Table SI. Plasmids used and generated in this study

Plasmid	Description	Source
pPS1152	*NPL3, URA3, Amp^R, 2μ*	Lee et al, 96 [1]
pAM420	*GFP-npl3-RK1-15, pGal. URA3, AmpR, 2μ*	McBride, 05 [2]
pPS879	*GFP-npl3 F160L, URA3, Amp^R, 2μ*	Lee et al, 96 [1]
pBS0A1	hnRNP A1-GFP	Zhu, 02 [3]
pJL215	pRS316 with *NPL3* promoter	This study
pJL226	hnRNP A1-6xHis	This study
pJL227	*NPL3*-6xHis	This study
pJL228	*npl3*-F160L-6xHis	This study
pJL229	*npl3*-RK1-15-6xHis	This study
pJL230	*npl3*-F160L L225S G241N E244K-6xHis	This study
pJL231	*npl3*-L225S G241N E244K-6xHis	This study

References:

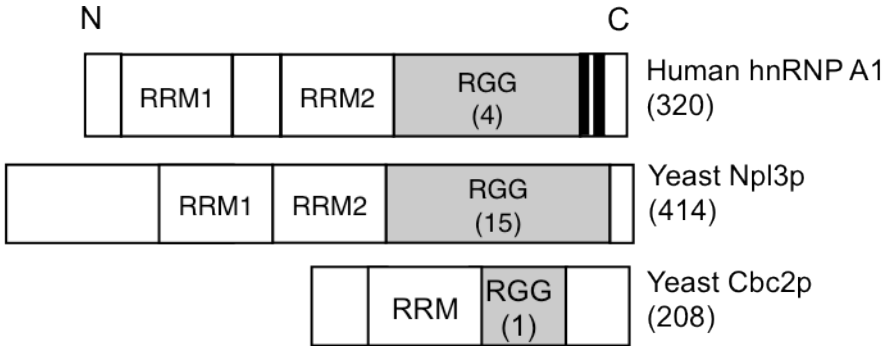
[1] M. S. Lee, M. Henry and P. A. Silver, A protein that shuttles between the nucleus and the cytoplasm is an important mediator of RNA export, Genes Dev 10 (1996) 1233-1246.
[2] A. E. McBride, J. T. Cook, E. A. Stemmler, et al., Arginine methylation of yeast mRNA-binding protein Npl3 directly affects its function, nuclear export, and intranuclear protein interactions, J Biol Chem 280 (2005) 30888-30898.
[3] D. Zhu, G. Xu, S. Ghandhi, et al., Modulation of the expression of p16INK4a and p14ARF by hnRNP A1 and A2 RNA binding proteins: implications for cellular senescence, J Cell Physiol 193 (2002) 19-25.

Table SII. Primers used to clone *NPL3* and *CBC2* genes. Oligonucleotides for gel shifts are also listed. 3137 sequences were derived from chromosome I-L

	Primer/Oligonucleotide	**Sequence (5' - 3')**
1	Npl3 NORF1 Sac For	AAAGAGCTCGGAGTGCACCAAATCTACCGCAGTG
2	Npl3 NORF1 Eag Mlu Rev	ATATCGGCCGACGCGTATCCTTATGGTTTTAGCGTAATTGC
3	Npl3 ATG Mlu For	AAAAACGCGTATGTCTGAAGCTCAAGAAACTCACGTAGAG
4	Npl3 6xHIS Bam Rev: (BamHI)	ATATGGATCCTTAGTGATGGTGGTGATGGTGCCTGGTTGGTG ATCTTTCACGTGGAGCATCTCTGGTTCTG
5	A1 ATG Mlu For	AAAAACGCGTATGTCTAAGTCAGAGTCTCCTAAAGAGCCCGA ACAGCTGA
6	A1 6xHIS Bam Rev	AAAAGGATCCTTAGTGATGGTGGTGATGGTGAAATCTTCTGC CACTGCCATAGCTACTGCTGCTGCTGGAA
7	3137 For	TGTGTGTGTGTGGGTGTGGTGTGGTGTGTGGTGTGG
8	3137 Rev	CCACACCACACACCACACCACACCCACACACACACA
9	3137 RNA	UGUGUGUGCGUGGGUGUGGUGUGGUGUGUGGUGUGG
10	C-MYC PmlI top	CACGTGGAACAGAAGCTGATCTCAGAGGAGGACCTGCACGTG
11	C-MYC PmlI bottom	CACGTGCAGGTCCTCCTCTGAGATCAGCTTCTGTTCCACGTG

Figure S1

A



B

```
RRM1     14  RKLFIGGLSFETTDESLRSHFEQWGTLTDCVVMRDPNTKRSRGFGFVTYA   63  Hs hnRNP A1
              .::.                                      :: ::
RRM1    125  TRLFVRPFPLDVQESELNEIFGPFGPMKEVKIL--------NGFAFVEFE  166  Sc Npl3
              RNP-2                                     RNP-1

RRM2    106  KKIFVGGIKEDTEEHHLRDYFEQYGKIEVIEIMTDRGSGKKRGFAFVTFD  148  Hs hnRNP A1
              .: .  .                                   : . . :
RRM2    200  YRITMKNLPEGCSWQDLKDLARENS-LETTFSS--VNTRDFDGTGALEFP  246  Sc Npl3
              : . ::                                    :   . .
RRM      46  STIYVGNLSFYTSEEQIYELFSKCGTIKRIIMGLDRFKFTPCGFCFIIYS   95  Sc Cbc2
              RNP-2                                     RNP-1
```

C

```
194  SSQRGRSGSGNFGGGRGGGFGGNDNFGRGGNFSGRGGFGGSRGGGGYGGSGDGYNGFGNDGSNFGGGGSYN
     :.. :  : : : ::  ::: :   : :::    :::::: ::: : : :  :: :.   :     ::  .
284  SNRGGFRGRGGFRGGFRGGFRGG--FSRGGFGGPRGGFGGPRGGYG-GYSRGGYGGYSRGGYGGSRGGYDS

      DFGNYNNQSSNFGPMKGG---NFGGRSLGPYGGGGQYFAKPRNQGG  306  Hs hnRNP A1
        : :..    .     :   :  :   : :::    .  ::   :
      PRGGYDSPRGGYSRGGYGGPRNDYGPPRGSYGGSRGGYDGPRGDYG  396  Sc Npl3
```

Figure S1. Mammalian hnRNP A1 and hnRNP-related proteins in yeast, Npl3 and Cbc2, are very similar RNA processing proteins. (A) They all share RNA recognition motifs and glycine/arginine/arginine-rich regions. hnRNP A1 also has nuclear localization sequences. **(B)** The amino acid sequence alignments of protein domains in human hnRNP A1, yeast Npl3, and yeast Cbc2 show highly conserved RNA recognition motifs. The RRMs, in particular, contain two well-conserved short submotifs, the RNP-1 octamer and the RNP-2 hexamer (Birney et al, *Nucl. Acid Res.*, 1993). **(C)** The RGG regions between hnRNP A1 and Npl3 are 38.5% identical and 45.3% similar as determined by the Matcher program (Mobyle, Pasteur Institute).

Figure S2

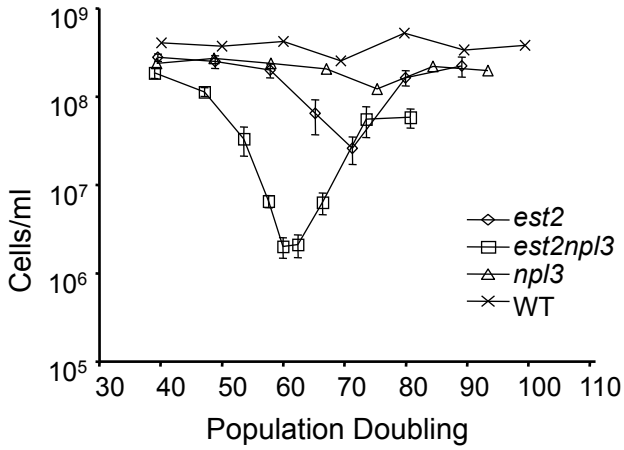


Figure S2. Deletion of *NPL3* accelerated the senescence of *est2* mutants. *est2::URA3* (from Open Biosystems) and *npl3::Kan*^{R} diploids were mated to generate yJL340 diploid cells. After sporulation and dissection, haploid cells were maintained in YPAD for the senescence assay. Population doublings were determined daily for 2x10^6 cells after 22 hours of growth. Error bars for represent standard error of the mean, $N = 5$ for *est2* and *est2 npl3* mutants and $N = 2$ for WT and *npl3* cells.

Figure S3

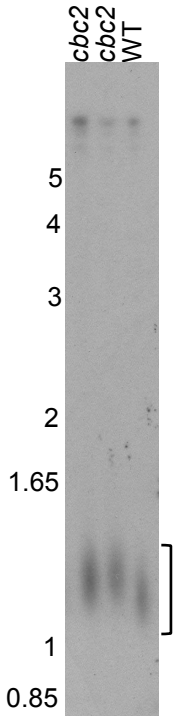


Figure S3. At steady state, *cbc2* mutants have longer telomeres than WT cells. DNA from *cbc2* and *CBC2* cells were extracted, digested, separated on gel, and blotted. Telomere fragments (indicated by bracket) were hybridized with ^{32}P labeled 784 bp PCR-generated Y' fragment. Ladder marker (in kb) is indicated on the left.