Expanded View Figures

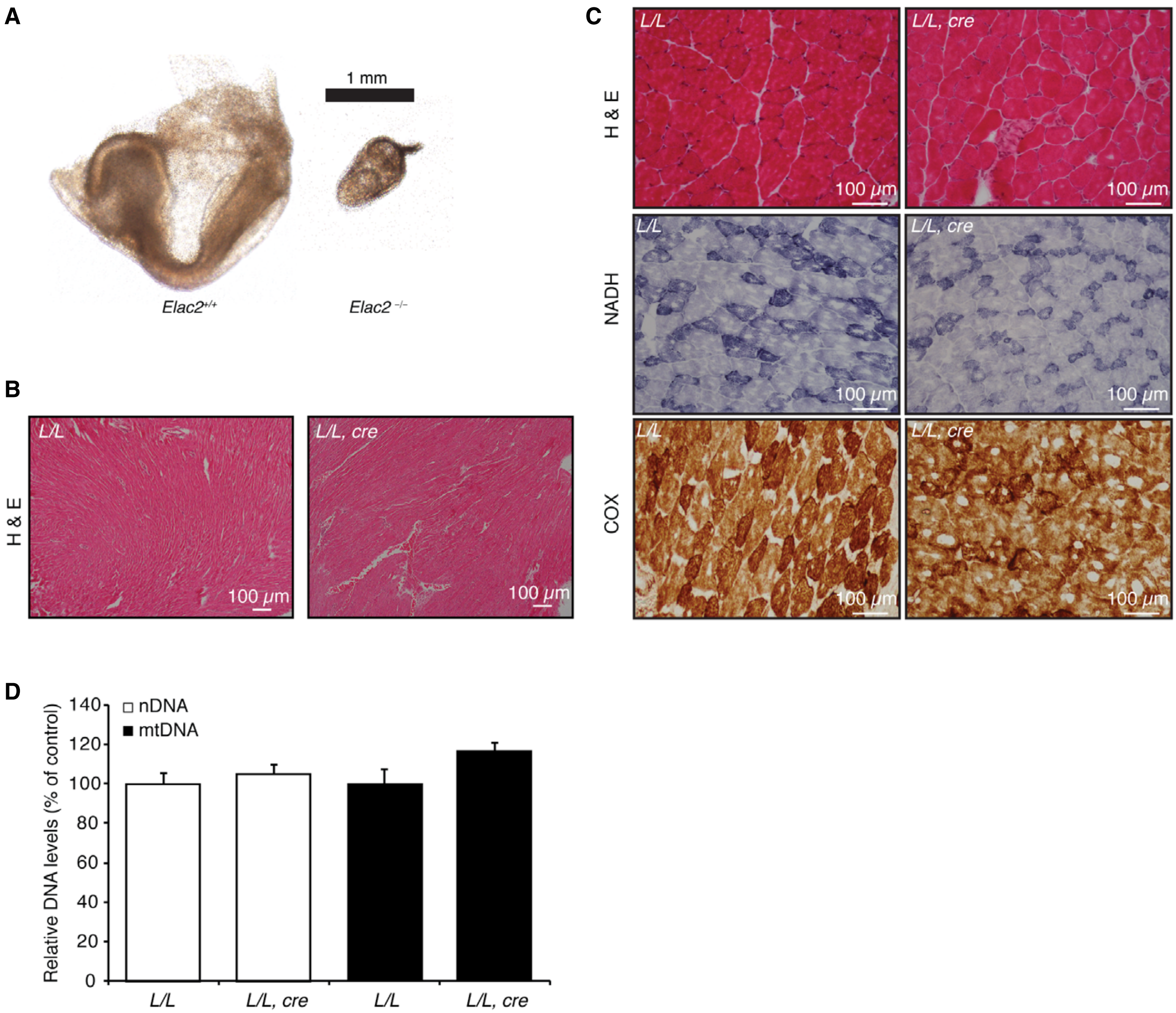

Figure EV1. Related to Fig 1: The effects of ELAC2 loss on survival and skeletal muscle.

A Morphology of the *Elac2*$^{+/+}$ and *Elac2*$^{-/-}$ embryos at day E8.5. Scale bar, 1 mm.

B Hematoxylin and eosin (H&E) of heart from control (*L/L*) and knockout (*L/L, cre*) mice. Scale bar is 100 μm.

C Hematoxylin and eosin (H&E), and NADH and COX staining of skeletal muscle from control (*L/L*) and knockout (*L/L, cre*) mice. Scale bar is 100 μm.

D Changes in mitochondrial and nuclear DNA were determined by qPCR on DNA isolated from 4-week-old control (*L/L*, n = 3) and knockout (*L/L, cre*, n = 3) mouse hearts. Values are means ± SEM.

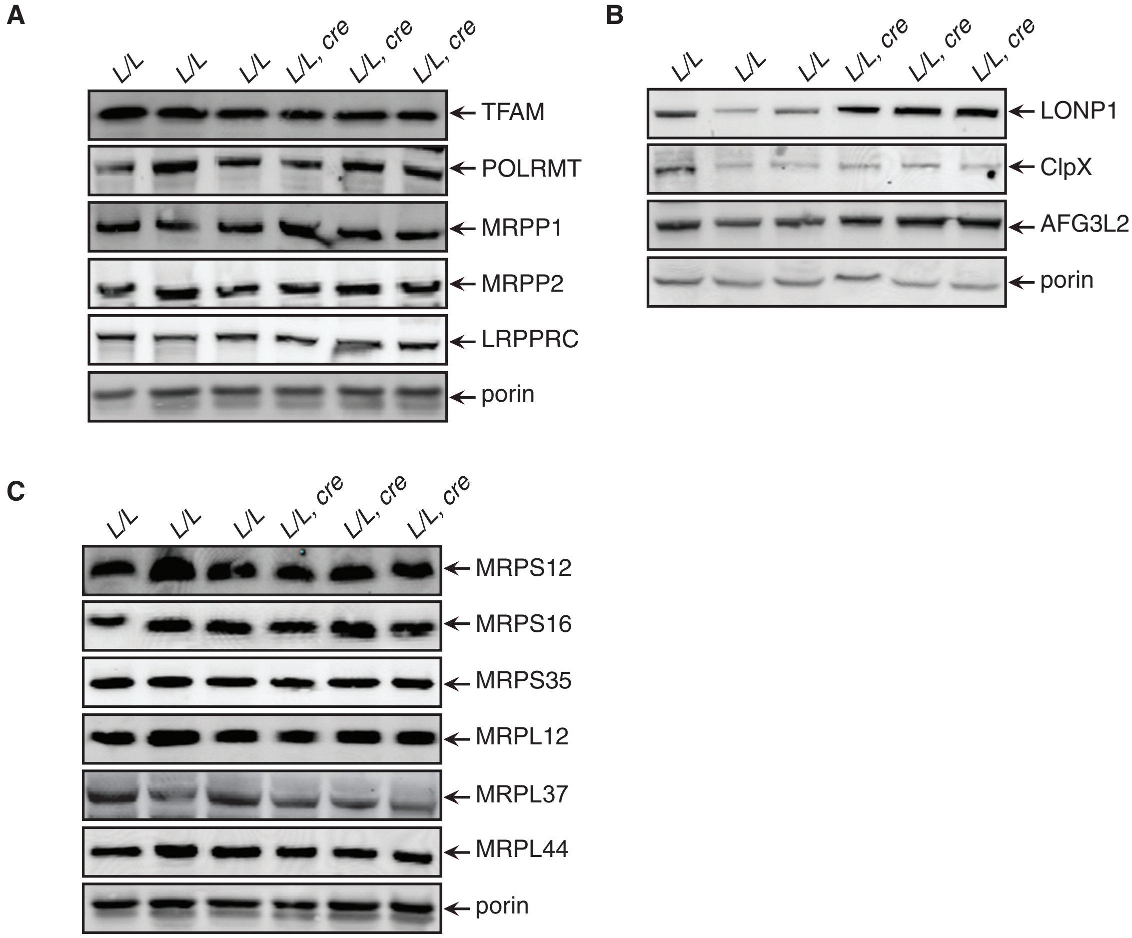

Figure EV2. Related to Fig 4: The effects of ELAC2 loss on RNA-binding proteins.

A–C The levels of nuclear-encoded mitochondrial RNA-binding proteins (A), proteases (B), and mitochondrial ribosomal proteins (C) were measured by immunoblotting in heart mitochondria from 4-week-old control (*L/L*) and knockout (*L/L, cre*) mice. Porin was used as a loading control.

Figure EV3. Related to Figs 5 and 6: Loss of *Elac2* causes a primarily transcriptional response.

A–D Reads mapping from the small RNA-Seq datasets were summarized to identify that mature tRNA sequences were reduced in most cases in the *Elac2* knockout mice (A), loss of the mascRNA derived from *Malat1* (B), a strong coordinated increase in cytoplasmic tRNA synthetases and several snoRNP components (C). Differential expression analyses on the RNA-Seq datasets identified an increased transcriptional regulation of mitochondrial ribosomal mRNAs and tRNA synthetases (D). Taken together, these data show that loss of ELAC2 and consequent impaired processing of non-coding RNAs causes a compensatory increase in ribosomal proteins and tRNA synthetases in response to reduced mature tRNA levels.

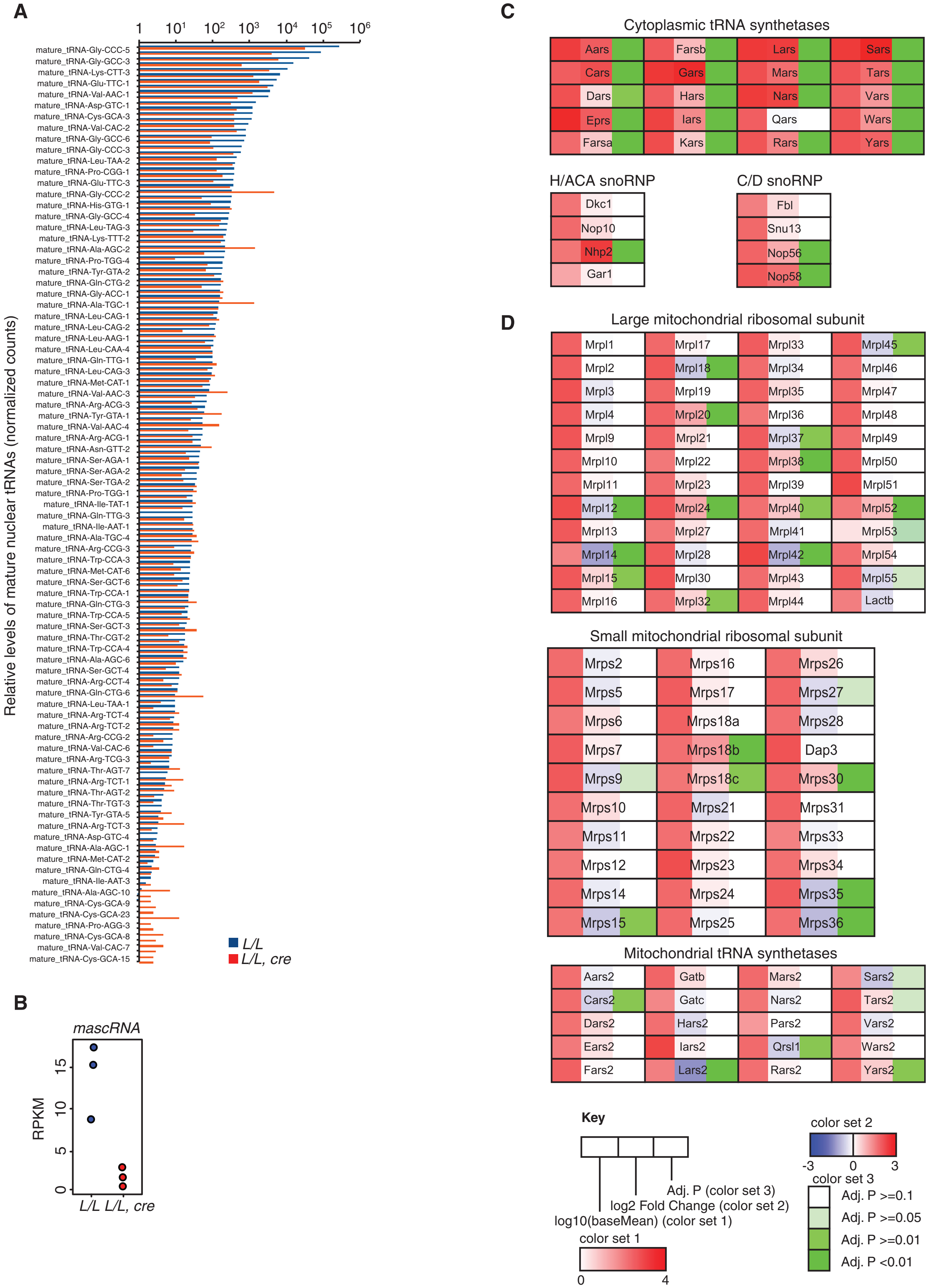
A
1 10^1 10^2 10^3 10^4 10^5 10^6
Relative levels of mature nuclear tRNAs (normalized counts)
mature_tRNA-Gly-CCC-5
mature_tRNA-Gly-GCC-3
mature_tRNA-Lys-CTT-3
mature_tRNA-Glu-TTC-1
mature_tRNA-Val-AAC-1
mature_tRNA-Asp-GTC-1
mature_tRNA-Cys-GCA-3
mature_tRNA-Val-CAC-2
mature_tRNA-Gly-GCC-6
mature_tRNA-Gly-CCC-3
mature_tRNA-Leu-TAA-2
mature_tRNA-Pro-CGG-1
mature_tRNA-Glu-TTC-3
mature_tRNA-Gly-CCC-2
mature_tRNA-His-GTG-1
mature_tRNA-Gly-GCC-4
mature_tRNA-Leu-TAG-3
mature_tRNA-Lys-TTT-2
mature_tRNA-Ala-AGC-2
mature_tRNA-Pro-TGG-4
mature_tRNA-Tyr-GTA-2
mature_tRNA-Gln-CTG-2
mature_tRNA-Gly-ACC-1
mature_tRNA-Ala-TGC-1
mature_tRNA-Leu-CAG-1
mature_tRNA-Leu-CAG-2
mature_tRNA-Leu-AAG-1
mature_tRNA-Leu-CAA-4
mature_tRNA-Gln-TTG-1
mature_tRNA-Leu-CAG-3
mature_tRNA-Met-CAT-1
mature_tRNA-Val-AAC-3
mature_tRNA-Arg-ACG-3
mature_tRNA-Tyr-GTA-1
mature_tRNA-Val-AAC-4
mature_tRNA-Arg-ACG-1
mature_tRNA-Asn-GTT-2
mature_tRNA-Ser-AGA-1
mature_tRNA-Ser-AGA-2
mature_tRNA-Ser-TGA-2
mature_tRNA-Pro-TGG-1
mature_tRNA-Ile-TAT-1
mature_tRNA-Gln-TTG-3
mature_tRNA-Ile-AAT-1
mature_tRNA-Ala-TGC-4
mature_tRNA-Arg-CCG-3
mature_tRNA-Trp-CCA-3
mature_tRNA-Met-CAT-6
mature_tRNA-Ser-GCT-6
mature_tRNA-Trp-CCA-1
mature_tRNA-Gln-CTG-3
mature_tRNA-Trp-CCA-5
mature_tRNA-Ser-GCT-3
mature_tRNA-Thr-CGT-2
mature_tRNA-Trp-CCA-4
mature_tRNA-Ala-AGC-6
mature_tRNA-Ser-GCT-4
mature_tRNA-Arg-CCT-4
mature_tRNA-Gln-CTG-6
mature_tRNA-Leu-TAA-1
mature_tRNA-Arg-TCT-4
mature_tRNA-Arg-TCT-2
mature_tRNA-Arg-CCG-2
mature_tRNA-Val-CAC-6
mature_tRNA-Arg-TCG-3
mature_tRNA-Thr-AGT-7
mature_tRNA-Arg-TCT-1
mature_tRNA-Thr-AGT-2
mature_tRNA-Thr-TGT-3
mature_tRNA-Tyr-GTA-5
mature_tRNA-Arg-TCT-3
mature_tRNA-Asp-GTC-4
mature_tRNA-Ala-AGC-1
mature_tRNA-Met-CAT-2
mature_tRNA-Gln-CTG-4
mature_tRNA-Ile-AAT-3
mature_tRNA-Ala-AGC-10
mature_tRNA-Cys-GCA-9
mature_tRNA-Cys-GCA-23
mature_tRNA-Pro-AGG-3
mature_tRNA-Cys-GCA-8
mature_tRNA-Val-CAC-7
mature_tRNA-Cys-GCA-15
L/L
L/L, cre
B
mascRNA
RPKM
0 5 10 15
L/L L/L, cre
C
Cytoplasmic tRNA synthetases
Aars Farsb Lars Sars
Cars Gars Mars Tars
Dars Hars Nars Vars
Eprs Iars Qars Wars
Farsa Kars Rars Yars
H/ACA snoRNP
Dkc1
Nop10
Nhp2
Gar1
C/D snoRNP
Fbl
Snu13
Nop56
Nop58
D
Large mitochondrial ribosomal subunit
Mrpl1 Mrpl17 Mrpl33 Mrpl45
Mrpl2 Mrpl18 Mrpl34 Mrpl46
Mrpl3 Mrpl19 Mrpl35 Mrpl47
Mrpl4 Mrpl20 Mrpl36 Mrpl48
Mrpl9 Mrpl21 Mrpl37 Mrpl49
Mrpl10 Mrpl22 Mrpl38 Mrpl50
Mrpl11 Mrpl23 Mrpl39 Mrpl51
Mrpl12 Mrpl24 Mrpl40 Mrpl52
Mrpl13 Mrpl27 Mrpl41 Mrpl53
Mrpl14 Mrpl28 Mrpl42 Mrpl54
Mrpl15 Mrpl30 Mrpl43 Mrpl55
Mrpl16 Mrpl32 Mrpl44 Lactb
Small mitochondrial ribosomal subunit
Mrps2 Mrps16 Mrps26
Mrps5 Mrps17 Mrps27
Mrps6 Mrps18a Mrps28
Mrps7 Mrps18b Dap3
Mrps9 Mrps18c Mrps30
Mrps10 Mrps21 Mrps31
Mrps11 Mrps22 Mrps33
Mrps12 Mrps23 Mrps34
Mrps14 Mrps24 Mrps35
Mrps15 Mrps25 Mrps36
Mitochondrial tRNA synthetases
Aars2 Gatb Mars2 Sars2
Cars2 Gatc Nars2 Tars2
Dars2 Hars2 Pars2 Vars2
Ears2 Iars2 Qrsl1 Wars2
Fars2 Lars2 Rars2 Yars2
Key
Adj. P (color set 3)
log2 Fold Change (color set 2)
log10(baseMean) (color set 1)
color set 1
0 4
color set 2
-3 0 3
color set 3
Adj. P >=0.1
Adj. P >=0.05
Adj. P >=0.01
Adj. P <0.01

Figure EV3.

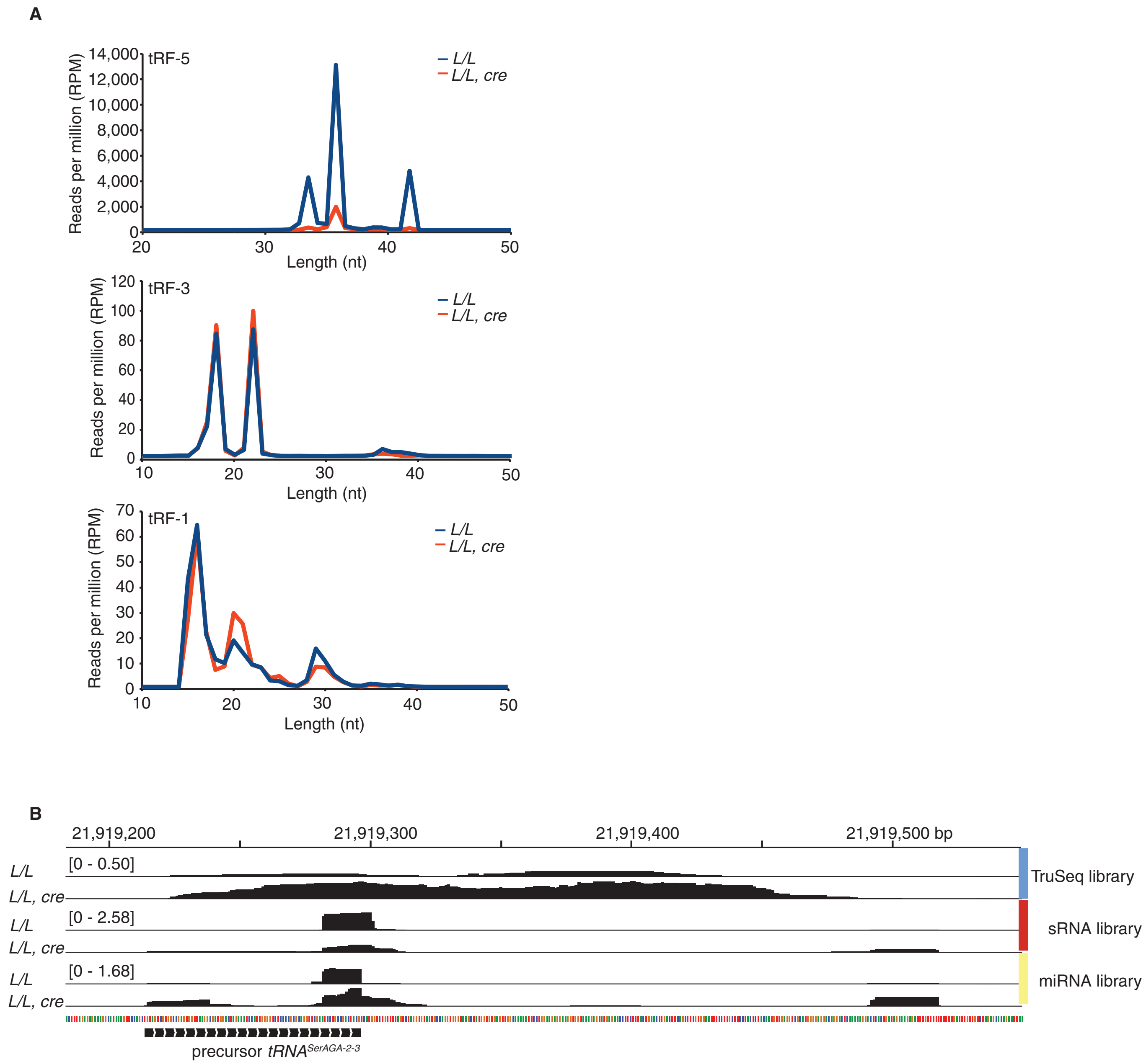

Figure EV4. Related to Fig 7: Loss of *Elac2* causes varied effects on tRNA fragment pools and increases in pre-tRNA-derived small RNAs.

A, B We identified that tRNA fragments derived from the 5′ and 3′ ends of mature tRNAs (tRF-5 and tRF-3, respectively) and 3′ pre-tRNA trailers (tRF-1) showed differential changes (A) and increases in pre-tRNA-derived small RNA associated with Pol III termination sites, including those derived from secondary termination sites from long pre-tRNA transcripts (B).

