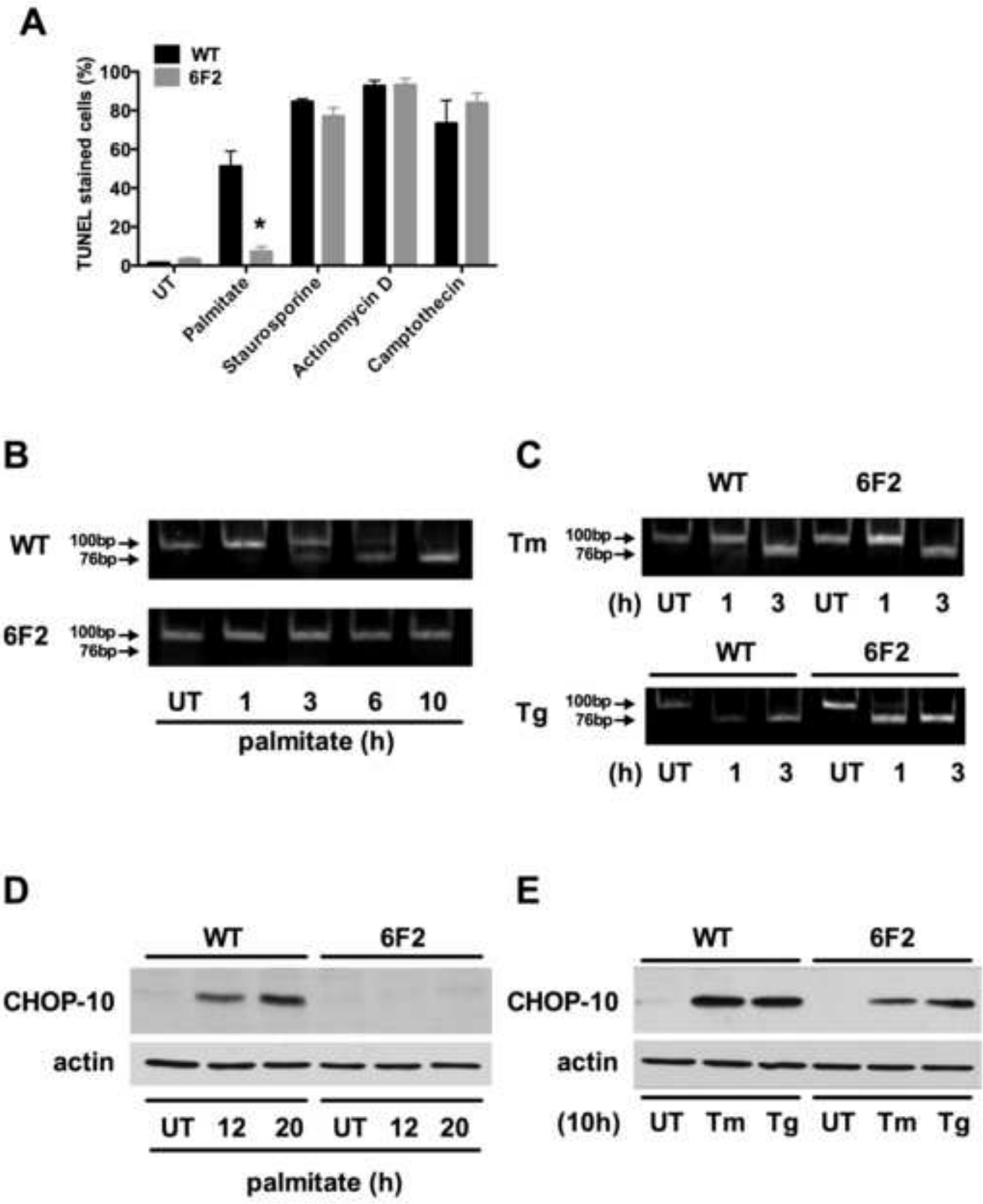
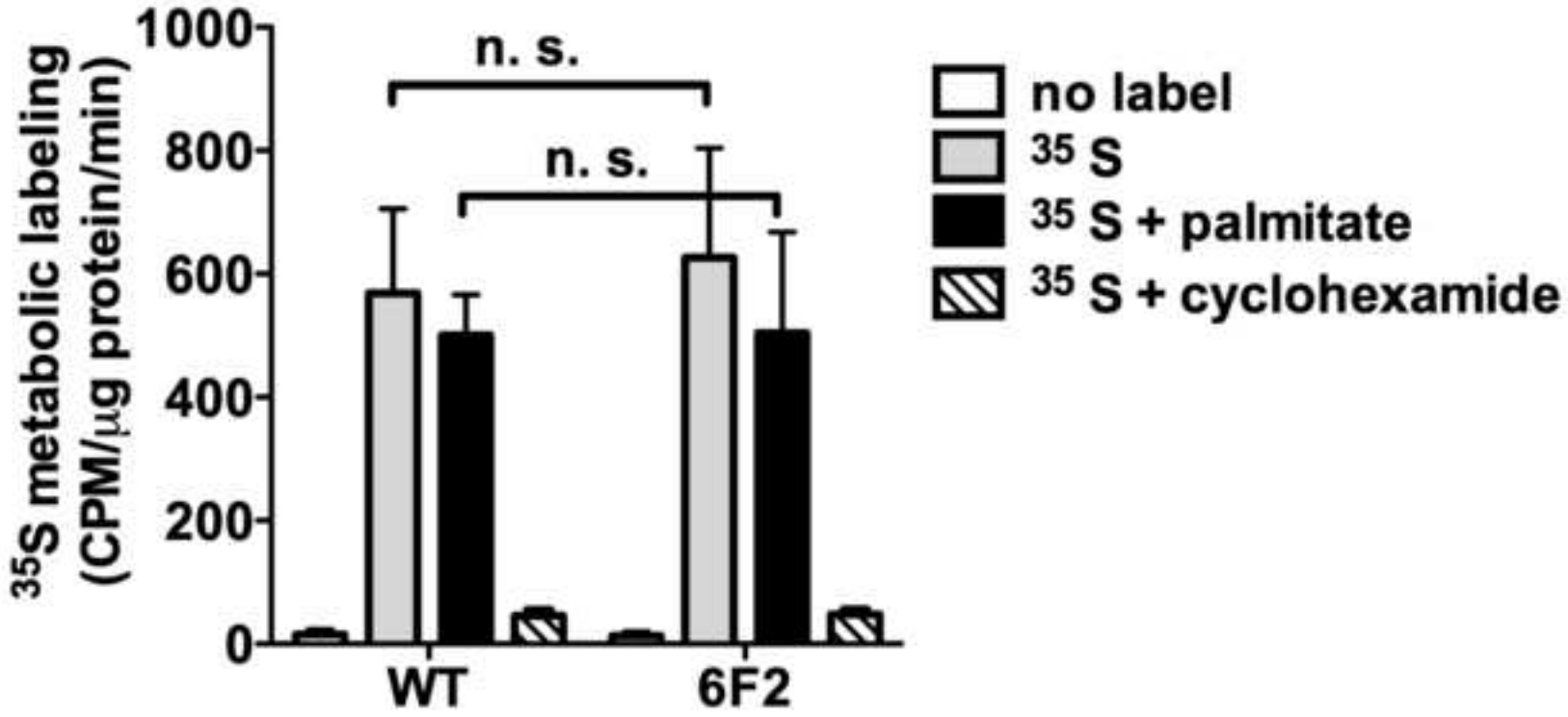
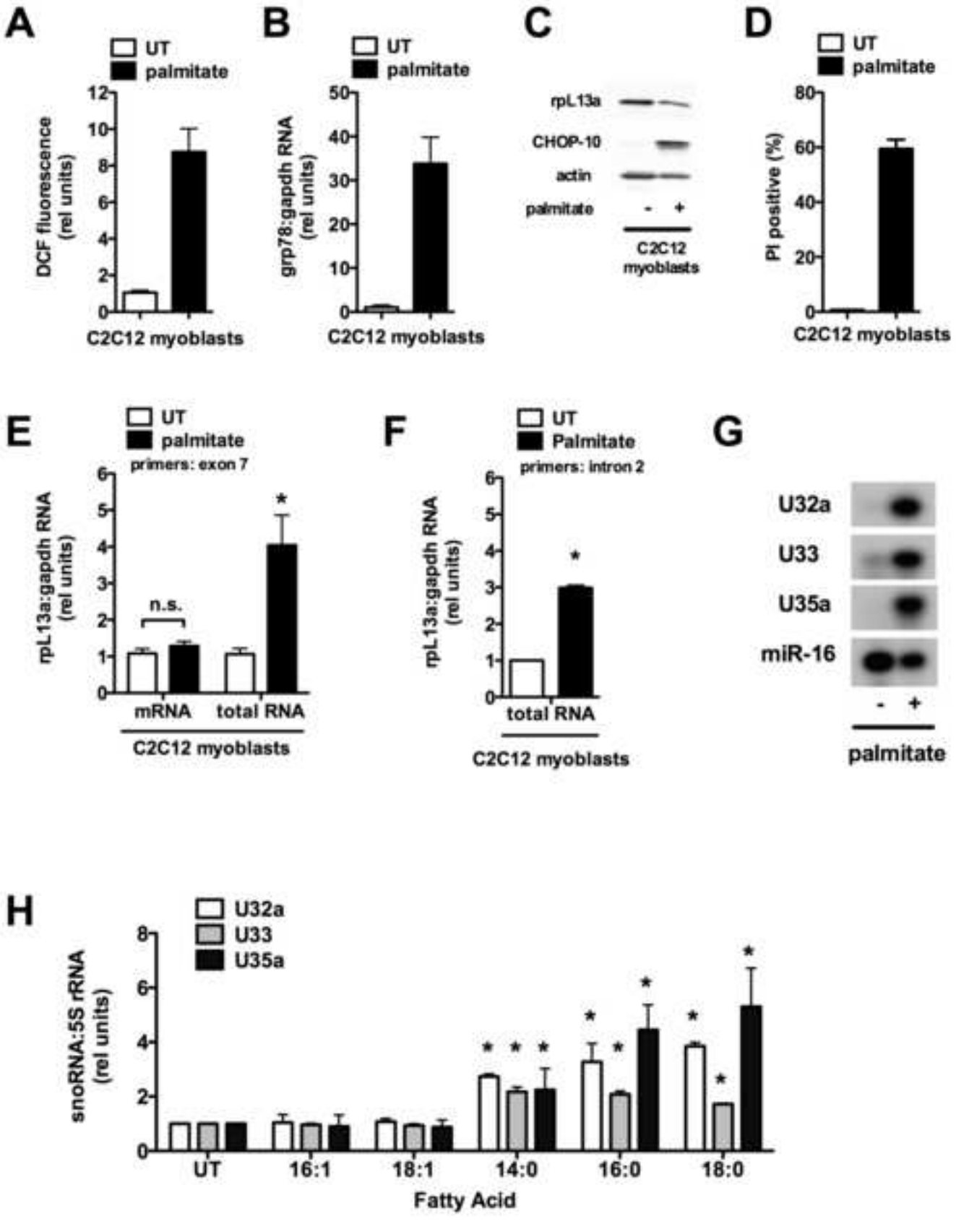
Supplemental Figure 1



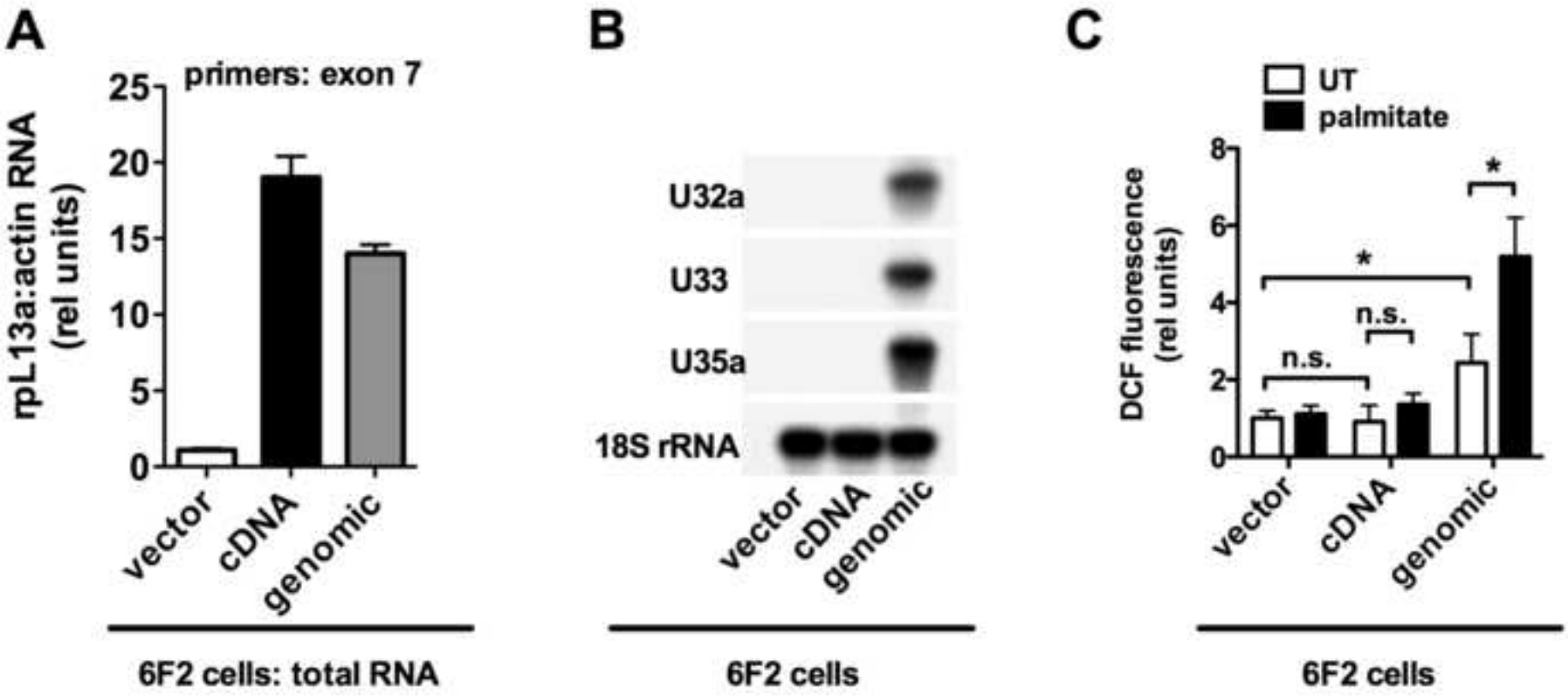
Supplemental Figure 2



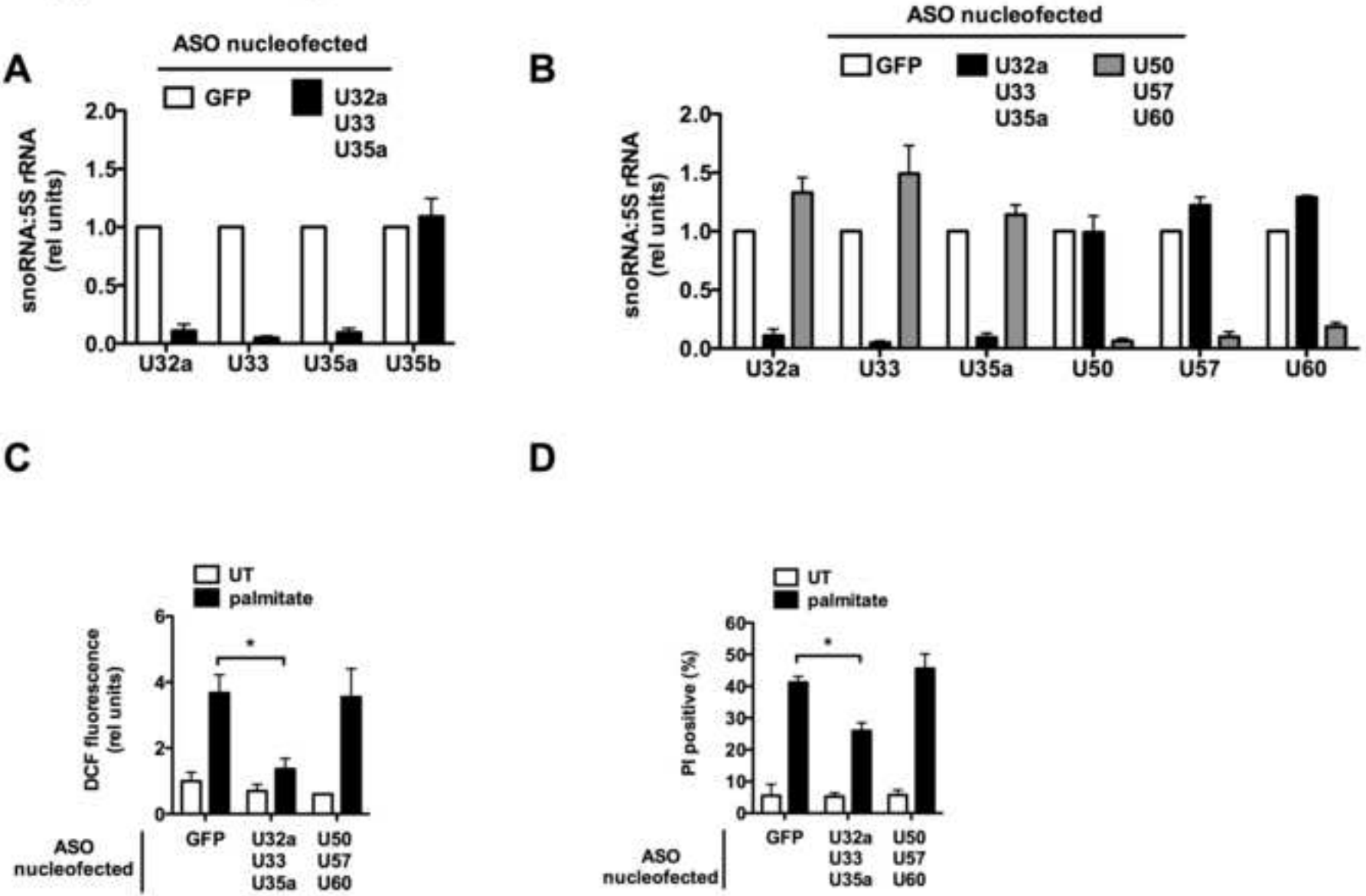
Supplemental Figure 3



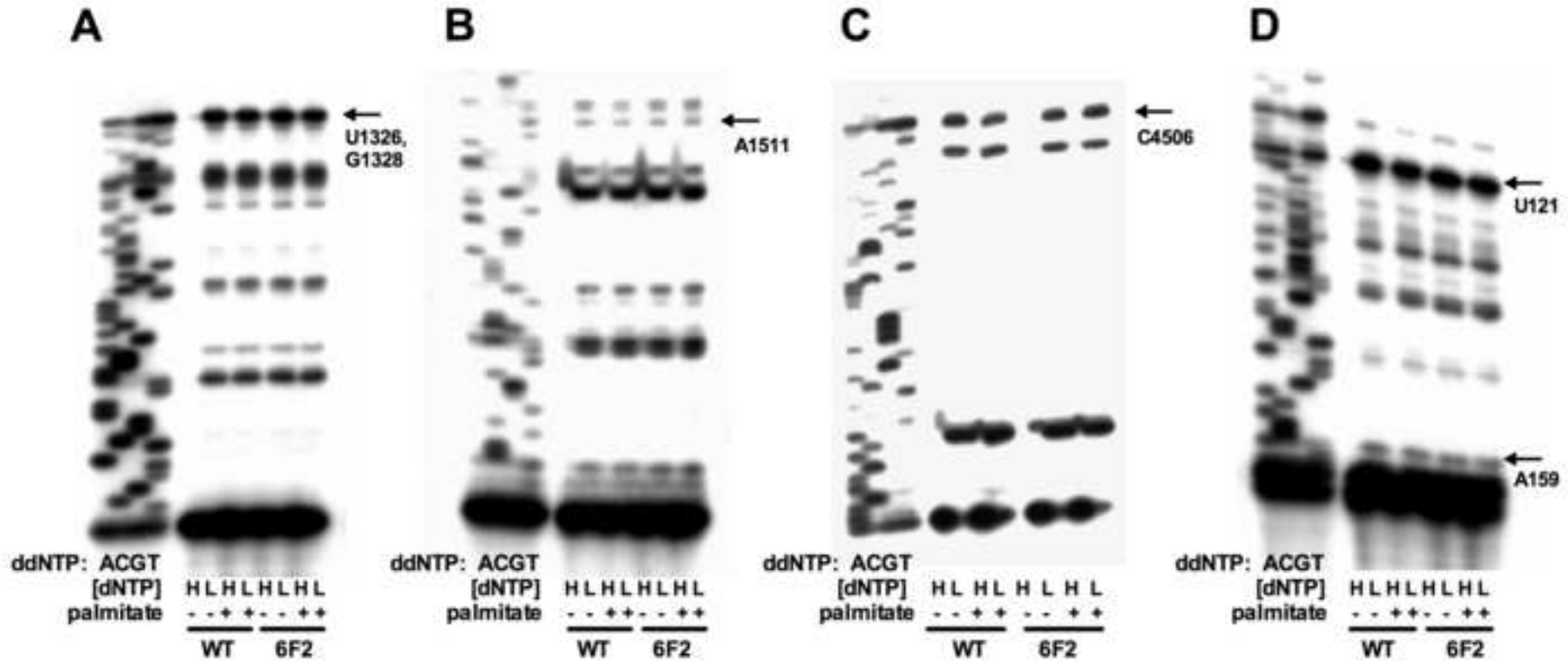
Supplemental Figure 4



Supplemental Figure 5



Supplemental Figure 6



SUPPLEMENTAL FIGURE LEGENDS

Supplemental Figure 1. 6F2 cells are protected from palmitate-induced apoptosis and ER stress. (Related to Figure 1)

(A) Wild type (WT) and mutant 6F2 CHO cells were untreated (UT) or supplemented with 500 μM palmitate for 48 h or with or 80 nM staurosporine, 2 μM actinomycin D, or 10 μM camptothecin for 24 h. Apoptosis was quantified by DNA fragment end labeling (TUNEL) and flow cytometry on 10^4 cells/sample. (B, C, D, E) WT and 6F2 cells were untreated (UT) or treated with 500 μM palmitate (B, D), or 2.5 μg/ml tunicamycin (Tm), or 1 μM thapsigargin (Tg) (C, E) for indicated times. (B, C) Cells were harvested and cDNA synthesized. PCR was performed using primers specific for a 100 nucleotide region of XBP-1 mRNA sequence containing the 26 nucleotides spliced out during ER stress induction. PCR products were separated by non-denaturing PAGE, followed by EtBr staining. (D, E) Protein lysates were analyzed by western blotting for CHOP-10 or actin.

Supplemental Figure 2. Global rates of protein synthesis are unchanged in 6F2 cells. (Related to Figure 2)

WT and 6F2 cells were untreated, or treated with 500 μM palmitate, or treated with cyclohexamide. Cells were then pulse-labeled for 30 min with or without ^{35}S-Cys/Met and TCA-precipitable proteins were collected. Radiolabel incorporation was quantified by scintillation counting and normalized to total cellular protein. Graph shows mean ± SE for 3 independent experiments. n.s., not significant.

Supplemental Figure 3. *rpL13a*-encoded box C/D snoRNAs are induced in palmitate- and H_2O_2-treated C2C12 myoblasts. (Related to Figure 3)

(A-G) C2C12 cells were untreated (UT) or supplemented with 500 μM palmitate for 24 h. (A) ROS generation was quantified by DCF labeling and flow cytometry. (B) *grp78* expression was analyzed by qRT-PCR and normalized to *gapdh* expression. (C) Protein lysates were prepared and analyzed by western blot using rpL13a, actin, and CHOP-10 antibodies. (D) Cell death was quantified by PI staining and flow cytometry. (E) cDNA

synthesis was primed with oligo-dT (mRNA) or random hexamers (total RNA). *rpL13a* expression was analyzed by qRT-PCR, using primers specific to exon 7, and normalized to *gapdh* expression. (F) cDNA synthesis was primed by random hexamers (total RNA). *rpL13a* expression was analyzed by qRT-PCR, using primers specific to intron 2, and normalized to *gapdh* expression. (G) Following treatment with 500 μM palmitate for 24 h, small RNA was harvested and used in RNase protection assay with ^{32}P- labeled murine-specific *rpL13a* snoRNA probes or miR-16 probe as control. Protected probe was separated by PAGE and analyzed by autoradiography. (H) C2C12 cells were untreated (UT) or supplemented with 500 μM of indicated fatty acid for 24 h. Small RNA was harvested and used in qRT-PCR analysis for snoRNAs U32a, U33, and U35a relative to 5S rRNA. All data expressed as mean ± SE (n = 3 to 6). * $p < 0.01$ for fatty acid treated vs. untreated cells.

Supplemental Figure 4. Transient expression of *rpL13a* genomic sequence restores palmitate-induced ROS generation in mutant 6F2 cells. (Related to Figure 4)

(A-C) 6F2 cells were transiently transfected with murine *rpL13a* genomic or cDNA sequences or with empty vector control plasmid. (A) Total RNA was harvested, cDNA synthesis was primed with random hexamers (total RNA), and *rpL13a* expression was analyzed by qRT- PCR, using primers specific to exon 7, and normalized to β-*actin* expression. (B) RNA was harvested and used in RNase protection assay with ^{32}P-labeled murine-specific *rpL13a* snoRNA probes or 18S rRNA probe as control. Protected probe was analyzed by PAGE and autoradiography. (C) Cells were untreated (UT) or supplemented with 500 μM palmitate for 15 h. ROS generation was quantified by DCF labeling and flow cytometry. All data expressed as mean ± SE for 3 independent experiments. * $p< 0.01$. n.s., not significant.

Supplemental Figure 5. ASO-mediated knockdown of unrelated snoRNAs does not confer resistance to palmitate-induced oxidative stress and cell death in C2C12 myoblasts. (Related to Figure 5)

(A-D) C2C12 myoblasts were nucleofected with the indicated ASOs, designed to

specifically target U32a, U33, U35a, U50, U57, or U60 snoRNAs or green fluorescent protein (GFP) as a control. Nucleofected cells were then untreated (UT) or supplemented with 500 μM palmitate for 24 h. (A, B) Total RNA was harvested and used for qRT-PCR analysis of snoRNAs relative to 5S rRNA. (C) ROS generation was quantified by DCF labeling and flow cytometry. (D) Cell death was quantified by PI staining and flow cytometry. All data expressed as mean ± SE for 3 independent experiments. * $p < 0.01$.

Supplemental Figure 6. 2'-O-methylation of rRNAs is unaffected in 6F2 cells. (Related to Figure 6)

WT and 6F2 mutant cells were treated with 500 μM palmitate and total RNA analyzed for pseudouridylation or 2'-O-methylation nucleotide modification of predicted sites using reverse transcriptase primer extension. Autoradiograms show primer extension assays and parallel sequencing for detection of (A) U32a and U33 target sites on 18S rRNA (G1328 and U1326, respectively); (B) U32a target site on 28S rRNA (A1511); (C) U35a target site on 28S rRNA (C5406); (D) unrelated snoRNA target sites on 18S rRNA as controls (snoRNAs Z17a & Z17b target U121; snoRNAs U45a & U45c target A159). For each panel, arrows point to bases in rRNA (numbered according to human rRNA sequence) that are modified and corresponding DNA sequence is shown in the left-most four lanes.

EXTENDED EXPERIMENTAL PROCEDURES

Materials

Palmitate was from Nu-Chek Prep. ^{14}C-palmitate and α-^{32}P-UTP were from Perkin Elmer Life Sciences. Staurosporine, camptothecin and actinomycin D were from Calbiochem. H_2O_2, tunicamycin, and thapsigargin were from Sigma. Fatty acid-free bovine serum albumin was from SeraCare. Propidium iodide, hygromycin D, CM-H_2DCFDA, and DHE were from Invitrogen. All synthetic oligonucleotides were from IDT unless otherwise specified.

Cell culture

CHO-K1 cells (American Type Culture Collection) and CHO-derived cell lines were maintained in high glucose (4.5 mg/ml Dulbecco's modified Eagle's medium and Ham's F-12 nutrient mixture (1:1)) medium with 5% non-inactivated fetal bovine serum, 2 mM L-glutamine, 50 units/ml penicillin G sodium, 50 units/ml streptomycin sulfate, and 1 mM sodium pyruvate. C2C12 myoblasts (American Type Culture Collection) were maintained in high glucose (4.5 mg/ml Dulbecco's modified Eagle's) medium with 10% heat-inactivated fetal bovine serum, 50 units/ml penicillin G sodium, and 50 units/ml streptomycin sulfate. For lipotoxicity experiments, cell culture media was supplemented with 500 μM palmitate complexed to BSA at a 2:1 M ratio, as described previously (Listenberger et al., 2001). To induce ER stress, cells were treated with 2.5 μg/ml tunicamycin or 1 μM thapsigargin. For ROS induction cells were treated with the indicated concentrations of H_2O_2 in PBS containing 0.5 mM $MgCl_2$ and 0.92 mM $CaCl_2$.

Generation of CHO cell mutants

Vesicular stomatitis virus G protein pseudotyped retrovirus was generated by transfecting 293GPG packaging cells with the ROSAβgeo retroviral promoter trap as previously described (Friedrich and Soriano, 1991;Borradaile et al., 2006). CHO cells were transduced with virus at a low multiplicity of infection (1 integration per 10 genomes on average) and mutants were isolated. Number of retroviral insertions within

the mutant cell genome was assessed by Southern blot analysis of genomic DNA using ^{32}P-labeled probe corresponding to the ROSAβgeo proviral sequence.

Cell death and apoptosis assays

Cell death was quantified by membrane permeability to propidium iodide (PI) staining and flow cytometry (Listenberger et al., 2001). Apoptosis was quantified by DNA fragment end labeling (TUNEL assay kit, Calbiochem) and flow cytometry. Analyses were performed on 10^4 cells/sample.

^{14}C Palmitate Uptake Assay

2 x 10^6 cells were resuspended in 1ml PBS containing 500 μM ^{14}C-palmitate complexed to 250 μM BSA and incubated for one minute at 37^{o}C. Cells were then washed with 10 ml PBS containing 0.1% BSA and 500 μM phloretin, filtered, and cell-associated ^{14}C was quantified by scintillation counting. A parallel aliquot of cells was used for quantification of protein by bicinchoninic acid assay (Pierce).

^{3}H Palmitate Oxidation

Palmitate oxidation rates were determined using [9,10-^{3}H]palmitic acid as previously described (Djouadi et al., 2003). WT and 6F2 cells were treated with PBS containing 500 μM ^{3}H-palmitic acid complexed to 250 μM BSA for 2 h. After incubation, PBS-palmitate media was collected and TCA-precipitated. Supernatant was applied to ion-exchange resin (Sigma: DOWEX 1x2 chloride form, 200-400 μm), washed once with distilled H_2O, and eluted ^3H_2O quantified by scintillation counting and normalized to total cellular protein, which was quantified by bicinchoninic acid assay (Pierce).

^{35}S-Cys/Met metabolic labeling

WT and 6F2 cells were untreated (UT) or treated with 500 μM palmitate complexed to 250 μM BSA for 12 h. Cells were then pulse-labeled for 30 min with ^{35}S-Cys/Met in the absence or presence of 10 μM cyclohexamide and TCA-precipitatable proteins were collected. Radiolabel incorporation was quantified by scintillation counting and normalized to total cellular protein, which was quantified by bicinchoninic acid assay

(Pierce).

Identification of trapped gene

The endogenous gene disrupted by retroviral insertion was identified by 5' rapid amplification of cDNA ends (RACE) using an oligonucleotide tag and ROSAβgeo sequences (SMART RACE cDNA amplification kit, Clontech). The 5' RACE product was TA-cloned, sequenced, and blasted against NCBI databases. Direct PCR was used to verify retroviral integration within the rpL13a gene (primer sequences in Supplemental Table 1).

Quantitative real Time PCR (qRT-PCR)

For quantification of total RNA or mRNA, RNA was isolated using TRIzol or TRIzol LS reagent (Invitrogen) and cDNA synthesis was primed oligo dT or random hexamer using SuperScript First-strand Synthesis System (Invitrogen). qRT-PCR was performed using SYBR Green PCR master mixture (Applied Biosystems) and an ABI Prism 7500 Fast Real-Time PCR System (40 cycles, 1 μM template-specific primers). Relative quantification of gene expression was performed using the comparative threshold method as described by the manufacturer.

For quantification of snoRNAs, small RNA was isolated using mirVana miRNA isolation kit (Ambion) or RNAzol®RT (Molecular Research Center). cDNA synthesis was primed using hairpin stem-loop oligos as previously described (Feng et al., 2009), with overhang complementarity to the 3' end of the processed snoRNA. First strand synthesis was performed as above. RT-qPCR and relative quantification was also performed as above, using a snoRNA-specific forward primer and a universal reverse primer from the stem-loop oligos.

snoRNA probe synthesis and RNase digestion

Hamster- and mouse-specific snoRNA probes were generated using Megashortscript kit (Ambion). dsDNA templates were generated for probe for each *rpL13a* snoRNA by PCR amplification of cloned hamster or mouse *rpL13a* genomic sequence templates using primers containing the T7 RNA polymerase promoter and used for *in vitro* RNA

transcription of ^{32}P-labeled snoRNA probes. miR-16 and 18S rRNA probes were synthesized using templates from mirVana miRNA Detection kit and Megashortscript kit, respectively (Ambion). RNA was isolated from cells using either Trizol reagent or mirVana miRNA isolation kit (Ambion) and hybridized to ^{32}P-labeled RNA probes (mirVana miRNA Detection kit) overnight at 42-52^{o}C, followed by RNase digestion and ethanol precipitation. RNA was separated by 10 or 15% polyacrylamide electrophoresis and visualized by autoradiography.

Generation of rpL13a genomic constructs

Hamster genomic, murine mRNA, and murine genomic *rpL13a* sequences were generated from CHO DNA, C2C12 cDNA, and a murine chromosome 7 BAC clone RP24-235B15 (CHORI), respectively, using Platinum *Taq* Hi-Fidelity polymerase (Invitrogen) and cloned into pcDNA3.1. For expression of genomic sequences from the endogenous *rpL13a* promoter, genomic sequence including 1 kb from the endogenous promoter were cloned into a modified pSilencer4.0Hygro vector from which the vector sequences for expression of the insert were removed. QuikChange II Site-directed Mutagenesis Kit (Stratagene) was used to create all mutant *rpL13a* genomic constructs. All constructs were confirmed by sequencing.

Expression of *rpL13a* constructs

Constructs were transfected into 6F2 mutant cells using Lipofectamine 2000 (Invitrogen). Stable clonal mutant lines were isolated using 200 µg/ml hygromycin and assayed for expression of mouse *rpL13a* genomic sequence by qRT-PCR. For expression of *rpL13a* WT and mutant constructs in C2C12 myoblasts, DNA was introduced by nucleofection (Amaxa), and populations of nucleofected cells were selected by growth in G418-containing media for 4 days prior to assays for RNA expression and ROS detection.

snoRNA knockdown *in vitro*

Anti-sense oligos (ASOs) were designed to specifically target murine U32a, U33, U35a, U50, U57, and U60 snoRNA sequences according to Ideue et al (Ideue et al., 2009).

For snoRNA "knock-down" experiments, 10^6 C2C12 myoblasts were nucleofected using Nucleofector Kit V (Amaxa) and a total of 600 pmol of ASO.

Reactive oxygen species detection *in vitro*

Cells were loaded with 1μM (C2C12 cells) or 3 μM (CHO cells) 5-(and-6)-chloromethyl-2',7'-dichlorodihydrofluorescein diacetate, acetyl ester (CM-H_2DCFDA, Invitrogen) in PBS containing 0.5 mM $MgCl_2$ and 0.92 mM $CaCl_2$ at 37 °C for 1 h. Cells were then rinsed with PBS, harvested by trypsinization, and resuspended in culture media. Mean fluorescence was determined by flow cytometry (10^4 cells/sample).

Immunoblot analyses

Whole cell protein lysates were prepared using RIPA buffer (50 mM Tris-Cl, 150 mM NaCl, 1% Nonidet P-40, 0.5% sodium deoxycholate, 0.1% SDS, and 5 mM EDTA) containing 1 mM phenylmethylsulfonyl fluoride, 1x Protease Complete inhibitor mixture (Roche), and 1x phosphatase inhibitors I and II (Sigma). 10μg protein was resolved by 12% SDS-PAGE gel electrophoresis and immunoblotted (nitrocellulose membrane, Millipore) using a polyclonal rabbit α-hamster *rpL13a* antibody (1:2000), monoclonal α-actin (Sigma), or monoclonal α-CHOP-10 antibody (Santa Cruz). Subcellular fractions were isolated by sequential detergent solubilization and analyzed using α-hsp 90 (Stressgen) and α-lamin B1 (Abcam) antibodies. Proteins were visualized using horseradish peroxidase-conjugated secondary antibodies (Jackson ImmunoResearch Laboratories, 1:10,000) and chemiluminescence (PerkinElmer Life Sciences). Band intensities were quantified by densitometry (Quantity One Basic Software).

Mapping of 2'-O-methyl modification by primer extension

Protocol was based on methods from Lowe and Eddy (Lowe and Eddy, 1999). Total RNA from Trizol extraction was annealed with ^{32}P end-labeled primers at 55^{o}C for 4 min. Primer extension reactions were carried out in the presence of 50 mM Tris-Cl (pH 8.6), 60 mM NaCl, 9 mM $MgCl_2$, 10 mM DTT, 1 mM dNTP, and using avian myeloblastosis virus reverse transcriptase for 30 min at 37^{o}C. For rRNA sequencing, ddNTPs were used in individual reactions. For 2'-O-methyl mapping, reactions were

carried in “High” (4 mM), or “Low” (0.004 mM) dNTP concentrations, and 5 mM $MgCl_2$. Reaction products were separated by 6% polyacrylamide electrophoresis (PAGE) and visualized by autoradiography.

In situ hybridization of snoRNA probes

Synthesis and labeling of sense and antisense RNA probes were adapted from Darzacq et al. (Darzacq et al., 2002). C2C12 myoblasts were fixed in PBS containing 3% paraformaldehyde for 10 min, rinsed with PBS and permeabilized with 0.1% Triton X-100 for 10 min at room temperature. Cells were dehydrated serially in 70% ethanol, 95% ethanol, and 100% ethanol and then air-dried. Sequence-specific oligonucleotide probes containing aminoallyl UTP were generated using the FISH Tag RNA Kit (Invitrogen) and labeled with an amine-reactive Alexa Fluor 594 dye. Fluorescent probe (0.5 ng/µl) was denatured in hybridization buffer (2x SSC, 50% formamide, 10% dextran sulfate, 20 mM vanadyl ribonucleoside complexes) for 10 min at 80°C. Cells were incubated with probe/hybridization buffer at 37°C for 10 hr, followed by sequential washes with 2x SSC, 1x SSC, 0.1x SSC all containing 1% SDS for 15 min each at 37°C. Nuclei were counter-stained using SYTOX Green (Invitrogen). Slides were mounted with SlowFade antifade reagent (Invitrogen). Images were captured on a ZEISS LSM 510 META confocal laser scanning microscope using constant pinhole size, detector gain, and offset for each probe.

Mouse model of LPS-mediated oxidative stress and *in vivo* snoRNA knockdown

Female FVB mice were obtained from Charles River Laboratories and housed in our facilities in accordance with our institutionally-approved protocol. Diet was standard chow, and food was withheld at the time of LPS injection. Between 10 and 16 weeks of age, LPS was administered at 8 mg/kg intraperitoneally (IP), and animals were sacrificed 12-24 h later. For *in vivo* knockdown experiments, LNA-modified ASOs were purchased from Exiqon and used to specifically target snoRNAs U32a, U33, and U35a. An ASO targeting GFP was used as a control. Mice were injected IP with a total of 2.5 mg/kg of LNA every other day for a total of three injections, and then dosed with LPS as above at 48 h after the last LNA injection. Individual snoRNA ASO concentrations were

1.25, 0.5, and 0.75 mg/kg per dose, targeting U32a, U33, and U35a respectively. Liver tissue was divided and either snap-frozen in liquid nitrogen (for RNA), fixed in 10% neutral buffered formalin, or frozen in O.C.T. Compound (Tissue-Tek). Experimental procedures were approved by the Washington University Animal Studies Committee and were conducted in accordance with USDA Animal Welfare Act and the Public Health Service Policy for the Humane Care and Use of Laboratory Animals.

In vivo detection of ROS

Formalin-fixed, paraffin-embedded (FFPE) samples or liver frozen in O.C.T were mounted on slides in serial sections by the WUSM Anatomic and Molecular Pathology Core Lab. Detection of superoxide was performed on frozen sections using dihydroethidium (DHE; Invitrogen, Cat# D11347). Sections were incubated with 2 µM DHE for 30 min at 37°C or pre-treated with 200 Units/ml PEG-SOD (Sigma) followed by co-incubation of 2 µM DHE and 200 Units/ml PEG-SOD for 30 min at 37°C to verify the specificity of staining as indicated. For each animal (n = 4 for GFP ASO; n = 5 for SNO ASO), intensity of staining was quantified in three independent fields from each of six sections using ImageJ software. Protein carbonyls were detected by immunoblot using the OxyBlot Protein Oxidation Detection Kit (Chemicon) according to the manufacturer's instructions. Blots were quantified by densitometry using actin as a loading control. Tissue oxysterols (7-ketocholesterol, 7-keto; 3β,5α,6β-cholestantriol, triol) per mg liver protein were quantified using LC/MS/MS as described (Porter et al., 2011).

Supplemental Table 1: Primer and oligonucleotide sequences		
application	**primer/oligo**	**Primer sequence**
direct PCR	rpL13a forward	5'-AT GGC GGA GGG GCA GGT TC-3'
	rpL13a reverse	5'-CA CCA GGA GTC CGT TGG TCG-3'
	ROSAβgeo reverse	5'-CTC AGG TCA AAT TCA GAC GG-3'
qRT-PCR	*rpL13a* exon 7 forward	5'-TGA GGT CGG GTG GAA ATA CC-3'
	rpL13a exon 7 reverse	5'-GGC CTT TTC CTT GCG TTT CT-3'
	rpL13a mouse intron 2 forward	5'-TTA CCT TTG CCT GGG AGT CCA TGA-3'
	rpL13a mouse intron 2 reverse	5'-TGC ATG GGT TGA TCT CAC AGT CGT-3'
	rpL13a hamster intron 2 forward	5'-TGG CAA GCA AGT GCT ACT GGG TAA-3'
	rpL13a hamster intron 2 reverse	5'-TGC ATG GGT TGA TCT CAC AGT CGT-3'
	grp78 forward	5'-GCC TCA TCG GAC GCA CTT-3'
	grp78 reverse	5'-AAC CAC CTT GAA TGG CAA GAA-3'
	β-actin forward	5'-GGC TCC CAG CAC CAT GAA-3'
	β-actin reverse	5'-GCC ACC GAT CCA CAC AGA GT-3'
	GAPDH forward	5'-TCA ACA GCA ACT CCC ACT CTT CCA-3'
	GAPDH reverse	5'-ACC CTG TTG CTG TAG CCG TAT TCA-3'
	36B4 forward	5'-ATCCCTGACGCACCGCCGTGA-3'
	36B4 reverse	5'-TGCATCTGCTTGGAGCCCACGTT-3
RNase protection & FISH	mouse U32 forward	5'-GTT TCA TTC ACC ATT TAC CTT TGC C-3'
	mouse U32 reverse	5'-GTA ATA CGA CTC ACT ATA GGG AGA GGT CCC AAG AAA GCA GGG GCT G-3'
	mouse U33 forward	5'-TGT AGG ACA GGG TAG GCT CTG G-3'
	mouse U33 reverse	5'-GTA ATA GCA CTC ACT ATA GGG AGA CTG AGA ACC CCG TCT GAC CCC-3'
	mouse U34 forward	5'-CTT AGA GCC TCT GCG TCC AGC-3'
	mouse U34 reverse	5'-GTA ATA CGA CTC ACT ATA GGG AGA CTA GCA AGA AGG CCA GCA GGG-3'
	mouse U35 forward	5'-GTT AGA GGT TAG GCT TGT GAG CC-3'
	mouse U35 reverse	5'-GTA ATA CGA CTC ACT ATA GGG AGA AGG ACT CAT CCC CAG CAC GGG-3'
	CHO U32 forward	5'-TAC TGG GTA AGT TTC ATT CAG-3'
	CHO U32 reverse	5'-GTA ATA CGA CTC ACT ATA GGG AGG AAG GAG TCC AGG AGG G-3'
	CHO U33 forward	5'-GGG TGC CAT GGA GAA TGG G-3'
	CHO U33 reverse	5'-GTA ATA CGA CTC ACT ATA GGG AAG CGC TCT TAG CCC AGA TC-3'
	CHO U34 forward	5'-GCA AGC CTA GCT TTC CAC AG-3
	CHO U34 reverse	5'-GTA ATA CGA CTC ACT ATA GGG CTG GGA AGG AGG CTG GTG G-3'

	CHO U35 forward	5’-TTG CAG AGT GGT CTA GGT GG-3’
	CHO U35 reverse	5’-GTA ATA CGA CTC ACT ATA GGG ACA TAT CCC CCT ATA CAG GAG-3’
FISH	mouse U3 forward	5’-TGT AGA GCA CCC GAA ACC AC-3’
	mouse U3 reverse	5’-TCC ACT CAG ACT GCG TTC C-3’
	mouse rpL13a intron 1 forward	5’-AAT TAA CCC TCA CTA AAAG GGA GCA ATA AAC AGG GTG GCT GT-3’
	mouse rpL13a intron 1 reverse	5’-GTA ATA CGA CTC ACT ATA GGG TCC TCA GAT GCT CAA GCA GA-3’
in vitro snoRNA knockdown	U32	5'-mG*mC*mG*mG*mU* G*C*A*T*G*G*G* T*T*G* mA*mU*mC*mU*mC-3'
	U33	5'-mU*mG*mG*mU*mA* G*T*G*C*A*T*G*T*A*G* mA*mG*mU*mC*mA-3'
	U35	5'-mU*mU*mA*mG*mC* C*T*T*T*G*G*C*A*T*T* mA*mU*mC*mG*mG-3'
	GFP	5'-mU*mC*mA*mC*mC* T*T*C*A*C*C*C*T*C*T* mC*mC*mA*mC*mU-3'
	U50	5'-mA*mG*mC*mC*mA* G*A*T*C*C*G*T*A*A*T* mU*mA*mU*mG*mG-3'
	U57	5’-mA*mA*mA*mA*mA*A*A*C*T*G*A*T*T*T*A* mA*mU*mG*mA*mG-3’
	U60	5'-mU*mG*mC*mA*mG*T*T*T*T*C*A*T*A*C*G* mA*mG*mG*mU*mG-3'
Primer extension	U32a, U33: 18S target	5’- GTAACTAGTTAGCATGCCAGAGTCTCG -3’
	U32a: 28S target	5’- GCTACGGACCTCCACCAGAG-3’
	U35a: 28S target	5’-TCGTACTGAGCAGGATTACCATGGC-3’
	Z17a, Z17b, U45a, U45b: 18S target	5’- CCCGTCGGCATGTATTAGCTCTAG-3’
qPCR for snoRNAs	U32a forward	5’-GAGTCCATGATGAGCAACACTCACC-3’
	U33 forward	5’-AGCTTGTGATGAGACATCTCCCACT-3’
	U35a forward	5’-GGCACATGATGTTCTTATTCTCACGATGGT-3’
	U35b forward	5’-GGCAAGTGATGTCTGTTCTCACGATG-3’
	U50 forward	5’-TCTATGATGATCCTATCCCGAAC-3’
	U57 forward	5’-GATGAACGAACTTGGCCTGACCTTC-3’
	U60 forward	5’-CCAAGCCCTGATGAATTAA-3’
	Universal Reverse primer	5’-TCCCGACCACCACAGCC-3’
in vivo snoRNA knockdown	GFP	5'- +T*+C*+A*+C*+C*T*T*C*A*C*C*C*T*C*T*+C*+C*+A*+C* +T-3'
	U32a	5' +G*+C*+G*+G*+T*G*C*A*T*G*G*G*T*T*G*+A*+T*+C*+T *+C 3'
	U33	5' +T*+G*+G*+T*+A*G*T*G*C*A*T*G*T*A*G*+A*+G*+T*+C* +A 3'

	U35a	5' +T*+T*+A*+G*+C*C*T*T*T*G*G*C*A*T*T*+A*+T*+C*+G* +G 3'

*, phosphorothioate linkage; m, 2’-O methyl modified base; +, LNA base