SUPPLEMENTAL INFORMATION

SUPPLEMENTAL FIGURE LEGENDS

Figure S1. *Lin28a* reactivation promotes hair regrowth, Related to Figure 1.

- **A.** Relative skin thickness of WT vs. iLin28a Tg mice over time. Measurements included the epidermis and dermis.
- **B.** Number of hair follicles per 10x magnification field.
- **C.** Relative follicle bulb diameters were measured.
- **D.** WT and iLin28a Tg mice underwent wax depilation at age p44 and images were taken 11 days later.
- **E.** Phospho-histone H3 immunohistochemistry staining performed on p24 WT and iLin28a Tg mice, which were both in anagen. Data are represented as mean +/- SEM, * P<0.05, ** P< 0.01.

Figure S2. Local *Lin28a* overexpression does not promote adult digit regeneration after distal amputation, Related to Figure 2. Data are represented as mean +/- SEM.

Figure S3. Topical *LIN28B* overexpression does not promote pinnal tissue repair, Related to Figure 3. Data are represented as mean +/- SEM.

Figure S5. *Lin28a* alters the bioenergetic state during tissue repair, Related to Figure 5.

- **A.** LC-MS/MS selected reaction monitoring (SRM) analysis of abundance in glycolysis intermediates in $Lin28a^{+/+};Lin28b^{-/-}$ (n=7), $Lin28a^{+/-}; Lin28b^{-/-}$ (n=8) and $Lin28a^{-/-}; Lin28b^{-/-}$ (n=5) embryos at E10.5 post-coitum. * P<0.1
- **B.** SRM analysis of several metabolic indicators in $Lin28a^{+/+};Lin28b^{-/-}$ (n=7), $Lin28a^{+/-}; Lin28b^{-/-}$ (n=8) and $Lin28a^{-/-};Lin28b^{-/-}$ (n=5) embryos at E10.5 post-coitum. * P<0.1, ** P<0.01
- **C.** Mitochondrial DNA (mtDNA) content analyzed by means of qRT-PCR. Relative amounts were normalized to WT MEFs.
- **D.** Mitochondrial distribution in WT and iLin28a Tg MEFs, as visualized by MitoTracker Red CMXRos staining. Data are represented as mean +/- SEM, * P<0.05, ** P< 0.01.

Figure S6. *Lin28a* enhances mitochondrial oxidation enzymes to promote tissue repair, Related to Figure 6.

- **A.** Schematic of glucose catabolism through glycolysis, the Krebs cycle and oxidative phosphorylation, and the drug inhibitors that modulate this pathway. 2DG, 2-deoxy-D-glucose. 3BP, 3-bromopyruvate. ROS, reactive oxygen species. GSH, glutathione. GSSG, glutathione disulfide. NAC, N-acetyl-cysteine. Anti-A, antimycin-A. Pfkp, phosphofructokinase, platelet type. Pdha1, pyruvate dehydrogenase alpha 1. Idh3b, isocitrate dehydrogenase 3 NAD(+)-specific, mitochondrial, beta subunit. Sdha, succinate dehydrogenase complex subunit A, flavoprotein. Ndufb3/8, NADH dehydrogenase (ubiquinone) 1 beta subcomplex 3/8.
- **B.** Western blots showing efficacy of siRNA knockdown of metabolic enzymes in WT and iLin28a Tg MEFs.
- **C.** qRT-PCR showing efficacy of siRNA knockdown of metabolic enzymes in WT and iLin28a Tg MEFs. A compensatory feedback increase in Ndufb3 and Ndufb8 transcription was observed, after siRNA knockdown of their protein expression.
- **D.** ^{13}C-glucose flux analysis of glycolytic and Krebs cycle intermediates in WT and iLin28a Tg MEFs treated with siRNAs against glycolysis and Krebs cycle enzymes.

E. SRM analysis of the ATP/AMP and GSH/GSSG ratios in WT and iLin28a Tg MEFs treated with siRNAs against electron transport chain enzymes.
F. Primary MEF proliferation over 5 days with and without antimycin-A treatment. Data are represented as mean +/- SEM, * P<0.05, ** P< 0.01.

Figure S7. iLin28a Tg mice do not exhibit improved repair after cardiac cryoinjury. Related to Figure 7.
A. Images of the adult heart 2 weeks after cryoinjury with metal probes cooled in liquid N_2.
B. Injured adult hearts 8 weeks after cryoinjury.
C. H+E and trichrome images of the injured region of the heart at 8 weeks.

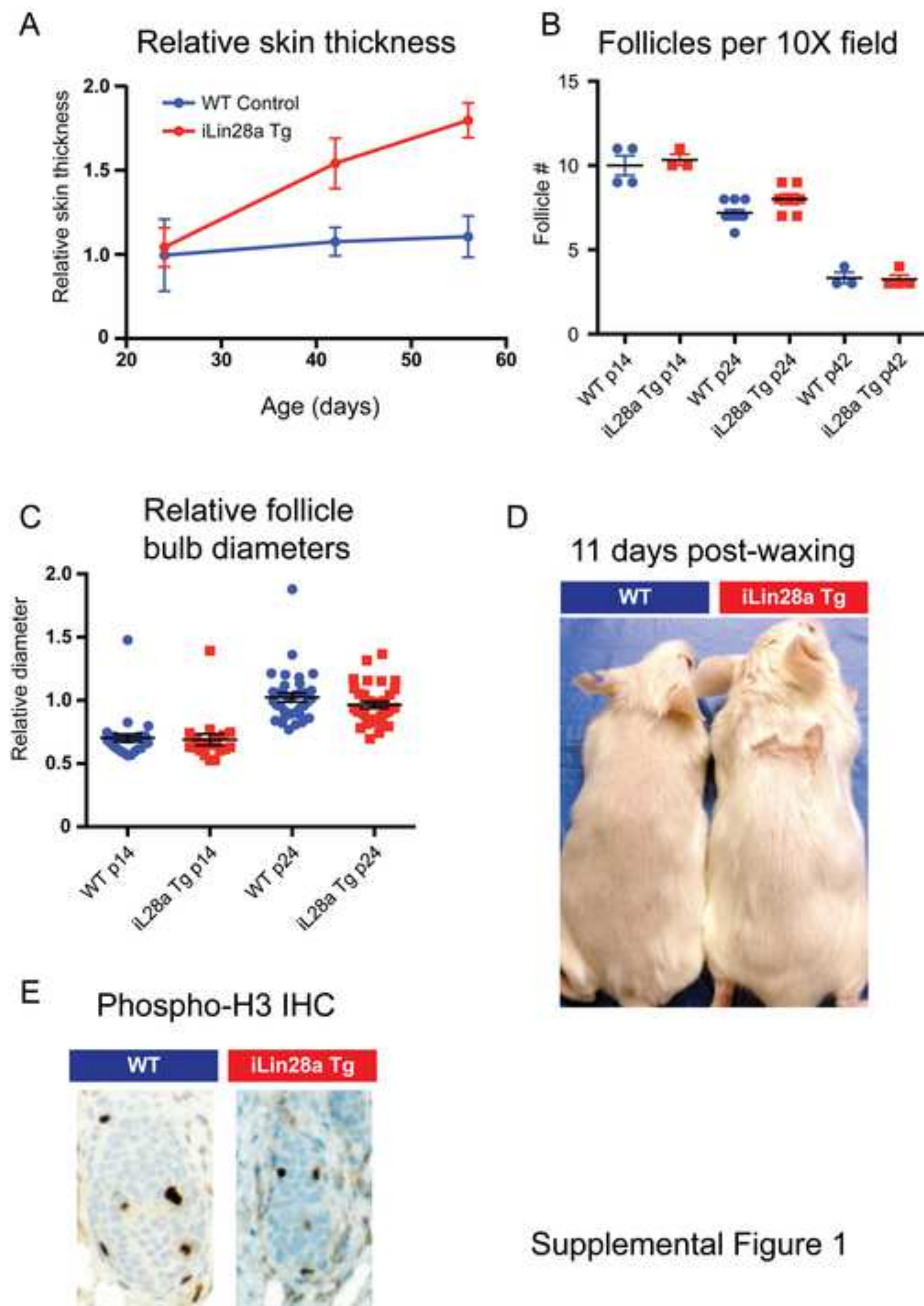
EXTENDED EXPERIMENTAL PROCEDURES

Cell culture. Primary mouse embryonic fibroblasts (MEFs) were derived from control or transgenic mouse embryos at E12.5, then cultured and tested up till passage 10. MEFs were cultured in DMEM (Gibco) supplemented with 10% FBS (Hyclone), 100uM NEAA (Gibco), 2 mM glutamine, 100U/100ug /ml penicillin/streptomycin (Gibco). For drug treatments, MEFs were incubated with 100uM 3-bromopyruvate or 50nM antimycin-A, dissolved in DMSO (vehicle). To check mitochondrial density, MEFs were incubated in CMXRos (Molecular Probes) according to manufacturer instructions, and imaged under a Nikon Eclipse Ti Live Cell Imaging microscope.
Transfections. $2x10^5$ MEFs were transfected with 50nM of a scrambled control, *let-7a* duplex (Ambion), or *let-7* LNA antimiR (Exiqon), or pooled siRNAs against mouse Pfkp, Pdha1, Idh3b, Sdha, Ndufb3, Ndufb8 (OriGene), using Lipofectamine RNAiMAX (Life Technologies) according to manufacturer instructions.
Scratch assay. MEFs were subjected to scratch migration assays, as previously described (Liang et al., 2007).
Cardiac cryoinjury. Mouse heart repair was tested with cryoinjury, as previously described (Robey and Murry, 2008).

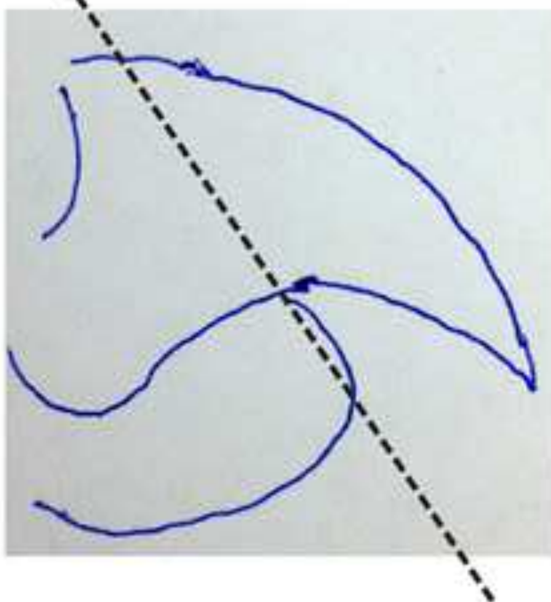
SUPPLEMENTAL REFERENCES

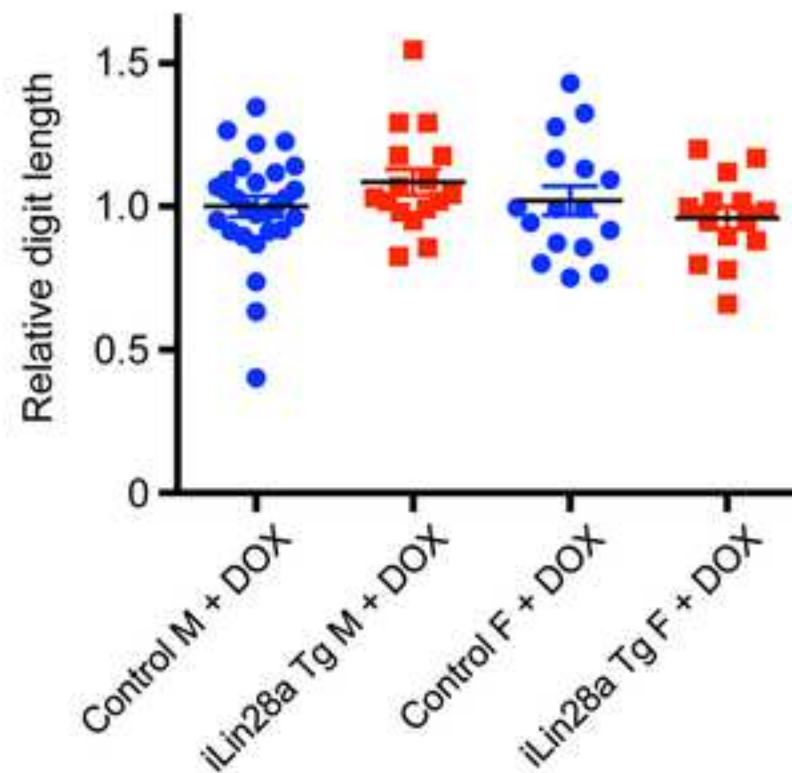
Liang, C. C. et al. (2007). In vitro scratch assay: a convenient and inexpensive method for analysis of cell migration in vitro. Nat. Protoc. 2, 329–33.

Robey, T. E., and Murry, C. E. (2008). Absence of regeneration in the MRL/MpJ mouse heart following infarction or cryoinjury. Cardiovasc. Pathol. 17, 6-13.



Supplemental Figure 1





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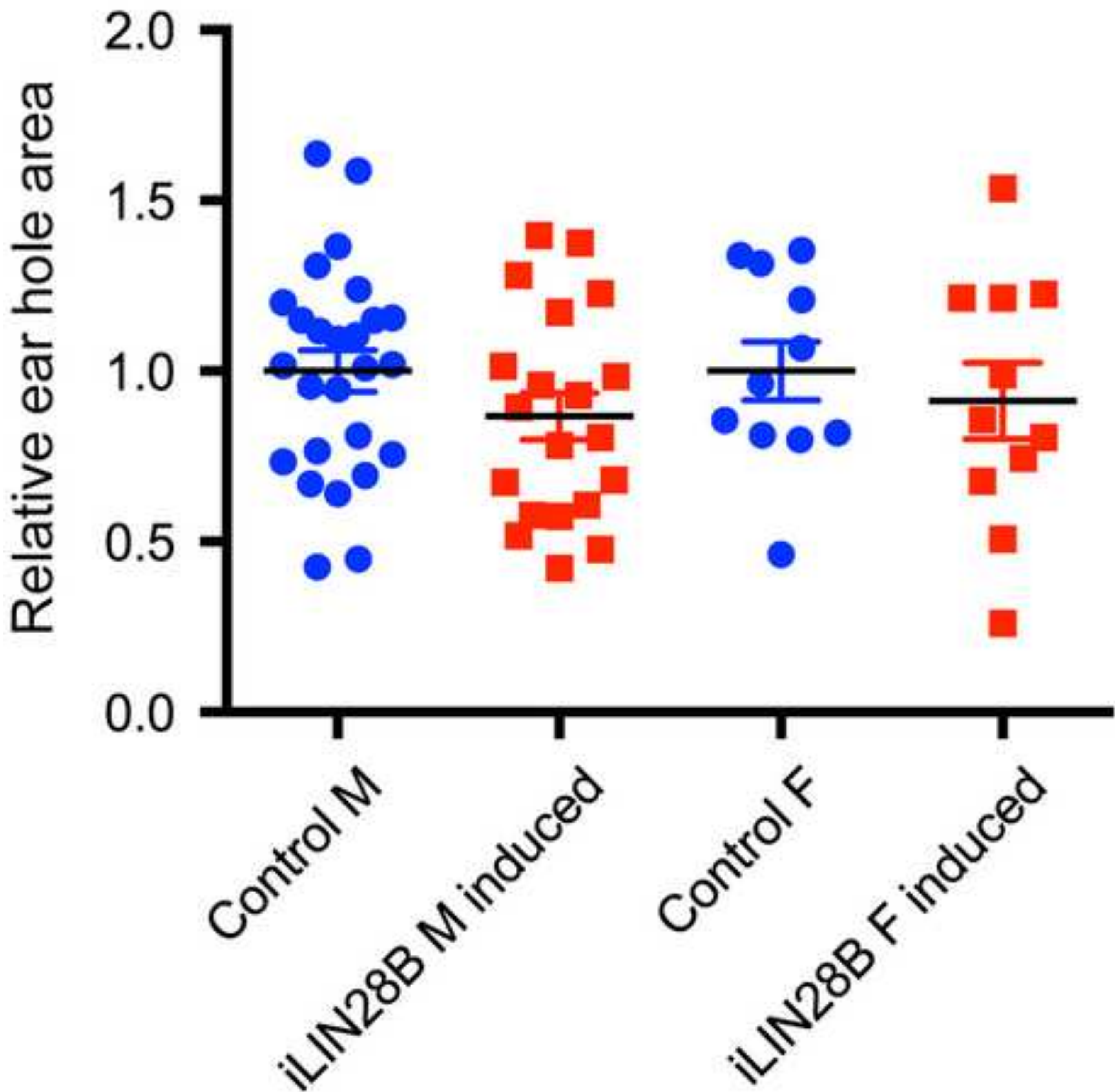

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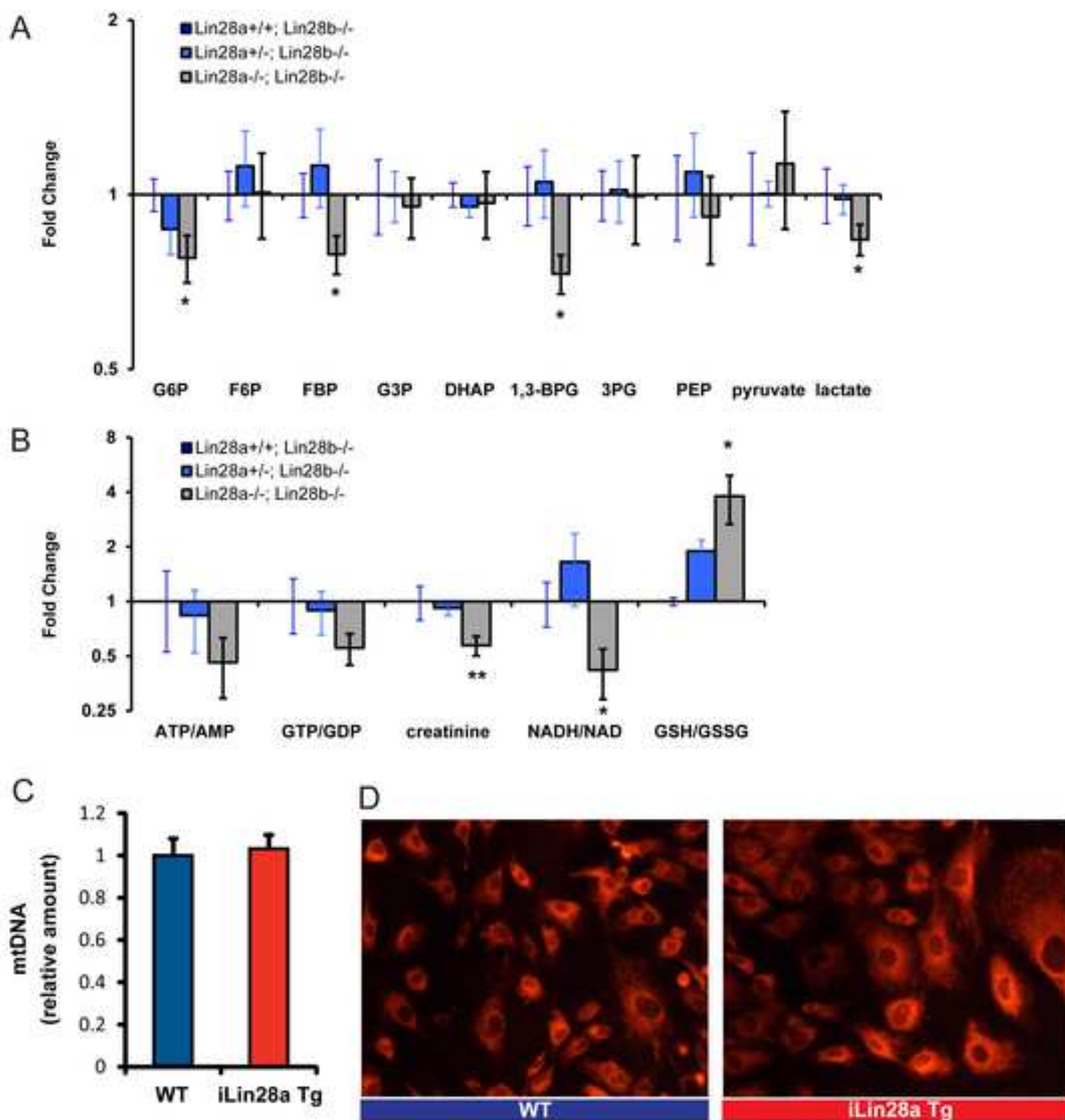

Supplemental Figure 5

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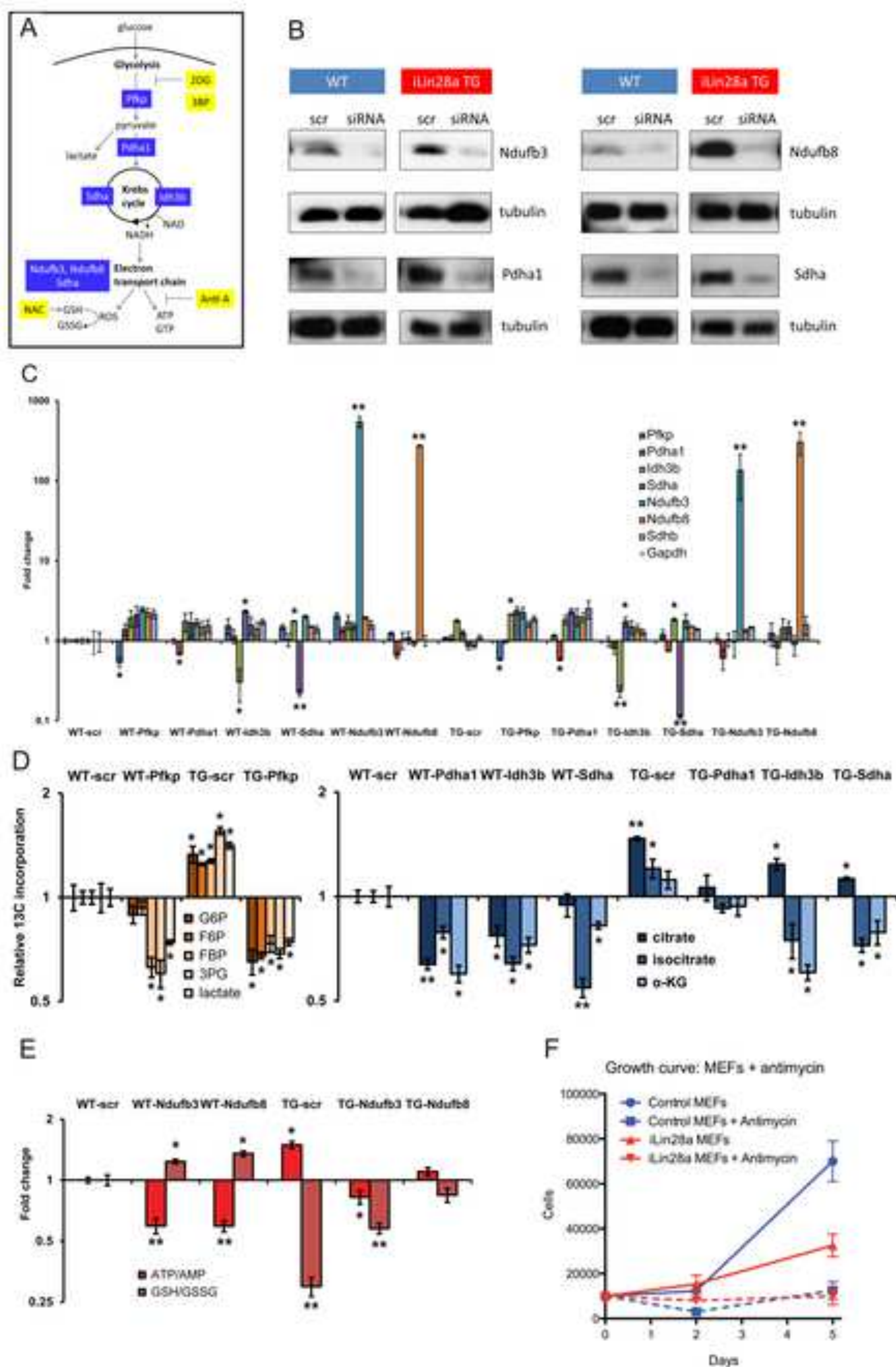

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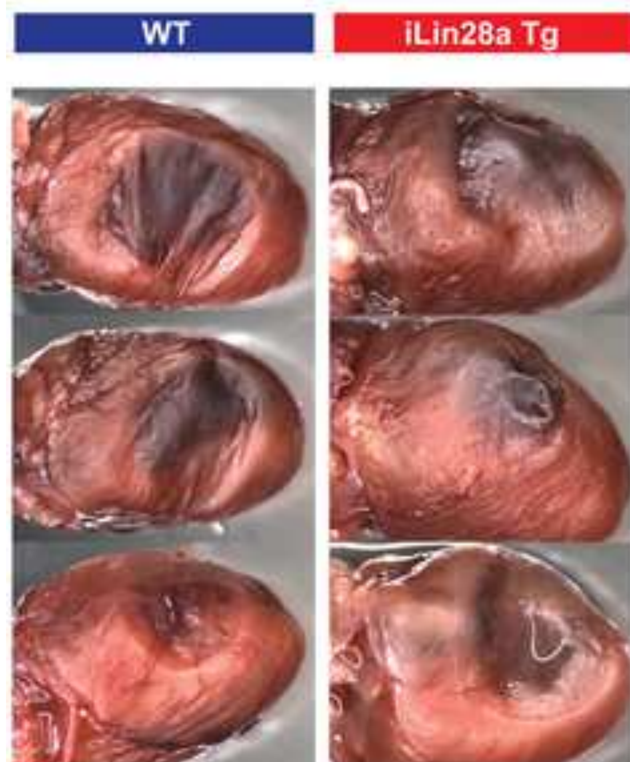

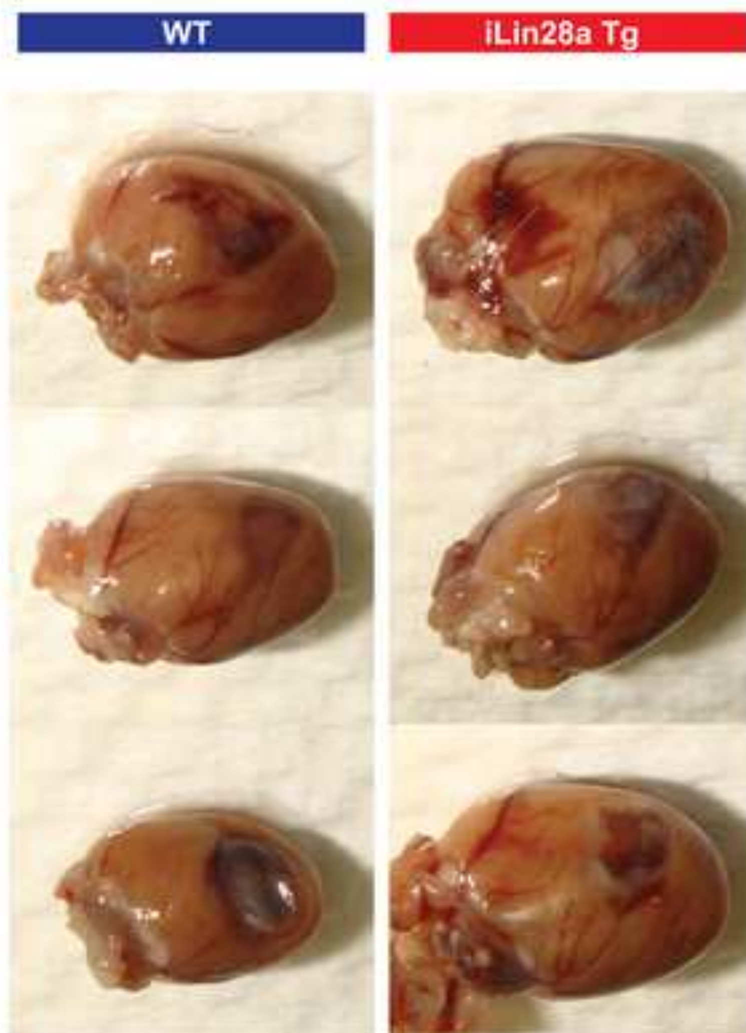

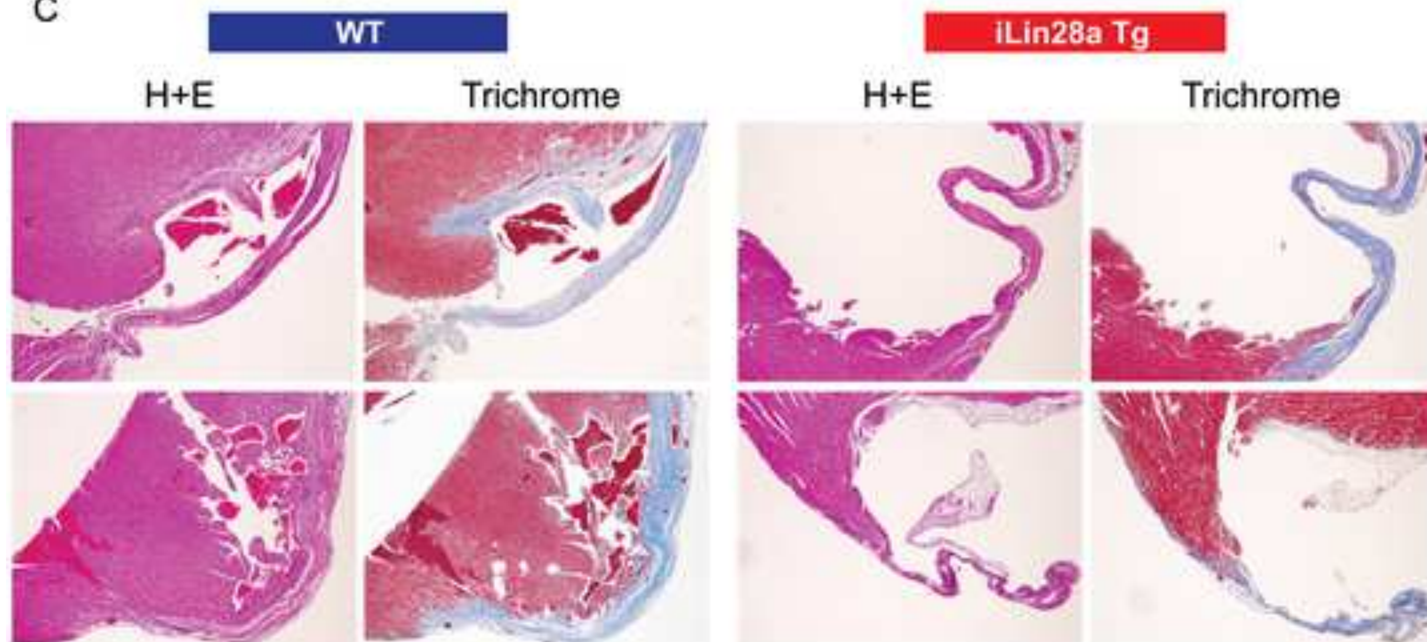

Supplemental Figure 7