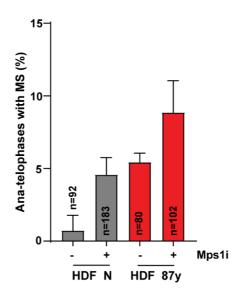
Appendix for Barroso-Vilares et al., Small molecule inhibition of aging-associated chromosomal instability delays cellular senescence (2020)

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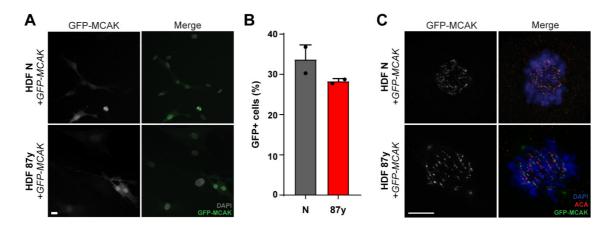
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Appendix Figure S1 (related to Fig 1). Increased frequency of chromosome missegregation in elderly vs. neonatal cells following Mps1 partial inhibition.

Percentage of ana-telophases with chromosome mis-segregation (MS) in neonatal vs. elderly cells treated with DMSO (-) or 500 nM AZ3146/Mps1i (+) for 4 hours as scored by Fluorescence *in situ* hybridization (FISH) using centromeric probes against chromosomes 7, 12 and 18.

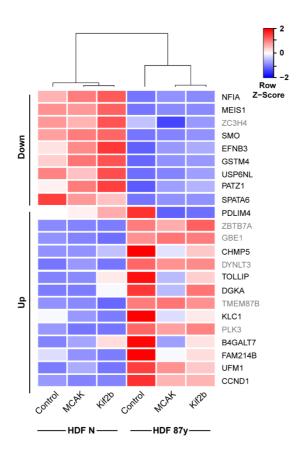
Data information: values shown are mean \pm s.d. of at least two independent experiments.



Appendix Figure S2 (related to Fig 2). GFP-MCAK overexpression in young and elderly cells.

- (A, B) Representative images (A) and percentages (B) of GFP-MCAK positive cells in total cell populations of neonatal (HDF N) or elderly (HDF 87y) after lentiviral transduction. Scale bar, $20\mu m$.
- (C) Representative images of mitotic cells overexpressing GFP-MCAK in neonatal and elderly cell populations transduced with lentiviruses. Scale bar, $5\mu m$.

Data Information: Values shown in (B) are mean \pm s.d. of two independent experiments, which are indicated as dots.



Appendix Figure S3 (related to Fig 3). Kinesin-13 overexpression rescues senescence core gene expression signature in elderly fibroblasts.

Heatmap of differentially expressed senescence core signature genes (see Dataset EV3). Down and Up refer to the expected changes reported for senescent cells. Gene symbols highlighted in grey indicate genes whose expression remained unaltered following Kinesin-13 overexpression. Z-score row color intensities indicate higher (red) to lower (blue) expression.

Appendix Table S1. List of primers used for lentiviral plasmid assembly and quantitative PCR.

Gene/Fragment name	Gene Symbol	Forward primer (5' to 3')	Reverse primer (5' to 3')	Experiment
BamHI-GFP-MCAK-NotI	-	TATCGCGGATCCATGGTGAGCA AGGGCGAGGAG	TATATAGCGGCCGCTCACTGGGGC CGTTTCTTGCTG	LP
NotI-GFP-Kif2b-MluI	-	TATATAGCGGCCGCATGGTGAG CAAGGGCGAGGAG	TATACGACGCGTTCACTCTACCTT GCTCTTCA	LP
BamHI-EOS-α-tubulin-NotI	-	CGCCGCGGATCCATGAGTGCGA TTAAGCCAGAC	TATATAGCGGCCGCTTAGTATTCC TCTCCTTCTTCC	LP
Aurora kinase B	AURKB	CGCAGAGAGATCGAAATCCAG	AGATCCTCCTCCGGTCATAAAA	qPCR
Beta-1,4-Galactosyltransferase 7	B4GALT7	CACTACCGGCTGTGCAATG	ACCCAGTTGTGATTCCCGAG	qPCR
Cyclin D1	CCND1	AGCTGTGCATCTACACCGAC	GAAATCGTGCGGGGTCATTG	qPCR
Cyclin-Dependent Kinase Inhibitor 1A	CDKN1A/P21	TGGACCTGGAGACTCTCAGG	CGGATTAGGGCTTCCTCTTGG	qPCR
Cyclic GMP-AMP synthase	CGAS	ACGTGCTGTGAAAACAAAGAAG	GTCCCACTGACTGTCTTGAGG	qPCR
Family With Sequence Similarity 214 Member B	FAM214B	CACCATCCAAGTGACCTTATTTA ACC	AGAAGTCAAAGGTCACAAGGAAC AT	qPCR
Hypoxanthine phosphoribosyltransferase 1	HPRT1	TGACCAGTCAACAGGGGACA	CTGCATTGTTTTGCCAGTGTCAA	qPCR
kinesin family member 2C	KIF2C/MCAK	CTCAGTTCGGAGGAAATCATGTC	TGCTCTTCGATAGAATCAGTCAC	qPCR
NDC80 kinetochore complex component	NDC80/HEC1	CCTCTCCATGCAGGAGTTAAGA	GGTCTCGGGTCCTTGATTTTCT	qPCR
Polo like kinase 1	PLK1	AAAGAGATCCCGGAGGTCCTA	GGCTGCGGTGAATGGATATTTC	qPCR
TATA-box binding protein	TBP	GAGCCAAGAGTGAAGAACAGTC	GCTCCCCACCATATTCTGAATCT	qPCR
Toll Interacting Protein	TOLLIP	GACTTCCTCCGCATCACGC	CCAACTTTGCCTGTACCACC	qPCR

LP - lentiviral plasmid assembly; qPCR - quantitative PCR