ONLINE SUPPLEMENT

cDNA and qPCR

For quantitative gene expression analysis, RNA was fircDNA was generated using M-MLV reverse transcriptase (Promega) under the following protocol and reaction conditions in Table S1. Primers obtained from Sigma and Probes designed and obtained from Roche Universal Probe Library are displayed in Table S2.

Table S1.

Component	**Volume (µL) per Sample**	**Temperature/Time**
RNA	~500 ng (x)	
Random Hexamer	0.5 (final 0.5 µg)	
Oligo-dT	1.85 (final 0.5 µg)	
Nuclease free water	Up to 14.7 µL	Incubate at 70 C for 5 min then return to ice for >1 min
5x Reaction buffer	5	
dNTPs (10 mM)	1.25	
RNase Inhibitor (40 U/µL)	0.625	
M-MLV (200 U/µL)	1	Incubate 37ºC – 60min, 42ºC – 10 min, 85ºC – 5 min, hold at 4ºC

Table S2.

Target	Forward Primer 5'-3'	Reverse Primer 5'-3'	Efficiency	Chemistry	Roche Probe
B2m	acactgaattcacccccact	tcacatgtctcgatcccagt	1.96	Sybr	
β-actin	aaggccaaccgtgaaaagat	gtggtacgaccagaggcatac		Taq	56
Nrf2	gcttttggcagagacattcc	atcagccagctgcttgtttt	1.88	Sybr	
Nqo1	ggtagcggctccatgtactc	agacctggaagccacagaaa	1.96	Sybr	
Ho1	gagcctgaatcgagcagaac	ctcggcttggatgtgtacct			
Gclc	aggctctctgcaccatcact	ctctgggttgggtctgtgtt			
Gclm	tcccatgcagtggagaagat	agctgtgcaactccaaggac	1.96		
Keap1	gatcggctgcactgaactg	ggcagtgtgacaggttgaag			
Fbx32	agtgaggaccggctactgtg	gatcaaacgcttgcgaatct	1.86	Sybr	
Trim63	cctgcagagtgaccaagga	ggcgtagagggtgtcaaact		Taq	17
Pppc3a	aggaacatttcactcacaacaca	tcacacacagctgggtaactg	1.91	Sybr	
Myh1	gaggaccaagtgagtgagctg	ctggcgtgagtattcacctg	1.99	Taq	63
Myh2	aactccaggcaaaagtgaaatc	tggatagatttgtgttggattgtt	1.98	Taq	75
Myh4	tggccgagcaagagctac	ttgatgaggctggtgttctg	2.01	Taq	17
Myh7	agatccgaaagcaactggag	ggatcttgccctcctcgt	1.98	Taq	25
Ppargc1a	acagctttctgggtggattg	tctgtgagaaccgctagcaa	1.93	Sybr	

Muscle CSA Measurements

Figure S1. Muscle cross sectional area (CSA) was assessed using CellProfiler software. Figure S1 provides representative images of the program utilized to analyze results. Briefly, frozen sections of GAST or SOL muscle were stained with anti-dystrophin antibodies followed by anti-rabbit-AlexaFluor 594 conjugated secondary antibody to visualize membranes of individual muscle fibers. Images were fed into the CellProfiler pipeline, the "ImageMath" function was applied to invert the intensities of the input image (A). Applying a "threshold" function raised levels of the input image (B), which was used to "subtract" from the inverted image to create distinctive gaps between each individual fiber (C). The subtracted image is used to identify primary objects (D). Those objects are manually edited to discard any outliers or inappropriately assigned fibers (E). The final selected objects are then measured by the CellProfiler program and individual fiber area was exported in pixels (F). The pixel values were changed to CSA in μm^2.

Figure S2. Frozen SOL muscle was doubled labeled with dystrophin to mark membranes (red) and myosin heavy chain (MHC) II (green) for fast, glycolytic fibers. Representative images of WT control (A), KO control (B), WT+STZ (C) and KO+STZ (D) are provided.

Figure S1.

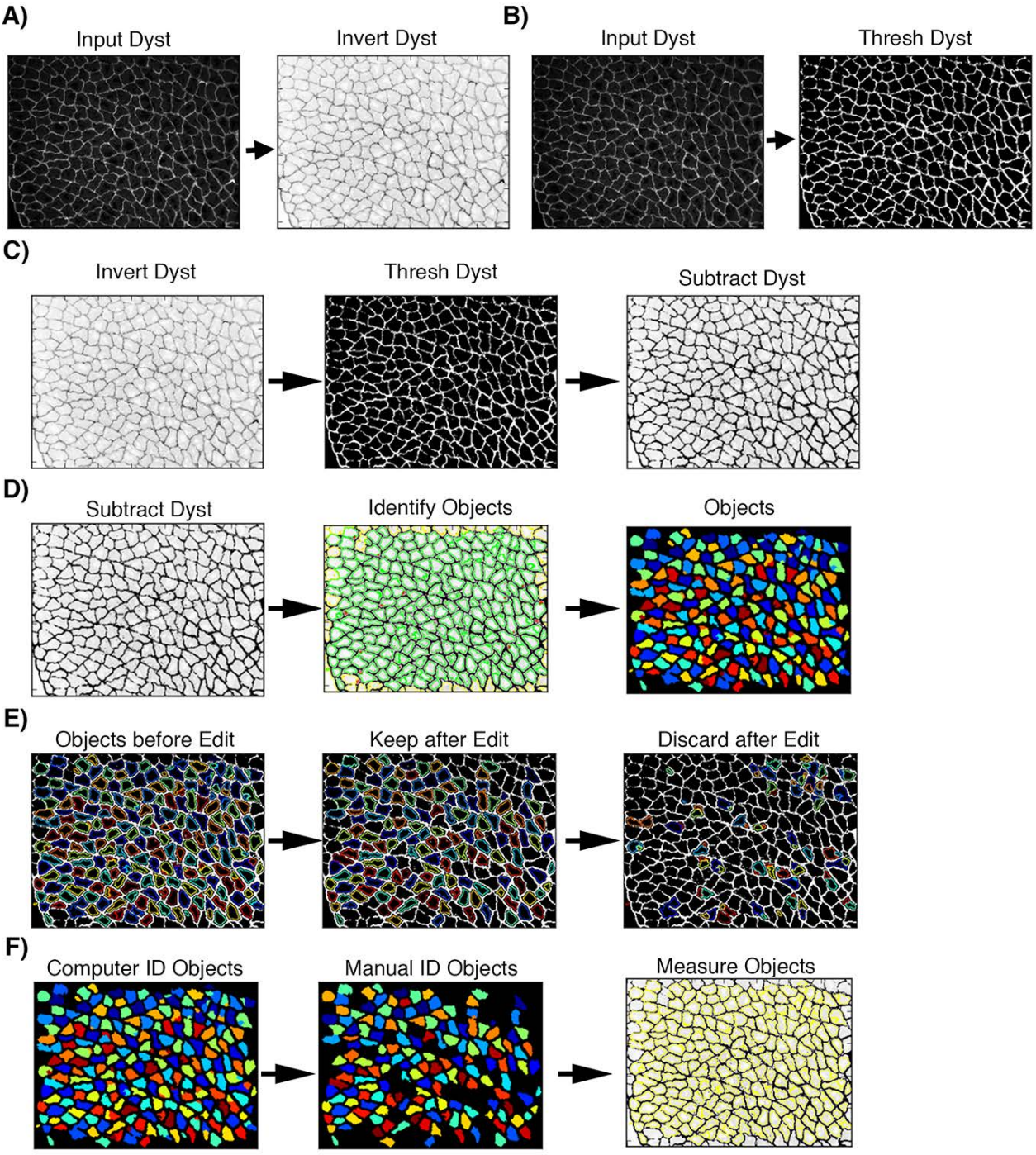
A)
Input Dyst
Invert Dyst
B)
Input Dyst
Thresh Dyst
C)
Invert Dyst
Thresh Dyst
Subtract Dyst
D)
Subtract Dyst
Identify Objects
Objects
E)
Objects before Edit
Keep after Edit
Discard after Edit
F)
Computer ID Objects
Manual ID Objects
Measure Objects

Figure S2.