

CPF-CF terminated snoRNAs shuttle through the cytoplasm via an mRNA guard protein-mediated surveillance mechanism

- SUPPLEMENTARY INFORMATION -

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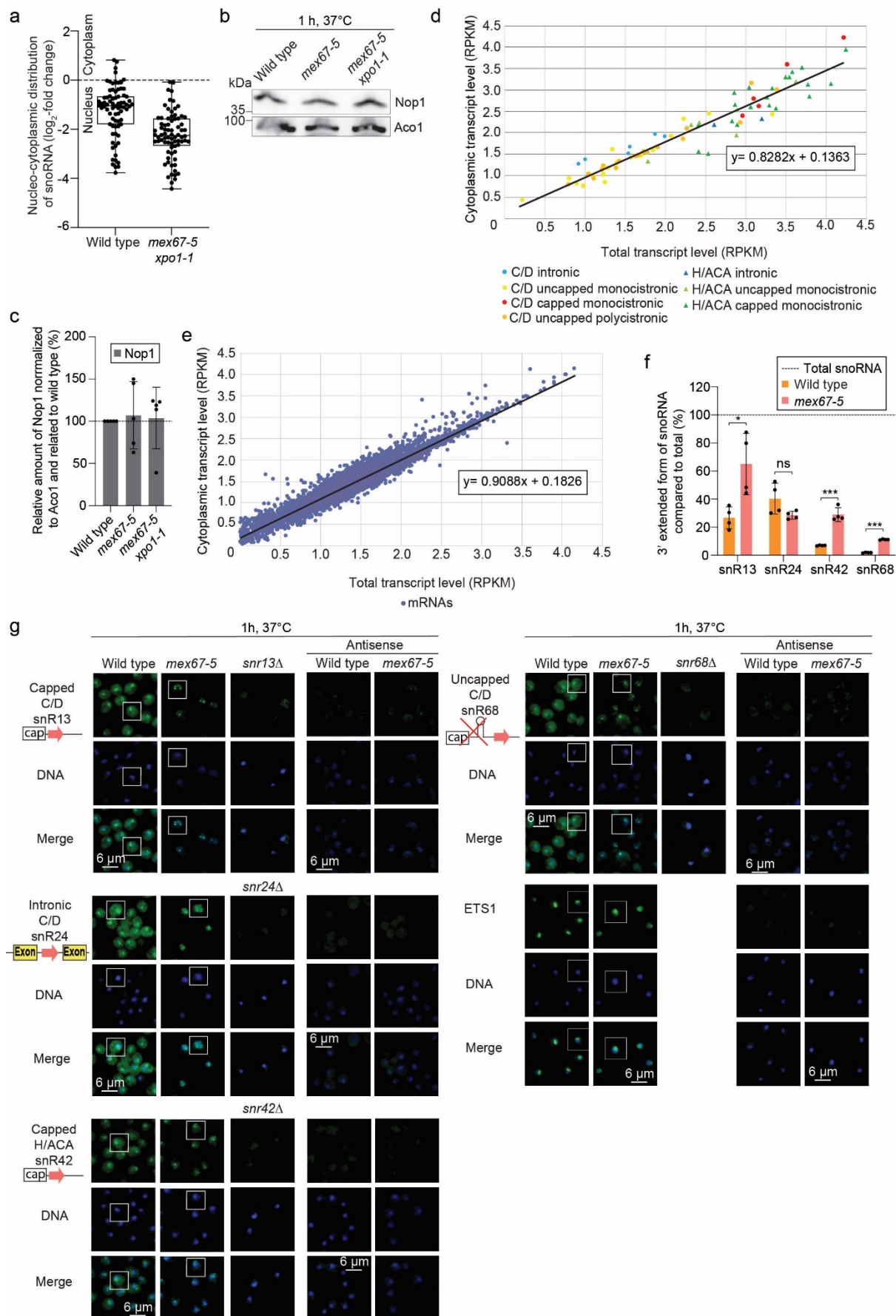
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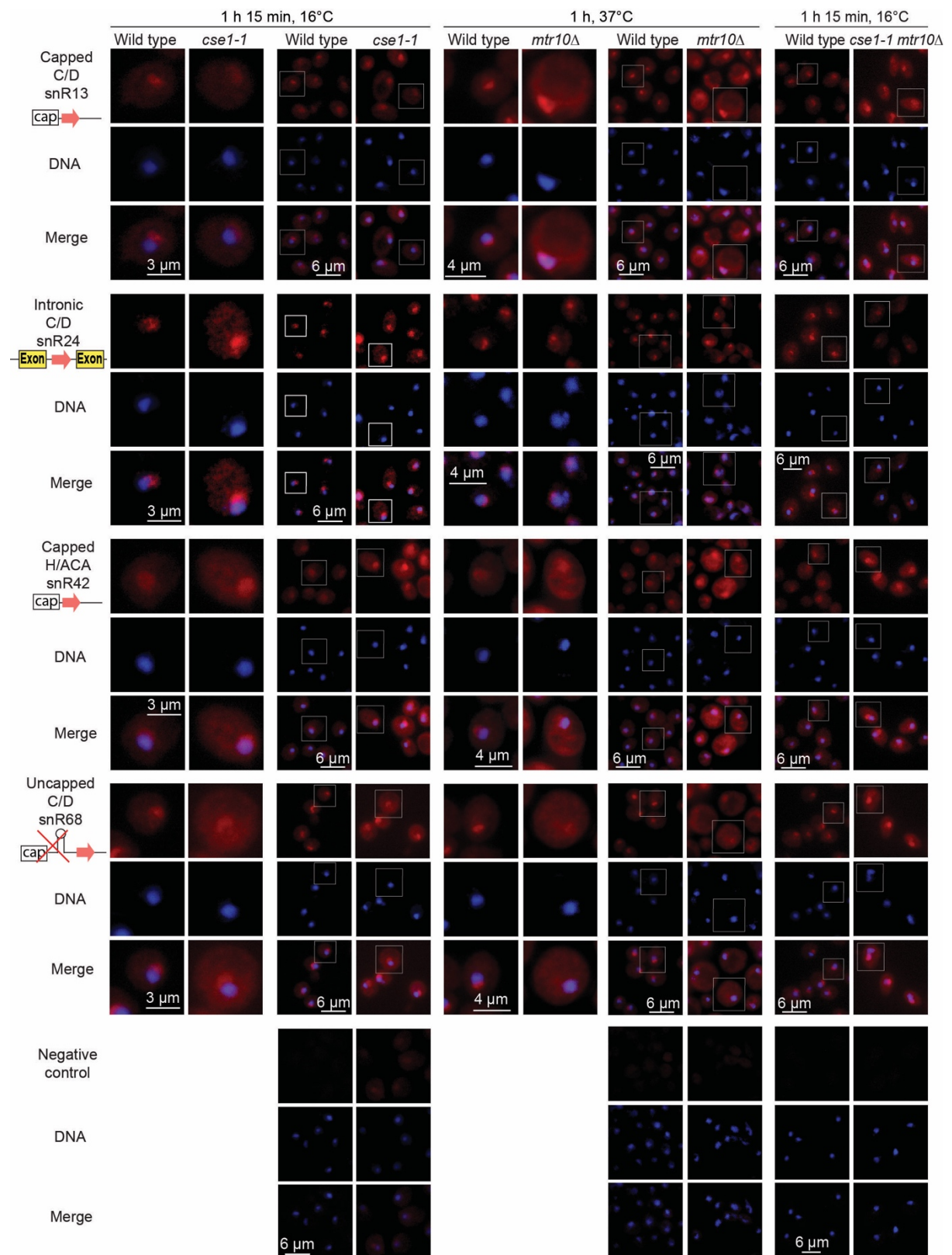
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Short title: snoRNA termination determines shuttling

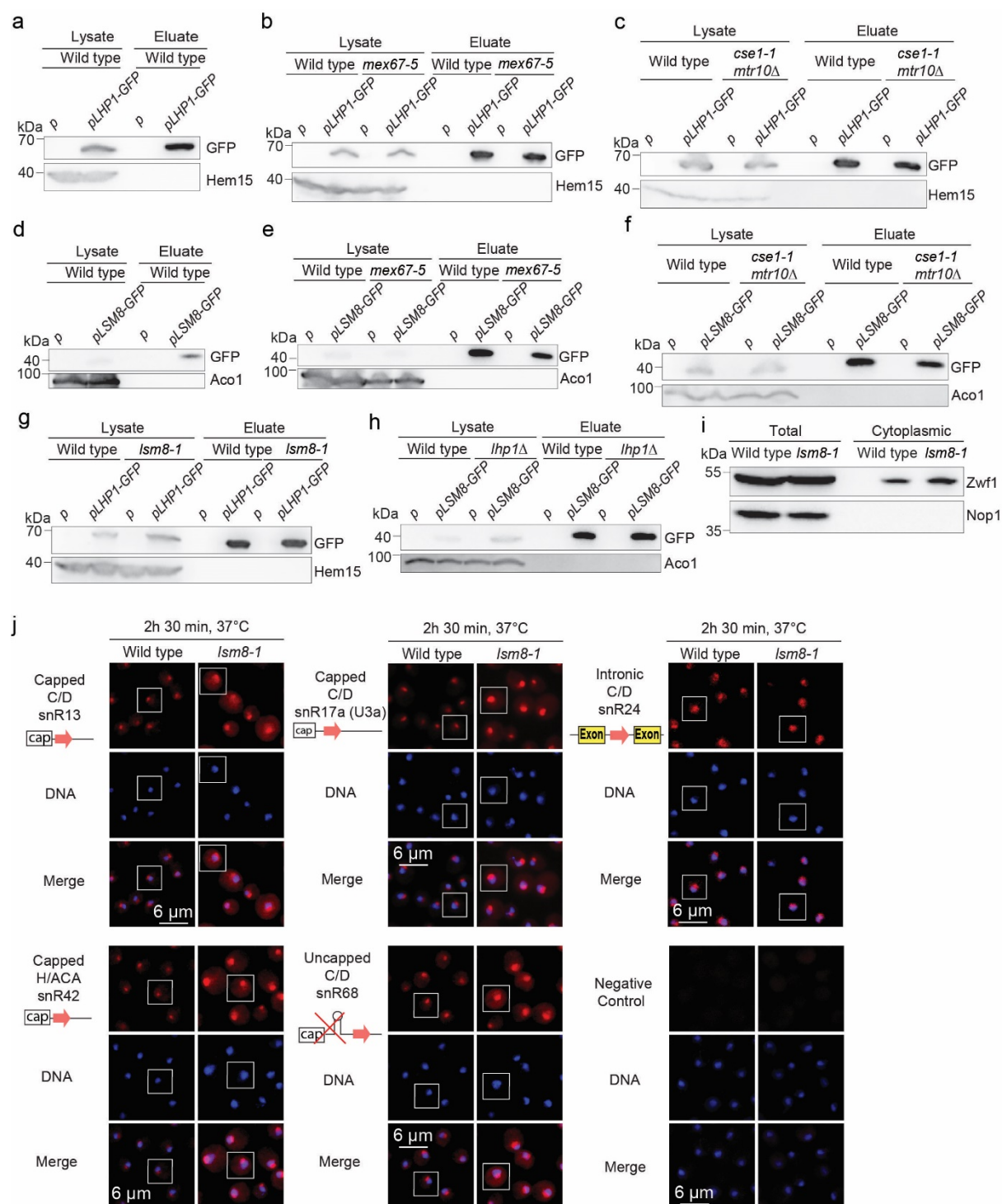
Keywords: snoRNA / RNA modification / rRNA / transcription termination / NNS / CPF-CF



Supplementary Fig. 1: (Related to Fig. 1) (a) The log₂-transformed fold change of the cytoplasmic reads related to total lysate read of snoRNA represents the nucleo-cytoplasmic distribution. The fold change <0 indicates that a snoRNA is more enriched in the nucleus and the fold change >0 indicates that a snoRNA is more enriched in the cytoplasm. Box plot indicates median (centre), 25th-75th percentile (box) and minimum-maximum values (whiskers) as well as all single data points. n=3. (b) Cells of wild type, *mex67-5* and *mex67-5 xpo1-1* were lysed after a temperature shift for 1 h to 37°C. Western blot analysis of Nop1 and mitochondrial Aco1 is shown. kDa=kilodaltons, n=5. (c) Quantification of the relative Nop1 amount normalized to Aco1 using the software Bio-1D is shown. n=5. (d, e) Total and cytoplasmic level of individual snoRNA classified by groups and mRNA obtained from RNA-sequencing data¹ were shown with an equation aligning the distribution. (f) qPCR analysis of total and immature 3'-extended snoRNAs of Fig. 1e was repeated with a standard curve that exhibits the efficiency of each primer pair. After adjusting the Ct value in regard to the primer efficiency, the relative percentage of the 3'-extended snoRNAs is shown against the total snoRNA amounts in wild type and *mex67-5* cells. (g) Overview figure with several cells of Fig. 1f is shown. snoRNA deletion mutants and antisense probes served as negative controls. n indicates the number of biological replicates. Data in bar plots (c, f) are presented as mean +/- SD. Two-tailed student's t-test was used to calculate *p* values. **p* < 0.05; ***p* < 0.01; ****p* < 0.001. Individual data points are represented by black circles.



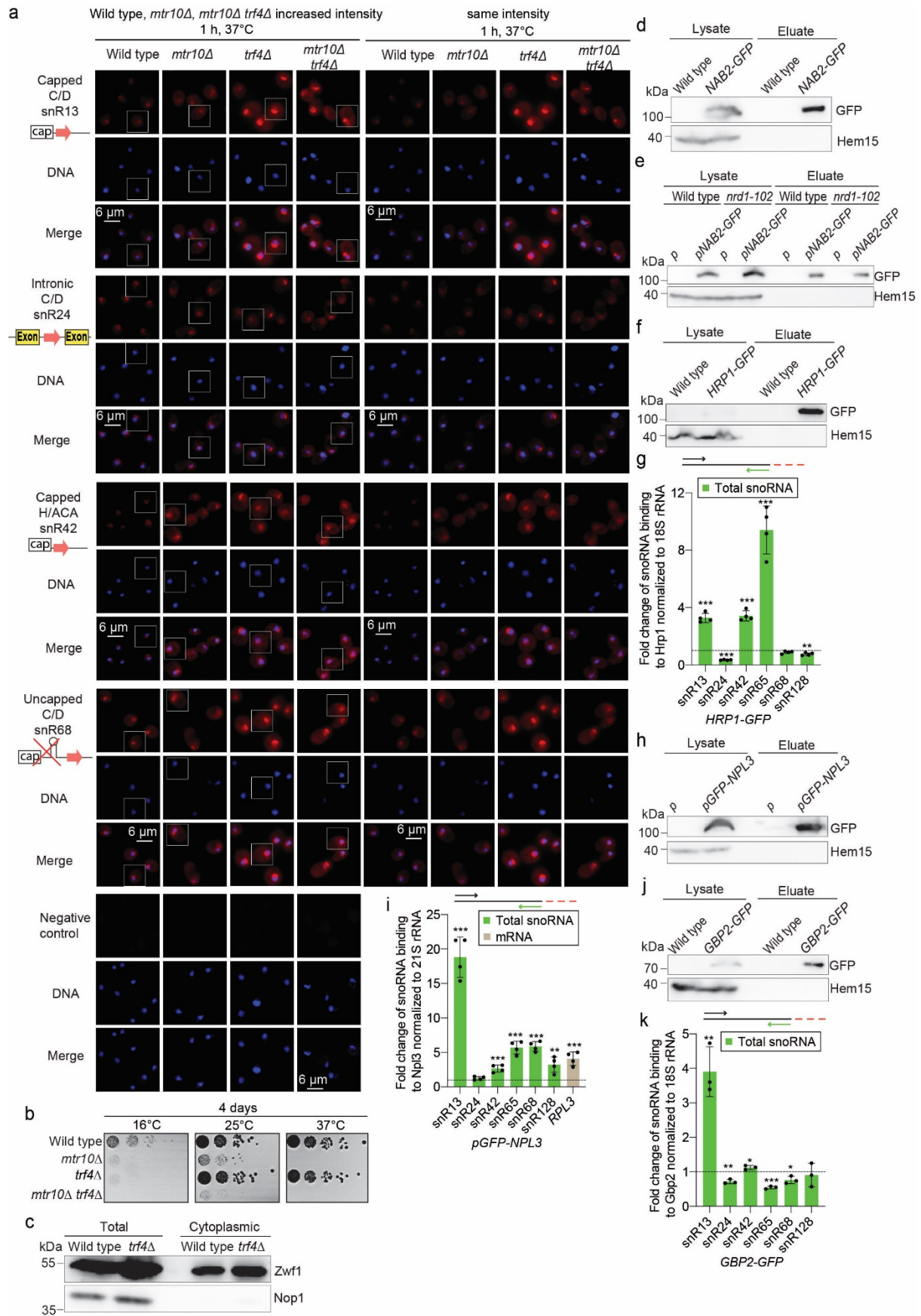
Supplementary Fig. 2: (Related to Fig. 2) FISH experiments were performed in wild type and *cse1-1* or *mtr10Δ* with a temperature shift of 1 h 15 min at 16°C in case of *cse1-1* and 1 h at 37°C in case of *mtr10Δ*. ~30 to 50 nt long Cy3 labelled probes were utilized to hybridize with indicated snoRNAs. The DNA was stained with DAPI. n=3. Overview figure with several cells of Fig. 2a is shown.



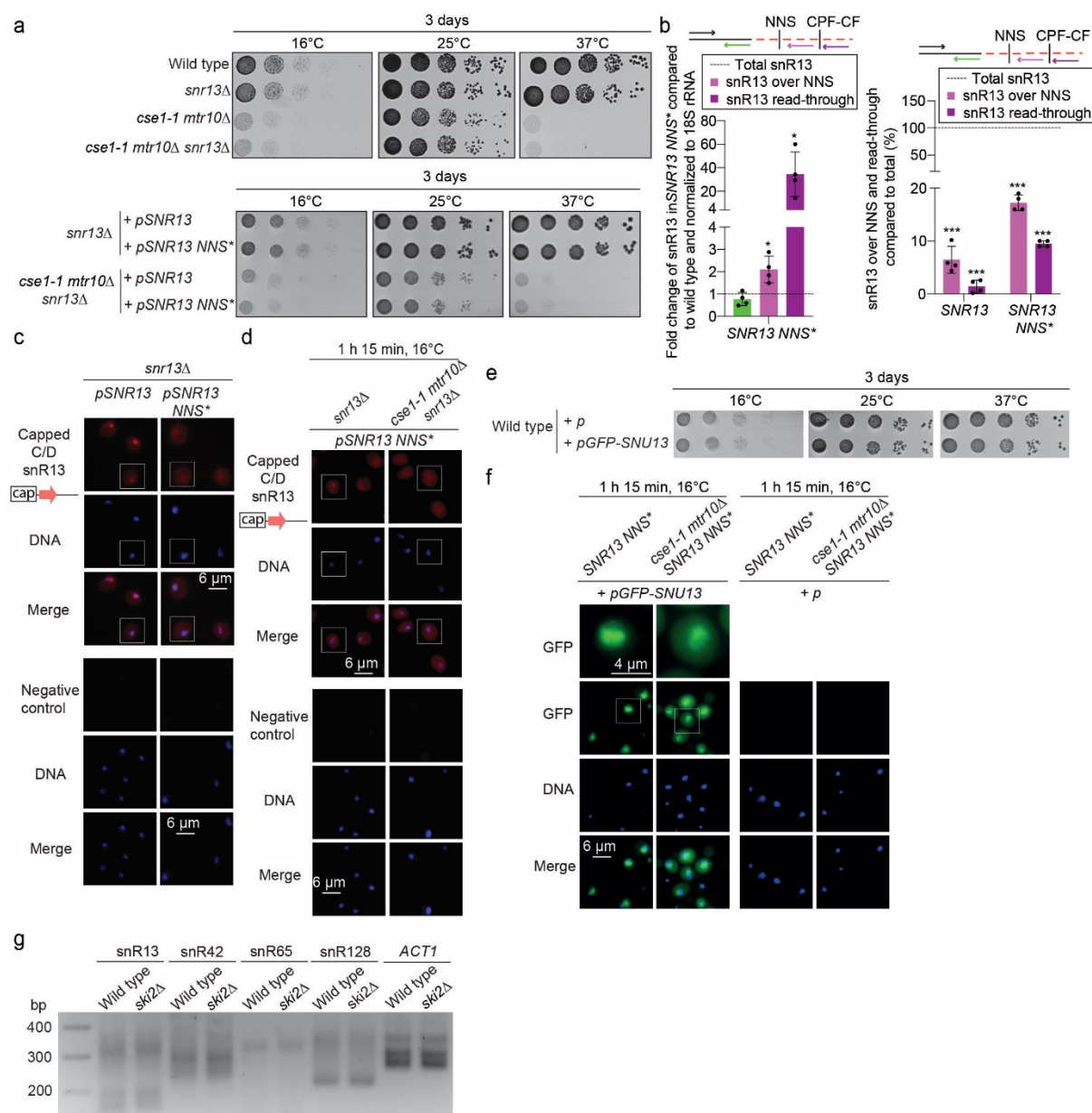
Supplementary Fig. 3: (Related to Fig. 3) (a-h) Precipitation of Lhp1 or Lsm8 was successful. Western blot analysis of Figure 3a-f. Hem15 or Aco1 served as a negative control. kDa=kilodaltons. (i) Fraction separation was successful. Western blot analysis of Fig. 3g. Zwf1 served as a cytoplasmic control and Nop1 as nuclear control. kDa=kilodaltons. (j) Overview figure with multiple cells of Fig. 3h is shown.



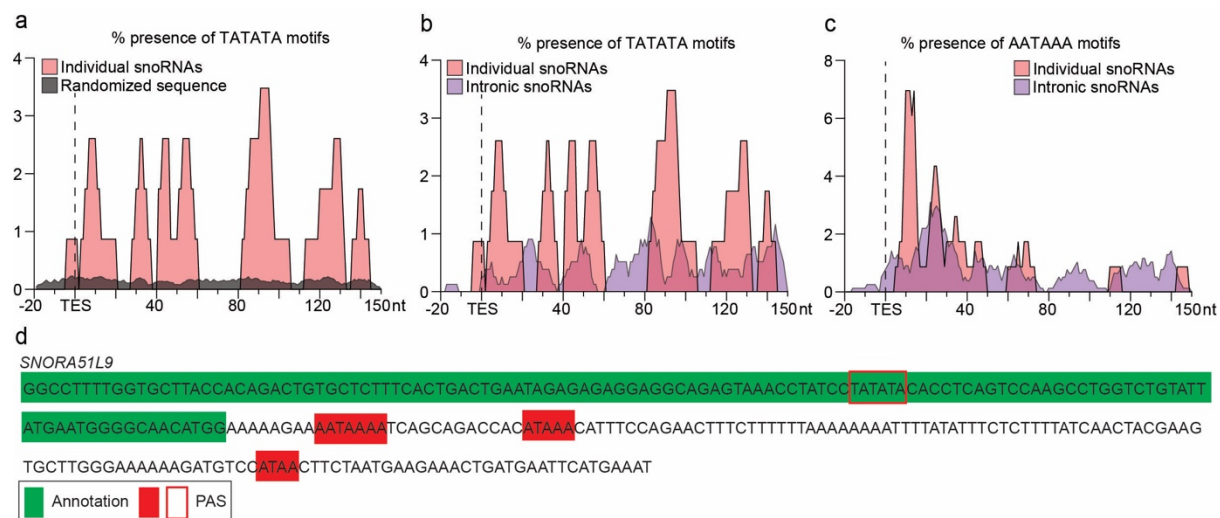
Supplementary Fig. 4: (Related to Fig. 4) Detail transcription termination map of snoRNAs used in this study. Sequence after the mature snoRNA end is shown and the motifs of NNS or CPF-CF termination as well as the assumed or identified termination sites were indicated.



Supplementary Fig. 5: (Related to Fig. 4) (a) Overview figure with several cells of Fig. 4b is shown including figure with the same intensity. (b) *TRF4* shows a genetic interaction with *MTR10* encoding the import receptor. 10-fold serial dilutions of the indicated strains were spotted onto YPD plates that were incubated at the indicated temperatures for four days. n=3. (c) Fraction separation was successful. Western blot analysis of Fig. 4e. Zwfl served as a cytoplasmic control and Nop1 as nuclear control. kDa=kilodaltons. (d, e) Precipitation of Nab2 was successful. Western blot analysis of Fig. 4h and 4i. Hem15 served as a negative control. kDa=kilodaltons. (f-k) SnoRNAs also bind to other guard proteins such as Hrp1, Npl3 and Gbp2. RIP experiments pulling down GFP-tagged Hrp1, Npl3 and Gbp2 were carried out in wild type. (f, h, j) Western blot analysis validates the precipitation of Hrp1(f), Npl3(h) and Gbp2(j) Hem15 served as a negative control. kDa=kilodaltons. (g, i, k) qPCR amplifying the total snoRNA is shown and the binding is normalized either to the 18S rRNA or 21S rRNA in the case of Npl3. (f, g) n=4. (h, i) n=4. (j, k) n=3. n indicates the number of biological replicates. Data in bar plots (g, i, k) are presented as mean \pm SD. Two-tailed student's t-test was used to calculate *p* values. **p* < 0.05; ***p* < 0.01; ****p* < 0.001. Individual data points are represented by black circles.



Supplementary Fig. 6: (Related to Fig. 5 and 6) (a) 10-fold serial dilutions of the indicated strains were spotted onto YPD plates or -ura plates that were incubated at the indicated temperatures for three days. $n=3$. (b) Total, CPF-CF terminated (over NNS) and read-through snR13 in *SNR13* and *SNR13-NNS** were quantified in qPCRs (left) and the relative amounts of CPF-CF terminated (over NNS) and read-through snR13 were compared to the total snR13 (right). $n=4$. (c, d) Overview figure for Fig. 5d and 5e is shown. (e) Wild type cells containing either empty vector (pHK85) or N-terminal GFP tagged Snu13 plasmid (pHK2159) were spotted in 10-fold serial dilution onto -his plates and incubated at the indicated temperatures for three days. $n=3$. (f) GFP microscopy experiments were conducted in *SNR13 NNS** expressed in the wild type or the *cse1-1 mtr10Δ* strains upon a temperature shift for 1 h 15 min to 16°C. The DNA was stained with DAPI. $n=3$. (g) Total RNA isolated from wild type and *ski2Δ* was used for 3'-end PCR. *ACT1* served as control. bp=base pairs, $n=3$. n indicates the number of biological replicates. Data in bar plots (b) are presented as mean \pm SD. Two-tailed student's t-test was used to calculate p values. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$. Individual data points are represented by black circles.



Supplementary Fig. 7: (Related to Fig. 7) (a) Percentual presence of TATATA motifs is shown around the transcript end site (-20 and +150 nt) for individual snoRNAs compared to a set of random sequences. (b) Percentual presence of AATAAA motifs is shown around the transcript end site (-20 and +150 nt) for individual and intronic snoRNAs. (c) Percentual presence of TATATA motifs is shown around the transcript end site (-20 and +150 nt) for individual and intronic snoRNAs. (d) Sequence of the mature SNORA51L9 is shown in green together with its downstream terminator sequence. Putative PAS sites are marked in red.

Supplementary Table 1: Yeast strains

Number	Name	Genotype	Source
HKY36	Wild type	<i>MATα ura3-52 leu2Δ1 his3Δ200</i>	2
HKY82	<i>MTR10</i> shuffle	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 trp1Δ0 ade2Δ0 mtr10::HIS3 + pRS316-URA3-MTR10</i>	3
HKY82	<i>cse1-1</i>	<i>MATα mtr10::HIS3 ura3Δ0; leu2Δ0 trp1Δ0 his3Δ1 ade2Δ0</i>	4
HKY314	Wild type	<i>MATα ura3Δ0 leu2Δ0 his3Δ met15Δ0</i>	Euroscarf
HKY332	<i>nrd1-102</i>	<i>MATα nrd1::kanMX ura3Δ0 his3Δ1 leu2Δ0 met15Δ0 +pnrd1-102 LEU2 CEN</i>	5
HKY502	<i>GBP2-GFP</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 met15Δ0 GBP2-GFP:HIS3MX6</i>	Invitrogen
HKY644	<i>mex67-5</i>	<i>MATα mex67::HIS3 ura3Δ0 leu2Δ0 his3Δ1 trp1Δ0 ade2Δ0 + pUN100-mex67-5 LEU2 CEN</i>	6
HKY682	<i>npl3Δ</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 npl3::KanMX4</i>	7
HKY1112	<i>trf4Δ</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 met15Δ0 trf4::kanMX4</i>	Euroscarf
HKY1266	<i>MEX67-GFP</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 met15Δ0 MEX67-GFP:HIS3MX6</i>	Invitrogen
HKY1475	<i>NAB2-GFP</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 met15Δ0 NAB2-GFP:HIS3MX6</i>	Invitrogen
HKY1610	<i>cdc28-13</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 met15Δ0 cdc28-13::kanMX4</i>	8
HKY1854	<i>lsm8-1</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 trp1Δ0 lys2Δ0 lsm8-1</i>	9
HKY1861	<i>HRP1-GFP</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 met15Δ0 HRP1-GFP:HIS3MX6</i>	Invitrogen
HKY1906	<i>lhp1Δ</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 lhp1::kanMX4</i>	Euroscarf
HKY2035	<i>cft2-1</i>	<i>MATα cft2-1::kanMX4 ura3Δ0 leu2Δ0 his3Δ1 met15Δ0</i>	Euroscarf
HKY2087	<i>cse1-1 mtr10Δ</i>	<i>MATα ura his cse1-1 mtr10::kanMX4</i>	10
HKY2328	<i>snr13Δ</i>	<i>MATα ura3-52 leu2Δ1 his3Δ200 snr13Δ</i>	This study
HKY2349	<i>snr42Δ</i>	<i>MATα ura3-52 leu2Δ1 his3Δ200 snr42Δ</i>	This study
HKY2524	<i>NAB2-GFP snr13Δ</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 met15Δ0 NAB2-GFP:HIS3MX6 snr13Δ</i>	This study
HKY2525	<i>HRP1-GFP snr13Δ</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 met15Δ0 HRP1-GFP:HIS3MX6 snr13Δ</i>	This study
HKY2528	<i>mex67-5 snr13Δ</i>	<i>MATα mex67::HIS3 ura3Δ0 leu2Δ0 his3Δ1 trp1Δ0 ade2Δ0 snr13Δ + pUN100-mex67-5 LEU2 CEN</i>	This study
HKY2565	<i>cse1-1 mtr10Δ snr13Δ</i>	<i>MATα ura his cse1-1 mtr10::kanMX4 snr13Δ</i>	This study
HKY2829	<i>mtr10Δ trf4Δ</i>	<i>MATα ura3Δ0 leu2Δ0 his3Δ1 ade2Δ0 mtr10::HIS3 trf4::kanMX4</i>	This study
HKY2962	<i>snr24Δ</i>	<i>MATα leu2Δ1 his3Δ200 snr24::URA3</i>	This study
HKY2972	<i>snr68Δ</i>	<i>MATα leu2Δ1 his3Δ200 snr68::URA3</i>	This study

Supplementary Table 2: Plasmids

Number	Genotype	Source
pHK85	<i>HIS3; CEN; AMP^R</i>	11
pHK86	<i>TRP1; CEN; AMP^R</i>	11
pHK87	<i>LEU2; CEN; AMP^R</i>	11

pHK88	<i>URA3; CEN; AMP^R</i>	11
pHK356	<i>URA3 CEN AMP^R P_{NAB2}: NAB2-GFP</i>	This study
pHK418	<i>P_{NPL3}: GFP-NPL3; LEU2; CEN; AMP^R</i>	12
pHK1711	<i>P_{LSM8}: LSM8-GFP; URA3; CEN; AMP^R</i>	13
pHK1748	<i>P_{LHP1}: LHP1-GFP; URA3; CEN; AMP^R</i>	13
pHK1894	<i>P_{SNR13}: SNR13; CEN; URA3; AMP^R</i>	This study
pHK1917	<i>P_{SNR13}: SNR13, GTAG -> GCCG (139/140*), GTAA -> GCCA (139/140*), TCTT -> TACT (147/148*) (160/161*); CEN; URA3; AMP^R</i>	This study
pHK2159	<i>P_{SNU13}: GFP-SNU13; HIS3; CEN; AMP^R</i>	This study
pHK2235	<i>P_{SNU13}: GFP-SNU13; TRP1; CEN; AMP^R</i>	This study

Supplementary Table 3: Primers for cloning

Construct	Number	Sequence	Name
pHK1711	HK3731	5'-CACAATATTTCAAGCTATACCAAGCATACAA TAAGCATCGATGTCAGCCACCTTGAAA-3'	<i>LSM8</i> forward
	HK3732	5'-CAGTGAAGAGTTCTTCTCCTTTGCTAGCCATA GCTCGAGATTTTGTCTTTGATTTCGTACACTT-3'	<i>LSM8</i> reverse
	HK3880	5'-GTCGCGCCATTTCGCCATTTCAGGCTGCGCAACT GTTGGGAATTCCGTCTCATGCTGCTT-3'	<i>LSM8</i> promoter forward
	HK3881	5'-CAACTCTTTTATTTAAGTAGTCTTTCAAGGTG GCTGACATGGTGCACGATAGTGTGTTCT-3'	<i>LSM8</i> promoter reverse
pHK1748	HK3747	5'-CACAATATTTCAAGCTATACCAAGCATACAATA AGCATCGATGTCTGAAAAACCACAACAAGAG-3'	<i>LHP1</i> forward
	HK3748	5'-CAGTGAAGAGTTCTTCTCCTTTGCTAGCCATA GCTCGAGACTCCTTGTGCTCCTCATCG-3'	<i>LHP1</i> reverse
	HK3905	5'-GTCGCGCCATTTCGCCATTTCAGGCTGCGCAACTG TTGGGAA-3'	<i>LHP1</i> promoter forward
	HK3906	5'-GTGGTTTCTCTTGCTCCTCTTGTTGTGGTTTTTCA GACATTACTTCTTTGCAGATGCTACTTTAG-3'	<i>LHP1</i> promoter forward
pHK1894 pHK1917	HK4414	5'-TCTATACTTTAACGTCAAGGAGAAAAAACTATA GAATTCAAGGAAGTTTTTCCCTTTTATATG -3'	<i>SNR13</i> forward
	HK4415	5'-ACAAAAGCTGGGTACCTTCACATGTTCCGCA GATTTTGATACTCCAGTATTATGGATGG -3'	<i>SNR13</i> reverse
	HK4601	5'-CTACCTTTTTTTACTTTTATCTGACCTTTTAAC TTCCCCGCCGAAAATACTAGTAATCCTTCT-3'	<i>SNR13</i> 1. NNS mutation forward
	HK4602	5'-CGCATTATATATGCAGCGCTACGATACAATG TAAGAAGGATTACTAGTATTTTCGGC-3'	<i>SNR13</i> 1. NNS mutation reverse
	HK4669	5'-TGACCTTTTAACTTCCCCGCCGAAAATACTAG CCATCCTTACTACATTGTAT-3'	<i>SNR13</i> 2. NNS mutation forward
	HK4670	5'-GAAAATTTTACGCATTATATATGCAGCGCTA CGATACAATGTAGTAAGGATGGCTAGTAT-3'	<i>SNR13</i> 2. NNS mutation reverse
	HK4653	5'-GTGGCGGCCGCTCTAGATGATTGTATTGTCAT TGTAGGAGGT-3'	<i>SNR13</i> promoter forward
	HK4654	5'-ACTCATATTCATCATATAAAAAGGAAAAAACTTC CTTGAATTCGACCTTTAAAAAATCCAATTAACA-3'	<i>SNR13</i> promoter reverse
pHK2159	HK6213	5'- TGGCGGCCGCTCTAGAACTAGTGGATCCCCCGG GCTGCAGGCAGCTTCTGAATCACTAAAG-3'	<i>SNU13</i> promoter forward
	HK6214	5'-GACAACTCCAGTGAAGAGTTCTTCTCCTTTGCTA GCCATTTTAAATATAGTTTTTATAGTATGAAATCTGG-3'	<i>SNU13</i> promoter reverse
	HK6215	5'- TGGGATTACACATGGCATGGATGAACATACAAA GGATCCATGTCTGCCCCAAACCCAAA-3'	<i>SNU13</i> forward

	HK6216	5'-TACCGGGCCCCCCTCGAGGTCGACGGTATCGAT AAGCTTATTCGGTTACCTAATAAGATAATCTC-3'	<i>SNU13</i> reverse
pHK2235	HK6683	5'- ACCATAACCACCTTTTCTTTTCTATTACTCTTGGC CTCCTACGACATTACTATATATATAATATAGGAAG-3'	<i>TRP1</i> forward
	HK6684	5'- ATTCACACCGCATACTGCAGCTTTAAATAATCG GTGTCACATTCTTAGCATTCTTGACG-3'	<i>TRP1</i> reverse

Supplementary Table 4: Primers for knock out generation

Strains	Number	Sequence	Name
HKY2328 HKY2565	HK4188	5'-AAAATTTTACGCATTATATATGCAGCGCTACGA TACAATGCCAGTCACGACGTTGTAAAACG-3'	<i>SNR13</i> knockout FW
	HK4229	5'-GCCATATTGGTCGTAACACAAAAATGCCCAATGG AGCCTGGGGATCTATCGATCTCGACAACCCG-3'	<i>SNR13</i> knockout RV
HKY2524 HKY2525	HK5005	5'-GCCATATTGGTCGTAACACAAAAATGCCCAAT GGAGCCTGGCTATCGATCTCGACAACCCCTCG-3'	<i>SNR13</i> knockout FW
	HK5006	5'-AAAATTTTACGCATTATATATGCAGCGCTACG ATACAATGGTGAATTCGAGCTCGGTACCCG-3'	<i>SNR13</i> knockout RV
HKY2528	HK5009	5'-GCCATATTGGTCGTAACACAAAAATGCCCAATGG AGCCTGGCGACAACCCCTCGAGGATCT-3'	<i>SNR13</i> knockout FW
	HK5010	5'-AAAATTTTACGCATTATATATGCAGCGCTACGAT ACAATGGTAAAACGACGGCCAGTGAA-3'	<i>SNR13</i> knockout RV
HKY2962	HK6445	5'-CAATCGAGTAGAAGAAGAAAAAGTGGATTTGT GTATGCCATGGATCTATCGATCTCGACAACCCG -3'	<i>SNR24</i> knockout FW
	HK6446	5'-ACATAACGAGTAAAGAGAAGAGCAGAGTAATG CTAAACCACCCAGTCACGACGTTGTAAAACG-3'	<i>SNR24</i> knockout RV
HKY2972	HK6447	5'-TTTCTTTTCAGTTTCCTCCGTTTATTTTTTGT TGGATTGGATCTATCGATCTCGACAACCCG-3'	<i>SNR68</i> knockout FW
	HK6448	5'-TTTTTCAGTTTAAAGTTTCCAATTTTTTTGTAAAG ATGAACCCCCAGTCACGACGTTGTAAAACG-3'	<i>SNR68</i> knockout RV

Supplementary Table 5: Primers for qPCR

Target	Number	Sequence	Name
<i>SNR13</i>	HK3530	5'-GAAGTTTTTTCCTTTTTATATGATGAA-3'	Forward
	HK3531	5'-GGTCAGATAAAAAGTAAAAAAGGTAGC-3'	Reverse
	HK3533	5'-TTACTAAGATTTTCTACGGGGAAG-3'	Reverse 3'-extended unprocessed form
	HK5084	5'- ATTTTACGCATTATATATGCAGCG-3'	Reverse over NNS termination site
	HK5013	5'-CGCTTCCGTGTCTCTTGTCC-3'	Reverse read-through
<i>SNR17a</i> (<i>U3a</i>)	HK3542	5'-GTTGATGAGTCCATAACCTTT-3'	Forward
	HK3545	5'-CAATTTAGAAAAGGAAAAAAGTGG-3'	Reverse 3'-extended unprocessed form
<i>SNR24</i>	HK2797	5'-TCAAATGATGTAATAACATATTTGCTACTTC- 3'	Forward
	HK2798	5'-CATCAGAGATCTTGGTGATAATTGG-3'	Reverse
	HK3405	5'-GAGAAGAGCAGAGTAATGCTAAACC-3'	Reverse 3'-extended unprocessed form
<i>SNR42</i>	HK2799	5'-TTGTGATGCTTTAGGGAGCC-3'	Forward
	HK2800	5'-ATCCTTTCTCTATCTCACCCTG-3'	Reverse
	HK3413	5'-CATACTATAAATACGTATATATAACAAAG TAACCATCAAG-3'	Reverse 3'-extended unprocessed form

	HK3786	5'-AATATTGATCTCAATGTCATCAGCTG-3'	Reverse over NNS termination site
SNR65	HK4753	5'-ACACAATTTATGCTAGATAGTATCTGAAAG-3'	Forward
	HK4754	5'-GTTTCAGAGATTTAAACTGTGCGAAAACACT-3'	Reverse
	HK4756	5'-CGCCCATAATCAAATCAGCTCATAC-3'	Reverse over NNS termination site
SNR68	HK3402	5'-ATCATGATGAGCATTATTTTACTGCG-3'	Forward
	HK3403	5'-CAGCAAATCTGTTAAGAGTCAATTTCC-3'	Reverse
	HK3411	5'-AGTTTTCCAATTTTTTTGTAAGATGAACC-3'	Reverse 3'-extended unprocessed form
	HK3787	5'-GACCATATTATCAAGAGCTTCCTGCT-3'	Reverse over NNS termination site
SNR128 (U14)	HK4618	5'-TCACGGTGATGAAAGACTGGTTC-3'	Forward
	HK4619	5'-AAGAGCGGTCACCGAGAGTA-3'	Reverse
	HK5164	5'-ACGCATTTTTTAAGCTAGCAGGATTCA-3'	Reverse over NNS termination site
18S rRNA	HK1396	5'-CATGGCCGTTCTTAGTTGGTGG-3'	Forward
	HK1397	5'-ATTGCCTCAAACCTCCATCGGC-3'	Reverse
21S rRNA	HK3089	5'-AGTTACGCTAGGGATAACAGGG -3'	Forward
	HK3090	5'- TGACGAACAGTCAAACCCTTC -3'	Reverse
RPL3	HK829	5'-CCAGGTTCTAAGTTCCACAAGCG-3'	Forward
	HK830	5'-CCATCTTGAGCGTACTTGGCAG-3'	Reverse
ZWF1	HK3060	5'-GGTGAAGCCGATGACTCTAAGG-3'	Forward
	HK3061	5'-GGCCAGATAGAAGAGACGGT-3'	Reverse

Supplementary Table 6: Primers for digoxigenin labeled RNA probes

Target	Number	Sequence	Name
SNR13	HK3303	5'-TAATAGGACTCACTATAGGGGAAG TTTTTCCTTTTATATGATG-3'	Forward antisense
	HK3304	5'-GGTCAGATAAAAGTAAAAAAAGGTAGC-3'	Reverse antisense
	HK3324	5'-GGAAGTTTTTTCCTTTTATATGATG-3'	Forward sense
	HK3325	5'-TAATAGGACTCACTATAGGGGTCAGATAAAAGTAAAAAAAGGTAGC-3'	Reverse sense
SNR24	HK3305	5'-TAATAGGACTCACTATAGGTCAAATGATGTAATAACATATTTGC-3'	Forward antisense
	HK3306	5'-TTCATCAGAGATCTTGGTG-3'	Reverse antisense
	HK2607	5'-TCAAATGATGTAATAACATATTTGC-3'	Forward sense
	HK2608	5'-TAATAGGACTCACTATAGGTTTCATCAGAGATCTTGGTG-3'	Reverse sense
SNR42	HK3307	5'-TAATAGGACTCACTATAGGGATGCTTTAGGGAGCCTATTG-3'	Forward antisense
	HK3308	5'-GGATTACCTCAGCACCAAC-3'	Reverse antisense
	HK2605	5'-GATGCTTTAGGGAGCCTATTG-3'	Forward sense
	HK2606	5'-TAATAGGACTCACTATAGGGGATTACCTCAGCACCAAC-3'	Reverse sense
SNR68	HK3309	5'-TAATAGGACTCACTATAGGATGATGAGCATTATTTTACTGCGT-3'	Forward antisense

	HK3310	5'-TTTCAGCAAATCTGTTAAGAGTC-3'	Reverse antisense
	HK3326	5'-ATGATGAGCATTATTTTACTGCGT-3'	Forward sense
	HK3327	5'-TAATAGGACTCACTATAGGTTTCAGCAAATCTGTTAAGAGTC-3'	Reverse sense

Supplementary Table 7: Primers labeled with Cy3 dye

Target	Number	Sequence
<i>SNR13</i>	HK3431	5'-CY3-GAGTTTTTCCACACCGTTACTGATTTGGCAAAGCCAAACAGCAACTCGA-CY3-3'
<i>SNR17a (U3a)</i>	HK3563	5'- CY3-GTTTCTCACTCTGGGGTACAAAGGTTATGGGACTCATCAACCAAGTTGGA-CY3-3'
<i>SNR24</i>	HK2802	5'- CY3-GTGATAATTGGTATGTCTCATTCGGAAGTCAAAGTTCCATCTGAAGTAGC-CY3-3'
<i>SNR42</i>	HK2801	5'- CY3-CGATGGTTTTTAAAGATGGATTACCTCAGCACCAACAGTTACACTATTCGG-CY3-3'
<i>SNR68</i>	HK3562	5'- CY3-GCCCCCGTCAATACGATAACGCAGTAAAAT-CY3-3'

Supplementary Table 8: Primers for analytical PCR

Number	Sequence	Name
HK2089	5'-ATTGGTGGTTCTATCTTGGC-3'	<i>ACT1</i> forward for 3'-end PCR
HK4375	5'- GTTCATCCATGCCATGTGTAATCC-3'	3'-end PCR adapter reverse
HK4683	5'-TTTGTATAGTTCATCCATGCCATG TGTAATCCTTTTTTTTTTTTTTTTTC-3'	cDNA for 3'-end PCR with adapter
HK4684	5'-TTTGTATAGTTCATCCATGCCATG TGTAATCCTTTTTTTTTTTTTTTTG-3'	cDNA for 3'-end PCR with adapter
HK4685	5'-TTTGTATAGTTCATCCATGCCATG TGTAATCCTTTTTTTTTTTTTTTTIA-3'	cDNA for 3'-end PCR with adapter
HK5334	5'-GATGAATATGAGTGCATTTGGCTCGAG-3'	<i>SNR13</i> forward for 3'-end PCR
HK5539	5'-GTGTAAGTGTGGTGCTGAGGTAATCC-3'	<i>SNR42</i> forward for 3'-end PCR

Supplemental Table 9: Antibodies for western blot and FISH

Antibody (organism)	Dilution (method)	Source
Anti-Aco1 (rabbit)	1:4000 (western blot)	U. Mühlenhoff, Marburg (Germany)
Anti-Digoxigenin-FITC (sheep)	1:200 (FISH)	Roche
Anti-GFP (mouse)	1:5000 (western blot)	Thermo Fisher Scientific
Anti-GFP (rabbit)	1:4000 (western blot)	ChromoTek GmbH
Anti-Hem15 (rabbit)	1:5000 (western blot)	U. Mühlenhoff, Marburg (Germany)
Anti-Mex67 (rabbit)	1:10000 (western blot)	Davids Biotechnology
Anti-mouse IgG-HRP (goat)	1:10000 (western blot)	Dianova
Anti-Nop1 (mouse)	1:4000 (western blot)	Santa Cruz
Anti-rabbit IgG-HRP (goat)	1:10000 (western blot)	Dianova
Anti-Yra1 (goat)	1:2000 (western blot)	Santa Cruz
Anti-Zwf1 (rabbit)	1:4000 (western blot)	U. Mühlenhoff, Marburg (Germany)

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