qPCR
P5CDh1 157/40	TATTCGGCGCTTTTCCACTG	ACAAAGGCACACCACACTTC	ChIP-qPCR
Cyt-c-p -496/-372	AAAGATGGTGTTCGCAGGTC	TGGCCGATCTTGTTGTTTCG	ChIP-qPCR
Zasp66 -847/-712	TCGAAAACTCTGCACATCGG	GGGCCACAGGATTTAATTGACC	ChIP-qPCR
Negative site	TGTGTATGCCTTGCTTGCAC	TCTATGCACACGCTCTACTGAG	ChIP-qPCR
Rbf	CTAATACGACTCACTATAGGGAGCG AATTCGACGAACGCATTC	CTAATACGACTCACTATAGGGA GCAGCCAAGCAGGTACACCTTG	dsRNA
GFP	CTAATACGACTCACTATAGGGAGCG TAAACGGCCACAAGTTCAG	CTAATACGACTCACTATAGGGA GACGAACTCCAGCAGGACCATG	dsRNA

* T7 sequence is underlined

Table S7: Primer sequences. Sequence of primers used to measure gene expression (RT-qPCR), genomic occupancy (ChIP-qPCR) and primers used to make dsRNA.